VIRULENCE OF ENTOMOPATHOGENIC NEMATODES AGAINST FRUIT FLY, BACTROCERA ZONATA LARVAE
[VIRULENCIA DE NEMATODOS ENTOMOPATOGÉNICOS CONTRA LA VUELA DE FRUTA, BACTROCERA ZONATA LARVAE]

Aatif, H. M., C. M. S. Hanif, M. Ferhan, K. Ali and M. Yasin. College of Agriculture, BZU, Bahadur sub Campus, Layyah. aatif.pak@bzu.edu.pk

The peach fruit fly, Bactrocera zonata (Saunders) (Diptera: Tephritidae), is an economically important insect pest of citrus fruits, especially Kinnow, which is widely cultivated in Pakistan. At larval stage, the fruit fly destroys fruits by interior feeding and results in high economic losses. A laboratory study was carried out to investigate the pathogenicity of four entomopathogenic nematode (EPN) species viz. Steinernema asiaticum, Heterorhabditis bacteriophora, Steinernema pakistansis, and Heterorhabditis indica at three different concentrations 50, 75, and 100 IJs/µL, against 3rd instar larvae of peach fruit fly. The larvae were released in Petri dishes containing sterilized soil treated with respective doses of EPNs. Mortality data was recorded after 2, 3, and 10 days post exposure. The results indicated that all the EPN species were effective at either concentration for controlling fruit fly as compared to control treatment. The exposure time and EPNs concentrations also had a significant influence on the mortality rates of fruit fly. The highest mortality rate was recorded after 10 days as compared to 2 and 3 days. The percent mortality of fruit fly larvae was found higher (96.47%) with application of H. bacteriophora at 100IJs/µL followed by S. pakistansis (96.46%) and S. asiaticum (91.16%). The lowest mortality (85.87%) was achieved due to application of H. indica. In crux, the use of EPNs to control fruit fly is a novel approach in Pakistan to reduce the fruit fly induced yield losses in citrus.

NEMATOLOGY IN SOUTH AMERICA
[NEMATOLOGIA EN AMERICA DEL SUR]
Aballay, E. Facultad de Ciencias Agronómicas, Universidad de Chile, Casilla 1004, Santiago, Chile. eaballay@uchile.cl

Nematology in South America has shown a stagnated growth over the last 10 years, based in a low or non-existent incorporation of new candidates or professionals to the different areas of the discipline in the region’s countries. However, a clear development has been observed in some new areas, with people joining the study of entomopathogenic nematodes or biological control of plant-parasitic nematodes. In all South American countries, nematologists work mainly in public institutions, and secondarily in private companies, but in some countries the number is very low or almost non-existent, which is the case of Uruguay and Paraguay, respectively. The other countries have a number of nematologists ranging from 6 to 15, except for Brazil, with the highest number. Political authorities do not show interest in the creation of new positions in nematology to be incorporated in government ministries, but only to replace some retired professionals. On the other hand, universities and state research centers have, in general, maintained the number of nematologists, but the total number has decreased in the last 20 years. An increase in the number of specialists may occur in private companies advocated to farming services, as soil sampling, nematode analysis, and new chemical
assessments, but there is no contribution to research or extension activities. A positive vision for the future of nematology would be the development of activities that call the attention of institutions, professionals, and companies, such as the search of new pest control methods through the use of entomopathogenic nematodes, nematode identification based on molecular methods, and biological control of plant parasitic nematodes. Conversely, there is little interest by employers in general in areas such as agricultural monitoring or classical taxonomy. The lack of specific nematology courses at universities has been identified as an important factor for the lack of development of this discipline, since students do not have the right vision of the importance of nematodes in agricultural systems.

**RHIZOBACTERIA TO IMPROVE SOIL SUPPRESSIVENESS IN ORCHARDS AND VINEYARDS [RIZOBACTERIAS PARA MEJORAR LA SUPRESIVIDAD DE LOS SUELOS EN HUERTOS FRUTALES Y VIÑAS]**

Aballay, E., and S. Prodan. Facultad de Ciencias Agronómicas, Universidad de Chile, Casilla 1004, Santiago, Chile. eaballay@uchile.cl

The presence of suppressive soils is an important characteristic that may help to keep plant-parasitic nematode populations under economic thresholds and avoid damages to root and other underground structures. The identification of the soil biotic and abiotic factors associated to this property may be very useful to protect and stimulate this soil characteristic or may help to isolate and introduce it in other soils, where the presence of pathogens is not under a natural control. The presence of rhizobacteria is one of the biological factors that may be related to this soil characteristic since it has been shown that its presence is associated with healthy root systems in grapevine plants growing under replant conditions, with no rootstocks, in soils from different areas. The relationship between these rhizobacteria and other soil properties or microorganisms present in that soil is not known, but it seems that these rhizosphere-associated bacteria are able to decrease nematode populations and/or root damage. Several genera and species have been isolated from root zones and also identified, which has shown that some of them are present in different places along Chile. Also, some metabolites have been identified. To corroborate their impact in soil suppressiveness, isolated rhizobacteria have been cultivated, mixed and used to inoculate root systems and soils of new plants, grapevines, cherries, and citrus, showing that nematodes and damages are fewer than non-inoculated plants. It may help to maintain these soils with a group of antagonists or growth promoters for a long term, since rhizobacteria population is associated with living roots. Several species from the genera Bacillus, Brevibacterium, Stenotrophomonas, and Pseudomonas have been associated with nematode suppressiveness.

**MANAGEMENT OF PHYTOPARASITIC NEMATODES IN GARLIC IN THE AREQUIPA REGION, PERU [MANEJO DE NEMATODOS FITOPARÁSITOS EN AJO EN LA REGIÓN DE AREQUIPA, PERÚ]**

Anculle, A. Department of Plant Health, Faculty of Agronomy, National University of San Agustin, Arequipa, Peru. albertoanculle@gmail.com

In Arequipa, Peru, the cultivation of garlic is very important because, annually, more than 4,300 ha are installed (56% of the national total). In 1985 garlic was imported from Chile and Argentina and in 1987 plants with damage produced by Ditylenchus dipsaci were found, which was reported as a new disease in the crop in 1992. The presence of this nematode generated great concern in the producers, because losses of up to 100% of production were recorded; lower income; soil contamination; attack to other crops and few management options. The activities for the management of this disease were centered, mainly in the Tambo Valley, where the greatest damage of D. dipsaci was recorded and where more than half of the garlic cultivated in Arequipa is installed annually. Activities for integrated management were also extended in other areas such as the Arequipa countryside and La Joya Irrigation. Several actions were carried out oriented to the cultural, biological, chemical management, to the determination of efficient hosts and to the evaluation of the level of economic damage of D. dipsaci whose results were exposed in national and international congresses. A capacity-building program was carried out for farmers in the Tambo valley with
the intervention of Peruvian and foreign experts. The results of these actions led to the use of a thermal treatment plant, as well as chemical treatment of the seed tooth, before planting; to the selection of mother bulbs; to change cultivation practices such as irrigation in the Tambo Valley; and to internalize the coexistence with the nematode. On the other hand, legal provisions were issued prohibiting the importation of garlic and onion from Chile and Argentina because of the risk of being carriers of some races of *D. dipsaci* and another declaring *D. dipsaci* as a quarantine important pest for the country, located only in garlic and onion in the Department of Arequipa, and that the import of onion and garlic bulbs must come from *D. dipsaci*-free areas. In 1998, a mapping of *D. dipsaci* was also carried out in production areas to determine free areas that serve as certified nurseries, the results of which contributed to the management of *D. dipsaci*. At present, although the nematode has not been eradicated, the damage levels in the field have drastically decreased, reaching an incidence of less than 1% on average. Farmers select bulbs with no symptoms for use as seed or buy seed from localities with very low incidence of the disease. They also apply heat treatment and nematicides to the seed tooth before planting. The subsequent application of nematicides is done both to the foliage and to the soil during cultivation. The sustainable management of the crop in Arequipa has been achieved thanks to a coordinated work between public and private institutions linked to the agricultural sector and garlic producers.

**SUPPRESSION OF THE ROOT-KNOT NEMATODE (MELOIDOGYNE SP.) WITH CHEMICAL-ORGANIC PRODUCTS IN ONION CROP [SUPRESIÓN DEL NEMATODO ROOT-NUDO (MELOIDOGYNE SP.) CON PRODUCTOS QUÍMICOS-ORGÁNICOS EN LA CULTIVA DE LA CEBOLLA]**


The management of root-knot nematodes is a complex process in any farming system of susceptible plants, since most of the products not fumigants do not have current registration for vegetables. In the last five years, very promising results were obtained with the application of flusulfone, a product of the fluoroalkenyle group, and offering a replacement alternative to the traditionally used chemical nematicide and generate high detrimental impact over the environment and human health. We evaluated three doses of FSF (175, 2.0, and 2.25 L / ha), one of fenamiphos (7.0 L / ha) and untreated plants were included as controls. All treatments were established under a design of complete blocks with three replications randomly. The flusulfone in doses of 2 to 2.25 L significantly reduced populations and damage caused by *Meloidogyne* sp. 60 DAS, plots treated with FSF in doses of 2 to 2.25 L.ha⁻¹ and fenamiphos (7 L.ha⁻¹) showed lower damage with 6.33-9.00% of damping on average, reducing the damping in onion plants by more than 260%. The dose of 1.75 FSF L.ha⁻¹ reduced the damage in a 114% compared to control plants. Commercial bulbs production was greater in plots treated with FSF (2.0-2.25 L.ha⁻¹) and fenamiphos (7 L.ha⁻¹) with 5.9, 6.0 and 5.9 kg.m⁻², increasing performance by 11-14% relative to the control, respectively. The main advantages of the FSF is that it acts as a true nematicide, its control effect can last up to 60 days with low populations, is applied through the irrigation system prior to planting, and is not phytotoxic to the crop if it is applied following the instructions on the label.

**PHYLOGENETIC RELATIONSHIPS AMONG MEXICAN POPULATIONS OF NACOBUS ABERRANS (NEMATODA, PRATYLENCHIDAE) REVEAL THE EXISTENCE OF CRYPTIC (COMPLEX) SPECIES [RELACIONES FILIOGÉNICAS ENTRE POBLACIONES MEXICANAS DE NACOBUS ABERRANS (NEMATODA, PRATYLENCHIDAE) REVELAN LA EXISTENCIA DE ESPECIES CRÍPTICAS (COMPLEJOS)]**

The plant-parasitic nematode *Nacobbus aberrans* is an agricultural pest of quarantine importance. The taxonomic status of the nematode has been the subject of controversy due to wide degree of variation exhibited by the species. This variability has led to the suggestion that *N. aberrans* is actually a species complex rather than a single species. Nevertheless, all previous studies revealed a limited genetic variability in Mexican populations. The objective of this study was to analyze, for the first time, the Cytochrome c Oxidase Subunit 1 (COI) gene to investigate its variability in 15 Mexican populations of *N. aberrans* and to analyze the phylogenetic relationships of *N. aberrans* with other plant-parasitic nematodes. COI sequences revealed significant intraspecific variation and the phylogenetic inference showed the formation of two separate groups with different levels of intragroup variability. These results indicate the possibility of the existence of more than one group of *N. aberrans* species in Mexico. COI phylogenetic analyses of *N. aberrans* with related plant-parasitic nematodes confirm its proximity to both cyst and root-knot nematodes.

**CONTROL OF PROARNA BERGIE (CHICHARRA) IN COMMERCIAL FIELDS OF ASPARRAGUS USING HETERORHABDITIS SP. [CONTROL DE PROARNA BERGIE (CHICHARRA) EN CAMPOS COMERCIALES DE ESPARRAGO UTILIZANDO HETERORHABDITIS SPP.]**


Commercial fields of asparagus located in La Libertad, Peru, have a high incidence of the phytophagous *Proarina bergi* Distant. (181 nymphs / m³), whose nymphal stages are located in the roots causing progressive weakening, lower production and wounds that favor the entry of soil pathogens. The chemical control of this pest is not efficient because the insect deepen into the soil, which implies using high doses and high application volumes with the risk of accumulation of pesticides. The entomopathogenic nematodes (NEPs) due to their wide range of action, high virulence and great search capacity are a promising alternative in the regulation of pests that perform part of their life cycle in the soil. In this context, the objective of this investigation was to evaluate the effect of the application of *Heterorhabditis* sp. on the control of *P. bergie*. 3000 juveniles of *Heterorhabditis* per hectare were applied. For the application the infected larvae were manually crushed by adding water, then the homogenized was filtered, taking it to a total volume of 200 liters and injected through the drip irrigation system using a stationary pump. After the application, irrigation was continued for 25 minutes to bring the juveniles closer to the root zone, in order to facilitate the search for the insect nymphs. The number of nymphs / m³ and the number of parasitized nymphs were recorded weekly. One month after the application, 26% of parasitized nymphs were obtained per sampling point, and this percentage progressively increased to levels that fluctuated between 48% and 62%.

**NEMATOLOGY IN CENTRAL AMERICAN COUNTRIES, A GENERAL OVERVIEW OF THE PRESENT SITUATION [NEMATOLOGÍA EN LOS PAÍSES DE CENTROAMÉRICA, UNA VISIÓN GENERAL DE LA SITUACIÓN ACTUAL]**

**Chaverri, F.,** V. Bravo, F. Ramírez, S. Berrocal, M. Orozco, and L. Córdoba. IRET, Universidad Nacional, 86-3000, Heredia, Costa Rica. fabio.chaverri@una.cr

Pesticide use in Central American agriculture has been promoted as an important tool for development for decades despite being a hazardous technology for human health and the environment in many cases. At the same time that a number of industrialized countries are undertaking significant steps to reduce pesticide consumption, developing countries are becoming a more important marketing target. In addition to high volume and toxicity of pesticides in use in Central America, tropical conditions and usage practices can also increase environmental and human health risks. Collateral damage of pesticides has been attributed primarily to
insufficient regulation and failure on technical assistance of pesticide users in developing countries, and it has been assumed that strengthening of regulations and education to users would lead to an acceptable level of pesticide safety. Recent studies by the IRET (National University of Costa Rica), estimate that Central America imports around 1,600 tons of nematicides per year, which accounts for about 5% of the total import of pesticides in the area. An important issue is that the agricultural area remains almost the same size over the last 5 years, around 7.6 x 10^6 ha (FAO-STAT). The use of chemical nematicides remains high in the Central American region, especially for exportation crops with intensive agricultural practices. The study of the water bodies downstream of these agricultural lands shows consistent results of pesticide concentrations and effects on aquatic organisms and communities. Many imported chemical nematicides are of special environmental and human health concerns, including 1,3-D, terbufos, cadusafos, ethoprop, among others. Some of them are included on the Hardly Hazardous Pesticides list (HHP). Recent studies among farmers show increased knowledge and risk awareness without substantial changes in pesticide handling. Continuous poisonings and environment contamination with pesticides were reported to the surveillance systems. Alternative approaches are needed, and successful examples exist like Integrated Pest Management and biological control programs. Although there are successful cases of substitutions of dangerous pesticides and alternatives, there are still failures in implementation such as lack of plant-parasitic nematology extension, research services, and educational programs. It is necessary to make profound changes in international and national agricultural policies and steer towards sustainable agriculture, hazardous substances as the chemical nematicides require different levels of control in their manufacturing, transportation, storage, handling, use, and disposal to manage the potential risk properly, especially in the Central American area, one of the most vulnerable zones to the effects of climate change around the world. The main emerging concern of the Central American region is the reduction in some companies of personnel specialized in nematology. This concern is based on the next future needs, especially because the main goal of this topic seems to be to develop innovative research projects, tools, and technologies for nematode surveillance over large geographic regions, applicable or adaptable to multiple crops and disease/pest complexes.

**CURRENT STATUS AND FUTURE PERSPECTIVES FOR NEMATOLOGY RESEARCH IN EUROPE [SITUACIÓN ACTUAL Y PERSPECTIVAS FUTURAS PARA LA NEMATOLOGIA EN EUROPA]**

Ciancio, A. Istituto per la Protezione Sostenibile delle Piante, CNR, Via G. Amendola 122-d, 70126 Bari, Italy. aurelio.ciancio@ipsp.cnr.it

European nematology science, as many other information-based systems, may be considered to evolve under two selective pressures. These are characterized by the need to: 1) transfer the knowledge produced to the new generations and 2) integrate/update the achievements produced by those of other scientific disciplines. However, more than one century of research on nematodes often has led european nematology to face two alternative options: extinction or adaptive change. Extinction has seen the loss of renowned nematology groups. Other research groups escaped extinction by adapting to critical issues such as reduced fund allocations, integrating themselves in broader, multi-disciplinary scientific initiatives. Similarly, the reduction of nematology journals has been balanced by publishing in journals less specialized, but with a broader audience. Research topics still focus on agriculture and zoology, including taxonomy, management and biocontrol, ecology, plant protection, genetics, and resistance. These themes have been wisely integrated by many -omics approaches, including genomics, transcriptomics, and metagenomics. Actual perspectives reflect the need of the society at large to find innovative answers to old problems, including the development of integrated approaches for safer food and industrial crop productions, as well as the search for effective and sustainable management systems. Research funding mostly shifted to trans-national initiatives, including EU-funded cooperative projects with nematode keywords. Future events such as the 2020 International Nematology Congress account for the sector vitality, and represent an opportunity to
reinforce international cooperation and scientific integration.

MICROBIOME-BASED SERVICES FOR SUSTAINABLE NEMATODE MANAGEMENT [SERVICIOS BASADOS EN MICROBIOMAS PARA NEMATODO SOSTENIBLE ADMINISTRACIÓN]

Ciancio, A., and L. C. Rosso. Mariantonietta Colagiero and Isabella Pentimone Istituto per la Protezione Sostenibile delle Piante, CNR, Bari, Italy. aurelio.ciancio@ipsp.cnr.it

Studies based on -omic approaches highlighted mechanisms involved in regulation of phytonematode by soil and rhizosphere microorganisms. Time series and population modelling may describe a single microcosm, but increasing complexity is needed to account for the soil interactions or functional redundancies. Regulatory functions often rely on a few microbial antagonists through density-dependent mechanisms, but external drivers such as nematicides applications may also affect belowground microorganisms and species composition profiles. The interactions of nematicides and nematodes with soil bacteria showed shifts in biodiversity, i.e. root-knot nematodes and fenamifos showing an enrichment in the soil metabolic capacities. Experimental data showed changes of bacterial diversity that occurred at finest taxonomic levels, involving unclassified OTUs whose clustering mirrored the soil conditions applied. Further studies on the hyphomycete Pochonia chlamydosporia highlighted its potential as a growth promoter, through a differential gene expression and root re-programming. Its metabolism as an egg parasite contrasts with the plant defense response it was found to elicit. This function reduces the nematode numbers on roots limiting the fungus food source, indicating a complex rhizosphere role. Specialized biocontrol agents such as Pasteuria spp. showed efficient nematode regulation. In field conditions, however, it is questionable whether a single biocontrol agent only may be functionally significant for nematode regulation and management. Biodiversity conservation and farm productivity have been often in conflict in intensive cropping systems or monocultures, and appropriate management assumptions are needed to sustain yields and food production in the long term.

APPLICATION OF NANOPORE TECHNOLOGY FOR FAST SEQUENCING OF VERY LONG DNA READS FROM PLANT-PARASITIC NEMATODES [APLICACIONES DE LA TECNOLOGIA NANOPORE PARA EL SECUENCIAMIENTO RAPIDO DE FRAGMENTOS DE ADN MUY LARGOS DE NEMATODOS FITOPARASITICOS]

Ciancio, A., L. C. Rosso, M. Colagiero, and I. Pentimone. Istituto per la Protezione Sostenibile delle Piante, CNR, Via G. Amendola 122-d, 70126 Bari, Italy. aurelio.ciancio@ipsp.cnr.it

Nanopore sequencing is a promising, portable, and easy-to-use technology allowing the sequencing of very long DNA fragments (from several kilo-bases up to megabases). To check its potential in plant protection, we tested a nanopore device for identification of plant-parasitic nematodes. The samples tested were DNA extracts obtained from five juveniles of Meloidogyne incognita or two juveniles of Xiphinema diversicaudatum. Hand-picked nematodes were extracted from soil, washed, ruptured, and digested in test tubes with a DNA extraction and amplification kit. The preparation was then processed as required by the manufacturer, adding the adapter leader and terminus loop sequences, without DNA fragmenting. The constructs were then sequenced for up to 48 hr through a MinION™ device (Oxford Nanopore, UK) using one flow cell per sample, with remote basecalling. The sequences were extracted from the FAST5 data files that passed the basecalling quality control (5700 for M. incognita and 733 for X. diversicaudatum). Preliminary online BLAST analyses showed a number of matching reads with sequences of Nematoda, for both samples. Sequences included a 3121 nt long read matching a region (9% query cover) in accession AF387097 from M. incognita, and a 12808 nt long read, matching a region (3% query cover) in accession JQ780335, for a mitochondrial NADH dehydrogenase of X. diversicaudatum. The potential of nanopore sequencing in plant-nematode diagnostics and genome analysis is high, although actually limited
by the database coverage and availability of sequenced genome data.

**OPPORTUNITY, CHALLENGE, AND EXPERIENCE REQUIRED TO PRODUCE A NEW GENERATION OF NEMATOLOGISTS [OPORTUNIDAD, RETO Y NECESIDAD EN LA FORMACIÓN DE NEMATOLOGOS JÓVENES POR LA ONTA]**

_Cid del Prado, V. I._ Colegio de Postgraduados, Mexico, 56230. icid@colpos.mx

In Latin America there are 74 regular members of the Organization of Tropical America Nematologists (ONTA) but only 11 are directly involved in teaching and research in Nematology. There is a great need in Latin America to develop a new generation of young nematologists with the training, skills, and passion to develop into effective teachers, researchers, plant health consultants, agribusiness personnel, and advisors to agricultural producers. An important role for the experts in our organization is to provide training courses, usually intensive short courses, in nematology in Latin American countries. My experience in teaching and participating in such courses in Central and South America convinces me that in every country there are students and early-career professionals who are eager to receive training in nematology and that there are many career opportunities for students who have participated in the courses. ONTA members have the necessary skills to provide appropriate training courses, especially when supported by official institutions and private companies. I recently taught such a course at the National University of Trujillo, Peru, where I enjoyed working with enthusiastic young participants who were committed to learning basic techniques and the recognition and identification of plant-parasitic nematodes. During the course, we took samples from a range of crops, including artichoke, asparagus, chili, fruit trees and vines. We identified the major plant-parasitic nematodes in the samples and discussed management approaches. The challenge to ONTA members is to offer official or capacitation courses, supported by instructional materials, to fulfill this essential educational activity.

**OPPORTUNITY, CHALLENGE AND NECESSITY TO SUPPRESS NEMATOIDE GALLS IN HORTICULTURE CROPS [OPORTUNIDAD, DESAFÍO Y NECESIDAD PARA SUPRIMIR GALLES DE NEMATOIDE EN CULTIVOS DE HORTICULTURA]**

_Cid del Prado V. I._ Colegio de Postgraduados, Mexico, 56230. icid@colpos.mx

Some of the major nematodes of economic importance in vegetable crops are the gall nematodes: _Meloidogyne_ spp. and _Nacobbus aberrans_, which are related to yield and growth reductions, deformation of the host crop, and economic loss. Differences in nematode damage recognized are plant cultivar, soil type, and climatic conditions. Damage, given as percent yield reduction, depends also on initial population density and the disease complexes involving other plant pathogens. There is considerable literature on cropping systems involving gall nematodes control; included are: cultural practices, biological control, added organic and inorganic amendments to soil to improve fertility and crop yield, the use of nematicides, and intercropping systems. Some of these amendments are known to contain substances toxic to nematodes when added to soil in sufficient quantity; they suppress respiration or movement of nematodes directly or in combination with chitinolytic ability to destroy nematode eggs. The activity of these compounds depends on the type of extract (methanolic, ethanolic, or watery) or the type of glucosinolate and the kind of breakdown products of it such as isothiocyanate or thiocianate. Numerous reviews have addressed the occurrence of glucosinolates in vegetables, primarily the family Brassicaceae (syn. Cruciferae); including _Brassica_ spp. and _Raphanus_ spp, with nematicidal effects. Many glucosinolate-containing genera in plants are currently being investigated for their fungicidal, bacteriocidal, nematocidal and allelopathic properties. It is of more than academic interest to re-examine some of the “non Brassica” glucosinolate-containing plants for their potential as nematicidal properties. Addressing these future challenges for integrated nematode management will depend on sufficient basic and applied nematological research program to insure the sustainability of horticultural crops.
CONTRIBUTION TO THE KNOWLEDGE OF THE GENUS *GEOMONHYSTERA* (NEMATODA: MONHYSTERIDA) [CONTRIBUCIÓN AL CONOCIMIENTO DEL GENUS *GEOMONHYSTERA* (NEMATODA: MONHYSTERIDA)]

Cid del Prado, V. I.¹, H. Ferris², and S. Subbotin³. ¹Colegio de Postgraduados, Montecillo 56230, México. ²Department of Entomology and Nematology, University of California, Davis, CA 95616, USA. ³Plant Pest Diagnostics Center, California Department of Food and Agriculture, Sacramento, CA 95832, USA. icid@colpos.mx

Species of the genus *Geomonhystera* are terrestrial nematodes with smooth to finely-striated cuticles, which may have somatic setae distributed along the body. The six triangular lips surrounding the stoma are not fused. The anterior sensilla are in three whorls, six circum-oral inner labial papillae, which are located at the bases of the lips, six outer labial setae, which may be articulated or segmented by a joint near the base, and four cephalic setae which may be similarly segmented at the base. The esophagus is cylindrical without terminal swelling and separated from the intestine by a cardium. The rectum is unusually strong and muscular, more than one anal body diameter long. The vulva is located at 75-85% of body length. Males are unknown in many species. The spicules are ventrally arcuate, usually less than 1.5 times anal body diameter but occasionally longer. The cuticle is wrinkled in the precloacal region of male. The tail is ventrally curved. A scanning electron microscope study of six new and one known species of *Geomonhystera* from Mexico and Ecuador revealed, in these species, very fine striation of the cuticle, the distribution and sizes of somatic setae, that the lips are separate, that the outer labial and cephalic setae are not segmented and that males, when present, have wrinkled cuticle in the precloacal region.

SUSCEPTIBILITY OF TRANSGENIC SOYBEAN CULTIVARS TO *MELOIDOGYNE JAVANICA* IN NORTHWEST ARGENTINA [SUSCEPTIBILIDAD DE LOS CULTIVARES DE SOJA TRANSGÉNICA A *MELOIDOGYNE JAVANICA* EN EL NOROESTE DE ARGENTINA]


Root-knot nematode (*Meloidogyne javanica*) is the most prevalent plant-parasitic nematode in soybean in Northwest Argentina, often causing significant losses to this crop. Recently, transgenic soybean cultivars (Intacta RR2 Pro®) were released with resistance to glyphosate and caterpillars and their planting area is increasing. The response of these new varieties against phytoparasitic nematodes is mostly unknown. Therefore, the objective of this study was to evaluate the reaction of Intacta RR2 Pro® soybean genotypes to *M. javanica* under greenhouse conditions. Ten Intacta RR2 Pro® soybean cultivars (CZ 5905, CZ 7905, DM 63i64, DM 8277, Ho 6620, MS 6.3, MS 6.9, SPS 6x8, SYN 7x1, SPS 7x8) were evaluated in a completely randomized design with six replicates per variety, four glyphosate resistant soybean (RR1) (DM 62r63, DM 8473, CZ 6505, NS 7809) also were included in this evaluation. Plants were inoculated with 1,017 eggs and second-stage juveniles of *M. javanica* per pot. A susceptible soybean cultivar was also inoculated as a check of inoculum viability. Thirty two days after inoculation the plants were removed, and the roots were carefully washed. The number of egg masses per plant was counted, and the egg mass index (EI) was assessed according to a 0-5 scale. The reaction of cultivars was determined according to Hadisoeganda & Sasser (1982). All evaluated soybean cultivars were susceptible to *M. javanica* (EI= from 4.7 to 5). These transgenic varieties of soybean are excellent hosts of *M. javanica*, so this situation should be considered in fields with the presence of this pest.

NEMATOPHAGOUS FUNGUS *POCHONIA CHAMYDOSPORIA* PERFORMS ROOT COLONIZATION AND PLANT GROWTH PROMOTION IN WINTER CROPS [NEMATOPHAGOUS FUNCHUS *POCHONIA CHAMYDOSPORIA* PERFORMA LA COLONIZACIÓN DE RAÍCES Y LA PROMOCIÓN DEL CRECIMIENTO DE
PLANTAS EN LOS CULTIVOS DE INVIERNO


The no-till agriculture system is widely used in soybean production to increase soil moisture and fertility, nutrient recycling, organic matter content and to avoid the dissemination of plant-parasitic nematodes. The production of large volumes of green mass in the off-season crops for straw formation is one of the main objectives of the use of some winter crops, so methods that promote vegetative growth of the plants are important tools to the sustainability of the no-tillage system. 

*Pochonia chlamydosporia* is a nematophagous fungus known for its ability to parasitize eggs and females of plant-parasitic nematodes, significantly reducing their populations. In addition, *P. chlamydosporia* is able to survive saprophytically in the soil and to colonize endophytically roots of several mono and dicotyledonous plant species, significatively promoting the vegetative growth of these crops. Therefore, the objective of this work was to evaluate whether the *P. chlamydosporia* var. *chlamydosporia*, isolate Pc-10, is able to colonize the poor nematode hosts, millet cultivars (ADR 300, ADR 500 and ADRG 9050), *Urochloa ruizienisis*, *Crotalaria spectabilis*, and *Stylosanthes* sp., and to promote the vegetative growth of these plants. An *in vitro* assay was performed to verify the root-colonization and a greenhouse experiment was conducted to evaluate the growth promotion of these plants by fungus. *Pochonia chlamydosporia* colonized the root systems of all the evaluated plants, and structures of the fungus could be documented within the cells of the roots, such as chlamydospores, conidia, and hyphae. The fungus increased the total fresh mass of the millet cultivars ADR 300 and ADR 500, and of the plant species *U. ruizienisis* and *C. spectabilis*, by 36.2, 32.3, 21.3, and 33.8%, respectively, and the total drymass of ADR 300 and ADR 500 by 28.4 and 17. 5%, *U. ruizienisis* by 16.1% and *C. spectabilis* by 30.1%. In addition, it increased the height of all crops. ADR 300, *U. ruizienisis* and *C. spectabilis* had their root volumes increased by 65. 6, 88.2 and 63.8%, respectively. Therefore, it was possible to conclude that *P. chlamydosporia* is able to colonize and promote the growth of all evaluated winter crops, increasing the benefits of the no-tillage system. This is the first report and proof of the colonization of this fungus in these plant species and the first visualization of chlamydospores inside plant root-cells.

POTATO CYST NEMATODE ERADICATION AND CONTAINMENT IN THE UNITED STATES: SUCCESSES AND STRUGGLES [NEMATODO QUITSE DE PAPA ERRADICACIÓN Y CONTENCIÓN DE NEMATODOS EN LOS ESTADOS UNIDOS: ÉXITOS Y LUCHAS]

Dandurand, L-M, J. B. Contina, I. A. Zasada, and X. Wang. Department of Entomology, Plant Pathology, and Nematology, 875 Perimeter Drive MS 2329, University of Idaho, Moscow, ID, USA; USDA ARS, 3420 NW Orchard Ave., Corvallis, OR 97330; USDA ARS, Robert W. Holley Center for Agriculture and Health, 538 Tower Road, Ithaca, NY. lmd@uidaho.edu

Phytosanitary measures for the exclusion, suppression, containment, or eradication of plant pests have been developed throughout the world to prevent the entry of potentially damaging pests. Phytosanitary measures work best for nematode species that have a narrow host range and a slow rate of reproduction such as the potato cyst nematodes (*PCN*), *Globodera pallida* and *G. rostochiensis*. For the US, examples of regulated nematode pests include the potato cyst nematodes, *G. rostochiensis*, which is regulated by 119 countries worldwide, and *G. pallida*, which is regulated by 80 countries worldwide. In the US, the presence of potato cyst nematodes poses a serious threat to the $4 billion potato industry, but stringent adherence to phytosanitary programs and the use of resistant potato cultivars have successfully contained *G. rostochiensis* to eight counties in New York, within fewer than 6,000 acres, despite its documented presence since 1941. The infestation of *G. pallida*, first found only in Idaho in 2006, continues to be contained to fewer than 3,000 acres which is less than 1% of the total acreage planted with potatoes in Idaho. Efforts in Idaho by both USDA APHIS (US Department of Agriculture Animal and Plant Health Inspection Service) and the Idaho State Department of...
Agriculture, have concentrated on containment, sanitation and eradication of *G. pallida*, and of the nine original fields infested with *G. pallida*, all of them no longer have active PCN infestations. The Idaho infestation has provided an opportunity to examine the spatial pattern and spread of *G. pallida*. The infestation is spatially clustered and the spread of this regional infestation grew in diameter from the original center as an ellipsoidal-shaped cluster. Results indicate that the presence of *G. pallida* in southern Idaho is unlikely to be associated with new introductions from outside the state of Idaho. For risk model development, the coefficients of tuber yield losses at different initial nematode population densities were determined and incorporated into the Decision Support System for Agrotechnology Transfer (DSSAT) model. Simulated tuber yield loss, in the absence of host resistance, was predicted to be in excess of 80% when *G. pallida* populations were moderately high (80 eggs/g soil). The use of phytosanitary measures and alternatives to fumigation for control of potato cyst nematodes will be discussed.

**IMPACT OF NEMATOPHAGOUS FUNGI ON EGGS AND JUVENILES OF MELOIDOGYNE SPP. AND FUTURE PERSPECTIVES OF ITS USE IN EXPORT CROPS IN PERU**

Delgado, M. A. and H. E. Guardia, Universidad Privada Antenor Orrego. Av. América Sur 3145, Trujillo, La Libertad, Perú. mdelgadoj@upao.edu.pe

Different species of *Meloidogyne* are widely disseminated in the agroindustrial areas of the Peruvian coast and are serious threats to agricultural production. Its rapid dispersion is associated with very susceptible crops (*Capsicum*, artichoke, grape, asparagus, sugar cane, etc.), temperatures between 14 and 30°C, sandy or sandy loam soils lacking in organic matter. For more than a decade, efforts have been made to maintain populations of nematodes below economic thresholds, through integrated management, in which chemical and biological control predominate, the latter being based on the use of *Purpureocillium lilacinus* (*P.l.*) which is one of the most tested nematophages and is formulated in various commercial products in the world. There is a great variability in the parasitic ability of *P.l.* about females and eggs of *Meloidogyne* spp. Currently, there are massive applications of this biocontroller in different agroindustrial companies in the north of Peru and recent field observations confirm that the effectiveness of this controller has decreased, and the profitability of its use has already been questioned. In this investigation the parasitic ability of a strain of *P.l.* of massive use in field was evaluated and it was grown in a medium based on Papa-Dextrose-Agar to obtain the inoculum, whose density was calibrated at three concentrations: $10^4$, $10^5$ and $10^6$ cfu/mL. The eggs of *Meloidogyne* spp were extracted from severely infested celery roots. The confrontations of *P. lilacinum* with *Meloidogyne* eggs were made in: a) Petri dishes with water agar (2%), b) Petri dishes with water-agar amended with 100 ppm of ampicillin, chloramphenicol, captan and pentachloronitrobenzene and c) Pots with 100cc of autoclaved agricultural land. Each plate received 50 and each pot 1,000 eggs of *Meloidogyne* and $10^4$, $10^5$ and $10^6$ cfu of *P. l./mL*, according to the treatment, which were arranged in a completely random design with 10 repetitions. The J2 population of *Meloidogyne* was evaluated at 7 days and at 25 days in pots. In media with agar, *P. lilacinum* reduced hatching in 16 to 37% of eggs, with an increased impact on amended agar. When the eggs and conidia of *P.l.* were discharged to sterile soil the parasitic depression of *P. lilacinum* reached only 16.6% on average, which proves that it is a less virulent strain, a situation that has already been reported by other researchers.

**INDIRECT DEPRESSIVE INTERACTION OF THERMO RESISTANT BACTERIA PRESENT IN THE ORGANIC AMENDMENT AVIBIOL ON POPULATIONS OF PLANT-PARASITIC NEMATODES**

INDIRECTA DE BACTERIAS TERMORRESISTENTES PRESENTES EN LA ENMIENDA ORGÁNICA AVIBIOL SOBRE POBLACIONES DE NEMATODOS FITOPARÁSITOS}
Avibiol is an organic amendment obtained from the anaerobic fermentation of manure from laying hens that promotes the root development of plants. New and in production banana plantations, treated with Avibiol incorporated to the soil, have shown that populations of *Meloidogyne, Radopholus, Helicotylenchus, Pratylenchus*, and *Rotylenchulus* decreased between 28 to 97%, compared with untreated plants. Tests developed "in vitro" showed that Avibiol has no depressive effect on eggs and juveniles of nematodes at concentrations applied in the field. The microbial population of this amendment was also investigated, and it was found $3.8 \times 10^4$ u.f.c. of heat-resistant bacteria / mL. Six bacterial isolates have been identified molecularly and correspond to *Solibacillus silvestris* (TR5-1), *Brevibacillus invocatus* (TR5-2), *Bacillus thuringiensis* (TR6-1), *Bacillus thuringiensis* serovar *kurstaki* (TR6-3), and *Bacillus* sp. (TR6-4). It is postulated that some or all of these isolates have the ability to develop competitively in the rhizosphere of the plants by taking root exudates, which act as electrochemical attraction substances for nematodes to be oriented toward areas of active root development. It is postulated that this nematode-root interaction is strongly weakened in such a way that nematodes die by starvation and not because of the direct effect of the Avibiol amendment.

**NON-FUMIGANT NEMATICIDES PROVIDE NEW OPPORTUNITIES FOR FLORIDA’S PLASTICULTURE [NEMATICIDAS NO FUMIGANTES PROPORCIONAN NUEVAS OPORTUNIDADES PARA LA PLASTICULTURA DE FLORIDA]**

Desaeger, J., and T. Watson. University of Florida – Entomology and Nematology Department, Gulf Coast Research and Education Center, Wimauma, FL, 33598 US

Plasticulture, or the use of drip irrigation and plastic mulch, is the most commonly used system to grow vegetables and strawberries in Florida. Fumigants remain the primary nematode management tool in plasticulture. However, fumigant labels have changed significantly in recent years, and the practice of fumigation is facing increased scrutiny due to regulatory and societal pressure. Fumigants are still largely favored by Florida growers, both because of the broad-spectrum control they provide, and due to the historic lack of non-fumigant nematicide alternatives. Fortunately, several new chemical and biological non-fumigant nematicides (e.g. flusulfone, fluopyram, fluaazindolizine, and *Burkholderia* spp.) have recently emerged, and may provide new opportunities to reduce nematode losses in Florida’s plasticulture. These new nematicides are currently being evaluated in a wide range of crops, including vegetables and strawberries, and will hopefully provide growers in Florida and elsewhere with some much-needed new and safer nematode management options. The new nematicides were evaluated by themselves, as well as in combination with fumigants, on new and double-cropped plastic beds. All applications were done via the drip irrigation system. Root-knot nematodes (*Meloidogyne* spp.), sting nematodes (*Belonolaimus longicaudatus*) and lesion nematodes (*Pratylenchus penetrans*) were the main target nematodes. In addition to the effect on plant-parasitic nematodes, we also measured the impact of these new nematicides on non-plant-parasitic / beneficial nematodes. Overall, new non-fumigant nematicides showed good potential to reduce root-knot nematodes, and showed less impact on non-plant-parasitic / beneficial nematodes. However, our field trials also demonstrated the limitations of these new non-fumigant nematicides in terms of weed and soil disease management. Therefore, in order to reduce the dependency of Florida growers on soil fumigation, it will be necessary to develop alternative sustainable and economically viable soil management plans for Florida’s plasticulture. The development of such IPM plans with reduced reliance on soil fumigants will be discussed.

**GENETIC MARKERS FOR SELECTION OF IMPROVED LINES OF THE ENTOMOPATHOGENIC NEMATODE HETERORHABDITIS BACTERIOPHORA [MARCADORES GENÉTICOS PARA LA SELECCIÓN DE LÍNEAS MEJORES DE LA NEMATODA ENTOMOFÁBICA HETERORHABDITIS BACTERIOPHORA]**


50th Annual Meeting of the Organization of Tropical Nematologists of America
The use of the entomopathogenic nematode (EPN) *Heterorhabditis bacteriophora* in large scale agriculture is often limited by environmental stresses. Breeding for nematode strains with enhanced stress resistance is thus a strategy aimed to make the use of EPNs more affordable, e.g. against the corn root borer *Diabrotica v. virgifera*. Here we report on approaches to enhance the longevity and desiccation tolerance of this EPN.

In the framework of the BIOCOMES EU project, academic and industrial partners have joined efforts to deeply study the genetic basis of environmental stress-resistance in *H. bacteriophora* dauer juveniles (DJ). Among the major outcomes of this research, a significant correlation between oxidative stress tolerance and survival in storage conditions has been found. A collection of *H. bacteriophora* wild type strains was characterized for their shelf life under oxidative stress and this property has been proposed as predictor to select for longer living nematodes. Hybrid strains and EMS-mutants with extended survival time have been tested for their general performance with satisfactory results. Genomic tools have been applied to gain insights into the mechanisms of DJ-longevity and desiccation stress tolerance in *H. bacteriophora*. Sequence information has been generated comprising of the expression of more than 20,000 different transcripts under infective- and stress-conditions. At least 11 highly-informative desiccation and DJ-longevity expression markers have been identified and validated under oxidative stress, chemical- and hygroscopic-desiccation conditions. In parallel, genotyping by sequencing (GBS) has identified more than 700 reproducible single nucleotide polymorphism (SNPs) for this species. Joining genotype and phenotype information by QTL- and association-analysis has subsequently allowed to find molecular markers linked to DJ-longevity in *H. bacteriophora*. These markers are being applied in breeding nematode lines with enhanced performance and could be applied in selection of new native lines.

*Heterorhabditis bacteriophora*, symbiotically associated with the bacterium *Photorhabdus luminescens*, is commonly used against insect pests. Dauer Juveniles (DJs) develop into self-fertilizing hermaphrodites, which lay eggs until juveniles hatch inside the uterus and feed on the body content of the mother (endotokia matricida). The life history traits (LHT) of *H. bacteriophora* were studied at 2.5 ×, 5 ×, 10 × and 20 × 10^9^ cells ml\(^{-1}\) of *P. luminescens* at 25°C using a hanging drop technique. The number of offspring produced per hermaphrodite increased from 50 at 2.5 × 10^9^ cells ml\(^{-1}\) to 269 at 20 × 10^9^ cells ml\(^{-1}\) of *P. luminescens*. At 25°C and 20 ×10^9^ cells ml\(^{-1}\) of *P. luminescens* approximately 40% of the offspring of the hermaphrodite originated from endotokia matricida and the others from eggs laid before entry into endotokia matricida. Almost 100% of the offspring of amphimictic females originated from endotokia. Data on LHT will be provided. Larvae of the invasive pest Western Corn Rootworm (*Diabrotica v. virgifera*) were infested with DJs, and the development was recorded daily by video. The occurrence of hermaphrodites, egg laying, the development to second amphimictic adults, and the endotokia matricida in the cadaver of last instars of the beetle will be commented based on the video.

**MASS PRODUCTION OF ENTOMOPATHOGENIC NEMATODES**

*Heterorhabditis bacteriophora*, Steinernema
feltiae and S. carpocapsae are currently produced in large scale bioreactors in liquid culture. The presentation will introduce into the relevant process parameters and consider the biological processes to be considered during the reproduction in monoxenic culture on the symbiotic bacteria Photorhabdus luminescens, Xenorhabdus bovenii and X. nematophilus, respectively. Downstream processing, formulation and packing systems need to be adapted to the biological potential of the biocontrol nematodes in order to guarantee high quality from ex works, to the distributor and then to the producer. A short introduction into quality control assays will be presented. The potential of biocontrol products based on EPN for the Latin-American agriculture will be discussed.

CONTROL STRATEGY FOR ROOT-KNOT NEMATODES IN TOMATO (Solanum lycopersicum) THROUGH BIOFUMIGATION AND THE USE OF Pochonia chlamydosporia IN MEXICO [ESTRATEGIA DE CONTROL PARA LOS NEMATODOS DE NUDO DE RAÍZ EN TOMATE (SOLANUM LYCOPERSICUM) A TRAVÉS DE LA BIOFUMIGACIÓN Y EL USO DE LA Pochonia chlamydosporia EN MÉXICO]  


In Mexico increased production of tomato (Solanum lycopersicum) has increased the problems caused by root-knot nematodes. For this reason, the present study evaluated the efficiency of different treatments of biofumigation complemented with the application of Pochonia chlamydosporia for the biological control of Nacobbus aberrans and Meloidogyne incognita. Four treatments with eight repetitions were evaluated, for a total of 32 experimental units (EU) with an experimental design of generalized randomized blocks. Treatments were: T1. Compost + cabbage (Brassica oleracea), T2. Cow dung + cabbage, T3. Poultry manure + cabbage and T4. Control. The treatments were incorporated into the soil, irrigated at field capacity, and covered for 30 days with plastic. Two weeks after the plastic cover was removed, tomato cv. Condor was transplanted and immediately (except the control) inoculated (except the control) with P. chlamydosporia at a concentration of 46 ml per plant (3x10^6 CFU/ml) monthly for 7 months. Before the biofumigation for each EU, a soil sample (200 g) was taken at a depth of 0-15 cm and after the biofumigation for each EU, soil samples were taken monthly for 7 months, quantifying the second-stage juveniles (J2) of N. aberrans and M. incognita. Seven months after transplantation, the following variables were evaluated: Galling index (GI), colonization of P. chlamydosporia in roots and root egg masses. As a result, the best treatment was biofumigation with poultry manure + cabbage, since it had a 50% decrease in GI compared to the control, a root colonization of 68.89% in egg masses and a colonization of more than 4 x10^7 CFU/g in root. The treatments of compost + cabbage and cow manure + cabbage showed a significant difference (SD) on the GI compared to the control, as they obtained a reduction of ±20% and in their colonization of egg masses was 63 and 55%. In J2 N. aberrans, all treatments showed a decrease in SD of ±24% compared to the control. However, they did not show SDs among them and on M. incognita J2 the treatment compost + cabbage and the treatment poultry manure + cabbage showed a decrease a SD of ±38% in contrast to the control.

CHEMICAL CONTROL OF MELOIDOGYNE SPP. IN GRAPEVINES (Vitis vinifera) [CONTROL QUIMICO DE MELOIDOGYNE SPP. EN VIDES (VITIS VINIFERA)]  

Esquivel, G., M. Tobar, E. Salas and M. Araya. Drokasa, Perú. AMVAC Chemical Corporation. 3Catedráctico Universidad Nacional, Escuela Ciencias Agrarias, Costa Rica. LIFE-RID. maraya@life-rid.com
In most of the grapevine plantations in Perú, phytonematodes usually occur in polycpecific communities, consisting mainly of a mixture of Meloidogyne incognita, Meloidogyne spp., and rarely Xiphinema index, X. americanum, Mesocricinema xenoplax, Tylenchulus semipenetrans, and Pratylenchus spp. They feed on plant roots causing extensive root damage, which reduces water and nutrient uptake, ending in yield reduction. Many of the commercial grapevine rootstocks are susceptible to nematodes including root-knot (Meloidogyne spp.). Application of insecticide-nematicides have been an important component and showed promise as alternatives for integrated grape pest management. Then, in two field trials using a complete randomized block design with 4 replicates the effect of increasing rates of 0, 6, 8, 10, and 12 L ha\(^{-1}\) of Mocap® 6EC (ethoprophos-AMVAC) on grape (Vitis vinifera cv Red Globe on Quebranta rootstock) root Meloidogyne spp. control was evaluated. To quantify nematode numbers in soil and roots, and the number of galls in a linear meter root, root and soil samples were taken just before treatment and at 30, 60 and 90 days after product application. In both trials, at 30, 60, and 90 days after the application, a decreasing linear effect on Meloidogyne spp. numbers in soil (\(P<0.0001\)) and roots (\(P \leq 0.0002\)) and number of galls (\(P<0.0001\)) was observed as rate increased. The average reduction was of 4.6, 4.9, and 5.2; and 5.5, 6.0, and 6.3 individuals per 100 g of soil, and 5.6, 9.9, and 9.9, and 4.9, 7.7, and 8.2 nematodes per 100 g of roots, and 2.0, 4.0, and 4.3, and 1.8, 3.9 and 4.9 galls per linear meter of root, by every liter of increase on the applied rate, at 30, 60, and 90 days post application, for the experiment at Ica and Lima department, respectively. Differences in biological efficacy among rates were found for soil (\(P<0.0001\)) and root (\(P<0.0001\)) nematode control, and number of root galls (\(P<0.0001\), increasing the control as the rate increased in both experiments. Efficacy in soil nematode control varied from 51 to 98% and 73 to 99%, in roots it varied from 61 to 85% and 61 to 87%, and in the number of root galls from 55 to 85%, and 78 to 81% for the experiment at Ica and Lima department, respectively. Then, the recommended rate was 10 L ha\(^{-1}\).

**RESPONSES OF** _**MELOIDOGYNE ENTEROLOBII**_ (= M. MAYAGUENESIS RAMMAH & HIRSCHMANN) **EN VARIOS ROOTSTOCKS DE** _**PSIDIUM SPP**._

Flores-Chaves\(^1\), J., M. Ávalos-Cerdas\(^1\), A. Bogantes-Arias\(^2\), D. A. Humphreys-Pereira\(^1\), and E. Mora-Newcomer\(^3\). \(^1\)Laboratory of Nematology-CIPROC, University of Costa Rica, San Pedro, Costa Rica, 2060. \(^2\)Estación Experimental Los Diamantes, Guápiles, Limón, Costa Rica. \(^3\)Estación Experimental Agrícola Fabio Baudrit Moreno, Escuela de Agronomía, Universidad de Costa Rica, San Pedro, Costa Rica, 2060.

Guava (Psidium guajava) is cultivated in Costa Rica for fresh consumption in the provinces of Alajuela and Puntarenas, and for industry mainly in Heredia, Cartago, and Limón. _Meloidogyne enterolobii_ causes severe damage on guava and can reduce yields up to 50%. This nematode species induces symptoms of yellowing followed by wilting and plant death. It was first reported on guava in 2012 with morphological and molecular methods. Since then, the nematode has been identified in all seven provinces of Costa Rica, associated with coffee, guava and acerola. Few management options are available as alternatives to nematicides. Recently, a guava breeding program was developed to search for resistance against _M. enterolobii_. Several _Psidium_ spp. (güisaro, cas and guava) were grown from seed under greenhouse conditions for 15 weeks. The _M. enterolobii_ inoculum was increased on tomato plants (cv. Hayslip) for two months. Ten replicates per _Psidium_ material were inoculated with 10,000 eggs + juveniles per plant. The reproduction factor, presence of symptoms, fresh root weight, and the galling index were evaluated after five months. A second assay was performed with the same plant materials and location. The highest fresh root weight was observed in one of the güisaro materials (_Psidium_ sp.). Cas (_P. friedrichsthalianum_) and one güisaro material showed the lowest galling index (GI=1) and reproduction factor. In contrast, most of the guava materials were susceptible to _M. enterolobii_. A study in the field was performed to analyze the population dynamics of this nematode. Four _Psidium_ spp. (10 months old) were inoculated with 10,000 _M. enterolobii_ eggs + juveniles in the greenhouse and transferred to the field after one
week. Root samples were collected every 6 months for 30 months. The results indicated low population densities of *M. enterolobii* in case (ranging from 0 to 17 nematodes/100 g of roots) and güisaro (ranging from 0 to 405 nematodes/100 g of roots). In contrast, the population levels were high in two guava materials (ranging from 873 to 42,624 nematodes/100 g and from 1,170 to 25,351 nematodes/100 g). Furthermore, severe damage was observed in the roots of the guava materials, with symptoms of “corchosis” and small galls. Cas and güisaro may provide rootstocks or a source of resistance genes against *M. enterolobii*. Grafting and breeding efforts are environmentally friendly options to reduce nematode populations.

**THE PLANT HEALTH CLINICS HELPING FARMERS TO SOLVED PHYTOSANITARY PROBLEMS IN PERÚ: THE POTATO CYST NEMATODE CASE [LAS CLÍNICAS DE SALUD VEGETAL PARA AYUDAR A LOS AGRICULTORES A RESOLVER PROBLEMAS FITOSANITARIOS EN EL PERÚ: EL CASO DE NEMATODOS DE CYST DE PAPAS]**

**Franco, J.**, J. Gómez and, Y. Colmenárez, CABI–Plantwise, Lima, Perú. javierfrancoponce@gmail.com

Plantwise, a Global program coordinated by CABI and through an agreement with INIA, has installed Plant Clinics (PCs) in 8 regions of Peru where the respective Experimental Stations of INIA are located. These PCs are assisted by a "Plant Doctor" and are established in public places. The attention to producers is made on previously established dates that bring their samples of plants with phytosanitary problems. The diagnosis and recommendations provided to farmers are recorded in a format or entered into an electronic version (e-Plant Clinics). Producers in the PCs receive management recommendations that reduce the population and incidence of detected pests and diseases. This registered information allows identifying the main crops and their phytosanitary problems. The information generated in the PCs confirms the importance of the service provide to small farmers in Peru, as a source of information of the major challenges faced by farmers at field level to design policies for research, promotion and dissemination of technology in base of that. However, in spite of PCs service for small producers, results of a study on on potato phytosanitary problems is presented. Out of 8 regions selected in Perú, results obtained during the last years in 4 departments located in the highlands of the Andean region (Cajamarca, Junin (Huancayo), Cuzco and Puno) have been identified with the highest number of potato samples. According to results obtained at PCs in these Andean departments the last 4 years, the "Andean weevil" (*Premnotripes* sp.) is still the most “visible” phytosanitary problem that worries potato producers and appears to causes the greatest yield losses. Other “visible” problems identified at PCs by presence of characteristic “visible” symptoms are late blight (*Phytophthora infestans*), *Epitrix* sp., *Alternaria*, and *Rhizoctonia solani*. No nematode problems were detected / associated to potato yield losses in these regions. However, contrary to these results obtained with potato samples brought by farmers at PCs, previous nematological surveys in potato-growing areas have shown wide distribution of “potato cyst nematodes” (*Globodera pallida* and *G. rostochiensis*). *G. pallida* predominates in most North and Central of Andean departments, while *G. rostochiensis* in Southern region of Peru (Altopiano region shared by Peru and Bolivia). Of the 1,118 samples evaluated, 64.9% presented the PCN between infestation levels from incipient to very high (1.02 - 2161 eggs / cc of soil). The levels of incipient, medium, and high infestation were found in 13.71%, 11.05% and 10.44%, respectively. 25.10% of the remaining fields were highly infested by the PCN. According to the levels of infestation indicated, these cause yield losses of 5% in 6,120 hectares, 13% in 4,722 hectares, 45% in 4,464 ha and 58% in 1,782 ha, respectively, that severely affect the production of native potatoes in the departments of the Central and South region of the country.

**MOLECULAR IDENTIFICATION OF MELOIDOGYNE SPP. INFECTING BEANS (PHASEOLUS VULGARIS) IN ARGENTINA [IDENTIFICACIÓN MOLECULAR DE MELOIDOGYNE SPP. INFECCION DE HABAS (PHASEOLUS VULGARIS) EN ARGENTINA]**
Argentina is the fifth largest producer of beans (*Phaseolus vulgaris*) on the American continent, with 99% of national production concentrated in the Northwest region. Root-knot nematodes (*Meloidogyne* spp.) have been reported as one of the most important pathogens infecting beans in many parts of the world, not only because of its direct damage, but also for creating access for numerous bacteria and fungi that affect its performance. Although there have been multiple associations observed between beans and root-knot nematodes (*Meloidogyne* spp.) in Argentina, many of these reports have not been well analyzed or documented, and very little is known about the species of root-knot nematodes involved in such associations. The objective of this work was to use a molecular approach to identify *Meloidogyne* spp. found infecting white beans in the Jujuy province, Argentina. In 2016, soil samples were collected from the rhizosphere of white bean var. Alubia, in the locality of Rio Blanco, department Palpalá (24°13'40"S/65°14'40.70"O). Seedlings of *Impatiens* sp. were transplanted into the infested soil samples and maintained in a greenhouse for 60 days. Configuration of the perineal patterns, morphometrics of selected characters of nematode females including body, stylet and tail length were consistent with those reported in the original description of *M. arenaria*. For the molecular analysis, nematode females were extracted from the roots and placed in 96% ethanol for further use. DNA was extracted from individual females and mitochondrial DNA (mtDNA) was amplified using the following primer sets: MORF (5’ - ATCGGGGTGTATTAATGG G -3’) and MTHIS (5’ -AAATTC AATGAAAATTAATAG C- 3’); TRNAH (5’ -TGA ATT TTT TAT TGT GAT TAA-3’) and MHR106 (5’ - ATT TCC TAA AGA CCT TTC TTA GT-3’). A fragment of approximately 740 bp and 550 bp was obtained with each of the above primer sets, respectively. To further confirm the nematode species identification, we used the *M. arenaria* species-specific SCAR primer set Far (5’- TCGGCGATAGAGGTAAATGAC-3’), Rar (5’- TCGGCGATAGACACTACAACT-3’). This primer set yielded a fragment of approximately 430 bp, which is identical to that previously reported for *M. arenaria*. DNA sequencing is still in progress. To our knowledge, this report constituted the first molecular identification of *M. arenaria* found infecting beans in Argentina.

**POPULATION DYNAMICS OF ECTOPARASITIC AND ENDOPARASITIC NEMATODES IN NORTH CAROLINA [ DINÁMICA DE POBLACIÓN DE NEMATODOS ECTOPARASÍTICOS Y ENDOPARASÍTICOS EN CAROLINA DEL NORTE]**


Sting nematode (*Belonolaimus longicaudatus*) and root-knot nematode (*Meloidogyne* spp.) are emerging problems for creeping bentgrass and bermudagrass putting greens in the transition zone. These nematodes cause severe putting green damage resulting in decreased playability. Sting nematode is an ectoparasitic nematode, feeding on the plant roots and causing severe damage at very low population numbers. Root-knot nematode is an endoparasitic nematode, feeding and reproducing within the turf root system and decreasing overall root function. However, little is known about either nematode’s population dynamics and soil movement throughout the year. A sampling study was initiated in 2014 to identify nematode population numbers throughout the year, and to understand the vertical distribution of the nematodes within a putting green soil column. Four golf courses were sampled throughout central North Carolina, and nematode samples were taken monthly at three different depths. *B. longicaudatus* populations were consistent across three golf courses, with numbers ranging from 40-250 nematodes per 500 cm² soil. Root-knot nematode was sampled at a single course, and numbers of stage 2 juveniles...
ranged from 20-120 nematodes per 500 cm³ soil. For sting nematode, populations were primarily located within the top 10 cm of the soil column during the winter, but the summer saw a population shift to the lower 20+ cm of the soil column. This was very different from root-knot nematode, where populations were localized within the top 10 cm throughout the entire year. Both sting and root-knot populations began to increase in April as temperatures start to rise. This shows very different population dynamics depending upon feeding tactics, with high variability between ectoparasitic and endoparasitic nematode species.

NEW SWEET BIOSENSORS: SYNTHESIS AND STRUCTURAL CHARACTERIZATION OF CARBON QUANTUM DOTS EMPLOYING GLUCOSE AS A PRECURSOR AND THEIR USE AS BIOMARKERS IN NEMATODES [NUEVOS Y DULCES BIOSENSORES: SINTESIS Y CARACTERIZACION ESTRUCTURAL DE PUNTOS CUANTICOS DE CARBONO EMPLEANDO GLUCOSA COMO PRECURSOR Y SU USO COMO BIOMARCADORES EN NEMATODOS]

Guerra, M.1, M. Revilla1, A. Meza1, G. López2, Y. Alvarado1, and E. San-Blas3 1 Laboratorio de Polímeros, Universidad del Zulia, Maracaibo, Venezuela C.P. 4001. 2 Centro de Investigación y Tecnología de los Materiales, Laboratorio de Caracterización Molecular y Biomolecular Instituto Venezolano de Investigaciones Tecnológicas, Maracaibo, Zulia, Venezuela. 3Laboratorio de Protección Vegetal, Instituto Venezolano de Investigaciones Científicas, Maracaibo, Venezuela C.P. 4001. mayamaru.guerra@gmail.com

In the last decade, the study of nanoparticles has gained very much attention, which has allowed the development of new tools in nanotechnology. One of the latest advances made in the area was the discovery of carbon dots (C-dots). They have very interesting properties such as chemical inertia, easy functionalization, good photoluminescence, non-toxic, and high biocompatibility. These properties have made the C-dots extremely important in the field of optoelectronics, bioimaging, biosensors, among others. In this work, C-dots were obtained by means of microwave-assisted synthesis from glucose as a precursor. C-dots were characterized by Fourier transform infrared spectroscopy with an attenuated total reflectance (FTIR-ATR), UV-Visible range, fluorescence, and scanning electron microscopy. The interaction of the C-dots with entomopathogenic nematodes (EPN) was evaluated. The results indicated that the synthesized C-dots were non-toxic to Steinernema and Heterorhabditis and no sub-lethal effects were observed in both genera when using different concentration of C-dots for 2 weeks. On the other hand, different developmental stages of NEP were observed under an epifluorescence microscope when treated with C-dots. Infective juveniles were soaked in a solution containing C-dots, and adults were fed using bacteria with C-dots. The resulting images confirmed a high affinity of C-dots to chitin. The cuticle of the infective juveniles was covered by C-dots and hard external structures were heavily marked (e.g., tooth like structure in Heterorhabditis) exhibiting a fluorescent green color when UV light was applied. In adults apart of chitin structures, the C-dots were traced in the intestinal tract. These results demonstrate that C-dots could become a reliable, cheap, and useful tool for bioimaging and can be use in the future to evaluate many aspects of the biology and ecology of nematodes.

POTATO CYST NEMATODE GLOBODERA SPP. OCCURRENCE, BIOLOGY AND MANAGEMENT IN NORWAY [NEMATODO QUISTE DE LA PATATA GLOBODERA SPP. OCURRENCIA, BIOLOGÍA Y CONTROL EN NORUEGA]

Holgado, R., C. Magnusson, B. Hammeraas, I. Rasmussen, K. Strandnaes, and R. Knudsen. Norwegian Institute of Bioeconomy Research, Norway. Norwegian Food Safety Authority, Norway. ricardo.holgado@nibio.no

In Norway, potato cyst nematodes (PCN) Globodera rostochiensis and G. pallida are quarantined pests. PCN was recorded in 1955, and subsequently legislative regulations have been implemented and extensive surveys have been performed. PCN management requires knowledge about the life cycle, occurrence of species,
pathotypes, and population dynamics. Until today, no nematicides have been used in Norway, for PCN management. Norwegian management of non-virulent *G. rostochiensis* (Ro1/Ro4) is by crop rotation combined susceptible and resistant potato varieties. Infestations by *G. pallida* or virulent *G. rostochiensis* results in 40-years ban on growing potato. Studies of selected Norwegian PCN populations using morphological and PCR amplification of ITS regions identified most populations as *G. rostochiensis*, with one exception, which belonged to *G. pallida*. PCR amplification and sequencing of the non-coding short circular mitochondrial (scmt) region confirmed the species identification, and revealed a close relationship of Norwegian PCN populations to European populations. The vap-1 gene proved to be present in each population, and showed different expression patterns of the vap-1 gene whiting populations. However, differences in allele frequencies between populations are minor.

Fields in quarantine for 32, 18, and 12 years were selected to study the decline in absence of plant host. These fields were infested by *G. rostochiensis* (Ro3), *G. rostochiensis* (Ro1) and *G. pallida* (Pa 2/3) respectively. Cysts from each soil were baited using susceptible potato cultivar. After 4 months, soils were analysed for new cysts. Viable PCN were found in all soil samples, demonstrating that *G. rostochiensis* could survive for 32 years and that *G. pallida* so far, has survived for 12 years. European reports indicated that PCN have one generation per year; however, there are reports that suggest PCN can complete or partially complete, a second generation. Our studies showed that both PCN species complete a second generation of J2s. Concerning PCN life cycle, young female development takes about 35 days, and 40 days for new cysts. However, there are populations that develop cyst after 29 days. If potato is used as a trap crop, it needs to be lifted no later than 35 days after planting. In Norway *Solanum sisymbriifolium* as trap crop is not a practical alternative for managing PCN and this is also the case for *Brassicaceae* mustard as green manures or biofumigants. Areas with certified seed potatoes have been under monitoring since 1956 and are so far free of PCN. To prevent introduction of new PCN populations, import and movement of seed-potato are prohibited. Using the EPPO protocol for resistant test, 26 potato varieties were tested. Score nine was recorded in 12 varieties, score eight in five varieties, score three in one variety, score two in three varieties and the lowest score one in five varieties. It appears that Norwegian farmers have several advantages concerning PCN management compared to many other countries.

**Globodera Pallida: an Unwelcome Visitor in the Potato-Growing Areas of Costa Rica and Its Interaction with Other Nematodes Associated with Potatoes**

Humphreys-Pereira, D. A., R. Sandoval-Ruíz and L. Flores-Chaves. 1Laboratory of Nematology-CIPROC, University of Costa Rica, San Pedro, Costa Rica, 2060.

Potatoes are the third most important staple food crop in Costa Rica, with a per capita consumption of 14.7 kg. There are two main potato-growing areas in the country, Cartago with 72.9% of the potato production and Alajuela (mainly Zarcero) with 25.4%. The province of San José contributes to the remaining 1.7% of production. Plant-parasitic nematodes such as *Globodera*, *Pratylenchus* and *Meloidogyne* are considered a problem for the potato industry worldwide, due to yield reduction and tuber quality deterioration. The pale potato cyst nematode (*G. pallida*) has been reported in Central America in Costa Rica and Panamá. The firsts symptoms caused by *G. pallida* on potatoes in Costa Rica were observed in 2005, but it was not until 2009 that it was characterized with morphological and molecular techniques. Since then, no additional research has been conducted on this nematode or other nematode species associated with potatoes in Costa Rica. Soil and root samples were collected in 46 potato fields at different altitudes (ranging from 1,609 to 3,199 m.a.s.l.) and soil textures mainly from the north region of Cartago. The most frequent nematode species associated with potato roots were *Pratylenchus* (68%) followed by *Meloidogyne* and *Globodera* with 29.5%. However, *Globodera* cysts were found in 67% of the potato fields at altitudes ranging from 1,645 to 3,199 m.a.s.l. Importantly, the frequency of...
Globodera cysts increased with the altitude, 25% between 1600-2100 m.a.s.l., 49% at 2,101-2,600 m.a.s.l. and 88% at 2,601-3,100 m.a.s.l. Similarly, the frequency of Pratylenchus increased with the altitude, from 25 to 54 and 77% at the same altitude ranges as Globodera. In contrast, the frequency of Meloidogyne decreased at higher altitudes with 40, 35, and 9%, respectively. Molecular techniques such as PCR-RFLP and species-specific primers allowed the identification of M. incognita and M. hapla on several potato samples. Pratylenchus samples were characterized based the 28S gene and identified as P. penetrans. Fifteen populations of Globodera (14 from Cartago and one from Zarcero) were identified at the species level by sequencing one mitochondrial gene (cob) and the ITS nuclear marker. The identity for all samples were positive for G. pallida. A unique haplotype was obtained from both markers and phylogenetic analyses showed a large monophyletic group formed with the Costa Rican haplotype and sequences from the northern region of Perú. A problem contributing to the wide distribution of G. pallida in Costa Rica is the high concentration of seed production in the region between the 2,601-3,100 m.a.s.l. where G. pallida has been identified from many of the farms in this region.

TYLENCHULOID SPECIES: A COMPONENT OF NEMATOFAUNA OF HARDWOOD FORESTS OF NORTH FLORIDA [ESPECIES TYLENCHULOIDES: UN COMPONENTE DE NEMATOFAUNA DE BOSQUES DE MADERA DURO DEL NORTE DE FLORIDA]

Inserra, R.N.1, S. A. Subbotin², A. Troccoli³ and P. J. Anderson¹. ¹Division of Plant Industry, DPI-FDACS, Gainesville, Florida 32614-7100, USA. ²Plant Pest Diagnostic Services, CDFA, Sacramento, California 95832, USA. ³CNR, Istituto per la Protezione Sostenibile delle Piante, Via G. Amendola 122/D, Bari 70126, Italy.

The hardwood forests of North Florida consist of numerous tree species belonging to at least 53 genera. The most representative of these genera include Acer (maples), Carya (hickories), Fraxinus (ash trees), Liquidambar (sweet gum), Magnolia (magnolias and tulip poplars), Myrica (wax myrtle, a shrub) and Quercus (oaks). The results of nematode surveys conducted in these natural areas in the last three decades indicate that tylenchuloid species occur commonly in this environment. Two species of Taphotylenchulus, T. arenarium and T. floridensis, parasitize roots of sweet gum (L. styraciflua) and oak (Quercus spp.), respectively. Another species in another genus, Tylenchulus palustris, parasitizes roots of pop ash (F. caroliniana). An additional tylenchuloid, reported as a parasite of L. styraciflua, was described originally as Sphaeronema whittoni and later as Tumiota whittoni. Molecular and morphological studies conducted cooperatively with taxonomists in the USA and Italy have more recently identified this species as a member of the genus Meloidoderita; however, this species is not typical of the genus in that its mature resistant stage fits the definition of a heteroderid cyst rather than that of a Meloidoderita cystoid.

SPATIAL DISTRIBUTION OF MELOIDOGYNE SPECIES IN THE PLANTAIN (MUSSA AAB) PRODUCING REGION OF CÓRDOBA, COLOMBIA [DISTRIBUCIÓN ESPACIAL DE ESPECIES DE MELOIDOGYNE EN LA REGIÓN PRODUCTORA DE PLÁTANO (MUSSA AAB) DE CÓRDOBA, COLOMBIA]

Jaraba-Navas, J. D., I. Suárez, and E. Combatt. Universidad de Córdoba, Facultad de Ciencias Agrícolas, Carrera 6 No. 76-103. Montería, Colombia. juan@fca.edu.co

Córdoba is the most important plantain producer state in the Caribbean Region of Colombia. In region the most important challengers to produce plantain are Black Sigatoka (Mycosphaerella fijiensis), Moko disease (Ralstonia solanacearumrace 2) and root-knot nematodes (Meloidogyne spp). Affected plants by Meloidogyne spp. have poor anchorage, lack of vigor, and are prone to topping under bunch weight. The objective of this study was to determine the regional spacial distribution of Meloidogyne spp. in the plantain producer area of Córdoba state. Regional spatial patterns of Meloidogyne species and infecting plantain crops in Córdoba state were analyzed from 70 plantain fields in the municipalities of Loricá, Los Córdobas, Moñitos, Puerto Escondido, San
Bernardo del Viento, Tierralta, and Valencia. Rhizosphere soil and roots samples were collected at each site. Soil texture, organic matter content, pH, and electrical conductivity were assessed from the soils and the presence of galling were estimated from the plant root. Morphologic and morphometric parameters were used to identify *Meloidogyne* spp. Forty-three out seventy plantain fields contained *Meloidogyne* spp in this state. Root-knot nematodes species were *M. incognita* and *M. arenaria*. Soils positives to *Meloidogyne* spp. were moderately acid to slightly alkaline, had 22 to 75% sand, 6 to 33% clay content and most of them had poor to rich organic matter content. Soil sand and clay content were highly correlated with principal components I and II, respectively. This correlation allowed us to classify soils from all the sampled sites into four groups of edaphic similarity in the plantain producing soils of Córdoba.

**MIGRATION OF SECOND-STAGE JUVENILES OF *MELOIDOGYNE FLORIDENSIS* TOWARDS TOMATO ROOTS [MIGRACIÓN DE JUVENILES DE SEGUNDA ETAPA DE *MELOIDOGYNE FLORIDENSIS* HACIA RAÍCES DE TOMATE]**

1Federal Rural University of Pernambuco, Recife, Pernambuco 52171-900, Brazil. 2University of Florida, Gainesville, Florida 32611, USA. 3Estação Experimental de Cana-de-açúcar do Carpina, Carpina, Pernambuco 55812-010, Brazil. hugginsdiego@gmail.com

Infected juveniles of plant-parasitic nematodes must move through soil to locate and infect roots, thus their migration is reported to be related to plant chemical cues. Our objective was to determine the number of second-stage juveniles (J2) of *M. floridensis* that moved upward in columns with and without plant stimuli. Migration columns were 14-cm long and constructed with three PVC rings (4.4-cm-d x 4-cm long) taped together on top of a 2-cm long inoculation ring. Each column was filled with pasteurized sandy soil maintained at 1.2 kg.dm⁻³ with 10% water content. Styrofoam cups containing 4-week-old tomato (cv. Cobra) seedlings were attached on top of each column. Cups without tomato served as controls. To prevent root growth into columns, a 35-μm nylon mesh was attached to the bottom of the cups. Freshly hatched J2 (1,000±100 per column) were added in the middle of the inoculation ring and the columns were placed in an environmental chamber at 20°C. To prevent J2 migration from the inoculation ring a 15-μm nylon mesh covered its base. The experiment was completely randomized with four replicates. The columns were dismantled 3 and 12 days after inoculation (DAI). Nematodes were extracted from the soil of each ring and cup separately and roots were stained to determine the number of J2 that penetrated. Juveniles were categorized as active or inactive; the latter were deteriorated or did not show any movement. Data were subjected to a repeated measure MANOVA and posterior chi-square analysis to compare the distribution of active and inactive J2 along the columns. The presence of tomato roots did not increase the rate of J2 migration compared with that occurring in columns without tomato (*P* ≥ 0.05). Less than 1.0% migrated over 13 cm regardless of stimuli. A mean of 5 J2/root system were recorded 12 DAI. The interaction between time and distance migrated was significant for both active and inactive nematodes (*P* ≤ 0.05). Three DAI 22% of active J2 migrated at least 1 cm, whereas 67% of inactive J2 remained in the inoculation ring. Twelve DAI 17.5% of active J2 migrated 1-5 cm, 15% migrated 5-9 cm and 4% migrated 9-13 cm. There was an increase of inactive J2 in the second ring (1-5 cm) 12 DAI, which suggests a downward movement of these inactive juveniles due to daily watering.

**DIVERSITY OF POPULATIONS OF THE ROOT-KNOT NEMATODE IN PERU [DIVERSIDAD DE POBLACIONES DEL NEMATODO DEL NODULO DE LA RAÍZ EN EL PERU]**

Lima-Medina, I.1, and R. Y. Bravo-Portocarrero1. Ingeniería Agronómica, Facultad de Ciencias Agrarias, Universidad Nacional del Altiplano. Av. Floral 1153, Puno-Perú. islimes2@gmail.com

Phytosanitary problems are observed in different crops of agricultural importance in Peru and being drastically affect their production and yield. Among these pathogens, the root-knot nematode, *Meloidogyne* spp., is undoubtedly the most important group of nematodes for tropical and
50th Annual Meeting of the Organization of Tropical Nematologists of America

Ma, X.¹, V. Richards² and P. Agudelo¹
¹Department of Plant and Environmental Sciences, Clemson University, 29630, SC, USA.
²Department of Biological Sciences, Clemson University, 29634, SC, USA.

We report mitochondrial genome sequences of lance nematodes Hoplolaimus columbus and H. galeatus, two species of agricultural importance in field crops and turfgrasses. Despite their economic significance, these nematodes are relatively under studied and few reference sequences exist. The mitochondrial genomes were assembled de novo using Whole Genome Amplification and Illumina MiSeq, and multiple assembly methods were compared. Annotations were performed with invertebrate mitochondrial genetic code 5 and nematode code 14, with protein-coding genes and rRNA genes being consistent, but with discrepancies in tRNA gene prediction between the two annotations. Both species have circularized mitochondrial genomes of approximately 25kb, including 12 protein-coding genes, two ribosomal RNA genes, and a large non-coding region. The atp8 gene is missing in both genomes. We also show phylogenetic relationships among 80 diverse nematode species using maximum likelihood, based on concatenated nucleotide sequences of 12 protein-coding genes (17kb). Gene order was diverse and phylogenetically informative, and analyses suggest that cox2 and nd5 genes are more informative than other mitochondrial genes in Tylenchida. These mitochondrial genome sequences will be useful in identifying informative regions for building references and for developing genetic tools for species identification and phylogenetic investigations. Of special significance is the potential contributions to the elucidation of the origin of H. columbus, first reported from South Carolina, USA in 1963, but believed to be an introduced species to the United States. These genomes are also a tool that enables comparative genomic approaches to studying parthenogenetic reproduction and its role in speciation of lance nematodes.

DE NOVO ASSEMBLY AND PHYLOGENETIC ANALYSIS OF MITOCHONDRIAL GENOMES OF HOPLOLAIMITUS COLUMBUS AND H. GALEATUS (NEMATODA: TYLENCHIDA)
[ASAMBLEA DE NOVO Y ANÁLISIS FARGENÉTICO DE GENOMOS MITOCONDRIALES DE HOPLOLAIMITUS COLUMBUS Y H. GALEATUS (NEMATODA: TYLENCHIDA)]

NEMATICIDAL EFFICACY AND ROOT PROTECTION PROVIDED BY SALIBRO™ IN GRAPES: PART 1: MATURE CHARDONNAY GRAPE VINES AFFECTED BY MELOIDOGYNE ETHIOPIICA,
Mesocriconema xenoplax, Paratylenceus sp., and Xiphinema americanum in Casablanca, Chile [Eficacia nematicida y protección de raíz proporcionada por Salibro™ en uvas: Parte 1: Vidas de uva de Chardonnay maduras afectadas por Meloidogyne ethiopica, Mesocriconema xenoplax, Paratylenceus sp. y Xiphinema americanum en Casablanca, Chile]

Magunacelaya, J.C.¹, S. González ¹, T. Ahumada¹, P. Riveros², M. L. Di Miro³ and J. A. Wiles⁴. ¹Pontificia Universidad Católica de Valparaíso, Chile. ²Corteva Agriscience™, Agriculture Division of DowDuPont™, Dow AgroSciences Chile S.A., Av. Américo Vespucio Sur 100, piso 6, Las Condes, Santiago, Chile. ³Corteva Agriscience™, Agriculture Division of DowDuPont™, DuPont Argentina, Av. madres de Plaza 25 de Mayo 3020 - Piso 11, S2013SWJ, Rosario, Provincia de Santa Fe, Argentina. ⁴Corteva Agriscience™, Agriculture Division of DowDuPont™, DuPont (U.K.) Limited, 4th Floor, Kings Court, London Road, Stevenage, SG1 2NG, United Kingdom.

Chardonnay is the wine grape with the highest sensitivity to Meloidogyne species in Chilean viticulture, especially in Casablanca soils with low organic matter and high sand content. The predominant species is M. ethiopica. The nematicidal efficacy of Salibro™, which contains the active ingredient fluazaindolizine, was evaluated at doses of 2.0, 2.0 + 2.0, 3.0, 4.0, and 5.0 L/ha, applied by drip application. Plots of 22-year-old Chardonnay vines, located in the Casablanca Valley in Chile, were evaluated for root damage, vegetative growth and yield, and soil samples were taken to assess nematode populations. All the Salibro™ treatments reduced M. ethiopica populations during the period of the trial, including the lowest dose of 2 L/ha. The highest reductions in phytoparasitic nematodes were evident when the Salibro™ dose was 4 and 5 L/ha, and the quality of functional roots (root structure and colour) improved in proportion to the increase in dose of Salibro™. Other phytoparasitic nematodes present in the vine rhizosphere were also reduced by Salibro™ treatments, such as Mesocriconema xenoplax, Paratylenceus sp., and Xiphinema americanum. In particular very good nematicidal effects were observed on Mesocriconema xenoplax, and Paratylenceus sp. In this year, the first season after treatment, the fruit harvest showed no statistical differences between treatments, but there was a trend that the higher doses of Salibro™ had slightly increased yields. As well as plant-parasitic nematodes other types of nematodes were enumerated in the soil rhizosphere samples. It was noteworthy that none of the Salibro™ treatments showed any impact on population levels of non-phytoparasitic nematodes. Overall, Salibro™ showed very good plant-parasitic nematode control and root protection in mature Chardonnay grape vines. It is common in grape vines that the benefits of improved root health and reduced infection by plant-parasitic nematodes is expressed in positive vegetative growth and yield in the following season.

Nematicidal efficacy and root protection provided by Salibro™ in grapes: Part 2: newly established Sauvignon Blanc grape vines and mature Chardonnay grape vines affected by Meloidogyne ethiopica, Paratylenceus sp., Helicotylenceus sp., and Xiphinema americanum in Casablanca, Chile [Eficacia nematicida y protección de raíz proporcionadas por Salibro™ en uvas: parte 2: Vines de uva de vinas blancas de Sauvignon establecidas recientemente afectadas por Meloidogyne ethiopica, Paratylenceus sp., Helicotylenceus sp., y Xiphinema americanum en Casablanca, Chile]

Magunacelaya J. C.¹, S. González ¹, T. Ahumada¹, P. Riveros², M. L. Di Miro³ and J. A. Wiles⁴. ¹Pontificia Universidad Católica de Valparaíso, Chile. ²Agriculture Division of DowDuPont™, Dow AgroSciences Chile S.A., Av. Américo Vespucio Sur 100, piso 6, Las Condes, Santiago, Chile. ³Agriculture Division of DowDuPont™, DuPont Argentina, Av. madres de Plaza 25 de Mayo 3020 - Piso 11, S2013SWJ, Rosario, Provincia de Santa Fe, Argentina. ⁴Agriculture Division of DowDuPont™, DuPont (U.K.) Limited, 4th Floor, Kings Court, London Road, Stevenage, SG1 2NG, United Kingdom.
After Chardonnay, Sauvignon Blanc is the second most important variety of grape that is affected by *Meloidogyne sp.* in Chile, especially in the Casablanca Valley where the soil is sandy and has low organic matter. The dominant species is *M. ethiopica*. This study was conducted in young Sauvignon Blanc vines (less than 3 years old) that were being established in a commercial vineyard. The nematicidal efficacy of Salibro™, which contains the active ingredient fluazaindolizine, was evaluated at doses of 2.0, 2.0 + 2.0, 3.0, 4.0 and 5.0 L/ha, applied by drip application. Plots were evaluated for root damage, vegetative growth and yield, and soil samples were taken to assess nematode populations. All the Salibro™ treatments considerably reduced *Meloidogyne ethiopica* populations during the study period. The quality of the vine roots (structure/colour assessment) improved with increasing rates of Salibro™, which was probably because the treatments could protect a greater volume of roots against phytoparasitic nematodes. Other phytoparasitic nematodes found in the rhizosphere samples in this study, such as *Paratylenchus sp.*, *Xiphinema americanum*, and *Helicotylenchus sp.*, were also affected and their populations were reduced in Salibro™ treatments. In this first season, there were no statistically significant differences in vegetative growth or fruit harvest weights between treatments, however as root health was clearly improved by Salibro™ treatments, the vines will be monitored to determine if vegetative or fruiting productivity may improve in the following season. In the soil samples, non-phytoparasitic nematodes were also counted, and none of the Salibro™ treatments affected the population levels of these nematodes. Overall, Salibro™ showed very good control of *M. ethiopica* and other plant parasitic nematodes and provided root protection in young, newly established, Sauvignon Blanc grape vines.

**THE CONUNDRUM OF A CAREER IN PLANT NEMATOLOGY: WHETHER TO REMAIN OR LEAVE? (EL ACERTIJO DE UNA CARRERA EN NEMATOLOGIA)**

**Manzanilla López, R. H.** Visiting Professor, Centro de Desarrollo de Productos Bióticos, Yautepec, Morelos, C. P.62731, Mexico. rosa.manzanilla@gmail.com

Nematology capacity building and retention of nematologists represents an important worldwide challenge to continue research in this area of crop protection. One key issue to succeed in this quest is the recognition of plant-parasitic nematodes as an important limitation to sustainable crop production by stakeholders like farmers, government and non-governmental bodies who are responsible for allocating nematology funding for training (undergraduate and graduate level), and for research programmes. Meanwhile, fewer and fewer permanent positions for nematologists are either retained or created in universities and research institutes. Job opportunities to increase retention of highly qualified graduates is of utmost importance for individuals to remain in or leave nematology or any other crop protection area. However, how can graduates increase their chances of establishing a career in nematology? So far, graduate training has been centred in training/formation on a specific subject under the supervision of an expert authority, but times are changing. Graduate research is generally carried out within a specific project that is usually sponsored by a funding agency. Increasingly, the output and quality of his/her scientific research is assessed within a research-publication framework according to journal impact factor and number of publications (H index). Furthermore, the present research funding system encourages strong competition, thus affecting collaboration between research groups that eventually reflect in fewer job opportunities. The rapid development of molecular tools and IT has prompted a changing and challenging environment where skills such as creativity, innovation, public communication, entrepreneurial skills and ‘know how to’ transfer new technology and create links with industry/business are required. In this scenario, early and mid-career graduates will continuously be required to solve riddles such as how to succeed in job and research funding applications, select those education courses/programmes that may be relevant to increase their chances to
position themselves better in the job market or even create a new job for themselves. We discuss how learned societies, like ONTA, can sponsor/mentor early-mid career nematologists to remain in nematology, for example, by creating a web of mentors, travel grants, short laboratory stays, work experience, publication training, among others.

THE FUTURE OF PLANT NEMATOLOGY (THE MEXICAN CASE) [EL FUTURO DE LA NEMATOLOGÍA VEGETAL (EL CASO MEXICANO)]

Marbán-Mendoza, N. and A. de J. Cabrera-Hidalgo. Laboratorio de nematodos fitopatógenos, Posgrado en Protección Vegetal, Universidad Autónoma Chapingo, Carretera México-Texcoco km 38.5, Chapingo, Edo. de México, C.P. 56230. nmarbanm@yahoo.com.mx

It is Plant Nematology shrinking in Mexico? If so, why? And much better, what can the Mexicans can do to curb this tendency. To answer these questions, we decided to count the number of active Plant Nematologists such as: Expert Plant Nematologists (teach, conduct research publish and get funding for research programs), Plant Nematologist (teach usually undergraduate courses, do field or laboratory work), Professional Plant Nematologist (do field and laboratory work). This task was done before 1990 and after considering this date as major brake because the fumigant Methyl Bromide was included in the Montreal Protocol to be eliminated a fact that promoted a great incentive for research and development. Results showed a clear stagnation of Plant Nematology. Fewer Permanent position are kept either in Government and non-Government institutions, practically no new courses and programs in Graduate programs has been developed in the last 25 years. Professional Nematologists remain pretty much stable due the plant sanitation norms that obligated growers to fulfill requirements for goodies exportations. We think that major changes must happen in the near future because Expert Nematologists are aging and younger and highly qualified people lack of job opportunities in major Universities. Needless to say that no undergraduate or graduate programs have been developed in the same period. In Peru, we are looking forward to receiving feedback from our peers on this major problem.

RESISTANCE OF CHILI PEPPER (CAPSICUM SPP.) TO MELOIDOGYNE ENTEROLOBII [RESISTENCIA DE PIMIENTA DE CHILE (CAPSICUM SPP.) A MELOIDOGYNE ENTEROLOBII]

Marques, M. I. S., and M. R. Rocha. 1IF Goiano, Campus Ceres, 2Universidade Federal de Goias, Escola de Agronomia, Goiania, GO, Brasil. Darochamararubia@gmail.com

The genus Capsicum comprises a very diverse group of sweet and hot peppers originating from the tropics of the American continent, and currently being grown in several countries. In Brazil it is cultivated in practically all regions of the country. The present species being found are: C. annuum, C. frutescens, C. baccatum and C. chinense. Meloidogyne enterolobii is considered one of the most important parasite that affects this crop due to its wide range of hosts and its ability to parasitize plants with resistance to other species of Meloidogyne. The purpose of this study was to evaluate the reaction of chili pepper genotypes in order to find resistance and to identify the mechanisms involved. In the first part of the study, 73 genotypes of Capsicum spp. were evaluated and 31 were identified as resistant to M. enterolobii by presenting reproduction factor lower than 1.0. From these results, two genotypes with contrasting behaviors of resistance (C. chinense, Bode roxa A) and susceptibility (C. baccatum, Cambuci) were selected for the following studies. In the second part of the study, the development and penetration of M. enterolobii in these two genotypes were evaluated. The second stage juveniles (J2) penetrated the roots of the resistant and susceptible genotypes, although in a smaller number in the resistant. M. enterolobii had its life cycle delayed in the resistant genotype with the presence of fewer specimens inside the roots. At 28 DAI, fully developed females were observed in Cambuci (susceptible genotype), whereas in Bode roxa A (resistant genotype), J4 were found at 35 DAI with some of them differentiating into males. Cambuci confirmed its susceptibility behavior with FR = 1.87 compared to the resistant Bode roxa A, with FR = 0.67. In the third part of this study, histological and cytometric analyses were performed. The resistance of the Bode roxa A was demonstrated by the occurrence of late hypersensitivity reaction
(RH), since the nematode was able to penetrate and develop in the vascular cylinder, presenting at 28 and 35 DAI females at the feeding site in less quantity than the susceptible genotype (Cambuci). The resistant genotype (Bode roxa A) inoculated had a larger cell area than the susceptible genotype (Cambuci). In the fourth study the physiological responses to \textit{M. enterolobii} parasitism were evaluated by comparative analysis of the enzymatic activity in resistant and susceptible genotypes, inoculated or non-inoculated. There was a greater enzymatic activity of $\beta$-1,3 glucanase (GLU) and lipoxygenase (LOX) in the leaves and the roots of the inoculated resistant genotype of \textit{Capsicum chinense} (Bode roxa A).

**MULTIMEDIA MATERIALS CURRENTLY AVAILABLE FOR FREE FOR TEACHING NEMATOLOGY [MATERIALES MULTIMEDIA ACTUALMENTE DISPONIBLES GRATIS PARA LA ENSEÑANZA DE NEMATOLOGÍA]**

McGawley, E. C., and C. Overstreet. Louisiana State University Agricultural Center, Department of Plant Pathology and Crop Physiology, 302 Life Sciences Bldg., Baton Rouge, LA 70803. emcgawley@agcenter.lsu.edu

The PC and Macintosh-compatible, Quicktime-formatted presentation by E. C. McGawley, C. Overstreet, M. J. Pontif, and A. M. Skantar entitled “Introduction to Nematodes” contains 123 multilayered slides that have 536 photographs, 159 illustrations, 17 tables, 16 videos, 10 animations, and a 16-19 page syllabus available in 8 languages: English, Spanish, Japanese, Arabic, Chinese, Russian, Italian, and Portuguese. This presentation has been on the websites of The Organization of Nematologists of Tropical America, The Society of Nematologists, and Nematode.net where it was downloaded for free over 5,500 times. Links to this presentation have been featured on the websites and newsletters of The European Society of Nematologists, The Russian Society of Nematologists, the Japanese Society of Nematologists, and The American Phytopathological Society. Because of the large size of this file, almost 4 GB, it is currently disseminated, still for free, via the websites “Dropbox” and “WeTransfer” following email requests made to emcgawley@agcenter.lsu.edu. Other free resources available include black and white and color posters illustrating “Nematode Anatomy and Morphology” (90 X 60 and 45 X 30CM formats) and “Common Genera of Plant Parasitic Nematodes” (90 X 60cm) as well as full-color, illustrated life cycles for cyst, root-knot, reniform, lesion, and dagger nematodes.

**USE OF THE SULFONAMIDE NEMATICIDE SALIBRO™ FOR THE CONTROL OF PLANT-PARASITIC NEMATODES IN POTATO CROPS IN THE MESO-ANDEAN REGION [USO DEL NEMATICIDIO SALIBRO™ DE SULFONAMIDA PARA EL CONTROL DE NEMATODOS PARASITICOS VEGETALES EN CULTIVOS DE PAPA EN LA REGIÓN MESO-ANDINA]**

Mejía, J.\textsuperscript{1}, H. R. Iruegas\textsuperscript{2}, and L. A. Apolinar\textsuperscript{2}. \textsuperscript{1}Corteva Agriscience\textsuperscript{TM}, Agriculture Division of DowDuPont\textsuperscript{TM}, DuPont de Colombia. Calle 113 # 7-21 Torre A Piso 14, Bogotá, Colombia. \textsuperscript{2}Corteva Agriscience\textsuperscript{TM}, Agriculture Division of DowDuPont\textsuperscript{TM}, DuPont Mexicana. Avenida Homero 206, Polanco, 11570. Ciudad de México, México. julian.mejia@dupont.com

Potato (\textit{Solanum tuberosum}) is one of the most important food sources in the Mesoamerica and Andean Region of the American continent. Plant-parasitic nematodes, mainly \textit{Meloidogyne} sp. and \textit{Globodera} sp., attack potato plant roots as soon as they develop and throughout the crop cycle, reducing yields and affecting tuber quality. Throughout the Meso- and Andean region, Mexico and Peru report the highest damage caused by nematodes in potato, and because of this, growers in these countries spend considerable resources to manage this threat. Few solutions are available, and particularly in Peru, potato growers often only have access to use older broad-spectrum insecticide/nematicides. The Agriculture Division of DowDuPont\textsuperscript{TM} is developing the new nematicide Salibro\textsuperscript{TM}, which is a 500SC formulation containing the novel active ingredient fluazaindolizine. Salibro\textsuperscript{TM} has a favorable toxicological and ecotoxicological profile, and has shown compatibility with biological control agents. Salibro\textsuperscript{TM} has been tested for the control of the root-knot nematode,
Meloidogyne incognita, and the cyst nematodes, Globodera rostochiensis and Globodera pallida. Salibro™ showed reduction in root and tuber damage, and improved yields. Effective rates of Salibro™ were also lower than existing older products employed as standards. Salibro™ will be a useful tool for inclusion in integrated nematode management programs in the main potato growing regions of Latin America. Data from trials in Mexico will be presented as well as suggestions of uses in potato production in Peru.

ORCHID MYCORRHIZAL FUNGAL CONTROL MELOIDOGYNE JAVANICA IN VITRO [ORQUÍDULAS MICORRIZADORAS CONTROL MELOIDOGYNE JAVANICA EN VITRO]

Mendes, B. L. 1, L. G. de Araújo 2, and M. R. da Rocha 1. 1 Nematology Laboratory, Federal University of Goiás, Goiânia, Brazil. 2 Genetics of Microorganism Laboratory, Federal University of Goiás, Goiânia, Brazil.

The root-knot nematode, Meloidogyne javanica, is a threat to agriculture in tropical countries, because it is polyphagous and highly virulent. The use of conventional synthetic nematicides generates negative effects on the environment and human health, since the residual effect of these products is high in soil. Therefore, it is interesting to insert sustainable control methods into integrated management of M. javanica, such as biological control. Orchid Mycorrhizal Fungal (OMF) Waitea circinata was isolated from Epidendrum nocturnum, a epiphytic orchid of the Brazilian Cerrado and has antagonistic action against leaf blast in rice crop, wherefore, the objective of this study was to evaluate the potential of the OMF W. circinata in antagonism to the M. javanica. The experiment aimed to evaluate the effect of concentrations of mycelial suspension on mortality and and hatching of second-stage juveniles (J2). We employed a completely randomized design, nine replications and six treatments; T1: control – autoclaved water; T2: 5 grams of mycelium per liter of water (gm/lw); T3: 10 gm/lw; T4: 15 gm/lw; T5: 20 gm/lw; T6: 25 gm/lw. The parasitic capacity of the OMF mycelium also were evaluated. The eggs were placed in contact with fungal mycelium discs. Data were analyzed by R software and after significant F-test, regression analysis was performed. The concentration of 25 gm/lw guaranteed 60% mortality of J2 in relation to the control treatment and presented positive linear regression. The concentration of 5 gm/lw reduced 86% hatching of J2. The mycelium of the OMF parasitized the eggs of M. javanica. This is the first report of the Orchid Mycorrhizal Fungus presented nematicidal characteristics, where the mycelial suspension killed juveniles of second stage, altered the embryogenesis, and the mycelium parasitized the eggs of the M. javanica.

FROM GENOTYPE TO PHENOTYPE, A WORLDWIDE COMPARISON OF GLOBODERA SPECIES [DE GENOTIPO A FENOTIPO, UNA COMPARACIÓN MUNDIAL DE ESPECIES DE GLOBODERA]

Mimee 1, B., M. Sabeh 1, P-Y. Véronneau 1, V. Blok 2 and É. Grenier 3. 1 St-Jean-sur-Richelieu Research and Development Centre, Agriculture and Agri-Food Canada, 430 boul. Gouin, St-Jean-sur-Richelieu, QC, J3B 3E6, Canada. 2 Cell and Molecular Sciences, The James Hutton Institute, Invergowrie, Dundee, DD2 5DA, United Kingdom. 3 Institute of Genetic, Environment and Plant Protection, INRA, Le Rheu, 35653, France.

The Globodera genus comprises several species of plant-parasitic nematodes. At least eight Globodera species feed on Solanaceae plants and they are all parasitic on tomato. Interestingly, only three of them are also pathogenic on potato, including the potato cyst nematodes (PCN), Globodera rostochiensis and G. pallida. These are among the most economically important pests of potato and quarantine organisms in many countries. The introduction and potential spread of these species in any part of the world has serious implications for potato production and export. We hypothesized that the ability of these hyperspecialized parasites to grow on a particular host relies on specific pathogenicity genes. The comparison of close relatives with different host preference, G. tabacum – G. rostochiensis and G. mexicana – G. pallida, has highlighted transcriptomic differences that could be linked with host specificity. This includes seven transcripts that were unique to PCN species. Genetic variations showing strong evidences for selection in PCN species were also identified. Several of them were located in genes coding for effector proteins involved in host evasion and
feeding site development. As a result of a long co-evolution, the development of these species on potato is also influenced by the presence of resistance genes in the host and virulence factors in the nematodes. The genotyping of more than 130 PCN populations showed geographic structuration among each species and revealed interesting diversification in relation to pathogenicity. For some groups, SNP markers that differentiate virulent from avirulent populations were highlighted and could be used for diagnostics in management programs. However, it was not possible to associate unique gene variants that perfectly fit each of the pathotypes in the current classification scheme. This suggests that multiple independent adaptation events could have occurred on some resistant hosts and/or that the actual pathotypes scheme should be revised in some places. This worldwide comparison of PCN populations confirmed that South America represents a high source of genetic diversity and putative virulence genes. It also supports the hypothesis that both PCN species share a single common introduction origin into Europe and then to North America.

FIRST REPORT OF POCYHONIA CHLAMYDOSPORIA PARASITIZING PRATYLENCHUS BRACHYURUS [PRIMER INFORME DE POCYHONIA CHLAMYDOSPORIA PARASITIZAR PRATYLENCHUS BRACHYURUS]

Monteiro¹, T. S. A., A. S. Gouveia², R. R. Coutinho¹ and L. G. Freitas¹. ¹Universidade Federal de Viçosa - Campus Viçosa, Departamento de Fitopatologia, 36570-900 - Viçosa/MG - Brasil. ²Universidade Federal de Viçosa - Campus Viçosa, Departamento de Bioquímica e Biologia Molecular, 36570-900 - Viçosa/MG - Brasil. thalita.s.a.monteiro@gmail.com

Pochonia chlamydosporia is a biological control agent of plant-parasitic nematodes and was reported parasitizing nematodes from the genera Meloidogyne, Heterodera, Globodera, Nacobus and Rotylenchulus. Pratylenchus brachyurus is a very important nematode in Brazil because it reproduces in mono an dicotyledon plants throughout the year, is present at high population densities in the soil, and is widespread in soybean, corn, cotton and sugarcane areas causing significant losses in these crops. The aim of this work was to evaluate the capacity of *P. chlamydosporia* (isolate Pc-10, the active component of Rizotec®) to parasite eggs and vermiform stages of *P. brachyurus*. The nematode inoculum (around 10 vermiform stages and 30 eggs) were desinfested by washing them with Nystatin, Penicillin, Cloranphenicol, Cloramin T and Ampicillin. Petri dishes with water-agar received either *P. chlamydosporia*, *P. chlamydosporia + P. brachyurus*, or *P. brachyurus*. Each treatment was replicated five times. Pictures started to be taken 10 days after the nematodes were put together with the fungus. The intense micelial development was detected in eggs and vermiform stages. It is the first report of *P. chlamydosporia* acting directly on *P. brachyurus*.

MANAGEMENT STRATEGIES OF MELOIDOGYNE SPP. IN GRAPE AND SUGARCANE CROPS IN THE NORTH COAST OF PERÚ [ESTRATEGIAS DE MANEJO DE MELOIDOGYNE SPP. EN LOS CULTIVOS DE VID Y CAÑA DE AZÚCAR EN LA COSTA NORTE DEL PERÚ]

Murguía, C. Department of Plant Health, Faculty of Agronomy, National University of Piura, Peru. cmurguiar@unp.edu.pe

Avocado, table grapes, pomegranate, asparagus, artichoke, bananas and mangoes are the most important exportation crops on the north coast of Peru. During the year 2017, vegetables, fresh fruit, frozen and preserved fruit were exported to 148 countries around the world for a value of US $5.488 billion. Sugarcane is a crop of dual purpose, both for obtaining sugar and producing ethanol. The export agroindustry of northern Peru was developed from four projects of irrigation: Poechos and San Lorenzo (Piura), Chavimochic (La Libertad) and Olmos (Lambayeque). The coastal climate is typically arid and dry, and the arable soil is mostly sandy, deserted and very poor in organic matter. These conditions are very favorable for phytoparasitic nematodes. The most important nematode genera associated to grape and sugarcane are Meloidogyne, Helicotylechus, Pratylenchus, Criconemoides, Tylenchorhynchus, and Xiphinema. Meloidogyne spp. is the most
widespread and abundant in all of the grape-producing areas. Disease management costs require investments of thousands of dollars per campaign to avoid significant production losses. Sugarcane cultivated in Olmos and Chira-Piura valleys is extensive, with concomitant damage to the roots related to *Meloidogyne* spp., *Pratylenchus* spp. and *Criconemoides*. Programs of nematode management include incorporation of manures to the soil. Unfortunately, this practice has decreased because of the high cost, poor quality, and contamination with phytoparasitic nematodes. Integrated and sustainable strategies for *Meloidogyne* spp. management and other phytonematodes are being validated for intensive (grapes) or extensive (sugarcane) agricultural systems under warm temperatures conditions, sandy soils and constant irrigation. The goal is to reduce the use of chemical nematicides using a biologically intensive approach that integrates the plant phenology and growth, rhizosphere microbiome, and soil properties. These approaches are being evaluated for management of *Meloidogyne* spp. and associated species.

THE “ROOT-KNOT NEMATODES” *MELOIDOGYNE* SPP.: SPECIES, DISTRIBUTION AND DAMAGE IN GRAPE CROP IN THE NORTH OF PERU

**Murguia, C.** Plant Health Department, Faculty of Agronomy, National University of Piura, Peru. cmurguiar@unp.edu.pe.

Peru is the fifth largest producer of table grapes in the world. During the production campaign of 2016/2017, 320,000 tons were exported to more than 40 countries at a value of US$ 646.3 million. There are 40% of the 17,600 cultivated hectares with this fruit in the north region of Peru. *Meloidogyne* spp. is widely disseminated in the main regions of grape production, specifically La Libertad, Lambayeque, and Piura. Population densities of *Meloidogyne* in the soil varied according to the level of damage on roots, age of plant, and texture of soil. Evaluations of the population dynamics of *Meloidogyne* spp. in field conditions confirm that rootstocks of cultivars Harmony, Freedom, MTG, VG, R110, and Dog Ridge are susceptible to nematode. Grape crops infested early by *Meloidogyne* spp. suffer loss of vigor and limited canopy, and in some cases, it is necessary to replant the crop. Species were determined by esterase phenotype (Est) for 14 populations isolated in different producer sectors of table grapes in the north of Peru. The isolate collection contained five phenotypes of esterase corresponding to the species: *M. morocciensis*. Est. A3, *M. arenaria* Est. A2, *M. incognita* Est. I2, *M. ethiopica* Est. E3, and *M. javanica* Est. J2. Mixtures of species of *Meloidogyne* were detected in 6 of the 14 populations. The main species (*M. morocciensis*, *M. arenaria*. and *M. incognita*) were detected in 64.3%, 50%, and 42% of the total number of populations, respectively. *Meloidogyne* spp. is the most important root parasite that affects grape production economically. More morphological and DNA studies are necessary for a greater consistency in the identification of species.

FOODWEB INDICATORS OF SOILS NATURALLY SUPPRESSIVE TO SOYBEAN CYST NEMATODE

**Neher, D.A.**, T. Nishanthan*,* Z. Grabau*, and S. Chen*. 1University of Vermont. 2University of Florida. 3University of Minnesota. Deborah.neher@uvm.edu

Soybean cyst nematode (SCN) *Heterodera glycines* has become a major pest problem in the soybean (*Glycine max*) production system in the United States, especially in the north central region. We designed an experiment to test the hypothesis that soil suppressiveness to *H. glycines* is correlated positively to management practices that favor later ecological succession and greater trophic diversity of nematode communities. A factorial combination of cultivation, crop rotation, and biocide application treatments were monitored for four years in two fields with a history of no-till and monoculture of susceptible soybean. Crop rotation had the greatest impact on nematode community index values followed by descending order of cultivation and biocides. Greater diversity of trophic groups and greater maturity index values were apparent in this case
of natural suppression of SCN. Covariable analysis showed that pH and salinity were not significant but the form of nitrogen and abundance of organic matter (mean 6.6%) affected nematode community composition. Furthermore, fungal parasitism and aminopeptidase (valine, leucine and alanine) and chitinase activity were greatest in in the treatment where no tillage, biocide, or crop rotation treatments were implemented to disturb suppressiveness. Cuticle degradation by these enzymes can directly reduce the SCN population. Amplicon sequencing of soil bacterial and fungal communities suggest that microbial community consortia are likely involved in biological control instead of single genera or species. For this specific case, disease suppression corresponded with complex food webs and any disturbance (rotation with corn, cultivation or biocide) reduced suppression.

**MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF A HETERODERA POPULATION FROM COSTA RICA | CARACTERIZACIÓN MORFOLÓGICA Y MOLECULAR DE UNA POBLACIÓN DE HETERODERA DE COSTA RICA**

Núñez-Rodríguez, L., L. Flores-Chaves, and D. A. Humphreys-Pereira, Laboratory of Nematology-CIPROC, University of Costa Rica, San Pedro, Costa Rica, 2060.

Cyst nematodes are one of the most economically important plant pathogens globally. In Costa Rica, cyst nematode research is predominately focused on *Globodera pallida*, which has been reported in the main potato-growing areas of the country. Currently, information related to morphological descriptions, inter- and intra-specific variability and the distribution of other cyst nematodes in Costa Rica is lacking. To address this issue, a study was conducted on nematodes associated with weeds in potato fields of the Central Valley of Costa Rica; this lead to the identification of a population of *Heterodera* sp. identified on the roots of *Rumex* sp., which was characterized with morphological and molecular methods. Morphological characters were measured and described from cysts and second-stage juveniles. The nuclear marker *ITS1*-5.8S-*ITS2* (728pb) and the mitochondrial marker *cox1* (a portion of 431pb) were amplified and sequenced from adult females and juveniles. The cysts showed a lemon form and dark brown coloring. The measurements of the cysts (n = 12) were: length 764.5 ± 61.4 (656.6-837.7) μm, width 515.5 ± 61.2 (445.5-621.1) μm and L / W ratio 1.49 ± 0.09 (1.3-1.6) μm. Second-stage juveniles (n = 19) showed the following characters: body length 567.6 ± 32.9 (510.5-636.5) μm, diameter at the middle of the body 25 ± 2.4 (21.7-29.2) μm, at 22.8 ± 1.2 (20.4-24.4) μm, stylet length 28.8 ± 0.43 (27.8-29.5) μm, stylet column 11.5 ± 0.44 (10.4-12.2) μm, stylet cone 13.8 ± 0.50 (12.8-14.8) μm, stylet nodule diameter 5.7 ± 0.22 (5.3-6.2) μm and OGDE 7.9 ± 0.40 (7.2-8.7) μm. The *ITS* sequences of *Heterodera* sp. from *Rumex* sp. were compared with sequences of other *Heterodera* species retrieved from GenBank. Divergence levels vary between 0.1% (1 nt) and 1.4% (10 nt), compared with *H. schachtii*, 0.1% (1 nt) and 1.0% (7 nt) compared with *H. trifolii*, and 0.3% (2 nt) and 0.4% (3 nt) compared with *H. daverti*. The *cox1* gene sequences differed more than sequence comparisons performed with the *ITS*. Divergence levels of 2.1% (8 nt) were identified in comparison with *H. daverti*; 3.3-3.6% (13-14 nt) compared with *H. trifolii*, and 8.7% (34 nt) compared with *H. schachtii*. The phylogenetic relationships of *Heterodera* sp. extracted from *Rumex* sp. and other *Heterodera* spp. were estimated using the Bayesian Inference analyses. The analysis of the *ITS* region grouped the identified *Heterodera* sp. within the *schachtii* group, with a high support value (PP = 100). The phylogenetic analysis based on the partial *cox1* gene determined that *Heterodera* sp. was in a clade with sequences of *H. daverti* (PP = 93). However, a subclade was formed only with *H. daverti* sequences (PP = 98), which may indicate the presence of cryptic species.

**IN VITRO BIOCONTROL POTENTIAL OF NATIVE PGPRs FROM COFFEA ARABICA RHIZOSPHERE AGAINST MELOIDOGYNE SPP. AND PHYTOPATHOGENIC FUNGI [POTENCIAL DE BIOCONTROL IN VITRO DE PGPRS NATIVAS DE LA RHIZOSFERA DE COFFEA ARABICA CONTRA MELOIDOGYNE SPP. Y HONGOS FITOPATOGENICOS]**
1Laboratorio de Ecología Microbiana y Biotecnología, Departamento de Biología, Facultad de Ciencias, Universidad Nacional Agraria La Molina, Lima-Peru. 2Istituto di Bioscienze e Biorisorse, Consiglio Nazionale delle Ricerche, Bari-Italy. dzuniga@lamolina.edu.pe

Strategies for biological control of root-knot nematodes (RKN, *Meloidogyne* spp.), include the use of microorganisms, such as Plant Growth Promoting Rhizobacteria (PGPR), with a broad range of antagonistic activity against different phytopathogens. In this study we tested *in vitro* five PGPR isolates proceeding from the rhizosphere of coffee plants sampled at Villa Rica, Cerro de Pasco-Peru (Longitude W 75°17’25” Latitude S 10°45’12.2”). The trials included antagonism assays vs phytopathogenic fungi. RKN populations 1, 2, 3, 4, and 5 were isolated from coffee roots whereas population 6 was isolated from chili roots. The isolates that displayed plant growth promotion and/or biocontrol capabilities were selected for molecular characterization, by sequencing their 16S rRNA ribosomal gene. *Meloidogyne* spp. identification was based on perineal patterns and molecular analysis of mtDNA, COII/16S, and rDNA ribosomal genes, through PCR amplification with specific primers. Molecular characterization of isolates M3ACT14, M4ACT5, and M4ACT1 based on 16S rRNA gene sequences classified them within the genus *Streptomyces*, closely related to *S. mirabilis* (100%), *S. humi* (99.7%), and *S. plumbiresistens* (98.9%), respectively. Four PGPR isolates produced indolacetic acid, with highest production rates for M4ACT5 (10.4 μg ml⁻¹), M4ACT1 (8.8 μg ml⁻¹), and M4ACT7 (7 μg ml⁻¹). Two isolates were able to solubilize CaHPO₄. Isolate M4ACT1 had higher rates of phosphate solubilization efficiency (27.1%, 15.8% and 33.9%) when incubated at 20, 28, and 35°C, respectively. Dual cultures with M4ACT1 and M4ACT5 showed high antagonistic activity (% of *in vitro* growth inhibition) against *Rhizoctonia* sp. (40.9%, 40.9%), *Fusarium* sp. (40.3%, 41.1%) and *Colletotrichum* sp. (41.3, 49.2%), respectively. COII/16S and 18S sequences obtained from RKN populations 1, 2, and 5 showed 97, 93, and 83% identity with *M. exigua*, whereas PCR from population 6 yielded a 18S rDNA gene sequence with a higher identity with *M. arenaria*, and the other populations gave undetermined amplicons. *In vitro* RKN antagonism assays, with population 1, showed a nematostatic effect (percent of immobilization) for M3ACT14 (100%), M4ACT5 (50.6%) and M4ACT7 (46.9%), all significantly higher compared to Triptic soy broth (TBS) (20.1%) and water (11.2%) controls, without inoculum (LSD test, *P* < 0.05). The nematicidal effect (mortality rate) was significantly higher for M3ACT14 (100%) and M4ACT7 (36.4%), compared to TBS (13.1%) and water (11.2%) controls (LSD test, *P* < 0.05). The selected PGPR showed a biocontrol potential for environment friendly management of *Meloidogyne* spp. and fungi on coffee. Further research is needed to develop appropriate field technologies for inoculation of infested roots and soil. This research was funded by 177-FONDECYT 2015 y Proyecto 007-PNIA 2016.

1Federal Rural University of Pernambuco, Recife, Pernambuco 52171-900, Brazil. 2University of Florida, Gainesville, Florida 32611, USA. akmsol22@hotmail.com

*Meloidogyne* spp. are considered among the world’s most important soilborne pathogens of agricultural crops. Their successful management requires a better understanding of their behavior within soil. Our objective was to investigate the upward mobility of second-stage juveniles (J2) of *M. enterolobii* in soil with and without tomato root stimuli to determine their ability to locate and penetrate host roots. Polyvinyl chloride columns were constructed with three 4.4-cm-d x 4-cm-long rings each taped together to one 2-cm long inoculation ring. All rings were filled with sandy soil with bulk density of 1.2 g.cm⁻³ and 10% water content. A Styrofoam cup with a 4-week-old tomato (cv. Cobra) seedling was taped to the top of half of the columns. The control contained no plant. A 35-μm nylon mesh was placed between
the Styrofoam cups and the upper ring to prevent root growth into the column, while the base of the inoculation ring was covered with a 15-μm mesh to keep J2 in the columns. Freshly hatched J2 (1,000±100) were injected into the inoculation ring and the columns placed into an environmental chamber set at 20°C. Columns were completely randomized and replicated four times. The columns were disassembled 3 and 12 days after inoculation (DAI). Nematodes were extracted from the soil of each ring and cup separately and roots were stained to observe nematode penetration. Nematodes were classified as active or inactive J2 based on whether there was visible movement or the J2 were deteriorated. Data were subjected to a repeated measures MANOVA and chi-square analysis to compare J2 distribution within the columns. There was no detectable effect of plant stimuli, but the interaction of time and distance J2 migrated was significant for both active and inactive nematodes. Eighty percent of J2 moved from the inoculation ring with 1.2% found at the top of the columns at 3 DAI. An inverse relationship was found at 12 DAI, where 23% of active juveniles migrated distances greater than 13 cm. There were fewer nematodes in the middle of the column, suggesting a rapid migration when the juveniles moved closer towards plant roots. Most of the inactive J2 were found in the inoculation ring (52% and 37% at 3 and 12 days after inoculation, respectively). J2 infected roots as early as 3 days (mean of 4 J2/root system), with greater numbers recorded 12 DAI (mean of 17 J2/root system). Inactive J2 at the top of the columns was 8% higher at the end of the experiment, indicating that even though the nematodes migrated more than 13 cm some were unable to infect tomato 12 DAI.

**IDENTIFICATION OF APHELENCHOIDES SPP. ASSOCIATED WITH STRAWBERRIES IN FLORIDA [IDENTIFICACIÓN DE APHELENCHOIDES SPP. ASOCIADO CON LAS FRESAS EN FLORIDA]**

Oliveira, C.1,2, J. Desaeger1, T. Watson1, S. Vau1, L. G. Freitas2, and R. N. Inserra1. 1Entomology and Nematology Dept, University of Florida, Gulf Coast Research and Education Center, Wimauma, FL, 33568, USA. 2Dept. Fitopatologia, Univ. Federal de Viçosa, Viçosa, Brazil. 3Division of Plant Industry, DPI-FDACS, Gainesville, Florida 32614-7100, USA. clemen.agro@gmail.com

Florida is the second largest strawberry producer in the USA and supplies the eastern United States during late fall and winter months. The summer crimp nematode, *Aphelechoides besseyi* is a foliar nematode that has damaged Florida strawberries in the past. After decades without any reports of foliar nematode infestations, *A. besseyi* has re-appeared in recent years in Central Florida strawberry farms. Nematode surveys have been conducted in these strawberry fields to monitor the populations of this nematode during the strawberry season. The results have shown that several unidentified foliar nematodes, morphologically different from *A. besseyi*, occur in leaves of both live and dead strawberry plants in these sites. Nematodes were extracted from strawberry leaves and cultured on the fungus *Monilinia fructicola* to obtain a sufficient number of specimens for morphological and molecular analyses. Comparisons of the morphological features of these populations with those of other species reported in the literature indicate that they belong to two species: *A. bicaudatus* and *A. fujanensis*. The detection of *A. bicaudatus* is a new record for Florida and that of *A. fujanensis*, is a new record for Florida and the United States. *Aphelenchoides fujanensis* was described in China and has been reported in Brazil. Molecular analyses of the cytochrome oxidase subunit I (COI), 18S rRNA, and 28S rRNA gene regions of these nematode species are still in progress.

**DOES THE STAGE OF PLANT AND INITIAL POPULATION LEVEL OF ROOT KNOT NEMATODE INFLUENCE SYMPTOM DEVELOPMENT ON SWEET PEPPER? [¿LA ETAPA DE LA PLANTA Y LOS NIVELES DE POBLACIÓN INICIAL DE LA INFLUENCIA DEL NÚMETODO DE RAÍZ DEL NUDO EN EL DESARROLLO DE LOS SÍNTOMAS EN SWEET PEPPER?]**

Oliveira, C. M. G., S. A. Oliveira, J. M. O. Rosa, J. Eulálio. Instituto Biológico, Laboratório de Nematologia, 13001970, PO Box 70, Campinas, SP, Brazil. marcelo@biologico.sp.gov.br

The aim of this study was to determine *Meloidogyne enterolobii* pathogenicity at three
different stages (one, three, and five leaf pairs) of sweet pepper (*Capsicum annuum* cv. Orázió) seedlings under greenhouse conditions. Each plant was cultivated in one pot filled with 3.8 L of substrate. The treatments consisted of five different inoculation levels: zero (control), 300, 1000, 3000 and 10000 *M. enterolobii* eggs and juveniles for each stage of seedling. The test was a completely randomized design with four replications. The plants were assessed 60 days after inoculation, plant shoot weight, final population of nematodes, and reproduction factor were measured. The results were fitted to Seinhorst model: \( Y = m + (1 - m). Z^{p^*}. T \). The tolerance limit (T) was 2,500 nematodes for plants with one and three leaf pair, and 8,500 nematodes for the five-leaf pair plant. Minimal yield (m) of plants, obtained under high population densities of the nematode, was 0.445, 0.809 and 0.965 for the one, three and five leaf pair. The plants with five leaf pair demonstrated a higher tolerance limit to the nematode infection and could be considered in the management in areas infected with *M. enterolobii*.

**DITYLENCHUS GALLAEFORMANS: A POTENTIAL BIOLOGICAL CONTROL AGENT AGAINST CLIDEMIA HIRTA**

Oliveira, S. A., P. M. Agudelo, and S. J. DeWalt. Clemson University, Clemson, SC, 29630 USA. solivei@clemson.edu

How important is coevolution between pathogens and their plant hosts in terms of resistance, tolerance, and symptoms development? *Clidemia hirta* (Melastomataceae) is a shrub native to Central and South America and the Caribbean. This plant has become an invasive species in several regions of the world, including Hawaii, where it competes with the native rain-forest species. Several management techniques have been applied against *C. hirta* in Hawaii, but none of them have been successful. We are investigating the nematode *Ditylenchus gallaeformans*, which causes severe galls on the leaves and shoots of various Melastomataceae species across Central and South America, with the goal of introducing it as a biological control agent of *C. hirta* and other invasive Melastomataceae in Hawaii. The objectives of this project are to: i) assess whether pathogenicity and virulence of the nematode depend on the host species, host genotype, or geographic source of the nematode; ii) study the microbiome associated with galls from different locations and hosts; iii) examine the amount of genetic variation in *D. gallaeformans* explained by host species and geographic location. For objective (i), we inoculated nematodes from Costa Rica and Trinidad on plants from Costa Rica, Trinidad, and Hawaii in several different experiments. Different inoculum levels were tested as well as different plant ages and shade/sun exposure. The results obtained with these experiments were not satisfactory. In two inoculation trials, some plants developed small galls; however, the galls stopped developing before causing any damage to the plant. In most of the inoculation trials, no symptoms ever developed. With the suspicion that there may be other organisms playing an important role in the infection, we decided to study the microbiome of these galls (objective ii). For the microbiome analysis, 4 samples per plant (2 healthy leaves and 2 galled leaves) were collected from 3 different plant hosts with 4 replicates each. The samples were collected in two different locations in Trinidad. We are comparing the bacterial 16S rRNA gene in healthy vs. galled leaves to look for bacteria diversity. For the phylogeographical study (objective ii), we sequenced three genes (COI, 12S, and IGS) from individual nematodes from Costa Rica, Trinidad, and Brazil. The sequences are going to be analyzed and the genetic distance between the populations are going to be assessed. Both the microbiome and the phylogeographical studies are in progress.

**EXPLOITING GENOMICS TO REVEAL THE MECHANISMS OF MELOIDOGYNE SPP. SUPPRESSION BY PASTEURIA PENETRANS**

Orr, J.1,2, P. Cock1, V. C. Blok1 and K. G. Davies2.1Cell and Molecular Sciences, The James Hutton Institute, Invergowrie, Dundee, DD2 5DA, UK. 2School of Life and Medical Sciences,
Pasteuria penetrans is a spore-forming obligatory parasite of Meloidogyne spp., contributing to highly specific suppression in soils. This suppression results from two modes of action: hindrance of root invasion by attachment of spores to the nematode cuticle, and sterilisation of infected adult females within roots. However, the genetic and molecular mechanics of these interactions are as yet unknown. Electron microscopy of spores and attachment inhibition studies suggest the involvement of electrostatic interactions, exosporium collagens, and N-acetylglucosamine (NAG) glycosylation in initial attachment to the cuticle. Using Multiple Displacement Amplification (MDA) and long read PacBio sequencing we have obtained a draft genomic sequence of Pasteuria penetrans an estimated 86% complete with minimal evidence of contamination. Analysis of this sequence provides insights into the properties of the bacterium including putative proteins which may be involved in attachment, host specificity, and parasitism. Interestingly, we have identified nine collagenous fibres structurally similar to those involved in adhesion of related spores, each with at least one predicted NAG binding site, and negative C-terminal electrostatic potential. Understanding these processes may lead to improvements in in vitro culture, prediction of efficacy, and ultimately in the effective application of Pasteuria spp. as biocontrol agents in the field.

DEVELOPING MANAGEMENT ZONES FOR NEMATODES IN SOYBEAN [DESARROLLO DE ZONAS DE GESTIÓN PARA NEMATODOS EN SOYA]

Overstreet, C., E. C. McGawley, and D. M. Xavier-Mis. Louisiana State University Agricultural Center, Department of Plant Pathology and Crop Physiology, 302 Life Sciences Bldg., Baton Rouge, LA 70803. coverstreet@agcenter.lsu.edu

The use of site-specific technology has been developed and utilized for nematode management in the Southern United States in cotton and currently is being investigated with soybeans. Meloidogyne incognita and Rotylenchulus reniformis are recognized as major nematode pathogens for both crops. Management zones are usually delineated using apparent electrical conductivity (ECa) because of the strong relationship with soil texture and nematode reproduction and pathogenicity. In trials conducted during the past several years, the response to management zones has been similar to that reported from cotton in soybean with M. incognita but not with R. reniformis. Trials conducted in a Commerce silt loam soil infested with only the reniform nematode and treated with the soil fumigant 1,3-dichloroprene did not result in significant yield increases and management
zones were not required. In a trial conducted in another location that had *R. reniformis* and *M. incognita* together, significant yield responses to a fumigant of as high as 1,345 kg/ha over the untreated control were reported in the management zone with lowest ECa values (designated zone 1) and yield responses declined as ECa values in the zones increased. Similar results were found in two producer fields infested with both nematodes and had yield increases from the fumigant of 1,311 and 1,204 kg/ha in zone 1. Yield responses to the fumigant were lower in both fields in management zones with higher ECa levels. These trials indicate that management zones can be successfully established in soybean especially when *M. incognita* is present.

**GLOBODERA SPECIES IN CHILE: CURRENT STATUS, GEOGRAPHICAL DISTRIBUTION AND PERSPECTIVES**

**Pacheco, H., B. Yangari, and O. Acevedo.**
Servicio Agrícola y Ganadero, Santiago, Chile.

In the context of the National Potato Health Program, the Agricultural and Livestock Service conducts specific surveys for pests that affect this crop. Cyst nematodes are among the priorities both for their quarantine status and for the serious damage they can cause in production. With the aim of increasing certainty in the diagnosis and being able to analyze a large number of samples, since 2010 molecular identification techniques have been used for *Globodera rostochiensis* and *G. pallida*. This has allowed to establish within the areas the geographical distribution of these nematode species; at the same time, the protection of the area free of quarantine pests has been achieved through the isolation of the infestation points detected. *G. rostochiensis* and *G. pallida* maintain the status of quarantine pest present in Chile. The geographic distribution is heterogeneous, distinguishing an area in which PCN are present naturally and the potato represents a subsistence crop; an infested area in which the potato is a commercial crop; and a "free area" in which both commercial production and the production of seed potatoes with the presence of PCN foci are developed. In the northern of country, 18 ° and 23 ° SL, some populations of *Globodera* have been detected that could not be identified as *G. rostochiensis* or *G. pallida* by PCR with species-specific primers. These populations are under study and it would correspond to *G. ellingtonae*. In the southern of country, 53 ° SL, *G. pallida* is widely distributed in small home gardens. In the infested lands, different control strategies have been developed, from the use of non-fumigant to biological products. Soon it is expected to have more sustainable management alternatives for the management of PCN in the different conditions exposed.

**BIOCONTROL OF PRATYLENCHUS BRACHYURUS IN SOYBEAN AND CORN WITH POCHONIA CHLAMYDOSPORIA, TRICHODERMA SP. AND DUDDTINGTONIA FLAGRANS**

**Pacheco, P. V. M., T. S. A. Monteiro, R. R. Coutinho, H. M. Balbino, and L. G. Freitas.**
Dept. of Plant Pathology, Universidade Federal de Viçosa, CEP 36570-900, Viçosa, MG, Brazil.

Currently *Pratylenchus brachyurus* is considered to be one of the most important phytosanitary problems in soybean and corn crops, mainly in the practice of succession and no-tillage, and nematophagous fungi may become a sustainable alternative for the management of this nematode. This study evaluated the use of *Pochonia chlamydosporia*, *Trichoderma* sp., and *Duddingtonia flagrans* for the biocontrol of *P. brachyurus* in soybean and corn. The first and second experiments were carried out with soybean and evaluated five isolates of *P. chlamydosporia* (Pc-3, Pc-4, Pc-10, Pc-35, and Pc-49), a *Trichoderma* sp. (T-10) at concentrations of 5,000 and 10,000 conidia.g⁻¹ soil, an isolate of *D. flagrans* (AC001), and a MIX with all isolates. In the experiment with corn, three isolates of *P. chlamydosporia* (Pc-3, Pc-10, and Pc-35) and one isolate of *Trichoderma* sp. (T-10) at the concentration of 5,000 conidia.g⁻¹ soil. The soil of each pot was infested with 1,000 specimens of *P. brachyurus*. After 60 days the nematode-related variables were evaluated in both experiments and
the agronomic characteristics in the second soybean experiment and in the corn experiment. In the first experiment, isolates Pc-3, Pc-10, Pc-35, and isolate T-10 at concentrations of 5,000 and 10,000 conidia.g⁻¹ of soil were efficient in reducing the total population of *P. brachyurus* in relation to the control treatment. The Pc-10 isolate resulted in the greatest reduction, 43.7%. In the second experiment, the isolates Pc-10, T-10 (5,000 and 10,000 conidia.g⁻¹ soil), and MIX reduced the total population of *P. brachyurus* compared to the control treatment by up to 58.7%. The T-10 isolate at the two concentrations evaluated and the MIX reduced the number of *P. brachyurus*.g⁻¹ of root by up to 56.8%, relative to the control treatment. The increase in fresh root weight was observed with the isolates Pc-3, Pc-4, T-10 (10000 conidia.g⁻¹ soil) and AC001. In corn, all isolates reduced the total population of *P. brachyurus* when compared to the control treatment. Reduction of the number of *P. brachyurus*.g⁻¹ of root was obtained with the isolates Pc-3, Pc-35 and T-10, emphasizing the isolated Pc-35 to promote a reduction of 73.2%.

*Duddingtonia flagrans* was not able to control *P. brachyurus* in any experiment. The isolates of *P. chlamydosporia* and *Trichoderma* sp. are shown to be efficient in controlling *P. brachyurus* in soybean and corn crops.


Peraza-Padilla, W., C. Cantalapiedra-Navarrete, T. Zamora-Araya, J. E. Palomares-Rius, P. Castillo, A. Archidona-Yuste. 1National University of Costa Rica, Laboratory of Nemathology, 86-3000, Heredia, Costa Rica. 2Institute for Sustainable Agriculture (ISA), CSIC (Spanish National Research Council). Menéndez Pidal Avenue, s/n, 14004 Córdoba, Spain. walter.peraza.padilla@una.cr

A new dagger nematode, *Xiphinema tica* n. sp., is described and illustrated from several populations extracted from soil associated with several crops and wild plants in Costa Rica. The new dagger nematode is characterised by a moderate body size (3276–4240 μm), a rounded lip region, ca 13.5 μm wide, separated from body contour by a shallow depression, amphidial fovea large, stirrup-shaped, a moderately long odontostyle ca 135 μm long, styllet guiding ring located at ca 122 μm from anterior end, vulva almost equatorial (50–54%), well-developed Z-organ, with heavy muscularised wall containing in the most of specimens observed two moderately refractive inclusions variable in shape (from round to star-shaped), with uterine spines and crystalloid bodies; female tail short, dorsally convex-conoid, with rounded end and a small peg, with a c' ratio ca 0.8, bearing two or three pairs of caudal pores and male absent. The unique and novel uterine differentiation based on the coexistence of a well-developed Z-organ mixed with uterine spines and crystalloid bodies in *Xiphinema* prompted us to update and include this combination of characters in the polytomy key of Loof and Luc (1990). Integrative diagnosis was completed with molecular data obtained, using D2-D3 expansion segments of 28S rDNA, ITS1-rDNA, partial 18S–rDNA and the partial mitochondrial gene cytochrome c oxidase subunit 1 (*cox1*). The phylogenetic relationships of this species with other *Xiphinema* spp. indicated that *X. tica* n. sp. was monophyletic to the other species from the morphospecies Group 4, *Xiphinema oleae*.

MOSQUITO CONTROL WITH PARASITIC NEMATODES ROMANOMERMIS IYENGARI AND STRELKOVIMERMIS SPICULATUS IN WATER OF BREEDING SITES OF MOSQUITO LARVAE [MOSQUITO CONTROL CON NEMATODOS PARASITICOS ROMANOMERMIS IYENGARI Y STRELKOVIMERMIS SPICULATUS EN AGUA DE SITIOS DE CRIA DE MOSQUITO LARVAE]

The mosquitoes are important vectors of Dengue, Malaria, Chikungunya, and Zika virus. An alternative to mosquito control is the biological control with parasitic nematodes. The objective of this research was to determine the potential of parasitic nematodes *Strelkovimermis spiculatus* and *Romanomermis iyengari* in mosquito larvae of *Culex quinquefasciatus* with in four different types of water, three types water of breeding sites of mosquito larvae (water stored in storage tank, water breeding sites of mojarras, water artificial lagoon and distilled water as a control). Three doses of nematodes (3:1, 5:1 and 10:1 nematodes for larvae) were evaluated, comparing the two nematode species. Three days after infestation a sample of 20 larvae was taken; each experimental unit (100 mosquito larvae), to deterimine the number of nematodes to emerge of each mosquito larvae (Mean infestation). The three doses of parasitic nematodes caused 100% of infestation, with dose de10 nematodes / larva. *R. iyengari* caused a MI of 8.18 (nem/larvae) and *S. spiculatus* a MI 4.55 (nem/larvae). *R. iyengari*, caused the highest MI in distilled water (control), water storage, pond water, and water from the artificial lake, with values of 8.73, 8.63, 8.13 nematodes / larva, respectively. *S. spiculatus* caused the highest MI in distilled water with a value of 5.48 nematodes / larva of mosquito and water mojarras pond, water storage tank, and pond water from the artificial lake near the river Atoyac, Oaxaca Mexico with MI 4.48, 4.31, and 3.95 nematodes / mosquito larvae, respectively

**SEASONAL POPULATIONS OF MELOIDOGYNE FLORIDENSIS, MELOIDOGYNE AREANRIA, AND RING NEMATODES IN TWO PEACH ORCHARDS, FLORIDA USA [POBLACIONES ESTACIONALES DE MELOIDOGYNE FLORIDENSIS, MELOIDOGYNE AREANRIA Y NEMATODOS DE ANILLO EN DOS PISOS DE MACHO, FLORIDA USA]**

Qiu, S.¹, J. A. Brito¹, and D. W. Dickson². ¹Division of Plant Industry, Gainesville, FL, USA. ²Univeristy of Florida, Gainesville, Florida, USA.

In Florida, peach is being promoted as an alternative crop for the declining citrus industry. *Meloidogyne* spp., are considered a limiting factor in peach production. The peach rootstock ‘Flordaguard’ was introduced as having resistance to *M. arenaria, M. floridensis, M. incognita,* and *M. javanica*. However, shortly after its release, reports began to surface that it was being damaged by *Meloidogyne* spp. The objectives of this study were to determine the seasonal population fluctuations of root-knot and ring nematodes in two peach orchards at different soil depths over a 17-month period, and to identify the plant-parasitic nematode species present. Orchard A, located in central Florida, was sampled at four soil depths (0-30; 31-60; 61-90; 91-120 cm). Orchard B, located in south Florida at two depths (0-30; 31-60 cm). Sampling at the latter was limited to the top 60 cm because of high water table. In both orchards the soil was acidic and sandy; silt and clay were less than 3% in all the soil depths. The nematodes identified in orchard A were *M. arenaria* (H3 phenotype) and *Mesocriconema ornatum*, whereas in orchard B, *M. floridensis* and *M. xenoplax*. Over all, in both orchards few roots were found below 30 cm, therefore data acquired from roots for all depths were combined. At orchard A, the number of root-knot nematodes (J2) and ring nematodes per 200 cm³ soil in the top 120-cm soil was higher in 2015 than that in 2016. The highest J2 in the soil was found at the top 30 cm. From April to June 2015, the number of J2 and ring nematodes increased at 30 cm, with the highest numbers occurring in June 2015 followed by a slow drop in August 2015. The number J2 and ring nematodes in the soil decreased following August 2015 and remained consistently low at all depths during the experiment. The highest number of eggs and J2 per gram of fresh root was found in August 2015, followed by a sharp decrease in December 2015 and an increase in March 2016. At orchard B, the number of J2 and ring nematodes per 200 cm³ soil in the top 120-cm soil was higher in 2015 than that in 2016. The highest J2 in the soil was found at the top 30 cm. Population densities of J2 in the soil peaked in June 2015 and again in March 2016. Ring nematodes had the highest number in June 2015 and April 2016. Both eggs and J2 per gram of fresh roots varied over the sampling period, and reached their peak in August 2015 and again in March 2016, respectively. In both peach orchards sampled, more root-knot and ring nematodes were found in the upper 30-cm soil depth, where most small feeder roots of peach occurred.
MORPHOLOGICAL AND MOLECULAR IDENTIFICATION OF MELODOGYNE SPECIES THAT AFFECT CAPSICUM ANNUM L. VARIETY "PIMIENTO DEL PIQUILLO", IN LA LIBERTAD, PERU [IDENTIFICACIÓN MORFOLÓGICA Y MOLECULAR DE ESPECIES DE MELODOGYNE QUE AFECTAN CAPSICUM ANNUM L. VARIEDAD “PIMIENTO DEL PIQUILLO”, EN LA LIBERTAD, PERÚ]


La Libertad, on the north coast of Peru, has climatic and edaphic conditions suitable for the cultivation of Capsicum annum L. variety "Pimiento del Piquillo", factors which allow high yields and quality. One of the phytosanitary problems that most affects this crop when it develops in sandy soils is the presence of nematodes especially of the Meloidogyne genus that cause chlorosis in the foliage, nodules or galls in the roots, less development and low yield. The objective of this research was to identify morphologically and molecularly the main Meloidogyne species that affect the crop in the area. For this, of the nodular roots were extracted females of which the perineal pattern was examined, the shape of the head of the males, the morphology of the juveniles (J2), the shape of the stylet of males and females. The molecular identification was made by amplification of ITS1 region by PCR and sequencing. Due to the morphological characteristics observed and the results of the molecular analysis, it was determined that the species that affect Capsicum annum L. variety "Pimiento del Piquillo" are M. incognita and M. javanica.

ALTERNATIVES FOR THE MANAGEMENT OF THE MELODOGYNE ARABICIDA-FUSARIUM OXYSPORUM COMPLEX IN COFFEE PLANTATIONS IN COSTA RICA [ALTERNATIVAS PARA EL MANEJO DEL COMPLEJO MELODOGYNE ARABICIDA-FUSARIUM OXYSPORUM EN CAFETALES DE COSTA RICA]

Rojas, M.1, D. Ramírez1, L. Salazar2, and G. H. Sera3. 1Instituto del Café de Costa Rica (ICAFE). 2Instituto Agronómico do Paraná (IAPAR). 3Laboratorio NEMAFITO Costa Rica. mrojas@icafe.cr

The Meloidogyne arabicida-Fusarium oxysporum complex causes significant losses in some coffee growing areas of Costa Rica. The symptoms were observed since 1974; the nematode was described by López and Salazar in 1989 and the relationship with the fungus was initially reported by Marbán et al. (1991). The disease induces corky roots, chlorosis, defoliation, fruit fall, and death of the plant. The chemical control has been ineffective and the disease has been managed with graft on Coffea canephora. The study objectives were to: a) evaluate the genetic resistance of Coffea arabica cultivars and b) assess the biological efficacy of two nematicides in nursery. Two trials were established in 2015 in Cartago, Costa Rica. In both, randomized complete block design was used, with five replications of 10 plants established in pots of 1.3 liters of substrate constituted by soil (50%), compost (25%) and rice husk (25%). Each plant was inoculated with at least 1,100 eggs + J2 of M. arabicida 120 days after sowing. In the genetic resistance test, 10 cultivars were evaluated: Caturra, Catuaí Vermelho IAC 99, IAPAR 59, IPR 98, IPR 99, IPR 100, IPR 102, IPR 103, IPR 106, and IPR 107. Seven months after the inoculation the radical nodulation index and the density of M. arabicida was evaluated. In the chemical control test, six treatments were evaluated: Absolute Control, Inoculated Control, Terbufos (0.2 g ai / plant) and Fluopyram (0.075 g ai / plant), both nematicides were applied 15 days before and 30 days after inoculation. In this test, the variables: height, number of leaves, aerial weight, root weight, radical nodulation index and density of M. arabicida was evaluated. In the chemical control test, the plants without inoculum showed less development at 180 days after the inoculation; the nodulation index was lower in the Absolute Control, Terbufos applied before
inoculation and in the two Fluopyram treatments. The lowest densities of *M. arabicida* were recorded in the Absolute Control (zero J2 / g of root), Fluopyram applied before (101 J2 / g of root) and after (94 J2 / g of root) of inoculate; in contrast to the Inoculated Control (14,109 J2 / g of root) or Terbufos applied after inoculation (16823 J2 / g of root). The Fluopyram is a good alternative for the management of the complex in preventive treatment and the cultivars IPR 100 and IPR 106 are good options for long-term cultivation.

**FUSARIUM OXYSPORUM IN CAFETALES OF COSTA RICA [FUSARIUM OXYSPORUM EN CAFETALES DE COSTA RICA]**

Rojas, M.1, D. Ramírez1, L. Salazar2 and G. H. Sera3. 1Instituto del Café de Costa Rica (ICAFE). 2Instituto Agronómico do Paraná (IAPAR). 3Laboratorio NEMAFITO Costa Rica. mrojas@icafe.cr

The *Meloidogyne arabicida*-Fusarium oxysporum complex causes significant losses in some coffee-growing areas of Costa Rica. The symptoms were observed since 1974; the nematode was described by López and Salazar in 1989 and the relationship with the fungus was initially reported by Marbán et al. (1991). The disease induces corky roots, chlorosis, defoliation, fruit fall, and death of the plant. The chemical control has been ineffective and the disease has been managed with graft on *Coffea canephora*. The study objectives were: a) evaluate the genetic resistance of *C. arabica* cultivars and b) assess the biological efficacy of two nematicides in nursery. Two trials were conducted, established in 2015 in Cartago, Costa Rica. In both, randomized complete block design was used, with five replications of 10 plants established in pots of 1.3 liters of substrate constituted by soil (50%), compost (25%) and rice husk (25%). Each plant was inoculated with at least 1100 eggs + J2 of *M. arabicida* 120 days after sowing. In the genetic resistance test 10 cultivars were evaluated: Catuра, Catuai Vermelho IAC 99, IAPAR 59, IPR 98, IPR 99, IPR 100, IPR 102, IPR 103, IPR 106 and IPR 107. Seven months after the inoculation the radical nodulation index and the density of *M. arabicida* was evaluated. In the chemical control test, six treatments were evaluated: Absolute Control, Inoculated Control, Terbufos (0.2 g ai / plant) and Fluopyram (0.075 g ai / plant), both nematicides applied 15 days before and 30 days after inoculation. In this test the variables were evaluated: height, number of leaves, aerial weight, root weight, radical nodulation index and density of *M. arabicida* at 60, 120, and 180 days after inoculation. In the genetic resistance test, the cultivars IPR 100 and IPR 106 showed more than 90% reduction in the reproduction factor compared to the susceptible variety Catuа Vermelho IAC 99, categorized as resistant and highly resistant, respectively. In the chemical control test the plants without inoculum showed less development at 180 days after the inoculation; the nodulation index was lower in the Absolute Control, Terbufos applied before inoculation and in the two Fluopyram treatments. The lowest densities of *M. arabicida* were recorded in the Absolute Control (zero J2 / g of root), Fluopyram applied before (101 J2 / g of root) and after (94 J2 / g of root) of inoculate; in contrast to the Inoculated Control (14,109 J2 / g of root) or Terbufos applied after inoculation (16823 J2 / g of root). The Fluopyram is a good alternative for the management of the complex in preventive treatment and the cultivars IPR 100 and IPR 106 are good options for long-term cultivation.


Rudon G1, W. Ramclam2, E. Salas3, D. Azofeifa4, and M. Araya5. 1Banana Growers’ Association, Big Creek, Belize. 2BELAGRO Agriculture Big Creek, Belize. 3Catedrático Universidad Nacional, Escuela Ciencias Agrarias, Costa Rica. 4AMVAC Chemical Corporation. 5LIFE-RID. maraya@life-rid.com

Banana is the most important crop in Belize, accounting for almost 13% of the agricultural gross national product. In 2017, about 84,906 tons produced on 2,791 ha were exported to the United Kingdom. Besides the constraints of banana market requirements and demands, banana...
production is limited by abiotic factors affecting yield, mainly reduced soil depth, poor soil texture and structure. Among the biotic factors limiting banana production, banana root nematodes are second after black Sigatoka disease caused by *Mycosphaerella fijiensis* Morelet, reducing leaf emission, bunch weight, and plantation longevity, and increasing the crop cycle duration. Then, an analysis of the plant-parasitic nematodes occurring in 19 commercial banana (*Musa* AAA) farms in Belize from 2005 to 2017 was undertaken. Nematode extraction was done using 25 g of fresh roots macerated in a kitchen blender, followed by recovering the nematodes with a 0.025 mm sieve. The data were subjected to frequency analysis in PC-SAS, and the absolute frequency for each genus was calculated as a percentage (number of samples containing a species / numbers of samples collected) * 100. Two plant-parasitic nematode species were detected and based on their frequencies and population densities their relative importance was established as: *Helicotylenchus multicinctus* > *Radopholus similis*. *Helicotylenchus multicinctus* was the most abundant species, accounting for 50 to 70% of the overall root nematode population throughout each study year. From a total of 7329 root samples, 7035 (96%) contained *H. multicinctus* and 6539 (89%) *R. similis*, and when the nematodes present in the samples were pooled, (total nematodes) only 180 (2.5%) of the samples were free of nematodes. A larger number of samples with a nematode population above the economic threshold suggested by the Belize Banana Grower Association (100 nematodes per gram of roots) were observed in all the years, the months and the two sampled districts where bananas are grown in Belize. The statistical differences (*P* < 0.0001) detected for the nematode frequencies among the years, months and districts, more likely were associated with the high number of samples included in each year, month and district, because the variations in the frequencies for each nematode genus were small.

**DISTRIBUTION OF *PRATYLENCHUS PENETRANS* IN SAND AND LOAMY SAND SOILS IN RELATION TO EDAPHIC FACTORS |DISTRIBUCCIÓN DE *PRATYLENCHUS PENETRANS* EN EL SUELO DE LA ARENA Y LOAMY EN RELACIÓN CON FACTORES EDÁFICOS**

*Saeed, I. A.*, G. D. Pack, and A. E MacGuidwin. Department of Plant Pathology, University of Wisconsin, Madison, Wisconsin 53706, USA. aem@plantpath.wisc.edu

*Pratylenchus* spp., particularly *P. penetrans* (Pp), are important pests of irrigated crops grown in the sandy soils of central Wisconsin. Estimates of nematode population densities are used to make decisions about crop rotation and the use of soil fumigation, nematicides, and seed treatments. The distribution of Pp is known to be aggregated, as are many soil characteristics. The objective of our project was to determine if there is a correspondence between variation in edaphic factors and nematode population densities within fields. We studied four types of soil characteristics; data that are: 1) observable by farmers such as elevation and relative soil moisture, 2) available from standard soil tests such as pH and soil macronutrients, 3) available from specialized soil tests such as micronutrients and heavy metals, and 4) available from contracted consultants such as apparent electrical conductivity. Nine commercial fields were sampled in the spring, four in 2016, and five in 2017. Eighteen to sixty geo-referenced locations were sampled per field. Nematodes were recovered from 100 cm³ subsamples of soil and the root fragments therein, using sieving/sucrose flotation and incubation techniques, respectively. One field was too sparsely populated with Pp to be used. Median population densities in the other fields ranged from 1 to 15 Pp cm⁻³ and the maximum number among locations within fields ranged from 12 to 73 cm⁻³. Standard soil tests were performed by our lab and specialized soil tests were performed by a commercial lab. Nematode data was rescaled using a log10(x+1) transformation and data for the soil variables was rescaled to fall between 0 and 1 for each field. The least absolute shrinkage and selection operator (LASSO) was used to reduce the number of independent variables to test in a general linearized model generated using PROC GLIMMIX (SAS Institute). Data representing all four types of soil characteristics collected from 224 locations in six fields suggested a model with five positive coefficients (altitude and ppm copper, manganese, sodium, and barium) and two negative coefficients (percent nitrogen and ppm potassium). Some models for individual fields
included different variables such as percent sand and subsoil apparent electrical conductivity. This study illustrates the challenge of finding a suite of variables with universal utility for locating "hotspots" of Pp within fields. The ideal correlates would be stable features that are inexpensive to measure. Heavy metals, such as those revealed by our study, fit the first criteria and merit further evaluation for their association with nematode abundance.

DYNAMICS OF SOIL NEMATODE COMMUNITIES IN TOMATO CROPS FROM A PRODUCTIVE REGION OF ARGENTINA [DINÁMICAS DE COMUNIDADES DE NEMATODOS DEL SUELO EN CULTIVOS DE TOMATE DE UNA REGIÓN PRODUTIVA DE ARGENTINA]

Salas, A. and M. F. Achinelly; CEPAVE, CCT-La Plata, CONICET-UNLP calle 60 y 120 s/n, Bs. As, Argentina. augustosalas@cepave.edu.ar

The objective of this work was to evaluate nematode communities through ecological indexes to determine those variables that are the best indicators of changes in soils subjected to tomato crops under two different agricultural management systems, in La Plata, Buenos Aires province, the main tomato producing region of Argentina. A seasonal crop and an intensive conventional crop with use of agrochemicals were considered for the study. Site A was characterized by tomato crops during spring and summer season and covered by weeds the rest of the year, using organic fertilizers as the only external income. Site B was represented by an intensive conventional culture with methyl bromide and Agrocelhone application as nematicide before the seedtime the first year and towards the end of the second year of sampling respectively. Soil samples were collected between September 2015 and October 2017. At each site, a single sample, totaling a minimum volume of 500 cm³, was collected; this comprised multiple soil cores (c. 20), taken to a depth of 20 cm using a steel auger (4-cm diameter). A total of nine samples were taken for each site. In lab the technique of centrifugation was performed for nematode isolation. The determination at the genus level and the count of nematodes was carried out to obtain the values of abundance per site. To identify changes in the structure of the nematode community, diversity and function indexes were analyzed and tested to search differences between sites. Following multivariate analysis were considered: ANOSIM to determine significant differences between sites, SIMPER to estimate the contribution of each taxon to average Bray Curtis dissimilarity and MDS to model the proximity (similarity) between the samples. The results showed significant differences between Richness (= N0), abundance, Margalef (d) and Σ MI, no differences being observed in MI, Shannon-Wiener, Pielou and other diversity indices analyzed. Significant differences were found between the sites using the ANOSIM test (global R= 0.5, p value=0.002). SIMPER analysis indicated that Helicotylenchus, Filenchus, and the opportunistic enrichment nematode Rhabditis were mainly responsible for the dissimilarity between sites. The MDS indicated evident separation of samples taken after the application of methyl bromide and Agrocelhone. ΣMI showed changes in the community structure of soil nematodes. This could be related to the inclusion of plant-feeding nematodes in this index. Similar values for MI between sites could be related to the abundance of bacteriophage nematodes due to enrichment of the environment with organic fertilizers (Site A) and disturbance for fumigants (Site B). As results of this work, we present the most sensible variables in nematode communities, in soils under horticultural management in the city La Plata. The lack of studies in this region encourages to keep on this type of analysis to estimate changes in the diversity of nematofauna as measure of ecological quality for these agricultural soils.

ENTOMOPATHOGENIC NEMATODES IN LATIN-AMERICA: THEIR HISTORY AND SCIENTIFIC PRODUCTION IN NUMBERS [NEMATODOS ENTOMOPATÓGENOS EN LATINOAMÉRICA: SU HISTORIA Y PRODUCCIÓN CIENTÍFICA EN CIFRAS]

San-Blas, E. Laboratorio de Protección Vegetal, Instituto Venezolano de Investigaciones Científicas, Calle 79 con Av. 8, Maracaibo, Venezuela C.P. 4001. esanblas@yahoo.com

The study of entomopathogenic nematodes (EPN) in Latin America (LATAM), is limited to a few groups that have generated an important number
of discoveries and advances comparable to the rest of the world. Many of those results are not published in international journals and are kept in thesis dissertations, congresses abstracts or proceedings, divulgation means or in the best of the cases in their original languages (Spanish or Portuguese) in national journals with difficult access. A data base was built using 302 scientific papers recovered from different sources and classified by country of origin, date of publication and theme. The first written experiences produced in LATAM were done by Dutki in Chile in 1954 and Tang in Peru in 1958 with the introduction of *Steinernema carpocapsae* to control different insect pests. Since then, the number of publications has been increasing. Before 1989, 23 papers were published in the present decade, 123 have been published so far. Fifty-seven percent of those papers were published in national journals and the rest in international ones. Brazil, Colombia, Cuba, Chile, and Peru were the most publishing countries up to 1989, by 26%, 18%, 17%, 9% and 4%, respectively. Between 1990 and 1999, Argentina (52%), Cuba (12%), México (11%), Colombia (11%), and Puerto Rico (11%) gathered the majority of the published papers. In the present decade Brazil (31%) has retained first place of publishing countries followed by México, Cuba, Colombia, and Venezuela (18% 13%, 13%, and 9% respectively). Up today more than 170 insect species have been targeted and tested against EPN in different labs from LATAM, but other organisms such as ticks and plant-parasitic nematodes have also been targeted. On the other hand, 16 out of 116 recognized EPN species have been isolated and described from LATAM. It is evident that the experience gained by Latin-American groups is vast, especially in biological control tests of tropical and endemic insects in laboratory conditions. However, the introduction of EPN in field trials and at commercial levels remains in the early stages. Augmenting EPN applications in the field, gaining the interest of policy makers, creating the conditions to innovate *in vitro* mass production, revisiting and broadening extension and educational programs at all levels, keeping national sampling projects, and enhancing the Latin-American research cooperation are obligate activities to improve the presence of EPN in the field. Nevertheless, there is a real consensus between farmers and consumers about the benefits of reducing chemical inputs in agriculture nowadays, and this represents an opportunity to EPN to become common, reliable, and available biocontrol products in LATAM.

**ASSESSING QUALITY AND PERFORMANCE OF ENTOMOPATHOGENIC NEMATODES EVALUATING THEIR ENERGY RESERVES BY USING INFRARED SPECTROSCOPY (FTIR-ATR)**

San-Blas, E.¹, M. Guerra², E. Portillo¹, N. Cubillán³ and G. Paba¹. ¹Laboratorio de Protección Vegetal, Instituto Venezolano de Investigaciones Científicas, Calle 79 con Av. 8, Maracaibo, Venezuela C.P. 4001. ²Laboratorio de Polímeros, Universidad del Zulia, Maracaibo, Venezuela C.P. 4001. ³Grupo de Investigación de Biotecnología de Microrganismos, Fisicoquímica Aplicada y Estudios Ambientales, Universidad del Atlántico, Barranquilla, Colombia. esanblas@yahoo.com

The main objective of this research was to evaluate the energy reserves of the entomopathogenic nematodes (EPN) *Heterorhabditis indica* and *Steinernema riobrave*, by using ATR/FTIR characterization, principal component analysis (PCA), two-dimensional correlation spectroscopic (2DCoS), and integration of absorption bands. In addition, the levels of such reserves were associated with survival and infectivity of infective juveniles (IJ) towards *Galleria mellonella* according to their storage temperature: T₁ (10°C), T₂ (20°C) and T₃ (30°C). These results suggest that lipids (especially triglycerides) represent the main source of energy in both species, whereas glycogen and trehalose act as alternative sources. In the case of *H. indica*, the highest percentages of infectivity were obtained by IJs from T₂, while those from T₃ were unable to infect *G. mellonella* larvae after 8 wk of storage. On the other hand, the IJs of *S. riobrave* were more virulent compared to those of *H. indica*; indeed, they were
more tolerant to low ($T_1$) and high temperatures ($T_3$). Thus, it was evidenced that energy reserves and infectivity are influenced by storage. Results similar to those presented in this study have been reported and conclude that Steinernematids survive much longer at high temperatures compared to Heterorhabditids ones. However, this depends on the species and the regulatory mechanisms involved in the acquisition of thermo-tolerance, such as saturation and desaturation of fatty acids and accumulation of stabilizing compounds of cell membranes, such as trehalose. Thus, we present pioneering results in the quality of EPN, which are necessary for their formulation and also to predict their performance as biological control agents.

**CONTROL OF MELOIDOGYNE ENTEROLOBII IN TOMATO BY THE NOVEL SULFONAMIDE NEMATICIDE SALIBRO™ [CONTROL DE MELOIDOGYNE ENTEROLOBII EN TOMATE POR LA NOVELA SULFONAMIDA NEMATICIDA SALIBRO™]**

*Silva, M. G.*¹, A. Ferreira¹, C. Vassallo², J. A. Wiles³, and T. C. Thoden⁴. ¹Corteva Agriscience™, Agriculture Division of DowDuPont™, DuPont do Brasil S.A. Alameda Itapecuru 506, Alphaville, Barueri, SP, 06454080, Brazil. ²Corteva Agriscience™, Agriculture Division of DowDuPont™, Dow AgroSciences, Boulevard Cecilia Grierson 355, Dique IV, Piso 25, Puerto Madero (C1107CPG), Ciudad Autónoma de Buenos Aires, Argentina. ³Corteva Agriscience™, Agriculture Division of DowDuPont™, DuPont (U.K.) Limited, 4th Floor, Kings Court, London Road, Stevenage, SG1 2NG, United Kingdom. ⁴Corteva Agriscience™, Agriculture Division of DowDuPont™, DuPont de Nemours (Deutschland) GmbH, Hugenottenalle 175, 63263 Neu-Isenburg, Germany. CVassallo@dow.com

Tomato yields can be far below their genetic potential due to root infection by root-knot nematodes. Integrated nematode control approaches, including biological control agents, nematicide applications, and resistant varieties, are being employed by growers to reduce nematode populations in the field. One root-knot nematode species increasing in importance is *Meloidogyne enterolobii*. This species appears to have the ability to overcome resistance mechanisms of some host plants, including tomato. In addition, some nematicides have been banned or are restricted in use in several countries because of their unfavorable toxicological and environmental profiles, which has reduced the control options available to growers. Fluazaindolizine is a novel sulfonamide nematicide active ingredient that is being developed and can become a new, effective, tool for nematode management in tomatoes and several other crops. The formulated product is Salibro™ 500SC. The objective of this study was to measure root damage and tomato yield after Salibro™ application for the control of *M. enterolobii*. The experiment was conducted in a commercial field area with high nematode pressure in São Paulo State, Brazil. Salibro™ was applied either by drip application or in furrow spray applications one day before tomato transplanting. A commercial biological product (containing *Bacillus subtilis* + *Bacillus licheniformis*) as well as an untreated control were included for comparison. At 30 and 60 days after treatment application (DAA), plants were collected at random from each treatment to determine nematode population development. The roots of each plant were evaluated for the presence of galls according to the standard gall index scale 0-10 (Bridge and Page, 1980) and also sent to an external laboratory for root extraction to obtain the number of nematodes per gram of root. No statistically significant differences amongst treatments were noted in root nematode counts, however Salibro™ treatments clearly reduced gall damage on tomato roots at 60 DAA. Salibro™ applied by drip application and in furrow application increased the yield by an average of 8.8 and 7.1 tons/ha, respectively, when compared with untreated check, and by 8.1 and 6.4 tons/ha respectively when compared with the biological product. This study demonstrated that Salibro™ increased tomato yield in a field area infested with *M. enterolobii* and will be a useful tool for management of this nematode species.

**PRODUCTION OF INFECTIVE JUVENILES (IJ) OF HETERORHABDITIS SP. USING GALLERIA MELLONELLA AND SPODOTERA FRUGIPERDA AS HOSTS [PRODUCCIÓN DE JUVENILES INFECTIVOS (IJ) DE HETERORHABDITIS]**
**50th Annual Meeting of the Organization of Tropical Nematologists of America**

*ABST 43*

**SP. USANDO COMO HOSPEDEROS GALLERIA MELLONELLA Y SPODOPTERA FRUGIPERDA**


The management of pests is based mainly on the application of synthetic insecticides, which unbalance the ecosystem, destroy beneficial organisms and, in some cases, allow the plague to develop populations resistant to insecticides. The entomopathogenic nematodes of the genus *Heterorhabditis* are an important option, since they have a wide range of hosts, high search capacity, and high virulence. The objective of this study was to evaluate the production level of infective juveniles (IJ) of *Heterorhabditis* sp. using *Galleria mellonella* and *Spodoptera frugiperda* as hosts. For this purpose, the two hosts were raised in the laboratory and when they reached the last stage of larval development, 50 larvae of each species were taken at random. Each larva was weighed, measured, and subsequently isolated in disposable deposits by placing paper towel in the base. Then, it was sprinkled with 20 juveniles (IJ). The evaluations were carried out after 24 hours observing a slow movement only in *S. frugiperda*. At 36 hours cream to soft orange change was observed only in *G. mellonella* and 48 hours later there was 98% mortality of larvae in *G. mellonella* and 78% in *S. frugiperda*. Twenty days later the total was obtained of infective juveniles (IJ) produced by larva being the average in *G. mellonella* of 326-280 juveniles and in *S. frugiperda* 246-490 juveniles. The results obtained allow us to conclude that *G. mellonella* is the most efficient host.

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**EFFECT OF ORGANIC NEMATICIDES TO CONTROL DRY ROT DISEASE OF YAMS (DIOSCOREA ROTAUNDATA) IN PUERTO RICO [EFFECTO DE LAS NEMATICIDAS ORGÁNICAS PARA CONTROLAR LA ENFERMEDAD DE ROTE SECO DE LOS YAMS (DIOSCOREA ROTAUNDATA) EN PUERTO RICO]**

**Soto-Ramos, C. M.,** M. Feliciano-Rivera, and J. M. Cardona. Department of Agro-environmental Science, University of Puerto Rico, Mayagüez Campus. P.O. Box 9030, Mayagüez, PR 00681-9030. casiani.soto@upr.edu

White yam (*Dioscorea rotundata*) belongs to the Dioscoreaceae family. Yam are the most important tuber root crops in Puerto Rico. Local production has been affected by the lack of pathogen-free seed, thus we are unable to supply local demand in Puerto Rico. *D. rotundata* is highly susceptible to nematode infection, affecting tubers from the field through storage, reducing seed quality and availability. *Pratylenchus coffeae*, is the causal agent of dry rot disease. During storage, weight loss up to 50% and promotion of other pathogen infection, had been accounted to nematode presence. Storage practices performed by farmers have not proved effective in improving seed quality. Thus, the objective of this research was to assess the effectiveness of two oil-based nematicides to control dry rot disease. The experiment was conducted using a randomized complete block design with four repetitions. Treatments included: S1-Sesamin EC®, S2-Sesamin EC® (x2), S3-Sesamin EC® + Oxamyl, N1- Nemakill™ and V1- Oxamyl) and a non-treated control. The population of *P. coffeae* in soil and tubers was determined three times during the cycle following the Christie and Pery protocol with modifications. Effectiveness of the treatments was evaluated based on nematode population, yield, and tuber weight loss after the storage period. Nemakill™ resulted in being the least effective treatment to reduce the population of *P. coffeae* at the end of the crop cycle. None of the Sesamin EC® doses and the labeled nematicide (Oxamyl) provided adequate control of the populations of *P. coffeae*. There were no significant differences in the yield between treatments. However, the positive control (Oxamyl) obtained the lowest yield, while the S2 treatment obtained the highest yield; with 10% difference. Nemakill™ treated tubers showed losses of more than 25% during the storage period, while the treatment S1, S2, and S3 obtained losses of 21%, 24% and 22%, respectively. The difference between the positive control (Oxamyl) and the negative control (C) was 1%. The range of weight loss was between 20-26%.
disease showed no significant differences between the treatments. Weight loss ranged from 5 to 17% compared to the natural seed weight loss. The ineffectiveness of Oxamyl proposes the use of other control alternatives to manage dry rot disease. Although the treatments were not effective in reducing the population of *P. coffeae*, it could be observed that the percentage of weight loss associated with the disease during storage was low. This research is a steppingstone to develop new sources of control for nematodes in tuber crops such as white yam.

POTATO CYST NEMATODES IN THE UK: UNDERSTANDING THE PROBLEM AND THE SEARCH FOR NEW CONTROL MEASURES [NEMATODO QUISTE DE PAPA EN EL REINO UNIDO: ENTENDIENDO EL PROBLEMA Y LA BÚSQUEDA DE NUEVAS MEDIDAS DE CONTROL]  

Thorpe, P.1, C. J. Lilley2, E. G. J. Danchin3, D. R. Laetsch4, M. Da Rocha5, C. Rancurel1, N. E. Holroyd5, J. A. Cotton5, A. Szitzenberg6, E. Grenier7, J. Montarry7, B. Mimec8, M. Duceppe8, I. Boyes9, J. M.C. Marvin2, L. M. Jones2, H. B. Yusup2, J. Lafond-Lapalme8, M. Esquierl7, M. Sabeh8, M. Rott8, H. Overmars10, A. Finkers-Tomczak10, G. Smant10, G. Koutsovoulos84, V. Blok1, S. Mantelin1, P. J. A. Cock1, W. Phillips11, B. Henriass12,13, P. E. Urwin13, M. Blaxter4, J. T. Jones1, 14, A. Reid15, J. Pickup15, E. Anderson16, and S. Eves-van den Akker17. 1James Hutton Institute, Dundee, DD2 5DA, UK. 2Centre for Plant Sciences, University of Leeds, Leeds, LS2 9JT, UK. 3INRA, Univ. Nice Sophia Antipolis, CNRS, UMR 1355-7254 Institut Sophia Agrobiotech, 06900 Sophia Antipolis, France. 4Institute of Evolutionary Biology, University of Edinburgh, EH9 3FL, UK. 5Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Cambridge, CB10 1SA, UK. 6School of Biological, Biomedical and Environmental Sciences, University of Hull, Hull, HU6 7RX, UK. 7INRA, UMR1349 IGEPP (Institute for Genetics, Environment and Plant Protection), F-35653 Le Rheu, France. 8Agriculture and Agri-food Canada, Horticulture Research and Development Centre, 430 Bboul. Gouin, St-Jean-sur-Richelieu, Quebec, J3B 3E6, Canada. 9Sidney Laboratory, Canadian Food Inspection Agency (CFIA), 8801 East Saanich Rd, Sidney, BC V8L 1H3, Canada. 10Laboratory of Nematology, Department of Plant Sciences, Wageningen University, Droevendaalsesteeg 1, 6708 PB Wageningen, The Netherlands. 11USDA-ARS Horticultural Crops Research Laboratory, Corvallis, Oregon, USA. 12CNRS UMR 7257, INRA, USC 1408, Aix-Marseille University, APMB, 13288 Marseille, France. 13Department of Biological Sciences, King Abdulaziz University, Jeddah, Saudi Arabia. 14School of Biology, University of St Andrews, North Haugh, St Andrews, KY16 9TZ, UK. 15Science and Advice for Scottish Agriculture, Edinburgh, EH12 9FJ, UK. 16Scottish Agronomy Ltd, Arlary Farm, Milnathort, Kinross, KY13 9SJ. 17Department of Plant Sciences, University of Cambridge, Cambridge CB2 3EA.

The potato cyst nematodes, Globodera rostochiensis and G. pallida, cost UK agriculture over £60 million per year and are a major constraint on the potato industry. Distinct populations of *G. pallida* exist in the UK. These populations differ in their ability to overcome sources of resistance and, therefore, cultivar choice is important. However, given that resistance-breaking populations are already present in UK fields, the search for future control mechanisms is similarly important. There is thus a need to both understand the current state of the potato cyst nematode problem, and prepare for the future. We have been employing genetic/genomic approaches to these ends. To understand the current state of the potato cyst nematode problem, we have developed a method to catalogue the genetic variation of *G. pallida* populations found in the UK. This diversity relates to the presumed historical introductions of small proportions of the total genetic diversity found for these species in South America. Nevertheless, we find that most fields contain a single genotype, one fifth contain a mix of genotypes, and less than 3% contain all three prevalent genotypes. Within mixed fields, we are able to quantify the relative abundance of each genotype, and describe a complex “pathoscape” within each field. In an effort to expedite the development of novel control mechanisms in the future, we launched a community-wide consortium to analyse the genome of *G. rostochiensis*. We have generated a draft genome assembly for *G. rostochiensis* to identify ‘parasitism genes’, to map gene expression through the life cycle focusing on key
parasitic transitions, and to explore the genetic variation underlying eight populations including four additional plant resistance-breaking pathotypes. Importantly, we identified a putative regulatory DNA motif enriched in the promoters of a subset of parasitism genes. This six-base pair DOrral Gland box (DOG box) is present in the promoter region of representatives from 26 of the 28 dorsal-gland effector families. Using the DOG box, we predicted a superset of putative parasitism genes, validate these predictions, and explore the implications for developing novel control mechanisms.

SALIBRO™ — A NOVEL SULFONAMIDE NEMATICIDE FOR THE CONTROL OF PLANT-PARASITIC NEMATODES IN BRAZIL [SALIBRO™ — UN NEMATICIDIO DE SULFONAMIDA NOVELA PARA EL CONTROL DE NEMATODOS PARASITICOS VEGETALES EN BRASIL]

Vassallo, C. N.¹, M. G. Silva², A. Ferreira², J. Mejia³, J. A. Wiles³, and T.C. Thoden⁴. ¹Corteva Agriscience™, Agriculture Division of DowDuPont™, Dow AgroSciences Argentina SRL, Boulevard Cecilia Grierson 355, Dique IV, Piso 25, Puerto Madero (C1107CPG), Ciudad Autónoma de Buenos Aires, Argentina. ²Corteva Agriscience™, Agriculture Division of DowDuPont™, DuPont do Brasil S.A. Alameda Itapecuri 506, Alphaville, Barueri, SP, 06454080, Brazil. ³Corteva Agriscience™, Agriculture Division of DowDuPont™, DuPont de Colombia. Calle 113 # 7-21 Torre A Piso 14, Bogotá, Colombia. ⁴Corteva Agriscience™, Agriculture Division of DowDuPont™, DuPont (U.K.) Limited, 4th Floor, Kings Court, London Road, Stevenage, SG1 2NG, United Kingdom. ⁵Corteva Agriscience™, Agriculture Division of DowDuPont™, DuPont de Nemours (Deutschland) GmbH, Hugenottenallee 175, 63263 Neu-Isenburg, Germany. cvassallo@dow.com

Several species of nematodes, such as root-knot, lesion, reniform, and cyst nematodes, have been causing significant losses of productivity in different important crops. Both increased awareness of the damage and yield loss that nematodes cause to crops and sustainable nematode control strategies are essential for growers to optimize crop productivity. There are several tools available for the control of nematodes, such as resistant or tolerant crop varieties, antagonist plants, biological control agents, and chemical nematicides. All of them must be considered as part of a management plan for these pests. Recently, some nematicides have been banned or restricted due to their toxicological and environmental profile, thereby reducing the alternatives for chemical control. New products are needed that combine efficiency and safety for users and the environment. Salibro™ (a 500SC formulation containing the active ingredient fluazaindolizine) is a novel sulfonamide nematicide that is being developed by Corteva AgriScience™, Agricultural Division of DowDupont™. This paper will describe field trials conducted to characterize the efficacy of Salibro™ for the control of some key plant-parasitic nematodes (Pratylenchus brachyurus, Pratylenchus zeae, Meloidogyne javanica, and Meloidogyne incognita) in soybean, sugarcane, potato, and tomato crops in Brazil. Different application methods such as in-furrow, drip, and broadcast were tested according to practices that may be employed by growers. Crop-relevant parameters were assessed to determine the efficacy of this new compound, including root and plant vigor, nematode root counts and crop yield. The results showed that Salibro™ provides excellent control of a range of plant-parasitic nematode species, providing root protection and improved yield. Salibro™ will be a promising, new, and effective tool for nematode management in Brazil.

PHYTOPARASITIC NEMATODES ASSOCIATED WITH REPRESENTATIVE CROPS OF PERU [NEMATODOS FITOPARÁSITOS ASOCIADOS CON CULTIVOS REPRESENTATIVOS DEL PERÚ]

Vera-Obando, N. Laboratorio de Ecología Microbiana y Biotecnología, Departamento de
Perú tiene un amplio rango de condiciones geográficas y climáticas favorables para el establecimiento de nematodos parásitos de plantas, que afectan una variedad amplia de cultivos causando pérdida de rendimiento. Nudos de raíz (RKN, Meloidogyne spp.), son ampliamente distribuidos en Perú, y estrategias de control de nematodos incluyen las características taxonómicas y la interacción específica con los cultivos. Se realizaron estudios para identificar especies de Meloidogyne de treinta poblaciones. Cada uno fue obtenido de un solo masa de huevos a través de caracterización morfológica y amplificación del ARN con primers específicos SCAR. Veinticinco poblaciones mostraron productos de amplificación con MIF/MIR y Inc-14k F/Inc-14k R primers para M. incognita, una población afectando tabaco de Lambayeque fue identificada como M. javanica con Fjav/Rjav primers, una población afectando uvas de Lima fue identificada como M. arenaria con Far/Rar primers, y M. hapla fue identificada en una población afectando aguaymanto de Cajamarca con las DHF / DHR primers. Todos estos resultados se ajustaron con la identificación morfológica según el patrón perin al. Dos poblaciones no mostraron productos de amplificación con los primers probados. Además, técnicas de identificación morfológica fueron realizadas para determinar cuáles nematodos estaban asociados con los cultivos representativos de Amazonas, Perú, así como su frecuencia y densidad de población.

Pratylenchus penetrans es un parásito económico importante de frutas blandas capaz de causar pérdida significativa de rendimiento. Las restricciones sobre fumigantes de suelo han generado interés en estrategias de manejo alternativas, particularmente aquellas que pueden suprimir los nematodos parasitarios del suelo a través de la inducción de suavidad del suelo. Este estudio evaluó los efectos de los enmiendas orgánicas sobre: (1) crecimiento temprano y rendimiento de cereza dulce, (2) P. penetrans dinámicas de población, (3) suelo biológica supresión de nematodos parasitarios del suelo, y (4) la abundancia de microorganismos colonizadores del rizosfera asociados con la supresión de enfermedad. Un guarnizo de manzana fue replantado en 2014 con cerezas dulces. Antes de plantar, se aplicaron las siguientes tratamientos de suelo: (1) compostaje, (2) mangle de madera, (3) compostaje y mangle de madera, (4) fumigación y (5) sin tratamiento. En la primera temporada de crecimiento sólo la fumigación aumentó el crecimiento de plantas en relación con el control sin tratamiento. En la segunda temporada de crecimiento la fumigación y la combinación de compostaje y mangle de madera
increased plant growth. In the third and fourth growing seasons, all soil treatments increased plant growth relative to that of the untreated control. Fumigation reduced *P. penetrans* populations at the time of planting; however, by the end of the first growing season, root and soil populations did not differ from that of the untreated control. Compost and bark chip mulch, applied alone or in combination, suppressed soil and root populations of *P. penetrans* over the first three growing seasons, and enhanced soil biological suppressiveness to plant-parasitic nematodes. Compost increased rhizosphere populations of antibiotic-producing bacteria, root colonization by arbuscular mycorrhizal fungi, and soil microbial activity. Bark chip mulch increased rhizosphere populations of total fungi and soil microbial activity. Overall, compost and bark chip mulch show potential to suppress *P. penetrans* and improve replant establishment of sweet cherry trees in old orchard sites.

**HISTOLOGICAL AND TRANSCRIPTOMIC CELL WALL RESPONSES OF UPLAND COTTON (*GOSSYPIUM HIRSUTUM*) AND SOYBEAN (*GLYCINE MAX*) ROOTS INFECTED BY RENIFORM NEMATODE (*ROTYLENCHULUS RENIFORMIS*) [RESPUESTAS HISTOLÓGICAS Y TRANSCRIPTÓMICAS DE PARED CELULAR DE ALGODÓN UPLAND (*GOSSYPIUM HIRSUTUM*) Y SOYA (*GLYCINE MAX*) INFECTADAS POR NEMATODO RENIFORME (*ROTYLENCHULUS RENIFORMIS*)]

Wei, L.¹, N. Redding¹, P. Agudelo¹, R. R. Powell², T. F. Bruce², and C. Wells³. ¹Department of Plant and Environmental Sciences. ²Clemson Light Imaging Facility, ³Department of Biological Sciences, Clemson University, Clemson, SC, 29634.

Parasitism of reniform nematode (*Rotylenchulus reniformis*, RN) on host plants induces the formation of a permanent feeding structure, the syncytium. Syncytia are regions of hypertrophic, interconnected pericycle cells with partially lysed cell walls and disorganized cytoplasm. We investigated how plant cell wall-associated genes were manipulated in RN-infected upland cotton and soybean roots. We also investigated modifications of cell wall components in RN-induced syncytia of cotton. Cotton and soybean plants were cultured in a split-root growth system where half of each root system was inoculated with 3,000 nematodes. At 3, 9, and 12 days after inoculation (DAI), root tissue was harvested for transcriptome sequencing to identify differentially expressed (DE) genes and significantly enriched/depleted gene sets (FDR < 0.05). Cell wall-associated DE genes and gene sets were identified based on gene ontology descriptions. In cotton, 45 DE genes and 29 enriched/depleted gene sets appeared to be involved in modification of cellulose, hemicellulose, pectin, and cell wall proteins, as well as in plant defense and cell-to-cell communications. Four DE genes and 6 significantly depleted gene sets were annotated as pectin-modifying proteins that may contribute to high levels of methyl-esterified pectin in RN-infected roots. Antibodies LM19 and LM20 were used to target low/non methyl-esterified pectic homogalacturonan (HG) and highly methyl-esterified pectic HG. Fluorescence imaging revealed that RN-induced syncytium walls had more methyl-esterified pectic HG than unmodified pericycle cell walls (*p* < 0.05). An LM15 antibody was also used to target xyloglucan in hemicellulose. The distribution of weak xyloglucan fluorescence overlapped with areas that also contained less methyl-esterified pectic HG, suggesting that pectate cross-links may play a greater load-bearing role than cellulose-xyloglucan network. In soybean, RN infection was associated with differential expression of 450 cell wall-associated genes and enrichment/depletion of 34 gene sets (FDR<0.05). The pattern of transcriptomic changes differed from that of cotton. In cotton, the largest number of DE genes were down-regulated at day 9 and 12 when obvious cell wall lysis was observed. In soybean, after an initial down-regulation at 3 DAI (190 DE genes), a larger number of DE genes was up-regulated at 9 and 12 DAI. Cell wall-associated gene sets were generally depleted in RN-infected cotton but significantly enriched in RN-infected soybean. Both transcriptomic data and immunolabeling revealed RN manipulation of cell wall genes and cell wall components in cotton. Soybean presented a different transcriptomic response to RN infection, and further histopathological studies are needed to
visualize how specific cell wall components are altered in RN-infected soybean roots.

**BIOLOGICAL NEMATICIDES FOR ROOT AND TUBER CROPS \[NEMATICIDAS BIOLÓGICAS PARA CULTIVOS DE RAÍCES Y TUBEROS\]**


Over 95% of the world’s annual production of yams comes from West Africa, including Nigeria and Benin. Of the various constraints affecting yam production, the root-knot nematode (*Meloidogyne* spp.; RKN) is one of the most destructive plant-parasitic nematodes. Pesticides are available for nematode control, but are expensive, not readily available to smallholder farmers in Africa, and toxic to the user and environment. Biological control of agricultural pests minimizes the risk of exposure, unlike toxic synthetic pesticides, and promises the potential to offer season-long control through systemic inoculation of the crop and root rhizosphere. Effective biopesticide solutions maintain durability through multiple modes-of-action which minimizes the development of pest resistance. Moreover, unlike synthetic pesticides, naturally occurring microbes could feasibly be produced and distributed locally with far less requirements for capital and technical infrastructure than synthetic pesticides. AgBiome has established a discovery and development pipeline focused on identification, development, and production of field-useable, stable microbes and is well-positioned to develop a nematicidal biological that satisfies the requirements of an effective RKN control product for small-farm yam production in West Africa.

**GLOBAL BIOLOGICAL INTERPRETATIONS OF THE NOVEL SELECTIVE SULFONAMIDE NEMATICIDE SALIBRO™ IN CONTROLLING PLANT-PARASITIC NEMATODES: LABORATORY TO FIELD \[INTERPRETACIONES BIOLÓGICAS MUNDIALES DEL NEMATICIDIO DE SULFONAMIDA SELECTIVA NOVELA SALIBRO ™ PARA EL CONTROL DE LOS NEMATODOS PARASITICOS DE LAS PLANTAS: LABORATORIO DE CAMPO\]**


1°Corteva Agriscience™ Agriculture Division of DowDuPont™, DuPont (U.K.) Limited, 4th Floor, Kings Court, London Road, Stevenage, SG1 2NG, United Kingdom. 2°Corteva Agriscience™ Agriculture Division of DowDuPont™, DuPont de Nemours (Deutschland) GmbH, Hugenottenalle 175, 63263 Neu-Isenburg, Germany. 3°Corteva Agriscience™ Agriculture Division of DowDuPont™, Dow AgroSciences, Boulevard Cecilia Grierson 355, Dique IV, Piso 25, Puerto Madero (C1107CPG), Ciudad Autónoma de Buenos Aires, Argentina. john.wiles@dupont.com

During the last ten years, an increasing number of companies have invested significantly in the research and development of new chemical and biological nematicides, which has led to a renaissance in nematicide research. At the same time there seems to have been a clear growth in awareness globally of the need for improved nematode control and management to mitigate yield losses in modern-day crop rotations. This paper will describe a range of studies conducted to characterize the potency, soil behavior, and selectivity of the new, novel, sulfonamide nematicide Salibro™, which contains the active ingredient fluazaindolizine. Interpretation will be provided as to how these characteristics have translated into effective root protection of various crops under field conditions in Latin America and around the world. Plant-parasitic nematodes exposed to Salibro™ show strongly reduced activity and mobility, coupled with a loss of plant infectivity. Across a range of key species of plant-parasitic nematodes (e.g. root-knot, dagger, and lesion) a recognizable sequence of symptomology is consistently observed. These effects appear irreversible and are only marginally affected by temperature. Additionally, Salibro™ has demonstrated a remarkable selectivity profile, not only to a wide range of non-target organisms, but also to other trophic guilds of nematodes, as well as compatibility with microbial organisms that have a role in reducing soilborne plant diseases and supporting plant growth. Investigations into soil behavior showed Salibro™ can be well distributed in soil, showing sufficient mobility and residuality to impact sensitive plant-
parasitic nematode life stages in crop root zones. These biological and physico-chemical features correlated well with observations on root protection in field studies in Latin America and globally in a range of important crops. The balanced properties of Salibro™, in terms of activity on key plant parasitic nematodes, soil behaviour, and favourable mammalian and ecotoxicological safety profile, will make Salibro™ a useful future tool for inclusion in nematode management programs.

MUTUALISM BETWEEN ENTOMOPATHOGENIC NEMATODES AND A SAPROPHYTIC FUNGUS
[MUTUALISMO ENTRE NEMATODOS ENTOMOPATOGÉNICOS Y UN HONGO SAPROFÍTICO]

Wu, S-Y.1,2, F. E. El-Borai1,2, J. H. Graham1, and L. W. Duncan1. 1University of Florida, Citrus Research and Education Center, 700 Experiment Station Rd., Lake Alfred, FL 33850, USA. 2Plant Protection Department, Faculty of Agriculture, EL-Zagazig University, Egypt. sywu@ufl.edu.

In field surveys, the saprophytic fungus, Fusarium solani, was isolated from high proportions of Galleria mellonella sentinel larval cadavers that did not have evidence of entomopathogenic nematode (EPN) reproduction. Because F. solani is not entomopathogenic, we tested the hypothesis that the fungus facilitates infection and death of soil arthropods by attracting EPNs and thereafter competes with the nematode in the insect cadaver. In two-choice olfactometer assays, infective juvenile (IJ) Steinernema diaprepesi migrated in greater numbers toward the arm containing agar plugs with F. solani mycelia and conidia compared to the side with only agar plugs. However, this tendency attenuated in proportion to the complexity (addition of insects, use of raw rather than sterile soil, etc.) of habitat. The attraction may be caused by 1-octen-3-ol that was abundant in F. solani mycelia, using headspace-solid phase micro-extraction (HS-SPME) and gas chromatography mass spectrometry (GC/MS). Fungivorous insects are reported to be attracted by 1-octen-3-ol. The IJ EPNs migrated preferentially toward 1-octen-3-ol in two-choice assays, regardless of whether insects were present or absent from both arms. When conidia of F. solani and IJ S. diaprepesi, alone or in combination, were added to soil microcosms containing larvae of the weevil Diaprepes abbreviatus, significantly more weevils were killed (83%) in the concomitant species treatment compared to treatments with only the EPN (58%) or the fungus (0%). The abundance of both organisms increased in concomitant as opposed to single species treatments. In a 12-month citrus orchard survey using qPCR to measure populations, there was a highly significant spatial association between EPNs and F. solani as measured by Spatial Analysis by Distance Indices (SADIE). Consilience of results from natural surveys and laboratory experiments supports the plausibility of a mutualistic relationship between F. solani and S. diaprepesi that enhances the ability of each organism to exploit arthropod resources.

THE NORTH AND SOUTH AMERICAN GLOBODERA ELLINGTONAE CONNECTION [LA CONEXIÓN DE ELLINGTONAE DE GLOBODERA DEL NORTE Y SUDAMÉRICA]

Zasada, I. A.1, H. Pacheco2, O. Acevedo2, L. M. Dandurand3, C. Hesse1, E. Grenier4, and I. Moreno2. 1Agriculture Research Service, USDA, Corvallis, Oregon 97330 USA. 2Servicio Agricola y Ganadero, Santiago, Chile. 3University of Idaho, Moscow, Idaho 83843, USA. 4INRA, UMR1349 IGEPP, Institute of Genetic Environment and Plant Protection, F35653 Le Rheu, France. Inga.zasada@ars.usda.gov

Globodera ellingtonae was described as a new species in 2012 from nematodes collected in Oregon and Idaho, USA. As part of this description, a comparison of ITS rDNA sequences in Genbank revealed that the populations in the USA were molecularly similar to a population collected from the Antofagasta Region of Chile in 2006. Subsequent analysis of a population from Argentina also demonstrated the presence of G. ellingtonae in South America. To further define the global distribution of G. ellingtonae and to collect populations for molecular and biological comparison, four populations of G. ellingtonae were collected from the Antofagasta Region, Chile in 2017; additionally, the original G. ellingtonae population collected in 2006 and maintained in culture was included in the analysis.
For each population, eggs were freed from cysts and 10 to 15 juveniles as well as the cyst fenestra were collected for morphological characterization. DNA was extracted from the remaining eggs and genomic DNA sequenced on an Illumina Hi-Seq platform. De novo assembly of genomes from each population were mined to retrieve commonly used phylogenetic markers for comparison to known *G. ellingtonae* and other *Globodera* species. Additionally, single-copy orthologous genes were used to generate a more robust phylogenomic tree from all available *Globodera* genomes. Results from this analysis, as well as an update on the current state of knowledge regarding the biology and pathogenicity of *G. ellingtonae* will be discussed.