

MOLECULAR CHARACTERIZATION OF HETEROGENEOUS *PASTEURIA* SPP. SPORES WITHIN A SINGLE *HETERODERA GLYCINES* CYST. **Dyrdahl-Young, R.<sup>1</sup>, R.M. Giblin-Davis<sup>2</sup>, W.L. Nicholson<sup>3</sup>, L.W. Duncan<sup>4</sup>, S. Joseph<sup>1</sup>, and T.M. Mengistu<sup>1</sup>.** <sup>1</sup>Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611, <sup>2</sup> Entomology and Nematology Department, University of Florida, Fort Lauderdale Research and Education Center, Davie, FL 33314, <sup>3</sup>Microbiology and Cell Science, University of Florida, Kennedy Space Center, Titusville, FL 32899, <sup>4</sup>Department of Entomology and Nematology, Citrus Research and Education Center, University of Florida, Lake Alfred, FL 33850.

*Pasteuria* is a genus of obligate endoparasite, spore-forming bacteria with specific isolates antagonistic towards different genera of plant-parasitic nematodes. *Pasteuria nishizawae*, which parasitizes *Heterodera glycines* was first reported in North America in 1994. However, poor understanding of basic biology about lifecycle, ideal soil conditions for sporulation, and host preferences of the bacterium make it difficult to culture sufficient endospores for research purposes. Establishment of homogenous spore populations is an essential first step to efforts to optimize conditions for endospore augmentation. Previous reports found heterogeneity in *P. penetrans* genotypes within a single nematode. In order to elucidate the potential diversity of spore lines, an 800 bp fragment of the 16S rRNA gene with a 200 bp highly variable region was amplified from endospores. This region is reportedly able to segregate *Pasteuria penetrans* on an intrapopulation level. Spores from ten different *H. glycines* cysts were isolated and the genomic DNA was extracted from each sample. The variable region of 16srRNA was amplified and cloned from each sample. Ten clones from each of the ten samples were sequenced. The presence of multiple genotypes from within a single nematode will be discussed.

PROJECT NEMATODA: A COLLECTION OF ORIGINAL SPECIES DESCRIPTION OF EVERY NEMATODE. **Eisenback, J.D.** Department of Plant Pathology, Physiology and Weed Science, Virginia Tech, Blacksburg, VA 24060.

A recent study concerning specimens in natural history museums reported that half are wrongly labeled, but who is to blame and what are the solutions? Perhaps the paucity of taxonomists and the lack of man-hours devoted to identifying organisms versus the overwhelming numbers of specimens that need to be described and labeled are the reason for this quandry. In the last 250 years more than 25,000 species of nematodes have been described, an average of 100 descriptions per year. Furthermore, descriptions are published in many different journals in numerous languages from all around the world. Each researcher has to collect these species descriptions even though not all libraries subscribe to every journal and some are not open access, yet all of them are absolutely necessary to identify nematodes to species or to compile monographs or other taxonomic aids. Theoretically, type specimens are the backbone of nematode taxonomy and descriptions are linked to these types. Ideally, type specimens should be consulted whenever an unknown is trying to be identified, but permanent type specimens are not very “permanent” and are generally too valuable to be lent to most requesters. Travelling to the museums with types is an option, but can be expensive, time consuming, and may not be extremely productive. Often, the drawings, photographs, and textural description substitute for the “type”. One possible solution to this dilemma is to make resources that are necessary for taxonomy easier to access and more widely available. Project Nematoda is an attempt to collect the physical description of every species of nematode that has ever been described. The Project uses the name in the original publication and arranges them alphabetically by the genus name. Currently more than 15,000 species descriptions have been collected and easy access to these resources will be available on the internet to everyone with a computer and a connection.

EVALUACIÓN A CAMPO DEL USO DE NEMATODOS ENTOMOPATÓGENOS EN EL CONTROL DE *LOBIOPA INSULARIS* (COLEOPTERA: NITIDULIDAE) EN CULTIVOS DE FRUTILLA DE LA REGIÓN HORTÍCOLA PLATENSE, BUENOS AIRES, ARGENTINA. **Eliceche, D.P., A. Salas, J.M. Rusconi, and M.F. Achinelly.** CEPAVE, Facultad de Ciencias Naturales y Museo, (UNLP), CONICET. Argentina.

*Lobiopa insularis* es una de las principales plagas en cultivos de frutilla en la región hortícola platense, causando daños directos en los frutos debido a la alimentación de larvas y adultos, e indirectos por la colonización de microorganismos que aprovechan dicho daño, reduciendo el uso para consumo y/o comercialización. El sitio de estudio, Colonia Urquiza (La Plata, Buenos Aires), comprende una zona hortícola con un manejo orgánico de los cultivos. *Lobiopa insularis* representa un problema para los horticultores de la región debido a la falta de enemigos naturales. El control se ejerce principalmente mediante el manejo cultural, a través de la cosecha de los frutos antes de su maduración, evitando así atraer al coleóptero. Para su comercialización, el fruto debe estar en un estado de madurez apropiado según el color, contenido de azúcares y consistencia. Teniendo en cuenta el índice de madurez establecido por el SENASA (75% del fruto color rojo), y que la frutilla es una fruta no climatérica, la cosecha temprana contribuye al manejo del coleóptero, aunque puede disminuir la calidad del fruto comercializable. La aparición del coleóptero se da cuando las frutillas se encuentran maduras y/o fermentadas, por tanto la cosecha tardía representaría un problema para el horticultor. Como alternativa al manejo cultural se propuso evaluar a campo la potencialidad del nematodo entomopatógeno *Heterorhabditis bacteriophora* aislado en el sitio de estudio, como agente de control de *L. insularis*. La susceptibilidad del coleóptero al nematodo ha sido previamente evaluada en condiciones de laboratorio, arrojando resultados satisfactorios. Se realizaron liberaciones aumentativas de juveniles infectivos (JI) del nematodo, por medio de aplicaciones de suspensiones acuosas cada 5 pasos, la dosis utilizada fue de 10.000 JI en 3 ml de

agua. La unidad de estudio fue una parcela conformada por 7 camellones con dos hileras de plantas cada uno. El cultivo de frutilla se encontraba alternado a intervalos irregulares con plantas de tomate, berenjena, cebolla de verdeo y malezas, irrigado por sistema de riego y manejo agroecológico del mismo. Se utilizaron dos camellones como puntos de aplicación y dos como control. El monitoreo de la plaga se realizó cada 15 días, a partir de la estimación del daño en el fruto, cuyos porcentajes variaron entre 0-10%. Una vez abandonada la cosecha, se realizaron nuevos monitoreos en los cuales se registró un daño promedio en los controles del 46%, mientras que en los camellones aplicados fue del 23%. A partir de estos datos preliminares podemos inferir que las diferencias entre los tratamientos son significativas, por tanto el uso del nematodo como agente de control de *L. insularis* podría representar una estrategia de control alternativa al manejo cultural, garantizando la producción de frutos con las características demandadas por el mercado.

GENOME SCANS ON EXPERIMENTALLY EVOLVED *GLOBODERA PALLIDA* POPULATIONS TO IDENTIFY MOLECULAR BASIS OF ADAPTATION TO *GPAV<sub>VRN</sub>* POTATO RESISTANCE. **Eoche-Bosy, D.<sup>1</sup>, M. Esquibet<sup>1</sup>, S. Fournet<sup>1</sup>, M. Gautier<sup>2,3</sup>, F. Legeai<sup>1,4</sup>, A. Bretaudeau<sup>1,4</sup>, E. Grenier<sup>1</sup>, and J. Montarry<sup>1</sup>.** <sup>1</sup>UMR IGEPP, INRA, 35653 Le Rheu, France, <sup>2</sup>UMR CBGP, INRA, IRD, Cirad, Montpellier SupAgro, 34988 Montpellier-sur-Lez, France, <sup>3</sup>Institut de Biologie Computationnelle, 34095 Montpellier, France, <sup>4</sup>IRISA, INRIA, 35042 Rennes, France.

In the current agronomical context of reduced use of pesticides, deciphering the genetic bases of pathogen adaptation to plant resistances is of major importance as it could help us to improve durability of these resistances. *Globodera pallida* is a major pest of potato and for which the promising resistance factor, QTL *GpaV<sub>VRN</sub>*, has been identified in *Solanum vernei* and introduced into several new resistant cultivars in Europe. However, a previous study having employed an experimental evolution protocol, in which *G. pallida* lineages evolved on resistant or sensible potato genotypes, showed that *G. pallida* was able to overcome the resistance from *S. vernei*. The aim of the present study was to investigate the genomic regions involved in the resistance breakdown using a genome scan approach on the lineages resulting from the experimental evolution. A first low throughput genome scan was performed using 202 microsatellite markers distributed along the genome, using three different tests of neutrality based on genetic differentiation and heterozygosity. We identified eight outlier loci, several of which were found by multiple outlier detection methods and/or in two independent adapted lineages, indicative of genomic regions putatively involved in the resistance breakdown where several effectors of interest were identified. We also showed that the same adaptive genetic pathways seem to be involved in overcoming potato genotypes that harbored the same resistant QTL but differed in their genetic background. These results have validated the feasibility of a genome scan approach on biological material coming from a short term experimental evolution and led us to target more precisely the genomic regions involved in the adaptation. Thus a second high-throughput genome scan was performed using NGS data obtained through a whole genome resequencing of pools of individuals (Pool-Seq) that belonged to some of the lineages coming from the experimental evolution. 1.6 million SNPs are currently being used for a genome scan performed with the BAYPASS program. Identified outlier loci will allow us to target more precisely the best candidate genes involved in the adaptation and will also provide a molecular tool to follow virulence allele frequencies within wild *G. pallida* populations, which would be useful in order to conceive efficient strategies for maximizing the durability of potato resistance.

THE EMERGENCE OF *MELOIDOGYNE HAPLANARIA* IN FLORIDA, AND THE EFFECT OF INITIAL DENSITIES POPULATIONS ON TOMATO. **Espinoza-Lozano, L.<sup>1,3</sup>, S. Joseph<sup>1</sup>, W. Crow<sup>1</sup>, L. Duncan<sup>2</sup>, J. Noling<sup>2</sup>, and T. Mekete<sup>1</sup>.** <sup>1</sup>Department of Entomology and Nematology, University of Florida, Gainesville, FL, 32608, <sup>2</sup>Department of Entomology and Nematology, University of Florida, Lake Alfred, FL 33850, <sup>3</sup>Centro de Investigaciones Biotecnológicas del Ecuador, Escuela Superior Politécnica del Litoral, Guayaquil, Ecuador.

Root-knot nematodes are globally considered one of the most devastating plant-parasitic nematodes and are responsible for significant economic losses on a multitude of crops including tomato. The use of resistant varieties is a key tactic in the arsenal of management tools to control root-knot nematodes. In tomato, a single dominant gene referred to as the *Mi* gene has been widely used in plant breeding efforts and varietal development, which confers resistance to a number of the most economically importance species of root-knot nematode found in Florida, including *Meloidogyne incognita*, *arenaria*, and *jananica*. *Mi*-virulent, resistance breaking isolates of these species, capable of reproduction and causing plant damage, have been detected in many areas of the world following the repeated use of these cultivars in field production. All too frequently, the discovery is also made that field populations of many different species of root-knot nematode are present in production fields, and those species that overcome the *Mi* resistance gene can proliferate and cause damage. *M. haplanaria* is an example of a root-knot nematode species recently reported in Florida affecting tomato crops that carry the *Mi* gene; this species was originally reported affecting peanut crops in Texas and Arkansas. Little is known about this nematode and its potential effect on tomato cultivars, including those conferred with the *Mi* gene. The main goal of the studies reported herein was to quantitatively describe the relationship between eight initial population densities of *M. haplanaria* on the resistant tomato cultivar “Sanibel” and the non-resistant cultivar “Rutgers”. Results from greenhouse and growth chamber studies will also be discussed.