measured in dose-response assays, was compared against the pea aphid, *Acyrthosiphon pisum*, that had been reared on different host plant species. *A. pisum* were reared on dwarf bean then inoculated with *P. neoaphidis* and returned to dwarf bean or inoculated and transferred to field bean, pea or lucerne. The smallest estimated median lethal concentration (LC_{50}) was 7.7 conidia mm^{-2} for aphids returned to dwarf bean, with LC_{50}s of 13.0 and 14.6 conidia mm^{-2} for aphids transferred to field bean or pea, respectively. The largest LC_{50} was achieved when aphids were transferred to lucerne: 2941.0 conidia mm^{-2}. In a subsequent experiment, *A. pisum* were reared on either pea or dwarf bean for four generations before bioassays. The LC_{50} for aphids reared and incubated on dwarf bean was 7.3 conidia mm^{-2}, compared to 13.3 and 15.3 conidia mm^{-2} when aphids were transferred between dwarf bean and pea, or pea and dwarf bean, respectively. The LC_{50} for aphids reared then incubated on pea plants was 27.9 conidia mm^{-2}. Overall, the virulence of *P. neoaphidis* was greatest when *A. pisum* was reared and maintained on dwarf bean, the plant used for long-term routine culturing of the aphid. In conclusion, virulence of *P. neoaphidis* was influenced by host plant and particularly by the plant species to which the host aphid had become adapted. Plant resources may affect the population dynamics of *P. neoaphidis* and could result in a greater impact on aphid herbivores that are not suffering physiological stress related to a change in host plant.

CONTRIBUTED PAPERS Tuesday, 8:00-10:00

NEMATODES 2

Contributed paper. Tuesday, 8:00. **64**

Entomopathogenic nematode behavioral responses to chemical cues from cadavers.

Paige Redifer, Brittany Gale, Allison McLain, Glen Stevens, Laura Grochowski
School of Natural Sciences and Mathematics, Ferrum College, Ferrum, VA, USA
Address for Correspondence: gstevens3@ferrum.edu

Entomopathogenic nematodes (EPN) are exposed to a range of cues in the soil. To the extent these cues are positively associated with the presence of insect hosts, one might hypothesize that EPN would respond positively to such cues. Decomposing animals release many different chemical compounds into the soil, attract large numbers of foraging insects, and produce large numbers of insect larvae. Thus, these chemical compounds may serve as an important cue for foraging EPN. We hypothesized the *Steinernema feltiae* and *Steinernema glaseri* IJs would respond generally positively to two particular compounds (putrescine and cadaverine) produced during animal cadaver decomposition. We further hypothesized that *S. feltiae* would respond more strongly to putrescine, and that *S. glaseri* would respond more strongly to cadaverine. We initially used standard agar-based "bulls-eye" attraction assays, and assessed *S. feltiae* and *S. glaseri* responses to diffusion discs soaked in 5 µl of 50, 100, 500, and 1000 µmol concentrations of each of the two compounds. We followed those agar trials with more realistic small sand column assays, assessing responses to the compounds when they were presented with additional stimuli such as host presence. On agar, responses differed between the different EPN species, chemical compounds, and concentrations, but the chemicals were never attractive and often strongly repellent. Responses were more complex in the sand columns; in particular, the compounds seem to attract more IJs to areas that also contained hosts.

Contributed paper. Tuesday, 8:15. **65**

The *Wolbachia* Endosymbiont as a Nematode Drug Target for Control of Human Filariasis, a Neglected Tropical Disease and Other insect Borne Pathogens

Barton E. Slatko
Molecular Parasitology Group. Genome Biology Division, N
EnglandBiolabs, Inc., Ipswich MA USA
Address for Correspondence: slatko@neb.com

Most human filarial nematode parasites and arthropods are hosts for a bacterial endosymbiont, *Wolbachia*. In filaria, *Wolbachia* are required for normal development, fertility and survival, whereas in arthropods, they are largely parasitic and can influence development and reproduction, but are generally not required for host survival. Due to their obligate nature in filarial parasites, *Wolbachia* have been a target for drug discovery initiatives using several approaches including diversity and focused library screening and genomic sequence analysis. *In vitro* and *in vivo* anti-*Wolbachia* antibiotic treatments have been shown to have adulticidal activity, a long sought goal of filarial parasite drug discovery. In mosquitoes, it has been shown that the presence of *Wolbachia* can inhibit the replication of certain viruses, such as Dengue, Chikungunya, Yellow Fever West Nile, and the infectivity of the malaria-causing protozoan, *Plasmodium* and filarial nematodes. Furthermore, *Wolbachia* can cause a form of conditional sterility that can be used to suppress populations of mosquitoes and additional medically important insects. Thus *Wolbachia*, a pandemic endosymbiont offers great potential for elimination of a wide-variety of devastating human diseases.

Contributed paper. Tuesday, 8:30. **66**

Differential PirAB expression of the entomopathogenic bacterium *Photorhabdus luminescens* (Enterobacteriaceae) based on tissue association and portal of entry to the insect host

Anaïs Castagnola[1,2]; Nathaniel Davis[3]; Belen Molina[4]; S. Patricia Stock[1]; John G. McMullen II[1]
[1]Department of Entomology, University of Arizona; [2]Center for Insect Science, University of Arizona; [3]Pima Community College; [4]Department of Ecology and Evolutionary Biology, University of Arizona
Address for Correspondence: anais@email.arizona.edu

Photorhabdus bacteria gain access to an insect host by their association with the free-living infective juvenile stage (IJ) of *Heterorhabditis* nematodes. Penetration of the insect can be achieved through three different portals of entry: a) digestive (mouth, anus), b) tracheal (spiracles) and c) integument. Studies have shown that *Photorhabdus* may colonize other tissues before they establish in the insect's hemocoel, the final destination for full release of bacterial symbionts and completion of their life cycle. It is likely that *Photorhabdus* employs effectors related to virulence factors in pathogens for adhesion, invasion, and intracellular growth in its host's cells. In this study we investigated tissue aggregations and virulence factors by measuring *Pir*AB toxin expression of *Photorhabdus luminescens* (TT01) in different insect tissues and concurrent to different portals of entry used by their nematode hosts.

Contributed paper. Tuesday, 8:45. **67 STU**

Candidate Virulence Loci in Pan-Genome of the Entomopathogenic Bacterium, *Xenorhabdus bovienii* (Gamma-Proteobacteria: Enterobacteriaceae)

John G McMullen II[1]; Gaelle Bisch[2]; Jean-Claude Ogier[2]; Sylvie Pagés[2]; Sophie Gaudriault[2]; S. Patricia Stock[3]
[1]University of Arizona, School of Animal and Comparative

Biomedical Sciences, 1117 E. Lowell St., Tucson, AZ; [2]Université Montpellier II/INRA, UMR 1333 Laboratoire DGIMI, Montpellier, France; [3]University of Arizona, Department of Entomology, 1140 E. South Campus Dr., Tucson, AZ
Address for correspondence: jgm2@email.arizona.edu

Xenorhabdus spp. has dual life styles: they are pathogenic to insects and mutualistic with *Steinernema* nematodes. The nematodes vector the bacteria from one insect to another. In return, bacteria provide a suitable environment in the insect cadaver for the nematodes to maturate and reproduce. Each *Steinernema* spp. carries one *Xenorhabdus* sp. Contrarily, a *Xenorhabdus* spp. may associate with more than one nematode host. The most promiscuous bacterium is *X. bovienii*, which associates with nine *Steinernema* spp. In this study, we performed a comparative genomic analysis of nine *X. bovienii* strains to depict novel virulence factors. Furthermore, virulence assays were performed considering three different lepidopteran hosts. Results revealed that four *X. bovienii* strains were attenuated, whereas the other five were virulent. The genomic platform MicroScope was used to identify known and candidate genes that contribute to their pathogenicity. Additionally, loci involved in their association with the nematodes were investigated. Two loci were identified as novel candidates involved in the bacterium's ability to interact with both nematode and insect hosts. The first region appears to be specific to interactions with nematode partners. The second region contains a type six secretion system (T6SS), which is known to contribute to bacterial pathogenicity. We hypothesize T6SS may contribute to the bacterium's ability to cause death in a wide range of insect hosts. Further molecular studies are undergoing to expand our understanding on the role of these loci and their mode of action in the dual lifestyle of this bacterium.

Contributed paper. Tuesday, 9:00. **69**

Molecular mechanism of the nematicidal activity of *Photorhabdus luminescens* LN2 against *Heterorhabditis bacteriophora* H06 nematodes

Xuehong Qiu and Richou Han
Guangdong Entomological Institute, 105 Xingang Road West, Guangzhou 510260, China
Address for Correspondence: hanrc@gdei.gd.cn

Photorhabdus luminescens subsp. akhurstii LN2 (Enterobacteriaceae) is a symbiont of entomopathogenic nematodes *Heterorhabditis indica* LN2 and showed nematicidal activity against *H. bacteriophora* H06 infective juveniles (IJs). The LN2 bacteria may secrete unidentified toxic factors lethal for the H06 nematodes. The trans-specific nematicidal activity of the bacteria against the non-symbiotic nematode may have an impact on competitive interactions when one insect host is co-infected by different nematode species. To explore the molecular mechanism of the trans-specific nematicidal activity of *P. luminescens* LN2 against *H. bacteriophora* H06, the complete genome of *P. luminescens* LN2 was sequenced; two mutagenesis libraries of *P. luminescens* LN2 were constructed using Tn5 transposon and rifampicin antibiotic respectively; the mutants from the libraries were tested for nematicidal activity and mutants negative for nematicidal activity were genetically and proteomically characterized. At least 9 putative proteins including DsbA, HlpA, RhlE, RplC, RpoB, NamA, NamB (a protein from T3SS), and 2 hypothetical proteins (similar to unknown protein YgdH and YggE of *Escherichia coli* respectively) were involved in the nematicidal activity of LN2 bacteria against H06 nematodes. This hypothesis was further confirmed by creating insertion-deletion mutants of corresponding genes. It seems that a big network system is involved in this nematicidal activity.

Contributed paper. Tuesday, 9:15. **70**

Natural products from entomopathogenic bacteria: Understanding the interaction of bacteria, insects and nematodes

Helge B. Bode
Merck Stiftungsprofessur für Molekulare Biotechnologie, Fachbereich Biowissenschaften, Goethe Universität Frankfurt, Germany
Address for Correspondence: h.bode@bio.uni-frankfurt.de

Entomopathogenic bacteria of the genera *Xenorhabdus* and *Photorhabdus* live in symbiosis with nematodes of the genera *Steinernema* and *Heterorhabditis*, respectively, and together they are able to infect and kill several different insect larvae. We have shown recently by chemical analysis and genome sequencing that these bacteria are able to produce a huge variety of different low molecular weight natural products. These compounds show insecticidal but also antibiotic and anticancer activity and novel bacterial signalling compounds have also been identified.
Recent work indicates that several of the bacterial natural products are addressing different parts of the insect immune system in order to make sure that the bacteria can evade it and kill the insect host. As the nematode immune system shows the same basic principles, it is of high interest how the natural products can differentiate between insect prey and nematode host. We will present our recent finding on natural products and their natural targets as well as ways to improve the production of these – probably also pharmaceutically useful – compounds.

CONTRIBUTED PAPERS Tuesday, 8:00-10:00

VIRUSES 3

Contributed paper. Tuesday, 8:00. **71**

Characterization and formulation of a Colombian isolate of *Erinnyis ello* granulovirus (L.) (Lepidoptera: Sphingidae)

Juliana Gómez[1], Gloria Barrera[1], Paola Cuartas[1], Carolina Ruiz[1], Adriana Santos[1], Liz Uribe[1], Guillermo León[2], Laura Villamizar[1]
[1]Centro de Biotecnología y Bioindustria (CBB), Corpoica, Bogotá, Colombia, . [2] Centro de Investigación "La Libertad" Corpoica, Puerto López, Colombia
Address for Correspondence: gbarrera@corpoica.org.co

Erinnyis ello (L.) is a polyphagous lepidopteran pest that may cause serious annual losses in the rubber industry. The use of granulovirus represents an interesting alternative as a biological control agent for this insect. One Colombian granulovirus isolate (VG010) was obtained from *E. ello* larvae in the field and was characterized at morphological, biological and molecular level. Occlusion bodies showed an oval morphology with a unique nucleocapsid, and a size of 302.9 ± 22 x 181.5 ± 16 nm. The VG010 viral genome size was estimated to be approximately 88.7 kb. Phylogenetic relationships based on selected gene sequences *lef-8, lef-9* and *gran* showed a close relationship between VG010 and another isolate from *E. ello* previously reported (M34-4), suggesting that these isolates are genotypic variants of the same viral specie. The mean lethal dose of VG010 against second instar *E. ello* larvae was 4.3×10^3 OBs/mL and the viral productivity ranged between 2.1×10^9 and 3.8×10^9 OBs/g of larval tissue. With this virus, a wettable powder formulation was