

Entomology

FRIENDS WITH BENEFITS; NATURAL ENEMIES AND POLLINATORS IN VIRGINIA CUCURBIT PRODUCTION. James M. Wilson & Thomas P. Kuhar, Department of Entomology, Virginia Tech, Blacksburg, Virginia 24061. Cucurbit production in Virginia provides the commonwealth with melons, pumpkins, summer squashes, and cucumbers, all of which are dependent upon adequate pollination for fruit set and development. Growers liberally apply fungicides in Virginia, and often will tank-mix broad-spectrum insecticides that are known to have negative effects on both natural enemy populations and pollinators regardless of pest pressure. Here we examine the potential role of narrow-spectrum insecticides and their interactions with the squash bug egg parasitoid, *Gryon pennsylvanicum* (Hymenoptera: Scelionidae). *G. pennsylvanicum* has been identified around the commonwealth and is capable of high levels of parasitization on eggs of the squash bug, *Anasa tristis* (Hemiptera: Coreidae) a pest of many cucurbit crops. Bioassays were conducted utilizing field collected squash bug egg masses with four narrow-spectrum insecticides and a broad-spectrum insecticide (used as a baseline). Squash bug egg masses were dipped in formulations of label-rate insecticides, and squash bug nymphs and parasitoid wasp adults were allowed to hatch. In 2015, there were no significant differences between treatments after a χ^2 goodness of fit test, in which the control replicate data were used as the “expected” values of background parasitization levels. A shift away from broad-spectrum insecticide use may reduce the risk of releasing secondary pest populations. Adapting effective narrow-spectrum insecticide use may help to mitigate the negative impacts of insecticides on pollinators and the natural enemy complex in cucurbit crop systems.

FLEA BEETLE SPECIES AND THEIR IMPACT ON CABBAGE AND EGGPLANT IN VIRGINIA. James A. C. Mason & Thomas P. Kuhar, Department of Entomology, Virginia Tech, Blacksburg VA 24061. Flea beetles (Coleoptera: Chrysomelidae), *Epitrix* spp. and *Phyllotreta* spp., are important economic pests of vegetable crops in North America, particularly eggplant and cabbage. An experiment was conducted in Whitethorne, Virginia on both “classic” eggplant and “Bravo” cabbage in 2015 to determine the impact of flea beetle feeding injury on crop yield. Beetle density and defoliation were assessed weekly for about one month by counting the number of beetles present on 10 plants per plot, and using a percent defoliation scale counting 10 randomly selected leaves per plot: 1= no defoliation; 2= 1-20% defoliation; 3= 21-40% defoliation; 4= 41-60% defoliation; 5= >60% defoliation. After one month, all plots were treated with a broad spectrum insecticide combination to eliminate any further insect injury.

Crop yield was assessed on eggplant by counting the number of eggplant harvested per plot, and cabbage by taking the total weight of all 16 cabbage heads per plot. Individual plants were monitored using the same defoliation scale by assigning an overall defoliation rating 1-5 to the entire plant, 10 plants were used for each category of defoliation. Yield of these individual plants was also assessed by counting the number of eggplant per plant, and the individual weights of the cabbage heads. The predominant flea beetle species found on eggplant was *Epitrix fuscula* and on cabbage were *Phyllotreta striolata* and *Phyllotreta cruciferae*. There was a significant negative relationship between defoliation rating and yield in both crops on a per plot basis and on an individual plant basis.

ATTRACT-AND-KILL TECHNOLOGY FOR HARLEQUIN BUG, *MURGANTIA HISTRIONICA* (HAHN), IS OPTIMIZED WITH NEW INSIGHTS ON BEHAVIORAL RESPONSE TO COLOR AND SEMIOCHEMICALS A. S. DiMeglio¹, T. P. Kuhar¹ & D. C. Weber², ¹Department of Entomology, Virginia Tech, Blacksburg, VA 24061 and ²USDA - ARS, Beltsville, MD, 20705. To develop an effective attract-and-kill trap for harlequin bug (HB), *Murgantia histrionica* Hahn, an economically important pest of brassicaceous crops in the United States, we conducted a series field and lab studies. In a lab color-choice study, we found dark green to be most attractive for nymphs and adults, and yellow and white the least attractive. We compared green and yellow panel traps in the field, both baited with the aggregation pheromone of HB, murgantiol, and showed that green panels caught significantly more HB and fewer non-target lady beetles than did the baited yellow panels. Investigating the effect of trap type with the addition of semiochemical attractants in the field, we found that a pyrethroid-treated ramp, square panel, and pyramidal trap all intercepted HB effectively in the field, with the pyramidal trap killing the most HB. Also, the combination of murgantiol with a low emission of benzyl isothiocyanate was more attractive than murgantiol alone for all trap types. To integrate this information into an effective attract-and-kill strategy, we explored the potential of using pyrethroid treated screen as a killing agent for the trap. We determined the LT₅₀ of HB nymphs on PermaNet®, a deltamethrin-treated screen, and showed this product to be promising as trap construction material for managing HB.

SESQUITERPENOID PHEROMONE BIOSYNTHETIC PATHWAYS IN STINK BUGS (PENTATOMIDAE). Jason Lancaster¹, Ashot Khimian², Dawn Gundersen-Rindal², Tom Kuhar³ & Dorothea Tholl¹, ¹Department of Biological Sciences, Virginia Tech, Blacksburg, VA 24061, ²USDA-ARS, Beltsville, MD, 20705 and ³Department of Entomology, Virginia Tech, Blacksburg, VA 24061. The stink bugs (Pentatomidae) harlequin bug (*Murgantia histrionica*), brown

marmorated stink bug (*Halyomorpha halys*), and southern green stink bug (*Nezara viridula*) are significant agricultural pests both in the United States and globally. The aggregation pheromones produced by these insects are known and their structures have been determined, however their use in pest management strategies is limited due to laborious synthetic chemistry and lack of knowledge of *de novo* biosynthetic pathways. This project aims to identify and functionally characterize farnesyl diphosphate synthase and sesquiterpene synthase genes in harlequin bug, brown marmorated stink bug and southern green stink bug. We hypothesize that Pentatomidae produce sesquiterpene aggregation pheromones *de novo* via a biosynthetic pathway utilizing a bisabolyl carbocation. We have cloned and expressed a gene from both harlequin bug and southern green stink bug whose encoding amino acid sequences are similar to other insect farnesyl diphosphate synthases but enzymatic assays produce primarily sesquiterpenes. We believe these genes are involved in production of the stink bug's aggregation pheromone and are currently investigating a similar gene in brown marmorated stink bug. This project will advance our understanding of the genetic mechanisms of terpene production in Pentatomidae and provide resources for novel, genetic based solutions.

OLD FOES, NEW LANDS: USING GCMS AND PCR TO EXAMINE PREDATOR-PREY RELATIONSHIPS IN THE INVASIVE POISON HEMLOCK PLANT. C. D. Alencar¹, K. F. Haynes², J. J. Obrycki², K. J. Athey², & J. D. Harwood², ¹Biology Department, University of Virginia, Charlottesville VA, 22904 and ²Entomology Department, University of Kentucky, Lexington KY, 40506. Poison hemlock, *Conium maculatum* (Apiaceae), is a long-established invasive plant of North America with a unique toxic chemistry. Previous research on this plant has focused on identifying potential biological control agents or describing the plant's suite of alkaloids. However, none have examined the role of higher trophic levels in the food web surrounding poison hemlock or the possibility that its unique chemistry imparts cascading effects. Generalist predators and food web interactions are an important component of studies investigating long-term invasion effects, as plant or animal introductions can alter ecosystem functioning. In this study, plant resources and predators foraging in poison hemlock were sampled at the foliar and ground levels. Predator-resource linkages were quantified using molecular gut-content and chemical analyses. Foliar *Harmonia axyridis* (Coccinellidae) contained aphid DNA and plant chemicals, while *Harpalus pensylvanicus* (Carabidae) only contained alkaloids, suggesting that ground predators obtain plant chemicals via alternative food resources. Feeding trials between *H. axyridis* and their potentially toxic prey, *Hyadaphis foeniculi* (Aphididae), showed quickened development in the exotic predator through immature stages when consuming aphids from poison hemlock compared to similar

alternative diets. This study revealed that three Eurasian species may be facilitating one another, illustrating the importance of continued examination of invasive species interactions.

NOVEL MODES OF PATHOGEN TRANSMISSION FROM MEDICALLY IMPORTANT ARTHROPOD VECTORS. Hameeda Sultana, Center for Molecular Medicine, Department of Biological Sciences, Old Dominion University, Norfolk, VA 23529. Molecular determinants and mechanisms of arthropod-borne flavivirus transmission to the vertebrate host are poorly understood. Research in my laboratory has shown that both tick and mosquito-borne flaviviruses use exosomes, the small membranous extracellular vesicles for transmission from arthropods to human host. Our studies have revealed that arthropod derived exosomes are important means of communication and transmission between the vector and the vertebrate host. We have found that Langkat virus (LGTV), a flavivirus member closely related to tick-borne encephalitis virus and mosquito-borne dengue viruses, are transmitted from vector to the vertebrate host through exosomes as novel modes of transmission. The exosomes containing LGTV and dengue viruses were viable, secured and highly virulent in all tests such as re-infection kinetics, trans-migration and viral plaque formation assays, suggesting exosomes as favorable modes of transmission. Both matured virions and replicative forms of arthropod-borne flaviviruses were found to be using exosomes for transmission. Our data also showed that arthropod derived exosomes facilitate infection of human cells that eventually produce exosomes loaded with flaviviruses. These transmission strategies used by flaviviruses to exit arthropods and infect human host were envisioned as best approaches to develop transmission-blocking vaccines against molecules or determinants that facilitate pathogen transmission.

PATHOGEN-VECTOR-HOST INTERACTIONS: NEW LESSONS LEARNED FROM THIS OLD CONNECTION. Girish Neelakanta, Center for Molecular Medicine, Department of Biological Sciences, Old Dominion University, Norfolk, VA 23529. In the United States, *Ixodes scapularis* ticks transmit various pathogens to humans that include *Anaplasma phagocytophilum*, the agent of Human anaplasmosis. Ticks ingest *A. phagocytophilum* while feeding on an infected animal. Upon entering, *A. phagocytophilum* establishes itself in the salivary glands and is then transstadially maintained through different developmental stages of these ticks. Our previous studies have elucidated that *A. phagocytophilum* manipulates arthropod cell signaling to survive in its vector. In the view for the development of an anti-vector vaccine as an effective means to block transmission of *A. phagocytophilum* from these ticks, understanding the role of conserved arthropod molecules in vector biology and interactions with pathogens

remains important. This study provides evidence for the role of some of the conserved arthropod molecules in tick-*A. phagocytophilum* interactions. RNAi knockdown studies and *in vitro* cell culture experiments elucidate the role of important arthropod conserved molecules in *A. phagocytophilum* transmission and survival in these ticks. Collectively, this study defines a new pathway not only to understand vector biology but may also lead to the development of better strategies to block transmission of *A. phagocytophilum* and perhaps other Rickettsial species from these ticks.

IMPACT ASSESSMENT OF *LARICOBIVS NIGRINUS* (COLEOPTERA: DERODONTIDAE), A PREDATOR OF HEMLOCK WOOLLY ADEGLID. A. Heminger¹, A. Mayfield², J. Elkinton³, G. J. Wiggins⁴, J. F. Grant⁴, J. Lombardo⁵, T. McAvoy¹, A. Tait⁶, B. Mudder² & S. Salom¹, ¹Virginia Polytechnic Institute and State University, Blacksburg, VA, 24061 ²USDA - Forest Service, Asheville, NC 28804 ³University of Massachusetts, Amherst, MA 01003 ⁴University of Tennessee, Knoxville, TN 37996, ⁵Dartmouth College, Hanover, NH 03755 ⁶North Carolina State University, Asheville, NC. *Laricobius nigrinus* is a predator of hemlock woolly adelgid (HWA). HWA is a serious pest of eastern and Carolina hemlocks causing hemlock dieback and death. HWA is being controlled through several different methods including through the use of predators such as *L. nigrinus*. In the fall, we set up nine field sites in six states from New Jersey to Georgia. Three treatments were set up at each of the sites, cage (excluded *L. nigrinus*), no cage which allows for predation, and open cage to assess cage microclimate. The first sample was taken in October/November, the second in February/ March, and the third in March/April. The first sample documented HWA/cm, the second HWA mortality and signs of *L. nigrinus*, and the third to assess presence of *L. nigrinus*. The second year of the study was conducted at the same field sites. Results show that larval impacts appear greater than adult only; plots further south showed slightly higher rates of *L. nigrinus* activity, and *L. nigrinus* predation was seen at six out of the nine sites and *L. nigrinus* larvae was recovered from five off the nine in year 1. The study was funded by USDA Forest Service.

STRUCTURE ACCESS AND SETTLING SITE PREFERENCES OF OVERWINTERING BROWN MARMORATED STINK BUGS (*HALYOMORPHA HALYS*). B. D. Chambers^{1,2}, T. P. Kuhar², A. R. Pearce¹, T. C. Leskey³ & G. Reichard¹, ¹Department of Building Construction, Virginia Tech, Blacksburg, VA 24061 ²Department of Entomology, Virginia Tech, Blacksburg, VA 24061 ³USDA-ARS, Kearneysville, WV 25430. The brown marmorated stink bug is an Asian invasive species that is spreading across the United States. In the

years since its introduction, it has become an important agricultural pest. It has also become a major nuisance in houses, where it often spends winters. This research examined some aspects of the entry and settling of these insects as they seek winter harborage in homes. One part tested movement directions of *H. halys* walking on building exteriors in the fall. A tendency towards upward movement was observed. A test of bugs disturbed from diapause in spring also suggested a tendency towards upward movement. To determine the limits on sizes of gaps in walls through which *H. halys* can pass, heat was used to encourage bugs to move through holes of various sizes. Passage was limited by rigid width and vertical compressibility. Males did not pass 3 mm tall or 7 mm wide holes, and females mostly did not pass 4 mm tall or 8 mm wide holes. The effect of the dead bugs often left over from previous years was examined with several tests. Non-tactile tests suggest the possibility that bugs may avoid clusters of dead bugs, but tactile tests suggested a willingness to aggregate with the dead.

INTERACTIONS OF HOST PLANT CHEMISTRY AND LEAF COLOR ON CABBAGE WHITE BUTTERFLY, *PIERIS RAPAE*. Wallis L. Hudson, Claire M. Ingram & Mary E. Lehman, Department of Biological and Environmental Sciences, Longwood University, Farmville VA 23909. *Pieris rapae* is known to detect the chemical content of plants that would be best suited for offspring development. Host plant chemistry and leaf color were manipulated to determine the main and interactive effects on ovipositional choices of *P. rapae*. Cabbage leaf disks were sprayed with deionized water (control) or 1.0 mM *p*-coumaric acid (PCO) or sinapic acid. The effects of these host chemicals and leaf color were first assessed individually and then in an interaction experiment. Females and cabbage leaf disks were placed in mesh enclosures and eggs were counted after a 24- hour period. When tested individually, PCO significantly stimulated oviposition and sinapic acid acted as a significant deterrent. In the interaction experiments, the main effects of both chemicals were not significant, but the main effect of leaf color was highly significant, with more eggs consistently laid on green than on white leaf disks. A significant interaction effect was only seen with sinapic acid. This study provides evidence that leaf color is an important determinant of ovipositional choices, an effect that may be strong enough to counteract or override the possible secondary cues of phenolic acid chemicals in host plants.