

Field 7. PROJECT SUMMARY: Sustainable Management of the Small Hive Beetle (*Aethina tumida*), an Emerging Pest of Honey Bees

Gruner, D.S. (PD), C.R.R. Hooks, and G.P. Dively (University of Maryland)

With this joint **research-extension** project (**\$38,661 PL89-106 + \$17,446 Smith-Lever**), we propose to develop and apply sustainable control practices for the ‘small hive beetle’ (SHB, *Aethina tumida*), which infests honey bee (*Apis mellifera*) colonies and vectors pathogens that may contribute to Colony Collapse Disorder. Honey bees – critical pollinators of a wide variety of fruit, nut and vegetable crops – are in protracted decline nationwide. The invasive SHB is rapidly expanding its range from the Southeast U.S., where it has been exceptionally destructive to colony health since 1998, into the mid-Atlantic and Northeast regions. Existing chemical controls are problematic for honey bee health because of exposure risks and associated costs. This project will develop, evaluate, and disseminate a multi-faceted, sustainable IPM strategy to disrupt the SHB life cycle. In functional hive experiments, we will evaluate two novel tactics – soil drenches of biopesticides and entomopathogenic nematodes to control wandering larvae – deployed in combination with in-hive trapping devices to capture invading adults (**Objective 1**). We will evaluate these IPM strategies through on-site demonstrations with cooperating master beekeepers and disseminate research results via eXtension education to help mid-Atlantic beekeeping associations rapidly implement recommendations (**Objective 2**). This project addresses thirteen NE-RIPM relevance criteria, three major priorities of the Northeast IPM center, and many specific directives from regional beekeeping organizations. Our approach will reduce environmental and human health risks by replacing hazardous pesticides with affordable traps, biorational organic-compatible pesticides, and augmentative biological control, and our demonstration and extension efforts will stimulate widespread adoption of IPM strategies across the region.

8. PROJECT NARRATIVE: Sustainable Management of the Small Hive Beetle (*Aethina tumida*), an Emerging Pest of Honey Bees

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(8.i) Problem, Background and Justification

Problem. Honey bees are essential for pollination of over 90 fruit and vegetable crops worldwide and are valued at \$14.6 billion in the United States (Morse and Calderone 2000) and over \$200 billion worldwide (Gallai et al. 2009). The value of pollination made up approximately 10% of the total value of food consumed globally by humans in 2005 (Gallai et al. 2009). However, pollinators worldwide are in decline (Berenbaum et al. 2007), which is leading to a concomitant decline in their essential pollination services (Biesmeijer et al. 2006, vanEngelsdorp et al. 2008). Specifically, honey bee (*Apis mellifera*) populations are in jeopardy; the dramatic loss of colonies since 2006 and the rise of ‘Colony Collapse Disorder (CCD)’ is the latest demonstration of this. As evidence of their population decline, the National Agriculture Statistics Service reported that there were 2.44 million honey-producing colonies in the United States as of February 2008, down from 4.5 million in 1980, and 5.9 million in 1947 (Berenbaum et al. 2007). Further, a recent survey indicated that between 0.75 and 1.00 million honey bee colonies died in the United States over the winter of 2007–2008 (vanEngelsdorp et al. 2008). Thus, overwintering losses during this brief period were estimated at 35.8%, an increase of 11.4% compared to the previous year. Since the early 1980’s pathogens and pests of American honey bee strains have risen dramatically with introduction of tracheal mites in 1984 (Sammataro et al. 2000), followed by *Varroa destructor* in 1987 (Delfinado-Baker and Houck 1989), and small hive beetle (SHB) *Aethina tumida* Murray (Coleoptera: Nitidulidae) in 1998 (Elzen et al. 1999). Since it was initially documented in the US, the distribution of the SHB continues to expand. Originating from sub-Saharan Africa, SHB has now been detected in >30 U.S. states (Neumann and Elzen 2004).

Although weak colonies are most susceptible to their colonization, under heavy infestations, SHB may force honey bees to abandon their hives and can overwhelm and directly kill even strong colonies (Elzen et al. 1999, Wenning 2001). The developing SHB larvae are the most destructive stage: larvae damage wax combs while feeding on pollen and brood, and ruin honey by defecating within the food cells, causing the honey to ferment and produce a foul odor (Lundie 1940, Hood 2004, Neumann and Elzen 2004). Furthermore, the honey becomes thin and runs out of the combs, rendering it unmarketable. Multiple honey bee viruses have been isolated from SHB specimens (Eyer et al. 2009a, Eyer et al. 2009b), and SHB is a suspected vector involved in the recent epidemic of Colony Collapse Disorder. Thus, SHB can cause significant economic harm to hobby beekeepers, sideliners, or commercial operators through reduced quality of product, direct destruction, or bee abandonment of hives. In 1998, Florida beekeepers alone experienced an estimated \$3 million loss due to SHB (Ellis et al. 2002).

The Mid-Atlantic Apiculture Research and Extension Consortium (MAAREC) of beekeepers from state beekeeper associations, regulators and researchers from Delaware, Maryland, New Jersey, Pennsylvania, Virginia, and West Virginia hold two meetings each year to discuss current and emerging issues of concern to beekeepers. Based on annual surveys by MAAREC during the

last 5 years, colony loss among experienced beekeepers in the Mid-Atlantic region has ranged from 40 to 100%. Regional professional beekeepers, at a recent meeting (MAAREC, 20 October 2008), identified SHB as a rapidly spreading honey bee pest of significant concern and expressed a strong preference for research geared to rapid implementation and extension delivery of improved control tactics for this pest. In a recent Pest Management Strategic Plan for Honey Bees in the Mid-Atlantic states (DE, MD, NC, NJ, PA, SC, VA, WV), as high priorities for SHB these stakeholders ranked integrated pest management and sustainable alternatives to current control tactics that are heavily reliant on pesticides toxic to bees (Gatton et al. 2008).

This project will evaluate the efficacy of two novel tactics – biopesticides and entomopathogenic nematodes (EPN) to control wandering larvae and pupae – deployed in combination with in-hive trapping devices to capture adults entering hives (**Objective 1**). We will disseminate research results through demonstration activities and extension education to help beekeepers and local beekeeping associations rapidly implement an IPM strategy for SHB control (**Objective 2**). Thus, our proposed research and extension efforts directly addresses stakeholder’s listed priority areas and aims to develop management tactics that can be instantly used for a pest that limits their economic growth and production efficiency. The control measures we propose to evaluate were meticulously selected based on potential economic cost to stakeholders, compatibility with current apiculture operations, potential for success, and the biology of the SHB.

Justification. Current control tactics: Current chemical controls for SHB involve two approaches: 1) treatment with CheckMite+ Strips inside the colony for adults; and 2) soil drench treatment of GardStar® (40% EC) for control of developing pre-pupae. In most mid-Atlantic states, CheckMite+Strips can be used under a Section 18 special exemption authorized by EPA. This product is formulated with the a.i. coumaphos (10%), an organophosphate pesticide, impregnated in a plastic strip attached to the corrugated side of cardboard are placed on the bottom board with the strips facing down. Beetles tend to hide on the bottom of the hive and seek shelter in the corrugated cardboard where they are exposed to a lethal dose of coumaphos. Sustained control efficacy has been inconsistent because the CheckMite label does not allow strips to remain in the colonies continuously (Elzen et al. 1999, Hood 2000, Wenning 2001). The GardStar® soil treatment takes advantage of a vulnerable stage of the beetle's life cycle - when mature larvae enter the soil near the hive to pupate. This product, an emulsifiable concentrate containing 40% permethrin, is applied as a soil drench around the hive using a sprinkling can or low-pressure sprayer. Larvae are killed when they come into contact with insecticide-treated soil. These treatments can be effective if applications are correctly timed to the beetle life cycle and properly administered (Pettis and Shimanuki 2000).

However, existing SHB control tactics are problematic for honey bee health, with potential for misuse and continuing indirect and direct costs. Coumaphos, used inside the hive, is lipophilic and moderately toxic to honey bees. This insecticide has been shown to decrease the survivorship of developing queens (Haarmann et al. 2002). Beeswax contaminated with coumaphos is an increasingly recognized problem for beekeeping and for the sale of beeswax. Permethrin (in Gardstar drenches) is highly toxic to bees and even small amounts spilled, sprayed onto a hive, or contaminating the water source can be dangerous to bees. Many beekeepers are reluctant to use chemical controls for SHB because of these risks, and neither product is approved for organic honey production. Also, CheckMite+ Strips may not be available

in the future because the emergency use registration of coumaphos is only temporary and must be renewed or eventually granted a general use registration. Furthermore, extended exposure of these active ingredients to SHB could lead to resistance development, as it has with Apistan treatments for mite control (Lodesani et al. 1995, Milani 1999). Thus, a safer, alternative management approach to SHB control is clearly warranted.

Since the introduction of SHB into the United States, various cultural/mechanical controls have been used by many beekeepers as the first lines of defense against SHB infestations. Several approaches have been tested including the use of modified hive entrances such as polyvinyl chloride (PVC) pipes (Hood 2004). Reducing hive entrances by using PVC pipes resulted in a decrease in the number of small hive beetle in the colonies. However, the efficacy of this technique was inconsistent, and these entrances had collateral effects on brood production, water drainage, and bottom board debris (Ellis et al. 2003a, Hood 2004).

The maintenance of strong, healthy colonies probably remains the best protection because this increases the likelihood that bees can rid themselves of parasitic adults and larvae in the event of an infestation. However, strong colonies can still be overpowered by rapid population growth rates of heavy infestations (Wenning 2001), and recent research established that SHB can vector important honey bee viruses linked to CCD (Cox-Foster et al. 2007, Eyer et al. 2009b, Johnson et al. 2009). Thus, shortcomings of current management tactics and the inability of honey bees to adequately protect colonies from SHB dictates a need to create and disseminate information on effective alternative management practices that will target SHB while reducing or eliminating detrimental impacts on honey bee vigor and survival.

Background. SHB biology: Although SHB originates from tropical regions, adult beetles can readily overwinter in colder climates such as the Northeast by living within the honey bee cluster (Elzen et al. 1999, Hood 2000). When the larvae reach maturity, they enter a wandering phase, leave the hive, and fall to the ground where they burrow into the soil to pupate (Figure 1). Adults emerge from the soil and fly in search of a host honey bee colony. The beetle is attracted to worker honey bee volatiles, composed essentially of alarm pheromones (Torto et al. 2007). Once inside, adults are capable of rapid population growth, with clutches of more than 200 eggs and a lifetime fecundity between 1000-2000 eggs per adult female (Hood 2004, Neumann and Elzen 2004). In the United States, damage to honey bee colonies typically occurs during the summer. Currently, there is no effective tool for monitoring adult beetles in managed honey bee colonies.

When “foreign” organisms (e.g. female SHB) enter a bee hive, guard bees aggressively chase and corral the unfamiliar organisms in an attempt to remove them (Ellis et al. 2003b). In the instance of SHB, their defensive strategy is to escape by entering a crevice too small for bees to follow. Thus, one potential strategy is to set up a “lure and kill” trap within the colony that only SHB can enter while trying to escape guard bees. Thus, one critical point of beetle control is to sever the life cycle as they enter the hive and before adults lay eggs (**Figure 1, A**). Effective isolation traps would reduce or eliminate the problems associated with contaminated honey, reduce viral transmission by adult beetles, and reduce the prodigious egg-laying and reproduction that can lead to massive aggregations, host absconding and colony failure. This is one of two critical points or “weak links” in the SHB life cycle that can be exploited through IPM tactics. The other key target is the wandering larval stage which must enter the soil to

pupate (**Figure 1, B**). Interruption of the beetle life cycle outside the honey bee colony would mitigate or eliminate colony problems related to current chemical control tactics, damage caused by the beetles and energy expenditure by adult honey bees corralling adult beetles or removing dead individuals with insecticide use within the colony.

A number of in-hive trapping devices that have shown great potential for managing SHB have been developed. These beetle traps reduce the number of adult beetles available to lay eggs without the risks of chemical use. One type (Freeman Trap, West Trap) is designed as a screened, mineral oil tray that rests on the bottom board of the colony (Freeman 2009). Another type deploys a similar trapping strategy (Hood Beetle Trap) but is attached to the bottom bar of a standard frame and filled with vinegar or mineral oil to attract and drown adult SHB (Hood and Miller 2003). Other types include the AJ Beetle Eater and Sonny-Mel SHB traps which are placed between or on the top of frames and consist as containers with small holes through which SHB can pass but not bees. Traps take advantage of the bees' aggressive behavior to chase SHB adults and the beetles' defensive response to seek any small space where bees cannot go. SHB enter the bottom screen or through the holes and become coated with mineral oil and are unable to escape. However, these traps have not been comparatively evaluated in the Mid-Atlantic region and thus stakeholders are unsure of their efficacy. Additionally, the effectiveness of these traps may be enhanced if they are integrated with other management strategies (e.g, biological control, biopesticides).

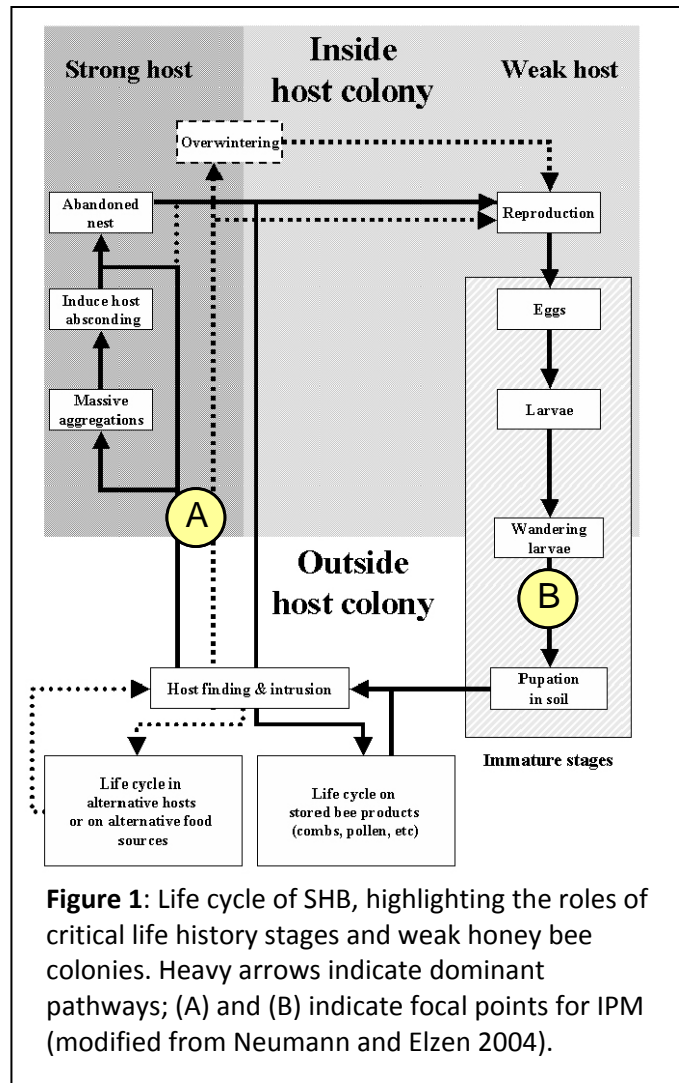


Figure 1: Life cycle of SHB, highlighting the roles of critical life history stages and weak honey bee colonies. Heavy arrows indicate dominant pathways; (A) and (B) indicate focal points for IPM (modified from Neumann and Elzen 2004).

Entomopathogenic nematodes and SHB: Entomopathogenic nematodes (EPN) in the families Heterorhabditidae and Steinernematidae can play a significant role in the biological control of soil borne insects in both natural and managed ecosystems (Kaya and Gaugler 1993, Gaugler et al. 1997, Denno et al. 2008). After locating and penetrating a suitable insect host in soils or roots, the third instar dauer larval stage (infective juveniles, 'IJs') inject symbiotic bacteria that rapidly kill hosts. EPN cycle through several reproductive generations inside a cadaver while feeding on bacterial byproducts of insect tissues, and eventually emerge into the soil environment in large numbers to seek new hosts (Kaya and Gaugler 1993). Under the strategic plan for SHB control in the Mid-Atlantic region, these obligate insect microparasites are a high priority for efficacy

research (Gatton et al. 2008). The role for EPN in integrated pest management programs has been magnified by the rapid evolution of pesticide resistance, regulatory restrictions and environmental cautions on use of chemical pesticides, and the emergence of pests favored indirectly by conservation tillage (Gaugler et al. 1997, Lacey et al. 2001, Georgis et al. 2006).

A single published laboratory study examined EPN potential for control of SHB (Cabanillas and Elzen 2006). Simple laboratory virulence assays indicated that all three EPN species tested (*S. riobravus*, *S. carpocapsae*, and *H. megidis*) infected and killed >50% of wandering larval and pre-pupal stages with fewer than 200 IJs per individual. In preliminary laboratory experiments with two species isolated locally in Maryland (*H. bacteriophora* and *S. carpocapsae*), we demonstrated strong control potential of wandering larvae (see Approach and Procedures). Similar studies on another nitidulid beetle also showed promise (Vega et al. 1994), and limited laboratory work demonstrated low virulence towards adult and larval *Apis mellifera* with direct contact not likely to occur with soil drenches (Lehnert and Cantwell 1978, Kaya et al. 1982, Baur et al. 1995, Zoltowska et al. 2003). However, laboratory environs do not replicate the abiotic variability, soil heterogeneity, and biotic complexity of field conditions. Laboratory experiments test conservatively for the suitability of hosts and demonstrate potential for control, but may overestimate ecological importance. Therefore, replicated field studies on target and nontarget organisms are needed under the conditions typically experienced by honey bee colonies in the Eastern U.S. Moist loamy-sandy soils, as found throughout the SE US and along the Eastern seaboard, favor populations of both EPN and SHB (Grant and Villani 2003, Ellis et al. 2004), a confluence that favors the success of control measures where SHB populations are high.

Native entomopathogenic nematodes are widely distributed naturally – species have been isolated from all continents except Antarctica (Hominick 2002) – and they are also inexpensive and widely available in commercial formulations (e.g., Rincon-Vitova Insectaries Inc.). Multiple species are therefore accessible to amateur and professional beekeepers. Because EPN species vary in their foraging behavior, survival and persistence abilities, habitat use, and host range (Gaugler 2002, Lewis et al. 2006), multiple species and strains must be experimentally compared for their efficacy against SHB under field conditions. Over the long term, the manipulation of native EPN species through cultural practices may be feasible, both for economic and conservation objectives (Lewis et al. 1998).

IPM approach for SHB: It is important to detect SHB infestations early by looking for adults under top covers or on bottom boards during all hive inspections. Infestations are more likely to occur when supering colonies, stacking empty or weak colonies on stronger ones, making weak splits, and exchanging combs, because these activities can inadvertently interfere with the protective behaviors of the bees. As preventative measures, apiaries should be free of equipment not in use and any food sources to which beetles may be attracted (honey, bits of comb, cappings, etc.). This also applies to the honey extraction area where stored equipment and filled supers should be protected against beetle infestations. Furthermore, supers with honey should be extracted quickly to reduce beetle damage.

In combination with preventative cultural practices described above, our goal is to test and develop a sustainable and economical feasible management approach of biological and chemical control tactics to keep SHB populations below levels that affect honey bee colony health and

production. No biopesticides or biological control protocols are available for SHB control at this time (Neumann and Elzen 2004). Our proposed control tactics will involve in-hive adult traps to reduce egg-laying in combination with soil-dwelling entomopathogenic nematodes and biopesticides to target wandering pre-pupal larvae entering the soil and adults emerging from the soil. As a soil drench for SHB control, we will test two organically certified biopesticides: the active ingredient spinosad applied as a liquid solution of Entrust® (Dow AgroScience) and a pyrethrum- and azadirachtin-based product called Azera® (McLaughlin Gormley King).

Spinosad is a novel insect control agent derived by fermentation of the Actinomycete bacterium, *Saccharopolyspora spinosa*. It received the 1999 Presidential Green Chemistry Challenge Award which recognizes chemicals that reduce negative impacts on human health and the environment (López et al. 2005). Spinosad controls a wide range of insect pests at very low application rates, and seed treatments and transplant drenches of Entrust have been effective against soil insects such as root maggots. The dual biopesticide formulation of AZERA has been developed primarily for the organic markets and was recently registered by EPA for a wide range of labeled uses. Pyrethrum is expected to give quick knockdown of SHB larvae entering soil, whereas azadirachtin will cause disruption of molting and pupation (Schmutterer 1988, Isman 1993). Azadirachtin has a safe use history and does not have significant toxicity to honey bees. Pyrethrum and spinosad are highly toxic to worker honey bees if orally exposed under worst case laboratory conditions. However, residue tests conducted under semi-field and field conditions on worker honey bees foraging on treated foliage indicated that dry product residues are harmless (Dively, unpublished data). Thus, if applied in the evening when bees are inactive, drench treatments do not pose an unacceptable risk of harm to honey bees. Both biopesticides have the potential to be effective and cost competitive as an alternative to the less environmentally friendly GardStar® soil treatment.

EPN have a broad host range and are safe for plants and vertebrates, easily applied with standard spray equipment, and compatible with many chemical pesticides (Georgis et al. 2006). The great majority of tested chemical pesticides do not affect mortality, activity, or virulence of infective juveniles (Rovesti and Deseö 1990, Shapiro-Ilan et al. 2006). Moreover, the integration of both control tactics in the soil may result in a synergistic interaction that could enhance overall SHB control. In studies on white grubs, the root-feeding larvae of scarabaeid beetles, the combination of EPN and imidacloprid caused mortality far greater than observed from either approach in isolation (Koppenhöfer and Kaya 1998). The mechanistic explanation for this phenomenon is that the sublethal effects of imidacloprid disrupt normal nerve function of the grubs, and thus reduce overall activity level and defensive evasive and grooming behaviors that deter nematode attachment and penetration (Koppenhöfer et al. 2000). Similar synergistic responses were observed with the anthranilic diamide, chlorantraniliprole, used in concert with *Heterorhabditis bacteriophora* to control the oriental beetle, *Anomala orientalis* (Koppenhöfer and Fuzy 2008). It is therefore plausible that inhibition of SHB pupation in the soil by azadirachtin (away from hives) may prolong and intensify their lethal exposure to beneficial nematodes.

(8.ii) Objectives and Impacts

Objectives: Our research goals are to develop, evaluate, and demonstrate a multi-faceted sustainable IPM strategy for SHB and provide valuable information regarding SHB management

that can be immediately adopted by beekeepers. **Objective 1** will: 1) evaluate entomopathogenic nematodes and biopesticides separately and in tandem for their ability to suppress wandering larval stages of the SHB; and 2) evaluate in-hive traps (both commercially available types and new, modified traps) for their ability to reduce SHB colonization and egg-laying. **Objective 2** will involve 1) on-site demonstrations with master beekeepers to evaluate IPM methods, and 2) dissemination of recommendations through various extension outreach activities to help beekeepers and local beekeeping associations rapidly implement a sustainable IPM strategy. Throughout the two-year project, a continuous commitment will be made to learn more about SHB and their effects on colony health. In accomplishing these objectives, we hope to increase the long-term viability and survival of apiary operations by enhancing SHB management and lowering management cost.

The objectives in the proposed project address 13 of the 14 specific RIPM relevance criteria and align with three goals of the NE IPM Center. Moreover, the objectives parallel general research and educational priorities established by the Pest Management Strategic Plan for honey bees in the Mid-Atlantic States (= DE, MD, NC, NJ, PA, SC, VA, WV), completed by MAAREC in March of 2008 (<http://www.ipmcenters.org/pmsp>), as well as specific priorities laid out for SHB in particular (*please see Relevance statement for extensive details*). These industry stakeholders have consistently ranked integrated pest management and non-chemical control tactics as high priorities for sustainable SHB control.

Anticipated impacts: Honey bees are essential for the pollination of all tree fruits, small fruits, and cucurbit crops valued in excess of \$40 million in Maryland (USDA-NASS 2007). Approximately 10,000 colonies in 1,625 apiaries are maintained by 1,352 beekeepers in the state. Of these colonies, only an estimated 6,444 are used for pollination services on 319 Maryland farms (unpublished data, MD Crop Statistics). The IPM practices developed by this project will reduce colony losses due to SHB, resulting in more efficient and sustainable apiculture operations providing pollination services and honey production. According to the Maryland agricultural census, 931 and 451 Maryland producers of cucurbit crops and fruit crops, respectively, will benefit from increased yields if their specialty crops are all pollinated with the recommended number of honey bee colonies per acre. This census does not include data on honey production due to disclosure restrictions; however, development of sustainable control practices for SHB will certainly enhance honey production. In particular, organic honey operations will benefit because these control tactics are all certified for organic production.

Our project will result in immediately adoptable IPM practices for all beekeepers. The biopesticides will be readily adopted if demonstrated to be inexpensive and efficacious as replacements for existing, more hazardous, SHB control tactics. The development of EPN-based biological control would provide beekeepers with a safe, long-term solution to SHB infestations, if commercially available species are shown to be as efficacious in the field as in the lab. We anticipate that implementation costs of the alternative control tactics will be less than current methods of SHB control. Another economic benefit that may accrue to beekeepers adopting less hazardous SHB control tactics is an increased demand for bee products. Opportunities to sell beeswax have diminished as knowledge of the pesticide residues in wax has increased. Adoption of in-hive trapping combined with reduced-risk pesticides and nematodes would result in fewer contaminants in wax. Additionally, as honey bee viruses have been isolated from SHB (Eyer et

al. 2009b), beetles therefore may aid the spread of virus. As the range for SHB continues to expand, colony losses may increase due to direct impacts of SHB and to indirect effects from increased prevalence of virus in colonies. As such, if SHB are involved in transmission of and maintenance of viral populations, adoption of sustainable IPM tactics for SHB control will have an even greater impact on the health of honey bees, because few methods currently exist to reduce the transmission of virus to honey bees.

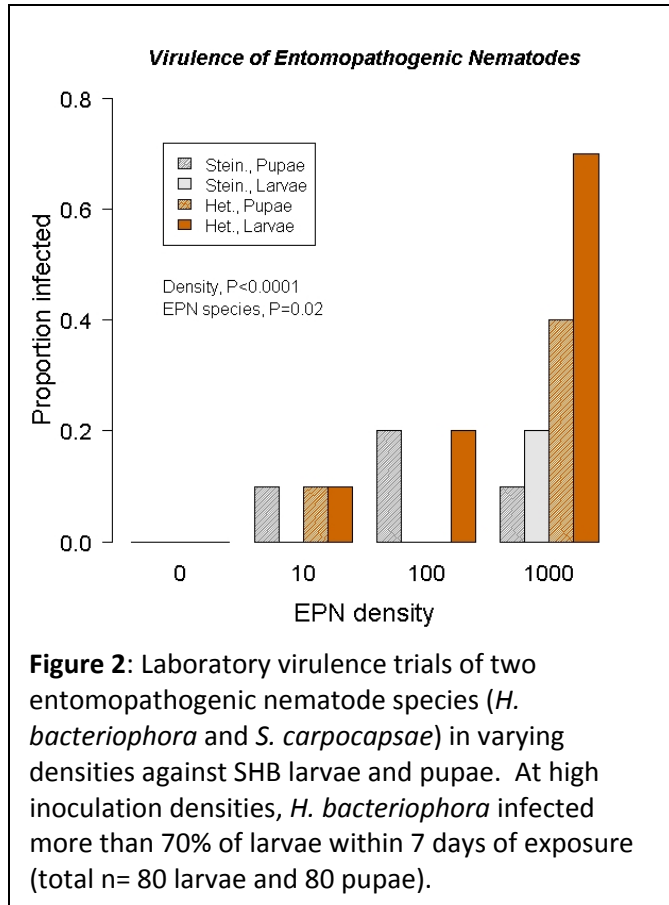
(8.iii) Approach and Procedures

Objective 1: *Evaluate efficacy of alternative control tactics applied as a multi-faceted sustainable IPM strategy for SHB control.*

We will test several cost-efficient, in-hive trapping devices to capture adults and reduce egg-laying. We will determine if beneficial nematodes can control SHB populations and if nematode enhancement of apiary soils is viable and feasible. Additionally, we propose to test biorational drench treatments, Azera® and Entrust®, to control pre-pupae. We intend to evaluate these tactics singly and together to determine their compatibility and possible synergism.

Novel In-Hive Trapping: In the summer of 2009, we evaluated three in-hive trapping methods to control small hive beetles. Sixteen colonies were established with bee packages and sister queens and initially fed sugar syrup to build-up populations. Four hives were equipped with each of three traps: AJ trap (one per hive), shelter straws filled with boric acid, and the Freeman bottom trap. Another four colonies without traps served as controls. Visual counts of active adult beetles on frames and number of beetles captured by each trap were recorded each week. Control colonies became heavily infested and stressed by SHB by mid July, resulting in the loss of two colonies. Boric acid straws captured no beetles and thus infestations did not differ from control colonies. Pooled counts of active beetles observed in control and boric acid colonies averaged 12.9 ± 2.19 s.e.m per hive. Counts of live beetles in the AJ trap and Freeman trap averaged 9.7 ± 1.72 and 4.6 ± 0.81 , respectively. The Freeman trap was the most effective, capturing approximately twice the number of beetles as the AJ trap. We will continue tests of AJ and Freeman traps, with the objective of reducing adult colonization and egg-laying, in combination with soil drench treatments to reduce the active larval and pupal stages outside the hive.

Entomopathogenic nematodes, laboratory studies: At the University of Maryland, we currently maintain robust colonies of SHB, following well documented laboratory procedures for mass rearing (Neumann et al. 2001, Mürrle and Neumann 2004). These laboratory colonies will be used to supply individuals for laboratory and field studies. We will use both commercially available EPN species and species isolated locally from Maryland agricultural sites to test against SHB larvae, pre-pupae, pupae, and adults by standard protocols in a series of replicated virulence trials in the laboratory (Woodring and Kaya 1998). Preliminary trials show promising results for high inoculation densities of *Heterorhabditis bacteriophora* on SHB immature stages, particularly larvae (**Figure 2**). These results mirror the only published report which showed higher virulence of *H. megidis* relative to *Steinernema* species (Cabanillas and Elzen 2006). We will repeat virulence trials against honey bee larvae, pupae and adults (Kaya et al. 1982, Baur et al. 1995) to assess the potential for non-target impacts. By varying the densities, timing,



nematode species and strains, and abiotic conditions of challenge trials, the results will help to optimize the combinations used in field efficacy trials described below.

Entomopathogenic nematodes and biopesticides, field studies: We will employ functional colony experiments to evaluate the effectiveness of entomopathogenic nematodes and soil drenches of biopesticides applied alone and in combination with nematodes to control SHB populations. Experiments will be conducted in 2010 and repeated in 2011 on the University of Maryland Hayden Farm, which is part of the USDA Beltsville Agricultural Research Center in Beltsville, MD. This almost contiguous area of about 6,600 acres of fields, pastures, orchards, and forests provides an ideal setting for isolating replicate apiaries under different treatment regimes. In the first year, the experimental design will consist of four randomized treatments

applied to the soil beneath ten replicate colonies for each treatment regime: 1) tap water drench as a control; 2) soil inoculations of entomopathogenic nematodes; 3) soil drenches of the biopesticides; and, 4) a combination of nematodes and biopesticides. Replicates of treatment regimes 3 and 4 will be split into two sub-treatments involving five replicates of each biopesticide (Azera and Entrust). The Freeman in-hive trapping device will be tested in five replicate colonies of treatment regimes 1 and 2. Variations on these treatments will be repeated in the second field season, but with adjustments to different traps, EPN species or application rates of the EPN and/or biopesticide.

A total of 40 colonies will be established in single deep hive boxes, starting with one drawn comb and nine foundation frames. Packages of 900 g of bees from source colonies free of SHB will be obtained from a commercial supplier (Wilbanks Apiaries; Claxton, GA, USA) and used to establish colonies by mid April 2010. For the first month, all colonies will be located in one apiary and fed sugar syrup and a pollen substitute (MegaBee patties) to allow colonies to build up before they are assigned to treatment groups. During mid May, hives will be inspected to visually assess and equalize bee and brood densities if necessary. At this time, a medium-depth super will be added to each hive, and colonies will be assigned to treatment groups and relocated to ten isolated apiaries. Each apiary will consist of four colonies representing one replicate of each treatment. Individual hives will be set on stands (0.5 m high) and spaced 10 m apart in each apiary. The apiaries will be temporarily located along the forest edges of agriculturally managed

fields, pre-sampled to verify low endemic populations of entomopathogenic nematodes, particularly the species used in the study.

Each colony will be manually infested each day with 50 adult SHB for 10 consecutive days. The small hive beetles will be reared in the laboratory. One week after the final infestation, releases of nematodes and drench treatments of biopesticides will be applied to soil surface areas (4 m^2) within the adjacent field and then colonies will be relocated to these treated sites. Drench treatments will be applied using a sprinkler can until the ground is thoroughly wet over the 4 m^2 areas. Commercial EPN formulations will be suspended in concentrated stock solution of known densities of infective juveniles. A general rule of thumb is that an inundation rate of 250,000 IJs per m^2 is required for successful pest suppression (Shapiro-Ilan et al. 2006). Therefore, we will apply nematodes at this density over the same 4 m^2 footprint used in pesticide trials. The density of this solution will be adjusted to match the total volume of fluid added in the biopesticide applications, and an equal volume of tap water will be applied for treatment control plots. In the last treatment, nematode and biopesticide solutions will be concentrated by a factor of two and combined for single applications at the same fluid volumes and area as in the other treatments. Applications of all four treatment levels (drench treatment, EPN, drench treatment + EPN, control) around the hives will be repeated every two weeks for two months through late July. The Freeman bottom trap will be placed in five replicate colonies of treatment regimes 1 and 2, after all hives are manually infested, and during the second year in off-site demonstration trials with master beekeepers.

Colonies will be inspected biweekly to visually estimate bee strength, brood development, and stores of food, expressed as a percentage of comb area covered; to examine for the presence and egg-laying status of the queen; and to count the number of adult SHB seen running across the combs when the hive is opened. On alternate weeks, observations of each colony will be made to record the number of foraging bees returning to the hive. Counts of bees with and without pollen pellets entering the hive entrance will be tallied over a 5-minute period in the morning between 9.00 and 11.00. SHB densities will be monitored using corrugated cardboard inserts (with the paper removed on one side), placed corrugated side down on the bottom board. These shelter traps will be checked regularly, along with evidence of beetle frass on the bottom board.

The experiment will be terminated in late July when final measurements will be recorded. Colonies that are unable to establish after the initial colony setup or those that died but not due to SHB stresses will be deleted from the final results. Absconding colonies will be recorded as a treatment effect. Data will include: percentage of combs covered with bees, capped brood, and stores of honey and bee bread; weight of a sub-sample of 100 bees; net weight gain of medium supers as a quantitative estimate of colony production; final density of SHB adults and larvae determined by aspirating and counting; and presence/ absence of a laying queen. Depending on results from the first year, we may repeat the same experiments with different EPN species, or may adjust the formulations of EPN and/or the biopesticide in an effort to improve or fine-tune the observed suppression of SHB. For data analysis, we will apply a flexible linear modeling approach dependent on the data; for example with binomial distributed response variables (e.g., infection prevalence of SHB by EPN), we will use the logit link-function in generalized linear model procedures. We will analyze data in the SAS and R statistical packages (SAS Institute 1996, <http://www.R-project.org>)

Objective 2: *Conduct on-site demonstrations and disseminate research results through various extension outreach activities to help beekeepers and local beekeeping associations rapidly implement a sustainable IPM strategy for SHB control.*

Demonstration, extension education, implementation, and evaluation: Over the past five years, federal, state and non-governmental organizations in the Mid-Atlantic region have been working to assemble partnerships of honey bee expertise. MAAREC is an example of such effort to bring together expertise to address current and emerging issues of concern to beekeepers. The Maryland Honey Bee Working Group (MHBWG) was also recently formed to identify research and extension needs for beekeepers and serve as a catalyst to coordinate efforts in addressing the crisis of CCD and other issues regarding honey bees and pollination. Clearly, interest in collaborative extension education and demonstration work in beekeeping is high, and there is an existing network of honey bee educators operating in the Mid-Atlantic region. Funding from this project will help to maximize these existing partnership and collaborative efforts to ensure that research findings will be rapidly implemented by the beekeeping industry.

Mid-Atlantic beekeepers are acutely aware of the adverse impact of SHB, viruses and chemical applications on their honey bee colonies. A total of 154 beekeepers from Virginia, Maryland, Delaware, Pennsylvania, and New Jersey completed a mail-in questionnaire survey designed to assess the importance of small hive beetle as a pest of honey bee colonies. Sixty-nine percent (69%) of the respondents reported SHB infestations in their colonies, of which 56, 38, and 6% were considered light, moderate, and heavy infestations, respectively. The impact of SHB also was evident by the frequency of management practices used to control the pest. Forty-six percent (46%) of the beekeepers reported that they have or are currently using traps, in-hive insecticides, soil drench treatments, or combinations of two or more methods to control SHB infestations. Almost half of the infestations were considered most stressful on overwintered colonies early in the spring. Forty-two percent (42%) of beekeepers experienced SHB problems for the first time within the last three years, indicating that SHB is increasing in economic importance.

Therefore, the level of interest in non-chemical and softer chemical control tactics for all honey bee pests, including SHB, is very high. Based on the first year of studies, we will conduct collaborative demonstrations of in-hive trap devices, biopesticide drench treatments, and soil inoculations of entomopathogenic nematodes to reduce SHB infestations. Working with master beekeepers and local clubs (see letters of support), demonstration trials in apiaries (including a few organic operations) will be conducted at locations where SHB currently exists throughout Maryland and the greater Mid-Atlantic region. The project, supplemented by funding from the State Extension IPM Program, will provide different traps and soil treatments for master beekeepers to test. We believe that this approach is important as an educational tool to test first-hand the relative efficacy of these trapping devices and soil drenches. These key beekeepers will then train other beekeepers through local associations.

Once the project has demonstrated a sustainable IPM strategy for SHB control, we will launch an extensive outreach effort to get the message out to the beekeeping community. This will begin in the first year but will ramp up in the second year and continue beyond the funding period. Presentations at winter and spring state and regional beekeeper meetings and local workshops

will include results of the research and instructions on how to implement IPM tactics for SHB control. Altogether, these events should enable the direct training of more than 400 beekeepers each year. Apiary inspectors in the mid-Atlantic region will learn about project findings at annual in-service training events held in each state and will be instrumental in promoting IPM practices for SHB control with every registered beekeeper.

The key information vehicle for honey bee management recommendations in the Mid-Atlantic region is the MAAREC website, where detailed information on the IPM strategy for SHB control will be posted. The potential applicability of this information to other production regions is great because the MAAREC website reaches beekeepers throughout the United States. In 2008, the site averaged a mean of 5,491 independent visits per month with a high of 8,995 visits in July, of which 78% were U.S. visitors and 21% international. Additionally, research-based information generated by this project will be posted as subject matter on the national eXtension website.

Objective #1: Test Novel IPM Strategies	2010												2011												2012				
	A	M	J	J	A	S	O	N	D	J	F	M	A	M	J	J	A	S	O	N	D	J	F	M	...	D			
Laboratory EPN virulence experiments																													
Eval. of EPN nontarget effects on bees																													
Establish 40 beehives at field site																													
Experim. infest hives with SHB; field tests of EPN, biopesticides, & traps																													
Sample brood, bees, colony health & SHB																													
Prep manuscripts for professional & trade journals																													
Objective #2: Demonstration, extension and implementation of IPM																													
develop, send, collect beekeeper surveys																													
collaborative demonstration hives																													
present research findings at state & regional beekeeper meetings																													
Educate apiary inspectors at annual in-service training events																													
prepare magazine, newspaper articles and web-based material (eXtension)																													
Post SHB management recommendations at MAAREC website																													
Develop online video training module																													
Prepare reports to RIPM via CRIS																													

Figure 3: Timetable for major proposed project activities, 2010-2012.

Written and web-based material will be produced to describe steps to implement IPM tactics for reduction in SHB populations. Project results will be disseminated via articles in beekeeping magazines, local agricultural-related newspapers, and on-line IPM newsletters (each state has IPM websites for this purpose). Additionally, a video learning module will be made available on-line and contain a complete educational package of information explaining SHB biology, ways the beetle’s lifecycle can be interrupted, and how to implement sustainable management tactics. This self-directed module will allow beekeepers to obtain information and educate themselves when it is convenient for them.

Ultimately, results of the project will be incorporated into existing IPM systems throughout the country through our participation in the Multistate Research Project NC 508, "Sustainable Solutions to Problems Affecting Honey Bee Health." Finally, we anticipate several high-profile

scientific papers of project results will be given at national apicultural research meetings and published in professional and trade journals. The timetable for all proposed approaches and procedures is summarized in **Figure 3**.

(8.iv) Evaluation Plans

Following the logic model (**Figure 4**), anticipated impacts associated with the project will be measured by three mechanisms: 1) an annual regional beekeeper survey, 2) yearly interviews of individual beekeepers, and 3) feedback from the representatives of MAAREC. During the first year of the project we will conduct a baseline survey (we will secure approval for human subjects prior to implementation) covering topics such as the presence/absence of SHB in colonies, SHB control tactics used, number of apiaries, total number of colonies, number of colony deaths and cause of death if known, and cost of pesticides and traps purchased. Following delivery of research results, questions on the direct cost of IPM tactics, labor costs from implementing IPM and/or using pesticides, and problems encountered while implementing IPM tactics will be added to the survey. The specific questions used will be vetted by the MAAREC group. The survey will be distributed through state and local beekeeping organizations and will be available electronically on the MAAREC website. Our largest obstacle is a low return rate. To counter this problem, a brief explanation of the value of their response will be included with the survey. The importance of surveys will be discussed at beekeeper meetings, including how the information helps us to better serve the beekeepers. We will consider including a drawing for beekeeping supplies and other incentives to increase response rate.

Master beekeepers – innovators, highly respected by other beekeepers – and officers from state beekeeping associations will be interviewed in person (when possible) or by phone or email. Specific emphasis will be problems encountered during implementation of IPM strategies along with suggestions on how to more effectively communicate with beekeepers. Suggestions for additional interviewees will be solicited from MAAREC and state apiary inspectors. At least one MAAREC meeting per year we will request feedback on our IPM program. Successes from this project will be included in the MAAREC recommendations for beekeeping practices.

Because the tactics evaluated herein are readily adoptable by beekeepers, we expect to see a reduction in the frequency of insecticide use for the control of SHB. Honey samples will be taken to evaluate this possible effect. Colony loss from SHB should be reduced through the implementation of IPM tactics tested, and we anticipate improvement in colony health and pollination services. The economic benefits to beekeepers implementing the proposed IPM can be realized quickly because multiple EPN species isolated from Maryland field sites and used in pilot experiments (Figure 2) are already commercially available, and we anticipate that the biopesticides will be on the market before the completion of this project. The percent of colonies lost during the growing season and over winter should decline, reducing the need to purchase bees. Losses due to damaged equipment, contamination of honey, and cleanup of honey houses and extracted honey frames should be reduced with improved control of SHB. Beekeepers should not incur additional labor costs as neither IPM strategy being tested is more labor intensive than current control tactics. It is possible that labor costs and input costs could decline if the EPN persist without repeated inundative releases. We will quantify these predictions using the goals set forth in **Table 1**.

Table 1: Measurable impact objectives

Goals	Measurable Impact Objectives
Field test three prongs of IPM: traps, EPN, biopesticides	Biologically significant enhancement of colony health, honey production and reductions in SHB at a comparable or reduced cost from present tactics.
Demonstrate alternative IPM tactics.	In the second year, demonstration hives will be established at four research farms and at apiaries of four master beekeepers. These sites will be used in two beginner short courses.
Disseminate project results	During the two years, we will educate and train 300 beginner and established beekeepers to use in-hive traps and biorational control.
Increased adoption of alternative IPM tactics	By the end of the second year, 200 beekeepers will implement IPM tactics for SHB control.
Reductions in colony losses due to SHB	By the end of the second year, colony losses attributable to SHB infestation will drop by 50%.
Increases in the number of apiculture operations	Number of new beekeepers will increase by 10%.
Increase the percentage of specialty crops pollinated by honey bee colonies	Currently, less than the recommended number of bee hives is used in only 60% of the bee-pollinated specialty crops. By the end of the project, 75% will be pollinated with the appropriate number of colonies per acre.
Increased demand for bee products	Opportunities to sell honey and beeswax will increase by 10%.

(8.v) Key Personnel

Project Director **Dr. Daniel Gruner** has worked with the ecology and management of entomopathogenic nematodes and soil ecosystems for 5+ years. Gruner will rear lab stocks of SHB and oversee laboratory experiments and the isolation, cultivation, formulation, and application of beneficial nematodes for field studies. Coordinating with Dr. Dively on the biopesticide applications and their formulation with nematodes, Gruner will oversee the successful completion of Objective 1 (experimental testing and verification of IPM methods), analyze these experimental data, and will co-author extension reports and manuscript(s) reporting the results from the project. **Dr. Cerruti Hooks**, a research entomologist and extension specialist in vegetable cropping systems, is a Co-Investigator for this project and will direct extension activities (Objective 2). Dr. Hooks will develop extension materials and bulletins arising from the research and, with cooperators Mike Embrey (Extension Apiculturist) and Dr. Delaney (Delaware), will coordinate annual beekeeper surveys, deployment of demonstration hives, workshops and presentations for stakeholders, and general outreach education of beekeepers. **Dr. Galen Dively**, Emeritus Entomologist, is a Co-Investigator for this project, and will work closely with Drs. Hooks and Gruner on both research and extension objectives of the project. Dively will work with all cooperators to bridge the experimental and extension objectives. Together, the team will arrange for study sites at the research farm, procure the hive equipment, bees and queens to establish the honey bee colonies, semiweekly drenches of biopesticide treatments at each replicate site, the collection and supplemental feeding of bees, assessment of SHB samples from treated colonies, and record keeping of the proposed colony and foraging data.

Figure 4: Logic Model for Sustainable Management of the Small Hive Beetle (*Aethina tumida*), an Emerging Pest of Honey Bees
Situation: In recent years Northeastern beekeeping has generally declined and the incidence of pests, such as *Varroa* and the small hive beetle (SHB), have risen in tandem. The SHB disrupts and destroys honey bee colonies and its range and impacts are on the rise, but current control measures have unacceptable costs and collateral impacts on honey bees and their products. We propose novel, biorational IPM and extension to address these problems.

Inputs	Outputs	Participants & audience	Activities	Outcomes			
				Short	Medium	Long	
<ul style="list-style-type: none"> • Project leaders/staff • Network of partners • local, state, and regional beekeeper associations • State Apiary inspectors • In-kind resources • Funding • Materials • Stakeholders' needs • Extension base 	<ul style="list-style-type: none"> • Formalize an ad hoc alliance with master bee keepers and several honey bee associations • Establish colonies on commercial apiaries & centers of education • Demonstrate results and information on new IPM strategies • Train stakeholders how to integrate biological, cultural, and bioinsecticides • Short course training to bee keepers • Develop & distribute educational material 	<p><i>Participants</i></p> <ul style="list-style-type: none"> • Program leader/staff • Master beekeepers • Beekeeper associations • Education center crew • Extension Partners <p><i>Audience</i></p> <ul style="list-style-type: none"> • Beekeepers • Apiary inspectors • Consumers • Stakeholders • Government agencies • Future partners • Scientific community 	<ul style="list-style-type: none"> • Extension articles, web-clippings, short courses • Participants learn best management practices for honey production • Participants become familiar with integrating soft chemicals, biological and cultural control • Participants consider how these new tactics might be incorporated into their current honey production scheme • Bee keepers learn new ways to manage bee colonies • Bee keepers gain confidence in their new skill sets, teach others • Bee keepers make better production decisions. 	<ul style="list-style-type: none"> • Increased number of bee keepers use IPM tactics • Increased number of farmers adopt best management practices for managing bees • Bee keepers and other stakeholders have easy access to new information • Bee keepers adopt practices that are economically and environmentally feasible • Decrease use of high risk pesticides • Agri-chemicals replaced by more sustainable, biorational options • Level of beekeeper interest in alternative SHB control tactics reach an all time high. 	<ul style="list-style-type: none"> • Formal statewide partnership of federal, state, and non-governmental honey bee experts assembled • Significant improvement in honey bee health in Northeast • Notable reduction in insecticide residue in honey • Permanent stakeholder involvement in honey bee applied research & extension program • Increased role of biological control in bee keepers IPM programs • Overall increase in honey production at lower operational and environmental cost • Honey bee industry in NE moves towards economic stability • Bee keepers who left the industry return and prosper. 	<p>External Factors: Bee keepers willing to change practices to protect their colonies and lower production costs. Bee keeping associations such as the Mid-Atlantic Apiculture Research and Extension Consortium promote adoption of a more integrated IPM program for SHB management.</p>	<p>Assumptions: Honey bee keepers are aware of the spread of SHB and the increased production costs it causes.</p> <p>Evaluation: Surveys and questionnaires to determine impact. New technol. (i.e., clicker - Interactive software) will be used at meetings, conferences</p>

9. REFERENCES CITED: Sustainable Management of the Small Hive Beetle (*Aethina tumida*), an Emerging Pest of Honey Bees

Gruner, D.S. (PD), C.R.R. Hooks, and G.P. Dively (University of Maryland)

- Baur, M. E., H. K. Kaya, Y. S. Peng, et al. 1995. Nonsusceptibility of the honey bee, *Apis mellifera* (Hymenoptera: Apidae), to steinernematid and heterorhabditid nematodes. *Journal of Nematology* 27:378-381.
- Berenbaum, M. R., P. Bernhardt, S. Buchmann, et al. 2007. *Status of Pollinators in North America*. National Academies Press, Washington, DC.
- Biesmeijer, J. C., S. P. M. Roberts, M. Reemer, et al. 2006. Parallel declines in pollinators and insect-pollinated plants in Britain and the Netherlands. *Science* 313:351-354.
- Cabanillas, H. E., and P. J. Elzen. 2006. Infectivity of entomopathogenic nematodes (Steinernematidae and Heterorhabditidae) against the small hive beetle *Aethina tumida* (Coleoptera: Nitidulidae). *Journal of Apicultural Research* 45:49-50.
- Cox-Foster, D. L., S. Conlan, E. C. Holmes, et al. 2007. A metagenomic survey of microbes in honey bee colony collapse disorder. *Science* 318:283-287.
- Delfinado-Baker, M., and M. A. Houck. 1989. Geographic variation in *Varroa jacobsoni* (Acari: Varroidae): application of multivariate morphometric techniques. *Apidologie* 20:345-358.
- Denno, R. F., D. S. Gruner, and I. Kaplan. 2008. Potential for entomopathogenic nematodes in biological control: a meta-analytical synthesis and insights from trophic cascade theory. *Journal of Nematology* 40:61-72.
- Ellis, J. D., K. S. Delaplane, R. Hepburn, et al. 2003a. Efficacy of modified hive entrances and a bottom screen device for controlling *Aethina tumida* (Coleoptera: Nitidulidae) infestations in *Apis mellifera* (Hymenoptera: Apidae) colonies. *Journal of Economic Entomology* 96:1647-1652.
- Ellis, J. D., R. Hepburn, K. S. Delaplane, et al. 2003b. The effects of adult small hive beetles, *Aethina tumida* (Coleoptera: Nitidulidae), on nests and flight activity of Cape and European honey bees (*Apis mellifera*). *Apidologie* 34:399-408.
- Ellis, J. D., R. Hepburn, B. Luckman, et al. 2004. Effects of soil type, moisture, and density on pupation success of *Aethina tumida* (Coleoptera : Nitidulidae). *Environmental Entomology* 33:794-798.
- Ellis, J. D., P. Neumann, R. Hepburn, et al. 2002. Longevity and reproductive success of *Aethina tumida* (Coleoptera: Nitidulidae) fed different natural diets. *Journal of Economic Entomology* 95:902-907.
- Elzen, P. J., J. R. Baxter, D. Westervelt, et al. 1999. Field control and biology studies of a new pest species, *Aethina tumida* Murray (Coleoptera, Nitidulidae), attacking European honey bees in the Western Hemisphere. *Apidologie* 30:361-366.
- Eyer, M., Y. P. Chen, M. O. Schafer, et al. 2009a. Small hive beetle, *Aethina tumida*, as a potential biological vector of honeybee viruses. *Apidologie* 40:419-428.
- Eyer, M., Y. P. Chen, M. O. Schafer, et al. 2009b. Honey bee sacbrood virus infects adult small hive beetles, *Aethina tumida* (Coleoptera: Nitidulidae). *Journal of Apicultural Research* 48:296-297.
- Freeman, J. 2009. A new hive beetle trap. http://freemanbeetletrap.com/yahoo_site_admin/assets/docs/ANEWBEEETLETRAP2.33153109.pdf

- Gallai, N., J.-M. Salles, J. Settele, et al. 2009. Economic valuation of the vulnerability of world agriculture confronted with pollinator decline. *Ecological Economics* 68:810-821.
- Gatton, H., S. Nessler, R. Fell, et al. 2008. Pest Management Strategic Plan for Honey Bees in the Mid-Atlantic States (DE, MD, NC, NJ, PA, SC, VA, WV).
<http://www.ipmcenters.org/pmsp/pdf/MidAtlanticHoneyBeePMSP.pdf>
- Gaugler, R., ed. 2002. *Entomopathogenic Nematology*. CABI Publishing, London.
- Gaugler, R., E. Lewis, and R. J. Stuart. 1997. Ecology in the service of biological control: the case of entomopathogenic nematodes. *Oecologia* 109:483-489.
- Georgis, R., A. M. Koppenhöfer, L. A. Lacey, et al. 2006. Successes and failures in the use of parasitic nematodes for pest control. *Biological Control* 38:103-123.
- Grant, J. A., and M. G. Villani. 2003. Soil moisture effects on entomopathogenic nematodes. *Environmental Entomology* 32:80-87.
- Haarmann, T., M. Spivak, D. Weaver, et al. 2002. Effects of fluvalinate and coumaphos on queen honey bees (Hymenoptera: Apidae) in two commercial queen rearing operations. *Journal of Economic Entomology* 95:28-35.
- Hominick, W. 2002. Biogeography. Pages 115-144 in R. Gaugler, ed. *Entomopathogenic Nematology*. CABI Publishing, London.
- Hood, W. M. 2000. Overview of the small hive beetle, *Aethina tumida*, in North America. *Bee World* 81:129-137.
- Hood, W. M. 2004. The small hive beetle, *Aethina tumida*: a review. *Bee World* 85:51-59.
- Hood, W. M., and G. A. Miller. 2003. Trapping small hive beetles (Coleoptera: Nitidulidae) inside colonies of honey bees (Hymenoptera: Apidae). *American Bee Journal* 143:405-409.
- Isman, M. B. 1993. Growth inhibitory and antifeedant effects of azadirachtin on six noctuids of regional economic importance. *Pesticide Science* 38:57-63.
- Johnson, R. M., J. D. Evans, G. E. Robinson, et al. 2009. Changes in transcript abundance relating to colony collapse disorder in honey bees (*Apis mellifera*). *Proceedings of the National Academy of Sciences* 106:14790-14795.
- Kaya, H. K., and R. Gaugler. 1993. Entomopathogenic nematodes. *Annual Review of Entomology* 38:181-206.
- Kaya, H. K., J. M. Marston, J. E. Lindegren, et al. 1982. Low susceptibility of the honey bee, *Apis mellifera* L. (Hymenoptera: Apidae), to the entomogenous nematode, *Neoaplectana carpocapsae* Weiser. *Environmental Entomology* 11:920-924.
- Koppenhöfer, A. M., and E. M. Fuzy. 2008. Effect of the anthranilic diamide insecticide, chlorantraniliprole, on *Heterorhabditis bacteriophora* (Rhabditida: Heterorhabditidae) efficacy against white grubs (Coleoptera: Scarabaeidae). *Biological Control* 45:93-102.
- Koppenhöfer, A. M., P. S. Grewal, and H. K. Kaya. 2000. Synergism of imidacloprid and entomopathogenic nematodes against white grubs: the mechanism. *Entomologia Experimentalis et Applicata* 94:283-293.
- Koppenhöfer, A. M., and H. K. Kaya. 1998. Synergism of imidacloprid and an entomopathogenic nematode: a novel approach to white grub (Coleoptera: Scarabaeidae) control in turfgrass. *Journal of Economic Entomology* 91:618-623.
- Lacey, L. A., R. Frutos, H. K. Kaya, et al. 2001. Insect pathogens as biological control agents: do they have a future? *Biological Control* 21:230-248.
- Lehnert, T., and G. E. Cantwell. 1978. Effects of microbial insecticides on honey bees: a review. *American Bee Journal* 118:674-675.

- Lewis, E. E., J. Campbell, C. Griffin, et al. 2006. Behavioral ecology of entomopathogenic nematodes. *Biological Control* 38:66-79.
- Lewis, E. E., J. F. Campbell, and R. Gaugler. 1998. A conservation approach to using entomopathogenic nematodes in turf and landscapes. Pages 235-254 in P. Barbosa, ed. *Conservation Biological Control*. Academic Press, London.
- Lodesani, M., M. Colombo, and M. Spreafico. 1995. Ineffectiveness of Apistan® treatment against the mite *Varroa jacobsoni* Oud in several districts of Lombardy (Italy). *Apidologie* 26:67-72.
- López, O., J. G. Fernández-Bolaños, and M. V. Gil. 2005. New trends in pest control: the search for greener insecticides. *Green Chemistry* 7:431-442.
- Lundie, A. E. 1940. *The Small Hive Beetle*. Union of South Africa, Department of Agriculture and Forestry, Pretoria, South Africa.
- Milani, N. 1999. The resistance of *Varroa jacobsoni* Oud. to acaricides. *Apidologie* 30:229-234.
- Morse, R. A., and N. W. Calderone. 2000. The value of honey bees as pollinators of U.S. crops in 2000. *Bee Culture* March:1-15.
- Mürrle, T. M., and P. Neumann. 2004. Mass production of small hive beetles (*Aethina tumida*, Coleoptera: Nitidulidae). *Journal of Apicultural Research* 43:144-145.
- Neumann, P., and P. J. Elzen. 2004. The biology of the small hive beetle (*Aethina tumida*, Coleoptera: Nitidulidae): gaps in our knowledge of an invasive species. *Apidologie* 35:229-247.
- Neumann, P., C. W. W. Pirk, R. Hepburn, et al. 2001. Laboratory rearing of small hive beetles *Aethina tumida* (Coleoptera, Nitidulidae). *Journal of Apicultural Research* 40:111-112.
- Pettis, J. S., and H. Shimanuki. 2000. Observations on the small hive beetle, *Aethina tumida* Murray, in the United States. *American Bee Journal* 140:152-155.
- Rovesti, L., and K. V. Deseö. 1990. Compatibility of chemical pesticides with the entomopathogenic nematodes, *Steinernema carpocapsae* Weiser and *Steinernema feltiae* Filipjev (Nematoda, Steinernematidae). *Nematologica* 36:237-245.
- Sammataro, D., U. Gerson, and G. Needham. 2000. Parasitic mites of honey bees: life history, implications, and impact. *Annual Review of Entomology* 45:519-548.
- Schmutterer, H. 1988. Potential of azadirachtin-containing pesticides for integrated pest control in developing and industrialized countries. *Journal of Insect Physiology* 34:713-719.
- Shapiro-Ilan, D. I., D. H. Gouge, S. J. Piggott, et al. 2006. Application technology and environmental considerations for use of entomopathogenic nematodes in biological control. *Biological Control* 38:124-133.
- Torto, B., D. G. Boucias, R. T. Arbogast, et al. 2007. Multitrophic interaction facilitates parasite-host relationship between an invasive beetle and the honey bee. *Proceedings of the National Academy of Sciences* 104:8374-8378.
- vanEngelsdorp, D., J. Hayes, Jr, R. M. Underwood, et al. 2008. A survey of honey bee colony losses in the U.S., fall 2007 to spring 2008. *PLoS ONE* 3:e4071.
- Vega, F. E., P. F. Dowd, and T. C. Nelson. 1994. Susceptibility of driedfruit beetles (*Carpophilus hemipterus* L.; Coleoptera: Nitidulidae) to different *Steinernema* species (Nematoda: Rhabditida: Steinernematidae). *Journal of Invertebrate Pathology* 64:276-277.
- Wenning, C. J. 2001. Spread and threat of the small hive beetle. *American Bee Journal* 141:640-643.

Woodring, J. L., and H. K. Kaya. 1998. *Steinernematid and Heterorhabditid Nematodes: a Handbook of Techniques*. Southern Cooperative Series Bulletin, volume 331. Arkansas Agricultural Experiment Station, Fayetteville, Arkansas.

Zoltowska, K., Z. Lipinski, and E. Lopienska. 2003. Beneficial nematodes: a potential threat to honey bees? *Bee World* 84:125-129.

Field 12. Other Attachments: Relevance Statement – N.E. Region**(a) Names of Institutions of PDs and major cooperators:**

Gruner, Daniel S. (PD), Cerruti R.R. **Hooks**, and Galen P. **Dively**: University of Maryland

(b) Title: Sustainable Management of the Small Hive Beetle (*Aethina tumida*), an Emerging Pest of Honey Bees**(c) Project Type: *Joint Research-Extension***

(d) Project Summary: With this joint **research-extension** project (**\$38,661 PL89-106 + \$17,446 Smith-Lever**), we propose to develop and apply sustainable control practices for the ‘small hive beetle’ (SHB, *Aethina tumida*), which infests honey bee (*Apis mellifera*) colonies and vectors pathogens that may contribute to Colony Collapse Disorder. Honey bees – critical pollinators of a wide variety of fruit, nut and vegetable crops – are in protracted decline nationwide. The invasive SHB is rapidly expanding its range from the Southeast U.S., where it has been exceptionally destructive to colony health since 1998, into the mid-Atlantic and Northeast regions. Existing chemical controls are problematic for honey bee health because of exposure risks and associated costs. This project will develop, evaluate, and disseminate a multi-faceted, sustainable IPM strategy to disrupt the SHB life cycle. In functional hive experiments, we will evaluate two novel tactics – soil drenches of biopesticides and entomopathogenic nematodes to control wandering larvae – deployed in combination with in-hive trapping devices to capture invading adults (**Objective 1**). We will evaluate these IPM strategies through on-site demonstrations with cooperating master beekeepers and disseminate research results via eXtension education to help mid-Atlantic beekeeping associations rapidly implement recommendations (**Objective 2**). This project addresses thirteen NE-RIPM relevance criteria, three major priorities of the Northeast IPM center, and many specific directives from regional beekeeping organizations. Our approach will reduce environmental and human health risks by replacing hazardous pesticides with affordable traps, biorational organic-compatible pesticides, and augmentative biological control, and our demonstration and extension efforts will stimulate widespread adoption of IPM strategies across the region.

(e) Problem, Background and Justification: Honey bees are essential for pollination of more than 90 fruit and vegetable cash crops and are valued in excess of \$40 million in Maryland, \$14.6 billion in the United States, and over \$200 billion worldwide. However, pollinators worldwide are in decline, leading to a concomitant decline in their essential pollination services. Specifically, honey bee (*Apis mellifera*) population declines are accelerating with the increased incidence of pest arthropods, viral and bacterial pathogens, and accumulation of pesticide residues. The dramatic loss of colonies since 2006 and the rise of ‘Colony Collapse Disorder (CCD)’ are the latest demonstration of this. The National Agriculture Statistics Service reported that there were 2.44 million honey-producing colonies in the United States as of February 2008, down from 4.5 million in 1980, and 5.9 million in 1947. Since the early 1980’s pathogens and pests of American honey bee strains have risen dramatically with introduction of tracheal mites in 1984, followed by *Varroa destructor* in 1987, and small hive beetle (SHB) *Aethina tumida* Murray (Coleoptera: Nitidulidae) in 1998.

Since it was initially documented in the US, the distribution of the SHB continues to expand. Originating from sub-Saharan Africa, SHB has now been detected in >30 U.S. states. Although

weak colonies are most susceptible to their colonization, under heavy infestations, SHB may force honeybees to abandon their hives and can overwhelm and directly kill even strong colonies. The developing SHB larvae are the most destructive stage: larvae damage wax combs while feeding on pollen and brood, and ruin honey by defecating within the food cells, causing the honey to ferment and produce a foul odor. Furthermore, the honey thins and runs out of the combs, rendering it unmarketable. Multiple honeybee viruses have been isolated from SHB specimens, and SHB is a suspected vector involved in the recent epidemic of Colony Collapse Disorder. Thus, SHB can cause significant economic harm to hobby beekeepers, sideliners, or commercial operators through reduced quality of product, direct destruction, or bee abandonment of hives. In 1998, the first reported year of U.S. infestation, Florida beekeepers experienced an estimated \$3 million loss from SHB. At recent regional meetings of professional beekeepers, the Mid-Atlantic Apiculture Research and Extension Consortium (MAAREC, 20 October 2008) and the Maryland State Beekeepers Association (MSBA, 20 June 2009), industry stakeholders, smallholders and hobbyists identified SHB as a rapidly spreading honey bee pest of significant concern and expressed a strong priority for research geared to rapid implementation and extension delivery of improved control tactics.

Current chemical controls for SHB involve two approaches, both of which have limited efficacy and carry unacceptable risk to individual hives and to honeybee populations: 1) treatment with coumaphos impregnated strips placed inside the colony for adults (CheckMite+ Strips); and 2) soil drench treatment of permethrin for control of developing pre-pupae (GardStar®). These control tactics are problematic for honey bee health, have potential for misuse, provide inconsistent control, and are not approved for organic honey production. Many beekeepers are reluctant to use these chemical measures because of these continuing indirect and direct costs.

This project addresses 13/14 of the **RIPM relevance** objectives, as summarized in the table:

NE-RIPM Relevance Review Criteria	Evidence of Relevance
1) Will reduce risks to the environment 2) Will reduce risks to human health 9) Reduces dependence on chemical pesticides	We will test safer alternatives to predominant chemical controls, all of which are certified compatible with organic production
3) Has stakeholder support and the priority is cited 4) Focuses on a pest, crop, or setting found in at least five states or cropping regions 6) Involves multiple states in an active partnership 12) Addresses an emerging pest, crop, or problem	Letters of support, survey results, and MAAREC priorities demonstrate multi-state partnership and support of industry stakeholders for an emerging pest of high regional concern (MD, DE, NJ, PA, VA).
5) Will fill a niche (no such tactics or approaches exist) 7) Will advance IPM in as soon as three (3) years 13) Is likely to be adopted by the target audience	Current controls were not developed for SHB and involve unacceptable costs and risks. The proposed IPM uses inexpensive, rapidly implementable strategies.
10) Has significant economic implications 11) Explains, justifies, and will serve an “underserved audience.” 14) Advances an IPM practice that is more cost-effective than the status quo	Beekeepers should not incur additional labor costs as neither IPM strategy being tested is more labor intensive than current control tactics. Labor costs and input costs will decline if EPN persist without repeated inundative releases.

This project also addresses three important priorities of the **Northeast IPM center** (http://northeastipm.org/regu_regional.cfm): 1) Promotion of public (and beekeeper) awareness of an invasive species (SHB): we will work closely with MAAREC stakeholders in the design and implementation of our demonstration and extension; 2) Research on biocontrol methods: we will field test a very promising avenue of biological control of the SHB using entomopathogenic nematodes, the effects of which may be enhanced in synergy with low concentrations of biopesticides; and 3) Testing of pest management materials allowable in an organic production system: all three elements of our proposed IPM strategy are compatible with organic standards.

Finally, our project objectives directly address research and educational priorities established by the **Pest Management Strategic Plan for honeybees in the Mid-Atlantic States** (= DE, MD, NC, NJ, PA, SC, VA, WV), completed by MAAREC in March of 2008 (<http://www.ipmcenters.org/pmsp>), which include: 1) Determine ways to improve overall colony health and productivity; identify factors leading to decline in honeybee health, such as nutrition, stress and pests; focus on new or current issues; 2) Reduce dependence on chemical control methods; 3) Create and disseminate surveys of honeybee diseases and pests to apiarists throughout the mid-Atlantic region; 4) Design best management practices (BMPs) related to pests; 5) Share BMPs with beekeepers; 6) Work to improve viability of beekeeping industry; encourage and educate new or potential beekeepers; 7) Improve information transfer techniques (e.g., eXtension); and 8) Develop and maintain interstate collaborations, such as MAAREC, because operational networks may find it easier to get new control methods in the pipeline or registered for beekeeper use. Further, goals of this project are in direct line with the Mid-Atlantic PMSP priorities developed specifically for SHB: 1) Develop a better trap using pheromones or lures; 2) Research nematodes as biological control agents; 3) Research and develop alternatives to coumaphos; 4) Research and develop alternatives to Gardstar soil treatment; and 5) Educate beekeepers on traps and best control options. These industry stakeholders have consistently ranked IPM and non-chemical control tactics as high priorities for sustainable SHB control.

(f) Project Objectives and Anticipated Outcomes: This project will evaluate the efficacy of two novel tactics – biopesticides and entomopathogenic nematodes (EPN) to control wandering larvae and pupae – deployed in combination with in-hive trapping devices to capture adults entering hives (**Objective 1**). We will disseminate research results through demonstration activities and extension education to help beekeepers and local beekeeping associations rapidly implement an IPM strategy for SHB control (**Objective 2**).

Because the tactics evaluated herein are readily adoptable by beekeepers, we expect to see a reduction in the frequency of insecticide use for the control of SHB and reduced residues in beeswax and honey. Colony loss from SHB should be reduced through the implementation of IPM tactics tested, and we anticipate improvement in colony health and pollination services. The economic benefits to beekeepers implementing the proposed IPM can be realized quickly because multiple EPN species are already commercially available, and the biopesticides will be on the market before the completion of this project. The percent of colonies lost during the growing season and over winter should decline, reducing the need to purchase bees. Losses due to damaged equipment, contamination of honey, and cleanup of honey houses and extracted honey frames should be reduced with improved control of SHB. We will evaluate these goals, predictions, and economic implications with feedback from master beekeepers, beekeepers at regional meetings, and through the use of annual directed surveys.