ENHANCING BIOLOGICAL CONTROL OF WHITE GRUBS BY NATIVE PARASITIC WASPS ON GOLF COURSES

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Executive Summary:

Goals:

1) Investigate the biology, behavior, and importance of native and introduced species of *Tiphia* wasps, the predominant parasitoids attacking white grubs on golf courses. More specifically, we will:

   a) Determine which endemic *Tiphia* species most commonly parasitize white grubs on golf courses, and the range of grub species and instars that are attacked.
   b) Document the biology, seasonal life cycle, and incidence of native *Tiphia*, including periods of adult emergence and activity, oviposition behavior of wasps on grub victims, timing and duration of larval development, and incidence of parasitism as affected by grub population density and location on golf courses.
   c) Investigate oviposition behavior of *Tiphia*, particularly cues by which the wasps locate grubs and discriminate among grub species under the turf.

2) Determine the feasibility of enhancing *Tiphia* populations via habitat management, particularly through conservation or augmentation of wildflowers or other carbohydrate sources used by the adult wasps. We seek to:

   a) Identify wild or cultivated plants that are frequented by the adult wasps as carbohydrate (nectar or honeydew) sources
   b) Test if higher parasitism occurs when such plants are incorporated into golf course landscapes.
   c) Determine if providing supplemental carbohydrate (sugar) sources for the adult wasps will increase parasitism rates of grubs.

Tiphia wasps are the dominant group of parasitic insects that attack white grubs in the soil. During 2000, we determined that two species of *Tiphia* are common on golf courses in Kentucky. Adults of *Tiphia vernalis*, a species introduced for control of Japanese beetles, were active from May 4 to June 1. *Tiphia pygidialis*, a native species that attacks masked chafer grubs, were active from August 1 to September 14. Yellow pan traps were effective for monitoring early-season activity of *T. vernalis*, whereas both species could be monitored with a 10% sugar water solution. Weekly sampling of the natural grub population showed that both *Tiphia* species parasitized primarily third instars. Parasitism of masked chafers averaged 15% at two golf courses, but was as high as 37% at some sites.

Cues used by *Tiphia* to locate grubs below ground were examined by use of an "ant farm", positioned horizontally to allow observation of wasp behavior in the soil. Wasps were found to locate their victims by following species-specific scent trails left by the grub as it moves through the soil. They showed an even stronger response to frass from their host grubs. This is the first study to show how *Tiphia* wasps

locate grubs underground. Ovipositional behavior of each wasp species was characterized. Both *T. vernalis* and *T. pygidialis* deliver a paralyzing sting and then manipulate the body of the grub in preparation for oviposition. Then, the female scrapes the grub to thin the cuticle where the egg will be laid. *T. vernalis* lays its egg on the underside of the grub, whereas *T. pygidialis* lays its egg on the back of the host grub. Eggs hatch in 3 to 5 days. Larval development is completed in about 21 days, after which a cocoon is spun. When offered Japanese beetle, masked chafer, or May beetle grubs, *T. vernalis* parasitized only Japanese beetles. *Tiphia pygidialis* were tested with the aforementioned grub species, plus two additional exotic species, European chafer and Oriental beetle. In general, only masked chafer grubs were attacked. In one case, however, *T. pygidialis* parasitized and completed larval development on a third-instar European chafer.

The relationship between the developing larval *Tiphia* and its host also was examined. Shortly after being parasitized, grubs quit feeding on grass roots and move down into the soil to depths as much as 20 cm. We speculate that *Tiphia* wasps manipulate juvenile hormone levels in parasitized grubs, causing them to prematurely descend. Hemolymph was collected from normal and parasitized grubs to test this hypothesis. Our experiments show that once parasitized, grubs no longer contribute to turf damage. Persons monitoring for parasitized grubs should look deeper in the soil where such grubs will be found.

Dilute sugar sprays were applied to small turf plots in an attempt to attract *Tiphia* wasps and increase parasitism of grubs. Although large numbers of wasps were attracted and observed feeding on the sprayed grass, no grubs were parasitized in sprayed plots. In adjacent, unsprayed turf, however, up to 37% of the grub population was parasitized. This indicates that sugar sprays applied near, but not directly on, grub-infested turf may increase the rate of parasitism. Twenty species of flowering perennials were planted and monitored to determine if they attract *Tiphia* wasps. No wasps were found on these plantings. Since sugar water sprays did not attract any wasps when applied on surrounding turf, it is likely that no wasps were present at the study site. The wildflower test will be repeated elsewhere.

PROGRESS REPORT: 2000
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OVERALL GOAL:

The long-term goal of this project is to develop a better understanding of the biology of *Tiphia* wasps, parasites of white grubs, and use this knowledge to develop methods to manage the populations of these beneficial insects on golf courses.

OBJECTIVE 1. *Investigate the biology, behavior, and importance of native and introduced species of Tiphia wasps, the predominant parasitoids attacking white grubs on golf courses.*

Two methods were tested for monitoring for *Tiphia* wasps; use of pan traps and dilute sugar water sprays. Pan traps consisting of 8oz. Solo plastic bowls filled with soapy water were used in an attempt to capture adult wasps. Five colors of bowls were used: red, purple, orange, yellow and white. Beginning in mid-April, pan traps were set out at four study sites on golf courses for 1 day each week. Yellow pan traps were most effective at collecting *Tiphia vernalis*, an introduced species that attacks Japanese beetle grubs, with catches beginning May 4. About 2 weeks after first flight, trap catches declined dramatically as wasps were found to be more attracted to oak and other flowering plant species in bloom. At this time a 10% sugar water solution sprayed on the lower foliage of oak trees was the most effective manner to monitor *T. vernalis* activity. *Tiphia vernalis* was active from May 4 to June 1. *Tiphia pygidialis*, a native species that attacks masked chafer grubs, was active from August 1 to September 14.

During the spring and fall *Tiphia* flights, masked chafer grubs were dug from several study sites to determine which instar(s) were being parasitized. Most of the grubs parasitized were in the third instar. After the fall *Tiphia* flight ended, grub digs were made at two golf courses to determine the amount of parasitism that occurred. At each site, a tape measure was stretched 10m and one $0.3m^2$ sample was taken every meter. The number of parasitized grubs and cocoons recovered from each hole was counted. This was repeated at each course for a total of 20 $0.3m^2$ samples per course. Based on this sampling method, we found that the parasitism rate of masked chafer grubs was 17% at one course and 15% at the other study site. However, work done earlier in the year showed parasitism rates as high as 37% at other study sites.

The below-ground process of host location by *Tiphia* wasps was examined. An ant farm-like observation chamber consisting of two panes of glass positioned horizontally and filled with soil was used to observe behavior of the wasps in response to certain cues. A Y-shaped trail was made in the soil, and each arm of the Y received a separate treatment. Treatments included grub body odor trails made by dragging a grub through the soil, vibrational cues made by the presence of a grub at the end of a trail, and frass collected from different species of grubs. Once treatments were applied to each arm of the Y trail, a wasp was introduced at the bottom of the Y and allowed to make a choice between the different paths. For each combination of choices, 30 wasps were tested. Wasps followed species-specific scent trails left by a grub as it moves through the soil. Vibrations made by grub movement did not elicit a response from the wasps.

Wasps responded to frass from both species of grubs tested, but a stronger response was shown for frass from their preferred host grubs.

The ovipositional behavior of each of the wasp species was characterized. The observation chamber previously described in the host location experiments was used to observe the egg laying process. A grub was placed into the chamber and a wasp then introduced from one end. A video camera was used to record each event. A total of 10 oviposition events were recorded to characterize this behavior. Upon encountering a suitable host grub in the soil, both *T. vernalis* and *T. pygidialis* deliver a paralyzing sting and then manipulate the body of the grub into a position suitable for oviposition. Then, the female laboriously scrapes the grub with the end of her abdomen to thin the cuticle where the egg will be laid. *T. vernalis* lays its egg on the underside of the grub, whereas *T. pygidialis* lays its egg on the back of its host grub. Eggs hatch in 3–5 days. Larval development is completed in 21–24 days, after which a cocoon is spun. The wasp will overwinter in this cocoon and emerge the next year as an adult.

No-choice tests were conducted with several grub species to study the range of grubs that each wasp will attack. Four ounce plastic cups were filled with sterilized, sifted soil and provisioned with two grubs of the same species. Dental wick, moistened with sugar water was provided as a food source for the wasp. A wasp was then placed into the cup and a lid placed on the cup. Ten wasps were tested against each species of grub. After 72 hours, the wasps were removed from the cups and each of the grubs examined for eggs. *T. vernalis* were tested against Japanese beetle, masked chafer, and May beetle grubs and found only to parasitize Japanese beetle grubs. *Tiphia pygidialis* were tested with the aforementioned grub species plus two additional exotic species, European chafer and Oriental beetle. In general, only masked chafer grubs were parasitized. In one case however, *T. pygidialis* parasitized and completed larval development on a third-instar European chafer.

The relationship between the developing larval *Tiphia* and its host grub was examined. Ten vertical ant farm-like chambers were used to examine the behavioral response of grubs to their developing parasite. Each ant farm was subdivided longitudinally into four chambers, filled with sifted soil and plugs of Kentucky Bluegrass (*Poa pratensis*) placed into the top to provide roots for the grubs to feed on. Each ant farm received two parasitized grubs and two non-parasitized grubs with each grub being randomly assigned to one of the four chambers. The arenas were then wrapped in black felt to darken the area with grubs and soil but to still allow the grass to grow. Every three days, the felt was removed and the depth of each grub was recorded. Measurements were made until parasitized grubs spun cocoons (~24 days). Shortly after being parasitized, grubs invariably ceased feeding on grass roots and moved down into the soil profile to depths as much as 20 cm. We speculate that *Tiphia* wasps manipulate juvenile hormone levels in parasitized grubs, causing them to prematurely descend. Hemolymph was collected from normal and parasitized grubs and frozen to test this hypothesis. Juvenile hormone analyses will be conducted this winter. Our experiments show that once parasitized, grubs no longer contribute to turf damage. Additionally, persons monitoring for the presence of parasitized grubs should look deeper in the soil where parasitized grubs will be found.

OBJECTIVE 2. *Determine the feasibility of enhancing Tiphia populations via habitat management, particularly through conservation or augmentation of wildflowers or other carbohydrate sources used by the adult wasps.*

Based on our observations that sugar water sprays attract large numbers of *Tiphia* wasps, we hypothesized that by spraying sugar water on turf would increase the rate of grub parasitism. Paired PVC rings (30 cm diam.) were driven into the ground 1 m apart. Each ring was provisioned with 20 masked chafer grubs. The turf within one ring from each pair was sprayed with ~20 ml of a 10% sugar water daily. After 7 days, the turf within each ring was destructively sampled and the grubs within each ring examined for signs of parasitism. Despite the fact that large numbers of wasps were observed feeding on the sprayed grass blades, no grubs were parasitized in the sprayed turf. In the rings not receiving sugar sprays, up to 37% of the grubs were parasitized. These results indicate that sugar sprays applied near, but not directly on grub infested turf may increase the rate of parasitism.

We also hypothesized that planting flowers used as a nectar source by *Tiphia* may lead to a localized increase in grub parasitism. As the first step in testing this hypothesis, we planted twenty kinds of flowering perennials at the U.K. Spindletop Research Farm, in a completely randomized design replicated three times with five plants per replicate. Species were selected in consultation with Sharon Bale (Dept. of Horticulture, Univ. of Kentucky) as appropriate for use in golf course landscapes. Flowers were transplanted in early June. Beginning in late July, the plants were monitored to determine if they attract *Tiphia* wasps. No wasps were found on these plantings. Since sugar water sprays did not attract any wasps when applied to turf around the garden, it is believed that no wasps were present the area. This experiment will be tried again next year in a different location.

Future Plans:

We will continue to study the basic biology of these two wasp species in 2001. Additional tests will be conducted to determine if planting of wildflowers, use of sugar sprays, or other tactics can increase the benefits that these wasps provide. Studies on the impact of turfgrass insecticides, including newer chemistries, on the performance of these wasps will be undertaken. Such information, together with our studies of the wasps' seasonal activity, may enable golf superintendents to modify timing of pest control actions so as to conserve beneficial *Tiphia* populations.

Recent Papers Supported In Part by My Previous USGA Grant

Invited Book Chapters:

1. Potter, D.A., R.C. Williamson, K.F. Haynes, and A.J. Powell, Jr. 2000. Cultural control, risk assessment, and environmentally responsible management of scarab grubs and cutworms in turfgrass. pp. 383–396 in: J.M. Clark and and M.P. Kenna (eds.). Fate and management of turfgrass chemicals. American Chemical Society, ACS Series 743. Washington, D.C.
2. Potter, D.A. 2000. The Japanese beetle. CAB International Global Crop Protection Compendium, Wallington, UK (electronic, published on CD)
3. Potter, D.A. and D.W. Held. 2001. June Beetles (*Phyllophaga* spp.) Encyclopedia of Insects, Academic Press. *In Press*.
4. Held, D.W. and D.A. Potter. 2001. The Japanese beetle. Encyclopedia of Insects; Academic Press. *In Press*.

Refereed Journal Articles

1. Kunkel, B.A., D.W. Held, and D.A. Potter. 1999. Impact of halofenozide, imidacloprid, and bediocarb on beneficial invertebrates and predatory activity in turfgrass. J. Econ. Entomol. 92: 922–930.
2. Lopez, R. and D.A. Potter. 2000. Ant predation on eggs and larvae of the black cutworm (Lepidoptera: Noctuidae) and Japanese beetle (Coleoptera: Scarabaeidae) in turfgrass. Environ. Entomol. 29: 116–125.
3. Lopez, R., D.W. Held, and D.A. Potter. 2000. Management of a mound-building ant, *Lasius neoniger* Emery, on golf putting greens and tees using delayed-action baits or fipronil. Crop Science 40: 511–517.
4. Bauerfeind, R.J., K.F. Haynes, and D.A. Potter. 2000. Responses of three *Cyclocephala* (Coleoptera: Scarabaeidae) s[ecies to hexane extracts of *Cyclocephala lurida* sex pheromone. J. Kansas Entomol. Soc. 72: 246–247.
5. Kunkel, B.A., D.W. Held, and D. A. Potter. 2000. Lethal and sublethal effects of bendiocarb, halofenozide, and imidacloprid on *Harpalus pennsylvanicus* DeGeer (Coleoptera: Carabidae) following different modes of exposure in turfgrass. J. Econ. Entomol. *In Press*.
6. Walston, A. T., D. W. Held, N. R. Mason, and D. A. Potter. 2000. Absence of interaction between endophytic perennial ryegrass and susceptibility of Japanese beetle grubs to *Paenibacillus popilliae*. J. Entomol. Sci. *In Press*.
7. Potter, D.A. and D.W. Held. 2001. Biology and management of the Japanese beetle. Annual Review of Entomology. Invited, In progress. Manuscript due in early 2001.

Invited Technical Articles in National Trade Journals (all originate from my laboratory):

1. Potter, D.A. 1999. Novel chemicals provide new grub control options. Grounds Maintenance 34(4): 12–17. (Full-Cover Feature Article)
2. Potter, D.A. and R.Lopez. 1999. Baits target nuisance ants. Golf Course Management August 1999: 64–67.
3. Potter, D.A. and M.F. Potter. 1999. Managing biting and stinging pests on golf courses. September 1999: 53–58.