Biology of *Tiphia pygidialis* (Hymenoptera: Tiphiidae), a Parasitoid of Masked Chafer (Coleoptera: Scarabaeidae) Grubs, with Notes on the Seasonal Occurrence of *Tiphia vernalis* in Kentucky

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ABSTRACT We investigated the biology of *Tiphia pygidialis* Allen, a previously unstudied native parasitoid of masked chafer, *Cyclocephala* spp. grubs, in central Kentucky and the seasonal dynamics of *Tiphia vernalis* Rohwer, an introduced parasitoid of Japanese beetle, *Popillia japonica* Newman. *T. pygidialis* was active from mid-August to early October, parasitizing third-instar masked chafers, whereas *T. vernalis* attacked overwintered third-instar *P. japonica* from mid-April to early June. Adult *T. vernalis* were attracted to modified Japanese beetle traps and yellow pan traps and to 10% sugar water sprayed on tree foliage. Spraying sugar water directly on turf most effectively monitored *T. pygidialis* wasps. Parasitism rates as high as 33 and 58% were observed for *T. pygidialis* and *T. vernalis*, respectively. In the laboratory, *T. pygidialis* larvae progressed through five instars to cocoon formation in ≈22 d. They overwinter as prepupae. Field-collected female wasps lived 32 ± 4 d, parasitizing 22 ± 6 grubs. In no-choice tests with eight species of native and exotic white grubs, *T. pygidialis* readily parasitized only *Cyclocephala* spp., including *C. lurida* Bland and *C. borealis* Arrow, which it normally encounters in Kentucky, but also *C. pasadenae* Casey, a western species not know to occur within the wasp's geographic range. Wasps did not discriminate between nematode-infected and healthy grubs, indicating potential for interference between these biological control agents.

KEY WORDS biological control, Scarabaeidae, parasitoid, turfgrass, host selection

WASPS IN THE GENUS *Tiphia* (Hymenoptera: Tiphiidae) are the predominant parasitoid taxa that attack root-feeding white grubs (Coleoptera: Scarabaeidae) (Clausen 1940). Approximately 100 species are present in North America (Krombein 1979), yet few studies have been conducted on their biology. Female *Tiphia* wasps, ≈1 cm long (Fig. 1A), burrow into the soil and locate subterranean hosts using species-specific kairomones present in grub body odor trails and frass (Rogers and Potter 2002). As far as is known, most *Tiphia* are host-specific, although some species may attack several congeneric grub species (Jaynes and Gardner 1924). Once a host is located, the wasp stings it, causing temporary paralysis. An egg is attached to the grub in a location that is species-specific (Clausen et al. 1927). The *Tiphia* larva feeds externally during its first four instars by piercing the grub's integument and imbibing host body fluids. During the fifth instar, the larval *Tiphia* devours all but the sclerotized portions of the grub (Fig. 1D) and then spins a silken cocoon (Fig. 1Ee) in which it overwinters, emerging the following year.

Most of what is known of the biology of these wasps comes from work with exotic species imported for release against the Japanese beetle, *Popillia japonica* Newman, during the 1920–1930s (Clausen 1978). At least seven species were released in the United States, but only two, *Tiphia popilliavora* Rohwer and *Tiphia vernalis* Rohwer, were known to establish and provide some control of *P. japonica.* Since the 1960s, almost nothing has been published on the spread of these introduced *Tiphia* and their contribution in controlling grub populations (Ladd and McCabe 1966, Clausen 1978).

In Kentucky, we have observed two species of *Tiphia* on golf courses and other turf sites. *Tiphia pygidialis* Allen, a native wasp, is active in late summer. Its known geographic distribution includes Connecticut, Maryland, Pennsylvania, West Virginia, Kentucky, Iowa, Kansas, Oklahoma, and Mississippi (Krombein 1979). Before our studies, no information on its biology, including host species, was known. We have also found *T. pygidialis* parasitizing third-instar masked chafers, *Cyclocephala* spp., during August and September. Since its introduction in 1924 (Clausen et al. 1933), *T. vernalis* has spread to many parts of the United States where *P. japonica* has become established (Clausen 1978). Although not previously documented in Kentucky, we have found *T. vernalis* to be locally abundant on golf courses and other turf sites in northern and central Kentucky, parasitizing overwin-

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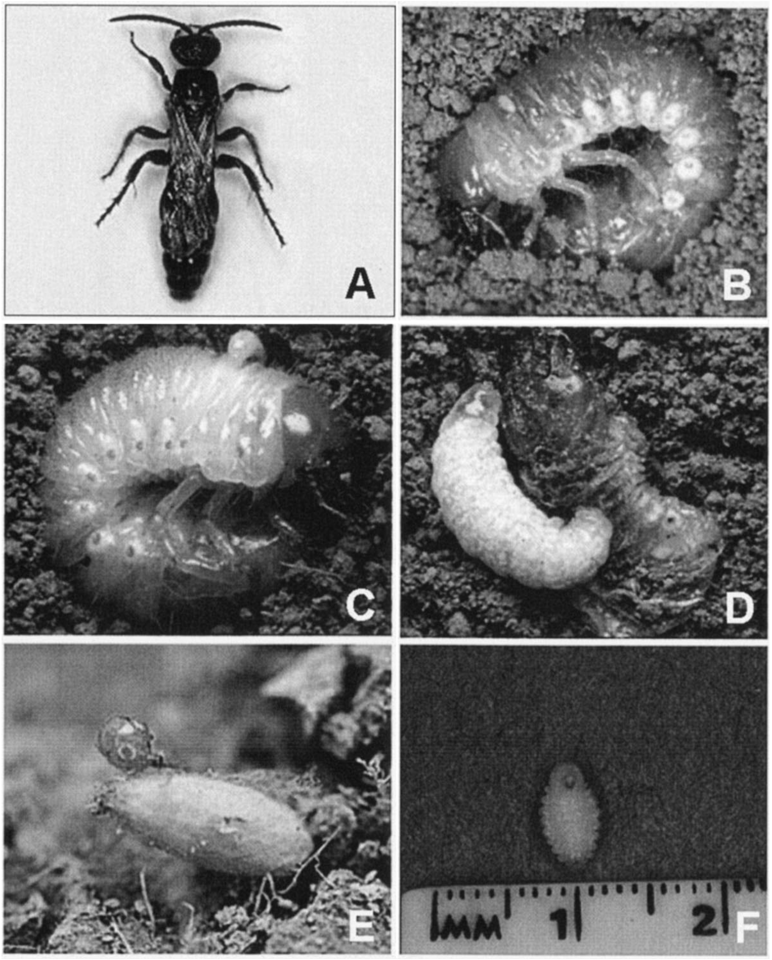

Fig. 1. Life stages of *T. pygidialis* on its *Cyclocephala* sp. host. (A) Adult female. (B) Egg attached dorsally between second and third thoracic segments of third-instar grub. (C) Third-instar *T. pygidialis* feeding on host grub. (D) Host grub being consumed by fifth-instar *T. pygidialis*. (E) Cocoon with remains of host head capsule attached. (F) Prepupa (overwintering stage) removed from cocoon.

tered third-instar *P. japonica* in the spring (Rogers and Potter 2002, 2003, 2004).

The objective of this study was to examine the biology of *T. pygidialis* in both the field and laboratory. We examined the adult flight period, evaluated methods to monitor wasp activity, and surveyed grub parasitism rates on golf courses in central Kentucky. We also examined the development of *T. pygidialis* larvae on their host grubs, longevity and fecundity of adult females, and factors affecting host selection and host range. Information on the seasonal biology and abundance of *T. vernalis* in Kentucky also was obtained.

Materials and Methods

Flight periods of *Tiphia* spp. in Kentucky. We determined the flight periods of *T. pygidialis* and *T. vernalis* by carefully searching for wasps visiting sugar sprays to feed. Beginning in early April, a 10% sugar water solution was sprayed on the foliage of trees, low-growing vegetation, and directly on 1-m^2 patches of turfgrass at two golf courses in central Kentucky and at the University of Kentucky Spindletop research farm every 7 d. The sprayed vegetation was carefully observed for 1 h for *Tiphia* feeding. Monitoring ended in mid-June, during host beetle flight, when grubs were no longer present, and resumed in early August, when second and third instars were again present. A hand-held vacuum (BioQuip, Rancho Dominguez, CA) was used to collect all wasps feeding on the sugar-sprayed vegetation, and species identification was confirmed by K. R. Ahlstrom (North Carolina Department of Agriculture, Raleigh, NC). Voucher specimens are deposited in the University of Kentucky Insect Collection.

Because of the large amount of time needed to monitor *Tiphia* flight using sugar water sprays, we evaluated two less time-consuming techniques. The first was the use of pan traps consisting of 355-ml plastic bowls (Solo, Urbana, IL) filled to the top with

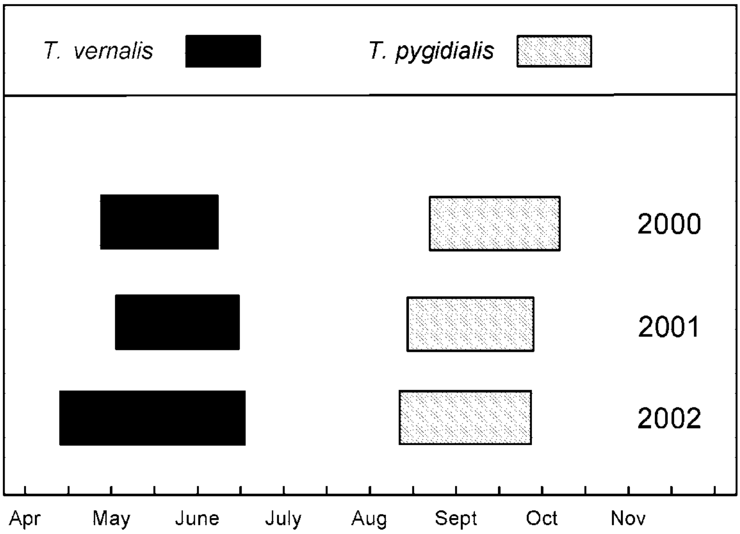

Fig. 2. Flight period of *T. vernalis* and *T. pygidialis* as indicated by their presence or absence at sugar water–sprayed vegetation during 3 consecutive yr in Kentucky.

water and ≈5 ml of liquid soap to reduce surface tension. We evaluated five colors of bowls: orange, purple, red, white, and yellow. Rows of five different colored pan traps were placed in grub-infested roughs at Andover Golf Course, Lexington, KY; Kearney Hills Golf Course, Lexington, KY; and the University of Kentucky Spindletop research farm once each week for 24 h during wasp flight of *T. vernalis* and *T. pygidialis* in spring and late summer, respectively. Each pan trap was separated by 1 m with four sets of five traps per study site. Seasonal totals for each color were pooled within sites. The number of wasps collected in each of the five colors of traps was compared by two-way analysis of variance (ANOVA), using sites as replicates, followed by means separation using Tukey's honest significant difference (HSD) test (Analytical Software 2000).

Pan traps interfered with mowing, especially at the golf course sites. We therefore examined whether modified Japanese beetle traps could be used to monitor *Tiphia* activity. Rationale for these tests was based on our observation that yellow pan traps were attractive, as well as serendipitous capture of *T. vernalis* in Japanese beetle traps being used in an unrelated study. Metal Japanese beetle trap tops (Ellisco-Philadelphia Can Co., Philadelphia, PA), freshly painted bright yellow, were used with a 473-ml glass mason jar (Kerr Group, Indianapolis, IN) screwed onto the trap top. The jars were used because the standard trap bottoms have holes through which the wasps could escape. The traps were hung ≈1 m above ground from metal posts. Four traps were hung in a row 10 m apart. Four rows of traps were set up in the roughs at the Pendleton County Country Club Golf Course, Pendleton Co., KY, and operated continuously from 1 April 2002 until flight of *T. vernalis* was over in June. To monitor for *T. pygidialis*, similar sets of traps were placed in stands of turf at Spindletop research farm where sugar water sprays had confirmed that wasps were present. Those traps were monitored from 1 August until 1 October 2002. The traps were emptied every 15 d, and the number and species of *Tiphia* captured were determined.

Grub Parasitism Rates. Northern and southern masked chafers, *Cyclocephala borealis* Arrow and *C. lurida* Bland, respectively, are abundant in Kentucky (Potter 1980), but their grubs are indistinguishable by known morphological characters. We therefore refer to them collectively as *Cyclocephala* spp. Parasitism of third-instar *Cyclocephala* spp. by *T. pygidialis* and of *P. japonica* by *T. vernalis* was surveyed at Kentucky golf courses in fall and spring, respectively, after flight of the wasps had ended. Parasitism of *Cyclocephala* spp. was sampled at Kearney Hills Golf Course and Lakeside Golf Course, both in Lexington, KY, in early October 2000 and 2002. A third golf course, The Bull in Richmond, KY, was sampled in October 2002. Parasitism rates of *P. japonica* were sampled at the Pendleton County Country Club Golf Course in mid-June 2001 and 2002 and at Spindletop research farm in 2003. At each golf course study site, 10 (0.3 m^2) turf samples, each separated by 3 m, were taken from the bluegrass, *Poa pratensis* L., roughs along four fairways, and all grubs and *Tiphia* cocoons were collected. The turf was excavated to a depth of 30 cm to account for characteristic movement of parasitized grubs deeper into the soil (Rogers et al. 2003). Similar samples taken at Spindletop research farm were collected from stands of bluegrass and tall fescue, *Festuca arundinaceae* Schreber. All grubs were examined for *Tiphia* eggs or larvae to estimate the percentage parasitism occurring at each site. Simple linear regression was used to determine the effect of grub density on parasitism rate (Analytical Software 2000).

Longevity and Fecundity of *T. pygidialis*. Because *T. pygidialis* are univoltine and difficult to overwinter in

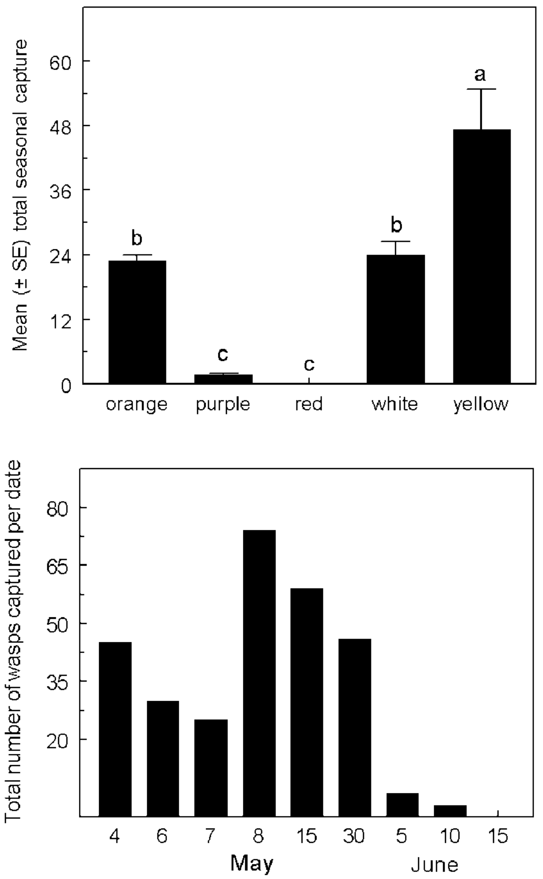

Fig. 3. *Tiphia vernalis* adults captured in colored pan traps placed in the roughs of two central Kentucky golf courses and the University of Kentucky Spindletop Research farm during 2000. (Top) Mean ± SE total number of wasps per site collected in each color pan trap. Values with the same letter are not significantly different (Tukey's HSD, $P > 0.05$). (Bottom) Total number of wasps collected on each date.

the laboratory, we examined longevity and fecundity of adult females collected in the field near the beginning of wasp flight in mid-August. Fifty field-collected female wasps were confined in individual 118-ml plastic cups half-filled with autoclaved soil. A film canister lid containing a piece of dental wick soaked in a 10% sugar water solution was provided on the surface of the soil as a food source for each wasp. One field-collected third-instar *Cyclocephala* sp. was provided each day for oviposition. We recorded wasp survival and whether the grub was parasitized daily. Parasitized grubs were replaced with nonparasitized ones as needed.

We also determined the number of grubs that a female can parasitize in a single day. Thirty females were provided sugar water but deprived of hosts for 7 d. The wasps were then confined in individual plastic containers (15 cm diameter by 6.5 cm height) half-filled with moist autoclaved soil. Each container was provisioned with 20 third-instar *Cyclocephala* spp. A sugar water–soaked dental wick was provided as before. Each day for 14 d, the grubs in each container were checked for *T. pygidialis* eggs, and the number that had been parasitized was recorded. Parasitized hosts were removed, and nonparasitized ones were added to maintain 20 grubs per container.

Development of Larval *T. pygidialis*. The development of *T. pygidialis* was examined on >200 parasitized *Cyclocephala* spp. grubs in the laboratory. Parasitized grubs bearing a *T. pygidialis* egg were obtained by confining female wasps with one third-instar *Cyclocephala* sp. in 118-ml plastic cups half-filled with autoclaved soil. Wasps were provided 10% sugar water as before. Each day, parasitized grubs were removed from the containers, placed into separate cups of autoclaved soil, and held at 22–24°C. Every 3 d, each parasitized grub was removed from its cup, and the head capsule width of the *T. pygidialis* larva was measured with an ocular micrometer to determine the number of instars and time required to reach each instar.

Host Selection by *T. pygidialis*. Host range of *T. pygidialis* was tested with eight species of turf-infesting scarabaeid grubs in five genera. These included third instars of five species that commonly occur with *T. pygidialis* in Kentucky: *C. borealis; C. lurida; P. japonica;* May beetle, *Phyllophaga* sp.; and green June beetle, *Cotinis nitida* L.; and three species that do not occur in Kentucky, including southwestern masked chafer, *Cyclocephala pasadenae* Casey; oriental beetle, *Exomala orientalis* (Waterhouse); and European chafer, *Rhizotrogus majalis* (Razoumowsky). To be certain of their identity, *C. borealis* and *C. lurida* were reared from eggs laid by field-collected females, which, unlike the grubs, can be readily separated (Potter 1980). The other Kentucky species were collected from turf. The remaining species were field-collected by colleagues in California, New York, or New Jersey and shipped overnight in soil.

For each of the eight grub species tested, 10 female *T. pygidialis* were confined individually in 118-ml plastic cups provisioned with a grub and half-filled with autoclaved soil. Sugar water solution (10%) was provided as before. Grubs were checked daily for 7 d for parasitoid eggs, and nonparasitized grubs were provided as needed.

A second experiment was conducted to determine if *T. pygidialis* would discriminate between the field-collected *Cyclocephala* spp. grubs it normally encounters in Kentucky (i.e., mixed populations of *C. borealis* and *C. lurida*) and a congeneric species, *C. pasadenae*, which is not found in Kentucky. Choice tests were conducted by confining 20 female *T. pygidialis* individually in 118-ml containers of soil with one locally collected third-instar *Cyclocephala* sp. and one third-instar *C. pasadenae.* Because those *Cyclocephala* grubs are difficult to separate morphologically, either the right or left antenna was removed from the respective grubs before they were confined together. Each day, over the next 5 d, the grubs were removed from the containers and checked for parasitism, with additional nonparasitized grubs provided to replace parasitized

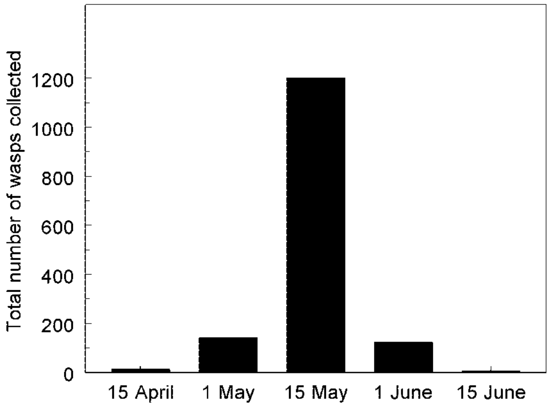

Fig. 4. Total number of *T. vernalis* adults captured in modified Japanese beetle traps elevated 1 m above the ground at a golf course in central Kentucky, 2002.

ones. Wasps' choice of hosts for oviposition was analyzed using the χ^2 test against the null hypothesis of a 1:1 ratio (Snedecor and Cochran 1991).

The effect of host instar on wasp oviposition choice was also examined. Twenty female *T. pygidialis* were confined in plastic containers (15 cm diameter by 6.5 cm height) of autoclaved soil with five second-instar and five third-instar *Cyclocephala* sp. Food (10% sugar water) was provided as before. After 48 h, the grubs were checked for parasitism. Wasps' choice of hosts based on instar of development was analyzed using the χ^2 test as before.

The effect of host health on a wasp's oviposition choice, or more specifically, whether wasps would parasitize hosts infected with an entomopathogenic nematode, was tested in both choice and no-choice tests. Ten female *T. pygidialis* were confined in individual plastic containers (15 cm diameter by 6.5 cm height) of autoclaved soil as previously described. Ten *Cyclocephala* spp. grubs that had been visibly infected in the laboratory by *Heterorhabditis bacteriophora* Poinar HP88 strain were placed into each container. Grubs were examined for parasitism after 3 d. In a second experiment, 10 wasps were each confined with 10 *H. bacteriophora*–infected third-instar *Cyclocephala* spp. and 10 healthy third-instar *Cyclocephala* spp. grubs. After 3 d, the grubs were removed to check for parasitism. Wasps' preference for healthy versus nematode-infected hosts was analyzed using the χ^2 test as before.

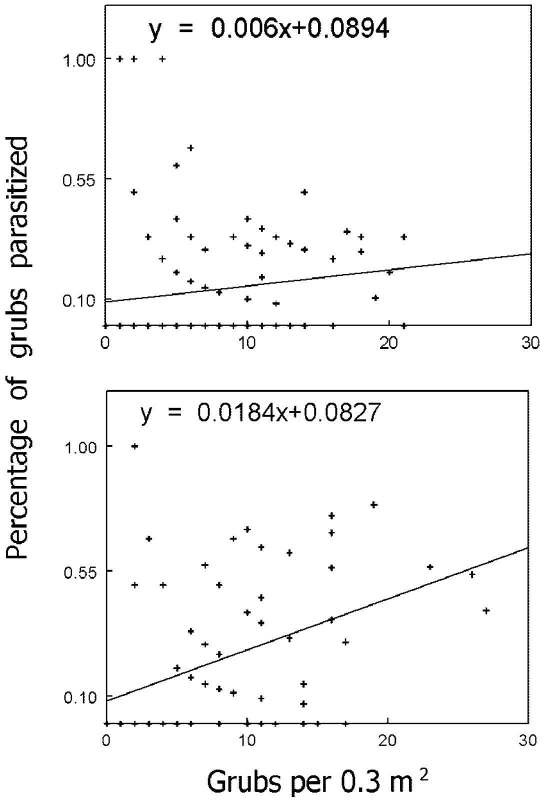

Fig. 5. Percentage of grubs parasitized versus grub density. (Top) *Cyclocephala* spp. grubs parasitized by *T. pygidialis* based on ten 0.3-m^2, 30-cm-deep turf samples examined in each of four roughs at five golf courses ($F = 2.6$; df = 1,199; $P = 0.11$; $R^2 = 0.01$). (Bottom) *Popillia japonica* grubs parasitized by *T. vernalis* based on ten 0.3-m^2, 30-cm-deep turf samples examined in each of four roughs at two golf courses ($F = 19.23$; df = 1,79; $P < 0.0001$; $R^2 = 0.19$).

Table 1. Rates of parasitism of masked chafer (*Cyclocephala* spp.) and Japanese beetle (*P. japonica*) grubs at central Kentucky golf courses based on ten 0.3-m^2 30-cm-deep turf samples examined in each of four roughs at each course in each year

Site	Year	Total no. of grubs	Parasitized grubs	Percent parasitism
Cyclocephala spp. grubs parasitized by *T. pygidialis* in autumn				
Kearney	2000	347	66	19.0
Lakeside	2000	184	26	14.1
Keamey	2002	54	18	33.3
Lakeside	2002	118	28	23.7
The Bull	2002	290	5	1.7
P. japonica grubs parasitized by *T. vernalis* in spring				
Pendleton	2001	230	82	35.6
Pendleton	2002	176	102	58.0
Spindletop	2003	146	21	14.4

Results

Flight periods of *Tiphia* spp. in Kentucky. The flight period of *T. vernalis* and *T. pygidialis* in Kentucky

Table 2. Development of *T. pygidialis* on third-instar *Cyclocephala* spp. in the laboratory at 22°C

Instar	*n*	Head capsule measurements (mm)		Time (d) to reach instar	
		Median	Range	Median	Maximum
1	221	0.25	0.2–0.3	6	2–10
2	208	0.35	0.3–0.45	11	6–15
3	120	0.5	0.35–0.5	14	11–17
4	87	0.75	0.7–0.75	17	15–21
5	52	0.9	0.85–1.0	21	17–25
Cocoon	33	—	—	22	19–27

varied slightly over the 3 yr of surveying (Fig. 2). Based on the presence or absence of wasps visiting sugar water sprays applied to tree foliage, we determined *T. vernalis* adults were active from mid-April to mid-June. *T. pygidialis* were attracted to sugar water sprays applied directly to turfgrass from mid-August to early October.

Relatively few *T. vernalis* were collected in the pan traps (Fig. 3). Yellow pan traps were most effective ($F = 26.67$; df = 4,14; $P = 0.0001$; Fig. 3). During early April, a few wasps identified as *T. letalis* Roberts also were collected in the yellow pan traps. The host species of *T. letalis* is unknown (Krombein 1979). Large numbers of *T. vernalis* were collected in the modified Japanese beetle traps (Fig. 4). During the 15-d period of peak wasp flight, >1,000 male and female *T. vernalis* were collected in these traps.

Pan traps were not effective for collecting *T. pygidialis.* In fact, during August and September 1999 and 2000, only one *T. pygidialis* female was collected in any of the pan traps at our study sites. Sugar water sprays applied directly to the turfgrass near these pan traps attracted hundreds of wasps, confirming presence of *T. pygidialis* in the area. No *T. pygidialis* were collected in the modified Japanese beetle traps during the late summer.

Grub Parasitism Rates. Parasitism rates of *P. japonica* by *T. vernalis* at our study site in Pendleton county were 35.6 and 58% when sampled in mid-June 2001 and 2002, respectively, and 14.4% at the University of Kentucky Spindletop farm in 2003 (Table 1). Parasitism within the individual roughs sampled at the Pendleton site ranged from 2 to 48% in 2001 and 40 to 80% in 2002. Parasitism of *Cyclocephala* spp. by *T. pygidialis* ranged from 14 to 33% at two Lexington area golf courses in 2000 and 2002, whereas only 1.7% of the *Cyclocephala* spp. population was parasitized at a third golf course in 2002 (Table 1). Parasitism of *Cyclocephala* spp. also varied between individual sampled roughs, ranging from 9 to 41% at the Kearney and Lakeside sites. There was a significant positive relationship between grub density and percentage of *P. japonica* parasitized by *T. vernalis* ($F = 19.23$; df = 1,79; $P < 0.0001$; Fig. 5) and a trend for higher parasitism rate of *Cyclocephala* spp. by *T. pygidialis* at higher grub densities ($F = 2.6$; df = 1,199; $P = 0.11$; Fig. 5).

Longevity and Fecundity of *T. pygidialis*. Field-collected female *T. pygidialis* held individually and provided one host *Cyclocephala* spp. grub per day survived an average of 32 ± 3.8 d (range, 16–44 d) and oviposited an average of 22 ± 6.3 eggs. Several females deposited multiple eggs on a host, laying >50 eggs over their lifespan in the laboratory. Wasps previously deprived of hosts for 7 d and then confined with 20 third-instar *Cyclocephala* spp. parasitized an average of 3 ± 0.6 grubs/d. During the first few days of being provided hosts, some wasps parasitized as many as seven grubs in 24 h. However, this was usually followed by a 1- to 2-d hiatus in egg-laying, after which the wasps resumed ovipositing only on 1–2 hosts of the 20 available each day for the remainder of the 14-d test.

Development of Larval *T. pygidialis*. Eggs of *T. pygidialis* were deposited dorsally on third-instar *Cyclocephala* spp. between the second and third thoracic segments (Fig. 1B). On eclosion, which occurred 6 ± 0.2 d after oviposition, the first-instar *T. pygidialis* pierces the integument of its host and begins feeding on hemolymph. Based on head capsule measurements, the larva progresses to the fifth instar in ≈21 d (Table 2). By then, the host grub has become flaccid (Fig. 1D). The wasp larva devours all but the sclerotized portions of its host before forming a cocoon (Fig. 1E). The parasitoid overwinters as a prepupa in the cocoon (Fig. 1F), developing to the adult stage the following spring.

Host Selection. In no-choice tests wherein 80 females were individually confined for 7 d with one of the eight grub species, *T. pygidialis* readily oviposited on all three *Cyclocephala* spp. including *C. pasadenae,* which does not occur in the wasp's known geographic range. For wasps confined with the other species of grubs, three eggs were laid on *R. majalis,* two eggs were laid on *E. orientalis,* and none were found on *C. nitida* or *Phyllophaga* spp. grubs.

When female *T. pygidialis* were given a choice between ovipositing on its normal hosts (either *C. borealis* or *C. lurida*) and *C. pasadenae,* there was no significant difference in host preference. Total numbers of eggs laid on Kentucky *Cyclocephala* spp. grubs versus *C. pasadenae* were 46 and 54, respectively ($\chi^2 = 0.64$, df = 1, $P = 0.42$). In choice tests with second- and third-instar grubs, female *T. pygidialis* showed no preference for oviposition (38 versus 46 total eggs laid, respectively; $\chi^2 = 0.91$, df = 1, $P = 0.34$).

In no-choice tests, *T. pygidialis* readily parasitized grubs that were visibly infected with the nematode *H. bacteriophora.* When given a choice, the wasps did not discriminate in oviposition on healthy versus nematode-infected hosts (22 versus 16 eggs, respectively; $\chi^2 = 1.11$, df = 1, $P = 0.29$), despite the fact that some infected grubs were in the final stages of nematode infection and had become flaccid.

Discussion

Adult activity of the two primary *Tiphia* species found on central Kentucky golf courses corresponded to the presence of third-instar hosts, the stage that most often is found parasitized (Jaynes and Gardner 1924). The flight period of spring-active *T. vernalis* coincided with resumption of root feeding by overwintered *P. japonica* and lasted until the grubs began to pupate in early June. Likewise, *T. pygidialis* emerged when early third-instar *Cyclocephala* spp. grubs were present in early to mid-August and were active until late September or early October.

Monitoring techniques differed in effectiveness for the two species of *Tiphia. T. vernalis* were only found feeding on sugar water sprays applied to tree foliage, whereas *T. pygidialis* only visited sprays applied directly to turfgrass. This difference likely is because of differences in the species' mating or feeding habits. *T. vernalis* reportedly feed primarily on aphid honeydew on the foliage of trees (Clausen et al. 1927, 1932). Indeed, we have observed hundreds of *T. vernalis* visiting aphid-infested leaves of oaks (*Quercus* spp.), maples (*Acer* spp.), and other woody plants, and also mating in such trees. We also have observed them at infestations of soft scales (Coccidae) and nectar-producing flowers of peony, *Peonia lactiflora* Pallas (Rogers and Potter 2004). *T. pygidialis* was never observed to feed and mate on the foliage of trees, but instead, we observed them mating directly on the turf. Wasps feeding at sugar sprayed vegetation likely are arrested by the food rather than being attracted from a distance (Rogers and Potter 2004). Female *T. pygidialis* may be arrested to feed on sugar sprayed turf as they search for grubs on which to oviposit.

Such differences in behavior should be considered when monitoring *Tiphia* wasp activity. Yellow pan traps placed on the turf collected far fewer *T. vernalis* than did modified Japanese beetle traps hung 1 m above ground level. The latter were probably more visible, resulting in the greater capture. The reason *T. pygidialis* were not captured using either pan traps or modified Japanese beetle traps is not known. One possibility is that *T. pygidialis* do not travel far to feed compared with *T. vernalis*, which travel to areas with trees to mate and feed. Thus, our traps may have been intercepting *T. vernalis* flying between mating or feeding sites and oviposition sites. Failure to collect *T. pygidialis* in the traps was likely not caused by low wasp numbers, because at the same time, numerous wasps were observed feeding on sugar water sprays applied to nearby turf.

Parasitism rates of *P. japonica* at the Pendleton County golf course were 35 and 58% in June 2001 and 2002, respectively. Those rates are similar to those determined by King and Parker (1950), who observed parasitism by *T. vernalis* ranging from 19 to 67%. Parasitism of third-instar *Cyclocephala* spp. ranged from 14 to 33% at two golf courses but was only 1.7% at a third course. Food availability for adult wasps may be one factor affecting parasitism rates at our study sites. The Pendleton course, in rural north central Kentucky, has fairways and roughs surrounded by mature trees and woodlots, which provide ample food sources (i.e., honeydew) for *T. vernalis.* Absence of food sources in the form of honeydew and nectar has been suggested as the reason *Tiphia* spp. often have failed to establish (Gardner 1938, Clausen 1978). Access to carbohydrate food greatly enhances the longevity of *T. pygidialis* and *T. vernalis* and can increase parasitism at varying distances from the food source (Gardner 1938, Rogers and Potter 2004). In this study, field-collected *T. pygidialis* lived an average of 32 d when provided with food and hosts in the laboratory. Some wasps survived >40 d. In a related study, wasps provided sugar water lived six times longer than those receiving only distilled water (Rogers and Potter 2004).

Insecticide usage may also affect parasitism rates (e.g., Cockfield and Potter 1984, Kunkel et al. 1999). For example, imidacloprid, a long-residual grub insecticide, has sublethal effects on *T. vernalis*, reducing parasitism rates by inhibiting the wasps' ability to locate hosts while in the soil (Rogers and Potter 2003). It is unclear what effect, if any, past or present insecticide applications may have had on *Tiphia* populations and grub parasitism rates at our study sites.

When confined with different grub species in no-choice tests, *T. pygidialis* parasitized only species of the genus *Cyclocephala,* including *C. pasadenae,* which is not known to exist within the wasp's geographic range. Earlier work with introduced species of *Tiphia* also suggested that they are relatively host-specific, parasitizing grubs within one or a few closely related genera (Clausen et al. 1927). *Tiphia* spp. use species-specific cues from grub body odor trails and frass to locate their hosts in the soil (Rogers and Potter 2002). Undoubtedly, use of such host-specific cues limits *Tiphia* spp. to encounters with only those grub species used a hosts.

Female *T. pygidialis* also did not discriminate between second- and third-instar hosts. Brunson (1938), in contrast, showed that the introduced *T. popilliavora* prefer to oviposit on third- rather than second-instar *P. japonica.* Eggs of that species laid on second instars developed into males, whereas those laid on third instars developed into female wasps (Brunson 1938). Because it is difficult to overwinter wasps in the laboratory, we did not determine whether *T. pygidialis* determines the sex of its offspring based on host instar.

Tiphia pygidialis readily oviposited on hosts infected with the nematode *H. bacteriophora* in both choice and no-choice tests. These nematode-infected grubs died before eclosion of the *T. pygidialis* eggs, resulting in mortality of the parasitoid. Similarly, White (1943) found that *T. vernalis* and *T. popilliavora* developed on third-instar *P. japonica* infected with *Paenibacillus* (=*Bacillus*) *popilliae* Dutky; however, the parasitized hosts may succumb to the pathogens before the larval *Tiphia* has completed its development. Thus, the presence of nematodes and bacterial pathogens may have indirect detrimental effects on *Tiphia* populations.

Because both *Tiphia* species and their hosts have only one generation per year, commercial development of the wasps for biological control is unlikely. However, the benefits provided by resident populations of our native and introduced species of *Tiphia* have largely been overlooked in the recent decades of reliance on chemical control of scarab larvae. As turf insecticides become more target selective, it may be easier to conserve or enhance *Tiphia* populations in home lawns and on golf courses. For example, *T. vernalis* might be recruited to home landscapes by incorporating plants with honeydew or nectar as food. More studies are needed in other regions of the United States to determine which *Tiphia* spp. are present and the extent to which they suppress white grub populations.

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