Behavior of *Telenomus podisi* (Hymenoptera: Platygastridae) Adults under Overwintering Conditions¹

Sriyanka Lahiri, David Orr², Clyde Sorenson, and Yasmin J. Cardoza

Department of Entomology and Plant Pathology, North Carolina State University, Campus Box 7613, Raleigh, North Carolina 27695 USA

Abstract To assess overwintering refuge preferences by *Telenomus podisi* Ashmead, artificial refuges of varying geometries and composition were provided to wasps when exposed to overwintering conditions in an environmental chamber. Field sampling of leaf-litter and tree bark was also used in an effort to determine site preference of overwintering wasps. Under artificial overwintering conditions, wasps preferred to hang inverted while quiescent, regardless of the refuge design, indicating behavior that avoids precipitation or extreme temperature fluctuations during overwintering in field conditions. Wasps preferred refuges with wider gaps between upper and lower surfaces, avoiding spaces that were narrower than their standing height. Parasitoids also preferred settling at least 60 mm from refuge edges. Results suggest that *T. podisi* has a preference for the type of overwintering refuge that leaf-litter may offer.

Key Words Platygastridae, overwintering, biocontrol

Insect dormancy has been defined as a physiological condition of growth retardation or arrest, primarily designed to overcome low temperatures (Mansingh 1971) or a seasonally recurring period in the life cycle of an organism during which growth, development, and reproduction are suppressed (Tauber et al. 1986). Southern insect populations show different cryo-protective adaptations compared with populations in the northern United States and Canada (Baust 1981) due to higher levels of glycerol accumulation in northern species (Baust and Lee 1982). Low temperature induces overwintering behavior in parasitoids, but it does not appear to induce a reproductive diapause for some parasitoids in climatically warmer areas such as the southeastern United States. In these climatic situations, parasitoids may take refuge during cold weather, but still be able to parasitize any hosts that may be available during winter. Trichogramma exiguum Pinto and Platner (Hymenoptera: Trichogrammatidae) were reported to be active in winter with emergence and oviposition observed even at 9°C in North Carolina (Keller 1986). Due to sunlight exposure, overwintering Trissolcus biproruli Girault (Hymenoptera: Platygastridae) has been reported to oviposit in southern New South Wales, Australia, in winter as the black body color helps the overwintering wasps to capture sunlight and increase body temperature as necessary for oviposition (James 1988).

J. Entomol. Sci. 52(1): 15-28 (January 2017)

¹Received 07 March 2016; accepted for publication 13 June 2016.

²Corresponding author (email: dorr@ncsu.edu).

Progeny of *Telenomus californicus* Ashmead (Hymenoptera: Platygastridae) (egg parasitoid of Douglas-fir tussock moth, *Orgyia pseudotsugata* McDunnough), from both fall and spring ovipositions emerge in late summer and overwinter as fertilized females (Torgersen and Ryan 1981). *Telenomus podisi* (Hymenoptera: Platygastridae) overwinter as adults in subtropical regions and, under natural conditions, adult longevity and fecundity decrease as the temperature experienced by immatures decreases (Doetzer and Foerster 2007). It is clear that platygastrids mostly overwinter as adults (Austin 1984, James 1988, Torgersen and Ryan 1981) with one exception: *Scelio bipartitus* Kieffer overwinters either as pharate adults or as larvae inside orthopteran host eggs (Baker and Dysart 1992).

In the southeastern United States, *T. podisi* appears to be the predominant egg parasitoid of several agricultural pest stink bugs, including *Nezara viridula* L., *Oebalus pugnax pugnax* F., and *Euschistus servus* Say (Hemiptera: Pentatomidae) (Tillman 2010). Understanding critical life-cycle components, like overwintering behavior, may benefit conservation biocontrol efforts utilizing both native parasitoids such as *T. podisi* and imported species targeting stink bugs and relatives such as kudzu bug, *Megacopta cribraria* F. (Hemiptera: Plataspidae). Studies highlighting the importance of parasitoid–host life-history synchrony point to the importance of overwintering survival of native and introduced parasitoids, depending on the local climate, if they are to establish and function as effective biological control agents (Al-Ghamdi et al. 1995, Babendreier et al. 2003, Coombs 2004, Hance et al. 2007).

Although many studies have investigated the cold hardiness of parasitoids (e.g., Colinet et al. 2007, Foerster and Doetzer 2006, Foerster et al. 2004, Langer and Hance 2000), very few have focused on potential hibernacula for overwintering adult parasitoids, especially for tiny wasps such as platygastrids. Such information could help in better management of agroecosystems by elucidating the needs of a crucial natural enemy. This study was undertaken to assess *T. podisi* behavior in response to different potential refuge architectures and materials when subjected to overwintering conditions in the laboratory followed by an attempt to corroborate laboratory observations with field collections of overwintering parasitoids.

Materials and Methods

Insect colonies. *Podisus maculiventris* Say (Hemiptera: Pentatomidae), used as egg hosts for *T. podisi*, were originally obtained from the laboratory colony of Dr. Walker Jones (USDA Biological Control of Pests Research Unit and National Biological Control Laboratory, Stoneville, MS). Under favorable conditions, *P. maculiventris* do not enter obligate diapause, as do some herbivorous stink bugs such as *Euschistus servus* Say (Hemiptera: Pentatomidae), thereby assuring an adequate host egg supply for a laboratory colony of *T. podisi* throughout the year. Nymphs were held separately from adults through the fifth stadium in 100 × 15-mm polystyrene petri dishes (P5856, Sigma-Aldrich, Milwaukee, WI) to avoid cannibalism and provided with moist filter paper (Fisherbrand[®] Filter Paper P8-creped, Fisher Scientific, Pittsburgh, PA) and fed *Tenebrio molitor* L. (Coleoptera: Tenebrionidae) pupae. Adults were maintained in 9.5-L plastic aquaria (Living World Small Pals Pen Plastic Carrier, Hagen, Mansfield, MA) containing a potted soybean plant, *Glycine max* (L.) Merrill (Fabaceae) (cv. Woodruff) originally

obtained from Michael Buffaloe (North Carolina State University Soybean Extension Technical Specialist) and fed *T. molitor* pupae ad libitum. The aquaria were maintained at 25 \pm 1°C, 14:10 h (L:D), and 60 \pm 5% relative humidity (RH). The walls of the aquaria were lined with cheesecloth (50 grade, American Fiber and Finishing, Burlington, MA) to provide an oviposition substrate. The portion of cheesecloth with deposited eggs were cut out daily and stored in 5-ml polypropylene test tubes at 25 \pm 1°C, on 14:10-h photoperiod regime and 60 \pm 5% RH in a growth chamber. Identification of the stink bug species was done by the senior author, according to the taxonomic key in Arnett (1993). Voucher specimens were stored in 70% ethanol at the North Carolina State University Insect Museum.

Tenebrio molitor larvae, that served as food for *P. maculiventris*, were originally obtained from PetSmart (Walnut Street, Cary, NC) and subsequently reared in 12-L plastic aquaria in the laboratory. Larvae and adults were fed oatmeal (Old Fashioned Oats, My Essentials[™], DZA Brands, LLC, Salisbury, NC) as needed in addition to four or five carrot slices (2 cm long) every 48 h as source of moisture. Frass was sieved monthly from aquaria containing \geq 45-d-old larvae to maintain hygienic conditions. Every 60 d, adults were transferred to clean 12-L aquaria with fresh oatmeal and carrots for oviposition. Aquaria were maintained at 25 ± 1°C, on a 14:10-h photoperiod regime and 60 ± 5% RH. Pupae were collected every 48 h for both colony renewal and feeding *P. maculiventris*. Identification of the mealworm species was done by the senior author according to the taxonomic key in Arnett (1993). Voucher specimens were stored in 70% ethanol at the North Carolina State University Insect Museum.

Sentinel *P. maculiventris* egg masses were pinned to the underside of leaves to collect *T. podisi* within soybean fields between June to September 2011 and 2012 (Johnston Co. and Wake Co., NC). Parasitized eggs were held in 100 × 15-mm polystyrene petri dishes (P5856, Sigma-Aldrich) with moistened single layer of filter paper. After eclosion, 24-h-old females were collected and transferred to 5-ml polystyrene test tubes streaked with clover honey (Foodlion LLC, Salisbury, NC) as source of nutrition and provided *P. maculiventris* egg masses for oviposition. Tubes were stoppered with a cotton plug to confine wasps, then held in test tube racks over a water trough inside a 9.5-L plastic aquarium (Living World Small Pals Pen Plastic Carrier, Hagen) to maintain ambient environmental conditions at 25 ± 1°C, 14:10-h photoperiod, and $60 \pm 5\%$ RH. Identification of the wasp species was done by the senior author, according to the taxonomic key in Johnson (1984). Voucher specimens were stored in 70% ethanol at the North Carolina State University Insect Museum.

Laboratory experiments. Six experiments were conducted to examine the overwintering refuge preferences of *T. podisi* in relation to refuge height, material, distance from refuge edge, and resting position inside refuge. Decisions on refuge sizes were based on measurements of female *T. podisi* taken under a stereomicroscope (Leica Wild MZ8, Leica Microsystems Inc., Buffalo Grove, IL) at $10 \times$ magnification as follows: (a) total body length from tip of antennae to resting wing tip (side view) of 10 live and 10 frozen *T. podisi* females, (b) the standing height estimated from 10 frozen *T. podisi* females, (c) the width of the head and abdomen at their widest from 10 frozen *T. podisi* females, and (d) height of 10 *P. maculiventris* eggs.

Polystyrene petri dishes (90 × 15 mm) (Fisher Scientific[™], Waltham, MA) were used as arenas for Experiments 1, 4, and 6. Experiments 2, 3, and 5 used 150 × 25-mm polystyrene dishes (Falcon[®], Radnor, PA) because these refuges required taller and wider dishes. Elmer's Glue-All[®] (Borden Inc., Columbus, OH) was used in refuge construction.

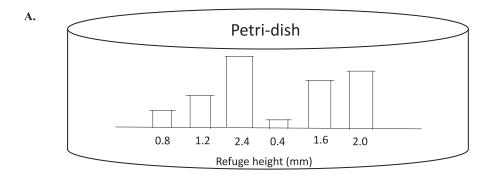
Each experimental arena was provided ten 24-h-old mated and starved *T. podisi* females and streaked with processed clover honey (Foodlion LLC). They were held in a growth chamber for the duration of the experiment at 70 \pm 1% RH. The temperature and photoperiod were gradually changed over a period of 5 d from 25°C to 5°C and 14-h to 10-h photophase, respectively, to simulate overwintering conditions and induce overwintering. On the sixth day, the number of wasps that had entered each refuge and their position (refuge top/side/bottom) was noted.

The objective of Experiment 1 was to determine the preference of overwintering *T. podisi* for various discrete refuge heights. Refuges using 0.3-mm-thick index cards and 0.1-mm-thick paper (Husky[®] Copy, Domtar Corporation, Fort Mill, SC) were constructed 10 mm long and 5 mm wide, and the following six heights in each arena: 0.4, 0.8, 1.2, 1.6, 2.0, and 2.4 mm (Fig. 1A). A strip of Parafilm (Parafilm M[®], Pechiney Plastic Packaging, Menasha, WI) was glued to the bottom of each refuge, and refuges then were glued in random order to a petri dish base 5 mm apart. The experimental arena (petri dish) was replicated five times, and one trial of the experiment was conducted.

The objective of Experiment 2 was also to determine the preference of overwintering *T. podisi* for discrete refuge heights, but over a broader range. Refuges were constructed as described in Experiment 1 but included the following range of heights in each arena: 0.5, 1.0, 2.0, 4.0, 8.0, and 16.0 mm (Fig. 2A). The experimental arena was replicated five times, and one trial of experiment was conducted.

The objective of Experiment 3 was to determine the refuge height preference of overwintering *T. podisi* when offered a gradient of refuge heights as opposed to discrete refuge heights in the previous two experiments. The refuges in the experimental arena were constructed using paper. Each petri dish arena contained a single 10-cm² paper refuge with a height of 4 mm at one end and 20 mm at the other (Fig. 3A). A strip of Parafilm was glued to the bottom of the refuge, which in turn was glued to a petri dish base. The experimental arena was replicated five times, and two trials of experiments were conducted.

The objective of Experiment 4 was to determine the preference of overwintering *T. podisi* for refuge material, two discrete refuge heights based on height of hatched *P. maculiventris* eggs, and presence of a cover on top of refuges. Refuges (10 mm long and 5 mm wide) were constructed from two types of materials, hatched *P. maculiventris* eggs and paper, and two height options of 1.0 or 2.0 mm (Fig. 4A). The average height of 10 *P. maculiventris* eggs was measured to be approximately 1.0 ± 0.05 (SD) mm (Leica Wild MZ8, Leica Microsystems Inc.), and this measurement provided a basis for building refuges from hatched *T. podisi* host eggs in a single layer as would be naturally laid in the field as well as occasional double-layered *P. maculiventris* egg masses. Only half of the material–height combination refuges received an index card cover on top. The experimental arena was replicated six times, and one trial of experiment was conducted.





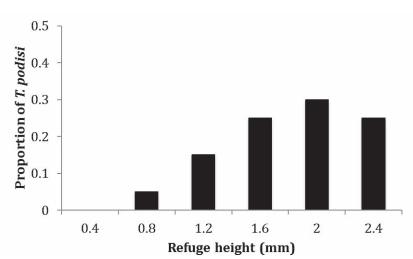
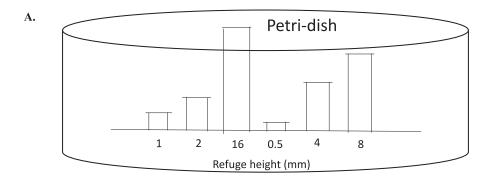


Fig. 1. (A) Experimental arena (petri dish) with artificial refuges provided in Experiment 1. (B) Refuge height preference by *Telenomus podisi* females (24 h old, mated, unfed; n = 20) when exposed to overwintering conditions (5°C; 10-h photophase; 75% RH). (Wald Chi-Square test; P = 0.0086).

The objective of Experiment 5 was to determine the preference of overwintering *T. podisi* for distance from edge of refuge. Paper refuges of the same cross-sectional area (10^2 mm) but of six different lengths, including 10, 20, 40, 60, 80, and 100 mm (Fig. 5A), were constructed. Each refuge was glued to a Parafilm strip, and then refuges were randomly arranged and glued to petri dish bases. The experimental arena was replicated five times, and three trials of experiments were conducted.

19



В.

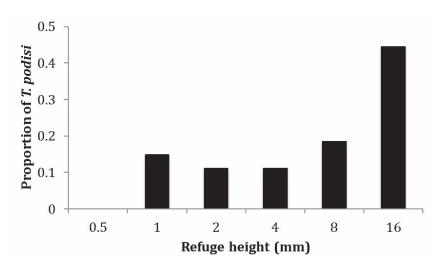
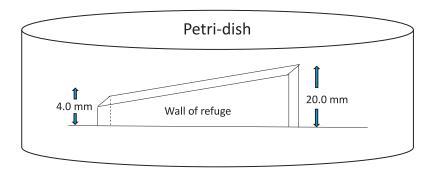


Fig. 2. (A) Experimental arena (petri dish) with artificial refuges provided in Experiment 2. (B) Refuge height preference by *Telenomus podisi* females (24 h old, mated, unfed; n = 27) when exposed to overwintering conditions (5°C; 10-h photophase; 75% RH). (Wald Chi-Square test; P = 0.0008).

The objective of Experiment 6 was to test whether the wasps were being attracted or repelled by Elmer's Glue-All (Borden). A pair of identical refuges (1,000 mm³) was used in each arena, with one glued to a Parafilm strip then glued to the petri dish base, and a second one placed in each dish without gluing. This experimental arena was replicated five times, and one trial of experiment was conducted.

А.



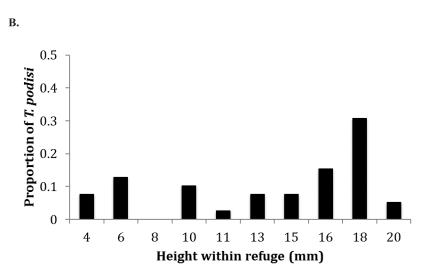
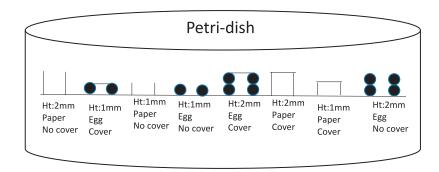


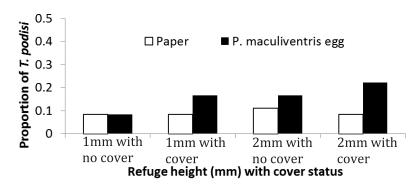
Fig. 3. (A) Experimental arena (petri dish) with artificial refuges provided in Experiment 3. (B) Refuge height preference by *Telenomus podisi* females (24 h old, mated, unfed; n = 39) when exposed to overwintering conditions (5°C; 10-h photophase; 75% RH). (Wald Chi-Square test; P = 0.0013).

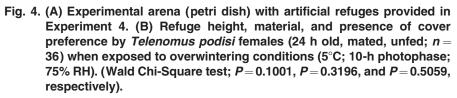
The objective of this survey was to determine whether parasitoid wasps specific to pentatomids prefer leaf-litter or tree bark for overwintering. Three sites with southfacing woodland borders adjacent to soybean fields previously identified as harboring stink bug and kudzu bug populations during the preceding crop growing season were selected near Clayton (Johnston Co., NC). The geographic locations of the three sites were N 35.686093°, W –78.316169° for Site 1, N 35.699303°, W

А.



В.





-78.336124° for Site 2, and N 35.700906°, W -78.337412° for Site 3. Adjacent woodlands were mixed forests consisting primarily of mature loblolly pine, *Pinus taeda* L. (Pinaceae), and sweet gum, *Liquidambar styraciflua* L. (Altingiaceae), trees. Pine and sweet gum were selected for sampling due to their prevalence and because the bark of older trees in both species becomes deeply furrowed, offering potential refuge sites for overwintering insects. Tree pairs were assigned for sampling, such that each pair consisted of one pine and one sweet gum tree that were never more than 3 m apart.



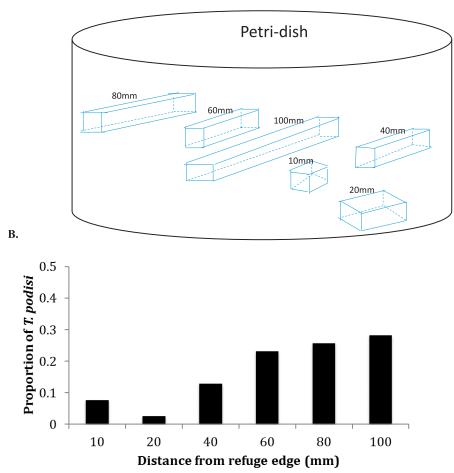


Fig. 5. (A) Experimental arena (petri dish) with artificial refuges provided in Experiment 5. (B) Preference of *Telenomus podisi* females (24 h old, mated, unfed; n = 39) for distance from refuge edge when exposed to overwintering conditions (5°C; 10-h photophase; 75% RH). (Wald Chi-Square test; P = 0.1028).

Leaf-litter samples were collected in plastic bags from a 0.09-m² area directly beneath north- and south-facing sides of caged trees, and between trees in each pair, at Sites 2 and 3 (4–22 February 2013) when temperatures were never above 5°C and then refrigerated until tallied in the laboratory. Samples were held in landscaping fabric cages (0.9-m height) (360-count Promat Professional, Lowe's Inc., Cary, NC) in the greenhouse with supplemental lighting to maintain a 14-h photophase for parasitoid extraction. Each cage was fitted with a funnel/bottle collection system on top to trap parasitoids as they moved upwards when

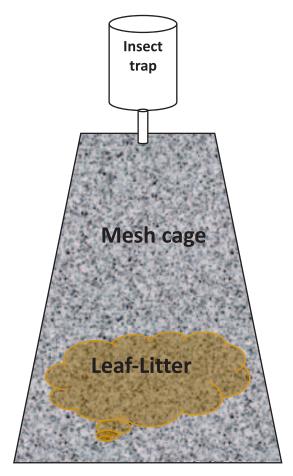


Fig. 6. Mesh cage with funnel-bottle collection system in greenhouse to collect overwintering parasitoids from leaf-litter collected at Sites 2 and 3 near Clayton, Johnston Co., NC (February 2013).

temperatures were warm enough for flight or crawling (Fig. 6). Emerging parasitoids were collected daily for 2 wk and preserved in 70% ethanol until identification. Density of parasitoids was calculated by dividing total number of parasitoids by total sample area (9.0 m^2).

To assess potential parasitoids overwintering in bark, tree bark was sampled as described in Lahiri et al. (2015). All emerging parasitoids were collected from the attached bottle traps (4–29 March 2013) and stored in 70% ethanol for identification according to taxonomic keys by Arnett (1993). Density of parasitoids from bark was calculated by dividing total number of parasitoids collected from bark by total sample area (21.07 m^2).

Data analysis. All laboratory experimental data were subjected to PROC GENMOD for conducting Wald Chi-Square tests to examine whether there was any

association between refuge types and *T. podisi* overwintering refuge choice (SAS Institute Inc. 2013). Significant tests were followed by two-sided Fisher's Exact Conditional test to test whether the proportion of wasps entering taller refuges was higher compared with those opting for shorter refuges. An alpha level of 0.05 was used for all statistical analyses.

Results

The mean (\pm SD) body length of female *T. podisi* was 2.01 \pm 0.06 mm in live females and 1.7 \pm 0.09 mm in frozen females. The mean (\pm SD) standing height of female *T. podisi* was 0.74 \pm 0.12 mm. The mean (\pm SD) width of head, thorax, and abdomen of female *T. podisi* was 0.39 \pm 0.004, 0.37 \pm 0.03, and 0.33 \pm 0.04 mm, respectively, at dorsal view. Mean (\pm SD) height of *P. maculiventris* eggs was 1.0 \pm 0.05 mm.

Results from Experiments 1, 2, and 3 indicated that there was a statistically significant effect of refuge height on overwintering preference of T. podisi $[0\chi^2](1,$ n=20 = 6.91, P=0.0086; $\chi^2(1, n=27) = 11.25$, P=0.0008 and $\chi^2(1, n=39) = 11.25$ 10.38, P = 0.0013, respectively] and that the proportion of wasps entering taller refuges ($H_0 = \mu_1 = \mu_2 = \mu_n$; $H_T = \mu_1 < \mu_2 < \mu_n$, where μ_1 represents the mean number of wasps overwintering in the shortest refuge and μ_n represents the mean number of wasps overwintering in the tallest refuge) was significantly higher compared with shorter refuges [t(1) = 7.56, exact-P = 0.0066; t(1) = 12.6, exact-P= 0.0004, and t (1) = 11.08, exact-P = 0.0009, respectively] (Figs. 1B, 2B, 3B). Approximately one-third of wasps settled in the tallest refuges, those that were 24fold the insect's standing height (Figs. 1B, 2B). In Experiment 4, there was no significant effect of refuge material (*P. maculiventris* egg versus paper) [χ^2 (1, n =36 = 2.70, P = 0.1001], refuge height (1 mm versus 2 mm) [χ^2 (1, n = 36) = 0.99, P = 0.3196], or presence versus absence of a cover [χ^2 (1, n = 36) = 0.44, P =0.5059] on where wasps preferred to settle (Fig. 4B). Data from Experiment 5 indicated that parasitoids prefer how far away from the edge of horizontal shelters they settled to become quiescent [χ^2 (1, n = 39) = 10.74, P = 0.0010], and the proportion of wasps selecting longer refuges over shorter refuges was higher [t(1)]= 11.52, exact-P = 0.0006] (Fig. 5B). When settling positions of wasps from all experiments were combined, 86% of wasps preferred hanging inverted from the tops of refuges as opposed to hanging on the side (8%) or standing on the bottom (6%) (Fig. 7). Elmer's glue had no significant effect (F = 0.07; df = 1, 8; P =0.7924) on wasp settling behavior, with 52% settling in refuges with glue and 48% in refuges without alue.

In the field study, density of all overwintering parasitoids collected from leaf-litter was found to be 3.44/m² and from bark to be 0.33/m² (Table 1). Because so few parasitoids were collected, the possibility of statistical analysis was precluded, so collected parasitoids were identified to the family level and results tabulated. Parasitoids belonging to various superfamilies included Chalcidoidea (five families), lchneumonoidea (two families), Platygastroidea (one family), Diaprioidea (one family), Cynipoidea (one family), and Chrysidoidea (one family). The number of parasitoids captured from leaf-litter and tree bark, belonging to the respective hymenopteran families, has been reported in Lahiri (2014). Only one platygastroid

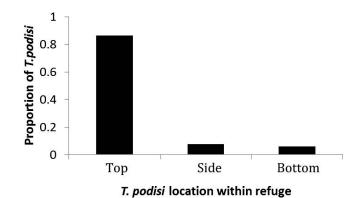


Fig. 7. Preference of *Telenomus podisi* females (24 h old, mated, unfed; n = 182) for top, side, or bottom of a refuge when exposed to overwintering conditions (5°C; 10-h photophase; 75% RH).

wasp was collected each from north-facing pine leaf-litter and tree bark samples. All parasitoids combined were 10-fold more numerous in leaf-litter than in bark.

Discussion

Telenomus podisi only selected artificial "overwintering" refuges that were 0.8 mm or taller that accommodated the wasp standing height of 0.74 ± 0.14 mm, but they strongly preferred much taller (Figs. 1, 2, 3) and longer (Fig. 5) refuges. The wasps also preferred hanging inverted from the tops of refuges. This, together with their avoidance for tight spaces, suggests that fresh hardwood leaf-litter (versus older, packed litter or tree bark) might be a choice overwintering refuge to protect against excessive moisture or extreme temperature fluctuations. Forest leaf-litter offers a protective environment to overwintering insects by acting as a buffer against severe winter temperatures due to insulating properties of litter (Byers 1984). Also, microbial respiration within leaf-litter (Gunnarsson et al. 1988) may be a steady source of heat and moisture, which might prove very beneficial for small overwintering parasitoids.

Johnson (1984) referred to a single unpublished observation by "Hopkins" (no citation, first name, or date given) of several female *T. podisi* apparently

Location	Number Samples	Total Number Parasitoids Collected	Parasitoid Density (number/m ²)
Leaf-litter	100	31	3.44
Tree bark	58	7	0.33

Table 1. Density of parasitoid wasps found in leaf-litter and bark from loblolly pine and sweet gum trees near Clayton, NC (January–March 2013).

overwintering in the hibernaculum (loose silk thread covering) of overwintering satin moth larvae, *Leucoma salicis* L. (Lepidoptera: Lymantriidae), on a tree branch. Based on this single record from the literature, it might be reasonable to assume that *T. podisi* prefer to overwinter in structures produced by other insects. However, in this study *T. podisi* did not appear to differentiate between refuges made of hatched host eggs or paper (Fig. 7).

Although statistical analysis was not possible because of the low numbers of wasps collected, the field study provided some indication of the different families of parasitoids taking refuge in the woodland borders adjacent to soybean fields in North Carolina. Higher densities of parasitoids collected from leaf-litter versus tree bark also reiterated the possibility of parasitoids preferring leaf-litter to tree bark for overwintering.

The small size of platygastroids, such as *T. podisi*, has limited overwintering observation of this species, and there has been only one mention of a single nonpublished observation of *T. podisi* overwintering in the literature (Johnson 1984). Active *Telenomus* adults have been collected early in spring near forest environments, suggesting that they were moving from these sites following overwintering (Kuzin et al. 1980). Other parasitoids overwintering in forest environments have been reported as well. For example, while studying the hibernating preferences of 39 species of adult ichneumonid wasps in Ohio, the underside of loose bark on moss-covered fallen logs suspended across deep ravines, lying at the base of north-facing slopes, was found to be most preferred (Dasch 1971). Minimal humidity and temperature fluctuations in these hibernacula were found to be the reason for such strong preference.

The present study noted an apparent higher density of overwintering parasitoids, particularly small chalcidoid parasitoids, in hardwood leaf-litter. This, combined with the laboratory studies, suggests this may be a preferred and probably abundant habitat for *T. podisi* and its relatives in the Southeast. It may be valuable for a future study to compare the value of hardwood versus pine leaf-litter as overwintering habitat for these parasitoids.

Acknowledgments

This study was supported in part by a USDA-Southern SARE Graduate Student Grant (GS11-104). We thank Walker Jones and Richard Evans (USDA-ARS) for providing *P. maculiventris*, Tim Britton for locating field sites, John Monahan for statistical analysis advice, Steve Frank for using his sewing machine to make cages, and Jennifer Call for assistance with the project.

References Cited

- Al-Ghamdi, K.M., R.K. Stewart and G. Boivin. 1995. Synchrony between populations of the tarnished plant bug *Lygus lineolaris* (Palisot De Beauvois) (Hemiptera: Miridae), and its egg parasitoids in Southwestern Quebec. Can. Entomol. 127: 457–472.
- Arnett, R.H. Jr. 1993. American Insects: A Handbook of the Insects of America North of Mexico. Sandhill Crane Press, Gainesville, FL.
- Austin, A.D. 1984. The fecundity, development and host relationships of *Ceratobaeus* spp. (Hymenoptera: Scelionidae), parasite of spider eggs. Ecol. Entomol. 9: 125–138.
- Babendreier, D., S. Kuske and F. Bigler. 2003. Overwintering of the egg parasitoid *Trichogramma brassicae* in Northern Switzerland. BioControl 48: 261–273.

- Baker, G.L. and R.J. Dysart. 1992. Development of *Scelio bipartitus* Kieffer (Hymenoptera: Scelionidae) in diapause eggs of *Gastrimargus musicus* (F.) (Orthoptera: Acrididae). Aust. J. Entomol. 31: 241–242.
- Baust, J.G. 1981. Biochemical correlates in cold-hardening insects. Cryobiology 18: 186– 198.
- Baust, J.G. and R.E. Lee. 1982. Environmental triggers to cryoprotectant modulation in separate populations on the gall fly, *Eurosta solidaginsis* (Fitch). J. Insect Physiol. 28: 431–436.
- Byers, J.A. 1984. Electronic multiprobe thermometer and multiplexer for recording temperatures of microenviroments in the forest litter habitat of bark beetles (Coleoptera: Scolytidae). Environ. Entomol. 13: 863–867.
- Colinet, H., P. Vernon and T. Hance. 2007. Does thermal-related plasticity in size and fat reserves influence supercooling abilities and cold-tolerance in *Aphidius colemani* (Hymenoptera: Aphidiinae) mummies? J. Thermal Biol. 32: 374–382.
- Coombs, M. 2004. Overwintering survival, starvation resistance, and post-diapause reproductive performance of *Nezara viridula* (L.) (Hemiptera: Pentatomidae) and its parasitoid *Trichopoda giacomellii* Blanchard (Diptera: Tachinidae). Biol. Control 30: 141–148.
- Dasch, C.E. 1971. Hibernating Ichneumonidae of Ohio (Hymenoptera). Ohio J. Sci. 71: 270– 283.
- **Doetzer, A.K. and L.A. Foerster. 2007.** Development, longevity and reproduction of *Trissolcus basalis* (Wollaston) and *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae) in natural conditions during autumn and winter, in Southern Parana, Brazil. Neotrop. Entomol. 36: 233–242.
- Foerster, L.A. and A.K. Doetzer. 2006. Cold storage of the egg parasitoids *Trissolcus basalis* (Wollaston) and *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae). Biol. Control 36: 232–237.
- Foerster, L.A., A.K. Doetzer and L.C.F. de Castro. 2004. Emergence, longevity and fecundity of *Trissolcus basalis* and *Telenomus podisi* after cold storage in the pupal stage. Pesq. Agropec. Bras., Brasília 39: 841–845.
- Gunnarsson, T., P. Sundin and A. Tunlid. 1988. Importance of leaf litter fragmentation for bacterial growth. Oikos 52: 303–308.
- Hance, T., J. van Baaren, P. Vernon and G. Boivin. 2007. Impact of extreme temperatures on parasitoids in a climate change perspective. Annu. Rev. Entomol. 52: 107–126.
- James, D.G. 1988. Fecundity, longevity and overwintering of *Trissolcus biproruli* Girault (Hymenoptera: Scelionidae), a parasitoid of *Biprorulus bibax* Breedin (Hemiptera: Pentatomidae). J. Aust. Entomol. Soc. 27: 297–301.
- Johnson, N.F. 1984. Systematics of Nearctic *Telenomus*: Classification and revisions of the *Podisi* and *Phymatae* species groups (Hymenoptera: Scelionidae). Bull. Ohio Biol. Surv. New Ser. 6(3): 1–113.
- Keller, M.A. 1986. Overwintering by *Trichogramma exiguum* in North Carolina. Environ. Entomol. 15: 659–661.
- Kuzin, A.A., V.S. Kudryavtsev and A.Y. Ponurovskii. 1980. Noxious bugs on wheat and their insect enemies. Zashchita Rastenii 10: 24–25.
- Lahiri, S. 2014. The potential for conservation biological control of pentatomids and plataspids in North Carolina. PhD Dissertation. North Carolina State Univ., Raleigh.
- Lahiri, S., D. Orr, C. Sorenson and Y.J. Cardoza. 2015. Overwintering refuge sites for Megacopta cribraria (Hemiptera: Plataspidae). J. Entomol. Sci. 50: 69–73.
- Langer, A. and T. Hance. 2000. Overwintering strategies and cold hardiness of two aphid parasitoid species (Hymenoptera: Braconidae: Aphidiinae). J. Insect Physiol. 46: 671–676.
- Mansingh, A. 1971. Physiological classification of dormancies in insects. Can. Entomol. 103: 983–1009.
- SAS Institute Inc. 2013. SAS/STAT® 9.3 User's Guide. SAS Institute Inc., Cary, NC.
- Tauber, M.J., C.A. Tauber and S. Masaki. 1986. Seasonal adaptations of insects. Oxford Univ. Press, New York. 411 pp.
- Tillman, P.G. 2010. Parasitism and predation of stink bug (Heteroptera: Pentatomidae) eggs in Georgia corn fields. Environ. Entomol. 39: 1184–1194.
- Torgersen, T.R. and R.B. Ryan. 1981. Field biology of *Telenomus californicus* Ashmead, an important egg parasite of Douglas-fir tussock moth. Ann. Entomol. Soc. Am. 74: 185–186.