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*Larra bicolor*, a parasitic wasp of mole crickets, is one of three natural enemies imported and introduced into Florida for biological control of mole crickets and has since expanded its range into Alabama and Mississippi and has been introduced into Georgia and Louisiana. The presence of nectar sources are important to the establishment of *L. bicolor*. The objectives of this study were to compare the recruitment of adult *L. bicolor* to ornamental plants, to evaluate potential passive monitoring techniques, and to determine the seasonal and diurnal activity of *L. bicolor*.

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# Biology of *Larra bicolor*, a Parasitoid of Mole Crickets, and Potential Ornamental Plants as Nectar Sources

David W. Held and C. M. Abraham

## SUMMARY

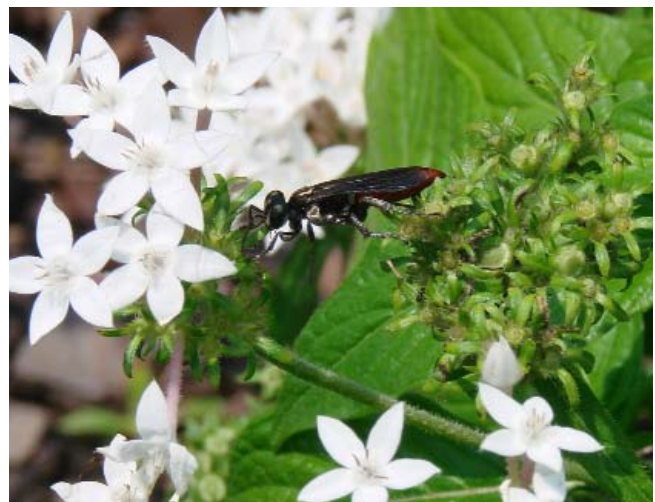
*Larra bicolor*, a parasitic wasp of mole crickets, is one of three natural enemies imported and introduced into Florida for biological control of mole crickets and has since expanded its range into Alabama and Mississippi and has been introduced into Georgia and Louisiana. The presence of nectar sources are important to the establishment of *L. bicolor*. The objectives of this study were to compare the recruitment of adult *L. bicolor* to ornamental plants, to evaluate potential passive monitoring techniques, and to determine the seasonal and diurnal activity of *L. bicolor*. Results of this study include:

- No wasps were observed on the patches of grass treated with sugar water or on the foliage of trees sprayed with sugar water.
- In a test to evaluate the effectiveness of pan-type traps, sets of plastic bowls (white, yellow, and clear) were placed 0.5 m apart around the edge of the flower garden containing flowering *Spermococe verticillata* and *Pentas lanceolata*. No wasps were captured in any of the bowls, although adults were active on flowers during these experiments.
- In 2007 and 2008, a garden of potential nectar sources was planted on the grounds of the Coastal REC, Biloxi, MS. In both years, *L. bicolor* wasps were only observed on flowers of pentas and spermacoe. The number of wasps found on spermacoe and white-flowered pentas was double the number found on either red- or pink-flowered pentas.
- Laboratory trials were conducted to determine longevity of wasps provisioned with flowering *S. verticillata* or pentas. In Trial 1, wasps provisioned with pentas flowers lived 4.75 days longer than those provisioned with spermacoe. In Trial 2, average survival of wasps ranged from 3-4.5 days, and there was no significantly different between treatments.
- Seasonal activity of *L. bicolor* was monitored on four sites in coastal Mississippi from 2006-2008. Wasps are active from June to November and were active later in the season on flowering plants. Daily activity begins at 0700 in August and 0900 hours in October and continues to near dusk.

*Larra bicolor* F. is one of three natural enemies imported and introduced into Florida for biological control of mole crickets (*Scapteriscus* spp., Orthoptera: Gryllotalpidae) (7). Of these, *L. bicolor* has naturally expanded its range into Alabama (1) and Mississippi (10) and has been introduced into Georgia and Louisiana (6).

*Larra bicolor* females hunt in pastures and managed grasses infested with mole crickets. Females enter the burrow forcing the mole cricket to emerge. On the surface, the mole cricket is stung which induces a short paralysis to allow oviposition (4). The egg requires 6-7 days to hatch. The wasp larva develops externally feeding for about 11 days before detaching and forming a cocoon (4). Pupation occurs in the soil with adults emerging about 6-8 weeks later. Between bouts of hunting, females are found on flowering

plants (2, 4) and the presence of nectar sources are important to the establishment of *L. bicolor* (11).



Adult *L. bicolor* feeding on the white flowers of *Pentas lanceolata*.

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Once the egg hatches (about a week), the wasp larvae feeds externally as an ectoparasite on the host mole cricket.

### Previous Research on *Larra bicolor*

Most of the published research on the biology and ecology of *L. bicolor* has been conducted in South America, Puerto Rico, or Florida. In Florida, *L. bicolor* has two to three asynchronous generations per year (6). Activity is typically determined by observing wasps on turfgrass or flowering plants (2, 11), although wasps are easily disturbed which complicates monitoring (5, authors observations).

Adult *Tiphia* spp., parasitoids of white grubs, were recruited to sugar solutions applied to the grass or to foliage on low-hanging limbs of trees and collected in pan-type traps (15). Identification of alternative monitoring methods may enable researchers and extension personnel to better document the presence and continued spread of *L. bicolor*.

### Nectar Sources are Important

Consumption of nectar or other carbohydrate sources (e.g., honeydew or sugar sprays) can increase adult longevity and fecundity as well as increase egg maturation (16). Arévalo and Frank (2) compared attractiveness of five flowering wildflowers to *L. bicolor*, but all were visited less than *Spermacoce verticillata* (Rubiaceae). *Spermacoce verticillata* is a non-native weed (13)

that readily reproduces from seeds and overwinters in the landscape in coastal Mississippi (personal observations). Populations of *L. bicolor* on flowers of *S. verticillata* are reportedly male biased (6), but there are apparently no data to support these claims.

In 2006, *L. bicolor* was observed feeding on *Pentas lanceolata* (Rubiaceae) in the landscape in coastal Mississippi. This suggests that other flowering ornamental plants or other plants in the



The wasp larva develops externally feeding for about 11 days before detaching and forming a cocoon.





Four sets of plastic bowls; white, yellow, and clear, were placed around the edge of the flower garden containing flowering *Spermacoce verticillata* and *P. lanceolata*. The bowls were filled with water and about 10 ml of liquid soap to capture the wasps. Despite wasps active on the flowers, no wasps were captured in the bowls.

Madder family may substitute as nectar sources for *L. bicolor*, especially where there are concerns about the pernicious nature of *S. verticillata*. Pentas are common in southern landscapes for

their heat tolerance and attractiveness to butterflies (3).

The objectives of this study were to compare the recruitment of adult *L. bicolor* to ornamental plants, to evaluate potential passive monitoring techniques, and to determine the seasonal and diurnal activity of *L. bicolor*.

### **Attempts to Monitor Activity of *Larrea bicolor***

Sugar sprays and pan-type traps (15) were evaluated for effectiveness to monitor adult *L. bicolor*. Sugar sprays were evaluated in three trials from June 30 to August 4, 2005 on two golf courses in coastal Mississippi where *L. bicolor* were active. Three small, 1 X 1-m plots of hybrid bermudagrass (*Cynodon dactylon* L. Pers × *C. transvaalensis* Burtt-Davy) were selected in out-of-play areas on each course. The grass was sprayed to runoff with a 10% sugar solution (15). In addition, a lower branch on the closest tree to each set of three plots was also sprayed.



Sixteen taxa of flowering ornamental plants were compared in a common garden experiment.

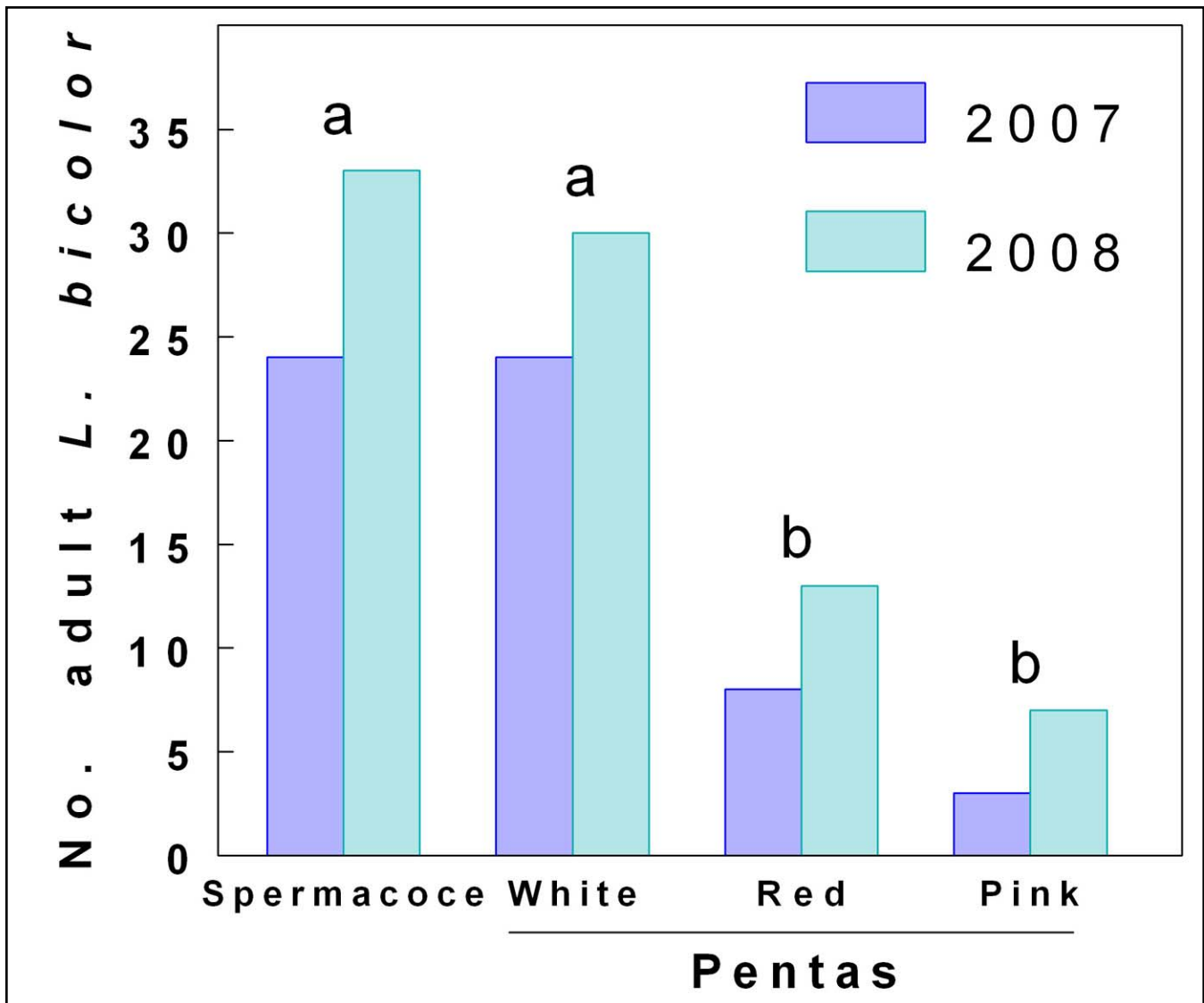
Treatments were applied around 1000 hours and monitored hourly for a 3-hour period immediately after treatment.

Another experiment was conducted to evaluate the effectiveness of pan-type traps on November 6, 2008. Set of plastic bowls (white, yellow, and clear) were placed 0.5 m apart around the edge of the flower garden containing flowering *S. verticillata* and *P. lanceolata* and filled with water and about 10 ml of liquid soap to reduce surface tension. At 0900, 1200, and 1500 hours, the number of adult *L. bicolor* captured in each bowl and the number of wasps counted in 2 minutes in each flowering plot were recorded.

Unfortunately, no wasps were observed on the patches of grass treated with sugar water or on the foliage of trees sprayed with sugar water. Likewise, no wasps were captured in any of the bowls, although adults wasps were active in the grass on the golf courses and on flowers during these experiments.

### Field Evaluation of Flowering Plants as Nectar Sources

In 2007 and 2008, a garden of potential nectar sources (Table 1) was planted on the grounds of the Coastal REC, Biloxi. Plants were



Data from 6 days in 2007 and 10 days in 2008 collected from the common garden experiment were analyzed. Plant species effect was significant. In both years, the number of wasps found on spermacoce and white-flowered pentas was double the number found on either red- or pink-flowered pentas.



Plant species	Common name	Family
<i>Achillea millefolium</i>	Yarrow	Asteraceae
<i>Asclepias curassavica</i>	'Silky Gold' Butterfly weed	Asclepidaceae
<i>Agastache urticifolium</i>	'Honeybee White' sweet hyssop	Lamiaceae
<i>Foeniculum vulgare</i>	Fennel	Umbellifereae
<i>Ixora coccinea</i>	Jungle geranium	Rubiaceae
<i>Lobularia maritime</i>	Sweet alyssum	Brassicaceae
<i>Pentas lanceolata</i>	'Butterfly Pink' pentas	Rubiaceae
<i>Pentas lanceolata</i>	'Butterfly White' pentas	Rubiaceae
<i>Pentas lanceolata</i>	'Butterfly Red' pentas	Rubiaceae
<i>Spermacoce verticillata</i>	False buttonweed	Rubiaceae
<i>Verbena bonariensis</i>	Purpletop verbena	Verbenaceae
<i>Verbena hybrida</i>	'Aztec White' verbena	Verbenaceae
<i>Verbena hybrida</i>	'Quartz Waterfall' verbena	Verbenaceae
<i>Verbena hybrida</i>	'Lady Scarlett' verbena	Verbenaceae
<i>Verbena hybrida</i>	'Blue Lagoon' verbena	Verbenaceae

**Table 1.** Flowering plants evaluated as nectar sources for *Larra bicolor*.



This test arena was used to determine the relative longevity of wasps held captive with flowering plants of spermacoce or pentas. Trial 1 compared longevity of field-collected wasps provisioned with a potted flowering plant of spermacoce or pentas to those provided no food (negative control). Trial 2 compared longevity of wasps provisioned with flowering potted plants of spermacoce or pentas to those provisioned with a sugar solution (positive control).

chosen based on observations of flower-feeding by *L. bicolor* (e.g., pentas), those reported to benefit other parasitic wasps or bees (e.g., fennel and sweet hyssop), members of the Madder family (i.e., *Ixora*), or ornamental plants with small, shallow flower that consistently flower all summer (e.g., verbena). Each plant species or cultivar was established in four replicated plots, 0.5 × 0.5 m. The entire garden was mulched and had drip irrigation. In 2008, annual plant taxa (i.e., pentas, hybrid and trailing verbenas, fennel, and jungle geraniums) were replanted, whereas the others were perennials.

Beginning in mid-May each year, plants were in full-flower and were inspected for adult *L. bicolor*. Numbers of wasps on flowering plants were recorded between 1100-1200 hours each day with a 15-minute observation period for each replicate. Plants species visited and the number of foraging wasps were recorded and summarized over 6 days in 2007 and 10 days in 2008. In both years, *L. bicolor* wasps were only observed on flowers of pentas and spermacoce. The number of wasps found on spermacoce and white-flowered pentas was double the number found on either red- or pink-flowered pentas.

## Comparison of Adult Longevity in the Laboratory

Laboratory trials were conducted to determine longevity of wasps provisioned with flowering *S. verticillata* or pentas. Plants for these experiments were grown from seeds in 15-cm plastic pots. Trial 1 compared longevity of field-collected wasps provisioned with a potted flowering plant of spermacoce or pentas to those provided no food (negative control). Trial 2 compared longevity of wasps provisioned with flowering potted plants of spermacoce or pentas to those provisioned with a sugar solution (positive control). Four replicates of each treatment were used in each trial. Wasps were field-collected within 12 hours of the start of each experiment. These experiments were conducted in plastic cylindrical test arenas with the bottom of each filled with dry sand to a depth of 20 cm. An empty plastic 15-cm pot was placed in the center so that flowering plants could be exchanged.

Initially, flowering plants were placed into each arena then replaced as needed to ensure adequate flowers throughout the experiment. Control treatments did not have flowering plants, but had a similarly sized plastic pot inverted over the empty pot in the center. In Trial 2, a cotton ball

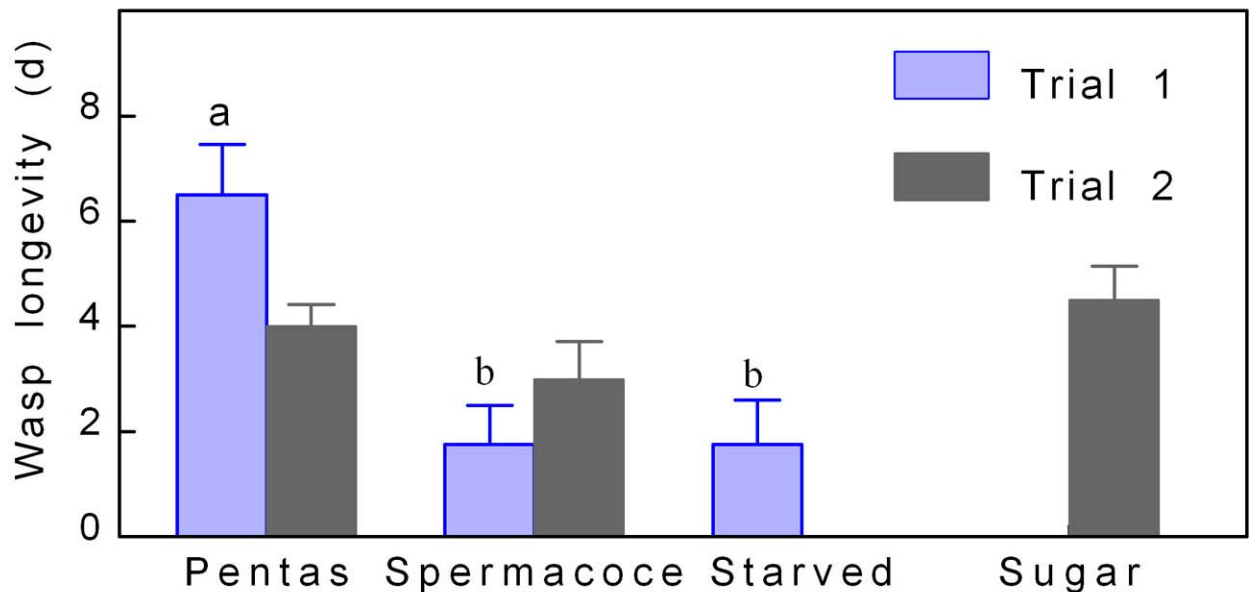
saturated with the sugar solution and placed on a plastic lid on top of the inverted pot. The sugar diet was changed every other day. A single field-collected wasp was introduced to each arena and wasp survival evaluated daily.

In Trial 1, wasps provisioned with pentas flowers lived 4.75 days longer than those provisioned with spermacoce. In Trial 2, average survival of wasps ranged from 3-4.5 days, and there was no significantly different between treatments.

## Seasonal Monitoring

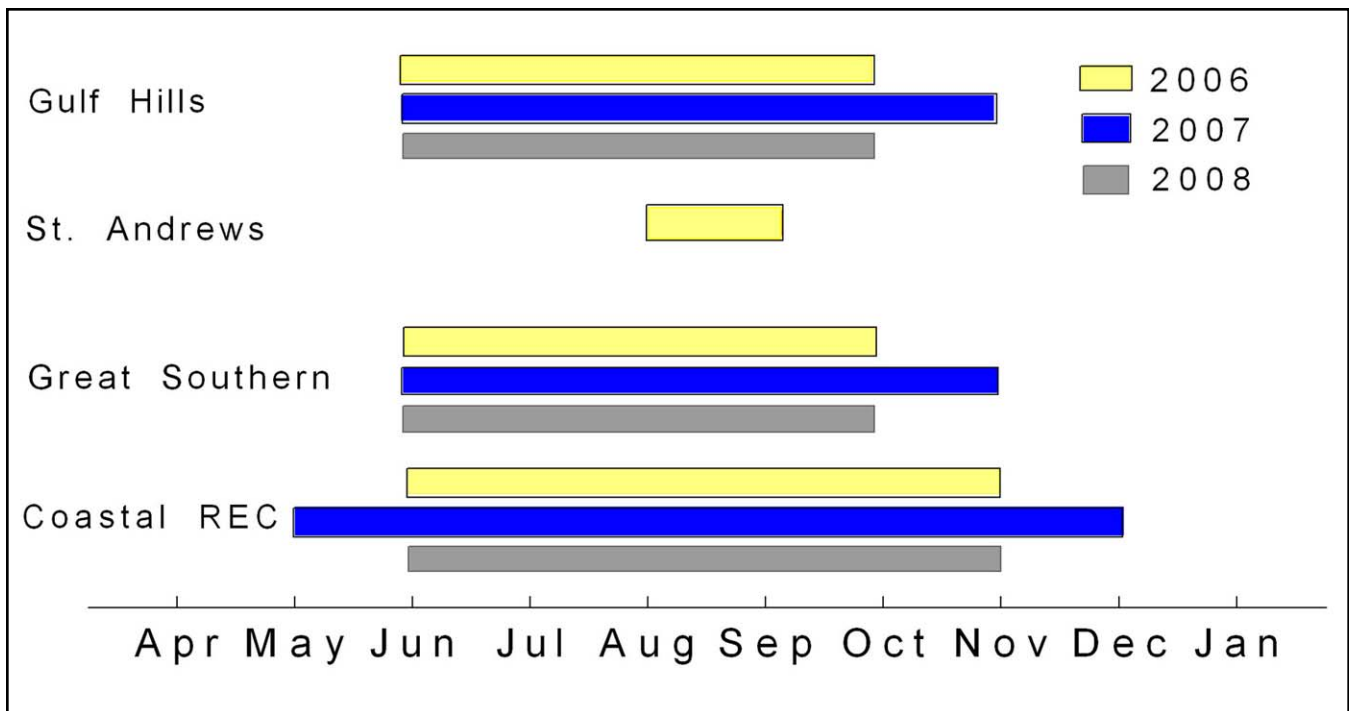
The seasonal abundance of *L. bicolor* was previously reported from Florida, Puerto Rico, and South America, therefore this experiment determined the seasonal activity in the northern Gulf Coast region. Three golf courses in coastal Mississippi (Harrison and Jackson counties) were visited and surveyed for *L. bicolor*. On each golf course, two sites were selected for monitoring based on activity of *L. bicolor* and mole crickets.

Observations were also made of wasps foraging on flowers *S. verticillata* and *P. lanceolata* in the previously mentioned garden. Since these wasps are noted nectar feeders, monitoring flowering plants would gauge the flower feeding activity of *L. bicolor* in addition to the hunting



Longevity (days) of *L. bicolor* wasps provisioned with a potted flowering plant of *Pentas lanceolata* 'Butterfly White' or *Spermacoce verticillata*. In Trial 1, wasps confined with pentas flowers had significantly greater longevity than those confined with Spermacoce flowers or no food. In Trial 2, wasps provided flowers or sugar water had comparable longevity.





Seasonal occurrence of *Larra bicolor* as indicated by presence or absence of wasps hunting in turfgrass infested with mole crickets (*Scapteriscus spp.*) on three golf course sites (Gulf Hills, St. Andrews, Great Southern) or foraging on flowers (Coastal REC) in coastal Mississippi.

activity. Each site was scouted for 15-20 minutes for wasp activity and presence of *L. bicolor* noted. Monitoring was done every 2 weeks on clear days between 1000-1500 hours from April 2006-December 2008.

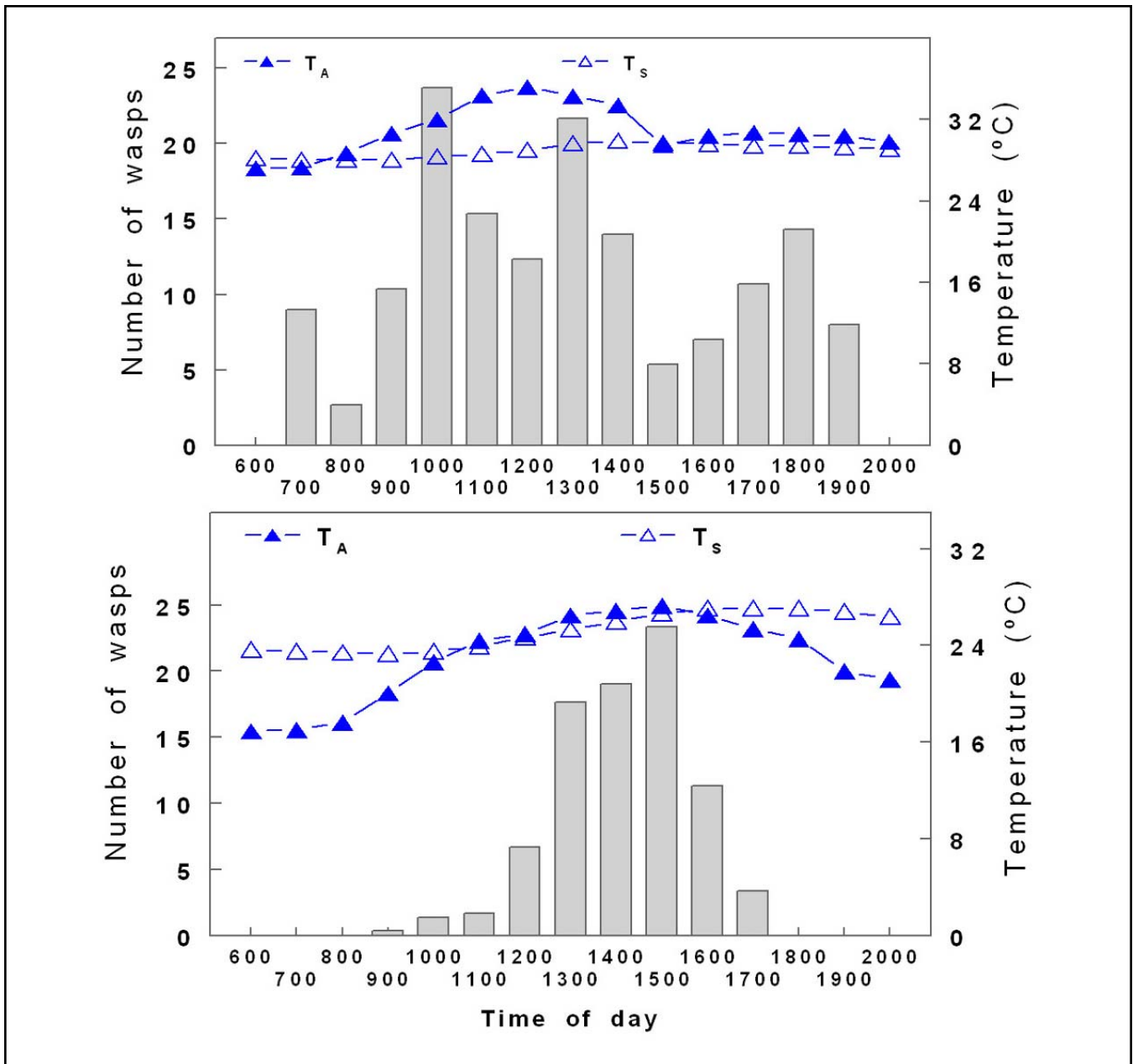
Across all sites, wasps were first observed in May and activity continued until December. This is similar to that reported from the Florida panhandle (Gadsden Co.,11). Wasps were observed on golf courses hunting in turf for mole crickets from June to November; however, wasps were active on flowers (Coastal REC) one month later each year. Seasonal activity was consistent on golf course sites except for one (St. Andrews). On that site, wasps were active in 2006 but not thereafter. Perhaps a reduction in mole cricket populations below a critical threshold, although unknown, or perhaps the direct impact of insecticides on the wasp population may explain this reduction. The impact of insecticides on *L. bicolor* has not been studied, but certain insecticides cause significantly mortality of other parasitoids (i.e., *Tiphia spp.*) foraging in treated turfgrass (14).

### Diurnal Activity

*Larra bicolor* are day-active so we wanted to document this diurnal activity as this wasp extends its range outside of Florida. Activity of wasps on flowers in the previously mentioned garden were recorded on August 7-10, 2007 and October 9-11, 2008 to compare summer and fall foraging. The number and sex of wasps on flowering plants in eight plots were taken hourly from 0500-2000 hours each day.

In August, plots of flowering plants were observed every two hours throughout the overnight period to determine if and when wasps abandoned nectar sources. Observations were made hourly for 20 minutes. Data were collected despite changes in weather such as rain or over-cast conditions. Air temperatures were recorded by a mini-weather station adjacent to the flower garden. Data from August 2007 and October 2008 were analyzed independently to determine the relationship between number of wasps and air and soil temperature. Also, male and females abundance was recorded for each time on each day.

Wasps were observed on flowers from



Daily activity of *Larra bicolor* (indicated by bars) over three days in August 2007 (upper graph) and October 2008 (lower) in relation to air ( $T_A$ ) and soil ( $T_S$ ) temperature.

about 0700 to 1900 hours (CST) each day in August and from 0900 to 1700 hours in October. This is consistent with daily flight activity reported from Brazil and Puerto Rico (5). Sunrise and sunset during August were 0520 hours and 1845 hours, and 0600 and 1730 hours during October, respectively. In August, air and soil temperatures were predictor variables of wasp activity but not in October.

Wasps were observed resting under foliage in the shaded interior of the flowering plants during the day. On two days in August, females

remained overnight underneath foliage on pentas. Between 0600-0700 hours, females that spent the overnight on pentas began grooming themselves before abandoning the plant. Most wasps, however, leave the plant at or near dusk regardless of time of year or temperatures. Dusk or overcast conditions may trigger wasps to seek refuge sites (5) before certain constraints (e.g., thermal or visual) prevent activity.

A maximum of 14 females and 31 males were observed at one time on flowers in August versus 3 females and 25 males in October. Males

outnumber females on flowers, which is consistent with previous observations (6). Fewer females later in the season may also explain why wasps were still observed on flowers when none were observed hunting later in the season. Females, however, were observed on flowers for distinct periods (3 to 9 hours) each day. The timing of this period, however, was inconsistent from day to day. In Puerto Rico, females hunt early in the day (0645 to 1200 hours EDT), about 1 hour after sunrise, and forage on flowers later (0745 to 1600 hours) from May to June (5).

In this study, females were often present in the morning, but the daily maximum number of females usually occurred later, about 1300 hours in August and October. In Puerto Rico, females hunt about one hour after sunrise, then forage on flowers later (5). Females likely need nectar early in the morning before hunting, returning later in the day to feed or rest as temperatures increase. Males do not approach females on flowers (4, author's observations). Mating, although rarely observed and poorly understood, reportedly occurs on the ground near hunting sites (1, 4).

### Conclusions

*Larra bicolor* was introduced to Florida for biological control of mole crickets (*Scapteriscus spp.*) and has spread to adjacent states. The objectives of this study were to determine the seasonal and diurnal activity of *L. bicolor* in its expanded range and to compare the ornamental plants as nectar sources. Experiments with colored, pan-type traps and sugar solutions to monitor for *L. bicolor* were unsuccessful and direct observations of wasps on flowers or hunting were used to monitor activity. The wasps may use floral volatiles (12) or a combination of volatile and visual cues to find nectar sources. Future field experiments are planned to evaluate floral extracts of flowers of pentas and spermacoce as attractants for *L. bicolor*. This may produce a reliable passive monitoring tactic or enable mass collection of wasps.

Seasonal activity of *L. bicolor* was monitored on four sites in coastal Mississippi from

2006-2008. Wasps are active from June to November and were active later in the season on flowering plants. Daily activity begins at 0700 in August and 0900 hours in October and continues to near dusk. Populations of wasps on flowers are typically male-biased, but females may be more abundant on flowers in the morning and again in late afternoon.

Among the various plants evaluated in common garden experiments, wasps only fed on pentas and spermacoce. In laboratory assays, longevity of wasps provisioned with pentas was comparable or better to those provisioned with spermacoce. As *L. bicolor* continues to expand its range, pentas, an annual ornamental, may be a more aesthetic and less problematic alternative to spermacoce as a nectar source for *L. bicolor* in the landscape.

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