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Observations of *Cerceris fumipennis* (Hymenoptera: Crabronidae) phenology and variation in its buprestid prey in Louisiana

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Abstract

The non-native emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae), threatens extirpation of susceptible ash (*Fraxinus* species; Lamiales: Oleaceae) in North America. *Cerceris fumipennis* Say (Hymenoptera: Crabronidae), a ground-nesting wasp that preys on Buprestidae in eastern North America, is used as a survey tool for the emerald ash borer in the northeastern U.S. and Canada. The recent detection of the emerald ash borer in Louisiana provides an opportunity to complement trapping surveys with the use of *C. fumipennis*, but knowledge of *C. fumipennis* in the region is lacking. From 2011 to 2014, we conducted searches at 155 sites and located *C. fumipennis* aggregations at 25% ($n = 39$) of these sites; 36% ($n = 14$) of these were located at forest harvests, an aggregation habitat not previously reported in the literature. We collected 1,559 buprestids representing 35 species from 2 aggregations in Louisiana between May and Aug 2012. Buprestid collections at these aggregations and observations of *C. fumipennis* activity at a 3rd aggregation indicated the number of buprestid species and individuals collected declined significantly from May to Jul. We collected significantly more *Agrilus difficilis* Gory (Coleoptera: Buprestidae) in the afternoon than morning hours and observed similar diurnal patterns among other buprestid species during the early weeks following aggregation activation. We also discuss evidence suggesting a portion of the regional *C. fumipennis* population is bivoltine. Although *A. planipennis* was not collected during this study, our results suggest that *C. fumipennis* is a feasible sampling tool and a useful addition to ongoing emerald ash borer surveys in the region.

Key Words: biosurveillance; Buprestidae; *Agrilus planipennis*; Coleoptera

Resumen

El barrenador esmeralda del fresno no nativo, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae), amenaza la extirpación de fresnos susceptibles (especies de *Fraxinus*; Lamiales: Oleaceae) en América del Norte. Se utiliza *Cerceris fumipennis* Say (Hymenoptera: Crabronidae), una avispa que hace su nido en la tierra y se alimenta de los Buprestidae en el este de América del Norte, como una herramienta para monitorear el barrenador esmeralda del fresno en el noreste de los EE.UU y Canadá. La reciente detección del barrenador esmeralda del fresno en Louisiana ofrece la oportunidad de complementar el monitoreo del barrenador en trampas con el uso de *C. fumipennis*, pero el conocimiento de *C. fumipennis* en la región es insuficiente. Desde el 2011 hasta el 2014, se realizaron búsquedas en 155 sitios, y agregaciones de *C. fumipennis* fueron encontradas en el 25% ($n = 39$) de estos sitios; el 36% ($n = 14$) de ellas estaban ubicadas en las cosechas forestales, un hábitat de agregación que no han sido reportado anteriormente en la literatura. Se recolectaron 1,559 buprestidos que representan 35 especies de 2 agregaciones en Louisiana entre mayo y agosto del 2012. Las colecciones de buprestidos en estas agregaciones y observaciones de la actividad *C. fumipennis* en la tercera agregación indican que el número de especies e individuos de buprestidos recolectados disminuyó significativamente de mayo a julio. Recolectamos significativamente más *Agrilus difficilis* Gory (Coleoptera: Buprestidae) en la tarde que en la mañana y observamos patrones diurnos similares en otras especies de buprestidos durante las primeras semanas después de la activación de las agregaciones. También discutimos la evidencia que sugiere que una parte de la población regional del *C. fumipennis* es bivoltina. Aunque *A. planipennis* no fue recolectado durante este estudio, nuestros resultados sugieren que *C. fumipennis* es una herramienta de muestreo factible y una adición útil para el sondeo del barrenador esmeralda del fresno en curso en la región.

Palabras Clave: biovigilancia; Buprestidae; *Agrilus planipennis*; Coleoptera

The accidental introduction of the Palearctic emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae), to North America in the 1990s has since resulted in the death of millions of ash trees (*Fraxinus* species; Lamiales: Oleaceae) in the eastern U.S., and near extirpation of some ash species in southeastern Michigan and northern Ohio (Herms & McCullough 2014). Emerald ash borer popula-

tions now occur in at least 25 U.S. states (APHIS 2015a; LDAF 2015) and the Canadian provinces Quebec and Ontario (CFIA 2015a), and the pest continues to spread each year by natural and anthropogenic means. Native wild ash populations hold significant ecological, cultural, and economic value, and because of their tolerance of a broad range of growing conditions and aesthetic properties, several ash species com-

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monly are planted in urban areas (Poland & McCullough 2006). Common ash species that occur in forested areas of eastern North America, such as green ash (*F. pennsylvanica* Marshall) and white ash (*F. americana* L.), are among *Fraxinus* species preferred by and susceptible to the emerald ash borer (Anulewicz et al. 2008); these species are found frequently in the southern U.S. where the emerald ash borer is only now becoming established.

Early detection of the emerald ash borer is paramount to the establishment of effective quarantines that restrict the movement of ash wood products and aid in slowing expanding emerald ash borer populations. Prior to the arrival of the emerald ash borer in North America, little was known of its chemical ecology or that of its congeners (Crook & Mastro 2010). It now is understood that the emerald ash borer depends largely on volatiles and visual cues to locate hosts and uses visual cues and short-range sex pheromones to locate mates (Crook & Mastro 2010; Silk & Ryall 2015). The lack of economic importance of most native Buprestidae has precluded the need for conducting such basic chemical ecology research, and thus little is known about them. Surveys in the U.S. for the emerald ash borer consist of purple prism traps and green multifunnel traps baited with host volatiles and suspended from the canopy of ash trees (APHIS 2015b), and the Canadian Food Inspection Agency uses a combination of green prism traps baited with host volatiles, visual surveys for symptomatic ash trees, and an ash branch sampling method for detection of the emerald ash borer in asymptomatic trees (Ryall et al. 2011; CFIA 2015b). Instead of relying solely on the currently available survey tools for the emerald ash borer, Marshall et al. (2005) proposed employing the predatory digger wasp *Cerceris fumipennis* Say (Hymenoptera: Crabronidae) to augment traditional emerald ash borer and other buprestid survey efforts.

Cerceris fumipennis is active during much of the emerald ash borer adult flight period and occurs from eastern Canada west to the Rocky Mountain Region and south to Texas and Florida (Scullen & Wold 1969), overlapping with the distribution of many common ash species and making it an ideal candidate for “biosurveillance” of the emerald ash borer. It is a ground-nesting species often found in aggregations in well-drained soils on open, sparsely vegetated sites such as sports fields, and the aggregations range in size from 2 to over 500 nests, although most contain fewer than 50 nests (Nalepa et al. 2012; Careless et al. 2014). Emergence and nest construction within aggregations commences in the late spring to early summer, depending on latitude, and whereas *C. fumipennis* is univoltine in the northern limits of its range, a partial or full 2nd generation is believed to occur in Florida and western Texas (Evans 1971; Mueller et al. 1992). Upon locating their prey, female wasps sting and usually successfully paralyze the adult beetle and carry it back to their nests by grasping the beetle using their legs and/or mandibles, and by sometimes “tucking” the beetle’s abdomen into a groove on their 5th abdominal sternum, a morphology unique to buprestid-seeking *Cerceris* species and not found in other species groups (Krombein 1981; Mueller et al. 1992; Nalepa & Swink 2015). Typically, a single female occupies a nest (Evans 1971; Evans & Rubink 1978), but ownership of nests within aggregations is a dynamic process. By marking and observing *C. fumipennis* at aggregations, nest usurpation was commonly observed, and occasionally a small portion of nests within an aggregation were shared by females but not provisioned with prey (Mueller et al. 1992) or, conversely, were provisioned with prey (Kurczewski & Miller 1984). This degree of sociality appears to occur infrequently among *C. fumipennis*, whereas other *Cerceris* species commonly cooperate in provisioning nests with prey (Alcock 1975; Alexander & Asis 1997).

Cerceris fumipennis does not sting humans, and thus it can be intercepted to collect their buprestid prey by using sweep nets, which startle females and cause them to drop their prey, and by creating nest collars, which slow or prevent the wasp from entering the nest with

its prey so that researchers can collect it (Careless et al. 2014). Using variations of these techniques, buprestid beetles (and rarely other insect families) have been collected for decades (Grossbeck 1912; Cartwright 1931; Scullen & Wold 1969; Evans 1971; Kurczewski & Miller 1984; Rutledge et al. 2011; Careless et al. 2014). At least 105 species of Buprestidae have been recorded as prey of *C. fumipennis* (Scullen & Wold 1969; Rutledge et al. 2011, 2013; Swink et al. 2013; Careless et al. 2014; Westcott & Thomas 2015). These collections have resulted in the addition of new state records and dozens of previously unrepresented Buprestidae species to regional insect museum collections (Hellman & Fierke 2014; Westcott & Thomas 2015). Nalepa et al. (2015) collected 5 times more buprestid species and nearly 10 times more total buprestid beetles from *C. fumipennis* aggregations than from the standard purple prism traps (APHIS 2015b) placed at the edge of forests surrounding these aggregations. Although a direct comparison between using *C. fumipennis* and traps and other means of detection is not possible, this work supports the notion of combining these various methods to detect Buprestidae, including the emerald ash borer.

In light of its efficiency in detecting buprestids and the accessibility of its aggregations to human collectors, protocols to use *C. fumipennis* as an emerald ash borer detection tool have been developed (Careless 2009a; Carrier & Jackson 2012) and used successfully by numerous volunteer groups in many areas for the detection of the emerald ash borer and other uncommonly collected Buprestidae in the northeastern U.S. (Bohne et al. 2011; Rosenholm 2012; Rutledge et al. 2013). However, their utility for biosurveillance of the emerald ash borer and other Buprestidae in the western Gulf Region of the U.S. is unknown. In an attempt to evaluate the potential of *C. fumipennis* in this region, we conducted an extensive search for *C. fumipennis* aggregations in Louisiana and eastern Texas. Once aggregations were located, we documented the annual initiation of aggregation activity at a subset of known aggregations. We assessed the daily patterns of foraging behavior at a small aggregation and intensively sampled prey items returned to 2 large aggregations to measure buprestid abundance (defined here as the number of prey items collected from *C. fumipennis*, not to be confused with actual Buprestidae abundance in the environment) and richness (number of species). Taken together, these data were collected to determine the optimal timing of sampling efforts, something not previously reported on *C. fumipennis* in this region.

Materials and Methods

AGGREGATION LOCATION AND ANNUAL INITIATION OF AGGREGATION ACTIVITY

Selection of habitat to search for aggregations was based on local knowledge and the use of Google Earth (Google Inc. 2009) to identify likely habitats such as baseball diamonds, parks, and other flat, open, non-vegetated areas. To document aggregation initiation, 3 aggregations located in 2011 were visited weekly beginning in Apr of each year (2012–2014) and the date of first activity was noted.

SEASONAL ABUNDANCE AND DIVERSITY OF BUPRESTID PREY AT STUDY SITES

Two relatively large aggregations were used to evaluate seasonal and daily Buprestidae abundance and diversity, namely the Highland Road Park (HRP) aggregation ($n > 500$ nests), located on a soccer field in Baton Rouge (30°20'55.37"N, 91°4'28.42"W), Louisiana, and the Verda Elementary School playground (VESP) aggregation ($n = 374$ nests), in

Verda (31°41'59.26"N, 92°46'19.74"W), Louisiana. To estimate daily activity patterns, 1 person at HRP and 2 people at VESP used a sweep net to collect as many wasps laden with prey as possible, twice daily from 9:30 to 11:00 AM and from 1:00 to 2:30 PM. To estimate seasonal activity patterns, the above described morning and afternoon samples were collected twice per week starting soon after the initiation of aggregation activity until most activity at the aggregation had ceased (VESP aggregation, 21 Jun 2012; HRP aggregation, 17 Jul 2012). All specimens were identified by 1 of the authors (T.C.M.) and are deposited in the Louisiana State Arthropod Museum, Louisiana State University in Baton Rouge, Louisiana.

To reduce effects on wasp behavior due to sampling prey items, the Verda Ballfield aggregation (VB; $n = 12$ nests) located at the cleared area at the home plate of an unused public baseball diamond in Verda (31°41'55.97" N, 92°46'29.07"W), Louisiana, was monitored passively and prey not taken from wasps. All nests were permanently labeled using a nail and aluminum tag (but wasps were not marked), and a single observer was stationed where all nests could be observed to record times of wasp departure and arrival, presence/absence of prey returned to nest, and any additional noteworthy occurrences such as presence of males, evidence of potential nest usurpation, or sharing between wasps. Monitoring was conducted twice per week from 9:30 to 11:00 AM and from 1:30 to 2:30 PM from 9 May 2012 until adult wasps were no longer active (2 Jul 2012).

STATISTICAL ANALYSES

All analyses were conducted using the Proc GLM procedure in SAS (Version 8; SAS Institute 2009). Validity of ANOVA assumptions for raw and transformed data were checked by visual assessment of residual plots, and prey counts were log or square root transformed to reduce variance heterogeneity where necessary. Subsequent pair-wise comparisons of least squares means were tested using the Tukey–Kramer procedure (SAS Version 8; SAS Institute 2009). A significance level of 0.05 was used in all statistical tests performed.

SEASONAL AND WITHIN-DAY ABUNDANCE AND SPECIES RICHNESS OF BUPRESTID PREY AT VESP AND HRP

An ANOVA of the total number of buprestid species (richness) and the total buprestid abundance was conducted separately for the VESP and HRP aggregations. Additional separate analyses were conducted on the total abundance of common species that met the criterion that no more than 1 collection effort totaled zero for the given species. Species analyzed included *Buprestis lineata* F. and *B. maculipennis* Gory

(VESP); and *Agrilus macer* LeConte, *A. difficilis* Gory, and *B. rufipes* Olivier (HRP).

VESP Analyses. Analyses of prey beetles collected at the VESP aggregation were conducted as a split-plot, where time period was considered the whole plot factor, day within period constituted whole plot error, and the time of day (AM vs. PM) was the subplot factor. The sampling period was subdivided into 3 periods to account for a systematic decrease in prey collection; period 1 (18–25 May 2012; 3 AM and PM sampling events), period 2 (29 May to 6 Jun 2012; 3 AM and PM sampling events), and period 3 (15 Jun to 1 Jul 2012; 4 AM and PM sampling events).

HRP Analyses. Frequent rain events at the HRP aggregation interrupted several planned sampling events; thus, morning and afternoon sampling observations did not always occur on the same day. Analyses of prey beetles collected at the HRP aggregation were analyzed as a 2-factor ANOVA, with factors time of day and period, and with the error used for tests given by day within period and time of day. The HRP collections were split into 2 periods to account for a systematic change in prey numbers, with period 1 representing prey collections from 14 May to 5 Jun 2012 (4 AM and 4 PM sampling events) and period 2 representing the collections from 14 Jun to 17 Jul 2012 (5 AM and 6 PM sampling events).

VB Aggregation: Analysis of Total Buprestids. Analysis of total buprestids observed returned to nests (totaled over all nests) at the VB aggregation was conducted as a split-plot with the error structure as described for the analysis of VESP. Collection dates were divided into 2 periods consisting of 3 collection days each to account for the systematic decrease in buprestid abundance; period 1 consisted of collections from 9 to 17 May 2012, and period 2 from 22 to 30 May 2012. Activity at the aggregation decreased drastically after this point; only 4 nests remained active in Jun.

Results

AGGREGATION LOCATION AND INITIATION OF AGGREGATION ACTIVITY

We searched 155 sites from 2011 to 2014 during times of the year when *C. fumipennis* was known to be active locally, and located nests occupied by *C. fumipennis* at 39 sites, yielding a 25% total search success rate. More than a third of these aggregations were found in the open areas at log loading sites following forest harvesting operations (Table 1). A remarkably large aggregation numbering in excess of 500 nests (HRP) was located on a soccer field in 2011, and a 2nd large ag-

Table 1. *Cerceris fumipennis* aggregation site description and size categories (number of nests) of aggregations located in Louisiana and eastern Texas, 2011 to 2014 ($n = 155$ total sites searched).

| Site type | Site category | | | | Total aggregations | Percentage of total |
|-------------------------|----------------|-------|-------|-----|--------------------|---------------------|
| | >50 | 25–50 | 15–24 | <15 | | |
| Forest harvesting sites | 0 | 3 | 0 | 11 | 14 | 36% |
| Playground | 1 ^a | 0 | 2 | 8 | 11 | 28% |
| Baseball diamond | 0 | 1 | 1 | 4 | 6 | 15% |
| Parking area | 0 | 1 | 0 | 3 | 4 | 10% |
| Other sports fields | 1 ^b | 1 | 0 | 0 | 2 | 5% |
| Vacant lot | 0 | 0 | 0 | 2 | 2 | 5% |
| Total | 2 | 6 | 3 | 28 | 39 | |
| Percentage of total | 5% | 15% | 8% | 72% | | |

^aVerda Elementary School playground, Verda, Louisiana; approx. 374 nests

^bHighland Road Park, Baton Rouge, Louisiana; >500 nests

gregation of 374 nests (VESP) was located on a playground in 2012; however, 80% of all aggregations found contained less than 25 nests (Table 1). After a chance encounter with a small *C. fumipennis* aggregation at a recent forest harvest site in 2010, a more extensive search conducted in 2012 at 32 separate log loading sites associated with 6 large forest harvests (clearcuts) resulted in a 48% aggregation location success rate. Anecdotal evidence suggested these aggregations were found primarily on sandy loam soils.

Weekly monitoring of aggregations in central and southern Louisiana (2012–2015) indicated that the aggregations became active from late Apr (2012) to late May (2013–2015). In 2012–2013 and 2013–2014, Louisiana experienced colder winters than in 2011–2012, which may explain the delayed activity in the latter years. The spring of 2015 was exceedingly wet; this may explain the later aggregation activation in 2015. The following observations at the VB aggregation and prey sampling at the HRP and VESP aggregations indicate there may be a partial 2nd generation in central Louisiana and possibly more in southern Louisiana. Based on prey sampling in central Louisiana, wasp activity dropped off and nearly ceased approximately 1 mo following first observation of aggregation activity before a lesser peak was observed during the 2nd month of activity, whereas activity at the HRP aggregation persisted into late Jul (Fig. 1). Additionally, male emergence, pursuit of females, mating, and the appearance of 7 new nests were ob-

served between 30 May and 8 Jun 2012 at the VB aggregation. A small portion of the new nests persisted for 11 d, but only 1 wasp returning with a buprestid prey item was observed before the nests became inactive (filled in with soil). The last *C. fumipennis* observed entering a nest at the VB aggregation with prey was on 14 Jun 2012, and the last observation of a wasp was recorded on 26 Jun 2012. All nests at the VB aggregation appeared filled with soil and inactive by 2 Jul 2012. Neither the VESP nor the VB aggregation was checked routinely for the remainder of 2012. The HRP aggregation was nearly inactive by mid-Aug; the last buprestid prey item ($n = 1$) was collected on 21 Aug 2012, after which no further observations of the aggregation were made.

SEASONAL AND WITHIN-DAY ABUNDANCE AND DIVERSITY OF BUPRESTID PREY AT VESP AND HRP

Thirty-five species of Buprestidae, totaling 1,559 specimens, and 1 species of Chrysomelidae, *Neoclamisus* sp., ($n = 7$ specimens) were collected from the HRP and VESP aggregations in 2012 (Table 2). Beetle capture rates peaked at 1.96 beetles per collector per minute on 15 May at HRP and at 0.24 beetles per collector per minute on 23 May at the VESP aggregation. Among the buprestid species collected, *Actenodes davidi* Nelson (from HRP) and *Chrysobothris caddo* Wellso and Manley (from HRP and VESP), have not previously been reported as

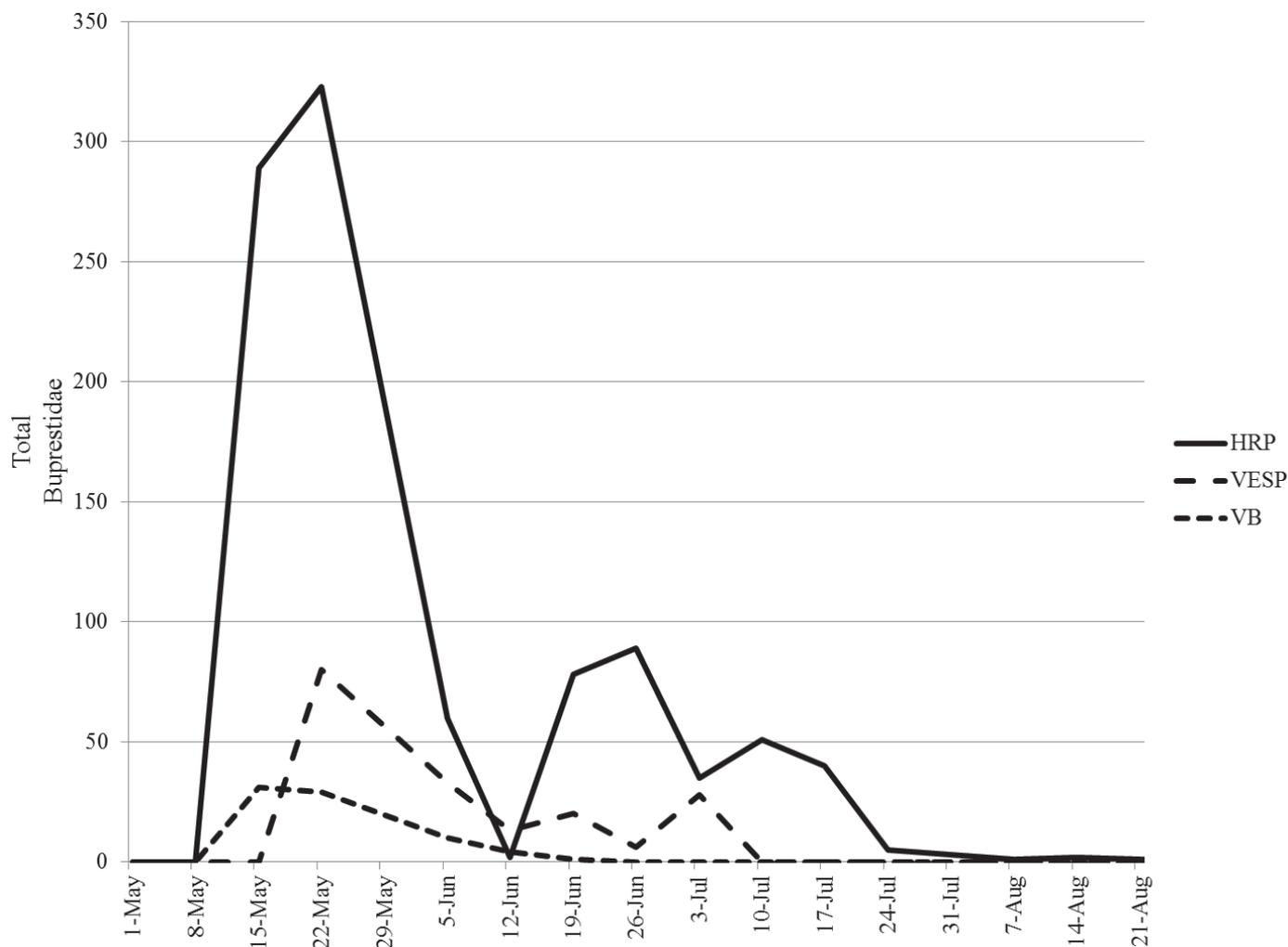


Fig. 1. Total Buprestidae prey taken per day from *Cerceris fumipennis* at aggregations located at Highland Road Park, Baton Rouge, Louisiana (HRP), Verda Elementary School playground (VESP) and observed at Verda Ballfield (VB), Verda, Louisiana, May to Aug 2012. Days reflecting zero beetles collected were not included in the ANOVA.

Table 2. Numbers of Buprestidae and Chrysomelidae collected at 2 large *Cerceris fumipennis* aggregations in Louisiana, May to July (Verda Elementary School playground, VESP) and May to Aug (Highland Road Park, HRP) 2012.

| Family and species | HRP | VESP |
|---|--------------|------------|
| Buprestidae | | |
| <i>Acmaeodera pulchella</i> (Herbst) | 0 | 2 |
| <i>Acmaeodera tubulus</i> (F.) | 0 | 1 |
| <i>Actenodes acornis</i> (Say) | 49 | 3 |
| <i>Actenodes davidi</i> Nelson ^a | 34 | 0 |
| <i>Agrilus arcuatus</i> (Say) | 5 | 0 |
| <i>Agrilus bilineatus</i> (Weber) | 11 | 0 |
| <i>Agrilus difficilis</i> Gory | 210 | 0 |
| <i>Agrilus fallax</i> Say | 8 | 0 |
| <i>Agrilus lecontei lecontei</i> Saunders | 2 | 0 |
| <i>Agrilus macer</i> LeConte | 532 | 0 |
| <i>Agrilus obsoleteguttatus</i> Gory | 2 | 0 |
| <i>Agrilus quadriguttatus quadriguttatus</i> (Gory) | 76 | 0 |
| <i>Brachys ovatus</i> (Weber) | 6 | 0 |
| <i>Buprestis apricans</i> Herbst ^b | 0 | 1 |
| <i>Buprestis consularis</i> Gory | 0 | 43 |
| <i>Buprestis lineata</i> F. | 0 | 96 |
| <i>Buprestis maculipennis</i> Gory | 0 | 121 |
| <i>Buprestis rufipes</i> Olivier | 139 | 48 |
| <i>Buprestis striata</i> F. | 0 | 1 |
| <i>Chrysobothris caddo</i> Wellso and Manley ^a | 10 | 5 |
| <i>Chrysobothris cribraria</i> Mannerheim | 2 | 7 |
| <i>Chrysobothris dentipes</i> (Germar) | 0 | 15 |
| <i>Chrysobothris quadriimpressa</i> (Gory & Laporte) | 0 | 9 |
| <i>Chrysobothris scitula</i> Gory | 1 | 0 |
| <i>Chrysobothris sexsignata</i> Say | 1 | 0 |
| <i>Chrysobothris shawnee</i> Wellso and Manley | 21 | 25 |
| <i>Chrysobothris viridiceps</i> Melsheimer | 0 | 5 |
| <i>Dicerca lurida</i> (F.) | 20 | 14 |
| <i>Dicerca obscura</i> (F.) | 2 | 0 |
| <i>Dicerca punctulata</i> Schönherr | 0 | 5 |
| <i>Poecilnota cyanipes</i> (Say) | 1 | 0 |
| <i>Poecilnota thureura</i> (Say) | 21 | 0 |
| <i>Spectralia gracilipes</i> (Melsheimer) | 3 | 0 |
| <i>Texania campestris</i> (Say) | 0 | 1 |
| <i>Xenorhipis brendeli</i> LeConte | 2 | 0 |
| Chrysomelidae | | |
| <i>Neochlamisus</i> sp. | 7 | 0 |
| Total | 1,165 | 401 |

^aNot previously reported as collected by *C. fumipennis* (Scullen & Wold 1969; Careless 2009b; Paiero et al. 2012; Swink et al. 2013; Hellman & Fierke 2014; Westcott & Thomas 2015)

^b*Buprestis apricans* collected 15 May 2012 prior to study initiation.

buprestid prey species, raising the list of buprestid species known to be taken by *C. fumipennis* to 107 (Scullen & Wold 1969; Careless 2009b; Paiero et al. 2012; Swink et al. 2013; Hellman & Fierke 2014; Westcott & Thomas 2015).

Analyses of total prey abundance and species richness did not indicate a significant interaction between the main effects period and time of day at either the HRP or the VESP aggregations. However, we collected significantly more total buprestids in the early sampling period compared with the later period(s) at both the VESP ($F = 16.64$; $df = 2, 7$; $P = 0.002$) and HRP ($F = 20.32$; $df = 1, 15$; $P < 0.001$) aggregations (Table 3). Furthermore, significantly more species were represented in collections from the early sampling period compared with later dates at each aggregation (VESP: $F = 12.95$; $df = 2, 7$; $P = 0.004$; HRP: $F = 7.49$; $df = 1, 15$; $P = 0.015$; Table 3). Although analyses of total prey

abundance indicated there was no significant difference between the AM and PM sampling times, there was some evidence that analyses of total buprestid abundance masked individual species collection patterns. We collected significantly more *A. difficilis* ($F = 6.40$; $df = 1, 15$; $P = 0.023$; Fig. 2) and observed a similar but non-significant pattern in *B. rufipes* collection (Fig. 3) in the afternoon than in the morning during the days soon after the HRP aggregation became active; however, later in the season, there was little difference in collections between AM and PM sampling. We collected significantly more *A. macer* at HRP ($F = 16.22$; $df = 1, 15$; $P = 0.001$) and *B. maculipennis* at VESP ($F = 9.54$; $df = 2, 7$; $P = 0.010$) in the early periods of aggregation sampling (Table 3), and although afternoon collections were greater during the early collection period, abundance did not differ significantly between AM and PM collection times.

VB AGGREGATION: ANALYSIS OF TOTAL BUPRESTIDS

Although analyses of total buprestids returned to the passively observed VB aggregation did not indicate significant differences between periods (averaged over time of day) or between AM and PM collections (averaged over periods), the data suggested a weak interaction between the time of day and the phenology of aggregation activity ($F = 5.54$; $df = 1, 4$; $P = 0.078$). Generally, more buprestids were collected earlier than later in the season ($F = 4.92$; $df = 1, 4$; $P = 0.091$), and observed means suggested larger catches during the afternoon compared with morning efforts in the early season period, similar to the trend seen at the HRP and VESP aggregations. However, *a priori* simple tests comparing AM and PM collections within each period were not significant, and a larger study with a greater number of sampling dates would be needed to confirm this tendency.

Discussion

Cerceris fumipennis is a widely distributed species that preys almost exclusively on beetles in the family Buprestidae and has been proven as a valuable complement to other buprestid collection methods in the eastern U.S. and Canada. This study is the first to report the results of field observations on *C. fumipennis* habitat characteristics and diurnal and seasonal effects on wasp activity and buprestid prey diversity and abundance in Louisiana, USA. The use of *C. fumipennis* to collect Buprestidae in Louisiana also resulted in the collection of 2 species of Buprestidae not previously known as *C. fumipennis* prey (Scullen & Wold 1969; Careless 2009b; Paiero et al. 2012; Swink et al. 2013; Westcott & Thomas 2015).

In Louisiana and eastern Texas, adult *C. fumipennis* became active after the foliage had flushed in the native trees and woody shrubs, and observations in 2014 and 2015 indicated activity overlapped with the blooming of the mimosa tree (*Albizia julibrissin* Durazz.; Fabales: Fabaceae) and *Gardenia* (Gentianales: Rubiaceae) bush, information that could aid in the timing of searches for *C. fumipennis* aggregations in the region. Our observations of aggregation habitat characteristics and daily activity are in general agreement with those previously reported (Evans 1971; Nalepa et al. 2012; Careless et al. 2014). Our overall success rate in locating aggregations at a variety of habitats (25%) was slightly greater than that reported by Nalepa et al. (2012) (22.5%), but these authors restricted their search to ball diamonds. Our location of numerous aggregations at forest harvest sites increased our success rate; and although this is evidence of a potentially reliable category of site type to search for *C. fumipennis*, unfortunately the aggregations at these sites did not persist the following year. All of the log-loading sites were prepared for replanting the following season using pre-

Table 3. Seasonal variation in total buprestid prey abundance, species richness, and the abundance of common buprestid species collected at the Verda Elementary School playground (VESP) and Highland Road Park (HRP) *Cerceris fumipennis* aggregations in central and southern Louisiana, May to Jul 2012. *F*-test results for the main effect period, and the mean counts by period (± 1 standard error), are reported.

| Site | Response variable | Test of period ¹ | Early | Mid | Late |
|------|-------------------------------|-------------------------------|-------------------------------|------------------|------------------|
| VESP | Total prey ² | $F_{2,7} = 16.64; P = 0.002$ | 39.8 \pm 2.9 A ³ | 17.7 \pm 4.3 B | 6.6 \pm 2.6 B |
| | Species richness ² | $F_{2,7} = 12.95; P = 0.004$ | 9.5 \pm 1.2 A | 6.0 \pm 0.6 AB | 3.4 \pm 0.5 B |
| | <i>Buprestis lineata</i> | $F_{2,6} = 2.99; P = 0.115$ | 8.0 \pm 1.5 A | 5.2 \pm 1.9 A | 2.1 \pm 0.4 A |
| | <i>B. maculipennis</i> | $F_{2,6} = 9.54; P = 0.010$ | 13.0 \pm 1.9 A | 4.8 \pm 1.4 AB | 1.8 \pm 0.3 B |
| HRP | Total prey ⁴ | $F_{1,15} = 20.32; P < 0.001$ | 106.6 \pm 22.2 A | NA ⁵ | 26.5 \pm 3.3 B |
| | Species richness ⁴ | $F_{1,15} = 7.49; P = 0.015$ | 10.5 \pm 1.5 A | NA | 6.5 \pm 0.3 B |
| | <i>B. rufipes</i> | $F_{1,15} = 9.65; P = 0.007$ | 11.1 \pm 2.7 A | NA | 4.2 \pm 1.1 B |
| | <i>Agrilus difficilis</i> | $F_{1,15} = 26.55; P < 0.001$ | 18.9 \pm 3.9 A | NA | 4.9 \pm 0.5 B |
| | <i>A. macer</i> | $F_{1,15} = 16.22; P < 0.001$ | 49.4 \pm 12.1 A | NA | 11.8 \pm 1.9 B |

¹Based on mixed model ANOVA *F*-test comparing sampling periods (averaged over AM and PM) over the course of the study.

²Mean total buprestid prey abundance and species richness collected by sampling period at VESP.

³Differing letters indicate significant differences (based on Tukey's test) within a row only. Analyses based on LSMEANS of square root-transformed values. Raw means (\pm standard error) are presented.

⁴Mean total buprestid prey abundance and species richness collected by sampling period at HRP.

⁵Mid-season not applicable; only 2 periods distinguished at HRP.

scribed fire, and in some cases, the soil was plowed into raised beds; this severe disturbance may explain the lack of aggregation activity the following year.

Whereas aggregations in the northern U.S. appear to be univoltine, aggregations in Louisiana may have a partial (and perhaps full) 2nd generation, as suggested by others in Florida (Kurczewski & Miller 1984) and southern Texas (Hook & Evans 1991). Based on prey collections and close observation of the VB aggregation, the initial period of wasp activity (i.e., the 1st generation) lasted 5 to 6 wk. Careless et al. (2014) reported a similar period of activity for the single generation in the northeastern U.S. The appearance of a few new nests at the VB and VESP aggregations, a small increase in observed prey return after this time period, and the observation of wasps as late as 26 Jun (9 wk following aggregation activation) at VB suggests a partial 2nd generation may have taken place. Prey collections at HRP support these observations; collections declined to zero over a roughly 1 mo period ending 12 Jun before a 2nd, smaller peak in abundance that lasted for another 5 to 6 wk (Fig. 1). Further evidence is the location of dozens of new nests (based on tumulus) at highly active aggregations at multiple forest harvest sites with friable soils (easy to establish new nests) in central Louisiana in early Jul when less than 10% of the initial aggregation nests at

VB and VESP remained active. The harvest sites would have been heavily vegetated the previous summer, suggesting that there would not have been an aggregation at these sites the previous year, but rather that the daughters or expelled females who had their original nests usurped (Mueller et al. 1992) at other aggregations in the area utilized these sites. Activity at the VESP and VB aggregations ceased in early Jul; however, activity continued into early Aug at the HRP aggregation. Although these observations could have been the result of variations in buprestid prey availability or a prolonged 1st generation of *C. fumipennis* due to dry weather conditions at VESP and heavy rainfall near HRP, they may also reflect a partial multivoltine reproductive strategy.

Careless et al. (2014) illustrated that marked wasps could successfully relocate their aggregation after being released up to 2 to 3 km from nests, which suggests the foraging range of individuals could be similar. This potential foraging radius (and the greater number of nests) may partly explain the greater number of buprestid species in collections from the HRP compared with the VESP aggregation. Although we did not specifically test the hypothesis, it could be that the differing flora surrounding the 2 aggregations played a role in the observed buprestid prey differences. HRP is located along a ridge that borders the Mississippi River alluvial floodplain, and foraging wasps could easily

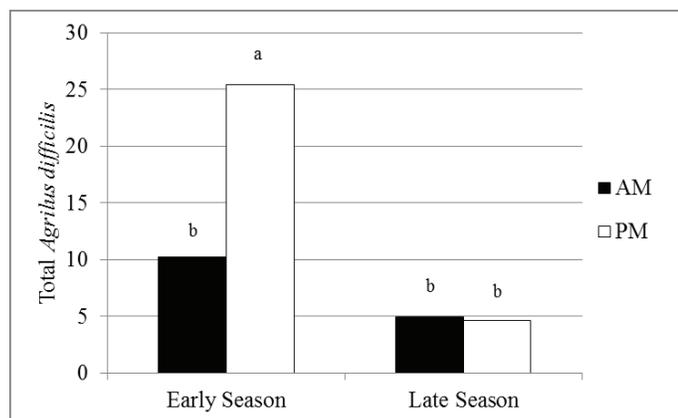


Fig. 2. Mean numbers of *Agrilus difficilis* taken from a *Cerceris fumipennis* aggregation at Highland Road Park, Baton Rouge, Louisiana (HRP), from 9:30 to 11:00 AM and from 1:00 to 2:30 PM, from 14 May to 5 Jun 2012 (early season) and from 14 Jun to 17 Jul 2012 (late season). Means without a letter in common differ significantly based on Tukey's test on square root-transformed counts.

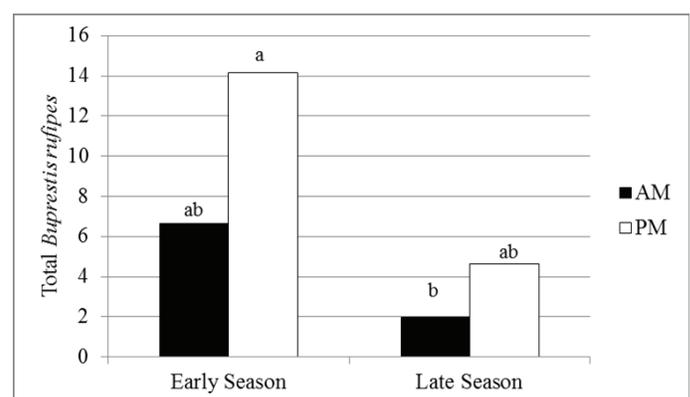


Fig. 3. Mean *Buprestis rufipes* taken from a *Cerceris fumipennis* aggregation at Highland Road Park, Baton Rouge, Louisiana (HRP), from 9:30 to 11:00 AM and from 1:00 to 2:30 PM, from 14 May to 5 Jun 2012 (early season) and from 14 Jun to 17 Jul 2012 (late season). Means without a letter in common differ significantly based on Tukey's test on square root-transformed counts.

reach prey found on the diverse hosts growing in these 2 very different soil types and moisture regimes. Such habitat differences leading to increased buprestid species richness has been reported by others (Swink et al. 2013; Hellman & Fierke 2014). The VESP aggregation is located along a ridge dominated by *Pinus taeda* L. (Pinales: Pinaceae) and a few hardwood species adapted to drier soils, and the total woody species diversity is lower than that at HRP. Other evidence suggests wasps will forage no further than necessary, given the investment made in carrying prey back to a nest (Nalepa et al. 2013), and some have reported wasps collecting many individuals of the same species, suggesting wasps may exploit an abundant food source (Swink et al. 2013; Careless et al. 2014). The lower prey diversity at VESP compared with that at HRP could be explained by such behavior, given that the most abundant prey species collected there, *B. lineata* and *B. maculipennis*, commonly colonize pine species, of which there are many in the immediate vicinity of the VESP aggregation. These differences all likely contribute to the fact that only 6 of the 34 buprestid species were collected at both aggregations. This emphasizes the importance of the location of aggregations, because the surrounding habitat will determine the buprestid community sampled by *C. fumipennis*.

Our observations on the daily habits of foraging wasps mostly agree with those reported by Careless et al. (2014), who found buprestid collection to be greatest near noon and just after midday. Our study sampled shorter time intervals, with a pause in collection and observation between 11:00 AM and 1:00 PM, but we observed trends toward greater prey collection in the afternoon sampling efforts, and this was more pronounced in the early period following aggregation activation. Greater activity in the afternoon could be due to the general diurnal abundance of Buprestidae. Although it may not be a behavior shared by all Buprestidae, Jennings et al. (2014) reported emerald ash borer adults were most active in the crowns of ash trees on sunny days between 11:00 AM and 3:00 PM. It is difficult to imagine that wasps would forage during the hotter portion of the day, requiring greater energy costs than flight during morning hours, if there were not significant benefits in prey acquisition.

Careless et al. (2014) found that intercepting and removing prey from wasps did not alter wasp foraging behavior on subsequent days. Our comparison of the observed VB and intensively sampled VESP and HRP aggregations is imperfect, but the similar diurnal and seasonal prey abundance patterns among the aggregations support the resilience of wasps when faced with sampling interference. Collectors should be wary of sampling efforts at smaller aggregations where sampling interference will likely have more pronounced effects on the aggregation in subsequent years. At these aggregations, prey items should be returned to the female's nest following identification of intercepted prey (Careless et al. 2014).

Perhaps the most important factor to consider when planning buprestid collecting efforts at a *C. fumipennis* aggregation is the effect of phenology. Regardless of the time of day when the effort is made, sampling during the first 2 wk of aggregation activity will result in greater numbers and diversity of prey items. Surveys will likely be most successful and cost effective when conducted soon after aggregation initiation and concentrated in the midday to early afternoon hours.

An extension of this study was the enlistment over the summers of 2013 to 2014 of members of 3 Texas Master Naturalist Program Chapters to monitor and collect buprestids at 3 aggregations located in eastern Texas, as has been done with similar groups in the northern U.S. (Bohne et al. 2011; Teerling 2012; Rutledge et al. 2013). Although these efforts did result in buprestid collections (but no emerald ash borer), there were few beetles collected due to challenges in locating aggregations of sufficient working size and gaining commitments from enough Chapter members to conduct surveys in the early days following aggrega-

tion initiation (when the efforts would be most productive). If these challenges can be overcome, involving local volunteer organizations in native and non-native Buprestidae sampling efforts could result in viable complementary surveys for species such as the emerald ash borer and serve as an aid to underfunded government agencies tasked with conducting emerald ash borer surveys. Such partnership also often aids such groups in satisfying the personal goals and interests of their members (Rosenholm 2012). Refining the knowledge of *C. fumipennis* life history in the southern U.S. and its use as a survey tool presents potential for continued contributions to science and for creating a public education and outreach tool regarding invasive insect species issues.

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