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ORIGINAL ARTICLE

Spatial distribution of *Vespa velutina* individuals hunting at domestic honeybee hives: heterogeneity at a local scale

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Abstract Since its recent introduction into Europe, the yellow-legged hornet, *Vespa velutina*, has become a major predator of the domestic honeybee, *Apis mellifera*, but little is known about its hunting behavior. We studied *V. velutina* hunting behavior by a capture-mark-recapture procedure in an experimental apiary. A total of 360 hornets were captured and tagged, and we determined: (i) the number of hornets visiting the apiary and the changes in time, (ii) the average number of individual visits per half-day and the time elapsed between consecutive recaptures, and (iii) the individual and global distribution of the hornets in the apiary. More than 50% of the marked hornets were recaptured at least once, this increased to 74% in considering the first marked individuals. We estimated 350 hornets visiting the patch daily with at least 1 visit per half-day. The number of marked hornets decreased over time while the number of unmarked ones increased, suggesting a turnover of individuals. The reduction of the delay between consecutive visits indicates that hornets became more efficient over time. Most of the hornets (88%) were recaptured in front of different hives but, overall, the global distribution was aggregative. Hornets were mainly recaptured in front of 1 hive which was neither the smallest nor the biggest colony, suggesting that the major cue used by hornets is not the amount of food. We hypothesize that the defensive behavior of the honeybee colony could explain our results which may be promising to further studies.

Key words *Apis mellifera*; capture-mark-recapture; learning; predation; Vespidae; yellow-legged hornet

Introduction

The eusocial vespids are considered to be opportunistic or generalist foragers, but may also be scavengers. They prey on diverse arthropods, carrion, or alternative protein sources such as meat or fish in open access stalls to feed their brood (Spradbery, 1973; Edwards, 1980; Matsuura & Yamane, 1990; Raveret Richter, 2000). Their food choice may be determined by prey quantity (Nakasuji *et al*., 1976), prey quality (Stamp & Meyerhoefer, 2004), or both (Stamp, 2001; Amrstrong & Stamp, 2003). The workers may revisit large prey, with small pieces from each visit taken back to the nest. In other words, workers may make several visits to resource-rich patches (Nakasuji *et al*., 1976; Raveret Richter & Jeanne, 1985; Richter, 2000; D'Adamo & Lozada, 2003; Amrstrong & Stamp, 2003). The assessment of patch size and quality and the decision to return to a site means that they must learn the relevant characteristics of patches. Previous studies have demonstrated the presence of such cognitive traits in the Vespidae (D'Adamo & Lozada, 2003; Toh & Okamura, 2003; Weiss *et al*., 2004; Lozada & D'Adamo,

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2006; Warkentin *et al*., 2006; Moreyra *et al*., 2012) and it has been argued that the plasticity of their cognitive traits could favor invasiveness in new environments (D'Adamo & Lozada, 2007, 2009, 2011; Lozada & D'Adamo, 2009, 2011).

The yellow-legged hornet, *Vespa velutina*, is an invasive species recently introduced into Europe from eastern China and observed for the first time in France in 2004 (Rortais *et al*., 2010). Like other *Vespa* species, a queen can produce thousands of individuals throughout the year, so abundant protein is required for feeding the brood; during spring to autumn several hundreds of hornets are present within the colony (see Monceau *et al.*, 2014). This explains the increase of predation pressure on domestic honeybees from July to November (Monceau *et al.*, 2013a, 2013b, 2014). In its area of origin, *V. velutina* preys on *Apis cerana* and the introduced European honeybee, *Apis mellifera.* Contrary to *A. cerana*, *A. mellifera* is more vulnerable to vespid predators, presumably because it did not coevolve with this hornet species (Ken *et al*., 2005; Tan *et al.*, 2007, 2010, 2012a, 2012b, 2013). In France, predation by *V. velutina* weakens native honeybee colonies toward the end of summer. For example, up to 20 hornets have been counted at the same time in front of single hives in our study areas (Monceau *et al.*, 2013b). When bee colonies are only slightly defended, hornets may enter the hives to obtain their dietary protein (from honeybee larvae) and carbohydrate (from honey). Consequently, apiaries represent an accessible and valuable dietary resource for *V. velutina* throughout the season.

While most published behavioral studies of this species have focused on differences in the defense behavior of *A. mellifera* and *A. cerana* when confronted with *V. velutina* (Ken *et al.*, 2005; Tan *et al.*, 2007, 2010, 2012a, 2012b, 2013), the hunting behavior of this hornet species has received little attention (Monceau *et al.*, 2013b). However, understanding the prey selection process and associated cues of *V. velutina* may represent significant information for the development and implementation of effective pest management strategies (Roitberg, 2007). To date, most of the behavioral observations on this invasive species are made by beekeepers who report differential level of attacks within an apiary between hives, suggesting that hornets may aggregate in front of particular hives. However, the relevance of these observations has not been assessed to date but represents a key parameter to understand *V. velutina* hunting behavior.

In this study, we proposed to describe quantitatively the hunting behavior of *V. velutina* (number of hornets and visits) and to assess the existence of an aggregation of *V. velutina* at a small spatial scale. We investigated these issues in an experimental apiary consisting of 6 homogeneous hives by performing a capture-mark-recapture (CMR) survey. Each captured hornet received a unique combination of colors which allows differentiating all individuals. Therefore, we worked at 2 levels: (i) in considering hornets individually to define their prey searching pattern, that is, if they focused on a specific hive or if they visited all hives within the apiary, and (ii) in considering all individuals visiting the hives to define the overall distribution of the hornets within the apiary, that is, if *V. velutina* aggregated in front of particular hives or if they are randomly distributed within the apiary. As some individuals performed several visits during the survey (see the results section), we also analyzed the time laps between recaptures to know if hornets may gain experience as a function of their increasing number of visits.

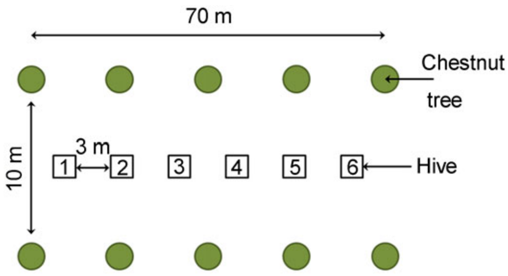

Fig. 1 The experimental apiary. (La Grande Ferrade, INRA Bordeaux-Aquitaine research centre, GPS: N 44°47′27.05″ W0°34′38.35″). The 6 hives (h1–h6) were equidistant and visually similar.

Materials and methods

Ethics statements

No permits were required for the described study, which complied with all relevant regulations.

Study site

The study was performed during August 2011, a period of dramatic increase of *V. velutina* predation in an experimental apiary of 9 beehives "La Grande Ferrade" installed in a 25-ha agricultural area that received no insecticide treatment (INRA, Villenave d'Ornon, GPS: N44°47′27.05″ W0°34′38.35″). We only considered 6 beehives (h1–h6), located in a single row of chestnut trees because they were in a close vicinity to each other (3 m apart, Fig. 1) and offered a homogenous site to test if the hornets visited all hives equally. All hives were painted with identical grey paint, and had the same shape and

Table 1 Number of frames in each hive that were fully covered by honeybees (maximum = 12) during spring (March 23), summer (July 22), and winter (December 1) of 2011. Hives were not opened in the autumn due to high predation pressure. The sample time in the present study was just after the summer visitation.

Hive	Spring	Summer	Winter
h1	6	11	Dead
h2	7	10	6
h3	9	11	3
h4	10	10	2
h5	9	10	6
h6	10	5	4

appearance. The 6 colonies were obtained from a professional beekeeper, and their activity and sanitary status were regularly monitored by counting the number of frames covered by honeybees (Table 1). At the time of the experiment, 5 beehives had 10 or 11 full frames and 1 beehive (h6) had 5 frames.

Marking technique

Several marking techniques are classically used to monitor insect populations, such as simple tags (e.g., paint marks), genetic techniques, and radioactive-isotope techniques (Hagler & Jackson, 2001). We used paint dots from weather-proof acrylic pencil brushes (Marabu GmbH and Co., Germany) because they are easy to use in field conditions (Hagler & Jackson, 2001). The durability, nontoxicity, and absence of effect on flight of the marks were first checked on ca. 50 hornets for at least 3 weeks (Monceau *et al.*, 2013c). This period was also used to train field experimenters in the quick and efficient application of small droplets of paint on the dorsal part of the hornets so as to avoid spiracle clogging. Fourteen different colors were used in different combinations: (i) 1 on the thorax, (ii) 1 on the thorax and 1 on the abdomen, or (iii) 1 on the thorax and 2 on the abdomen (Fig. 2). These combinations allowed us to monitor up to 2 954 individuals (Fig. 2).

Fig. 2 Codes used for marking *Vespa velutina* for population monitoring by capture-mark-recapture. One, 2, or 3 painted marks of different colors were placed on the thorax and abdomen.

Catching and marking procedure: overall design

Captures and recaptures were performed on 5 days: D1 (01/08/11), D2 (02/08/11), D3 (03/08/11), D8 (08/08/11), and D9 (09/08/11). On D1, hornets were captured, marked, and recaptured between 08:30 am and 12:00 noon for the morning session ($D1_{am}$) and between 02:00 pm and 05:00 pm for the afternoon session ($D1_{pm}$). It is impossible to accurately record the color combinations using binoculars (resighting) due to the size and velocity of the hornets. Thus, individuals were physically recaptured to allow the color combination to be correctly read, although it may have elicited an alert display (but see discussion). To prevent from or reduce such a risk, hornets were not caught daylong on D2 and D3 but only between 9:00 am and 12:00 noon. Thus, on D2 and D3, hornets were captured, marked, and recaptured only during morning sessions. On D8 and D9, a total of 100 and 60 hornets (respectively) were captured in front of the hives in the morning to count the number of remaining marked individuals in the apiary. Table 2 presents the number of marked and recaptured hornets for each session.

Hornets were gently captured with insect nets in front of the hives. The captures were performed in parallel by 3 experimenters (2 hives each). Newly captured individuals were marked as described above, and the hive number and capture date and time were recorded for all newly captured and recaptured hornets. All individuals were immediately released from the location where they were captured, except at D8 and D9 (marked but not unmarked individuals were released). Individuals recaptured less than 5 min after a previous capture were not counted to avoid pseudoreplication because when released, some hornets directly returned to the hive entrance and were then directly recaptured.

Number of V. velutina *visiting the 6 hives and daily variations*

The number of *V. velutina* (*N*) visiting the 6 hives was estimated by the unbiased Lincoln–Petersen index (Pollock *et al.*, 1990):

$$N = \frac{n_1 \times n_2}{m_2},$$

where n_1 is the number of caught, marked, and released individuals on D1; n_2 is the number of caught individuals (marked and unmarked) on D2; and m_2 is the number of caught individuals marked on D1 ($D1_{am} + D1_{pm}$) and recaptured on D2.

Table 2 Number of unmarked and recaptured (marked) *Vespa velutina* during the experiment from D1 to D9, and proportion of recaptured individuals from the initial sample marked. Unmarked individuals were marked at D1, D2, and D3 but not at D8 and D9.

Day	Unmarked individuals	Number of recaptured individuals (total number of recaptures)[†]						Proportion of recaptured (%)
		$\mathrm{D1_{am}}$	$\mathrm{D1_{pm}}$	D2	D3	D8	D9	
$\mathrm{D1_{am}}$	188	73 (125)	67 (114)	94 (207)	67 (129)	3 (3)	2 (2)	73.94
$\mathrm{D1_{pm}}$	42	–	6 (6)	12 (17)	12 (17)	0 (0)	2 (2)	59.52
D2	64	–	–	13 (17)	14 (22)	1 (1)	0 (0)	32.81
D3	66	–	–	–	19 (26)	1 (1)	0 (0)	28.79
D8	95	–	–	–	–	–	–	–
D9	56	–	–	–	–	–	–	–

[†] The total number of recaptures is superior or equal to the number of recaptured individuals because some individuals were recaptured more than once.

The variance of N was calculated as:

$$\mathrm{Var}(N) = \frac{(n_1+1)\times(n_2+1)\times(n_1-m_2)\times(n_2-m_2)}{(m_2+1)^2\times(m_2+2)},$$

and the 95% confidence interval as:

$$95\%\mathrm{CI} = [N - 1.96\times\sqrt{\mathrm{Var}(N)}; N + 1.96\times\sqrt{\mathrm{Var}(N)}].$$

The same index was applied for D2 and D3.

The variation in the number of caught hornets (marked vs. unmarked) during the course of the survey was analyzed using an ANCOVA, based on the F-ratio statistics.

Number of daily visits and interval between 2 consecutive recaptures

The average number of visits performed per half-day was calculated based on the individuals marked at $\mathrm{D1_{am}}$. It was obtained in dividing the total number of recaptures by the number of different hornets recaptured in the apiary for each session ($\mathrm{D1_{am}}$, $\mathrm{D1_{pm}}$, D2, and D3). The average number of visits was then compared between sessions using a Chi-square test.

To analyze the time elapsed between 2 successive visits, we considered a subgroup of hornets recaptured at least 5 times from D1 to D9 and which performed at least 2 visits during the same session. Here, we made the assumption that successive recaptures were related to individual successive visits. An ANOVA for repeated measurements was used to test the variation of the duration of the interval between consecutive visits on the same day (log-transformed) and the total number of visits (log-transformed) by considering linear and quadratic effects. Statistical significance was assessed using the F-ratio statistics.

Distribution of V. velutina *on the 6 hives*

The subgroup of hornets recaptured at least 5 times from D1 to D9 was used to test the distribution of their visits to the 6 hives using Fisher's exact tests. If significant, a binomial test was used to determine if the more visited hive was significantly the most visited (unilateral test for probability capture greater than 1/6).

The distribution of *V. velutina* on the 6 hives was estimated using a nonparametric index of dispersion (I_d) developed for a known size of resource patch (Thiéry *et al.*, 1995). In this case, this index was used to test the distribution of marked hornets among the 6 beehives. This index was computed considering the 3 days as replicates, that is, D1 ($\mathrm{D1_{am}} + \mathrm{D1_{pm}}$), D2, and D3, based on the number of hornets marked and recaptured separately, and then pooled:

$$I_\mathrm{d} = \frac{1}{\sqrt{N}} \times \sum_{i=1}^{N} \frac{\Delta i - E(\Delta i)}{\sqrt{V(\Delta i)}},$$

with the distribution:

$$\Delta = \sum_{i=1}^{n} yi,$$

the mean of the distribution:

$$E(\Delta) = \frac{t}{n} \times (n+t-1)$$

and the variance of the distribution:

$$\mathrm{V}(\Delta) = \frac{2t}{n} \times (t-1) \times (n-1),$$

where n is the number of replicates (3); y_i is the number of hornets on hive i within the n hives ($n = 6$), and t is

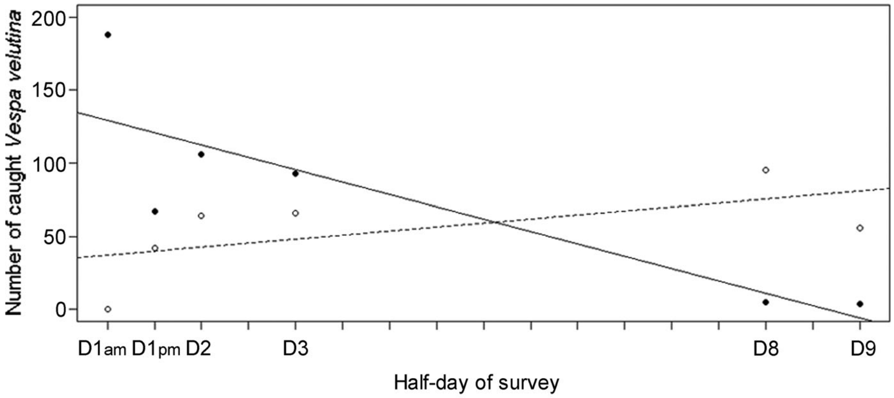

Fig. 3 Number of *Vespa velutina* caught during the course of the survey. Marked hornets are presented with black dots/plain line and unmarked hornets with white dots/dash line. For the initial sampling day ($\mathrm{D1_{am}}$), all caught hornets (188) were marked (i.e., 0 unmarked individuals). At $\mathrm{D1_{pm}}$, 67 individuals marked at $\mathrm{D1_{am}}$ were recaptured and 42 unmarked individuals were caught and marked, etc. See Table 2 for sample size details.

the total number of hornets. Then, the I_d distribution was compared to a normal distribution. The null hypothesis (i.e., random distribution of hornets among hives) was rejected when I_d was greater than 1.96 (aggregative distribution) or less than -1.96 (regular distribution) at $\alpha = 0.05$. Hives attracting more hornets were identified from the 95% CI based on the percentage (p) of hornets caught:

$$95\%\mathrm{CI} = \left[p - 1.96 \times \sqrt{\frac{pq}{n}}; p + 1.96 \times \sqrt{\frac{pq}{n}}\right],$$

where $q = 100 - p$ and n is the number of hornets.

All statistical analysis employed R software (R Development Core Team, 2008).

Results

Number of V. velutina *visiting the 6 hives and daily variations*

We marked a total of 360 hornets from D1 to D3 and according to the Lincoln–Petersen index, ca. 350 hornets visited the patch daily during the experiment (D1–D2: 354 hornets, 95%CI: 328–380 and D2–D3: 351 hornets, 95%CI: 214–487). Among 360 marked hornets during the survey, 204 (56.67%) were recaptured at least once, and if considering only the individuals marked at $\mathrm{D1_{am}}$, that is, the individuals being first marked and thus having the longest period of time to revisit the apiary, this percentage increased up to 74% (Table 2).

Overall, there was no difference between the number of marked and unmarked individuals caught (ANCOVA: $F_{1,8} = 1.35, P = 0.28$) nor during the course of the survey ($F_{1,8} = 3.31, P = 0.11$). However, this was mainly due to the fact that the number of marked individuals decreased while the number of unmarked individuals increased ($F_{1,8} = 12.64, P < 0.01$, Fig. 3).

Number of daily visits and interval between 2 consecutive recaptures

For each session, the number of visits the hornets made after being marked was similar (Chi-square test: $\chi^2_3 = 2.43$, $P = 0.49$): the hornets made in average 1.88 ± 0.23 visits (average range: 1.70–2.20) per session, that is, half-day.

Fifty individuals (ca. 14% of the overall marked sample) were recaptured more than 5 times (range: 5–15) between D1 and D9. The interval between 2 consecutive visits per individual decreased linearly as a function of successive recaptures (ANOVA for repeated measurements: linear effect: $F_{1,175} = 4.54$, $P = 0.03$; quadratic effect: $F_{1,175} = 1.28$, $P = 0.26$, Fig. 4) and was different between individuals (linear effect: $F_{1,47} = 7.54, P < 0.01$; quadratic effect: $F_{1,47} = 0.17, P = 0.68$).

Distribution of V. velutina *on the 6 hives*

Only 6 of the 50 hornets recaptured at least 5 times from D1 to D9 were caught in front of 1 hive most of the time (Fisher exact test: $P < 0.05$ for all comparisons; binomial test for recapture probability greater than 1/6, $P < 0.01$, Table 3). The other individuals were caught in front of all the hives in the apiary (Fisher exact test: $P > 0.05$ for all comparisons).

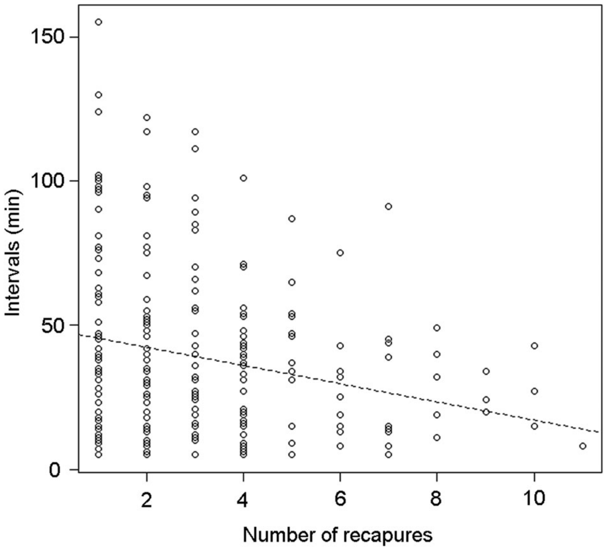

Fig. 4 Time elapsed between consecutive recaptures of *Vespa velutina* workers (intervals in minutes). Only hornets recaptured at least 5 times from D1 to D9 and which performed at least 2 visits during the same session were considered for this analysis. The dashed line stands for the linear regression.

The analysis of the distribution of the hornet recaptures indicated that overall, hornets were not randomly distributed in the apiary (first capture: $I_\mathrm{d} = 6.15$, $P < 0.05$, $n = 360$; recaptures: $I_\mathrm{d} = 10.41$, $P < 0.05$, $n = 680$; pooled: $I_\mathrm{d} = 630.02$, $P < 0.05$, $n = 1040$). They were predominantly aggregated on h5 at the benefit of h2, h3, and h6, the remaining hives (h1 and h4) receiving an intermediate number of visits (Fig. 5).

Discussion

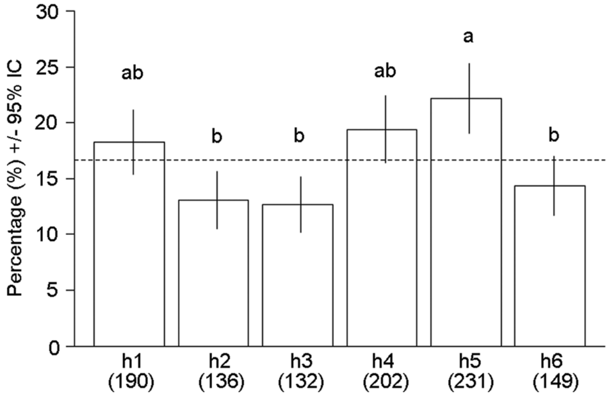

Fig. 5 Percentage of *Vespa velutina* workers in front of hives h1–h6. Percentages are assorted with their 95% confidence intervals (vertical lines), and sample size (in parentheses on the abscissa). A homogeneous distribution (16.67%) is indicated by the dashed line. Columns with the same letter were not significantly different.

The quality of capture-mark-recapture data mainly depends on the recovery rate and the initial number of marked individuals (Ackerman *et al.*, 1982). Obviously, the proportion of individuals recaptured at least once depends on the moment they have been marked during the survey. In our case, more than 50% hornets were recaptured at least once if considering the whole sample but if considering the individuals who had the greatest exposure time (i.e., those captured at $\mathrm{D1}_\mathrm{am}$), this percentage increased to more than 70%. Compared to other studies of flying insects, this recovery rate was in the high range (e.g., 0.09% for the parasitoid wasp *Nasonia vitripennis*, Grillenberger *et al.*, 2009; less than 2% for the mosquitoes *Anopheles arabiensis* and *A. vititipennis*, McCall *et al.*, 2001; Ulloa *et al.*, 2002; 13.02% for the parasitoid *Gryon gallardoi*, Canto-Silva *et al.*, 2006; 22.8% for the Red Mason bee *Osmia rufa*, Steffan-Dewenter & Schiele, 2004; 33%–53.75% for the solitary bee *Andrena hattorfiana*, Franzén *et al.*, 2009; 33%–100% for Euglossine bees, Ackerman *et al.*, 1982). Although the effect of the experimenters on the site cannot be completely discarded, these

Table 3 Distribution of the 6 *Vespa velutina* recaptured mainly on 1 hive as a percentage of recaptured in front of each hive and the total number of recaptures.

Individual	Hive 1	Hive 2	Hive 3	Hive 4	Hive 5	Hive 6	Total
1	**87.50***	0.00	12.50	0.00	0.00	0.00	8
2	37.50*	0.00	**62.50**	0.00	0.00	0.00	8
3	12.50*	0.00	12.50	**75.00**	0.00	0.00	8
4	0.00	**83.33***	0.00	0.00	0.00	16.67	6
5	7.14	0.00	21.43*	7.14	**57.14**	7.14	14
6	16.67	0.00	0.00	**83.33***	0.00	0.00	6

Bold numbers indicate hives where hornets were recaptured most often. Asterisks indicate hives where they were first caught.

high recovery rates suggest that the CMR procedure did not repel hornets from hunting honeybees in this apiary or at least that hornets gradually adapted to the situation.

Rapid changes of the number V. velutina *visiting the apiary*

In this experimental apiary, up to 20 hornets were scored at the same time in front of single hives at autumn (mid-September) when the predation pressure was at its highest level (Monceau *et al.*, 2013b), which estimates over 120 hornets visiting daily our 6 hives at this period. Our results indicated that at least 350 *V. velutina* visited the apiary each day in early August. Obviously, this number depends on the population size (i.e., the number of hornets from different colonies visiting the apiary). However, the localization of *V. velutina* colonies is often realized *a posteriori*, the nests being cryptic until they reach a large size or until leaves fall at autumn. In the present case, 5 nests were recorded within 1 km of our experimental site. Moreover, the foraging range of *V. velutina* is still unknown and the number of workers chasing in apiaries per nests is also unknown. Previous research indicates that several species of Vespidae can fly over long distances (Matsuura & Yamane, 1990). For example, the Japanese giant hornet (*Vespa mandarinia*), was recorded at apiaries 1–2 km away from the nest and individuals can fly up to 8 km from the nest (Matsuura & Yamane, 1990). This may also be true for *V. velutina*, so we cannot reliably assess the number of colonies that visited our apiary without further investigation by molecular or chemical analyses.

The presence of hornets in the apiary lasted at least for 9 days. Overall, the number of marked individuals decreased while the number of unmarked individual increased. *V. velutina* forms large colonies whose size enlarges dramatically from June to November. Consequently, there is an increase in larval nutritional requirements, an increase of predation and thus an increase in the number of hunting hornets in the apiary (Monceau *et al.*, 2013a, 2013b). The decline in the number of recaptured individuals cannot be attributed to degradation of the paint, because these paints remain stable for at least 3 weeks in captive hornets (Monceau *et al.*, 2013c). Thus, the observed decrease in recaptures at D8 and D9 may be explained by 2 nonmutually exclusive hypotheses. First, it could result from a change in tasks among individuals. Division of labor in *Vespa* spp. has not been well studied compared to *A. mellifera* (see Johnson, 2010 for a review of polyethism in honeybees) and there is limited data available. There is no clear-cut pattern of age-related division of labor in *Vespa* spp. (Matsuura, 1984; Jeanne, 1991; Volynchik *et al.*, 2009). Recent research showed that nest defense is mostly ensured by the oldest individuals of the colony (Monceau *et al.*, 2013c) but the distribution of each age-classes and their relation to the different tasks is not known to date. Second, the low number of recaptures at D8 and D9 could be attributed to a high mortality rate of the individuals which chase honeybees. Worker longevity varies among different *Vespa* species (*V. simillima* has maximal longevity less than 30 days and *V. tropica* workers may live for more than 55 days, Matsuura, 1984) but data on the longevity of *V. velutina* is not available. Accurate data on division of labor and life span in *V. velutina* are thus required to favor of one or the other hypothesis.

Number of daily visits and interval between consecutive visits

On average, the hornets visited the apiary at least once per half-day session and some individuals were recaptured up to 15 times from $D1_{am}$ to D9. Among the 204 hornets marked and recaptured during the survey, 50 individuals (24.50%) were recaptured more than 5 times, suggesting that they were able to memorize cues to return to the apiary. Learning can benefit an individual who performs a task multiple times and can also enhance fitness (Dukas, 2008a) and improvements in the foraging performance of eusocial species over time can benefit the whole colony, as documented in bees (Dukas & Visscher, 1994; Ohashi *et al.*, 2006, 2008; Schippers *et al.*, 2006; Saleh & Chittka, 2007; Dukas, 2008b). Although the progressive decrease of the time lapse between consecutive visits may partially result from the handling habituation of the capture, it is also suggestive of an increase in the hornet foraging efficiency. Improvements in foraging efficiency may result, for example, from an increase in the ability to locate the food source (Dukas, 2008c) or to acquire the food (Weiss *et al.*, 2004; Warkentin *et al.*, 2006; Dukas, 2008b). In this study, further work is however required to confirm our preliminary findings, because hornets were not monitored between consecutive recaptures in the apiary and thus their behavior during this period is unknown.

Distribution of V. velutina *in the apiary*

Basically, the hunting behavior of social vespids can be summarized as waiting for the prey, catching the prey, processing the prey, and returning to the nest (Richter, 2000), and *V. velutina* is not an exception to the rule. Indeed, at the individual level, most of the *V. velutina* were recaptured in front of different hives suggesting that hornets visited all hives within the apiary. While only a few *V. velutina* were recovered in front of 1 specific hive, the distribution of all individuals visiting the apiary appeared to be aggregative.

In apiaries, *V. velutina* try to catch honeybees and then pillage the hives for brood and honey when the colonies are weakened. Thus, the hive attractiveness may be based on the amount of brood, stored resources, or the aggression of the honeybee colony. Giray *et al.* (2000) reported a negative relationship between colony defense efficiency and foraging in *A. mellifera*, that is, the less aggressive colonies are expected to hold the largest resources. However, in our study, 5 of the 6 hives had equivalent vigor and the other hive was not less attractive than these 5 hives. Especially, hornets mainly aggregated in front of h5 which was neither the smallest nor the biggest colony of the apiary. Alternatively, *V. velutina* may adjust its behavior to the aggressiveness of its prey. Breed and Rogers (1991) demonstrated that defense behavior in *A. mellifera* is genetically based and recent studies show significant variation among colonies in this trait (Kastberger *et al.*, 2009; Wray *et al.*, 2011). The aggregation of *V. velutina* individuals on specific hives may result from low bee defensiveness (i.e., the brood and the storage are easily accessible for pillage) or alternatively from high aggressiveness, which coerces chasing (i.e., to be more effective for weakening the colony). This later hypothesis is less likely, because *V. velutina* preys less intensively on *A. cerana* in Asia than on *A. mellifera* in France, even though the latter species is less defensive (Ken *et al.*, 2005; Tan *et al.*, 2007, 2010, 2012a, 2012b, 2013).

Conclusions

Alien predators like *V. velutina* can thrive in a new environment because of their novelty in the local food webs and the absence of competitors (Snyder & Evans, 2006; Sih *et al.*, 2010). Although beekeepers also consider *Vespa crabro*, the native and direct competitor, to be a pest, its significant damage on honeybee colonies has still to be proven and is far less than that from *V. velutina*. Indeed, the invasive hornet forms larger colonies than the native one and thus requires more proteins to feed its brood. *V. velutina* predation is thus much more intense than that from *V. crabro*. However, *V. velutina* predation is less severe than the massive group predation of *V. mandarinia* as it does not destroy honeybee hives within a few days when it displays group predation (Matsuura, 1984; Matsuura & Yamane, 1990).

Finally, we found that at a small spatial scale, the distribution of *V. velutina* is heterogeneous suggesting that the prey choice does not occur randomly. Finding the factor responsible for the aggregation of *V. velutina* at the entrance of specific hive could help to adapt management plans to protect apiaries. According to our results, this aggregated distribution is not related to the honeybee colony size but could reflect the variability in defensiveness between colonies. Such a hypothesis should receive full attention because of the genetic basis of defensive behaviors and their potential high heritability (Breed & Rogers, 1991; Breed *et al.*, 2004). Thus, a selection of honeybee colonies based on their level of defensiveness could be a promising strategy to reduce the impact of *V. velutina*.

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Disclosure

The authors have no conflicts of interest, including specific financial interests and relationships and affiliations relevant to the subject of this manuscript.

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