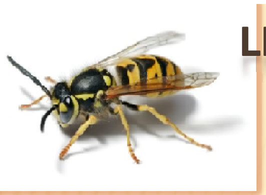

LEARNING FLIGHTS IN *VESPULA GERMANICA* WHILE RELOCATING A FOOD SOURCE

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Abstrac: The German yellowjacket, *Vespula germanica* (F.) (Hymenoptera: Vespidae), is a highly invasive wasp that exhibits efficient mechanisms while foraging. When foraging on carrion resources, this wasp usually makes repeated visits to the feeding site until depleting the resource. We studied wasps' learning flights in different conditions. Number of learning flights were recorded in different wasps' visits and when displacing a food source. Moreover, we analyzed the effect of adding conspicuous cues in wasps learning flights behavior. Wasps were trained to feed from a certain array. At each feeding visit, the studied wasp fed and collected food from the dish, then departed to the nest and returned a few minutes later. Training session consisted on three or four feeding trials (depending on treatment). The numbers of learning flights were registered during wasp departure for all experiments. We found that as more previous experience with food, less were the mean number of learning flights. Furthermore, when food was displaced from the original feeding site, the number of learning flights increased after the following visit. We also found that when feeding in contexts presenting conspicuous cues, wasps performed less learning flights than in contexts without these references. We conclude that number of learning flights depends on previous wasps ' experiences with a certain location, as well as on the conspicuity of landmark cues.

Introduction: *Vespula germanica*(F.) (Hymenoptera: Vespidae) is a highly invasive social wasp which has successfully colonized diverse environments around the World. It is Native to Eurasia and it became in a few years one of the most abundant insects in the Andean Region. *V. germanica* has complex foraging strategies which imply intricate learning mechanisms. They display different abilities when feeding in open or closed habitats, revisit a previously visited location more frequently when foraging in closed habitats than when foraging in open ones. As they systematically return to the site where a food resource couldn´t be transported on only one trip, *V. germanica* wasps learn global and local landmarks which serve them as guides to relocate the information. This is acquired by specialized flights (i.e. learning flights) where the wasp learns and stores the information needed of the place to which it will return. The source relocation behavior has been studied under different environmental conditions, supporting the evidence of diverse learning mechanisms for this invasive species. Nevertheless, there are few studies regarding learning flights. In this work we study wasps' learning flights in different contexts that imply or not enriched contexts. Moreover, we analyzed learning flights in consecutive visits, and the effect of changing the food source location. We expect to find that 1) as more previous experience with food, less is the mean number of learning flights, 2) if food is displaced 60 cm from the original feeding site, the number of learning flights will increase in the next visit, 3) when feeding in enriched contexts, wasps will do less learning flights than in contexts without these references.

Methods: **All experiments were carried out under natural conditions near San Carlos de Bariloche, Argentina, during the V. germanica wasp's most active period (February - April) in 2006, 2007 and 2008. Experiments were conducted near the pebbly shoreline of a lake. In all experiments an individual forager was allowed to feed from a white plastic dish containing 20 g of minced fresh meat. When one forager spontaneously arrived at the dish and was feeding, it was distinguishably marked with a dot of washable paint on the abdomen for further identification. Any other wasp visiting the dish was removed. At each trial, the studied wasp collected food from the dish, departed to the nest, and returned a few minutes later. An individual wasp was used for only one experiment and one treatment. Each experiment consisted on 2, 3 or 4 feeding trials (depending on treatment). Thus, one feeding trial consisted on one discrete visit to the feeding dish. After each feeding trial, the number of learning flights was recorded in all experiments. Comparisons among the number of learning flights in different wasps' visits were done utilizing Friedman ANOVA and Kendall Coef. of Concordance. Paired comparisons were conducted by Wilcoxon matched pair tests. Comparisons of number of learning flights between enriched and non-enriched contexts were conducted by Mann Whitney U Test.**

Experiment 1: Each wasp was allowed to feed 4 consecutive times from a dish with food placed at the centre of a square of 30 cm side length, delimited by four cylinders of the same color, 2 cm in diameter and 60 cm in height.

Experiment 2: Each wasp was allowed to feed 2 consecutive times from a dish with food, the cylinders were arranged as in exp.1, and 4 dishes were placed at the four edges of the square, 1 of these dishes contained food, the wasp fed consecutively in the same place twice (position A) and when it left the nest after its second visit and before it returned for its third visit, the dish with food was displaced from the original feeding location to the opposite point on the array (position B) the wasp was allowed to feed 2 more times consecutively.

Experiment 3: Each wasp was allowed to feed 3 consecutive times from a dish as in exp.1. This array was located in an open habitat in enriched contexts whit five wooden sticks surrounded this array, 1.5 m high with blue flagging 50 cm long draping from the top. These sticks were placed at a distance of 1 m from the array. In the control group the array was not surrounded by the wooden sticks i.e., non enriched contexts.

Results

Experiment 1: The number of learning flights significantly varied according to the number of feeding visits (FV) (X2=52. 81, N= 32, p<0.0001). Significantly more learning flights were observed after the first feeding visit than after the second one (Z=3.99, N= 32, p<0.0001). Similarly, more learning fights were found after the second than after the fourth one (Z= 4.06, N= 32, p<0.0001) (Fig. 1). However, non significant differences existed between the number of learning flights after the third and the fourth visit (Z= 0.47; N= 105, p > 0.6), reaching an asymptotic level..

Experiment 2: Wasps fed two consecutive times from a certain feeder position (position A) before feeding twice from a different position (position B). Significant differences were observed between the number of learning flights performed after four feeding visits ($X^2_{(N=13,\ df=3)}$ = 22.450, P< 0. 00005), (Figure. 2). As in exp 1, the number of learning flights was significantly less after the second visit than after the first one (Z=2.54, N=13, p<0.01). However, after the third visit, (food position changed) the number of learning flights increased, reaching similar levels as those observed after the first one, as non significant differences were found between them (Z=1.78, N= 13, p>0.05). This indicates that the change of the feeder position generated more learning flights. Moreover, significantly less learning flights were found after the fourth feeding visit than after the second one (Z=2.19, N=13, p< 0.05). As expected, significantly more learning flights were observed after the third feeding visit than after the fourth one (Z=3.00, N=13, p<0.005).

Experiment 3: Significant differences were found when comparing the number of learning flights after the first, second and third visit, between enriched and non enriched contexts (N1,2= 41, 39, Z=1.99, p< 0.05; N1,2= 40, 39, Z= 2.21, p< 0.05; N1,2= 39, 38, Z= 2.45, p< 0.05, respectively) (Fig. 3). Significant higher number of learning flights were observed after the first visit than after the second one, and after the second than after the third one within each environment (Z= 5.05, N=39, p< 0.05; Z=4.75, N=40, p<0.05; Z= 5.30, N=38, P<0.05; Z= 4.64, N=38, p<0.05 for enriched and non enriched contexts respectively).

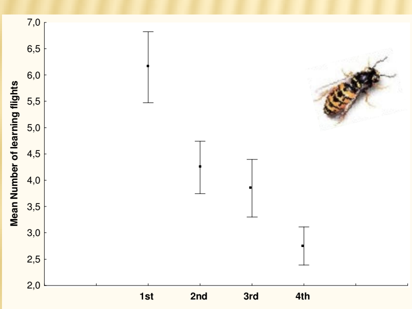

Fig 1. Mean numbers of learning flights after each feeding visit (FV)

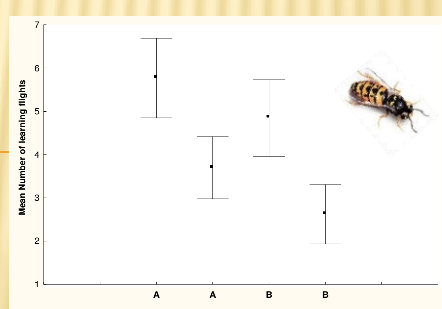

Fig 2. Mean number of learning flights according to number of feeding visits and changing feeder location.

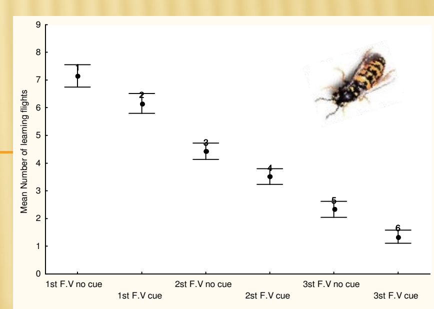

Fig 3. Mean number of learning flights according to feeding visits (FV) in contexts with and without cues

Conclusion: *Vespula germanica* wasps learn to associate contextual cues in relation to certain food position, and this is reflected in the number of learning flights. As wasps have more experience in feeding from a certain site, they decrease the number of learning flights. However, food displacement from the original feeding site re-increased the number of learning flights reaching first visit levels. As previously demonstrated in other studies, during learning flights wasps acquire information about contextual landmarks, which will help foragers to find the food source. Interestingly in enriched contexts with conspicuous cues, wasps performed less learning flights than in non enriched ones. These results show that local cues are used by *V. germanica* wasps to orient them to relocate a food source. Number of learning flights seems to be a good indicator of learning in this species.