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Why do honeybees reject certain flowers?

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Summary. Honeybees often approach flowers of *Lotus corniculatus* and then fly away without attempting to extract nectar. These rejected flowers contained 41% less nectar than my random sample. The accepted flowers contained 24% more nectar than my random sample. The differences among these three flower-groups were due to differences in the percent of empty flowers in each group rather than the differences in the absolute amount of nectar. Honeybees increased their foraging efficiency by accepting less empty flowers and rejecting more empty flowers than would be expected if they foraged randomly. There are two possible mechanisms for this discrimination-behavior: either the bees are smelling nectar odor or they are smelling bee scent left by previous visitors to the flower. My results are inconsistent with the hypothesis that bees are basing their decision on nectar smell and suggest that they are using bee scent as a means of identifying empty flowers.

When honeybees forage, the do not attempt to extract nectar from every flower that they encounter. As bees approach a flower, they often briefly touch or hover over a flower and then fly away (Frankie and Vinson 1977; Marden 1984). Presumably this is a form of discrimination-behavior in which the bee is considering the flower as a possible source of food for some reason decides not to accept it.

This form of discrimination-behavior has been noted by various researchers. Zimmerman (1982) found that pollen-collecting bumblebees reject flowers that do not have enough pollen visible to make collecting profitable. Bell et al. (1984) showed that while visiting *Impatiens capensis*, honeybees, bumblebees and wasps more frequently rejected flowers in the female stage and preferentially visited the male-stage flowers. Flowers in the male stage contain more nectar and it has been hypothesized that flower-preference results from the bees' ability to sense the amount of nectar in the flower. Heinrich (1979) showed that nectar-gathering bumblebees preferentially chose flowers that contained relatively large amounts of nectar and rejected flowers with little nectar. He was able to detect differences in odor between clover inflorescences that had been visited and those that had not, and hypothesized that the bees were also able to smell the nectar and thus chose the most rewarding flowers.

In this study I will attempt to explain why worker honeybees accept or reject particular flowers of *Lotus corniculatus* while foraging for nectar. Specifically, I will address the following questions: (1) Does the discrimination-behavior increase the bees' foraging efficiency? (2) What mechanism is the bee using to discriminate between accepted and rejected flowers?

Methods

Description of Lotus corniculatus and the study site

Lotus corniculatus. L. (Leguminosae) is a weedy perennial, native to Europe, that honeybees forage for nectar and pollen. It occurs along roadsides and in disturbed places throughout much of the United States. In northern California it flowers from May to September, with peak flowering in July and August.

The papilionaceous flowers have nectar hidden deep within the corolla at the base of the fused filaments. The flowers are bright yellow while pollen and nectar are present, but turn red when these rewards are no longer available. This color change is presumably a post-pollination phenomenon (see Gori 1983). Bees generally ignored red flowers and no red flower was found to contain nectar or pollen (personal observation).

Fieldwork was conducted in a recently harvested redwood forest in Eureka, California and in a cleared field in Arcata, California. Each site contained several clumps of *L. corniculatus* growing in discrete patches of approximately 2 m in diameter. Both sites were located approximately at sea level, 10 to 15 km from the coast. All work was conducted from June to August, 1984.

Nectar measurements

Sampling was done twice a day on ten days. The first session each day, "morning", was started within 30 min of the first bee-visit to the area. Consequently most of the flowers sampled either had opened that morning or had not been visited since the previous day. The starting time of the morning session was between 1000 and 1200 h depending on the weather. The second session, "afternoon", started after 4 to 5 h of foraging had taken place, between 1500 and 1600 h. This session ended shortly before the end of the foraging day.

Nectar volume was measured to the nearest 0.5 mm (0.015 μl) using a 1 μl capillary tube and concentration was

Table 1. Nectar quantity and quality of the four groups of flowers for the full day and broken down into the morning and afternoon sessions

Session	Group	n	% empty flowers	Mean volume (μl × 100)	Mean volume excluding empty flowers (μl × 100)	Mean % conc. excluding empty flowers (μl × 100)
Full day	standing crop	490	53.7	2.06	4.45	42.0
	accept	490	44.1	2.58	4.61	44.0
	reject	490	70.2	1.22	4.11	45.1
	visited	110	73.3	0.77	–	–
Morning	standing crop	250	46.4	2.73	5.10	41.1
	accept	250	41.2	3.07	5.22	42.1
	reject	250	66.4	1.60	4.76	43.5
Afternoon	standing crop	240	61.2	1.36	3.51	43.4
	accept	240	47.1	2.07	3.91	46.5
	reject	240	74.2	0.83	3.22	47.2

Session	Type of test[a]	Groups compared	n	Significance of nectar vol. means	Significance of nectar vol. means excluding empty flowers
Statistical evaluation					
Full day	K-W	standing vs accept vs reject	490	$p<0.001$	$P=0.752$
	mult. comp.	standing vs accept	490	$p<0.001$	
	mult. comp.	standing vs reject	490	$p<0.001$	
	mult. comp.	reject vs accept	490	$p<0.001$	
	M-W	reject vs visited	150	$p=0.17$	
Morning	K-W	standing vs accept vs reject	250	$p<0.001$	$P=0.969$
	mult. comp.	standing vs accept	250	$p>0.1$	
	mult. comp.	standing vs reject	250	$p<0.001$	
	mult. comp.	reject vs accept	250	$p<0.001$	
Afternoon	K-W	standing vs accept vs reject	240	$p<0.001$	$P=0.441$
	mult. comp.	standing vs accept	240	$p<0.001$	
	mult. comp.	standing vs reject	240	$p<0.001$	
	mult. comp.	reject vs accept	240	$p<0.001$	

[a] K-W = Kruskal-Wallis 1-way ANOVA; M-W = Mann-Whitney test; mult. comp. = non-parametric multiple comparisons using ranked sums from the K-W test

determined to the nearest percent with a Bellingham and Stanley handheld refractometer modified by the factory for small volumes. Because of the small size of the flowers, it was necessary to remove the corollas in order to make the measurements. Flowers collected were categorized as follows:

(1) "standing crop" – a random selection of flowers.

(2) "accepts" – the first flower on any inflorescence that a bee attempted to probe. Bees were chased from these flowers after landing but before inserting their proboscises. Such flowers were categorized as "accepted" because honeybees never landed on a flower without attempting to extract nectar. All bees observed were foraging for nectar; I never saw honeybees foraging for pollen only.

(3) "rejects" – flowers that were touched by the bee's antennae or legs in flight but not probed. Bees occasionally rejected flowers by briefly hovering over a flower and then going on without touching it. This type of rejection occurred much less frequently, consequently this type of flower was not measured. Only yellow flowers were included in the survey. I could not detect any differences in odor or appearence, in the visable and ultraviolet range, among flowers in the three groups.

The sample size was 25 for each group of flowers for each session. However, during two of the afternoon sessions the bees stopped foraging before 25 flowers of each group could be measured. The sample size was 17 and 23 on these two occasions.

On three of the sampling-days an additional group of flowers ("visited") was collected. Flowers were collected immediately after a bee visit and nectar volume was measured.

Results

Nectar measurements

The mean concentrations of nectar for the three groups are shown in Table 1. There is no difference in concentration among the standing crop, accept and reject flower groups ($P=0.223$) for the full day. Broken down into morning and afternoon sessions, there is still no difference (morning $P=0.648$; afternoon $P=0.317$).

Table 1 shows the comparison of nectar volumes for the three groups. The mean volumes of nectar for the three flower-groups are significantly different when data are analyzed for the full day as well as when they are divided into morning and afternoon sessions, with the accepts having the most nectar, followed by the standing crop and then the rejects. Average volumes differ for all pairwise comparisons within the full day, afternoon session, and morning session. However, the standing crop vs accept comparison in the morning session is not different. These results mean that, for the full day, the bees obtained significantly more nectar than if they chose flowers randomly (24% more). However in the morning my random selection of flowers was as successful as the bees' choices.

There is a large number of empty flowers in each group. Even in the morning standing crop, when most of the sampled flowers had not been visited that day, 46% contained no nectar. When empty flowers are excluded, the volumes of the three groups are not different (Table 1). The differences in volume among the three groups is due to the differences in the percent of flowers in each group that contained no nectar, with rejects having the most empty flowers followed by standing crop and then accepts. The only exception is in the morning standing crop vs accept comparison which shows no difference in the percent of empty flowers ($X^2 = 1.17$; $P > 0.25$). Thus, the bees increased their nectar intake because they were able to distinguish between empty and non-empty flowers; they did not discriminate among differences in the *absolute* amount of nectar.

Visited flowers were not always thoroughly emptied. The nectar volume in visited flowers is not different from that of the rejects (Table 1). Only the rejects that were sampled at the same time as the visited group were used in this test.

Discussion

My study supports the theory that the bees' discrimination-behavior increases foraging efficiency. When foraging on *L. corniculatus*, honeybees are able to choose flowers that have more than the average amount of nectar and reject those flowers that have less. Each day the bees harvested 24% more nectar than they would by random selection. Some of this gain in efficiency could be due to area-restricted foraging in nectar rich areas (Waddington 1980) rather than discrimination-behavior. Although area-restricted foraging could explain why the accepted flowers contained more nectar than the standing crop, it doesn't explain why bees reject flowers, why rejected flowers contain less nectar, or why rejected and accepted flowers are often found next to each other.

It appears that as honeybees approach a flower, they decide to accept or reject that flower on the basis of how the flower smells. This decision is made either by smelling the flower while hovering over it or by briefly touching it with their antennae or feet.

However, it is not clear what the bees are smelling. Marden (1984) and Bell et al. (1984) found that bees accepted flowers with relatively high amounts of nectar and rejected those with less nectar, and concluded that the bees were smelling nectar. Another possibility deserves consideration. Since the absence of nectar is frequently caused by the extraction of the nectar by a bee, any other changes made by the visiting bee must also be investigated. It is possible that bees leave a scent when they visit a flower, so that flowers without nectar may also contain bee scent. There is too much evidence for scent marking by bees to ignore this possibility.

Many researchers have found that bees use scent markings to communicate with conspecific bees. Ribbands (1955) suggested that the scents that honeybees use to mark food locations might also be used to mark visited flowers; these would then be avoided by subsequent foragers. Bumblebees (Cederberg 1977) and honeybees (Butler 1969) leave scent trails leading to their nests. These scents are applied by the bees' feet and presumably could also be left on flowers. Cameron (1981) found that bumblebees left scent marks on artificial flowers that were recognized by subsequent foragers. The best evidence to date of bees leaving scents as markers on visited flowers is from the study by Frankie and Vinson (1977) on *Xylocopa*. They found that females left scents while visiting flowers that resulted in rejection of those flowers by conspecific females for a period of 10 min. They were also able to remove nectar from unvisited flowers and showed that bees still visited those flowers, suggesting that *Xylocopa* females were using pheromones, and not nectar scent, as an indicator of the presence of nectar.

In the following section I will discuss some of the results of my study in terms of two possible hypotheses for how honeybees decide which flowers to accept and which to reject: (1) the bees accept and reject flowers based on nectar smell, and (2) the bees accept flowers with no bee scent and reject flowers with bee scent. These two hypotheses are not mutually exclusive, but the results of my study support the hypothesis that honeybees use bee scents left on flowers by other foragers as an indicator of the presence or absence of nectar.

The first piece of evidence that supports the idea that honeybees were not able to smell nectar is the observation that honeybees did not distinguish among different *absolute* amounts of nectar, but only between the presence or absence of nectar. Many researchers (e.g. Ribbands 1955) have demonstrated that honeybees will use their ability to distinguish between minute differences in amounts of odoriferous substances to increase their food intake. If the bees could smell nectar, they should have used this ability.

There are additional observations that are inconsistant with the idea that honeybees are smelling nectar. It is difficult to explain why honeybees accept flowers that contain no nectar 44% of the time and reject flowers with nectar 30% of the time (Table 1). Some of this can be explained if the time of foraging is considered.

When the foraging day is broken down into morning and afternoon sessions, the results show that the bees' behavior pays off only in the afternoon. This may be due to differences between the morning and afternoon standing crop. Two differences are evident: (1) the morning standing crop had more flowers that contained nectar, and (2) the majority of the morning standing crop flowers had not been visited since the previous day, while many of the afternoon crop flowers had recently been visited. These differences have to be investigated in light of the two possible mechanisms. If bees are smelling nectar, then why are they less successful in the morning when more nectar is present? If anything, the signal would be stronger and the bees would be more successful. If bees are smelling bee scent, presumably the scent wears off with time (Frankie and Vinson 1977; Cameron 1981). In the morning, many of the flowers contain no nectar but have not been visited recently. Additionally, when I covered flowers prior to anthesis and measured nectar volume in flowers that had been opened at least one day, 8.4% ($n=119$) contained no nectar even though they had never been visited. If the decision is based on bee scent, bees would accept these flowers even though they contained no nectar. This may explain why bees accept so many flowers with no nectar throughout the day (if the bees are smelling nectar, this result is hard to explain). In the afternoon, many of the empty flowers had been visited recently. This explains why the bees are more successful in the afternoon than in the morning. This type of flower could be part of the accepted empty flowers. Why then did the bees reject flowers with nectar present? Flowers that had been visited were not significantly different from rejected flowers. It is possible for flowers to contain bee scent and nectar. Rejected nectar-containing flowers can be explained as recently visited but not throughly emptied flowers.

An alternative explanation for the bees' less efficient foraging is that the bees cannot accurately locate the source of the odor, whether the odor is nectar or bee scent. Since the flowers often touch each other, accurate discrimination may be impossible. This explanation still does not explain the discrepancy between the foraging results in the morning and the afternoon.

Additional evidence against the hypothesis that honeybees are smelling nectar comes from an examination of the possibility of odoriferous nectar from the viewpoint of the plant. Heinrich (1979) was able to smell clover nectar but I know of no other reports of odoriferous nectar. At face value, the evolution of odoriferous nectar seems unlikely. If a plant advertises its nectar, then pollinators will only visit flowers that contain nectar. Flowers that have been recently visited would not reap the benefits of pollinator visits. It may make more sense for plants to make the pollinator deliver without a guarantee of payment. Casper and La Pine (1984) suggested three plant attributes that would make it advantageous for a plant to advertise the presence of nectar and/or pollen. Although they were studying the change of corolla color as a way of advertising the presence of rewards to pollinators, the arguments they present are applicable to odoriferous nectar. Odoriferous nectar could be explained if: (1) the plant's entire pollen load is removed in one visit, or (2) the plant must compete for pollinators, or (3) the plant needs only one visit to fertilize all of its ovules. At least two of the three conditions do not seem to apply to *L. corniculatus* and the other condition was shown not to occur in another study: (1) many visits are necessary to remove all of the pollen from a single flower (personal observation), (2) Stephenson (1984) found that reproductive output was not pollinator limited in fields of *L. corniculatus* (this, of course, does not prove that pollinators were not the limiting factor in my study), and (3) Morse (1958) showed that *L. corniculatus* flowers are open from 3 to 10 days and require 12 to 25 visits for maximum seed set. On the basis of this evidence, *L. corniculatus* does not seem a likely candidate for odoriferous nectar. However, the possibility of competition for pollinators sometime in the past can not be ruled out as a possible selective pressure for the evolution of odoriferous nectar. Plants that advertise their reward make foraging less of a gamble for pollinators. If two plants occur together and only one advertises the presence of food, the plant that advertises could receive substantially more pollinator visits and thus produce more offspring.

The results of my study give an ultimate explanation for the discriminating behavior but do not give a proximate one. The critical experiments would be to (1) remove nectar from an unvisited flower and (2) inject nectar into flowers immediately after they have been visited. Observation of these two types of manipulated flowers, as bees accept or reject them, would give evidence as to the mechanism involved.

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References

Bell G, Lefebvre L, Giraldeau L-A, Weary D (1984) Partial preference of insects for the male flowers of an annual herb. Oecologia (Berlin) 64:287–294

Butler CG (1969) Nest-entrance marking with pheromones by the honeybee *Apis mellifera* L., and by a wasp, *Vespula vulgaris* L. Anim Behav 17:142–147

Cameron SA (1981) Chemical signals in bumble bee foraging. Behav Ecol Sociobiol 9:257–260

Casper BB, La Pine TR (1984) Changes in corolla color and other floral characteristics in *Cryptantha humilis* (Boraginaceae): Cues to discourage pollinators? Evolution 38:128–141

Cederberg B (1977) Evidence for trail marking in *Bombus terrestris* workers (Hymnenoptera, Apidae). Zoon 5:143–146

Frankie GW, Vinson SB (1977) Scent marking of passion flowers in Texas by females of *Xylocopa virginica texana* (Hymenoptera: Anthophoridae). J Kans Entomol Soc 50:613–625

Gori DF (1983) Post-pollination phenomena and adaptive floral changes. In: Jones CE, Little RJ (eds) Handbook of experimental pollination biology. Van Nostrand-Reinhold, New York, pp 31–49

Heinrich B (1979) Resource heterogeneity and patterns of movement in foraging bumblebees. Oecologia (Berlin) 40:235–246

Marden JH (1984) Remote perception of floral nectar by bumblebees. Oecologia (Berlin) 64:232–240

Morse RA (1958) The pollination of birdsfoot trefoil (*Lotus corniculatus* L.) in New York State. In: Arnold JW (ed) Proceedings of the 10th International Congress of Entomology, Montreal, Canada, 4:951–953

Ribbands CR (1955) Scent perception of the honey bee. Proc R Soc Lond (Biol) 143:367–379

Stephenson AG (1984) The regulation of maternal investment in an indeterminate flowering plant (*Lotus corniculatus*). Ecology 65:113–121

Waddington KD (1980) Flight patterns of foraging bees relative to density of artificial flowers and distribution of nectar. Oecologia (Berlin) 44:199–204

Zimmerman M (1982) Optimal foraging: Random movement by pollen collecting bumblebees. Oecologia (Berlin) 53:394–398

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