

STIMULI ELICITING MATING BEHAVIOUR OF BUMBLEBEE (*BOMBUS PRATORUM* L.) MALES

by

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INTRODUCTION

The males of many species of bumblebees fly along established routes connecting special places (*e.g.* part of a twig or leaf, or base of a tree) they have marked with a pheromone from their mandibular glands (*e.g.* DARWIN, 1885, 1886; FRANK, 1941; HAAS, 1946 and 1952; STEIN, 1962). It is supposed, although not proved, that females are attracted to these places and mating facilitated (see FREE & BUTLER, 1959). FRISON (1927) and CUMBER (1953) suspended young queens by threads along flight routes and found that males were attracted to them. I have used a similar technique to study the factors that attract males to queens.

METHODS AND RESULTS

Investigations were made during June, 1963, at a flight route (about a metre above ground level) established by *Bombus pratorum* males along the edge of a wood. The visiting places were 3 to 10 m apart (mean of 7 m). On two occasions a *B. pratorum* queen was seen to fly along this flight route, pausing at visiting places but it was not seen to meet males.

The first time that a *B. pratorum* queen was suspended, by a black thread round her waist, at a scenting place, a male alighted on her within a few seconds, and within a short time a group of 8 males had formed on her. Such an intensive reaction to a queen was not seen subsequently, probably because males learned that queens presented in this way were unwilling to mate.

Effect of location.

To see whether the location of a queen along a flight route affected the behaviour of males toward her, a queen was suspended, for ten minute periods, alternately at a position approximately halfway between two visiting places and within about 100 mm of one or other visiting place. In this and later experiments, the males that flew within 150 mm of the queen were

counted and their behaviour recorded. The same queen was not used throughout an experiment, which was always spread over more than one day.

When queens were suspended *near* visiting places, 60% of the males reacted to them and 34% alighted on them, whereas queens suspended *between* visiting places were nearly always ignored (Table 1).

TABLE 1

Effect of location of queens in the flight route on the behaviour of B. pratorum males. (Combined results from eleven observation periods at each type of location)

Location	Behaviour of males towards queens				Mated or attempted to mate for:	
	Ignored	Inspected	Touched	Alighted	< 30 sec. duration	> 30 sec. duration
At scenting places	35	9	13	23	3	4
Between scenting places	36	0	1	0	1	0

Comparison of attractiveness of queens and workers.

A *B. pratorum* virgin queen (about 20 mm long), a medium size worker (about 15 mm long) and a small worker (about 10 mm long) were suspended near three visiting places and watched simultaneously. When ten males had visited each visiting place, the positions of the queens and workers were exchanged. Queens received more responses than medium sized workers, which in turn were touched or alighted on more often than small workers (Table 2).

TABLE 2

Comparison of attractiveness of B. pratorum queens and workers. (Combined results from 20 observation periods during which each category was visited by 10 males)

	Behaviour of males toward queen and workers			Mated or attempted to mate for:	
	Inspected	Touched	Alighted	< 30 sec. duration	> 30 sec. duration
Small worker	8	14	3	0	0
Medium size worker	9	21	7	1	0
Queen	6	52	1	5	2

Effect of size and shape.

Queens in the above experiment could have been more attractive because they were larger. To test the effect of size alone black balsa wood models, consisting of a 'thorax', 'waist' and 'abdomen' were used. Three sizes, 15, 25 and 30 mm long and 7, 10 and 12 mm wide respectively, were watched

simultaneously at visiting places (Table 3). The response to the medium and larger size models was about equal but both elicited more responses than the smaller model ($P < 0.05$ for each comparison).

TABLE 3

Comparison of attractiveness of models of different sizes to B. pratorum males. (Combined results of 17 observation periods during which each type of model was visited by 10 males)

Size of model	Behaviour of males toward models		
	Inspected	Touched	Alighted
Small	8	4	1
Medium	14	9	3
Large	15	6	5

Effect of removing head and wings.

Virgin queens were killed by cold and presented with either their heads removed, their wings removed, or both wings and head removed. Initial responses to these queens were similar, but males attempted to mate six times with queens with heads but no wings, only once with queens without head or wing and not at all with queens without heads but with wings (Table 4). In all the attempts to mate the male adopted the normal mating position.

TABLE 4

Effect of presence or absence of wings and head on the mating behaviour of B. pratorum males. (Combined results of 15 observation periods, during which each type of queen was visited by 10 males)

	Behaviour of males toward queens			Mated or attempted to mate for:	
	Inspected	Touched	Alighted	< 30 sec. duration	> 30 sec. duration
queens without heads	15	9	10	0	0
queens without wings or heads	15	10	12	1	0
queens without wings	20	4	11	5	1

Effect of colour.

Some dead queens had their orange and yellow bands stained black so that their bodies were entirely black; others had black stain applied to their black parts only so they appeared of normal colouring, but had about the same amount of black stain as the all-black queens. The attractiveness of the two types when suspended near scenting places was compared simultaneously during ten observation periods. Males responded to the all-black queens 42 times and to the normally coloured queens 26 times.

Next, large balsa wood models (30×15 mm), of the type used previously, but painted a) all black, b) black with three orange stripes and c) all blue, were compared simultaneously (Table 5). The black models and models with stripes elicited more reaction from the males than the blue models ($P < 0.05$ for each comparison).

TABLE 5

Comparison of attractiveness of models of different colours to B. pratorum males. (Combined results of 10 observation periods, during which each type of model was visited by 10 males)

Model	Behaviour of males towards models	
	Inspected	Touched
All black	10	7
Black with orange stripes	11	4
All blue	6	0

Effect of movement.

Virgin queens were either secured with threads in such a way that they moved little, or suspended from a small branch which was kept slowly swaying back and forth. During ten observation periods the immobile and mobile queens received 49 and 51 responses respectively and males mated twice with each type.

In a similar experiment live, suspended queens were compared with queens recently killed by cold; the live and dead queens elicited 46 and 56 responses respectively, including 2 and 1 attempted matings during the course of 12 observation periods.

Effect of age and condition of queen.

The attractiveness of the following were compared: a) virgin queens 2-3 days old, b) mated queens 7-14 days old, c) old queens that had previously headed a colony. Each type elicited a similar number of responses but, whereas males attempted to mate with the young queens, none mated with the old mother queens, (Table 6).

TABLE 6

Effect of age and condition of the queen on the mating behaviour of B. pratorum males. (Combined results of 11 observation periods, during which each type of queen was visited by 10 males)

	Behaviour of males towards queens			Mated or attempted to mate for:	
	Inspected	Touched	Alighted	< 30 sec. duration	> 30 sec. duration
Young virgin	8	18	4	4	5
Young mated queen	7	7	4	2	10
Old mother queen	7	19	8	0	0

In a further 13 observation periods, the attractiveness of virgin queens 12 hours old, 24-36 hours old and 4-5 days old, was compared. Each was about equally attractive, eliciting 59, 53 and 64 responses respectively.

Effect of odour of queen.

Three cylindrical cages of perforated zinc (each 50 × 20 mm) were put near each of three scenting places; one cage contained a virgin queen, the second contained 4 males and the third contained 4 workers. Most of the males visiting the scenting places ignored the cages. During the 15 observation periods, 6 males inspected the cage with workers but did not alight, and 2 similarly inspected the cage with males. However, of the 10 males that inspected the cage containing a queen, 9 alighted and 6 of these ran up and down the outside of the cage apparently following the movement of the queen inside.

Individual workers were kept in small cages with queens for 24 hours so that they might acquire something of the queens' odours. However, the attractiveness of workers treated in this way resembled that of workers kept in isolation for 24 hours (68 and 54 responses by males during which they alighted 27 and 20 times respectively). Similar experiments in which males and balsa wood were confined with queens gave similar results.

Comparison of the attractiveness of queens of different species.

The attraction of *B. pratorum*, *B. hortorum* and *B. terrestris* queens suspended near scenting places along a *B. pratorum* flight route were simultaneously compared. The *B. terrestris* queens received more responses than the *B. hortorum* (59 : 33) and as many as the *B. pratorum* queens (59 : 48), but the *B. pratorum* males alighted on, or attempted to mate, more with *B. pratorum* queens than *B. hortorum* or *B. terrestris* queens (Table 7), although one spent a considerable time attempting to mate with a *B. hortorum* queen.

TABLE 7

Comparison of the attractiveness of bumblebee queens of different species to *B. pratorum* males. (Combined results of 13 observation periods during which each type of queen was visited by 10 males)

	Behaviour of males towards queens			Mated or attempted to mate for:	
	Inspected	Touched	Alighted	< 30 sec. duration	> 30 sec. duration
<i>B. pratorum</i> queen	8	19	15	6	0
<i>B. hortorum</i> queen	6	22	4	0	1
<i>B. terrestris</i> queen	21	33	5	0	0

Attractiveness of visiting places.

Attempts were made to increase the attractiveness of *B. hortorum* visiting places to *B. hortorum* males by adding freshly crushed heads of *B. hortorum* males to them, and to decrease their attractiveness by adding freshly crushed *B. pratorum* heads, but without success.

DISCUSSION AND CONCLUSIONS

Because queens seem to attract males only near scenting places, which free flying queens have been seen to visit, it seems probable that males are attracted to females at such sites. However, even if mating begins at a visiting site it is unlikely to continue there, as the rapid accumulation of males on a queen when the pairing couple cannot move elsewhere would probably hinder copulation.

Males of *B. pratorum* seeking a mate are primarily attracted to black objects of a size range similar to that of their queens. This size preference helps them to distinguish queens from males and other smaller insects. Indeed, *B. terrestris* queens, probably because of their large size, elicited as much initial response as *B. pratorum* queens and more than *B. hortorum* queens. The yellow and orange bands seem of little importance in attracting males. Perhaps this is associated with the greater preponderance of yellow on the bodies of male than female *B. pratorum*; the same would be true of *B. lapidarius* and *B. lucorum*.

The odour of a queen seems important in inducing males to seize her. In common with other insects with low visual acuity (*e.g.* the butterflies *Danaus gilippus berenica*, BROWER *et al.* 1965; and *Argynnis paphia*, MAGNUS, 1950) mistakes made during the visual approach can be rectified by scent stimuli at close quarters. Thus, the odour differences between bumblebee queens of different species (FREE 1958) enable males to distinguish between them. Probably interspecific mating is avoided, partly because the different species of bumblebees tend to establish their flight routes at different heights above the ground (HAAS, 1949) and partly because the scents used to mark the flight routes (see CALAM, 1969) are species-specific. Crushed heads of another species probably failed to alter the attractiveness of visiting sites because when males have learned a site their orientation to it is primarily visual.

Perhaps an odour change associated with age or ovary development enables males to distinguish between old mother queens and young queens. The ability to make such a distinction is necessary should males mount young queens in the nest (see FREE & BUTLER, 1959). The apparent importance of the head in releasing mating behaviour suggests that it might be the source of any pheromones concerned.

Although it seems improbable that any courtship is involved in the mating behaviour of bumblebees, it is not known whether a queen's response to the male plays any part in facilitating mating, or whether any maturation of behaviour patterns is involved. Perhaps queens are most receptive to males when following a flight route.

SUMMARY

1. Males were attracted to queens presented near a visiting place, but usually ignored those offered elsewhere along a flight route.

2. A black object the size of a queen successfully elicited inspection and attempts to mate; the orange and yellow bands on a queen's body and her movements seemed not to do so.

3. The odour of a queen is important in inducing males to alight on her. Odour differences probably account for the greater attractiveness of young than old queens.

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SOMMAIRE

1) Les mâles étaient attirés par les reines, placées près d'un lieu de visite mais en général ils n'ont tenu aucun compte des reines qui étaient offertes, ailleurs, le long d'une route de vol.

2) Un objet noir de la même taille qu'une reine incita les mâles à examiner la reine et à tenter l'accouplement, les raies orange et jaune tracées sur le corps d'une reine et des mouvements n'ont pas produit d'effet.

3) L'odeur d'une reine est importante pour faire descendre les mâles sur la dite reine. Les différences d'odeur expliquent probablement que les jeunes reines attirent davantage les mâles que les vieilles reines.