Chemical communication

Butterfly anti-aphrodisiac lures parasitic wasps

To locate their hosts, parasitic wasps can 'eavesdrop' on the intraspecific chemical communications of their insect hosts[1–3]. Here we describe an example in which the information exploited by the parasitic wasp *Trichogramma brassicae* is a butterfly anti-aphrodisiac that is passed from male to female *Pieris brassicae* butterflies during mating, to render them less attractive to conspecific males[4–6]. When the tiny wasp detects the odour of a mated female butterfly, it rides on her (Fig. 1) to her egg-laying sites and then parasitizes the freshly laid eggs. If this fascinating strategy is widespread in nature, it could severely constrain the evolution of sexual communication between hosts.

In two-choice olfactory bioassays (for methods, see supplementary information), females of the parasitic wasp *T. brassicae* showed a clear preference for odours of mated *P. brassicae* females or males (Fig. 2a). The wasps were more strongly attracted by the scents of male and mated female butterflies than of virgin females, but they did not discriminate between male and mated female butterflies. When offered separately against clean air, odours from male or mated female butterflies attracted the wasps significantly, but odours from virgin females did not (Fig. 2a).

In similar behavioural trials, we determined the concentration of the anti-aphrodisiac, benzyl cyanide[5], that is attractive to the wasp. When given solvent-treated virgin females as an alternative, wasps were significantly attracted by odour from virgin female butterflies treated with 2 μg ($P=0.008$, Wilcoxon's matched pairs signed rank test) and 1 μg ($P=0.016$, Wilcoxon's matched pairs signed rank test) benzyl cyanide. This result indicates that *T. brassicae* is using the anti-aphrodisiac benzyl cyanide from *P. brassicae* as a foraging cue.

In further two-choice bioassays, we investigated whether wasps mount a mated female butterfly in response to the anti-aphrodisiac. When wasps were exposed to butterflies, they preferred to climb on to mated females rather than on to males or virgin females (Fig. 2b). Discrimination between males and mated females in this assay was probably based on close-range cues other than benzyl cyanide. But when virgin female butterflies painted with 2 μg benzyl cyanide dissolved in hexane were offered against virgin female butterflies painted with hexane alone, wasps preferred to mount the butterflies treated with benzyl cyanide significantly more often than the controls (Fig. 2b).

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Figure 1 *Trichogramma brassicae* wasp (roughly 0.5 mm long), hitches a lift on a mated, female butterfly (*Pieris brassicae*).

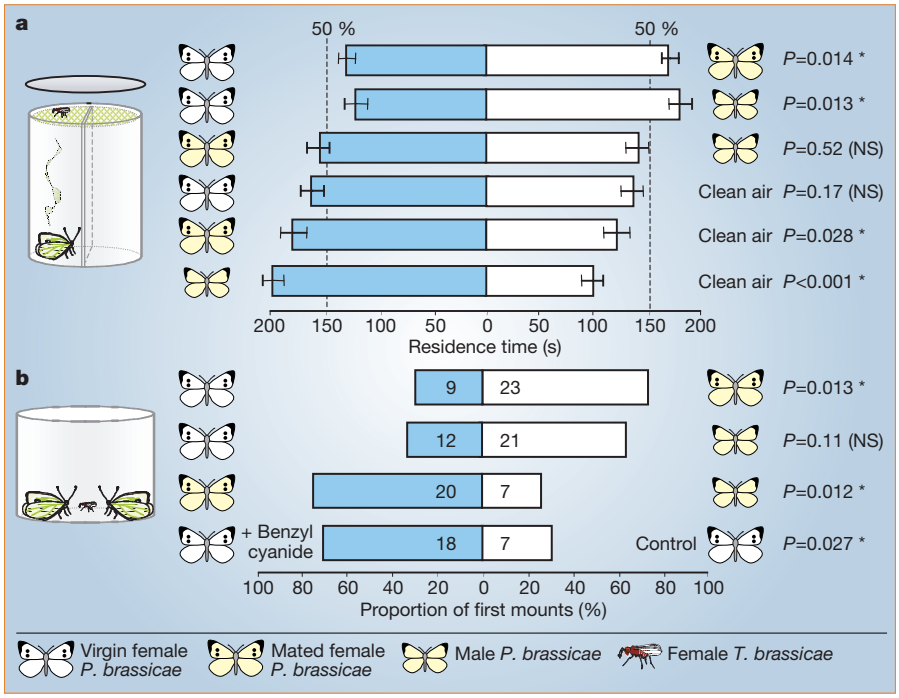

Figure 2 Response of *Trichogramma brassicae* parasitic wasps to signals of its host, the large cabbage-white butterfly *Pieris brassicae*. **a,** Mean residence time (±s.e.m.) spent by wasps in two odour fields in a static olfactometer; $n=50$ wasps tested per combination (Wilcoxon's matched pairs signed ranks test). **b,** Proportion of first mounts (%) by *T. brassicae* wasps on adult butterflies. The number of responding wasps is shown inside each bar; $n=40$ wasps tested per combination (χ^2 test). *Trichogramma* size is disproportionately large as drawn (see Fig. 1). Asterisks indicate significant differences within the choice test; NS, not significant.

We used a flight chamber to determine whether *T. brassicae* wasps are transported by the mated female butterfly to the host plant, the Brussels sprout (*Brassica oleracea* var. *gemmifera*), where they subsequently parasitize her freshly laid eggs. In total, 28 mated female butterflies were each seen carrying a single wasp; 14 wasps (50%) were found on the butterfly after it had landed on the host plant and had started laying eggs. Four of these wasps reached the host plant by descending from the butterfly, and two of them parasitized the new eggs after leaving the butterfly. Transportation (phoresy) on female butterflies therefore led to successful parasitism by *T. brassicae* in at least 7.1% of cases.

Phoresy is an ambush strategy[7,8] that has been only rarely observed in *Trichogramma* spp. (ref. 9; and N. E. F. and M. E. H., unpublished observations). But host location by chemical espionage in combination with phoresy, which we describe here, may have evolved frequently in egg parasitoids. From an egg parasitoid's point of view, it is most adaptive in those species with limited flight, a narrow time window for successful parasitism[10], and/or a preference for hosts that lay egg clutches. From the host's perspective, egg parasitoids represent significant mortality factors[11] and so exert selection on butterfly intraspecific communication, seriously constraining its evolution. The link between genetic variation in this important trait and local parasitoid selection pressure warrants further investigation.

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Supplementary information accompanies this communication on *Nature*'s website.

Competing financial interests: declared none.