

Honeybees consider flower danger in their waggle dance

Kevin R. Abbott*, Reuven Dukas

Department of Psychology, Neuroscience and Behaviour, McMaster University

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Like most animals, honeybees, *Apis mellifera*, possess a suite of antipredatory adaptations used to defend their colony against intruders and to avoid flowers associated with predation risk. Honeybees also possess a remarkable ability to communicate the direction, distance and relative profitability of flower patches to hive mates using the well-studied waggle dance. Here we show that honeybees returning from foraging on dangerous flowers are less likely to perform the waggle dance and engage in fewer waggle runs than foragers returning from equally rewarding, safe flowers. Our results indicate that experienced foragers effectively steer naïve recruits away from dangerous flowers and raise interesting questions as to how information about the reward and risk properties of patches are integrated into the waggle dance. © 2009 The Association for the Study of Animal Behaviour. Published by Elsevier Ltd. All rights reserved.

The honeybee waggle dance is a well-studied and surprisingly sophisticated example of animal communication. The waggle dance was first deciphered by von Frisch (1967), who determined that honeybee foragers communicate the location of profitable flower patches to hive mates using the waggle-run portion of the dance, where the direction and distance to a patch are indicated by the angle and duration of the waggle run, respectively. Subsequent work has demonstrated the flexibility of this mode of communication. For example, honeybees are more likely to dance and perform more waggle runs when returning from food sources that contain more concentrated sucrose solutions (von Frisch 1967; Seeley 1994; Seeley et al. 2000). It thus appears that foragers integrate a number of relevant foraging parameters into a measure of profitability, and that the probability of dancing, and the number of waggle runs performed are positively correlated with this measure of profitability (von Frisch 1967; Waddington 1982; Seeley 1994). While honeybees tune the waggle dance in a way that, under natural settings, would lead to greater recruitment of foragers to patches with better reward properties, it is unknown whether honeybees can also tune the waggle dance in response to predation risk at flowers, which may render a patch less valuable to their colony (Clark & Dukas 1994).

Several recent studies have documented that honeybees avoid flowers containing cues of elevated predation risk. For example, honeybees avoided flowers where they had previously experienced a simulated predator attack, flowers that harboured ambushing spiders and flowers that contained dead conspecifics (Dukas 2001a; Suttle 2003; Reader et al. 2006). Furthermore, in a large-scale field experiment with crab spiders (*Misumena vatia*), which are common flower-dwelling ambush predators (Morse 2007), milkweed (*Asclepias syriaca*) patches that contained crab spiders received significantly fewer honeybee visits than did control patches with no crab spiders (Dukas & Morse 2005). Since individual honeybees avoid locations with elevated risk of predation, it seems plausible that experienced foragers might modify the waggle dance in response to the perceived risk at flowers in order to facilitate the avoidance of predation risk by naïve recruits.

To determine whether perceived danger affects the waggle dance, we trained honeybees to visit two equally profitable artificial flowers. One flower was safe whereas the other contained a cue of predation risk. We predicted that bees returning from foraging trips on the dangerous flower would perform fewer waggle runs than foragers arriving from the safe flower.

METHODS

The experiment was conducted in the summer of 2008 at the Wildlife Research Station in Algonquin Park, Ontario, Canada (see Dukas 2008 for description of field site and basic set-up). We

* Correspondence: K. R. Abbott, Animal Behaviour Group, Department of Psychology, Neuroscience and Behaviour, McMaster University, 1280 Main Street West, Hamilton, ON L8S 4K1, Canada.

E-mail address: abbotkr@mcmaster.ca (K.R. Abbott).

trained honeybees from a two-frame observation hive, with approximately 2000 workers, to forage on two artificial flowers that were approximately 250 m from the colony and 60 m apart. Flowers consisted of jars of scented sucrose solution inverted over grooved Plexiglas plates (von Frisch 1967). To aid in recruitment, and to facilitate the faithfulness of foragers to a single flower, one flower was consistently scented with anise extract, and the other was consistently scented with mint extract. Whenever additional foragers were required, we habituated established foragers to a 1.25 M sucrose solution for at least 1 h before increasing the concentration to 2.5 M. This increase in sucrose concentration is known to promote dancing by established foragers and, thus, the recruitment of new foragers (von Frisch 1967; Higo et al. 1995; Seeley 1996). The dance response of foragers to increased nectar concentration (i.e. probability of dancing and number of waggle runs performed) was relatively weak throughout the experiment, probably because of an abundance of natural sources of nectar. All the bees used in this experiment were faithful to a single flower, and preliminary analyses indicated that the number of waggle runs performed was similar for the anise and mint flowers (two-tailed Mann–Whitney U test: $U = 108$, $N_{\text{mint}} = 15$, $N_{\text{anise}} = 17$, $P = 0.4$).

Trials were run on warm ($>20^\circ\text{C}$), sunny afternoons (1400–1800 hours) with one to four individually marked focal bees that had been feeding on a 1.25 M sucrose solution for at least 1 h. To equalize the number of foragers on the two feeders as closely as possible, we removed excess bees from the more active feeder before the start of the trial. At the beginning of each trial, the concentration of the sucrose solution at each flower was increased to 2.5 M to encourage dancing. Two recently killed stimulus bees (Dukas 2001a) were placed on the experimental flower so that they were conspicuous but did not interfere with foraging. For the duration of the 30 min trials, observers at the flowers recorded the number of bee visits and notified hive observers by radio when focal bees that had completed a full flower visit lasting at least 1 min departed for the hive. Two hive observers, blind to flower treatment, recorded the number of waggle runs performed by the focal bees. The dance behaviour of a single focal bee was observed up to three times per trial. A focal bee was only tested in one trial and only bees that had not previously visited a flower containing a dead bee were tested. We could not quantify recruitment because many recruits approached the dangerous flower but did not land, and such bees could often not be identified.

We ran eight matched trials, with anise and mint flowers serving on half the trials as the safe flower and on half the trials as the dangerous flower in random order. Technical difficulties in three trials interfered with counting the number of visitors, which reduced the sample size in the rate-of-visits analysis by two (see below; in one of these trials, the rate could still be calculated based on a 23 min observation period, rather than a 30 min observation period). Owing to small sample sizes and non-normal data, we used nonparametric statistical tests.

RESULTS

On average, bees returning from safe flowers performed about 20 times more waggle runs than bees returning from dangerous flowers (two-tailed Mann–Whitney U test: $U = 46.5$, $N_{\text{safe}} = 18$, $N_{\text{dangerous}} = 14$, $P = 0.001$; Fig. 1). We also conducted a matched comparison using trials as the independent experimental units. This analysis indicated that approximately 30 times more waggle runs were performed by bees visiting the safe flower than the dangerous flower (two-tailed Wilcoxon signed-ranks test: $T = 0$, $N = 8$, $P = 0.012$; Fig. 1).

The above analyses included many cases where the focal bee did not dance on one or more observations. This allowed us to examine

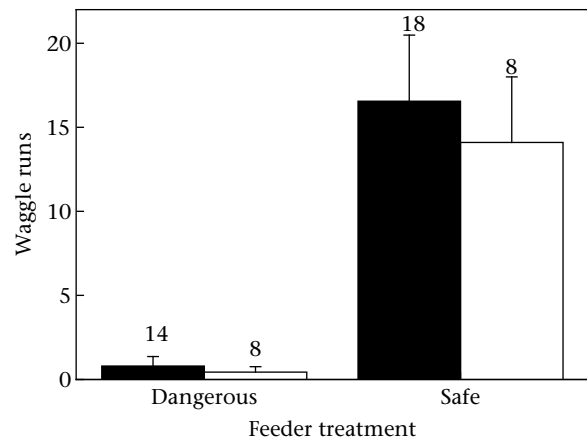


Figure 1. Mean \pm SE number of waggle runs as a function of flower danger. ■: mean number of waggle runs when each bee was considered an independent statistical unit. □: mean number of waggle runs when each trial was considered the independent unit. Sample sizes are depicted above the bars.

whether the experimental manipulation also affected the probability of dancing. An analysis including only the dance data from the first foraging bout for each bee in a trial revealed that bees that had visited the safe flower were eight times more likely to perform at least one waggle run than bees that had visited the dangerous flower (10 of 18 safe-flower bees danced and 1 of 14 dangerous-flower bees danced; two-tailed Fisher's exact test: $P = 0.008$).

Consistent with previous findings indicating that honeybees avoid flowers associated with danger (Dukas 2001a; Suttle 2003; Reader et al. 2006), the safe flower received significantly higher rates of bee visits than did the dangerous flower (mean \pm SE = 0.68 ± 0.11 versus 0.41 ± 0.07 visits/min; two-tailed Wilcoxon signed-ranks test: $T = 0$, $N = 5$, $P = 0.043$) even though the number of bees visiting each flower was equalized before the start of each trial.

DISCUSSION

Our results demonstrate that the presence of a cue of predation risk depresses the dance behaviour of honeybees. Previous studies have shown that the waggle dance is modulated by the reward properties of flowers (von Frisch 1967; Waddington 1982; Seeley 1994; Seeley et al. 2000; Afik et al. 2008; Seefeldt & De Marco 2008). Our study indicates that predation risk, a property unrelated to the quality, quantity, accessibility, or distribution of reward, also affects honeybees' dance behaviour.

The simplest interpretation of our results is that honeybees, which are well known to show antipredatory behaviour at flowers (Dukas 2001a; Suttle 2003; Dukas & Morse 2005; Reader et al. 2006), also extend their individual response to cues of predation to their waggle dance. That is, the mechanism that allows individual honeybees to evaluate danger and to prefer safe over dangerous flowers also increases their tendency to perform the waggle dance when returning from safe, rather than dangerous, flowers. An unlikely alternative is that the fewer waggle runs for dangerous flowers than for safe flowers merely indicate bees' perception of lower profitability owing to increased vigilance. This alternative is not consistent with either the data indicating antipredatory behaviour of honeybees cited above, or the very large, 20–30-fold decrease in waggle runs for the dangerous flower than for the safe flower (Fig. 1). Nevertheless, it is possible that bees in the two conditions perceived different levels of profitability. The interacting effects of cues of predation risk and perceived profitability on dance behaviour would be a suitable target for future research. Regardless

of the exact mechanism involved, however, any reduction in recruitment to dangerous patches should reduce the predation costs incurred by recruits and thus increase colony fitness (Clark & Dukas 1994).

We have assumed that the foragers perceived the dead bees as a cue of predation risk and acted accordingly. There are two other ways in which the dead bees could have been perceived that might affect our interpretation. The first is as a novel object, and our results could be the result of neophobia. Dukas (2001a) showed, however, that honeybees that avoided a recently killed conspecific did not avoid a comparable novel object. Alternatively, foragers may have perceived the dead bees as competitors (i.e. they may not have perceived them as dead) and our results could be the result of an aversion to cues of competition risk. However, the artificial flowers used in our experiment were designed for the simultaneous use by numerous honeybees (von Frisch 1967; Seeley 1996), so the presence of two live bees at the feeder should not reduce the perceived quality of that feeder. Furthermore, crush-killed honeybees emit alarm pheromone that is highly conspicuous to humans as well as conspecifics, which show characteristic alarm behaviour (Dukas 2001a). Indeed, Abbott (2006) found that when bumblebees (*Bombus* spp.) were given a choice between an artificial flower containing a freshly crush-killed conspecific (killed as in this experiment and emitting alarm pheromones) and one containing a freeze-killed conspecific (not emitting alarm pheromones), the foragers preferred the flower with the freeze-killed bee.

Our study suggests that honeybees integrate two distinct types of information, patch profitability and predation risk, into a single signal, the number of waggle runs. This suggests a promising line of research investigating the mechanisms underlying such integration at the neural and behavioural levels. For example, using the proboscis extension reflex paradigm, Hammer (1993) identified a honeybee interneuron that produces more action potentials when either sucrose or an odour previously paired with sucrose is presented. A neuroeconomic study (Hammer 1997; Sanfey et al. 2006) that combines this approach with cues of predation risk could lead to the identification of either individual neurons or neural networks that are involved in the integration of information about food quality and predation risk and its translation into a communication output.

The waggle dance is a remarkable example of social learning about food as well as other resources, such as water and nest sites (von Frisch 1967; Seeley 1996; Dyer 2002; Visscher 2007). A variety of animals also show social learning about predators (Griffin 2004). Although our results do not address explicit social learning about predation risk, they do illustrate that implicit social learning about danger can be achieved merely by modulating the social transmission of information about food, which results in a disproportionate recruitment of naïve individuals to safe sites. Similar forms of implicit social learning of danger may allow ants to avoid foraging sites associated with predation risk (Nonacs 1990) or nesting sites associated with intraspecific aggression (Franks et al. 2007), and allow rats to avoid poisonous food (Galef 1985). Such indirect communication about predation risk may have had an important yet underappreciated effect on the dynamics and spatial distribution of predators and prey (Sih 1998), and on insect–plant interactions (Dukas 2001b).

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References

- Abbott, K. R. 2006. Bumblebees avoid flowers containing evidence of past predation events. *Canadian Journal of Zoology*, **84**, 1240–1247. doi:10.1139/Z06-117.
- Afik, O., Dag, A. & Shafir, S. 2008. Honeybee, *Apis mellifera*, round dance is influenced by trace components of floral nectar. *Animal Behaviour*, **75**, 371–377. doi:10.1016/j.anbehav.2007.04.012.
- Clark, C. W. & Dukas, R. 1994. Balancing foraging and antipredator demands: an advantage of sociality. *American Naturalist*, **144**, 542–548.
- Dukas, R. 2001a. Effects of perceived danger on flower choice by bees. *Ecology Letters*, **4**, 327–333.
- Dukas, R. 2001b. Effects of predation risk on pollinators and plants. In: *Cognitive Ecology of Pollination* (Ed. by L. Chittka & J. Thomson), pp. 214–236. Cambridge: Cambridge University Press.
- Dukas, R. 2008. Life history of learning: performance curves of honeybees in settings that minimize the role of learning. *Animal Behaviour*, **75**, 1125–1130. doi:10.1016/j.anbehav.2007.08.029.
- Dukas, R. & Morse, D. H. 2005. Crab spiders show mixed effects on flower-visiting bees and no effect on plant fitness components. *Ecoscience*, **12**, 244–247.
- Dyer, F. C. 2002. The biology of the dance language. *Annual Review of Entomology*, **47**, 917–949.
- Franks, N. R., Dornhaus, A., Hitchcock, G., Guillem, R., Hooper, J. & Webb, C. 2007. Avoidance of conspecific colonies during nest choice by ants. *Animal Behaviour*, **73**, 525–534. doi:10.1016/j.anbehav.2006.05.020.
- von Frisch, K. 1967. *The Dance Language and Orientation of Bees*. Cambridge, Massachusetts: Harvard University Press.
- Galef, B. G. 1985. Direct and indirect behavioral processes for the social transmission of food avoidance. In: *Experimental Assessments and Clinical Applications of Conditioned Food Aversions* (Ed. by P. Bronstein & N. S. Braveman), pp. 203–215. New York: New York Academy of Sciences.
- Griffin, A. S. 2004. Social learning about predators: a review and prospectus. *Learning & Behavior*, **32**, 131–140.
- Hammer, M. 1993. An identified neuron mediates the unconditioned stimulus in associative olfactory learning in honeybees. *Nature*, **366**, 59–63. doi:10.1038/366059a0.
- Hammer, M. 1997. The neural basis of associative reward learning in honeybees. *Trends in Neurosciences*, **20**, 245–252. doi:10.1016/S0166-2236(96)01019-3.
- Higo, H. A., Winston, M. L. & Slessor, K. N. 1995. Mechanisms by which honey-bee (Hymenoptera, Apidae) queen pheromone sprays enhance pollination. *Annals of the Entomological Society of America*, **88**, 366–373.
- Morse, D. H. 2007. *Predator upon a Flower: Life History and Fitness in a Crab Spider*. Cambridge, Massachusetts: Harvard University Press.
- Nonacs, P. 1990. Death in the distance: mortality risk as information for foraging ants. *Behaviour*, **112**, 23–35.
- Reader, T., Higginson, A. D., Barnard, C. J., Gilbert, F. S. & The Behavioural Ecology Field Course 2006. The effects of predation risk from crab spiders on bee foraging behavior. *Behavioral Ecology*, **17**, 933–939. doi:10.1093/beheco/arl027.
- Sanfey, A. G., Loewenstein, G., McClure, S. M. & Cohen, J. D. 2006. Neuroeconomics: cross-currents in research on decision-making. *Trends in Cognitive Sciences*, **10**, 108–116. doi:10.1016/j.tics.2006.01.009.
- Seefeldt, S. & De Marco, R. J. 2008. The response of the honeybee dance to uncertain rewards. *Journal of Experimental Biology*, **211**, 3392–3400. doi:10.1242/jeb.017624.
- Seeley, T. D. 1994. Honey bee foragers as sensory units of their colonies. *Behavioral Ecology and Sociobiology*, **34**, 51–62.
- Seeley, T. D. 1996. *The Wisdom of the Hive*. Cambridge, Massachusetts: Harvard University Press.
- Seeley, T. D., Mikheyev, A. S. & Pagano, G. J. 2000. Dancing bees tune both duration and rate of waggle-run production in relation to nectar-source profitability. *Journal of Comparative Physiology A*, **186**, 813–819. doi:10.1007/s003590000134.
- Sih, A. 1998. Game theory and predator–prey response races. In: *Advances in Game Theory and the Study of Animal Behaviour* (Ed. by L. A. Dugatkin & H. K. Reeve), pp. 221–238. Oxford: Oxford University Press.
- Suttle, K. B. 2003. Pollinators as mediators of top-down effects on plants. *Ecology Letters*, **6**, 688–694. doi:10.1046/j.1461-0248.2003.00490.x.
- Visscher, P. K. 2007. Group decision making in nest-site selection among social insects. *Annual Review of Entomology*, **52**, 255–275. doi:10.1146/annurev.ento.51.110104.151025.
- Waddington, K. D. 1982. Honey bee foraging profitability and round dance correlates. *Journal of Comparative Physiology A*, **148**, 297–301. doi:10.1007/BF00679014.