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Honeybee Navigation: Nature and Calibration of the "Odometer"

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There are two theories about how honeybees estimate the distance to food sources. One theory proposes that distance flown is estimated in terms of energy consumption. The other suggests that the cue is visual, and is derived from the extent to which the image of the world has moved on the eye during the trip. Here the two theories are tested by observing dances of bees that have flown through a short, narrow tunnel to collect a food reward. The results show that the honeybee's "odometer" is visually driven. They also provide a calibration of the dance and the odometer in visual terms.

It is well known that honeybees navigate accurately and repeatedly to a food source, as well as communicate to their nestmates the distance and direction in which to fly to reach it, through the "waggle dance" (*1*). However, the cues by which bees gauge the distance to the goal have been controversial. Early work suggested that flight distance is estimated in terms of energy consumption (*2*). More recent studies suggest that the primary cue is the integral, over time, of the image motion that is experienced en route (*3–6*). Here we put the two theories to a stringent test by recording dances of bees that have been trained to fly into a short, narrow tunnel to collect a food reward.

The experimental bees (*Apis mellifera ligustica* Spinola) were housed in a two-frame observation hive, with transparent walls on both sides. One frame was positioned above the other so that all comb faces were visible for observing and filming dances. The hive was located on the inside wall of a building, with an exit to the outside. Typically, six bees were individually marked and used for each experiment.

In one series of experiments, individually marked bees were trained to forage at a feeder carrying sugar solution placed in a wooden tunnel 6.4 m long, 11 cm wide, and 20 cm high. The tunnel was positioned outdoors near the hive. The far end was closed, and bees could enter and leave the tunnel only at the near end. The top of the tunnel was covered with black insect-screen cloth, which permitted observation and provided the bees with a view of the sky.

In experiment 1, the tunnel was positioned with its entrance 35 m from the hive, and was oriented along the direction to the hive (Fig. 1A). The walls and floor of the tunnel were lined with a random visual texture (*7*). The feeder was placed at the entrance to the tunnel. Bees returning from the feeder performed predominantly round dances: The probability of a round dance was 85.2% (Fig. 1B). This is consistent with the fact that *A. mellifera ligustica* performs mainly round dances when visiting food sources that are within 50 m of the hive (*8*). However, when the feeder was placed 6 m inside the tunnel (experiment 2), the bees performed primarily waggle dances: The probability of a waggle dance was 90.0% (Fig. 1B) (*9*). This change from round dances to waggle dances occurred while the distance flown by the bees had increased by a mere 6 m, from 35 m in experiment 1 to 41 m in experiment 2. Clearly, in experiment 2, the feeder was still at a distance at which bees normally perform round dances when flying outdoors.

Why were the bees performing waggle dances in experiment 2? One possibility is that flight in the narrow tunnel generated a large integrated optic flow on the eye, mim-

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icking the effect of a long flight in natural outdoor conditions. The distances to the walls and the floor would typically be much smaller than those to nearby objects or the ground during free flight in an open environment. Therefore, if the bee moves forward by, say, 1 cm, it would experience a greater angular

motion of the image when flying in the tunnel than when flying outdoors. The magnified image motion in the tunnel might cause the bees to infer a journey considerably longer than 6 m.

The possibility that image motion is indeed the cue for estimating flight distance

was tested in experiment 3 (Fig. 1A). Here the tunnel and the feeder were positioned as in experiment 2, but the walls and floor were lined with axially oriented stripes (10). This tunnel provided negligible image motion cues, because the stripes were parallel to the direction of flight. Bees returning from this tunnel produced predominantly round dances: The probability of a round dance was 86.7% (Fig. 1B), even though these bees had flown exactly the same physical distance—41 m—as those in experiment 2. Evidently, the lack of image motion in the axial-striped tunnel caused the bees to infer that they had flown a very short distance.

In a further experiment (experiment 4), the tunnel carried a random texture, as in experiments 1 and 2, but was positioned such that the entrance was only 6 m from the hive exit (Fig. 1A). The tunnel was pointed toward the hive, and the feeder was placed 6 m inside the tunnel. Bees returning from this tunnel performed mainly waggle dances: The probability of a waggle dance was 87.5% (Fig. 1B), even though the feeder was now only 12 m from the hive. We conclude from these experiments that distance flown is inferred on a visual basis, the primary cue being the extent of image motion experienced by the eye.

How is image motion translated into a percept of distance flown? To examine this question, we carried out another series of experiments in which we recorded the dances of marked bees returning to our hive from feeders placed at various locations in the outdoor environment of the campus of the Australian National University. The feeders were positioned at distances of 60, 110, 150, 190, 225, 340, and 350 m from the hive (11). Data were obtained from two different hives, H1 and H2, each at a different location on campus, to check for possible colony-specific or route-specific differences (12). Bees returning from these feeders performed primarily waggle dances at all of the distances tested. The mean durations of the waggle phases (13) of the dances for each of these feeder distances are plotted in Fig. 2 [see Table 1 and (14)]. The waggle duration increases approximately linearly with distance flown, as is well known for distances up to 1 km (15). Linear regression on the data yields a correlation coefficient of 0.998. The slope of the regression line is 1.88 ms of waggle duration per meter of distance traveled. This value is in close agreement with classical published data for comparable flight distances (15). For any given distance, there were no significant differences between the durations of the waggle phases recorded from the two colonies. We infer from this that the bees from the two hives flew through essentially similar outdoor environments (see below).

How do the dances of bees returning from the tunnels in experiments 2 and 4 compare with those of bees flying in the open outdoor

Fig. 1. (**A**) Layout for experiments using tunnels. Each tunnel represents a separate experiment (1, 2, 3, or 4). The dot in the tunnel shows the position of the feeder in each case. (**B**) Probability of waggle (W) round (R) dance for experiments 1 to 4. N and n represent the numbers of bees and dances analyzed, respectively, in each experiment.
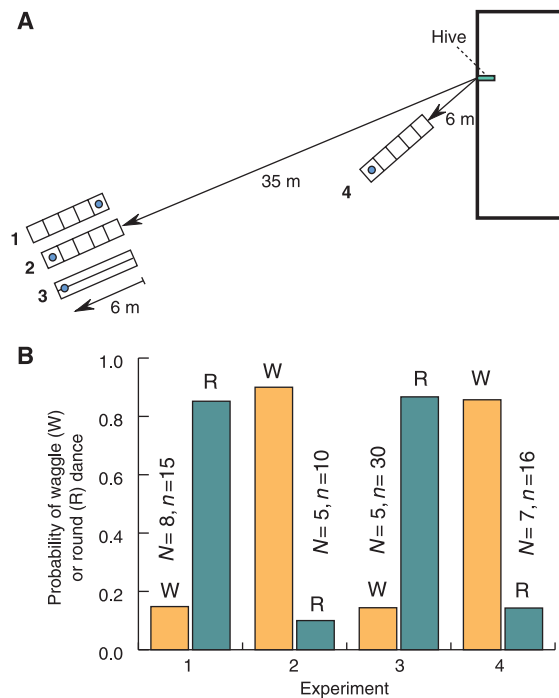

Fig. 2. Mean waggle durations of dances elicited by outdoor feeders at various distances d. The straight line is a linear regression on the data, defined by the expression $\tau = 95.91 + 1.88d$. Also shown are the mean waggle durations measured in the tunnel experiments (experiments 2 and 4) and their equivalent outdoor flight distances as read off from the regression line.
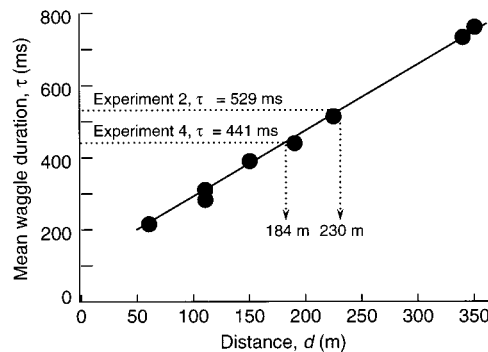

Table 1. Details of measurements of waggle dances in various experiments. SD, standard deviation of mean waggle durations measured for different bees.

Experiment	Hive	Waggle duration (ms) (mean ± SD)	Number of bees analyzed	Number of dances analyzed	Number of waggle phases analyzed
Outdoor feeder at 60 m	H1	217.5 ± 65.2	3	10	92
Outdoor feeder at 110 m	H1	283.0 ± 66.0	5	10	93
Outdoor feeder at 110 m	H2	312.4 ± 44.4	6	14	181
Outdoor feeder at 150 m	H1	390.9 ± 62.5	3	10	92
Outdoor feeder at 190 m	H1	441.2 ± 60.0	6	7	65
Outdoor feeder at 225 m	H2	514.4 ± 74.8	8	21	345
Outdoor feeder at 340 m	H2	733.3 ± 116.4	10	23	222
Outdoor feeder at 350 m	H1	762.6 ± 129.4	5	9	87
Tunnel experiment 2	H2	528.8 ± 67.6	4	9	216
Tunnel experiment 4	H2	441.2 ± 57.0	7	16	138

environment? We found no qualitative differences. In either case, bees signaled the direction of the feeder in their waggle dances, regardless of the time of day: The direction of the waggle axis shifted in step with the sun's movement in the sky.

However, bees returning from the tunnels massively overestimated the distance that they had traveled. In experiment 2, the bees flew 35 m in a natural outdoor environment, followed by 6 m in the tunnel—a total of 41 m to the feeder. But analysis of their dances revealed a mean waggle duration as long as 529 ms. This corresponds to a flight of 230 m in an outdoor environment, as revealed by projection of this duration on the regression line of Fig. 2. Evidently, a flight of 6 m in the tunnel was perceived as equivalent to a flight of 195 m outdoors. In experiment 4, the bees flew 6 m outdoors, followed by 6 m in the tunnel. But this was signaled by a waggle dance of 441 ms, which, from the regression line, represents a distance of 184 m outdoors. In this case, the 6-m flight in the tunnel was perceived as equivalent to an outdoor flight of 178 m (see Table 1). Averaging the two results, we calculate that a 6-m flight in the tunnel is equivalent to a journey of 186 m in the outdoor environment. In other words, the tunnel magnifies the perception of distance flown by a factor of 31.

We can use the data from the tunnel experiments to calibrate the honeybee's odometer and the dance in terms of the visual input to the eye. This is because the dimensions of the tunnel are known and, unlike the situation in an open outdoor environment, flight in the tunnel is tightly constrained. In the tunnel, bees tend to fly along an axis roughly midway between the two walls (16) and halfway between the floor and the ceiling. The distance to each wall is therefore 5.5 cm, and the height above the floor is 10 cm. Assuming that distance flown in the tunnel is gauged primarily on the basis of the image motion that is experienced by the lateral fields of view of the eyes (6), we calculate that 1 cm of forward motion in the tunnel would cause the image of the wall to move backward by an angle of 10.3° in the lateral visual field. Therefore, 6 m of forward motion in the tunnel would generate 6180° of image motion. We have already shown that a 6-m flight in the tunnel corresponds to a flight of 186 m in a natural outdoor environment. From the slope of the regression line in Fig. 2, we calculate that 186 m of outdoor flight is encoded by a waggle duration of 350 ms. Therefore, 1 ms of waggle in the dance encodes (6180/350) = 17.7° of image motion in the eye (17).

Traditionally, the honeybee's odometer has been calibrated in terms of the waggle duration, the number of waggles, or the waggle length, per meter of distance flown (15). However, it is now clear that the distance flown is not perceived directly in distance units, but rather in terms of the amount of image motion that is experienced by the eye. The image motion that is generated during 1 m of forward flight would depend strongly on the average distance of foliage and the ground during the bee's journey, which could vary from one environment to another. Therefore, the traditional calibration cannot be an "absolute" one because it is environment-dependent. The correct way of specifying the calibration of the odometer would be in terms of the amount of image motion that is required to generate a given waggle duration. As derived above, this calibration is 17.7° of image motion per millisecond of waggle. We propose that this is the fundamental, absolute calibration of the honeybee's visual odometer. We surmise that, in nature, visual odometry of this kind is reliable because new recruits tend to take the same route as experienced foragers.

The total angular image motion that a bee experiences while flying a particular route depends only on how far she flies, and not on the speed of flight. The reason is as follows. Assume that the bee moves forward by a small distance Δd cm at a speed of V cm/s. The induced angular velocity of the image in the lateral field of view of the eye, A (in degrees per second), would depend on the distance of the scene being viewed. However, for a given scene, A would be proportional to V. Thus, $A = k \cdot V$, where k is a scene-dependent constant. On the other hand, the time Δt taken to fly the distance Δd is inversely proportional to V. That is, $\Delta t = \Delta d/V$. Therefore, during this interval, the angular motion of the image on the eye (in degrees) would be $\alpha = A \cdot \Delta t = k \cdot \Delta d$, which depends only on the distance traveled and is independent of flight speed. Thus, the total angular motion of the image is a robust measure of the distance traveled along a particular route. Indeed, there is some evidence that bees are able to estimate the distance traveled to a goal fairly accurately, regardless of how rapidly they fly to it (6), although this needs more careful examination.

If we assume that odometry in outdoor flights also relies primarily on image motion, then we can obtain from our data a rough estimate of the average distance of objects and the ground as experienced by the bees when they flew in the outdoor environment. Given that the tunnel amplifies the perception of distance flown by a factor of 31, and that the distance to each wall of the tunnel was 5.5 cm, we deduce that the average object distance outdoors was ~31 × 5.5 = 170 cm. Although this value is difficult to verify precisely, it is quite plausible for our environment. Thus, it is reasonable to infer that cues based on image motion play an important role even in outdoor flights of several hundred meters.

Bisetzky (18) elicited waggle dances from bees that were trained to walk through short tunnels. It is not clear what cues the bees were using in these experiments: The visual cues were not defined or controlled. Kirchner and Braun (19) were able to elicit waggle dances from tethered, flying bees that were exposed to moving visual patterns. They found that the duration of the waggle increased with flight duration. However, this finding does not distinguish between visual and energy-based cues, because both would increase with flight duration. The visual cues were not manipulated independently.

Our study suggests that honeybees use cues based primarily on image motion to monitor flight distances of hundreds of meters in natural outdoor environments. In the future it should be possible to study mechanisms of navigation and path integration conveniently, and under controlled conditions, by training bees to fly through short tunnels arranged in various configurations.

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