James C. Nieh

The honey bee shaking signal: function and design of a modulatory communication signal

Abstract This study explores the meaning and functional design of a modulatory communication signal, the honey bee shaking signal, by addressing five questions: (I) who shakes, (II) when do they shake, (III) where do they shake, (IV) how do receivers respond to shaking, and (V) what conditions trigger shaking. Several results confirm the work of Schneider (1987) and Schneider et al. (1986a): (I) most shakers were foragers (at least 83%); (II) shaking exhibited a consistent temporal pattern with bees producing the most signals in the morning (0810–1150 hours) just prior to a peak in waggle dancing activity; and (IV) bees moved faster (by 75%) after receiving a shaking signal. However, this study differs from previous work by providing a long-term, temporal, spatial, and vector analysis of individual shaker behavior. (III) Bees producing shaking signals walked and delivered signals in all areas of the hive, but produced the most shaking signals directly above the waggle dance floor. (IV) Bees responded to the signal by changing their direction of movement. Prior to receiving a signal, bees selected from the waggle dance floor moved, on average, towards the hive exit. After receiving a shaking signal, some bees continued moving towards the exit but others moved directly away from the exit. During equivalent observation periods, non-shaken bees exhibited a strong tendency to move towards the hive exit. (V) Renewed foraging activity after *food dearth* triggered shaking signals, and, the level of shaking is positively correlated with the *duration* of food dearth. However, shaking signal levels also increased in the morning before foraging had begun and in the late afternoon after foraging had ceased. This spontaneous afternoon peak has not previously been reported. The shaking signal consequently appears to convey the general message "reallocate labor to different activities" with receiver context specifying a more precise meaning. In the context of foraging, the shaking signal appears to activate (and perhaps deactivate) colony foraging preparations. The generally weak response elicited by modulatory signals such as the shaking signal may result from a high receiver response threshold which allows the receiver to integrate multiple sources of information and which thereby increases the probability that receiver actions will be appropriate to colony needs.

Key words Honey bee · Shaking signal · Dorsoventral abdominal vibration · Modulatory communication · Signal design

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Introduction

What is the function of signals that appear in a wide variety of contexts yet do not elicit strong responses in any context? How is the design of such signals adaptive? These questions underlie the study of modulatory signals, an important, yet relatively unexplored component of communication in social organisms.

Hölldobler and Wilson (1990, p. 253) define modulatory communication as communication systems that are "outwardly inefficient....[and] influence the behavior of receivers, not by forcing them into narrowly defined behavioral channels but by slightly shifting the probability of the performance of other behavioral acts." Modulatory communication is most frequently seen in complex animal societies where "a flexible program is required if the work force is to distribute its energy investment among the different tasks in an effective manner" (Hölldobler and Wilson 1990, p. 253). Although modulatory signals play an intriguing role in mediating complex social interactions, clear signal effects have been difficult to establish, and our understanding of these signals has consequently been slow to advance. First described by Haydak (1929), honey bee shaking signals

are frequently seen in a wide variety of contexts, are thought to play a role in food collection and colony reproduction, and are reported to produce diverse, weak effects on receivers (Allen 1959a,b; Fletcher 1978; Schneider et al. 1986a). Yet it has been difficult to fit all of these data within a single conceptual framework and arrive at an overall message for the shaking signal. The goal of this paper is to infer the general message of the shaking signal and to use this knowledge to better understand the function and design of modulatory communication signals.

A honey bee performs the shaking signal to another bee by vigorously and rhythmically shaking her body dorso-ventrally as her metathoracic legs grip the comb and her prothoracic and mesothoracic legs grip the receiver. The receiver remains still, does not appear to resist, and vibrates with the body of the shaker. Occasionally, a bee will simultaneously shake multiple bees. Shakers can also shake the comb by holding onto it with all six legs. While a bee shakes, she remains in one spot on the comb. When she is not shaking, she walks on the comb normally without showing any vibration in her gait. Gahl (1975) filmed shaking bees with a high-speed film camera and determined that, on average, a shaking signal lasted for 1.2 ± 0.3 s and produced vibrations at 16.3 ± 5.8 Hz.

This shaking behavior has been called a "jerking dance" or a "vibration dance" (von Frisch 1967; Schneider et al. 1986a). However, because shaking is a discrete behavior, not a continuous one such as the waggle or tremble dance (Seeley 1992), I will only refer to the signal itself. Allen (1956) provided the first systematic observations of this behavior, and I return to her usage by calling each shaking episode a "shaking signal", not a "dorso-ventral-abdominal vibration" (DVAV) (Milum 1955) because the whole body of the shaker, not just the abdomen, vibrates.

The goal of this study was to elucidate the design features and general message of the shaking signal. I have consequently organized it around five questions: (I) who shakes, (II) when do they shake, (III) where do they shake, (IV) how do receivers respond to shaking, and (V) what conditions trigger shaking. Under special circumstances, workers will shake queen bees (Allen 1958). Because this study focuses on shaking signals delivered between workers, I will only address queen shaking in the Discussion.

To establish a framework for these studies, I will begin by reviewing what is known about the shaking signal.

Who are the shakers ?

Milum (1955) first hypothesized a link between foraging and shaking based on observations of bees alternating between waggle dancing and producing shaking signals. Working with *Apis mellifera scutellata*, Schneider (1989) noted that 30–40% of marked foragers performed shaking signals upon returning to the hive. Moreover, the maximum morning level of shaking activity declined when exiting foragers were removed. These observations provided further evidence that most shakers are foragers. But are all shakers foragers? Longer observations of individual bees producing shaking signals may be more revealing.

Who is shaken?

Shakers appear to target bees younger than themselves. Allen (1959b) reported that shaken workers ranged in age from 0 to 55 days, with bees of age 14–21 days having the highest probability of being shaken. Gahl (1975) reported that receivers varied in age from 0 to 26 days.

Where and when do they shake?

The temporal and spatial distributions of shaking are intriguing. Seeley et al. (in press) observed bees previously identified as foragers shaking in the early morning before any bees had left the hive to begin foraging. Schneider et al. (1986a) found that bees performed the majority of shaking signals on the dance floor (the hive region where most waggle dances occur) and that overall levels of shaking were much higher in the spring and summer than in fall and winter. Allen (1959b) reported the same seasonal trend. Within each day, shaking began before there was much flight activity and continued at reduced levels even after bees had stopped waggle dancing and flying for the day (Allen 1959b; Schneider et al. 1986a). Thus more information about the function of shaking signals may be gleaned from carefully analyzing the changing distribution of signals in all areas of the hive at different times of day.

How do bees respond to shaking?

Foraging-age bees that received shaking signals moved significantly more often into the waggle dance floor than into other areas of the hive, moved faster than non-shaken bees of similar age, and spent more *total time* and increased the *overall rate* of performing the following tasks: grooming another bee, attending the queen, engaging in trophallaxis, ventilating, building and trimming cell cappings, building and shaping comb, performing waggle dances, and entering and remaining in a brood or food cell for greater than 5 s (Schneider et al. 1986a; Schneider and McNally 1991). Young bees not of foraging age (1–13 days old) increased the time that they spent in a brood cell (but not in a food cell) and their rate of visiting food cells and cell capping (Schneider 1987) after receiving a shaking signal. Thus bees evidently increased their overall activity levels after receiving a shaking signal.

An earlier study on the movement of bees before and after they were shaken (Schneider et al. 1986a), provided indirect evidence for changes caused by the shaking signal because shaken bees were compared with control bees. To obtain direct evidence, I observed individuals before and after they had received a shaking signal and conducted an error vector analysis to test for changes in walking direction. I also replicated the work of Schneider et al. 1986a; Schneider 1991; and Schneider and McNally 1991, and tested for changes in (1) walking speed, (2) the time spent on various tasks, and (3) the rate of performing various tasks.

What causes bees to shake?

Initial foraging success may trigger the production of shaking signals. Schneider (1989) reported that 52% of successful nectar foragers and 67% of pollen foragers began shaking within 5 min of entering the hive. Furthermore, peaks in shaking activity generally preceded peaks in waggle dancing activity throughout the year (Schneider et al. 1986a). When Schneider et al. (1986b) controlled access to food by using a flight cage, they observed a peak in shaking activity within 30 min of foraging. Seeley et al. (in press) placed an observation hive in a natural environment with virtually no natural food sources and withheld artificial food for several days. When they subsequently provided a feeder, they observed that foragers performed shaking signals, not waggle dances, upon returning from their first few trips to the feeder. Foragers did not begin to waggle dance until their third or fourth trip back from the feeder. These data demonstrate that sudden access to food outside the hive after a period of dearth can cause a forager to produce shaking signals. Seeley et al. (in press) consequently proposed that the message of shaking signal is "prepare for greater activity."

To test this hypothesis, I conducted flight cage experiments to precisely manipulate foraging while recording shaking and waggle dancing activity inside the hive. These results point to a general shaking signal message "reallocate labor for different activities or activity levels," and support the message "prepare for greater foraging activity" in specific contexts.

Methods

Study site and bee colonies

I conducted these experiments from June to August in 1993, 1994, and 1995 at the Liddell Field Station in Ithaca, New York. During this period, I used four different colonies of *Apis mellifera ligustica*.

I housed two colonies (A and B) in three frame observation hives and two colonies (C and D) in two frame observation hives. All hives had only one opening through which bees could enter and exit and were constructed so that bees could enter and exit from only one side of the hive, thereby establishing an exit side (side with direct access to the exit) and a non-exit side (Fig. 1, side without
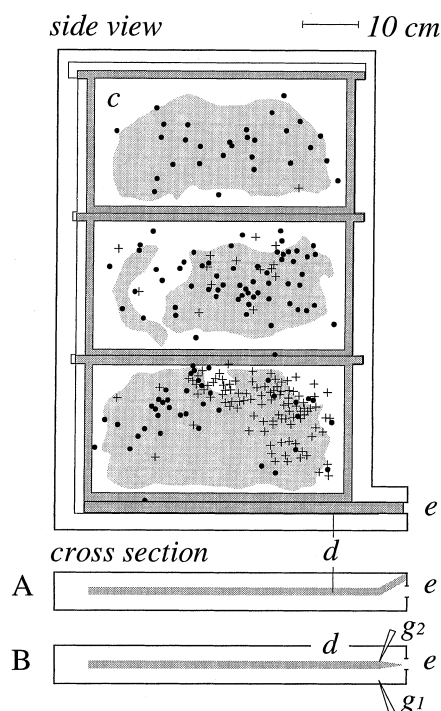

Fig. 1A,B The distribution of shaking signals (*black dots*) and waggle runs (*crosses*) for two 20-min censuses of colony A conducted between 1105 and 1205 hours on 18 June 1993. The brood areas are the irregular shapes shaded in gray inside the combs (*c*). The entrance (*e*) leads into the hive which is divided by a wooden strip (*d*) shown here in the side view and cross section for **A** colony A and **B** colony B. In colony A, a slanted piece of wood blocks off access to one side. In colony B, two gates (*g1* or *g2*) can be closed to direct traffic to only one side of the combs. In this illustration, *g1* is open and *g2* is closed. Thus the waggle dance floor is on the side of *g1*. Note that the only way for a bee to cross over from one side to the other is to pass through the spaces between the combs and the hive walls on the top and on the left, opposite the entrance (as shown here). Bees could not pass through the combs or through gaps between the combs

direct access to the exit, technique discussed in Seeley 1989). Because I used observation hives with two sides to model the more complex structure of a natural hive, the (1) exit side and (2) non-exit side respectively model regions that are (1) near the exit and (2) away from the exit in a natural hive.

In colonies A, C, and D, the accessible side was fixed. In colony B, I could insert or withdraw a metal gate to block off any side to entering and exiting bees to change the position of the dance floor (Fig. 1). This enabled me to separate the effects of dance floor and hive side on shaking signal production.

Each colony contained 4 000–13 000 bees as censused by placing a 13 × 9 grid drawn on a piece of glass over the observation glass on each side of the hive; counting the number of bees within ten randomly chosen 5.3 × 5.7 cm rectangles on each side at 0800 hours; and then multiplying by the total number of rectangles. I chose grid squares based upon coordinates randomly generated from a Casio fx-5000F calculator.

Individual shaker behavior

For observations of individual shakers, I replaced the glass over the bottom and middle comb on the exit side of colony A with a mesh screen made of black tulle fabric through which I could paint the thorax of a selected shaker. This allowed me to track her as she moved onto different sides of the hive. To choose a new shaker, I scanned from the top comb on the exit side as I would for a

population census and chose the first shaker that I observed. If the shaker produced at least two shaking signals in the first 5 min, I followed her as long as possible on both sides of the hive. I consequently gathered data only on the behavior of bees *strongly motivated to produce shaking signals*. Many bees produced only one shaking signal or shook other bees only occasionally. Their contribution to colony-wide patterns of shaking activity is counted through censusing, but is not reflected in the analysis of individual shaker behavior.

I collected behavioral data on 28 shaking bees. I used a data-logger program running on a Macintosh PowerBook 170 to record which side of the hive she was on; the time spent monitoring her; and the number of times she performed 11 behaviors (1) shook another bee, (2) shook the comb, (3) performed waggle dances, (4) groomed herself, (5) inserted only her head into a cell (cell inspection), (6) placed her entire body into a cell (cell entry), (7) paused and did nothing for more than 3 sec, (8) followed a waggle dancer, (9) received food from another bee, (10) gave food to another bee, and (11) antennated another bee without receiving or giving food.

Schneider et al. (1986a) observed more shaking signals on the exit side of the hive than on the non-exit side. I consequently used one-tailed Wilcoxon paired-sample signed-ranks tests (test statistic = W) to determine if bees producing shaking signals spent more time and produced more signals on the exit side than on the non-exit side. I used two-tailed tests to determine if bees producing shaking signals (1) entered cells, (2) groomed, (3) received food, (4) unloaded food, or (5) followed waggle dancers at different rates on the two sides of the hive.

Spatial and temporal distribution of shaking signals in the hive

With colony A, I measured the level of shaking activity over space and time. At the beginning of each day, I randomly chose which side to begin scanning. With a second grid composed of 15 identical 23×10.2 cm rectangles (five over each comb), I scanned down each rectangle in 4 s (keeping time with a Seiko DM-20 digital metronome set at 0.25 Hz) and counted the number of shaking signals and waggle dances. I began at the topmost comb, moving from right to left across each comb and then down to the far right of the next comb. Each scan lasted 1 min, and I made five complete scans on one side before switching to scan the opposite side. I consequently alternated sides every 5 min. I then calculated the number of shaking signals and waggle dance dances that occurred on the (1) top, (2) middle, and (3) bottom comb on each side of the hive.

For some censuses, I obtained a visual record of shaking signal and waggle dance locations by marking the location of each shaking signal and waggle dance on the glass, tracing the brood comb area, removing the glass, and photographing it against an illuminated slide table. In these cases, I counted shaking signals and waggle dances directly from the glass.

I scanned both sides of the observation hive for 20 min (four 5-min scans on each side) at various times over 13 days from 23 June to 9 July 1993 (scans were not made on all days). My earliest scans began at 0430 hours and my latest scans ended at 2250 hours. When possible, I made scans under natural light since the colony was facing a window. However, to avoid the possibility of light influencing bee behavior at night or before dawn, I illuminated the colony with a gallium-aluminum-arsenide light-emitting diode array generating negligible amounts of heat with a peak light intensity around 667 nm (analyzed on a Aries FF250 Spectrograph calibrated with a HeNe laser producing 632.8 nm) and filtered to only pass wavelengths longer than 640 nm (Roscolux medium red filter #27). These wavelengths are above the range of light sensitivity of honey bee photoreceptors (Autrum and von Zwehl 1964), but within the range of human vision.

To analyze the overall distribution of waggle dance and shaking signal activity, I pooled data collected throughout the day. I first compared the census results from all combs on the exit side with the census results from all combs on the non-exit side. I then separately

compared the census results for each comb (top, middle, and bottom) on each side of the hive, testing for differences with two-tailed Mann-Whitney tests (test statistic = M). To simultaneously examine temporal and spatial effects, I performed Kruskal-Wallis tests (non-parametric alternative to ANOVA, test statistic = K) with the following variables: (1) date, (2) time, (3) hive side, (4) comb, (5) shaking activity, and (6) waggle dancing level. Where appropriate, I calculated partial correlation coefficients to eliminate conflating correlations between predictor variables (Sokal and Rohlf 1981).

Switching the location of the dance floor/hive exit

I switched the dance floor/exit area from one side of hive B to the other by pushing in a metal gate to block access to one side and pulling out a metal gate on the opposite side (Fig. 1). After each switch, I immediately began to census the number of shaking signals and waggle dances using the procedure previously described. Because temporal bias could have accrued from repeatedly censusing the same side at the beginning of each trial or at the beginning of each day, I made multiple dance floor/exit area switches each day, and I alternated the first side that I opened on each day. On the first day, I began with side 1 open and side 2 closed. After a few switches, I left the hive overnight with side 2 open and side 1 closed. On the next day, I began with side 2 open and side 1 closed. Within each day, I alternated the side (exit side or non-exit side) on which I began censusing; censused that side for 10 min; switched to the opposite side for another 10 min census; then switched back to the initial side to repeat this procedure. Thus I censused each side for 20 min in alternating 10 min blocks.

I tested for differences between the observed and expected shaking signal and waggle dance distributions using two-tailed *G*-tests (observed distribution = distribution after switching; expected distribution = distribution prior to switching).

Changes in the behavior of bees receiving the shaking signal

In 1995, I compared the behaviors of bees before and after they received a shaking signal to test the following hypotheses: (H1) bees move faster after being shaken, (H2) bees perform various behaviors at higher rates after being shaken, (H3) bees spend more time performing various behaviors after being shaken, and (H4) bees are more likely to move to different areas of the hive after being shaken. I used colony C (4 000–5 000 bees) for these comparisons and examined the frequency with which shakers performed the 11 behaviors listed above.

I placed a glass sheet marked with a 1×1 cm grid over the exit side of the hive, selected a bee on the *bottom comb* from a grid square specified by randomly generated coordinates, and followed this bee until she was shaken. I only selected bees from the bottom comb because the density of bees and the level of shaking activity were much lower on the top comb. To avoid behavioral disruption, I did not mark the selected bee. I entered the time that she crossed each grid square into the datalogger. Bees were abandoned if they were not shaken within 30 min. I recorded the coordinates of (1) her initial position, (2) the position where she was shaken, and (3) her final position. On average, I followed bees for approximately equal times intervals before and after they were shaken. I selected and followed bees between 0608 and 1642 hours. Most bees were followed before noon (32 bees before noon and 6 after noon).

To analyze a bee's rate of movement before and after being shaken, I calculated her average speed (number of squares crossed by her thorax per min) for the time she spent in the hive *after* being shaken and compared this with her average speed in an equal duration of time *before* she was shaken.

I also calculated her average direction of movement *before* she received a shaking signal by determining the vector which pointed from her initial position to the position at which she was shaken. I calculated her average direction of movement *after* she was shaken by determining the vector that pointed from the position at

which she was shaken to her final position. To determine if bees were more likely to head for the hive exit after being shaken than before being shaken, I calculated error angles based upon the differences between their ideal exit bearings and their pre- and post-shaking bearings. The ideal exit bearing was the bee's bearing if she headed directly to the exit along the shortest possible path and is defined as 0°. I calculated each bee's ideal exit bearings from her initial position (ideal pre-shaking exit bearing) and from the position at which she was shaken (ideal post-shaking exit bearing). I consequently generated two error angle distributions: pre-shaking errors and post-shaking errors. I also generated a third distribution for bees who were monitored but did not receive any shaking signals. I calculated error angles for these bees using the procedure described for pre-shaking error angles.

I calculated the magnitude and direction of the mean vector (circular statistic) for each error angle distribution under the assumption of unimodality. I then used the Raleigh test to determine if these distributions were significantly non-uniform (Greenwood and Durand 1955). If a distribution was significantly non-uniform ($P < 0.050$) and appeared bipolar, I applied the double angle transformation described by Batschelet (1965). I accepted bipolarity if the transformation resulted in a larger mean vector magnitude (larger r), reduced the angular deviation, and resulted in a more significant departure from a uniform circular distribution (Raleigh test). I then applied the procedures of Batschelet (1965) to calculate the magnitude and direction of the mean vector for a bipolar distribution. For each unimodal distribution, I tested to see if the direction of the mean error vector equaled zero with Stephens (1962a) polar vector test. Finally, I used a modified F-test (described by Stephens 1962b) to determine whether the mean vector magnitudes and the variances of the two error distributions were significantly different. Although parametric, this test is robust even when the assumption of circular normality is violated (Stephens 1962b).

Manipulating food availability

In the summer of 1994, I studied the effects of pollen and sucrose solution foraging on temporal patterns of shaking with colony D (5 000–6 000 bees housed in a two-frame observation hive). The exit from this hive led to a 3.7 m long × 3.7 m wide × 1.8 m high screened tent.

I censused the number of shaking signals, waggle dances, and tremble dances on each side with a modification of the previously described scanning procedure. Since colony D contained only two frames, I censused each 23 × 10.2 cm rectangle in 6 s to make a complete scan of each side in 1 min. I alternated between sides each 5 min and scanned both sides for a total of 10 min. Because I was interested in the total level of shaking, waggle dancing, and tremble dancing, I pooled counts from both sides. I censused at 1-h intervals, except when I observed a surge of activity. In these cases, I censused each 15 min. I usually collected data between 0600 and 2000 hours, although I did not census the colony over this entire time range on all days.

The colony could only forage inside the flight cage. Sucrose solution (2.5 M sucrose scented with 100 μl commercial anise extract per liter of sugar solution) was presented in a 10-cm-diameter grooved feeder capable of simultaneously feeding more than 80 bees (feeder design discussed in von Frisch 1967) on a block 0.3 m above the ground and 1 m from the hive entrance. I obtained pollen pellets from traps placed on other hive entrances; ground these into a powder, and stored the powdered pollen in a freezer until it was needed. Pollen was presented in a 15-cm-diameter dish on the same block used to present sucrose solution. I never presented pollen and sucrose solution simultaneously. When I presented food, I allowed the bees to feed *ad libitum*. To determine the amount of food consumed, I weighed the sucrose solution and pollen feeders before and after feeding the bees.

After installing the flight cage, I did not feed the bees for four days. On the 5th day, I introduced a sucrose solution feeder for

105 min. In the last 15 min of this period, large numbers of foragers fed at the feeder. Thereafter, foragers immediately began to feed as soon as the feeder was set out. I obtained the same results after an initial introduction of pollen.

I separately exposed the foragers to sucrose solution and pollen for periods of time ranging from 5 to 255 min. I define the maximum value of the food-elicited shaking signal peak as the greatest number of shaking signals counted on both sides in one 10-min interval while food was present.

One *peak* of shaking occurred in the early morning and one in the late afternoon when no food was in the flight cage. I defined these peaks as an increase in shaking resulting in a 10-min count that was at least 1 SD above the average level of shaking before food was present and 1 h after food had been removed.

All averages are reported as the mean ± 1 SD.

Results

The behavior of highly motivated individual shakers

Highly motivated shakers remained in the hive producing shaking signals for an average of 31.9 ± 43.9 min (performance time = time from the first to the last observed shaking signal, $n = 42$, range: 1.3–256.8 min). Because I randomly selected these shakers, I did not necessarily observe them from the time that they initially began to shake. Thus the true average performance time may be 63.8 min. (Assuming that I selected shakers at random times throughout their performances, then, on average, I should have selected them halfway through their complete performances.)

The majority of these highly motivated shakers were foragers. Of the 42 shakers that I followed, 14 carried pollen in their corbiculae, 14 spent time waggle dancing, 19 spent time following a waggle dance, and 1 unloaded food to another bee and then left the hive. Some shakers performed more than one of these behaviors. Thus 83.3% of the shakers were foragers because they performed forager-specific behaviors. All of the remaining shakers (16.7%) left the hive but did not perform any forager-specific behaviors.

Figure 2 shows the average distribution of behaviors performed by 28 shakers. A detailed breakdown of these behaviors and their rates for each side of the hive is given in Table 1. The 5 most frequent behaviors were: shaking another bee (80.6 ± 13.2%); shaking the comb (7.8 ± 5.7%); grooming (3.1 ± 2.5%); receiving food from another bee (2.1 ± 6.7%); and waggle dancing (2.0 ± 7.4%). Thus bees delivered only a small proportion of shaking signals directly to the comb.

On average, highly motivated shakers spent 60% more time on the exit side than on the non-exit side of the hive ($P = 0.007$, Table 1). These shakers produced slightly more signals on the exit side (57%, $P = 0.073$). However, shakers produced shaking signals at a significantly higher average rate (21% higher, $P = 0.046$) on the non-exit side than on the exit side. The same trends appear when one separately considers shaking signals delivered to bees and to the comb, although none of these differences are significant ($P > 0.055$). Thus
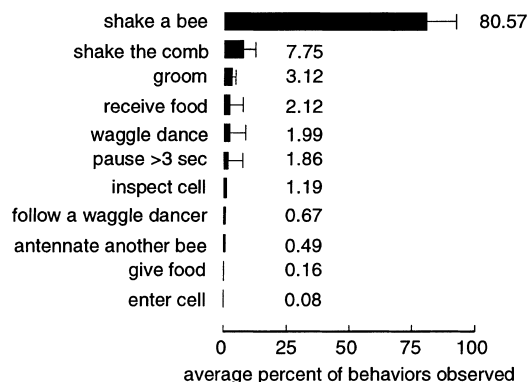
shake a bee — 80.57
shake the comb — 7.75
groom — 3.12
receive food — 2.12
waggle dance — 1.99
pause >3 sec — 1.86
inspect cell — 1.19
follow a waggle dancer — 0.67
antennate another bee — 0.49
give food — 0.16
enter cell — 0.08

average percent of behaviors observed

Fig. 2 The average probability distribution of 11 different behaviors ordered from most to least frequent. The average for each behavior is given to the *right* of each *horizontal column*. The *error bars* represent 1 SD. These data were collected from 28 bees producing shaking signals that were followed for an average of 56.0 ± 39.1 min each (total observation time = 6.03 h, total number of behaviors observed = 21 693)

highly motivated shakers spent more time on the exit side but produced shaking signals at higher rates on the non-exit side. The greater time they spent on the exit side compensated for this rate difference to result in more *total signals* being produced on the exit side.

The rates of other behaviors also differed between the two sides (Table 1). On the exit side, shakers had a significantly higher grooming rate (77% higher, $P = 0.016$). On the non-exit side, shakers appeared to inspect cells and enter cells more frequently, although these differences were not significant ($P \geq 0.067$). Thus bees who repeatedly produced shaking signals spent long periods of time walking around both sides the hive. They had a higher grooming rate, spent more time, and produced more signals on the *exit side*; but produced signals at a higher rate on the *non-exit side*.

Overall spatial patterns of waggle dancing and shaking

The following results give the overall spatial distributions of shaking signals produced by bees over all time periods *at all levels of signaling motivation*. Many bees were not included in the previous analysis because they produced only one shaking signal or produced them at a very low rate (<2 shaking signals/5 min). However, their contributions are considered in the following spatial analysis.

Foragers performed significantly more waggle dances (90.4%) on the exit side than on the non-exit side ($M_{30,30} = 1210$, $P < 0.001$). In each 20-min census, I counted 50.7 ± 40.7 waggle dances on the exit side and 5.40 ± 6.03 waggle dances on the non-exit side.

The "waggle dance floor," the area in the hive with the highest concentration of waggle dances, was located on the bottom comb on the exit side. On the exit side, foragers performed 91.3% of waggle dances on the bottom comb, 8.0% on the middle comb, and 0.7% on

Table 1 How does the behaviour of shakers differ between the two sides of the hive? The exit side is denoted by e and the non-exit side by ne. I also give the ratio of the exit side to non-exit side for each variable as well as the maximum value, the number of behaviours observed and the total time spent in observation of bees to collect the data on each variable. Ha gives the alternative hypotheses (Ho: $e = ne$) for each test, followed by the test statistic, P value, and sample size

Variable	Dance floor		Max	Exit side/ non-exit side	No. behaviours	Total time observed	Ha	Wilcoxon paired-sample signed-ranks		Total no. trials
	Exit side (e)	Non-exit side (ne)				(h)		W	P	
Time on each side (min)	34.50 ± 26.37	21.54 ± 21.15	125.45	1.602		26.15	e > ne	2.4821	0.007	28
All shaking signals	200.82 ± 175.81	152.71 ± 160.13	808	1.315	10006	26.09	e > ne	1.4574	0.073	28
Shaking on bees	185.30 ± 165.20	140.43 ± 145.39	720	1.320	9119	26.09	e > ne	1.4574	0.073	28
Shaking on comb	17.08 ± 17.53	13.40 ± 17.36	88	1.275	780	26.09	e > ne	0.7268	0.234	25
All shaking signals/min	6.37 ± 3.25	8.01 ± 2.97	14.57	0.795	10006	26.09	e ≠ ne	-1.9941	0.046	27
Shaking on bees/min	5.82 ± 3.02	7.28 ± 2.61	13.86	0.800	9119	26.09	e ≠ ne	-1.9220	0.055	27
Shaking on comb/min	$0.55 \pm .56$	$0.73 \pm .95$	4.29	0.748	780	26.09	e ≠ ne	-1.0811	0.140	27
Groomings/min	$0.22 \pm .14$	$0.13 \pm .12$	0.53	1.769	318	26.00	e ≠ ne	2.4001	0.016	26
Cell inspections/min	$0.10 \pm .09$	$0.21 \pm .27$	1.18	0.465	174	23.26	e ≠ ne	-1.8343	0.067	22
Cell entries/min	$0.01 \pm .01$	$0.04 \pm .05$	0.13	0.221	10	23.26	e ≠ ne	-1.0142	0.155	7
Antennations/min	$0.001 \pm .002$	$0.002 \pm .003$	0.01	0.500	49	23.26	e ≠ ne	-0.9103	0.810	14

the top comb (Fig. 1). I observed the same pattern on the non-exit side. Here, foragers performed 74.7% of all waggle dances on the bottom, 17.3% on the middle, and 8.0% on the top comb.

Shakers performed more shaking signals on the exit side (87.5%) than on the non-exit side ($M_{34,34} = 1697$, $P < 0.001$). On average, in each 20-min census, I counted 35.8 ± 30.4 shaking signals on the exit side and 5.1 ± 7.7 shaking signals on the non-exit side. On the exit side, bees produced 30.4% of shaking signals on the bottom comb, 50.3% on the middle comb, and 19.3% on the top comb (Fig. 1). On the non-exit side, bees produced 39.2% of shaking signals on the bottom comb, 33.7% on the middle comb, and 27.1% on the top comb.

Temporal and spatial changes in the pattern of waggle dancing and shaking

I obtained the following results by considering the number of shaking signals produced in five time intervals spaced throughout the day (Fig 3).

Waggle dancing

Foragers produced the most waggle dances in the time interval 1150–1530 hours; and, within this interval, they produced the most dances on the bottom comb (Fig. 3). More waggle dances occurred on the exit side than on

the non-exit side ($K_{1,186} = 7.97$, $P = 0.005$); and, on the exit side, the bottom comb received the most dances and the top comb the least dances ($K_{1,93} = 40.69$, $P < 0.0001$, Fig. 3). Thus the waggle dance floor was located on the bottom comb of the exit side. On the non-exit side, most waggle dances also occurred on the bottom comb ($K_{1,93} = 18.07$, $P < 0.0001$).
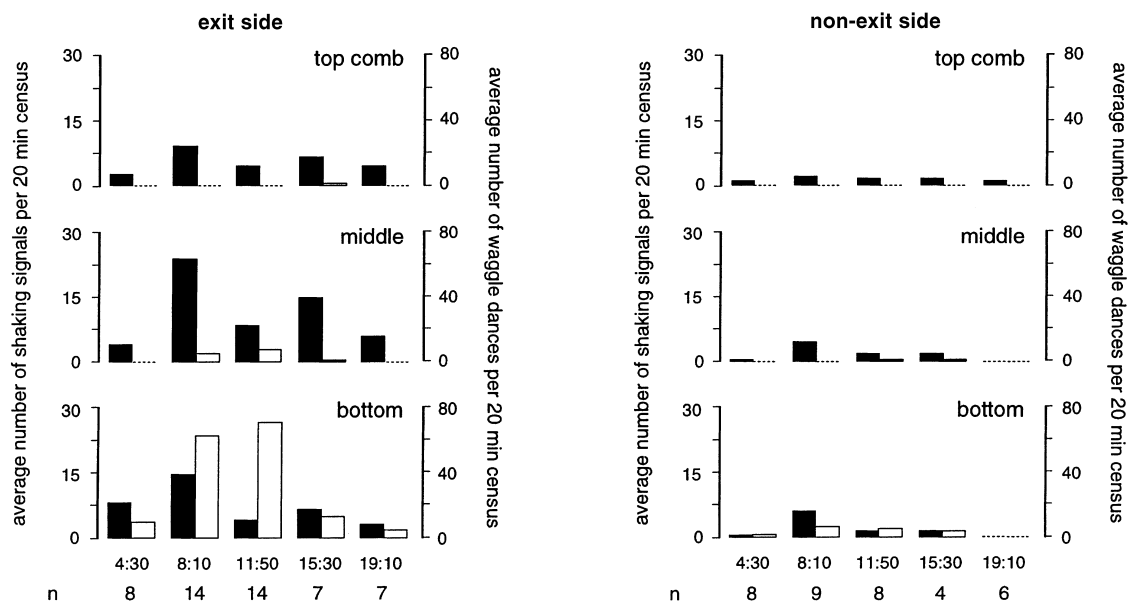
Shaking

Between 0430 and 0810 hours, bees produced the most shaking signals on the waggle dance floor and, in subsequent time intervals, produced the most signals in the region immediately above the dance floor (middle comb, exit side, Fig. 3). Bees produced the most shaking signals in the time interval 0810–1150 hours (just before the average daily peak in waggle dancing). During this time interval, bees produced the most exit-side signals on the middle comb and the most non-exit-side signals on the bottom comb. These two regions had the greatest direct access to the dance floor.

The variable "time of day" accounts for a significant portion of the variability in shaking on both sides of the hive (Fig. 3; $K_{73,252} = 193.13$, $P < 0.0001$). The most shaking occurred between 0810 and 1910 hours. I found no significant relationship between "date" and the level of shaking on each side of the hive (partial correlation coefficient: $r = 0.0023$, $df = 249$, $P > 0.050$).

Overall, the greatest shaking activity on the exit side occurred on the middle comb and least on the top comb ($K_{2,150} = 10.39$, $P = 0.006$). On the non-exit side, almost all shaking occurred on the top comb ($K_{2,102} = 9.95$, $P = 0.007$).

Fig. 3 The spatial and temporal distribution of the average number of shaking signals and waggle runs on both sides of the hive. *Solid bars* represent shaking signals. *Open bars* represent waggle runs. *Dashed lines* in each time interval represent a census value of 0. Each time interval represents 3 h and 40 min. Only the start time of each interval is shown. The number of scans (*n*) made in each interval is also shown

Switching the location of the dance floor/hive exit

As the previous analysis demonstrates, most shaking signals occurred on the dance floor/exit side of the hive. This may have occurred (H0) because of an arbitrary preference of the shakers or (H1) because of a consistent association between shakers and the dance floor/exit side. To test these hypotheses, I used a modified observation hive housing colony B (Fig. 1). Within 20 min after I switched the exit side, I observed a majority of all shaking signals (67.9 ± 36.0%) and all waggle dances (76.5 ± 41.1%) on the *new* exit side. Within 40 min, I observed 74.9 ± 27.2% of shaking signals and 78.3 ± 38.1% of waggle dances on the new exit side. I conducted nine trials over 3 days, beginning each day at 1010, 1200, and 1125 hours respectively. In eight out of nine trials over 3 days, shakers delivered significantly
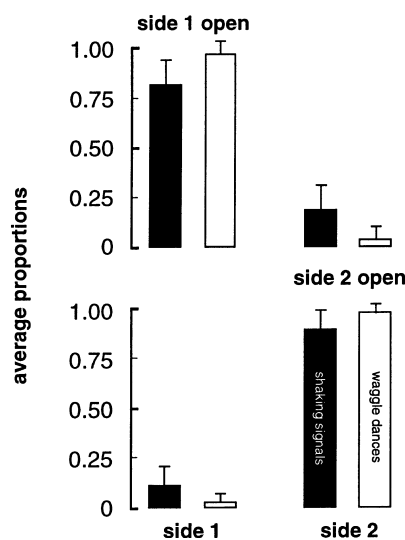
more shaking signals on the newly opened exit side of the hive (Fig. 4, $G_{individual} > 6.86$, 1 df, $P < 0.009$). Thus H1 is accepted with p $\ll$ 0.001 ($G_{pooled} = 272.5$, 8 df). The association between shaking and the dance floor/exit side is not arbitrary.

The behavior of bees before and after being shaken

Bees moved significantly faster after being shaken than before they were shaken (Wilcoxon signed-ranks test, $W = 3.22$, $n = 41$, $P < 0.001$). In 25 out of 41 cases, individual bees increased their speed after being shaken. On average, bees moved 3.04 ± 3.01 cm/min before being shaken and 5.32 ± 5.29 cm/min after being shaken. These speed changes were highly variable, but on average, bees increased their rate of movement by 75% after receiving a shaking signal. I measured speed in approximately equal time intervals before and after an individual bee was shaken. These time intervals ranged from 28 to 1841 s, averaging 333 ± 340 s.

On average, bees spent significantly less time inside cells after being shaken (64% less time, $P = 0.041$, Table 2). Bees increased their average rates of grooming (by 59%), receiving food (by 79%), unloading food (by 700%), and following waggle dances (by 358%) after being shaken, although none of these increases is significant ($P \geq 0.052$, Table 2). However, bees exhibited an overall increase (≥ 0.50 in all cases) in the rate of performing these behaviors after being shaken (Table 2). This overall rate increase is significant (one-sample, one-tailed T-test, $P = 0.031$).

The direction of a bee's movement before and after she is shaken

I obtained five major results from the analysis of movement directions:

1. The pre-shaking distribution of error angles is significantly different from a uniform circular distribution (Raleigh test based on unimodal data, $n = 38$, $z = 4.973$, $P < 0.010$) and has a mean vector bearing of

Fig. 4 Results of the dance floor/exit side switching experiment. The mean proportions of nine trials in which the exit side was repeatedly switched are shown. *Solid rectangles* represent the mean proportion of shaking signals and *open rectangles* represent the mean proportion of waggle dances occurring on the specified side. The *error bars* represent 1 SD

Table 2 Individuals rates of behavior and the time spent performing a behavior before and after being shaken. The proportion of individuals who increased their level of a particular behavior after receiving a shaking signal is also shown. For example, out of 26 individuals who made cell entries, 53.8% (14 individuals) had a higher rate of cell entry after receiving a shaking signal than before

Rate (per min) of	Before	After	Wilcoxon signed-ranks 1-tailed tests			Proportion where after > before
			W	P	n	
Entering cells	0.463 ± 0.783	0.450 ± 0.693	686	0.482	26	0.538
Grooming	0.147 ± 0.257	0.234 ± 0.319	554	0.052	25	0.560
Receiving food	0.080 ± 0.228	0.143 ± 0.357	205.5	0.123	15	0.600
Unloading food	0.004 ± 0.026	0.032 ± 0.169	8	0.177	3	0.667
Following a waggle run	0.012 ± 0.059	0.055 ± 0.331	18	0.500	4	0.500
Average time spent (s)						
Inside cells	48.958 ± 74.612	17.842 ± 48.677	153.5	0.041	11	0.182
Grooming	6.471 ± 5.442	7.386 ± 5.096	75.5	0.200	14	0.429

$315 \pm 64°$ (Fig. 5). This mean bearing is significantly different from $0°$ error (Stephens polar vector test, $R_{\text{test}} = 13.747$, $P < 0.010$). (An error bearing of $0°$ indicates that the bee moved directly towards the hive exit.)

2. The post-shaking distribution of error angles is also significantly different from a uniform circular distribution (Raleigh test based on unimodal data, $n = 38$, $z = 0.3903$, $P < 0.050$). However, inspection of this distribution (Fig. 5) reveals a potential bimodality. The test for bimodality (Raleigh test on double angle transformed data) reveals a far more significant difference between the transformed data and a uniform distribution ($n = 38$, $z = 5.8937$, $P < 0.005$). Thus the bimodal

distribution is a much better fit (mean vector bearings are $177 \pm 31°$ and $357 \pm 31°$).

3. The pre-shaking distribution of error angles is not significantly bimodal ($z = 1.008$, $P < 0.10$). Thus the distribution of error angles shifted from unimodal to bimodal after bees received a shaking signal. Additionally, the pre-signal and post-signal distributions have significantly different mean vector bearings (polar vector test with transformed data, $x = 11.121$, $P < 0.010$).

I followed bees for 480 ± 401 s (max = 1841 s, min = 28 s) before they were shaken and 545 ± 480 s (max = 2024 s, min = 79 s) after they were shaken. The distribution of "after" time intervals is not significantly different from the distribution of "before" time intervals ($W = 1342$, $n = 38$, $P = 0.63$). Per individual, the average after/before time ratio is 1.24 ± 1.08 (max = 4.06, min = 0.16). Thus both error angle distributions are derived from data taken at approximately equal average time intervals.

Bearings of non-shaken bees

Since the changes in the error angle distributions may have resulted from the passage of time and not from shaking signals, I also calculated the error angle distribution for bees who never received signals and who I followed *for time periods equivalent to the time spent following shaken bees.*

On average, I followed non-shaken bees for 875 ± 614 s (max = 2418 s, min = 197 s, $n = 29$). I followed shaken bees for 1030 ± 685 s, (max = 2992 s, min = 107 s, $n = 38$). These two time interval distributions are not significantly different from each other ($M_{29,38} = 919.5$, $P = 0.318$). *Thus I followed all bees (shaken and non-shaken) for 963 ± 660 s.*

4. The distribution of error angles for non-shaken bees is significantly unimodal (see Fig. 5; Raleigh test, $n = 29$, $z = 20.417$, $P < 0.0001$). The non-shaken vector bearing is $13 \pm 33°$ and is not significantly different from $0°$ error ($F_{1,28} = 3.742$, $P = 0.063$). In fact, 17 non-shaken bees (59%) exited the hive while being monitored. By comparison, only 3 shaken bees (8%) exited the hive while being monitored for periods of time *equivalent to the time spent monitoring non-shaken bees.* Thus significantly more non-shaken bees than shaken bees exited during the 963 ± 660 s monitoring periods ($\chi^2 = 38.97$, 1 *df*, $P < 0.0001$).

5. The pre-shaking and post-shaking error angle distributions have mean bearings that are significantly different from $13°$, the mean non-shaken error bearing, (pre-shaking: $F_{1,37} = 47.145$, $P < 0.001$; double angle transformed post-shaking: $F_{1,37} = 9.859$, $P = 0.003$, Fig. 5). Before receiving a shaking signal, bees moved, on average, towards the hive exit (mean error bearing $= 315°$). After receiving a shaking signal, some bees moved directly away from the exit and some continued towards the exit (mean error bearings are $357°$ and

Path of a shaken bee

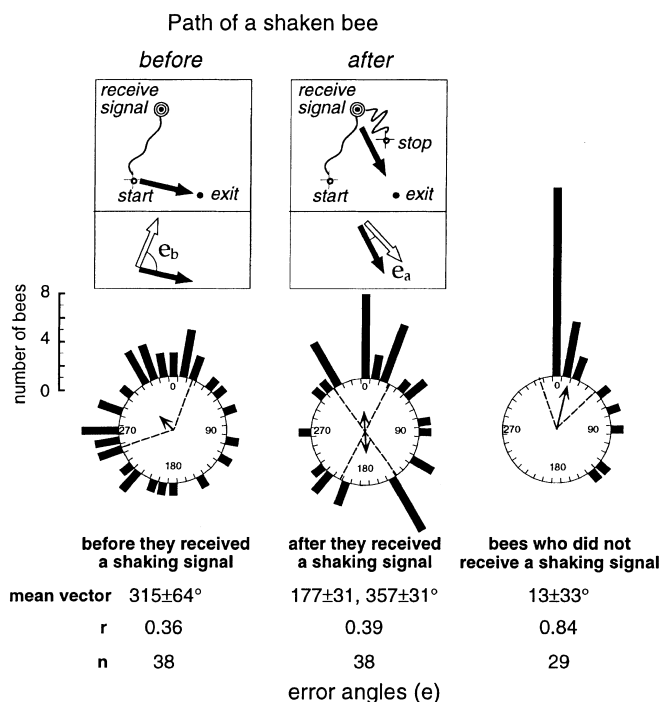|	before they received a shaking signal	after they received a shaking signal	bees who did not receive a shaking signal
mean vector	$315 \pm 64°$	$177 \pm 31°$, $357 \pm 31°$	$13 \pm 33°$
r	0.36	0.39	0.84
n	38	38	29

error angles (e)

Fig. 5 Error angle distributions for bees before and after they received a shaking signal and for bees who never received a shaking signal. The insets above the circular distributions of the *before* (e_b) and *after* (e_a) error angles depict how these error angles were measured. These diagrams show the path of a bee receiving a shaking signal mapped onto the hive dance floor (see Fig. 1). The path of the bee is shown along with the start point, signal reception point, and stop point. The location of the hive exit is also shown. The *open arrow* shows the bee's actual direction of movement and the *solid arrow* shows the ideal exit direction. In the example shown, $e_b = 85°$ and $e_a = 20°$. A mean error angle of $0°$ corresponds to a bee moving (on average) directly towards the hive exit. The direction and magnitude of the mean error vector is shown by the *arrow* in the center of each distribution. The magnitude of the mean error vector is drawn proportionally, such that $r = 1$ corresponds to the circle's radius. The direction of the mean error vector is reported ± 1 angular deviation (± 1 angular deviation is also shown as *dashed lines* on each circular distribution). These data are also given numerically below each histogram. The histogram of error angles for bees after they were shaken is bimodal (see Results), and I therefore show the mean error vectors and angular deviations originally obtained from the double angle transformation, then re-transformed back into a single angle (Batschelet 1965)

177°). In contrast, control bees who never received a shaking signal exhibited an extremely strong tendency (vector magnitude: $r = 0.84$) to move towards the exit (mean error bearing $= 13°$).

The relationships between food availability and waggle dancing, tremble dancing, and shaking

On average, waggle dancing, tremble dancing, and shaking signal levels in colony D peaked 22 ± 33, 26 ± 33, and 22 ± 17 min respectively after I presented sucrose solution or pollen inside the flight cage ($n = 22$). There are no significant differences between any of these response times ($M_{22,22} > 440.5$, $P > 0.183$). Bees did not tremble dance when foraging solely on pollen. Figure 6 shows typical response curves.

The height of the waggle dancing peak is positively correlated with the amount of sucrose consumed ($r^2 = 0.55, n = 20, F_{1,18} = 21.54, P < 0.001$) but not with the amount of pollen consumed ($r^2 = 0.05, n = 14, F_{1,12} = 0.58, P = 0.460$). The height of the tremble dancing peak is positively correlated with the amount of sucrose consumed ($r^2 = 0.29, n = 23, F_{1,21} = 8.65, P = 0.008$).

Although shaking peaked within 15 min after I placed sucrose solution or pollen in the flight cage (Fig. 6), the height of the shaking peak is not correlated with the amount of sucrose consumed ($r^2 = 0.12$, $n = 24$, $F_{1,22} = 2.94, P = 0.101$) or the amount of pollen consumed ($r^2 = 0.01, n = 23, F_{1,21} = 0.17, P = 0.685$). It is also not correlated with the duration of sucrose presentations ($r^2 = 0.03, n = 25, F_{1,23} = 0.65, P = 0.429$) or the duration of pollen presentations ($r^2 = 0.01, n = 23$, $F_{1,21} = 0.17, P = 0.685$).

However, time gaps between foraging influenced colony-wide levels of shaking signal production. I define "time gap" as the time between food presentations *when no food was present*. The height of the shaking peak was greatest for the first presentation of sucrose or pollen and generally declined for subsequent presentations during the same day (Fig. 6A). The shaking peak only recovered its former magnitude if the time gap between pollen presentations was sufficiently long (Fig. 6B, 7).

I define "recovery ratio" as the maximum height of a subsequent food-elicited shaking peak divided by the height of the first food-elicited shaking peak of the day. The recovery ratio is positively correlated with the time gap between pollen presentations (Fig. 7; $r^2 = 0.71$, $n = 15, F_{1,13} = 31.43$, $P < 0.001$). As the time gap increased, the recovery ratio increased. On any given day, the heights of secondary shaking peaks were between 84% and 116% of the first peak's height for consecutive presentations of pollen spaced 3.1 h apart (95% regression confidence interval). This was only true for pollen foraging. The recovery ratio is not correlated with the time gap between sucrose presentations (Fig. 7; $r^2 = 0.05, n = 16, F_{1,14} = 0.65, P = 0.43$). However, this may be due to the smaller range of time gaps used for sucrose presentations.
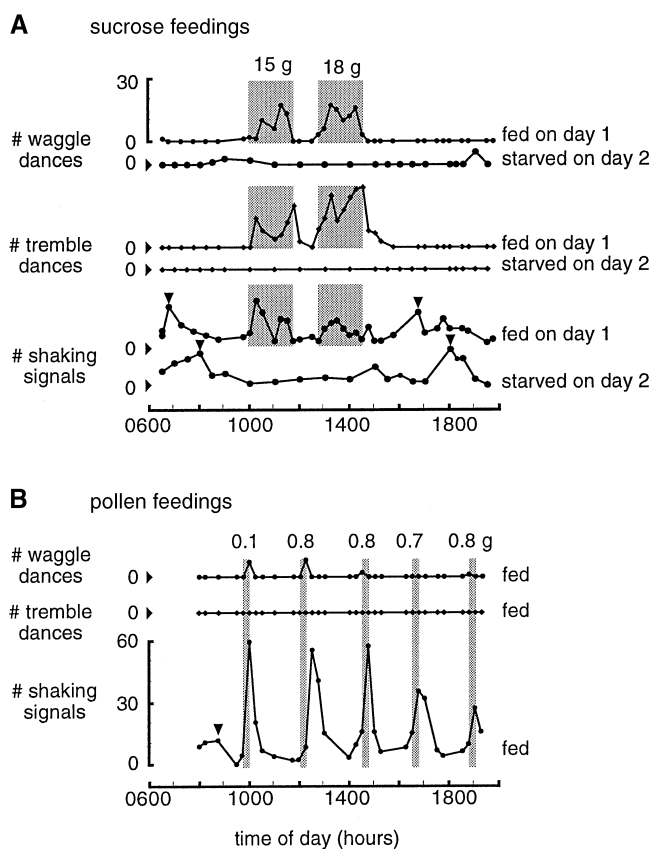

Fig. 6A,B Response of foragers to food inside the flight cage. **A** Shaking signals can be activated when foragers collect 2.5 M sucrose solution inside the flight cage. The census data for waggle dancing, tremble dancing, and shaking signals on 2 consecutive days are shown. The *vertical scale* is identical for all plots. The *zero point* is indicated to the *left* of all plots. *Inverted triangles* point to the major morning and afternoon shaking peaks that were not elicited by foraging. *Stippled bars* indicate when I provided food. The amount of sucrose solution (g) that bees collected *ad libitum* is shown above each feeding period. I presented food twice on 7 July 1994. Notice that the peak level of shaking activity occurs within 20 min of sucrose solution presentation and then decays as the sucrose solution continues to be present. On 8 July 1994, I provided no sucrose solution, yet I observed three waggle runs at 0900 hours and six waggle runs at 1900 hours. **B** Bees also begin to shake after foraging on pollen alone. I collected the pictured data on 28 July 1994. Bees were fed five times with sequential time gaps of 2.25, 2.0, 1.75, and 2.0 h. The amount of pollen (g) that bees collected *ad libitum* is shown above each feeding. On this day, I also observed a major morning shaking peak that was not elicited by foraging (indicated by the *inverted triangle*). I did not observe a major afternoon shaking peak in the absence of foraging. Possible explanations for the absence of such a peak are given in the Discussion

Shaking signals that are not triggered by pollen or sucrose solution foraging

I observed a major *morning* shaking peak before any food was present in the flight cage and a major *afternoon* shaking peak after food had been removed for at least 1 hr. These peaks were not triggered by the immediate availability of food. Figure 6A shows a typical pattern. On 7 July 1994, peak levels of shaking activity occurred at 0645 and at 1700. Both peaks occurred while no bees
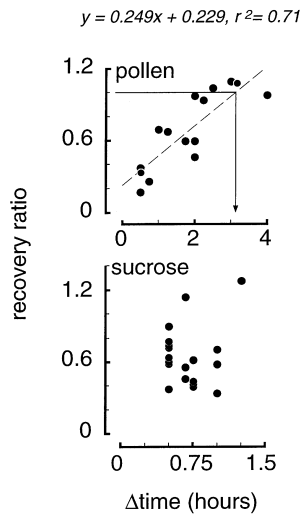
$y = 0.249x + 0.229, r^2 = 0.71$

pollen

sucrose

recovery ratio

Δtime (hours)

Fig. 7 Relationship between the recovery ratio and the time gap between pollen presentations. The recovery ratio is the maximum height of the subsequent shaking peak divided by the maximum height of the prior shaking peak. The *dashed regression line* and the *regression equation* for the pollen plot are shown. The *solid vertical line* marks the critical time interval which yields a recovery ratio of unity (as calculated from the regression equation). There is no significant correlation between the recovery ratio and the time gap between sucrose presentations

$y = 0.641x + 5.137, r^2 = 0.67$

afternoon peak (# shaking signals)

morning peak (# shaking signals)

A

$y = -0.353x + 20.026, r^2 = 0.77$

time of afternoon peak

time of morning peak

B

Fig. 8A,B Relationship of morning and afternoon shaking peaks that occur in the absence of foraging. **A** The height of the major afternoon shaking peak is positively correlated with the height of the major morning shaking peak. The *dashed regression line* and *regression equation* are shown. **B** The time at which the major morning shaking peak occurs is negatively correlated with the time the major afternoon shaking peak occurs. The *dashed regression line* and the corresponding *regression equation* are shown

were foraging. On the next day, I starved the colony, and continued to observe an early morning shaking peak (0800 hours) and an afternoon peak (1800 hours). On this day, both peaks were followed by a few waggle dances, although a careful inspection of the flight cage revealed no possible food sources or leaks through which foragers could escape from the flight cage. I observed a morning and an afternoon shaking peak on all 12 days that I monitored the colony. The earliest morning peak occurred at 0700 hours and the latest afternoon peak occurred at 1800 hours. I observed a few waggle dances in the absence of food on only 5 of the 12 days (6, 7, 8, 9, and 11 July).

The morning and afternoon shaking peaks are correlated in magnitude and time. The height of the afternoon peak is positively correlated with the height of the morning peak (Fig. 8A; $r^2 = 0.66, n = 12, F_{1,10} = 19.35, P = 0.001$). As the height of the morning peak increased, the height of the afternoon peak *on the same day* increased. There is no correlation between the height of the major afternoon peak *on the previous day* and the height of the morning peak *on the subsequent day* ($r^2 = 0.02, n = 7, F_{1,5} = 0.10, P = 0.77$).

However, the time of the afternoon peak is negatively correlated with the time of the morning peak (Fig. 8B; $r^2 = 0.59, n = 12, F_{1,10} = 14.15, P = 0.004$). Thus as the major morning peak occurred later in the day, the major afternoon peak occurred earlier in the day; and the two peaks drew closer together in time.

Over the 22 days that colony D was confined to foraging in a flight cage, the onset of the morning shaking peak came progressively later and the onset of
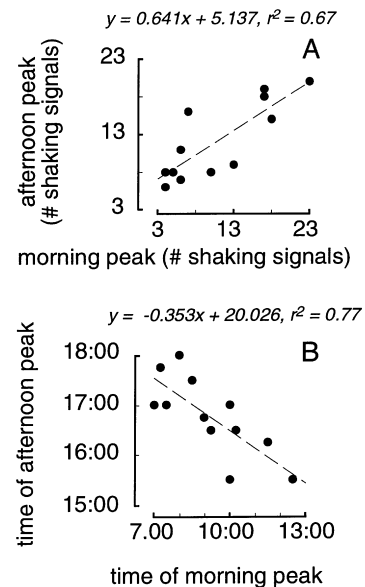
the afternoon shaking peak came progressively earlier. A significant positive correlation exists between date and morning peak time ($r^2 = 0.52, n = 12, F_{1,10} = 10.95, P = 0.008$) and a significant negative correlation exists between date and afternoon peak time ($r^2 = 0.42, n = 12, F_{1,10} = 7.27, P = 0.022$).

Discussion

Taken together, these results support the following general message for the shaking signal: "reallocate labor for different activities or activity levels,"and support the message "prepare for greater foraging activity" in specific contexts. I will summarize the evidence for these messages by readdressing the 5 questions posed at the beginning of this paper.

I. Who shakes? Most, if not all, of the bees producing shaking signals were foragers. All of the shakers that I monitored eventually left the hive and 83.3% performed behaviors exclusive to foragers. This is consistent with the message "prepare for greater foraging-related activity" since only foragers possessed direct information about the status of external food sources.

II. When do they shake? Within each day, levels of shaking changed significantly over time in a non-linear fashion (Figs. 3, 6). Bees produced the most shaking signals in the early morning (0810–1150 hours) just prior to the peak in waggle dancing activity (1150–1530

hours). This temporal order suggests that shaking signals communicate information related to foraging. Bees also performed shaking signals in the absence of foraging (Fig. 6, 8B). These spontaneous peaks may be linked to foraging experienced on previous days.

III. Where do they shake? Shakers traveled and produced signals on both sides of the hive, but spent the most time on the side directly accessible to the exit. This focus of shaking activity on the exit/dance floor side also suggests a close link between shaking and foraging related activities.

The region in which bees produced the most shaking signals changed over time. In the early morning (0430–0810 hours), more shaking occurred on the dance floor at a time when relatively few bees were waggle dancing. Later in the day (0810–1530 hours), the level of waggle dancing increased and bees produced the most shaking signals on the comb above the dance floor (Fig. 3). Thus one apparent function of the shaking signal is to communicate the presence of food outside the hive after a period of foraging inactivity. When not foraging, foragers tend to congregate on the quiet dance floor (von Frisch 1967). This area is consequently the natural focus of bees trying to activate colony foraging. Thus as more foragers returned to colony A and began to fill the dance floor with waggle dances, the number of inactive foragers on the dance floor shrank and the locus of shaking moved up to the middle comb.

IV. How do receivers respond to shaking? On average (and with considerable individual variation), bees moved 75% faster after receiving a shaking signal. These speed increases were measured in the morning and early afternoon and are thus consistent with the specific message "prepare for greater foraging-related activity" since the sudden availability of food after a period of dearth requires the rapid mobilization of foragers and bees engaged in foraging-related activities.

After receiving a shaking signal, bees spent significantly less time inside cells (on average 64% less time), and displayed higher rates of grooming, receiving food, unloading food, and following waggle dances. On average, a greater proportion of bees (57%) exhibited higher rates of these behaviors after receiving a shaking signal ($P = 0.031$). Although these studies did not separate foraging-age from non-foraging-age receivers and thus examined shaking signal effects on bees of random ages, they confirm the findings of Schneider (1987, 1990) that bees increase their rates of performing different behaviors after receiving shaking signals.

The analysis of bee movement yields a novel result. Before receiving a shaking signal, bees selected from the dance floor tended to move towards the hive exit. After receiving a shaking signal, some bees moved *towards* the hive exit and some moved *away* from the hive exit. By comparison, *non-shaken* bees *followed for comparable periods of time* exhibited a strong tendency to move towards the hive exit (Fig. 5).

V. What conditions trigger shaking? The level of shaking rose within 15 min after I placed pollen or su-crose solution in the flight cage (Fig. 6). The magnitudes of shaking peaks triggered by pollen foraging were restored to their maximum level only when I removed the pollen for at least 3 h. Thus the amount of shaking depended upon a critical period of food dearth (Fig. 7). The largest peak of food-elicited shaking signals is therefore typically observed in the morning (Allen 1959b; Schneider et al. 1986a; and Schneider 1986, 1989) because night provides a natural, extended period of dearth for *Apis mellifera* foragers.

The initially high level of shaking decayed with the continued presentation of a food source, although waggle dancing increased (Fig. 6). The level of signaling also appeared to correspond inversely with the colony's foraging readiness: signaling was greatest in the early morning when foraging-related activity inside the hive was lowest (Fig. 3). Thus foraging-triggered shaking levels may have been initially high to prepare the colony for greater foraging activity. After reaching an optimal level of activation, foragers began to produce more waggle dances. In a natural environment with virtually no natural food sources, Seeley et al. (in press) observed individuals switching from shaking to waggle dancing when they successfully foraged after a long period of dearth.

Perhaps most interesting are the morning and afternoon peaks in shaking that occurred in the absence of any foraging. Schneider et al. (1986a) proposed that the morning peak represents preparation for foraging based upon past experience, and that shaking signals consequently regulate foraging on two time scales: (1) as an immediate response to food availability, and (2) as a long-term response reflecting the availability of food on previous days. Since flowers will predictably produce nectar or pollen at certain times of the day (von Frisch 1967), the experience of previous days can be used to predict the time at which nectar or pollen will be available on subsequent days (if weather conditions are not too variable) and thus shaking signals would give the colony a competitive foraging advantage.

But what of the afternoon shaking peak? An afternoon *peak* in shaking levels *in the absence of foraging* has never been previously documented. Allen (1959b) and Schneider et al. (1986a) observed shaking in the late afternoon when no bees were flying outside the hive. However in both cases, it was possible to dismiss this shaking activity as a residual or delayed extinction of shaking initiated by foraging earlier in the day. This is particularly plausible since it is now clear that many shakers can remain inside the hive performing shaking signals for more than 30 min. However, residual shaking cannot explain the spontaneous shaking *peak* that occurred in the late afternoon after all foraging had ceased. This phenomenon does not fit within the message, "prepare for greater foraging activity," and I consequently propose a broader message, "reallocate labor for different activities or activity levels."

Two interesting clues suggest a more specific function for these spontaneous afternoon shaking signals.

1. The time of the afternoon shaking peak is negatively correlated with the time of the morning shaking peak (Fig. 8B). Thus as the morning peak occurred later in the day, the afternoon peak occurred earlier in the day. During these experiments, I provided food between 1139 and 1556 (averages of times at which food was first provided and last available on each day). On the first day of data collection (7 July 1994) the spontaneous morning peak occurred at 0645 hours and the spontaneous afternoon peak occurred at 1700 hours. On the last day of data collection (26 July 1994) the spontaneous morning peak occurred at 1230 hours and the afternoon peak at 1530 hours. Thus the time of the morning and afternoon shaking peaks may have changed to mark the average times at which food was first and last available. The positive correlation between date and the time of the morning peak and the negative correlation between date and the time of the afternoon peak indicate that both peaks moved successively closer to bracket the time interval in which foraging actually occurred. These peaks therefore appear to represent a long-term response to the timing of foraging success. I will refer to this as the "work-whistle hypothesis" since, like a factory work whistle, shaking signals would signal the beginning and close of the "work day".

2. A second clue lies in the positive correlation between the magnitudes of morning and afternoon shaking peaks that were not triggered by foraging (Fig. 8A). The work-whistle hypothesis predicts such a correlation since the activation of foraging in the morning should be paired with a proportional inactivation of foraging in the afternoon.

Under this hypothesis, the spontaneous afternoon shaking peak may allow bees to predict the end of the typical foraging day and reallocate their labor to different tasks or lower their activity level to conserve colony energy. If they are typically unable to gather food after time x on each day, then on subsequent days they could save energy by reallocating resources to other activities after time x. The work-whistle hypothesis predicts that spontaneous afternoon shaking signals will cause receivers to reduce their direct participation in foraging (but not necessarily in secondary activities such as storing food). Studies on the pre- and post-signal behavior of bees during the spontaneous afternoon shaking peak would test this prediction and increase our understanding of these signals.

In closing, I will return to the general issue of signal design and function. What is the adaptive value of having a modulatory communication system with a message as general as "reallocate labor for different tasks"? Since foraging requires the coordination of several types of labor (e.g. foraging, food unloading, food storing, pollen packing), a diverse range of effects should be elicited by a signal designed to activate foraging preparations. A *general message* can thus have different *specific meanings* when receiver context supplies a specific meaning and thus a specific effect. We thus observe the shaking signal exerting a wide range of effects in a wide variety of contexts: foraging, not foraging (this paper), swarming (Allen 1959a), suppression of queen emergence (Bruinsma et al. 1981), and aggression among queenless workers (Schneider and McNally 1991).

Why are the effects of modulatory communication so difficult to detect? I propose a signal redundancy hypothesis: a high response threshold to modulatory signals enables a receiver to integrate information from multiple sources and thus increases the accuracy of information guiding the receiver's responses.

Several authors have reported that the shaking signal elicits no effect or a subtle age-specific effect (Milum 1955; Allen 1959a,b; von Frisch 1967; Schneider 1987). Their data suggest that bees possess high response thresholds to shaking signals. Such high thresholds could enable the colony to integrate individual information about the availability of food sources and thus achieve a more *reliable* colony-wide mobilization (or demobilization) of foraging. Imagine a single forager leaving in the early morning, finding a food source, and then returning and producing shaking signals. Her signals will have a limited effect since shaking signals tend to elicit weak responses. However, she may also have erred and overestimated the food source's potential value. If receivers had a low response threshold, then the colony would be prematurely roused to action by this error and waste energy inappropriately activating bees. As more foragers discover food sources outside the hive and return, they will produce more shaking signals. This increase in signaling now results from the combined information of several foragers and is thus more reliable than the information of any single forager, since the probability that they have all made errors is exponentially lower. The probability of all n foragers making errors can be modeled as m^n, where m is the probability of error per forager. As n increases, the probability of a bee receiving only inappropriate signals becomes quite small.

A high receiver threshold therefore allows the colony to integrate the information of many foragers and optimally allocate its labor. Although foragers who produce shaking signals instead of foraging cost the colony energy, this cost is probably quite small compared to the benefits of mobilizing labor for foraging-related tasks and increasing the net colony energy intake. Under normal foraging conditions, shaking signals peak during the morning and then decrease throughout the day. Thus the time spent on signaling is small relative to the total time spent foraging.

The signal redundancy hypothesis can also explain a remarkable feature of shaking signals delivered to queens. The queen is not normally shaken, but she often begins to receiving shaking signals several days before a swarm departs (Milum 1955; Hammann 1957; Allen 1958, 1959a; Fletcher 1975; Schneider 1990, 1991). These signals evidently prepare her to fly out with the swarm since she is shaken most intensely if the swarm leaves

without her and must then return (Allen 1959a). She is thus signaled to reallocate her labor from the inside to the outside of the hive, from laying eggs to flying with the swarm. However, she needs to carefully time her departure. If she flies outside prematurely, she may die or become lost without the swarm's protection. If she flies too late, the swarm must return to the hive and prepare again for departure, thereby wasting time and needlessly taking on risk by leaving the protective hive. Because thousands of workers depend upon the timing of her preparations, she needs extremely accurate information. Since the shaking signal appears to enhance the timing accuracy of her decision to depart, the signal redundancy hypothesis predicts that her response threshold and the number of signals that she receives will be in proportion to her need for accurate information, i.e. she should have a high response threshold and receive many signals. In fact, she receives *thousands* of shaking signals at an increasing rate over several days prior to her departure (Allen 1958, 1959a; Fletcher 1975). Allen (1959a) reports a maximum rate of 252 signals (from 21 different bees) delivered to the queen in 1 h! This remarkable level of signal redundancy may be best explained by her need for accurate, integrated information.

The need to coordinate activities involving multiple tasks, to integrate information and to maximize its accuracy is not unique to honey bees. We may therefore discover that modulatory signals with diverse, context-dependent meanings and high redundancy are present in a broad range of social animals.

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