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TRANSMISSION OF VIBRATION ACROSS HONEYCOMBS AND ITS DETECTION BY BEE LEG RECEPTORS

D. C. SANDEMAN*, J. TAUTZ† AND M. LINDAUER

Lehrstuhl für Verhaltensphysiologie und Soziobiologie, Theodor-Boveri-Institut (Biozentrum) der Universität, Am Hubland, D-97074 Würzburg, Germany

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Summary

Vibration of the rims of open cells in a honeycomb, applied in the plane of the comb face, is transmitted across the comb. Attenuation or amplification of the vibratory signal depends on its frequency and on the type of comb. In general, framed combs, both large and small, strongly attenuate higher frequencies, whereas these are amplified in small open combs. The very poor transmission properties of the large framed combs used in commercial hives may explain the bees' habit of freeing an area of comb from the frame in those areas used for dancing. Extracellular electrical recordings from the leg of a honeybee detect large action potentials from receptors that monitor extension of the tibia on the femur. Measurements of threshold displacement amplitudes show these receptors to be sensitive to low frequencies. The amplification properties of unframed combs extend the range of these receptor systems to include frequencies that are emitted by the bee during its dance, namely the 15 Hz abdomen waggle and 250 Hz thorax vibration.

Key words: honeybee, waggle dance, comb vibration, communication, *Apis mellifera carnica*.

Introduction

Foraging honeybees returning to the hive are able to communicate the location of food sources and nest sites during the execution of a 'dance' on the surface of the comb in the dark hive. For sites more than about 100 m from the hive, the dance contains a straight run during which the dancing bee swings its body from side to side at a frequency of approximately 15 Hz and, with its thoracic musculature, produces bursts of vibrations of approximately 250 Hz (Esch, 1961; Wenner, 1962; Michelsen *et al.* 1986).

Dancing and receiving bees probably use a number of cues, including acoustic and olfactory, but there is little direct evidence to suggest the use of comb vibration by dancers despite its importance as a communication pathway for begging bees and foragers and for piping queens and workers (Michelsen *et al.* 1986; Kirchner, 1993). Recently it was discovered that forager bees dancing on open, empty combs recruit three times as many nest mates to feeding sites as those that dance on capped brood combs (Tautz, 1996). This result can be interpreted as indicating that dancing bees could indeed rely on vibratory signals transmitted across the comb surface, but that the important component of vibration may be a displacement of the cell walls in the plane of the comb face and not at right angles to it. This idea is supported by video recordings of dancing bees (von Frisch and Lindauer, 1975) and by our own very high-speed video recordings (200 frames s^{-1}) that show how, not just the abdomen, but the entire body of the bee is thrown first one way and then the other against the rims of cells on which it stands. The motion resembles someone sitting in a boat and rocking it from side to side. Follower bees arranged along each side of the dancer also have the tarsi of their front legs placed firmly on the rims of the cell walls, usually no more than one cell away from the dancer and sometimes on the very wall supporting the dancer itself.

Two aspects of the dance were of particular interest to us. First, the frequencies involved in the dance are relatively low and, second, the motion of the bee is more likely to produce a displacement of the cell walls that is in the plane of the comb face rather than at right angles to it. If such low-frequency vibrations are indeed transmitted through the comb, relevant receptor systems in follower bees will probably be found in the legs. The presence of very sensitive vibration receptors in insect legs has long been known (Autrum, 1941; Autrum and Schneider, 1948; Shaw, 1994), and the morphology of one of these receptors in bees, the subgenual organ, has been described (Schön, 1911; McIndoo, 1922). The mechanical behaviour of the bee subgenual organ to vibrations applied to the leg show it to be maximally displaced, and so presumably most sensitive, to frequencies between 300 and 600 Hz (Kilpinen, 1995; Rohrseitz and Kilpinen, 1996).

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*Present address: Biological Science, University of New South Wales, NSW 2052, Australia (e-mail: sandeman@acsusun.acsu.unsw.edu.au).
†To whom reprint requests should be addressed.

While more than sensitive enough for the detection of comb vibrations (the begging call produces displacement of about 1.5 µm, Michelsen *et al.* 1986), the subgenual organ would seem to be tuned to frequencies that would exclude any displacement of the comb caused by the dancing (15 Hz) and would attenuate the intermittent 250 Hz thoracic vibrations of the dancing bee. The subgenual organ, however, although perhaps the most sensitive, is not the only receptor organ in the bee leg that could monitor displacements of the leg joints. Debaisseux (1938) describes both femoro-tibial and tibio-tarsal chordotonal organs, fairly large systems that, by their insertions, could respond to articulation of knee and foot joints.

We set out to examine the phenomenon of low-frequency (10–500 Hz) displacement of honeycomb cell walls in the plane of the comb face and to search for receptor systems in bee legs that may be tuned to detect these. In this paper, we describe electrophysiological responses of mechanoreceptors in the bee leg to low-frequency comb vibration in the plane of the comb face and laser vibrometry measurements of the transmission of low-frequency vibration across honeycomb. We conclude that framed combs do not provide optimal conditions for the transmission of the vibratory signals produced by dancing bees and draw attention to a behavioural response of bees that may well be used by them to improve the poor transmission properties of framed combs.

Materials and methods

Before making recordings, honeybees (*Apis mellifera carnica*) were immobilised by placing them in a freezer (–4 °C) for 2–3 min. The bee was killed by decapitation and a leg removed at its base. Extracellular electrical discharges from the isolated bee leg were recorded by inserting two steel pins through the femur and connecting the pins (ground and active) to an amplifier (WPI DAM50). The pins also served to anchor the preparation to a Sylgard block which was mounted in a manipulator. The leg was then positioned so that the tarsus rested either on an electromagnetic shaker (Bruel and Kjaer 4810), to determine the threshold to displacement of the electrical activity, or on the rim of a honeycomb cell wall, to test its response to displacement of the cell wall in the plane of the comb face (Fig. 1). To ensure that the tarsus did not slip off the shaker or cell wall, a heated insect pin was used to melt a little wax beneath the terminal claws of the tarsus. No wax was applied to the upper part of the tarsus, tibia or femur. Preparations made in this way last 30–120 min before changes in the size of receptor discharges or their sensitivity to displacement appear. Weakening preparations were immediately abandoned.

Electrical discharges from the femur were fed into a window discriminator (WPI121) and to an audio system and loudspeaker. Thresholds at different frequencies were determined by ear and by observing raw and window-discriminated data on an oscilloscope (Fig. 1). Amplitudes of applied displacements were measured directly at the tarsus with a laser vibrometer (Polytec). Small reflective flags (0.5 mm×0.5 mm) were attached to surfaces measured by the laser vibrometer.

Combs used in these experiments were small (120 mm long by 90 mm deep) and constructed by bees along a wooden slat measuring 175 mm long, 20 mm wide and 8 mm thick. Such combs resemble those made by bees in the wild and differ from those found in commercial hives in that the comb is supported only along one edge and is not enclosed by a rigid wooden frame. For one series of measurements, we enclosed honeycomb in a small wooden frame measuring 120 mm by 128 mm and constructed of the same material as the support for ‘open’ comb. The edges of the comb were fused onto this frame by melting beeswax down between comb and frame. We also measured the transmission of vibration across standard commercial frames

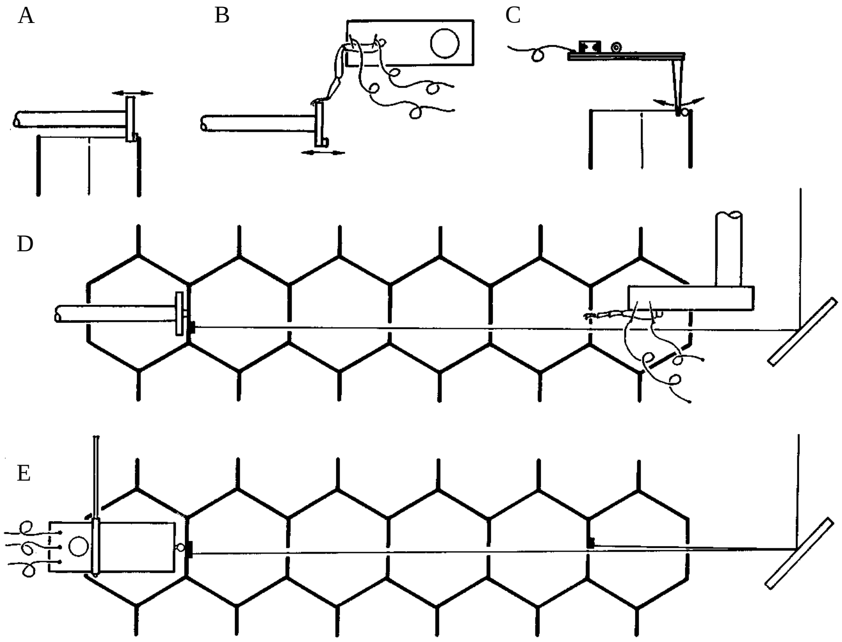

Fig. 1. Vibration of the comb in the plane of its face was induced by resting a probe against the side of the cell wall and driving this with an electromagnetic vibrator (A,D) or with a piezo-electric beam (C,E). The piezo-electric beam was pivoted and counterweighted so that the probe rested against the cell wall (C). Displacement threshold of leg stretch receptors to different frequencies of vibration was measured by supporting the tip of the tarsus on the vibrator. The leg was pinned through the femur to a block of Sylgard (Dow Corning), the pins acting as the electrodes that recorded the responses of the stretch receptors (B). Transmission of the signal across the comb was monitored with the leg preparation (D) or measured directly with a laser at the source of the vibration and at points distant from it (E).

that measured 40 cm×20 cm×2 cm and had been used over the summer months by bees. Small combs were supported by attaching a clamp to the wooden slat on which they had been built and fastening the clamp to a stand on a vibration-free table. Large commercial combs were supported by resting the wooden frame on four pillars that stood on a vibration-free table. The combs we used contained only empty, open cells.

Hexagonal cells in honeycomb are oriented in the hive so that an apex and two walls of the hexagon are vertical relative to gravity. We limited ourselves here to displacing the rims of the vertical cell walls from the side and in the plane of the comb face. This approximates the situation of a dancing bee making her waggle run vertically up a comb in the hive.

Displacement of honeycomb cell walls in experiments with leg mechanoreceptors was produced by the same Bruel and Kjaer shaker used to determine their threshold responses. Displacement of the cell walls in experiments on the transmission of vibration across the comb was produced by a bilayered piezo-electric transducer (BM/ML 60/40/300, Piezomechanik, München). The beam of the piezo-electric transducer was supported on a pivot and weighted so that a 1 mm diameter stimulus probe attached to the beam pressed lightly against the thickened rim of the cell wall (Fig. 1). The force exerted by this system was measured with a Bruel and Kjaer force transducer (8001 impedance head) and found to be 1 mN at 10 Hz. The piezo-electric beam bends in an arc when driven by an electrical signal. The probe attached to the beam therefore displaced the cell wall both in the plane of the comb face and also at right angles to it. Direct vibrometer measurement showed the displacement in the plane of the comb face to be 2.5 times larger than that at right angles to it.

Displacement amplitude of the cell rim was measured with the laser vibrometer first at the point of the stimulus and then on a rim, four cells away (Fig. 1E). Input displacement amplitudes were set at 2 µm for each frequency and the resulting output then measured. Although time-consuming, this procedure allowed us to exclude frequency-dependent properties of the transducer system from our measurements. In every case, measurements were made only after the signal had reached and maintained a stable amplitude for about 30 s. Each series of measurements was repeated 4–5 times.

Results

Mechanoreceptive responses

Extracellularly located pin electrodes are unselective and record the responses of a large number of mechanoreceptors when inserted through the femur. If positioned nearer the inner side of the femur, two or three particularly large, slowly adapting tonic units were found that were sensitive to extension of the tibia on the femur. Flexion of the tibia on the femur silenced the units (Fig. 2). Removal of the tarsus at its joint with the tibia had no observable effect on the large units, which exhibited virtually the same sensitivity to displacement with or without the tarsus in place.

The femur in our preparations was fixed horizontally by the recording needles, the tibia extending down at right angles to the femur. The tarsus was angled outwards, with the terminal joints resting horizontally on the shaker (Fig. 1B). Displacement was therefore transmitted to the tibia–femur joint along the entire leg in approximately the way it would be in a follower bee facing the side of a dancing bee. We used the prothoracic legs in these experiments, but the same mechanoreceptors can be found in both meso- and metathoracic legs. The responses of the metathoracic leg receptors were less sensitive than those of the pro- or mesothoracic legs, perhaps because of their larger size and the accompanying decrease in the angular excursion between femur and tibia due to the greater length of the tibia and tarsus.

The threshold to displacement for sinusoidal stimuli showed the mechanoreceptors in legs to be fairly broadly tuned and most sensitive to frequencies between 30 and 100 Hz. At their best frequencies, they can detect displacements of about 2 µm (Fig. 3). Flexing the tibia results in a large decrease in the sensitivity of the receptors to displacement (Fig. 4).

To test the responses of leg mechanoreceptors to displacement of the comb in the plane of its face, we positioned the tip of the shaker probe (1 mm square) against the rim of the honeycomb cell wall (Fig. 1D). The soft wax rims of open cells adhered to the probe so that relatively large displacements of the probe (up to 90 µm) could be made without the wall separating from the probe. In these experiments, we used the vibrometer to measure the displacement amplitude of the cell

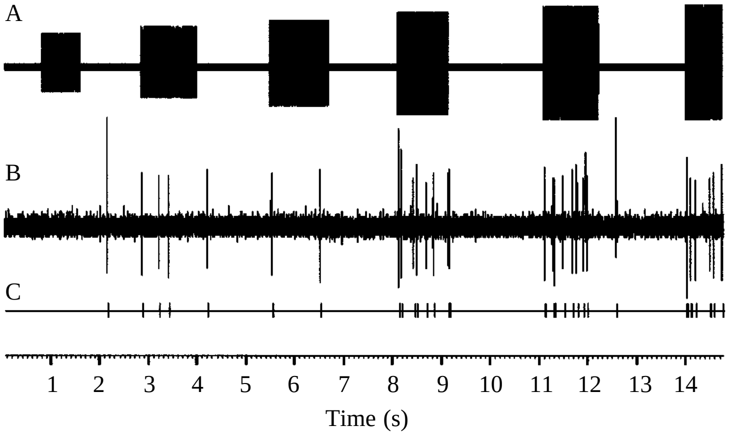

Fig. 2. Electrophysiological recordings from the femur of a bee leg showing responses of large units to increasing amplitudes of vibration applied to the tarsus. (A) The signal amplitude (150 Hz here); (B) the signals recorded from the leg nerve; and (C) the signal filtered by the window device. Threshold was defined as that amplitude at which the responses could be unequivocally associated with the stimulus; the fourth stimulus from the left, in the example shown here.

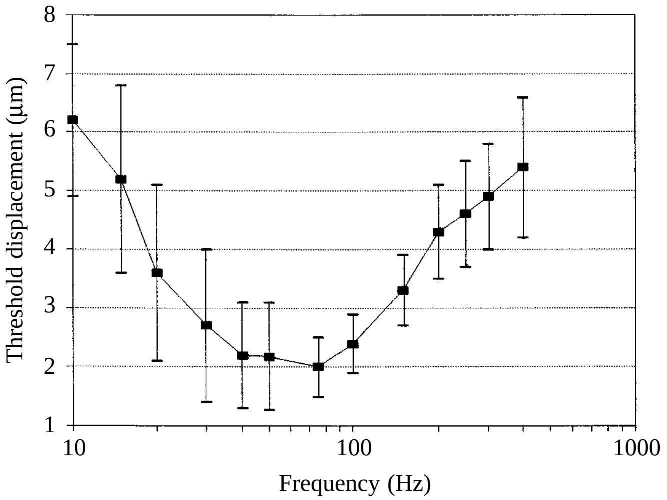

Fig. 3. The threshold displacement of nine large units from nine different animals to displacement of the leg. The tarsus in these experiments was fixed to an electromagnetic shaker and its displacement measured directly with the laser vibrometer. There is considerable variation at the lower and higher frequencies and less around the most sensitive frequencies between 30 and 100 Hz. Bars indicate the standard deviation.

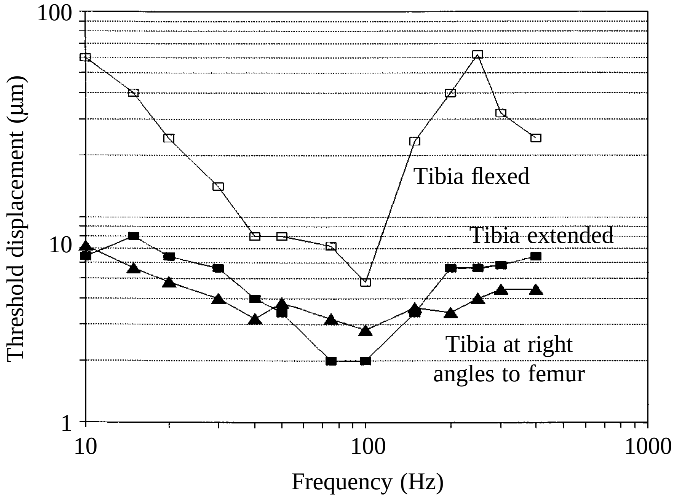

Fig. 4. The effect of flexion and extension of the leg on the threshold displacement of a single large unit in one animal. Extension of the tibia (■) on the femur does not have a large effect on its sensitivity to displacement. Flexion of the tibia (□) on the femur was accompanied by a marked decrease in sensitivity of the organ to displacement. ▲, tibia at right angles to femur.

rim necessary to discharge the leg mechanoreceptor. Thus, threshold curves are related to the displacement amplitude of the cell rim.

Placing the tarsus on the cell rim opposite to the one being shaken produces a threshold curve that approaches that obtained with the tarsus resting on the shaker. This is to be expected, given the short distance between the shaker and the leg of the bee. Changes in the stimulus signal amplitude imposed by the filtering and resonant properties of the comb gradually appear as the leg is moved further away from the stimulus (Fig. 5).

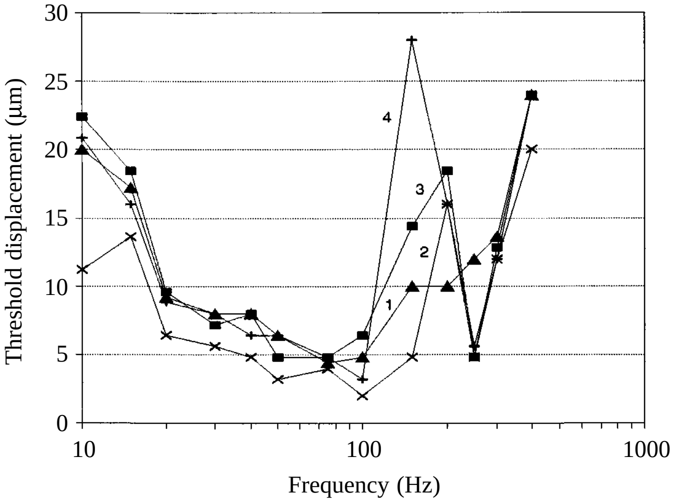

Fig. 5. Changes in the threshold displacement of a large unit that occur as the stimulus point is moved further away from the cell on which the tarsus of the bee was resting in a small open comb. 1 (▲), the tarsus was on the cell rim on the opposite side of the same cell to which the vibration was applied. 2 (×), two intervening cells, 3 (■), three intervening cells, and 4 (+), four intervening cells. We find little difference between the threshold displacement of the organ to stimulation between 20 and 100 Hz, but above 100 Hz the results indicate a complex interaction of resonances and damping. The first indication of an amplification of the signal at around 250 Hz can be seen here in the relatively low threshold to this frequency.

The collected results from nine preparations, in which the legs were all three or four cells away from the stimulus, confirm the results of the single preparation in Fig. 5 and show the apparently broadened bandpass of the receptors when subjected to vibrations that have passed through the comb (Fig. 6). On small wild combs, the threshold to vibration around 20 Hz and 250 Hz is lowered, but it is raised for signals at 150 Hz or above 300 Hz. From the responses of the mechanoreceptors, it would appear that vibration frequencies around 150 Hz and above 300 Hz are being strongly damped by the comb, whereas those around 20 Hz and 250 Hz are being amplified by it.

Transmission of vibration across small open combs

To explore the filtering and resonant properties of small open combs further, we undertook a series of input–output measurements with the laser vibrometer. The rim of a cell was displaced through 2 μm in the plane of the comb face and the resulting displacement of a cell rim, four cells distant, was measured. Each measurement was repeated several times for frequencies ranging from 10 Hz to 500 Hz (Fig. 1E). The small comb contained 18 horizontal rows of cells from its lower edge to the upper edge support on which it had been constructed. The displacement was, as before, from the side of the comb and in the plane of the comb face. Measurements were carried

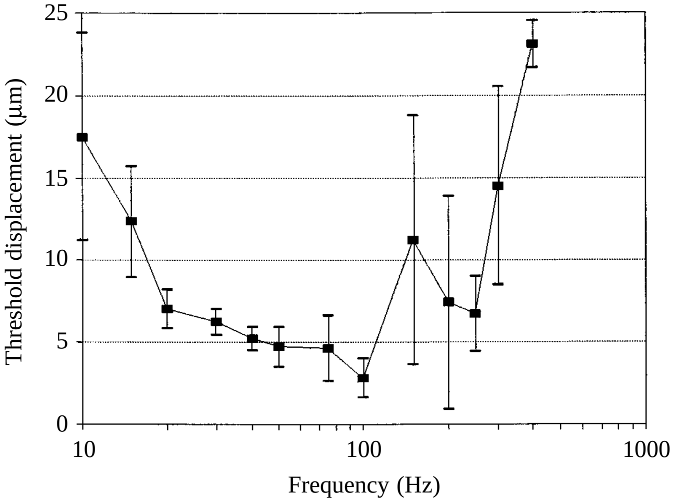

Fig. 6. Threshold displacements of nine different units from nine different animals where the tarsus was three or four cells away from the applied vibration on a small open comb. The sensitivity declines steeply to 20 Hz and becomes slightly more sensitive at 100 Hz. Signals of 150 Hz are damped, whereas those around 250 Hz are amplified. Bars indicate the standard deviation.

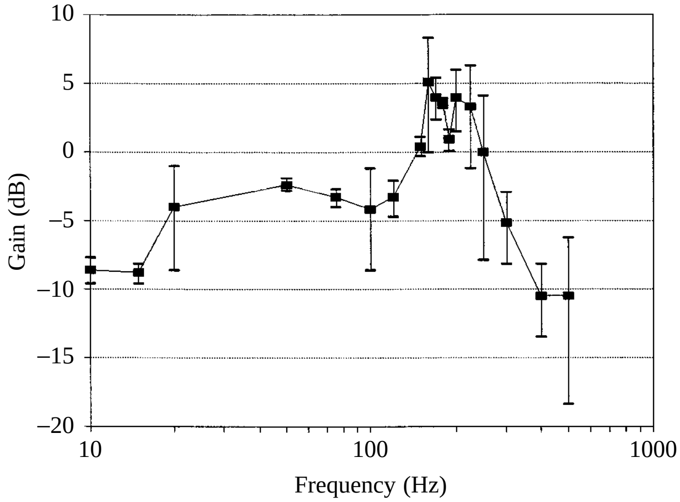

Fig. 7. Transmission of vibrations across a small open comb revealed by input/output measurements of the rims of the cells. 0 dB defines the level at which the amplitude of the output signal is the same as the input amplitude. Vibrations below 150 Hz are all attenuated, but those above 150 Hz and below 250 Hz are amplified. Bars indicate the standard deviation; N=5.

out on cells in rows 1 (lower edge), 4, 7, 11 (central) and 13 (upper edge, near support).

Transmission of vibration across cells located near the centre of the comb (rows 4, 7 and 11) supported the indications of the electrophysiological results: vibration from 10 Hz to 15 Hz is attenuated but less so between 20 Hz and 75 Hz. Vibration is damped around 150 Hz but amplified around 250 Hz (Fig. 7).

Comparing these results with the measurements obtained from rows 1 (lower edge) and 13 (upper edge) reveals a further complexity. 10 Hz vibration transmitted along the edge of the comb is highly damped, whereas that at 20 Hz is amplified. 100 Hz vibration is more heavily damped, as is that at 300 Hz and 400 Hz. 250 Hz signals are almost doubled in size (Fig. 8). Signals travelling along the comb near its support provide us with yet another profile. All lower frequencies are heavily damped. Vibrations around 150 Hz are amplified, but those above 200 Hz are attenuated and no signals above 400 Hz reach the fourth cell from the stimulus (Fig. 9).

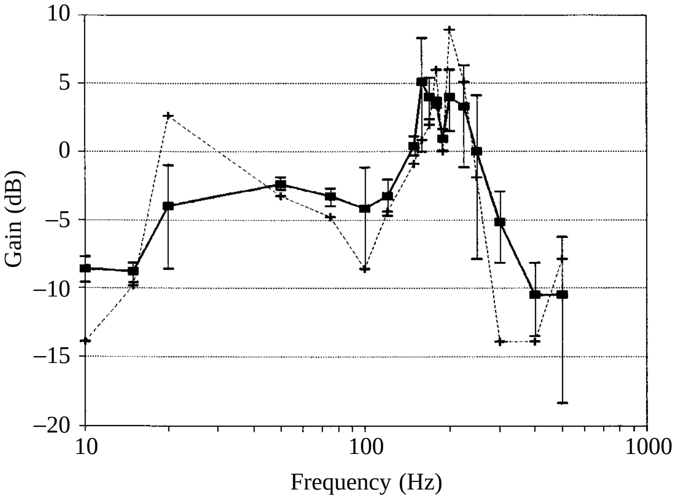

Fig. 8. A comparison of the input/output characteristics of the cell rows in the centre of the small open comb (solid line) with those lying along the lower, open edge (dashed line). The displacement was applied to the side of the comb, in the plane of its face. Signals around 10 Hz and 100 Hz are attenuated, whereas those around 20 Hz and 200 Hz are amplified. Amplification of the signals is greater at 20 Hz and 200 Hz in the cells along the lower edge than in the centre, i.e. the horizontal row to which the signal was applied. Bars indicate the standard deviation; N=4.

Transmission of vibration in framed combs

Attenuation of signals near the support in small combs could be explained by an increase in mechanical impedance imposed by the relatively inflexible wooden slat on which the comb had been built. Comparisons between wild and framed combs have shown impedances in framed combs to be ten times higher than those of the wild combs (Michelsen *et al.* 1986).

Repeating our measurements on a small (120 mm×120 mm) framed comb confirmed the damping effect of the frame. The transmission profile along the edge of the framed comb is very similar to that found for the cells along the support in the unframed wild comb. Transmission in the centre of the framed comb is poor, with all frequencies being attenuated, although the basic bandpass features of the small comb are still apparent (Fig. 10). Transmission of vibration in large framed commercial combs is even worse than in small framed combs with frequencies of around 20 Hz being passed with considerable attenuation and signals above 100 Hz no longer measurable.

The poor vibrational performance of large commercial

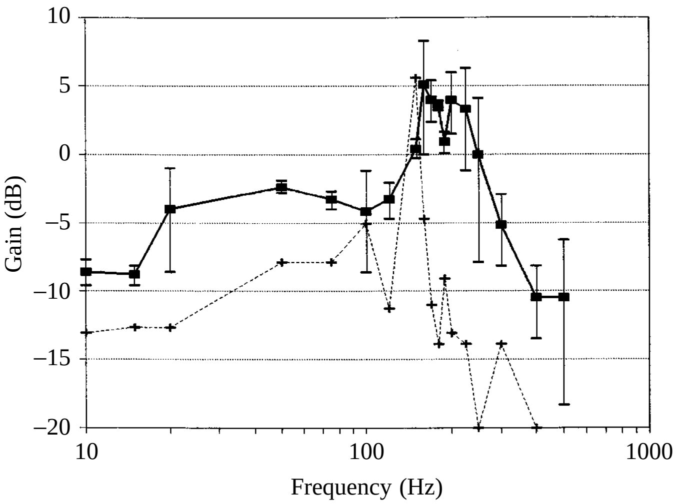

Fig. 9. As Fig. 8 except that the dashed line shows the characteristics of the cell row that lies close to the supporting slat at the top of the small open comb. Apart from the resonance peak around 150 Hz, signals at all frequencies are attenuated in the cells along the support. Bars indicate the standard deviation; *N*=4.

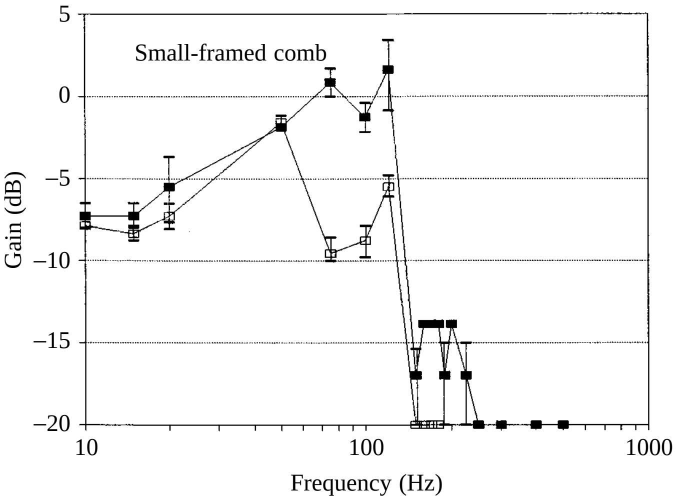

Fig. 10. The effect on transmission of vibration in small combs when they are bounded on all sides by a wooden frame. Although the overall frequency characteristics of the small open comb are still recognisable, the signals are strongly attenuated. Filled squares, cell row along supporting frame; open squares, cell row in centre of comb. Bars indicate the standard deviation; *N*=5.

combs and far better performance of combs with free edges led us to direct our attention to combs that had been used by bees over the summer and in which they had removed cells between the comb and the frame (Fig. 11). Apparently a common feature in bee hives and long known to one of us (M.L.), the areas of wax removal are usually found in frames on which the foragers dance. We measured the transmission of vibration along cell rows in such a comb, both in areas where it was fused to the frame and where the comb had been freed from the frame. Transmission along cell rows in areas fused to the frame is, as in all large frames, tuned to frequencies around 20 Hz and no frequencies beyond 120 Hz are conducted far from the source. However, 20 Hz signals passing across cells opposite an area of comb freed from the frame are amplified, and although 250 Hz signals are attenuated, the bandpass profile of the small wild combs can again be recognised (Fig. 12).

Displacements of the rim of a cell in an area of comb freed of the frame can be recorded in cells up to 30 cells away (Fig. 13), suggesting that bees dancing in a frameless area could broadcast signals over a much greater distance than those dancing on framed combs.

The transmission pathway

While much has been written about the dimensional uniformity of honeycomb, less has been said about the heterogeneity within a single cell. The rims of the cells, for example are many times thicker than the walls (rims ±300 µm; walls ±70 µm) (Fig. 14). The rims are also soft and sticky, whereas the walls are papery and do not melt easily when heated with a hot probe. The thickness of the rims led us to suspect that vibration may be carried by the rims and that the

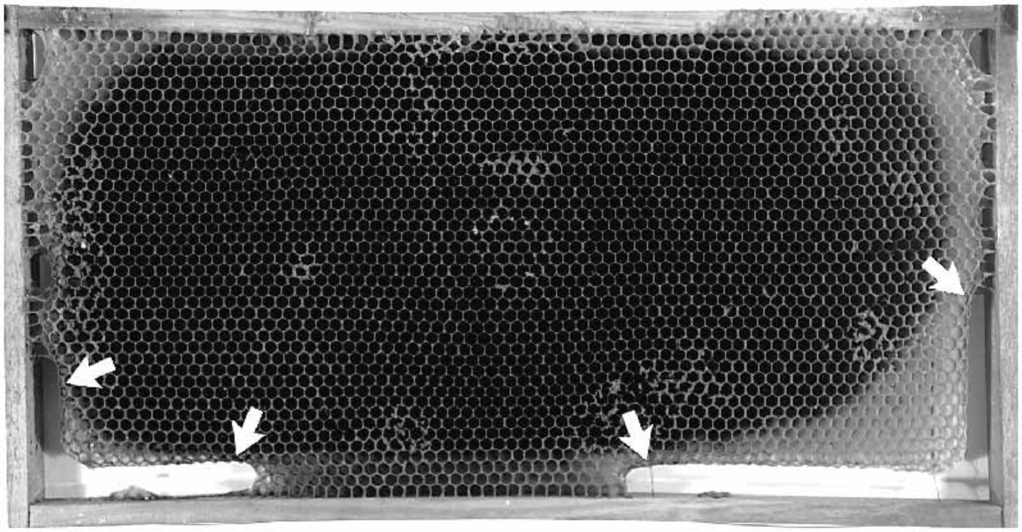

Fig. 11. A large comb taken from an active hive at the end of summer. The bees have removed cells between comb and support at the lower left- and right-hand corners of the frame (regions between the arrows). Forager bees conduct their dances in these areas.

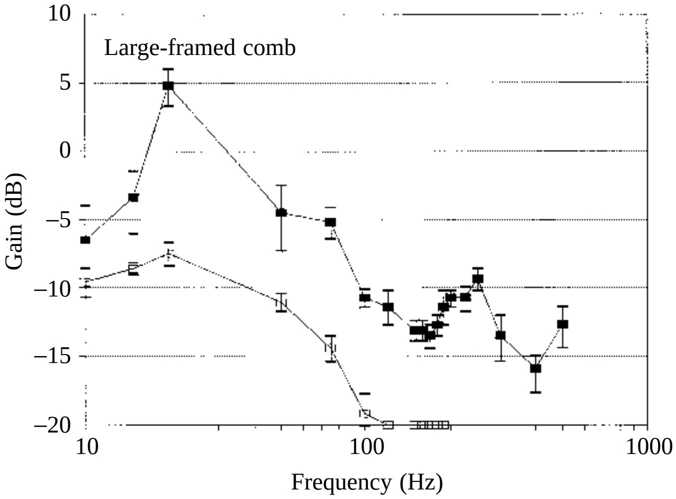

Fig. 12. The transmission of vibration in a large comb along a row of cells in an area where the comb was free of the frame (filled squares) and along a row of cells in an area where the comb was fused to the frame (open squares). In both cases, signal with frequencies around 20 Hz are transmitted with least attenuation. The higher-frequency signals are transmitted only along cell rows in the area free of the frame which, as in the small open combs, resonate around 250 Hz. Signals above 200 Hz were not transmitted in cell rows where the comb is fused to the frame. Bars indicate the standard deviation; *N*=5.

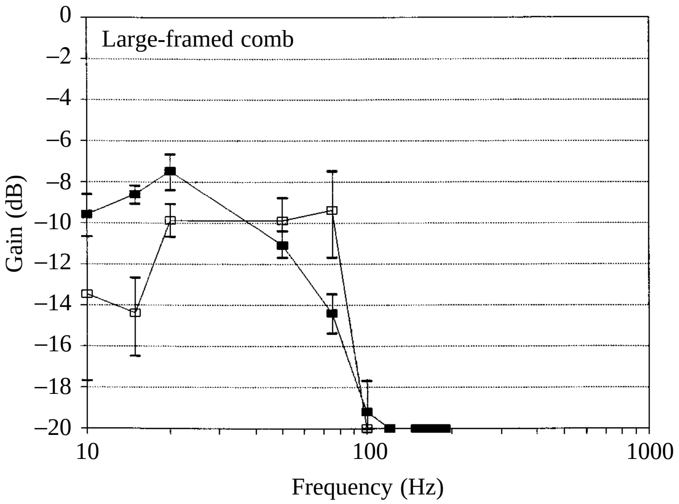

Fig. 13. Transmission of signals across a large comb when the signal is applied in an area where the comb is fused to the frame or free of the frame. The filled squares show the nature of the transmission when both input and output were in an area where the comb was fused to the frame and where the input was four cells away from the output. The open squares show the transmission of vibrations where the input was in an area free of the comb, and the output was in an area of comb fused to the frame, but 30 cells away from the input. Thus, applying vibration to an area free of the frame results in transmission of the signal across a much greater distance. Bars show standard deviation; *N*=5.

walls of the cells beneath them provide a fairly flexible support. To test this supposition, we applied vibration to the rim of a cell in a small wild comb and measured the transmission of the displacement over four cells, as before. With a hot needle we then removed the walls beneath the rims on each side of the one being displaced and re-measured the transmission properties of the comb. No change in the amplitude of the transmitted signal was recorded. Removal of the cell rims adjacent to the one receiving the stimulus, however, attenuated the transmitted signals (Fig. 15).

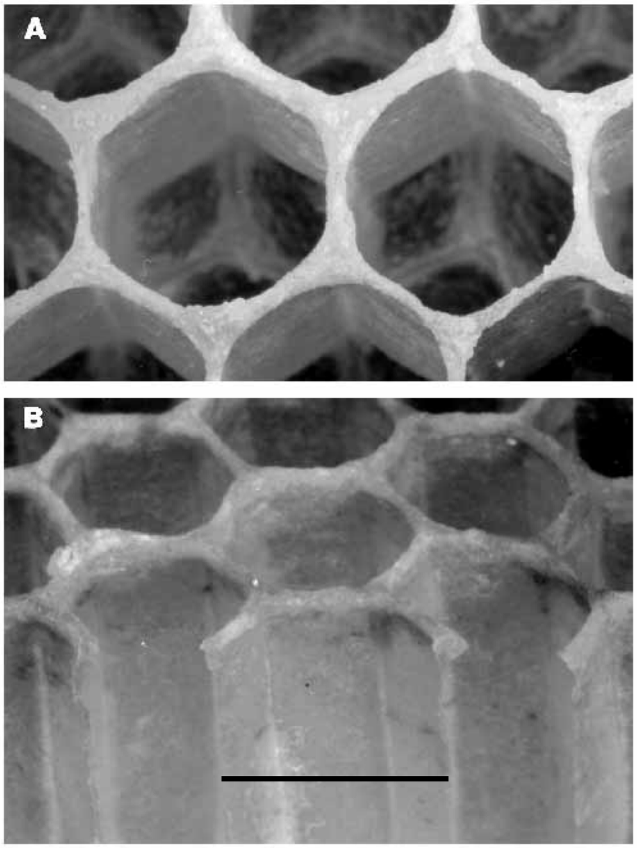

Fig. 14. Photograph of open, empty honeycomb, showing buttressed corners of the cells (A) and, in longitudinal section (B), the thickened rims of the cell walls. Scale bar, 5 mm.

Discussion

Our results show that small (2 µm) displacements, in the plane of the comb face, of the rim of open cells in honeycombs are transmitted across the comb and that under certain conditions such displacements are amplified. We have also shown that receptors in the legs of bees are able to detect displacements at frequencies that are close to those produced by bees during their dances. The consequences of enclosing honeycombs in frames, the possible nature of the leg receptors, and the effect of the thick rims of open-celled honeycomb on transmission of vibratory signals, are discussed below.

The bandpass of honeycomb to displacement of the cell walls in the plane of the comb face

Of the three different types of comb used, only the open,

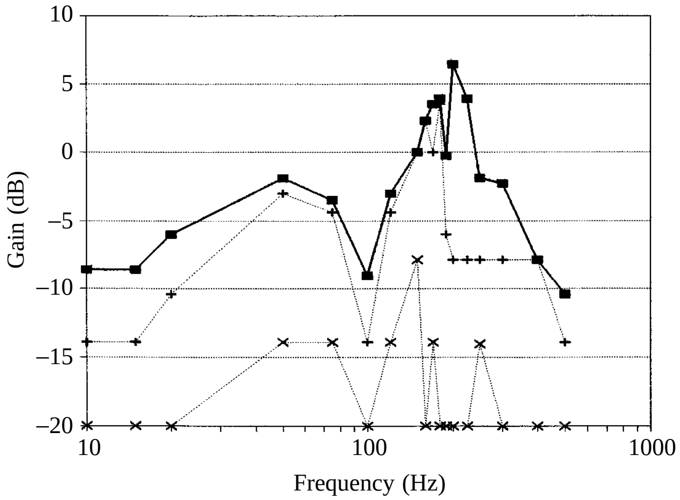

Fig. 15.Transmission of vibration across a small open comb when all rims between the vibrated rim and the rest of the comb were intact (solid line), when the anterior lateral rims were destroyed (dashed line, + symbols), and when all rims on either side of the vibrated rim were destroyed (dashed line, × symbols). Breaking the rims (but not the walls) of cells adjacent to the wall being displaced leads to a significant attenuation of the transmitted signals.

unframed comb provided a relatively unimpeded path for signals at frequencies around 250 Hz. Unframed combs actually amplified the signals transmitted across areas not restricted by the support, with the result that the sensitivity of the receptor organs from which we recorded was extended over a broader range of frequencies (Fig. 6). Amplification of the signals in the comb was an unexpected result and indicates the presence of resonances in small open combs. Our measurements did not allow us to determine the source (or sources) of the resonance, which could be limited to specific regions of the comb or be a property of the entire structure. Some of the amplification we found in the open combs may have been caused purely by the geometry of the situation and the possible action of the thick rims of the cell walls (see below).

Combs bounded by frames on all sides still carry a low-frequency signal (about 15 Hz), but high-frequency displacements could not be detected four cells away from the source (Figs 10, 12, 13).

Our results on the transmission of vibration in the plane of the comb face accord with and confirm the results of Michelsen *et al.* (1986) on the transmission of vibration at right angles to the plane of the comb face. Vibrations at right angles to the comb face are produced by bees that ‘beg’ food from returning foragers. Begging bees press their thoraces down onto the comb while producing a 320 Hz vibration of the thoracic musculature. Displacements of up to 1.5 µm at right angles to the plane of the comb face can be measured a few centimetres away from begging bees. Such vibrations are heavily damped in framed combs. The highly complex and variable nature of honeycomb and the changes in impedance that occur across the combs has already been reported by Michelsen *et al.* (1986), who point out the difficulties in obtaining values that would be representative for honeycomb in general, given the extreme variation caused by the shapes and the influence of inflexible supports on adjacent areas of the comb.

From the biological point view, however, one has to consider that, if the bees are indeed using the comb as a transmission pathway, the signals they are using must be robust enough to carry the essential message across less than perfect lines of communication. That the message content is influenced by imperfections of the comb is clearly indicated by the behavioural demonstration that bees dancing on open combs recruit more effectively than those dancing on capped combs (Tautz, 1996). The interesting aspect of this finding is that the bees dancing on capped combs were still able to recruit, but not as well as those on the open combs, suggesting that the signals were weakened rather than abolished. Examining the bandpass characteristics of the three combs we used (small open, small framed and large framed) in this light we see that, although the high frequencies are clearly attenuated in the framed combs, the overall bandpass profiles are the same in all cases. Also of importance in terms of the information transfer is the accuracy with which the bees need to indicate the direction of the food source and the adaptive disadvantage of directing recruits too narrowly to a food source (Seeley, 1986).

The habit of begging bees of pressing their thoraces against the surface of the comb may provide us with another clue to the waggle dance. High-speed videos of dancing bees show that, during the dance, bees alter their gait and take only a single ‘step’ (i.e. each leg moves once) during even the longest dances (Tautz *et al.* 1996). The consequence of the altered gait is that for most of the time the tarsi are holding fast to the rims of cells. This observation leads us to suggest that the bee, which has a relatively small mass and is supported on slender legs, is using the waggle to improve the transfer of the bursts of thoracic vibration to the comb. The bee swings its entire body weight against the legs clutching the rims of the walls, and produces a dynamically changing mechanical tension in them, possibly preparing them for an efficient propagation of the 250 Hz vibration.

Impedance of the comb

Measurements of the impedance of the different combs provide us with useful information on the environment in which the bees are operating. Open wild combs have an impedance that is about one-tenth that found in the framed combs routinely used in commercial hives (Michelsen *et al.* 1986). A curious, and so far unexplained, response of bees to such framed combs is to gnaw part of the comb away from the frame (Fig. 11). The effect on the transmission of the displacement in the plane of the comb face is shown in Figs 12, 13. Transmitted displacements in the free area of the comb contain both low and high frequencies, and the bandpass profile resembles that of the small open comb. In the same comb, but in an area attached to the frame, only low-frequency displacements are transmitted, and these are significantly attenuated. The bees behavioural response to the framed combs is remarkable because the areas that are freed from the frame are closest to the entrance to the hives; in other words, where the dances take place. The behaviour of the bees suggests that they are able to determine that the impedance of the framed honeycomb is less than optimal for the transmission of dance signals and to take the appropriate action.

An aspect to be considered in the transmission of vibratory signals in open wild combs, particularly where the signals are amplified, is that two dancers close to one another would, in such a case, interfere with one another. In the natural situation, however, there are many bees concentrated on the dance floor and the system may be self-regulating in that follower bees absorb and damp out the vibrations from the bee dancing closest to them.

The receptors

Impaling the limbs of insects with needles as a means of recording the electrical impulses from the axons in the leg nerves has been used many times, and the technical reason why it is so effective has been clearly explained by Shaw (1994). A disadvantage of the method is that recordings contain signals from many neurones and it is difficult in intact animals to be certain whether action potentials are afferent or efferent. Removing the leg excludes efferents but changes the receptor systems into purely passive organs which, in the absence of muscle tonus, may have become less sensitive. Hence, the recordings from the isolated legs may be taken as a ‘worst case’ situation as far as the absolute sensitivity is concerned. It is also possible that the frequency characteristics of the sensory organs are altered.

We have not identified the receptors from which we recorded. Their relative insensitivity would suggest to us that we are not recording from the subgenual organs because in insects these organs are among the most sensitive known to exist in the animal kingdom (see review by Shaw, 1994) and those of the bees are no exception (Kilpinen and Michelsen, 1994). In addition, the subgenual organs appear to be sensitive to frequencies that are significantly higher than those recorded here (Rohrseitz and Kilpinen, 1996). Other receptor organs are known in the legs of insects, and possible candidates are the chordotonal organs that monitor the tibio-femoral and tibio-tarsal joints (Debaisseux, 1938).

Whatever the identity of the receptors, their characteristics would seem to fit them admirably for the task of detecting the low frequencies that are associated with the dancing bees. The question that remains is whether they are sensitive enough. At best, they were shown to detect directly applied displacements of about 2 μm at frequencies between 20 and 100 Hz (Fig. 3), which is considerably less sensitive than that known for leg receptors in other arthropods (Barth, 1985; Shaw, 1994). The dance frequencies lie slightly below and above this, however, making the resonant properties of the small comb so interesting because, when the leg is placed some distance away from the stimulus on the comb, the bandwidth of the receptors is broadened to be almost flat from 20 Hz to almost 300 Hz (Fig. 6).

We do not yet know what effect muscle activity could have on the sensitivity of the receptor organs, but an indication that the sensitivity is variable and dependent on the relative amount of stretch that is applied to the joints comes from our measurements of the threshold displacements of isolated legs in flexed or extended positions: flexing the tibia on the femur produces an approximately tenfold decrease in absolute sensitivity of the receptor organ compared with a tibia at right angles, or extended, relative to the femur (Fig. 4). The sensitivity of the receptor organ could be under the control of the animal.

Honeycomb as a ‘web’

There is a large difference between the thickness and nature of the rims of the cell walls and the walls themselves (Fig. 14). Destroying the walls beneath the rims has very little effect on the transmission of their displacements, whereas removing adjacent rims and walls severely attenuates the signals (Fig. 15). This result leads us to suggest that honeycomb could be likened to a web (the rims) ‘floating’ on a substratum of thin and flexible walls. Such a structure could explain the differences in amplification of displacements applied to the side of an open comb, in the plane of its face (Fig. 8). Under these conditions, amplification of the signal along the lower, open edge of the comb is greater than that directly horizontal to the applied signal. If the rims of the cells form a fairly rigid ‘curtain’, pivoted at the upper edge, then displacements from the side are bound to be larger along the lower, free edge and smaller at the point of attachment to the support. Signal transmission along such a structure could also be expected to be frequency-dependent. However, such a ‘curtain’ will not explain the amplification we measure in the cell row, horizontal to the point at which the signal is applied. Resonance appears to be the only possible source of amplification here. The mechanism underlying this amplification can only be unravelled by a careful cell-by-cell analysis of honeycomb.

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References

AUTRUM, H. (1941). Über Gehör und Erschütterungssinn der Insekten. *Z. vergl. Physiol.* **28**, 580–637.

AUTRUM, H. AND SCHNEIDER, D. (1948). Vergleichende Untersuchungen über den Erschütterungssinn der Insekten. *Z. vergl. Physiol.* **31**, 77–88.

BARTH, F. G. (1985). Neuroethology of spider vibration sense. In *Neurobiology of Arachnids* (ed. F. G. Barth), pp. 203–207. Berlin, New York, Heidelberg: Springer Verlag.

DEBAISSEUX, P. (1938). Organes scolopidiaux des pattes d’insectes. II. *La Cellule* **47**, 77–202.

ESCH, H. (1961). Über die Schallerzeugung beim Werbetanz der Honigbiene. *Z. vergl. Physiol.* **45**, 1–11.

KILPINEN, O. (1995). A method for visualising the oscillations of the subgenual organ in the honeybee. *Proceedings of the 23rd Göttingen Neurobiology Conference: Learning and Memory* (ed. N. Elsner and R. Menzel), p. 271. Stuttgart, New York: Thieme Verlag.

KILPINEN, O. AND MICHELSEN, A. (1994). Vibration sensitive neurons in the honeybee, *Apis mellifera. Proceedings of the 24th Göttingen Neurobiology Conference: Sensory Transduction* (ed. N. Elsner and H. Breer), p. 324. Stuttgart, New York: Thieme Verlag.

KIRCHNER, W. H. (1993) Acoustical communication in honeybees. *Apidologie* **24**, 297–307.

MCINDOO, N. E. (1922). The auditory sense of the honeybee. *J. comp. Neurol.* **34**, 173–199.

MICHELSEN, A., KIRCHNER, W. H. AND LINDAUER, M. (1986). Sound and vibrational signals in the dance language of the honeybee, *Apis mellifera*. *Behav. Ecol. Sociobiol.* **18**, 207–212.

ROHRSEITZ, CH. AND KILPINEN, O. (1996). Vibration transmission characteristics of legs in honeybees and other insects. *Proceedings of the 24th Göttingen Neurobiology Conference: Brain and Evolution* (ed. N. Elsner and H.-U. Schnitzler). Stuttgart, New York: Thieme Verlag.

SCHÖN, A. (1911) Bau und Entwicklung des tibialen Chordotonalorgans bei der Honigbiene und bei Ameisen.

SEELEY, T. D. (1986). Social foraging by honeybees: how colonies allocate foragers among patches of flowers. *Behav. Ecol. Sociobiol.* **24**, 181–199.

SHAW, S. R. (1994). Re-evaluation of the absolute threshold and response mode of the most sensitive known ‘vibration’ detector, the cockroach’s subgenual organ: A cochlea-like displacement threshold and a direct response to sound. *J. Neurobiol.* **25**, 1167–1185.

TAUTZ, J. (1996). Honeybee waggle dance: recruitment success depends on the nature of the dance floor. *J. exp. Biol.* **199**, 1375–1381.

TAUTZ, J., ROHRSEITZ, K. AND SANDEMAN, D. C. (1996). The entire waggle run in the honeybee dance is only one slow-motion stride. *Nature* **382**, 32.

VON FRISCH, K. AND LINDAUER, M. (1975). Entfernungs- und Richtungsnachweis bei der Honigbiene- Rund- und Schwänzeltänze. Film C135, IWF Göttingen.

WENNER, A. (1962). Sound production during the waggle dance of the honeybee. *Anim. Behav.* **10**, 79–95.