RESEARCH ARTICLE

The recruiter's excitement – features of thoracic vibrations during the honey bee's waggle dance related to food source profitability

Michael Hrncir[1],*, Camila Maia-Silva[2], Sofia I. Mc Cabe[3] and Walter M. Farina[3]

[1]Departamento de Ciências Animais, Universidade Federal Rural do Semi-Árido, CEP: 59625-900, Mossoró-RN, Brazil, [2]Departamento de Biologia, Universidade de São Paulo, Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto, CEP: 14040-901, Ribeirão Preto-SP, Brazil and [3]Grupo de Estudio de Insectos Sociales, IFIBYNE-CONICET, Departamento de Biodiversidad y Biología Experimental, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales, C1428EHA, Buenos Aires, Argentina

*Author for correspondence (michael.hrncir@gmx.at)

SUMMARY

The honey bee's waggle dance constitutes a remarkable example of an efficient code allowing social exploitation of available feeding sites. In addition to indicating the position (distance, direction) of a food patch, both the occurrence and frequency of the dances depend on the profitability of the exploited resource (sugar concentration, solution flow rate). During the waggle dance, successful foragers generate pulsed thoracic vibrations that putatively serve as a source of different kinds of information for hive bees, who cannot visually decode dances in the darkness of the hive. In the present study, we asked whether these vibrations are a reliable estimator of the excitement of the dancer when food profitability changes in terms of both sugar concentration and solution flow rate. The probability of producing thoracic vibrations as well as several features related to their intensity during the waggle phase (pulse duration, velocity amplitude, duty cycle) increased with both these profitability variables. The number of vibratory pulses, however, was independent of sugar concentration and reward rate exploited. Thus, pulse number could indeed be used by dance followers as reliable information about food source distance, as suggested in previous studies. The variability of the dancer's thoracic vibrations in relation to changes in food profitability suggests their role as an indicator of the recruiter's motivational state. Hence, the vibrations could make an important contribution to forager reactivation and, consequently, to the organisation of collective foraging processes in honey bees.

Key words: honey bees, *Apis mellifera*, waggle dance, profitability, thoracic vibrations, communication, laser vibrometry.

INTRODUCTION

The waggle dance of the honey bee *Apis mellifera* is, without doubt, one of the best studied communication systems in the animal kingdom. The stereotyped figure-of-eight-shaped movements, performed by successful foragers on the vertical combs inside the nest (von Frisch, 1923), depend on both the distance (duration of waggle phase) and the direction (orientation of dancer's body relative to gravity during waggle phase) of the visited food patch (von Frisch, 1946; von Frisch, 1965). Naïve recruits who follow the dances are capable of deciphering the vector information provided by the dancer's movements and use it when searching for the advertised patch (Esch et al., 2001; Riley et al., 2005; Menzel et al., 2011). Decoding the waggle dance, however, poses a sensory challenge for the forager's nestmates because of the darkness inside the nest. One possible source of information used by dance followers are pulsed thoracic vibrations (frequently referred to as 'dance sounds'), produced by the foragers exclusively during the waggle phase of the dance (Fig. 1) (Esch, 1961a; Esch, 1961b; Wenner, 1962). On average, each waggle movement is accompanied by two vibratory pulses (Rohrseitz, 1998; Hrncir et al., 2006). Hence, the number of pulses increases linearly with the duration of the waggle run and, consequently, may provide information about the distance to the food source (Esch, 1961b; Wenner, 1962). In addition, wing vibrations along with the thoracic vibrations have been shown to

generate a spatially narrow jet airflow that dance followers can use to determine the movement direction of a waggling forager (Michelsen, 2003).

The position of a food patch is not the only information potentially provided by the honey bees' waggle dance. Several parameters related to the profitability of the collected food, such as sugar concentration, have been demonstrated to influence the probability at which dancing occurs as well as the 'liveliness' ('Lebhaftigkeit') of the foragers' dances (von Frisch, 1942; von Frisch, 1965; Lindauer, 1948; Seeley and Towne, 1992; Farina, 1996; Cogorno et al., 1998). In approaches to quantify the dance liveliness, it has been shown (Seeley et al., 2000; De Marco and Farina, 2001) that dancing honey bees tune the number of waggle runs per hive stay as a function of the collected sugar concentration (see also Seeley and Towne, 1992). Also, the return phase of the waggle dance (Fig. 1) becomes shorter with increasing food quality, thus resulting in more rapid dance cycles (Seeley et al., 2000). Another important parameter concerning food source profitability is the rate at which sugar is delivered to the foragers. In natural settings, bees frequently exploit flowers that produce nectar at low rates (Núñez, 1977; Vogel, 1983). This fact significantly increases the time a forager spends collecting food in the field, and bees often return to the nest with only partly filled crops (Núñez, 1966; Núñez, 1982; Varjú and Núñez, 1991; Núñez and Giurfa, 1996; Fernández et al., 2003). In
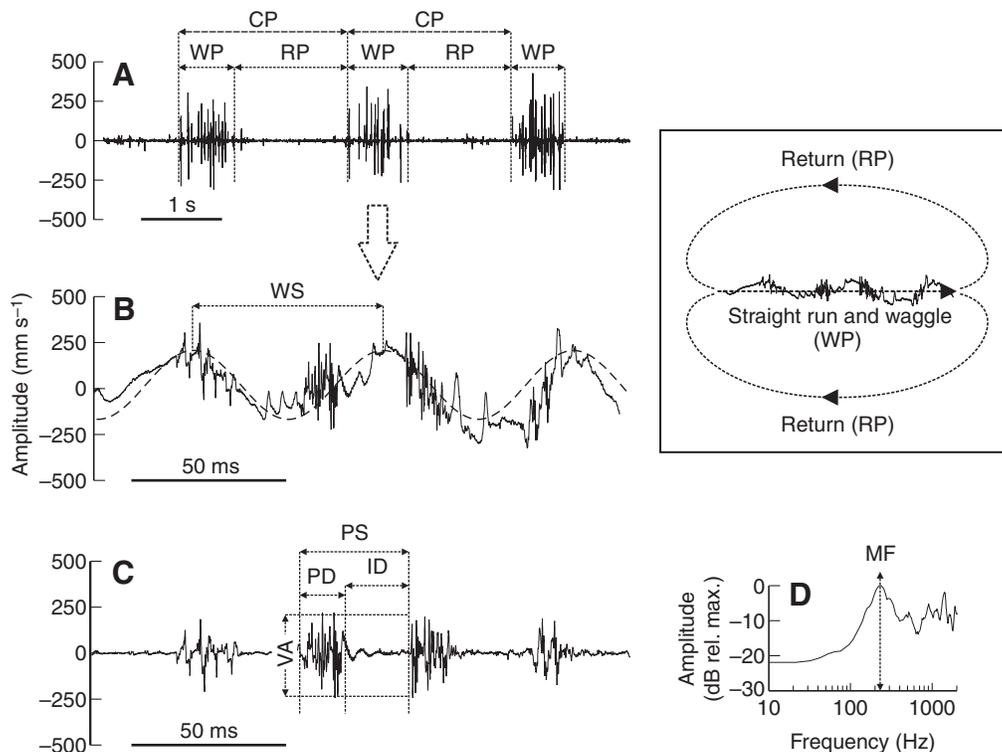
Fig. 1. The honey bees' waggle dance. (A) A dance cycle (CP, cycle phase) is divided into a waggle phase (WP) and a return phase (RP), during which the forager circles back to the starting point of the waggle run (inset). During the waggle phase, foragers often generate thoracic vibrations ('dance sounds'). Thus WP can be separated into two main mechanical components: (B) the waggle movement itself (emphasised by dashed line; WS, waggle sequence), and (C) the pulsed thoracic vibrations (frequencies <20 Hz filtered out digitally for better visualisation). PD, duration of a single pulse; VA, velocity amplitude (peak to peak, p–p) of a vibratory pulse; ID, duration of the interval between two subsequent pulses; PS, pulse sequence (PD+ID). (D) Frequency spectrum of a vibratory pulse indicating the main frequency (MF=0 dB).

line with the findings for sugar concentration, increasing reward rates have been shown to increase the probability of dancing as well as the rate of waggle run production (Cogorno et al., 1998).

Food source profitability has a strong impact on several features of the recruitment mechanisms of social insects [honey bees (Núñez, 1970; Seeley, 1986; Farina, 1996; Waddington, 2001), stingless bees (Aguilar and Briceño, 2002; Hrncir et al., 2004a; Hrncir et al., 2004b; Schmidt et al., 2006; Schmidt et al., 2008; Hrncir, 2009), ants (Roces, 1993; Cassill, 2003)]. In particular, as described above, this variable strongly influences several features of the honey bee's waggle dance (von Frisch, 1965; Cogorno et al., 1998; Seeley et al., 2000; De Marco and Farina, 2001). Hence, an important question concerning the food source communication of these animals needs to be asked: do those dance parameters that have been postulated to carry reliable information about the distance to a feeding site (duration of waggle phase, number of vibratory pulses) actually remain unperturbed by food source profitability? The present study tried to tackle this problem by asking (1) which parameters of the foragers' waggle dance (dance behaviour, thoracic vibrations during the dance) change with changing resource profitability and (2) which parameters remain constant in spite of varying food profitability and, consequently, could provide reliable information about the location of the feeding site to dance followers.

MATERIALS AND METHODS
Study site and bees

These experiments complied with the 'Principles of Animal Care', publication no. 86-23, revised 1985 by the National Institutes of Health, and with the current laws of the country in which the experiments were performed.

The experiments were performed in March 2008 and March 2009 using two queenright colonies (H1, H2) of the honey bee *Apis mellifera ligustica*, Spinola 1806, (one colony each experimental season) held at the experimental field of the University of Buenos Aires, Argentina (34°33′S; 58°26′W). Colony H1 (2008, ~3500 adult individuals) was kept in a two-frame observation hive. Colony H2 (2009, ~2000 adult individuals) was kept in a one-frame observation hive. The observation hives were set up in a flight chamber (6×3×2 m³) (see Arenas et al., 2008), which prevented potential interference from other bee colonies during the recordings. One lateral side of the flight chamber remained open so that foraging in the surrounding environment was not restricted.

Experiments

Because of low night-time temperatures in both experimental seasons (10–15°C), which accounted for a reduced flight and foraging activity of the bee colonies in the morning, experiments (training followed by trial) were performed exclusively between 10:00 h and 17:00 h. Bees who abandoned foraging in the course of a trial, which frequently happened at low sugar concentrations or low solution flow rates, were excluded from the analysis.

Training

Using the training procedure described by von Frisch (von Frisch, 1965), we trained foragers to collect unscented sucrose solution (12.5% weight on weight, w/w) at a feeding station located at a

distance of ~180 m, 60 deg west of the nest entrance. Prior to each trial, we marked one forager with non-toxic paint (Tinta Plástica; Acrilex: São Bernardo do Campo, Brazil) on her thorax while collecting at the feeding station. For the marking we used exclusively white paint to allow for an optimal reflection of the laser beam during the vibration recordings (see below). All non-marked foragers were captured with a plastic suction tube and released after the trial. In order to avoid pseudo-replications, the marked forager was killed by freezing after the trial.

Experiment 1 – sugar concentration

After marking the focal forager and capturing all unmarked individuals at the feeding site, we changed the training feeder for an alcohol-cleaned experimental food source, an inverted acrylic cup on a grooved plate that provided food *ad libitum*. As food we offered unscented sucrose solutions containing 12.5%, 25% or 50% sucrose w/w. In total, 15 foragers (N=7 in 2008; N=8 in 2009) participated in this experimental series. Eight bees (N=4 in both experimental seasons) collected sucrose solution in a sequence of increasing concentration (12.5–25–50% w/w), and seven bees (N=3 in 2008; N=4 in 2009) collected sucrose solution in a sequence of decreasing concentration (50–25–12.5% w/w). Each sugar concentration was offered for at least 1 h, during which we observed the probability at which returning foragers performed waggle dances in the nest, and recorded their thoracic vibrations during the waggle dances (see below). Recruits arriving at the feeding site during a trial were captured with a suction tube and released after the trial.

Experiment 2 – solution flow rate

In this experimental series, the artificial food source consisted of a feeder (henceforth, rate feeder) that provided unscented sucrose solution (50% w/w) at different flow rates [described in detail by Núñez (Núñez, 1970; Núñez, 1971; Núñez, 1982)]: a synchromotor activated a pumping system connected to a syringe filled with sucrose solution. By using different synchromotors, the pumping velocity could be adjusted, thus supplying the solution at different rates. The sucrose solution was delivered through a fine polyethylene tube to a lacuna in an acrylic feeding platform. Prior to initiating the trial, the marked foragers, who had been trained to collect at an *ad libitum* feeder, had to learn to imbibe the sucrose solution from the lacuna in the feeding platform. During this extra training, which took half an hour on average, the lacuna was constantly filled with 50% w/w sucrose solution in order to maintain the motivation of the forager to collect at the food source. After this training phase, the feeder pump was turned on only when the marked forager landed on the feeding platform and turned off immediately when she left for the hive. The following solution flow rates were used: in experimental season 2008: 4.5, 9.0 and 17.5 µl min^{-1} (N=12 foragers; 6 foragers increasing series, 6 foragers decreasing series); in experimental season 2009: 2.5, 5.5, 12.0 and 17.5 µl min^{-1} (N=10 foragers; 5 foragers increasing series, 5 foragers decreasing series). Depending on the success of recording the foragers' vibrations in the nest, each reward rate was offered during four to seven foraging trips. At the feeding site, we recorded the visit time of the bees, defined as the time spent by an individual at the rate feeder between her arrival and departure for the nest, and estimated her crop load through multiplying the visit time by the solution flow rate (Núñez, 1966; Fernández et al., 2003). At the hive, we observed the probability at which returning foragers performed waggle dances, and recorded their thoracic vibrations during the waggle dances (see below). Recruits arriving at the feeding site during a trial were captured with a suction tube and released after the trial.

Laser vibrometry and signal analysis

The thoracic vibrations produced by the honey bee foragers during the waggle dances were recorded as velocities using a contactless portable laser–Doppler vibrometer (PDV100; Polytec: Waldbronn, Germany). Recording the vibrations directly on the thorax of the dancers avoided interference by background noise such as that produced by ventilating nestmates or other vibrating bees (Hrncir et al., 2004a; Hrncir et al., 2004b). The use of laser vibrometry kept the signal-to-noise ratio very high, which allowed a much more precise measurement of the temporal parameters contained in the signals than is possible when recording airborne sounds (Hrncir et al., 2004a).

In experimental season 2008, the observation hive (H1) was set up in its normal, vertical position (von Frisch, 1965). The laser vibrometer was mounted on a tripod positioned lateral to the hive, and the foragers' movements could be followed with the laser beam by turning the head of the tripod. It was, however, difficult to keep the laser beam focused on the waggle dancers because of the constantly and rapidly changing distance between the bees and the lens of the vibrometer. Consequently, the vibration recordings were restrained to a rather small area of the dance floor. To facilitate recording, in experimental season 2009 we positioned the observation hive (H2) horizontally. On horizontal combs and under diffuse or low light conditions, as was the case within the flight chambers used in the present study, honey bees have been shown to perform dances in random direction and with shorter waggle phases compared with oriented dances (Esch, 1964). As our study did not investigate the vector information coded in the waggle dances, we accepted the disorientation of the foragers as trade-off for better vibration recordings. The laser vibrometer was mounted on a four-wheeled cart, and a mirror directed the laser beam onto the dancing bee (Hrncir et al., 2004a). With this setup, movements of the forager could be followed easily and without losing focus by moving the cart along with the dancer.

The laser vibrometer was connected to a portable microcomputer (Toshiba Satellite M65, 2.0 GHz) *via* a custom-made voltage reducer (Insight Equipamentos: Ribeirão Preto, Brazil) and a stereo-soundcard (PSC 805; Philips: Amsterdam, The Netherlands). The signals were recorded on the computer's hard disk using the software Soundforge 7.0 (Sony Digital Pictures Inc., Madison, WI, USA) at a sampling rate of 44,100 Hz and 16 bits. Signal analysis was performed using the software SpectraPro 3.32 (Sound Technology Inc., Campbell, CA, USA), Microsoft® Office Excel® 2007 and SigmaPlot 10.0 (Systat Software Inc.: Point Richmond, CA, USA). Fast Fourier transformation analysis (FFT analysis, SpectraPro 3.32) was performed at a frequency resolution of 10 Hz.

The following parameters of the pulsed thorax vibrations produced during the waggle dances were analysed: PD, duration of a pulse; VA, velocity amplitude of a pulse; MF, dominant frequency of a pulse (peak frequency in the spectrogram); ID, duration of the interval without vibration between two subsequent pulses; PR, pulse rate (number of pulses per second, 1/PS; PS=PD+ID); and DC, duty cycle (PD×PR) (Fig. 1). In addition to the pulsed thoracic vibrations generated during the dances, the thorax vibrates along with the waggle movements (Esch, 1961b) (Fig. 1), which allowed us to analyse the waggle frequency (WF) of the foragers' dances as well as the duration of the waggle phase (WP) and of the return phase (RP). From these last two measurements we determined the duration of the dance cycles (cycle phase, CP=WP+RP). Because the foragers frequently changed the face of the comb during their stay in the hive, we were not able to reliably count the number of dance circuits per foraging trip, another important feature of the honey bees' dance behaviour related to food

profitability (Raveret-Richter and Waddington, 1993; Cogorno et al., 1998; Seeley et al., 2000; De Marco and Farina, 2001).

Statistics

The nest-internal behaviour of every single forager was recorded during four to seven returns to the nest at each sugar concentration (experiment 1) or solution flow rate (experiment 2). For each bee we calculated the probability of producing waggle dances (dance probability, PrD=hive stays with waggle dances×100/number of observed hive stays), and the probability of generating thoracic vibrations during the waggle dance (vibration probability, PrV=waggle dances with vibrations×100/number of recorded waggle dances). For each sugar concentration or solution flow rate, we calculated the mean value of each analysed signal parameter from between 18 and 241 pulses (mean: 125 pulses; total: 12,376 pulses) produced during 2–13 waggle dances (mean: 7 waggle phases; total: 737 waggle phases) for each individual (N=37). Likewise, we calculated the mean feeding times and crop loads for each forager in experiment 2. The statistical analysis was performed with these representative mean values to avoid different weightings in the analysis due to different sample sizes (Hrncir et al., 2004a). We investigated potential differences in the signal pattern and the feeding times between experimental steps using two-way repeated measures ANOVA (Tukey's *post hoc* test), considering food quality (sugar concentration or reward rate), sequence (increasing sugar concentration/solution flow; decreasing sugar concentration/solution flow) and the interaction between them (quality×sequence) as possible sources of variation. Student's *t*-test was used to assess potential behavioural differences between the experimental seasons (feeding behaviour and nest-internal behaviour at a solution flow of 17.5 μl min^{-1}). Statistical analysis was performed using the software SigmaStat 3.5 (Systat Software Inc.). The level of significance for differences was taken as $P \leq 0.05$ (experiment 1) and as $P_C \leq 0.025$ (experiment 2, correction of α-level because of multiple use of data sets) (Zar, 1999). Throughout the text, values are presented as means ± s.e.m. N refers to the number of bees, n to the number of measured pulses.

RESULTS

We analysed different mechanical features of the waggle dances of honey bee foragers collecting sucrose solutions of different sugar concentration (experiment 1) or offered at different reward rates (experiment 2). During the entire study, the feeding site visited by the focal bees was located 180 m and 60 deg west of the nest. Consequently, any variation in the foragers' signals during the respective trials had to be due to factors other than food source location.

Effect of sugar concentration on the behaviour of waggle dancers

Because some foragers in experiment 1 did not produce waggle dances at the lowest sugar concentration (12.5% w/w; 2 bees out of 7 in 2008, 3 bees out of 8 in 2009), which critically diminished the sample size, we pooled the data from the two seasons for the statistical analysis (Table 1). We observed a significant variation in several features of the bees' behaviour within the nest during the trials (Figs 2, 3). This variation could be attributed to the sugar concentration offered at the feeder, and was independent of the sequence in which we offered the sugar concentrations at the food source (increasing series, decreasing series) (Table 1). In nine out of 12 studied mechanical features of the waggle dance (PrD; CP; RP; PrV, PD, VA, MF, ID, DC), we observed statistically significant differences ($P \leq 0.05$) between the lowest (12.5% w/w) and highest (50% w/w) sugar concentration offered (Figs 2, 3; Table 1). PrD, PrV, PD, VA, MF and DC increased, whereas CP, RP and ID decreased with increasing sugar concentration (Figs 2, 3) (Table 1). WP, PR and WF were not affected by sugar concentration (Figs 2,3; Table 1).

Food uptake at different solution flow rates

In addition to the sugar concentration, an important parameter determining the overall quality of a food source is the solution flow rate because it influences both the time a collecting bee spends at the patch and the forager's crop load. In both experimental seasons (2008, 2009), the observed variation of the time bees spent at the feeders could be attributed to the solution flow rate, and was

Table 1. Experiment 1 – sugar concentration

| | Sugar concentration (w/w) | | | Source of variation (two way RM ANOVA) | | | | | | |
| | | | | Concentration | | Sequence | | Concentration × sequence | |
	12.5%	25%	50%	F	P	F	P	F	P
PrD (%)	33.6±7.5[a]	78.6±4.7[b]	96.7±1.8[b]	**31.5**	**<0.001**	0.3	0.624	0.0	0.970
CP (s)	2.41±0.07[a]	2.19±0.06[a]	1.89±0.07[b]	**13.4**	**<0.001**	0.0	0.834	1.1	0.349
WP (s)	0.52±0.01[a]	0.52±0.01[a]	0.53±0.01[a]	0.5	0.624	0.2	0.706	1.7	0.201
RP (s)	1.89±0.07[a]	1.67±0.06[b]	1.36±0.07[c]	**14.6**	**<0.001**	0.0	0.907	1.1	0.364
PrV (%)	57.4±7.3[a]	87.4±5.8[b]	100.0±0.0[b]	**10.5**	**0.001**	0.8	0.383	0.1	0.879
PD (s)	0.013±0.001[a]	0.014±0.001[a]	0.017±0.001[b]	**7.6**	**0.005**	2.7	0.122	1.1	0.366
VA (mm s^{-1})	272.8±15.2[a]	340.6±11.5[b]	395.0±17.2[b]	**10.9**	**0.001**	0.1	0.761	0.6	0.572
MF (Hz)	213.4±2.5[a]	225.0±3.4[a]	235.7±3.5[b]	**21.3**	**<0.001**	0.4	0.558	0.1	0.888
ID (s)	0.025±0.002[a]	0.022±0.002[a,b]	0.019±0.001[b]	**8.5**	**0.003**	0.6	0.457	1.7	0.223
DC	0.38±0.02[a]	0.43±0.02[b]	0.51±0.03[c]	**24.3**	**<0.001**	0.1	0.717	0.1	0.922
PR (Hz)	27.8±1.5[a]	30.5±1.8[a]	29.1±0.8[a]	1.12	0.349	2.3	0.148	1.5	0.254
WF (Hz)	14.2±0.3[a]	14.5±0.2[a]	14.7±0.2[a]	1.5	0.252	0.8	0.389	0.9	0.427

Foragers collected sucrose solutions of varying sugar concentration offered *ad libitum* in either increasing or decreasing sequence. Mean values ± s.e.m. of 15 individuals. The parameters (dance behaviour, thoracic vibrations) were statistically analysed using two way repeated measures ANOVA, considering sugar concentration, sequence and the interaction between them as possible sources of variation (*F*- and *P*-values). Statistically significant test results ($P \leq 0.05$) are emphasised in bold type. Same superscript letters (a–c) indicate statistically inseparable data sets (Tukey's *post hoc* test, $P > 0.05$).

PrD, probability to perform dances; CP, cycle phase duration; WP, waggle phase duration; RP, return phase duration; PrV, probability of producing thoracic vibrations; PD, pulse duration; VA, velocity amplitude; MF, main frequency; ID, interval duration; DC, duty cycle; PR, pulse rate; WF, waggle frequency (see Fig. 1).
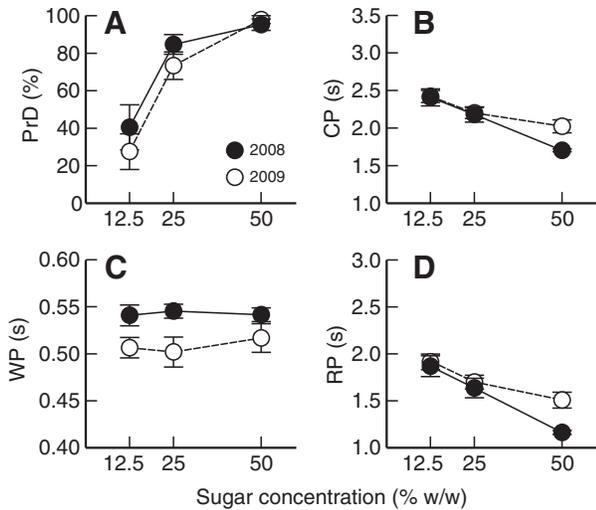
Fig. 2. The effect of sugar concentration on the honey bee's waggle dance. We analysed the dance probability (PrD; A), as well as the duration of the cycle phase (CP; B), the waggle phase (WP; C), and the return phase (RP; D) of the waggle dances of foragers collecting unscented sucrose solution of varying sugar concentration (12.5–25–50% w/w) offered *ad libitum* at an artificial food source (180 m, 60 deg west of the hive). Graphs show mean values ± s.e.m. of 7 foragers (2008; filled circles) and of 8 foragers (2009; open circles). Note differences in waggle phase duration between 2008 and 2009. For statistics (pooled data of the two experimental seasons), see Table 1.

independent of the sequence in which we provided different flow rates at the food source (increasing series, decreasing series) (Tables 2, 3). At higher flow rates, the foragers spent less time at the feeder, yet achieved a higher final crop loads (Fig. 4; Tables 2, 3). In both experimental seasons, the highest solution flow rate used was 17.5 μl min^{-1}. At this reward rate, foragers in 2008 imbibed

sugar solution significantly longer than foragers in 2009 ($\alpha_C \leq 0.025$; t-tests: $t = -3.7$, $P < 0.001$, d.f. = 20; Fig. 4).

Effect of solution flow rate on the behaviour of waggle dancers

In line with our findings in Experiment 1 (sugar concentration), we observed a significant variation ($P_C \leq 0.025$) of several features of their nest-internal behaviour when honey bee foragers collected at feeders that offered sucrose solution (50% w/w) at different flow rates (Figs 5, 6). In all but one case, this variation could be attributed to the solution flow rate and was independent of the sequence in which we changed the reward rates at the feeder (increasing series, decreasing series) (Tables 2, 3). In experimental season 2009, the observed variation in ID was in part due to the sequence in which we changed the solution flow rate (Table 3).

In eight out of the 12 studied mechanical features of the waggle dance (PrD, CP, RP, PrV, PD, VA, ID, DC), we measured statistically significant differences ($P_C \leq 0.025$) between low and high solution flow rates, but not between high flow rates (2008: no differences between 9.0 μl min^{-1} and 17.5 μl min^{-1}; 2009: no differences between 12.0 μl min^{-1} and 17.5 μl min^{-1}) (Figs 5, 6; Tables 2, 3). In agreement with our findings in Experiment 1, PrD, PrV, PD, VA and DC increased, whereas CP, RP and ID decreased with increasing food profitability (Figs 5, 6; Tables 2, 3). WP, MF, PR and WF were not significantly influenced by the solution flow rate offered at the feeder (Figs 5, 6; Tables 2, 3). In 2008, the foragers' dances were characterised by significantly longer WP, shorter RP and shorter CP than those observed in 2009 (reward rate 17.5 μl min^{-1}: t-tests: $t_{WP} = -4.6$, $P < 0.001$; $t_{RP} = -3.3$, $P = 0.004$; $t_{CP} = 2.9$, $P = 0.010$, d.f. = 18; Fig. 5). Also, foragers in 2008 generated significantly longer pulses (PD) at a higher PR and, consequently, shorter intervals between subsequent pulses (ID) and a higher DC than bees in 2009 (reward rate 17.5 μl min^{-1}: t-tests: $t_{PD} = 2.4$, $P = 0.024$; $t_{ID} = -5.9$, $P < 0.001$; $t_{PR} = 4.4$, $P < 0.001$; $t_{DC} = 5.7$, $P < 0.001$, d.f. = 20; Fig. 6). PrD, PrV, VA and WF did not differ significantly
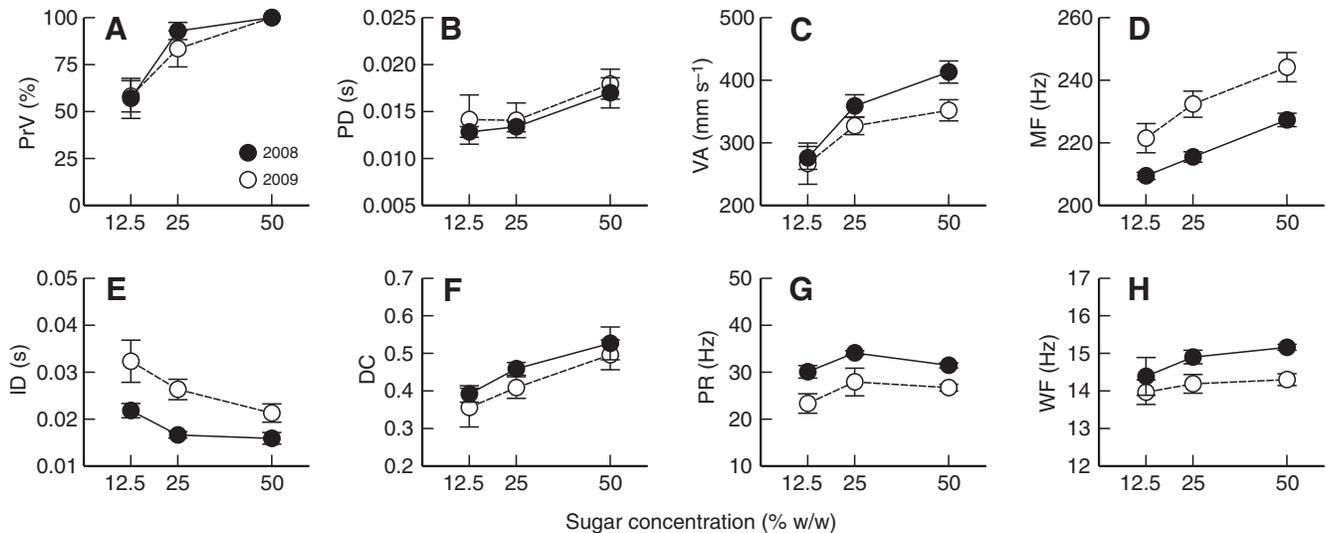

Fig. 3. The effect of sugar concentration on the honey bee's dance sounds. Foragers collected unscented sucrose solution of varying sugar concentration (12.5–25–50% w/w) offered *ad libitum* at an artificial food source (180 m, 60 deg west of the hive). We analysed the probability of producing thoracic vibrations during the waggle dance (PrV; A), as well as the duration of vibratory pulses (PD; B), their velocity amplitude (VA p–p; C) and main frequency component (MF; D), the duration of the intervals between two subsequent pulses (ID; E), the duty cycle (DC, proportion of the waggle phase in which foragers vibrate their thorax; F), the pulse rate (PR, vibratory pulses per second; G), and the frequency of the waggle movements (WF; H). Graphs show mean values ± s.e.m. of 7 foragers (2008; filled circles) and of 8 foragers (2009; open circles). For statistics (pooled data of the two experimental seasons), see Table 1.

Table 2. Experiment 2 – solution flow rate, experimental season 2008

| | Solution flow rate ($\mu l\,min^{-1}$) | | | Source of variation (two way RM ANOVA) | | | | | |
| | | | | Flow | | Sequence | | Flow × sequence | |
	4.5	9.0	17.5	F	P	F	P	F	P
Time (min)	10.6±0.8[a]	6.7±0.5[b]	4.0±0.4[c]	**72.7**	**<0.001**	0.2	0.703	0.3	0.758
Load (µl)	47.7	60.6	70.5	–	–	–	–	–	–
PrD (%)	47.2±11.8[a]	80.7±8.4[b]	91.9±5.6[b]	**8.5**	**0.002**	0.0	0.894	0.7	0.491
CP (s)	2.40±0.08[a]	1.83±0.06[b]	1.80±0.05[b]	**37.9**	**<0.001**	0.0	0.955	0.2	0.786
WP (s)	0.54±0.01	0.54±0.01	0.55±0.01	0.1	0.899	0.8	0.399	0.9	0.415
RP (s)	1.86±0.08[a]	1.29±0.06[b]	1.25±0.05[b]	**37.6**	**<0.001**	0.0	0.976	0.4	0.673
PrV (%)	77.9±3.6[a]	91.8±3.2[b]	97.8±1.6[b]	**16.8**	**<0.001**	0.3	0.621	0.7	0.510
PD (s)	0.015±0.001[a]	0.018±0.001[b]	0.019±0.001[b]	**9.2**	**0.001**	0.3	0.621	0.6	0.581
VA ($mm\,s^{-1}$)	301.6±14.5[a]	367.2±16.6[b]	390.4±15.7[b]	**13.0**	**<0.001**	0.0	0.997	0.0	0.998
MF (Hz)	229.9±7.0	234.9±4.7	236.0±5.1	2.3	0.123	0.1	0.821	0.4	0.677
ID (s)	0.019±0.001[a]	0.015±0.001[b]	0.015±0.001[b]	**14.8**	**<0.001**	0.7	0.436	0.8	0.473
DC	0.45±0.02[a]	0.55±0.02[b]	0.57±0.01[b]	**19.2**	**<0.001**	0.5	0.476	1.0	0.389
PR (Hz)	30.7±0.7	30.6±0.7	30.6±0.6	0.0	0.997	0.1	0.790	0.0	0.953
WF (Hz)	14.5±0.2	14.7±0.1	14.6±0.2	0.0	0.966	0.0	0.921	0.4	0.706

Foragers collected 50% w/w sucrose solution offered at varying solution flow rates in either increasing or decreasing sequence. Means ± s.e.m. of 12 individuals. The parameters (dance behaviour, dance sounds) were statistically analysed using two way repeated measures ANOVA, considering flow rate, sequence and the interaction between them as possible sources of variation (F- and P-values). Statistically significant test results ($P \le 0.05$) are emphasised in bold type. Same superscript letters (a–c) indicate statistically inseparable data sets (Tukey's *post hoc* test, P>0.05).
Abbreviations of the waggle dance parameters same as in Table 1 (see also Fig. 1). Time, visit time; Load, estimated mean crop load.

between the experimental seasons (reward rate $17.5\,\mu l\,min^{-1}$: *t*-tests: $t_{PrD}=-0.03$, $P=0.971$; $t_{PrV}=0.6$, $P=0.558$; $t_{VA}=1.6$, $P=0.133$; $t_{WF}=0.3$, $P=0.792$, d.f.=20; Fig. 6).

DISCUSSION

The honey bee's waggle dance is certainly one of the most intriguing examples of communication in the animal kingdom. In a recent publication, Grüter and Farina proposed that the waggle dance should be considered a multicomponent signal as it comprises more than one information component (Grüter and Farina, 2009): the dance (1) attracts surrounding hive bees, both food processors and unemployed collectors, to the active forager; (2) reactivates private navigational information in inactive foragers; (3) provides information about the availability of a profitable food source; (4)

indicates the position (distance, direction) of a food patch; and (5) provides chemosensory information about the visited food source (Grüter et al., 2008; Grüter and Farina, 2009). All of these components, which can be either redundant (i.e. lead to the same or an enhanced response) or not (i.e. provide multiple messages, each leading to a particular response) (Rowe, 1999), contribute to improving the efficiency of collective foraging in honey bees.

In our study, several features of the thoracic vibrations generated by the foragers during their waggle dances varied according to changes in sugar concentration and solution flow rate encountered by the bees at the feeding site (PrV, PD, VA, ID, DC; Figs 3, 6; Tables 1–3). Those features of the thoracic vibrations, however, that putatively carry information about the distance of the visited food patch remained constant despite changes in food profitability (pulse

Table 3. Experiment 2 – solution flow rate, experimental season 2009

| | Solution flow rate ($\mu l\,min^{-1}$) | | | | Source of variation (two way RM ANOVA) | | | | | |
| | | | | | Flow | | Sequence | | Flow × sequence | |
	2.5	5.5	12.0	17.5	F	P	F	P	F	P
Time (min)	11.1±0.7[d]	6.9±0.5[e]	3.6±0.3[f]	2.3±0.4[f]	**81.9**	**<0.001**	0.5	0.506	0.2	0.923
Load (µl)	27.6	37.6	42.9	39.3	–	–	–	–	–	–
PrD (%)	26.7±9.4[d]	76.7±10.0[e]	96.7±3.3[f]	92.2±4.1[f]	**40.0**	**<0.001**	3.8	0.087	2.7	0.067
CP (s)	2.88±0.24[d]	2.44±0.08[e]	2.05±0.06[f]	2.04±0.07[f]	**18.0**	**<0.001**	0.9	0.358	0.5	0.687
WP (s)	0.49±0.01	0.50±0.01	0.50±0.01	0.50±0.01	1.7	0.209	1.0	0.337	0.1	0.975
RP (s)	2.39±0.24[d]	1.95±0.08[e]	1.55±0.07[f]	1.54±0.07[f]	**18.0**	**<0.001**	1.1	0.320	0.5	0.692
PrV (%)	45.6±5.1[d]	70.9±5.3[e]	94.5±2.5[f]	95.8±3.2[f]	**22.8**	**<0.001**	3.7	0.079	1.3	0.303
PD (s)	0.012±0.001[d]	0.014±0.001[d]	0.016±0.001[e]	0.016±0.001[e]	**11.6**	**<0.001**	2.0	0.190	1.3	0.310
VA ($mm\,s^{-1}$)	267.2±16.1[d]	316.6±13.2[e]	348.0±11.8[f]	357.0±13.4[f]	**34.2**	**<0.001**	1.1	0.317	2.0	0.160
MF (Hz)	231.3±9.9	239.2±7.2	244.9±4.0	246.8±6.6	2.9	0.065	3.1	0.112	0.6	0.655
ID (s)	0.035±0.007[d]	0.026±0.001[e]	0.023±0.001[e]	0.024±0.002[e]	**9.8**	**<0.001**	**24.2**	**<0.001**	**3.6**	**0.038**
DC	0.33±0.03[d]	0.40±0.03[d]	0.46±0.01[e]	0.46±0.02[e]	**10.0**	**<0.001**	3.7	0.080	0.5	0.680
PR (Hz)	24.8±0.4	25.9±0.7	27.0±1.0	25.8±1.0	1.5	0.254	1.1	0.313	0.6	0.597
WF (Hz)	14.7±0.3	14.7±0.5	14.5±0.1	14.5±0.2	0.4	0.787	0.0	0.996	1.9	0.166

Foragers collected 50% w/w sucrose solution offered at varying solution flow rates in either increasing or decreasing sequence. Mean values ± s.e.m. of 10 individuals. The parameters (dance behaviour, dance sounds) were statistically analysed using two way repeated measures ANOVA, considering flow rate, sequence and the interaction between them as possible sources of variation (F- and P-values). Statistically significant test results ($P \le 0.05$) are emphasised in bold type. Same superscript letters (d–f) indicate statistically inseparable data sets (Tukey's *post hoc* test, P>0.05).
Abbreviations of the waggle dance parameters same as in Table 1 (see also Fig. 1). Time, visit time; Load, estimated mean crop load.
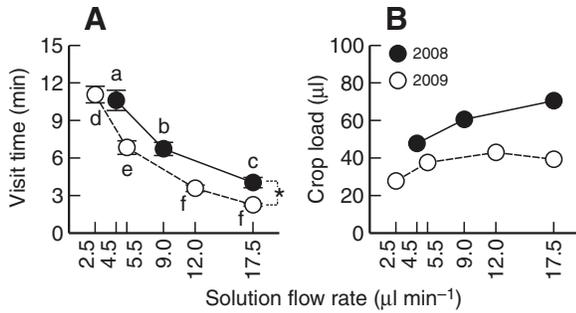
Fig. 4. Food uptake at different solution flow rates. Foragers collected unscented sucrose solution (50% w/w) offered at different solution flow rates at a rate feeder (180 m 60 deg west of the hive) (2008: N=12, filled circles; 2009: N=10, open circles). From the visit times (time between arrival at the rate feeder and departure for the nest; A), we estimated the crop load of each forager (B) through multiplying the visit time by the solution flow rate (Núñez, 1966; Fernández et al., 2003). Mean values ± s.e.m. of visit times (A) and mean values only of the estimated crop loads (B) are shown. Visit times were statistically analysed using two-way repeated measures ANOVA. Same superscript letters (2008: a–c; 2009: d–f) indicate statistically inseparable data sets (Tukey's *post hoc* test, $P>0.05$). For details see Tables 2 and 3. Asterisk in A indicates significant differences (*t*-test: $P≤0.05$; see Results for details) concerning visit times between foragers collecting at a reward rate of 17.5 μl min^{-1} in 2008 and 2009.
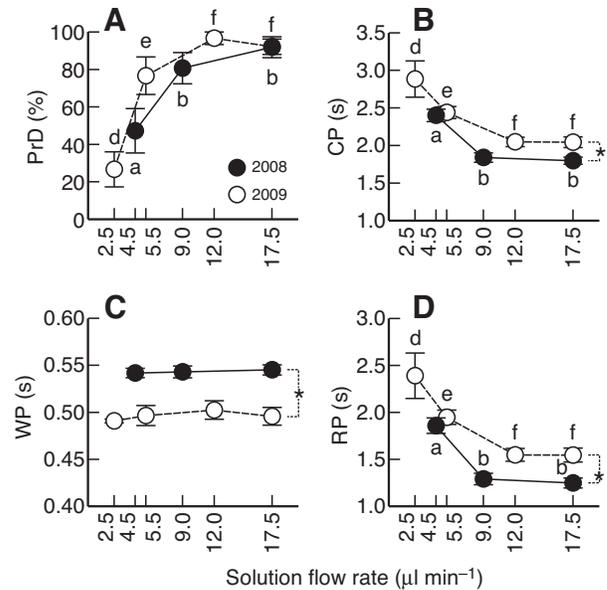

Fig. 5. The effect of solution flow rate on the honey bee's waggle dance. We analysed the dance probability (PrD; A), as well as the duration of the cycle phase (CP; B), the waggle phase (WP; C), and the return phase (RP; D) of the waggle dances of foragers collecting unscented sucrose solution (50% w/w) offered at different solution flow rates at a rate feeder (180 m 60 deg west of the hive). Graphs show mean values ± s.e.m. of 12 foragers (2008; filled circles) and of 10 foragers (2009; open circles). Parameters were statistically analysed using two way repeated measures ANOVA. Same superscript letters (2008: a–c; 2009: d–f) indicate statistically inseparable data sets (Tukey's *post hoc* test, $P>0.05$). For details, see Tables 2 and 3. Asterisks indicate significant differences (*t*-tests: $P≤0.05$; see Results for details) between foragers collecting at a reward rate of 17.5 μl min^{-1} in 2008 and in 2009. Note differences in WP duration between 2008 and 2009.

rate, waggle frequency; Figs 3, 6; Tables 1–3). The thoracic vibrations, therefore, could be a source of multiple information for hive bees, who cannot visually decode the movements of the dancer because of the darkness on the dance floor (Towne, 1985; Dyer, 2002; Hrncir et al., 2006).

The recruiters' excitement – enhancing the attractiveness of the dancer

With increasing profitability of a food source (sugar concentration, solution flow rate), the excitement of foragers rises, in that their dances become more frequent and rapid (von Frisch, 1965; Waddington, 1982; Farina, 1996; Waddington, 2001; Cogorno et al., 1998; Seeley et al., 2000) (Figs 2, 5). This increase in signal occurrence enhances both the redundancy of potential messages and the attractiveness of the dancers. In our study, the probability of producing thoracic vibrations during the waggle phase increased with food profitability [Figs 3, 6; for similar findings on round dances see Waddington and Kirchner (Waddington and Kirchner, 1992)]. These vibrations have been postulated to account for the recruitment success of foragers (Esch, 1962; Dyer, 2002), predominantly because they attract surrounding bees towards the dancer (Tautz, 1996; Tautz and Rohrseitz, 1998; Hasegawa and Ikeno, 2011) so that these potential recruits can receive other information encoded in the waggle dance (Grüter and Farina, 2009).

Not only did the occurrence of the signal increase with increasing food source profitability but also its intensity increased (Figs 3, 6; Tables 1–3). Foragers generated thoracic vibrations of significantly higher velocity amplitude when collecting at high-profit food sources than when collecting at low-profit food sources (behavioural threshold low-profit–high-profit food source in this study: experiment 1: concentration ≥25% sugar w/w; experiment 2: solution flow ≥9 μl min^{-1}; Figs 3, 6; Tables 1–3). Thoracic vibrations of the dancers are transmitted to potential receivers either through substrate vibrations (Autrum and Schneider, 1948; Sandeman et al., 1996; Kilpinen and Storm, 1997; Tautz and Rohrseitz, 1998; Nieh

and Tautz, 2000) or through air particle movements (Heran, 1959; Michelsen et al., 1987; Michelsen, 2003; Tsujiuchi et al., 2007; Hasegawa and Ikeno, 2011). In both cases, the strength of attraction over distance depends on the strength of the stimulus vibration (Morse, 1981; Hrncir et al., 2008). Thus, the observed increase in both amplitude and duty cycle of the thoracic vibrations (proportion of the waggle phase in which the bees vibrate their thorax) may enhance the attractiveness of the dancer with increasing food source profitability. Consequently, more bees would congregate around the waggle dancer, thus facilitating the transmission of information and increasing her recruitment success [see previous studies on increasing recruitment success with increasing sugar concentration (von Frisch, 1965; Seeley, 1986; Seeley and Towne, 1992; Fernández and Farina, 2002) or solution flow rate (Cogorno et al., 1998; Fernández et al., 2003)].

The recruiters' excitement – reliable distance information in the waggle dance

Once attracted to a dancer, hive bees may obtain a variety of messages from the waggle dance (Grüter and Farina, 2009), such as information about the position of the food patch visited by the forager. The duration of the waggle phase is highly correlated with the distance to a food source (von Frisch, 1965; Dyer, 2002), and, importantly for providing accurate spatial information, is not influenced by food profitability (Figs 2, 5; Tables 1–3) (Seeley et al., 2000). The putative carriers of distance information are the thoracic vibrations of the waggle dancers (Esch, 1961b; Wenner,
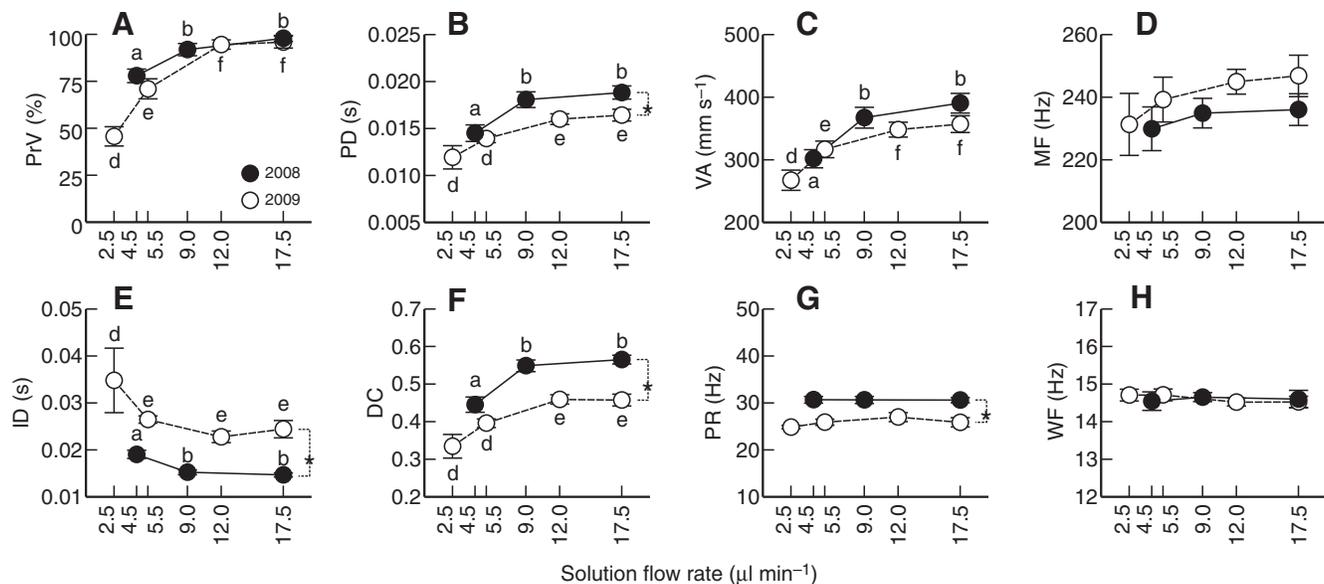
Fig. 6. The effect of solution flow rate on the honey bee's dance sounds. Foragers collected unscented sucrose solution (50% w/w) offered at different solution flow rates at a rate feeder (180m, 60 deg west of the hive). We analysed the probability of producing thoracic vibrations during the waggle dance (PrV; A), as well as the duration of vibratory pulses (PD; B), their velocity amplitude (VA, p–p; C) and main frequency component (MF; D), the duration of the intervals between two subsequent pulses (ID; E), the duty cycle (DC, proportion of the waggle phase in which foragers vibrate their thorax; F), the pulse rate (PR; vibratory pulses per second; G), and the frequency of the waggle movements (WF; H). Graphs show mean values ± s.e.m. of 12 foragers (2008; filled circles) and of 10 foragers (2009; open circles). Parameters were statistically analysed using two way repeated measures ANOVA. Same superscript letters (2008: a–c; 2009: d–f) indicate statistically inseparable data sets (Tukey's *post hoc* test, $P>0.05$). For details, see Tables 2 and 3. Asterisks indicate significant differences (*t*-tests: $P \le 0.05$; see Results for details) between foragers collecting at a reward rate of 17.5 μl min^{-1} in 2008 and in 2009.

1962). Independently of food source distance, a dancer produces on average two vibratory pulses per waggle movement (Esch, 1961b; Wenner, 1962; Rohrseitz, 1998; Hrncir et al., 2006) (see also Tables 1–3: pulse rate ~30 Hz at a waggle frequency of ~15 Hz). In our study, the waggle phase duration (WP), the pulse rate (PR) and, consequently, the number of vibratory pulses during the waggle phase (PR×WP) were not significantly influenced by sugar concentration or reward rate (Figs 3, 6; Tables 1–3). Thus, given the pulse rate, apparently, is not significantly influenced by food source profitability (sugar concentration and solution flow rate), the number of vibratory pulses linearly increases with the duration of the waggle phase. Consequently, the number of pulses could indeed be used by dance followers as reliable information about food source distance, as suggested in previous studies (Esch, 1961b; Wenner, 1962).

In both experimental seasons, the observation hive and the feeding station were located at exactly the same position and, consequently, the distance between hive and food source and also the flight route were the same during our entire study. Even so, the duration of WP differed significantly between the years 2008 and 2009 (2008: 0.54–0.55 s; 2009: 0.49–0.51 s; Figs 2, 5). As the visual environment met by the bees when collecting at our feeding site was, in principle, the same in the two experimental seasons, these observed differences in WP cannot be attributed to differences in the optic flow experienced by the foragers during flight (Esch et al., 2001; Tautz et al., 2004). A possible explanation, however, relates to differences in our experimental setup: in 2008, the observation hive was oriented vertically (normal position), whereas it was oriented horizontally in 2009 in order to improve vibration recordings. Under these latter circumstances, honey bees are known to perform disoriented waggle dances with significantly shorter waggle phases (Esch, 1964). Very similar to our observations, Esch recorded waggle phase durations

of 0.54 s in vertically oriented hives and of 0.50 s in horizontally oriented hives when the food source was located at a distance of 170 m from the nest (Esch, 1964). Our experimental disturbance of the bee hives might also be responsible for the significant differences between the experimental seasons concerning several parameters of the thoracic vibrations (Fig. 6). Most importantly, regarding distance communication, the pulse rate decreased from ~31 Hz in 2008 to ~25–27 Hz in 2009 (compare Tables 2 and 3). Consequently, the number of vibratory pulses dropped from ~16–17 to 12–13 (calculated values from WP×PR). If dance followers used pulse number as measure of food source distance, the disoriented waggle dances on vertical combs in fact would provide inaccurate information about both the direction and the distance of a patch.

The recruiters' excitement – reflecting the motivational state of foragers

Most of the studied mechanical features of the waggle dance varied considerably with both sugar concentration and solution flow rate (Figs 2, 3, 5, 6; Tables 1–3). In particular, we observed significant differences in the recruiters' excitement between low-profit and high-profit food sources (behavioural threshold in our study: concentration $\ge 25\%$ sugar w/w; solution flow $\ge 9\,\mu$l min^{-1}; Figs 3, 6; Tables 1–3), but not among low-profit sources (solution flows: 2.5 and 5.5 μl min^{-1}; Fig. 6; Table 3), or high-profit sources (sugar concentrations: 25% and 50% w/w; solution flows: 9.0 and 17.5 μl min^{-1}; 12.0 and 17.5 μl min^{-1}; Figs 3, 6; Tables 1–3). These findings suggest that the waggle dances, and, more importantly, the thoracic vibrations as potential information carriers, indicate the motivational state of a dancing forager rather than signalling the exact profitability of a food source.

The motivational state of a honey bee forager depends on her subjective evaluation of a food source (Núñez and Giurfa, 1996;

Waddington, 1985; Waddington, 2001; Chittka, 2004). In addition to profitability, parameters influencing this subjective evaluation are the nutritional state of the colony (Lindauer, 1948; Seeley, 1986), the abundance of food sources in the environment (Lindauer, 1948; Seeley, 1986; Seeley, 1994), and the bee's previous foraging experience (Raveret-Richter and Waddington, 1993; De Marco and Farina, 2001; De Marco et al., 2005). Concerning the last parameter, Raveret-Richter and Waddington (Raveret-Richter and Waddington, 1993) reported that waggle dance intensity (measured as dance circuits min^{-1}) showed a greater change when foragers experienced a decreasing sequence of sugar concentrations than when experiencing an increasing sequence. Interestingly, with one exception (ID in experiment 2 performed in 2009; Table 3) none of the parameters investigated in the present study were significantly affected by the sequence in which we changed sugar concentration or reward rate at the feeding site (Tables 1–3). This discrepancy between our findings and those of Raveret-Richter and Waddington (Raveret-Richter and Waddington, 1993) could be due to either differences in experimental setup (this study: each forager experienced either an increasing or a decreasing profitability sequence; Raveret-Richter and Waddington: each forager experienced first an increasing and subsequently, on the next day, a decreasing profitability sequence) or differences in analysis (this study: all recorded dances analysed; Raveret-Richter and Waddington: only the fastest dances analysed).

In a manner analogous to the thoracic vibrations, food source profitability influences the thoracic temperature of foragers [see studies on sugar concentration (Stabentheiner and Hagmüller, 1991; Stabentheiner et al., 1995; Sadler and Nieh, 2011) and reward rate (Farina and Wainselboim, 2001)]. Physiologically, a bee's thoracic temperature depends on her metabolic rate (Moffatt, 2001), which increases with increasing sugar concentration and reward rate at a food source (Moffatt and Núñez, 1997), and also with her motivational state (Balderrama et al., 1992; Moffatt, 2000; Sadler and Nieh, 2011). Although honey bees are capable of detecting temperature differences as small as 0.25°C (Heran, 1952), it is unlikely that dance followers use thoracic temperature as a source of information on the excitement of the dancers (Germ et al., 1997). Even so, hive bees have been shown to adjust their activity level (measured as thoracic temperature) according to that of the foragers (Farina and Wainselboim, 2001), which indicates that bees are indeed capable of perceiving the motivation level of others.

At the individual level, the motivational state of a bee determines whether she continues foraging or to stays in the nest (Núñez and Giurfa, 1996). At the colony level, the excitement of a waggle dancer provides important information about the availability of profitable food sources. Unemployed, experienced foragers are stimulated by the simple presence of a dancing bee to resume their collecting activity at previously visited food patches, irrespective of vector or odour information provided by the waggle dancer (Grüter et al., 2008). These experienced foragers predominantly rely on private navigational information to reinitiate foraging (Grüter et al., 2008). When their foraging activity has been interrupted, however, inactive foragers need some sort of sensory input to resume foraging tasks (Biesmeijer and Seeley, 2005). Here, the motivational level of successful foragers, transmitted through chemical and/or mechanical signals, could be an important stimulator for inactive individuals. Non-glandular pheromones produced by waggle dancers have been shown to increase the foraging activity of honey bee colonies (Thom et al., 2007). The elevation of the foragers' body temperature according to their motivational state (Stabentheiner and Hagmüller, 1991; Stabentheiner et al., 1995; Farina and Wainselboim, 2001;

Sadler and Nieh, 2011) might increase the passive release and, thus, the presence of these volatiles on the dance floor. In addition, or alternatively, inactive foragers might use the thoracic vibrations generated by the waggle dancers to access their motivational state. The fact that dancing foragers fail to recruit nestmates when their dances are not accompanied by pulsed thoracic vibrations (Esch, 1962) underlines the importance of these vibrations for the coordination of foraging processes in honey bee colonies.

Motivation-dependent nest-internal recruitment behaviour of foragers, apparently, is a common trait among the eusocial Apidae (honey bees, stingless bees and bumble bees). In stingless bees (Apidae; Meliponini), returning foragers generate pulsed thoracic vibrations predominantly during trophallactic contacts with their nest mates (Hrncir et al., 2006; Hrncir, 2009). The excitement of these vibrations increases with increasing sugar concentration [longer pulses, shorter intervals, higher duty cycle: *Melipona seminigra* (Hrncir et al., 2004b), *M. rufiventris* (Hrncir et al., 2006), *Scaptotrigona* aff. *depilis* (Schmidt et al., 2006), *Nannotrigona testaceicornis* (Schmidt et al., 2008)] and decreases with increasing energetic costs [*M. seminigra* (Hrncir et al., 2004a)] or decreasing crop load [*M. seminigra* (Hrncir, 2009)]. In bumble bees (Apidae; Bombini), so far, modulations of motivation-dependent locomotor patterns of foragers have only been studied with regard to sugar concentration. Here, walking patterns like running velocity [*Bombus terrestris* (Dornhaus and Chittka, 2005)] or the length of a forager's running path inside the nest [*B. occidentalis* (Nguyen and Nieh, 2011)] have been shown to change with the changing quality of the collected food. In both stingless bees and bumble bees, the nest-internal recruitment behaviour of foragers putatively plays a key role in mobilising the colony's foraging force (Dornhaus and Chittka, 2004; Dornhaus and Chittka, 2005; Renner and Nieh, 2008; Hrncir, 2009; Nguyen and Nieh, 2011). Hence, motivation-modulated locomotor patterns of foragers, both body vibrations and walking patterns, that influence the decision of inactive individuals of whether or not to initiate foraging, might be a common starting point for the different food-communication systems found among the eusocial corbiculate bees.

REFERENCES

Aguilar, I. and Briçeño, D. (2002). Sounds in *Melipona costaricensis* (Apidae: Meliponini): effect of sugar concentration and nectar source distance. *Apidologie* **33**, 375-388.

Arenas, A., Fernández, V. M. and Farina, W. M. (2008). Floral scents experienced within the colony affect long-term foraging preferences in honeybees. *Apidologie* **39**, 714-722.

Autrum, H. and Schneider, W. (1948). Vergleichende untersuchungen über den erschütterungssinn der insekten. *Z. Vergl. Physiol.* **31**, 77-88.

Balderrama, N. M., de Almeida, B. L. O. and Núñez, J. A. (1992). Metabolic rate during foraging in the honeybee. *J. Comp. Physiol. B* **162**, 440-447.

Biesmeijer, J. C. and Seeley, T. D. (2005). The use of waggle dance information by honey bees throughout their foraging careers. *Behav. Ecol. Sociobiol.* **59**, 133-142.

Cassill, D. (2003). Rules of supply and demand regulate recruitment to food in an ant society. *Behav. Ecol. Sociobiol.* **54**, 441-450.

Chittka, L. (2004). Dances as windows into insect perception. *PLoS Biol.* **2**, e216.

Cogorno, G. D., Fernández, P. C. and Farina. W. M. (1998). Honeybees' dancing behaviour is modulated according to the sugar flow rate of the exploited nectar source. In *New Neuroethology on the Move. Proceedings of the 26th Göttingen Neurobiology Conference* (ed. N. Elsner and R. Wehner), p. 314. Stuttgart, New York: Georg Thieme Verlag.

De Marco, R. J. and Farina, W. M. (2001). Changes in food source profitability affect the trophallactic and dance behavior of forager honeybees (*Apis mellifera* L.). *Behav. Ecol. Sociobiol.* **50**, 441-449.

De Marco, R. J., Gil, M. and Farina, W. M. (2005). Does an increase in reward affect the precision of the encoding of directional information in the honeybee waggle dance? *J. Comp. Physiol. A* **191**, 413-419.

Dornhaus, A. and Chittka, L. (2004). Information flow and regulation of foraging activity in bumble bees (*Bombus* spp.). *Apidologie* **35**, 183-192.

Dornhaus, A. and Chittka, L. (2005). Bumble bees (*Bombus terrestris*) store both food and information in honeypots. *Behav. Ecol.* **16**, 661-666.

Dyer, F. C. (2002). The biology of the dance language. *Annu. Rev. Entomol.* **47**, 917-949.

Esch, H. (1961a). Ein neuer Bewegungstyp im Schwänzeltanz der Bienen. *Naturwissenschaften* **48**, 140-141.

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

Esch, H. (1962). Über die Auswirkung der Futterplatzqualität auf die Schallerzeugung im Werbetanz der Honigbiene. *Verh. Dtsch. Zool. Ges.* **26**, 302-309.

Esch, H. (1964). Beiträge zum Problem der Entfernungsweisung in den Schwänzeltänzen der Honigbiene. *Z. vergl. Physiol.* **48**, 534-546.

Esch, H. E., Zhang, S., Srinivasan, M. V. and Tautz, J. (2001). Honeybee dances communicate distances measured by optic flow. *Nature* **411**, 581-583.

Farina, W. M. (1996). Food-exchange by foragers in the hive – a means of communication among honey bees? *Behav. Ecol. Sociobiol.* **38**, 59-64.

Farina, W. M. and Wainselboim, A. J. (2001). Changes in the thoracic temperature of honeybees while receiving nectar from foragers collecting at different reward rates. *J. Exp. Biol.* **204**, 1653-1658.

Fernández, P. C. and Farina, W. M. (2002). Individual recruitment in honeybees, *Apis mellifera* L. The effect of food source profitability on the rate of bees arriving at the feeding place. *Acta Ethol.* **4**, 103-108.

Fernández, P. C., Gil, M. and Farina, W. M. (2003). Reward rate and forager activation in honeybees: recruiting mechanisms and temporal distribution of arrivals. *Behav. Ecol. Sociobiol.* **54**, 80-87.

Germ, M., Stabentheiner, A. and Kastberger, G. (1997). Seasonal and daily variation of honeybee dancing temperature under constant feeding conditions. *Apidologie* **28**, 385-398.

Grüter, C. and Farina, W. (2009). The honeybee waggle dance: can we follow the steps? *Trend. Ecol. Evol.* **24**, 242-247.

Grüter, C., Balbuena, M. S. and Farina, W. M. (2008). Informational conflicts created by the waggle dance. *Proc. R. Soc. B.* **275**, 1321-1327.

Hasegawa, Y. and Ikeno, H. (2011). How do honeybees attract nestmates using waggle dances in dark and noisy hives? *PLoS ONE* **6**, e19619.

Heran, H. (1952). Untersuchungen über den temperatursinn der honigbiene (*Apis mellifica*) unter besonderer berücksichtigung der wahrnehmung strahlender wärme. *Z. Vergl. Physiol.* **34**, 179-207.

Heran, H. (1959). Wahrnehmung zur regelung der flugeigengeschwindigkeit bei *Apis mellifica* L. *Z. Vergl. Physiol.* **42**, 103-163.

Hrncir, M. (2009). Mobilizing the foraging force – mechanical signals in stingless bee recruitment. In *Food Exploitation by Social Insects: Ecological, Behavioral, and Theoretical Approaches* (ed. S. Jarau and M. Hrncir), pp. 199-221. Boca Raton, London, New York: CRC-Press, Taylor & Francis Group.

Hrncir, M., Jarau, S., Zucchi, R. and Barth, F. G. (2004a). Thorax vibrations of a stingless bee (*Melipona seminigra*). I. No influence of visual flow. *J. Comp. Physiol. A* **190**, 539-548.

Hrncir, M., Jarau, S., Zucchi, R. and Barth, F. G. (2004b). Thorax vibrations of a stingless bee (*Melipona seminigra*). II. Dependence on sugar concentration. *J. Comp. Physiol. A* **190**, 549-560.

Hrncir, M., Barth, F. G. and Tautz, J. (2006). Vibratory and airborne-sound signals in bee communication (Hymenoptera). In *Insect Sounds and Communication: Physiology, Behaviour, and Evolution* (ed. S. Drosopoulos and M. F. Claridge), pp. 421-436. Boca Raton: CRC-Press, Taylor & Francis Group.

Hrncir, M., Schorkopf, D. L. P., Schmidt, V. M., Zucchi, R. and Barth, F. G. (2008). The sound field generated by tethered stingless bees (*Melipona scutellaris*): inferences on its potential as a recruitment mechanism inside the hive. *J. Exp. Biol.* **211**, 686-698.

Kilpinen, O. and Storm, J. (1997). Biophysics of the subgenual organ of the honeybee, *Apis mellifera*. *J. Comp. Physiol. A* **181**, 309-318.

Lindauer, M. (1948). Über die Einwirkung von Duft- und Geschmackstoffen sowie anderer Faktoren auf die Tänze der Bienen. *Z. vergl. Physiol.* **31**, 348-412.

Menzel, R., Kirbach, A., Haass, W. D., Fischer, B., Fuchs, J., Koblofsky, M., Lehmann, K., Reiter, L., Meyer, H., Nguyen, H. et al. (2011). A common frame of reference for learned and communicated vectors in honeybee navigation. *Curr. Biol.* **21**, 645-650.

Michelsen, A. (2003). Signals and flexibility in the dance communication of honeybees. *J. Comp. Physiol. A* **189**, 165-174.

Michelsen, A., Towne, W. F., Kirchner, W. H. and Kryger, P. (1987). The acoustic near field of a dancing honeybee. *J. Comp. Physiol. A* **161**, 633-643.

Moffatt, L. (2000). Changes in the metabolic rate of the foraging honeybee: effect of the carried weight or of the reward rate? *J. Comp. Physiol. A* **186**, 299-306.

Moffatt, L. (2001). Metabolic rate and thermal stability during honeybee foraging at different reward rates. *J. Exp. Biol.* **204**, 759-766.

Moffatt, L. and Núñez, J. A. (1997). Oxygen consumption in the foraging honeybee depends on the reward rate at the food source. *J. Comp. Physiol. B* **167**, 36-42.

Morse, P. M. (1981). *Vibration and Sound.* New York: The Acoustical Society of America.

Nguyen, H. and Nieh, J. C. (2011). Colony and individual forager responses to food quality in the New World bumble bee, *Bombus occidentalis*. *J. Insect Behav.* doi:10.1007/s10905-011-9277-5.

Nieh, J. C. and Tautz, J. (2000). Behaviour-locked signal analysis reveals weak 200-300 Hz comb vibrations during the honeybee waggle dance. *J. Exp. Biol.* **203**, 1573-1579.

Núñez, J. A. (1966). Quantitative Beziehungen zwischen den Eigenschaften von Futterquellen und dem Verhalten von Sammelbienen. *Z. vergl. Physiol.* **53**, 142-164.

Núñez, J. A. (1970). The relationship between sugar flow and foraging and recruiting behaviour of honey bees (*Apis mellifera* L.). *Anim. Behav.* **18**, 527-538.

Núñez, J. A. (1971). A simulator for learning studies in the bee *Apis mellifera* L. *Acta Cient. Venez.* **22 (Suppl. 2)**, 101-106.

Núñez, J. A. (1977). Nectar flow by melliferous flora and gathering flow by *Apis mellifera ligustica*. *J. Insect. Physiol.* **23**, 265-275.

Núñez, J. A. (1982). Honeybee foraging strategies at a food source in relation to its distance from the hive and the rate of sugar flow. *J. Apicult. Res.* **21**, 139-150.

Núñez, J. A. and Giurfa, M. (1996). Motivation and regulation of honey bee foraging. *Bee World* **77**, 182-196.

Raveret-Richter, M. and Waddington, K. D. (1993). Past foraging experience influences honey bee dance behaviour. *Anim. Behav.* **46**, 123-128.

Renner, M. and Nieh, J. C. (2008). Bumble bee olfactory information flow and contact-based foraging activation. *Insect. Soc.* **55**, 417-424.

Riley, J. R., Greggers, U., Smith, A. D., Reynolds, D. R. and Menzel, R. (2005). The flight paths of honeybees recruited by the waggle dance. *Nature* **435**, 205-207.

Roces, F. (1993). Both evaluation of resource quality and speed of recruited leafcutting ants (*Acromyrmex lundi*) depend on their motivational state. *Behav. Ecol. Sociobiol.* **33**, 183-189.

Rohrseitz, K. (1998). Biophysikalische und ethologische aspekte der tanzkommunikation der honigbienen (*Apis mellifera carnica* Pollm.). Doctoral thesis, University of Würzburg, Germany.

Rowe, C. (1999). Receiver psychology and the evolution of multicomponent signals. *Anim. Behav.* **58**, 921-931.

Sadler, N. and Nieh, J. C. (2011). Honey bee forager thoracic temperature inside the nest is tuned to broad-scale differences in recruitment motivation. *J. Exp. Biol.* **214**, 469-475.

Sandeman, D. C., Tautz, J. and Lindauer, M. (1996). Transmission of vibration across honeycombs and its detection by bee leg receptors. *J. Exp. Biol.* **199**, 2585-2594.

Schmidt, V. M., Zucchi, R. and Barth, F. G. (2006). Recruitment in a scent trail laying stingless bee (Scaptotrigona aff. depilis): changes with reduction but not with increase of the energy gain. *Apidologie* **37**, 487-500.

Schmidt, V. M., Hrncir, M., Schorkopf, D. L. P., Mateus, S., Zucchi, R. and Barth, F. G. (2008). Food profitability affects the intranidal recruitment behaviour in the stingless bee *Nannotrigona testaceicornis*. *Apidologie* **39**, 260-272.

Seeley, T. D. (1986). Social foraging by honeybees: how colonies allocate foragers among patches of flowers. *Behav. Ecol. Sociobiol.* **19**, 343-354.

Seeley, T. D. (1994). Honey bee foragers as sensory units of their colonies. *Behav. Ecol. Sociobiol.* **34**, 51-62.

Seeley, T. D. and Towne, W. F. (1992). Tactics of dance choice in honey bees: do foragers compare dances? *Behav. Ecol. Sociobiol.* **30**, 59-69.

Seeley, T. D., Mikheyev, A. S. and Pagano, G. J. (2000). Dancing bees tune both duration and rate of waggle-run production in relation to nectar-source profitability. *J. Comp. Physiol. A* **186**, 813-819.

Stabentheiner, A. and Hagmüller, K. (1991). Sweet food means hot dancing in honeybees. *Naturwissenschaften* **78**, 471-473.

Stabentheiner, A., Kovac, H. and Hagmuller, K. (1995). Thermal behavior of round and wagtail dancing honeybees. *J. Comp. Physiol. B* **165**, 433-444.

Tautz, J. (1996). Honeybee waggle dance: recruitment success depends on the nature of the dance floor. *J. Exp. Biol.* **199**, 1375-1381.

Tautz, J. and Rohrseitz, K. (1998). What attracts honeybees to a waggle dancer? *J. Comp. Physiol. A* **183**, 661-667.

Tautz, J., Zhang, S., Spaethe, J., Brockmann, A., Si, A. and Srinivasan, M. (2004). Honeybee odometry: performance in varying natural terrain. *PLoS Biol.* **2**, 915-923.

Thom, C., Gilley, D. C., Hopper, J. and Esch, H. E. (2007). The scent of the waggle dance. *PLoS Biol.* **5**, 1862-1867.

Towne, W. F. (1985). Acoustical and visual cues in the dances of four honey bee species. *Behav. Ecol. Sociobiol.* **16**, 185-187.

Tsujiuchi, S., Sivan-Loukianova, E., Eberl, D. F., Kitagawa, Y. and Kadowaki, T. (2007). Dynamic range compression in the honey bee auditory system toward waggle dance sounds. *PLoS ONE* **2**, e234.

Varjú, D. and Núñez, J. (1991). What do foraging honeybees optimize? *J. Comp. Physiol. A* **169**, 729-736.

Vogel, S. (1983). Ecophysiology of zoophilic pollination. In *Physiological Plant Ecology III – Encyclopedia of Plant Physiology* (ed. O. L. Lange, P. S. Nobel, C. B. Osmond and H. Ziegier), pp. 559-624. Berlin, Heidelberg, New York: Springer Verlag.

von Frisch, K. (1923). Über die sprache der bienen, eine tierpsychologische untersuchung. *Zool. Jahrb.* **40**, 1-186.

von Frisch, K. (1942). Die werbetänze der bienen und ihre auslösung. *Naturwissenschaften* **30**, 269-277.

von Frisch, K. (1946). Die tänze der bienen. *Österr. Zool. Zeitsch.* **1**, 1-48.

von Frisch, K. (1965). *Tanzsprache und Orientierung der Bienen.* Berlin, Heidelberg, New York: Springer Verlag.

Waddington, K. D. (1982). Honey bee foraging profitability and round dance correlates. *J. Comp. Physiol. A* **148**, 297-301.

Waddington, K. D. (1985). Cost-intake information used in foraging. *J. Insect Physiol.* **31**, 891-897.

Waddington, K. D. (2001). Subjective evaluation and choice behavior by nectar- and pollen-collecting bees. In *Cognitive Ecology of Pollination* (ed. L. Chittka and J. D. Thomson), pp. 41-60. Cambridge: Cambridge University Press.

Waddington, K. D. and Kirchner, W. H. (1992). Acoustical and behavioral correlates of profitability of food sources in honey bee round dances. *Ethology* **92**, 1-6.

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

Zar, J. H. (1999). *Biostatistical Analysis*, 4th edn. Englewood Cliffs: Prentice Hall.