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Adaptive evolution of honeybee dance dialects

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Efficient communication is highly important for the evolutionary success of social animals. Honeybees (genus *Apis*) are unique in that they communicate the spatial information of resources using a symbolic 'language', the waggle dance. Different honeybee species differ in foraging ecology but it remains unknown whether this shaped variation in the dance. We studied distance dialects—interspecific differences in how waggle duration relates to flight distance—and tested the hypothesis that these evolved to maximize communication precision over the bees' foraging ranges. We performed feeder experiments with *Apis cerana*, *A. florea* and *A. dorsata* in India and found that *A. cerana* had the steepest dialect, i.e. a rapid increase in waggle duration with increasing feeder distance, *A. florea* had an intermediate, and *A. dorsata* had the lowest dialect. By decoding dances for natural food sites, we inferred that the foraging range was smallest in *A. cerana*, intermediate in *A. florea* and largest in *A. dorsata*. The inverse correlation between foraging range and dialect was corroborated when comparing six (sub)species across the geographical range of the genus including previously published data. We conclude that dance dialects constitute adaptations resulting from a trade-off between the spatial range and the spatial accuracy of communication.

1. Introduction

Since Karl von Frisch discovered in the 1940s that honeybees use the waggle dance to communicate the spatial locations of resources among nest-mates, this unique behaviour has served as a primary model for the study of animal communication [1–5]. When returning from a successful foraging trip, a worker honeybee may display to her nest-mates a figure-eight movement pattern of two-way alternating dance circuits (figure 1a). A circuit is composed of the waggle phase, a straight run accompanied by abdomen-wagging which contains the direction and distance information of the goal, and the return phase, which is a stereotyped inter-signal behaviour. While the body orientation of the dancer during the waggle phase encodes the direction of the resource in the field, the duration of the waggling linearly correlates with its distance, with longer waggle durations being associated with further flight distances [1,6]. Astonishingly, most honeybee research has been conducted with only a few European lineages of a single species, the Western honeybee, although there is a huge diversity of geographical subspecies of *Apis mellifera* and there are several other honeybee species, that are native to tropical and eastern Asia [7–9]. The extant diversity of *Apis* bees includes populations occurring in tropical to temperate habitats, variation in colony size and nesting behaviour, and a wide variation of worker body sizes, all of which are likely to influence the bees' foraging ecologies. At the same time, all honeybees share the same basic mechanisms of dance communication, offering the opportunity for comparative studies [10].
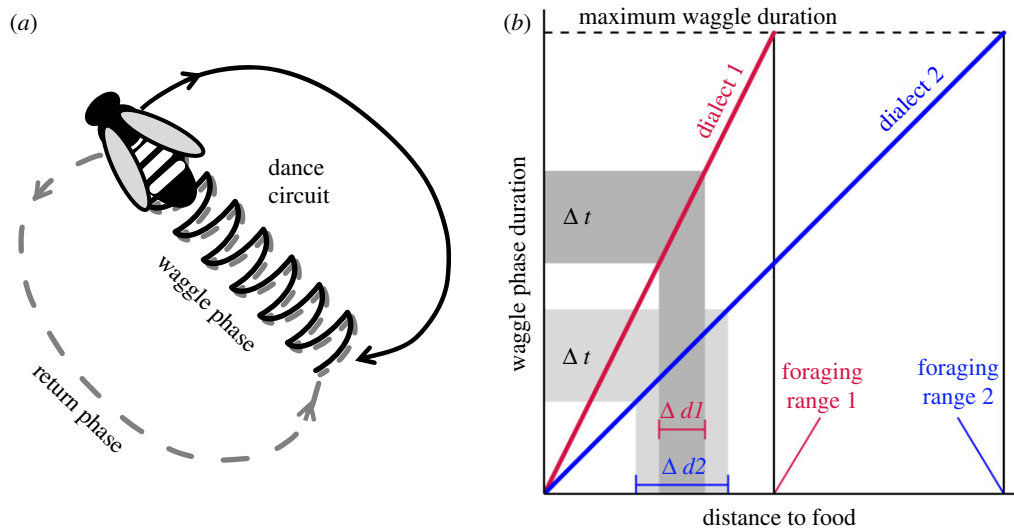
Figure 1. (a) Scheme of the figure-eight movement pattern of the waggle dance. The waggle phase contains the direction (orientation of the dancer) and distance information (duration of wagging). One waggle phase plus one subsequent return phase makes a dance circuit. Dancing bees usually perform several consecutive circuits in a single dance. (b) Graphical representation of the adaptive-tuning hypothesis for the evolution of distance dialects. A steeper dialect (dialect 1) allows for a more precise distance indication as a given change in waggle duration (Δt) indicates a smaller range of flight distances ($\Delta d1$ versus $\Delta d2$). A lower dialect (dialect 2) allows to communicate over a larger foraging range (foraging range 2 versus foraging range 1). (Online version in colour.)

Research on *A. mellifera* has shown that the usefulness of the dances' spatial information for enhancing foraging success depends on the resource environment (i.e. the tempo-spatial distribution of food patches and the variability in food quality) [11–17]. Conversely, it is an enduring question whether the dance communication system is adapted evolutionarily according to ecological variation between co-occurring species or between subspecies that live in different environments [3,10].

Studying three Asian honeybee species in Sri Lanka and geographical subspecies of the western honeybee, *Apis mellifera*, respectively, Lindauer [18] and Bosch [19] found that these communicate with (sub)species-specific distance codes, so-called distance dialects. A distance dialect is the slope of the linear function relating the duration of the waggle phase to the distance of a resource in the field. The original explanation for the existence of distance dialects, as provided by the 'adaptive-tuning hypothesis' [20], was that they represent ecological adaptations that evolved to maximize communication precision over the bees' typical foraging ranges [1,3,10] (figure 1b). The basic premise is that there is a limitation to the maximum waggle duration of dances. On the one hand, dances with very long waggle durations could be *ineffective* because dancing bees might fail to produce, and dance followers might struggle to understand, the distance signal on the crowded dance floor [3,10]. On the other hand, dances with very long waggle durations might become *inefficient* as increasing time costs might outweigh the benefits of communication [21]. Honeybees with 'steeper dialects' have a more precise distance communication because a given change in waggle duration encodes a smaller range of foraging distance in the field. However, the drawback of steep dialects is that the bees are restricted in the spatial range over which they can communicate, since with steep dialects they quickly reach the maximum possible waggle duration. Irrespective of the intrinsic (e.g. body size constraints on maximum flight range) and ecological factors (availability and spatial distribution of resources; competition) that determine how far the bees typically need to fly, the resulting prediction is

that honeybees with smaller foraging ranges have steeper distance dialects [3,10].

The distance code can be experimentally determined by examining dances of bees trained to artificial feeders at known distances from the nest [1]. Conversely, when a researcher knows the distance calibration function of the bees, dances for natural food sources can be 'eavesdropped' and decoded to infer the indicated foraging distances (forage mapping technique [22]). Two prior studies, each applying these methods to three sympatric honeybee species occurring in South Asia, *A. cerana*, *A. florea* and *A. dorsata*, did not confirm the prediction of the adaptive-tuning hypothesis. Punchihewa *et al.* [23] confirmed the dialect differences originally described by Lindauer [18] in Sri Lanka, but the inferred differences in the foraging ranges were small and did not correlate with the dialects. By contrast, Dyer & Seeley [20] found all three species to communicate with similar distance codes but to have markedly different foraging ranges in Thailand.

Apart from the above described seeming contradiction, there are two reasons why these results might not yet be conclusive. Instead of determining the duration of the waggle phase, it was common practice in earlier studies to take the duration of dance circuits (the duration of the waggle phase plus the subsequent return phase; figure 1a) as the measure of the distance signal. Circuit duration also correlates with flight distance and is easily determined by direct observation in the field, while measuring waggle duration requires video recordings and analyses. Although circuit duration is a good predictor of foraging distance, it might not be suited for a dialect comparison between species unless these had similar return phase durations, a prerequisite that has never been validated. Another issue arose when it was shown that honeybees estimate distance by the amount of image motion perceived during flight [24–27]. As the same bees can communicate distance differently depending on the flight path, any study of dialect comparison should control for the visual features of the terrain.

Based on these considerations, we here revisit the unsolved problem of honeybee distance dialects, and ask whether the

bees bear adaptations of the dance that are related to their foraging ecology. We empirically studied three sympatric honeybee species, the Asiatic honeybee *A. cerana*, the dwarf honeybee *A. florea* and the giant honeybee *A. dorsata*, in Bangalore, India, and consulted published data for three additional honeybee populations to test the prediction that distance dialect slopes are inversely correlated to the bees' foraging ranges.

2. Material and methods

(a) Study sites, species and dance observation

All experiments and observations were conducted at the adjacent campuses of the National Centre for Biological Sciences (NCBS) and the University of Agricultural Sciences (UAS) in Bangalore, India, during the dry season between January and March 2017 and between February and April 2018. Wild colonies of all three honeybee species native to the region, *A. cerana*, *A. florea* and *A. dorsata*, naturally inhabit the area. We used different observation techniques to account for species differences in nesting behaviour [28] (electronic supplementary material, figure S1). *A. cerana* colonies build multiple comb nests in cavities and the foragers dance on the perpendicular combs in the dark. We bought colonies from local beekeepers and transferred them into custom-built observation hives fitting six parallel comb frames of the local standard size (outer frame dimensions: 23 cm × 16 cm × 3 cm). They had a lateral window, which allowed us to observe the outer side of the first comb to which homecoming bees were guided via a wedge [22,28]. A lightproof cloth covering the hive ensured that dancing bees would not be influenced by direct sunlight but allowed diffuse light to enter from below, which enabled video recording. *A. florea* and *A. dorsata* colonies build exposed single-comb nests attached to twigs, tree branches or buildings which they protect with a curtain of bees. In both species, foragers dance at the nests' peripheries in the open. We spotted colonies of *A. florea* nesting in trees or bushes on the campuses. In late afternoon, when flight activity had ceased, we wetted the nests with a water sprayer, cut the supporting twigs and mounted them onto open boxes of plywood using metal wires. With this set-up, the dances, which are performed on a concave wax crown on top of the nest, could be recorded from above by a downward-facing camera fixed to a tripod. The nests of *A. dorsata* are directly attached to big tree branches or buildings. Transferring a colony requires cutting the comb off its original substrate and mounting it to some artificial attachment. However, few colonies are sufficiently accessible for such a procedure, and the partial destruction of the nest can seriously disturb the bees which is bad for the study and the researchers (*A. dorsata* colonies are notorious for fierce coordinated defence reactions). We, therefore, relied on colonies nesting at buildings on the campuses that could be approached via windows, stairways or rooftops. We recorded the dances, which are performed on top of the bee curtain, using cameras mounted onto extension arms. In total, we used data from four colonies of each *A. cerana* (colony IDs: C1–C4), *A. florea* (colony IDs: F1–F4) and *A. dorsata* (colony IDs: D1–D4). Videos were recorded at Full HD (1920 × 1080 pixel) and at 50 or 25 Hz frame rate for the distance training experiment and the natural dance observation, respectively, using commercially available video cameras (Sony HDR-CX240, Panasonic HC-X929, Panasonic HC-V707). We analysed the videos manually using the program UTILIUS FAIRPLAY 5 (ccc software, Leipzig, Germany).

(b) Distance communication

We determined the relationships between foraging distance and different components of the waggle dance by training bees to visit artificial feeders at known distances from the hives and examining their corresponding recruitment dances [1]. To rule out any confounding effects of the visual features of the terrain on the comparison, we trained bees of all three species in the same area of the Botanical Garden of the UAS (electronic supplementary material, figure S2). As an artificial food source, we provided a gravity feeder containing scented sucrose solution (star anise; 1–2.5 mol l^{-1}, adjusted to the bees' propensity to forage and dance) placed on a coloured stool. The test locations, which we had determined *a priori* with the use of a hand held GPS device, were at distances of 100, 200, 300, 400 and 500 m from the nests. To initially make the bees visit our feeders, we had to use slightly different procedures. In the case of *A. cerana*, we simply placed the bait in front of the hive entrance where it was soon discovered by the bees. To get *A. florea* foragers, we dipped a small stick into the sugar solution, presented it to the bees on top of the nest until some crawled onto it, and transferred the bees to the feeder. In the case of *A. dorsata*, we filled a piece of wax comb with sugar solution, pinned it onto a 2 m-long plastic pole and presented it to the bees via a window. Once some bees had climbed onto the comb, we pulled the pole back and carefully transferred the comb with the bees into a small box. With the box, we went outside to the feeder stand and released the bees, of which some would eventually learn the place and come back independently. Once we made a move to a test location we individually labelled foragers with paint marks of different colours on the thorax and/or the abdomen (Uni POSCA Paint Markers, Uni Mitsubishi Pencil, UK), kept the feeder for at least 1 h and video recorded the dances at the nests.

Prior studies generally established that different honeybee colonies from the same local population communicate distance with a similar code [1,23]. Indeed, in *Apis mellifera*, a considerable fraction of a population's variation in distance communication is represented by the variation between individuals of the same colony [29,30]. Thus, we expected that the distance code of any test colony should give a good approximation of its population's distance code. We performed the experiment twice for the species *A. cerana* (colonies C1 and C4) and *A. florea* (colonies F1 and F3) but relied on a single *A. dorsata* colony (colony D2) nesting at a building next to the Botanical Garden to determine its population's distance dialect. During *A. dorsata* training, a mass bloom of trees (*Tabebuia spec.*) near the nest hampered our attempts to train foragers beyond 400 m.

For the analyses we screened the recorded videos to identify all unique individuals that danced for the feeder stations and analysed sets of 6–11 (mean ± s.d.: 8 ± 1.5) dances per feeder distance, each performed by a different bee. This resulted in 43 dances of *A. cerana* (C1: 27 dances, C4: 16 dances), 43 dances of *A. florea* (F1: 31 dances, F3: 12 dances) and 30 dances of *A. dorsata*. We determined the duration of a waggle phase as the time lapse between the first still image in which a focal bee had clearly moved its abdomen laterally or dorsoventrally and the next still image in which the bee had stopped wagging its abdomen again. By going through the videos frame by frame we determined the duration with a temporal resolution of approximately 0.02 s. To obtain a dance's mean waggle phase duration we averaged the duration of three to six (mean ± s.d.: 5.54 ± 0.86) consecutive waggle phases within that dance. We also determined the circuit durations of the same dances by averaging the duration of 3–19 (mean ± s.d.: 7.3 ± 2.9) consecutive circuits, and inferred the return phase durations by subtracting waggle duration from circuit duration.

(c) Foraging distances

We made video recordings of bees dancing for natural food sources, surveyed random sets of these dances and inferred the indicated foraging distances using the distance calibration functions obtained from the respective feeder training experiments

[22]. As we worked in an agro-urban environment, we also refer to nectar and pollen from plants growing in gardens and plantations as 'natural' food sources. Honeybees also forage and dance for water and it is not possible to differentiate by observation if a bee is dancing for a nectar or water source. However, as the gardens on the campuses were watered daily, all observation colonies had access to it in their immediate surroundings (within 20–30 m distance), guaranteeing that the forage distance distributions could not have been biased by differences in water access. Colonies of each species were videoed in parallel or close to parallel for 3–12 h per day during their daytime activity between 06.00 and 18.00 h. This was done on a total of 16 days in February and March 2017 and in March and April 2018. Besides using the colonies with which we had determined the distance dialects, we decoded dances of two to three other colonies per species, resulting in 43 colony observation days (see electronic supplementary material, table S1 for a timetable of the natural dance observations). As we only observed one side of the first comb of *A. cerana* colonies, we probably did not keep track of all dances performed by the bees. However, since we assumed that those dances that were in sight of the camera represented a random set, we screened the complete video sequences and analysed all recorded dances (we avoided to decode more than one dance performed by the same bee during a single home stay, though). In *A. florea* and *A. dorsata*, we kept track of all dances as foragers dance at the nests' peripheries in the open. Here, we analysed dances occurring during the first 5 min of every 0.5 h of daytime that were covered by the videos (i.e. dances performed between 09.00–09.05 h, 09.30–09.35 h, 10.00–10.05 h, etc.). This resulted in similar sample sizes for the three species (*A. cerana*: 400 dances; *A. florea*: 372 dances; *A. dorsata*: 373 dances). Both waggle phase duration and circuit duration highly correlate with foraging distance [1]; so that human observers can use both measures for forage mapping (waggle duration, e.g. [31]; circuit duration, e.g. [22,32–34]). We decided to infer foraging distance from dances' circuit durations because this is much more time efficient: to determine a dance's average circuit duration, one needs to make a single time measurement of a counted number of consecutive circuits. In comparison, to determine a dance's average waggle phase duration, one needs to separately time multiple waggle phases that are interspersed by return phases, which multiplies the worktime needed per dance. We averaged the duration of two to 24 (mean ± s.d.: 7 ± 3.02) consecutive circuits per dance and inferred the indicated foraging distance by using each species' inverse of the empiric linear function relating circuit duration to foraging distance. Because we linearly extrapolated to decode dances with circuit durations indicating foraging distances beyond the 400–500 m covered by the feeder training experiments, there is some uncertainty in the inferred distances beyond that range. However, as almost 90% of all observed dances indicated foraging distances within 500 m from the hives, any uncertainties regarding the exact maximum foraging distances would have a minimal effect on our conclusions regarding the typical foraging ranges.

(d) Global test of adaptive distance dialects

To make a general test of the relationship between foraging range and distance dialect we searched for other studies in which researchers determined distance dialects (based on waggle phase duration) and foraging ranges for the same populations of honeybees. Three other datasets were available: for *A. mellifera* from southern England [31,35], for *A. mellifera* from the Okavango delta in Botswana [36] and for *A. cerana* from Japan [37]. We consulted the median of observed foraging distances as a measure of a population's typical foraging range. This distance often lies within the range for which distance-dance calibration

functions were empirically determined and hence for which distances were inferred with high certainty.

(e) Statistical analysis

All statistical analyses described in the following were performed in R 3.5.1 [38]. Data figures were created using the graphics package ggplot2 [39].

(i) Distance dialects

For both *A. cerana* and *A. florea*, we first made a comparison of two linear regression models, one only containing the factor distance and the other containing the interaction between the factors distance and colony ID (to account for colony-specific distance dialect slopes) using the Akaike information criterion for small sample sizes (AICc, package MuMIn [40]). In both species, models not accounting for colony-specific dialect slopes had smaller AICc values (*A. cerana*: ΔAICc = 1.172; *A. florea*: = 4.972). Hence, we concluded that in each case most of the variation was caused by between-bee differences, not by between-colony differences. This allowed us to pool the dances of both test colonies in *A. cerana* and *A. florea* for the dialect comparison between species. It also supported our assumption that the single *A. dorsata* test colony should be representative of its population's dialect. As we sampled from different individuals, the dances were independent. A visual data inspection revealed that the distance–waggle duration, distance–circuit duration and distance–return duration relationships were linear in all species, variances were homogeneous and data for any species–distance combination appeared to be drawn from normal distributions. This allowed us to model the data using simple linear regression (lm function from the stats package). To test whether the slopes of the distance–waggle duration functions, the distance–circuit duration and the distance–return duration functions of the three species differed statistically, we pooled the respective datasets of the three species and analysed the effect of the interaction between species and foraging distance using multiple linear regression models.

(ii) Foraging distances

We grouped the inferred foraging distances into equally sized distance classes (distance class size: 20 m), thereby creating the dummy response variable 'number of dances' (count data), and the dummy explanatory variable 'distance class' (treated as a continuous variable). By pooling the three datasets, we created the variable 'species'. We then analysed whether the species differed with respect to the observed number of dances for different distance classes using generalized linear mixed models with negative binomial error structures and the observation colonies as random factors ('glmmTMB' function from the 'glmmTMB' package; [39]). We produced two models, one with an interaction term between our two predictors (species and distance class) and another without an interaction between the two, and used a likelihood ratio test (using the ANOVA function in the stats package) to determine the effect of the interaction between species and distance class.

(iii) 'Global test'

Using data of six honeybee populations, we tested whether there was an inverse correlation between dialect slope and median foraging range (log transformed to meet linearity) using Pearson's correlation (one-sided test).

3. Results

The feeder training experiments revealed strong interspecific differences in the waggle duration–flight distance relationship— there were distance dialects (LM: $F_{2,110} = 23.658$, $p < 0.0001$;
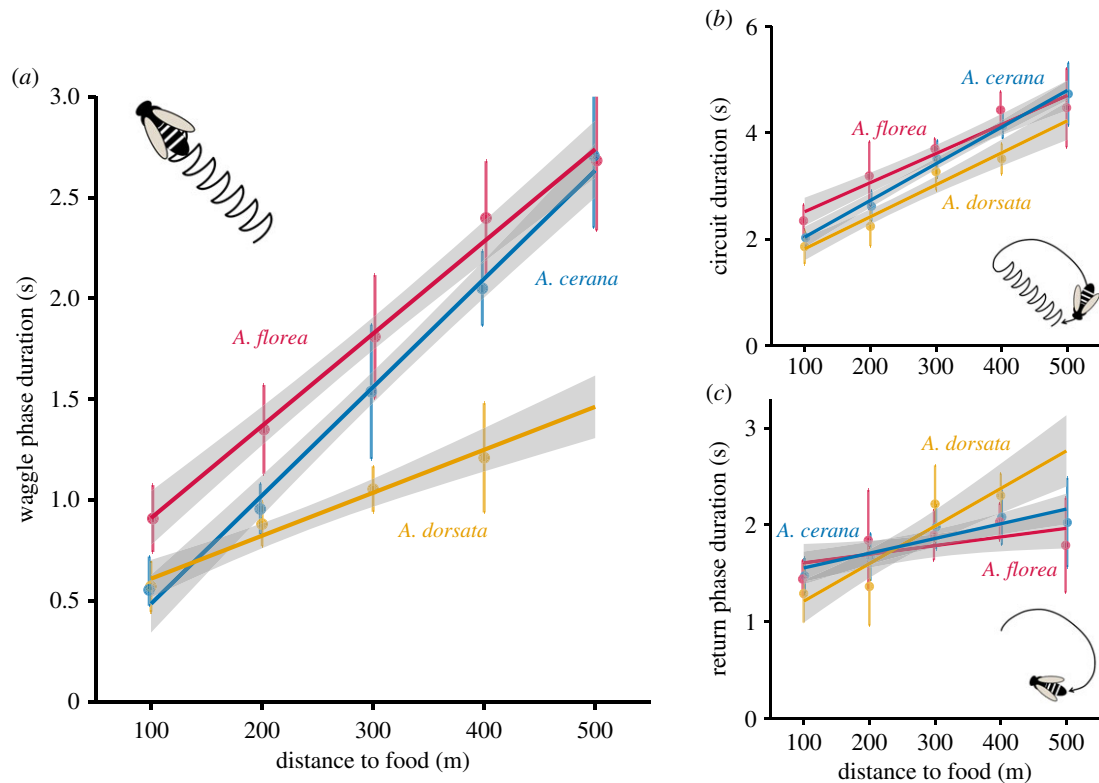
Figure 2. Comparison of the relationships between flight distance and the duration of three components of the waggle dance in the Asian honeybee species *A. cerana*, *A. florea* and *A. dorsata* in Bangalore, India. Dots and bars give means and standard deviations of 6–11 dances per distance. Regression lines and 95% confidence intervals stem from simple linear regression models. (*a*) Waggle phase duration (*A. cerana*: $y = 0.0054x - 0.0514$, $R^2 = 0.897$; *A. florea*: $y = 0.0046x + 0.4532$, $R^2 = 0.87$; *A. dorsata*: $y = 0.0021x + 0.3968$, $R^2 = 0.702$). (*b*) Circuit duration (*A. cerana*: $y = 0.0069x + 1.3502$, $R^2 = 0.886$; *A. florea*: $y = 0.0055x + 1.9701$, $R^2 = 0.713$; *A. dorsata*: $y = 0.006x + 1.2171$, $R^2 = 0.78$). (*c*) Return phase duration (*A. cerana*: $y = 0.0015x - 1.4016$, $R^2 = 0.333$; *A. florea*: $y = 0.0009x + 1.5169$, $R^2 = 0.107$; *A. dorsata*: $y = 0.0039x + 0.821$, $R^2 = 0.581$). See text for statistics. (Online version in colour.)

figure 2*a*). We found *A. cerana* to have the dialect with the steepest slope (5.4 s of wagging per kilometre flight distance), *A. florea* to have an intermediate dialect (4.6 s km^{-1}) and *A. dorsata* to have the lowest dialect (2.1 s km^{-1}).

When considering the distance-related increase in circuit duration (waggle duration + return phase duration), the interspecific dialects were obscured (LM: $F_{2,110} = 2.440$, $p = 0.092$; figure 2*b*). Compared to the slope differences in waggle duration, the slopes of the circuit duration–distance functions were more similar and the species differences were partly interchanged. *A. cerana* still had the steepest slope (6.9 s of circuit duration per kilometre), but the circuit duration slope of *A. dorsata* (6 s km^{-1}) was steeper than that of *A. florea* (5.5 s km^{-1}). This is because the duration of the return phase was not a constant but increased differently with distance in the three species (LM: $F_{2,110} = 9.459$, $p < 0.0001$; figure 2*c*). The return phase duration of *A. dorsata* (3.9 s of return phase per kilometre) increased 2.6 times more rapidly than that of *A. cerana* (1.5 s km^{-1}) and 4.3 times more rapidly than that of *A. florea* (0.9 s km^{-1}) (see also electronic supplementary material, figure S3 and table S2).

Decoding waggle dances for natural food sources revealed that the three species slightly differed in the distribution of foraging distances (GLMM: d.f. = 2, $\chi^2 = 5.268$, $p = 0.072$). The species' differences in foraging ranges were inversely correlated with the differences in distance dialects: *A. cerana* had the smallest foraging range with 50% of observed dances advertising food sources within only 94 m from the hive and with a mean ± s.d. foraging distance of 145 ± 165 m. *A. florea* had an

intermediate foraging range (median: 148 m, mean ± s.d.: 225 ± 297 m) and *A. dorsata* had the largest foraging range (median: 197 m, mean ± s.d.: 374 ± 516 m). Other parameters of the foraging distance distributions, like the 90th percentile and the maximum foraging range, differed similarly between the three species (figure 3).

For a global test of whether distance dialects comprise ecological adaptations, we consulted all available studies in which dialect and foraging range had been determined for the same population of honeybees. Covering six datasets from across the geographical range of the genus, the analysis included our data representing tropical populations of the three major Asian honeybee species, data for both temperate and tropical populations of *A. mellifera* from England [31,35] and Botswana [36], respectively, and a dataset for temperate *A. cerana* from Japan [37]. There was a general inverse relationship between the slope of the distance dialect and the distance at which the bees typically forage (figure 4). The pattern is unlikely to have arisen by chance (Pearson's correlation: $r = -0.782$, $p = 0.033$). Interestingly, *A. mellifera* and *A. cerana* converged in that populations living in tropical climate zones had steeper dance dialect slopes and smaller foraging ranges than their temperate relatives.

4. Discussion

The notion that honeybee (sub)species communicate via different distance codes is as old as the discovery of the dance language itself. However, the question of what
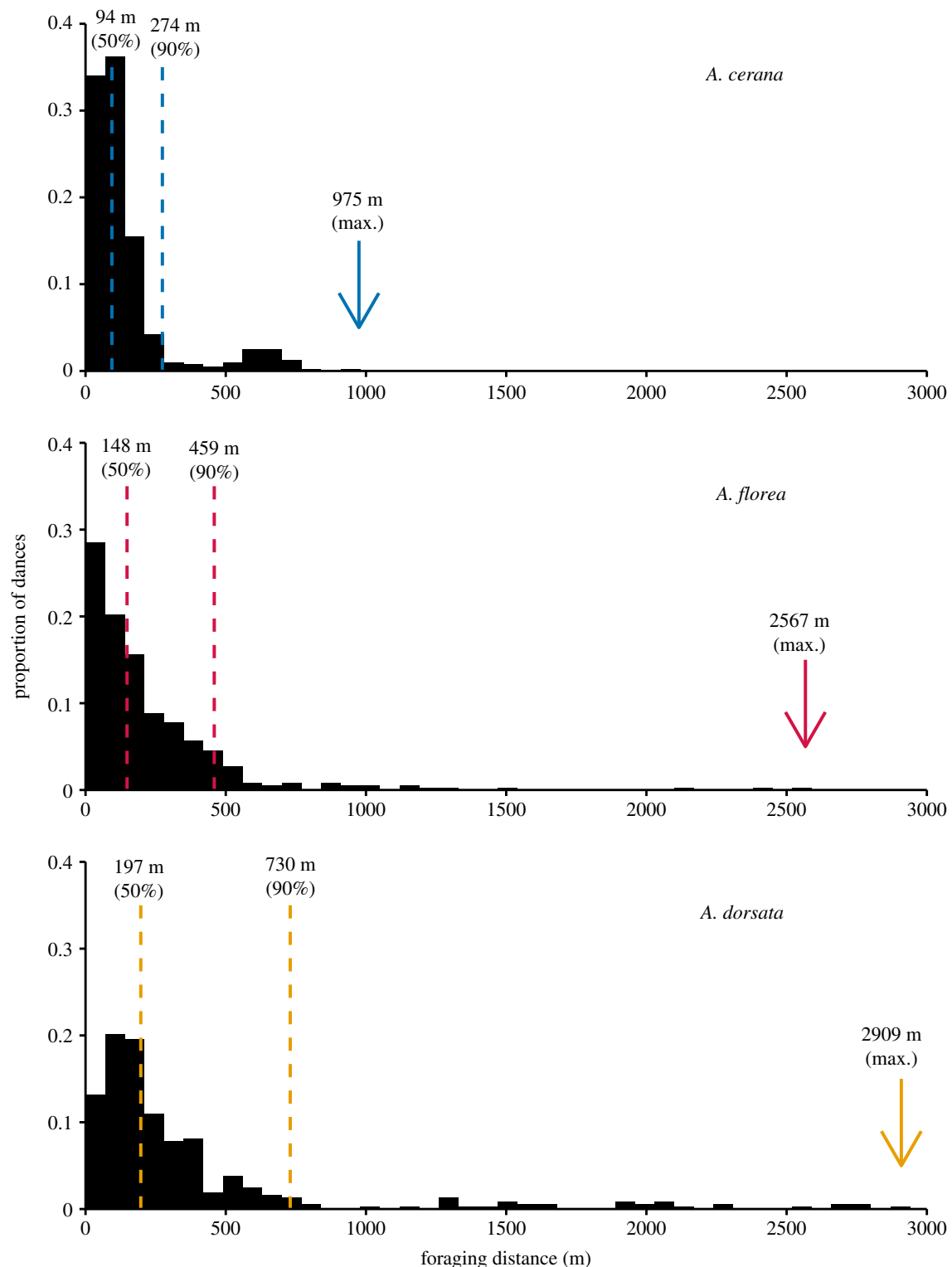
Figure 3. Comparison of the foraging ranges of *A. cerana*, *A. florea* and *A. dorsata* in Bangalore, India. Foraging distances were inferred from recruitment dances for natural food sources performed by bees in observation colonies (*A. cerana*: $n = 400$ dances, four colonies; *A. florea*: $n = 372$ dances, four colonies; *A. dorsata*: $n = 373$ dances, four colonies). Dashed lines give the spatial range within which 50% and 90% of observed foraging took place, arrows indicate the maximum inferred foraging distances. See text for statistics. (Online version in colour.)

causes these 'distance dialects' has remained unsolved until today. This is partly because existing studies seemed conflicting, and partly because insights from the physiology of the dance raised the concern that dialects might be an artefact caused by variation of the experimental environments. Here, we demonstrate that distance dialects exist among the three sympatric Asian honeybee species *A. cerana*, *A. florea* and *A. dorsata* in Bangalore, India. As we recorded the bees on video, we were able to measure different distance-related

components for the same dances. This revealed that there are clear distance dialects when considering the duration of the waggle phase, but that the dialects are obscured when considering the duration of dance circuits (waggle phase plus return phase). This is crucial given that in former studies researchers regularly measured circuit duration as a proxy of the distance signal. Further, we show that steeper distance dialects are associated with smaller foraging ranges, and, along with data from other studies, that populations of the
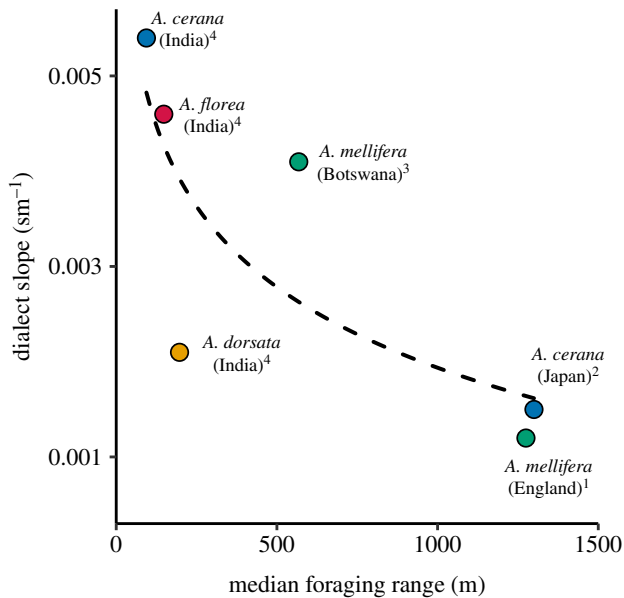
Figure 4. Distance dialect slope versus median foraging range for six honeybee populations. [1]Data from [35] (dialect slope) and [31] (foraging range); [2]data from [37]; [3]data from [36]; [4]this study. See electronic supplementary material, table S3 for data values and data sources. Dotted line: back-transformed regression of dialect slope versus log-transformed foraging range. See text for statistics. (Online version in colour.)

same species can have divergent foraging ranges and distance dialects. These findings support the hypothesis that distance dialects comprise ecological adaptations, being selected towards maximizing communication precision with respect to the bees' foraging range.

The finding that distance dialects in the waggle dance are real is important since the recognition that honeybees use optical flow to estimate flight distance had raised the concern that the dialects were merely a result of bees experiencing different visual information during experimentation [25,42]. By conducting distance training experiments with Indian A. cerana, A. florea and A. dorsata foragers in the same terrain, we ruled out that differences in the visual environment during the bees' flights could be responsible for the observed differences in distance communication. Further, the considerable magnitude of dialect differences between honeybee (sub)species across the geographical range of the genus (figure 4) suggests that earlier studies also reported true variation of distance communication in the waggle dance.

Our results also explain why the two previous studies using the same three sympatric honeybee species in Sri Lanka [23] and in Thailand [20], respectively, may have reported conflicting results on distance dialects. The researchers measured the duration of dance circuits, assuming differences in the circuit duration–distance relationships would mirror differences in the actual distance dialects based on waggle duration. In India, however, the species differences were hidden when we measured circuit duration because the species also differed in how the return phase duration of the dance relates to foraging distance. We suspect that also in Sri Lanka and Thailand the return phase may have masked the actual dialect differences between the three species.

The second major finding is that distance dialects are inversely correlated to the bees foraging ranges. This holds true for both our direct comparison of three species in India as well as for the comparison of six (sub)species: the smaller

the spatial range of communication, the steeper the bees' dialect slope. The bees' dialects determine their spatial communication accuracy because a given interval of waggle duration corresponds to a smaller or a larger interval of flight distances depending on the slope of the distance–waggle duration function (steeper = more precise; figure 1b). Certainly, the absolute spatial precision of a honeybee's waggle dance does not only depend on the distance dialect. It is a product of the combined (im)precisions of both its distance and its direction component. However, the direction (im)precision of dances (i.e. the degree of angular scatter of wagging directions around the average bearing) is largely subject to behavioural constraints [43] and has been shown to be similar across honeybee species [44]. Therefore, differences between honeybees in the spatial precision of their dances are mostly driven by variation in their distance dialects. Providing that the steepness of the dialect is interpretable as precision, our data support the hypothesis that dialects are shaped by natural selection to maximize communication precision given the distances the bees typically forage at and which need to be communicated by the dance. An important point in favour of this scenario is that we can exclude the possibility that the dialects are a result of different physical constraints. On the one hand, body size proportionally scales with eye size among workers of different honeybee species [45]. Eye size determines physiological properties such as light sensitivity and spatial resolution [46], and might thus affect the visually driven odometer. On the other hand, it is established that foraging ranges of bees are generally positively correlated to body size [47]. However, among the three honeybee species in India, it was not the smallest species, A. florea, but the medium-sized species, A. cerana, that had the steepest distance dialect and the smallest foraging range. The most compelling evidence that neither distance dialects nor foraging ranges are affected by physiological eye or body size constraints is the variation observed between populations of the same species. Temperate and tropical subspecies of both A. mellifera and A. cerana show divergent dialects and foraging ranges, with tropical populations having around four times steeper dialect slopes and clearly smaller foraging ranges than their temperate relatives (figure 4). Finally, the fact that distance dialects can diverge between populations of the same species or converge between different species rules out another possible explanation for distance dialects: they are not the result of random divergence over time (i.e. neutral evolution).

The optimum distance dialect of a given honeybee population will depend on several ecological variables, including the bees' energetic needs, the spatio-temporal distribution of resources or the competition for these resources. When colonies need to hoard large honey stores to survive periods of cold or draught, or when resources are scarce, the required habitat area can be large. These bees need a dialect that allows for a large communication range. Examples are temperate populations of A. mellifera and A. cerana which regularly communicate and forage over distances of more than 5–10 km from the nest [22,31–33,37], and which convergently evolved low distance dialect slopes. In tropical regions, the bees often find enough resources in their closer surroundings so that they do not need to fly far. Then, especially when the resources are clumped in patches and hard to locate, precise guidance of recruits will be advantageous, and selection will favour honeybees with steep

distance dialects. Where the co-occurrence of multiple honey-bee species creates high competition [48,49], species with more precise dialects might additionally benefit from being faster in recruiting to and exploiting rewarding food sources closer to their nests. Other species could avoid competition by foraging over larger areas, which in turn requires having lower dialects. Intriguingly, between the three sympatric species *A. cerana*, *A. florea* and *A. dorsata*, the same qualitative foraging range differences were reported for Sri Lanka (*cerana*: approx. 600 m, *florea*: approx. 900 m, *dorsata*: 1000 m, maximum ranges [23]), Thailand (*cerana*: 905 m, *florea*: 1323 m, *dorsata*: 3810 m, 95% ranges [20]) and India (*cerana*: 980 m, *florea*: 2570 m, *dorsata*: 2910 m, maximum ranges). It is conceivable that variation in the spatial foraging strategies of these honeybees contributes to their coexistence [50]. In turn, coexistence of honeybee species might affect the dialect they evolve.

Considering the ecological adaptation of distance dialects, a remaining question is that of the spatial scales of dialect variation within taxonomic units. Many honeybee (sub)species extend over large areas, encompassing regions of contrasting landscapes [7]. There are cases where admixed populations extend over relatively strict natural borders of two adjacent landscape types (e.g. savannah/forest transitions [51]). Honeybees living in human-modified landscapes (e.g. the bees from Bangalore) are facing very different environments compared to those that live in large adjacent natural areas. Food availability and hence the required foraging range are likely to be very different in such contrasting landscapes. And different looks of the terrains might create contrasting 'visual currencies' so that a visually driven odometer adapted to one landscape type might not fit to the other. It would be interesting to test whether there is variation in the distance dialects among honeybee colonies originating from various sites along landscape transition zones. Given the widespread beekeeper-mediated hybridization of geographically distant lineages of the western honeybee, *A. mellifera* [7,52], an applied research question is to what extent the dance dialects of this species are getting disconnected from the environments to which they are adapted. A large proportion of honeybee colonies used by beekeepers today are hybrids of several subspecies (e.g. *A. m. mellifera*, *A. m. ligustica*, *A. m. carnica*), whose original distance dialects were described as differing greatly [19]. Von Frisch [1] showed that foragers of Carnolian *A. m. carnica* and Italian *A. m. ligustica* forced to live in mixed-subspecies colonies misunderstand the distance signal of the interspecific hive mates. It is conceivable that this problem remains when the bees are genetically mixed, which might have adverse consequences for the functioning of their spatial communication system [29].

In the history of research on the honeybee waggle dance, the most controversial question was whether the spatial information encoded by dancing bees is functional, i.e. (i) whether dance followers can actually use the spatial information to fly towards an advertised location in the field and (ii) whether the communication of direction and distance of a resource—in addition to the mere alert about its presence, type and quality—enhances a honeybee colony's fitness. Today there is no doubt that the general answer to these questions is yes. On the one hand, the flight paths of dance followers could be tracked using harmonic radar technique, demonstrating that the spatial information of the dance indeed affects the recruits' flight behaviour in the field [53]. On the other hand, studies using honeybee colonies in which the spatial information of the dance was either experimentally disrupted or undisturbed found that spatial communication can enhance both the quantity and the quality of the resources collected by honeybee colonies [54,55]. In studying distance communication dialects between honeybee species, we looked at the variation of the waggle dance that arises on evolutionary timescales. Our findings indicate that dance dialects exist both between and within honeybee species, and that they are adapted to be as precise as possible given the bees' foraging ranges, which in turn vary greatly subject to ecological factors. This highlights the role of the environment in shaping the waggle dance and provides further evidence for the evolutionary significance of spatial communication in honeybees.

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