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RESEARCH ARTICLE

Vector-based and landmark-guided navigation in desert ants inhabiting landmark-free and landmark-rich environments

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SUMMARY

Two species of desert ants – the North African *Cataglyphis fortis* and the central Australian *Melophorus bagoti* – differ markedly in the visual complexity of their natural habitats: featureless salt pans and cluttered, steppe-like terrain, respectively. Here we ask whether the two species differ in their navigational repertoires, in particular, whether in homing they place different emphasis on their vector-based and landmark-based routines. In trying to answer this question, we applied the same experimental paradigms to individual foragers of either species on either continent: training and/or testing with and/or without artificial landmark arrays. We found that the open-terrain species *C. fortis* runs off its (path integration) home vector much more readily even in unfamiliar landmark settings than the cluttered-terrain species *M. bagoti*. These data support the hypothesis that *C. fortis* has a higher propensity to rely on vector-mediated navigation, whereas in the same experimental situations *M. bagoti* more easily switches to landmark-guided behaviour. In the actual navigational performances, such species-specific propensities are most likely shaped by environment-dependent individual experiences.

Key words: desert ant, *Cataglyphis fortis*, *Melophorus bagoti*, path integration, landmark guidance, interspecific comparison.

INTRODUCTION

Cataglyphis fortis Forel 1902 and *Melophorus bagoti* Lubbock 1883 are two species of desert ants (subfamily Formicinae) that, on different continents, occupy the same general ecological niche as thermophilic scavengers (Wehner, 1987). They inhabit either featureless desert plains [in the case of the North African *C. fortis* (Dillier and Wehner, 2004)] or densely cluttered environments [in the case of the central Australian *M. bagoti* (Muser et al., 2005)]. Although the Saharan salt pans are only sparsely overgrown and are often devoid of any vegetation, cluttered Australian semi-deserts are characterized by irregular arrays of grass tussocks, small shrubs and loosely scattered trees. Contrary to the featureless *C. fortis* habitat, where the ants travel through barren terrain over long distances, *M. bagoti* foraging grounds provide many nearby and more distant landmarks used for navigation (Wehner, 2003; Kohler and Wehner, 2005; Narendra, 2007a; Narendra et al., 2007a; Narendra et al., 2008; Cheng et al., 2009). Hence, different navigational strategies might be favoured by species that have to find their ways in habitats that differ so strongly in their visual complexity.

Solitarily foraging desert ants are equipped with an amazingly rich navigational repertoire. In particular, *Cataglyphis* and *Melophorus* species are visually guided ants par excellence (Wehner, 2008; Cheng et al., 2009; Wehner, 2009). Path integration is the ant's major means of navigation, i.e. it is in use the entire time ants are foraging (Sassi and Wehner, 1997; Andel and Wehner, 2004; Knaden and Wehner, 2005). When an ant leaves its nest for the first time, path integration is its only means of navigation. Later, however, the ant becomes familiar with landmarks in its surroundings. Different studies with North African (Wehner and

Räber, 1979; Wehner et al., 1996; Bisch-Knaden and Wehner, 2003; Bregy et al., 2008; Merkle and Wehner, 2008) and central Australian desert ants (Kohler and Wehner, 2005; Wehner et al., 2006; Narendra, 2007a; Narendra, 2007b; Sommer et al., 2008; Graham and Cheng, 2009a; Graham and Cheng, 2009b; Wystrach et al., 2011), wood ants of holarctic regions (Nicholson et al., 1999; Graham and Collett, 2002; Collett et al., 2003; Fukushi and Wehner, 2004) and tropical ants (Macquart et al., 2006) have shown that ants are able to acquire and use rich navigational memories of landmark-defined places and routes.

The two species of ants examined in the present study were selected for the species-specific type of landscape in which they occur, because these two types define the two ends of a wide spectrum of more or less cluttered desert environments ranging from extremely landmark-poor to extremely landmark-rich landscapes. By selecting these two species (*C. fortis* and *M. bagoti*), we aim to determine whether the two species differ in how strongly they depend on their path-integration home vector (HV strategy) or on landmark guidance (LG strategy) when returning from a feeder, and whether potential differences are species-specific and/or environment-dependent traits.

A previous study on a North African *Cataglyphis* species, *C. bicolor* in this case, showed that individual ants can be trained to switch from vector-based to landmark-guided behaviour (Wehner, 1970). In this study, the ants were trained to a landmark-defined feeder. Although in the first training run the landmark-defined course and the vector-based course coincided, in subsequent runs the landmark-defined course leading to the feeder was made to always deviate from the vector-based course. In this competition paradigm, the ants first adhered to the vector-defined course, but after having

been exposed to this paradigm several times, they consistently chose the landmark-defined course (Wehner, 1970). This result, obtained in a *Cataglyphis* species that primarily inhabits low-shrub semi-desert areas, raised the question whether open-habitat or cluttered-habitat species would have a higher or lower propensity, respectively, to adhere to their vector-based strategy even if they were presented with unfamiliar landmark situations. Furthermore, drawing upon previous displacement experiments, we already know what happens when the two desert ant species considered in the present study are displaced from a feeder to a near-by point in their natural habitat. Upon release, *C. fortis* would run off its home vector completely and then start a systematic search around the fictive position of the nest (Wehner and Srinivasan, 1981), whereas *M. bagoti* would run off only approximately half the distance indicated by its home vector before starting to search (Narendra, 2007a). This difference between the two groups of displaced ants is not due to differences in the ants' path-integration strategy *per se*, as ants of either species cover the full feeder–nest distance when they perform their outbound and inbound runs within channels that block the view of the landmark panorama and provide a uniform, uninformative, but familiar visual surround [at least for distances of up to 12 m; *C. fortis* (Sommer and Wehner, 2004) and *M. bagoti* (Narendra, 2007a; Narendra et al., 2007a)]. Hence the difference in travelling the full length of the home vector (in *C. fortis*) or not (in *M. bagoti*) must be dependent on the degree to which the two groups of displaced ants relied on their HV strategy. After displacement, *C. fortis* encountered a visual scene that was nearly identical to that in its nest area, but *M. bagoti* was confronted with an unfamiliar landmark situation. The amount of the home vector actually paid out by *M. bagoti* after displacement to novel territory is certainly not a constant [44% in Narendra's study (Narendra, 2007a)], but most likely depends on the amount of mismatch between the landmark panoramas at the training and test site. When wood ants *Formica japonica* (Fukushi, 2001; Fukushi and Wehner, 2004) and rainforest ants *Gigantiops destructor* (Beugnon et al., 2005) were tested in somewhat similar experimental paradigms, they relied on their home vector even less than *M. bagoti* did in Narendra's displacement experiments (Narendra, 2007a). These results fall into line with the negative correlation between landmark density and the proportion of the home vector run-off: a high density of highly conspicuous landmarks prevails in the woodland environments of the two latter species.

Taken together, the data referred to above raise the question whether the differences found in the various species are species-specific traits, and/or whether they are due to the different experiences that the individuals have made in their more-or-less cluttered and landmark-dominated foraging terrains. We try to answer this question in desert ants by testing *C. fortis* and *M. bagoti* on the two continents on which they occur – (northern) Africa and (central) Australia, respectively – under identical experimental conditions. To accomplish this task, we had to select a *M. bagoti* colony that was located in an open desert field devoid of any nest- or route-defining landmarks, and had to subject the *M. bagoti* foragers of this colony located in a *C. fortis*-like habitat to the very same experimental paradigm as *C. fortis* in its natural habitat. If such ideal experimental conditions could be established, and if differences were found in how *C. fortis* and the open-field *M. bagoti* relied on HV mediated or LG mediated homing behaviour, then these differences must be species-specific traits, as the open-field *M. bagoti* foragers have never before experienced the cluttered type of environment that is characteristic for this species.

In detail, *C. fortis* and *M. bagoti* foragers were trained to an artificial feeder and the homeward runs were subsequently recorded either in the training field itself or in an unfamiliar test field. In the 'symmetric paradigm', the landmark situation was the same in the training and test conditions; in both cases, the ants were provided with either no landmarks at all [–/–] or a conspicuous array of black cylinders [+/+]. In the 'asymmetric paradigm', the ants experienced the landmark array either only during their inbound (homing) runs [–/+] or only during their outbound (foraging) runs [+/-].

MATERIALS AND METHODS

Experimental sites and ant species

Experiments with *C. fortis* were carried out in a flat, featureless salt pan near Maharès, Tunisia, between July and September 2006 and in June 2008. *Melophorus bagoti* was studied ~18 km south of Alice Springs, Australia, between December 2007 and January 2008 and between December 2008 and March 2009. Although the nests of *M. bagoti* are usually surrounded by grass tussocks, bushes and trees, in the present study we selected an open area (a hot-air balloon field near Alice Springs Airport) devoid of natural landmarks on the field such as trees and shrubs, so that landmark arrays could be established artificially and in the very same way as it was done in the experiments with *C. fortis* in North Africa. The balloon-launching field was surrounded by a skyline of trees of fairly uniform height. The nest was towards one edge of the oval field. The panorama could provide some additional compass information to the ants.

Experimental setup

Foraging ants were trained to shuttle back and forth between the nest and an artificial feeding site (containing cookie crumbs or mealworms) at a distance of 10 m from the nest. Ants were trained either within a landmark alley or in the absence of any landmarks. The landmark alley consisted of black cylinders (0.15 m high and 0.10 m wide), which were arranged in two rows flanking the route from the nest to the feeder (Fig. 1A,B). In order to prevent ants from learning that the end of the alley marked the goal positions, the alley was extended beyond both the nest and the feeder. The within-row and between-row distances between every two cylinders were 1 m. One row of cylinders was shifted by 0.5 m relative to the other, so that each cylinder of one row faced the gap between two cylinders of the other row. Ants that had arrived at the feeder were marked with a day-specific colour dot on their gaster. After at least one day of training, the homing behaviour of the ants was studied either in the training area or in a distant and landmark-free test field that contained a rectangular grid of lines (mesh width: 1 m) painted on the floor (in the case of *C. fortis*) or established by strings fixed by tent pegs (in the case of *M. bagoti*). The test site was also located in the open balloon field for *M. bagoti*. The inbound runs of ants captured at the feeder were recorded on graph paper until the ants started to perform search loops (in the test field) or until they entered their nest (in the training area). First, we compared the home runs performed by ants in the landmark-free training (home) area [–] with the inbound runs of displaced ants trained and tested without landmarks [–/–]. This comparison should show how 'landmark-free' the training area actually was. In the symmetric experimental paradigm, ants were trained and (after displacement) tested with the same landmark conditions: either without [–/–] or with the artificial landmarks [+/+]. In the asymmetric paradigm, ants were either trained without landmarks and subsequently tested with the landmark alley [–/+] or *vice versa* [+/-]. This paradigm allowed us to study the influence that the landmarks would have on the

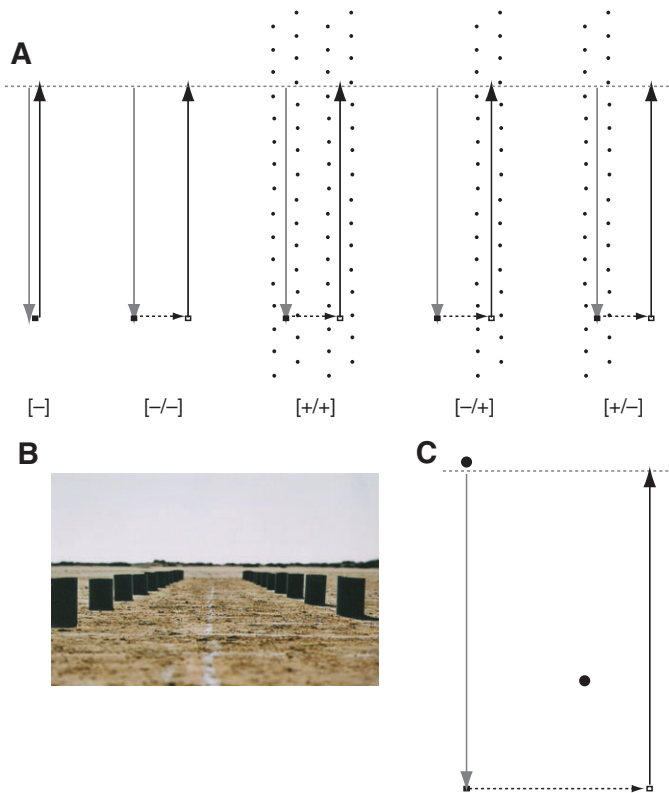


Fig. 1. Experimental setup. (A) Different training and test situations. The ants were tested either under symmetric ($[-/+]$ and $[+/-]$) or asymmetric conditions ($[-/+]$ and $[+/-]$). Additionally, the homeward runs in the training field were recorded ($[-]$). The filled square depicts the feeding site in the training area and the point of release in the test field is marked with the open square. The dashed line indicates the level of the nesting site in the training area or that of the fictive nest in the test field. The nest–feeder distance is 10 m and the within-row and between-row distances of the cylinders (black circles) are 1 m. (B) Photo of the landmark alley used in the experiment with *Cataglyphis fortis* at the field site near Maharès (Tunisia). (C) Experimental paradigm with the nest-defining landmark. The nest–feeder distance is 15 m (dashed line indicates the level of the nest), the filled square depicts the feeding site in the training area and the point of release in the test field is marked with the open square. Tests were performed with the landmark (black circle) placed 3 m lateral to the feeder–nest line at one-third of the feeder–nest distance.

ants' reliance on their path integrator. We only tested ants that, in their inbound runs, still carried their food item. This assured us that all ants exhibited high return-to-nest motivational states. Each ant was tested only once. To assure that ants could derive correct compass information from the polarization gradients in the sky (Wehner and Müller, 2006), tests were performed only under open-sky conditions.

An experimental paradigm that had formerly been used on *C. fortis* (Bregy et al., 2008) was also conducted for comparison on *M. bagoti*. In this paradigm, the nest entrance was marked by a black cylinder (0.41 m high and 0.45 m wide) and the artificial feeder was placed 15 m distance from the nest and cylinder (Fig. 1C). Again, foraging ants were trained in their home area for at least one day before their inbound runs were recorded in the distant test field. During these tests, the nest-defining landmark was placed at a position lateral to the ants' vector course; during the control tests, the nest-defining landmark was placed at the nest position (for details, see Bregy et al., 2008). We added a control for innate

(untrained) tendencies to head to the landmark. Ants in this group were trained to home without any landmarks. In the test, the experimental landmark was added lateral to the ants' vector course.

Data analyses

The recorded paths were digitized using a graphics tablet (Digikon 3, Kontron, Eching, Germany) and GEDIT Graphics Editor and Run Analyser (MbasoSoft GEDIT for Windows, v. 2.5, 1995; written by N. Antonsen, Zürich, Switzerland). Paths were later analysed from the release point (RP) up to the point where the ants started their searches for the nest (search point, SP), i.e. where they changed their direction by at least 60 deg and did not revert to the previous direction for the next 3 m, or when they crossed their previous path after this change had occurred (360 deg loop). We used an angle larger than 30 deg (Merkle et al., 2006) because the ants would often turn by that amount but then keep running in the home direction. We used an angle less than 180 deg [specified by Bregy et al. (Bregy et al., 2008)] because it was clear that the ants sometimes started searching with a turn less than that amount. We used a distance after turn longer than the 0.5 m specified by Narendra (Narendra, 2007a) because the runs of the ants, especially those of *M. bagoti*, exhibited tortuousness and would sometimes 'wiggle' for >0.5 m while heading in the home direction. In the end, we arrived at a criterion that accords well with subjective judgements. A random subset of runs was judged by eye by a researcher blind to the experimental conditions. The Pearson correlation coefficient between the formal analyses and eyeball judgement on d_0 (location of the start of the search) was 0.8 (except for a single ambiguous run, it would have been 0.9).

Beeline distance d_0 [i.e. the projection of the RP–SP line (d) onto the nest–feeder line] and straightness index [ratio between d and the total path length (l), calculated by GEDIT] were further analysed statistically by using the statistical software R (version 2.12.1). Because not all data were distributed normally (Kolmogorov–Smirnov test), the non-parametric Kruskal–Wallis test with multiple comparisons (*post hoc*) was applied. The variances of d_0 were additionally analysed with the O'Brien's test for homogeneity of variances (SAS, 2002). The directions of the runs were determined at half the nest–feeder distance (5 m) by measuring the angles from the recorded paths. To do this, the angle between the true homeward direction (feeder–nest line) and the direct line between the point of release and the point of intersection (between the homing path and a circle with a radius of 5 m drawn around the point of release) was measured. Oriana 3, statistical software for circularly distributed data (Batschelet, 1981), was used throughout. The directionality of the data was examined using the Rayleigh test (all $P < 0.001$) and Mardia–Watson–Wheeler tests were applied to detect differences between the mean directions and/or angular variances of two experimental groups. Because of multiple pairwise testing, a Bonferroni correction was applied. We used a corrected significance level of $P = 0.003$. A given mean direction was considered to deviate statistically from the true homeward direction if the latter direction lay outside the 95% confidence interval of the sample mean. Because no group tested deviated from the home direction, we can conclude that differences detected by the Mardia–Watson–Wheeler test are due to the angular variances. The accuracy of the runs was determined by comparing the angular means of the test groups, whereas the directional scatter of the homing runs was analysed by comparing the angular variances of the groups. The inbound runs of *M. bagoti* ants faced with the nest-defining landmark were analysed graphically. In order to compare the proportion of ants heading towards the landmark, a Fisher's exact test was performed.

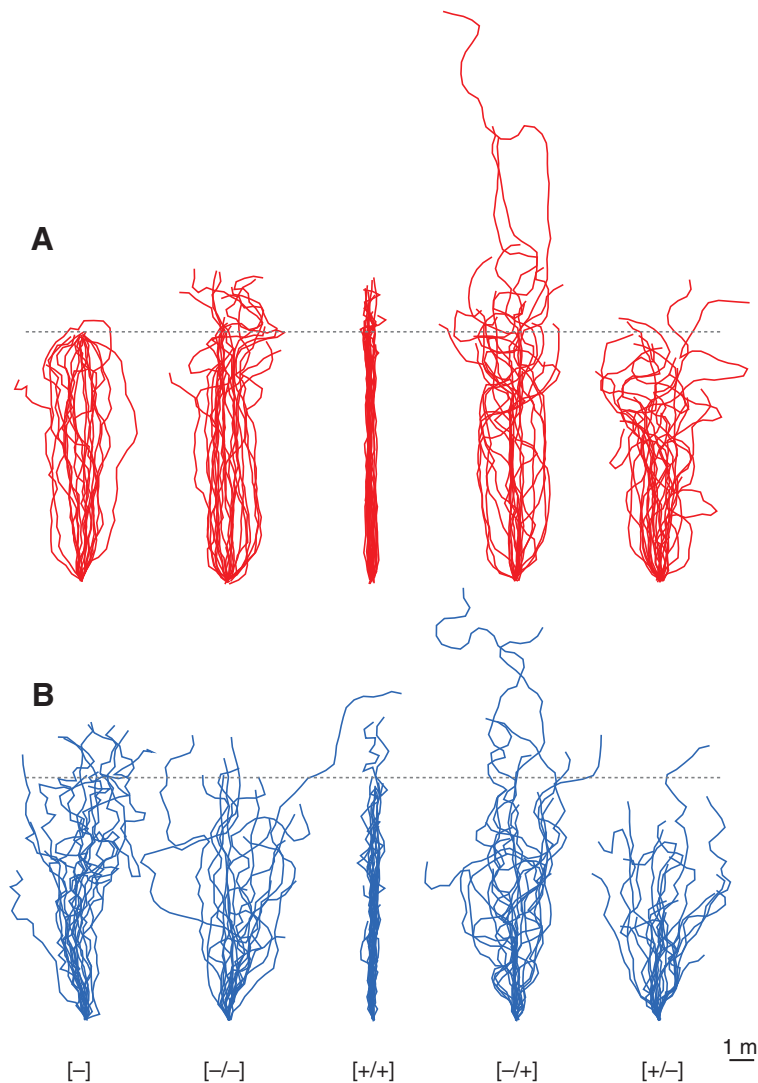


Fig. 2. Homing runs of *C. fortis* (A) and *Melophorus bagoti* (B) ants under the different test conditions (training situation/test situation). The dashed line indicates the level of the fictive nest position. The homing runs were clipped at the start of the searching behaviour. *Cataglyphis fortis*: $N=20$ ants [-], $N=23$ ants [-/-], $N=22$ ants [+/+], $N=22$ ants [-/+], $N=25$ ants [+/-]. *Melophorus bagoti*: $N=18$ ants [-], $N=21$ ants [-/-], $N=22$ ants [+/+], $N=20$ ants [-/+], $N=23$ ants [+/-].

As the main focus of this paper is to compare two species of desert ants, *C. fortis* and *M. bagoti*, intraspecific as well as interspecific comparisons were performed in all cases.

RESULTS

Intraspecific comparison

Cataglyphis fortis

Homing runs of *C. fortis* returning to their nest in the landmark-free training field ([-] series in Fig. 2) and running towards the fictive nest position in the landmark-free test field ([-/-] series in Fig. 2) did not differ significantly with regard to all parameters tested, i.e. the beeline distance d_0 , the straightness index and their directional distribution (see Figs 3–5). When ants performed their training as well as their home runs within the familiar landmark alley [+/+], the inbound test runs were well channelled within this landmark corridor (Fig. 2). Ants returning within the landmark array ran along the midline of the alley of cylinders. The angular variance of the runs was much smaller in the [+/+] than in the [-/-] situation ($P<0.003$; Fig. 5). Although d_0 was not significantly different between the two symmetric paradigms (Fig. 3), the index of straightness clearly was ($P<0.05$; Fig. 4). Among the two symmetric conditions and [-], the variances of d_0 also differed significantly (O'Brien's test, $F_{2,62}=10.90$, $P<0.001$; Fig. 6).

Next, we asked how ants behaved when they experienced unfamiliar landmarks in the test situation *versus* when the habitual

landmarks were missing (asymmetric paradigm). Ants walking back within an unfamiliar landmark alley [-/+] had significantly longer homeward runs than observed in the symmetric [-/-] case ($P<0.05$; Figs 2 and 3). The indices of straightness were significantly lower ($P<0.05$; Fig. 4), whereas the directional distributions did not differ significantly between the two conditions (Fig. 5). Compared with ants returning within the known alley [+/+], the homing ants tested with the unfamiliar alley [-/+] did differ significantly in indices of straightness ($P<0.05$; Fig. 4), directional distribution ($P<0.003$; Fig. 5) and d_0 ($P<0.05$; Fig. 3). Most of the ants that had learnt the landmark alley during training did not pay out their entire inbound vector when subsequently tested in the absence of the landmarks ([+/-] series in Fig. 2). They aborted their homing paths prematurely: the beeline distance d_0 was significantly shorter than when the landmarks were present ($P<0.05$; Fig. 3), and the data points were distributed over a larger range (Fig. 6). The homeward runs were also shorter when compared with those in the [-/-] situation ($P<0.05$; Fig. 3). In the absence of the known alley, the ants performed quite tortuous runs, so that the index of straightness was significantly lower than in the [+/+] and [-/-] cases (both $P<0.05$; Fig. 4) cases. The interindividual variation in homing direction was increased when the habitual landmarks were absent in the tests ([+/-] condition, $P<0.003$). That is, the directional scatter of the runs was bigger than in the [+/+] case, but not bigger than in the [-/-] case

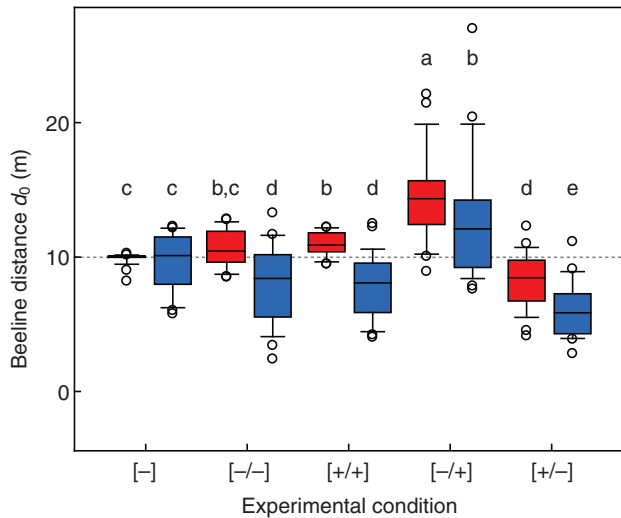


Fig. 3. Beeline distance d_0 travelled by *C. fortis* (red) and *M. bagoti* (blue) under different test conditions (training situation/test situation). The point of release (or the feeder in the training situation) is placed at 0m and the fictive nest position (or the nest in the training situation) is reached at 10m (dashed line). Box plots show medians, 25 and 75% quartiles, whiskers (90th and 10th percentiles) and outliers (circles). Intraspecific and interspecific comparisons were performed with non-parametric Kruskal–Wallis analyses and multiple comparisons. Lowercase letters indicate significant differences ($P < 0.05$) between the groups. Groups with same letters are not significantly different. For sample sizes see Fig. 2.

(Fig. 5). The variances in d_0 were larger in the asymmetric conditions than the symmetric conditions, with O'Brien's test for homogeneity of variances showing significant heterogeneity of variance across the five conditions ($F_{4,107} = 7.51$, $P < 0.001$).

Melophorus bagoti

As in *C. fortis*, the inbound runs of *M. bagoti* returning either in the training [-] or in the test field [-/-] did not differ significantly in their angular distributions (Fig. 5), but their beeline distances d_0 were shorter in the [-/-] than in the [-] situation ($P < 0.05$; Fig. 3) and the indices of straightness differed ($P < 0.05$; Fig. 4). In contrast to *C. fortis*, although displaced *M. bagoti* exhibited somewhat higher interindividual differences in d_0 than *M. bagoti* that were not displaced (Fig. 6), the differences in variances across the symmetric conditions were not significant (O'Brien's test, $P = 0.222$). In large part, this came about because the variance in the training runs of *M. bagoti* was large in contrast to that of *C. fortis* (discussed further in Interspecific comparison). The d_0 of ants trained and tested with landmarks [+/+] did not differ significantly from the [-/-] condition (Fig. 3). However, the angular variance parameter and the index of straightness differed between the two situations (for index of straightness see Fig. 4, $P < 0.05$). As in *C. fortis*, the directional scatter of the homing runs was much smaller when the ants could return within the known landmark array [+/+] than when they could not [-/-] ($P < 0.003$; Fig. 5).

In the asymmetric paradigm [-/+], the inbound runs of the *M. bagoti* foragers were significantly longer ($P < 0.05$; Figs 2 and 3) and the indices of straightness were significantly lower ($P < 0.05$; Fig. 4) than in the [-/-] case, whereas the angular variances did not differ significantly between these two test situations (Fig. 5). Compared with the homing runs of ants tested within the known landmark alley [+/+], [-/+] ants did differ significantly in their d_0 ($P < 0.05$;

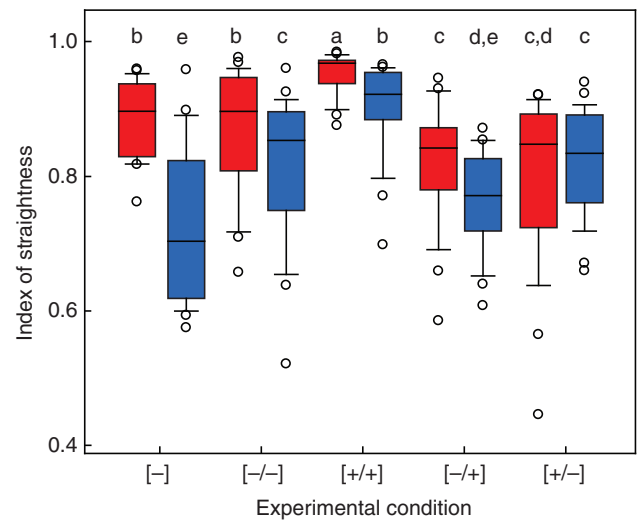


Fig. 4. Index of straightness of *C. fortis* (red) and *M. bagoti* (blue) foragers. For explanation of box plots and sample sizes see Figs 2 and 3. Lowercase letters indicate significant differences ($P < 0.05$) between the groups. Groups with same letters are not significantly different. The values for the index of straightness range from (nearly) 0 to 1, where 1 indicates a perfectly straight run.

Fig. 3), indices of straightness ($P < 0.05$; Fig. 4) and angular distributions ($P < 0.003$; Fig. 5). In the absence of the known landmark alley [+/-], the homing runs were significantly shorter than in the presence of the alley [+/+] ($P < 0.05$; Fig. 3). Furthermore, the runs were more tortuous ($P < 0.05$; Fig. 4) and showed a higher angular variance than those recorded in the presence of the familiar landmarks ($P < 0.003$; Fig. 5). That is, the directional scatter of the runs was bigger in the absence of the known landmarks [-/-] than in the presence of them [+/+]. The d_0 were also significantly shorter in the absence of the familiar alley [+/-] than in the [-/-] case ($P < 0.05$; Fig. 3). The [+/-] and [-/-] situations did not differ in their straightness indices (Fig. 4) or directional distributions (Fig. 5). The variances in d_0 were higher in the asymmetric conditions than in the symmetric conditions, especially in the [-/+]. Across the five conditions, variances were heterogeneous (O'Brien's test, $F_{4,99} = 2.65$, $P = 0.038$).

Interspecific comparison

Even though qualitatively *C. fortis* and *M. bagoti* behaved in quite similar ways under all conditions described above, there were marked quantitative interspecific differences. In general, the homing trajectories spread over a much larger range in *M. bagoti* than they did in *C. fortis*. This was born out especially by the parameters d_0 and index of straightness (d/l). The directional distributions did not differ between the two species and therefore will not be mentioned any more.

Although in the [-] control situation, 90% of the *C. fortis* ants returned to the nest directly, in *M. bagoti* this was the case in only 11% of the tested individuals. Only two out of 20 *C. fortis* ants started their search loops before they entered the nest. In *M. bagoti* this was the case in 16 out of a total of 18 ants. Even though in the training inbound runs ([-]) d_0 did not differ between the two species (Fig. 3), the points where the ants started the systematic search were concentrated much more strongly in *C. fortis* than in *M. bagoti* (Fig. 6). The standard deviation in d_0 in the [-] condition was 4.7 times bigger in *M. bagoti* than in *C. fortis*. Furthermore, in *C. fortis*

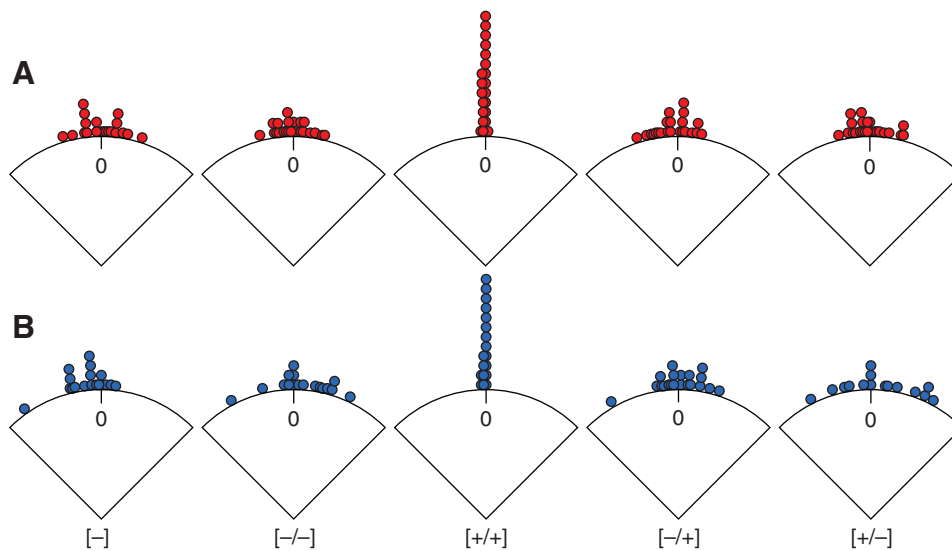


Fig. 5. Angular distribution of the homing runs of *C. fortis* (A) and *M. bagoti* (B) at half the nest–feeder distance (5 m). 0 deg indicates the homing direction. Each circle represents one ant. For sample sizes see Fig. 2.

the home runs ([-]) were significantly straighter than in *M. bagoti* ($P < 0.05$; Fig. 4).

When trained and tested without the landmark alley ([-/-]), *M. bagoti* started its search loops much earlier than *C. fortis*. Its d_0 was significantly shorter than that of *C. fortis* ($P < 0.05$; [-/-] in Fig. 3). In addition, as shown by the distribution of the end points of the homing runs (Fig. 6), the interindividual variation in *M. bagoti* was much higher than in *C. fortis*. The latter result is confirmed by series [-/-] (Fig. 2), in which the index of straightness differed significantly between the two species ($P < 0.05$; Fig. 4): the homeward runs of *C. fortis* were much straighter than those in *M. bagoti*.

Even in the [+/>] symmetric paradigm, in which ants were trained and tested within the landmark alley, *M. bagoti* stopped its inbound runs significantly earlier than *C. fortis* ($P < 0.05$; Fig. 3). As in the [-] and [-/-] situations, the [+/>] runs of *M. bagoti* were distributed over a much larger area and correspondingly exhibited significantly smaller indices of straightness ($P < 0.05$; Fig. 4) than *C. fortis* [+/>] runs.

Let us now turn to the asymmetric experimental paradigm and ask how the two species differed in their reliance on their path-integration vector even if the landmark information in the test differed from that in the training situation. When faced with an unknown landmark alley ([+/>]), both species exhibited longer home runs than in the [-/-] situation. *Cataglyphis fortis* walked for longer distances ($P < 0.05$; Fig. 3) in a straighter line ($P < 0.05$; Fig. 4) than *M. bagoti*. Likewise, in the [+/>] case, d_0 was significantly longer in *C. fortis* than in *M. bagoti* ($P < 0.05$; Fig. 3).

How do the two species behave when a landmark defining the nest itself rather than the route leading to the nest occurs earlier on an ant's path-integrated home run than normally? This topic has been studied in *C. fortis* (Bregy et al., 2008). Here we present the corresponding data for *M. bagoti* and compare both species in how they cope with this competitive situation between path integration and landmark guidance. When the ants were faced with a nest-defining landmark after they had run off only one-third of their home vector, 15 out of 20 *M. bagoti* headed towards the landmark (Fig. 7). For comparison, only seven out of 15 ants in Bregy et al.'s experiment (Bregy et al., 2008) headed towards the landmark, a significantly smaller proportion (Fisher's exact test, $P = 0.025$). In the control situation (landmark located at the nest position), all tested

ants in the present study ran towards the fictive nest position (see Fig. 7). In *M. bagoti*, we could not see an innate bias to head towards the landmark when trained without the landmark and tested afterwards with the unknown nest-defining landmark (data not shown).

DISCUSSION

How do two species of ants naturally inhabiting either landmark-free (desert plain) or landmark-rich (steppe-like) environments differ in their navigational repertoire, in particular, in how much they rely on their home vector *versus* landmarks, including searching for landmarks as guides? This is the central question we have addressed in the present study. We already know that landmark guidance (LG-mediated behaviour) can take precedence over fully paying out the home vector (HV-mediated behaviour) (Wehner, 2008; Cheng et al., 2009; Wehner, 2009), and that even when this is the case, the path integrator keeps running in the background (Sassi and Wehner, 1997; Andel and Wehner, 2004; Knaden and Wehner, 2005). It is neither switched off nor reset whenever the ant is foraging. Resetting occurs only after the ant has returned to the nest and vanished into it (Knaden and Wehner, 2006). Now we ask whether the switch from HV- to LG-mediated behaviour occurs more readily in *M. bagoti*, living in cluttered environments, than in *C. fortis*, living in open environments.

Do *C. fortis* and *M. bagoti* put different weights on their HV and LG routines? Does one species perform better than the other in path integration? Do they exhibit species-specific adaptive specializations (*sensu* Shettleworth, 2010)? To address these questions, we had to select colonies of both species that were located in the same type of habitat, in our case in bare desert environments devoid of any reliable landmark cues. This was easy for *C. fortis*, because such featureless terrains define the typical habitat of this species (Dillier and Wehner, 2004), but it was difficult for *M. bagoti*, a species normally inhabiting densely cluttered desert habitats, which are rich in grass tussocks and loosely scattered trees (Muser et al., 2005; Cheng et al., 2009), i.e. rich in visual cues that have previously been shown to be used as navigational aids (Kohler and Wehner, 2005; Narendra 2007b; Narendra et al., 2007b; Sommer et al., 2008; Graham and Cheng, 2009a; Graham and Cheng, 2009b; Wystrach et al., 2011). The main result was that ants of the two species, which had their colonies in the same kind of landmark-free environment

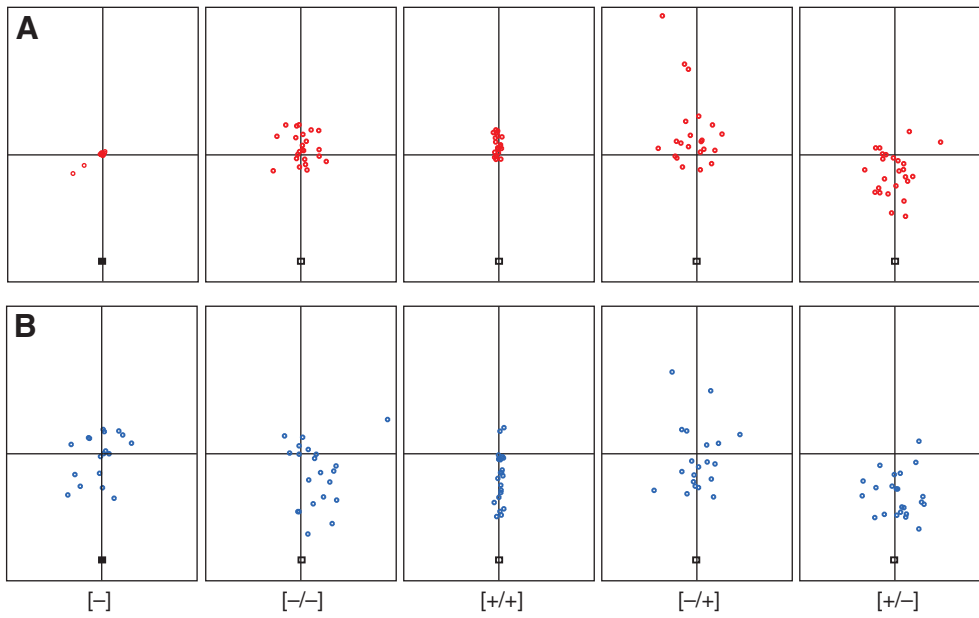


Fig. 6. Positions at which *C. fortis* (A) and *M. bagoti* (B) started their systematic search runs (for entire homing runs see Fig. 2). Homing runs of ants in their training (home) area ([-]), in the symmetric paradigm ([-/-] and [+/+]) and in the asymmetric paradigm ([-/+]) and [+/-]. The filled square indicates the feeding site in [-] and the open square depicts the fictive feeding site in the four conditions on the test field. The intersections of the horizontal and vertical lines mark the nest ([-]) or the fictive position of the nest in the test field. The nest–feeder distance is 10 m. For sample sizes see Fig. 2.

and were tested under the same experimental conditions, exhibited marked differences in their homing performances.

Cataglyphis fortis adhered much more strongly and consistently to its home vector than *M. bagoti*. Differences were found when foragers of the two species performed their homeward runs in their natural home-range (training) area, which was, in the present experimental setup, devoid of any landmarks (see series [-] in Fig. 2). First, although 90% of *C. fortis* ants returned straight to the nest and thus fully ran off their home vector, nearly 90% of the *M. bagoti* workers broke off their direct homebound paths long before they had reached the nest entrance. Second, these breaking-off points were distributed over a much larger area in *M. bagoti* than in *C. fortis* (Figs 3 and 6). Both these points suggest poorer performance in *M. bagoti*, perhaps coupled with a stronger tendency to switch from HV-based navigation to another mode, such as systematic searching. Third, *C. fortis* ran home along a rather straight line, but *M. bagoti* returned to its nest in a tortuous way (Fig. 4). Tortuous paths may indicate poor performance in path integration. But they may also indicate scanning or searching behaviour, a tendency to use another strategy than path integration. When the usual scene between their feeder and nest is changed, *M. bagoti*'s paths become more tortuous (Wystrach et al., 2011). Taken together, this unexpectedly poor performance and poor reliance of *M. bagoti* on its home vector even within its training (home) area ([-]) is all the more surprising, as the ants had not been disturbed at all by any displacement or experimental handling process, they just returned from the feeder to the nest on the very same desert ground over which they had performed their normal foraging journeys, facing the same accustomed contextual panoramic landmarks. The panorama of trees for this nest of *M. bagoti* was distant and could not be used to pinpoint the nest location, so the ants of this nest had to rely especially on path integration. In this context, it is worth mentioning that during repeated training the ants' inbound runs do not become more accurate (Narendra et al., 2007a; Merkle and Wehner, 2009).

When *C. fortis* has run off its home vector, it starts to search for nest-defining landmarks (Knaden and Wehner, 2005) and, if such landmarks are not available, switches on its systematic search routine and searches for the (in this case, fictive) position of the nest (Wehner

and Srinivasan, 1981). In some conditions in our study, *M. bagoti* started its search for familiar landmarks at an earlier stage of its home vector than *C. fortis*. The search behaviour itself has been studied less, but in a previous study, search patterns were evident after the ants had run off a fraction of the homing vector [see fig. 4 in Narendra (Narendra, 2007b)]. The ants in this case were displaced only a small distance (1.5 to 10 m laterally). The searching sometimes led the ants to the route corridor they had been trained to run along, and sometimes to take a course in the nest direction,

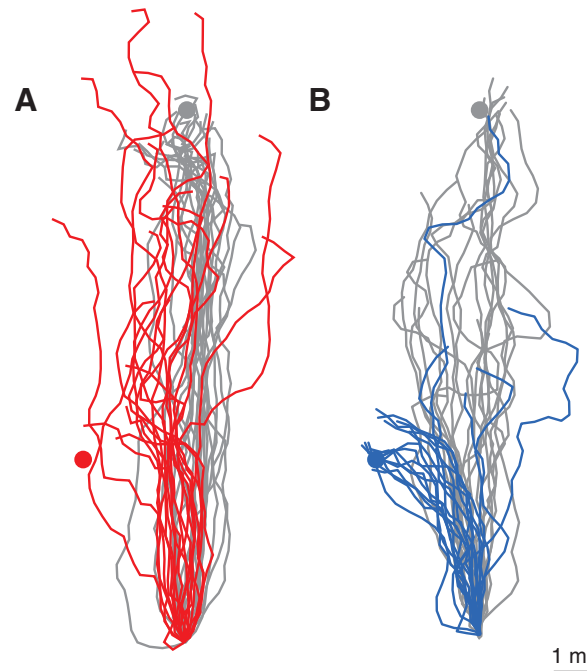


Fig. 7. Test runs of *C. fortis* (A) and *M. bagoti* (B). Foragers of both species were tested with the nest-defining landmark placed lateral of the vector course ($N=20$ ants; *C. fortis*, red runs; *M. bagoti*, blue runs) and at the training position ($N=20$ ants; control situation, grey runs). Data for *C. fortis* are adapted from Bregy et al. (Bregy et al., 2008).

presumably by means of view-based matching. In one recent study, the ants were allowed to run almost to their nest before being displaced to a distant test field (Schultheiss and Cheng, 2011). The ensuing search patterns then resembled those found in *Cataglyphis* in two major ways. The search was centred on the starting point, and it expanded with the passage of time. We thus interpret that breaking off of the home vector in both species as indicative of the ants' switching to landmark-based and/or systematic search strategies.

The significant differences apparent already in the ants' natural homing behaviour are further corroborated by the experimental series in which the homebound ants had been displaced to a test field, even by the series of the symmetric paradigm, in which the stimulus situations in the training and test fields coincided. When the ants were displaced from the feeder to a landmark-free test area (series [−/−] in Fig. 2), *C. fortis* behaved as if it were returning home in its habitual landmark-free training area. The homing distances did not differ between the [−/−] and [−] situations (Figs 3, 4 and 5), but *M. bagoti* undershot the 10 m homing distance by 16%. That it did not pay out its full home vector could indicate that the far-off landmark panorama of the balloon field or differences in the ground structure (Seidl and Wehner, 2006) had exerted some influence. In any case, the balloon field was the only area free of nest- and route-defining landmarks and inhabited by a colony of *M. bagoti*. But even if there had been slight influences of panoramic or ground-structure cues as indicated by the [−/−] paradigm, the relatively 'poor' performance of *M. bagoti* in the [−] paradigm, in which all these potential cues had been identical between test and training runs, is even more telling. Furthermore, the two species differed even when trained and tested within the landmark alley (series [+/+] in Fig. 2), which strongly channelled the ants' inbound runs, but did not specify the position of the nest (because the length of the alley exceeded the nest–feeder distance). Both species exhibited a strong 'channeling response' (Heusser and Wehner, 2002) by running along the midline of the alley and thus reaching the highest straightness indices of all experimental paradigms (Fig. 4). Certainly, landmark information helps to increase the ants' navigation efficiency (Benhamou, 2004; Narendra, 2007b). Nevertheless, the ants behaved in the species-specific ways described above. Again, although *C. fortis* ran off its 10 m path integration vector in the [+/+] situation as completely as in the landmark-free situation [−/−] (see also Merkle et al., 2006; Sommer and Wehner, 2004), *M. bagoti* stopped its runs governed by its home vector much earlier (Figs 3 and 6). The runs of *M. bagoti* were also far more variable in their length, and more tortuous (Fig. 4).

The differences between *C. fortis* and *M. bagoti* ants mentioned so far are further corroborated by employing site-specific rather than route-specific landmarks. When *M. bagoti* foragers experienced a nest-defining landmark after they had run off only one-third of their home vector, they immediately headed towards the landmark (Fig. 7), whereas *C. fortis* drifted only slightly, if at all, towards it and largely ran its home vector until it had finally reached the fictive position of its nest (Fig. 7).

Taken together, all experiments clearly show that there are distinct differences in the relative weights placed by the two species on their HV and LG navigational routines. The question of course remains whether these differences are species specific or environment dependent. In trying to tackle this question, we selected colonies that in both species were located on bare, flat desert ground devoid of any vegetation, stones or pebble, and had been at these locations for at least 3 years [for *C. fortis* (Dillier and Wehner, 2004), for *M. bagoti* (C.B., K.C. and R.W., personal observations)]. Given the

short life expectancies of the *Cataglyphis* and *Melophorus* foragers, a few days rather than weeks (Schmid-Hempel and Schmid-Hempel, 1984; Muser et al., 2005), all individuals used in our study had experienced featureless nest surroundings during their entire lifetimes, in the case of *M. bagoti* at least as featureless as was possible in this species. In any case, the clear differences between the two species even in the [−] paradigm (Figs 2–6), as well as the one documented in Fig. 7, indicate that species specificity is at least a strong component in the different significances accorded to HV and LG routines by the two species tested in this study, and in the differences in capabilities at estimating distance travelled (see also Narendra et al., 2007a). Of course it does not rule out the possibility that individually acquired traits 'ride on' species-specific ones, further canalizing or accentuating species differences. Hence the data presented in the current experimental study support the hypothesis that species such as *M. bagoti*, which inhabit cluttered, landmark-rich environments, have a higher propensity to abandon HV-mediated behaviour in favour of LG-mediated routines than is the case in open-space species such as *C. fortis*. Given the actual landmark scenery encountered by individual workers in their foraging terrain, one species might have evolved to switch more readily from HV to LG routines than the other species. The kind of competition experiment (Wehner, 1970) mentioned in the Introduction would provide a further paradigm to test this hypothesis. To guard against phylogenetic dependencies, such tests could most readily be performed within the genus *Cataglyphis* itself, in which some species – such as *C. fortis* – inhabit open terrain, whereas others – such as *C. bicolor* and *C. noda* – are bound to cluttered environments similar to those occupied by *M. bagoti*.

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