



**HAL**  
open science

# An intrinsic oscillator underlies visual navigation in ants

Leo Clement, Sebastian Schwarz, Antoine Wystrach

► **To cite this version:**

Leo Clement, Sebastian Schwarz, Antoine Wystrach. An intrinsic oscillator underlies visual navigation in ants. 2022. hal-04299190v1

**HAL Id: hal-04299190**

**<https://hal.science/hal-04299190v1>**

Preprint submitted on 26 Nov 2022 (v1), last revised 22 Nov 2023 (v2)

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

1                    **An intrinsic oscillator underlies visual navigation in ants**

2

3    Leo Clement<sup>\*</sup>, Sebastian Schwarz and Antoine Wystrach

4

5    Centre de Recherches sur la Cognition Animale, CNRS, Université Paul Sabatier, Toulouse

6    31062 cedex 09, France

7

8

9

10

11    \*Address of correspondence:

12    Léo Clément

13    Université Paul Sabatier

14    Centre de Recherches sur la Cognition Animale, CNRS

15    31062 Toulouse

16    Email: [clement.leo@univ-tlse3.fr](mailto:clement.leo@univ-tlse3.fr)

17    Phone: +33561558444

18

19

20

21 **Abstract**

22

23 Controlling behavior implies a constant balance between exploration – to gather information  
24 – and exploitation – to use this information to reach one’s goal. However, how this tradeoff is  
25 achieved in navigating animals is unclear. Here we recorded the paths of two  
26 phylogenetically distant visually navigating ant species (*Myrmecia croslandi* and  
27 *Iridomyrmex purpureus*) using a trackball-treadmill directly in their habitat. We show that  
28 both species continuously produce regular lateral oscillations with bursts of forward  
29 movement when facing the general direction of travel, providing a remarkable tradeoff  
30 between visual exploration across directions and movement areas. This dynamical signature  
31 is conserved across navigational contexts but requires certain visual cues to be fully  
32 expressed. Rotational feedback regulates the extent of turns, but is not required to produce  
33 them, indicating that oscillations are generated intrinsically. Learnt visual information  
34 modulates the oscillation’s amplitudes to fit the task at hand in a continuous manner: an  
35 unfamiliar panorama enhances the amplitude of oscillations in both naïve and experienced  
36 ants, favoring visual exploration; while a learnt familiar panorama reduces them, favoring  
37 exploitation through. The observed dynamical signature readily emerges from a simple  
38 neural-circuit model of the insect’s conserved pre-motor area known as the lateral accessory  
39 lobe, endorsing oscillations as a core, ancestral way of moving in insects. We discuss the  
40 importance and evolution of self-generated behaviors and how such an oscillator has been  
41 exapted to various modalities, behaviors and way of moving.

42

43 **Keywords:** insect navigation, time series analysis, intrinsic oscillator, lateral accessory lobe.

44

## 45 **Introduction**

46

47 Navigating through space implies both to acquire information (exploration) and to use this  
48 information to move in the correct direction (exploitation). A way to acquire information is to  
49 sample the environment by actively moving. Such ‘active-sampling’ is common across the  
50 animal kingdom spanning from invertebrates (Dittmar et al., 2010; Gomez-Marin et al., 2011;  
51 Lehrer, 1996) to vertebrates (Dawkins and Woodington, 2000; Otero-Millan et al., 2008;  
52 Wachowiak, 2011; Wesson et al., 2008; Wolf et al., 2017). However, active sampling entails  
53 movements that are typically different than moving toward the goal and thus require the animal  
54 to solve a tradeoff. Achieving a balance between sampling actions (exploration) and goal-  
55 directed actions (exploitation) lies at the core of the control of behavior but how it is achieved  
56 and evolved in animals remains unclear.

57           One way to solve the exploration/exploitation tradeoff is to move in order to  
58 maximize the information gain, for instance by using the so-called infotaxis (Hernandez-Reyes  
59 et al., 2021; Vergassola et al., 2007). Climbing up an information gradient can lead directly  
60 towards the goal if the source of information is emitted by the goal itself, such as when targeting  
61 sound or odor sources. However, it is unclear how such a strategy could apply when one moves  
62 without perceiving its final goal, such as during visual route following.

63           Another way of sampling the world is through the intrinsic production of  
64 regular alternations between left and right turns along the path: lateral oscillations. Lateral  
65 oscillations are observed in wide range of taxa (vertebrates: DeBose and Nevitt, 2008; Wolf et  
66 al., 2017; invertebrates: Freas and Cheng, 2022; Iwano et al., 2010; Kanzaki et al., 1992; Namiki  
67 and Kanzaki, 2016; Wystrach et al., 2016).

68 Oscillatory paths have been mainly studied in the context of olfaction, whether during plume  
69 following in moths (Kuenen and Baker, 1983; Olberg, 1983; Kanzaki et al., 1992; Kuenen and

70 Baker, 1983; Namiki et al., 2014), trail following in ants (Hangartner, 1967) or odor gradient  
71 climbing in *Drosophila* larvae (Wystrach et al., 2016) and *Caenorhabditis elegans* (Izquierdo  
72 and Lockery, 2010). The intrinsic production of oscillations enables an efficient sampling of  
73 odor across locations and models show that oscillations modulated by odor perception enables  
74 to reach the source in a remarkably effective way (Adden et al., 2020; Izquierdo and Lockery,  
75 2010; Wystrach et al., 2016).

76 Beyond that, modeling also shows that the production of oscillations can be equally effective  
77 during visual navigation tasks. For instance, having the amplitude of oscillation modulated by  
78 familiarity of visual scenes can produce robust visual navigation behavior such as route-  
79 following (Kodzhabashev and Mangan, 2015) or homing (Le Möel and Wystrach, 2020). It  
80 also captures particular behavioral signatures observed in ants (Murray et al., 2020). Moreover,  
81 visually guided insects such as ants (Graham and Collett, 2002; Lent et al., 2013, 2010;  
82 Jayatilaka et al., 2017; Murray et al., 2020), wasps (Stürzl et al., 2016; Voss and Zeil, 1998) or  
83 bumblebees (Philippides et al., 2013) do display oscillations, whose expression appears to be  
84 coupled with visual perceptual cues. But whether these lateral oscillations are produced  
85 internally, how exactly they interact with visual cues, and how they participate in the  
86 exploration/exploitation tradeoff during a visual navigational task remains unclear.

87           Here, we focused on the expression of oscillations in two ecologically and  
88 phylogenetically distant ant species known to use vision for navigation (*Myrmecia croslandi*  
89 and *Iridomyrmex purpureus*). These insects are known to rely on two main strategies to guide  
90 their foraging journeys to an inconspicuous goal (Nardendra et al., 2013; Card et al., 2016, see  
91 also SI1). The first strategy, commonly called path integration (PI), allows individuals to  
92 continuously estimate the distance and compass direction that separates them from their nest  
93 (or other starting points) during their foraging trip (Collett and Collett, 2017; Heinze et al.,  
94 2018; Muller and Wehner, 1988). The second, commonly called view-based navigation,

95 involves the learning and subsequent recognition of learnt visual panorama (Collett and  
96 Cartwright, 1983; Wehner, 1979; Zeil, 2012). To see how these navigational strategies do  
97 interact with oscillation, and how they participate to the exploration /exploitation tradeoff  
98 during a visual navigational task, we mounted ants of both species on a trackball device  
99 (Dahmen et al., 2017). This device enabled us to record their motor behavior and control the  
100 visual cue perceived directly in their natural environment. We characterized in detail whether  
101 the obtained trajectories show a regular oscillatory pattern of movements and how these  
102 patterns are influenced by the presence or absence of: (1) visual input, (2) learnt visual  
103 terrestrial cues, (3) path integration homing vector and (4) rotational visual feedback. In both  
104 species our results revealed the presence of a conserved pattern of oscillations generated  
105 intrinsically, which comprises both angular and forward velocity components. The amplitude  
106 of this dynamic signature is modulated by visual information in a way that is adapted to the  
107 navigational tasks at hand. Finally, a simple neural circuit model of reciprocal inhibition  
108 between left and right pre-motor area can readily explain the production of these movement  
109 dynamics.

110

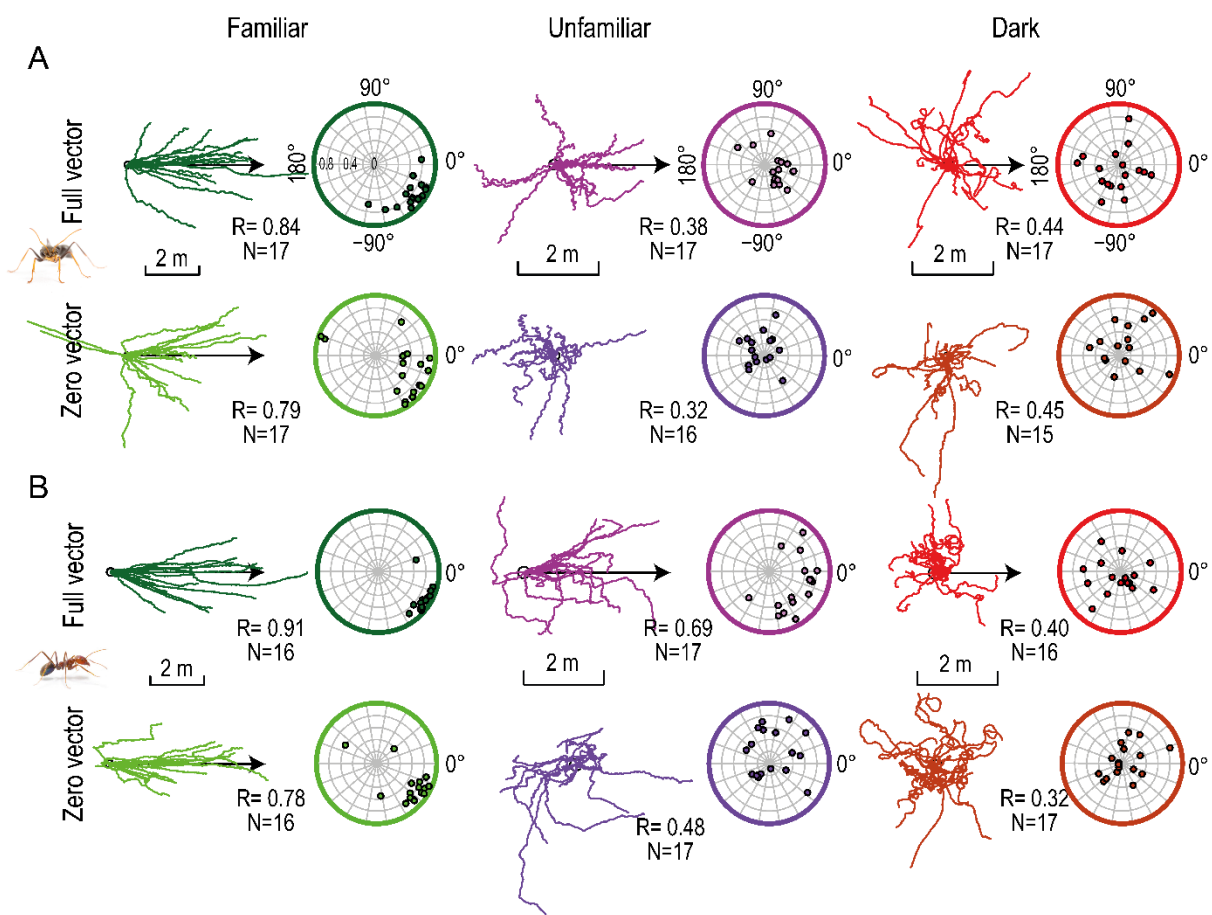
## 111 **Results**

112

### 113 **Ants mounted on the trackball display their natural navigational behavior**

114 We first investigated whether ants mounted on the trackball display the expected, natural  
115 navigational behavior for different experimental conditions. When released on the ground in  
116 their natural environment, both species studied here (*Myrmecia croslandi* and *Iridomyrmex*  
117 *purpureus*) are known to rely on learnt terrestrial cues as well as, to a lesser extent, on PI (Card  
118 et al., 2016 see Fig. S1 for preliminary experiment on *I. purpureus*; Murray et al., 2020;  
119 Narendra et al., 2013; Zeil et al., 2014). Here we mounted ants mounted on the trackball in a

120 way that enabled them to physically rotate and control their actual body orientation (Dahmen  
121 et al., 2017). The trackball was placed in three distinct visual conditions: (1) along the  
122 individual ant's familiar route (F) therefore in presence of familiar terrestrial cues; (2) in an  
123 unfamiliar location (U) 50 meters away from their usual route; or (3) without any visual  
124 input: in complete darkness (D). For each of these visual conditions, ants were tested either  
125 with (full-vector (FV) ants) or without (zero-vector (ZV) ants) path integration information.  
126 While FV ants were captured at their feeding place (from their foraging tree (*M. croslandi*) or  
127 from their trained feeder (*I. purpureus*)) and thus possess a PI homing vector pointing in the  
128 food-to-nest compass direction, ZV ants were caught just before entering their nest and thus no  
129 longer possess a PI homing vector.



130

131 **Figure 1. Ants on the trackball display natural navigational behaviors.** Data presented  
132 for both *M. croslandi* (A) and *I. purpureus* (B), tested in a familiar (green) or an unfamiliar  
133 panorama (purple) and in the dark (red). For each condition, individuals were tested either  
134 with (full vector) or without (zero vector) path integration information. The first column of

135 each condition shows the paths oriented to the theoretical direction of the nest (arrow). In the  
136 second column, each dot indicates the average circular vector calculated over the entire path  
137 of an individual, showing the mean direction and the average vector length (i.e., a point  
138 closer to the periphery indicates straighter paths). The R values indicate the length of the  
139 average resulting vector of the population. The direction (in familiar terrain) or theoretical  
140 direction (for FV in unfamiliar terrain) of the nest is 0°. Ants photography credit: Ajay  
141 Narendra.  
142

143 For both species and irrespectively of the PI state (FV or ZV), ants mounted on the trackball  
144 within their familiar visual route (i.e., in the presence of learnt terrestrial cues) displayed paths  
145 that were oriented toward the nest direction (Fig. 1A, B first column; Rayleigh test with nest  
146 as theoretical direction ps: *M. croslandi*: FV & ZV < 0.001; *I. purpureus*: FV & ZV < 0.001),  
147 proving that they recognized and used the familiar terrestrial cues to orient. When tested in  
148 unfamiliar surroundings, FV ants of both species were oriented toward the theoretical direction  
149 of the nest as indicated by their PI (Fig. 1A, B second column, first row; Rayleigh test with PI  
150 as theoretical direction ps: *M. croslandi*: FV < 0.001; *I. purpureus*: FV < 0.001); but ZV ants  
151 showed random orientations (Rayleigh test ps: *M. croslandi*: ZV=0.443; *I. purpureus*:  
152 ZV=0.672). This confirms that the distant release points were indeed unfamiliar to the ants and  
153 that both species relied on their PI when tested as FV ants. When tested in total darkness, ants  
154 displayed randomly orientated paths in all conditions (Fig. 1A, B third column; Rayleigh test  
155 ps: *M. croslandi*: FV=0.354; ZV=0.360; *I. purpureus*: FV=0.213, ZV=0.866), showing that the  
156 chosen dark condition was effective in preventing ants using terrestrial or celestial cues for  
157 orientation. Overall, these results demonstrate that both species use learnt terrestrial cues as  
158 well as their PI to navigate when mounted on the trackball system (Fig. 1A, B).

159 Independent of the PI state, the ants' travel directions were more constant in familiar  
160 terrain as compared to the other conditions (R values in Fig. 1A, B; Familiar vs. Unfamiliar &  
161 Familiar vs. Dark, both species: ps < 0.001), showing that the recognition of learnt terrestrial  
162 cues was most effective to maintain a constant direction of travel. With regards to the effect of  
163 PI, FV ants tended to keep their direction more constant than ZV ants but this effect was small,



164 and reached significance only in unfamiliar terrain for *I. purpureus* (Fig. 1; FV vs. ZV  $p =$   
165 0.049) but not in familiar terrain (Fig. 1; FV vs. ZV both species:  $p_s \geq 0.0589$ ). Finally, there  
166 was no difference between the PI state for both species in the dark condition (Fig. 1; FV vs.  
167 ZV  $p_s = 1$ ). Therefore, both species do use their PI to orient in unfamiliar terrain, but it seems  
168 that PI has a limited impact on their path's straightness.

169         These results are consistent with what is observed in these species when naturally  
170 walking on the floor (Card et al., 2016; Narendra et al., 2013).

171

### 172 **Ants display regular lateral oscillations**

173 To determine whether ants display regular lateral oscillations – that is, alternate between left  
174 and right turns at a steady rhythm – we first used our video recordings to track the ant's change  
175 in body orientation when mounted on the trackball. The body angular velocity signal is  
176 independent of the ant's actual forward movement and thus a direct reading of its motor control  
177 for turning. We conducted a Fourier analysis on autocorrelation coefficients of the angular  
178 velocity time series to obtain a 'power spectral density' (see Fig. S2 for detailed method). The  
179 obtained Fourier's magnitudes are a measure of the regularity of the rhythms in the time series,  
180 with high magnitude values indicating regular oscillation at the corresponding frequency (Fig.  
181 S2). We thus selected the frequency corresponding to the highest magnitude (peak), that is, the  
182 most salient rhythm in the signal. In all experimental conditions, the obtained peak fell in the  
183 expected range of 0.2 to 1.5 Hz, as observed in other insect species (Lonnendonker, 1991;  
184 Wystrach et al., 2016). It should be noted that this rhythm is 10 to 50 times slower than ants'  
185 typical stepping frequency (Zollikofer, 1994) and thus are not the consequence of their  
186 rhythmic walking gait per se but the product of an additional oscillatory mechanism. The  
187 obtained Fourier magnitudes were higher than those obtained with the spectral density of a

188 Gaussian white noise (Fig. 2A, B dashed line; Wilcoxon one-tail test:  $p \leq 2.144e-03$ ), showing  
189 that ants displayed lateral oscillations with a higher regularity than random.

190

### 191 **The navigational context modifies the regularity of oscillations**

192 We analyzed whether our different test conditions influenced the magnitude of the  
193 individuals' spectral density peaks, which indicates how regular the oscillations are. The  
194 statistical model revealed no interaction between the effects of the visual panorama type and  
195 the state of the path integrator (*M. croslandi*: AIC=-198.8,  $F_{2,101}=0.434$ ,  $p=0.647$ ; *I.*  
196 *purpureus*: AIC=-167,  $F_{2,90}=0.232$ ,  $p>0.79$ ). The additive effect model (i.e., without  
197 interaction, *M. croslandi*: AIC=-215.8; *I. purpureus*: AIC=-170.5) explains the variation of  
198 the magnitude peaks relatively well ( $R^2$  for *M. croslandi*: marginal=56% & conditional=67%;  
199 *I. purpureus*: 16%). The subsequent Anova revealed that the visual panorama type has a  
200 significant effect on the regularity of oscillations in both species (panorama effect for *M.*  
201 *croslandi*:  $F_{2,101}=83.663$ ,  $p < 0.001$ ; *I. purpureus*:  $F_{2,90}=10.038$ ,  $p < 0.001$ ); however, there  
202 was no overall effect of the PI state (vector effect: *M. croslandi*:  $F_{1,101}=1.291$ ,  $p>0.200$ ; *I.*  
203 *purpureus*:  $F_{1,90}=0.012$ ,  $p>0.900$ ).

204 For *M. croslandi*, the post-hoc analysis revealed that the magnitude peak varied  
205 significantly across the three visual conditions (Fig. 2A, F vs. U:  $p<0.001$ ; F vs. D:  $p=0.016$ ;  
206 U vs. D:  $p<0.001$ ). These differences in peaks magnitudes show that the oscillations are most  
207 regular in unfamiliar terrain, intermediate in familiar environment and least regular in the  
208 dark (mean $\pm$ se:  $U_{(FV+ZV)}=171\pm 19$ ;  $F_{(FV+ZV)}=68\pm 6$ ;  $D_{(FV+ZV)}=50\pm 3$ ). Interestingly, the effect of  
209 individuality is significant in *M. croslandi* ( $p=0.015$ ), indicating that some individuals have a  
210 better consistency in their oscillations than others, across conditions.

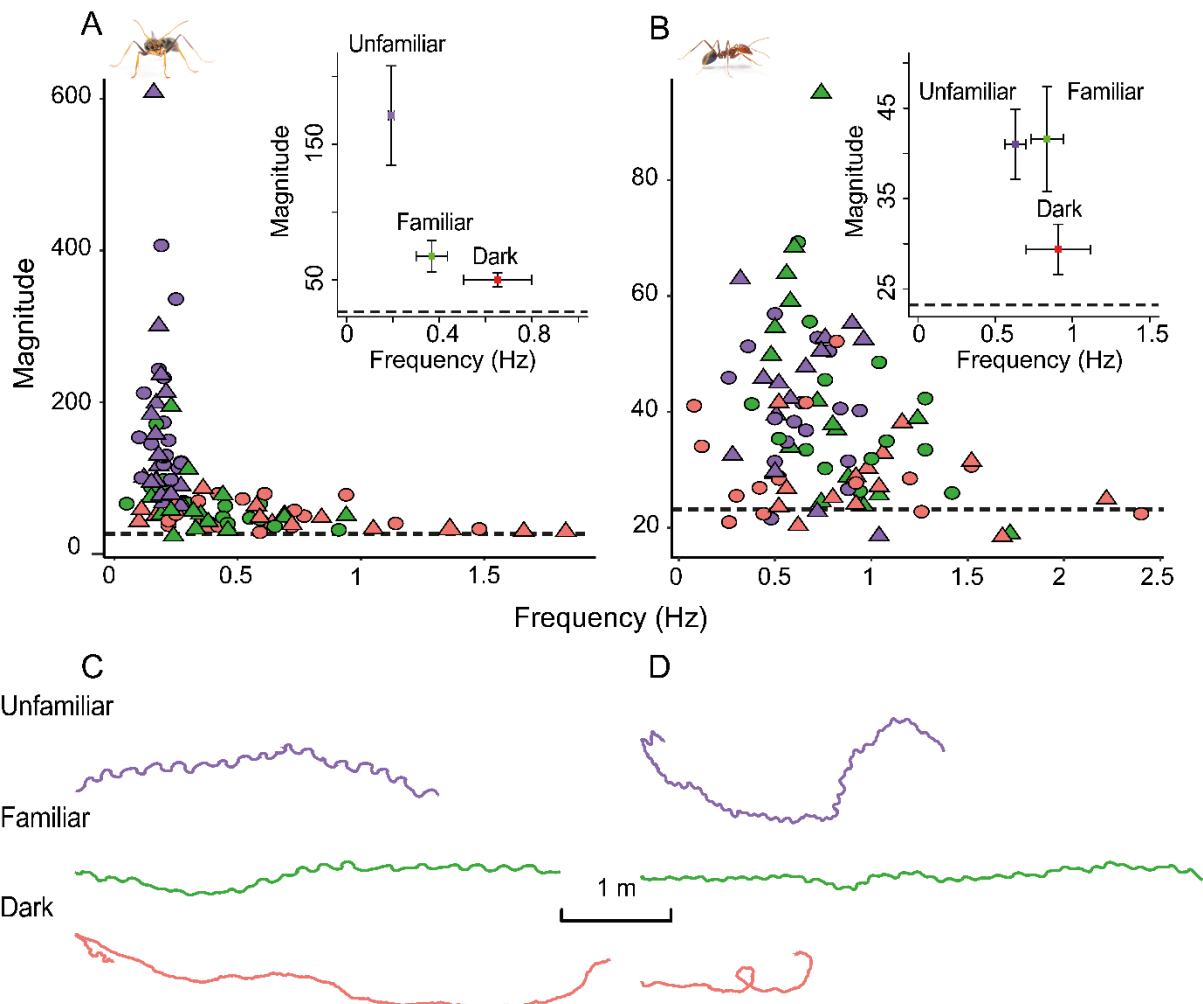
211 For *I. pupureus*, the post hoc analysis reveals that the regularity of the oscillations  
212 does not differ between familiar and unfamiliar terrain (Fig. 2B,  $p>0.96$ ,  $df=86$ , mean $\pm$ se:

213  $F_{(FV+ZV)} = 41.59 \pm 3$ ;  $U_{(FV+ZV)} = 41 \pm 2$ ) but both yield more regular oscillations than in the dark  
214 ( $p < 0.001$ ,  $\text{mean} \pm \text{se}$ :  $D_{(FV+ZV)} = 29.4 \pm 1.45$ ).

215 Overall, these differences confirm the existence of regular oscillations in both  
216 unfamiliar and familiar terrain, regardless of the PI state. In the dark however, peak  
217 magnitudes are significantly weaker, closer to Gaussian noise and with a wider frequency  
218 range, suggesting that the Fourier's peak in some individuals may result from noise (Fig. 2A,  
219 B). If oscillations persist in the dark, their expression is at least greatly inhibited (Fig.2).

220

221



222

223 **Figure 2. Oscillation characteristics vary across visual conditions.** The graphs show the  
224 frequency and spectral density magnitude of the dominant oscillation (highest magnitude) in  
225 *M. croslandi* (A) and *I. purpureus* (B) (for method see method see Fig. S2 A, C, D, E). High  
226 frequencies indicate a fast-oscillatory rhythm and high magnitudes indicate a strong presence  
227 of this oscillation. Individuals were tested within a familiar panorama (green), an unfamiliar  
228 panorama (purple) or in the dark (red). Symbols indicate whether the state of the path

229 integration vector was full (round) or set to zero (triangles). Inserts show the mean frequency  
230 against the mean magnitude of each visual condition with the associated 95% confidence  
231 interval around the mean. The dashed black line represents the mean of the spectral density  
232 peak magnitudes resulting from 200 Gaussian white noise signals. The second row show an  
233 example path for each condition for *M. croslandi* across 100 s (c) and *I. purpureus* across 83  
234 s (D). Ants photography credit: Ajay Narendra.

235  
236

## 237 **The navigational context modulates the oscillation frequency**

238 *Myrmecia* foragers tested in familiar panorama showed no differences in oscillation  
239 frequencies across PI states (Wilcoxon test for repeated measures; F: FV vs. ZV  $p=0.635$ ; U:  
240 FV vs. ZV  $p=0.343$ ). Again, independent of the PI state, the oscillatory frequencies were  
241 significantly higher in familiar than in an unfamiliar visual panorama ( $p=0.023$ ,  $\text{mean}\pm\text{se}$ :  
242  $F_{(\text{FV}+\text{ZV})}=0.37\pm0.036$  Hz;  $U_{(\text{FV}+\text{ZV})}=0.19\pm0.008$  Hz; Fig. 2A, C). *Iridomyrmex purpureus*  
243 showed the same tendency to oscillate quicker in familiar than in unfamiliar terrain  
244 ( $\text{mean}\pm\text{se}$ :  $F_{(\text{FV}+\text{ZV})}=0.83\pm0.056$  Hz;  $U_{(\text{FV}+\text{ZV})}=0.63\pm0.037$  Hz; Fig. 2B, D), but this effect did  
245 not reach significance ( $p_s \geq 0.180$ ).

246 Thus, the presence of familiar visual cues tends to increase the frequency of oscillations,  
247 at least for *M. croslandi* (Fig. 2A, C), but the state of the PI vector (FV or ZV) had no  
248 observable effect ( $\text{mean}\pm\text{se}$  in F: FV= $0.38\pm0.05$  Hz, ZV= $0.34\pm0.04$  Hz; U: FV= $0.19\pm0.01$ Hz,  
249 ZV= $0.18\pm0.0009$  Hz).

250

## 251 **The navigational context modulates the angular and forward velocity of oscillations**

252 To investigate the dynamics of the ant oscillatory movements in more detail, we combined  
253 for each ant its body angular velocity signal – obtained from video recording – with the  
254 forward movement signal – obtained from the trackball movements. We reconstructed  
255 ‘average cycles’ for each individual by pooling the angular and forward velocity recordings  
256 3s before and 3s after the moments where ants switch from a left to a right turn (when the  
257 time series of the angular velocity crosses zero from negative to positive; see Fig. 3—figure

258 supplements 1). That way, we can quantify each individual's average dynamic of angular and  
259 forward velocity and compare them across conditions.

260 For *M. croslandi*, the visual surrounding had a strong effect ( $p < 0.001$ ): ants  
261 displayed higher angular velocities (Fig. 3A, inset boxplot upper row; mean $\pm$ se:  
262  $U_{(FV+ZV)} = 129 \pm 4.8$  deg/s;  $F_{(FV+ZV)} = 67.5 \pm 4.4$  deg/s) and higher forward velocities (Fig. 3A,  
263 inset boxplot second row, mean $\pm$ se:  $U_{(FV+ZV)} = 4.8 \pm 0.34$  cm/s;  $F_{(FV+ZV)} = 2.3 \pm 0.2$  cm/s) in an  
264 unfamiliar environment. There was however neither an effect of the PI state on turn or  
265 forward velocity ( $p > 0.05$ ) nor an interaction between the visual surrounding and the PI state  
266 (mean peak of angular velocity model: AIC=160.9, interaction  $p > 0.05$ ; amplitude of forward  
267 velocity model: AIC=109.9, interaction  $p = 0.22$ ). Therefore, the amplitude of lateral  
268 oscillation within the path are higher in an unfamiliar panorama than in a familiar one  
269 independently of the PI state (Fig. 2 C). Interestingly, *M. croslandi* ants showed strong  
270 individual idiosyncrasies across conditions (forward velocity peak amplitude: random effect,  
271  $p < 0.05$ , R2 marginal=0.39, R2 conditional=0.61; angular velocity peak amplitude: random  
272 effect,  $p > 0.05$ , R2 marginal=0.5; R2 conditional=0.66).

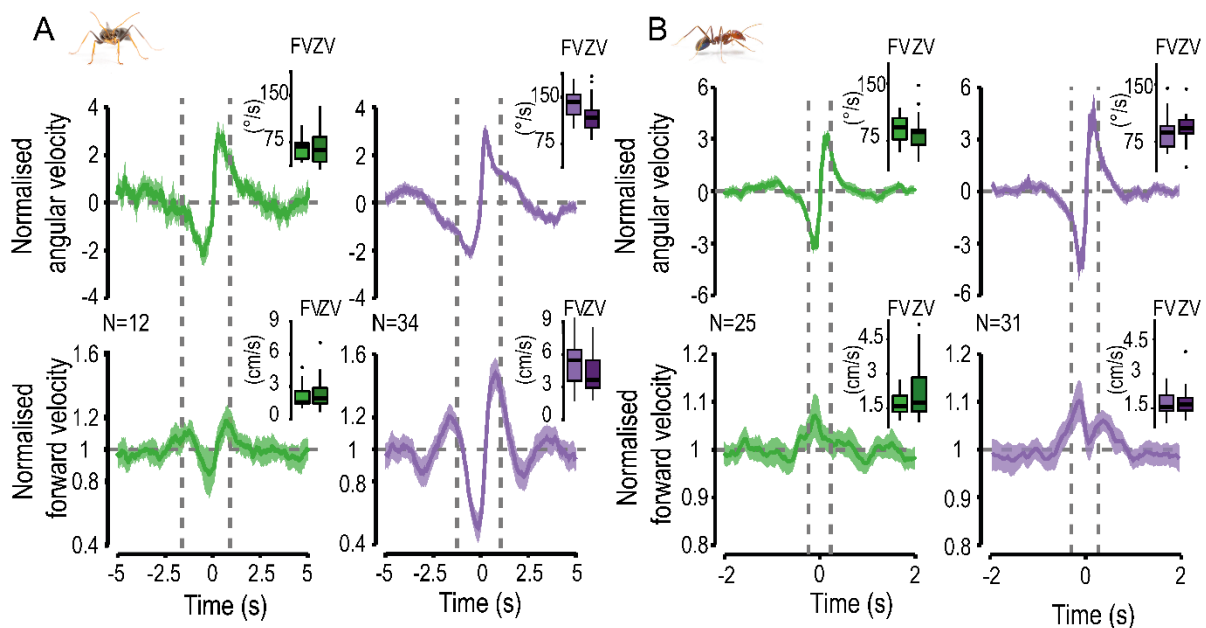
273 For *I. purpureus* ants, none of the models reached significance ( $p_s > 0.05$ ); showing  
274 that the dynamics of angular velocities and forward velocity underlying the oscillations are  
275 rather constant across navigational context (Fig. 3B insert). Showing no clear effect on the  
276 amplitude of oscillation display within the path (Fig. 2D).

277

## 278 **The conserved dynamics of lateral oscillations**

279 To test for the existence of conserved oscillatory dynamics across individuals, each  
280 individual average cycle (Fig. 3—figure supplements 1 for method example) was normalized  
281 and averaged to obtain a cycle at the population level. To ensure that we pooled ants  
282 oscillating at similar frequency ranges, we separated data from familiar and unfamiliar terrain

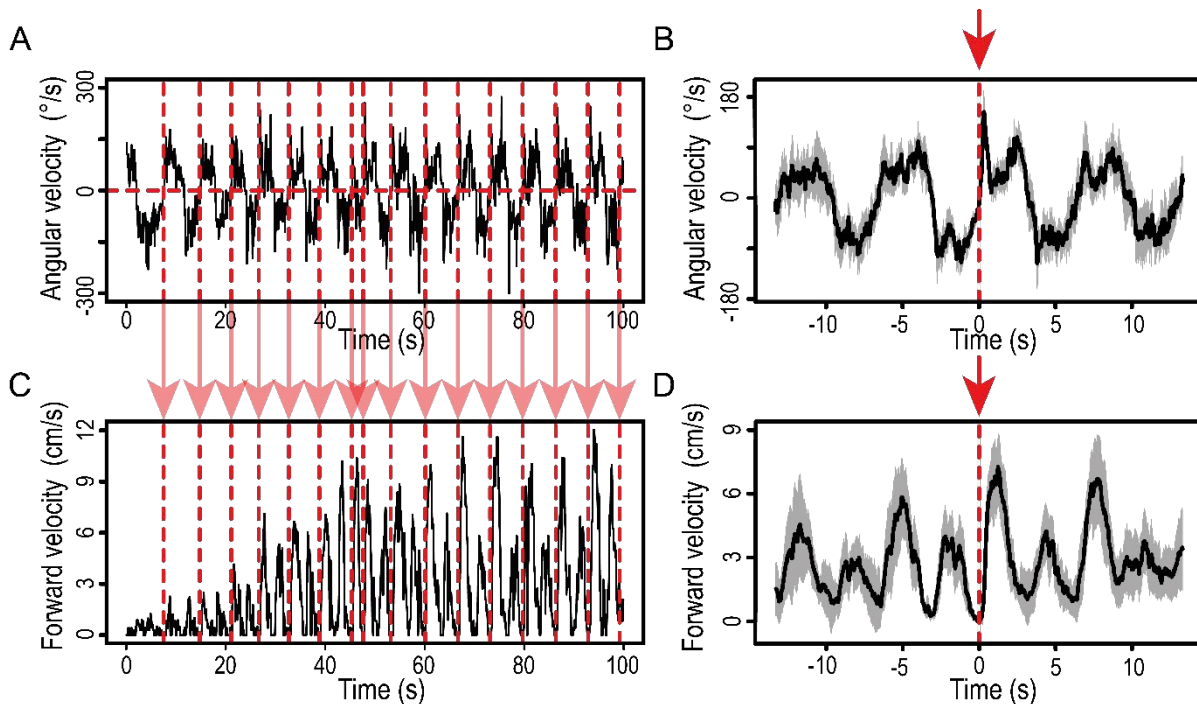
283 and selected only individuals showing a peak frequency within 0.1 to 0.27 Hz for *M.*  
284 *croslandi* (N=12) and within 0.26 to 1.04 Hz for *I. purpureus* (N=25). These frequency  
285 ranges correspond to high magnitude, ensuring that they are not a consequence of noise. The  
286 emerging population-averaged oscillation patterns show the existence of movement dynamics  
287 that are consistent across individuals of both species, for both familiar and unfamiliar  
288 environments (Fig. 3, Fig. 4). Forward velocity co-varies with angular velocity in a particular  
289 way. Forward velocity is quite low when the ants reverse their turning direction (i.e., when  
290 angular velocity crosses zero). In contrast, we observe two significant peaks of forward  
291 velocity, which happen briefly (up to 1s) while the ant is sweeping to the left or right.  
292 Remarkably, these peaks coincide well with the moment when the ants' body orientation is  
293 aligned with its overall direction of travel (Fig. 3, dashed lines) even though during these  
294 moments the ants' angular velocity is quite high. In other words, ants display large sweeps  
295 from one side to the other and do not maximize the time they spent facing their goal  
296 direction, which could be achieved by reducing angular speed at this moment. However, they  
297 increase their forward velocity precisely while doing so.



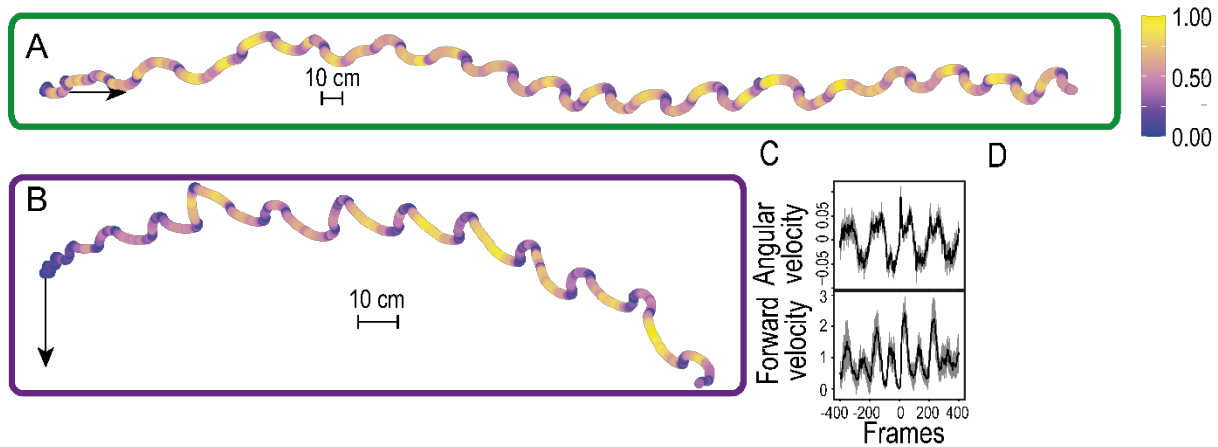
298

299

300 **Figure 3: Population-average cycle of oscillations shows how angular and forward**  
301 **velocities co-vary.** Angular velocities (top-row) and forward velocity (bottom row) co-vary  
302 in a way that seem conserved across species (*M. croslandi* (A) and *I. purpureus* (B)) when on  
303 familiar route (green) or in unfamiliar terrain (purple). Population cycles have been  
304 reconstructed by merging full- (FV) and zero vector (ZV) data and normalizing the data  
305 amplitude within the individual's average cycle (see Fig. 3 —figure supplements 1). Color  
306 areas around the mean curves represent the 95% confidence interval, based on the inter-  
307 individual variation. Dashed lines represent the moment when the ants are facing their overall  
308 direction of travels. Insert boxplots show the actual distribution of the non-normalized mean  
309 peak of angular speed and forward velocity amplitude for the individuals' average cycle (FV  
310 on the left, ZV on the right). Ants photography credit: Ajay Narendra.  
311



312  
313 **Figure 3—figure supplements 1: Methodology to obtain an average oscillation cycle.** (A)  
314 We used the angular velocity signal to flag moments when the signal crosses 0 and goes up to  
315 positive angles (vertical dashed red lines). (B) We extracted portions of the signal of  $\pm 3$ s  
316 (large enough to contain a full oscillation cycle) around each of the flags, aligned them at flag  
317  $= t_0$  and averaged them to obtain the individual's average cycle ( $\pm 95\%$  confidence interval in  
318 grey) of the angular velocity signal. (C, D) The individual's average cycle for the forward  
319 velocity signal was achieved using the angular velocity flags (vertical dashed red lines as in  
320 A).  
321  
322  
323



324

325 **Figure 4: Dynamics of forward velocity are the same in familiar and unfamiliar**  
326 **panoramas.** Reconstructed path (100 s) of *M. croslandi* individuals recorded in a familiar  
327 (A) and in an unfamiliar (B) panorama. Each path has been colored according to the forward  
328 velocity data (yellow forward movement; blue no forward movement). On both paths, the  
329 arrow indicates the theoretical direction of the nest. (C) Averaged cycle of the individual  
330 recordings in B, showing that forward and angular velocities' peaks happen simultaneously.  
331 (D) Video of the individual corresponding to the paths shown in B.

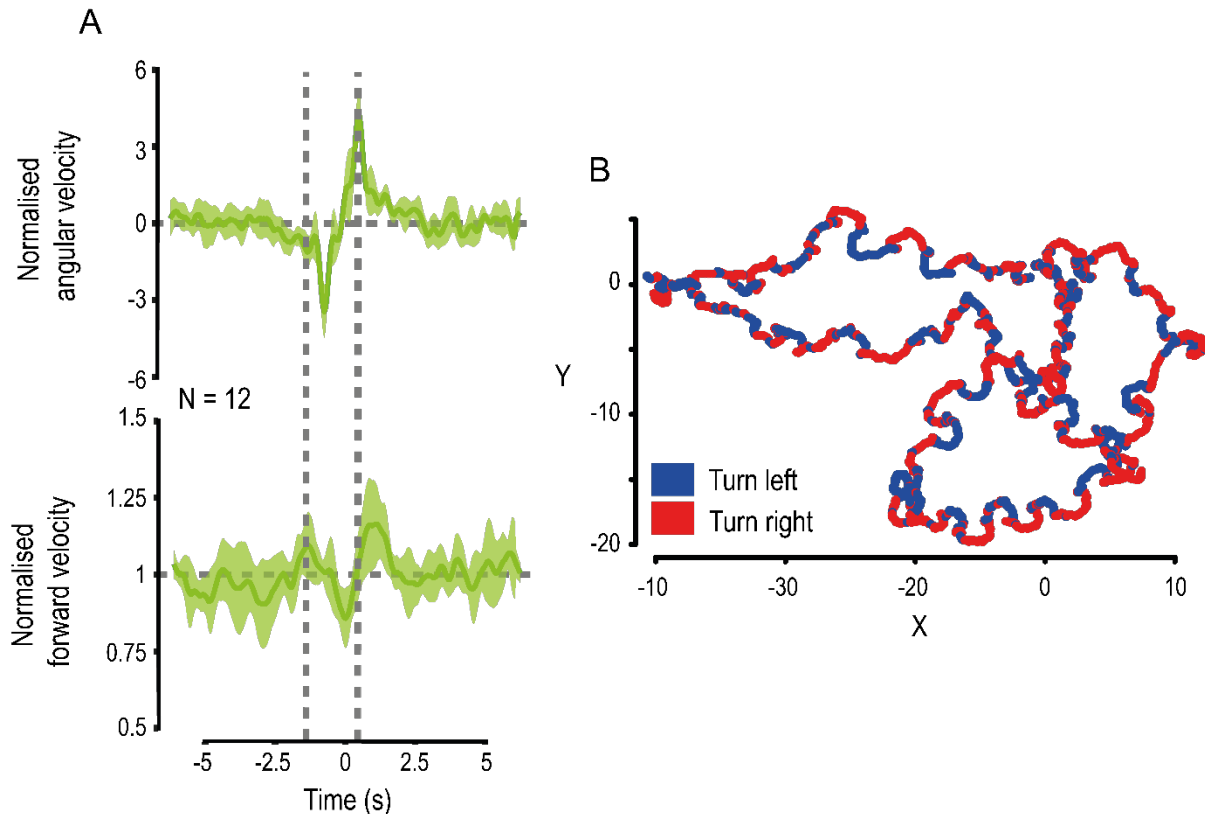
332

333 To ensure that these dynamics are not a consequence of being mounted on the  
334 trackball set-up, we replicated the same analysis using tracks of *M. croslandi* ants recorded  
335 directly on the ground while displaying learning walks around their nest (courtesy of Jochen  
336 Zeil, see Zeil and Fleishmann 2019). The natural learning walks also showed regular  
337 oscillations as well as the same dynamical relationship between angular and forward  
338 velocities (Fig. 5). Ants walking on the ground also tend to increase their forward velocity  
339 when they are aligned with their general direction of travel.



340

341 **Figure 5. Ant learning walks show similar dynamics of oscillation.** Ants path and head



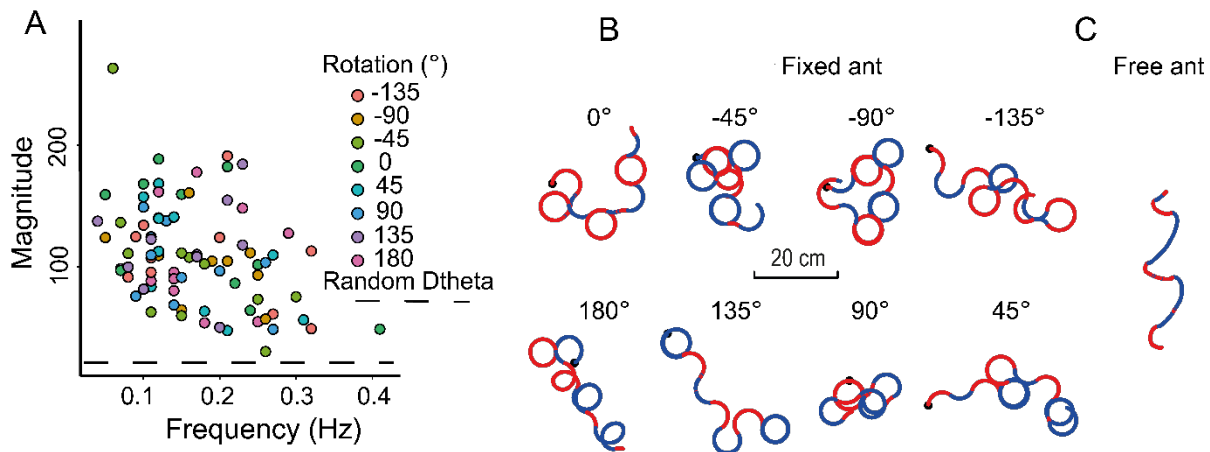
342 orientation were recorded (25 Hz) during their learning walks directly on the ground. (A)  
343 Angular velocity (top-row) and forward velocity (bottom row) co-vary in a similar way than  
344 when recorded on the track ball (see Fig. 3). Population cycles have been normalized by the  
345 data amplitude within the individuals mean cycle (see Fig. 3—figure supplements 1 for  
346 example). Color areas around the mean curves represent the 95% confidence interval, based  
347 on the inter-individual variation. (B) Example path of an individual showing clear alternation  
348 between right (red) and left (blue) turn.

349

### 350 **Oscillations are modulated by rotational feedback**

351 To test whether oscillations result from an intrinsic oscillator rather than a servo-mechanism  
352 based on sensory information, we investigated whether ants deprived of rotational feedback  
353 (either via optic-flow or compass cues) would still oscillate. For that, ants were mounted on  
354 the trackball in a way that prevented them from physically rotating their body on the ball:  
355 when the ant tries to turn, it is the ball that counterturns. In other words, the fixed ant  
356 experiences neither a change of body orientation nor visual rotational feedback when trying  
357 to turn (see Fig. S2B). For this experiment, we used *M. croslandi* ants in unfamiliar terrain as

358 this condition produced the clearest oscillations in our previous experiments (Fig. 2). To  
359 control for potential effects of facing directions (depending on celestial or terrestrial visual  
360 cues), we tested each ant fixed in eight subsequent different orientations with a 45° rotational  
361 shift.



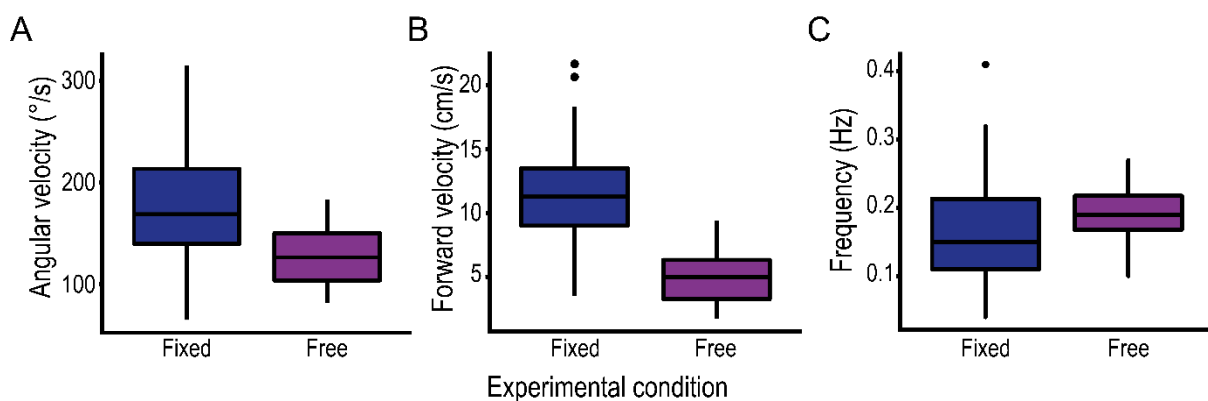
362

363 **Figure 6. Ants still oscillate in the absence of rotational feedback.** Zero vector ants were  
364 tethered on the trackball in a way that prevented actual body rotation; and tested in an  
365 unfamiliar environment. (A) Distribution of the individual Fourier dominant peak frequency  
366 and magnitude based on the angular velocity's spectral density time series (as in Fig. 2,  
367 method see Fig. S2 B, C, D, E). High frequency indicates a fast-oscillatory rhythm, and a  
368 high magnitude indicates a strong presence of this oscillatory rhythm. The color symbols  
369 represent the rotation relative to the theoretical nest direction (0°). The dashed black line  
370 represents the mean magnitude obtained from 200 Gaussian white noise signals (18.75). (B)  
371 Example paths of different individuals fixed in different orientations. (C) Example path from  
372 an ant that was free to rotate on the trackball, recorded in unfamiliar terrain in previous  
373 experiments. (B, C) In both situations, ants alternate regularly between right (blue) and left  
374 (red), but turns are sharper in the absence of rotational feedback.  
375

376 Despite the absence of visual rotational feedback, the obtained peak magnitudes of the  
377 angular velocity spectral density (Fourier analysis on autocorrelation coefficient, see  
378 Methods) were much higher than expected from a Gaussian white noise ( $p < 0.001$ ) showing  
379 clearly the presence of a regular alternation between left and right turns (Fig. 6). The ants'  
380 body orientation relative to the world had no effect on the magnitudes (orientation:  
381  $F_{7,83} = 0.3729$ ,  $p = 0.914$ ) nor did the other random parameters (individual  $p = 0.933$ , sequence  
382  $p = 0.473$ ). The mean oscillation frequencies (mean  $\pm$  se:  $0.17 \pm 0.007$  Hz) were quite close to  
383 what we observed when these ants were free to rotate in an unfamiliar environment

384 (Wilcoxon test for repeated measures:  $p=1$ ,  $N=88$ , Fig. 6). Thus, ants can display regular  
385 turning oscillations without rotational feedback, whether as a result of optic flow or a change  
386 of orientation relative to directional cues such as the visual panorama, wind, celestial  
387 compass cues or the magnetic field. Consequently, we are left with the conclusion that these  
388 oscillations are generated intrinsically.

389 Interestingly, the absence of rotational feedback led to higher angular  
390 velocities (mean: fixed=176 deg/s; free=128 deg/s;  $p<0.001$ ), higher forward velocity (mean:  
391 fixed=11 cm/s; free=5 cm/s;  $p<0.001$ ) and slightly slower turn alternation (i.e., lower  
392 frequency: fixed=0.16 Hz; free=0.19 Hz,  $p=0.0189$ ) than ants that were free to physically  
393 rotate on the ball (Fig. 7). Rotational sensory feedback is thus involved in limiting the  
394 amplitude of the oscillations.



395

396 **Figures 7: Absence of visual rotational feedback increases the amplitude of oscillation.**  
397 Distribution of *M. croslandi* individuals' average oscillation dynamics in unfamiliar terrain  
398 when recorded on the track ball either in fixed mode preventing rotational feedback (blue) or  
399 free (purple). Peak of angular velocity (A), peak of forward velocity (B) and frequency (C)  
400 of the individual's average oscillation cycle show that ants move faster, turn faster and slightly  
401 longer in the absence of rotation feedback.

402

403

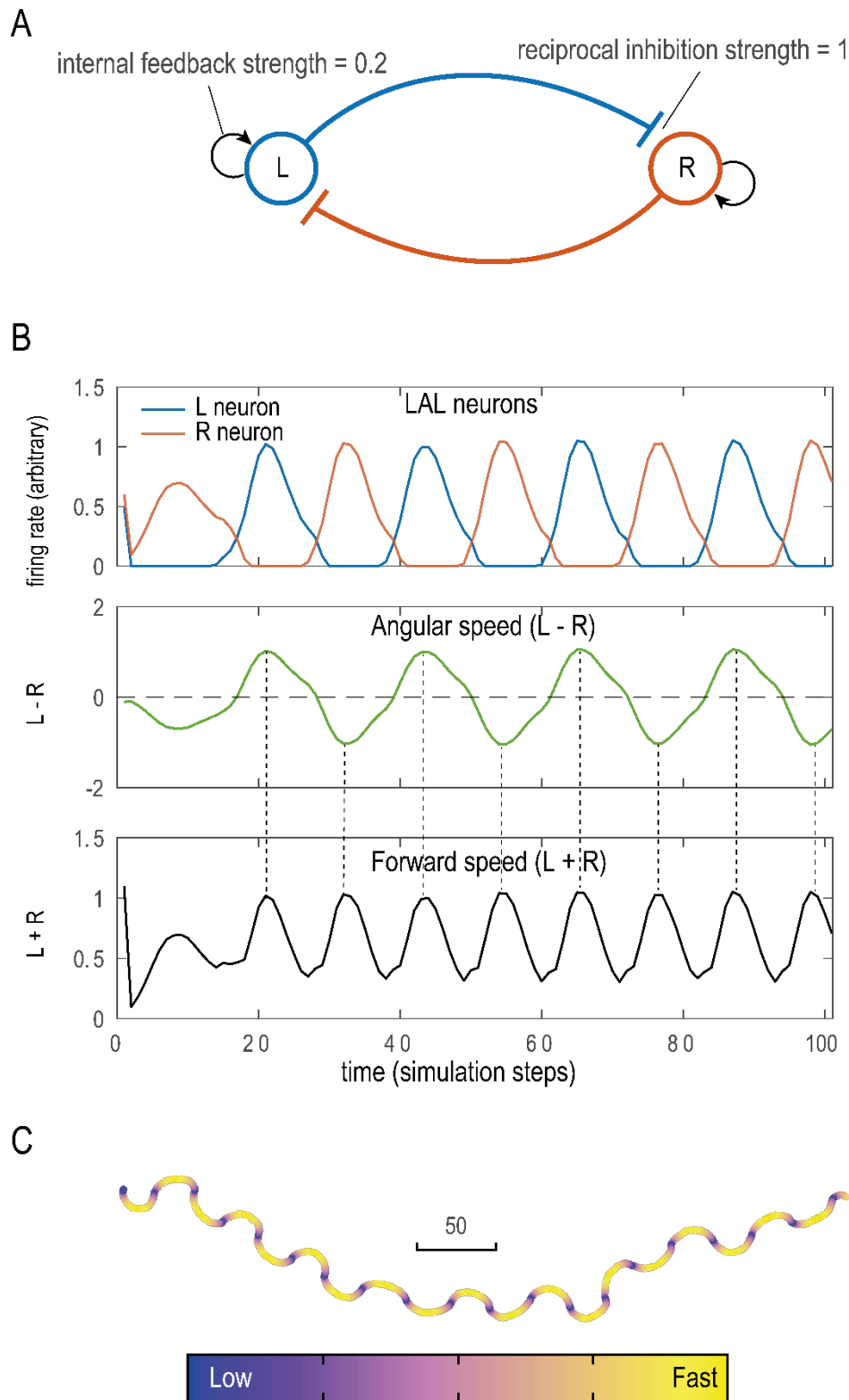
404 **The observed movement signature emerges readily from a simple neural circuit.**

405

406 We designed a simplistic neural model based on the circuitry of the insect pre-motor area so  
407 called lateral accessory lobes (LAL) (Namiki and Kanzaki, 2016a; Steinbeck et al., 2020).

408 The purpose of this model is not to match observed data quantitatively (Adden et al., 2020)

409 but simply to test whether the co-varying relationship observed between angular and forward  
410 velocity can readily emerge from this type of circuit. Activation of the neurons in the left and  
411 right pre-motor areas are known to mediate left and right turns respectively in insects (Berni,  
412 2015; Berni et al., 2012; Iwano et al., 2010; Kanzaki, 2005; Namiki and Kanzaki, 2016b;  
413 Steinbeck et al., 2020). Furthermore, these left and right regions are known to reciprocally  
414 inhibit each other (Iwano et al., 2010; Berni et al., 2012; Namiki and Kanzaki, 2016b;  
415 Steinbeck et al., 2020). Modelling two reciprocally inhibiting neurons with internal feedback  
416 –that tries to maintain a basal activity (Fig. 8)– is sufficient to obtain the typical oscillatory  
417 activity between left and right LAL (Iwano et al., 2010; Steinbeck et al., 2020), and thus  
418 provides an explanation for the regular oscillations between left and right turns observed in  
419 insects. Interestingly, we show here that simply assuming that forward velocity is controlled  
420 by the sum of left and right output – while angular velocity results from their difference  
421 (Adden et al., 2020; Steinbeck et al., 2020; Wystrach et al., 2016)– is sufficient for the  
422 observed covariation to emerge (Fig. 8). The agent accelerates while facing its general  
423 direction of travel and slows down at the end of its sweeping movement when its orientation  
424 extends most to the left and the right (Fig. 8C, 4). What’s more, the emergence of this  
425 particular relationship is robust to parameter change (Figure 8—figure supplements 1) and  
426 thus a stable feature of either these types of circuits.  
427

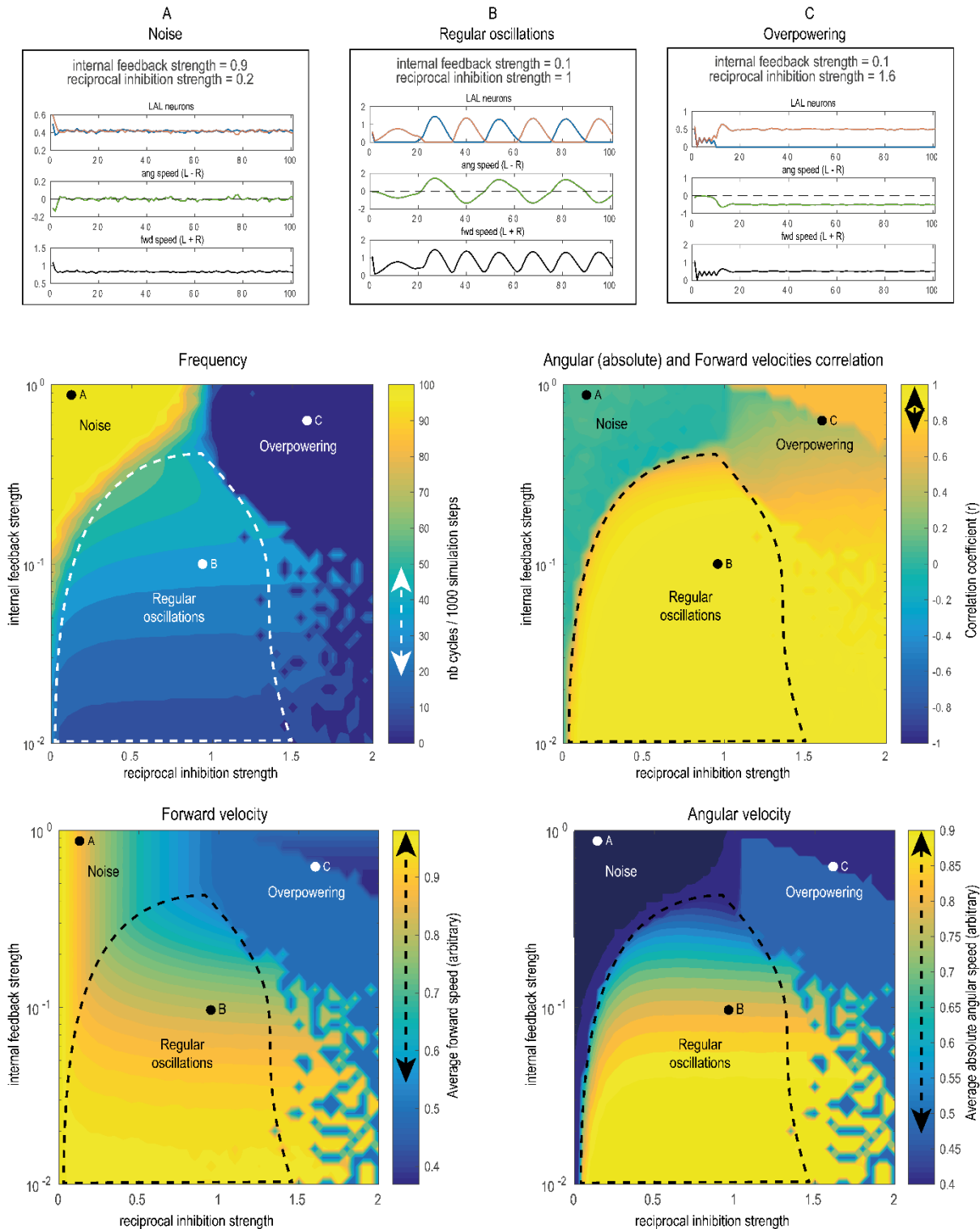


428

429 **Figure 8. Reciprocal inhibition between two units produces the observed relationship**  
430 **between angular and forward velocity.**

431 (A) Scheme of the model abstracted from the lateral accessory lobe (LAL). The Right (R) and  
432 Left (L) hemisphere neurons reciprocally inhibit each other (blue and red connection), while  
433 trying to sustain a basal firing rate through internal feedback (black arrow). (B) This results in  
434 the emergence of stable anti-phasic oscillatory activity between the R and L neurons (B,  
435 upper row). Assuming that angular velocity is controlled by the difference (B, middle row) –

436 and forward speed by the sum (B, bottom row) – between left and right activation is  
 437 sufficient to elicit the movement dynamics observed in ants (see Fig. 3C). As ants, the model  
 438 results in a zigzagging path where velocity drops during turn reversal, and increases when  
 439 facing the overall direction of travel (Fig. 3, 4). While the model enables the modulation of  
 440 amplitude and frequency of the oscillations, the forward/angular velocities relationship is  
 441 quite robust to parameter change (Fig.8—figure supplements 1).  
 442



443 **Figure 8—figure supplements 1. The covariation between angular and forward velocity**  
 444 **is robust to parameter change.**  
 445

446 Exploration of our abstract LAL model's (figure 8) sensitivity to change in the two free  
447 parameters (inhibition strength and internal feedback strength). Regular oscillations between  
448 LAL neurons emerge in a wide space of parameters (functional range delineated by the  
449 dashed line, an example for a given parameter set (dot) is shown in (B)). While frequencies  
450 (right of middle row), Forward and Angular velocities (bottom row) can vary roughly across  
451 a factor 2 across the functional parameter range, the correlation between absolute angular and  
452 forward velocity is constrained around 1 (dashed arrows in the color bars indicate the  
453 variation in the functional range). This positive covariation indicates that the agent slows  
454 down (low forward velocity) when reversing turning direction (low absolute angular velocity)  
455 and accelerates (high forward velocity) in between, that is when sweeping past its general  
456 direction of travel. Outside of this functional range, regular oscillations disappear for extreme  
457 parameter sets. For instance, if one neuron overpowers the other due to too strong reciprocal  
458 inhibitory strength (C, overpowering); or inversely, due to the absence of reciprocal  
459 inhibition (strength <0.01) or a too strong internal adaptation preventing the neurons to  
460 modulate their baseline activity, and thus resulting in noise (A).  
461

## 462 **Discussion**

### 463 **An internally generated movement at the core of ant visual navigation**

464 We recorded the detailed locomotor movements of two distantly related ant species – *M.*  
465 *croslandi* and *I. purpureus* – using a trackball-treadmill device directly in their natural  
466 environment. Both ant species displayed a similar rhythmical pattern of movement combining  
467 continuous lateral oscillations with a synchronized variation of forward speed: ants slow down  
468 – sometimes until a complete halt (see video Fig. 4D) – at the end of each left and right sweep,  
469 that is, when their body is facing away from their overall direction of travel. Conversely, they  
470 display a burst of forward speed in between, that is, when their body is aligned with their overall  
471 direction of travel, even though during these moments angular speed is rather high (Figs. 3,  
472 5A, S3). This generates oscillatory trajectories that optimizes the distance covered when facing  
473 the general direction of travel – and thus the effective distance covered – while simultaneously  
474 scanning left and right directions.

475 This particular movement signature is conserved whether ants are on their familiar route, in an  
476 unfamiliar panorama (Fig. 3) or when displaying their natural learning walks around the nest  
477 (Fig. 5A). Also, this movement signature is still expressed when the ant body is artificially  
478 fixed on our trackball in a given orientation (Fig. 6; Fig. S3), which shows that it results from

479 an internal process rather than a servomechanism based on external cues. Previous observations  
480 assumed that ants accelerate when facing their goal direction as the response to the recognition  
481 of familiar views when aligned in this particular direction (Baddeley et al., 2012; Kamhi et al.,  
482 2020; Wystrach et al., 2013). Here we show that this acceleration is a product of an endogenous  
483 process (that is also expressed in unfamiliar terrain), and thus stresses the often disregarded  
484 importance of internally generated movement within the ‘sensorimotor’ loop (Brembs, 2021;  
485 Gomez-Marin et al., 2011; Schroeder et al., 2010; Wolf et al., 2017; Yuste et al., 2005).

486

### 487 **Modulation of the exploration/exploitation balance**

488 The described internally generated movement dynamic provides an embedded solution for the  
489 compromise between exploration and exploitation. External visual cues can then modulate the  
490 exploration/exploitation balance to the task at hand by simply adjusting the amplitude of this  
491 endogenous dynamic. Higher amplitude oscillations optimize exploration in an unfamiliar  
492 environment, while lower amplitudes favor ‘exploitation’ (straighter paths) on a familiar route.  
493 Interestingly, this movement signature is equally useful, and used, during the acquisition of  
494 learnt visual information, when naïve ants display their learning walk around the nest  
495 (Jayatilaka et al., 2018) (Fig. 5). Optimising the acquisition of information requires to bias the  
496 balance toward ‘exploration’ in the same manner as searching for familiar cues in an unfamiliar  
497 environment does; thus it is not surprising to see here also high amplitude oscillations (Fig. 4,  
498 3). The apparent different needs for the acquisition of information on the one hand and the use  
499 of that information on the other hand, are thus solved with the same solution. The weaker  
500 modulation observed in *Iridomyrmex* may be explained by their strong use of chemical trails  
501 (Carde et al., 2016), whose absence on the trackball may favor exploration in both visually  
502 familiar and unfamiliar conditions.



503 Finally, we see in both species that the internally generated oscillations can be strongly – if not  
504 entirely – inhibited when not needed, such as when moving in the dark (Fig. 2). Indeed,  
505 oscillations are not always useful, and it may be advantageous to repress them in situations like  
506 when inside the nest, or when trying to escape an aversive situation. It is thus expected that the  
507 animal’s internal state as well as the presence/absence of relevant contextual information –  
508 such as a visual panorama by opposition to no panorama at all – modulates the expression of  
509 the internally produced oscillations in a similar vein as the male fly’s internal state continuously  
510 tunes up or down the gain of circuits promoting visual pursuit (Hindmarsh Sten et al., 2021).  
511 However, what specific type of visual information promotes the expression of oscillations  
512 remains to be seen.

513

#### 514 **Neural Implementation**

515 In the insect brain, regular lateral oscillations seem to result from reciprocal contra-  
516 lateral inhibitions between left and right hemispheric premotor areas, so-called Lateral  
517 Accessory Lobes (LAL). These circuit results in the internal production of asymmetrical and  
518 rhythmical excitation between left and right motor commands, so called flip-flop neurons  
519 (Berni, 2015; Berni et al., 2012; Iwano et al., 2010; Kanzaki, 2005; Namiki and Kanzaki,  
520 2016b; Steinbeck et al., 2020). Lateral oscillations are thus generated endogenously by  
521 structure analogous to central generator patterns (CPG). CPGs sustain a wide range of  
522 rhythmic functions such as chewing, breathing or various locomotor movements and can be  
523 typically modulated by sensory input (Marder and Calabrese, 1996; McAuley et al., 1999;  
524 Schroeder et al., 2010; Wolf et al., 2017; Yuste et al., 2005). Here however, rather than limb  
525 movements, the CPG-like in the insects’ LAL controls the displacement of the whole animal  
526 across space and thus, directly contributes to the navigational task.

527 The systematic positive covariation between angular and forward velocity observed here  
528 (Figs. 3, 5, S3) are unlikely to result from physical body constraints (where one could expect

529 the opposite, that is, a negative correlation between angular and forward velocities) and thus  
530 suggests that the internal dynamics of the LAL control both turning and forward velocity  
531 simultaneously. Remarkably, the observed relationship between forward and angular velocity  
532 readily emerges from our toy model when simply assuming that forward speed results from  
533 the overall excitation across both sides of the LAL; while turning velocities results from the  
534 difference between left and right excitation (Fig. 8). Bursts of forward speed appear when one  
535 side largely dominates over the other, that is when the ant is at the maximum speed of its  
536 angular sweep and thus roughly aligned with its overall direction of travel. Inversely, a break  
537 of forward speed occurs when the dominancy is reverted between LAL neurons, that is at the  
538 moment where ant reverts their turning direction (Fig. 8), as we can see in our ant data (Fig.  
539 3, 4, 5, S3). Certainly, we do not exclude that more complex circuitry could produce other  
540 regimes of covariations but our simple model shows that this particular relationship between  
541 forward and angular speed can readily emerge and is robust to modulation of the system. In  
542 contrast, the oscillations' amplitude and frequencies are somewhat sensitive to parameter  
543 change, at least across a factor of 2 (Fig. 8—figure supplements 1). Therefore, we can easily  
544 envision how various inputs into the LAL (such as pictured in Fig. 9) could modulate the  
545 amplitude and frequency of the emerging oscillations as we observed in our various  
546 conditions, without altering the fundamental relationship between forward and angular  
547 velocities. Understanding how such inputs actually operate in insects opens the door to a  
548 complexity that would be too vast to tackle here, but given the rapid development of  
549 neurobiological tools and knowledge about the circuitry of the lateral accessory lobes, this  
550 may constitute a realistic endeavour in the near future.

551

### 552 **Evolutionary consideration**

553

554 Several clues point to the idea that displaying such oscillations is an ancestral way of moving,  
555 that predates at least the holometabolous insects' common ancestor (around 300 Myr ago).

556 Indeed, comparable oscillations around 0.3-1Hz are observed in the two ant species studied  
557 here but also in moths (Kanzaki et al., 1992; Kanzaki and Mishima, 1996; Kuenen and Baker,  
558 1983; Olberg, 1983) *Drosophila* larvae (Wystrach et al., 2016), Colorado beetles  
559 (Lonnendonker, 1991) and other insects (Namiki and Kanzaki, 2016a). Furthermore, the LAL  
560 that likely produces these oscillations is an ancestral brain structure, largely conserved across  
561 insects (Namiki and Kanzaki, 2016b, 2016a; Steinbeck et al., 2020).

562 Some have suggested that neural systems evolved at first to coordinate movements  
563 endogenously (Keijzer et al., 2013; Yuste et al., 2005) whereas the modulation of these  
564 movements by external sensory information originated only as a second step (Keijzer et al.,  
565 2013). This view fits the observation that various multi-modal sensory cue converge to the  
566 same conserved region (the LAL) to produce a remarkable variety of navigational behaviors  
567 (Namiki et al., 2014; Namiki and Kanzaki, 2016b; Steinbeck et al., 2020). In crawling  
568 *Drosophila* larvae oscillations are modulated by variations in odor concentration, resulting in  
569 attraction toward (or repulsion from) the odor source (Wystrach et al., 2016). In flying male  
570 moths, it is the detection of a female's pheromone that modulates oscillations and enables the  
571 insect to track the odor plume. In walking ants also, zigzagging paths seem to be modulated  
572 by olfactory cues when following chemical trails (Hangartner, 1967). In wasps, the presence  
573 of a proximal visual object appears to trigger zigzag flights, which may provide useful visual  
574 depth information to the insect (Voss and Zeil, 1998). And finally, in visually navigating ants,  
575 oscillations are modulated by the recognition of familiar views to support route-following,  
576 visual exploration and the acquisition of visual memories in naïve individuals.

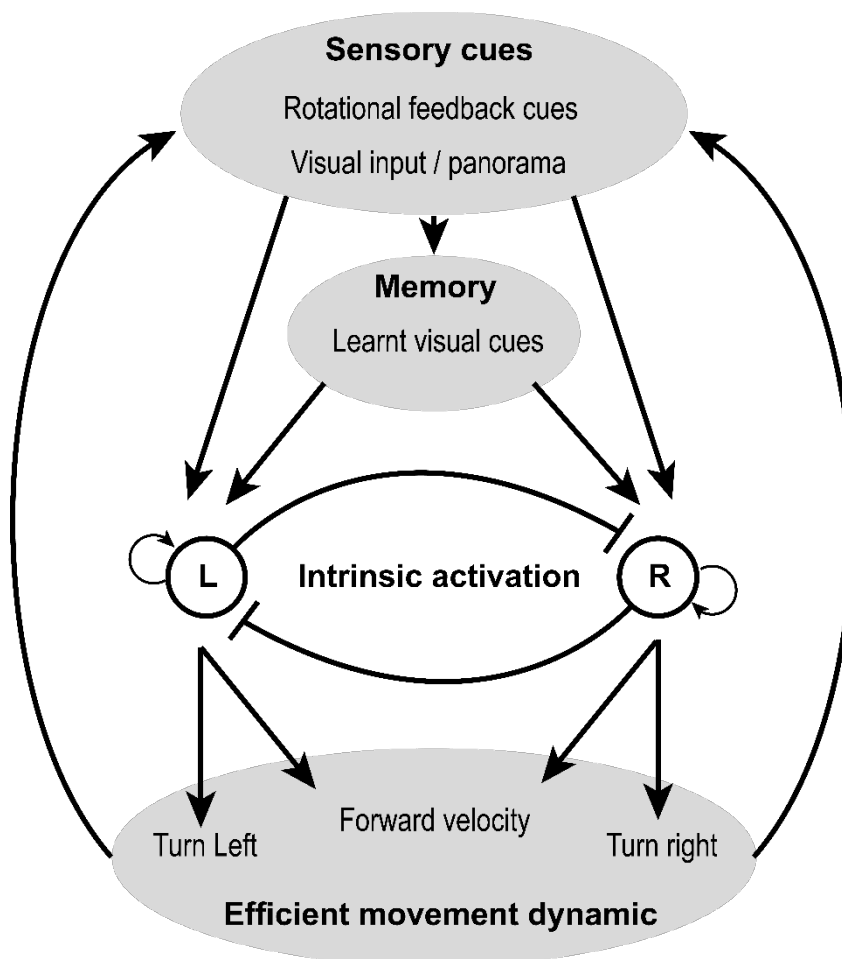
577 The modulation of oscillations by rotational visual cues, as we showed here in ants (Figs. 6,  
578 7) might be ancestral to insects as we equally observe it in moths (Namiki et al., 2014;  
579 Pansopha et al., 2014) (Namiki et al., 2014; Pansopha et al., 2014). This likely evolved  
580 through connections between horizontal optic-flow detectors in the visual lobes (Busch et al.,

581 2018) and the LAL (Fig. 9), providing a useful feedback-control to calibrate the amplitude of  
582 turns, and more generally participate in the widespread optomotor response. The modulation  
583 of oscillations by the presence/absence of visual cues, and more particularly  
584 familiar/unfamiliar visual cues however, must have appeared later with the evolution of  
585 visual route-following and homing in hymenopteran, through direct or indirect connections  
586 between the Mushroom bodies – the seat of navigational visual memories (Buehlmann et al.,  
587 2020; Kamhi et al., 2020; Webb and Wystrach, 2016) – and the LAL (Fig. 9).

588

589 Finally, the forward speed variation within the oscillation cycles presented here – which can  
590 emerge readily from the LAL connectivity (Fig. 8) – has not been reported in other species  
591 and thus could be a derived adaptation in ants. This may be an adaptation for walking. Even  
592 though ants can decouple their body orientation from their direction of travel if needed  
593 (Schwarz et al., 2017), walking sideways may be inefficient and preferably avoided due to  
594 physical constraints on leg movements. Thus, slowing down when looking on the side may be  
595 a compromise between the preference for walking forward (rather than sideways) and  
596 avoiding drifting away from the desired travel direction. Alternatively, this dynamic may be  
597 an adaptation for visual cue recognition in navigation, by opposition to the use of olfactory  
598 cues. When tracking an odor plume or trail, movements off track to the left and right (and  
599 crossing in between), provide useful information regarding the direction to follow. However,  
600 such sideway displacement off the beeline become useless for visual recognition (gaze  
601 *orientation* is what matters (Zeil 2012; Wystrach 2021)) and thus costly as they increase the  
602 overall distance walked. In addition, pausing when looking on the side must improve the  
603 efficiency of visual recognition due to gaze stabilization, which accommodates remarkably  
604 well the idea that views are learnt and recognized when oriented left and right, rather than  
605 toward and away from the goal (Stürzl et al., 2016; Wystrach et al., 2020).

606 These two hypotheses (i.e., adaptation to walking vs. visual recognition) could be tested  
607 straightforwardly by investigating the existence of such burst of forward speed during route-  
608 following in (1) walking ant species that do not use vision or in (2) flying insects that do use  
609 vision and are known to oscillate (e.g., bees or wasps) (Egelhaaf et al., 2012; Stürzl et al.,  
610 2016; Voss and Zeil, 1998). Overall, it is remarkable how a basal endogenous circuit  
611 producing a rhythm can be exapted to receive such a diversity of modalities and produce such  
612 a diversity of behaviours (Wystrach, 2021).  
613  
614



615  
616 **Figure 9. An intrinsic oscillator at the core of visual navigation.**

617 We propose a simple scheme to encompass our results. In this view, sensory input (in this  
618 case various innate and learnt visual information) act on behavior only indirectly, through the  
619 systematic modulation of an intrinsic oscillator. The latter ensures that an efficient movement

620 dynamic is preserved across navigational contexts, which would not necessarily be the case if  
621 various and potentially conflicting sensory information were directly modulating movement.  
622 This scheme also highlights the idea that action is not merely the product of perception. At  
623 the core of behavior lies an intrinsic, self-generated dynamic, which are modulated, rather  
624 than controlled, by sensory perception.  
625

## 626 **Conclusion**

627 Tracking a pheromone plume using olfaction, walking along a route using learnt visual cues  
628 or performing a learning walk as a naïve ant may appear as quite different tasks. However,  
629 our results support the idea that the solution to these challenges is based on a shared  
630 mechanisms: namely, the intrinsic production of left/right oscillations in a conserved pre-  
631 motor area (Kanzaki et al., 1992; Kodzhabashev and Mangan, 2015; Le Möel and Wystrach,  
632 2020; Murray et al., 2020; Pansopha et al., 2014; Steinbeck et al., 2020). This internally  
633 generated movement dynamic provides a remarkably optimized solution to the compromise  
634 between exploration and exploitation. Various sensory cues, through both innate or learnt  
635 pathways, can alter the exploration/exploitation balance to fit the task at hand by simply  
636 adjusting the amplitude of this endogenous dynamic. Evolution appears to have favored a  
637 general-purpose steering mechanism and it is notable how such a simple and repetitive  
638 oscillatory movement, generated intrinsically, accommodates such a diversity of input and be  
639 useful for such a diversity of tasks across species, modalities and ways of moving (i.e.,  
640 crawling, flying or walking).

## 641 **Methodology**

### 642 **Study Animal & Experimental site**

645 All experiments took place within an open grassy woodland at the National Australian  
646 University, Canberra from Feb. to Mar. 2019. We had the opportunity to work with two  
647 Australian endemic ant species: *Myrmecia croslandi* and *Iridomyrmex purpureus*. *Myrmecia*  
648 *croslandi* workers are known to forage solitarily, with each individual either hunting on the

649 ground in the vicinity of the nest or navigating routinely back and forth toward the same  
650 favorite foraging tree throughout her life span (Jayatilaka, 2014). These ants rely mainly on  
651 learnt terrestrial visual cues to navigate but are also able to resort to PI when the visual  
652 environment does not provide guidance (Jayatilaka et al., 2014, 2018; Narendra et al., 2013;  
653 Zeil et al 2014). Eight nests of *M. croslandi*, that foraged on two distinct trees between 6.0  
654 and 30.0 m away from the colony, were used in this study. *Iridomyrmex purpureus* ants form  
655 large colonies and forage along pheromone trails that lead to food patches. Despite  
656 employing pheromone trails for recruitment, this species also uses both learnt visual  
657 information and path integration for navigation (Card et al., 2016). We also performed  
658 navigational experiments with two *I. purpureus* nests to ensure that this species relies indeed  
659 on both learnt visual terrestrial cues and path integration, which they did surprisingly well  
660 (see Fig. S1). For the main experiment, a feeder was placed 7.0 m away from the nest and  
661 foragers were free to familiarize themselves with the route for 72 h before being tested.

662

### 663 **Trackball system and data extraction**

664

665 During tests, ants were mounted on a trackball device (Dahmen et al., 2017). This device  
666 consists of a polystyrene ball held in levitation in an aluminum cup by an air flow. The  
667 trackball has two sensors placed at 90° to the azimuth of the sphere, which record the  
668 movements of this sphere and translate them into X and Y data retracing the path of the ant.  
669 The X and Y acquisition of the trackball rotations happened at a 30 Hz frequency (i.e., 30  
670 data points per second), enabling us to reconstruct the ant's movements with high precision.  
671 Additionally, a camera (640×480 pixels) recording from above provided details of the ant's  
672 body orientation, also at 30 Hz. We also analyzed *M. croslandi* learning walks recorded  
673 directly on the natural ground. Head directions were obtained via video recordings at 25 Hz  
674 (provided by Jochen Zeil).

675 In this study, we used two different trackball configurations to record the ants' motor  
676 responses: further referred to as 'free ant' and 'fixed ant' experiments, respectively. Free  
677 ant experiment: two small wheels prevented the polystyrene ball from rotating in the  
678 horizontal plane, however, all other degrees of freedom of the ball rotation were accessible  
679 (Fig. S1 A; 'closed-loop' in Dahmen et al., 2017). Ants were attached on top of the ball by  
680 putting magnetic paint on their thorax and a micro-magnet fixed at the bottom end of a single  
681 dental thread that was in turn attached to a 0.5 mm pin. Crucially, the pin was placed within  
682 a glass capillary. This procedure enabled the ants to execute physical rotations on the ball (the  
683 ball is not rotating horizontally) but prevented any translational movement. Ants could thus  
684 execute body rotations and control the direction in which they faced but any attempt to go  
685 forward or backward resulted in ball rotations.

686 Fixed ant experiment: the two small wheels were no longer in place. Hence, the ball  
687 could now turn in any direction, including the horizontal plane (Fig. S1B; 'open-loop' in  
688 Dahmen et al., 2017). Ants were tethered directly to a needle with a micro magnet (glued at  
689 the end of the needle) and magnetic paint on their thorax. The top end of the needle was  
690 glued to a small piece of paper sheet (0.5×2.5 cm). Consequently, the fixed ant could no  
691 longer rotate, and the experimenter could choose in which direction she was facing. Any  
692 attempt to move, including turning, by the ants resulted in ball rotations. This trackball  
693 configuration was only conducted with the more robust *M. croslandi*.

694

#### 695 **Free ant experiments: protocol**

696 At the start of each test, the ant was mounted on the trackball device but surrounded by an  
697 opaque cylinder (30×30 cm) that prevented the ant from seeing any cues from the visual  
698 scenery around her. Once the ant was in place on the device, the whole apparatus was moved  
699 to the desired test location in the field. Afterward, the surrounding cylinder was removed,



700 revealing the visual scenery to the ant and data recording began. To ensure a high level of  
701 homing motivation, only individuals who had previously received a 40% sucrose solution  
702 (for *M. croslandi*) or a food item (for *I. purpureus*) were tested. The recording period was 3.5  
703 min for the robust *M. croslandi* and 1.5 min for the flimsier *I. purpureus*.

704 To test the impact of terrestrial visual cues on the oscillation behavior, ants were  
705 tested under three distinct conditions.

706 Familiar (F): ants were tested along their habitual route, which therefore presents a  
707 familiar visual view.

708 Unfamiliar (U): ants were tested at least 50 m away from the habitual route, which  
709 therefore presents an unfamiliar panorama.

710 Dark (D): ants were tested in total darkness, within an opaque cylinder (30×30 cm)  
711 covered with a red Plexiglas plate that transmitted only the low red wavelengths, which ants  
712 cannot perceive (Aksoy and Camlitepe, 2018; Ogawa et al., 2015).

713 To test the impact of PI on oscillation behavior, ants were tested as either full- or zero  
714 vector ants. Full vector (FV): ants were caught at the start of their inbound trip to the nest  
715 (i.e., at the foraging tree for *M. croslandi* and at the feeder for *I. purpureus*). FV ants have an  
716 informative homing PI vector, which points in the food-to-nest compass direction.

717 Consequently, FV ants can rely on both PI vector and the learnt visual scenery while being  
718 tested. Zero vector (ZV): homing ants were captured just before entering the nest, that is, at  
719 the end of their inbound trip. Hence, their PI homing vector is reduced to zero and thus no  
720 longer directionally informative. Consequently, ZV ants can only rely on the learnt visual  
721 scenery while being tested. For each of the three visual conditions FV and ZV ants were  
722 tested, resulting in a total of six conditions.

723 For *I. purpureus*, at least 16 ants were tested in each of the six conditions (F: FV&ZV  
724 =16; U: FV&ZV =17; D: FV=16 & ZV=17). Since *I. purpureus* forms very populous

725 colonies with an abundance of foragers, all individuals were tested only once, in one of the  
726 six conditions. The data obtained are therefore statistically independent. On the contrary, *M.*  
727 *croslandi* forms sparsely populated colonies and individuals usually make only one foraging  
728 trip per day (Jayatilaka 2014, a, b). Thus, it is time-consuming and challenging to capture,  
729 mark and follow individuals throughout foraging trips. Individuals were therefore captured  
730 (either at the foraging tree (FV) or before reaching their nest (ZV)) and tested successively in  
731 a pseudo-random order in all three visual panoramas: Familiar (F), Unfamiliar (U) and in the  
732 Dark (D). Both the sequence in which the visual conditions were experienced and the  
733 individuality were included in the statistical models as it is likely that the state of the PI  
734 vector will be modified across successive tests. Overall, 32 *M. croslandi* ants were tested  
735 with some individuals tested as ZV or FV ants on two different days. At least 16 *M. croslandi*  
736 ants were tested in each of the six conditions (F: FV&ZV =17; U: FV&ZV =17; D: FV=17 &  
737 ZV =16). Overall, 101 recordings were obtained.

738 In the ‘free ant’ experiments, the ant’s body axis can turn without the ball movement.  
739 We used the recorded videos to manually track the ants’ body orientation through time using  
740 the free software Kinovea. We removed the first 3s of recording of each ant as the removal of  
741 the opaque ring may have disrupted the behavior. Overall, the analyzed recording length was  
742 100s (except for two ants: 84 and 99s) for *M. croslandi* individuals and 50s (except for two  
743 ants: 49s) for *I. purpureus* ants. For details of the Fourier analysis (see below); nine paths of  
744 *I. purpureus* were discarded (F FV=1, U FV = 1, U ZV =2, D FV =2, D ZV = 3) as the ants  
745 displayed too many pauses.

746

#### 747 **Fixed ant experiments: protocol**

748 To test if oscillations are due to an intrinsic oscillator or caused by the fact that ants try to  
749 keep a bearing toward an external stimulus, we conducted an additional experiment on *M.*

750 *croslandi* ants. We recorded foragers without an informative integration vector (ZV) in the  
751 same unfamiliar environment (U) as before. But this time, ants were tested in eight different  
752 orientations, covering the 360° azimuth by bins of 45°; and with one direction corresponding  
753 to the food-to-nest compass direction. Each ant was tested in all eight orientations in a  
754 pseudo-random sequence. To change the ant's orientation, the experimenter would first place  
755 the opaque cylinder (30×30 cm) around the trackball to prevent individuals from perceiving  
756 any visual panorama, then rotate the whole set-up (trackball and mounted ant) and finally  
757 remove the opaque cylinder to re-start data collection. Ants were recorded for at least 15s up  
758 to 20s in each orientation. Eleven ants were tested in all eight orientations. Since the ants  
759 were fixed, trackball rotations along the horizontal axis provided a direct measure of the  
760 angular velocity of the attempted turn generated by the ant. Angular velocity time series were  
761 then extracted from the trackball data. At the end, the available length of the recordings for  
762 the analysis had 512 frames (~17s) except for 5 individuals (294,394,456,474,494 frames). It  
763 should be noted here that prior to the Fourier analysis (see below) we added a series of 0s  
764 (zero padding) at the end of the time series until it had a length of 3000 data points to match  
765 the same recording length obtained in the free ant experiment). This permits to increase the  
766 precision in the frequency range obtained from the Fourier analysis.

767

#### 768 **Fourier analysis:**

769 To reveal the occurrence of regular lateral oscillations, we choose to focus on the angular  
770 velocity value (Fig. S1C), which constitutes a direct reading from the left/right motor control.  
771 This time series was processed through three successive steps to obtain its 'spectral density',  
772 according to the Wiener-Khinchin theorem. First, the signal was parsimoniously smoothed  
773 with a moving median running of a length of five frames (0.17s) to reduce the influence of  
774 the recording noise (Fig. S1D, dashed blue line). Then, the recorded time series was passed

775 through an autocorrelation function (Fig. S1D). Finally, a Fourier transform was performed  
776 on these autocorrelation coefficients, providing the power spectral density (Fig. S1E). With  
777 this approach, the magnitudes obtained are independent from angular amplitudes of the  
778 oscillations and thus can be directly compared across individuals. A high magnitude indicates  
779 a strong oscillation for a given frequency. For each individual, the dominant frequency (i.e.,  
780 presenting the highest peak magnitude) and its magnitude were extracted (Fig S1E, dashed  
781 blue lines). To check whether these magnitudes indicate a significant regular oscillation, we  
782 compared them to the average spectral density magnitude resulting from 200 Gaussian white  
783 noise signals of the same length (within each species and experiment). Gaussian white noise  
784 signals were obtained by drawing a sequence of random values drawn from a normal law.  
785 These simulated signals were then processed through the exact same operations as the ants'  
786 angular velocity recording: namely smoothing, autocorrelation, Fourier transformation and  
787 extraction of the highest peak magnitude. The mean of the 200 highest magnitudes obtained  
788 was then compared to the real ants' equivalent magnitudes with a Wilcoxon one-tail test. As  
789 each experimental group has been compared with this mean magnitude of the simulated  
790 angular velocity signal, the p-value is subsequently adjusted using the Bonferroni correction.  
791

### 792 **Average oscillatory cycle**

793 To extract the average dynamics of an oscillation cycle, the mean cycles at the individual and  
794 population levels were reconstructed as follows. First, we smoothed the angular velocity time  
795 series of each individual by running twice a median with a window length of 31 frames for  
796 *M. croslandi* (for both free and fixed experiments), 25 frames for data during learning walks  
797 on the natural ground and seven frames for *I. purpureus*. This window length is much smaller  
798 than one oscillatory cycle and thus smoothens the data without altering the cycle general  
799 dynamics (Fig. 3—figure supplements 1A). Second, we indexed moments when the time

800 series crosses 0 (from – to +) as  $t_0$ ; and extracted a window of  $\pm 90$  frames around the  $t_0$   
801 indices ( $\pm 60$  frames for *I. purpureus*; Fig. 3—figure supplements 1A). We then reconstructed  
802 a mean cycle for each individual by averaging the individual's extracted windows, aligned at  
803  $t_0$  (Fig. 3—figure supplements 1B). The individuals' average forward speed dynamics during  
804 one cycle was obtained in the same way by using the same indices  $t_0$  obtained from the  
805 angular velocity data (Fig. 3—figure supplements 1C). For each ant, the mean angular  
806 velocity peak and amplitude of forward velocity cycles were extracted for analysis. Finally,  
807 we reconstructed the average cycle at the level of the population by averaging the mean cycle  
808 of all individuals. Note that to do so, each individual's mean cycle was first normalized to  
809 show similar amplitudes ( $\text{mean\_cycle\_normalised} = \text{mean\_cycle}/\text{mean}|\text{mean\_cycle\_values}|$ ).  
810 The goal was here to observe the cycle dynamics through time and not to estimate the inter-  
811 individual variation in amplitude, which was analyzed previously using the individual's mean  
812 cycle amplitude. Note that the criterion used to align the time series (the change from a left to  
813 a right turn) necessarily creates an artefact in the averaged angular velocity obtained.  
814 Namely, an average change from left turn to right turn at  $t_0$ . However, several factors indicate  
815 the relevance of such pooling at the population level: (1) the period of the average cycle  
816 corresponds to the mean frequency obtained from the Fourier transform. (2) we can observe a  
817 significant reversal of the angular velocity before and after the mean oscillation cycle. (3) the  
818 associated forward velocities co-vary in a significant way, indicating the existence of  
819 conserved dynamics.

820

### 821 **Analysis of the ants' direction of movement**

822 We reconstructed the ant paths that derived from the trackball recording to determine the  
823 mean direction of movement ( $\mu$ ) as well as the mean circular vector length ( $r$ , a measure of  
824 dispersion) of each individual. The mean directions ( $\mu$ ) were analyzed using a Rayleigh test

825 (from R package: Circular) that also includes a theoretical direction (analogous to the V-test).  
826 To test whether the angular data are distributed uniformly as a null hypothesis or if they are  
827 oriented toward the theoretical direction of the nest as indicated by the state of the PI or the  
828 familiar panorama, the average vectors lengths ( $r$ ) were analyzed via a Wilcoxon-Mann-  
829 Whitney test with a Bonferroni correction for multiple testing.

830

### 831 **Model**

832 For the free ant experiment, two types of models were tested: the first considering the  
833 interaction between visual panorama and vector modalities, the second with simple additive  
834 effect. It should be noted here that in case of deviation of the residuals of these models from  
835 normality and or homoscedasticity, the response variables have been transformed. Only the  
836 model presenting the lowest Akaike information criterion was retained and subsequently  
837 analyzed via an analysis of variance (Anova), followed by a post-hoc analysis of Tukey's  
838 rank comparison. Finally, for *M. croslandi* whose individuals were tested several times, the  
839 models are mixed models that consider these two random variables. For some models, we  
840 removed the sequence effect because of a singularity problem of this factor (i.e., the  
841 information given by this variable is already contained in other variables). For fixed ant  
842 experiments (which were tested only as ZV ants in an unfamiliar environment), the model  
843 analyzed the effect of the condition of the subsequent rotations. All data were processed and  
844 analyzed using free software R version 3.6.2.

845

### 846 **Acknowledgments**

847

848 We want to thank the Australian National University and particularly Jochen Zeil, Zoltán  
849 Kócsi and Trevor Murray for their advice and technical support. We also thank Hansjürgen  
850 Dahmen for providing us with the trackball system and Ajay Narendra for his incredible

851 knowledge about the local Australian ants. We are grateful to all these people for their helpful  
852 advice. We also thank Jochen Zeil, Paul Graham and Michael Mangan for their helpful  
853 feedback on the manuscript. Finally, we thank ants tested in these experiments for their  
854 participation.

855

## 856 **Additional information**

857 Funding

858 Funder: European Research Council, Grant reference number: EMERG-ANT 759817,

859 Author: Antoine Wystrach

860

861 Author contributions

862 LC Conception and Design of experiment, Data collection, Analysis and interpretation of

863 data, Drafting and revising the article. SS Conception and Design of experiment, Data

864 collection, Drafting and revising the article. AW Conception and Design of experiment, Data

865 collection, Analysis and interpretation of data, Drafting and revising the article, Supervision

866 of project.

867

868

## 869 References

- 870 Adden A, Stewart TC, Webb B, Heinze S. 2020. A neural model for insect steering applied to  
871 olfaction and path integration (preprint). *Neuroscience*.  
872 doi:10.1101/2020.08.25.266247
- 873 Aksoy V, Camlitepe Y. 2018. Spectral sensitivities of ants – a review. *Animal Biol* **68**:55–73.  
874 doi:10.1163/15707563-17000119
- 875 Baddeley B, Graham P, Husbands P, Philippides A. 2012. A Model of Ant Route Navigation  
876 Driven by Scene Familiarity. *PLoS Comput Biol* **8**:e1002336.  
877 doi:10.1371/journal.pcbi.1002336
- 878 Berni J. 2015. Genetic Dissection of a Regionally Differentiated Network for Exploratory  
879 Behavior in *Drosophila* Larvae. *Current Biology* **25**:1319–1326.  
880 doi:10.1016/j.cub.2015.03.023
- 881 Berni J, pulver SR, Griffith LC, Bate M. 2012. Autonomous Circuitry for Substrate  
882 Exploration in Freely Moving *Drosophila* Larvae. *Current Biology* **22**:10.
- 883 Brembs B. 2021. The brain as a dynamically active organ. *Biochemical and Biophysical*  
884 *Research Communications* **564**:55–69. doi:10.1016/j.bbrc.2020.12.011
- 885 Buehlmann C, Wozniak B, Goulard R, Webb B, Graham P, Niven JE. 2020. Mushroom  
886 Bodies Are Required for Learned Visual Navigation, but Not for Innate Visual  
887 Behavior, in Ants. *Current Biology* **30**:3438–3443.e2. doi:10.1016/j.cub.2020.07.013
- 888 Busch C, Borst A, Mauss AS. 2018. Bi-directional Control of Walking Behavior by  
889 Horizontal Optic Flow Sensors. *Current Biology* **28**:4037–4045.e5.  
890 doi:10.1016/j.cub.2018.11.010
- 891 Card A, McDermott C, Narendra A. 2016. Multiple orientation cues in an Australian trunk-  
892 trail-forming ant, *Iridomyrmex purpureus*. *Aust J Zool* **64**:227. doi:10.1071/ZO16046
- 893 Dahmen H, Wahl VL, Pfeffer SE, Mallot HA, Wittlinger M. 2017. Naturalistic path  
894 integration of *Cataglyphis* desert ants on an air-cushioned lightweight spherical  
895 treadmill. *J Exp Biol* **220**:634–644. doi:10.1242/jeb.148213
- 896 Egelhaaf M, Boeddeker N, Kern R, Kurtz R, Lindemann JP. 2012. Spatial vision in insects is  
897 facilitated by shaping the dynamics of visual input through behavioral action. *Front*  
898 *Neural Circuits* **6**. doi:10.3389/fncir.2012.00108
- 899 Freas CA, Cheng K. 2022. The Basis of Navigation Across Species. *Annu Rev Psychol*  
900 **73**:217–241. doi:10.1146/annurev-psych-020821-111311
- 901 Gomez-Marin A, Stephens GJ, Louis M. 2011. Active sampling and decision making in  
902 *Drosophila* chemotaxis. *Nat Commun* **2**:441. doi:10.1038/ncomms1455
- 903 Hangartner W. 1967. Spezifität und Inaktivierung des Spurphäromons von *Lasius fuliginosus*  
904 Latr. und Orientierung der Arbeiterinnen im Duftfeld. *Springer* **57**:34.  
905 doi:10.1007/BF00303068
- 906 Hindmarsh Sten T, Li R, Otopalik A, Ruta V. 2021. Sexual arousal gates visual processing  
907 during *Drosophila* courtship. *Nature* **595**:549–553. doi:10.1038/s41586-021-03714-w
- 908 Iwano M, Hill ES, Mori A, Mishima Tatsuya, Mishima Tsuneko, Ito K, Kanzaki R. 2010.  
909 Neurons associated with the flip-flop activity in the lateral accessory lobe and ventral  
910 protocerebrum of the silkworm moth brain. *J Comp Neurol* **518**:366–388.  
911 doi:10.1002/cne.22224
- 912 Jayatilaka P, Murray T, Narendra A, Zeil J. 2018. The choreography of learning walks in the  
913 Australian jack jumper ant *Myrmecia croslandi*. *J Exp Biol* **221**:jeb185306.  
914 doi:10.1242/jeb.185306



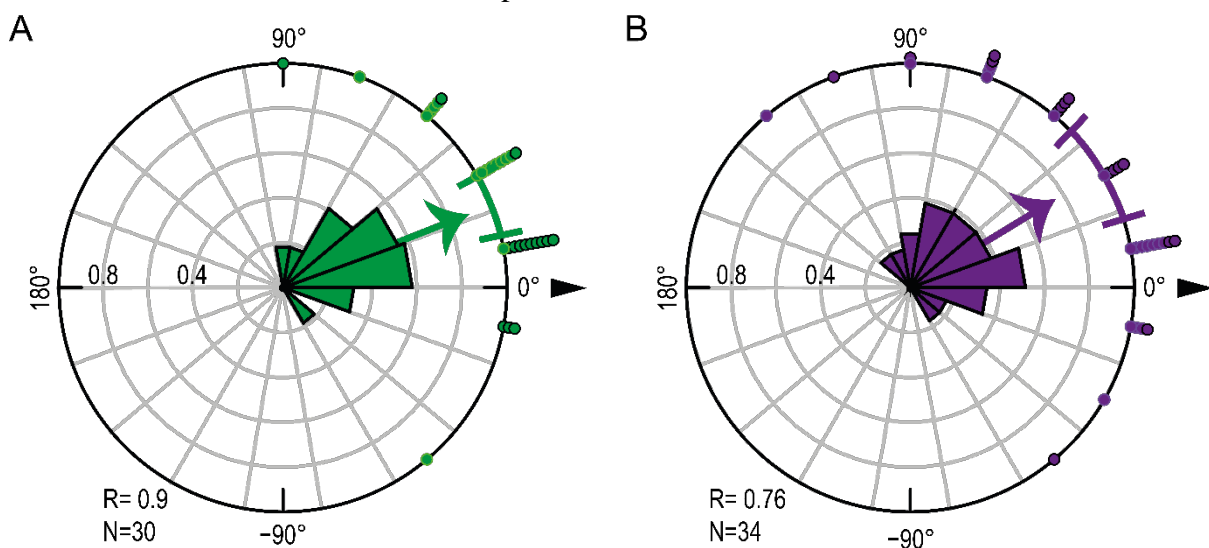
- 915 Jayatilaka P, Raderschall CA, Narendra A, Zeil J. n.d. Individual foraging patterns of the jack  
916 jumper ant *Myrmecia croslandi* (Hymenoptera: Formicidae) 10.
- 917 Jayatilaka PWA. 2014. Individual foraging careers of the Jack Jumper ant, *Myrmecia*  
918 *croslandi*. *thesis submitted for the degree of Doctor of Philosophy of the Australian*  
919 *180*.
- 920 Kamhi JF, Barron AB, Narendra A. 2020. Vertical Lobes of the Mushroom Bodies Are  
921 Essential for View-Based Navigation in Australian *Myrmecia* Ants. *Current Biology*  
922 **30**:3432-3437.e3. doi:10.1016/j.cub.2020.06.030
- 923 Kanzaki R. 2005. Neural Basis of Odor-source Searching Behavior in Insect Brain Systems  
924 Evaluated with a Mobile Robot. *Chemical Senses* **30**:i285–i286.  
925 doi:10.1093/chemse/bjh226
- 926 Kanzaki R, Mishima T. 1996. Pheromone-Triggered ‘Fiipflopping’ Neural Signals Correlate  
927 with Activities of Neck Motor Neurons of a Male Moth, *Bombyx mori*. *Zoological*  
928 *Science* **13**:79–87. doi:10.2108/zsj.13.79
- 929 Kanzaki R, Sugi N, Shibuya T. 1992. Self-generated Zigzag Turning of *Bombyx mori* Males  
930 during Pheromone-mediated Upwind Walking(Physiology). *Zoological science* **9**:515–  
931 527.
- 932 Keijzer F, van Duijn M, Lyon P. 2013. What nervous systems do: early evolution, input–  
933 output, and the skin brain thesis. *Adaptive Behavior* **21**:67–85.  
934 doi:10.1177/1059712312465330
- 935 Kodzhabashev A, Mangan M. 2015. Route Following Without Scanning In: Wilson SP,  
936 Verschure PFMJ, Mura A, Prescott TJ, editors. *Biomimetic and Biohybrid Systems*.  
937 Cham: Springer International Publishing. pp. 199–210. doi:10.1007/978-3-319-  
938 22979-9\_20
- 939 Kuenen LPS, Baker TC. 1983. A non-anemotactic mechanism used in pheromone source  
940 location by flying moths. *Physiol Entomol* **8**:277–289. doi:10.1111/j.1365-  
941 3032.1983.tb00360.x
- 942 Le Möel F, Wystrach A. 2020. Opponent processes in visual memories: A model of attraction  
943 and repulsion in navigating insects’ mushroom bodies. *PLoS Comput Biol*  
944 **16**:e1007631. doi:10.1371/journal.pcbi.1007631
- 945 Lonnendonker U. 1991. DYNAMIC PROPERTIES OF ORIENTATION TO A VISUALLY  
946 FIXATED TARGET BY WALKING COLORADO BEETLES. *Journal of*  
947 *Experimental Biology* **158**:16.
- 948 Murray T, Kócsi Z, Dahmen H, Narendra A, Le Möel F, Wystrach A, Zeil J. 2020. The role  
949 of attractive and repellent scene memories in ant homing ( *Myrmecia croslandi* ). *J*  
950 *Exp Biol* **223**:jeb210021. doi:10.1242/jeb.210021
- 951 Namiki S, Iwabuchi S, Pansopha Kono P, Kanzaki R. 2014. Information flow through neural  
952 circuits for pheromone orientation. *Nat Commun* **5**:5919. doi:10.1038/ncomms6919
- 953 Namiki S, Kanzaki R. 2016a. Comparative Neuroanatomy of the Lateral Accessory Lobe in  
954 the Insect Brain. *Front Physiol* **7**. doi:10.3389/fphys.2016.00244
- 955 Namiki S, Kanzaki R. 2016b. The neurobiological basis of orientation in insects: insights  
956 from the silkmoth mating dance. *Current Opinion in Insect Science* **15**:16–26.  
957 doi:10.1016/j.cois.2016.02.009
- 958 Narendra A, Gourmaud S, Zeil J. 2013. Mapping the navigational knowledge of individually  
959 foraging ants, *Myrmecia croslandi*. *Proc R Soc B* **280**:20130683.  
960 doi:10.1098/rspb.2013.0683
- 961 Ogawa Y, Falkowski M, Narendra A, Zeil J, Hemmi JM. 2015. Three spectrally distinct  
962 photoreceptors in diurnal and nocturnal Australian ants. *Proc R Soc B* **282**:20150673.  
963 doi:10.1098/rspb.2015.0673

- 964 Olberg RM. 1983. Pheromone-triggered flip-flopping interneurons in the ventral nerve cord  
965 of the silkworm moth, *Bombyx mori*. *J Comp Physiol* **152**:297–307.  
966 doi:10.1007/BF00606236
- 967 Pansopha P, Ando N, Kanzaki R. 2014. Dynamic use of optic flow during pheromone  
968 tracking by the male silkworm, *Bombyx mori*. *Journal of Experimental Biology*  
969 **217**:1811–1820. doi:10.1242/jeb.090266
- 970 Schroeder CE, Wilson DA, Radman T, Scharfman H, Lakatos P. 2010. Dynamics of Active  
971 Sensing and perceptual selection. *Current Opinion in Neurobiology* **20**:172–176.  
972 doi:10.1016/j.conb.2010.02.010
- 973 Schwarz S, Mangan M, Zeil J, Webb B, Wystrach A. 2017. How Ants Use Vision When  
974 Homing Backward. *Current Biology* **27**:401–407. doi:10.1016/j.cub.2016.12.019
- 975 Steinbeck F, Adden A, Graham P. 2020. Connecting brain to behaviour: a role for general  
976 purpose steering circuits in insect orientation? *J Exp Biol* **223**:jeb212332.  
977 doi:10.1242/jeb.212332
- 978 Stürzl W, Zeil J, Boeddeker N, Hemmi JM. 2016. How Wasps Acquire and Use Views for  
979 Homing. *Current Biology* **26**:470–482. doi:10.1016/j.cub.2015.12.052
- 980 Voss R, Zeil J. 1998. Active vision in insects: an analysis of object-directed zig-zag flights in  
981 wasps (*Odynerus spinipes*?, Eumenidae). *Journal of Comparative Physiology A:*  
982 *Sensory, Neural, and Behavioral Physiology* **182**:377–387.  
983 doi:10.1007/s003590050187
- 984 Webb B, Wystrach A. 2016. Neural mechanisms of insect navigation. *Current Opinion in*  
985 *Insect Science* **15**:27–39. doi:10.1016/j.cois.2016.02.011
- 986 Wolf S, Dubreuil AM, Bertoni T, Böhm UL, Bormuth V, Candelier R, Karpenko S,  
987 Hildebrand DGC, Bianco IH, Monasson R, Debrégeas G. 2017. Sensorimotor  
988 computation underlying phototaxis in zebrafish. *Nat Commun* **8**:651.  
989 doi:10.1038/s41467-017-00310-3
- 990 Wystrach A. 2021. Movements, embodiment and the emergence of decisions. Insights from  
991 insect navigation. *Biochemical and Biophysical Research Communications*  
992 **S0006291X21007348**. doi:10.1016/j.bbrc.2021.04.114
- 993 Wystrach A, Lagogiannis K, Webb B. 2016. Continuous lateral oscillations as a core  
994 mechanism for taxis in *Drosophila* larvae. *eLife* **5**:e15504. doi:10.7554/eLife.15504
- 995 Wystrach A, Le Moël F, Clement L, Schwarz S. 2020. A lateralised design for the interaction  
996 of visual memories and heading representations in navigating ants (preprint). *Animal*  
997 *Behavior and Cognition*. doi:10.1101/2020.08.13.249193
- 998 Wystrach A, Mangan M, Philippides A, Graham P. 2013. Snapshots in ants? New  
999 interpretations of paradigmatic experiments. *Journal of Experimental Biology*  
1000 **216**:1766–1770. doi:10.1242/jeb.082941
- 1001 Yuste R, MacLean JN, Smith J, Lansner A. 2005. The cortex as a central pattern generator.  
1002 *Nat Rev Neurosci* **6**:477–483. doi:10.1038/nrn1686
- 1003 Zeil J. 2012. Visual homing: an insect perspective. *Current Opinion in Neurobiology* **22**:285–  
1004 293. doi:10.1016/j.conb.2011.12.008
- 1005 Zeil J, Narendra A, Stürzl W. 2014. Looking and homing: how displaced ants decide where  
1006 to go. *Phil Trans R Soc B* **369**:20130034. doi:10.1098/rstb.2013.0034
- 1007 Zollikofer CPE. 1994. STEPPING PATTERNS IN ANTS. *Journal of Experimental Biology*  
1008 **192**:107–118.  
1009
- 1010

1011 **Supplemental information 1**

1012 To this date, the navigational skills of *Iridomyrmex purpureus* have been investigated in only  
1013 one study showing that these individuals are able to use terrestrial as well as celestial cues for  
1014 orientation (Card et al 2016). Before using this species in our study, we performed similar  
1015 experiments and found that this ant relies indeed on both sources of information to navigate.  
1016 First, to test whether *I. purpureus* was able to use visual familiarity foragers were captured at  
1017 the end of their inbound trip where the vector of integration was reduced to zero (ZV) and re-  
1018 released (tested) along their route. Second, to test if *I. purpureus* was able to use celestial  
1019 information foragers were captured at the feeder at the start of their inbound trip (FV) and re-  
1020 released (tested) in an unfamiliar location ~50 m away (see Methodology).

1021 For tests, ants with a food item were released at the center of a goniometer divided in  
1022 18 sectors (20° each) either in a familiar or unfamiliar surroundings. The taken directions of  
1023 the tested ants were recorded at two points: 15 cm and 27 cm. We used the “corrected



1024 direction” for our analyses, which is the direction taken between these two points. Note, that  
1025 after several tested individuals, the goniometers was rotate (180°) to prevent the use of  
1026 chemical trails.

1027

1028 **Figure S1: The meat ant *Iridomyrmex purpureus* uses both visual and celestial cues to**  
1029 **navigate.**

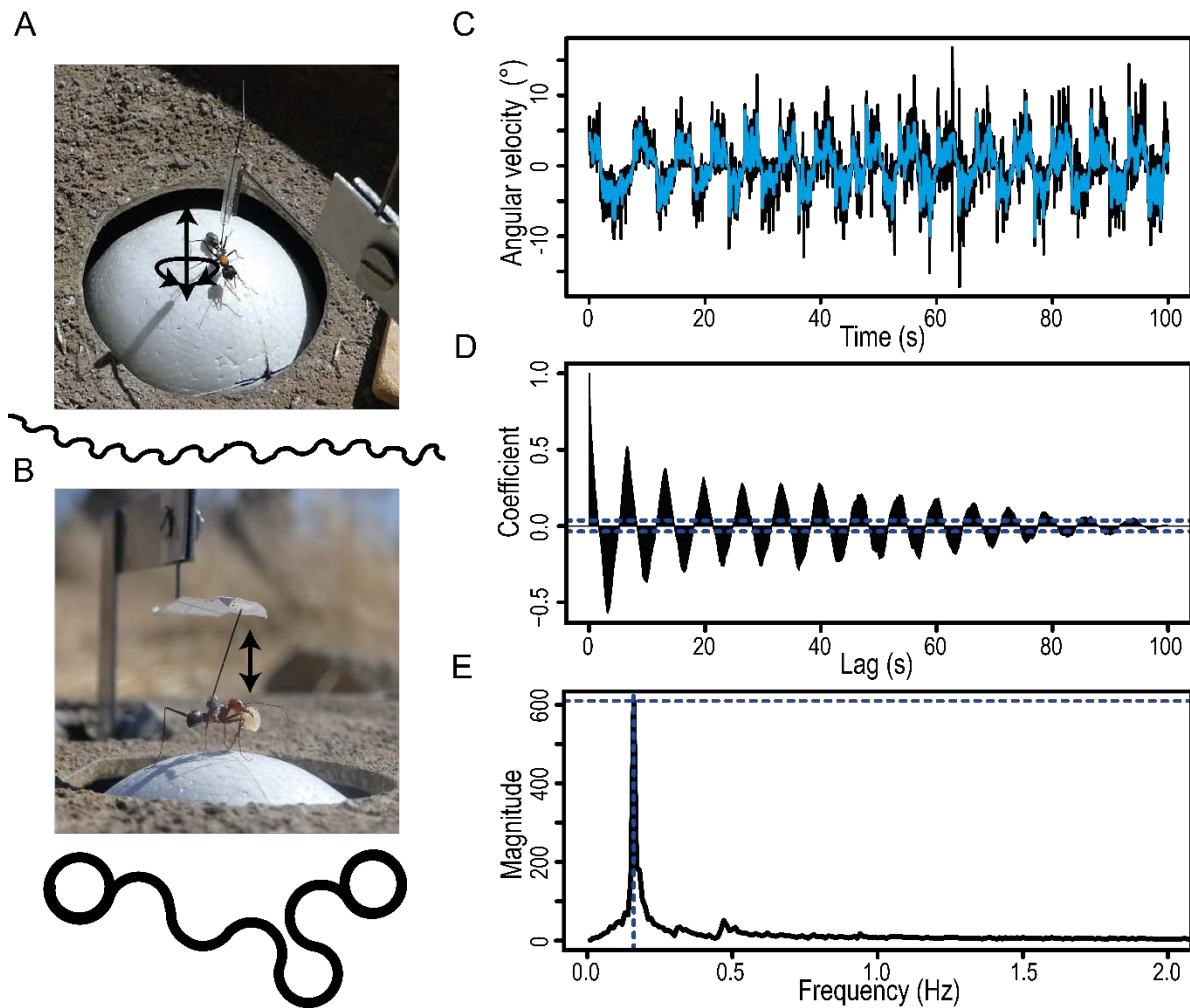
1030 (A) Circular histogram depicting the directional decisions of tested zero-vector ants (without  
1031 informative integration vector). (B) Circular histogram depicting the directional decisions of  
1032 tested full-vector ants (with informative integration vector). For A and B, nest direction is  
1033 indicated by the  $0^\circ$  direction (black triangle). The arrow corresponds to the mean vector of  
1034 the distribution. The 95% confidence interval of the mean is displayed as a colored arc. Each  
1035 dot corresponds to one corrected direction obtained from one individual. Dots with a black  
1036 rim are data recorded before rotation of the goniometers and the dots without black rim after  
1037 rotation.

1038

1039 First, the results show that ZV ants of *I. purpureus* use the familiar visual surrounding to  
1040 orient toward their nest (Fig. S1A; Rayleigh test with nest as theoretical direction  $0^\circ$ ;  
1041  $p < 0.001$ ). Secondly, in unfamiliar panorama results clearly show that FV ants followed the  
1042 direction indicated by celestial cues ( $0^\circ$  on the Fig. S1B; Rayleigh test  $p < 0.001$ ). Therefore,  
1043 this species does not only rely on chemical trails but also on the visual panorama and the path  
1044 integration during foraging.

1045

1046 **Supplemental information 2**



1047

1048

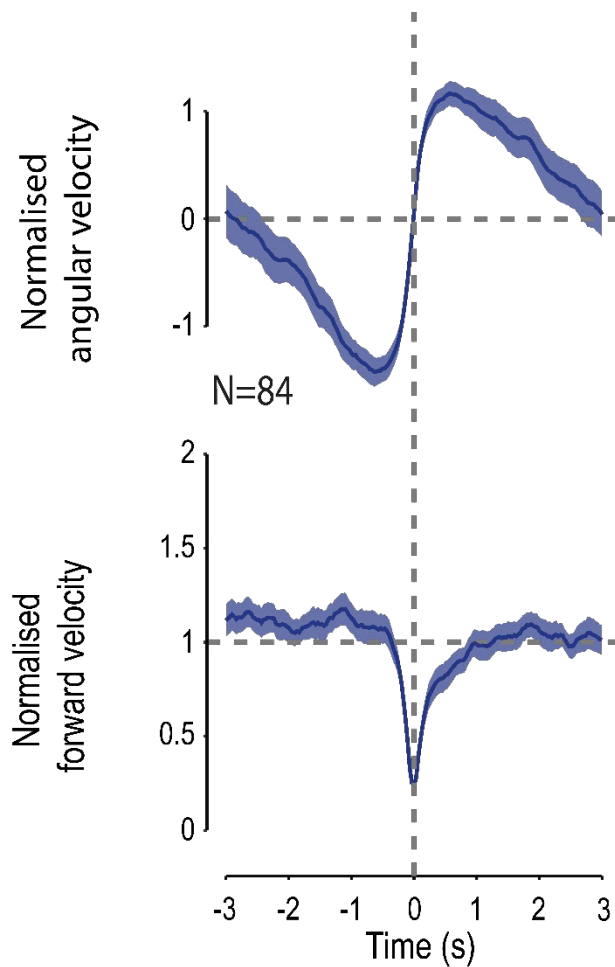
1049 **Figure S2: Trackball set-ups, recording and processing of the ants' trajectory.** (A)  
1050 Trackball set-up for 'free ant experiment' (top view). Two wheels prevent the sphere from  
1051 rotating along the horizontal plane. Ants are free to rotate their body along the yaw axis to  
1052 control in which direction they perceive the world. (B) Trackball set-up for 'fixed ant  
1053 experiment' (side view). The two wheels have been removed, so the ball can turn in every  
1054 direction. Ants are fixed in a way that prevent body rotations along the yaw axis, so they are  
1055 forced to keep their bearing in the imposed direction. For A and B an example path has been  
1056 plotted below the picture. (C) Angular velocity signal over time (recorded at 30 readings/sec)  
1057 in an individual of the species *M. croslandi* (black) with smoothed signal superimposed  
1058 (blue). (D) Autocorrelation carried out on the entire smoothed angular velocity signal. (E)  
1059 Fourier transformation of the autocorrelation coefficients signal (shown in D) provides the  
1060 'spectral density'. This approach has the advantage to provide magnitudes that are directly  
1061 comparable between individuals. For each individual, the frequency peak with the highest  
1062 magnitude was extracted, indicating a strong oscillation of the angular velocity signal at that  
1063 frequency (dashed blue lines).

1064

1065

1066

1067 **Supplemental information3:**



1068

1069

1070 **Figure S3: Absence of the rotational visual feedback does not modify the dynamical**  
1071 **signature of oscillation behavior.**

1072 Angular velocities (top-row) and forward velocity (bottom row) co-varies in a way that seem  
1073 conserved across experimental conditions (Fig. 3 & Fig. 5). Population cycles have been  
1074 reconstructed by merging all rotational condition as there is no differences of amplitude of  
1075 mean peak of angular velocity and mean forward amplitude cycles between rotational  
1076 conditions (Anova:  $Df = F_{7,84} = 1.0103$ ,  $p \geq 0.432$ ). Each individual signal has been normalized  
1077 before pooling. Colored areas around mean curves represent the 95% confidence interval,  
1078 based on the inter-individual variation.

1079

1080