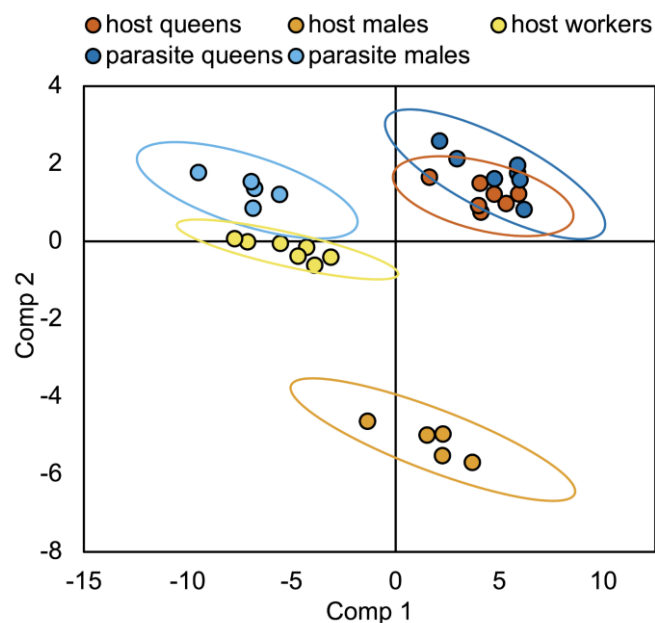


# The inquiline ant *Myrmica karavajevi* uses both chemical and vibroacoustic deception mechanisms to integrate into its host colonies

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## Supplementary material



**Supplementary Figure 1.** Representation of standardized components 1 and 2 of partial least squares discriminant analysis for log-transformed concentrations (ng/cm<sup>2</sup>) of cuticular hydrocarbons found on the cuticle of parasite and host castes (qS = *M. scabrinodis* queen; mS = *M. scabrinodis* males; wS = *M. scabrinodis* workers; qK = *M. karavajevi* queen; mK = *M. karavajevi* males). The two components accounted respectively for 61%, 13% of the total variation.

**Supplementary Table 1.** List of the 15 vibroacoustic parameters measured on the pulses, i.e., call fundamental unit, produced by each individual ant.

<b>Vibroacoustic parameters</b>	<b>Description</b>	<b>Unit</b>
Q25%	Lower quartile of the energy spectrum	Hz
Q50%	Central quartile of the energy spectrum	Hz
Q75%	Higher quartile of the energy spectrum	Hz
SDQ50	Frequency standard deviation	Hz
Fpeak	Frequency peak	Hz
EFpeak	Energy of the frequency peak	Pa <sup>2</sup> s <sup>-1</sup>
%EFpeak	Relation of the frequency peak energy to the call total energy	%
A	Mean amplitude	dB
P	Power	dB <sup>2</sup>
E	Energy	Pa <sup>2</sup> s <sup>-1</sup>
I	Intensity value	dB
RMS	Root-mean-square signal level	Pa
Δt	Duration of the pulse	s
PR	Pulse Rate, calculated as 1/tstart(x)-tstart(x+1))	s-1
CG	Centre of gravity	Hz

**Supplementary Table 2** Results of Kruskal-Wallis test and Wilcoxon-Mann-Whitney Test on rescue order and time (s) of brood items within *M. scabrinodis* colonies. PQWP = parasite queen white pupae; HQWP = host queen white pupae; HWBP = host worker brown pupae.

<b>Variables</b>	<b>Kruskall-Wallis test</b>	<b>Rescue order</b>	<b>Mann-Whitney Test</b>	<b>qKWP - qSWP</b>	<b>qKWP - wSBP</b>	<b>qSWP - wSBP</b>
<b>Rescue order</b>	H	18.9850	U	-2.3635	-4.0042	-2.7165
	p-value	<0.0001	p-value	<b>0.0178</b>	<b>&lt;0.0001</b>	<b>0.0073</b>
<b>Rescue time</b>	H	10.1350	U	<b>-1.3058</b>	<b>-2.9948</b>	<b>-2.1701</b>
	p-value	0.0063	p-value	<b>0.1982</b>	<b>0.0031</b>	<b>0.0307</b>

**Supplementary Table 3.** Results of linear models on the vibroacoustic parameters calculated on the stridulations emitted by *M. scabrinodis* queens and workers and *M. karavajevi* queens. Likelihood ratio (LR) statistics and results of Tukey multiple comparisons are reported for the delta time ( $\Delta t$ , s), pulse rate (PR,  $s^{-1}$ ), root-mean-square signal level (RMS, Pa), third frequency quartile (Q75%, Hz), the frequency standard deviation (SDQ50, Hz), frequency peak (Fpeak, Hz), the relation of the frequency peak energy to the call total energy expressed as a percentage (%EFpeak, %). qS = *M. scabrinodis* queen; wS = *M. scabrinodis* worker; qK = *M. karavajevi* queen queen.

		LR	Tukey	qS - qK	wS - qS	wS - qK
<b><math>\Delta t</math></b>	Chisq	17.021	t value	4.131	1.247	2.901
	p-value	<b>0.0002</b>	p-value	<b>&lt;0.001</b>	0.4283	<b>0.0125</b>
<b>PR</b>	Chisq	5.4498	z value	-1.782	2.187	0.375
	p-value	0.0655	p-value	0.181	0.078	0.925
<b>RMS</b>	Chisq	68.727	z value	1.811	7.316	9.028
	p-value	<b>&lt;0.0001</b>	p-value	0.171	<b>&lt; 0.0001</b>	<b>&lt; 0.0001</b>
<b>Q75%</b>	Chisq	105.06	z value	10.161	2.068	12.201
	p-value	<b>&lt;0.0001</b>	p-value	<b>&lt; 0.0001</b>	<b>0.101</b>	<b>&lt;0.0001</b>
<b>SDQ50</b>	Chisq	229.53	z value	19.490	6.831	26.228
	p-value	<b>&lt;0.0001</b>	p-value	<b>&lt; 0.0001</b>	<b>&lt; 0.0001</b>	<b>&lt; 0.0001</b>
<b>Fpeak</b>	Chisq	13.275	z value	-3.434	0.488	-2.953
	p-value	<b>0.0013</b>	p-value	<b>0.0024</b>	0.8773	<b>&lt; 0.0001</b>
<b>%EFpeak</b>	Chisq	4.8531	z value	-0.803	2.175	1.342
	p-value	0.0883	p-value	0.702	0.080	0.375

**Supplementary Table 4.** Results of generalized linear models of *M. scabrinodis* worker responses to different vibroacoustic stimuli. Likelihood ratio (LR) statistics and results of Tuckey multiple comparisons are reported for the sum of positive behaviours (total) and each behaviour separately (walking, antennating, staying). qS = host queen; qK = parasite queen; wS = host worker; WN = white noise.

		LR	Tuckey	qS - qK	qS-wS	qS-WN	qK-wS	qK-WN	wS-WN
<b>total</b>	Chisq	104.4400	z value	1.278	4.661	8.22	3.457	7.341	4.613
	p-value	<b>&lt;0.0001</b>	p-value	0.56768	<b>&lt; 0.001</b>	<b>&lt; 0.001</b>	<b>0.00284</b>	<b>&lt; 0.001</b>	<b>&lt; 0.001</b>
<b>walking</b>	Chisq	43.7570	z value	1.504	2.821	5.846	1.349	4.65	3.491
	p-value	<b>&lt;0.0001</b>	p-value	0.42853	<b>0.02389</b>	<b>&lt; 0.001</b>	0.52522	<b>&lt; 0.001</b>	<b>0.00255</b>
<b>antennating</b>	Chisq	45.6760	z value	0.213	2.751	4.661	2.557	4.554	2.991
	p-value	<b>&lt;0.0001</b>	p-value	0.9963	<b>0.0273</b>	<b>&lt;0.001</b>	<b>0.0469</b>	<b>&lt;0.001</b>	<b>0.0132</b>
<b>staying</b>	Chisq	36.0440	z value	0	0.004	0.004	0.004	0.004	0
	p-value	<b>&lt;0.0001</b>	p-value	1	1	1	1	1	1

**Supplementary Table 5.** Results of Kruskal-Wallis test and Wilcoxon-Mann-Whitney test on the main features of the stridulatory organs of host and parasite castes, i.e. the width and height of the pars stridens and the number of ridges counted along 100  $\mu\text{m}$ . qS = host queen; qK = parasite queen; wS = host worker.

Variables	Kruskall-Wallis test	Rescue order	Mann-Whitney Test	qK - wS	qS - wS	qK - qS
Width of pars stridens ( $\mu\text{m}$ )	H p-value	11.359 0.003414	U p-value	0.91287 0.4256	2.8823 <b>0.0022</b>	-2.7386 <b>0.0028</b>
Length of pars stridens ( $\mu\text{m}$ )	H p-value	14.235 0.0008107	U p-value	-2.7386 <b>0.0055</b>	2.8823 <b>0.0017</b>	-2.7386 <b>0.0035</b>
nr of ridges along 100 $\mu\text{m}$	H p-value	13.069 0.001453	U p-value	2.7767 <b>0.0014</b>	-2.4031 <b>0.0274</b>	2.7767 <b>0.0041</b>

Supplementary Table 6. Mean ( $\pm$  SD) values of body surface ( $\text{mm}^2$ ) for each caste (for details about calculation of body surface see von Beeren *et al.* 2011).

Body surface ( $\text{mm}^2$ )	<i>M. karavajevi</i> queens (N=7)	<i>M. karavajevi</i> males (N=5)	<i>M. scabrinodis</i> queens (N=7)	<i>M. scabrinodis</i> males (N=5)	<i>M. scabrinodis</i> workers (N=7)
Mean ( $\pm$ SD)	40.8 $\pm$ 3.5	35.2 $\pm$ 2.7	84.7 $\pm$ 5.1	77.9 $\pm$ 0.5	43.0 $\pm$ 6.1

## References

von Beeren, C.; Schulz, S.; Hashim, R.; Witte, V. Acquisition of chemical recognition cues facilitates integration into ant societies. BMC Ecol. 2011, 11, 1–13. doi.org/10.1186/1472-6785-11-30. doi.org/10.1186/1472-6785-11-30.