



Digital supplementary material to

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**Insights into social organization and population genetics of introduced *Odontomachus troglodytes* (Hymenoptera: Formicidae) in Taiwan**

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Appendix, as digital supplementary material to this article, at the journal's web pages:

Fig. S1: Rapid antennation (RA) duration box chart for each encounter (encounters over 10 are grouped together), plotted using R to illustrate the overtime bout duration pattern among the five groups.

Fig. S2: Combined rapid antennation (RA) duration across both populations.

Tab. S1: Colony IDs, collected counties, and the latitudes and longitudes of collected nests. Latitude and longitude data are represented in WGS84 (World Geodetic System 1984).

Tab. S2: Characteristics of the developed microsatellite markers.

Tab. S3: Model selection for rapid antennation duration analysis using quasi-information criterion (QIC).

Tab. S4: Pairwise relatedness values among *O. troglodytes* individuals in Chiayi and Kaohsiung populations.

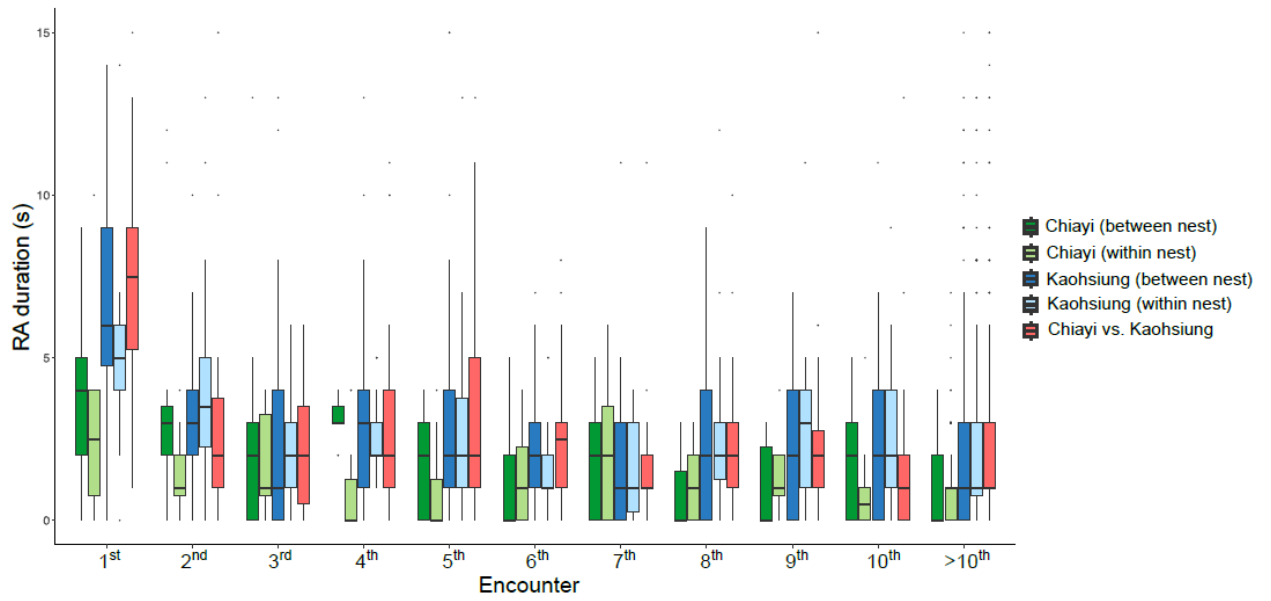


Fig. S1: Rapid antennation (RA) duration box chart for each encounter (encounters over 10 are grouped together), plotted using R to illustrate the overtime bout duration pattern among the five groups. Dark green: Duration of nests-to-nest in Chiayi; Light green: Duration of within nests in Chiayi; Dark blue: Duration of nests-to-nest in Kaohsiung; Light blue: Duration of within nests in Kaohsiung; Red: Chiayi vs. Kaohsiung.

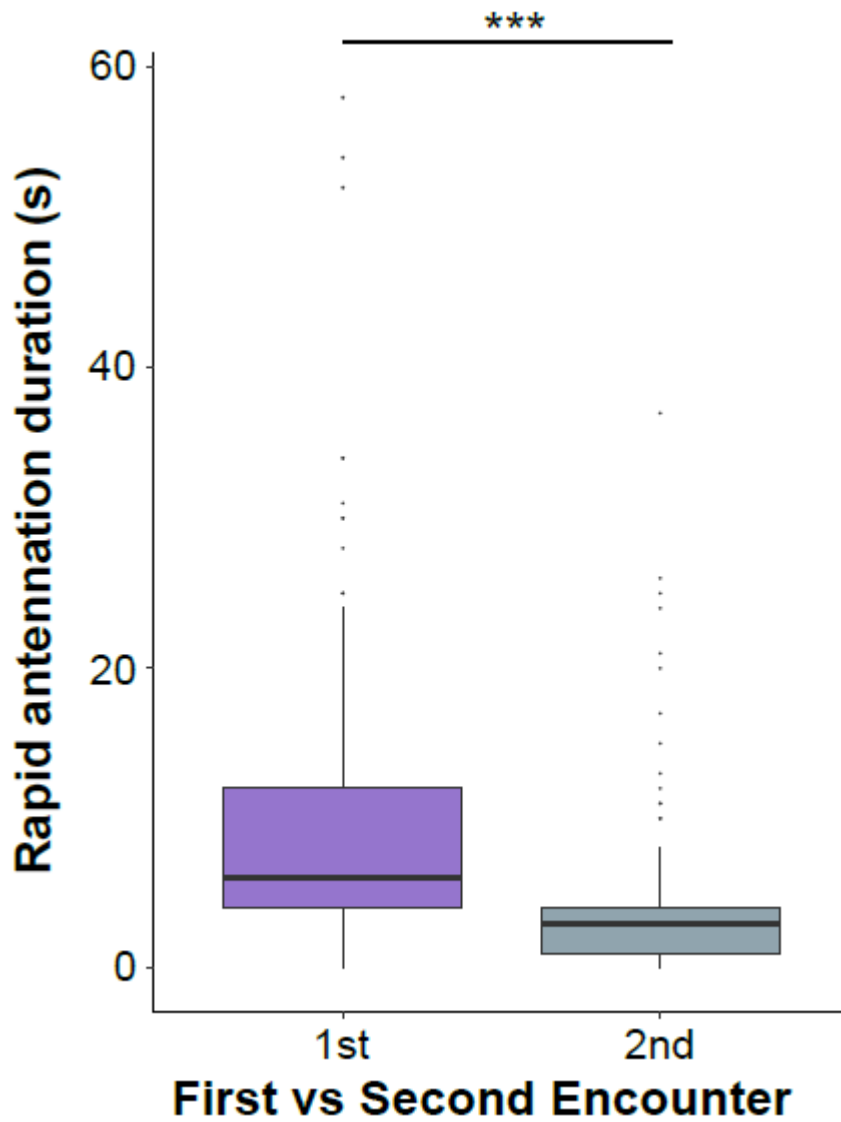


Fig. S2: Combined rapid antennation (RA) duration across both populations. RA duration during the first and second encounters is shown for all assays pooled from Chiayi and Kaohsiung. This summary figure illustrates the overall pattern of decreased RA duration during second encounters. Statistical differences were tested using Generalized Estimating Equations (\*\* $p < 0.001$ ). Note: This figure presents pooled data and does not reflect population-level differences, which are presented in Figure 3.

Tab. S1: Colony IDs, collected counties, and the latitudes and longitudes of collected nests. Latitude and longitude data are represented in WGS84 (World Geodetic System 1984).

<b>Nest</b>	<b>County</b>	<b>Latitude</b>	<b>Longitude</b>
<b>CY01</b>	Chiayi	23.49076	120.47226
<b>CY02</b>	Chiayi	23.49076	120.4723
<b>CY03</b>	Chiayi	23.49069	120.47228
<b>CY04</b>	Chiayi	23.49069	120.47276
<b>CY05</b>	Chiayi	23.49076	120.47264
<b>KH01</b>	Kaohsiung	22.61907	120.37898
<b>KH02</b>	Kaohsiung	22.61907	120.37921
<b>KH03</b>	Kaohsiung	22.61898	120.37931
<b>KH04</b>	Kaohsiung	22.59272	120.37104
<b>KH05</b>	Kaohsiung	22.59255	120.37126
<b>KH06</b>	Kaohsiung	22.55818	120.38945
<b>KH07</b>	Kaohsiung	22.55820	120.38947

Tab. S2: Characteristics of the developed microsatellite markers.

Marker	Repeat motif	Primer sequence (5'-3')	Ta (°C)	Na	He	Ho	Size range (bp)
OT1	(CT) <sup>16</sup>	F: AACAGGGTGC GTTCTCTCTC R: TAATTTTCGTGGCACGGCAC	58	2	0.241	0.179	112-114
OT2	(GTC) <sup>12</sup>	F: CGTAATTAACGCCTCGACGC R: AATCCGTCCGCTTATGGTGC	58	2	0.373	0.27	112-115
OT3	(AG) <sup>13</sup>	F: CTTTGTAACCCCGACGAAGC R: TCTCCCTCTCTCCCTCTCAC	58	—	—	—	
OT4	(GA) <sup>11</sup>	F: ACGAGTCGGTATGTGCCTTC R: AACGACACGAAAACAAGCGG	58	2	0.214	0.047	113-115
OT5	(CA) <sup>11</sup>	F: ACCGTCACACTGTCGTTACG R: GATTCAACGAACCGTCTCGC	58	3	0.603	0.365	109-134
OT6	(AG) <sup>14</sup>	F: GAGAGAAACGGGAAAGGGGG R: TACCGACTGCTACGTTGCTC	58	—	—	—	
OT7	(CT) <sup>14</sup>	F: AACCTGCAGGGAGACGTTAC R: GAGAGAGAGCGAGAGAGAGAAG	58	—	—	—	
OT8	(CT) <sup>11</sup>	F: AGAAAACGCGCCTCTCTCTC R: GAGTCGCCGATTGAAATG	58	3	0.616	0.382	112-116
OT9	(AG) <sup>12</sup>	F: AACGAGACAAAGTAGGGAGTGG R: TCTCTCTCTTTCTCCCTCCC	58	—	—	—	
OT10	(AGTG) <sup>11</sup>	F: ACGTGAGTGAGTGAGTGAGTG R: ACACACACACACACACACAC	58	—	—	—	
OT11	(CT) <sup>11</sup>	F: TCTGCGTGTCAGACAGGTTTC R: GATGAATCAACGCGGCAGAC	58	1	0	0	120
OT12	(CA) <sup>14</sup>	F: TTTCCCGTTGGAGTATGGCC R: AAGGCCGGAGACTTTTGGAG	58	3	0.549	0.418	207-211
OT13	(AT) <sup>12</sup>	F: GTGAAACTGCCGTCGTTCTG R: GTGAAACTGCCGTCGTTCTG	58	—	—	—	
OT14	(TC) <sup>13</sup>	F: TACGTCACACCAGCTGGAAC R: GAATCGATTGACCTCCGGG	58	—	—	—	
OT15	(TG) <sup>11</sup>	F: AGCAGCGTCCCTTAAGTACG R: TCCAAGCGCGGTAACAG	58	2	0.108	0.085	215-221
OT16	(CA) <sup>12</sup>	F: ACACGCACACACTCTCTC R: ATTACGAGCGACGAGAGCTG	58	2	0.237	0.142	212-214
OT17	(CT) <sup>12</sup>	F: TTTCGGGCAATGCGTTTCTG R: GCTAATACCGACAGGGGAGC	58	2	0.211	0.151	216-219
OT18	(CT) <sup>13</sup>	F: CCACGCTCGCTTTGATAAC R: TACAACGACCGCTAATGG	58	3	0.502	0.437	218-226
OT19	(TA) <sup>12</sup>	F: AATTTCTCCCGATTCTCTG R: TGAAGTACAGCTCGTACGAC	58	—	—	—	
OT20	(AG) <sup>15</sup>	F: GGCTGGAGAGGACATGATGG	58	2	0.204	0.183	212-214

R: GGTCTCACCTCCTCTTTCGC

Empty entries in columns indicate NA values for markers that could not be amplified and were excluded from subsequent analyses.

He: Expected heterozygosity; Ho: Observed heterozygosity; Na: Number of alleles in workers; Ta: Annealing temperature; Size range: Product size of amplified alleles.

Tab. S3: Model selection for rapid antennation duration analysis using quasi-information criterion (QIC).

Model	QIC	Parameters
Encounter Sequence × Group	69934.51	10
Encounter Sequence + Group	70873.92	6
Encounter Sequence × Category	70669.42	6
Encounter Sequence + Category	71244.84	4
Encounter Sequence only	71750.12	2

Group levels: Chiayi (between-nests), Chiayi (within-nest), Kaohsiung (between-nests), Kaohsiung (within-nest), and cross-regional Chiayi–Kaohsiung pairings. Category levels: within-nest (workers from the same nest), between-nest (workers from different nests within the region), and between-region (workers from Chiayi versus Kaohsiung).

Tab. S4: Pairwise relatedness values among *O. troglodytes* individuals in Chiayi and Kaohsiung populations. Relatedness was estimated using the Queller & Goodnight (1989) estimator and grouped by pair type (Worker–Worker or Queen–Worker) and nest category (within-nest or between-nest).

Region	Pair Type	N	Mean	SD
Kaohsiung	Worker-Worker (within)	30	0.188	0.514
Kaohsiung	Worker-Worker (between)	160	-0.081*	0.431
Kaohsiung	Queen-Worker (within)	48	0.074	0.452
Kaohsiung	Queen-Worker (between)	192	-0.051	0.453
Chiayi	Worker-Worker (within)	24	-0.013	0.426
Chiayi	Worker-Worker (between)	96	-0.013	0.439
Chiayi	Queen-Worker (within)	24	0.221*	0.410
Chiayi	Queen-Worker (between)	72	-0.159 **	0.450

N = number of pairwise comparisons. Values shown are mean relatedness  $\pm$  standard deviation (SD). Values that significantly deviate from zero (based on Wilcoxon signed-rank tests) are indicated with asterisks: \*  $p < 0.05$ , \*\*  $p < 0.01$ .