

1 **Supplemental materials for**

2
3 **Variation in soldier investment is linked to the evolution of termite**
4 **soldier defense strategies**

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6
7 This file includes the

- 8 - Legend of Supplemental materials S1–S4 (Tables uploaded as separate CSV
- 9 files)
- 10 - Supplemental materials S5–S7

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13 **Supplemental material Table S1:** The list of genera used in this study, with the
14 information of foraging strategy.

15 **Supplemental material Table S2:** Species-level data used in the broad dataset, with
16 information on soldier proportion, defensive strategy, soldier morphology, nesting type,
17 and collection method (ec: entire colony, fg: foraging group, lc: laboratory colony, sc:
18 sample taken from a mound, pg: peripheral gallery, and ?: unknown).

19 **Supplemental material Table S3:** Species-level data used in the robust dataset, with
20 information on soldier proportion, defensive strategy, soldier morphology, nesting type.

21 **Supplemental material Table S4:** Genus-level dataset used for supplementary analyses
22 excluding all ambiguous trait classifications (see Supplemental material S5).

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Supplemental material S5

Analysis of additional dataset. Examination of the effects of ambiguous classification genera for classification

Here we analyze the correlation between defense strategy and soldier proportion in the case of excluding all genera with ambiguous classification of defense or foraging strategies. Even if we excluded all ambiguous genera, the result of analysis was significant and showed the same trend as main analysis (Table S1).

Table S1. The results of PGLM

	Estimate	Standard Error	z value	lower boot CI	upper boot CI	p value
main analysis	-0.2272	0.0637	-3.57	-0.3636	-0.1197	< .001
exclude all ambiguous genera	-0.1613	0.0757	-2.13	-0.3944	-0.0409	0.03

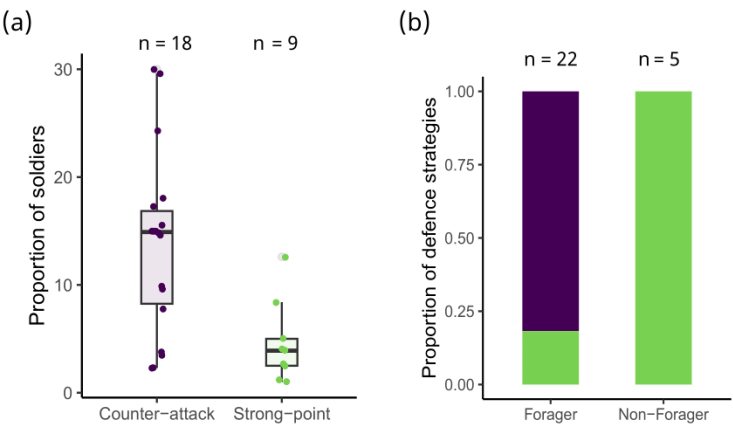


Figure S1. The relationship between termite defensive strategy and soldier proportion in the case of excluding all ambiguous genera. (a) Comparison of soldier proportion between the two defensive strategies. One dot represents one genus. (b) Proportion of defense strategies employed in foraging and non-foraging termites. Purple and green bars indicate defense strategy (green; strong-point, purple; counter-attack strategy).

Supplemental material S6

Analysis of two additional datasets. Examination of the effects of different coding of four genera belonging to Cubitermitinae.

Here we analyze the correlation between defense strategy and soldier proportion in the case of assuming all Cubitermitinae are strong-point strategists. The results of analysis were significant and showed the same trend as in the main analysis (Table S1).

Table S1. The results of PGLM

	Estimate	Standard Error	z value	lower boot CI	upper boot CI	p value
main analysis	-0.2272	0.0637	-3.57	-0.3636	-0.1197	< .001
all strong-point	-0.2714	0.0719	-3.78	-0.3745	-0.1488	< .001

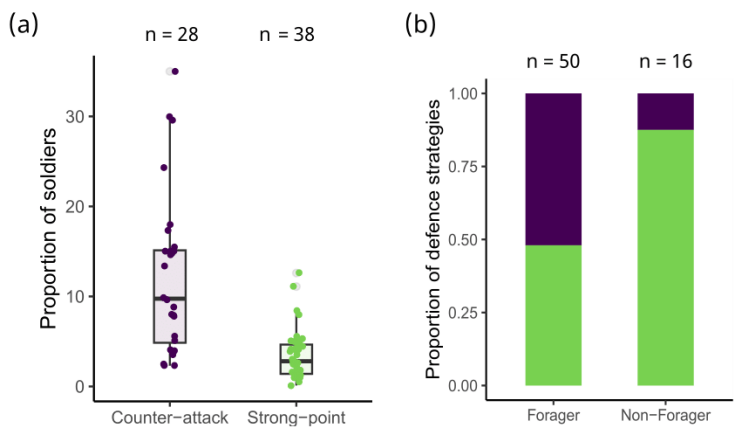


Figure S1. The relationship between termite defensive strategy and soldier proportion in the case of all Cubitermitinae were strong-point strategists. (a) Comparison of soldier proportion between the two defensive strategies. One dot represents one genus. (b) Proportion of defense strategies employed in foraging and non-foraging termites. Purple and green bars indicate defense strategy (green; strong-point, purple; counter-attack strategy).

Supplemental material S7

Examination of the robustness of Pagel test results

Here we analyze the evolutionary correlations between defense and foraging strategies using Pagel test. Pagel test (Pagel 1994) assesses the correlation between two discrete traits while correcting for phylogenetic non-independence among taxa. We also conducted Fisher's exact test to evaluate the relationship between defense and foraging strategies when lineage effects were not considered.

Table S1. The results of Pagel tests

	Pagel test		Fisher's exact test	
	likelihood ratio	p value	odds ratio	p value
main analysis	11.97	0.02	8.64	< 0.01
cubitermitinae all strong-point	8.35	0.08	Inf.	< 0.01
exclude all ambiguous genera	9.14	0.06	7.38	< 0.01

Fisher's exact test showed significant relationships in all supplemental analyses. Pagel test yield significant results only in the main analysis, although the supplementary analyses highlighted the same trend with marginal p-values (0.06 and 0.08).