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Citation for final published version:

Leahy, Lily, Chown, Steven L., Riskas, Hannah L., Wright, Ian J., Carlesso, Amelia G., Hammer, Ian J., Sanders, Nathan J., Bishop, Tom R., Parr, Catherine L. and Gibb, Heloise 2025. Metabolic traits are shaped by phylogenetic conservatism and environment, not just body size. *Proceedings of the National Academy of Sciences of the United States of America* 122 (29) , e2501541122. 10.1073/pnas.2501541122

Publishers page: <https://doi.org/10.1073/pnas.2501541122>

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3 Main Manuscript for

4 Metabolic traits are shaped by phylogenetic conservatism and 5 environment, not just body size

6

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22 **Author Contributions:** LL, HG, IW conceptualized and designed the study with input from SLC,
23 NJS, TRB, and CLP. LL analyzed the data and wrote the manuscript with contributions from HG,
24 SLC, IW, IJH, NJS, TRB, CLP. LL, HLR, and AGC collected field data and performed metabolic
25 assays. SLC and IJH provided equipment and technical support. Primary project funding was
26 acquired by HG and IW.

27 **Competing Interest Statement:** There are no conflicts of interest.

28 **Data availability:** Data and code is available at Dryad Data Repository
29 <https://doi.org/10.5061/dryad.d7wm37qb7>

30 **Classification:** Biological sciences, Ecology

31 **Keywords:** Metabolic rate, discontinuous gas exchange, macroecology, ants

32

35 **Abstract**

36 Metabolic rate dictates life's tempo, yet how ecological and environmental factors integrate to
37 shape metabolic traits remains contentious. Considering metabolic traits of 114 species of ants
38 from seven subfamily clades along a 1500-km climatic and soil phosphorus availability gradient in
39 Australia, we tested four hypotheses relating to variation in metabolic rate due to niche
40 conservatism, temperature, aridity, and ecological stoichiometry. We also tested the contested
41 hygric hypothesis, which predicts that insect ventilation patterns can be modified to reduce water
42 loss in arid environments. Mass-independent metabolic rate was phylogenetically conserved. The
43 ant clade *Myrmecia* had metabolic rates 3–10x higher than other species, likely related to their
44 large eye size, a correlate of cognitive complexity. Metabolic rate was higher in ants from warm,
45 arid sites relative to those from wet, cool sites. A weak positive interaction between soil
46 phosphorus and body mass indicated that, at sites with low soil phosphorus, smaller ants respired
47 at higher rates than expected based on their mass—consistent with ecological stoichiometry
48 theory. Larger ants, regardless of clade, were more likely to exhibit discontinuous gas exchange
49 (DGC) with increasing aridity, likely reflecting a water conservation strategy. Phylogenetic
50 conservatism of metabolic rate and a moderate influence of environment suggest that, in addition
51 to biophysical geometric constraints, metabolic rate has evolved to match the energetic demands
52 required of ecological strategies to address environmental stressors. For larger insect species
53 confronting their metabolic limits, discontinuous gas exchange may promote resilience in a world
54 that is becoming hotter and more arid.

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65 **Significance Statement**

66 Whether metabolic traits are shaped purely by biophysical constraints related to body size, or are
67 subject to evolutionary optimisation and environmental selection, remains a highly controversial
68 topic in metabolic theory. Using a macroscale empirical test on a diverse insect group, we find
69 phylogenetic conservatism in mass-independent metabolic rate, a weak but positive influence of
70 temperature and aridity, and steeper allometric scaling at sites with high soil phosphorus.
71 Metabolic rates have thus evolved to meet the energetic demands of ecological and
72 environmental stressors beyond their relationship with body size. Discontinuous gas exchange,
73 hypothesised to reduce desiccation, was positively correlated with aridity. Behavioural and
74 physiological modulation of ventilation patterns in dry environments will benefit insects in a
75 warming and drying world.

76

77 **Introduction**

78
79 From individual cells to entire organisms, metabolic rate indicates the tempo with which resources
80 are extracted and converted to energy and materials (1). Both metabolic rate and the manner by
81 which organisms exchange gases vary across multiple ecological and evolutionary scales (2, 3).
82 By driving how organisms interact with their environment, variation in metabolic traits shapes
83 macroecological patterns across the globe (1, 4). Understanding metabolic trait variation is
84 therefore of fundamental importance in biology. Yet despite over a century of intensive research
85 there is still substantial and ongoing debate on the causes of variation in these universal
86 measures of life's tempo (5, 6).

87
88 The development of metabolic theory has centered on debates surrounding the allometric scaling
89 of metabolic rate. Biophysical models (termed "Newtonian" approaches in Glazier et al. 2022)
90 emphasise that metabolic scaling with body mass is a product of mechanistic physical constraints
91 on the movement of gases or materials within organisms (7, 8). These models suggest that
92 selection on metabolic rate is not independent of selection on body size (9). Mass-independent
93 models (termed "Darwinian" approaches (5)) emphasise that variation in metabolic rate is driven
94 by evolutionary selection to meet the energy demands of different ecological and life history
95 strategies (10, 11). Consequently, metabolic rate and metabolic scaling relationships might be
96 expected to be shaped by external selective forces such as climate, net primary productivity, and
97 soil biogeochemistry (12, 13).

98
99 Previous studies have provided limited insight into the phylogenetic and macroecological drivers
100 of metabolic traits due to methodological limitations. Empirical studies have lacked power and
101 scale by focusing on only a few species or a single causal mechanism (e.g., (14-16)), while meta-
102 analyses at larger biological and biogeographic scales are limited by high methodological
103 variation among studies (e.g., (17, 18)). Here, we address these methodological shortcomings
104 using ants as a model taxon and leveraging data from 114 field collected species using a
105 consistent methodology of flow-through respirometry at a standard temperature. Ants are a
106 globally abundant and diverse social insect taxon that have been central to theoretical and
107 empirical advances in the field of metabolic evolutionary ecology (19-21).

108
109 Using an insect taxon to explore these central debates in metabolic theory enabled us to
110 investigate a second highly contentious issue relating to gas exchange patterns: the adaptive
111 origin of discontinuous gas exchange in insects (22, 23). Discontinuous gas exchange has
112 evolved independently five times amongst the insects, but there is substantial variation in its
113 occurrence both within and amongst insect taxa (3). The hygric hypothesis posits that

114 discontinuous gas exchange cycles (DGC) are an adaptation to reduce respiratory water loss,
115 which could have important implications in a drying and warming world (23, 24). Respiratory
116 water loss is highest under both hot and dry conditions, captured by the metric vapor pressure
117 deficit, often termed “the drying power of the air” (24).

118

119 A meta-analysis that covered multiple insect orders (40 species), found correlative support for the
120 hygric hypothesis, showing that species from warm and arid zones more frequently exhibit DGC
121 than do species from mesic regions (25). Experimental studies, however, have found mixed
122 support for the hygric hypothesis, both at the intraspecific (26, 27) and interspecific level (14, 23).
123 In general, interspecific experimental studies have been limited by small sample sizes (<10
124 species) (14, 28). Here, we extend the hygric hypothesis, predicting that the likelihood that
125 insects engage in DGC depends on the aridity at the location of the source population and body
126 size. We propose two scenarios: 1) smaller insects may desiccate faster, thereby triggering DGC,
127 2) larger insects respire higher total volumes of CO₂ per unit time, such that spiracles may be
128 open for longer periods, leading to greater respiratory water loss (29). In the latter case, larger
129 individuals may be more likely to engage in DGC, and/or have a higher ventilation frequency per
130 unit time during DGC, as demonstrated in *Scarabaeus* dung beetles (14).

131

132 We investigate interspecific variation in two metabolic traits: standard metabolic rate (SMR) and
133 ventilation pattern (discontinuous gas exchange) to test five non-mutually exclusive causal
134 hypotheses relating to phylogenetic conservatism and environmental factors underpinning
135 variation in metabolic rate and gas exchange pattern (Figure 1, Table 1). We used worker ants
136 from 114 species of 7 clades collected across a 1500 km latitudinal gradient in south-eastern
137 Australia, from sites ranging from semi-arid to subtropical climates and with naturally varying soil
138 phosphorus levels (Figure 1, Table S1). We measured CO₂ production of individual worker ants
139 as a proxy for metabolic rate and assessed the occurrence and frequency of discontinuous gas
140 exchange for each species.

141

142 **Results**

143

144 *Metabolic rate is conserved across major clades*

145 Metabolic rate varied 293-fold (2.38 - 698.87 μ W) across our 114 species and body mass varied
146 by almost 2000-fold (0.018 - 34.93 mg). Our data set was heavily skewed towards small-medium
147 ants (median = 0.5 mg; Figure S1a, b), typical of the global body size distribution of ants (34).

148 Metabolic rates have not been previously recorded for species smaller than 0.1 mg (35), yet
149 these species make up a large proportion of global ant diversity. Our experimental set-up

150 facilitated the first metabolic assays of smaller bodied ants (14% species < 0.1 mg) with high data
151 quality (Section S5: signal-to-noise ratio, Figure S4, S5).

152 In support of the niche conservatism hypothesis, we found strong phylogenetic signal in metabolic
153 rate ($\lambda = 0.70$, $p < 0.001$) and in mass-independent metabolic rate (residuals of the metabolic rate
154 – body mass relationship: $\lambda = 0.32$, $p = 0.02$). Body mass was also highly phylogenetically
155 conserved ($\lambda = 0.93$, $p < 0.001$). After accounting for variation due to body mass, metabolic rate
156 was higher for the large visual predators of the Myrmeciinae (bull-ants) than other taxa: 3.8 ± 1.4
157 (mean \pm SE) times higher than for Ectatomminae, 4.5 ± 2.3 for Dorylinae, and 8.9 ± 5.98 times
158 higher than for Ponerinae (LME: Table 2, Table S2: $p = 0.002$, Figure 2a).

159 Metabolic rate scaled with body mass as $MR = 1.5M^{0.49}$ in a linear regression model including
160 only body mass (Table S3). Excluding the smallest 25th percentile of ants (<0.18 mg) raised the
161 metabolic scaling exponent (b) from 0.49 (95% CI 0.40 – 0.57) to 0.68 (95% CI 0.57 – 0.79)
162 (Figure S2, Table S3). The elevated metabolic rate of the small ant subset was not due to
163 methodological constraints (Figure S4, S5) or high activity during the metabolic assays (Table S4)
164 but was driven by small-bodied species from multiple clades which had higher mass-independent
165 metabolic rates (residuals of MR~body mass relationship: Figure 2b).

166 *Aridity, temperature, and soil phosphorus influence metabolic rate*

167 To address each metabolic rate hypothesis presented in Figure 1c-e (Table 1), we constructed a
168 global model with fixed effects representing body mass, microclimate, and soil phosphorus
169 relationships and accounting for phylogeny (Section S9, Table 2). In the global model, the P-
170 limitation hypothesis was weakly supported by an interaction between body mass and soil
171 phosphorus (PGLS model: 0.12 (95% CI: -0.003 – 0.24), $p = 0.0576$, Table S6). In line with
172 predictions, smaller ants had elevated metabolic rates in lower phosphorus soil sites (Figure 3c).
173 This model was more parsimonious than a model including only body mass or a null model (Table
174 2). The global model showed high variance inflation factors (>5) for temperature and vapor
175 pressure deficit due to the correlation of these variables (Table S5). We therefore constructed two
176 additional models, each excluding one of the correlated variables, either vapor pressure deficit or
177 temperature, but retaining other predictors (see Table S6). These models indicated that
178 microclimate affected metabolic rates, but in the opposite direction from the predictions of both
179 the metabolic cold adaptation and metabolic rate aridity hypotheses. Species from warm (0.07
180 95% CI: 0.007 – 0.13, $p = 0.03$) and/or dry (0.6, 95% CI: 0.02 – 1.17, $p = 0.04$) sites (higher
181 temperature and vapor pressure deficit) had higher metabolic rates than species from sites that
182 were cool and/or humid (Table S6; Figures 3a-b). There was no interaction between body mass
183 and aridity (vapor pressure deficit) (Table S6).

184 To investigate the relative effect of temperature and vapor pressure deficit on metabolic rate,
185 given model uncertainty, we calculated the importance of each variable as the model-averaged
186 standardised effect size across a candidate set of models produced from the global model (36)
187 (Section S9). Aridity (vapor pressure deficit) had a much higher average predictor importance,
188 indicating a larger overall effect size than temperature across the candidate set of models (Table
189 S7).

190 *Discontinuous gas exchange increases with aridity, particularly for larger ants*

191 Around half (55%) of all species exhibited discontinuous gas exchange cycles (DGC) during
192 metabolic assays. We found no evidence for phylogenetic signal in DGC ($\lambda = 0$, $p = 1$), hence
193 non-phylogenetically informed GLMM models are reported (Table S8). We found general support
194 for the hygric hypothesis and our second prediction that larger ants were more likely to engage in
195 DGC with increasing VPD (Table S8: $p = 0.05$). In more arid sites, more individuals from larger
196 species engaged in a DGC ventilation pattern compared with smaller species, whereas in more
197 humid sites the proportion of individuals engaging in DGC per species was lower overall and did
198 not vary with body mass (Figure 4a, Table S8: $p = 0.015$, Table S9). For those species exhibiting
199 DGC, larger ants had lower ventilation frequency per hour (VF), indicating they take fewer but
200 larger breaths per unit time compared with smaller ants (Figure 4b, Table S8: $p = 0.0001$).

201 **Discussion**

202 Metabolic traits drive energy flow through ecosystems and underpin global ecological patterns,
203 yet much is still unknown about how metabolic traits vary among species and environments (1,
204 37). A lack of large-scale empirical tests has hindered efforts to disentangle the multiple
205 controversies in the field (5, 6). We provide new insights by testing several hypothesised
206 mechanisms of metabolic trait variation using a diverse insect group and a consistent
207 experimental method at a macroecological scale.

208 We found support for the niche conservatism hypothesis, which posits that metabolic rate evolves
209 to support conserved templates of ecological and life history strategy. Given the universal
210 relationship between metabolic rate and mass, it is impossible to disentangle conservatism of
211 these two traits by testing the phylogenetic signal of either trait independently (see Cameron and
212 Marshall (38) for commentary). We tested for phylogenetic signal in mass-independent metabolic
213 rate and found evidence that this trait is phylogenetically conserved. This supports our hypothesis
214 that, although primarily driven by mass, variation metabolic rate has likely evolved to meet the
215 energetic demands of diet, activity, sociality, and life history strategies.

216 As further evidence in support of niche conservatism, we found strong differences in metabolic
217 rate amongst subfamily clades. Subfamily clades generally have shared ecological traits relating
218 to diet and foraging strategies, although interspecific variation in ecological traits can be high in
219 some lineages (39, 40). The bull ants, Myrmeciinae, had higher mass independent metabolic
220 rates than most other clades. These largely predatory ants have complex sensory functionality
221 with large compound eyes and ocelli (usually a feature of flying insects (41) for solitary navigation
222 over long distances). The visual system is one of the most metabolically costly systems in the
223 brain. For example, in cavefish, the cost of vision was estimated to be as much as 15% of resting
224 metabolic rate (1g mass) (42). Expensive ecological traits such as complex vision also contribute
225 to provisioning of that energy, for example by improving foraging efficiency or predation rates (43,
226 44).

227 The universality of the metabolic scaling coefficient ($b = 0.75$) has been controversial (5). We
228 found that body mass size class significantly influenced the scaling coefficient, presenting a
229 challenge to the premise of the $\frac{3}{4}$ scaling coefficient as a natural “law” (2). With the removal of
230 the smallest ant species (which have not been previously tested), the scaling exponent increased
231 from rather flat ($b = 0.5$) to more in line with what has been previously reported for ants (~ 0.68)
232 (19, 35). For social insects, an evolutionary pathway to miniaturisation has been concomitant with
233 changing energy demands due to increased colony size and high energetic costs of fast tempo
234 foraging rates (45, 46). The metabolic-level boundaries hypothesis suggests that different
235 biophysical processes (mass-surface area versus mass-volume relationships) are operating
236 depending on baseline energy demands (metabolic level). This extension of dynamic energy
237 budget theory (7) predicts that metabolic scaling should be shallower (further from 1) for
238 organisms with higher metabolic level/energy demand as has been demonstrated in birds,
239 mammals, and reptiles (47). This could explain changes in allometric scaling due to the high
240 mass independent metabolic rates of several miniature species (<0.2 mg) tested here. Again, this
241 presents a case that both biophysical and evolutionary processes are shaping metabolic scaling
242 rather than a single universal mechanism.

243 Metabolic rates were biogeographically structured across climatic and environmental gradients,
244 indicating that phenotypic variation is influenced by external factors beyond body mass and
245 phylogeny (13). We found, however, that the relationship between climate and metabolic rate was
246 the opposite from that predicted. Ants from dry and/or warm sites had higher metabolic rates than
247 those from wet and/or cool sites, counter to both the metabolic cold adaptation (MCA) hypothesis
248 and the aridity-metabolic rate hypothesis. The MCA hypothesis considers enhanced metabolic
249 rates as a life-history adaptation to a short-growing season (48). Most tests of this hypothesis are

250 from latitudes above 50° N or S (16, 18, 49). However, at 39°S, even our southern-most sites
251 may not have been cool enough to support a counter-gradient response in metabolic rate.

252 We also predicted that because aridity accelerates desiccation-risk associated with respiration, a
253 depressed metabolic rate would be favoured in arid environments. However, metabolic rate
254 increased in drier sites. Similarly elevated metabolic rates have been found for Fijian bees in arid
255 environments (50). The aridity hypothesis has largely been tested in mammals and birds, but the
256 mechanisms underlying metabolic adaptation may vary depending on the heat and water loss
257 dynamics of ectothermic and endothermic taxa (7). Insects may have depressed metabolic rate in
258 wetter and in cooler environments due to limited daily windows of warm foraging conditions, while
259 they may have various adaptive mechanisms to cope with arid conditions (e.g., results here on
260 DGC and cuticular desiccation resistance (24)).

261 Our proposed P-limitation hypothesis was weakly supported. While we did not test the
262 mechanisms underlying the P-limitation hypothesis, namely, that increased metabolic rate is a
263 physiological strategy to balance C:P ratios when organisms are under P-limited diets (12), a
264 correlation between metabolic rate and soil phosphorus supported this hypothesis. Further, the
265 effect was largest for the smallest species at the lowest P sites. Smaller invertebrates are
266 expected to be affected more by P-limitation as the %P in RNA is inversely related to body size
267 (33). Given the increasing global deposition of phosphorus associated with agriculture and
268 industry, understanding the links between metabolic rate and ecological stoichiometry is a
269 research priority (51). This is the first macroecological correlative test of this hypothesis, but
270 further work is needed to more precisely understand the physiological mechanisms operating at
271 this scale.

272 Lastly, we found a positive relationship between aridity and discontinuous gas exchange in line
273 with the hygric hypothesis. DGC is a heritable trait that extends survival under food and water
274 stress, suggesting there is selection on this trait in response to environmental pressures (52).
275 Here, DGC was not linked with phylogenetic history but varied systematically with the aridity of
276 field collection sites. In a meta-analysis, DGC was found to significantly reduced water loss in the
277 insect clades Coleoptera, Blattodea, and Orthoptera (53), but some single species and
278 experimental studies have not linked DGC with reduced water loss (26, 54). We found that not all
279 individuals within a species exhibited DGC under experimental conditions, suggesting a degree of
280 phenotypic plasticity (55). Larger ant species from arid sites engaged in more DGC and took less
281 regular but larger breaths per unit time. Smaller organisms face desiccation stress due to greater
282 surface area to volume ratios, but may use other mechanisms to reduce water loss such as
283 cuticular resistance (24). Meanwhile, larger ants have greater total CO₂ emission volumes,

284 despite having lower mass-specific metabolic rates, and may therefore have a greater propensity
285 for switching to DGC to reduce respiratory water loss (56).

286 Strong evolutionary conservatism and systematic phenotypic variation in mass independent
287 metabolic rate are not consistent with an exclusively biophysical mechanism. Rather, we suggest
288 that metabolic rate is under selection to track the energy demands of ecological strategy, in this
289 case for traits related to resource acquisition and assimilation. Correlative relationships between
290 environmental factors and metabolic rate further highlight the importance of recent evolution in
291 response to external drivers of metabolic rate variation. These findings have important
292 implications in our rapidly changing world. Insects with high resting metabolic rates could be
293 negatively affected by metabolic shifts in response to global warming, with subsequent flow on
294 effects for ecosystem services (57). However, our findings also suggest that insects could
295 modulate their respiration (using DGC) to adapt to increasing aridity, potentially increasing their
296 ability to persist in changing environments.

297

298 **Materials and Methods**

299 *Study sites and ant sampling*

300 Ants were collected from six different locations spanning the east coast and inland regions of
301 south-eastern Australia (Figure 1a), covering a gradient in precipitation (421–1283 mm Mean
302 Annual Precipitation) and temperature (13–20 °C Mean Annual Temperature)(58). Each location
303 was sampled over the course of a week between June 2022 and April 2023 (Section S1 for
304 details)(58). At each location, two sites differing in soil phosphorus levels were selected, and
305 within each site, four 10 x 10 m plots were established approximately 200 m apart (yielding eight
306 plots per site). Soil cores were taken to verify soil phosphorus status (see Section S1). Over three
307 days of sampling per site, we aimed to collect as many live ant species as possible (Section S1).
308 For metabolic assays, we tested a minimum of 10 individuals per colony from 1–3 colonies per
309 species at each site, targeting a total of 30 individuals per species.

310 *Metabolic assays*

311 Ants were maintained at 20 °C in plastic nesting containers and given water and honey soaked in
312 cotton balls every two days. The time between field collection and the start of metabolic testing
313 ranged from 3 to 14 days across sites (58). To minimize the influence of digestion on metabolic
314 rate, ants were fasted but supplied with water for 48 hours prior to testing. Carbon dioxide
315 production (VCO₂) was used to assess metabolic rate and was measured at a consistent 22 °C
316 using eight Sable Systems International (SSI) multiple animal versatile energetic (MAVEN)

317 systems, each connected to a Li-Cor 7000 CO₂/H₂O infrared gas analyzer (Li-Cor, Lincoln,
318 Nebraska, USA)(58). Ants were placed in individual 2 ml or 3 ml cassettes (depending on the size
319 of the individual) and placed in the MAVEn system that was housed in an incubator in the dark to
320 reduce activity. Ants were not observed during assays to reduce stress but observations before
321 and after indicated walking, grooming, and resting behaviour inside cassettes. In some cases, ants
322 were observed running or seemed stressed prior to assays (see below for data treatment). To
323 examine activity during assays, automated activity readings of each individual ant were measured
324 simultaneously using infrared light detectors.

325

326 Following experiments, ants were instantly placed in a freezer at -20 °C and wet mass was
327 measured the following day. Ants were then dried at 50 °C for 48 hours and then weighed for dry
328 mass (wet and dry mass included in data repository). Data from assays was extracted using the
329 software Expedata (SSI) (Section S3) and metabolic rate was converted from minimum VCO₂
330 (μL/hr⁻¹) to microwatts following equations in Chown, *et al.* (19) (Section S4, data provided in both
331 units in data repository). Data were inspected and cleaned for technical errors and outliers
332 (Section S5). Cases of very high activity, deaths, and systematically identified outliers, given the
333 colonies average metabolic rate, were presumed to indicate stressed individuals and were
334 removed from the dataset (Section S5). Cleaned data generated a final dataset of 2805
335 individuals of 214 colonies. Signal-to-noise ratios were assessed and were deemed sufficient to
336 detect a VCO₂ signal against baseline noise (Section S5). We found no relationship between
337 activity during assays and metabolic rate (Section S6). Activity was therefore not included in
338 downstream analyses.

339 *Ventilation patterns*

340 Insects are known to exhibit three forms of gas exchange: discontinuous gas exchange cycle
341 (DGC), cyclic gas exchange and continuous gas exchange (see Figure S6 and Section S7 for
342 definitions). We calculated three metrics to indicate DGC occurrence and frequency. We
343 produced a binary (0,1) categorical value for whether DGC was being exhibited by a species. We
344 then calculated the proportion of individuals per species for each site and plot that were
345 conducting DGC, which ranged from 0 (no individuals exhibiting DGC) to 1 (all individuals
346 exhibiting DGC). We then calculated ventilation frequency per hour (VF), for those individuals
347 exhibiting a DGC pattern. Ventilation frequency was determined by first counting the number of
348 complete closed and open phases in an individual's VCO₂ trace, then multiplying this number by
349 six (i.e., six 10-minute periods in an hour) to give vent frequency in cycles per hour.

350 *Microclimate variables*

351 To test our climate hypotheses (Figure 1 c-f), we modelled microclimate at each site to represent
352 the thermal and hydric environment that ant species directly experience in the field (58). We
353 estimated hourly temperature and relative humidity for the 15 years preceding the study using
354 NicheMapR (59). From this, we calculated mean annual microclimate temperature and vapor
355 pressure deficit (VPD), with VPD representing the aridity gradient in downstream analyses
356 (Section S8). We also calculated warmest quarter (summer months) microclimate temperature
357 and VPD. Warmest-quarter variables were highly correlated with mean annual variables
358 (correlation = 0.75 and 0.94 for temperature and VPD respectively). Further, in the southern
359 hemisphere temperate climates, ants are relatively active all year round with total ant activity
360 showing a gentle seasonal cycle (60). Therefore, mean annual microclimate values were chosen
361 to be more representative of general climate conditions and are used in downstream analyses.

362 *Analysis*

363 For all analyses, we treated species at each site as unique 'site-species' as a conservative
364 approach to recognise that taxonomic identity of many species is not resolved in Australia
365 resulting in 139 site-species from 114 taxonomic species. Metabolic traits and body mass were
366 averaged at the site-species level. All statistics were performed in R version 4.1.1 (61). Details of
367 models and coding packages Section S9. Briefly, we tested for phylogenetic signal in metabolic
368 traits using Pagel's λ . We then used linear mixed effects models (LME) with location ($n = 6$)
369 included as a random effect to investigate present day phenotypic variation in metabolic rate
370 among subfamilies and used generalized linear mixed effects models (GLMM) with the same
371 random effects structure to investigate DGC metrics.

372 We used phylogenetic generalised least squares models (PGLS) to partition out the influence of
373 evolutionary history among species for testing environmental hypotheses (Section S9). In a global
374 model, we tested metabolic rate as a function of temperature (MCA hypothesis), mass and VPD
375 interaction (aridity hypothesis), and mass and soil phosphorus interaction (P-limitation
376 hypothesis). This model had high variance inflation factor values ($VIF > 5$) for temperature and
377 VPD due to correlations between these variables (Table S4). We therefore also constructed two
378 additional models with predictor variables as follows: 1) temperature and mass:soil phosphorus
379 interaction, and 2) mass:VPD interaction and mass:soil phosphorus interaction. These three
380 models performed comparatively (within 2 AICc). Effect sizes and p-values for all three models
381 are reported. To compare the relative effect of temperature and vapor pressure deficit (given their
382 correlation and subsequent model uncertainty) we then calculated predictor importance for each
383 variable over a set of models produced from the global model, following the approach applied in
384 Gonçalves-Souza, *et al.* (62) (see Section S9 for details).

385 For PGLS models we used a species-level, time-calibrated phylogeny from Economo, *et al.* (63)
386 and a Brownian motion model of evolution (Section S10). We also tested whether body mass
387 systematically varied across sites and locations, which could influence our results and did not find
388 this to be the case (ANOVA: body mass ~ site, F-value = 1.02, $p = 0.42$, $R^2 = 0.002$).

389 Acknowledgments

390 We thank and acknowledge the Wurundjeri people of the Kulin Nation, the Wotjobaluk, the
391 Ngiyampaa, the Durrumurragal, the Gubbi Gubbi, the Boonwurung, Bunurong, and Gunaikurnai
392 people on whose lands this field work was conducted. We thank Emily House for access to Glen
393 Echo. Field work was carried out under Permit SL102675 NSW Department of Planning, Industry
394 and Environment, Permit AA-0000328 Parks Victoria, and under animal ethics committee permit
395 AEC22001. We thank Alan Andersen for ant identification. This work was supported by Australian
396 Research Council funding DP210101630 to HG and IJW and DP190100341 to SLC.

397 References

- 398
- 399 1. J. H. Brown, J. F. Gillooly, A. P. Allen, V. M. Savage, G. B. West, Toward a metabolic
400 theory of ecology. *Ecology* **85**, 1771-1789 (2004).
 - 401 2. D. S. Glazier, Beyond the '3/4-power law': variation in the intra- and interspecific scaling
402 of metabolic rate in animals. *Biological Reviews* **80**, 611-662 (2005).
 - 403 3. E. Marais, C. J. Klok, J. S. Terblanche, S. L. Chown, Insect gas exchange patterns: a
404 phylogenetic perspective. *Journal of Experimental Biology* **208**, 4495-4507 (2005).
 - 405 4. S. J. Brandl, J. S. Lefcheck, A. E. Bates, D. B. Rasher, T. Norin, Can metabolic traits
406 explain animal community assembly and functioning? *Biological Reviews* **98**, 1-18
407 (2022).
 - 408 5. D. S. Glazier, Variable metabolic scaling breaks the law: from 'Newtonian' to
409 'Darwinian' approaches. *Proceedings of the Royal Society B* **289**, 20221605 (2022).
 - 410 6. J. F. Harrison *et al.*, White paper: an integrated perspective on the causes of hypometric
411 metabolic scaling in animals. *Integrative and Comparative Biology* **62**, 1395-1418 (2022).
 - 412 7. S. A. L. M. Kooijman, *Dynamic energy budget theory for metabolic organisation*
413 (Cambridge university press, 2010).
 - 414 8. G. B. West, J. H. Brown, B. J. Enquist, A General Model for the Origin of Allometric
415 Scaling Laws in Biology. *Science* **276**, 122-126 (1997).
 - 416 9. J. L. Maino, M. R. Kearney, R. M. Nisbet, S. A. Kooijman, Reconciling theories for
417 metabolic scaling. *Journal of Animal Ecology* **83**, 20-29 (2014).
 - 418 10. A. K. Pettersen, C. R. White, D. J. Marshall, Metabolic rate covaries with fitness and the
419 pace of the life history in the field. *Proceedings of the Royal Society B: Biological*
420 *Sciences* **283**, 20160323 (2016).
 - 421 11. C. R. White, D. J. Marshall, Optimisation and constraint: explaining metabolic patterns in
422 biology. *Journal of Experimental Biology* **226**, jeb245426 (2023).
 - 423 12. T. Ruiz *et al.*, A microcalorimetric approach for investigating stoichiometric constraints on
424 the standard metabolic rate of a small invertebrate. *Ecology letters* **21**, 1714-1722 (2018).
 - 425 13. S. L. Chown, K. J. Gaston, Exploring links between physiology and ecology at macro-
426 scales: the role of respiratory metabolism in insects. *Biological Reviews* **74**, 87-120
427 (1999).
 - 428 14. A. L. Davis, S. L. Chown, C. H. Scholtz, Discontinuous gas-exchange cycles in
429 *Scarabaeus dung* beetles (Coleoptera: Scarabaeidae): mass-scaling and temperature
430 dependence. *Physiological and Biochemical Zoology* **72**, 555-565 (1999).

- 431 15. P. D. Jeyasingh, Plasticity in metabolic allometry: the role of dietary stoichiometry.
432 *Ecology letters* **10**, 282-289 (2007).
- 433 16. Q. Willot, M. Ørsted, H. Malte, J. Overgaard, Cold comfort: metabolic rate and tolerance
434 to low temperatures predict latitudinal distribution in ants. *Proceedings of the Royal*
435 *Society B* **290**, 20230985 (2023).
- 436 17. C. R. White, P. Cassey, T. M. Blackburn, Allometric exponents do not support a universal
437 metabolic allometry. *Ecology* **88**, 315-323 (2007).
- 438 18. A. Addo-Bediako, S. L. Chown, K. J. Gaston, Metabolic cold adaptation in insects: a
439 large-scale perspective. *Functional Ecology* **16**, 332-338 (2002).
- 440 19. S. L. Chown *et al.*, Scaling of insect metabolic rate is inconsistent with the nutrient supply
441 network model. *Functional Ecology* **21**, 282-290 (2007).
- 442 20. I. J. Aitkenhead *et al.*, Tracheal branching in ants is area-decreasing, violating a central
443 assumption of network transport models. *PLoS computational biology* **16**, e1007853
444 (2020).
- 445 21. J. R. Lighton, Slow discontinuous ventilation in the Namib dune-sea ant *Camponotus*
446 *detritus* (Hymenoptera, Formicidae). *Journal of Experimental Biology* **151**, 71-82 (1990).
- 447 22. J. R. Lighton, Respiratory biology: why insects evolved discontinuous gas exchange.
448 *Current Biology* **17**, R645-R647 (2007).
- 449 23. P. G. Matthews, The mechanisms underlying the production of discontinuous gas
450 exchange cycles in insects. *Journal of Comparative Physiology B* **188**, 195-210 (2018).
- 451 24. A. Addo-Bediako, S. L. Chown, K. J. Gaston, Revisiting water loss in insects: a large
452 scale view. *Journal of Insect Physiology* **47**, 1377-1388 (2001).
- 453 25. C. R. White *et al.*, Evolutionary responses of discontinuous gas exchange in insects.
454 *Proceedings of the National Academy of Sciences* **104**, 8357-8361 (2007).
- 455 26. M. Bosch, S. L. Chown, C. H. Scholtz, Discontinuous gas exchange and water loss in the
456 keratin beetle *Omorgus radula*: further evidence against the water conservation
457 hypothesis? *Physiological Entomology* **25**, 309-314 (2000).
- 458 27. F. D. Duncan, B. Krasnov, M. McMaster, Novel case of a tenebrionid beetle using
459 discontinuous gas exchange cycle when dehydrated. *Physiological entomology* **27**, 79-83
460 (2002).
- 461 28. S. L. Chown, A. L. Davis, Discontinuous gas exchange and the significance of respiratory
462 water loss in scarabaeine beetles. *Journal of Experimental Biology* **206**, 3547-3556
463 (2003).
- 464 29. J. J. Wiens, C. H. Graham, Niche conservatism: integrating evolution, ecology, and
465 conservation biology. *Annu. Rev. Ecol. Evol. Syst.* **36**, 519-539 (2005).
- 466 30. B. K. McNab, The Influence of Food Habits on the Energetics of Eutherian Mammals.
467 *Ecological Monographs* **56**, 1-19 (1986).
- 468 31. C. R. White, The influence of foraging mode and arid adaptation on the basal metabolic
469 rates of burrowing mammals. *Physiological and Biochemical Zoology* **76**, 122-134 (2003).
- 470 32. B. G. Lovegrove, The zoogeography of mammalian basal metabolic rate. *The American*
471 *Naturalist* **156**, 201-219 (2000).
- 472 33. J. J. Elser *et al.*, Growth rate–stoichiometry couplings in diverse biota. *Ecology Letters* **6**,
473 936-943 (2003).
- 474 34. M. Kaspari, Global energy gradients and size in colonial organisms: worker mass and
475 worker number in ant colonies. *Proceedings of the National Academy of Sciences* **102**,
476 5079-5083 (2005).
- 477 35. J. Z. Shik, C. Hou, A. Kay, M. Kaspari, J. F. Gillooly, Towards a general life-history model
478 of the superorganism: predicting the survival, growth and reproduction of ant societies.
479 *Biology Letters* **8**, 1059-1062 (2012).
- 480 36. B. S. Cade, Model averaging and muddled multimodel inferences. *Ecology* **96**, 2370-
481 2382 (2015).
- 482 37. P. A. Marquet *et al.*, Scaling and power-laws in ecological systems. *Journal of*
483 *Experimental Biology* **208**, 1749-1769 (2005).

- 484 38. H. Cameron, D. Marshall, Estimating the relationship between fitness and metabolic rate:
 485 which rate should we use? *Philosophical Transactions of the Royal Society B* **379**,
 486 20220491 (2024).
- 487 39. M. D. Weiser, M. Kaspari, Ecological morphospace of New World ants. *Ecological*
 488 *Entomology* **31**, 131-142 (2006).
- 489 40. C. E. Sosiak, P. Barden, Multidimensional trait morphology predicts ecology across ant
 490 lineages. *Functional Ecology* **35**, 139-152 (2021).
- 491 41. A. Narendra, W. A. Ribi, Ocellar structure is driven by the mode of locomotion and activity
 492 time in *Myrmecia* ants. *Journal of Experimental Biology* **220**, 4383-4390 (2017).
- 493 42. D. Moran, R. Softley, E. J. Warrant, The energetic cost of vision and the evolution of
 494 eyeless Mexican cavefish. *Science advances* **1**, e1500363 (2015).
- 495 43. S.-J. Fu *et al.*, The behavioural, digestive and metabolic characteristics of fishes with
 496 different foraging strategies. *Journal of Experimental Biology* **212**, 2296-2302 (2009).
- 497 44. S. Bury, Energy expenses on prey processing are comparable, but paid at a higher
 498 metabolic scope and for a longer time in ambush vs active predators: a multispecies
 499 study on snakes. *Oecologia* **197**, 61-70 (2021).
- 500 45. K. S. Mason, C. L. Kwapich, W. R. Tschinkel, Respiration, worker body size, tempo and
 501 activity in whole colonies of ants. *Physiological Entomology* **40**, 149-165 (2015).
- 502 46. A. T. Burchill, C. S. Moreau, Colony size evolution in ants: macroevolutionary trends.
 503 *Insectes Sociaux* **63**, 291-298 (2016).
- 504 47. K. A. Nagy, I. A. Girard, T. K. Brown, Energetics of free-ranging mammals, reptiles, and
 505 birds. *Annual review of nutrition* **19**, 247-277 (1999).
- 506 48. A. Clarke, Seasonal acclimatization and latitudinal compensation in metabolism: do they
 507 exist? *Functional Ecology* **7**, 139-149 (1993).
- 508 49. C. R. White, L. A. Alton, P. B. Frappell, Metabolic cold adaptation in fishes occurs at the
 509 level of whole animal, mitochondria and enzyme. *Proceedings of the Royal Society B:*
 510 *Biological Sciences* **279**, 1740-1747 (2012).
- 511 50. C. R. da Silva *et al.*, Physiological traits and their relationships vary along an aridity
 512 gradient within and among Fijian bee species. *bioRxiv* 10.1101/2022.07.27.501487,
 513 2022.2007.2027.501487 (2022).
- 514 51. F. Jiao, X.-R. Shi, F.-P. Han, Z.-Y. Yuan, Increasing aridity, temperature and soil pH
 515 induce soil CNP imbalance in grasslands. *Scientific reports* **6**, 19601 (2016).
- 516 52. N. G. Schimpf, P. G. D. Matthews, C. R. White, Cockroaches that exchange respiratory
 517 gases discontinuously survive food and water restriction. *Evolution* **66**, 597-604 (2012).
- 518 53. S. O. Oladipupo, A. E. Wilson, X. P. Hu, A. G. Appel, Why do insects close their
 519 spiracles? A meta-analytic evaluation of the adaptive hypothesis of discontinuous gas
 520 exchange in insects. *Insects* **13**, 117 (2022).
- 521 54. J. R. Lighton, R. J. Turner, The hygric hypothesis does not hold water: abolition of
 522 discontinuous gas exchange cycles does not affect water loss in the ant *Camponotus*
 523 *vicinus*. *Journal of Experimental Biology* **211**, 563-567 (2008).
- 524 55. J. S. Terblanche, S. Clusella-Trullas, S. L. Chown, Phenotypic plasticity of gas exchange
 525 pattern and water loss in *Scarabaeus spretus* (Coleoptera: Scarabaeidae):
 526 deconstructing the basis for metabolic rate variation. *Journal of Experimental Biology*
 527 **213**, 2940-2949 (2010).
- 528 56. H. A. Woods, J. N. Smith, Universal model for water costs of gas exchange by animals
 529 and plants. *Proceedings of the National Academy of Sciences* **107**, 8469-8474 (2010).
- 530 57. L. Elizalde *et al.*, The ecosystem services provided by social insects: traits, management
 531 tools and knowledge gaps. *Biological Reviews* **95**, 1418-1441 (2020).
- 532 58. L. Leahy *et al.*, The economic strategies of superorganisms. *bioRxiv*
 533 10.1101/2025.02.21.639603, 2025.2002.2021.639603 (2025).
- 534 59. M. R. Kearney, P. K. Gillingham, I. Bramer, J. P. Duffy, I. M. D. Maclean, A method for
 535 computing hourly, historical, terrain-corrected microclimate anywhere on earth. *Methods*
 536 *in Ecology and Evolution* **11**, 38-43 (2020).

- 537 60. A. N. Andersen, Diversity, seasonality and community organization of ants at adjacent
 538 heath and woodland sites in southeastern Australia. *Australian Journal of Zoology* **34**, 53-
 539 64 (1986).
- 540 61. R Core Team (2023) R: A Language and Environment for Statistical Computing. (R
 541 Foundation for Statistical Computing, Vienna, Austria).
- 542 62. T. Gonçalves-Souza *et al.*, Species turnover does not rescue biodiversity in fragmented
 543 landscapes. *Nature*, 1-5 (2025).
- 544 63. E. P. Economo, N. Narula, N. R. Friedman, M. D. Weiser, B. Guénard, Macroecology and
 545 macroevolution of the latitudinal diversity gradient in ants. *Nature Communications* **9**, 1-8
 546 (2018).

547

548 **Figure Legends**

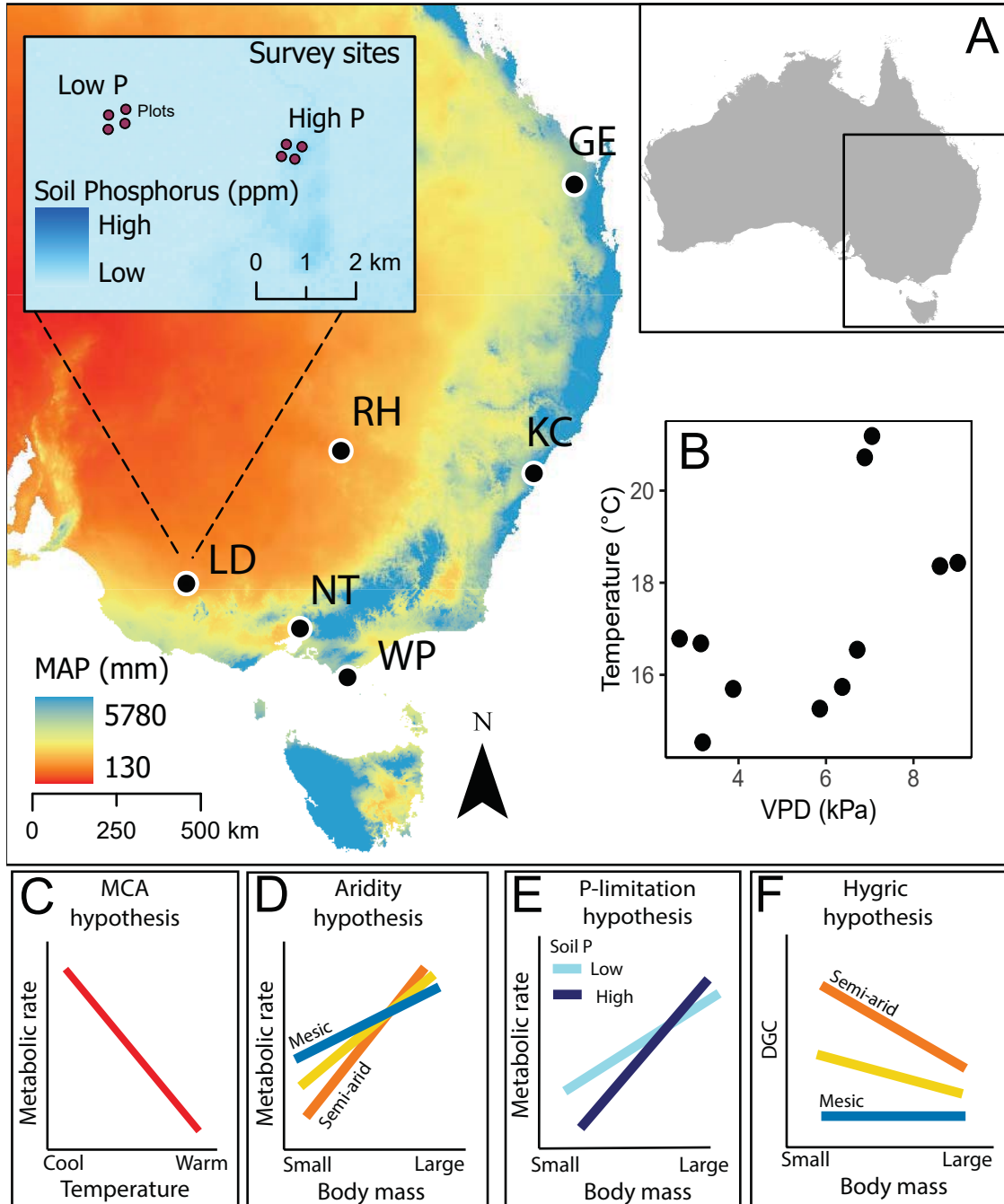
549 **Figure 1.** Study design and hypotheses. A) Map of six locations representing precipitation and
 550 temperature gradients in south-eastern Australia; inset indicates sites (low and high phosphorus)
 551 and plot design (total sites = 11), full location names Table S1. B) Mean annual microclimate
 552 temperature and vapor pressure deficit (VPD), a measure of aridity, at each site. C-E) Conceptual
 553 figures of the climate and environment hypotheses, MCA = metabolic cold adaptation. F) The
 554 hygric hypothesis: DGC = discontinuous gas exchange cycle, see Table 1 for predictions relating
 555 to body mass-aridity interaction extension of the hygric hypothesis, only prediction (a) is shown.

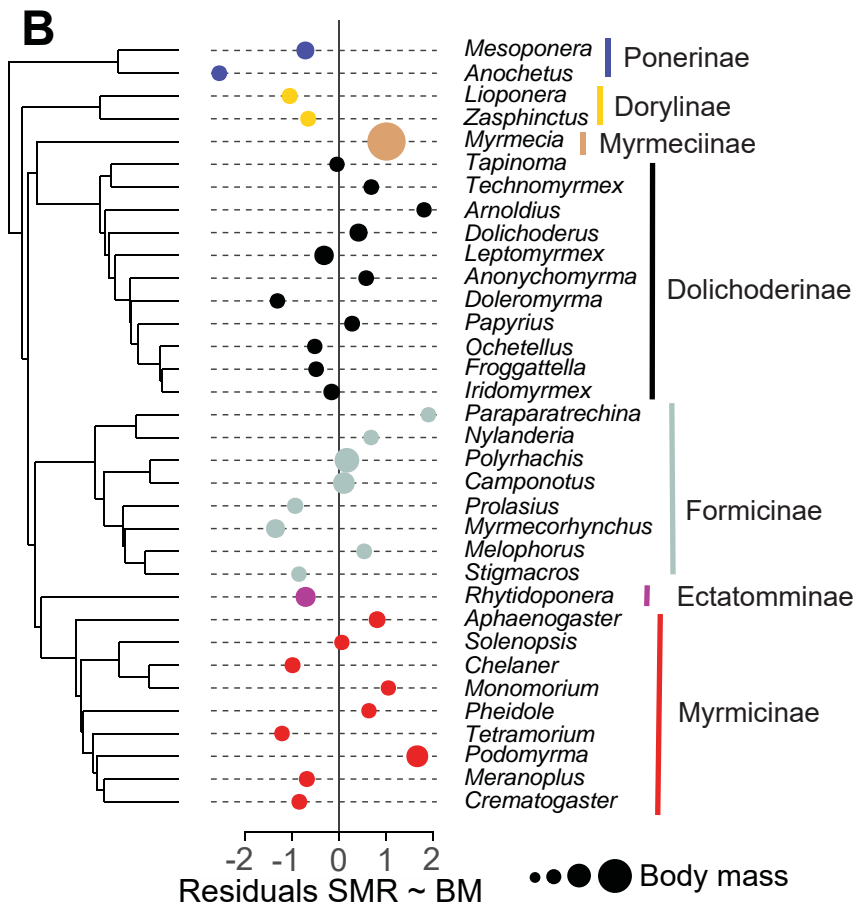
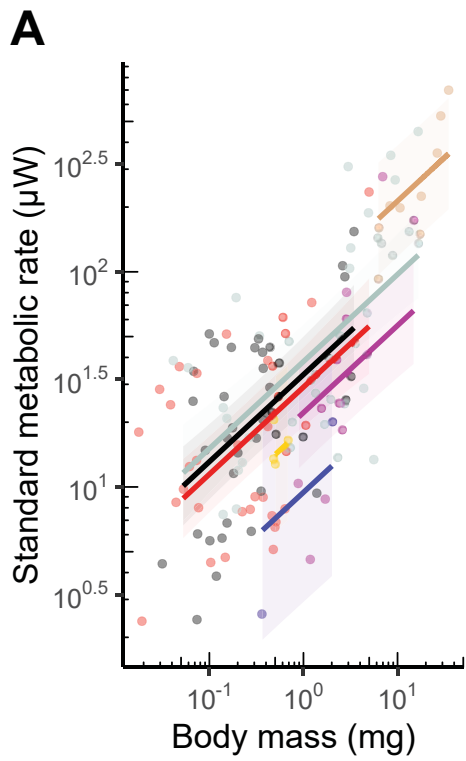
556 **Figure 2.** Relationship between standard metabolic rate and dry body mass of ant subfamily
 557 clades. (A) Metabolic rate-body mass relationship coloured by subfamily clade, lines and 95% CI
 558 are estimated marginal means (LME: $R^2 = 0.59$, Table 1); species raw values as points ($n = 139$
 559 site-species); (B) Mass independent metabolic rate mapped onto phylogeny: mean residuals from
 560 standard metabolic rate (SMR) ~ body mass (BM) (LME: $R^2 = 0.54$, Table 1), for 34 genera
 561 coloured by subfamily clade and sized by mean body mass (scaled between 1-5 for visualisation).

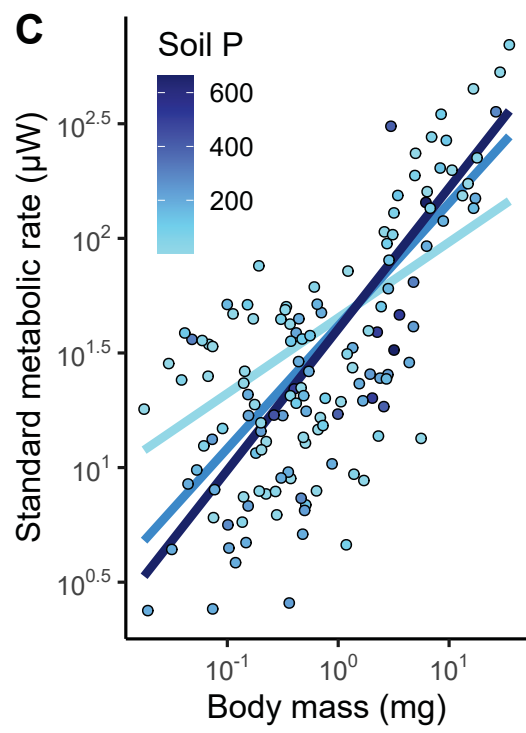
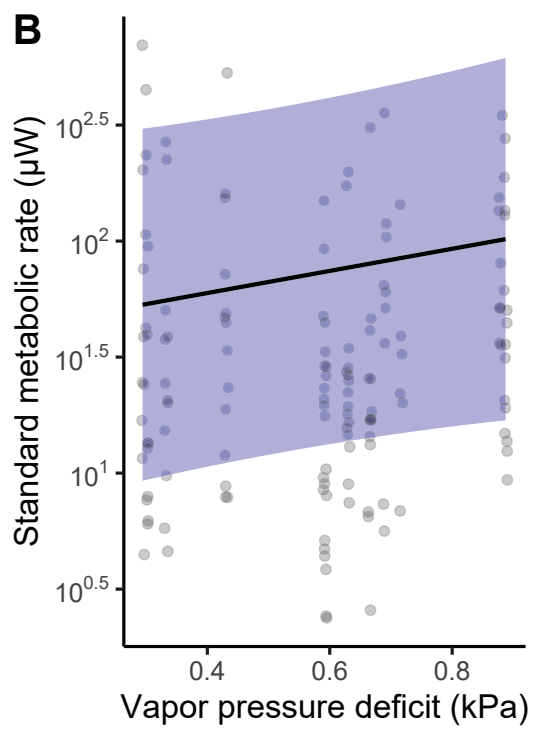
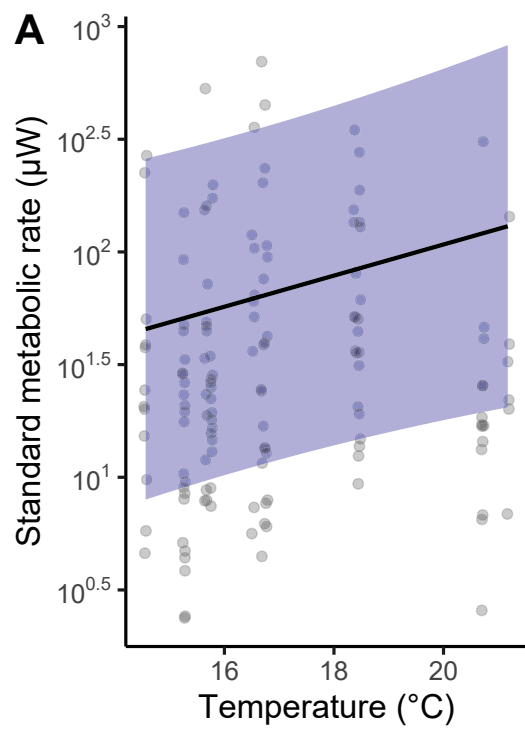
562 **Figure 3:** Relationships between \log_{10} SMR (μW), \log_{10} ant dry mass (mg), and environmental
 563 predictors. Showing estimated marginal means and 95% confidence intervals from model fits
 564 (Table 2, Table S6) and raw species values as points ($n = 139$ site-species). (A) Testing the
 565 metabolic cold adaptation hypothesis: $\text{SMR} \sim \text{body mass} * \text{soil phosphorus} + \text{mean annual}$
 566 $\text{microclimate temperature}$ (PGLS: Pseudo $R^2 = 0.68$). (B) Testing the aridity hypothesis: $\text{SMR} \sim$
 567 $\text{body mass} * \text{soil phosphorus} + \text{body mass} * \text{mean annual vapor pressure deficit}$ (PGLS: Pseudo R^2
 568 $= 0.69$), vapor pressure deficit = aridity. (C) Testing the P-limitation hypothesis: $\text{SMR} \sim \text{body}$
 569 $\text{mass} * \text{soil phosphorus} + \text{body mass} * \text{mean annual vapor pressure deficit} + \text{mean annual}$
 570 temperature (PGLS: Pseudo $R^2 = 0.69$), mass-metabolic slopes for predicted minimum (light
 571 blue), mean (blue), max (dark blue) soil P values (ppm), 95% CI not shown in (C) to simplify
 572 presentation.

573 **Figure 4:** Relationships between discontinuous gas exchange, body mass, and aridity (vapor
574 pressure deficit - VPD). (A) Hygric hypothesis (GLMM: $\chi^2 = 33.01$, $p < 0.001$), showing
575 relationship between the proportion of individuals displaying DGC per species and \log_{10} ant dry
576 mass (mg), lines and 95% CI are estimated marginal means from logistic regression for the
577 minimum, mean, and maximum VPD (kPa) (B) Ventilation frequency per hour as a function of
578 body mass for DGC exhibiting species, lines are estimated marginal means with 95% CI from
579 linear regression model (OLS: $R^2 = 0.20$). Species raw values shown as points (A = 139 site-
580 species, B = 78 site-species).

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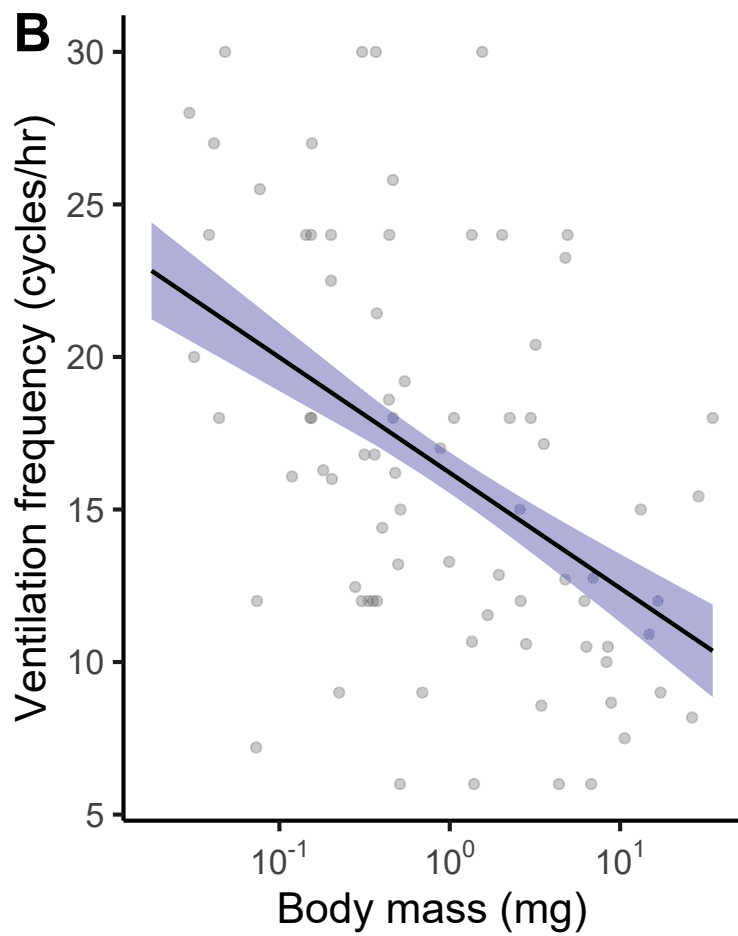
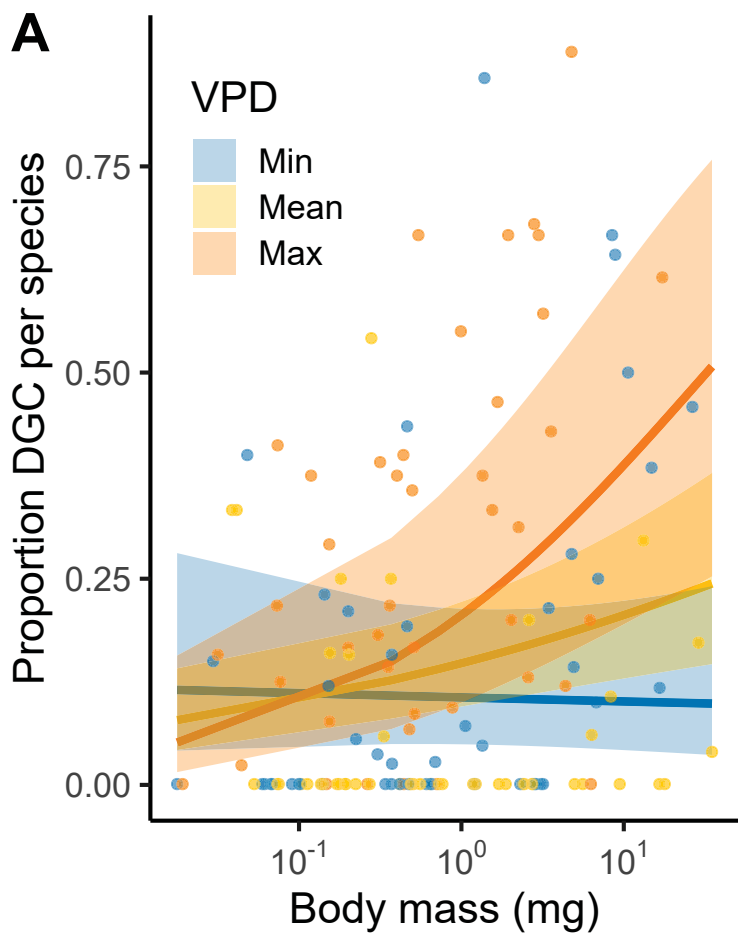


Table 1: Metabolic trait hypotheses relating to metabolic rate and ventilation patterns, with summarised mechanisms and our predictions for this study.

Hypothesis	Mechanism	Predictions	References
Niche conservatism hypothesis	Metabolic rate has evolved to meet the energetic demands of conserved ecological traits and life history strategies in addition to biophysical constraints related to body mass.	Mass independent metabolic rate varies across ant subfamily clades. Metabolic rate and mass-independent metabolic rate show strong phylogenetic signal.	(11, 64, 65)
Metabolic cold adaptation (MCA) (Figure 1C)	Cold-adapted species must complete key life tasks in a short warm season which requires elevated metabolic rates.	Cold environment ants have higher metabolic rates compared to warm environment ants, when metabolic rate is measured at the same temperature.	(46, 47)
Metabolic aridity hypothesis (Aridity) (Figure 1D)	Metabolic rate is depressed in arid environments due to desiccation risk and lower net primary productivity.	Higher desiccation for smaller ant species lowers metabolic rate, steeper metabolic-scaling with increasing aridity.	(66, 67)
P-limitation hypothesis (Figure 1E)	For small-bodied animals, dietary limitation of phosphorus increases metabolic rate due to the costs of maintaining C:P homeostasis under an imbalanced diet. Phosphorus could be limiting for smaller organisms in low soil P environments.	Smaller ant species will have elevated metabolic rate in low soil P environments equating to shallower metabolic-scaling relationships with decreasing soil phosphorus.	(12, 50, 68)
Hygic hypothesis (Figure 1F)	Discontinuous gas exchange cycles (DGC) are an adaptation to reduce respiratory water loss by limiting how often spiracles are open in dry environments.	DGC related to interaction between body mass and aridity. Opposing predictions in arid habitats: (a) smaller ants more likely to exhibit DGC due to increased desiccation risk (small surface area to volume ratio) (Figure 1F), or (b) larger ants more likely to engage in DGC due to higher total CO ₂ emissions volume and higher respiratory water loss rates.	(19, 24)

Table 2: Testing metabolic rate hypotheses (Table 1). Response variable is \log_{10} SMR. M = dry mass (mg), Clade = subfamily clade (n = 7), Temp = mean annual microclimate temperature ($^{\circ}$ C), VPD = mean annual microclimate vapor pressure deficit, representing aridity (kPa), soil.P = soil phosphorus (ppm). LME models include 'location' (n = 6) as a random effect.

Hypotheses	LME	df	AIC	R²m, R²c
Niche	$\sim \log_{10}M + Clade$	10	1280.6	0.53, 0.59
Conservatism				
Mass	$\sim \log_{10}M$	4	1288.9	0.47, 0.54
Phylogenetic - PGLS				Pseudo R²
MCA, aridity, p-limitation	$\sim \log_{10}M * \log_{10}soil.P + \log_{10}M * VPD + Temp$	8	1344.7	0.69
Mass	$\sim \log_{10}M$	3	1347.7	0.64