

# Difference of gut bacterial community and wing morphology in *Chlaenius pallipes* (Coleoptera: Carabidae) by habitat condition

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## INTRODUCTION

Carabid beetles are frequently used as indicators of biodiversity and habitat quality. Studies using carabid beetles as an indicator species typically compare carabid beetle communities and assemblages in different environmental habitats, such as in disturbed and undisturbed habitats.

However, identification of the response to environmental changes at the individual and population levels of carabid beetles is needed to assess habitat conditions in areas with low diversity of carabid beetle species or to determine the conditions of rare species.

Body size, wing shape, food quality, and including gut bacterial community can be used to indicate the health condition of an individual or a population of carabid beetles. For this, it is necessary to understand the correlation between these components.



## OBJECTIVES

We tested the assumption that when *Chlaenius pallipes* is present in the stable habitat will cause the species to have more diverse gut bacteria. We also expect that this environment will be associated with a change in *C. pallipes*' morphological shape (body and wing shape) and affect the food resources ( $\delta^{15}\text{N}$ ) of the species.

Considering the ecological traits of the species, the gut bacterial communities of carabid populations inhabiting stable (terraced paddy field, TP) or unstable habitats (large-sized paddy field, LP) were compared.



## CONCLUSION

We found that although each individual has a large variation in the gut bacterial community composition, the gut bacterial community can be used to evaluate the condition of each habitat through consistent investigation.

Habitat assessment based on changes in the number of carabid beetle species and their composition requires relatively long-term research; however, the gut bacterial community of carabid beetles can help identify short-term environmental changes.



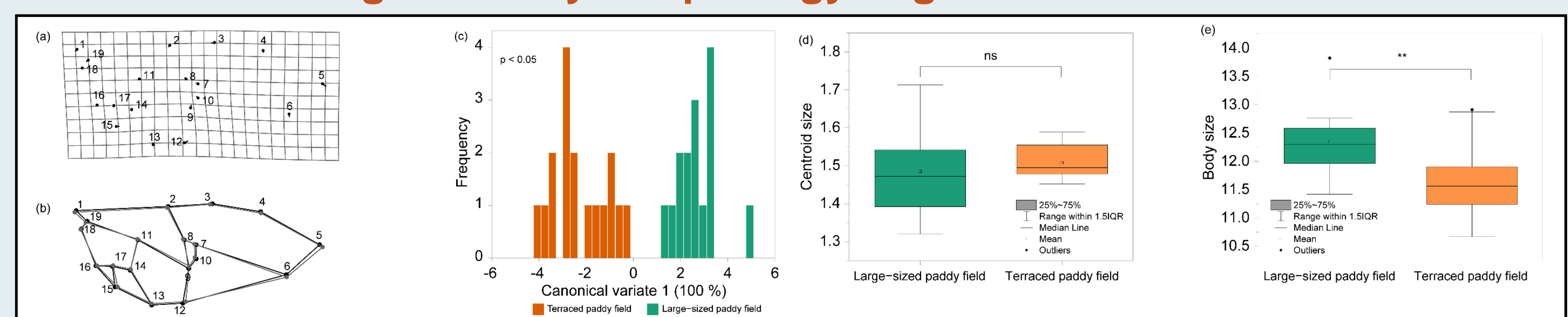
## RESULTS

### Difference of wing and body morphology in ground beetles



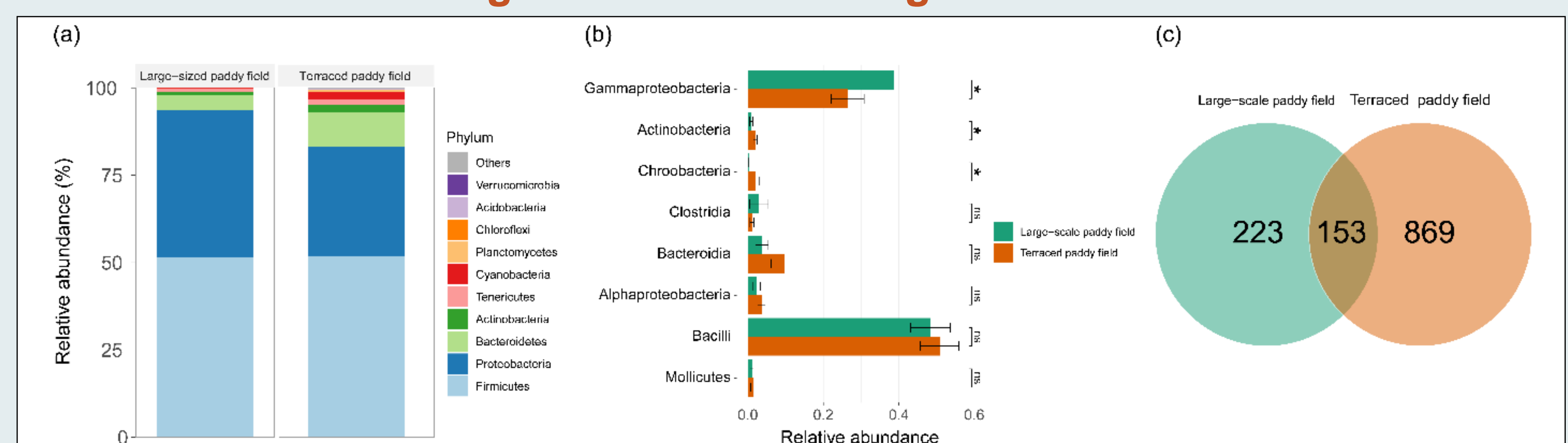
The freeze dried beetle samples were used for nitrogen isotope analysis. The stable nitrogen isotope ratio of *C. pallipes* in the TP environment ranged from 4.45 to 5.72, while the ratio of *C. pallipes* in the LP environment varied from 1.18 to 7.37. *C. pallipes* had a significantly higher stable nitrogen isotope ratio in the TP environment, compared to that of the LP (unpaired t-test  $p > 0.01$ ).

### Difference of wing and body morphology in ground beetles



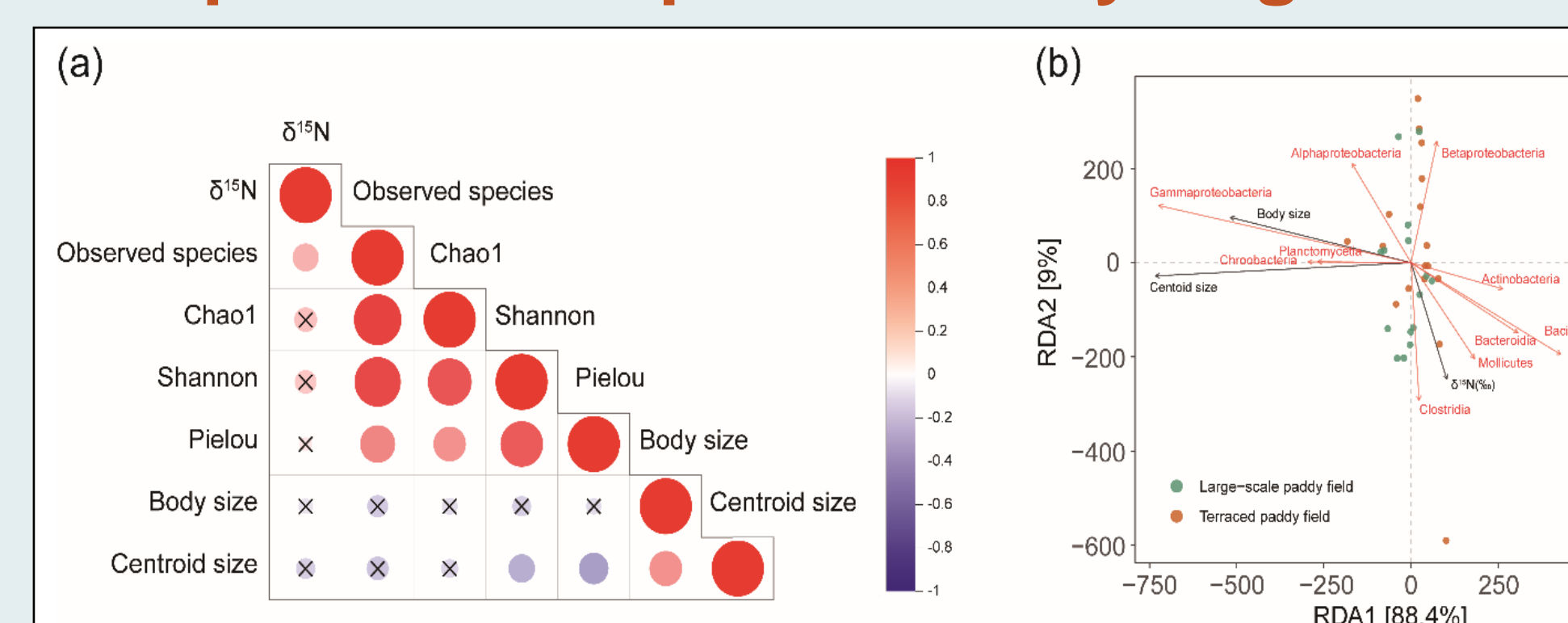
Landmark based geometric morphometrics were used to analysis the wing morphology of carabid beetles. Circles in the deformation grids and grey wireframe graphs indicate the wing shape of individuals with the lowest CV values (a, b). The positive end of CV1 showed the individuals that were grouped in the LP environment, whereas those individuals in the TP environment were grouped toward the negative CV1 axis (c). The wing size, which was estimated by its centroid size, was not significantly different (t-test,  $p > 0.05$ ) between the beetles from the LP and TP environments (d). However, body size was significantly larger in the TP group than in the LP group (e) (t-test,  $p < 0.01$ ).

### Relative abundance of gut bacterial taxa in ground beetles



The gut bacterial community is mainly dominated by Firmicutes, Proteobacteria, and Bacteroidetes. In the gut bacterial community of *C. pallipes* in the TP environment, the proportion of Bacteroidetes was higher than that in LP, while the proportion of Proteobacteria was higher in LP group (Fig. a). At the class level, Gammaproteobacteria in the LP group (36.4%) were significantly higher than in the TP group (26.9%) (Kruskal-Wallis test,  $p < 0.05$ , Fig. b). A total of 153 gut bacterial Operational Taxonomic Units (OTUs) (12.2% of the total OTUs) of *C. pallipes* overlapped between the LP and TP groups (Fig. c).

### Comparison in alpha diversity of gut bacterial community



Significant positive correlations were obtained by Kendall's correlation test for both the stable nitrogen isotope ratio and the observed species richness ( $\tau = 0.25$ ,  $p < 0.01$ ). Centroid size, was negatively correlated with the evenness indices ( $\tau = -0.25$ ,  $p > 0.05$ ;  $\tau = -0.32$ ,  $p > 0.05$ ).

RDA at the class level was performed to understand the main environmental factors, such as  $\delta^{15}\text{N}$  (‰), body length, and centroid size of the hind wing, that contributed to changes in the bacterial community structure. Centroid size ( $r^2 = 0.27$ ,  $p < 0.01$ ) was the highest among the experimental factors that represented the coefficient of determination for bacterial species distribution. Centroid size was positively correlated with Chroobacteria and Planctomycetia and body size was associated with Gammaproteobacteria. Interestingly,  $\delta^{15}\text{N}$  was positively correlated with Clostridia and negatively correlated with Alphaproteobacteria presence.

