

General comments

This manuscript with reference ID bg-2023-87 presents an optimality-based approach investigating the drivers of leaf trait variation along elevational gradients across China. To this end, the authors make use of data available from the China Plant trait database (version 2) to parameterize and test an eco-evolutionary optimality model for leaf nitrogen per unit mass (N_{mass}). Results obtained with a Bayesian phylogenetic linear mixed model suggest that variation in leaf stoichiometric traits are mainly controlled by species identity and phylogeny, thus indicating that accounting for community level responses and shifts in species turnover may allow for a more dynamic representation of ecosystem processes in Earth System models. Albeit the fact that this conclusion is not novel, the analysis appears to be sound and the manuscript is concise and very well written. Hence, I conclude that the article should be of great interest to the academic readership of the journal, and subject to minor amendments and modifications (see recommendations in the specific comments provided below), could be considered for publication.

Authors: We appreciate this comprehensive and encouraging evaluation.

Specific comments

The study by Xu and colleagues presents an interesting analysis investigating the drivers of leaf trait variation across environmental gradients. While the manuscript is generally well written and the findings are presented in a concise and informative way, I would suggest adding some further clarifications with regard to (i) statistical analysis, (ii) intra-specific trait variation, and (iii) parameters obtained from remote sensing.

Reviewer: First, there appears to be a potential spatial bias in the analysis (L71-74) investigating trait variation across large spatial scales, such as the large-scale environmental gradients across China. For instance, multiple regression on distance matrices (MRM) could be applied to quantify the relative amount of trait variation in response to space and environmental factors in ecological data (Lichstein, 2006) and to relate phylogenetic or functional beta diversity to spatial and environmental distance (Swenson, 2014).

Authors: We have implemented MRM to account for the spatial effect on trait variation, and added this it into the results (Line 151-154; Supplementary data Table S2). We found that leaf stoichiometric traits were not significantly related to spatial distance, but were strongly explained by climatic factors. We have summarized this finding as follows (Lines 204-206):

“MRM analysis also showed that trait variations were strongly explained by climatic factors, but not significantly related to geographic distance – indicating that the purely spatial effect on trait values is weak (Supplementary data Table S2).”

Reviewer: Second, the lack of phenotypic plasticity in leaf stoichiometry (L237-239) and the associated conclusion that leaf stoichiometric traits might be mainly controlled by species identity and phylogeny without proper consideration of intraspecific trait variation (ITV) could be misleading as the mechanisms driving trait variation across environmental gradients have been reported to shift across large spatial gradients (Ackerly & Cornwell, 2007). Whereas, across larger spatial scales abiotic factors, such as temperature and precipitation, represent

key determinants of ecosystem processes, at smaller spatial scales other biotic factors, such as competition among coexisting tree species, strongly affect ecosystem structure and functioning via the composition of the local species pool (Hofhansl, 2021). Hence, biotic factors can have equally strong impacts on trait expression as the dominant abiotic driver (Albert, 2010; Jung 2010, Violle 2012). As a result, an increasing number of studies documented the importance of ITV and thus it would be great to see a discussion on the potential of including ITV in optimization-based models, such as the one applied in this study.

Authors: We fully agree about the importance of intraspecific variation along trait gradients. To address this comment, we have implemented trait-gradient analysis in order to quantify intraspecific trait variation (Lines 174-180; 196-200), and added discussion of this result (Lines 340-346):

“Trait-gradient analysis showed that in addition to species turnover, intraspecific trait variation plays a role in determining trait shifts at a regional scale (Fig. 3). The intraspecific slopes for C_{mass} , N_{mass} and their ratio were calculated for 19, 19 and 42 species respectively. Only 9, 8 and 16 of these species showed significant slopes. The intraspecific slopes for N_{mass} and C:N ratio ranged from 0.7 to 2.1 and 0.6 to 1.9, respectively. The slopes for C_{mass} ranged from 0.8 to 1.4 except for one species (*Asparagus dauricus*) that had a negative slope.”

“Systematic variation in community-mean leaf stoichiometric traits along climate gradients can be achieved through progressive species replacement at a macroclimatic scale, and intraspecific trait variability at a regional scale (Liu et al., 2019; Yang et al., 2016). Some species in this study apparently adjusted their leaf stoichiometry along major environmental gradients, possibly via genetic adaptation over multigenerational timescales. Due to the lack of intraspecific data within communities, we could not assess the degree of variation among conspecific plants in the same environment. Intraspecific variation within communities may however increase functional diversity and promote species coexistence (Westerband et al., 2021) and potentially provide a buffer against climatic variation and change (Ahrens et al., 2021). Further studies are needed to better understand intraspecific trait variation (Moran et al., 2016) in order to assign appropriate timescales for the dynamic responses of traits to environmental changes in Earth system models.”

Reviewer: Third, the lack of a significant relationship of leaf stoichiometry with LAI and soil fertility (L252-253) both obtained from remote sensing estimates, and the controversial finding that nitrogen allocation to metabolic and structural components was related to leaf area index (L324-325), made me wonder if it would actually require data obtained from in-situ measurements (that match the spatial and temporal extend of the trait data) in order to identify these effects.

Authors: No LAI or soil information was collected during sampling, so it was necessary to rely on remotely sensed LAI and global soil products – a strategy which, we agree, carries significant uncertainty. We have pointed this out in our revised discussion (Lines 278-280; 336-340):

“Uncertainty in our soil fertility data may was inevitably introduced due to our reliance on a gridded soil map (Shangguan et al., 2013). More studies including *in situ* soil measurements

are needed to more comprehensively investigate the effect of soil properties on plant stoichiometry.”

“...our predicted N_{mass} was constrained within a narrow range, despite the well-captured variations in M_a and $V_{\text{cmax}25}$. The predicted N_{mass} in tropical forests with high LAI were systematically underestimated due to the low intercept (Supplementary data Table S1). We recognize that our method to estimate N_{mass} may overlook additional functions of N in leaves, such as chemical defences, perhaps causing greater variation than predicted. This requires further investigation.”

Reviewer: Overall, I would appreciate a more thorough discussion on some of the topics indicated above and would therefore recommend revising the manuscript based on the findings presented in the scientific literature (see additional references to be considered below) and how these results could be used to improve the dynamic representation of plant tissue stoichiometry in Earth System models.

Authors: We have added discussion about the role of intraspecific trait variation in particular. Please see above.

Reviewer: L316: correct typo: “our EEO-based approach thus suggests ...”

Authors: We have corrected this in the text.

Reviewer: L613-615: Please add a description for the (i) labels “Cmass”, “Nmass”, “C:N ratio”; (ii) colour code (red-blue gradient); and (iii) test statistics used in respective panels of Figure 1 A/B/C.

Authors: We have added this information in the caption.