Biogeosciences Discussions

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Reply on RC1

Marina Corrêa Scalon et al.

Author comment on "Contrasting strategies of nutrient demand and use between savanna and forest ecosystems in a neotropical transition zone" by Marina Corrêa Scalon et al., Biogeosciences Discuss., https://doi.org/10.5194/bg-2022-63-AC1, 2022

In general, reviewer #1 asked for more details and information regarding concepts in the introduction, details that were missing or unclear in the methods and statistical analysis. We believe all major and minor comments were addressed – we are providing all required information in the text and added 2 new supplementary tables with statistical results from the analyses. Also, there were indeed some mismatches between the text and figures and we acknowledge both reviewers for noticing it. Figures were redrawn and all minor recommendations were accepted. Please find our point by point answer to the reviewers comments.

Referee comment: Introduction needs some information. Particularly in relation to introduce important concepts for the better understanding of this study such as nutrient concentration, nutrient resorption, nutrient demand vs nutrient uptake, and then nutrient uptake efficiency vs nutrient use efficiency. Furthermore, the hypotheses will should clarify whether the study focuses on the species scale, on the community scale or both.

Reply: New information was added and hypotheses' scale were better clarified. Specifically, we now define: "*By definition, nutrient use efficiency is the amount of production per nutrient unit (Chapin, 1980) and can be estimated as the ratio of. NPP per unit of nutrient demand (Bridgham et al., 1995). Nutrient demand is the sum of nutrient accumulated in above and belowground biomass and nutrient returns to the soil via litterfall including resorption efficiency, while nutrient uptake excludes nutrient resorption efficiency. Nutrient resorption is defined as the process from which plants withdraw nutrients from senescent leaves prior to leaf abscission, and its efficiency is calculated as the proportional resorption from green to senesced leaves (Killingbeck 1996)*."

Referee comment: Materials and methods need some clarification. Particularly in relation to the experimental design of field sampling. Information is needed on how many replicates were sampled. Are there only two replicates per vegetation type (cerrado and cerradão), and they are referred to as plots in the manuscript? Where were soil samples collected, under the tree canopy or outside? How was species abundance measured, which method was followed? Could you describe species abundance and basal area? All species are found in the all replicates? Also, all sampled species are trees? I recommend adding this information in table S1, and this table add on the manuscript. Information is also needed on how you have measured net primary productivity. It is really important

parameter in this study, and there is very little information in the methods. Also, were senescent leaves collected from the same individuals collected previously in January 2008 Information on analytical techniques of soil data is also needed.

Reply: We understand the reviewer concern with the study scale, especially because we are scaling up from species to community and ecosystem functioning. We only have one replicate per vegetation type, i.e., for NPP calculations the sample unit is one. For that reason, we used error propagation techniques throughout the subplots and community weighted means to represent the community (as you already noted in your comment below).

We clarified now species selection criteria and inserted Table 1 (old Table S2) in the main document, with importance value index *(* IVI) and relative dominance values for each species. We also included all information required on this regard (5 individuals were used and the same individuals were sampled in both seasons): "*Calculation of relative dominance was based on previous census collected in the area (2009, 2012, 2014), by dividing the species dominance (i.e., total basal area of the species) and the sum of the dominance of all species multiplying by 100. The IVI was calculated as the sum of the relative frequency, the relative density and the relative dominance of each species within the community. We choose only adult trees with at least 5cm in diameter at breast high (dbh) in the cerrado and 10 cm dbh in the cerradão, following standardized criteria for individual inclusion in forest and savanna vegetation. For each species, 5 individuals were chosen, from which we collected samples…"*

For soil samples, we included only data from two soil depths – the text was changed accordingly with more details added as well: "*Within each 1ha plot, 25 subplots were delimited, from where soil samples at 0-10 and 10-20 cm depth were collected in all 4 corners and in the centre of each subplot, totalling 200 samples for each vegetation type. Soil chemistry data were analysed according to EMBRAPA procedure (EMBRAPA, 1997) and were provided by ForestPlots database Lopez-Gonzalez et al. 2011 (Table S1)."*

Primary productivity measurements were briefly described – more detailed description of NPP calculations can be found in associated published literature cited. It now reads: "*Here we quantified the major components of NPP, including the canopy (leaves, twigs and reproductive parts), wood (stem, coarse roots and branches), and fine roots during 2014 to 2016. Data were collected following GEM protocols (Malhi et al. 2021) and methods are described in detail in Mathews et al. (2014) and Malhi et al. (2021). Briefly, for the canopy NPP component estimation, litter traps sampled biweekly together with monthly canopy leaf area index were used. For wood component estimation, annual census and dendrometers measuring growth rates were converted into woody biomass production. Fine root production was measured with ingrowth cores installed and sampled every three months"*.

Referee comment: Information on some statistical methods needs to be improved. In particular, the reason behind the use of community weighted mean to scale up species values to community for nutrient concentrations, which affects nutrient demand, nutrient use, and nutrient use efficiency and nutrient uptake efficiency parameters at community scale. Species selection would produce a strong bias in the community value, especially when target species belong to different families. Could you justify species selection in the methods, indicating for example their abundance in plots. Furthermore, the use of community weighted mean is only suitable for use with many replicates to avoid Type I

error, or to include random effects on the models. For this reason, more information is needed on the statistical methods. Did you include any random factors on the MANOVAs and ANOVAs? What are the variables, the fixed terms and the random terms? And what R function did you use?

Reply: Information was added. We now specify how calculations of CWM were performed as well as dependent and independent variables used in MANOVA. Species selection criteria were already explained in the section below – see comment #2. We used function 'manova' from 'stats' package, however we do not see the necessity to cite base R packages, or functions.

"*To up-scaled species value for the whole communities, we calculated the community-weighted mean (CWM) for each organ using species relative dominance to weight the nutrient concentration (Muscarella & Uriarte, 2016). To compare community-weighted nutrient concentration average means and resorption efficiencies between the two vegetation types and between different organs, we performed a two-way multivariate analysis of variance (MANOVA) followed by univariate analysis of variances (ANOVA's) and Tukey HSD post-hoc test. The independent variables were site and plant organ while the dependent variables were the different nutrient concentrations (N, P, K, Ca, Mg) or resorption. Data normality and homogeneity of variances assumptions were previously checked with Shapiro-Wilk multivariate normality test using package 'mvnormtest' (Jarek and Jarek, 2009) and Levene test using package 'car' (Fox et al., 2012), respectively. All statistical analyses were performed in R software version 4.0.1 (R Core Team, 2019)*."

Referee comment: Discussion section would be clearer if separate paragraphs were used to discuss each hypothesis, indicating the key results of this study. In this sense, the authors dedicate the first and the second paragraphs to discuss a higher P content in wood by Cerrado species than Cerradão species as a key result when they did not report any statistically significant test value in the results for sapwood and heartwood (line 178, Fig1, Fig S1), as they did for inner bark. Could they justify this or report a test value in the results?

Reply: We re-ordered Discussion section to follow the three suggested hypothesis. Indeed, for some unknown reason the asterisks were not displayed in Fig 1. Differences are now shown in the figure and tests were acknowledge in the text. At the species scale, there was significant difference between cerrado and cerradão species for P content in wood. At the community scale, even though there was no difference between CWM nutrient concentration, there was also remarkable difference in P demand for the wood component and therefore we wanted to highlight this finding in the discussion.

Referee comment: Throughout the manuscript try to homogenize concepts as plots, area or sites, and to differentiate between species scale or community scale.

Reply: Done. We used sites throughout the ms to refer to the community.

Referee comment: Species name would be in italic format in the text, such as *Hirtella glandulosa* and *Emmotum nitens* in line 117.

Reply: Done.

Referee comment: Plant nutrient concentrations would be in mg/g instead of %.

Reply: Done.

Referee comment: You should clarify the statements of results and discussion in lines 229-230 and 278-280, because they can be misinterpreted. Ca uptake should always match demand and never resorption because the differences between nutrient demand vs nutrient uptake, and nutrient uptake efficiency vs nutrient use efficiency is based on use or not the nutrient resorption efficiency, which for Ca is zero.

Reply: We agree. Our intention was to compare between sites (cerrado vs. cerradão). We rephrased both statements only mentioning Mg differences, since Ca was used in the calculation for resorption metrics, to avoid confusion.

Referee comment: Are there significant differences between sites on N uptake efficiencies? On the figure 4 is indicated, but not on the text (line 224).

Reply: There is not (P-value is 0.078). Figure asterisks were placed incorrectly, and we are very sorry about that. We double checked this issue in new figures.

Referee comment: I recommend modifying Figure 4 and deleting the last row, because it is a repeat of Figure 3. Also, could you please provide test value for the Nutrient Use efficiency of P, because it does not seem significant in Figure 4, as well as for the nutrient uptake efficiency of K?

Reply: We added supplementary table with results from z-tests (transformed p-values). Deleted the last row of Figure 4, as it has redundant information. Asterisks are now displayed correctly.

Referee comment: I recommend reducing the importance of statements related to fine root production, because the sampling carried out is not accurate and other non-target species, such as grasses, could be measured.

Reply: Done. It now reads: "*However, cerrado species demanded more P and K to the fine roots, suggesting that generally cerrado species require more nutrient for fine roots production. Fine roots are important component of the biomass, reflecting in a higher uptake of limiting nutrients in soil (Loiola et al., 2016), and are essential to overcome the strong water deficit during the dry season in the Cerrado (Oliveira et al., 2005). Therefore, the efficient fine root production in the cerradão vegetation may be an important adaptation to readily absorb water and nutrients during the strongly seasonal rainfall period (February et al., 2013). However, since our sampling strategy may have measured non-target species, such as grasses, our results could be biased.*"

Referee comment: I recommend to avoid any reference to figures or tables in the discussion, because they should be indicated on the results.

Reply: Done. All reference to tables and figures in the discussion were removed.