How deep do we dig for surface soil? A comparison of patterns of microbial C:N:P stoichiometry between topsoil and subsoil along an aridity gradient

General comments

This paper focus on the sampling depth for analysing microbial C: N: P stoichiometry at 0-10 cm or 10-20 cm soil depth. It is an interesting study made in permanent grassland with an aridity index, but the interpretation and presentation of the data should be improved.

The paper will profit from clearer hypothesis that can be tested, and more clear wording and presenting of the results. I do also suggest putting the correlation analyses given in 3.1 into a table, which would make it more accessible for the reader. Your question: "How deep do we dig for surface soil?" Should be clearly answered in the conclusion.

Specific comments

Normally subsoil is used for the soil under the surface soil/ topsoil that are less affected by plant roots and tillage operations. However, I assume there were no tillage at the sites referred to in the present paper. The root distribution and rooting depth for the different sites are not given, but in permanent grassland most of the rooting and microbial activity is in the upper soil layer. I would still be reluctant to use the "subsoil" as a term for the soil layer at 10-20 cm depth as the roots would likely go deeper than 10 cm. Surface soil and topsoil are in many cases used as synonyms and the heading is therefore confusing. I suggest in stead: How deep do we dig for surface soil? A comparison of patterns of microbial C: N: P stoichiometry between an upper and lower soil layer along an aridity gradient.

When you present hypotheses, it should be possible to test them and to either confirm or reject them and the result of the testing of the hypotheses should be clearly presented in the conclusion.

(i) microbial C:N and C:P ratios increase and the microbial N:P ratio decreases across an aridity gradient because of differences in nutrient-use efficiency.

The first part of this hypotheses "microbial C:N and C:P ratios increase and the microbial N:P ratio decreases across an aridity gradient», you have actually tested in the present paper, but the result is not clearly written in the conclusions. In Figure 2, C:N, C:P and N:P ratios are given along an aridity Index (Gradient). Because of the very low relationships between the ratios and the aridity index, this part of the hyphothesis cannot be confirmed. R²=0.1 is very low. In discussion you write: "microbial C:N and C:P ratios increase and the microbial N:P ratio decreases across an aridity gradient» I do not agree with this statement. Because of the low R², a P<0.05 does not say much. If you look at figure 2, you see that the variation in within C:N and C:P sites at the same aridity is much larger than the impact of Aridity index. I would rather call it a trend, then to state a significantly impact.

The second part of the first hypothesis "because of differences in nutrient-use efficiency.", you do only discuss and do not test. I would leave that out from the hypothesis.

(ii) Due to variations in resource supply among different soil depths, the effects of driving factors on microbial C:N, C:P and N:P ratios might decrease with soil depth.

This hypothesis you have not tested and cannot do, as you do not know if variations in resource supply among different soil depths actually do effect driving factors on microbial C:N, C:P and N:P ratios. What you can test is: "Microbial C:N, C:P and N:P ratios do vary with soil depth." In the results 3.1 lines 222 to 223 you write: "Moreover, the microbial C:N ratio in the subsoil was significantly higher than that in the topsoil (Fig. 2b)." I assume you must mean table 2? If this is the case, such a hypothesis could be confirmed for C:N ratio, and rejected for C:P and N:P ratios. Obs,. You write in

the abstract (line 32-34): "We found that the microbial C:N, C:P and N:P ratios varied with soil depth.» According to table 2, they do not.

(iii) to adapt to the imbalance of resources, microbial C:N, C:P and N:P ratios vary between soil depths and at a depth of 10 cm, which could influence the research on the vertical patterns of microbial stoichiometry.

I do not understand what you mean by this hypothesis. You should convert it to a hypothesis that can be tested and clearly present the result of the hypothesis. Do you mean "Microbial C:N, C:P and N:P ratios do vary with soil depth. At 0-10 cm depth the ratios are more influenced of an aridity gradient and other ecological factors than at 10-20 cm soil depths"?

In 3.1 you refer to "environmental gradient" in the title, but you do not refer to what you mean with «environmental gradient.»

You do focus on the impact of Latitude, but I do not understand for which purpose. And again the degree of explanation is low ($R^2 = 0.14$) and the variation is large.

Because this study is done on three grassland types (meadow steppe, typical steppe and desert steppe) with corresponding soil types, I do miss the discussion on impact of grassland types, plant roots and rooting pattern on the microbial stoichiometry.

Because aridity gradient (index) is central in this study it should be given how it was calculated (Line 171-172).

Figure A3 need some introduction. How did you develop this?

Technical corrections

Line 181 and line 189, You must explain what a universal conversion factor is, what the units are and give a reference to where you got it from.

Line 185 Which principal method is used? Cloroform fumigation? Hedley and Stewart (1982) is not given in the reference list.

Line 201 Was the log10 transformed ratios normally distributed?

Line 223, Should it be table 2, not Fig. 2b?