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Letter to the Editor

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Title: Improvement of soil quality by vegetation restoration in newly reclaimed croplands

Dear Editor,

We are thankful to you for editorial assessment and review of our manuscript. We gratefully acknowledge the input of editor/reviewers and, certainly, this rigorous review helped us improve the manuscript. Now we have modified the manuscript as per the raised concerns, and detailed corrections/responses are listed point by point in "**Response to Editor/Reviewer's Comments**". All revisions are yellow highlighted in the revised manuscript. We do hope that our revisions will fulfill the publication requirements of your esteemed journal.

Best Regards,

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On behalf of all co-authors

Response to Reviewer 1 Comments

Point 1: While the paper provides an extensive analysis of the consequences of planting different crops/plants species as a soil restoration measure, the resulting understanding remains rather descriptive, describing observed changes, but not translating the results into a conceptual advance in understanding the contexts, interactions and trade-offs involved. And I mean this particularly from the perspective soil health, in a multifunctional perspective. Many microbial groups and metabolite groups are mentioned, and reference is made to biochemical pathways, but I expect an explicit link from them to soil functioning and restoration. As a reader of SOIL I think the current discussion falls below my level of expectation, although I do support the publication of this data and results in another place.

Response: *We are thankful to the Editor and Reviewer for hardworking on our manuscript, and providing us with the opportunity to improve the manuscript. In our revised version, we have addressed all the comments raised by the reviewers, and a point-by-point response to each comment is appended herewith. We hope that the revised version of the manuscript will be considered for publication in "SOIL".*

Point 2: It is a complicated study in terms of techniques used, I am familiar with most, but metabolomics I only have a cursory understanding of, cannot judge the quality here. I suggest to get a reviewer who can.

Response: *We are thankful to the Reviewer for his/her diligence and responsibility. And we hope that the revised version of the manuscript will be considered for publication in "SOIL".*

Point 3: The starting condition is poorly described, what were the sites reclaimed from? What was the nature of the degradation, the disturbance? Without this we cannot judge the meaning of the reported effects. The organic matter levels seem to be unexpectedly high for my understanding (5-10%!) on what is described as a sandy loam in the first 20 cm.

Response: *Many thanks for your valuable suggestion. The related information has been added in the revised manuscript. The objective of this study was to find suitable plant species to restore soil during reclamation of abandoned croplands in subtropical China, which was carried out in the field (cropland that had been abandoned for eight years, and was reclaimed in 2021) by investigating the effects of three different plant species (vegetable, corn, and peach) on soil quality. Furthermore, it is not very good high for soil organic matter level (6.03 g/kg), which is approximately equivalent to 0.603%.*

Point 4: The experimental setup is one of a practical applied experiment. In the present ms all effects are subscribed to the different plant species put in place in the treatments. But at the same time the treatments vary in planting density, but also type, timing and amount of fertilizer used. The control, received no plant, but also no fertilizer. In addition, the planting types are not described in very much detail, what variety was used? For vegetables we even completely do not know which species/crops were used. In places a plural form is used suggesting multiple species were used, but did they grow together in the same year, or was it a rotation? What type of management was done in addition to the fertilization, was there any irrigation, and pesticides were they used? How was the land prepared for the crop planting? Any tillage or other site preparation measures?

Response: *Many thanks for your valuable suggestion, which will significantly improve the quality of this manuscript.*

Revised: "The experiment contained four different treatments, which were carried out by planting vegetables (during October to March, cultivar "heixiaopang", a kind of leaf vegetable, purchased from Qingdao North-South Seed Industry Co., Ltd., Qingdao, China; and during April to September, cultivar "hangqie 2010", a kind of eggplant, provided by Hangzhou Academy of Agricultural Sciences, Hangzhou, China), corn (cultivar "qianjiangnuo 3", provided by Hangzhou Academy of Agricultural Sciences, Hangzhou, China), peach (cultivar "zhongtao 5", provided by Hangzhou Academy of Agricultural Sciences, Hangzhou, China), and the control without any plants in the field reclaimed from abandoned croplands (Figure 1a–d). Each treatment had five plots, and each plot consisted of an area of about 125 m2 (length 25 m × width 5 m). Furthermore, the planting density of vegetables, corn, and peach was 25 cm × 25 cm (for leaf vegetable) or 40 cm × 50 cm (for eggplant), 30 cm × 50 cm, and 4 m × 5 m, respectively. For vegetables and corn treatments, at early spring and autumn, the top 0–20 cm soil was mixed with sheep manure at 15 t/ha and chemical compound fertilizer (nitrogen: phosphorus: potassium = 15: 15: 15, without micronutrients) at 0.75 t/ha before planting (twice per year, equal to 2.63 t nitrogen input per ha every year). For peach treatment, sheep manure at 30 t/ha and chemical compound fertilizer at 1.50 t/ha was applied at early winter (once per year, equal to 2.63 t nitrogen input per ha every year). In addition, no fertilizer was used for the control. Other field management practices were set according to the local farmers' actual habits, such as tillage, irrigation, and so on".

Point 5: Regarding analysis, why were OTUs used to describe microbial diversity and not ASVs? How many samples were run together on a Miseq plate? What was the read count per sample, before and after bioinformatics? Also you say, ‘Principal component analysis (PCA) was conducted using Bray-Curtis metrics’, but PCA exclusively works with Euclidean distances, so what did you do? PCoA? Or PCA? What do you mean with ‘structural variation’ (L201), why not simply variation? Also, analyses and calculations are done at phylum, family and genus level, but the choice comes across as quite arbitrary – what is the rationale for doing one analysis at genus level and another at family level? How was the heatmap constructed, what correlation matrix is lying underneath? When was a bacterium considered dominant (L203)? Finally, an analysis is made of cross-correlations among microbial taxa and metabolite groups – but I wonder, what do these correlations mean? Quite some abiotic things change across planted vs unplanted controls, to what extend are these relationship confounded? How does fertilization yes/no and rhizodeposition from plant roots yes/no influence microbiomes and associated metabolite profiles? Also for the metabolites, they are extracted from soil, but to what extend do they originate from microbes or plants? And since the treated plots have more plants then the control, what does that tell us?

Response: *Many thanks for your valuable suggestion. Actually,*

1) OTUs were obtained through third–generation high-throughput sequencing, while ASVs were obtained through second–generation high-throughput sequencing. Furthermore, 20 samples were run on a Miseq plate (four treatments, and five replicates for each treatment).

2)“Indeed, there were 58,895, 64,511, 68,697, 80,006, and 80,027 raw reads for vegetables treatment; 79,851, 80,002, 79,966, 79,980, and 79,981 raw reads for corn treatment; 80,064, 68,024, 79,888, 79,956, and 80,115 raw reads for peach treatment; 79,950, 79,862, 80,190, 68,020, and 79,925 raw reads for the control. After bioinformatics analysis, there were 46,324, 50,652, 49,650, 54,637, and 59,487 high-quality reads for vegetable treatment; 59,863, 60,815, 64,090, 54,349, and 57,234 high-quality reads for corn treatment; 56,810, 51,238, 61,138, 59,511, and 58,630 high-quality reads for peach treatment; 60,074, 60,392, 55,475, 52,594, and 59,162 high-quality reads for the control”.

3) Yes, Principal component analysis (PCA) was conducted here, the sentence has been revised as follow: “Principal component analysis (PCA) was conducted using Euclidean distances to assess variation in soil bacterial communities”.

4) In this study, we mainly focused on the variation of relative abundances at the phylum and genus level, so the heatmap constructed at the family was deleted here.

5) The relative abundances at the phylum and genus level were carried out based on the top 10 bacteria, so in line 203, it should be “the top 10 bacteria.

6) Revised: “Previous study showed that vegetation types were the main factors that drove the variation in microbial community (Zhang et al., 2025b), and soil metabolites played a crucial *role in regulating interactions between microbes and plants. Furthermore, the metabolism of rhizosphere soil was greatly affected by plant roots than the bulk soil (Wang et al., 2019; Raaijmakers et al., 2009). Thus, based on the cross-correlations among soil physicochemical properties, microbial taxa, and metabolite groups, it can be inferred that soil nutrients of reclaimed croplands could be improved by three different types of plants (such as vegetable, corn, and peach), and these changes played an important role in regulating soil bacterial communities and soil metabolite spectrum. All these complicated interactions were important for reclamation of abandoned croplands".*

Point 6: In many places the taxonomy of bacteria is not correctly represented. Often all taxa are referred to as genera, but they actually represent a mixture of different taxonomic levels, that also makes me wonder about the analyses, did you merge them, if so how? And is that valid? I think in a regression trees approach where taxonomy is the predictor you could do it meaningfully in one go. This makes me worried about the identification of the metabolites as well. I also don't appreciate the use of the word vegetation type here, in ecology vegetation type (phytosociological entities) have a very specific meaning of a naturally shaped community. Here it is not even a community, it is one crop species, artificially put in place, and maintained that way. I would talk about plant or crop species effects, or cropping types.

Response: *Many thanks for your valuable suggestion. We completely agreed with the reviewer that it is arbitrary for the presentation of bacterial classifications. Indeed, all the taxonomy of bacteria has been revised as suggested by the reviewers, and the word vegetation type has also been revised into "three different plant species".*

Point 7: One thing that worries me is how the measured properties are evaluated as an improvement or not, why is it better to have more OTUs? Often these things are done intuitively, which I understand, but we need to be critical, why is that really better? In intensive arable system usually bacterial diversity is higher then in more extensive systems, is that good? I don't think so, I think the soil health is often better in the extensive systems, that have less bacteria, but more fungi, and as a consequence improved organic matter (quality), water infiltrating and holding capacity, and less nutrient leaching.

Response: *Many thanks for your valuable suggestion, which significantly improved the quality of this manuscript. Indeed, as shown in previous studies, the microbial community (OTUs) is highly related with soil health such as organic matter (quality). Indeed, the presentation of this manuscript has been revised based on the suggestion from the reviewers.*

Based on the data obtained from this study, more OUTs and alpha-diversity indexes were got in three different types of plants (vegetables, corn, and peach) treatments than the unplanted control. This increase in microbial community might be related to the soil nutrients. In our research, more sheep manure was provided in three plants treatments, and the organic matter contained in sheep manure was good for the growth of microbes.

Point 8: The conclusion largely repeats the results. No need to mention RDA and other experiment technical aspects – tell me 1) what is the message the data told you, 2) what are the wider implications for the research field, for practice, and perhaps beyond.

Response: *Many thanks for your valuable suggestion. The conclusion has been revised and improved as required by the reviewers.*

Revised: "This study indicated that three plants, in particular vegetables could significantly improve the soil quality of newly reclaimed croplands compared with the unplanted control by investigating the effect of three different types of plants in the soil physicochemical properties, bacterial community, and metabolic diversity during reclamation of abandoned croplands after a three year experiment. The soil improvement by vegetables may be mainly due to a significant increase in the richness, complexity and structure of the bacterial community in the reclaimed croplands, while the bacterial genera significantly enriched were unclassified_Vicinamibacterales, uncultured_gamma_proteobacterium, unclassified_Vicinamibacteraceae, and unclassified_A4b. Furthermore, a total of 1,103 metabolites were identified as amino acid derivatives, pyridine derivatives, small peptide, and so on in vegetables vs control, with 89 of upregulation and 41 of downregulation. In addition, there was a significant correlation between the top 20 SDMs and the enriched bacterial genera, and soil properties (especially MBC, AP, SBD, SOM, and TN) were related to variations in bacterial community composition. Overall, this study indicated that vegetables can be used as a good plant species for soil amelioration during reclamation of abandoned croplands".

Point 9: Title: can you include what the croplands were reclaimed from?

Response: *Many thanks for your valuable suggestion. The title has been revised as required by the reviewers.*

Revised: "Impacts of three different plant species on soil physicochemical properties, bacterial communities, and metabolites during reclamation of abandoned farmlands".

Point 10: L28: why is lowering of OM by peach considered a soil improvement? This needs an explanation to be understood.

Response: *Many thanks for your valuable suggestion. This presentation has been revised as suggested by the reviewers.*

Revised: "This study evaluated the impact of vegetables, corn, and peach on soil physicochemical properties, bacterial communities, and metabolites during reclamation of abandoned croplands in subtropical China. Results showed that all three plant species raised phosphorus, potassium (except peach), and microbial carbon, along with the increased root-zone soil bacterial diversity, and the significantly altered soil metabolites. In particular, vegetables gave the most consistent gains in soil nutrients, microbial abundance and diversity, and metabolites species, making it the best option for soil restoration".

Point 11: L30: metabolites extracted from what?

Response: *Many thanks for your carefulness. The metabolites extracted from root-zone soil, which has been added in the revised manuscript.*

Point 12: L30: do these gains have a cost somewhere to? Many of the processes involved are governed by (e.g. stoichiometric) trade-offs, that requires some consideration.

Response: *Many thanks for your valuable suggestion. Indeed, after eight years abandoned, vegetables gave the most consistent gains in soil, microbes, and metabolites during the first three years of reclamation, however, this might be, at least partially, attributed to the continuous application of fertilizers such as sheep manure.*

Point 13: L31: what do you mean with 'a gain in soil'? also for microbes and metabolites – what increased? Abundance, richness, diversity, particular components (species, compounds)?

Response: *Many thanks for your valuable suggestion. Indeed, it is not very clear for this presentation, which has been improved by replacing with "soil nutrients, microbial abundance and diversity, and metabolites species".*

Point 14: L32-33 this sentence is not complete. Also, why focus on one vegetation type? Why not a mixture, or several types that is spread over the landscape in a mosaic?

Response: *Many thanks for your valuable suggestion. This presentation has been revised and improved as suggested by the reviewers.*

Revised: "To find suitable plants as a good means of restoring reclaimed croplands in subtropical China, this study investigated the effect of vegetables, corn, and peach on soil properties, bacterial communities, and metabolites of reclaimed croplands after three years restoration".

In addition, to obtain more profits, most reclaimed farmlands were normally used for large-scale production lately in China by sowing a specific plant species, such as vegetables, corn, fruit trees, and so on. Thus, this study focuses on one plant species.

Point 15: L34-35: reclaimed from what? What were they before? What is the degradation like?

Response: *Many thanks for your valuable suggestion. This information has been included in the revised manuscript as required by the reviewers.*

Revised: "restoring abandoned croplands (used to be farmland, but be abandoned and unclutivated many years, with wilderness of grass and weeds)".

Point 16: L40-42, you need to tell something before about the situation in the control so readers can put these effects in context.

Response: *Many thanks for your valuable suggestion. The required content has been added in this revised manuscript.*

Revised: "used to be farmland, but be abandoned and unclutivated many years, with wilderness of grass and weeds".

Point 17: L45-47: you start with talking about relative abundances, but the evidence you give is about overall diversity levels. Please make consistent. And add some comma's in the right places to help your reader.

Response: *Many thanks for your valuable suggestion. The presentation has been revised as required by the reviewers. In addition, some comma's in the right places have been also added as required.*

Revised: "three plant species all increased soil bacterial abundance and diversity, with 6.21–10.54% increase in operational taxonomic units (OTUs), 6.22–10.53% increase in Chao1 and 2.30–3.11% increase in Shannon indices".

Point 18: L47: redundancy discriminant analysis (RDA), RDA in the literature stands for Redundancy Analysis, what the discriminant is doing in your term is unclear to me. You have linear discriminant analysis (LDA) or redundancy analysis (RDA) – mathematically related approaches, but with a different objective. From the figure I think you did RDA.

Response: *Many thanks for your carefulness, it should be redundancy analysis (RDA), which has been corrected in the revised in manuscript.*

Point 19: L51 I don't think the 'while' is appropriate here, it suggests something else is coming. While we observed X, we also saw Y.

Response: *Many thanks for your valuable suggestion. This presentation has been revised as required by the reviewers.*
Revised: "and redundancy analysis (RDA) revealed that…".

Point 20: L122 what do you mean with 'site' here? How many sites were there? How far apart? Differences in slope, rainfall, etc?

Response: *Many thanks for your valuable suggestion. The related information has been added as required by this reviewer.*
The site here means the experimental plots, there are four treatments including vegetables, corn, peach, and the blank control, each treatment has five plots, so twenty plots are here. All experimental plots were arranged within 1–ha area, all plots were under the same conditions: 6 m above sea level, with a subtropical monsoon climate (average annual temperature of 17.8°C and precipitation of 1,454 mm). All these revisions were yellow highlighted in the revised manuscript.

Point 21: L123-125: I want to see what the variation is in these base variables.

Response: *Many thanks for your valuable suggestion. This required information has been included in the revised manuscript.*
At the beginning of the experiment in 2021, five soil samples were collected using the 5-point sampling method in this area. Test showed that the soil had the following basic physical properties: pH 7.86 (7.45–8.20), soil organic matter 6.03 g/kg (4.40–7.60), total nitrogen 0.42 g/kg (0.12–0.71), available phosphorus 12.30 mg/kg (11.70–12.70), available potassium 378.70 mg/kg (371.61–387.74).

Point 22: L127: change the word restoration here to planting/sowing or so (what fits), because you don't know if the treatment will not restore anything, so while it is the goal, you cannot be sure that it does what you think until you tested it.

Response: *Many thanks for your valuable suggestion. We completely agreed with the reviewer that this presentation is a bit arbitrary. Indeed, the word "restoration" has been replaced by "planting" as suggested by the reviewers.*

Point 23: L127-8 describe this newly reclaimed cropland.

Response: *Many thanks for your valuable suggestion. This presentation has been revised as required by the reviewers.*

Revised: "in the field reclaimed from abandoned croplands".

Point 24: L130-131 okay so you have a compound treatment, combining plant species and fertilizer application in one combined measure. Can you rescale the amounts of fertilizer to equivalent per hectare? Also, can you give a sense of the N-input thus realized?

Response: *Many thanks for your valuable suggestion. Indeed, the amounts of fertilizer have been replaced by "t/ha". And, the amounts of fertilizer have also been converted into N-input per ha every year.*

Point 25: L133 what is in the compound fertilizer? Please give NPK amounts, and micronutrients if included.

Response: *Many thanks for your valuable suggestion. The information of the compound fertilizer has been added in the revised manuscript. Indeed, the NPK amounts in the compound fertilizer is 15: 15: 15, and without micronutrients in it.*

Point 26: L136: the control was sampled to the same depth? Now it reads a bit confusing, I know in the control there is no root zone, but we need to be sure you used the same setup.

Response: *Many thanks for your carefulness. This presentation has been revised as required by the reviewers.*

Revised: "about 1.0 kg of fresh soil for each treatment was sampled from the root zones of the plants (5–20 cm soil layer) and then packed in a sterile bag using a shovel, and the soil of 5–20 cm layer was collected in the control".

Point 27: L138: what do you mean with quickly? 1h, 1day, 1 week? Make precise.

Response: *Many thanks for your carefulness. This presentation has been revised as suggested by the reviewers.*

Revised: "all soil samples were transported to the laboratory using an ice box within two hours for further analysis".

Point 28: L165: what do you mean with 'clean read'.

Response: *Many thanks for your carefulness. Actually, after the sequencing process, raw reads were further filtered, denoised, and concatenated to obtain the high-quality reads, often called as 'clean read'. In order to avoid confusion, this presentation has been revised as required by this reviewer.*

Revised: "In order to ensure data quality, the subreads of the original sequencing data were preprocessed using SMRT Link (v8.0) by removing low-quality reads (minPasses < 5, minPredictedAccuracy < 0.9), and then merged to obtain circular consensus sequences (CCS) that identified using lima (v1.7.0), while the CCS without primers or average length < 1200 bp (or > 1650 bp) were cutoff with cutadapt (v2.7). After the chimera was further removed by USEARCH (v10), the high-quality CCS was then clustered into operational taxonomic units (OTUs) using USEARCH (v2.4.3) with a 97% similarity cutoff, and annotated by blasting against the Silva Release 138 Database using the RDP classifier".

Point 29: L429: also here the sentence after 'while' seems unfinished, the greater what was observed?

Response: *Many thanks for your carefulness. The sentence has been revised as required by this reviewer.*

Revised: "vegetation restorations increased OTUs, Chao1, and Shannon indices, and the greater increase was observed during the initial seven years".

Point 30: L431: what is ACE?

Response: *Many thanks for your carefulness. ACE means abundance-based coverage estimator, one kind of alpha indexes. This presentation has been revised by including the full name in the revised manuscript.*

Point 31: L447: please write out Ras here and elsewhere.

Response: *Many thanks for your carefulness. Actually, RAs here and elsewhere have been replaced by "relative abundances".*

Point 32: L461: "The improvement of microbes in soil quality" I don't get what you mean here. Please clarify.

Response: *Many thanks for your carefulness. This presentation has been improved as required by this reviewer.*
Revised: "The contribution of microbes to soil quality may be mainly attributed to a total of 17 bacterial biomarkers, which were found in all soil samples".

Point 33: L463-471 how likely is it that the strains you found in your plots, do the same as other members of their genus/family in another location?

Response: *Many thanks for your valuable suggestion. The strains found in our plots are in the same phylum/family/genus as other members obtained from another location, therefore, we infer that they might have the same function.*

Point 34: L498: what is FA and GP.

Response: *Many thanks for your carefulness. Indeed, FA and GP are fatty acid and glyceryl phosphatide, respectively. The presentation has been revised by including their full name in the revised manuscript.*

Point 35: L502-504: but do they have a function in the soil itself?

Response: *Many thanks for your valuable suggestion. Previous studies have reported that organic amendments with high proportion of heterocyclic compounds promote soil microbiome shift and microbial use efficiency of straw-C. The reference has been added in the revised manuscript.*

Point 36: L518: write out AK too. Okay, and is that study relevant in this case? I don't know it, but context matters hugely, so be careful with extrapolations.

Response: *Many thanks for your valuable suggestion. AK has been replaced by "available potassium". Also, we completely agreed with the reviewer that it should be careful with extrapolations due to context matters hugely. Indeed, the presentation has been improved as suggested by this reviewer.*

Point 37: L505: essential is too strong in my opinion, plants can also grow on mineral nitrogen.

Response: *Many thanks for your valuable suggestion. It may be arbitrary for this presentation. Furthermore, "essential" has been replaced by "important".*

Point 38: L523: what do you mean by connection here, I assume correlation? Or do you infer a functional dependence? Does microbe A produce compound X? if the latter, you need stronger evidence then an RDA or heatmap.

Response: *Many thanks for your carefulness. It should be "correlation", and this presentation has been improved in the revised manuscript.*

Point 39: L525: what do you mean with CAR and FFA? Write out.

Response: *Many thanks for your carefulness. The presentation has been revised.*

Point 40: L550: Vicinamibacterales is a family, not a genus. Check the others too and correct mistakes. Is A4b a complete name?

Response: *Many thanks for your carefulness. The presentation has been corrected as required by this reviewer. And A4b is a complete name, belonging to the order Anaerolineae.*

Revised: "unclassified_Vicinamibacterales, uncultured_gamma_proteobacterium, unclassified_Vicinamibacteraceae, and unclassified_A4b.

Point 41: L552: identified as what?

Response: *Many thanks for your valuable suggestion. This sentence has been revised as required by this reviewer.*

Revised: "a total of 1,103 metabolites were identified as organic acid, amino acid, heterocyclic compounds, and so on in vegetables vs control, with 89 of upregulation and 41 of downregulation".

Point 42: L526: there is not one organic acid, nor one nucleotide – including derivatives these seem like large heterogenous groups to talk about, is it meaningful to lump them?

Response: *Many thanks for your valuable suggestion. This presentation has been revised as required by this reviewer.*

Revised: "exhibited significant correlations with the metabolites including amino acid derivatives, benzene and substituted derivatives, bile acids, and so on".

Point 43: Fig 1. Please add a picture of the control. When were photos taken and the data, how many years into the study is this?

Response: *Many thanks for your valuable suggestion. Indeed, a picture of the control has been added as required by the reviewer.*

Point 44: Table 2. it is unclear what the values represent? Community composition quantified how, in what units? Is the post-hoc analysis done across the table, or can I only look across columns, or rows?

Response: *Many thanks for your valuable suggestion. The related information has been added as required by this reviewer.*

Table 2 shows the differences in bacterial community structure among the four treatments at phylum, class, order, family, and genus level, respectively. The data in the same column show the information of one specific treatment (for example, the data in second column show the types of phylum, class, order, family, and genus, respectively, in vegetable treatment). And the data in the same row show the difference of four treatments (for example, the data in second row show the types of phylum among vegetable, corn, peach, and the control treatment).

Point 45: Fig. 3. In b that's not only genus level, it's a mix of taxonomic levels. Please correct this.

Response: *Many thanks for your carefulness. This presentation has been corrected as suggested by this reviewer.*

Point 46: Fig 4. In b the rendering is way to small to make any sense of whats in the data.

Response: *Many thanks for valuable suggestion. The presentation of Fig.4 has been improved as suggested by this reviewer.*

Point 47: Fig. 5. OPLS-DA is an abbreviation for orthogonal partial least squares discriminant analysis, not what you call it. Is it the same technique or not? If so, please use the common term. How do you read that donut plot? Is it a relative abundance visualization or something else? If the former, can you switch to stacked barchart, its easier to see the pattern. If the latter, more guidance on how to read the result is needed.

Response: *Many thanks for your carefulness. Yes, OPLS-DA is an abbreviation for orthogonal partial least squares discriminant analysis, which has been revised as suggested by this reviewer. Furthermore, the donut plot in Figure 5d has been replaced by "histogram" based on the relative abundance.*

Point 48: Fig. 6. What is VIP here? The labels and plots are too small to read, Improve the readability.

Response: *Many thanks for valuable suggestion. VIP here means "Variable importance in projection", which has been improved by added the full name in this revised manuscript. Furthermore, Figure 6 has also been improved as required by this reviewer.*

Point 49: Fig. 7. Typically RDA stands for Redundancy analysis. I don't know what Redundancy discriminant analysis is, I don't think it exists, unless you mean Linear Discriminant Analysis? But the plot suggests classic RDA was used. Please fix for clarity. Please spell out the abbreviations for soil abiotic variables at first mention. What is SDM, please write out?

Response: *Many thanks for valuable suggestion. You are right, it should be "Redundancy analysis (RDA)". In addition, the presentation has been improved in this revised manuscript as suggested by this reviewer by spelling out the abbreviations for soil abiotic variables and SDM at first mention.*