The re- submitted manuscript titled "Drivers and assemblies of soil eukaryotic microbes among different revegetation types in a semi-arid mountain in China" is yet ready for publication.

See comments on authors responses below

## Responses:

All suggestions have been modified based on your suggestions

- 1. We have revised this text. (Line 42-45) ok
- 2. That sentence has been changed. (Line 73-74) ok
- 3. We have used the hitherto to replace the connector. (Line 81) ok
- 4.We have updated some new reports and replaced some older reports. e.g., Murase et al., 2015, Dong et al., 2014, Czechowski et al., 2016. ok
- 5. This sentence includes supplementary information for the sampling area. If you think it is not connected with the rest of the paragraph, we will delete the sentence. We have deleted the part of introduction. (Line 110-111)

we established study plots, which were the main component of the ecological systems classified as the forest, shrub, grass, and farmland soil habitat types in October 2017

Do you mean?

we established study plots, which represented were the main component of the ecological systems classified as the forest, shrub, grass, and farmland soil habitat types in October 2017

6. The purpose of this picture was to introduce the sampling tool. We apologize for the poor picture quality, and we have deleted the picture. The soil sample and root samples were collected at a soil depth of 5–10 cm. We have revised the sampling methods. (Line 113-117)

Soil samples were collected at a soil depth of 5–10 cm and were taken along the "S" model from five sampling points, then pooled together as one large sample for each plot (understood). After cleaning up the impurities, 200 g term organic matter in the shallow soil was also collected (this is still not clear, what is the depth for this sample? how do you collect organic matter from a soil?). Root samples in the vicinity of the soil samples were also collected with a soil auger (inner diameter = 4cm) at a depth of 5–10 cm.

- 7. The misspellings in Table S1 have been revised. ok
- 8. The reference format was modified to that of Schloss et al. (2009). (Line 160) ok
- 9. We have revised the text regarding OTU. (Line 190-191) ok
- 10. The number of decimals reported has been revised. (Table 2) ok
- 11. We have used the "furthermore" to replace the "however". (Line 272) ok
- 12. The data units have been revised to superscripts. (Table 1) ok
- 13. The Table 2 legend has been modified to "Geochemical characteristics of the soil samples and other sequence data information in the present study". (Table 2) ok
- 14. The purpose of this picture was to introduce the sampling tool. We apologize for the poor picture quality, and we have deleted the picture. The soil and root samples were collected at a soil depth of 5–10 cm. We have revised the sampling methods. (Line 113-117) ok
- 15. The Table S1 legend has been modified to "More detailed information about the PCR steps. Table S1) ok
- 16. Text regarding the indicator species analysis has been added. (Line 199-205)

Note: Using PCA (principal component analysis) to compress the element data of term organic matter layer, the comp.1 and comp.2 axes (Cumulative Proportion > 80%) are defined as TOM 1 (term organic matter layer) and TOM2. Do not understand the use of the word term

- 17. We have added precipitation data; annual precipitation varied from 440 860 mm. (Line 95-96) ok
- 18. We have revised the soil classification, and the most common soil was Ustalf (USDA classification).. (Line 98) ok
- 19. We have revised the sampling method, and the Figure 4 has been modified. (Line 113-117)

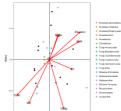


Figure 4.

See comment 6. Also, still not clear the difference between TOC and SOC

20. Soil water content =  $[(fresh soil quality) - (quality after drying)]/(fresh soil quality) <math>\times 100\%$ 

The water measurement method has been revised. (Line 126-128)

Thoroughly dried soil was used to calculate soil water content by comparing the quality difference between dry and fresh soil (This is called gravimetric water content and the way it is was calculated is incorrect. Please see a recognized soil science book, for example Nature and Properties of soil to get the correct calculation method). A soil auger were used to collect the term organic matter (five samples in a diagonal line for each plot), and the elemental analysis of term organic matter layer (is this referring to an organic horizon?) was measured by inductively coupled plasma mass spectrometry (Agilent Technologies, Palo Alto, CA, USA).

21.All "humus" have been deleted. We have used the term organic matter instead of humus in the paper. Meanwhile, we added the analysis of term organic matter. (Line 128-131)

This is not clear. I strongly recommend revising to better define the use of the organic matter. As I said before, are you referring to an O horizon or rather generalizing about a surface layer enriched in organic matter?

The elemental analysis of term organic matter layer was measured by ICPMS (Inductively Coupled Plasma Mass Spectrometry, Agilent Technologies, America). (As far as I understand ICP-MS is used to elemental analysis in liquid samples (unless an adaptation to solid samples is made). Then, I am not sure how soil organic matter was determined by this instrument) Using PCA (principal component analysis) to compress the element data of term organic matter layer, the comp.1 and comp.2 axes (Cumulative Proportion > 80%) were defined as TOM (term organic matter layer)1 and TOM2.

The results of PCA:

Comp.1 Comp.2

Standard deviation 2.7713530 1.4128615

Proportion of Variance 0.6400331 0.1663481

Cumulative Proportion 0.6400331 0.8063812

Comp.1 Comp.2

[1] 0.3952405 -1.8919874

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[2] -0.3597412 -2.5095802
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- [4] 3.0096398 0.9263883
- [5] 3.4258977 0.9270077
- [6] 3.6254524 0.8662455
- [7] 1.1249281 0.4899736
- [8] 0.7988064 0.6603870
- [9] 0.9342136 0.5245014
- [10] -4.4231814 1.6669533
- [11] -.0478505 1.0726758
- [12] -4.3822783 -0.1426623
- 22. The method for collecting the root samples has been modified. (Line 113-117)

## See previous comment on this

23.PE is Portable Executable. PE is the general name of the executable file under the system. (Line 161) ok

24. This sentence has been rewritten, and text regarding the indicator species analysis has been added. (Line 199-205)

The indicator species analysis revealed soil eukaryotic microbe characteristic (p < 0.05) were different among the soil habitat types; there were a total of 98 indicators were (revise) found in forest soil, rhizarian taxa (35 indicators) had a dominant position; 49 indicators were detected in farmland soil, metazoa taxa (19 indicators) comprised the main group; 74 indicators were found in shrub soil (25 indicators in metazoa taxa), and 48 indicators were observed in grass soil (35 indicators in rhizarian taxa) (Supplementary Table S4).

25. The network analysis information has been added. (Line 227-232)

Although the text added supports what is shown in the network analysis. This information can simply be obtained from an occurrence/abundance excel file. By deeply interpreting figure 3 there could be much more interesting information to pull out.

26. The main objective of this section was to compare the differences in soil eukaryotic microbes among different soil habitat types. We have revised this sentence and explained the results in more

<sup>[3] -0.1011271 -2.5899028</sup> 

detail; some simple descriptions have been deleted. (Line 246-251) ok

- 27. As your comment, some new descriptions have been added. (Line 313-318) ok
- 28. We have revised this sentence and added the meaning of the results. (Line 14-16) ok
- 29. Traditional studies, based for example on the estimates of cell volumes, ratios of protist account for fungal biomass as low as 10<sup>-3</sup>. However, a far higher ratio of protist to fungal sequences is obtained using high-throughput technology, so we added the comparison. If readers are confused here, we will delete the sentence. (Abstract) if you explain in the manuscript what you have described here than the information is understood and relevant to include in the manuscript
- 30. This sentence has been revised. (Line 26)

## See previous comment on this

31. All "humus" words have been deleted. We have used the term "organic matter" instead of "humus" in the paper.

## See previous comment on this

- 32. The "aquifer environment" has been added. We also revised the environmental comparisons and contrasts in the manuscript. (e.g., in aquifer environmental habitats, in agricultural habitats, in sandstone habitats, in semi-arid soil habitats). (Line 39) ok
- 33. We have added some new reports about the effects of organic fertilizers. (Line 42) ok
  Dong WY, Zhang XY, Dai XQ, Fu XL, Yang FT, Liu XY, Sun XM, Wen XF, and Schaeffer S. 2014.
  Changes in soil microbial community composition in response to fertilization of paddy soils in subtropical China. Applied Soil Ecology 84:140-147.

Murase J, Hida A, Ogawa K, Nonoyama T, Yoshikawa N, and Imai K. 2015. Impact of long-term fertilizer treatment on the microeukaryotic community structure of a rice field soil. Soil Biology & Biochemistry 80:237-243

34. This error has been corrected. (Line 45) ok

Cutler et al. (2013) shown the substrate characteristics may impact on the abundance/biomass of eukaryotic microbial communities and do not influence diversity in sandstone habitats.

- 35.We have added a hypothesis to the last paragraph of the Introduction and confirmed the hypothesis in the conclusion. (Line 85-86) ok
- 36. This sentence has been revised. (Line 246-249) ok
- 37. We have deleted the vague words and revised this sentence. (Line 246-251) ok

- 38. That was a comparison between protozoa and fungi, thus the descriptions were used to explain the reason why the percentage of protists was higher than fungi. (Line 267-271) understood
- 39. "Single OTUs" has been revised. We wanted to express that only a few OTUs existed in one habitat type. Thus, "single" has been deleted. (Line 278) ok
- 40. All "humus" have been deleted. We have used the "term organic matter" instead of "humus" in the paper and have revised the sampling method.

See previous comment on this