Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Effects of Revegetation on the composition and diversity of bacterial and fungal communities of sandification land soil, in Southern China.

Haozhi Long[1], Xiukun Wu[2], Yilin Wang[1], Junqing Yan[1], Xiaoyan Guo[1], Xuejiao An[1], Qinghua Zhang[1], Zhimin Li[1], and Guanghua Huo[1]

[1]Jiangxi Agricultural University
[2]Northwest Institute of Eco-Environment and Resources

October 31, 2020

Abstract

This study aimed to research the effects of forest (Pinus elliottii, slash pine) and shrub (Vitex trifolia) plantation on the soil microbial community in sandification land by using the Illumina Miseq sequencing of 16S rRNA and ITS rRNA genes and combined with the soil properties analysis to explore the driving factors. Finally, the results showed that the dominant bacterial phylum was Chloroflexi, Actinobacteria, Proteobacteria, and Acidobacteria, the shrub and forest plantation significantly increasing the proportion of Acidobacteria, while decreasing the proportion of Proteobacteria. For the fungal community, forest plantation was significantly changing the community structure at the phylum level that shifted from Ascomycota to Basidiomycota, and the ectomycorrhizal fungi take the most abundant with genus Rhizopogon predominant. The soil bacterial Chao1 and Shannon indices were significantly increased after revegetation, while the soil fungal Shannon diversity in the forest land that significantly correlated with soil total organic carbon and C/N was lower than that in the sandy and shrub land ($p<0.05$). Besides, the bacterial and fungal communities were significantly affected by soil water content among all analyzed soil properties. Our results suggest that the revegetation significantly increasing the soil bacterial diversity that correlated with soil water content, total organic carbon, and available phosphorus, but there was no significant change in community structure. In contrast, slash pine plantation changes the fungal community structure and diversity dramatically, and such changes should be attention because the shift of fungi community structure and diversity may lead to the decline of Pinaceae plantation.

Introduction

Land sandification is a kind of land degradation and it is a significant environmental problem across many countries (Liu *et al.* , 2012b, Zhang *et al.* , 2019, Huang *et al.* , 2020). In China, most degraded or desertification lands are distributed in the northern region, that is, the arid and semiarid regions (Chen *et al.* , 2004, Zhang *et al.* , 2019). While in southern China, including the Poyang Lake watershed which belongs to the humid area also have the sandification land (Zhu & Cui, 1996, Chen *et al.* , 2004), due to the joint influence of climate and human factors (Tang *et al.* , 2018).

Revegetation is the major effort to control and restore degraded ecosystems (Liu *et al.* , 2012b, Strickland *et al.* , 2017, Xue *et al.* , 2017, Zhang *et al.* , 2019). Especially the pine trees were wildly used in afforestation due to their good adaptation and environmental resistance (Xue *et al.* , 2017, Ning *et al.* , 2020, Zhao *et al.* , 2020). In northern China, *Pinus sylvestris* (Zhao *et al.* , 2020) and *Pinus tabulaeformis*(Liu *et al.* , 2019a, Liu *et al.* , 2019b, Wang *et al.* , 2019) were planted to control soil degradation or desertification. Likewise, since the 1980s, slash pine (*Pinus elliottii*) has been widely planted to control and restore sandy lands in the Poyang Lake watershed (Zou, 1990, Tang *et al.* , 2018). However, reforestation mainly relies on single-species monoculture, and pure forest plantation would cause soil quality decline and plantation

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

degradation in both degraded and non-degraded land (Liu *et al.* , 2012b, Wu *et al.* , 2015, Zhao *et al.* , 2020). Meanwhile, the shrub *Vitex trifolia* was also chosen to make effort to control fixing sandy dune due to its adaptation to sandy land and drought soil conditions (He, 1992). Moreover, the major studies including in this region focus on the aboveground plant community, and the belowground microbial community was ignored (Cao *et al.* , 2017, Chang *et al.* , 2017, Strickland*et al.* , 2017).

Soil microbial communities play an important role in terrestrial ecosystems, especially in degraded and desertified soil (Harris, 2009, Xue *et al.* , 2017, Zhang *et al.* , 2019). They are directly involved in plant nutrient acquisition and soil nutrient cycling. Virtually, the soil microbial community could be an indicator of ecosystem health and sustainability (Strickland *et al.* , 2017). For instance, the dynamics of fungal community structure and trophic mode may lead to the decline of forest plantation (Zhao *et al.* , 2020). To date, the research of soil microbial community in degradation land is mainly concentrated in northern China that is consistent with the distribution of desert and sandy land in China (Chen *et al.* , 2004), while the study of the microbial community in sandy land in southern China remains rare. Moreover, different Pinaceae have different rhizosphere microbial communities (Ning *et al.* , 2019), and even in the same region, the opposite pattern appears, that is, the shift of the proportion of Ascomycota and Basidiomycota (Liu *et al.* , 2019a, Wang *et al.* , 2019).

The Houtian area is located upstream of Poyang lake and has the typical subtropical moist monsoon climate with distinct dry and wet seasons. Before the 1990s, it was with heavy land sandification. Now, there are sandy, shrub, and forest land distributed in this area. In this study, we investigate the microbial community structure in the Houtian area by using Illumina Miseq sequencing of the 16S rRNA and ITS rRNA gene, and analysis the soil physicochemical properties. The objectives of this study were to explore: 1) the effect of revegetation on the microbial community composition in the sandification land of Poyang lake watershed at a fine-scale; 2) the factors driving the microbial community structure and diversity in sandification land in the humid area.

Materials and methods

Sampling site description

"Houtian desert" is a typical sandification land in the Houtian area and it is located about 28 kilometers at the southern of Nanchang which is the capital of Jiangxi Province, China. The annual rainfall is 1698.8 mm, and 68.7% of the rainfall occurs from March to June, the dry season is from July to September. Atmospheric temperature ranges from 1 to 39°C with a mean annual temperature of 17.7 °C. To understand the effect of revegetation on the microbial community of the degraded soil, soil samples were designed into three land types including sandy land soil, shrub (Vitex trifolia simplicifolia) land soil, and forest (slash pine, *Pinus elliottii*) land soil which were collected in July 29^{th}, 2017 (Fig. 1 and Table S1). The stand age of the slash pine plantation was more than 10 years based on archives of the local forest service and the google earth history map (Fig. S1).

The samples were collected from approximately 20 centimeters below the surface using the sterile shovels after removed the litter layer (Liu*et al.* , 2012a, Long *et al.* , 2017). Sandy soil was set with six quadrats, shrub soil and forest soil were set with five quadrats, respectively. Five parallel samples were collected from each quadrat then mixed. Altogether, 16 soil samples were collected. Subsamples for the soil chemical and physical analyses were placed into sealable plastic bags and stored at 4°C until further processed. Subsamples for the microbiological analysis were transferred into a sterile sampling bag and kept frozen on the ice during transport to the laboratory and analyzed then immediately.

Soil Physical and chemical properties analysis

Soil water content (WC), pH value, total organic carbon (TOC), and total nitrogen (TN) content were measured by the methods described previously (Liu *et al.* , 2012, Tai *et al.* , 2013). The available phosphorus (AP) and available potassium (AK) were measured after ammonium acetate extraction as described previously (Tai *et al.* , 2013). The C/N index was calculated via the TOC content division by TN content in

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

this study.

DNA extraction, sequencing and analysis of 16S and ITS rRNA genes

DNA of each soil sample was extracted by using the PowerSoil DNA isolation Kit (MoBio Inc., Carlsbad, CA, USA) according to the manufacturer's instructions. We used the primers 338F (5'-ACTCCTACGGGAGGCAGCA-3') and 806R (5'-GGACTACHVGGGTWTCTAAT -3') to amplify the V3-V4 region of the bacterial 16S rRNA gene and the primers ITS5F (5'-GGAAGTAAAAGTCGTAACAAGG-3') and ITS1R (5'-GCTGCGTTCTTCATCGATGC-3') to amplify the ITS1 region of the ITS rRNA gene. Purified amplicons were then run on the Illumina Miseq platform (Shanghai Personal Biotechnology Co., Ltd., China). The raw sequencing reads were quality trimmed and normalized to the same depth by using Mothur software (Schloss *et al.* , 2009). The sequences were clustered into the operational taxonomic units (OTUs) at 97% similarity by using the UCLUST tool of Quantitative Insights Into Microbial Ecology (QIIME) software (Edgar, 2010). The represented sequences for each OTU were taxonomically assigned according to the best matches in the Greengenes ribosomal database (Release 13.8,http://greengenes.secondgenome.com/, (DeSantis *et al.* , 2006)) for 16S rRNA and the UNITE database for ITS rRNA ((Release 5.0,https://unite.ut.ee/, (Koljalg *et al.* , 2013)). The Shannon diversity index and the Chao1 estimator were calculated using Mothur.

The raw data were submitted to the NCBI Sequence Read Archive (SRA) database and the SRA accession number for 16S and ITS sequences were PRJNA648283 and PRJNA648326, respectively.

Fungal functional groups

Fungal OTUs were assigned into functional guilds using the FUNGuild platform (http://www.stbates.org/guilds/app.php) (Nguyen *et al.* , 2016). The predicted results with "highly probable" or "probable" assignments were accepted for further analysis, and the fungi identified with combined trophic mode and combined guild were integrated as described previously (Zhao *et al.* , 2020).

Statistical analysis

All of the statistical analyses were performed by using R software (version 3.5.3), CANOCO program (version 5.0), and SPSS software (version 16.0, SPSS Inc., Chicago, IL, USA). One-way analysis of variance (ANOVA) was used to assess the significance of effects of revegetation on sandy soil properties, microbial community structure, and diversity by using SPSS. Venn diagram was generated by using OTUs among the three land types. The Redundancy analysis (RDA) was performed to elucidate the relationships between the soil properties and microbial communities using the CANOCO program (Tai *et al.* , 2013, Wang*et al.* , 2019). The data of bacterial and fungal communities were analyzed before the RDA assay using the detrended correspondence analysis (DCA) to confirm that the gradient lengths fit a linear model. Spearman's rank correlation analysis was used to determine the correlations between microbial diversity, richness, and soil properties by using SPSS (Zhao *et al.* , 2020).

Results

Soil characteristics and Alpha diversity indexes of microbial communities

The characteristics of 16 soil samples were analyzed concerning WC, pH, TOC, TN, AP, and AK (Table 1). The water content of the investigated samples ranged from 5.2 g/kg to 34.4 g/kg. Unexpectedly, the WC in sandy land samples significantly higher than shrub land and forest land. The soil pH value in all samples was below 7.00, the revegetation increased the pH value, but not significantly. The soil TN value ranged from 0.155 g/kg to 0.676 g/kg and the growth of vegetation increase the TN content. Likewise, the TOC of shrub land and forest samples were higher than sandy land. The C/N was highest in forest samples. Furthermore, The AP content in shrub land and forest samples were higher than sandy land. Although the AK have differences among each land type, the ANOVA analysis showed that there were no significantly different from one another.

With the growth of plants, the Chao1 index and Shannon index of soil bacteria are all significantly increased compared with the sandy land ($p<0.05$) (Table 2). While the soil fungal alpha diversity showed a different

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

trend. The fungal Chao1 index of shrub land samples significantly higher than sandy land samples whereas forest samples were lower than sandy land samples. The growth of shrub did not significantly change the fungal Shannon indices compared with sandy land samples. Yet, the forest significantly decreased the Shannon index compared with the sandy land samples ($p<0.05$) (Table 2).

The Microbial community composition and function in each land type

The taxa from the fungal community were fell into six phyla with Ascomycota and Basidiomycota dominant (Fig. 2). Ascomycota was the most dominant phylum in sandy land and shrub land samples (76.09 and 74.06%, respectively), while Basidiomycota was the most dominant in forest samples (62.92%). Zygomycota in shrub land samples (10.97%) was significantly higher than sandy land and forest land samples (2.17 and 3.27%, respectively). Chytridiomycota in sandy soil (1.51%) was significantly higher than shrub land and pine plantation land soil (0.50 and 0.10%, respectively), although it takes a small proportion in each land type samples. The other two phyla have no significant difference between land types.

The taxa from the bacterial community were fell into thirty-two phyla, and Chloroflexi, Actinobacteria, Proteobacteria, and Acidobacteria with the dominant. Chloroflexi was the most dominant phylum in this region (Fig. 2B1, Fig. S3), and there was no significant difference between the three land types. Also, the Actinobacteria phylum has no difference among each land type. The phylum that their relative abundance has significant differences between land types including Proteobacteria, Acidobacteria, Planctomycetes, Nitrospirae, Bacteroidetes, Verrucomicrobia, Deinococcus-Thermus, Fusobacteria, and WS2 phylum. Proteobacteria phylum in sandy land samples (25.79%) was significantly higher than shrub land and forest land samples (13.22% and 15.72%, respectively). While Acidobacteria in sandy land samples (7.45%) was significantly lower than shrub land and forest samples (15.55% and 13.75%, respectively). Planctomycetes and Nitrospirae phylum showed the highest relative abundance in the shrub land samples (5.49% and 1.03%, respectively), intermediate in the forest samples (3.11% and 0.89%, respectively), and lowest in the sandy land samples (1.76% and 0.37%, respectively). Bacteroidetes showed that the highest relative abundance in the sandy land (1.44%), intermediate abundance in the forest (0.40%), and lowest abundance in the shrub land (0.18%). Verrucomicrobia phylum in the forest (0.34%) and shrub land samples (0.18%) were higher than sandy land (0.01%). Deinococcus-Thermus and Fusobacteria phylum was significantly higher in sandy land (0.05% and 0.03%, respectively). On the contrary, WS2 phylum was significantly higher in forest samples (0.01%), although take a small proportion.

The Venn diagram showed that OTUs of fungi and bacteria differed among the three land types (Fig. 2, A2, and B2). The number of specific fungal OTUs was 453 in sandy land, 321 in shrub land, and 188 in forest land, and the number of specific bacteria OTUs was 469 in sandy land, 497 in shrub land, and 414 in the forest. Besides, the OTUs of fungi and bacteria that shared among all three land types were 151 and 1318, respectively.

Functional groups of soil fungal communities

In sandy land, the relative abundance of Pathotroph, Saprotroph, Symbiotroph were 27.45%, 45.89%, and 12.22%, respectively (Fig. 3). Compared with the sandy land, the relative abundance of each trophic mode in shrub land did not change significantly (Fig. 3). Notably, with the plantation of slash pine, the relative abundance of the Pathotroph and Saprotroph significantly decreased while the Symbiotroph significantly increased with Ectomycorrhizal take the most abundant (Fig.3, Table S2).

The impact of soil properties on microbial community structure and diversity.

Redundancy analysis (RDA) was used to determine the relationships among seven soil property factors and microbial community structure (Fig. 4). The results showed the soil WC significantly driving both fungal ($F=2.5$, $p=0.001$) and bacterial ($F=3.8$, $p=0.001$) community. Besides, TOC also showed a strong effect on the bacterial community ($F=1.9$, $p=0.026$) (Table S3). For the fungi community, all the seven soil properties explained 52.3% of the variance (Table S3), with axis 1 explaining 16.98% of the variance and axis 2 explaining 11.73% (Fig. 4A). For the bacteria community, all the seven soil properties explained 58.8% of

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

the variance (Table S3), with axis 1 explaining 23.38% of the variance and axis 2 explaining 13.62% (Fig. 4B).

The results of Spearman's correlation analysis showed in Table 3 that the Fungal Chao1 index was significantly positively correlated with the AK. The fungal Shannon diversity index TOC and C/N were significantly negatively correlated with. The bacterial Chao1 index was a negative correlation with WC and a positive correlation with AP at a significant level ($p<0.05$). The bacterial Shannon diversity index was significantly negatively correlated with WC and significantly positively correlated with TOC and AP.

Discussion

In this study, shrub and forest plantation did not change the dominant phyla of the soil bacterial community (Fig. 2). Compared with the sandy land, the relative abundance of Proteobacteria decreased in shrub and forest land although the content of TOC, TN, and AP increased after revegetation (Table 1). This result conflict with a recent report that the relative abundance of Proteobacteria decreased after afforestation but the nutrient factors were decreased (Wang *et al.* , 2019). Yet, the drought could provoke changes in plant metabolism and suppress bacterial growth, and with a greater negative impact on didems including Proteobacteria (Xu & Coleman-Derr, 2019). Hence, the soil water content in this study may the major factor lead to Proteobacteria abundance decreased (Fig. 4). However, phylum Acidobacteria which preferring nutrient-poor environments (Wang *et al.* , 2019) also belong to didems while it was increased in shrub and forest land in this study (Fig. 2). This suggests that the soil in the study area is still depauperate although the soil nutrients increased after the revegetation.

Conspicuously, the proportion of Chloroflexi in this study (more than 25%) and the global surveys (4.3%) was not commensurate (Islam*et al.* , 2019). But it was also more than 14% in the same area of wetland soil (Chen *et al.* , 2019). Furthermore, recently, (Xu & Coleman-Derr, 2019) reviewed that drought leads to enrichment of monoderm bacteria (including Chloroflexi) within the roots of many plant species across many environments. Likewise, the soil water content relatively low in this study (Table 1). Besides, during the sampling period, the air temperature was above 38degC in the daytime and the sandy soil temperature was higher than 50degC(data, not shown). Coincidently, Chloroflexi were abundant in hot springs (60–22.1degC) in Qinghai-Tibet plateau (Neufeld, et al., 2013), and various strains from the Chlorolfexi have been isolated and characterized form hot springs and geothermal soils (Islam *et al.* , 2019). Virtually, Chloroflexi could survive in a poly extreme environment and as major components (Wierzchos *et al.* , 2018). Furthermore, bacteria from the phylum Chloroflexi with metabolic diversity and adapted to both oxic and anoxic environments that are widespread and abundant in free-living microbial communities (Islam *et al.* , 2019). Thus, high temperatures, low soil water content, and poor nutrient condition may lead to the enrichment of Chloroflexi in this study.

The response of the fungal community to Slash pine plantation in the current study was stronger than the bacterial community validates results reported previously that the fungal communities were more sensitive to the afforestation than bacterial communities (Gunina*et al.* , 2017, Xue *et al.* , 2017, Zhong *et al.* , 2018, Wang *et al.* , 2019). Furthermore, our results revealed that the effect of forest plantation much stronger than the shrub. The fungal community composition in sandy and shrub land was similar to the wetland samples in the same area (Chen *et al.* , 2019), and that in the loess soil zone (Liu *et al.* , 2019a). The dominant fungal phylum changed from Ascomycota to Basidiomycota after slash pine plantation (Fig 2. A1). In contrast, in 78-year-old tree plantations in Australia, slash pine significantly increased the dominant taxa of Ascomycota (Zhou*et al.* , 2017). This may due to the restored soil available nutrient after long-term growth (Zhou *et al.* , 2017, Wang *et al.* , 2019), and the transition with the increasing stand age (Liu*et al.* , 2019b, Zhao *et al.* , 2020). Also, with the stand age increasing, the proportion and quantity of each functional group will change and may lead to the decline of the pure plantation (Wu*et al.* , 2015, Zhao *et al.* , 2020). Thus, we speculate the conflict fungal community pattern in the same region (Liu *et al.* , 2019a, Wang *et al.* , 2019) may due to the different stand age of pine plantation (Liu *et al.* , 2019a, Wang *et al.* , 2019). Follow studies are needed to verify this hypothesis in different stand age of Slash pine plantation.

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Pinaceae is not only the well-known EM fungi plant taxon (Liu *et al.* , 2019b, Ning *et al.* , 2019, Wang *et al.* , 2019, Zhao*et al.* , 2020), but also the most important factor determining the C/N ratio higher than other vegetations (Xue *et al.* , 2017) (Table 1). Although the C/N ratio in this study was much lower than previous studies in slash pine plantation soils (Wu *et al.* , 2015, Zhou*et al.* , 2017) and other Pinaceae plantation soils (Liu *et al.* , 2019b, Wang *et al.* , 2019), it was significantly negatively correlated with fungal Shannon diversity (Table 3). The fungal diversity was significantly decreased after slash pine plantation in this study (Table 2), this could be explained by the increased C/N ratio and another reason that monodominance of woody vegetation driving the lower fungal diversity in EM-dominated habitats (Bahram *et al.* , 2020). Indeed, with the slash pine plantation, *Rhizopogon* , a common EM fungus in temperate coniferous forests (Ning *et al.* , 2019, Zhao*et al.* , 2020), which can improve survival and drought tolerance of conifer seedlings (Zhao *et al.* , 2020) showed the dominant (Fig. S2), and the relative abundance of Pathotroph was inhibited (Fig. 3).

Besides, the bacterial diversity was significantly correlated with WC, TOC, and AP (Table 3), which partially coincident with the previous study in the sediments of Poyang Lake region that the bacterial community compositions were driven by total phosphorus and inorganic phosphorus (Sheng *et al.* , 2016). Although the microbial community in plantation soil was influenced by many factors, including soil C, N, P, K content and C/N ratio (Wu *et al.* , 2015, Liu *et al.* , 2019a, Liu *et al.* , 2019b, Wang *et al.* , 2019), in this study, the RDA analysis showed that the bacterial and fungal communities were all driving by WC. This may due to the relatively low WC and soil nutrients. Of course, the sampling time point and the stand age of slash pine in this study are single, thus, we still need to verify this hypothesis in the future work.

Conclusions

In this study, the results showed that the dominant soil bacterial phylum was Chloroflexi, Actinobacteria, Proteobacteria, and Acidobacteria. Compared with the sandy land, the proportion of Proteobacteria in the shrub and forest land decreased while Acidobacteria increased. For the fungal community, Slash pine plantation not only significantly changed the community structure at the phylum level that the dominant phylum reversed from Ascomycota to Basidiomycota, but also significantly decreased the Shannon diversity that significantly correlated with the C/N ratio. Besides, slash pine plantation had increased the proportion of ECM fungi with the*Rhizopogon* predominant. Combined with other studies previously, the shift of the fungal community could lead to degradation of the plantation. Moreover, the RDA analysis results showed that the soil bacterial and fungal communities were significantly affected by WC. Thus, the shift of soil microbial community of slash pine plantation in different seasons and different stand ages needed to investigate in the following research.

Acknowledgments

This study was supported by the Foundation of Key Laboratory of Extreme Environmental Microbial Resources and Engineering, Gansu Province (EEMRE201803), the Science and Technology Project of Jiangxi Education Department (GJJ150402), and the Jiangxi Provincial Natural Science Foundation (20192BAB204011 and 20202BABL213041).

References

Bahram, M., Netherway, T., Hildebrand, F., Pritsch, K., Drenkhan, R., Loit, K., Anslan, S., Bork, P. & Tedersoo, L. (2020). Plant nutrient-acquisition strategies drive topsoil microbiome structure and function. *New Phytologist* , doi: 10.1111/nph.16598.

Cao, Y., Lu, Y., Zhu, Y., Yang, J., Wang, X. & Li, S. (2017). Experimental Study and Demonstration of Pinus elliottii on Poyang Lake Desertification Area. *Ecology and Environmental Sciences* , *26* (5), 741-746. doi: 10.16258/j.cnki.1674-5906.2017.05.003

Chang, L., Cai, J., Wu, Q., Jin, Q., Zhou, H. & Hu, Q. (2017). Leaf Traits of Vitex rotundifolia along Desertification Gradient in a Sandy Hill of the Poyang Lake.*Journal of Desert Research* , *37* (1), 81-85. doi: 10.7522/j. issn. 1000-694X. 2015. 00195.

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Chen, H., Su, Z., Yang, P. & Dong, G. (2004). Preliminary reconstruction of the desert and sandy land distributions in China since the last interglacial period. *Science in China Series D* , *47* , 89-100. doi: 10.1360/04zd0010.

Chen, M., He, S., Li, J., Hu, W., Ma, Y., Wu, L. & Gang, G. (2019). Co-occurrence patterns between bacterial and fungal communities in response to a vegetation gradient in a freshwater wetland. *Canadian journal of microbiology* , *65* , 722-737. doi: 10.1139/cjm-2019-0147.

DeSantis, T. Z., Hugenholtz, P., Larsen, N., Rojas, M., Brodie, E. L., Keller, K., Huber, T., Dalevi, D., Hu, P. & Andersen, G. L. (2006). Greengenes, a chimera-checked 16S rRNA gene database and workbench compatible with ARB. *Appl Environ Microbiol* , *72* , 5069-5072. doi: 10.1128/AEM.03006-05

Edgar, R. C. (2010). Search and clustering orders of magnitude faster than BLAST. *Bioinformatics* ,*26* , 2460-2461. doi: 10.1093/bioinformatics/btq461.

Gunina, A., Smith, A. R., Godbold, D. L., Jones, D. L., Kuzyakov, Y. J. (2017). Response of soil microbial community to afforestation with pure and mixed species. *Plant and Soil* , *412* , 357-368. doi: 10.1007/s11104-016-3073-0.

Harris, J. (2009). Soil microbial communities and restoration ecology: facilitators or followers?*Science* , *325* , 573-574. doi: 10.1126/science.1172975.

He X (1992) Community Features of Vitex trifolia var. simplicifolia and Its Adaptation to Sandy Land in Houtian Area. *Chinese Journal of Ecology* , *4* , 36-40. http://en.cnki.com.cn/Article_en/CJFDTotal-STXZ199204008.htm.

Huang, J. , Zhang, G. , Zhang, Y. , Guan, X. , Wei, Y. , Guo, R. (2020). Global desertification vulnerability to climate change and human activities. *Land Degradation & Development* , *31* , 1380-1391. doi: 10.1002/ldr.3556.

Islam, Z. F., Cordero, P. R. F., Feng, J., Chen, Y., Bay, S. K., Jirapanjawat, T., Gleadow, R. M., Carere, C. R., Stott, M. B., Chiri, E., Greening, C. (2019). Two Chloroflexi classes independently evolved the ability to persist on atmospheric hydrogen and carbon monoxide. *ISME J* , *13* , 1801-1813. doi: 10.1038/s41396-019-0393-0.

Koljalg, U., Nilsson, R. H., Abarenkov, K., Tedersoo, L., Taylor, A. F. S., Bahram, M., Bates, S. T., Bruns, T. D., Bengtsson-Palme, J., Callaghan, T. M., Douglas, B., Drenkhan, T., Eberhardt, U., Duenas, M., Grebenc, T., Griffith, G. W., Hartmann, M., Kirk, P. M., Kohout, P., Larsson, E., Lindahl, B. D., Lucking, R., Martin, M. P., Matheny, B., Nguyen, N. H., Niskanen, T., Oja, J., Peay, K., Peintner, U., Peterson, M., Poldmaa, K., Saag, L., Saar, I., Schubler, A., Scott, J. A., Senes, C., Smith, M. E., Suija, A., Taylor, D. L., Telleria, M. T., Weiss, M., & Larsson, K-H. (2013). Towards a unified paradigm for sequence-based identification of fungi.*Molecular Ecology* , *22* , 5271-5277. doi: 10.1111/mec.12481.

Liu, D., Wang, H., An, S., Bhople, P. & Davlatbekov, F. (2019a). Geographic distance and soil microbial biomass carbon drive biogeographical distribution of fungal communities in Chinese Loess Plateau soils. *Sci Total Environ* , *660* , 1058-1069. doi: 10.1016/j.scitotenv.2019.01.097.

Liu, G., Hu, P., Zhang, W., Wu, X., Yang, X., Chen, T., Zhang, M. & Li, S. (2012a). Variations in soil culturable bacteria communities and biochemical characteristics in the Dongkemadi glacier forefield along a chronosequence. *Folia Microbiol (Praha)* , *57* , 485-494. doi: 10.1007/s12223-012-0159-9.

Liu, G., Chen, L., Shi, X., Yuan, Z., Yuan, L., Lock, T. R. & Kallenbach, R. L. (2019b). Changes in rhizosphere bacterial and fungal community composition with vegetation restoration in planted forests. *Land Degradation & Development* ,*30* , 1147-1157. doi: 10.1002/ldr.3275.

Liu, Y., Wei, X., Guo, X., Niu, D., Zhang, J., Gong, X. & Jiang, Y. (2012b). The long-term effects of reforestation on soil microbial biomass carbon in sub-tropic severe red soil degradation areas. *Forest Ecology and Management* ,*285* , 77-84. doi: 10.1016/j.foreco.2012.08.019.

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Long, H., Wang, Y., Chang, S., Liu, G., Chen, T., Huo, G., Zhang, W., Wu, X., Tai, X., Sun, L. & Zhang, B. (2017). Diversity of crude oil-degrading bacteria and alkane hydroxylase (alkB) genes from the Qinghai-Tibet Plateau. *Environ Monit Assess* , *189* , 116. doi: 10.1007/s10661-017-5798-5.

Nguyen, N. H., Song, Z., Bates, S. T., Branco, S., Tedersoo, L., Menke, J., Schilling, J. S. & Kennedy, P. G. (2016). FUNGuild: An open annotation tool for parsing fungal community datasets by ecological guild. *Fungal Ecology ,20* , 241-248. doi: 10.1016/j.funeco.2015.06.006.

Ning, C., Xiang, W., Mueller, G. M., Egerton-Warburton, L. M., Yan, W. & Liu, S. (2019). Differences in ectomycorrhizal community assembly between native and exotic pines are reflected in their enzymatic functional capacities. *Plant and Soil* , *446* , 179-193. doi: 10.1007/s11104-019-04355-9.

Ning, C., Egerton-Warburton, L. M., Mueller, G. M., Xiang, W., Yan, W. & Liu, S. (2020). Shifts in ectomycorrhizal fungal community composition during the early establishment of native and exotic pine seedlings. *Applied Soil Ecology* , *157* , 103722. doi: 10.1016/j.apsoil.2020.103722.

Schloss, P .D., Westcott, S. L., Ryabin, T., Hall, J. R., Hartmann, M., Hollister, E. B., Lesniewski, R. A., Oakley, B. B., Parks, D. H., Robinson, C. J., Sahl, J. W., Stres, B., Thallinger, G. G., Van Horn, D. J., & Weber, C. F. (2009). Introducing mothur: open-source, platform-independent, community-supported software for describing and comparing microbial communities. *Appl Environ Microbiol* , *75* , 7537-7541. doi: 10.1128/AEM.01541-09.

Sheng, P., Yu, Y., Zhang, G., Huang, J., He, L., & Ding, J. (2016). Bacterial diversity and distribution in seven different estuarine sediments of Poyang Lake, China. *Environmental Earth Sciences* ,*75* (6), 479. doi: 10.1007/s12665-016-5346-6.

Strickland, M. S., Callaham, M. A., Gardiner, E. S., Stanturf, J. A., Leff, J. W., Fierer, N., & Bradford, M. A. (2017). Response of soil microbial community composition and function to a bottomland forest restoration intensity gradient.*Applied Soil Ecology, 119* , 317-326. doi: 10.1016/j.apsoil.2017.07.008.

Tai, X. S., Mao, W. L., Liu, G. X., Chen, T., Zhang, W., Wu, X. K., Long, H. Z., Zhang, B. G. & Zhang, Y. (2013). High diversity of nitrogen-fixing bacteria in the upper reaches of the Heihe River, northwestern China. *Biogeosciences, 10* (8), 5589-5600. doi: 10.5194/bg-10-5589-2013.

Tang, L.-L., Cai, X.-B., Gong, W.-S., Lu, J.-Z., Chen, X.-L., Lei, Q., & Yu, G.-L. (2018). Increased Vegetation Greenness Aggravates Water Conflicts during Lasting and Intensifying Drought in the Poyang Lake Watershed, China. *Forests, 9* (1). doi: 10.3390/f9010024.

Wang, K., Zhang, Y., Tang, Z., Shangguan, Z., Chang, F., Jia, F, Chen, Y., He, X., Shi, W. & Deng, L. (2019). Effects of grassland afforestation on structure and function of soil bacterial and fungal communities. *Science of the Total Environment, 676* . doi: 10.1016/j.scitotenv.2019.04.259.

Wierzchos, J., Casero, M. C., Artieda, O., & Ascaso, C. (2018). Endolithic microbial habitats as refuges for life in polyextreme environment of the Atacama Desert.*Current Opinion in Microbiology, 43* , 124-131. doi: 10.1016/j.mib.2018.01.003.

Wu, Z., Haack, S. E., Lin, W., Li, B., Wu, L., Fang, C., & Zhang, Z. (2015). Soil Microbial Community Structure and Metabolic Activity of Pinus elliottii Plantations across Different Stand Ages in a Subtropical Area. *PLoS One, 10* (8), e0135354. doi: 10.1371/journal.pone.0135354.

Xu, L., & Coleman-Derr, D. (2019). Causes and consequences of a conserved bacterial root microbiome response to drought stress. *Current Opinion in Microbiology, 49* , 1-6. doi: 10.1016/j.mib.2019.07.003.

Xue, L., Ren, H., Li, S., Leng, X., & Yao, X. (2017). Soil Bacterial Community Structure and Co-occurrence Pattern during Vegetation Restoration in Karst Rocky Desertification Area. *Frontiers in Microbiology, 8* . doi: 10.3389/fmicb.2017.02377.

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Zhang, Y., Cao, C., Cui, Z., Qian, W., Liang, C., & Wang, C. (2019). Soil bacterial community restoration along a chronosequence of sand-fixing plantations on moving sand dunes in the Horqin sandy land in northeast China. *Journal of Arid Environments, 165* , 81-87. doi: 10.1016/j.jaridenv.2019.04.003.

Zhao, P. S., Guo, M. S., Gao, G. L., Zhang, Y., Ding, G. D., Ren, Y., & Akhtar, M. (2020). Community structure and functional group of root-associated Fungi of Pinus sylvestris var. mongolica across stand ages in the Mu Us Desert.*Ecol Evol, 10* (6), 3032-3042. doi: 10.1002/ece3.6119.

Zhong, Y., Yan, W., Wang, R., Wang, W., & Shangguan, Z. (2018). Decreased occurrence of carbon cycle functions in microbial communities along with long-term secondary succession. *Soil Biology and Biochemistry, 123* , 207-217. doi: 10.1016/j.soilbio.2018.05.017.

Zhou, X., Guo, Z., Chen, C., & Jia, Z. (2017). Soil microbial community structure and diversity are largely influenced by soil pH and nutrient quality in 78-year-old tree plantations. *Biogeosciences, 14* (8), 2101-2111. doi: 10.5194/bg-14-2101-2017.

Zhu, Z., & Cui, S. (1996). The problem of desertification in south China. *Journal of Desert Research, 16* (4), 331-337. http://www.desert.ac.cn/CN/Y1996/V16/I4/331.

Zou, X. (1990). A Study on Eolian Sand in Humid Subtropical Zone in China-Taking Houtian Area in Xinjian County, Jiangxi Province as An Example. *Journal of Desert Research, 10* (2), 43-53. http://www.desert.ac.cn/CN/Y1990/V10/I2/43.

Tables

Table 1. Soil physicochemical properties of each land type

Land type	Physicochemical factor	Physicochemical factor	Physicochemical factor	Physicochemical factor	Physicoc
	WC (g/kg)	pH value	TOC (g/kg)	TN (g/kg)	AP (mg/
Sandy	26.27±2.22a	5.36±0.15	0.461±0.027b	0.226±0.024	2.843±0.
Shrub	7.64±1.48b	5.65±0.30	1.173±0.392a	0.370±0.078	5.730±0.
Forest	12.40±1.62b	5.51±0.21	0.972±0.184ab	0.270±0.049	4.123±0.

The soil properties of each land type were given as means±SE. Different minuscule letters in the column indicate the values that are not significantly different from one another (ANOVA,p <0.05).

Abbreviations: WC, soil water content; TOC, total organic carbon; TN, total nitrogen; AP, available phosphorus; AK, available potassium.

Table 2. Alpha diversity indexes of the microbial community in different groups.

Land type	Fungi	Fungi	Bacteria	Bacteria
	Chao1	Shannon	Chao1	Shannon
Sandy land	226.52±47.34	5.50±0.52a	991.59±51.42b	6.03±0.19b
Shrub land	279.53±18.91	5.31±0.25a	1527.34±86.51a	8.01±0.37a
Forest land	191.63±10.21	3.28±0.21b	1558.51±89.88a	7.48±0.17a

Indexes' values are means±SE. Different minuscule letters in the column indicate the values that are significantly different from one another (ANOVA, p <0.05).

Table 3. Spearman's rank correlation analysis for microbial diversity, richness and soil properties

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

	Fungi	Fungi	Fungi	Fungi	Bacteria	Bacteria	Bacteria	Bacteria
	Chao1 Coefficients	Chao1 p	Shannon Coefficients	Shannon p	Chao1 Coefficients	Chao1 p	Shannon Coefficients	Shannon p
WC	-0.124	0.694	0.238	0.374	**-0.562***	**0.024**	**-0.774****	**0.000**
pH	0.224	0.405	0.238	0.374	0.240	0.371	0.096	0.725
TOC	-0.026	0.922	**-0.526***	**0.036**	0.347	0.188	**0.509***	**0.044**
TN	0.478	0.061	-0.001	0.996	0.172	0.532	0.380	0.146
AP	0.144	0.594	-0.212	0.431	**0.515***	**0.041**	**0.568***	**0.022**
AK	**0.563***	**0.023**	0.224	0.404	-0.336	0.203	-0.197	0.463
C/N	-0.341	0.196	**-0.659****	**0.006**	0.024	0.931	-0.338	0.200

Note: ** p<0.01, * p<0.05.

Abbreviations: WC, soil water content; TOC, total organic carbon; TN, total nitrogen; AP, available phosphorus; AK, available potassium.

Figures

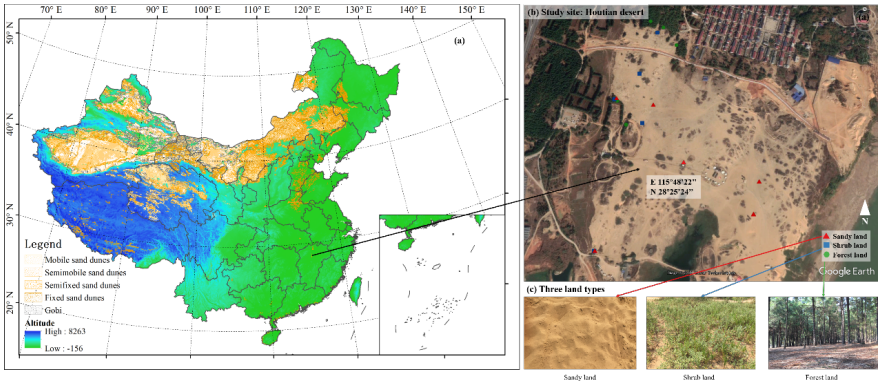

Fig. 1

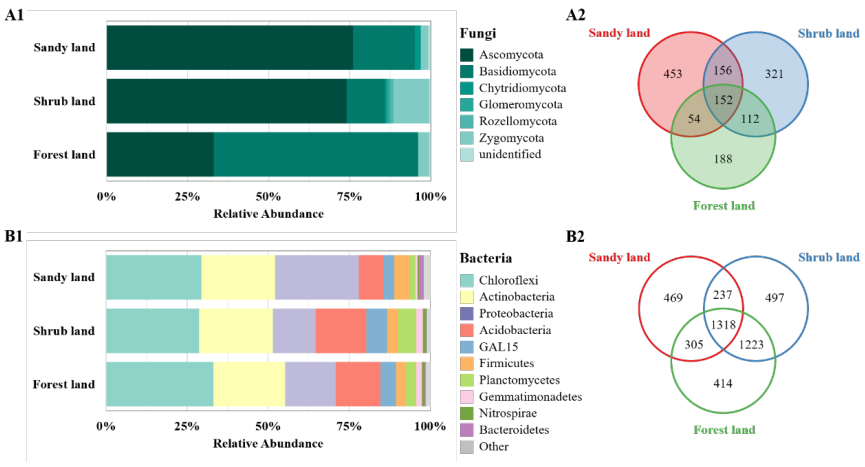

Fig. 2

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

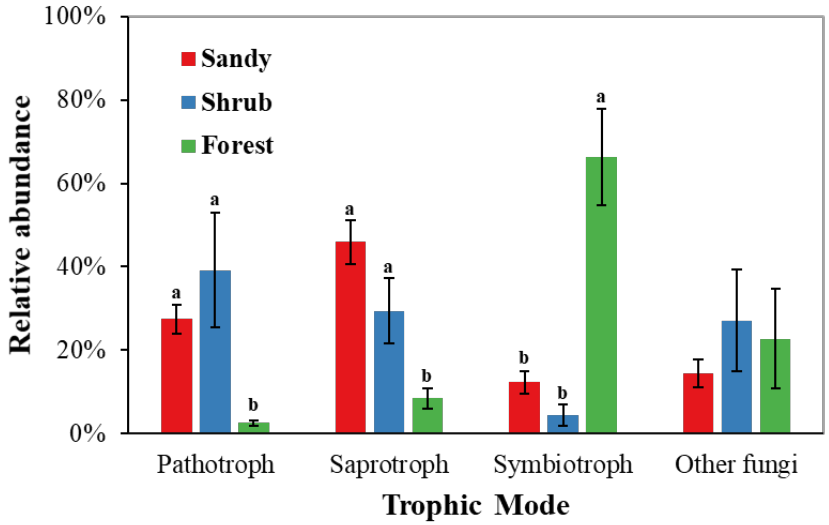

Fig. 3

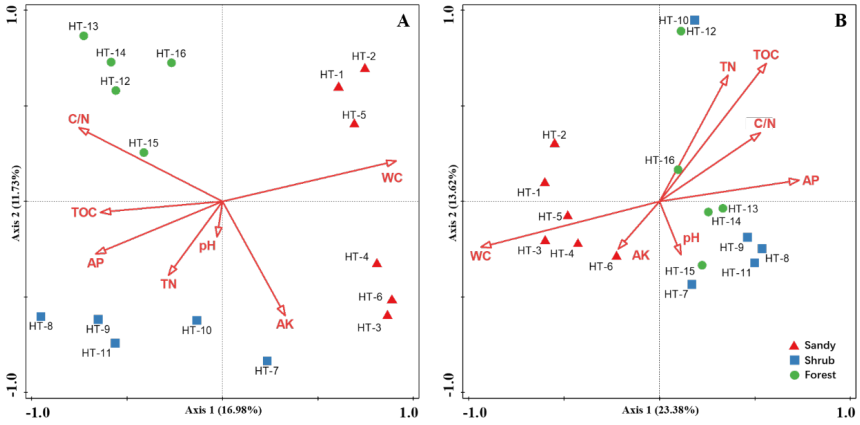

Fig. 4

Figure legends

Figure 1. Distribution of the modern desert and sandy land in China (a), study site and plot distribution (b), and three land types (c) in Houtian area.

Figure 2. The relative abundance of fungal (A1) and bacterial (B1) phyla in the three land types. Venn diagram of the exclusive and shared OTUs of fungal (A2) and bacterial (B2) found among the different land types.

Figure 3. The relative abundance of the fungal functional groups in the three land types.

Different minuscule letters indicate the values that are significantly different from one another (ANOVA, $p<0.05$).

Figure 4. Redundancy analysis to show correlation of the fungal (A) and bacterial communities (B) with soil physicochemical properties under three land types.

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Supplementary Materials

Table S1. The description of samples from the "Houtian Desert".

Sample ID	Longitude (E)	Latitude (N)	Dominant plant	Land type
HT-1	115°48'22.53"	28°25'24.77"	None	Sandy land
HT-2	115°48'29.49"	28°25'20.33"		
HT-3	115°48'30.54"	28°25'23.75"		
HT-4	115°48'10.64"	28°25'17.19"		
HT-5	115°48'17.29"	28°25'33.26"		
HT-6	115°48'12.26"	28°25'34.36"		
HT-7	115°48'10.56"	28°25'17.15"	*Vitex trifolia simplicifolia*	Shrub land
HT-8	115°48'15.89"	28°25'30.94"		
HT-9	115°48'17.75"	28°25'43.28"		
HT-10	115°48'11.96"	28°25'34.22"		
HT-11	115°48'15.31"	28°25'37.44"		
HT-12	115°48'13.74"	28°25'30.86"	*Pinus elliottii*	Forest land
HT-13	115°48'20.50"	28°25'41.24"		
HT-14	115°48'18.63"	28°25'43.98"		
HT-15	115°48'12.37"	28°25'34.00"		
HT-16	115°48'16.71"	28°25'45.10"		

Table S2. Functional groups and relative abundance of RAF in three land types.

Trophic mode	**Guild**	**Sandy land (%)**	**Shrub land (%)**	**Forest land (%)**
Symbiotroph	Arbuscular mycorrhizal	0.70	1.74	0.01
	Ectomycorrhizal	5.69	0.96	65.98
	Endophyte	2.15	1.34	0.16
	Lichenized	3.68	0.28	0.21
	Total	12.22	4.32	66.35
Saprotroph	Dung saprotroph	0.63	0.43	-
	Leaf saprotroph	-	-	-
	Soil saprotroph	1.70	6.95	0.22
	Wood saprotroph	3.75	3.20	1.30
	Undefined saprotroph	35.28	12.40	6.71
	Other saprotrophic fungi	4.53	6.40	0.19
	Total	45.89	29.38	8.42
Pathotroph	Animal pathogen	10.47	21.33	1.24
	Plant pathogen	12.63	17.17	0.40
	Other pathotrophic fungi	4.35	0.70	0.87
	Total	27.45	39.19	2.51
Other	Ericoid mycorrhizal	0.07	0.10	7.00
	Other fungi	14.37	27.00	15.72
	Total	14.44	27.10	22.72

Table S3. Forward selection results of the RDA analysis by using CANOCO 5.0

Fungal community (OTUs)	Fungal community (OTUs)	Fungal community (OTUs)	Fungal community
Explains %	Contribution %	pseudo-F	P

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

	Fungal community (OTUs)	Fungal community (OTUs)	Fungal community (OTUs)	Fungal community
WC	**15.2**	**29.1**	**2.5**	**0.001**
AK	8.1	15.6	1.4	0.09
CN	6.8	13.1	1.2	0.203
pH	6.4	12.3	1.1	0.293
TOC	4.9	9.4	0.8	0.659
TN	6.0	11.5	1.0	0.437
AP	4.7	9.0	0.8	0.671
	Bacterial community (OTUs) Explains %	Bacterial community (OTUs) Contribution %	Bacterial community (OTUs) pseudo-F	Bacterial communi P
WC	**21.5**	**36.6**	**3.8**	**0.001**
TOC	**10.1**	**17.1**	**1.9**	**0.026**
pH	5.2	8.8	1.0	0.448
AK	6.6	11.2	1.3	0.216
CN	5.0	8.6	1.0	0.454
TN	6.8	11.6	1.4	0.155
AP	3.6	6.0	0.7	0.775

Abbreviations: WC, soil water content; TOC, total organic carbon; TN, total nitrogen; AP, available phosphorus; AK, available potassium.

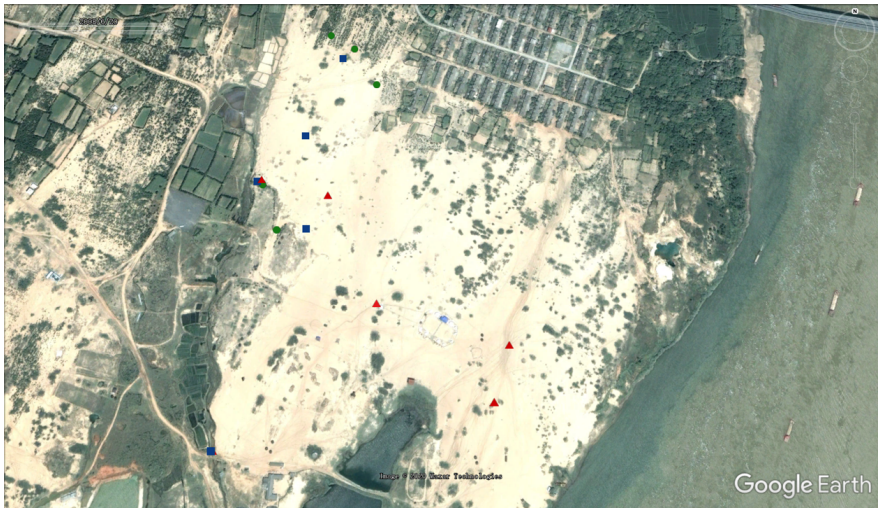

Figure S1. Google earth history map of sampling area in 2008. Red triangle means sandy land, Blue square means shrub land, and Green circle means forest land.

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

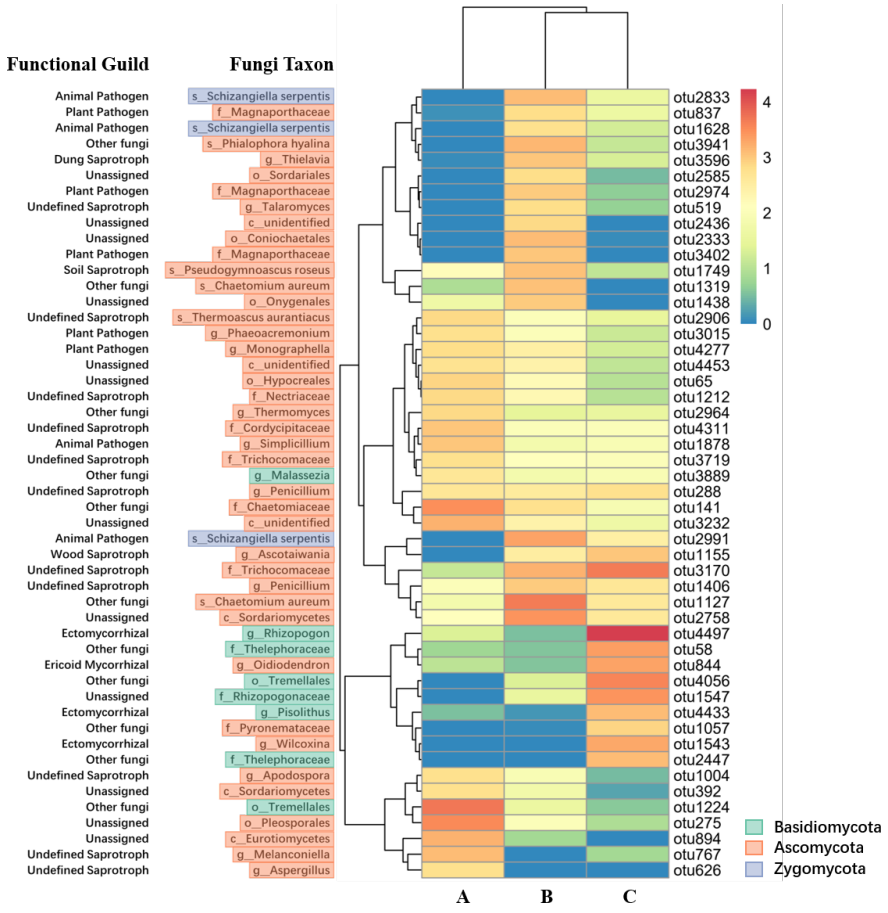

Figure S2. Heatmap displaying the abundance of top 50 OTUs of fungal across all the three land types after standardization: [p=log (n+1, 10), n means the average sequences number]. A, B, and C indicate sandy land, shrub land, and forest land, respectively. The fungi taxon and functional guild corresponding with each OTU are shown at the left and the letter before the taxon indicates the level of assignment (c=class, o=order, f=family, g=genus, s=species).

Posted on Authorea 31 Oct 2020 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.160411866.63653650/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

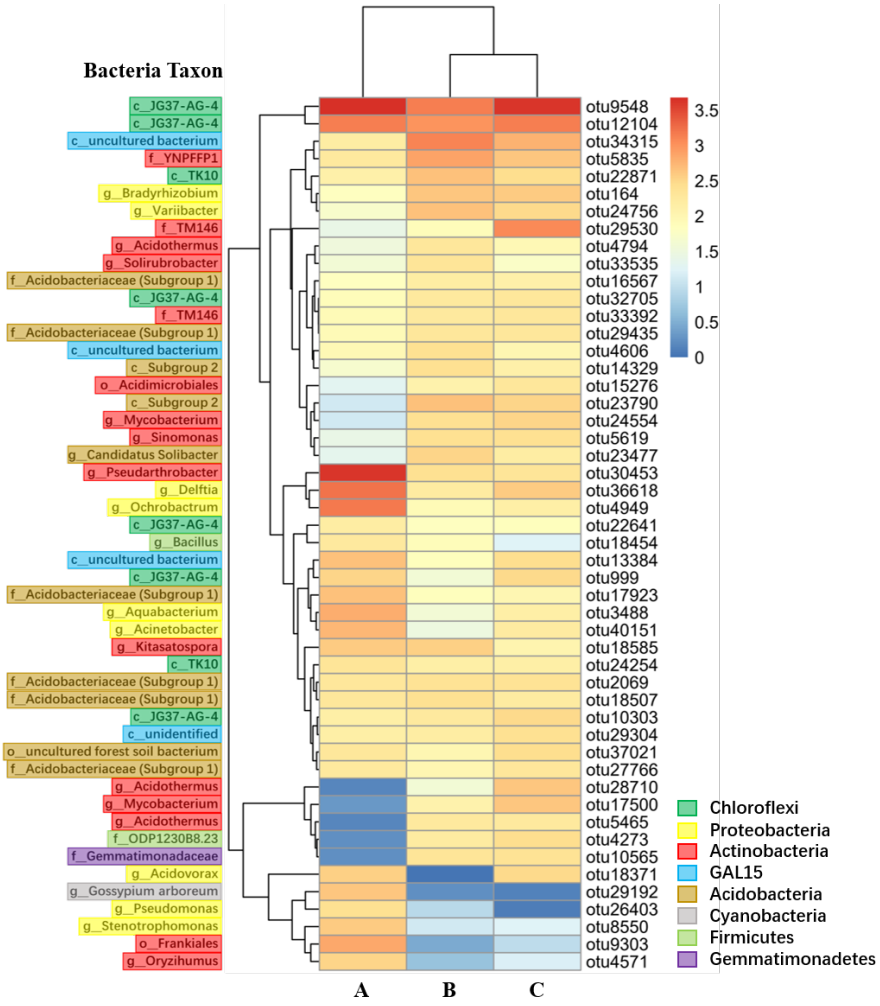

Figure S3. Heatmap displaying the abundance of top 50 OTUs of bacterial across all the three land types after standardization: [p=log (n+1, 10), n means the average sequences number]. A, B, and C indicate sandy land, shrub land, and forest land, respectively. The bacteria taxon corresponding with each OTU is showed at the left and the letter before the taxon indicates the level of assignment (c=class, o=order, f=family, g=genus).