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一个红壤剖面微生物群落的焦磷酸测序法研究

Pyrosequencing approach to study microbial composition in a red soil profile

中文关键词:红壤 焦磷酸测序 微生物多样性 古菌 细菌 真菌

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中文摘要:

利用定量PCR和454焦磷酸测序法，研究了湖南湘阴县一典型红壤剖面微生物相关基因的多度及微生物（古菌、细菌、真菌）群落结构。结果显示，随剖面深度增加，土壤黏粒含量增多，有机质和全氮含量、碳氮比则下降。每克干土微生物基因拷贝数也趋于下降，其值为：$10^{7.09}$～$10^{9.30}$（古菌16S rDNA），$10^{8.10}$～$10^{9.70}$（细菌16S rDNA），$10^{6.54}$～$10^{7.95}$（真菌18S rDNA），$10^{7.24}$～$10^{8.61}$（古菌*amoA*基因），$10^{4.76}$～$10^{6.25}$（细菌*amoA*基因），$10^{5.94}$～$10^{7.88}$（*nirK*基因），$10^{6.81}$～$10^{9.21}$（*nirS*基因），$10^{7.03}$～$10^{9.46}$（*nosZ*基因）。焦磷酸测序得到了6 459条古菌16S rRNA基因序列，平均长度为496 bp；28 626条细菌16S rRNA基因序列，平均长度为448 bp；4 683条真菌18S rRNA基因序列，平均长度为534 bp。OTU（97%相似度）分析表明，微生物群落α-多样性与所测土壤理化性质均无显著相关。Jaccard差异度分析表明同一剖面各土壤层次间微生物群落结构更为相似，而不同位点的三个表层土之间的差异较大；Mantel检验发现，与微生物群落变化相关的主要土壤因子是黏粒含量。在所有土样中，古菌以泉古菌门中的热变形菌纲（89%）为主，其分布与土壤黏粒含量相关。细菌的主要类群为酸杆菌门（33%）、变形菌门（17%）、绿弯菌门（12%）、厚壁菌门（10%）和放线菌门（7%），分类地位不明确的细菌约占11%。其中，酸杆菌门和变形菌门的相对多度在表层土中高于非表层土；而绿弯菌门和厚壁菌门的相对多度则在非表层土中更高，与土壤深度呈显著正相关。所有真菌序列分属于三个门，即子囊菌门（87%）、担子菌门（9%）和球囊菌门（4%），在纲一级的分类水平上，各样品间群落结构无明显差异。

英文摘要:

Abundances and community structure of archaea, bacteria and fungi in a red soil profile located at Xiangyin County, Hunan province, was investigated with the quantitative real-time PCR (qPCR) and 454 pyrosequencing approaches. Results showed that with increasing soil depth, soil clay increased in content, while organic matter, total nitrogen, and carbon to nitrogen ratio declined. Correspondingly, the numbers of gene copies per gram of dry soil also decreased within the range of$10^{7.09}$～$10^{9.30}$ for archaeal 16S rDNA, $10^{8.10}$～$10^{9.70}$ for bacterial 16S rDNA, $10^{6.54}$～$10^{7.95}$ for fungal 18S rDNA, $10^{7.24}$～$10^{8.61}$for archaeal *amoA*gene, $10^{4.76}$～$10^{6.25}$ for bacterial *amoA*gene, $10^{5.94}$～$10^{7.88}$for *nirK* gene, $10^{6.81}$～$10^{9.21}$for *nirS* gene, and $10^{7.03}$～$10^{9.46}$ for *nosZ* gene. Pyrosequencing generated 6 459 archaeal 16S rDNA sequences with an average length of 496 bp, 28 626 bacterial 16S rDNA sequences with an average length of 448 bp, and 4 683 fungal 18S rDNA sequences with an average size of 534 bp. OTU (97% similarity) analysis revealed that the α-diversity was in the order bacteria>fungi>archaea, but there was no significant correlation between microbial α-diversity and soil properties determined. Analysis of the Jaccard dissimilarity indicated that microbial communities in different layers of the same soil profile were closer, compared with the dissimilarity between three surface replicates. Mantel test showed that clay content was the main soil factor explaining the variation of microbial communities. In all the soil samples, archaea was dominated (89%) with Thermoprotei (belonging to Crenarchaeota), of which the distribution was significantly related to soil clay content. The bacterial community consisted mainly of Acidobacteria (33%), Proteobacteria (17%), Chloroflexi (12%), Firmicutes (10%)， Actinobacteria (7%) and Bacteria incertae sedis (11%) . Acidobacteria and Proteobacteria were found to be more abundant in the surface soils than in other soil layers, while the situations of Chloroflexi and Firmicutes were opposite. All fungal sequences belonged to three phyla, i.e., Ascomycota (87%), Basidiomycota (9%) and Glomeromycota (4%). This work demonstrated the great potential of pyrosequecing technique in revealing microbial diversity and presented background information of microbial communities in the red soil.

袁超磊,贺纪正,沈菊培,戴宇,张丽梅.一个红壤剖面微生物群落的焦磷酸测序法研究[J].土壤学报,2013,50(1):138-149.Yuan Chaolei,He Jizheng,Shen Jupei,Dai Yu and Zhang Limei.Pyrosequencing approach to study microbial composition in a red soil profile[J].Acta Pedologica Sinica,2013,50(1):138-149

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