RESEARCH ARTICLE

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Effects of continuous cropping of sweet potatoes on the bacterial community structure in rhizospheric soil

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Abstract

Background: Continuous cropping obstacles from sweet potatoes are widespread, which seriously reduce the yield and quality, causing certain economic losses. Bacteria of rhizospheric soil are the richest and are associated with obstacles to continuous cropping. However, few studies have examined how continuous sweet potato cropping affects the rhizospheric soil bacterial community structure.

Results: In the study, the Illumina MiSeq method was used to explore the variations in rhizospheric soil bacterial community structure of different sweet potato varieties after continuous cropping, as well as the correlation between soil characteristics and the bacterial community. The results showed that (1) the dominant bacterial phyla in rhizospheric soils from both Xushu 18 and Yizi 138 were *Proteobacteria*, *Acidobacteria*, and *Actinobacteria*. The most dominant genus was *Subgroup 6_norank*. The relative abundance of rhizospheric soil bacteria varied significantly between the two sweet potato varieties. (2) The richness and diversity indexes of bacteria were higher in Xushu 18 rhizospheric soil than in Yizi 138 soil after continuous cropping. Moreover, beneficial *Lysobacter* and *Bacillus* were more prevalent in Xushu 18, while Yizi 138 contained more harmful *Gemmatimonadetes*. (3) Soil pH decreased after continuous cropping, and redundancy analysis indicated that soil pH was significantly correlated with the bacterial community. Spearman's rank correlation coefficient analysis demonstrated that pH was positively associated with *Planctomycetes* and *Acidobacteria*, but negatively associated with *Actinobacteria* and *Firmicutes*.

Conclusions: After continuous cropping, the bacterial community structure and physicochemical properties of sweet potato rhizospheric soil were changed, and the changes from different sweet potato varieties were different. The contents of *Lysobacter* and *Bacillus* were higher in the sweet potato variety resistant to continuous cropping. It provides a basis for developing new microbial fertilizers for sweet potatoes to alleviate the continuous cropping obstacle.

Keywords: Bacterial community, Continuous cropping, Rhizospheric soil, Sweet potato

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Background

Sweet potato [*Ipomoea batatas* (L.) Lam.] is a major food crop that is cultivated worldwide. It is a high-yield, highly efficient, and strongly adaptable crop that is resistant to drought and barrenness [1–3]. In addition, sweet potato is rich in nutrients and has cancer preventing properties, making it increasingly popular [4]. Therefore, sweet potato planting areas increase annually. In China, because of limited cultivatable land, sweet potatoes are grown continuously in the same fields. However, continuous cropping can lead to decreased yield and quality, severe plant death, and a decreased or non-existent harvest [5, 6]. In recent years, the phenomenon of sweet potato continuous cropping obstacles has become serious, and it has huge adverse impact on the sweet potato industry [7].

Continuous cropping obstacles have a certain relationship with soil enzyme activity, soil microbial community, and root exudates [8–10]. Soil microorganisms are the key factors related to changes in soil quality, fertility, and productivity [11–13]. The rhizosphere is the soil region near the plant roots, where interactions between soil microorganisms and plant root systems are very strong [14, 15]. Rhizospheric soil microorganisms are closely related to the absorption and transformation of soil nutrients. Therefore, their community structure is a major factor affecting plant growth, development, and health [16–19]. Many reports have indicated that continuous cropping changes the structure of rhizospheric soil microbes [20–23]. These changes have further led to severe continuous cropping obstacles [9]. Therefore, the relationship between continuous cropping and the soil microorganisms has become a research hotspot [7].

Bacteria are the most abundant in rhizospheric soil and are the important component [24, 25]. Increasing reports have noted that continuous cropping causes changes in rhizospheric soil bacterial community structure. Previous studies on konjac and *vanilla* revealed that continuous cropping changed bacterial communities, resulting in a decrease of beneficial bacteria and an increase of harmful bacteria [26, 27]. Moreover, the study found that the bacteria decreased after *Panax notoginseng* continuous cropping for 3 years. And the rhizospheric soil bacterial diversity of the healthy *Panax notoginseng* was greater than that of diseased strains. Canonical correspondence analysis found that P, pH, and soil organic matter had the greatest impacts on the bacterial community [28]. Furthermore, Na et al. [29] and Lei et al. [30] revealed that the number of bacterial species and the α -diversities of the rhizospheric soil bacterial communities of *Lycium barbarum* L. and *Sophora flavescens* decreased significantly after continuous cropping.

The detrimental effects of continuous cropping on microbial community structures in rhizospheric soil have

been demonstrated for a variety of different crops. However, studies focusing on the continuous cropping of sweet potatoes have mainly focused on the prevention and treatment of pests and diseases [31]. Accordingly, little is known about how this process affects the bacterial community structure and the physical and chemical properties (especially the medium and micro elements) in the rhizospheric soil of sweet potato. Various medium and micro elements are not only indispensable components of plants, but also participate in various metabolic reactions and act as catalysts for various enzymes [32]. Moreover, it remains unclear whether there are differences in the bacterial community structure of rhizospheric soil after the continuous cropping of different sweet potato cultivars. For the first time, we have used two sweet potato varieties and high-throughput techniques to study bacterial community structure changes of rhizospheric soil after continuous cropping, and analyzed correlations between soil characteristics and bacterial community changes after continuous cropping. It aimed to reveal how continuous cropping affects the rhizosphere soil bacterial community structures of two sweet potato varieties, and to provide insights into the application of biological control the continuous cropping obstacles of sweet potatoes.

Results

Physicochemical properties of sweet potato rhizospheric soil

Available Mn in the Xushu 18 (X18) and Yizi 138 (Y138) rhizospheric soils decreased by 32.68% and 31.14%, respectively, at the beginning of planting in 2016 than in 2015, and by 27.35% and 31.10% in the pre-harvest period (Table 1). The soil pH of the X18 and Y138 were decreased by 2.72% and 3.11%, respectively, at the beginning of planting in 2016 than in 2015; changes in pH were not significant in the pre-harvest period. At the beginning of planting, available Ca in the X18 and Y138 rhizospheric soils increased by 29.80% and 38.97%, respectively; available Zn increased by 56.11% and 43.19%, respectively in 2016 than in 2015. And available Ca increased by 30.75% and 26.47%, respectively, while available Zn increased by 29.46% and 30.81%, respectively, in the pre-harvest period in 2016 than in 2015. Available Fe in the X18 and Y138 soils decreased by 18.61% and 17.08%, respectively, in the pre-harvest period in 2016 than in 2015, but was not significantly different at the beginning of planting. Available B in the X18 rhizospheric soil decreased by 20.63% at the beginning of planting, while the change in the Y138 soil was not significant.

Rhizospheric soil bacterial α -diversity

The average coverage of all samples was 96.19% (Table 2). Rarefaction curves were close to plateau (Fig. 1),

Table 1 The physicochemical properties of different soil samples

Sample	Ca (g kg ⁻¹)	B (mg kg ⁻¹)	Fe (mg kg ⁻¹)	Mn (mg kg ⁻¹)	Zn (mg kg ⁻¹)	pH
X18-1	0.34±0.01c	0.77±0.07a	4.64±0.67c	2.60±0.28a	1.65±0.13d	8.57±0.06a
X18-2	0.35±0.01c	0.71±0.08ab	5.86±0.30a	2.54±0.06a	1.96±0.01c	8.60±0.00a
X18-3	0.45±0.01b	0.61±0.06b	4.78±0.16bc	1.75±0.07b	2.58±0.09a	8.33±0.06b
X18-4	0.45±0.00ab	0.63±0.05b	4.77±0.01bc	1.85±0.05b	2.54±0.02a	8.50±0.10a
Y138-1	0.34±0.02c	0.78±0.05a	4.52±0.24c	2.56±0.01a	1.73±0.06d	8.57±0.06a
Y138-2	0.36±0.01c	0.69±0.04ab	5.31±0.53ab	2.71±0.26a	1.66±0.17d	8.60±0.00a
Y138-3	0.47±0.02a	0.69±0.08ab	4.70±0.18bc	1.76±0.12b	2.48±0.00a	8.30±0.00b
Y138-4	0.46±0.01ab	0.72±0.09ab	4.41±0.03c	1.87±0.04b	2.17±0.03b	8.53±0.06a

Values are mean±standard deviation of triplicate determinations. Different letters in the same column indicate significant differences of the same sweet potato varieties at different sampling times at a level of $P<0.05$ using Duncan's multiple range tests; Ca:available calcium; B: available boron; Fe: available iron; Mn : available Manganese; Zn: available zinc. X18: Xushu 18; Y138:Yizi 138; 1 and 2 represent sampling of early planting and early harvest in 2015; 3 and 4 represent sampling of early planting and early harvest in 2016, respectively

indicating that our sequencing depth was good. The reads ranged from 24,695 – 37,688 for samples, and the operational taxonomic units (OTUs) ranged from 3,137 – 3,734. Chao, Shannon, and Simpson indexes were computed based on the bacterial OTUs. Unlike the Shannon index, the larger the Simpson value, the lower the community diversity. The Chao and Shannon values of the X18 and Y138 soils were higher in the pre-harvest period than at the beginning of planting, while the opposite was true of the Simpson index, indicating that two communities had higher species richness and diversity in the pre-harvest period.

After X18 continuous cropping, the Chao index decreased by 2.53% and the Shannon and Simpson indices were basically unchanged at the beginning of planting in 2016 than in 2015; in the pre-harvest period, the Chao and Shannon indices increased by 5.95% and 1.59%, respectively, and the Simpson index decreased by 17.86%. After Y138 continuous cropping, the Chao and Shannon indices increased by 2.80% and 1.66%, respectively, and the Simpson indices decreased by 22.22% at the beginning of planting in 2016 than in 2015; in the pre-harvest

period, the Chao and Shannon indices increased by 2.53% and 1.16%, respectively, and the Simpson index decreased by 14.29%. These indicated that the diversity and richness of rhizosphere soil bacteria of X18 and Y138 also increased after continuous cropping.

X18 had higher Chao and Shannon indices than Y138, which were contrary to the Simpson index. In other words, the bacterial richness and diversity were higher in X18 rhizospheric soil than in Y138 soil.

Community composition analysis of rhizospheric soil bacteria

At the phylum level (Fig. 2), the dominant bacterial phyla of all samples were *Proteobacteria* (28.5%–34.9%), *Acidobacteria* (10.4%–21.1%), *Actinobacteria* (11.3%–18.1%), *Planctomycetes* (5.2%–9.9%), *Chloroflexi* (4.6%–9.1%), *Bacteroidetes* (3.4%–6.1%), *Gemmatimonadetes* (3.0%–7.4%), and *Firmicutes* (1.4%–10.9%). Among them, *Proteobacteria* was the most abundant phylum, followed by *Acidobacteria* and *Actinobacteria*.

After X18 and Y138 continuous cropping, *Proteobacteria* decreased by 17.30% and 8.05%, respectively, in

Table 2 MiSeq sequencing results and α -diversity index of sweet potato rhizospheric soil samples

Sample ID	Reads	0.97	Chao	Coverage	Shannon	Simpson
		OTU				
X18-1	37688	3326	4626 (4440, 4842)	0.969407	6.69 (6.68, 6.71)	0.0033 (0.0032, 0.0034)
X18-2	28047	3405	4955 (4743, 5201)	0.954327	6.91 (6.89, 6.93)	0.0028 (0.0027, 0.0029)
X18-3	35422	3207	4509 (4324, 4724)	0.967139	6.67 (6.65, 6.68)	0.0034 (0.0033, 0.0035)
X18-4	33236	3734	5250 (5046, 5485)	0.960434	7.02 (7.01, 7.04)	0.0023 (0.0022, 0.0023)
Y138-1	32513	3137	4388 (4209, 4596)	0.964876	6.64 (6.62, 6.66)	0.0036 (0.0035, 0.0037)
Y138-2	24695	3230	4808 (4590, 5061)	0.948654	6.89 (6.87, 6.91)	0.0028 (0.0027, 0.0029)
Y138-3	35866	3156	4511 (4316, 4739)	0.968187	6.75 (6.74, 6.77)	0.0028 (0.0028, 0.0029)
Y138-4	33436	3602	5074 (4873, 5307)	0.961897	6.97 (6.95, 6.98)	0.0024 (0.0024, 0.0025)

X18: Xushu 18; Y138:Yizi 138; 1 and 2 represent sampling of early planting and early harvest in 2015, 3 and 4 represent sampling of early planting and early harvest in 2016, respectively; OTU: operational taxonomic unit; The numbers within parentheses are the lower and upper limits in statistics of the corresponding date, respectively

Fig. 1 Rarefaction curves of all samples. 1 and 2 represent the early planting and pre-harvest samples in 2015; 3 and 4 represent the early planting and pre-harvest samples in 2016, respectively. The same as following

Fig. 2 Relative abundance of rhizospheric soil bacterial phyla

the pre-harvest period. *Acidobacteria* showed a decreasing trend and finally increased slightly, while *Actinobacteria* showed the opposite trend. After continuous cropping, the *Firmicutes* content of X18 soil increased by 71.54% and 97.66% in the early planting and early harvest stages, respectively, and by 4.10% and 129.57%, respectively in Y138 soil. Moreover, In both varieties, the *Firmicutes* contents was higher at the beginning of planting than in the early harvest period of the same year. However, the opposite trend was observed for *Planctomycetes*. The *Planctomycetes* content of X18 in the early planting and early harvest stages decreased by 30.36% and 7.35%, respectively, while that of Y138 decreased by 40.51% and 19.04%, respectively. Further, the *Chloroflexi* and *Gemmatimonadetes* contents showed increasing trends. In X18 and Y138 rhizospheric soils, *Chloroflexi* increased by 81.09% and 96.69%, respectively, and *Gemmatimonadetes* increased by 103.11% and 122.56%, respectively. *Gemmatimonadetes* was higher in Y138 rhizospheric soil than in X18 soil, especially in 2016.

At the genus level (Fig. 3), *Subgroup 6_norank* (6.59%–14.74%), *Nitrosomonadaceae_uncultured* (1.83%–6.40%), and *Anaerolineaceae_uncultured* (1.75%–3.63%) were the top three dominant bacteria genera in all rhizospheric soils of X18 and Y138. Other major genera were

Bacillus (0.65%–4.14%), *MSB-1E8_norank* (0.87%–3.83%), *Tepidisphaeraceae_norank* (1.71%–2.56%), *Xanthomonadales_norank* (0.62%–2.08%), and *Lysobacter* (0.55%–2.06%). After two years of continuous cropping, *Subgroup 6_norank* displayed decreasing trends in the X18 and Y138 rhizospheric soils, decreasing by 54.34% and 52.66%, respectively, and then increased slightly in the 2016 pre-harvest period. However, *Nitrosomonadaceae_uncultured* and *Anaerolineaceae_uncultured* were present at low levels at the beginning of planting, then were increased in the pre-harvest period of the same year. *Bacillus* and *Lysobacter* showed the opposite trend. Moreover, in every sampling period, *Lysobacter* was higher in the rhizospheric soil of X18 than in that of Y138. This was also true of *Bacillus*, except for the beginning of planting in 2015. Furthermore, in 2016, the reduction of *Lysobacter* in X18 and Y138 rhizosphere soil was 1.3 and 2.4 times greater than in 2015.

Venn analysis of rhizospheric soil bacteria

Venn diagram revealed the overlapped and unique OTUs of all samples (Fig. 4). After two years of continuous cropping, the OTUs shared by all samples was 507. In the four sampling periods, there were 95, 158, 127, and 202 unique OTUs in the rhizospheric soils of X18,

Fig. 3 Relative abundance of rhizospheric soil bacterial genera

Fig. 4 Number of common and unique OTUs based on Venn analysis

and 89, 124, 141, and 159 unique OTUs in the rhizospheric soils of Y138. As continuous cropping year increased, the specific OTUs in X18 soil increased by 33.68% and 27.85% in the early planting and early harvest periods, while the OTUs in Y138 soil increased by 58.43% and 28.23% in these periods.

The number of OTUs specific to X18 was greater than that specific to Y138 (except for the beginning of planting in 2016), indicating that continuous cropping led to changes in the bacterial communities in X18 and Y138 rhizosphere soil. These differences were largest during the early harvest period of 2016.

Heatmap, clustering, and principal component analysis (PCA) of rhizospheric soil bacteria

The results of heatmap and clustering analysis of 40 phyla in all samples are illustrated in Fig. 5, and clearly demonstrate the differences in rhizospheric soil bacterial composition between the X18 and Y138 varieties. The samples grouped into two clusters and samples from the same consecutive cropping times clustered together. In addition, X18 and Y138 samples from the same sampling periods grouped together.

The OTUs of X18 and Y138 were subjected to PCA. The extracted two principal components explained 72.48% of the total variation (Fig. 6). As continuous cropping time increased, samples from different sampling times became farther apart. However, at the same sampling times, X18 and Y138 samples were relatively close to each other. With continuous cropping, the distance between these samples also gradually increased, which indicated that differences between their bacterial communities were also increasing. These results were consistent with the results of the heatmap and cluster analyses in Fig. 5. Overall, the results suggested that (i) continuous cropping led to bacterial community structure changes in X18 and Y138 rhizosphere soil; (ii) rhizospheric soil bacterial community structures of X18 and Y138 were similar in the same sampling period.

Relationship between bacterial phyla and physicochemical characteristics of sweet potato rhizospheric soil

The results of redundancy analysis (RDA) on top ten bacterial phyla and environmental factors of X18 and Y138 rhizospheric soil are showed in Fig. 7. RDA1 and RDA2 explained 56.57% and 28.05% of the total

Fig. 5 Microbial community heatmap and cluster analysis of the bacterial phyla

Fig. 6 Principal component analysis (PCA) of OTUs

variation, respectively. The magnitude of the effects of soil properties on bacterial community structure had the following order, soil pH > Ca > Mn > Zn > B > Fe. The results showed that soil pH ($r^2=0.9737$, $Pr=0.004$) and available Ca ($r^2=0.8815$, $Pr=0.011$) were significantly correlated with the bacterial community. It indicated that pH was a strong predictor of the X18 and Y138 rhizospheric soil bacterial community compositions.

In addition, the results of Spearman's correlation coefficient analysis were as follows (Fig. 8). pH was positively correlated with *Planctomycetes* ($R=0.97$) and *Acidobacteria* ($R=0.93$), but had negative correlations with *Actinobacteria* ($R = -0.79$) and *Firmicutes* ($R = -0.72$); available Ca was positively related to *Actinobacteria* ($R=0.89$) and *Gemmatimonadetes* ($R=0.86$), and was inversely correlated with *Acidobacteria* ($R = -0.79$), *Planctomycetes* ($R= -0.75$), and *Nitrospirae* ($R= -0.72$). At the same time, it can be seen from Fig. 8 that the soil physicochemical properties were divided into two groups, with available Ca and available Zn clustered into one group and the rest clustered into another, indicating that

available Ca and available Zn had similar effects on the bacteria, which were different from the rest.

Discussion

To obtain a more comprehensive understanding of the rhizospheric soil bacterial community structure of sweet potatoes, the Illumina MiSeq method was used in this study [33, 34]. The V4–V5 highly variable region was selected as the sequencing region because a previous study demonstrated that this region was the best sequencing region among nine highly-variable regions [35].

We found that the Shannon index increased with continuous cropping, indicating an increase in bacterial diversity in the rhizospheric soil. This was aligned with previous studies on *Andrographis paniculata* and potatoes [36, 37], but contrary to *Sophora flavescens* and *Lycium barbarum* L. under continuous cropping conditions [29, 30]. The differences may be due to variations in the duration of continuous cropping, soil environmental conditions, plant types, etc.

Fig. 7 Redundancy analysis of the 10 dominant bacterial phyla and soil physicochemical properties. Ca: available calcium; B: available boron; Fe: available iron; Mn: available manganese; Zn: available zinc. The same as following

The most dominant bacterial phylum in the X18 and Y138 rhizospheric soil was *Proteobacteria*, which was in accordance with published reports [8, 24, 36, 38]. The main function of *Proteobacteria* is to decompose organic matter and promote plant growth [39, 40]. The most dominant genus was *Subgroup 6_norank*, which accorded with Yin et al. [24]. As continuous cropping time increased, the *Proteobacteria* content decreased, which was in agreement with Liu et al. [41], while was inconsistent with the report on *A. paniculata* [36]. *Acidobacteria* showed a tendency to decrease gradually and then increase slightly. *Actinobacteria* tended to increase gradually and then decrease slightly, which was almost consistent with Yin et al. [24] and our previous study [42], but differed from the results of Li et al. [8] and Wu et al. [27]. These discrepancies might be due to differences in plant species, plant genotypes, continuous cropping durations, and soil types, which also cause different changes in soil microorganisms [14, 43–45].

We also found that some microbial changes were affected by the sweet potato growth stage. For instance,

the content of *Firmicutes* was high at the beginning of planting and decreased in pre-harvest period. This indicates that the growth of sweet potato has a certain influence on it. After the sweet potatoes were harvested, its content recovered. These changes may also be related to the season. Li et al. [46] found that seasonal changes have important impacts on soil microbes, but that different microbes vary with season differently.

As continuous cropping time increased, the content of *Lysobacter* decreased. It is well known that *Lysobacter* is an important biocontrol bacteria with strong bacteriolytic and bacteriostatic effects [47]. For example, *Lysobacter enzymogenes* OH 11 was found to exert a significant bacteriostatic effect on sweet potato soft rot pathogen. Conversely, *Gemmatimonadetes*, a harmful bacteria that can lead to N loss and reduce crop growth [40], increased with continuous cropping.

Sweet potato continuous cropping changed the bacterial community structure, which was aligned with previous reports [8, 26, 27]. These changes further increased the continuous cropping obstacles of X18 and Y138.

Fig. 8 Correlations between the 10 dominant bacterial phyla and soil properties. *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$

The average yields of the two varieties both decreased, X18 decreased from 12.36 to 10.64 t hm^{-2} , and Y138 decreased from 9.15 to 2.66 t hm^{-2} .

Consequently, maintaining a balance of bacterial community structure is very important for prevention and control continuous cropping obstacles. In practice, microbial fertilizers and soil modifiers [48–50] can be applied to maintain the microecological balance.

We observed differences in rhizospheric soil bacterial community composition between the two sweet potato varieties after continuous cropping. The number of OTUs specific to X18 increased more than those specific to Y138, and the community structures became increasingly different. According to α -diversity analysis, the rhizospheric soil of X18 had higher bacterial richness and diversity. Thus, the community structure of X18 was relatively stable, its tolerance to soil environmental changes was greater, and its resistance to continuous cropping obstacles was stronger, while the community structure of Y138 was more susceptible to continuous cropping. Moreover, *Lysobacter* and *Bacillus* were higher in the X18 rhizosphere soil than in the Y138 soil. Many studies have shown that *Bacillus* may resist some soil-borne diseases [14, 27, 40]. Higher *Lysobacter* and *Bacillus* contents would therefore be more conducive to the growth of X18. In contrast, the rhizospheric soil of Y138 contained more harmful *Gemmatimonadetes*. Therefore, it is also important to choose varieties of sweet potato

that are resistant to continuous cropping in production practice.

After sweet potato continuous cropping, rhizospheric soil physicochemical properties were also changed. Moreover, it was found that pH had the greatest impact on the bacterial community structure, which was consistent with several published reports [8, 51, 52]. Soil pH can affect soil microbial physiological metabolism, alter competitive relationships within microbial communities, and inhibit the growth of non-adapted microbes [29], all of which affect microbial community structure. In terms of the three bacterial phyla with the highest relative content, *Acidobacteria* was positively correlated with pH, which was in line with Yang et al. [40]; *Actinobacteria* was negatively correlated with pH, which was in agreement with previous studies [27, 53]. However, the results were different from those reported by Li et al. [8], which might have been due to difference in plant age or genotype [54].

Similar to bacteria, soil fungi are important decomposers. However, some fungi are strongly associated with plant diseases [55, 56]. The results of our previous study indicated that fungal diversity and abundance in rhizospheric soil were significantly increased after continuous cropping. Furthermore, the contents of *Fusarium*, *Verticillium*, and *Colletotrichum*, the pathogens of sweet potatoes, were increased. These changes affected the balance of the fungal community structure [7].

Overall, after continuous cropping, the microbial community structure of sweet potato rhizospheric soil was changed, which might be a crucial aspect contributing to continuous cropping obstacles. Thus, maintaining the microecosystem balance of the rhizosphere soil is crucial to relieve the continuous cropping obstacles. In addition, continuous cropping obstacles are related to many factors. It is necessary to combine with many other methods to solve this problem. Therefore, we will perform further in-depth research on the mechanisms of sweet potatoes continuous cropping obstacles.

Conclusions

The continuous cropping of sweet potatoes obviously affected the bacterial community structure, physical and chemical properties of the rhizosphere soil. The bacterial diversity increased; however, the changes in the soils of two sweet potato varieties were different. The contents of *Lysobacter* and *Bacillus* were higher in the sweet potato variety that was resistant to continuous cropping. It provides a basis for the development of specialized microbial fertilizers for sweet potatoes. Therefore, further research is needed to see if *Lysobacter* and *Bacillus* could be used to produce microbial fertilizers to mitigate continuous cropping obstacles of sweet potatoes.

Methods

Study area and materials

Our study was conducted at the Dishang test station of Institute of Cereal and Oil Crops, Hebei Academy of Agriculture and Forestry Sciences, China (37°56′24.62″ N, 114°42′46.96″ E) [7]. There was a warm temperate sub-humid continental monsoon climate and the local soil type is Mottlic Hapli-Ustic Argosols (cinnamon soil). Two sweet potato varieties were used as experimental materials, one was X18 that was resistant to continuous cropping and the other was Y138 that was susceptible to continuous cropping. Both varieties were collected from the Institute of Cereal and Oil Crops, Hebei Academy of Agriculture and Forestry Sciences.

Experimental design and sample collection

Sweet potatoes were planted in May and harvested in October each year, and were continuously planted for 2 years (2015–2016). After each harvest, the fields were left fallow for the rest of the year. A random block design was used, with three experimental replicates. Details of the plot can be found in our previous report [7]. Agronomic management was the same for all fields over the two years. Rhizospheric soil was collected 30 days after planting and 7 days before the harvest, for a total of four sampling periods. Rhizospheric soil samples were taken by the method of Sun et al. [57]. For each sweet potato variety at each sampling point, the rhizospheric

soils of 15 sweet potatoes located in an “S” shape were collected from each plot and mixed thoroughly to generate one composite soil sample. All samples were quickly taken to the laboratory in a cooler. Each sample was filtered through a 2-mm sieve, and then divided into two portions. One portion was stored at −80°C for DNA analysis, the other portion was air-dried for soil characteristics analysis.

Soil physicochemical property analysis

Soil pH was determined by the electrode method (2.5:1 water: soil ratio) [40], and available Ca, B, Fe, Mn, and Zn were measured using the atomic absorption method according to Du and Gao [58].

DNA extraction, amplification, and sequencing

The DNA was extracted with an E.Z.N.A.® Soil DNA Kit following the instructions. The V4–V5 regions were amplified by polymerase chain reaction (PCR) with 515F 5′-barcode-GTGCCAGCMGCCGCGG-3′ and 907R 5′-CCGTCAATTCMTTTRAGTTT-3′, where barcode was an 8-base sequence unique to each sample. PCR reactions and amplicons extraction and purification were performed by the method of Yang et al. [40]. Purified PCR products were quantified using Qubit®3.0, and every 24 amplicons with different barcodes were equally mixed. Illumina Pair-End library construction and sequencing were performed according to our previous report [7]. Complete data sets were deposited in the NCBI Sequence Read Archive database under accession number SRP214716.

Processing and statistical analysis of sequencing data

To remove low quality sequences, quality control and sequence quality filtering were applied according to Yang et al. [40]. OTUs were clustered with 97% similarity cutoff with UPARSE (version 7.1 <http://drive5.com/uparse/>) and chimeric sequences were identified and removed with UCHIME. RDP Classifier was used to analyze the phylogenetic affiliations of each 16S rRNA gene sequence against the SILVA (<http://www.arb-silva.de>) database with a confidence threshold of 70% [40].

Chao, Shannon, and Simpson indexes were calculated with Mothur v.1.21.1 [59]. PCA was performed with R-forge (the PCA graphics was generated with Vegan 2.0 package). Shared and unique OTUs were analyzed using Venn diagrams by VennDiagram. RDA was performed to analyze the correlations between environmental factors and bacterial community composition. Spearman's rank correlations coefficients and Heatmap figures were performed with Vegan packages in R. The data of the physicochemical properties of rhizospheric soil were analysed by one-way analysis of variance (ANOVA) ($P < 0.05$) in SPSS v21.0 [40].

Abbreviations

X18: Xushu18; Y138: Yizi138; OTUs: Operational taxonomic units; PCA: Principal component analysis; RDA: Redundancy analysis; ANOVA: One-way analysis of variance

Acknowledgements

We thank Prof. Xiumin Zhang from Hebei University and Prof. Shulong Chen from the Plant Protection Institute, Hebei Academy of Agricultural and Forestry Sciences for their help with our manuscript.

Authors' contributions

ZM and ZJG designed this experiment. LL and WJ performed samples collection. YH, JX, XW, ZT, RJ and ML executed the experiment. ZYG, MH and ZT analyzed the data. ZYG, YH and MH finished the manuscript. All authors read and approved the final manuscript.

Funding

This work was supported by the National Natural Science Foundation of China (grant number 31771731); The Modern Agriculture Industrial Technology System Innovation Team Project of Hebei Province (grant number HBCT2018080203); The Modern Agricultural Science and Technology Innovation Project of Hebei Province (grant number 2019-4-2-7); China Agriculture Research System (CARS-10-C1, Sweet potato) and the Natural Science Foundation of Hebei Province, China (grant number C2019301127). The funding bodies played no role in the design of the study or collection, analysis, and interpretation of data or in writing the manuscript.

Availability of data and materials

The datasets generated and analysed during the current study are available in the NCBI Sequence Read Archive repository under accession number SRP214716 (<https://submit.ncbi.nlm.nih.gov/subs/sra/SUB7627087/overview>).

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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Received: 18 August 2020 Accepted: 9 February 2021

Published online: 01 April 2021

References

- Dai QW, Niu FX, Sun J, Cao J. Development and trend analysis of Sweetpotato processing industry in China. *Agricultural Engineering Technol.* 2015;35:27–31. <https://doi.org/10.16815/j.cnki.11-5436/s.2015.35.003>.
- Mateus JR, Marques JM, Dal'Rio I, Vollu RE, MRR C, Seldin L. Response of the microbial community associated with sweet potato (*Ipomoea batatas*) to *Bacillus safensis* and *Bacillus velezensis* strains (Article). *Antonie van Leeuwenhoek, Int J Gen Mol Microbiol.* 2019;112:501–12. <https://doi.org/10.1007/s10482-018-1181-y>.
- Song CJ, Wang JC. Research progress on high yield of sweetpotato. *Tillage and Cultivation.* 2007;2:45–7. <https://doi.org/10.13605/j.cnki.52-1065/s.2007.02.027>.
- Sheikha AFE, Ray RC. Potential impacts of bioprocessing of sweet potato: Review. *Critical Rev Food Sci Nutr.* 2015;57:455–71. <https://doi.org/10.1080/10408398.2014.960909>.
- Jia ZD, Xie YZ, Yin QH, Guo XD, Ji XQ. Study progress and perspective of black rot resistant germplasm in sweetpotato. *J Plant Genet Resources.* 2010;11:424–7 432.
- Zhou Z, Ma DF. Research status and prospect of sweetpotato stem nematode. *Rain Fed Crops.* 2003;23:288–90.
- Gao ZY, Han MK, Hu YY, Li ZQ, Liu CF, Wang X, et al. Effects of continuous cropping of sweet potato on the fungal community structure in rhizospheric soil. *Front Microbiol.* 2019;9:10. <https://doi.org/10.3389/fmicb.2019.02269>.
- Li H, Wang JQ, Liu Q, Zhou ZF, Chen F, Xiang D. Effects of consecutive monoculture of sweet potato on soil bacterial community as determined by pyrosequencing. *J Basic Microbiol.* 2019;59:181–91. <https://doi.org/10.1002/jobm.201800304>.
- Sun CS. Study on the effects of *Astragalus membranaceus* (fisch.) bunge quality and rhizosphere soil microorganisms under continuous cropping with the *Astragalus membranaceus* (fisch.) bunge rotation mode. Dissertation, Inner Mongolia: Inner Mongolia university; 2017.
- Huang WJ, Sun DL, Fu JT, Zhao HH, Wang RH, An YX. Effects of continuous sugar beet cropping on rhizospheric microbial communities. *Genes.* 2020;11:13. <https://doi.org/10.3390/genes1101013>.
- Beckers B, Op De Beeck M, Weyens N, Boerjan W, Vangronsveld J. Structural variability and niche differentiation in the rhizosphere and endosphere bacterial microbiome of field-grown poplar trees. *Microbiome.* 2017;5:25. <https://doi.org/10.1186/s40168-017-0241-2>.
- Blagodatskaya E, Kuzyakov Y. Active microorganisms in soil: Critical review of estimation criteria and approaches. *Soil Biol Biochem.* 2013;67:192–211. <https://doi.org/10.1016/j.soilbio.2013.08.024>.
- Sun L, Gao JS, Huang T, Kendall JRA, Shen QR, Zhang RF. Parental material and cultivation determine soil bacterial community structure and fertility. *FEMS Microbiology Ecology.* 2015;91:1–10. <https://doi.org/10.1093/femsec/fiu010>.
- Shang QH, Yang G, Wang Y, Wu XK, Zhao X, Hao HT, et al. Illumina-based analysis of the rhizosphere microbial communities associated with healthy and wilted Lanzhou lily (*Lilium davidii* var. *unicolor*) plants grown in the field. *World J Microbiol Biotechnol.* 2016;32:1–15. <https://doi.org/10.1007/s11274-016-2051-2>.
- Li M, Yang FZ, Wu XY, Yan H, Liu Y. Effects of continuous cropping of sugar beet (*Beta vulgaris* L.) on its endophytic and soil bacterial community by high-throughput sequencing. *Ann Microbiol.* 2020;70:39. <https://doi.org/10.1186/s13213-020-01583-8>.
- Berendsen RL, Pieterse CMJ, Bakker PAHM. The rhizosphere microbiome and plant health. *Trends Plant Sci.* 2012;17:478–86. <https://doi.org/10.1016/j.tplants.2012.04.001>.
- Bron PA, Van BP, Kleerebezem M. Emerging molecular insights into the interaction between probiotics and the host intestinal mucosa. *Nat Rev Microbiol.* 2012;10:66–78. <https://doi.org/10.1038/nrmicro2690>.
- Fierer N, Lauber CL, Ramirez KS, Zaneveld J, Bradford MA, Knight R. Comparative metagenomic, phylogenetic and physiological analyses of soil microbial communities across nitrogen gradients. *ISME J.* 2012;6:1007–17. <https://doi.org/10.1038/ismej.2011.159>.
- Jin Z. The activities of the rhizosphere soil culturable microbial and soil nutrient under potato continuous and rotation system. Dissertation, Inner Mongolia: Inner Mongolia Agricultural University; 2012.
- Guo LL, Yin WL, Guo DL, Hou XJ. Variations of bacterial biodiversity in rhizosphere soils of oil tree peony cropping continuously for different years. *Scientia Silvae Sinicae.* 2017;11:131–41. <https://doi.org/10.11707/j.1001-7488.20171115>.
- Huang LF, Song LX, Xia XJ, Mao WH, Shi K, Zhou YH, et al. Plantsoil feedbacks and soil sickness: from mechanisms to application in agriculture. *J Chem Ecol.* 2013;39:232–42. <https://doi.org/10.1007/s10886-013-0244-9>.
- Qiao PL, Wu FZ, Zhou XG. Effects of continuous cropping on soil microbe communities and enzyme activity. *J Shenyang Agricultural Univ.* 2013;5:524–30. <https://doi.org/10.3969/j.issn.1000-1700.2013.05.003>.
- Sun WQ, Kang YL, Liu JG, Jiang GY. Influence of continuous cropping of processing tomato on diversity of microflora in the rhizosphere soil. *Acta Agriculturae Boreali-occidentalis Sinica.* 2017;26:1099–110. <https://doi.org/10.7606/j.issn.1004-1389.2017.07.020>.
- Yin JZ, Li L, Jie WG, Cai BY. Effect of continuous cropping on bacterial flora structure in soybean rhizosphere soil. *Biotechnol Bull.* 2018;34:230–8. <https://doi.org/10.13560/j.cnki.biotech.bull.1985.2017-0728>.
- Tian WH, Yi XL, Liu SS, Zhou C, Wang AY. Effect of transgenic cotton continuous cropping on soil bacterial community. *Ann Microbiol.* 2020;70:61. <https://doi.org/10.1186/s13213-020-01602-8>.

26. Wu JP, Jiao ZB, Zhou J, Guo FL, Ding ZL, Qiu ZM. Analysis of bacterial communities in rhizosphere soil of continuously cropped healthy and diseased konjac. *World J Microbiol Biotechnol*. 2017;33:1–8. <https://doi.org/10.1007/s11274-017-2287-5>.
27. Wu X, Zhao QY, Zhao J, Xun WB, Li R, Zhang RF, et al. Different continuous cropping spans significantly affect microbial community membership and structure in a vanilla-grown soil as revealed by deep pyrosequencing. *Microbiol Ecol*. 2015;70:209–18. <https://doi.org/10.1007/s00248-014-0516-0>.
28. Tan Y, Cui YS, Li HY, Kuang AX, Li XR, Wei YL, et al. Diversity and composition of rhizospheric soil and root endogenous bacteria in *Panax notoginseng* during continuous cropping practices. *J Basic Microbiol*. 2017; 57:337–44. <https://doi.org/10.1002/jobm.201600464>.
29. Na XF, Zheng GQ, Xing ZC, Ma JP, Li ZH, Lu JH, et al. Effects of monocropping on diversity and structure of bacterial community in rhizosphere of replanted *Lycium barbarum* L. *Acta Pedologica Sinica*. 2017; 54:1280–92. <https://doi.org/10.11766/trxb201702070036>.
30. Lei HY, Liu A, Hou QW, Zhao QS, Guo J, Wang ZJ. Diversity patterns of soil microbial communities in the *Sophora flavescens* rhizosphere in response to continuous monocropping. *BMC Microbiol*. 2020;20:272. <https://doi.org/10.1186/s12866-020-01956-8>.
31. Johnson AC, Gurr GM. Invertebrate pests and diseases of sweetpotato (*Ipomoea batatas*): a review and identification of research priorities for smallholder production. *Ann Appl Biol*. 2016;168:291–320. <https://doi.org/10.1111/aab.12265>.
32. Li ZG, Wang C, Yang JF, Chao Z, Zheng WQ, Yu H. Characteristics analysis of soil and plant medium and micro element of consecutive cropping black pepper. *Chin J Tropical Crops*. 2017;38:2215–20. <https://doi.org/10.3969/j.issn.1000-2561.2017.12.003>.
33. Ai S, Zhang LJ, Xiao PY, Zhang XF, Xing ZL. Application and progress of high-throughput sequencing technology in the field of environmental microorganisms. *J Chongqing Univ Technol (Natural Science)*. 2018;32:111–21. [https://doi.org/10.3969/j.issn.1674-8425\(2\).2018.09.018](https://doi.org/10.3969/j.issn.1674-8425(2).2018.09.018).
34. Cheng YB, Ma QB, Ren HL, Xia QJ, Song EL, Tan ZY, et al. Fine mapping of a phytophthora-resistance gene *RpsWY* in soybean (*Glycine max* L.) by high-throughput genome-wide sequencing. *Theoretical Appl Genet*. 2017;130: 1041–51. <https://doi.org/10.1007/s00122-017-2869-5>.
35. Sun DL, Jiang X, Wu QL, Zhou NY. Intragenomic heterogeneity of 16S rRNA genes causes overestimation of prokaryotic diversity. *Appl environment microbiol*. 2013;79:5962–9. <https://doi.org/10.1128/aem.01282-13>.
36. Li JR, Chen XZ, Li SM, Zuo ZM, Zhan RT, He R. Variations of rhizospheric soil microbial communities in response to continuous *Andropogon paniculata* cropping practices. *Botanical Studies*. 2020;61:18. <https://doi.org/10.1186/s40529-020-00295-1>.
37. Zhao J, Zhang D, Yang YQ, Pan Y, Zhao DM, Zhu JH, et al. Dissecting the effect of continuous cropping of potato on soil bacterial communities as revealed by high-throughput sequencing. *Plos One*. 2020;15(5):e0233356. <https://doi.org/10.1371/journal.pone.0233356>.
38. Zhu L, Zeng CL, Li YQ, Yu BQ, Gao F, Wei W, et al. The characteristic of bacterial community diversity in soybean field with continuous cropping based on the high-throughput sequencing. *Soybean Science*. 2017;36:419–24. <https://doi.org/10.11861/j.issn.1000-9841.2017.03.0419>.
39. Li X, Sun ML, Zhang HH, Xu N, Sun GY. Use of mulberry-soybean intercropping in salt-alkali soil impacts the diversity of the soil bacterial community. *Microbiol Biotechnol*. 2016;9:293–304. <https://doi.org/10.1111/1751-7915.12342>.
40. Yang L, Tan LL, Zhang FH, Gale WJ, Cheng ZB, Sang W. Duration of continuous cropping with straw return affects the composition and structure of soil bacterial communities in cotton fields. *Canadian J Microbiol*. 2018;64:167–81. <https://doi.org/10.1139/cjm-2017-0443>.
41. Liu X, Zhang JL, Gu TY, Zhang WM, Shen QR, Yin SX, et al. Microbial community diversities and taxa abundances in soils along a seven-year gradient of potato monoculture using high throughput pyrosequencing approach. *PLoS One*. 2014;9:e86610. <https://doi.org/10.1371/journal.pone.0086610>.
42. Gao ZY, Hu YY, Liu LF, Wang YN, Jiao WJ, Ma ZM, et al. Effects of continuous cropping on the microbial community structure of rhizospheric soil of sweetpotato. *J Nuclear Agricultural Sci*. 2019;6:1240–7. <https://doi.org/10.11869/jissn.100-8551.2019.06.1248>.
43. Zhao F, Zhao MZ, Wang Y, Pang FH. Biodiversity of bacteria and fungi in rhizosphere of strawberry with different continuous cropping years. *Microbiology China*. 2017;44:1377–86. <https://doi.org/10.13344/j.microbiol.china.170170>.
44. Berg G, Smalla K. Plant species and soil type cooperatively shape the structure and function of microbial communities in the rhizosphere. *FEMS Microbiol Ecol*. 2009;68:1–13. <https://doi.org/10.1111/j.1574-6941.2009.00654.x>.
45. Liu F, Hewezi T, Lebeis Sarah L, Pantalone V, Grewal Parwinder S, Staton Margaret E. Soil indigenous microbiome and plant genotypes cooperatively modify soybean rhizosphere microbiome assembly. *BMC Microbiol*. 2019; 19(1):201. <https://doi.org/10.1186/s12866-019-1572-x>.
46. Li NJ, Zeng QP, He BH, Zhou F. Seasonal variations of soil microbial PLFAs and soil enzyme activity under the citrus plantation in Mt. Jinyun, Chongqing. *Environmental Science*. 2017;38:309–17. <https://doi.org/10.13227/j.hjlx.201605017>.
47. Chen ZH, Wang MZ, Chen BL, Huang JZ. The progress of research and application on *Lysobacter* and its bioactive compounds. *Academic Periodical of Farm Products Processing*. 2013;1:5–10. [https://doi.org/10.3969/j.issn.1671-9646\(X\).2013.01.002](https://doi.org/10.3969/j.issn.1671-9646(X).2013.01.002).
48. Cai Y, Hao MD, Zang YF, He XY, Zhang LQ. Effect of long-term fertilization on microbial diversity of black loessial soil based on 454 sequencing technology. *J Nuclear Agricultural Sci*. 2015;29:0344–50. <https://doi.org/10.11869/j.issn.100-8551.2015.02.0344>.
49. Qiao XR. Effect of different amendments on soil microbial ecology. Dissertation, Zhejiang, Zhejiang University. 2011.
50. Tao L, Chu GX, Liu T, Tang C, Li JH, Liang YC. Impacts of organic manure partial substitution for chemical fertilizer on cotton yield, soil microbial community and enzyme activities in mono-cropping system in drip irrigation condition. *Acta Ecologica Sinica*. 2014;34:6137–46. <https://doi.org/10.5846/stxb201301290184>.
51. Fernández-Calviño D, Bååth E. Growth response of the bacterial community to pH in soils differing in pH. *FEMS Microbiol Ecol*. 2010;73:149–56. <https://doi.org/10.1111/j.1574-6941.2010.00873.x>.
52. Qiao SS, Zhou YN, Liu JX, Jing JH, Jia T, Li C, et al. Characteristics of soil bacterial community structure in coniferous forests of Guandi Mountains, Shanxi Province. *Scientia Silvae Sinicae*. 2017;53:89–99. <https://doi.org/10.11707/j.1001-7488.20170211>.
53. Alami MM, Xue J, Ma Y, Zhu D, Gong Z, Shu S, et al. Structure, diversity, and composition of bacterial communities in rhizospheric soil of coptis chinensis franch under continuously cropped fields. *Diversity*. 2020;12:57. <https://doi.org/10.3390/d12020057>.
54. Marques JM, da Silva TF, Vollu RE, Blank AF, Ding GC, Seldin L, Smalla K. Plant age and genotype affect the bacterial community composition in the tuber rhizosphere of field-grown sweet potato plants. *FEMS Microbiol Ecol*. 2014;88(2):424–35. <https://doi.org/10.1111/1574-6941.12313>.
55. Dong LL, Xu J, Feng GQ, Li XW, Chen SL. Soil bacterial and fungal community dynamics in relation to *Panax notoginseng* death rate in a continuous cropping system. *Scientific Reports*. 2016;6:31802. <https://doi.org/10.1038/srep31802>.
56. Shang HS. Modern immunology of plants. Beijing: China Agriculture Press; 2013.
57. Sun JZ, Xue QH, Tang M, Cao SM, Xing SL. Study on the effect of actinomycetes on microflora of replanted strawberry's root domain and the bio-control effectiveness. *Journal of Northwest A & F University. Nat Sci Ed*. 2009;37:153–8. <https://doi.org/10.13207/j.cnki.jnwf.2009.12.021>.
58. Du S, Gao XZ. Soil analysis technical specification. 2nd ed. Beijing: China. Agriculture Press; 2006.
59. Schloss PD, Westcott SL, Ryabin T, Hall JR, Hartmann M, Hollister EB, et al. Introducing mothur: open-source, platform-independent, community-supported software for describing and comparing microbial communities. *Appl Environmental Microbiol*. 2009;75:7537–41. <https://doi.org/10.1128/aem.01541-09>.

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