

Soil Sickness Problems in Continuous Cropping System

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ABSTRACT

In recent years, the large-scale agricultural activities and the excessive use of soil resources has led to many problems in agricultural production. The vigorous implementation of the monoculture has resulted in serious decline in soil fertility, soil quality, soil diseases and pests. Although facility agriculture is an advanced modern agricultural method, soil acidification, salinization, nutrient imbalance, microbial community destruction and other problems still exist. The continuous cropping of field crops, vegetables and medicinal plants has led to the problems of soil sickness, which adversely affected their quality and yield. Continuous cropping obstacle has become the major problem for crop industry development. This review discusses the soil sickness problems in continuous cropping from two aspects : Changes in soil physico-chemical properties and Soil microflora diversity.

Key words: Bacteria, continuous cropping problem, field crops, fungi, medicinal plants, nutrient elements, soil microorganisms, soil sickness, vegetables.

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1. INTRODUCTION

The soils are an important component of terrestrial ecosystems, however, the unreasonable land use has resulted in soil salinization, acidification, nutrients loss and overall degradation of soil structure and changed microbial ecology (13,32). The land degradation is caused by the incorrect agricultural management practices (unsustainable planting systems, excessive use of chemical fertilizers, repeated cultivation of same crop and overgrazing). Long-term monoculture systems (continuous cultivation of the same crop or its close relatives on the same land), leads to continuous cropping problem, which is a form of land degradation (104). The mechanisms of this phenomenon are complex and lead to decline in soil quality and microbial diversity. In continuous cropping, many perennial and annual crops face problems of restricted plant growth, vitality, reduced crop yield and quality (3,13,51). Long-term monoculture also seriously affects the soil physico-chemical properties e.g., reduces the soil pH to acidic range, which adversely affects the soil availability of nitrogen (N), phosphorus (P) and potassium (K) nutrients (3,82). This results in the imbalance of soil supply of available nutrients to plants.

Owing to soil microorganisms ability to respond sensitively to environmental changes, they plays significant role in many processes of soil ecosystems and are used as ideal biological indicators of soil health and quality (6,10). The long-term continuous cropping affects the soil microbial community structure and diversity (21,41). For example, long-term continuous cropping increases the diversity and abundance of harmful microbial communities and decreases the diversity and abundance of beneficial microbial communities in the soil (11,72,94). Autotoxicity is an important cause of continuous cropping problem, which leads to the accumulation of autotoxins, the substances secreted by plant roots in soil (53). In addition to inhibiting the plant growth and development, the accumulation of autotoxins also affects the rhizosphere ecosystem by inducing changes in microbial population, soil enzyme activity and nutrients recycling (8,38).

As explained above, continuous cropping can lead to changes in the soil environment, which can lead to continuous cropping problem. Therefore, understanding the effects of continuous cropping on soil physical, chemical, and biological properties is crucial to further optimize the soil conditions, mitigate and eliminate the adverse effects of continuous cropping problems and improve the sustainability of continuous cropping systems (3). This paper reviews the effects of continuous cropping of field crops on the changes in soil physico-chemical properties and soil microbial community.

2. CHANGES IN SOIL PHYSICO-CHEMICAL PROPERTIES

Soil quality index refers to the soil processes and properties that are sensitive to soil function changes (92). Soil quality indicators can be used to evaluate the sustainability of land use and soil management practices (75). According to the American Society for Soil Science (1995), soil quality is defined as "the capacity of a particular soil to function within the boundaries of a natural or managed ecosystem to maintain plant and animal productivity, to maintain or enhance water and air quality, and to support human health and habitation".

These requirements can be met by maintaining or enhancing the interrelated chemical, physical and biological properties of the soil. For example, deterioration of soil physical conditions can adversely affect the chemical and biological properties of the soil (18). The soil fertility includes all kinds of total and available nutrients in soil and reflects the nutrients supply capacity of soil.

(i). Soil pH: Continuous cropping changes the soil physico-chemical properties; however, the type of changes vary with crops. For example the continuous cropping of strawberry (*Fragaria × ananassa* Duch.), Black pepper (*Piper nigrum* Carl Linnaeus.), coffee (*Coffea arabica* Carl Linnaeus.), corn (*Zea mays* Carl Linnaeus.), soybean (*Glycine max* Carl L. Merr.) gradually decreases the pH with increase of continuous cropping years. Whereas, the soil pH was not affected by alfalfa (*Medicago sativa* L.), barley and Chinese Chives (*Allium tuberosum* Rottler ex Sprengle.) (15,23,29,59,98,104, 105,111). However the sesame, increased the soil pH during short-term continuous cropping (84).

(ii). Soil nutrients: The content of available phosphorus (AP) in the soil of strawberry was increased in continuous cropping in second year and stabilized in the sixth year of continuous cropping. Total nitrogen (TN) content increased from the second year and became stable from the 10th year. The contents of ammonia nitrogen (NH₄⁺-N) decreased significantly from the 6th year, and the contents of available potassium (AK) and soil organic matter (SOM) decreased significantly from the 2nd year, and then decreased slowly and becomes stable (15). The SOM content of black pepper in continuous cropping decreased year by year, while available nitrogen and AP content increased gradually with continuous cropping years (98). The SOM content decreased gradually, and the contents of soil EC, AP, Fe and Zn increased with the increase of coffee continuous cropping years (111). Soil total carbon (TC), TN, available phosphorus, exchangeable potassium and exchangeable calcium in continuous cropping of maize decreased gradually with the increase of continuous cropping years (23). Soil available nutrients such as N, P and K decreased significantly in soybean continuous cropping (59). In the continuous cropping alfalfa soil, the contents of soil moisture, TC, TN, NO₃⁻-N and available potassium decreased first and then rose with the increase of planting years. The content of total phosphorus (TP) increased, while the AP content decreased. The content of NH₄⁺-N did not change significantly (104). The contents of TP and total potassium (TK) in barley continuous cropping soil did not change, while the concentrations of TN and nitrate nitrogen (NO₃⁻-N) declined with the increase of continuous cropping years (105). In the soil of continuous Chinese Chives cropping, the content of SOM did not change significantly, but the contents of TP, AP, TK and AK increased significantly with the increase of continuous cropping years. TP content reached the highest point at the fifth year of continuous cropping, while the AK content peaked at the third year of continuous cropping (29). In the short-term continuous cropping of sesame seeds (*Sesamum indicum* Carl Linnaeus.), soil TN content and C/N in the second crop year were increased by 0.05 g/kg and 0.39, respectively, than the first

crop year. In comparison to the first crop year, the contents of exchangeable Ca and Cu in soil increased by 146.8 mg/kg and 0.53 PPM respectively. Soil AP content was significantly increased from 18.0 mg/kg in the first crop year to 25.8 mg/kg in the second crop year, and it also had similar pattern in the contents of exchangeable Mg and trace elements Fe, Zn and Mn. However, the content of soil organic carbon decreased after continuous cropping (84).

3. CHANGES IN MICROBIAL COMMUNITY STRUCTURE

The continuous cropping of the same crop in the same land could adversely affect the crop yield and quality, a phenomenon known as ‘soil sickness’, which is very problematic issue in agriculture (103). Long-term continuous cropping often leads to the change in microbial community structure in plant rhizosphere soil and increase the soil borne plant pathogens in the rhizosphere soil (100). In severe cases, it adversely affects the growth and production of crops (17). The abundance and diversity of microorganisms in soil are the key factors in soil microbiology, which plays important role in maintaining the soil fertility and soil ecological balance (86). Soil microbiome serve as an indicator of soil quality due to its sensitivity to subtle changes in the environment resulting from environmental stresses or natural changes (67,73). The soil microbial diversity is crucial for maintaining the soil health (2). It is key driver of terrestrial ecosystems, as is critical to many soil biological, chemical and physical processes [soil structure formation, mineral nutrients recycling, transformation of organic matter and accumulation or removal of toxins (35,116)].

Studies have shown that the imbalance in the structure of soil microbial community is caused by long-term continuous cropping, the major problems in continuous cropping (72). The soil microbial community structure varies with the years of continuous cropping and crop species. Soil fungus are key regulators of carbon flow, plant population dynamics and soil physical environment in terrestrial ecosystems (97). Fungal diversity is beneficial to plant production by suppressing the pathogens (90). The bacteria are sensitive to external disturbance (26,33,67,78). A decrease in the bacterial/fungal ratio leads to an imbalance in soil microflora and deterioration of soil quality (113). A decrease in soil microbial diversity is responsible for the development of soil-borne plant diseases (60). The allelopathic effects of beneficial microbes (mycorrhizal fungi, rhizobia and plant-growth-promoting rhizobacteria and pathogenic microbes) could influence the plant growth as they are involved in many important processes for plant growth and nutrients availability (1,16,35).

Continuous cropping of different crops causes several changes in the soil microbial community structure. In general, soil bacterial diversity decreased after continuous cropping of some crops [peanut (13), American ginseng (19), soybean (50,59), tea (52), Notoginseng Radix Et Rhizoma (80), cucumber (57,112) and cotton (101,108), while fungal diversity (e.g., *Fusarium* dominant) increased after continuous cropping of strawberry (15,36), peanut (13), American ginseng (19), sorghum (93), tomato (44,115), Heterophylly Falsestarwort Root (95) and soybean (93)], while, Fungal diversity increased in other crops (Table 1). The accumulation of *Fusarium* load in soil decreases the beneficial fungi [*Trichoderma* sp., a glomeromycotan fungus and *Mortierella elongate* (29,50,95)]. With the increase in

continuous cropping years the fungal quantity in rhizosphere and bulk soil rose significantly. For example, the relative abundances of *Martrell sp.* (12,44,55), *Paecilomyces sp.* (59), *Thelebolus*, and potentially pathogenic fungi *Fusarium oxysporum* and *Lectera longa* (55,95), and verticillium wilt pathogens (109) were increased, while the quantities of bacteria [*Actinobacteria*, *Gemm-3*, *Acidimicrobiia*, *Halobacteria* (108) and *Pseudomonas* (98)] and actinomycetes decreased significantly (35,46,108,110,116).

Table 1. Effects of continuous cropping on Bacterial and Fungal diversity

Microbial Diversity	Continuously cropped crops
Bacterial diversity- Decreased	American ginseng (19), Black pepper (98), Cotton (108), Notoginseng Radix Et Rhizoma (80), Peanut (35,37,116), Potato(58), Sorghum (93), Soybean (59), Strawberry (49), Tea (85), Tobacco (77,88) and Tomato (44).
Fungal diversity- Increased	Cotton (46), Cucumber (114), Heterophylly Falsestarwort (53), Notoginseng Radix Et Rhizoma (79), Peanut (15,50), Rehmanniae Radix (96), Soybean (59), Strawberry (48) and Tea (53).

In general, Bacterial diversity decreased after continuous cropping of some crops, while, Fungal diversity increased in other crops (Table 1).

3.1 FIELD CROPS

3.1.1 Soybean

(i). Bacteria: Continuous cropping is common in soybean (*Glycine max* L.) production. Liu, Z.X. *et al.* (59) found that microbial diversity and abundance changed after continuous cropping of soybean. In the rhizosphere soil, the microbial diversity in crop rotation with maize was higher than in soybean continuous cropping. The fungi/bacteria ratio was significantly higher in the rhizosphere soils of continuously cropping for 3 and 5 years than 13 years cropping, indicating that short-term continuously cropped soybean reduces the bacterial abundance and raises fungal abundance. The soil pH and C/N were the primary soil factors to change the bacterial and fungal community structures in the bulk soils, respectively. Redundancy analysis showed that soil pH made the greatest contribution to the change in bacterial community structures, while the soil C/N ratio was the most important factor in changing the fungal community structures. The *Bradyrhizobium spp.* is beneficial to biological nitrogen fixation (5), phosphate solubilization (68), production of hormones [auxins, cytokinins, gibberelins and ethylene (9,76,83)] and control of pathogens (1,4,87). The genus *Gemmatimonas* is a diverse group, it contribute to soil organic carbon sequestration (30,89) and decomposition of cellulose and lignin (99). *Mortierella* degrades the aromatic hydrocarbons and decompose plant litter (19,63).

(ii). Fungi: Li, *et al.* (44) found that there were 106 OTUs in 2-3 years continuously cropped soybean soil samples and 44 OTUs in 1-year cropped soybean soil samples. Soybean continuous cropping not only affects the diversity of fungi in rhizosphere soil, but also increased the abundance of *Thelebolus* and *Mortierellales* fungi. *Thanatephorus*, *Fusarium* and *Alternaria* were dominant pathogenic fungal genera in rhizosphere soil from continuously cropped soybean fields. The *Fusarium spp.* causes soybean root rot in

northeastern China, and their secretions reduces the abundance of beneficial fungi and bacteria in soil. *Alternaria* spp. induce several diseases [early blight, black spot, brown patch, leaf spot, stem wilt and brown spot] through the production of toxins (74). Liu, *et al.* (55,56) also showed that the rhizospheric soil fungal abundance was significantly higher in continuous cropping (CC) than in crop rotation (CR) with maize. The continuous cropping increased the relative abundances of the pathogenic fungi (*Fusarium oxysporum* and *Lectera longa*) as well as the beneficial fungi (*Mortierella* sp. and *Paecilomyces lilacinus*). The bacterial abundance in the rhizosphere of CC was significantly lower than CR, hence, the fungi/bacteria ratio was higher in the rhizospheric soil of CC than CR. The CC of soybean decreased the bacterial abundance but increased the fungal abundance in both rhizospheric and bulk soils. There are two possible explanations for this phenomenon: CC of soybean decreases the soil pH, which benefits the fungal growth but inhibits the bacterial growth (7,62,91), i.e. associated with enrichment of allelochemicals in the CC soybean soils, as a positive correlation between the allelochemical daidzein content and soil fungal abundance in monocultured soybean fields (31).

3.1.2 Peanut

Peanut (*Arachis hypogaea* L.) is an important oil crop and its long-term continuous cropping changes the microbial community. The quantities of fungus in rhizosphere and bulk soil rose significantly with the increase in continuous cropping years, while the quantities of bacteria and actinomycetes decreased (35,37,116). The results have been described for short-term continuous cropping (1-2 Years) and long-term continuous cropping (11-12 Years). Chen, *et al.* (13) found that continuous cropping years strong influenced on the microbial communities as well as the soil physico-chemical properties. The soil pH and contents of AP and AK in soil changes in long-term continuous cropping of peanut. Additionally, as compared with short-term continuous cropping, the long-term continuous cropping changes the soil bacterial diversity, abundance and composition. The abundance of some taxa [*Planctomycetes*, *Nitrospirae*, *Bacteroidetes*, *Armatimonadetes*, *Latescibacteria*, *JL-ETNP-Z39*, *Thermotogae*, and *Caldiserica*] was decreased. Some dominant taxa showed an upward trend with the increase of continuous cropping years.

(i). Bacteria: In genera level, the dominant genus *Bacillus* abundance increased in the long-term continuous cropping years. Abundance of the dominant species (*Bacillus aryabhatai*, *Bacillus funiculus*) and the relatively abundant species (*Bacillus luciferensis*, *Bacillus decolorationis*) all increased with long-term continuous cropping. Thus identified members of *Bacillus* had good adaptability to the soil environment under long-term peanut continuous cropping. *Bacillus* species are most common biocontrol agents and also have plant growth-promoting properties (27,69). These improves the growth of soybean, wheat and Italian cocklebur and also improves with the mobilization and biofortification of zinc (42,66).

(ii). Fungi: The genus *Nitrospira* is related to nitrogen cycle (61). Additionally, the long-term continuous cropping of peanut also changed the soil pH and contents of AP and AK in soil. Chen *et al.* (13) found that the fungal populations changed significantly

and increased over time under continuous peanut cropping. The abundance and diversity of clones affiliated with *Eurotiales*, *Hypocreales*, *Glomerales*, *Orbiliales*, *Mucorales* and *Tremellales* showed an increasing trend with continuous cropping, but the clones affiliated with *Agaricales*, *Cantharellales*, *Pezizales* and *Pyxidiophorales* decreased in abundance and diversity over the time. Chen *et al* (12) reported that at the species level, the *Fusarium solani*, *Fusarium oxysporum*, *Neocosmospora striata*, *Acrophialophora levis*, *Aspergillus niger*, *Aspergillus corrugatus*, *Thielavia hyrcaniae*, *Emericellopsis minima*, and *Scedosporium aurantiacum* taxa abundances significantly increased in the long-term continuous cropping. In contrast, *Talaromyces flavus*, *Talaromyces purpureogenus*, *Mortierella alpina*, *Paranomyces uniporus* and *Volutella citrinella* were reduced in the long-term continuous cropping than in short-term continuous cropping. It reflects the genotype homogenization in the soil. The environmental and genetic uniformity of the agricultural ecosystem may promote the emergence of new pathogens (77), these unclassified fungi that appeared after intensive monocropping, may be related to new pathogenic species. Li, *et al* (50) found that the accumulation of a fungal pathogen load in the soil at the expense of the beneficial fungal flora (*Trichoderma sp.*, Glomeromycotan fungus, and *Mortierella elongata*) of the plant may be the possible reason for the decrease in the yield caused by continuous planting of peanuts. Combined with previous studies (12,50) indicated that fungal community structure was affected by the length of the continuous cropping period and there were significant differences between the short-term and the long-term continuous cropping soils, but it was less affected by the peanut variety and the growth stage. Simplification of fungal diversity and modifications in the soil microbes could lead to soil sickness in long-term peanut continuous cropping fields. The increased abundance of pathogens and the decreased abundance of beneficial fungi could be the main cause of the yield decline and poor growth of monocropped peanuts.

3.1.3 Cotton

(i). Bacteria: Zhang, *et al* (108) compared the composition of bacterial communities in the cotton (*Gossypium spp.*) root zone soil by analyzing multiple years of continuous cropping of cotton and corn-cotton rotation. He found that bacterial diversity decreased after long-term continuous cropping compared to 1 year of cotton planting and reached the minimum after 30-years of continuous cropping, with bacterial-diversity decreasing from 0.431 to 0.292. At the class level, cotton cultivation significantly reduced the number of *Actinobacteria*, *Gemm-3* and *Acidimicrobiia*. The *Halobacteria* nearly disappeared after continuous cotton cropping. *Alphaproteobacteria* and *Gemmatimonadetes* increased than others and *Thaumarchaeota*, *Gemm-1*, *Deltaproteobacteria*, and *Acidobacteria-6* increases greatly. These findings suggests that continuous cotton cropping reduces the organic matter in the soil, which significantly reduced the *Actinobacteria* population (107). Whereas, the *Acidobacteria*, *Nitrospirae* and other bacteria increased in the continuous cropping soil due to the increased soil pH [caused by continuous cropping and extensive use of nitrogen fertilizer in the cotton cultivation] (70). Li, *et al* (46) studied the

number of culturable microorganisms in the soil of continuous cropping cotton field with different years, and found that continuous cropping of cotton reduced the number of culturable microorganisms in the soil, and the number of microbes in such soil varied with different years, the number of bacteria and actinomycetes in soil decreased in continuous cropping cotton field of 5, 10 and 15 years than that of 1 year. In contrast, Yang, *et al.* (101) found that bacterial species (*Chlorobacteria*, *Acidobacteria*) richness and diversity and that of *Actinomycetes* increased for 10-15 years. Tian, *et al.* (81) applied a high-throughput sequencing method and compared the bacterial community structure as well as diversity of rhizosphere soil of the transgenic cotton line (25C-1) and its parent cotton line (TH-2). They found that the proportion of Proteobacteria in rhizosphere soil of 25C-1 cotton strain was higher compared to the TH-2 cotton strain. The proportion of *Actinobacteria* in rhizosphere soil of 25C-1 cotton strain was also significantly higher than TH-2 strain. Previous studies have shown that a reduction in the organic matter in the soil due to continuous cotton cropping significantly reduced the *Actinobacteria* population (107). However Li, *et al.* (55) found that the content of total nitrogen and organic matter, the abundance of Proteobacteria and Actinobacteria increased in the soil after continuous cropping of genetically modified cotton. This may be a different effect of genetically modified cotton on the soil.

(ii). Fungi: Zhang, *et al.* (109) found that during long-term continuous cropping of cotton, the number of verticillium wilt pathogens in the soil increased rapidly with prolonged continuous cropping time, reached maximum after 10 years. Li, *et al.* (46) found that the number of fungus in cotton field ameliorated with the increase in continuous cropping years and there were significant differences among the continuous cropping years. The maximum fungal population was only 4.53×10^3 cfu/g at the seedling stage in 1-year cotton field, but increase sharply after 10-years of continuous crop, and reached the highest point (1.91×10^4 cfu/g) at the boll opening stage in the 15-years cropped cotton field, which was 130.02 % higher than that in the same period of 10-years of continuous crop.

3.1.4. Tea

(i). Bacteria: Wang, (85) found that long-term continuous cropping of tea plants (*Camellia sinensis* L.) changed the bacterial community structure, reduced microbial diversity and decreased the beneficial plant-related bacteria in continuous cropping soil. At the genus level, the relative abundance of beneficial bacteria, such as *Pseudomonas*, *Rhodanobacter*, *Bradyrhizobium*, *Mycobacterium* and *Sphingomonas*, significantly decreased in the 20-year tea orchard soils. The abundance of bacteria and fungi in soil of 8-year and 10-year tea plantations were significantly higher than those in 34-year and 56-year tea plantations. The results showed that the abundance of bacteria decreased with the increase of continuous cropping years. Among them, the abundance of *Acidobacteria*, *Actinomycetes*, *Chlorobacteria*, *Proteobacteria* and *Bacteroidetes* in the soil of long-term continuous cropping tea plant was low.

(ii). Fungi: Li, *et al.* (53) found that the long-term continuous ratooning cultivation of tea plants changed the fungal communities in the rhizosphere, enriched saprotrophs and plant pathogens (*Alternaria* spp.) and reduced the beneficial fungi (symbiotrophs). The

FUNGuild revealed that the symbiotrophs in the 1-year tea soil was markedly higher than those in the 0-, 10- and 20-year soils. The saprotrophs in the 20-year soils were approximately two-folds higher than in the 0-, 1- and 10-year soils. The pathotroph-saprotroph- symbiotroph fungi were higher in the 10- and 20-year soils than in 0- and 1-year soils as expected.

3.1.5. Sorghum

Wu, *et al* (93) found that bacterial α -diversity, Chao1 index was significantly higher in sorghum-maize rotation than that in continuous sorghum cropping, while Shannon index had no significant difference. Chao1 index showed no significant difference in fungal α -diversity, and Shannon index was significantly higher in sorghum continuous cropping than in sorghum-maize rotation.

3.1.6. Medicinal Plants

3.1.6.1. Ginseng: (I). American ginseng (*Panax quinquefolius* L.): Dong, *et al* (19) found by high-throughput sequencing analysis of 16S rRNA and 18S rRNA of soil samples, that soil bacterial and fungal communities varied in the diversity and composition after continuous cropping of American ginseng. Compared with no continuous cropping, after 3 years of continuous cropping, bacteria and fungi decreased significantly by 47.7 % and 45.5 % , respectively. In soils used to cultivate American ginseng, the relative abundances of *Acidimicrobiales*, *Actinomycetales*, *Gaiellales*, *Solirubrobacterales*, *Sphingobacterales*, *Bacillales*, *Nitrospirales*, *Sphingomonadales*, *Burkholderiales*, *Myxococcales*, and *Syntrophobacterales* declined by 0.1-49.6 % and the relative abundances of *Arthoniales*, *Pleosporales*, *Lecanorales*, and *Sordariales* declined by 61.4-274 %.

(II). Notoginseng Radix Et Rhizoma (*Panax notoginseng* Chen ex C.H.)

(i). Bacteria: Tan, *et al* (80) found that with Notoginseng Radix Et Rhizoma continuous cropping the quantities of rhizospheric bacteria decreased, but did not affect the endophytic bacteria. The most dominant phyla in continuous cropping were *Proteobacteria*, *Cyanobacteria*, *Actinobacteria* and *Acidobacteria*. The genera *Pseudomonas*, *Rhodoplanes*, *Candidatus Solibacter* and *Streptomyces* were dominant in *P. notoginseng* rhizospheric soils and roots.

(ii). Fungi: Tan, *et al* (79) used the Illumina MiSeq to investigate the rhizospheric and root endophytic fungi in response to continuous Notoginseng Radix Et Rhizoma cropping practices. The results demonstrated that fungal diversity increased in the roots and in rhizosphere. In rhizospheric soil, the abundance from high to low followed the order : *Emericella*, *Fusarium*, *Plectosphaerella*, *Mycocentrospora*, *Cylindrocarpon*, *Penicillium*, *Aspergillus*, *Staphylotrichum*, *Trichoderma*, *Chaetomium* and *Trechispora*. Among the endophytic root fungi, the abundance from high to low followed the order : *Emericella*, *Fusarium*, *Plectosphaerella*, *Cylindrocarpon*, *Mycocentrospora*, *Staphylotrichum*, *Mortierella*, *Chaetomium*, *Gibberella* and *Penicillium*.

3.1.6.2. Rehmannia Radix (*Rehmannia glutinosa* Gaertn): Wu *et al* (96) found that after continuous cropping of Rehmannia Radix, the quantities of bacterial biomass improved with the increase of continuous cropping years and reached the peak at end of second year of continuous cropping. Bacteria, Gram (+), Gram (-), actinomycetes in the second and third years monoculture soils were significantly higher than in control and newly planted soils. Similar to certain specific PLFAs, the sum of fungal PLFAs significantly increased with the increasing years of monoculture, but decreased in the third year monoculture soil.

3.1.6.3. Heterophylly Falsestarwort Root (*Pseudostellaria heterophylla* Pax ex Pax et Hoffm): Wu, *et al* (95) found that continuous cropping of Heterophylly Falsestarwort Root significantly increased relative abundance of *Fusarium*, *Trichocladium*, *Myrothecium* and *Simplicillium* but significantly decreased the relative abundance of *Penicillium*. *Fusarium* is a pathogen causing root rot in traditional Chinese herbs (95,25). Moreover, the significant increase of *Fusarium oxysporum* was also confirmed by the quantitative PCR analysis. This study demonstrated that consecutive continuous cropping of Heterophylly Falsestarwort Root altered the fungal community in the rhizosphere, including enrichment of host-specific pathogenic fungi at the expense of plant-beneficial fungi.

3.1.7 Other crops

The changes in microbial community after continuous cropping of black pepper, potato, tobacco and other crops are discussed.

(i). Black pepper: Xiong, *et al.* (98) found that black pepper decreased the abundance of soil bacteria and changed the structure of soil microbial community during continuous cropping. The relative abundances of Bacteroidetes and Firmicutes phyla decreased with long-term continuous cropping, and at genus level, the *Pseudomonas* abundance was significantly reduced after 21 years continuous cropping.

(ii). Potato: Liu, *et al* (58) found that the diversity (Hshannon) and richness (Schaol) index of bacterial community reduced linearly in potato (*Solanum tuberosum* L.) continuous cropping for 7 years. Among them, the abundance of *Acidobacteria* and *Nitrospirae* were linearly decreased with the increase of continuous cropping years.

(iii). Tobacco: Long-term continuous cropping of tobacco (*Nicotiana tabacum* L.) significantly altered the soil microbial community, the bacterial diversity and decreased the evenness index with the extension of continuous cropping years. The abundance of beneficial bacteria (*Arthrobacter* and *Lysobacter*) was reduced significantly, however, the relative abundance of *Morphomyces* sp. increased, which could dissolve inorganic phosphorus in the soil (72,88).

3.2 GREENHOUSE CROPS

3.2.1 Cucumber

(i). Bacteria: Cucumber (*Cucumis sativus* L.) is usually grown in continuous cropping system in greenhouses. However, it is not sustainable in the long term. Liu, *et al* (57) found that the β -diversity of bacterial community, but not α -diversity, significantly changed with

consecutive years of cucumber cultivation and is positively linked to cucumber yield. The bacterial community members at phylum level, in prolonged cucumber cultivation increased the mean relative abundances of Chloroflexi and Gemmatimonadetes, while decreased the average relative abundance of Nitrospirae. At genus level, continuous cucumber cultivation decreased relative abundances of some beneficial microbes (i.e. *Bacillus*, *Solirubrobacter*, and *Rubrobacter*) and N-cycling related microbes (i.e. *Nitrospira* and *Azoarcus*), while increased the relative abundance of some functional microbes (i.e. *Agromyces*, *Thermomicrobium*, *Desulfotomaculum*, *Sphaerobacter*, and *Mycobacterium*). Due to high inputs of agricultural chemicals and manures year after year, prolonged greenhouse production may lead to the accumulation of soil pollutants (i.e. heavy metals, PAHs, pesticide residues and antibiotics), which probably increased the average relative abundances of these functional genera mentioned above. The bacterial community members were greatly influenced by significant changes in soil physico-chemical environment due to the long-term intensive greenhouse production i.e., soil physico-chemical variables significant affected the community structure. In brief, soil available phosphorus and organic carbon contents were the main contributors, followed by total nitrogen, available potassium, and C/N ratio. Zhao, *et al* (115) found that soil pH significantly decreased, whereas, soil EC, OM, and nutrient concentrations (TN, NO₃⁻-N, TP, AP, AK) significantly increased. The continuous cropping of cucumber for 8 or more years reduced the relative abundance of the dominant bacterial phyla Actinobacteria but increased the abundances of Acidobacteria and Firmicutes significantly than the cucumber planted for one season. Acidobacteria, normally identified as an oligotrophic (or K-selected) bacterial group in the soils (22), decreases with increasing N fertilization rates (106). However, the relative abundance of Acidobacteria significantly increased with the continuously continuous cropping years in this study. This was probably because the high N fertilization rates led to soil acidification, increasing the population of this oligotrophic bacterial group in this intensive continuous cropping system. Moreover, some groups of Acidobacteria are known as acidophiles (71), which also demonstrated a negative correlation trend with soil pH. Moreover, Zhou, *et al* (115) found that the predominant fungal phylum Ascomycota increased significantly with increasing cultivation years of cucumber.

(ii). Fungi: Cucumber continuous cropping significantly increased fungal pathogen community, especially *Fusarium*, in greenhouse pot experiments (114). Cropped the cucumber in pots under greenhouse conditions for nine successive cropping cycles. Structures and sizes of rhizosphere fungal and *Fusarium* (Ascomycota, Fungi) communities, both ubiquitous and ecologically important in soils, were analysed. The findings of this study indicated that cucumber showed slow growth in the seventh planting and the rhizosphere fungal and *Fusarium* communities were larger in the 7th planting cycle than other cycles. In addition to this, the findings also showed that the population sizes rather than the diversity of fungi and *Fusarium* communities were linked to the soil sickness associated with cucumber cultivation.

3.2.2 Strawberry

(i). Bacteria: Li, *et al* (49) found that Long-term continuous cropping of strawberry changed the bacterial abundance, the proportions of *Novosphingobium*, *Rhodoplanes*, *Povalibacter*, *Cellvibrio* and *Stenotrophobacter* decreased after continuous cropping of strawberry. While, the relative abundances of *Pelagibius*, *Thioprofundum*, *Allokutzneria* and *Bacillus* increased. The RDA results showed that the environmental variables were negatively associated with *Bacillus*, *Allokutzneria*, *Pelagibius* and *Thioprofundum* that increased and were positively related to *Novosphingobium*, *Povalibacter*, *Rhodoplanes* and *Stenotrophobacter*; *Sphingomonas* and *Nitrospira* were positively correlated with pH, organic matter, urease, invertase and alkaline phosphatase and were negatively correlated with $\text{NH}^{4+}\text{-N}$, AK, AP and TN. Chen, *et al* (15) found that the relative abundance of bacterial and fungal groups in different soils varied significantly with the years of continuous cropping. In the bacterial community, for example, *Bacillus* abundances were more in the second strawberry continuous cropping year than in other years but then gradually decreased with continuous cropping years. The abundance of *Bacillus* decreased significantly beginning in the 10th year. The abundance of *Sphingomonas* decreased slightly after 2-year and 4-year but increased significantly beginning in the 10th continuous year, and the abundance of *Sphingopyxis* increased significantly from the 8th strawberry continuous cropping year.

(ii). Fungi: Within the fungal community, the relative abundance of *Fusarium* and *Humicola* increased significantly beginning in the sixth year. Huang, *et al* (36) also reported that the dominant genera of beneficial bacteria and fungi (*Bacillus* and *Trichoderma*) were in the 1-year field, while the dominant genera of fusarium were in the 10-year field. There were significantly positive correlations ($p < 0.01$) between the abundances of beneficial species (*Bacillus subtilis* and *Trichoderma harzianum*) and soil microbial biomass C. On the other hand, the abundance of *Fusarium oxysporum* was positively ($p < 0.01$) correlated with soil moisture and EC. Li, *et al* (48) found that the soil environment changes significantly after continuous cropping of strawberry. At the same time, soil fungal diversity increased with consecutive cropping years. With the changing trend in soil physico-chemical properties, the soil nutrients content began to decline after the 4th year of continuous cropping. Continuous cropping reduced the AK, pH and $\text{NH}^{4+}\text{-N}$ content of the soil and altered the fungal community in the soil environment. Specifically, the soil-borne disease pathogens *Fusarium* and *Guehomyces* were significantly increased after strawberry continuous cropping, and the abundance of nematicidal (Arthrobotrys) fungi decreased from 4th year of continuous cropping.

3.2.3 Tomato

The large-scale production of tomato (*Lycopersicon esculentum* Mill.) in China in specific areas lead to the continuous cropping obstacle, increasing year by year. The quantities of bacteria and actinomycetes in continuous cropping soil decreased significantly, while the quantities of fungi increased. The microbial community structure was changed from bacterial type to fungal type (40,102). Lei, *et al* (43) study showed that the total number of soil microorganisms reduced significantly with the extension of crop years, and decreased

by 53.95 % than control (no continuous cropping) after 4 years of cropping. With the increase of crop years, the number of bacteria and actinomycetes were decreased significantly by 59.49 % and 19.36 %, respectively, after 4 years of continuous crop. The quantities of soil fungi decreased first and then increased. Through pot experiment, Sun, *et al* (78) reported that with the increase of continuous cropping years, the quantities of bacteria in continuous cropping for 2 and 3 years dropped by 12.1 % and 16.8 %, respectively, compared with that in continuous cropping for 1 year. While, the number of fungi in continuous cropping for 2 and 3 years increased by 20 % and 60 %, respectively, than in continuous cropping for 1 year. However, the quantities of actinomycetes increased after continuous cropping, and was 1.04 and 1.12 times higher in 2-year and 3-year soils than in 1-year soils, respectively.

5. FUTURE AREAS OF RESEARCH

- (i). More attention need be paid to the relationship between soil microorganisms and soil physico-chemical properties (soil pH, organic matter, etc.). Study the effects of soil physico-chemical properties and soil microorganisms on long-term continuous cultivation to provide a basis for future research on sustainable agricultural systems.
- (ii). Determine the impact of continuous cropping on abiotic factors (soil physico-chemical properties and autotoxic substances) and biological factors (soil microbial and nematode community). Analyze comprehensively why these obstacles occur after the continuous cropping and for the prevention and control of continuous cropping problems.
- (iii). Use the metagenomics to study the dynamic changes of functional genes during long-term continuous cropping of crops to improve our understanding of the mechanism of crop related soil sickness.

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DECLARATION

We declare that all authors of this Ms. have made substantial contributions. We did not exclude any author who substantially contributed to this Ms. We have followed our ethical norms established by our respective institutions.

CONFLICT OF INTEREST

The authors announce that they have no conflict of interest.

ETHICAL APPROVAL

The authors declare that the study was carried out following scientific ethics and conduct. However, this study did not involve any use of animals, hence no ethical approval has been obtained from the concerned committee.

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