

Effects of crop rotations on microbial community in rhizosphere soil of cucumber seedlings and its feedback

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ABSTRACT

We studied the effects of 7-crop rotations and continuous - monocropping systems on soil microorganism and its feedback. The results showed that absolute abundance of soil bacteria (*Pseudomonas* and *Bacillus*) in tomato - celery - cucumber - cabbage and cucumber - tomato - cucumber - cabbage rotation were significantly higher than control (CK). Absolute abundance of soil fungi in tomato - celery - cucumber - cabbage, kidney bean - celery - cucumber - cabbage, cucumber - kidney bean - cucumber - cabbage and cucumber - tomato - cucumber - cabbage rotation were significantly higher than CK. Dry weight of cucumber seedlings was significantly positively correlated with bacterial (*Pseudomonas* and *Bacillus*) abundance, and negatively correlated with fungal count. The results of inoculation with *Fusarium oxysporum* f.sp. *cucumerinum* showed that plant dry weight of cucumber seedlings in tomato - celery - cucumber - cabbage, cucumber - kidney bean - cucumber - cabbage, cucumber - tomato - cucumber - cabbage rotation soil was significantly higher than other treatments, and their disease index was significantly lower than other treatments. There was no significant difference in dry weight of cucumber seedlings in rotation and CK in the soil sterilization test. The results of plant - soil feedback experiment showed that soil microbial changes caused by different rotation patterns had a positive feedback effect on growth of cucumber seedlings.

Key words: Absolute abundance, *Bacillus*, bacteria, cabbage, celery, crop rotation, cucumber, disease Index, kidney bean, microbial community abundance, Plant-soil feedback, rhizosphere soil, seedlings, soil fungi, Soil microorganisms, tomato

INTRODUCTION

Cucumber (*Cucumis Sativus* L.) is commonly planted vegetable in the world. However, with continuous demand, its continuous cropping is widespread (24), despite the fact that its continuous cropping is not recommended. Continuous cropping for many years decreased the cucumber yield and aggravates soil-borne diseases. There is a significant negative correlation between the yield and continuous cropping years (38,48). After continuous cropping, microbial diversity of cucumber decreases, with the decline in beneficial bacteria (*Pseudomonas*, *Bacillus*) and increase in harmful fungi *Fusarium oxysporum* f.sp. *cucumerinum* (FOC), which adversely affects the cucumber growth, yield and quality (27,50). These soil sickness problems seriously restrict the sustainable development of vegetable production worldwide.

The continuous planting of the same crop or the same family on the same land for many years will lead to undesirable phenomena such as slow growth, decreased yield, decreased quality and serious diseases, called Soil Sickness (46,44), which has become a major factor restricting the sustainable development of agriculture. Soil quality is critical to

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healthy growth of crops (6), but, long-term continuous cropping deteriorates the soil physical and chemical properties and soil biological environment, causing soil microbial imbalance, which is not conducive to growth and development of crops (16,20,29). The long-term continuous cropping has significant impacts on the soil microbial abundance and community structure, decreasing the microbial diversity. Soil microbial types change from "bacterial type" to "fungal type". With extension of continuous cropping years, soil borne diseases become more and more serious year by year (15,43,40). In addition, long-term continuous cropping decreases large number of beneficial microorganisms and causes accumulation of pathogenic microorganisms, which destroys the microbial community structure (41,50). Now researchers have proposed crops rotations to prevent and control the continuous cropping problems, increase soil bacterial diversity, reduce the occurrence of diseases and thereby improve the cucumber yield. Among them, crop rotation with different crops is simple, safe and effective method to reduce the soil sickness problem.

Crop rotation is a pattern of planting different crops in same field in certain order. Different crops have variable needs for soil nutrients, and crop rotation with legumes improves the soil quality and fertility and ensures the effective use of land resources (31). It significantly increases the abundance of soil bacteria and improve soil community structure. Generally speaking, most pathogenic microorganisms have characteristics of specific parasitism. In crop rotation, the residual pathogens of previous crops are unable to find their hosts, thus gradually reducing their population in soil, to reduce the risk of soil borne diseases infecting crops. The crop rotation reduces the abundance of pathogenic bacteria and thereby effectively alleviates the soil borne diseases, improves the related enzyme activities and also regulates changes in soil microbial community and improves productivity of protected vegetable ecosystem (45). Abdel-Monaim and Abo-Elyousr (1) studied the effects of garlic on damping down and root rot of lentils by field rotation test and laboratory bacteriostatic test. The garlic rotation in field with lentil could significantly reduce the incidence rate and disease index of garlic and garlic root exudates significantly reduced the dry weight of pathogen mycelium. Li *et al* (22) showed that crop rotation used root exudates of different crops to kill or inhibit some harmful microorganisms, which cause diseases.

Soil microorganisms play a critical role in maintaining soil quality and regulating plant growth and development (7,19). The abundance and community structure of soil microorganisms depend on variety of factors (cropping systems, fertilization practices, crop varieties and land use types, etc.), especially in greenhouse cultivation (26). Compared with crop production in open field, greenhouse cultivation is usually characterized by high planting intensity, excessive fertilization, high humidity and relatively high soil temperature, which can change the physical and chemical environment of soil and feed back to soil microflora (27). Microbial ecological imbalance is one of main reasons leading to continuous - cropping problems (43,49). Compared with single cropping, rotation systems improve the abundance of soil microorganisms and changes in the community structure, thereby improving growing environment of crops (1). Therefore, it is necessary to monitor the changes of soil microbial community in greenhouse vegetable production and explore its potential relationship with continuous cropping problems. To this end, we used seven- crop rotations in our experiments to study their effects on (i). growth of cucumber seedlings, (ii). soil microorganisms and (iii). to find optimal rotation beneficial to cucumber growth.

MATERIALS AND METHODS

Four experiments (Pot culture, FOC inoculation, Soil sterilization, plant soil feedback) were conducted in greenhouse, Horticultural Station, Northeast Agricultural University, Harbin, China (45° 41' N, 126° 37' E), from June to August 2018. The soil samples up to 30 cm depth were collected from the 12-years old cucumber rotation experiment in June 2018. The detail of crops and order of crop rotation is given in Table 1.

The physico-chemical properties of soil were determined as previously described by Bao (2). Briefly, ammonia nitrogen (NH_4^+ -N), nitrate nitrogen (NO_3^- -N) and available phosphorus (AP) of the soil were measured by SKALAR (origin: USA, model San++) flow analyzer; available potassium (AK) by M3 extraction-flame photometry; while pH and EC (1:2.5, w/v) mS cm^{-1} were determined in soil-water leachates (1:5 ratio) using pH meter (model FE20, Shanghai) and conductivity meter (model FE30, Shanghai). The chemical properties of the soil are shown in Table 2.

Table 1. Test Experimental crops and varieties

#	Crop	Botanical name	Variety
1	Cucumber	<i>Cucumis Sativus</i> L.	Jinyan 4
2	Celery	<i>Apium graveolens</i> L.	Solid celery
3	Cabbage	<i>Brassica oleracea</i> L.	Dongnong 901
4	Kidney Bean	<i>Phaseolus vulgaris</i> L.	August green
5	Tomato	<i>Solanum lycopersicum</i> L.	Fende

Table 2 Chemical properties and number of tested soils

Crop rotation (spring-autumn-spring-autumn)	pH	EC ($\text{mS}\cdot\text{cm}^{-1}$)	NH-N ($\text{mg}\cdot\text{kg}^{-1}$)	NO_3^- -N ($\text{mg}\cdot\text{kg}^{-1}$)	AP ($\text{mg}\cdot\text{kg}^{-1}$)	AK ($\text{mg}\cdot\text{kg}^{-1}$)
Tomato - kidney bean - cucumber - cabbage (I)	6.57	0.28	19.72	152.05	106.42	263.63
Tomato - celery - cucumber - cabbage (II)	6.60	0.26	18.83	99.52	132.21	280.61
Kidney bean - tomato - cucumber - cabbage (III)	6.56	0.29	19.32	233.66	124.79	283.32
Kidney bean - celery - cucumber - cabbage (IV)	6.54	0.31	19.37	193.41	125.21	255.48
Cucumber - kidney bean - cucumber - cabbage (V)	6.58	0.27	19.94	69.97	129.50	298.26
Cucumber - celery - cucumber - cabbage (VI)	6.54	0.30	21.35	201.56	115.63	247.33
Cucumber - tomato - cucumber - cabbage (VII)	6.59	0.30	18.31	114.24	122.79	270.08
Cucumber - cucumber - cucumber - cucumber (CK)	6.52	0.25	20.66	284.66	80.34	297.92

I. Pot experiment

Cucumber (cv. Jinyan 4) seeds were soaked in warm water (55 °C) for 30 min and then germinated in sand in pots kept in growth chamber at 26 °C. Seedlings with two cotyledons were transplanted to plastic pots (10 cm x10 cm) containing 300 g of crop rotation or cucumber continuous cropping soil. There were 8-treatments consisting of 7-crop rotations and a cucumber continuous cropping system as control. Each treatment had 10 pots and the treatments were repeated thrice in complete randomized design. The pots were maintained in greenhouse (28 °C day/18 °C night, relative humidity of 70 %, 16 h light/8 h dark). Distilled water was applied to maintain the soil water content at about 60 % water holding capacity. Thirty days after transplanting, samples of cucumber seedlings biomass and rhizosphere soil were collected.

Seedlings biomass and collection of rhizosphere soil samples

Cucumber seedlings were harvested at 20 and 30 days after transplantation and the total plant dry weight was measured after oven drying at 70 °C to constant weight. For rhizosphere soil sample collection, cucumber roots were gently removed from the pots, and the soils loosely attached to cucumber roots were carefully removed by manual

shaking. Then, soil tightly adhering to the roots was removed from the root surface with a sterile brush and sieved (2 mm mesh). One portion of these fresh sampled soils was stored at -80°C for DNA extraction and other portion was used in plant-soil feedback experiment.

DNA extraction and quantitative PCR analysis

Total DNA was extracted from 0.25 g rhizosphere soil with Power Soil DNA extraction kit (MO Biological Laboratory, Carlsbad, USA). Each composite soil sample was extracted in triplicate and the extracted DNA solution was mixed together. The V3-V4 hyper variable region of 16S rRNA gene was used as the bacterial-specific fragment with the 338F/518R (5'-ACTCCTACGGGAGGAGCAG-3'/5'-ATTACCGCGGCTGCTGG-3') primer sets for bacteria (42). The 20 μl reaction system consists of 2 μl DNA, 9 μl SYBR Mixture, 0.2/0.2 μl (10 mM) upper/lower primer, and rest of volume is filled with deionized water. Pre-denaturation at 95°C for 5 min, denaturation at 95°C for 50 s, annealing at 62°C for 30 s, extension at 72°C for 1 min, 30 cycles, final extension at 72°C for 10 min. The length of amplified fragment of bacteria DNA was about 230 bp. The ITS1 region of fungal ITS gene was used as the fungal-specific fragment with the ITS1F/ITS2(5'-CTTGGTCATTTAGAGGAAGTAA-3'/5'-TCCTCCGCTTATTGATATGC-3') primer sets for fungi (3). The 20 μl reaction system consists of 2 μl DNA, 9 μl SYBR Mixture, 0.25/0.25 μl (10 mM) upper/lower primer, and rest of volume is filled with deionized water. Reaction conditions: pre-denaturation at 94°C for 5 min, denaturation at 94°C for 1 min, annealing at 58°C for 1 min, extension at 72°C for 1 min, 31 cycles, final extension at 72°C for 10 min. The length of amplified fragment of fungal DNA was about 750 bp. A standard curve was prepared using a 10-folds dilution series of plasmids containing bacteria, fungi, *Pseudomonas* and *Bacillus*. The threshold cycle (Ct) value of each sample was compared with standard curve to determine initial copy number of target gene. All expansions were in triplicate. Sterile water was used instead of templates as negative controls. The specificity of the product was confirmed by melt curve analysis and agarose gel electrophoreses.

II. FOC inoculation experiment

The *Fusarium oxysporum* f.sp. *cucumerinum* (FOC) was isolated and identified from a *Fusarium*-wilted cucumber plant in greenhouse. The strain was raised on potato-dextrose-agar (PDA) medium and conidia were obtained as described before (25). The soil used and procedures of cucumber germination, transplanting and treatment application was same as above pot experiment, however there were 20 pots for each replicate. Two days after transplanting, half of the cucumber seedlings (10 seedlings per replicate) were inoculated with FOC with a root-dipping method as previously described (18). Briefly, cucumber seedlings at 2-leaf stage were removed from soil and washed with sterile water. Then, root tips were cut off with a sterilized scissor and dipped in a FOC conidial suspension (10^6 CFU/ml) for 10 min. Thereafter, these inoculated cucumber seedlings were transplanted back in their original pots. Distilled water was applied to maintain the soil water content at about 60 % water holding capacity. Pots were randomly arranged and routinely managed. Seedlings for biomass and disease index were sampled at 20 and 30 days after inoculation. *Fusarium* wilt disease index was calculated using a scale containing six grades as suggested by Liu (25).

III. Soil sterilization experiment

Some part of soils collected from different crop rotations was sterilized thrice by autoclave (121°C , 30 min) in polythene sterilization bags and considered as sterilized soil, while fresh collected soil was considered as non-sterilized soil (17). Cucumber seeds were germinated and raised as described in above pot experiment. At 2-cotyledons stage, seedlings were transplanted into plastic pots (10 cm x 10 cm) containing 300 g soil

(sterilized, non-sterilized). Each treatment has 10 pots and all treatments were repeated thrice in complete randomized design. Sterilized distilled water (autoclave 121°C, 30 min thrice) was used to maintain soil water content at about 60 % water holding capacity. Cucumber seedling samples for measurement of biomass were collected at 10 and 20 days after transplantation.

IV. Plant-soil feedback experiment

The method of adding soil inoculum to sterilized normal soil was used to evaluate the effects of soil biota on cucumber seedling growth (5,21). Soil collected from greenhouse experiment was sterilized as described above and used as 'background soil', while fresh soil collected from different rotation systems was used as soil inoculum. Soils were mixed as under: (i). 94 % background soil + 6 % soil inoculum and (ii). 94 % background soil + 6 % sterilized soil inoculum. The soil inoculum was evenly mixed with the background soil and cultured in dark at 20 °C for 3 days to fully incubate the microorganisms in the mixed soil. The cucumber seedlings with 2-cotyledons were transplanted to plastic pots (10 cm x 10 cm) containing 300 g of mixed soils. There were eight treatments that were further repeated thrice with 10 pots per replicate. Sterilized distilled water was used to maintain soil water content at about 60 % water holding capacity. To determine biomass, cucumber seedlings were collected at 10 and 20 days after transplanting.

Statistical analysis

All data were checked for normality (Shapiro-Wilk's test) and homogeneity of variances (Levene's test). Data of microbial abundances from quantitative PCR analysis were logarithmically transformed. Then, these data were analyzed with one-way ANOVA, and means were compared between treatments by the Tukey's Honestly significant difference (HSD) test. The OriginPro 8.5 software was used to draw graph. Result differences were considered statistically significant at $P < 0.05$.

RESULTS AND DISCUSSION

Rhizosphere microflora of cucumber seedlings

Soil bacterial abundance in treatment II and VII were significantly higher than CK (Figure 1A), but in treatments II, III, IV and V, these were lower than control (Figure 1B). Soil *Pseudomonas* abundance in treatment II, IV, V and VII was significantly higher than CK (Figure 1C). Soil *Bacillus* abundance in treatment II, III, IV, V and VII was significantly higher than CK (Fig. 1D).

In different rotations the cucumber seedlings shoot, root, and plant dry weight were positively correlated with bacterial (*Pseudomonas* and *Bacillus*) abundance. While these were negatively correlated with fungal abundance (Table 3).

Different crop rotations significantly change the soil microhabitat of cucumber seedlings (36). According to qPCR results, bacterial absolute abundance of II (tomato - celery - cucumber - cabbage) and VII (cucumber - tomato - cucumber - cabbage) were significantly higher than CK (Fig. 2A), however it was lower than CK in treatment II (tomato - celery - cucumber - cabbage), III (kidney bean - tomato - cucumber - cabbage), IV (kidney bean - celery - cucumber - cabbage) and V (cucumber - kidney bean - cucumber - cabbage) ($p < 0.05$) (Figure 2B). Soil *Pseudomonas* spp. abundance in treatments II, IV, V and VII was significantly higher than CK ($p < 0.05$) (Fig. 2C). All treatments had significantly higher abundance of *Bacillus* spp. except treatment I than control ($p < 0.05$) (Fig. 2D). Correlation analysis showed that the cucumber seedlings

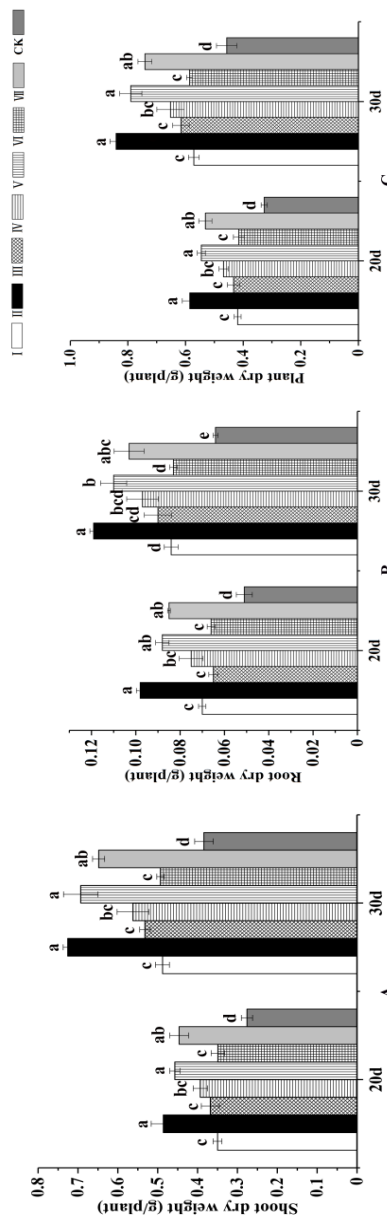


Figure 1. Effects of different rotation soil on cucumber seedlings shoot (A), root (B) and whole plant (C) dry weight. Data with different letters in each column indicate significant differences between treatments at 0.05 level.

Crop rotations : I: tomato - kidney bean - cucumber - cabbage, II: tomato - celery - cucumber - cabbage, III: kidney bean - tomato - cucumber - cabbage, IV: kidney bean - celery - cucumber - cabbage, V: cucumber - kidney bean - cucumber - cabbage, VI: cucumber - celery - cucumber - cabbage, VII: cucumber - tomato - cucumber - cabbage, CK: cucumber - cucumber - cucumber, same below.

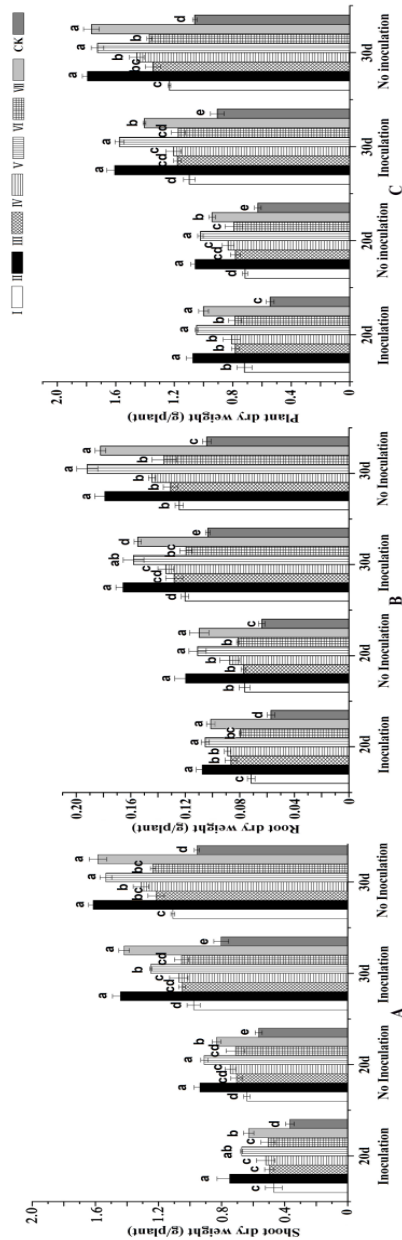


Figure 3. Effects of *Fusarium oxysporum* f.sp. *cucumerinum* on cucumber seedlings shoot (A), root (B) and whole plant (C) dry weight.

Table 3. Correlation coefficient between dry weight of whole cucumber seedlings and soil microorganisms

#	Bacteria abundance	Fungi abundance	<i>Pseudomonas</i> abundance	<i>Bacillus</i> abundance
Shoot dry weight	0.874**	-0.465*	0.875**	0.989**
Root dry weight	0.888**	-0.553*	0.867**	0.949**
Plant dry weight	0.716**	-0.459*	0.687**	0.679**

*Correlation is significant at the 0.05 level. **Correlation is significant at the 0.001 level.

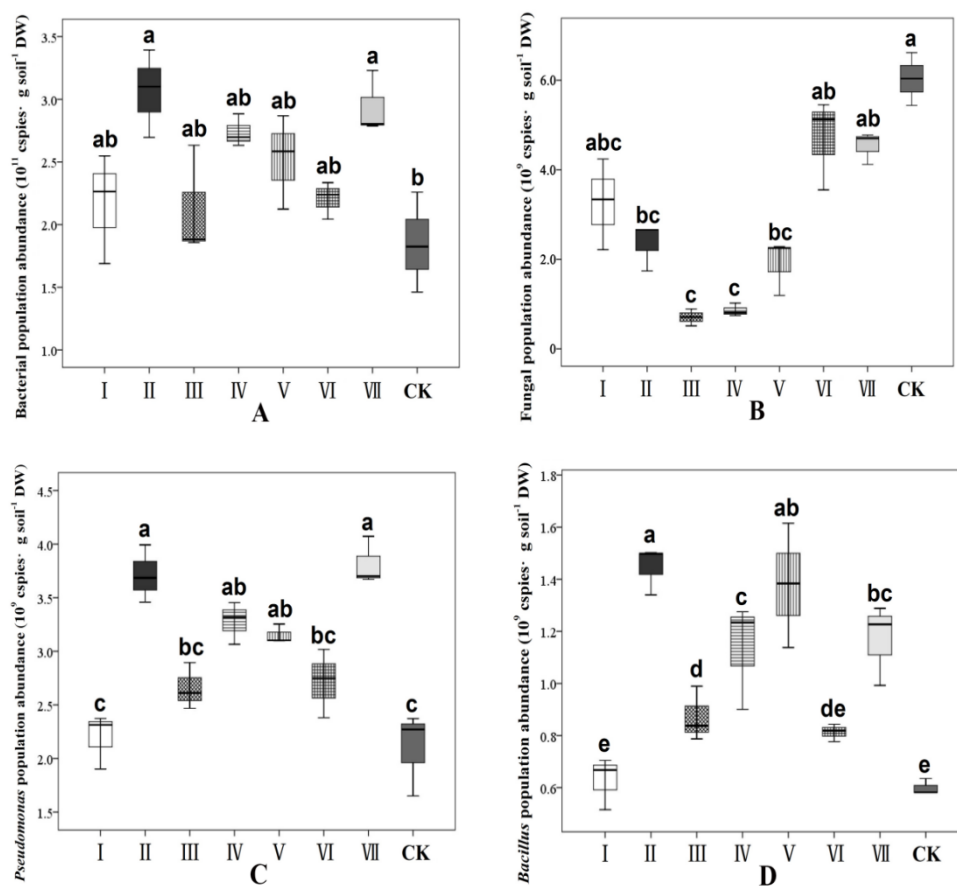


Figure 2. Effects of crop rotations on (A) Bacterial, (B) Fungal, (C) *Pseudomonas* and (D) *Bacillus* abundance in cucumber seedlings soil at 30 days after planting.

shoot, root, and whole plant dry weight were positively correlated with bacterial abundances of *Bacillus* and *Pseudomonas* spp. communities, while negatively correlated with fungal abundance (Table 3). These results are consistent with previous findings that crop rotation changes the soil microbial abundance (23,51). The plant dry weight of cucumber seedlings was significantly positively correlated with bacterial abundance,

Pseudomonas abundance and *Bacillus* abundance, while it was opposite to fungal abundance. It showed that after different crop rotations, increasing bacterial abundance and reducing fungal abundance helped in accumulation of dry matter in cucumber seedlings.

Cucumber seedlings Dry Weight

At 20 d and 30 d growth, the dry weights of shoots, roots, whole plants of cucumber seedlings were significantly higher than control and crop rotations II, V and VII (Fig. 2 A,B,C).

Inoculation v/s No Inoculation

- (i). **Shoot dry weight:** Comparison of the inoculation and no inoculation treatments II, V and VII showed an increase in shoot dry weight by 103.8, 83.1, 71.0 % and 65.9, 61.2, 47.3 % at 20 d respectively over CK. Comparison of the inoculation and no inoculation treatments II, V and VII showed an increase in shoot dry weight by 79.6, 55.7, 76.7 % and 68.9, 60.4, 65.7 % at 30 d respectively over CK (Fig. 2A).
- (ii). **Root dry weight:** Comparison of the inoculation and no inoculation treatments II, V and VII showed an increase in root dry weight by 88.4, 84.6, 77.5% and 87.3, 73.7, 71.9 % at 20 d respectively over CK. Comparison of the inoculation and no inoculation treatments II, V and VII showed an increase in shoot dry weight by 60.2, 52.6, 49.7 % and 71.6, 84.1, 74.8 % at 30d respectively over CK (Fig. 2B).
- (iii). **Whole plant dry weight:** Comparison of the inoculation and no inoculation treatments II, V and VII showed an increase in whole plant dry weight by 97.1, 92.5, 83.6 % and 68.1, 62.4, 49.8 % at 20 d respectively over CK. Comparison of the inoculation and no inoculation treatments II, V and VII showed an increase in shoot dry weight by 77.4, 74.0, 55.0 % and 69.1, 62.7, 66.6 % at 30 d respectively over CK (Fig. 2C).

Long term intensive continuous monocropping causes serious soil borne diseases, especially fungal diseases in greenhouse (35). The long-term continuous monocropping have significant impacts on number and community structure of microorganisms in soil, resulting in decrease in microbial diversity, shift of soil microbial types from "bacterial" to "fungal", and increase of soil-borne diseases year by year with extension of continuous cropping years (40,43). Ideal crop rotations can effectively promote the nutrients absorption of crops through different growth habits and root competition among crops, which is conducive to crop growth and development. The results of inoculation with FOC showed that plant dry weight of cucumber seedlings in II (tomato - celery - cucumber - cabbage), V (cucumber - kidney bean - cucumber - cabbage) and VII (cucumber - tomato - cucumber - cabbage) rotations soil was significantly higher than other treatments. This is because the four crops selected in early stage of rotation in this experiment were from 4-Families and Genera, hence, the root exudates of various plants were different. This improves the soil microbial community structure, improve the environment inhibitory to pathogenic bacteria, creating a better growth environment for cucumber seedlings (22). In addition kidney bean, a leguminous plant, has good effects on nitrogen fixation. It takes roots deeper in soil, absorbs nutrients from deeper soil, and at the same time recovers nutrients to top soil effectively (39). Celery is leafy green vegetable, its root system is shallower than cucumber, hence, has weak relationship with cucumber in nutrients competition, which helps the cucumber root system to absorb nutrients in soil, for better cucumber growth.

Disease index of cucumber seedlings

After 20 days of inoculation with *Fusarium oxysporum* f.sp. *Cucumerinum* (FOC), some cucumber plants began to show mild symptoms of wilt. The disease index of control fusarium wilt was significantly higher than crop rotations treatment at 20 d and 30 d after inoculation. The disease index of cucumber *Fusarium* wilt gradually increased with

extension of inoculation time (Figure 3).

Through ideal crop rotations the occurrence of soil borne diseases can be effectively minimised (22). The results showed that the ideal crop rotation significantly reduced the disease index of *Fusarium* Wilt of cucumber seedlings, which may be due to increase of bacteria (such as *Pseudomonas* and *Bacillus*) and decrease in harmful fungi in such rotation systems. It is known that some soil bacteria have antagonistic effects on pathogenic fungi. For example, two kinds of *Pseudomonas* isolated from rhizosphere of wheat inhibited the mycelial growth of some plant pathogenic fungi (10), increased abundance of *Bacillus* flora also had similar effects on *Fusarium graminearum* (13). Or it is because pathogen cannot find host in soil after the crop rotation, which effectively inhibits the occurrence of *Fusarium* Wilt (12). In addition, Chen found that reduction in number of pathogenic fungi accumulated in soil was related to change in soil microenvironment in different crops. This may be one of important reasons for reduction of cucumber *Fusarium* wilt incidence rate in crop rotation (8).

Effects of soil sterilization on cucumber seedlings

At 10 d and 20 d after cucumber planting, shoot, root and plant dry weight of non-sterilized cucumber seedlings in different rotations were significantly higher than non-sterilized control. Shoot, root and plant dry weight of non-sterilized treatments in crop rotations II, V and VII were significantly higher than in non-sterilized treatments I, III, IV and VI. Shoot and root dry weight of cucumber seedlings in various rotation treatments and CK were significantly higher than sterilization treatments. There were no significant effects of sterilization on dry weight of shoot, root and plant in various rotations treatments in 10 days and 20 days (Figure 4).

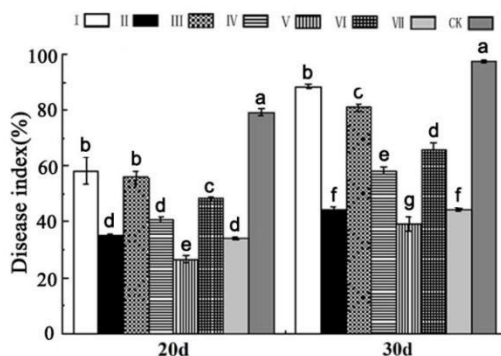


Figure 4 Effects of FOC on disease index of cucumber seedlings.

Soil microbial diversity has certain impact on crop growth and development (9,34). The diversity of soil microbial community structure plays a positive role in crop growth, which helps to solubilize the insoluble nutrients in soil and facilitates the absorption of nutrients by crops (28,37). Soil sterilization test showed that there was no significant difference in dry weight of cucumber seedlings between different crop rotation systems and control.

Effects of soil microorganism on cucumber growth in different rotations

At 10d and 20d of cucumber planting, shoot, root and plant dry weight of non-sterilized rotation treatments were significantly higher than those of sterilized rotation

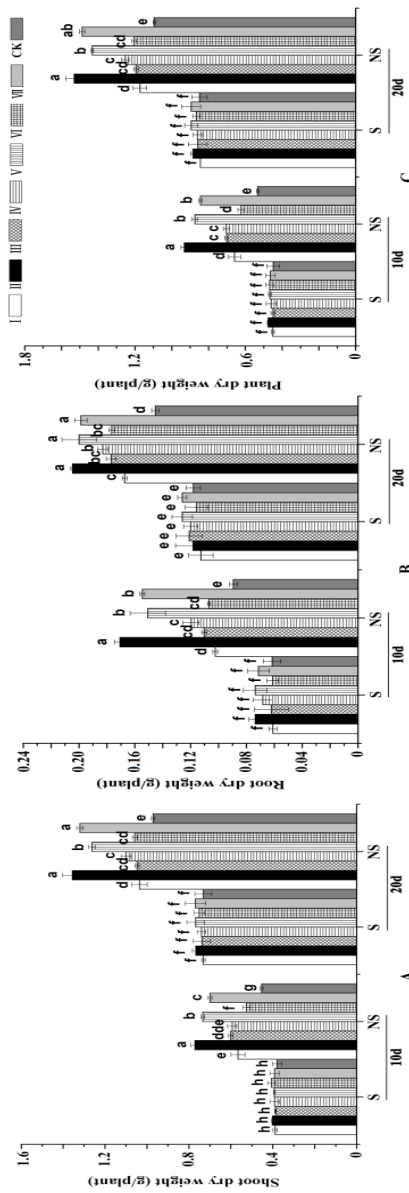


Figure 5. Effects of soil sterilization in different crop rotations on cucumber seedlings shoot (A), root (B) and whole plant (C) dry weight. S: Sterilized soil. NS: Non-sterilized soil in different crop rotations.

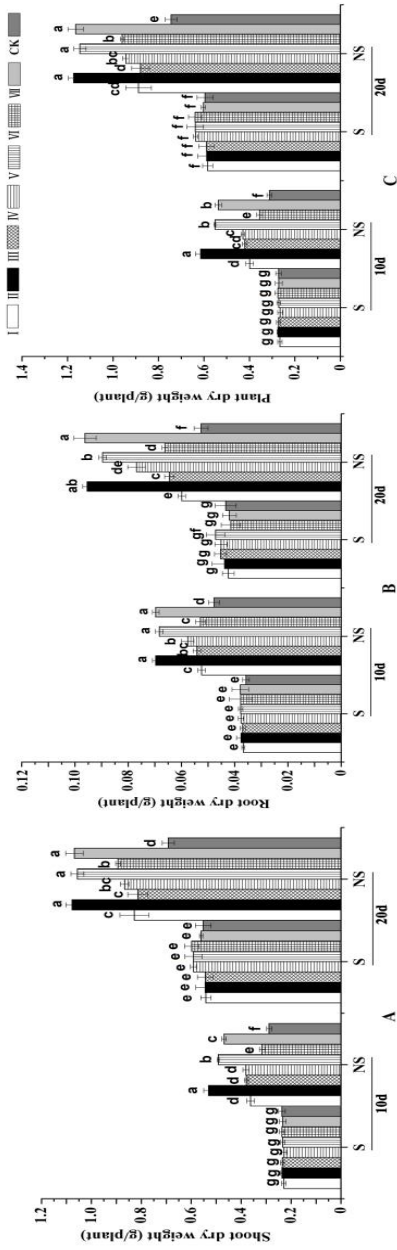


Figure 6. Effects of soil microorganism of different crop rotation on cucumber seedlings shoot (A), root (B) and whole plant (C) dry weight. S: Sterilized soil. NS: Non-sterilized soil in different crop rotations.

treatments. Shoot, root and plant dry weight of non-sterilized treatment II, V and VII were significantly higher than those of non-sterilized treatments I, III and VI at 10d. Shoot, root and plant dry weight of non-sterilized treatments II, V, VII was significantly higher than those of non-sterilized treatments I, III, IV and VI, and shoot, root and plant dry weight of non-sterilized cucumber seedlings in different rotations and CK were significantly higher than corresponding sterilized treatments at 20d. After sterilization, there was no significant difference in dry weight of cucumber seedlings between shoot, root and plant in two sampling periods (Figure 5).

Plant-soil feedback experiments showed that compared with sterilized soil, inoculation of soil microbes in continuous monocropping and different rotations increased the dry weight of cucumber plants, which may be due to the enrichment of soil microorganisms to promote the uptake of nutrients by crops (14,37), or because the reproduction of pathogenic microorganisms in soil was inhibited. When abundance of some beneficial microorganisms is high, their communities inhibits the pathogenic microorganisms, hence, they cannot harm the crops (11), thus creating a soil microbial environment more conducive to cucumber growth. After soil sterilization, elimination of soil microbial flora affected the transformation of nutrients in soil, inhibited the uptake of soil nutrients by cucumber, and thereby inhibited the growth and development of cucumber (34), which further proved the important role of soil microbial diversity in crop nutrients uptake.

CONCLUSIONS

The different crop rotations, significantly changed the abundance of soil microbial communities, however, it was balanced. The growth of cucumber seedlings could be predicted by measuring the abundance of soil microorganisms. The dry matter of cucumber seedlings was closely related to abundance of soil microbial communities. The crop rotations significantly improved the resistance of cucumber seedlings to *Fusarium* wilt. The plant dry weight was similar in sterilized soil, crop rotations and control i.e. soil sterilization was not beneficial to cucumber plants growth. Plant - soil feedback experiment showed that crop rotations changed the soil microbial patterns, which had positive feedback effect on cucumber seedling growth. The three-crop rotations: tomato - celery - cucumber - cabbage, cucumber - kidney bean - cucumber - cabbage, and cucumber - tomato - cucumber - cabbage rotations improved the cucumber seedlings biomass, regulated the soil microbial community abundance, increased the number of beneficial bacteria, reduced the cucumber disease, and built good ecological environment for cucumber growth.

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