

Effects of Continuous Cropping of *Polygonatum odoratum* on Microbial Functional Diversity and Selected Environmental Factors in the Rhizosphere Soil

Manjiang YU, Yihong HU, Fan XIE, Xiongmei ZHU, Chenzhong JIN*, Jing YE*

Key Laboratory of Green Control of Crop Pests, Hunan University of Humanities, Science and Technology, Loudi 417000, China

Abstract [Objectives] This study was conducted to investigate the mechanism of continuous cropping obstacles in *Polygonatum odoratum*. [Methods] Three treatments were established: continuous cropping (two consecutive crops), first-crop control (with *Phaseolus vulgaris* as the preceding crop), and blank control. The effects of continuous cropping on the functional diversity of soil microorganisms, soil enzyme activities, and soil nutrient coordination in the rhizosphere soil of *P. odoratum* during different growth stages were investigated. [Results] (1) Continuous cropping increased the carbon source metabolic capacity, Shannon diversity index, and richness of rhizosphere soil microorganisms by 3.2%–14.7%, 0.9%–3.5%, and 1.3%–12.5%, respectively, but the differences were not significant. (2) Principal component analysis indicated that during the middle stage of rhizome expansion, continuous cropping significantly altered the characteristics of microbial carbon metabolism, and the microbial communities utilizing carbohydrates, amino acids, polymers, carboxylic acids and amines as carbon sources exhibited vigorous metabolism. (3) Continuous cropping significantly reduced the activities of urease, polyphenol oxidase, and acid phosphatase in rhizosphere soil, with decreases of 24.4%–39.5%, 3.2%–14.8%, and 7.9%–18.2%, respectively. The activities of catalase and invertase sometimes exceeded and sometimes fell below those of the first crop, showing no consistent pattern. (4) Under continuous cropping conditions, nutrient imbalance occurred in the rhizosphere soil, characterized by nitrogen deficiency and phosphorus surplus. (5) Grey correlation analysis indicated that available phosphorus content, alkali-hydrolyzable nitrogen content and polyphenol oxidase activity in rhizosphere soil were the main factors influencing microbial functional diversity. [Conclusions] This study provides a theoretical basis for understanding the formation mechanism of continuous cropping obstacles in *P. odoratum*.

Key words *Polygonatum odoratum*; Continuous cropping; Rhizosphere soil; Microbial function

DOI:10.19759/j.cnki.2164–4993.2025.05.004

Due to limited arable land per capita in China, highly intensive farming practices and the specific ecological and climatic requirements of certain crops, crop rotation is often challenging. Consequently, continuous monoculture or multiple cropping with the same crop is common. Continuous cropping obstacles occur to varying degrees in a range of crops, including grain and oil crops (such as soybean, potato, corn, and sesame), greenhouse vegetables (such as tomato and pepper), economic crops (such as tobacco), and medicinal plants (such as *Panax notoginseng*, *Pseudostellaria heterophylla*, and *Rehmannia glutinosa*)^[1–2]. *P. odoratum* is a perennial herbaceous medicinal plant of the Liliaceae family, commonly used as a component in traditional Chinese medicine. Under continuous cropping conditions, the plants exhibit stunted growth, yellowing leaves, and a high incidence of leaf spot disease and root rot, resulting in a significant decline in both yield and quality^[3]. These effects adversely impact planting profitability and industry development. Therefore, resolving the continuous cropping obstacles of *P. odoratum* holds significant theoretical value and practical importance. Continuous cropping obstacles result from the combined effects of the rhizosphere microecology, soil, and the plant itself. The causes of

these obstacles include: imbalance in the rhizosphere microecology, autotoxic effects of root exudates, deterioration of soil physical and chemical properties, and soil nutrient imbalance or depletion^[4–5]. Research indicates that continuous cropping leads to a decrease in the bacteria/fungi ratio in the soil where *R. glutinosa* is cultivated, a reduction in the population of beneficial microbial communities, a large number of pathogenic bacteria and fungi, and a transformation from "bacterial-type" soil to "fungal-type" soil^[6]. Previous studies on the effects of continuous cropping of *P. odoratum* on the rhizosphere microecology have been limited and not sufficiently in-depth. In this study, rhizosphere soil samples from four growth stages of a second cropping cycle (continuous cropping) of *P. odoratum* were investigated, with soil from the first cropping cycle (non-continuous cropping) and fallow soil as controls, aiming to elucidate the impact of continuous cropping on the functional diversity of soil microorganisms, soil enzyme activities, and nutrient coordination. This study provides a theoretical basis for understanding the mechanisms behind the continuous cropping obstacles in *P. odoratum*.

Materials and Methods

Materials

Experimental materials The tested material was the *P. odoratum* cultivar 'Zhushiwei'. The experimental area was located in Gutang Village, Gutang Town, Lianyuan City, Hunan Province. In October 2015, a *P. odoratum* field that had been transplanted

Received: July 5, 2025 Accepted: September 7, 2025

Manjiang YU (1998–), male, P. R. China, master, devoted to research about agricultural biotechnology.

* Corresponding author.

in October 2011 and harvested in October 2013 and October 2015 (with seeds simultaneously retained) was selected as the continuous cropping treatment group (CC, representing two cropping cycles). Meanwhile, adjacent vegetable land and fallow land were selected as the non-continuous cropping treatment group (FC, with *Phaseolus vulgaris* as the preceding crop) and the blank control group (CK), respectively. A large-plot design was adopted without replicates. Each plot covered an area of 150 m². Only weeding was performed in the control plot, while all other treatments received routine field management.

Sample collection Before planting (October 5, 2015), basic topsoil samples (0–15 cm depth) were collected from each treatment using a multi-point sampling method, with three replicates. Sampling was also conducted during the flowering stage (May 12, 2016), the middle stage of rhizome expansion (July 9, 2016), and the withering stage (September 25, 2016) of the *P. odoratum* plants. During each sampling, 15 plants were selected, and the rhizosphere soil adhering to the roots was collected using the brushing method. The soil samples from the 15 plants were mixed to form one composite sample, and this sampling process was repeated three times to yield three replicates. One half of each soil sample was air-dried naturally for the determination of enzyme activities and available nutrient content, while the other half was stored at –80 °C in a freezer for subsequent Biolog analysis.

Methods

Biolog experiment Fresh soil samples equivalent to 10 g of oven-dried soil were weighed, and 90 ml of sterile NaCl solution (0.85%) was added. The containers were sealed with sealing film and shaken at 180 r/min for 30 min. The soil suspension was then diluted to 10^{–3}. Subsequently, 150 µl of the dilution was inoculated into each well of the Biolog-eco plates. The plates were incubated at 28 °C, and the optical density at 590 nm was measured every 24 h using a Biolog automated microbial identification system.

Determination of soil enzyme activities and available nutrients

Soil urease activity was determined using the sodium phenolate colorimetric method. The determination of polyphenol oxidase activity adopted the pyrogallol colorimetric method. Catalase activity was assessed by the potassium permanganate titration method. Invertase activity was determined using the 3,5-dinitrosalicylic acid colorimetric method. Acid phosphatase activity was measured by the sodium phenyl phosphate method^[7]. Soil alkali-hydrolyzable nitrogen content was determined by the alkali hydrolysis diffusion method. The determination of available phosphorus content adopted HCl-NH₄F extraction-molybdenum-antimony anti-colorimetric method. Available potassium content was determined by the NH₄OAC extraction-flame photometry method^[8].

Data statistics and analysis The calculation method for the average color change rate of the wells in the Eco plates;

$$\text{AWCD (average well color development)} = \sum \frac{(C - R)}{n}$$

In the formula, *C* is the optical density value of the substrate well; *R* is the optical density value of the control well; and *n* is the number of carbon source substrates in the Eco plate (*n* = 31).

The metabolic intensity of carbon source was estimated using the curve integration method^[9]:

$$\text{Trapezoidal area } S = \sum \left[(v_i + v_{i-1}) \times \frac{t_i - t_{i-1}}{2} \right]$$

In the formula, *v* is the AWCD value at time *i*.

In this study, the optical density values from the Biolog-Eco plates at 72 h was used to characterize the microbial carbon metabolic diversity, including carbon source utilization richness, the Shannon diversity index, and principal component analysis (PCA).

$$\text{Shannon index} = - \sum p_i \ln p_i \quad P_i = \frac{C - R}{\sum (C - R)}$$

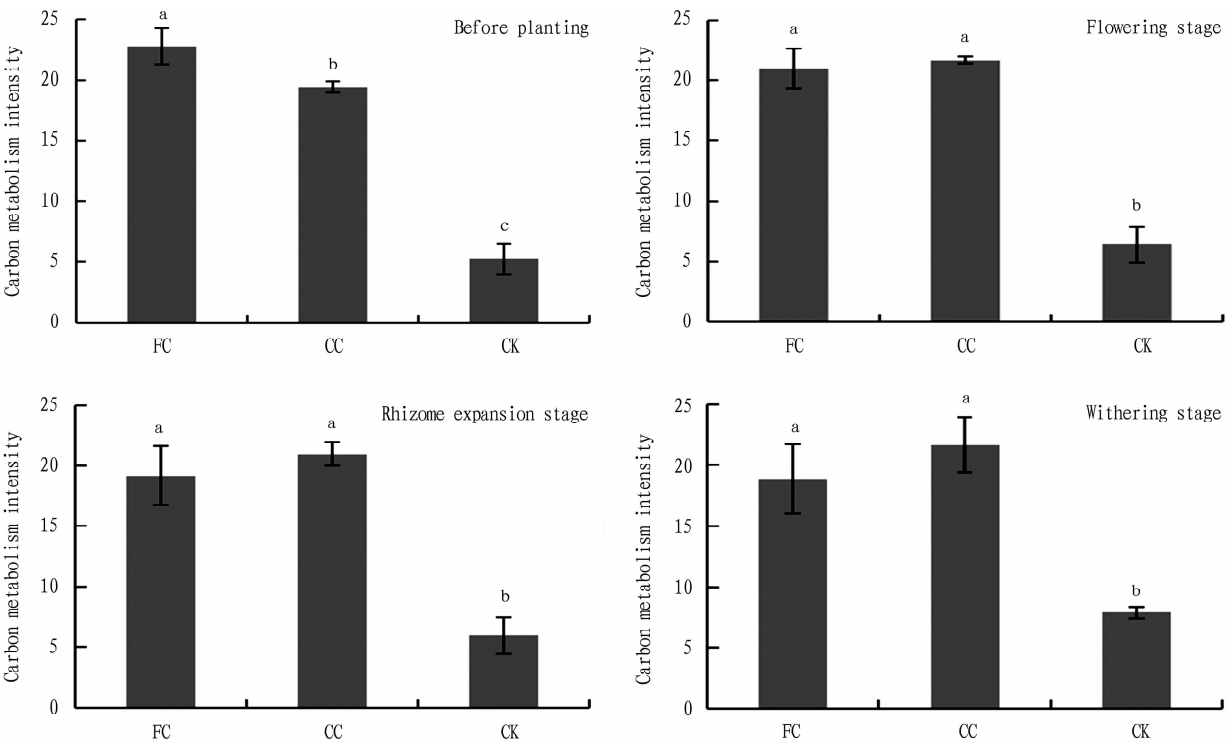
In the formula, *p_i* is the ratio of the relative absorbance value of the *i*th well to the sum of the relative absorbance values of the entire plate.

Richness was defined as the number of wells with an optical density value (*C* – *R*) greater than 0.25.

Results and Analysis

Effects of continuous cropping on the carbon metabolic intensity of rhizosphere soil microorganisms

There were significant differences in the carbon metabolic intensity of soil microorganisms between the fallow land and the *P. odoratum* cultivated fields. As shown in Fig. 1, before continuous cropping of *P. odoratum*, the microbial carbon metabolic intensity of rhizosphere soil was highest in the non-continuous cropping group (FC), significantly higher than that in both the continuous cropping group (CC) and the blank control group (CK). The carbon metabolic intensity in the continuous cropping group was significantly higher than that in the blank control group. During the flowering, rhizome expansion, and withered stages of *P. odoratum*, there was no significant difference in the microbial carbon metabolic intensity of rhizosphere soil between the non-continuous cropping (FC) and continuous cropping (CC) groups, but both were significantly higher than the blank control group (CK). The carbon metabolic intensity in the continuous cropping group was 3.2%, 9.3%, and 14.7% higher than that in the non-continuous cropping group during the three stages, respectively, but these differences were not statistically significant. However, it was significantly higher than the blank control group by 238.6%, 254.2%, and 173.8%, respectively, showing statistically significant differences. These results indicated that continuous cropping of *P. odoratum* affected the carbon metabolic intensity of rhizosphere soil microorganisms.



FC: Continuous cropping; CC: Non-continuous cropping; CK: Blank control.
Different lowercase letters on the bars indicate significant differences at the level of 0.05. The same below.

Fig. 1 Carbon metabolism intensity of microbial community

Effects of continuous cropping on the diversity indices of carbon source utilization by rhizosphere soil microorganisms

The cultivation of *P. odoratum* influenced the Shannon diversity index and richness of carbon source utilization by rhizosphere soil microorganisms, but no significant difference was observed between the continuous and non-continuous cropping groups. As shown in Table 1, in the continuous cropping treatment group, both the Shannon diversity index and richness of carbon source utilization by rhizosphere soil microorganisms were higher than those in the non-continuous cropping group at all stages: before planting, flowering stage, rhizome expansion stage, and withering stage. The Shannon diversity index increased by 0.9% , 0.9% ,

3.5% and 1.9% , respectively, while richness increased by 2.0% , 1.3% , 8.8% and 12.5% , respectively. However, none of these increases reached a significant level. In both the continuous and non-continuous cropping groups, the Shannon diversity index and richness of carbon source utilization by rhizosphere soil microorganisms were significantly higher than those in the blank control group in all four stages. Cultivating *P. odoratum* significantly increased both the Shannon diversity index and richness of carbon source utilization by rhizosphere soil microorganisms. However, there was no significant difference between the continuous cropping treatment group and the non-continuous cropping treatment group.

Table 1 Diversity of microbial function

Treatment	Before planting		Flowering stage		Rhizome expansion stage		Withering stage	
	Shannon diversity index	Richness	Shannon diversity index	Richness	Shannon diversity index	Richness	Shannon diversity index	Richness
FC	3.30 ±0.06 a	29.7 ±0.6 a	3.33 ±0.00 a	30.3 ±0.6 a	3.17 ±0.06 b	27.3 ±1.2 a	3.19 ±0.05 a	26.3 ±3.1 a
CC	3.33 ±0.06 a	30.3 ±1.2 a	3.36 ±0.02 a	30.7 ±0.6 a	3.28 ±0.06 a	29.7 ±1.5 b	3.25 ±0.04 a	29.0 ±1.7 a
CK	2.83 ±0.11 b	8.3 ±4.7 b	2.77 ±0.05 b	8.3 ±1.2 b	2.76 ±0.05 c	7.7 ±3.1 c	2.92 ±0.14 b	9.3 ±11.2 b

Effects of continuous cropping on the carbon metabolism patterns of rhizosphere soil microorganisms

The results from the 31 carbon sources formed multivariate vectors describing the metabolic characteristics of the microbial communities, which are not easily compared directly. Table 2 presents the statistical results after applying Principal Component

Analysis (PCA) to reduce the dimensionality of the carbon source absorbance data from before planting and the flowering, rhizome expansion, and withering stages. As shown in Table 2, the first two principal components extracted explained 70.29% , 84.70% , 66.00% , and 61.78% of the total variance, respectively.

Table 2 Contribution rates of principal components

Stage	PC1 eigenvalue	PC2 eigenvalue	PC1 contribution rate//%	PC2 contribution rate//%	Cumulative contribution rate//%
Before planting	20.04	1.75	64.64	5.65	70.29
Flowering stage	24.07	1.16	77.66	3.75	84.70
Rhizome expansion stage	18.14	2.32	58.52	7.49	66.00
Withering stage	16.56	2.59	53.43	8.35	61.78

The carbon source utilization patterns of soil microorganisms in the fallow land were distinctly different from those in the first cropping and continuous cropping groups at all four stages. As shown in Fig. 2, during the rhizome expansion stage, the scores of the continuous cropping treatment group on the PC1 axis were extremely significantly higher than those of the non-continuous cropping treatment group ($F = 14.073$, $\text{Sig} = 0.002$). Similarly, the scores of the continuous cropping treatment group on the PC2 axis were also extremely significantly higher than those of the

non-continuous cropping group ($F = 14.392$, $\text{Sig} = 0.002$). During other three stages, there were no significant differences in the scores on either the PC1 or the PC2 axis between the continuous cropping treatment group and the non-continuous cropping treatment group. At all four stages, there were extremely significant differences in the scores on both the PC1 and PC2 axes between the fallow land and both the continuous and non-continuous cropping treatment groups.

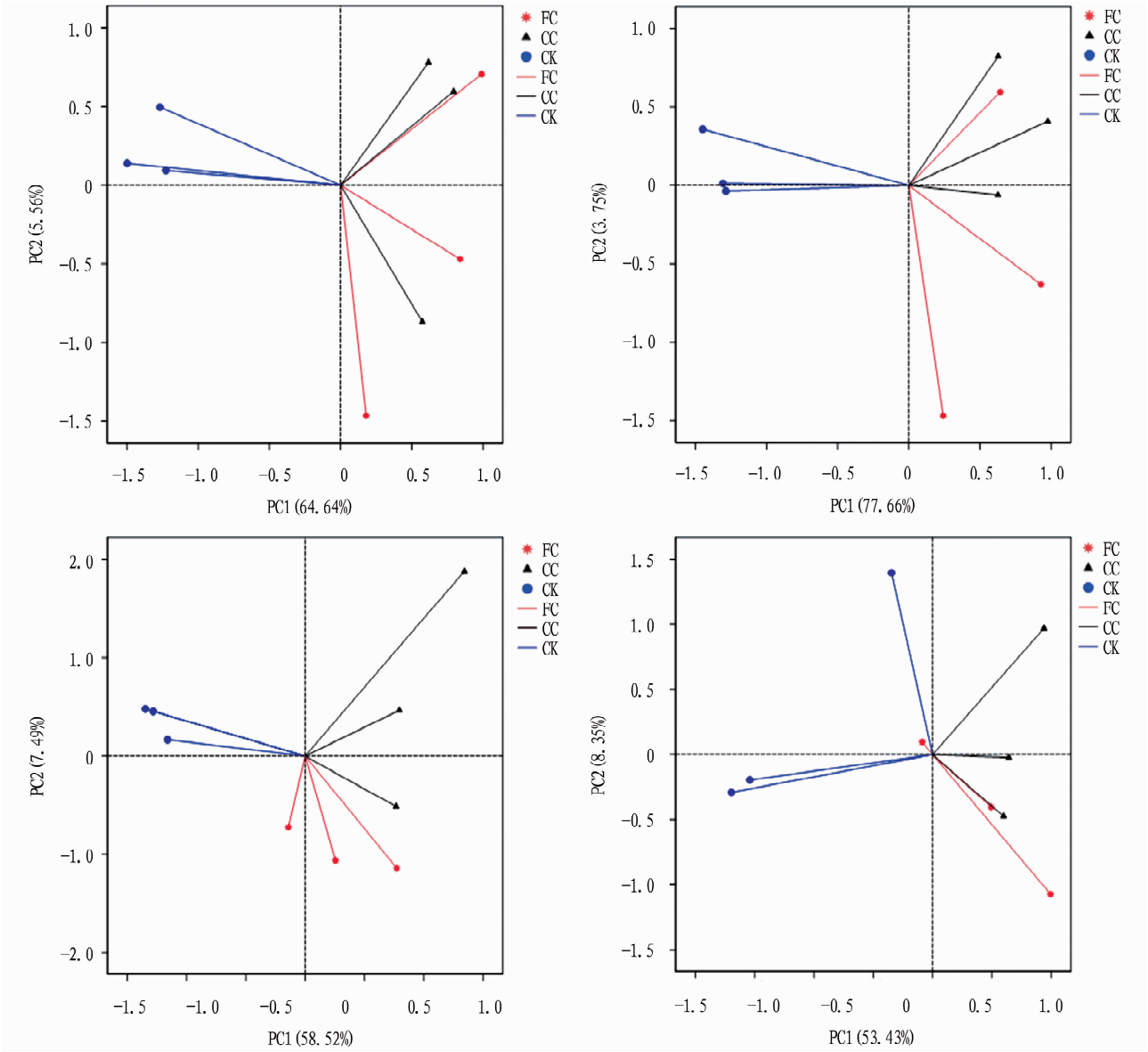


Fig. 2 Principal component analysis of microbial carbon metabolism

Based on the different metabolic pathways of microorganisms towards the 31 carbon sources in the Eco plates, the carbon source substrates were categorized into six types: carbohydrates (12 kinds), amino acids (6 kinds), carboxylic acids (5 kinds), polymers (4 kinds), phenolic compounds (2 kinds), and amines (2 kinds). Based on the principal component analysis results mentioned above, during the rhizome expansion stage, 28 carbon sources had a significant impact (loading value > 0.6) on PC1, while only three carbon sources, 2-hydroxybenzoic acid (phenolic acids), 4-hydroxybenzoic acid (phenolic acids), and α -ketobutyric acid (carboxylic acids), had no great impact on PC1. No carbon sources had a significant impact on PC2. It indicated that during the rhizome expansion stage, the microbial communities in the rhizosphere soil of continuous cropping exhibited vigorous metabolism of carbon sources such as carbohydrates, amino acids, polymers, carboxylic acids, and amines (Table 3).

Table 3 Loading values of principal components of correlative carbon sources

PC1-related carbon source	Loading value
Methyl β -D-glucoside	0.880
D-Galactonic acid γ -lactone	0.847
L(+)-Arginine	0.801
Methyl pyruvate	0.610
D-xylose	0.848
D-Galacturonic acid	0.797
L-Asparagine	0.924
Tween 40	0.782
i-Erythritol	0.721
L-Phenylalanine	0.661
Tween 80	0.705
D-Mannitol	0.875
L-Serine	0.906
α -Cyclodextrin	0.652
N-Acetyl-D-glucosamine	0.923
γ -Hydroxybutyric acid	0.759
L-Threonine	0.760
Glycogen	0.684
D-Glucosamic acid	0.800
Itaconic acid	0.664
Glycyl-L-glutamic acid	0.676
D-Cellobiose	0.849
α -D-Glucose-1-phosphate	0.861
phenylethylamine	0.804
α -Lactose	0.771
D,L- α -Glycerophosphate	0.909
D-Malic acid	0.767
Putrescine	0.678

Effects of continuous cropping on rhizosphere soil enzyme activities

Continuous cropping of *P. odoratum* significantly affected

rhizosphere soil enzyme activities. As shown in Table 4, the urease activity in the rhizosphere soil of the continuous cropping treatment was significantly lower than that in the non-continuous cropping treatment during the three stages (flowering stage, rhizome expansion stage, and withering stage), with decreases of 33.1%, 39.5%, and 24.4%, respectively. The acid phosphatase activity in the continuous cropping treatment was significantly lower than that in the non-continuous cropping treatment during the three stages, with decreases of 18.2%, 17.9%, and 7.9%, respectively. The polyphenol oxidase activity in the continuous cropping treatment was significantly lower than that in the non-continuous cropping treatment during the flowering and rhizome expansion stages, decreasing by 14.8% and 5.6%, respectively. During the withering stage, it decreased by 3.2% compared with the non-continuous cropping treatment, but this difference was not statistically significant. The catalase activity in the continuous cropping treatment increased by 10.5% and 10.2% during the flowering and withering stages compared with the non-continuous cropping treatment, but these differences were not statistically significant. However, it decreased significantly by 12.8% during the rhizome expansion stage. The invertase activity in the continuous cropping treatment increased significantly by 104.7% and 42.4% during the flowering and withering stages compared with the non-continuous cropping treatment, respectively, but decreased significantly by 39.1% during the rhizome expansion stage. These experimental results indicated that continuous cropping of *P. odoratum* had a substantial impact on rhizosphere soil enzyme activities.

Effects of continuous cropping on available nutrients in rhizosphere soil

Continuous cropping of *P. odoratum* significantly affected the available nutrients in the rhizosphere soil. As shown in Table 5, the alkali-hydrolyzable nitrogen content in the rhizosphere soil of the continuous cropping treatment decreased by 8.5%, 7.5%, and 3.2% during the flowering, rhizome expansion, and withering stages, respectively, compared with the non-continuous cropping treatment. Significant differences were observed during the flowering and rhizome expansion stages. The available phosphorus content in the rhizosphere soil of the continuous cropping treatment increased significantly by 60.5%, 30.2%, and 56.1% during the flowering, rhizome expansion, and withering stages, respectively, compared with the non-continuous cropping treatment. The available potassium content in the continuous cropping treatment showed minor changes during the flowering and withering stages compared with the non-continuous cropping treatment, with no significant differences, but increased significantly by 30.6% during the rhizome expansion stage. These experimental results indicated that continuous cropping of *P. odoratum* led to substantial changes in the available nutrient contents of the rhizosphere soil.

Table 4 Rhizosphere soil enzyme activities

Stage	Treatment	Urease//mg/g	Polyphenol oxidase mmol/(g·h)	Catalase//ml/g	Invertase mg/(g·h)	Acid phosphatase nmol/(g·h)
Flowering stage	FC	586.2±26.3 a	57.9±0.9 a	16.2±1.0 b	17.0±0.1 c	28.0±0.1 a
	CC	392.1±32.2 b	49.3±0.8 b	17.9±1.0 b	34.8±0.2 a	22.9±0.8 b
	CK	380.3±42.1 b	50.2±1.5 b	19.7±0.6 a	20.0±0.4 b	22.0±0.6 b
Rhizome expansion stage	FC	477.3±23.2 a	48.3±0.9 b	19.5±1.0 a	18.4±0.3 a	24.0±1.0 a
	CC	288.6±17.1 b	45.6±0.4 c	17.0±0.2 b	11.2±0.2 b	19.7±0.4 b
	CK	156.3±5.4 c	49.6±0.5 a	4.3±0.5 c	7.6±0.1 c	12.9±0.4 c
Withering stage	FC	394.4±38.2 a	49.3±0.3 a	15.7±1.0 b	14.6±0.2 c	23.9±0.5 a
	CC	298.2±25.7 b	47.7±1.2 a	17.3±1.0 b	20.8±0.2 a	22.0±0.3 b
	CK	289.7±7.8 b	39.3±1.3 b	20.4±1.1 a	20.2±0.2 b	15.5±0.3 c

Table 5 Rhizosphere soil available nutrients

Stage	Treatment	Alkali-hydrolyzable nitrogen//mg/kg	Available phosphorus//mg/kg	Available potassium//mg/kg
Before planting	FC	63.6±2.3 a	122.5±989 b	20.5±2.1 a
	CC	56.1±1.5 a	170.6±5.2 a	21.1±2.4 a
	CK	35.1±1.1 b	53.5±1.0 c	19.1±1.6 a
Flowering stage	FC	91.5±0.9 a	161.4±4.4 b	34.0±2.0 b
	CC	83.7±3.0 b	259.1±2.7 a	34.7±1.9 b
	CK	68.5±2.0 c	90.9±4.2 c	46.8±0.9 a
Rhizome expansion stage	FC	93.6±1.0 a	152.2±6.8 b	25.5±0.7 b
	CC	86.6±1.0 b	198.1±3.6 a	33.3±1.3 a
	CK	52.7±1.0 c	52.1±1.5 c	27.0±0.6 b
Withering stage	FC	72.0±2.0 a	144.5±9.9 b	22.5±1.5 a
	CC	69.7±3.0 a	225.6±5.2 a	25.1±2.0 a
	CK	45.2±1.2 b	58.5±1.6 c	22.1±2.0 a

Correlation between microbial parameters and environmental factors

Due to the differing units of measurement for microbial parameters and environmental factors, each indicator was normalized using the mean value method. Grey correlation analysis was performed with $\Delta_{\min}=0$ and a resolution coefficient $\beta=0.5^{[10]}$. As shown in Table 6, both rhizosphere soil enzyme activities and soil

nutrient contents showed high correlation with the carbon metabolism capacity and diversity of the microbial community. The most influential factor on microbial carbon metabolism capacity and richness was the soil available phosphorus content, while the most influential factors on the Shannon index were polyphenol oxidase activity and alkali-hydrolyzable nitrogen content.

Table 6 Correlation between microbial parameters and some environmental factors

Treatment	Carbon metabolism intensity							
	Urease	Polyphenol oxidase	Catalase	Invertase	Phosphatase	Alkali-hydrolyzable nitrogen	Available phosphorus	Available potassium
Before planting	0.591	0.552	0.586	0.550	0.615	0.640	0.740	0.516
Flowering stage	0.638	0.598	0.532	0.601	0.624	0.647	0.709	0.495
Rhizome expansion stage	0.594	0.490	0.761	0.568	0.602	0.633	0.772	0.525
Withering stage	0.542	0.568	0.465	0.481	0.619	0.641	0.738	0.529
Treatment	Shonnon index							
	Urease	Polyphenol oxidase	Catalase	Invertase	Phosphatase	Alkali-hydrolyzable nitrogen	Available phosphorus	Available potassium
Before planting	0.628	0.799	0.606	0.581	0.728	0.744	0.511	0.709
Flowering stage	0.634	0.747	0.637	0.541	0.755	0.837	0.475	0.527
Rhizome expansion stage	0.516	0.789	0.477	0.533	0.698	0.703	0.529	0.784
Withering stage	0.733	0.860	0.703	0.669	0.732	0.693	0.529	0.815
Treatment	Richness							
	Urease	Polyphenol oxidase	Catalase	Invertase	Phosphatase	Alkali-hydrolyzable nitrogen	Available phosphorus	Available potassium
Before planting	0.626	0.572	0.614	0.587	0.649	0.657	0.712	0.545
Flowering stage	0.644	0.557	0.499	0.569	0.581	0.603	0.663	0.462
Rhizome expansion stage	0.617	0.495	0.761	0.585	0.686	0.685	0.791	0.536
Withering stage	0.616	0.663	0.582	0.607	0.680	0.684	0.682	0.638

Conclusions and Discussion

Microorganisms are the fundamental source of soil vitality and the drivers of key biogeochemical cycles of elements. The microbial population and its community structure can serve as important indicators for assessing soil health. The functions of soil microorganisms primarily include organic matter decomposition, element cycling, and ecological security regulation, with material decomposition being the most critical function^[11]. Functional diversity is mainly reflected in species richness and evenness. To investigate the mechanisms behind the continuous cropping obstacles in *P. odoratum*, a treatment group of continuously cropped *P. odoratum* was compared with a control group of non-continuously cropped *P. odoratum* and a blank control group. In this study, the effects of continuous cropping on the functional diversity of rhizosphere soil microorganisms, soil enzyme activities and soil nutrient coordination during different growth stages were investigated. The experimental results indicated that: (1) continuous cropping increased the carbon source metabolic capacity, Shannon diversity index, and richness of rhizosphere soil microorganisms by 3.2% – 14.7%, 0.9% – 3.5%, and 1.3% – 12.5%, respectively, but these increases were not statistically significant. (2) Principal component analysis revealed that during the middle stage of rhizome expansion, continuous cropping significantly altered the characteristics of microbial carbon metabolism. Microbial communities utilizing carbohydrates, amino acids, polymers, carboxylic acids, and amines as carbon sources exhibited vigorous metabolic activity. (3) Continuous cropping significantly reduced the activities of urease, polyphenol oxidase, and acid phosphatase in rhizosphere soil, with decreases of 24.4% – 39.5%, 3.2% – 14.8%, and 7.9% – 18.2%, respectively. The activities of catalase and invertase sometimes exceeded and sometimes fell below those of the first crop, showing no consistent pattern of change. (4) Under continuous cropping conditions, rhizosphere soil nutrients were imbalanced, characterized by nitrogen deficiency and phosphorus surplus. (5) Grey correlation analysis indicated that the available phosphorus content, alkali-hydrolyzable nitrogen content and polyphenol oxidase activity in the rhizosphere soil were the main factors influencing microbial functional diversity.

This study demonstrated that continuous cropping of *P. odoratum* reduced the activities of urease, polyphenol oxidase, and acid phosphatase in the rhizosphere soil. During the growth period, the continuous cropping treatment showed decreases of 24.4% – 39.5%, 3.2% – 14.8%, and 7.9% – 18.2%, respectively, compared with the first crop, which is consistent with the findings of Ying *et al.*^[12]. In contrast, the activities of catalase and invertase were sometimes higher and sometimes lower than those in the first crop, but overall showed an increasing trend. Under continuous cropping conditions, a severe imbalance of nitrogen and phosphorus nutrients occurred in the rhizosphere soil, characterized by a decrease in alkali-hydrolyzable nitrogen content and an absolute

surplus of available phosphorus, which aligns with the findings of Yang *et al.*^[13]. The reason for the severe surplus of available phosphorus might be related to the preferential application of pig manure in fertilization. A long-term positioning study conducted in Jinxian County, Jiangxi Province, revealed that the total phosphorus content in pig manure is approximately 4.5%, and long-term application in red paddy soils leads to a substantial accumulation of phosphorus in the soil^[14].

Due to the complexity and holistic nature of the rhizosphere micro-environment, simple correlation analysis or net correlation analysis is insufficient to accurately express the relationships among multivariate variables. In this study, microbial functional diversity and the rhizosphere environment were treated as a grey system, attempting to identify the main influencing factors on microbial functional parameters. The results indicated that the most significant factor affecting microbial carbon metabolic capacity and richness was soil available phosphorus content, while the most influential factors on the Shannon index were polyphenol oxidase activity and alkali-hydrolyzable nitrogen content.

References

- [1] KANG YL, SUN WQ, LIU JG, *et al.* Study on the effect of continuous cropping on microbial community structure in rhizosphere soil of processing tomato by PLFA method[J]. Chinese Journal of Eco-Agriculture, 2017, 25(4): 594 – 604. (in Chinese).
- [2] LI X, DING C, HUA K, *et al.* Soil sickness of peanuts is attributable to modifications in soil microbes induced by peanut root exudates rather than to direct allelopathy[J]. Soil biology and biochemistry, 2014, 78: 149 – 159.
- [3] QIN Y, MA K, LIU P. Effects of continuous cropping of potato on soil microbial diversity[J]. Chinese Journal of Eco-Agriculture, 2015, 23(2): 225 – 232. (in Chinese).
- [4] WANG WP, MAO RZ, CHEN JB, *et al.* Analysis of functional diversity of soil microbial communities under different cultivation patterns at different growth stages of maize[J]. Chinese Journal of Eco-Agriculture, 2015, 23(10): 1293 – 1301. (in Chinese).
- [5] ZHANG ZY, LIN WX. Allelopathy and autotoxicity of medicinal plants and continuous cropping obstacles[J]. Chinese Journal of Eco-Agriculture, 2009, 17(1): 189 – 196. (in Chinese).
- [6] WU LK, HUANG WM, WANG JY, *et al.* Diversity analysis of rhizosphere microflora of wild *Rehmannia glutinosa* grown in monocropping for different years[J]. Acta Agronomica Sinica, 2015, 41(2): 308 – 317. (in Chinese).
- [7] SU SM, REN LX, HUO ZH, *et al.* Effects of Intercropping watermelon with rain fed rice on Fusarium wilt and the microflora in the rhizosphere soil[J]. Scientia Agricultura Sinica, 2008(3): 704 – 712. (in Chinese).
- [8] BAO SD. Soil agrochemical analysis[M]. Beijing: China Agricultural Science and Technology Press, 2000. (in Chinese).
- [9] CHEN FL, ZHENG H, YANG BS, *et al.* Effects of exotic species slash pine (*Pinus elliottii*) litter on the structure and function of the soil microbial community[J]. Acta Ecologica Sinica, 2011, 31(12): 3543 – 3550. (in Chinese).

(Continued on page 21)

K content was recorded during the growth stage, classified as level II (abundant), while the lowest content was found at the seedling stage, classified as level III (medium). Significant differences were observed at the growing and maturity stages under different treatments, while at the seedling stage, the differences were not significant. Comprehensive analysis indicated that under the EK treatment, soybean roots secreted substances such as organic acids and sugars, which activated insoluble potassium in the soil and converted it into available K. Meanwhile, the presence of potassium-solubilizing bacteria in the soybean rhizosphere soil decomposed and released fixed potassium, thereby increasing the available K content. Compared with the CK treatment, the soil available K content under the EK treatment increased by 5.97%, 18.04%, and 17.59% during the seedling, growing, and maturity stages, respectively.

Soil available K effectively reflects the supply capacity of potassium fertility in the soil. Its content directly indicates the effectiveness of soil potassium fertilizer and its potential impact on plant growth and quality. In peach orchards without soybean intercropping, the application of potassium fertilizer can be appropriately increased. Additionally, it is essential to integrate other management measures, such as straw returning, irrigation, and pest control, to comprehensively enhance the available K content.

Table 3 Soil available K content at different growth stages under different treatments

Growth and development stage	Treatment method	Available K//mg/kg	Standard deviation	Difference
Seedling stage	CK	134.78	14.21	Aa
	EK	143.34	9.23	Aa
Growing stage	CK	124.52	9.75	Bab
	EK	151.92	11.01	Aa
Maturity stage	CK	119.78	6.94	Bb
	EK	145.34	6.84	Aa

Conclusions and Discussion

The impact of soybean intercropping on soil physicochemical properties was primarily reflected in the increase levels of alkali-

hydrolyzable N and available K and the decrease level of available P in the soil during the seedling, growing, and maturity stages. Overall, this approach optimized and enhanced soil fertility in the peach orchards, providing a viable pathway for soil improvement in peach orchards located in karst landform areas. Based on the research findings, practical production can involve timely intertillage and pruning before the soybean maturity stage to maintain rhizobial activity, as well as targeted topdressing with available P fertilizers to balance soil nutrient supply and further enhance the synergistic effects of the intercropping system.

References

[1] Eating peaches in summer achieving the effects of whitening and anti-aging? Japanese media reveals the efficacy of peaches[J]. Xinxueguanbing Fangzhi Zhishi, 2014(17): 76.

[2] CHEN JP. Therapeutic effect of peach[J]. Medicine and Health Care, 1995(3): 21.

[3] CUI NB, LIU W. The impact and countermeasures option of global soybean trade pattern change on China's soybean industry[J]. Soybean Science, 2019, 38(4): 629–634.

[4] QI XP, JIANG BJ, WU TT, *et al.* Genomic dissection of widely planted soybean cultivars leads to a new breeding strategy of crops in the post-genomic era [J]. Crop J., 2021, 9(5): 1079–1087.

[5] DU JB, HAN TF, GAI JY, *et al.* Maize-soybean strip intercropping: achieved a balance between high productivity and sustainability[J]. Journal of Integrative Agriculture, 2018,17(4): 747–754.

[6] LUO H, WANG J, SONG Y, *et al.* Research status and prospect of maize-soybean interplanting model [J]. Crop Research, 2020, 34(5): 502–506.

[7] MA ZM, PAN ZR, YANG H, *et al.* Effect of maize-soybean strip intercropping on soil fertility[J]. Journal of Sichuan Agricultural University, 2024, 42(5): 1028–1035.

[8] CHEN LY, WANG ZB, HU LP. Effects of interplanting six leguminous green manure plants in apple orchard on soil carbon and nitrogen characteristics[J]. Acta Agrestia Sinica, 2021, 29(4): 671–676.

[9] XU Q. Effects of straw mulching on temporal and spatial distribution characteristics of nitrogen and phosphorus in farmland soil in black soil region[D]. Heilongjiang: Northeast Agricultural University, 2021.

[10] ZHANG YG, YU HM, XU YC, *et al.* Effect of soybean intercropping in peach orchards on peach tree growth[J]. Journal of Zhejiang Agricultural Sciences, 2017, 58(12): 2199–2200, 2204.

Editor: Yingzhi GUANG

Proofreader: Xinxiu ZHU

(Continued from page 18)

[10] SUN YG. Research on grey correlation analysis and its application[D]. Nanjing: Nanjing University of Aeronautics and Astronautics, 2007. (in Chinese).

[11] LIN XG, HU JL. Scientific connotation and ecological service function of soil microbial diversity[J]. Acta Pedologica Sinica, 2008(5): 892–900. (in Chinese).

[12] YING Y, DING W, ZHOU Y, *et al.* Influence of *Panax ginseng* continuous cropping on metabolic function of soil microbial communities[J].

Chinese Herbal Medicines, 2012, 4(4): 329–334.

[13] YANG YH, CHEN DM, JIN Y, *et al.* Effect of different fertilizers on functional diversity of microbial flora in rhizospheric soil under tobacco monoculture[J]. Acta Agronomica Sinica, 2011, 37(1): 105.

[14] LIU KL, LI DM, HUANG QH, *et al.* Ecological benefit and carrying capacity evaluation of long-term application of pig manure in red soil paddy field[J]. Scientia Agricultura Sinica, 2014, 47(2): 303–313. (in Chinese).