

Optimization of Water and Fertilizer Management of *Panax notoginseng* Based on Changes in Soil Microbial Biomass Carbon and Nitrogen and Enzyme Activities

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Abstract

At present, there is a lack of understanding of the dynamic characteristics of the carbon, nitrogen, and enzyme activities of the soil microbial biomass of *Panax notoginseng* under irrigation and fertilization. We set up a field experiment with 3 irrigation and 4 fertilization levels for a total of 12 treatments plus a control in the micro-sprinkler *Panax notoginseng* farmland in Luxi County, Yunnan Province, from 2018 to 2020. The results showed that the soil microbial biomass carbon, nitrogen, and enzyme activities increased from the rooting period to the flowering period, and then decreased in the fruiting period. The soil microbial biomass carbon, nitrogen, and enzyme activities of *Panax notoginseng* reached the maximum values in the flowering period, while the minimum values were reached in the rooting period. There were significant positive correlations between the soil microbial biomass carbon and nitrogen and enzyme activities during different growth periods under different water and fertilizer optimization management. This study revealed the tendency of the carbon, nitrogen, and enzyme activities of the soil microbial biomass of *Panax notoginseng* with the growth periods. It also analyzed the optimal management of different water and fertilizer regimes and the C_i values of soil microbial biomass carbon, nitrogen, and enzyme activities in different growth periods to determine the best water and fertilizer treatment methods in each growth period through the entropy value method combined with the TOPSIS method. The study found that the C_i values changed with the change in the fertilization level under the same irrigation level. The C_i values decreased and then increased with increasing irrigation water under the same fertilization level. These results showed that, in terms of the effect of different water and fertilizer treatments on the regulation of the soil microbial biomass carbon, nitrogen, and enzyme activities in each growth period, the irrigation volume of 10 mm and fertilizer application of $8.00 \text{ kg} \cdot 667^{-1} \cdot \text{m}^{-2}$ during the rooting period of *Panax notoginseng* were most effective, while an irrigation volume of 15 mm and fertilizer application of $8.00 \text{ kg} \cdot 667^{-1} \cdot \text{m}^{-2}$ were more suitable for the seedling, flowering, and fruiting periods.

1. Introduction

Soil microbial biomass (SMB), an active component of soil organic matter, plays a key role in the regulation of the soil ecosystem and its function, serving as one of the most important biological indicators for the evaluation of the soil's comprehensive quality. Soil microbial biomass carbon (SMBC) and soil microbial biomass nitrogen (SMBN) determine soil fertility and are important sources of soil organic carbon and available nitrogen. Soil enzymes are derived from soil microorganisms, plant root exudates, soil animals, and biological residues, and are sensitive to different fertilization responses. They are among the most active components in soil ecosystems and play a vital role in nutrient cycling (nitrogen fixation, phosphorus adsorption and desorption, potassium release, etc.), as well as soil structure maintenance and crop yield. They form a comprehensive soil fertility biological index. As a catalyst for soil organic matter decomposition and nutrient conversion cycling, soil enzymes have a close relationship with the soil nutrient content and microbial biomass carbon and nitrogen. Soil's comprehensive fertility is determined by the soil microbial biomass carbon and nitrogen, and enzyme activity.

Fertilization and irrigation have significant effects on the SMBC, SMBN, and soil enzyme activities. Irrigation improves soil enzyme activities, and a study on the soil microbial characteristics, soil enzyme activities, and nutrient regulation of winter wheat fields with water-saving irrigation showed that the soil microbial characteristics and soil enzyme activity were regulated by irrigation and were closely related to the cyclic transformation of soil carbon and nitrogen nutrients. Li et al. found that N application significantly increased the SMBC and SMBN. Xu et al. studied the effects of fertilization on the soil organic carbon (SOC) content, SMBC, and SMBN, and found that the 30% organic matter + 70% inorganic fertilizer, 60% organic matter + 40% inorganic fertilizer, and straw and inorganic fertilizer treatments produced higher yields of both early and late rice than inorganic fertilizer alone and no fertilizer. HAO et al. studied the effects of the long-term application of inorganic fertilizer and organic fertilizer on the organic matter and microbial biomass of three subtropical rice soils, and the results showed that the mixed application of inorganic fertilizer with manure or straw improved the organic matter and microbial biomass of the subtropical paddy soil. Bach et al. showed that the microbial activity in a fertilized grassland was higher than that in non-fertilized soil, and the addition of nitrogen to the fertilized grassland enhanced the soil microbial biomass and enzyme activity.

There have been many studies on the effects of different farmland management methods on the SMBC, SMBN, and enzyme activities, but the quantitative study of the medicinal plant *Panax notoginseng* has not received much attention. Studies on the effects of fertilizer–irrigation interactions on the inter-root secretions of *Panax notoginseng* and its SMBC, SMBN, and enzyme activities are also scarce, thus affecting the study of soil microbiological traits in *Panax notoginseng* farmlands. At the moment, research on the interactions of water and fertilizer is primarily focused on regulating the improvement in soil saline–alkali land in dryland farmland in the north, as well as crop yield and quality. *Panax notoginseng* research is primarily focused on single factors, such as the cultivation mode, continuous cropping obstacle of *Panax notoginseng*, accumulation and adsorption of heavy metals in soil, and the fertilizer element ratio. Few studies have been conducted to investigate the effects of optimal water and fertilizer management on the SMBC, SMBN, and enzyme activities of *Panax notoginseng*, as well as the regulation of the effects of the combination of irrigation and fertilization on the plant growth, yield, and quality of *Panax notoginseng*. The SMBC, SMBN, and enzyme activities characterize soil fertility. In this study, a complete randomized block experiment design was adopted for different water and fertilizer interactions, and three years of growth of *Panax notoginseng* from 2018 to 2020 was used to conduct different water and fertilizer optimization management experiments. This experiment analyzed the effects of different water and fertilizer management regimes on the soil microbial carbon, nitrogen, and enzyme activities of *Panax notoginseng*, and investigated the mechanism of the effect of *Panax notoginseng* management on the SMBC, SMBN, and enzyme activities to determine the most suitable water and fertilizer management program for different growth periods. The results can provide technical support for the efficient use and management of water and fertilizer in *Panax notoginseng* farmland, as well as important practical significance for guiding the high-yield and quality cultivation of *Panax notoginseng*.

2. Materials And Methods

2.1. Test Design and Sample Collection

The experiment was conducted in the typical planting area of *Panax notoginseng* in Dali Shu Village, Luxi County, Yunnan Province, from 2018 to 2020, at the geographical location of 24°15' N, 103°30' E. It belongs to the north subtropical monsoon climate. The growth period of *Panax notoginseng* was divided into four periods, i.e., the rooting period (November-January), seedling period (February-April), flowering period (May-July), and fruiting period (August-October). According to the high-yield and high-efficiency fertigation system of local *Panax notoginseng* cultivation, the experimental design consisted of two control factors, the amount of irrigation (W) and the amount of fertilizer (F) applied. The irrigation was set to three levels, i.e., W1 (irrigation volume of 5 mm), W2 (irrigation volume of 10 mm), and W3 (irrigation volume of 15 mm), where irrigation levels of 5 mm, 10 mm, and 15 mm corresponded to 40%, 60%, and 80% of the field water-holding capacity, respectively. The amount of fertilizer was set to four levels, i.e., F1 (3.20 kg.667⁻¹.m⁻²), F2 (4.80 kg.667⁻¹.m⁻²), F3 (6.20 kg.667⁻¹.m⁻²), and F4 (8.00 kg.667⁻¹.m⁻²). Through random complete combination, 12 treatments were set, i.e., W1F1, W1F2, W1F3, W1F4, W2F1, W2F2, W2F3, W2F4, W3F1, W3F2, W3F3, and W3F4. Additionally, a control treatment (CK) was added, and CK was neither irrigated nor fertilized. Each treatment had three replicates with a total of 39 plots, and all plots were covered with 5 mm pine needles on the surface. Each plot was 16.70 m long and 1.50 m wide. *Panax notoginseng* was planted with spacing of 10 cm between rows. Each plot was irrigated by a micro-sprinkler system, where the irrigation volume was mainly controlled by a water valve and a water meter on the water pipe in the system. At the beginning of each growth period, the water required for the different treatments and the fertilizer required for the growth period were mixed and delivered to a pipe in each test area and irrigated semimonthly. To avoid the effect of natural rainfall on the test, a double layer of shade netting and plastic film covered the experimental site. The fertilizer was German-American water-soluble organic fertilizers with N ≥ 21%, P₂O₅ ≥ 21%, K₂O ≥ 21%, humic acid ≥ 6%, B ≥ 0.1%, and Mo ≥ 0.007%.

The sampling was conducted on the third day after irrigation and fertilization in December of each year, and March, June, and September of the following year from 2018 to 2020. During the experiment, a 1 m × 1 m sampling point was set in each residential area to remove soil surface litter and gravel around the sampling points at sampling depths of 0–20 cm, 20–40 cm and 40–60 cm, respectively. The soil samples were added to self-sealing bags for labeling, sealed, and stored, and then taken back to the laboratory. The fresh soil sample was divided into two parts. One part was used to measure the microbial carbon and nitrogen contents. The other part was laid out on kraft paper to air-dry naturally and then remove roots, leaves, and gravel, and then the soil enzyme activities were tested after grinding and sieving. The basic physical and chemical properties of the soil before the test are shown in Table 1.

Table 1
Basic physical and chemical properties of soil.

Organic Carbon	Total Nitrogen	Total Potassium	Total Potassium	Available Potassium	Nitrate Nitrogen	Ammonium Nitrogen	Available Potassium	pH	Field Water-Holding Capacity %
g•kg ⁻¹	g•kg ⁻¹	g•kg ⁻¹	g•kg ⁻¹	mg•kg ⁻¹	mg•kg ⁻¹	mg•kg ⁻¹	mg•kg ⁻¹		
14.33	0.98	0.37	14.79	316.8	8.35	19.15	11.65	6.34	42.28

2.2. Determination Method

The chloroform-fumigation extraction method was used for the determination of the SMBC and SMBN. The chloroform-fumigated and non-fumigated soils were extracted with 0.5 mol•L⁻¹ K₂SO₄ solution and a soil:liquid ratio of 1:4. The organic carbon content in the extraction solution was determined by FeSO₄ solution titration with a conversion coefficient of 0.38, and the organic nitrogen content in the extraction solution was determined by the ninhydrin colorimetric method with a conversion coefficient of 5.00. Three replicates of each fumigated and non-fumigated sample were analyzed and the mean values were calculated.

The soil enzyme activity was determined by referring to the Soil Enzyme and Its Research method. Catalase (Cat) was measured by KMnO₄ titration, involving titration until the purple color faded to light pink, in milliliters per 1 g•h⁻¹ soil consuming 0.1 mol•L⁻¹ KMnO₄ to express the catalase activity. Urease (Ure) was measured using the phenol–sodium hypochlorite colorimetric method, with a spectrophotometer to measure the absorbance value of the sample at 578 nm for result calculation, and the urease activity was expressed in milligrams of NH₃⁻-N produced in 1 g of soil after 1 d of incubation. Sucrase (Sue) was measured by colorimetry with 3,5-dinitrosalicylic acid; the sample solution was orange–yellow due to the generation of 3,5-nitrosalicylic acid. After dilution with distilled water, the absorbance value at 508 nm on the spectrophotometer was measured and calculated, and sucrase activity was expressed in milligrams of glucose formed in 1 g of soil after 1 d of culturing. The acid phosphatase (Acp) activity was determined using the disodium phosphate colorimetric method; after the sample solution turned blue, it was diluted with distilled water and its absorbance value was measured at 660 nm and calculated in milligrams of phenol released from 1 g of soil 1 d later. Each soil sample was measured 3 times and the results were averaged.

2.3. Soil Microbial Biomass Carbon and Nitrogen and Enzyme Activity Evaluation Calculation

In this study, the entropy method was used to determine the weight of each indicator. Due to the differences between each indicator, in order to eliminate the dimension of indicator data and the positive and negative effects of the indicators, the original data were standardized to form a new data column. The SMBC, SMBN, and soil enzyme activities were analyzed using the TOPSIS method, which can assess sample quality by calculating the relative distance between the sample and the best or worst solution. Steps (1) and (2) are the entropy method to calculate the index weight, and step (3) is the TOPSIS evaluation calculation method:

Data standardization

$$u_{ij} = \frac{x_{ij} - \min(x_{ij})}{\max(x_{ij}) - \min(x_{ij})},$$

$$u_{ij} = \frac{\max(x_{ij}) - x_{ij}}{\max(x_{ij}) - \min(x_{ij})},$$

1

where u_{ij} is the value of indicator of its sample ($i = 1, 2, \dots, n$; $j = 1, 2, \dots, n$); $\max(x_{ij})$ and $\min(x_{ij})$ are the maximum and minimum values of indicator x_{ij} , respectively, and u_{ij} is the normalized x_{ij} , which are the standardized indicators of the soil microbial carbon and nitrogen contents and enzyme activity under different water and fertilizer optimal management regimes.

Calculate the weight of sample data under indicator for that indicator a_{ij} :

$$a_{ij} = u_{ij} / \sum_{i=1}^n u_{ij}$$

2

TOPSIS evaluation method

Construction of original evaluation matrix:

There are m evaluation objects and n evaluation indicators; the indicators and the corresponding entropy weights W_j are multiplied, and the data are represented as the original matrix $X = (X_{ij})_{m \times n}$, where X_{ij} is the original data of the j th indicator of the i th treatment:

$$Z_{ij} = \frac{X_{ij}}{\sqrt{\sum_{j=1}^n X_{ij}^2}}$$

3

where $i = (1, 2, \dots, m)$, $j = (1, 2, \dots, n)$, and $m = 13$, i.e., the 13 water and fertilizer treatment trials. $n = 6$ indicators of the soil enzyme activities and microbial biomass carbon and nitrogen.

Normalization matrix $Z = (Z_{ij})_{m \times n}$. The maximum and minimum values of each column constitute the optimal and inferior vectors, respectively:

$$Z^+ = (Z_{\max 1}, Z_{\max 2}, \dots, Z_{\max n})$$

$$Z^- = (Z_{\min 1}, Z_{\min 2}, \dots, Z_{\min n})$$

(4)

The distances between the i th treatment and the optimal and inferior solutions are:

$$D_i^+ = \sqrt{\sum_{j=1}^n (Z_{\max j} - Z_{ij})^2} \quad D_i^- = \sqrt{\sum_{j=1}^n (Z_{\min j} - Z_{ij})^2} \quad (5)$$

The i th treatment fits to the optimal solution C_i are:

$$C_i = \frac{D_i^-}{(D_i^+ + D_i^-)}$$

6

Microsoft Excel 2016 and SPSS26.0 were used for the statistical analysis of the data, and a one-way ANOVA was used to test the differences in the soil microbial biomass carbon and nitrogen contents and enzyme activity under different water and fertilizer optimal management regimes ($\alpha = 0.05$). The entropy method was used to calculate the weight and rank the soil microbial biomass carbon and nitrogen contents and enzyme activity. Mantel analysis was performed using the R language, and Origin2020b software was used for mapping.

3. Result Analysis

3.1. Effects of Different Water and Fertilizer Optimal Management Regimes on Soil Microbial Biomass Carbon and Nitrogen Change Characteristics

Under the same water and fertilizer optimal management regime, the difference in the soil microbial biomass carbon and nitrogen content was significant between different growth periods ($p < 0.05$), and the trend was an increase and then decrease over the growth periods. The carbon-to-nitrogen ratio of the microbial biomass in the soil showed a trend of decreasing and then increasing with the advancement of the growth period, with the highest value in the rooting period and the lowest value in the flowering period. As shown in Table 2, the SMBC and SMBN contents were the highest in the flowering period, at $1.06 \text{ g} \cdot \text{kg}^{-1}$ and $66.22 \text{ g} \cdot \text{kg}^{-1}$, respectively, and the lowest in the rooting period, at $0.39 \text{ g} \cdot \text{kg}^{-1}$ and $14.99 \text{ g} \cdot \text{kg}^{-1}$, respectively. From high to low, the SMBC and

SMBN contents decreased in the different periods in the following order: flowering period, fruiting period, seedling period, and rooting period. The maximum SMBC content in the W3F4 treatment was $1.06 \text{ g}\cdot\text{kg}^{-1}$, which was 34.18%, 24.71%, and 16.48% higher than those in the rooting period, seedling period, and fruiting period, respectively, at the same time; the difference between the flowering period and rooting period was the largest, which was $0.27 \text{ g}\cdot\text{kg}^{-1}$. The minimum SMBC content was $0.39 \text{ g}\cdot\text{kg}^{-1}$ in the rooting period under the CK treatment, and the difference between the flowering and rooting periods under this treatment was $0.25 \text{ g}\cdot\text{kg}^{-1}$, second only to that of the W3F4 treatment, and the smallest difference between the flowering and rooting periods was $0.11 \text{ g}\cdot\text{kg}^{-1}$ for W1F3. The SMBN content reached the maximum of $66.21 \text{ mg}\cdot\text{kg}^{-1}$ in the flowering period of W3F4, which was 48.41%, 29.49%, and 15.47% higher than those in the rooting period, seedling period, and fruiting period respectively. The minimum value of the SMBN content under W1F1 treatment was $14.99 \text{ mg}\cdot\text{kg}^{-1}$ in the rooting period, the maximum difference between the W3F4 flowering period and rooting period was $21.60 \text{ mg}\cdot\text{kg}^{-1}$, and the minimum difference between the W2F1 flowering period and rooting period was $9.24 \text{ mg}\cdot\text{kg}^{-1}$. The difference between the rooting period and flowering period was the smallest, at 1.82, under the W3F4 treatment, and the largest, at 9.52, under W1F3, which was mainly due to the higher microbial biomass nitrogen content in the flowering period. As a result, the SMBC and SMBN contents under the W3F4 treatment varied the most over the four growth periods, while their carbon-to-nitrogen ratio was the most stable. This also showed that W3F4 was more effective in regulating the carbon and nitrogen contents of the soil microbial biomass.

The soil microbial biomass carbon and nitrogen contents in the same growth period had significant differences with different water and fertilizer management regimes, and the SMBC and SMBN contents increased with the increase in the irrigation amounts and fertilizer application amounts. It could be seen from Table 2 that the SMBC and SMBN contents of W3F4 in all growth periods were greater than those of the other treatments. Except for CK treatment, W1F1 exhibited the lowest SMBC content in each growth period. In the rooting period, the content under W3F4 increased by $0.29 \text{ g}\cdot\text{kg}^{-1}$, 2.03 times that under CK compared with that under W1F1; in the seedling period, the content under W3F4 increased by $0.30 \text{ g}\cdot\text{kg}^{-1}$, 1.93 times that under CK compared with W1F1; in the flowering period, the content under W3F4 increased by $0.42 \text{ g}\cdot\text{kg}^{-1}$, 1.66 times that under CK compared with W1F1; and in the fruiting period, the content under W3F4 increased by $0.31 \text{ g}\cdot\text{kg}^{-1}$, 1.86 times that under CK compared with W1F1. The contents of SMBN in each growth period were higher than that in W1F1. In the rooting period, the content under W3F4 increased by $29.63 \text{ mg}\cdot\text{kg}^{-1}$ from that under W1F1, 2.37 times that under CK at the same time. In the seedling period, the content under W3F4 increased by $33.81 \text{ mg}\cdot\text{kg}^{-1}$ from that under W1F1, 2.47 times that of CK. In the flowering period, the content under W3F4 increased by $39.52 \text{ mg}\cdot\text{kg}^{-1}$ from that under W1F1, 2.23 times that under CK. In the fruiting period, the content under W3F4 increased by $37.17 \text{ mg}\cdot\text{kg}^{-1}$ from that under W1F1, 2.48 times that under CK. This shows that treatment W3F4 significantly increased the SMBC and SMBN contents in all four growth periods of the treatment.

Table 2

Characteristics of the changes in the soil microbial biomass carbon and nitrogen under different optimal water and fertilizer management regimes.

Growth Period	SMBC g·kg ⁻¹ SMBN mg·kg ⁻¹	W1F1	W1F2	W1F3	W1F4	W2F1	W2F2	W2F3	W2F4	W3F1	W3F2	W3F3	W3F4	CK	p
Rooting period	SMBC	0.50 ± 0.03 Ld	0.56 ± 0.07 Kd	0.65 ± 0.06 Fd	0.70 ± 0.05 Dd	0.56 ± 0.04 Jd	0.59 ± 0.05 ld	0.64 ± 0.05 Hd	0.67 ± 0.03 Ed	0.65 ± 0.04 Gd	0.75 ± 0.04 Cd	0.79 ± 0.04 Bd	0.79 ± 0.04 Ad	0.39 ± 0.04 Md	0.000 **
	SMBN	14.99 ± 1.19 Md	18.97 ± 1.27 Kd	23.35 ± 1.30 ld	28.43 ± 1.27 Fd	22.76 ± 1.22 Jd	25.73 ± 1.25 Hd	30.26 ± 1.35 Ed	36.01 ± 1.43 Bd	27.27 ± 1.24 Gd	31.12 ± 1.26 Dd	35.12 ± 1.39 Cd	44.62 ± 1.41 Ad	18.86 ± 1.16 Ld	0.000 **
Seedling period	SMBC	0.55 ± 0.05 Lc	0.65 ± 0.05 lc	0.70 ± 0.03 Gc	0.75 ± 0.04 Ec	0.59 ± 0.02 Kc	0.63 ± 0.03 Jc	0.71 ± 0.04 Fc	0.70 ± 0.05 Dc	0.70 ± 0.03 Hc	0.79 ± 0.03 Cc	0.83 ± 0.04 Bc	0.85 ± 0.04 Ac	0.44 ± 0.03 Mc	0.000 **
	SMBN	17.33 ± 1.25 Mc	21.47 ± 1.24 Kc	30.01 ± 1.35 lc	31.15 ± 1.36 Hc	26.41 ± 1.24 Jc	31.48 ± 1.33 Gc	33.44 ± 1.34 Ec	40.81 ± 1.36 Bc	31.72 ± 1.37 Fc	35.31 ± 1.34 Dc	38.48 ± 1.35 Cc	51.14 ± 1.39 Ac	20.74 ± 1.22 Lc	0.000 **
Flowering period	SMBC	0.64 ± 0.03 La	0.71 ± 0.04 Ja	0.76 ± 0.04 Ha	0.83 ± 0.04 Ea	0.70 ± 0.03 Ka	0.73 ± 0.03 Ia	0.79 ± 0.04 Ga	0.85 ± 0.04 Da	0.80 ± 0.03 Fa	0.89 ± 0.03 Ca	0.97 ± 0.03 Ba	1.06 ± 0.04 Aa	0.64 ± 0.03 Ma	0.000 **
	SMBN	26.70 ± 1.35 Ma	33.26 ± 1.38 Ja	41.30 ± 1.35 Ha	46.25 ± 1.37 Ea	32.00 ± 1.26 Ka	36.35 ± 1.35 Ia	41.20 ± 1.36 Ga	55.15 ± 1.41 Ca	45.70 ± 1.36 Fa	50.15 ± 1.34 Da	55.79 ± 1.39 Ba	66.22 ± 1.37 Aa	29.74 ± 1.26 La	0.000 **
Fruiting period	SMBC	0.60 ± 0.03 Lb	0.65 ± 0.02 Jb	0.73 ± 0.03 Hb	0.75 ± 0.03 Fb	0.63 ± 0.04 Kb	0.69 ± 0.02 lb	0.76 ± 0.03 Eb	0.81 ± 0.04 Db	0.75 ± 0.03 Gb	0.85 ± 0.04 Cb	0.86 ± 0.03 Bb	0.91 ± 0.04 Ab	0.49 ± 0.03 Mb	0.000 **
	SMBN	20.18 ± 1.38 Mb	24.47 ± 1.35 Kb	31.57 ± 1.37 lb	40.58 ± 1.26 Eb	28.82 ± 1.35 Jb	33.79 ± 1.36 Hb	37.97 ± 1.24 Fb	42.70 ± 1.33 Cb	36.94 ± 1.34 Gb	41.18 ± 1.24 Db	50.65 ± 1.34 Bb	57.35 ± 1.37 Ab	23.10 ± 1.26 Lb	0.000 **
Note 1: W1F1, W1F2, W1F3, etc. are the 13 treatments designed in this experiment.															
Note 2: Different capital letters indicate significant differences between different water and fertilizer control treatments in the same growth period ($p < 0.05$) and different lowercase letters indicate significant differences between different growth periods under the same water and fertilizer control treatment ($p < 0.05$). Note 3: ** indicate significant difference at 0.01 levels.															

3.2. Effect of Different Water and Fertilize Optimal Management on Soil Enzyme Activity Change Characteristics

Under the same optimal management regime of water and fertilizer, the activities of soil urease, sucrase, acid phosphatase, and catalase showed significant differences between the different growth periods ($p < 0.05$), which increased first and then decreased with the prolongation of the growth period of *Panax notoginseng*, the activities ranged from high to low in the following order: flowering period, fruiting period, seedling period and rooting period. From Fig. 1a, it can be seen that the urease activity ranged from 2.86 to 9.52 mg·kg⁻¹·d⁻¹, with increases of 30.81% and 46.08% from the rooting period to the flowering period, respectively, and the activity under W3F4 was 2.14 times higher than that under CK in the rooting period and 1.82 times higher in the flowering period. Figure 1b shows that the sucrase activity ranged from 17.12 to 68.79 mg·kg⁻¹·d⁻¹, and increased by 31.94% and 44.43%, respectively, from the rooting period to the flowering period; the rooting period activity under W3F4 was 1.78 times that under CK, and the flowering-stage activity was 1.83 times higher. Figure 1c shows the acid phosphatase activity, which ranged from 2.14 to 8.2 mg·kg⁻¹·d⁻¹, with 34.08% and 59.01% increases from the rooting to the flowering periods, respectively, and 2.10 times higher activity in the rooting period and 2.53 times higher activity in the flowering period under W3F4 than those under CK. Figure 1d shows that the catalase activity was between 5.1 and 9.42 mg·kg⁻¹·h⁻¹, and increased by 12.44% and 37.86%, respectively, from the rooting period to the flowering period; the rooting-period activity under W3F4 was 1.67 times that under CK, and the flowering-stage activity was 1.35 times higher.

Under different water and fertilizer optimal management regimes, the soil urease, sucrase, acid phosphatase, and catalase activities in the same growth period showed significant differences ($p < 0.05$). The soil urease, sucrase, acid phosphatase, and catalase activities increased with the increases in the irrigation amount and fertilizer application amount. As shown in Fig. 1, the soil urease, sucrase, acid phosphatase, and catalase activities under W3F4 in the flowering period were the highest, increasing by 44.26%, 61.51%, 42.56%, and 32.25%, respectively, as compared with the minimum value under W1F1. Under the same water and fertilizer optimal management regimes, the soil enzyme activities increased with the increase in the fertilizer application amount, and the activities of urease, sucrase, acid phosphatase, and catalase under W3F4 were the highest, which were 1.95, 1.69, 2.32, and 1.47 times those under CK. This shows that the W3F4 treatment was more likely to increase the soil enzyme activities.

3.3. Effect of Soil Microbial Biomass Carbon and Nitrogen on Enzyme Activity under Different Water and Fertilizer Optimal Management Regimes

The Mantel analysis method was used to obtain the correlation and explanation values for the relationships between soil microbial biomass carbon and nitrogen and soil enzyme activity under different water and fertilizer management conditions. It can be seen from Fig. 2a that the significant Mantel values of soil acid phosphatase, catalase, and microbial carbon were the lowest (Mantel's $p < 0.01$). Soil sucrase, acid phosphatase, catalase, and microbial nitrogen had the lowest values, while the value for sucrase (Sue) was less than 0.05 and that for urease was less than 0.05. The maximum interpretation values of the soil microbial biomass carbon and nitrogen by acid phosphatase and catalase (Mantel's $r \geq 0.5$) indicated that the soil microbial biomass carbon and nitrogen contents determined the soil acid phosphatase and catalase activities, and the maximum interpretation value of soil microbial biomass N by sucrase (Mantel's $r \geq 0.5$) indicated that the soil microbial biomass N content determined the soil sucrase (Sue) activity. However, the significant and interpreted Mantel values of urease and the soil microbial biomass carbon and nitrogen were 0.25–0.5, which were important factors affecting the microbial biomass carbon and nitrogen. The results in Fig. 2b showed that the soil enzyme activities and soil microbial biomass carbon and nitrogen had different degrees of correlation. The correlation coefficients of the soil acid phosphatase and catalase activity with the soil microbial biomass carbon and nitrogen were between 0.90 and 1.00, indicating that there was a highly significant positive correlation between the soil acid phosphatase and catalase activities and the soil microbial biomass carbon and nitrogen contents ($p < 0.01$), which was consistent with the results in Fig. 2a. The correlation coefficient of the urease and sucrase activities with the soil microbial biomass carbon and nitrogen were 0.80–0.95, indicating that the urease and sucrase activities had a significant positive correlation with the soil microbial biomass carbon and nitrogen ($p < 0.05$).

3.4. Effect Evaluation of different Water and Fertilizer Optimal Management Regimes on Soil Microbial Biomass C and N and Enzyme Activities of *Panax notoginseng* Farmland

To comprehensively and systematically reflect the effects of different water and fertilizer optimal management regimes on the soil microbial biomass carbon and nitrogen and enzyme activities of *Panax notoginseng* farmland, and avoid the difficulty in evaluating the quality of water and fertilizer management regimes due to a large number of test indexes, the entropy value method combined with TOPSIS method was used to analyze the indicators of the SMBC, SMBN, and enzyme activities in different fertility periods under different water and fertilizer optimization management regimes of *Panax notoginseng*. This method can determine the fit degree C_i and its ranking of the optimal solutions of the SMBC, SMBN, and enzyme activities under different optimal management regimes of water and fertilizer in different growth periods in order to find the optimal water and fertilizer management regimes in different growth periods. The greater the value of the optimal solution fit C_i , the better the microbial biomass carbon, nitrogen, and enzyme activities of *Panax notoginseng* soil for that treatment. As shown in Table 3, under the same irrigation treatment, the C_i values of the soil microbial biomass carbon, nitrogen, and enzyme activities of *Panax Notoginseng* followed the order of F4, F3, F2, and F1 from high to low. Under the same fertilization treatment, the C_i values decreased and then increased with the increase in irrigation, and the advantages and disadvantages of the microbial biomass carbon, nitrogen, and enzyme activity of *Panax notoginseng* soil were W2, W1, and W3, in descending order. The C_i values of the SMBC, SMBN, and enzyme activities in the rooting period were the highest under the W2F4 treatment (0.836), followed by that under the W3F4 treatment (0.794), and the lowest under the W1F2 and W1F1 treatments (0.214 and 0.133). The maximum C_i value in the seedling period was obtained under W3F4 (0.806), followed by W2F4 (0.801). W1F2 and W1F1 were the worst treatments for the SMBC, SMBN, and enzyme activities, and the C_i values were 0.263 and 0.111, respectively. The C_i value of the W3F4 treatment was the highest (0.733), followed by that of the W2F3 treatment (0.568). The SMBC, SMBN, and enzyme activity C_i values under W2F1 and W1F1 were the lowest, which were 0.216 and 0.189, respectively. The worst soil microbial carbon, nitrogen, and enzyme activities were obtained under W2F1 and W1F1 the C_i values were 0.273 and 0.068, respectively. According to the analysis of the optimum regime C_i for the SMBC, SMBN, and enzyme activities in different growth periods under different optimal water and fertilizer management regimes, the W2F4 treatment should be used in the rooting period and the W3F4 treatment should be used in the seedling, flowering, and fruiting periods to regulate the microbial biomass carbon, nitrogen, and enzyme activities of *Panax notoginseng* soil with the best effect.

Table 3
Comprehensive evaluation of the water and fertilizer control treatments in different growth periods

Treatment	Rooting Period					Seedling Period					Flowering Period
	Weight	Positive	Negative	Relative	Rank	Weight	Positive	Negative	Relative	Rank	Weight
	W_j	Ideal	Ideal	Proximity C_i		W_j	Ideal	Ideal	Proximity C_i		W_j
		Solution	Solution				Solution	Solution			
		Distance D_i^-	Distance D_i^+				Distance D_i^-	Distance D_i^+			
W1F1	0.074	0.366	0.056	0.133	13	0.075	0.361	0.045	0.111	13	0.076
W1F2	0.076	0.323	0.088	0.214	12	0.078	0.296	0.106	0.263	12	0.077
W1F3	0.078	0.249	0.166	0.400	8	0.078	0.225	0.168	0.428	8	0.077
W1F4	0.078	0.144	0.267	0.651	4	0.078	0.185	0.221	0.545	5	0.076
W2F1	0.075	0.296	0.126	0.299	10	0.075	0.279	0.122	0.304	10	0.076
W2F2	0.074	0.268	0.139	0.342	9	0.074	0.235	0.158	0.402	9	0.075
W2F3	0.075	0.157	0.240	0.605	5	0.075	0.148	0.242	0.621	4	0.075
W2F4	0.075	0.066	0.336	0.836	1	0.074	0.081	0.319	0.801	2	0.074
W3F1	0.078	0.262	0.185	0.414	7	0.077	0.246	0.195	0.443	7	0.076
W3F2	0.079	0.197	0.219	0.526	6	0.078	0.215	0.191	0.472	6	0.077
W3F3	0.079	0.092	0.316	0.774	3	0.078	0.132	0.282	0.681	3	0.077
W3F4	0.077	0.097	0.374	0.794	2	0.076	0.087	0.362	0.806	1	0.077
CK	0.068	0.358	0.109	0.233	11	0.069	0.337	0.126	0.272	11	0.074

4. Discussion

4.1. Effect Mechanism of Optimal Water and Fertilizer Management Regimes on Soil Microbial Biomass Carbon and Nitrogen for *Panax notoginseng*

This study found significant differences ($p < 0.05$) in the SMBC, SMBN, and enzyme activities during different growth periods under the same water and fertilizer optimal management regimes, and the values tended to increase first and then decrease with the growth of *Panax notoginseng*, which may have been due to the different physiological activities of *Panax notoginseng* during different growth periods. The SMBC and SMBN contents ranged from high to low in the flowering, fruiting period, seedling period, and rooting period, with the highest in the flowering period and the lowest in the rooting period. Bargali, K et al. found that the soil microbial biomass carbon of different forest types showed significant seasonal changes, with the minimum value occurring in the winter and the maximum value occurring in the rainy season. Lepcha, N.T. et al. studied the effects of land-use types and seasonal changes in the eastern Himalayas on the soil microbial biomass carbon and nitrogen and found that the soil microbial biomass carbon and nitrogen of different land-use types showed strong seasonal differences ($p < 0.001$), with the highest values occurring in the rainy season and the lowest values occurring in the winter. These findings are consistent with the trends found in this study. In this study, the highest soil microbial biomass carbon and nitrogen contents occurred in the flowering season and the lowest occurred in the root-growth period because the dry and wet seasons in the study area were distinct, with a rainy summer and dry winter and spring, abundant rainfall in the summer, humid air, and low evaporation. The soil temperature and humidity were suitable for the growth, reproduction, and decomposition activities of soil microorganisms, and the increase in microbial activity promoted the conversion and accumulation of the soil microbial biomass carbon and nitrogen contents. Huang et al. found that the interaction between an increased amount of irrigation and season had significant effects on the soil microbial respiration, SMBC, and SMBN, with significant increases in the summer and autumn, but no effects in the spring and winter. Rehydration in the summer and autumn increased the bacterial biomass, as well as the bacterial and fungal diversity and abundance, which was in contrast with the finding by Li et al. that the soil microbial biomass carbon and nitrogen in the Mediterranean agro-ecological system had the lowest values in the summer and the highest values in the autumn because of the different climatic types in the two regions. The Mediterranean region is hot and dry in the summer and warm and rainy in the winter, in contrast to the rainy summer and dry winter and spring of the studied region.

This study found significant differences ($p < 0.05$) between the SMBC, SMBN, and enzyme activities in the same growth period under different water and fertilizer optimal management regimes, which may be due to differences in the amounts of irrigation and fertilizer applied, and the SMBC and SMBN contents increased with the increases in the irrigation and fertilizer application amounts. A study showed that long-term fertilization improved the SMBC and dehydrogenase activities, and organic fertilizers had a greater effect on the SMBC and dehydrogenase activities than mineral fertilizers. This may be due to the fact that organic fertilizers can better increase the organic matter content of soil and improve the soil physicochemical properties compared with mineral fertilizers. Increases in the lower soil elemental and microbial biomass stoichiometry SMBC:SMBP (soil microbial biomass phosphorus) and SMBN:SMBP and LAP (leucine aminopeptidase) were mainly due to the combined use of manure and mineral fertilizers, accelerating SOC and N mineralization. The C_i comparison showed that the optimum treatment was W2F4 for the rooting period and W3F4 for the seedling, flowering, and fruiting periods; in addition, the W1F1 treatment was the least effective in regulating the soil microbial biomass in terms of carbon and nitrogen during all growth periods, while the W1F2

treatment was not suitable for the rooting and seedling periods and the W2F1 treatment did not achieve good results for the flowering and fruiting periods. This showed that 5 mm irrigation and $3.20 \text{ kg} \cdot 667^{-1} \cdot \text{m}^{-2}$ fertilizer application was not suitable for regulating the SMBC, SMBN, and enzyme activities during the *Panax notoginseng* growth periods.

4.2. Effect Mechanism of Water and Fertilizer Optimal Management Regimes on Soil Enzyme Activity of *Panax notoginseng*

Soil enzymes are key components for the catalytic decomposition of soil nutrients, and their activity in soil can be used as a measure of soil health. This study found significant differences in the soil urease, sucrase, acid phosphatase, and catalase activities during different growth periods under the same water and fertilizer optimal management regimes ($p < 0.05$), and increasing and then decreasing trends were observed with the prolongation of the growth period of *Panax notoginseng*, which ranged from large to small in the flowering period, fruiting period, seedling period, and rooting period. This is consistent with the findings of Pei et al., who reported that soil enzyme activities had obvious seasonal variation characteristics, with the soil sucrase activity decreasing in the order of summer > autumn > spring > winter and the urease activity decreasing in the order of summer > spring > autumn > winter. The results showed that the two soil enzyme activities were the highest in the summer and the lowest in the winter similar. Zhu et al. reported that the soil enzyme activities had obvious seasonal variation characteristics, with the peak values of soil catalase, phosphatase, and urease in the reed community appearing in the exuberant growth period, and the trough values appearing in the germination period and the leaf-development period. In this study, the flowering period of *Panax notoginseng* was in the summer and it grew vigorously, while the rooting period was in the winter, which was the germination period. Therefore, the soil enzyme activities of *Panax notoginseng* were the highest in the flowering period and the lowest in the rooting period. The reason was that summer was the rainy season, with plenty of rainfall. The soil temperature and humidity were suitable for the growth and reproduction of microorganisms, and the enzyme activities in the soil increased with the increase in microbial activities.

This study found that, under different water and fertilizer optimal management regimes, the soil urease, sucrase, acid phosphatase, and catalase showed significant differences in the same growth period ($p < 0.05$). The contents of soil urease, sucrase, acid phosphatase, and catalase increased with the increase in the irrigation amount and fertilizer application amount, and the soil urease, sucrase, acid phosphatase, and catalase were the highest in the W3F4 treatment during the flowering period. In terms of the soil enzyme activities, W3F4 should be selected as the most suitable treatment in all growth periods, while the W1F1 treatment is the least suitable for improving the *Panax notoginseng* soil enzyme activities.

4.3. Mechanism of Interaction of Water and Fertilizer on SMBC, SMBN, and Enzyme Activity of *Panax notoginseng* in Farmland

Water fertilizer coupling can achieve the reasonable coordination of irrigation and fertilization in terms of time, quantity, and method according to the demands for water and fertilizer during crop growth. Vepsilinen et al. suggested that chemical fertilizers affected soil enzyme activity by improving the soil's physical and chemical properties and microflora and that an appropriate ratio of water to fertilizer had an important effect on improving the soil fertility and crop yield. Reasonable irrigation can improve soil enzyme activity. Too much or too little soil moisture is not conducive to the growth and reproduction of soil microorganisms, resulting in a reduction in soil enzyme activity. This study found different distribution characteristics of the SMBC, SMBN, and enzyme activities' optimal regime fits under different optimal water and fertilizer management regimes during different fertility periods. This study found a significant correlation between the SMBC, SMBN, and soil enzyme activities ($p < 0.05$), a highly significant positive correlation between the soil acid phosphatase and catalase and SMBC and SMBN ($p < 0.01$), and urease and sucrase were significantly and positively correlated with the SMBC and SMBN ($p < 0.05$). This was similar to the finding by Eivazi, F. et al. that the activities of soil urease, sucrase, and alkaline phosphatase had highly significant positive correlations with the SMBC, SMBN, and phosphorus. This may have been due to the fact that soil enzymes are mainly derived from soil microorganisms, plant roots, and soil fauna; therefore, there may be a closer relationship between the soil enzyme activity and soil microbial biomass. Yang et al. found a highly significant positive correlation between the soil microbial biomass C, N, and P and soil urease, sucrase, and alkaline phosphatase activities. The study also revealed that soil invertase, acid phosphatase, and catalase were the main factors affecting the regulation of the SMBC and SMBN, while urease was an important factor affecting the SMBC and SMBN. In this experiment, W2 was 60% of the field water-holding capacity and W3 was 80% of the field water holding capacity. In the rooting period, the W2F4 treatment had the best regulation effect on the SMBC, SMBN, and enzyme activities, which was the same as Tang et al.'s conclusion that the soil enzyme activities of *Zanthoxylum bungeanum* Maxim. under fertilization coupling treatment were the highest when the soil water content was 62.0% of the field water-holding capacity. The regulation effect of W2 ranked second in the seedling period and flowering period, and W3 ranked in the first two places in the fruiting period, with W3F4 being the most suitable treatment in the seedling period, flowering period, and fruiting period, followed by W2F4, W2F3, and W3F3 respectively.

5. Conclusions

The characteristics of the SMBC, SMBN, and enzyme activities of *Panax notoginseng* in farmland were studied through the optimal management of water and fertilizer. The main conclusions were as follows:

Under the same optimal management regimes for water and fertilizer, the differences in the SMBC and SMBN contents and soil enzyme activities were significant between different growth periods ($p < 0.05$). The SMBC and SMBN contents and soil enzyme activities increased firstly and then decreased over time, and gradually increased from the rooting period to the flowering period; there were significant differences in the SMBC and SMBN contents and soil enzyme activities between the same growth period and different water and fertilizer optimal management regimes ($p < 0.05$), and the SMBC and SMBN contents and soil enzyme activities increased with increases in the irrigation amount and fertilizer application amount.

There was a significant correlation between the SMBC and SMBN and soil enzyme activities ($p < 0.05$); the correlation coefficients of acid phosphatase and catalase with the SMBC and SMBN were in the range of 0.90–1.00 under the optimal water and fertilizer management regime, while those of urease and sucrase with the SMBC and SMBN were in the range of 0.80–0.95. The C and N contents of the soil microbial biomass influenced the soil enzyme activities.

The comprehensive evaluation of the SMBC, SMBN, and enzyme activities under the optimal water and fertilizer management regimes showed that controlling irrigation at 10 mm and fertilizer application at $8 \text{ kg.667}^{-1}.\text{m}^{-2}$ during the rooting period of *Panax notoginseng*, and using 15 mm irrigation and setting the fertilizer application amount at $8 \text{ kg.667}^{-1}.\text{m}^{-2}$ during the seedling, flowering and fruiting periods could effectively regulate the SMBC, SMBN, and enzyme activities, which was beneficial to the growth and yield improvement of *Panax notoginseng* plants.

Declarations

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Author contributions

Conceptualization, Tuo, Y. and Liu, X.; methodology, Tuo, Y.; software, Shen, F.; validation, Liu, X., Zheng, Y. and Wang, Q.; formal analysis, Tuo, Y.; investigation, Wang, F.; resources, Tuo, Y.; data curation, Wang, Z., Shi, X. and Ding, M.; writing original draft preparation, Tuo, Y.; writing review and editing, Tuo, Y.; visualization, Tuo, Y.; supervision, Xiang, P.; project administration, Yang, Q.; funding acquisition, Tuo, Y.. All authors have read and agreed to published the manuscript.

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Availability of Data and Materials

All data supporting the conclusions of this article are provided within the article (and its additional files).

Declarations

Ethics Approval and Consent to Participate

Not applicable.

Consent for Publication

Not applicable.

Competing interests

All authors declare no competing interest.

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Figures

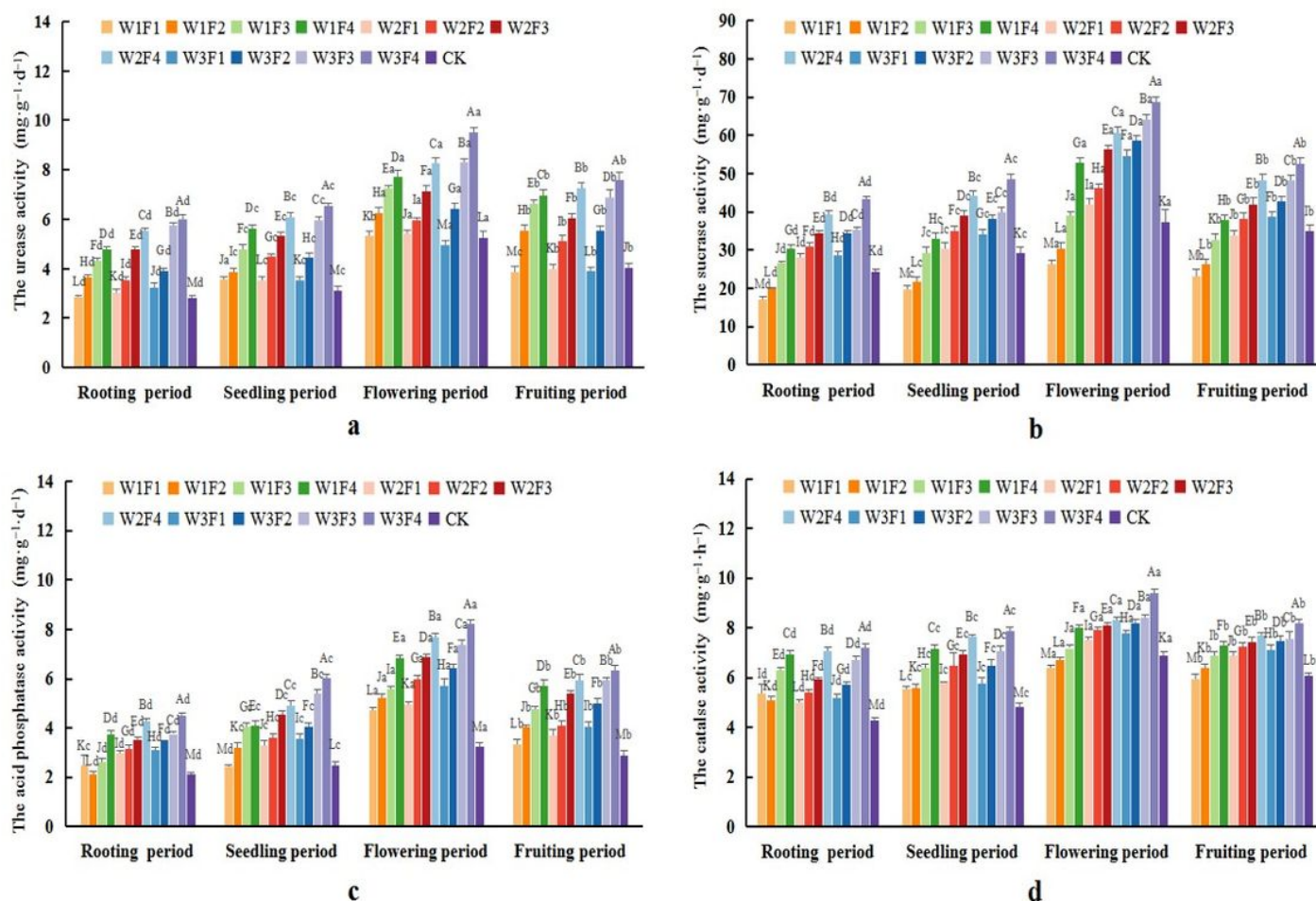


Figure 1

Characteristics of the changes in the soil enzyme activity under different optimal water and fertilizer management regimes.

Note 1: W1F1, W1F2, W1F3, etc. are the 13 treatments designed in this experiment.

Note 2: Different capital letters indicate significant differences between the different water and fertilizer control treatments in the same growth period ($p < 0.05$) and different lowercase letters indicate significant differences between the different growth periods under the same water and fertilizer control treatments ($p < 0.05$).

Note 3: (a) Urease activity; (b) Sucrase activity; (c) Acid phosphatase activity; (d) Catalase activity.

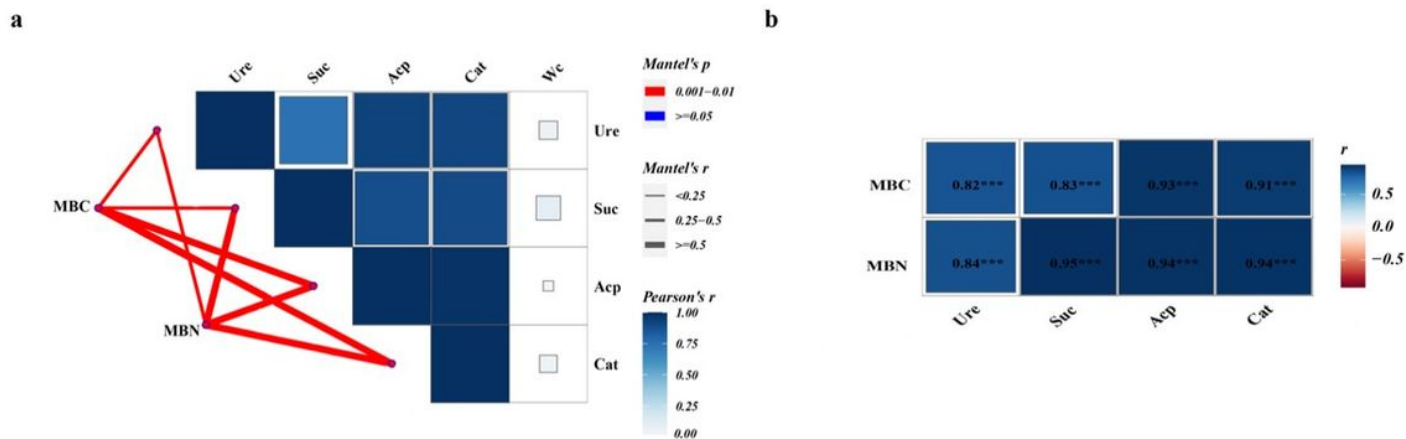


Figure 2

Mantel analysis of the soil enzyme activity with the soil micro biomass carbon, nitrogen, and their ratios.

Note 1: MBC is microbial biomass carbon, MBN is microbial biomass nitrogen, Ure is soil urease, Suc is soil sucrase, Acp is soil acid phosphatase, and Cat is soil catalase.

Note 2: (a) Mantel analysis; (b) Correlation between SMBC, SMBN and enzyme activities.

Note 3: *** indicate a significant correlation at the 0.001 levels.