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RESPONSES OF CARBON, NITROGEN AND PHOSPHORUS STOICHIOMETRY IN DIFFERENT ROOT ORDERS OF *ZANTHOXYLUM ARMATUM* SEEDLINGS TO FOUR SOIL TYPES*

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ABSTRACT

The effects of alluvial soil (AS), alkaline purple soil (ALS), red soil (RS) and yellow soil (YS) on the biomass, nutrient content and the stoichiometric ratio of roots were studied in *Zanthoxylum armatum* seedlings. Results showed that soil types, root order and their interaction had significant effects on biomass, C, N, P content and C:N, C:P ratios of *Z. armatum* ($P < 0.05$). The total biomass of roots in ALS was the largest and significantly higher than in the other soil types ($P < 0.05$), and the biomass of different root orders increased with the increase of root order, reaching the maximum in the fourth-order roots, where it was significantly higher than in the other root orders ($P < 0.05$). The root C content and C:P ratio were the largest in YS, the N content and N:P ratio were the largest in RS, and the P content was the largest in ALS. Root biomass, C content, C and N distribution ratio, C:N and C:P ratios showed the same trend, and increased with the increase of a root order, but the N and P content decreased with an increased root order. In AS, ALS and YS, the root N:P ratio was less than 12, indicating that the root growth was restricted by N to varying degrees. In RS, the root N:P ratio ranged from 5.8 to 12.1, indicating that the root growth in RS was restricted by N and P. The results will help to provide a theoretical basis and reference for the selection of soil type, cultivation and fertilization management of *Z. armatum*.

Keywords: soil types, root order, nutrient elements, accumulation and distribution, stoichiometric ratio, winged prickly ash.

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INTRODUCTION

Soil is the key to the survival of plants. It may provide water, nutrients and other conditions for plants to survive, and plays a crucial role in plant colonization, competition, growth and development (HOU et al. 2011). Physical and chemical properties of soil affect the growth status of plants, and the plant growth in turn acts on the soil to gradually improve its growth environment. Different soil types have different physical and chemical properties, which may affect nutrients' uptake, thus changing the morphology, growth and substance metabolism of plants (ANTONIO et al. 2019). The effects of different soil types on biomass allocation and nutrient allocation of seedlings may help us to understand their growth characteristics, but also to promote seedling establishment and growth by regulating the changes of soil nutrients (WANG et al. 2013). Thus, it is significant to study the relationship between plant growth and different soil types for evaluating plant regeneration and succession and forest restoration. Roots are not only the support for the growth of aerial parts, but also provide necessary nutrients and water for the growth and development of plants, and maintain the circulation and exchange function of matter and energy among soil and aerial parts of plants and the atmosphere (SONG et al. 2012). Soil types may affect the growth, development, distribution, morphological characteristics and function of roots due to differences in physical and chemical properties and fertility (WANG et al. 2017a). In order to adapt to the heterogeneity of soil types, plants adjust the distribution, morphology, configuration and physiological characteristics of roots during the development process, and promote the accumulation and distribution of water and nutrient elements to achieve the goal of optimal competition for soil nutrients and water. The root is the link connecting plants and soil, and it is the most active part underground of plants and the most sensitive plant organ to the soil environment (LIU et al. 2020). The application of branch levels (root order) to research of the C, N and P stoichiometric ratio may better predict the interaction among nutrient elements, as well as the structure and function of roots (PREGITZER 2002). Studies on the ecological stoichiometric characteristics of different root orders in different soil types will not only help to understand the relationship between the supply and demand of nutrient elements in soil and plants, but also provide a scientific basis for nutrient management.

Carbon (C), nitrogen (N) and phosphorus (P) are important essential elements found in soil for plant growth and development, and the most universal and important limiting nutrients in the ecosystem (TANG et al. 2015). Roots absorb N and P nutrients from the soil, and these elements are then used in transformations via photosynthesis, chlorophyll synthesis and during the physiological growth and metabolism of plants on the ground. The content and cycle changes of N and P nutrients in the plants-soil system play an important role in regulating the growth and physiology of plants (ZENG et al.

2016). Thus, a sufficient supply of N and P nutrients has an important effect on the growth of plant roots and the stability of the ecosystem. Recently, plant roots of different forms and functions have attracted much attention among scholars. Nowadays, the structure and functions of root are clearly understood, but the plant and soil nutrient interaction is still in the stage of preliminary research (HE et al. 2017). Early studies have shown that nutrient absorption, transformation and transport in some plant species on a single soil type received much attention, but studies comprising different soil types have not yielded unambiguous results yet.

Z. armatum is a small deciduous tree, belonging to the *Zanthoxylum* in the Rutaceae family, and is has significant economic value as an important food condiment, spice, and medicinal plant. It is characterized by a developed root system, resistance to drought and poor quality soil, fast growth, early fruit-bearing, simple cultivation and management, and high yield (PHUYAL et al. 2018). In southwest China, *Z. armatum* is planted in a wide area on various soil types. Significant differences in quality traits of *Z. armatum* fruit from four different soil types have been observed, alongside differences in particular traits and yields (PUROHIT et al. 2017). Thus, the aim of this study has been to improve our knowledge of the biomass and C, N, and P stoichiometry characteristics in *Z. armatum* roots. However, results obtained with plants grown in pot experiments may not be directly compared to those in field conditions, and provide data concerning the site selection and fertilization management of *Z. armatum* during cultivation.

MATERIAL AND METHODS

Study area overview

The experimental site is No. 5 teaching building of Sichuan Agricultural University, Chengdu, China. The region lies in the mid-latitude inland subtropical monsoon climate with mild climate, four distinct seasons and abundant rainfall. The average annual rainfall is 896.1 mm, the rainy season is mainly from June to September, the annual average temperature is 16.4°C, the annual average relative humidity is 84%, the average annual sunshine hours are 1104.5 h, and the annual frost-free period is 282 days.

Experimental material

Z. armatum seedlings were purchased from Dan-ling, Sichuan, China, in April 2018. Alluvial soil (AS), alkaline purple soil (ALS), red soil (RS) and yellow soil (YS) belong to the main types of soil widely distributed and representative in Sichuan, China. They were collected in Chengdu Campus of Sichuan Agricultural University, Chengdu, China (30°42'N, 103°51'E, altitude 542 m), Ji-feng Town, Zhong-jiang County, De-yang, Sichuan, China

(31°03'N, 104°68'E, altitude 900 m), San-xing Village, Feng-le Town, Shi-mian, Sichuan, China (29°32'N, 102°54'E, altitude 878 m), and Bai-sheng Village, Bao-lin Town, Qiong-lai, Sichuan, China (30°21'N, 103°30'E, altitude 552 m), respectively. The basic physical properties of the four soils were analyzed (Table 1). The soil was classified, crushed, screened and tilled for 2 days under strong light for further use.

Table 1

The basic physical and chemical properties of three types of soils (means \pm SE, $n=5$)

Soil type	pH	Organic matter (g kg ⁻¹)	Bulk density (g cm ⁻³)	Total nitrogen (g kg ⁻¹)	Total phosphorus (g kg ⁻¹)	Total potassium (g kg ⁻¹)
AS	5.12 ^a ±0.14	35.51 ^a ±0.74	1.29 ^a ±0.04	1.67 ^a ±0.09	0.83 ^a ±0.03	2.49 ^b ±0.06
ALS	8.67 ^a ±0.23	20.99 ^b ±0.91	1.25 ^b ±0.08	1.65 ^a ±0.15	0.30 ^b ±0.01	3.56 ^a ±0.10
RS	5.14 ^a ±0.16	16.64 ^a ±0.66	1.30 ^a ±0.06	1.14 ^b ±0.07	0.07 ^a ±0.00	3.15 ^a ±0.08
YS	4.79 ^b ±0.12	33.50 ^a ±0.64	1.32 ^a ±0.07	1.89 ^a ±0.11	0.08 ^a ±0.00	2.21 ^b ±0.09

Values followed by the same letter in the same column are not significantly different according to the Tukey's test ($\alpha=0.05$).

Seedling cultivation and root collection

Seedlings with the height of 13.3 cm and ground diameter of 0.27 cm were selected and transplanted into pots containing 10 kg of different types of soil (pot diameter 28 cm, pot height 22 cm, and the soil layer height 15 cm). The experiment consisted of 4 treatments with 18 replicates per treatment and a total of 72 pots. During the experiment, relative soil moisture was kept at about 60~80%, and weed control and pest control management were carried out uniformly. After culturing for 5 months, 5 seedlings were randomly selected from each soil type, and the roots were washed with deionized water. The division of root order was based on the root initiation point. The unbranched roots at the farthest end of the root axis were first-order root, first-order root were attached to second-order root, second-order root were attached to third-order root, and third-order root were attached to fourth-order root. Roots that did not branch from higher roots were also divided into first order roots. After grading, the roots of different root orders were put into the corresponding vessels, marked for grading, and their biomass was determined.

Determination of carbon, nitrogen and phosphorus content

Dried root of different order was placed in a mill (FW80, Taisite, China), ground and passed through a sieve to obtain plant samples. Plant samples were mineralized in concentrated H₂SO₄-HClO₄, and the following methods were applied: the potassium dichromate oxidation method, semimicro-Kjeldahl method and Mo-Sb colorimetric method to determine the C, N and P content, respectively (WU et al. 2019).

Data and statistical analysis

The element allocation ratio of different root order was calculated according to the biomass and element content of different root order, and the calculation formula was as follows: element allocation ratio = (element content \times biomass) / total element accumulation. Data processing and statistical analysis were completed with Excel 2010 and SPSS 20.0 software. A one-factor analysis of variance (one-way ANOVA) and the least significant method (LSD) were used to determine parameters in different soil types or root orders, while a two-factor analysis of variance (two-way ANOVA) and the least significant method (LSD) were employed to test differences between soil types, different root orders and the interaction of root of biomass, C, N and P content, and the impact of the stoichiometric ratio ($\alpha=0.05$). The Pearson test was used to analyze the correlation between root nutrient indexes and nutrient indexes in different soil types. Canoco 5.0 software was used to analyze the correlation between soil nutrients and root biomass and nutrient indexes. The tables were drawn with Origin 9.1 software. Data in the tables are expressed as "mean \pm standard error".

RESULTS

Effects of four soil types on biomass

As shown in Figure 1 and Table 2, soil type, different root order and their interaction had significant effects on the biomass ($P<0.05$). Root biomass showed similar trend under different soil types. The order of root biomass were ALS>YS>AS>RS, and the value in ALS was 45.5% higher than those of in RS. Moreover, the maximum of root biomass of different branch orders was also observed in ALS. The biomass of different root order increased with the increase of root order, and showed increase trend, and reached the maximum at the fourth-order root, which was significantly higher than those of other root, with the maximum difference of 72.8%.

Effects of soil types on C, N and P contents

As shown in Table 2, soil type, different root order and their interaction had significant effects on the C content ($P<0.05$), and soil type and different root order had significant effects on the C content of *Z. armatum* ($P<0.05$), but their interaction had no significant effects on P content ($P>0.05$). As shown in Figure 2, the order of C content in the roots were YS, AS, ALS, and RS, respectively. The order of N content were RS, YS, ALS, and AS, respectively, and the order of P content were ALS, AS, RS and YS, respectively. The root C content of *Z. armatum* was significantly different cultured in different soil types, but the root P content was not significantly different cultured in different soil types. In the comparison of root element contents

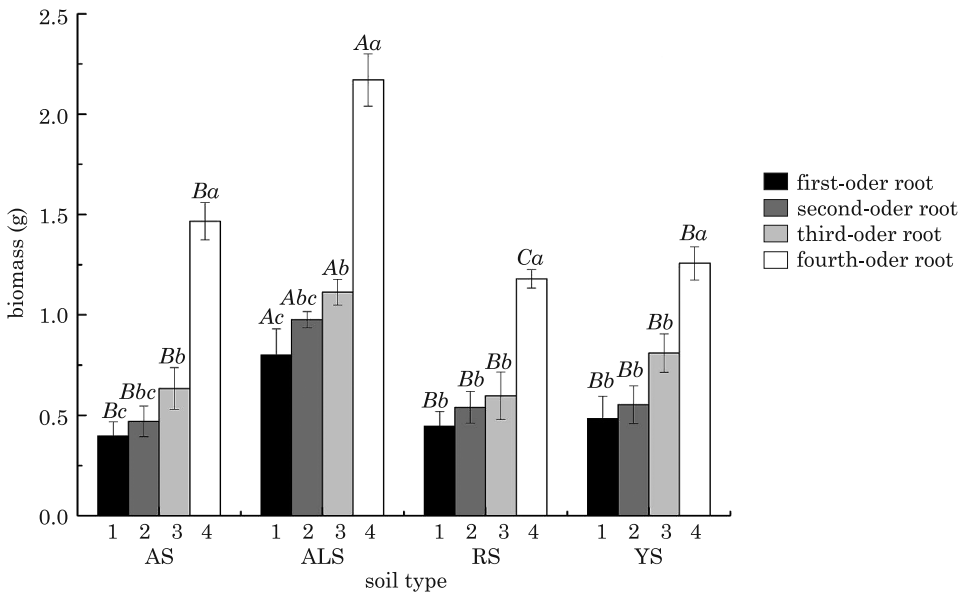


Fig. 1. Effects of four soil types on biomass in different root orders of *Zanthoxylum armatum*. Data are shown as means \pm SE ($n=5$). Different uppercase letters indicate significant difference in different soil types ($P<0.05$). Different lowercase letters indicate significant difference in different root orders ($P<0.05$)

Table 2

Two-way ANOVA of soil types and different root order on biomass, C, N and P contents, allocation proportion and stoichiometric ratios of *Zanthoxylum armatum*

Source of variation	Soil type			Root order			Soil type \times Root order		
	df	F	P	df	F	P	df	F	P
Biomass	3	101.32	<0.001	3	283.22	<0.001	9	7.23	<0.001
C content	3	29.60	<0.001	3	53.10	<0.001	9	9.18	<0.001
N content	3	65.43	<0.001	3	63.81	<0.001	9	8.29	<0.001
P content	3	30.36	<0.001	3	25.93	<0.001	9	1.78	0.111
C allocation proportion	3	0.41	0.748	3	5372.51	<0.001	9	58.09	<0.001
N allocation proportion	3	0.00	1.000	3	408.95	<0.001	9	31.32	<0.001
P allocation proportion	3	0.00	1.000	3	386.49	<0.001	9	7.78	<0.001
C:N	3	69.69	<0.001	3	107.17	<0.001	9	8.20	<0.001
C:P	3	19.86	<0.001	3	19.86	<0.001	9	2.90	0.013
N:P	3	70.96	<0.001	3	2.89	0.051	9	7.24	<0.001

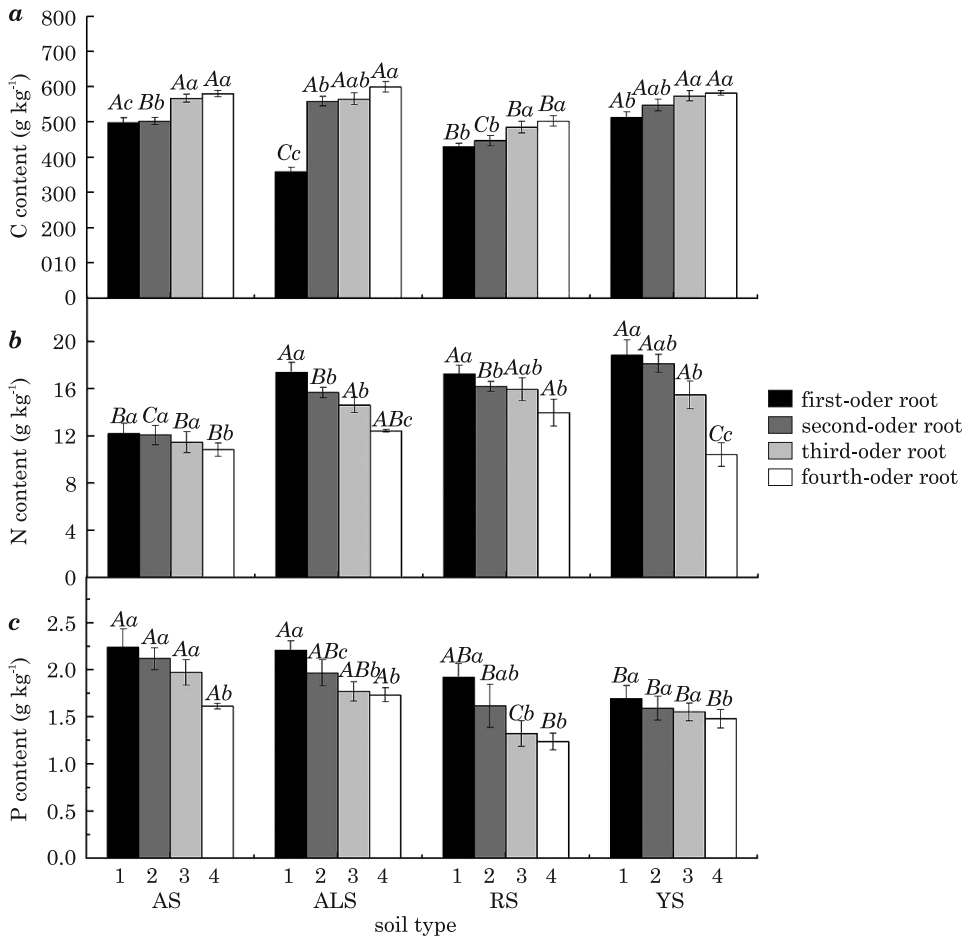


Fig. 2. Effects of four soil types on C content (a), N content (b), P content (c) in different root order of *Zanthoxylum armatum*. Data are shown as means \pm SE ($n=5$). Different uppercase letters indicated significant difference in different soil types ($P<0.05$). Different lowercase letters indicated significant difference in different root order ($P<0.05$)

in different root order cultured in the same soil, C content increased with the increase of root order, and C content reached the maximum at the fourth-order root, with the maximum increment of 40.3%. The trend of N and P content was similar, but the record showed a reverse trend to that of in C content. The N and P content decreased with the increase of the root order, and the fourth-order root was the smallest, with the maximum reduction of 44.7% and 35.4%, respectively.

Effects of soil types on C, N and P allocation proportion

As shown in Figure 3 and Table 2, different soil type had no significant effect on the C, N and P distribution in roots ($P>0.05$), but different root or-

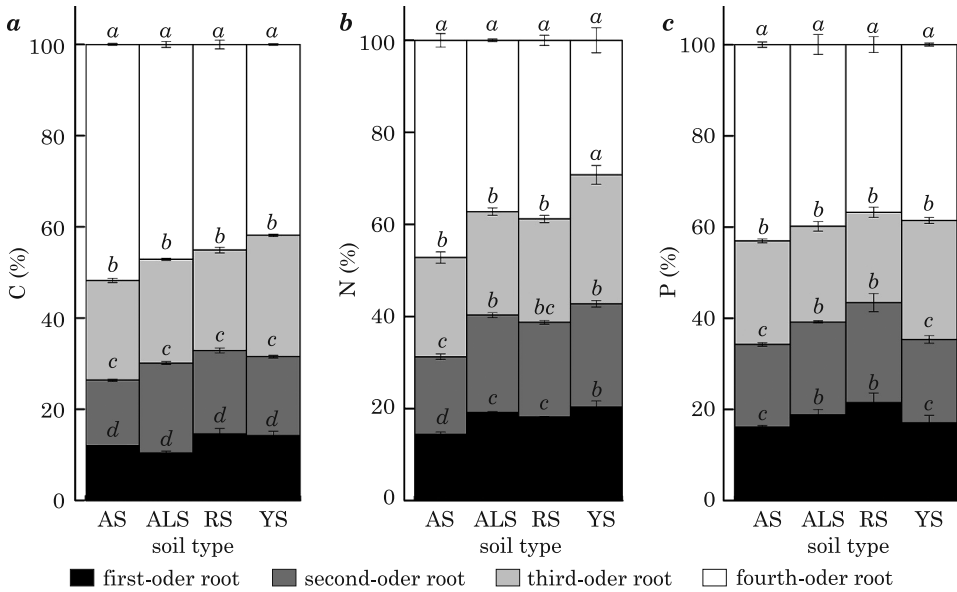


Fig. 3. Effects of soil types on C percentage (a), N percentage (b), P percentage (c) in different root order of *Zanthoxylum armatum*. Data are shown as means \pm SE ($n=5$). Different uppercase letters indicated significant difference in different soil types ($P < 0.05$). Different lowercase letters indicated significant difference in different root order ($P < 0.05$)

der and their interaction caused significant differences in the C, N and P distribution in roots ($P < 0.05$). The response of the C, N and P distribution ratio in roots cultured in different soil types was inconsistent, but the difference of the N distribution ratio among different soil types was considerable. However, the differences in the C and P distribution ratios in roots among the different soil types were not obvious. The C and N distribution ratios in plants grown in the same soil showed a similar change in different root orders, and increased with the increase of a root order, reaching the maximum in the fourth-order roots. The P distribution in RS showed different trends, and the values reached the maximum in the fourth-order root.

Effects of soil types on C, N and P stoichiometry

As shown in Figure 4 and Table 2, soil type, different root order and their interaction had significant effects on the C:N and C:P ratios in roots ($P < 0.05$), and the soil type had significant effects on the N:P ratio in roots ($P < 0.05$), but there was no significant difference in the N:P ratio between different root orders ($P > 0.05$). The results showed that the order of the C:N ratio in the roots were AS, YS, ALS, and RS, the order of the C:P ratio in the roots were YS, RS, AS, ALS, and the order of the N:P ratio in the roots were RS, ALS, YS, and AS. In different soil types, the C:P ratio in different root orders showed a big difference, but the N:P ratio did not differ much. The root stoichiometric ratio among different root orders of plants cultured

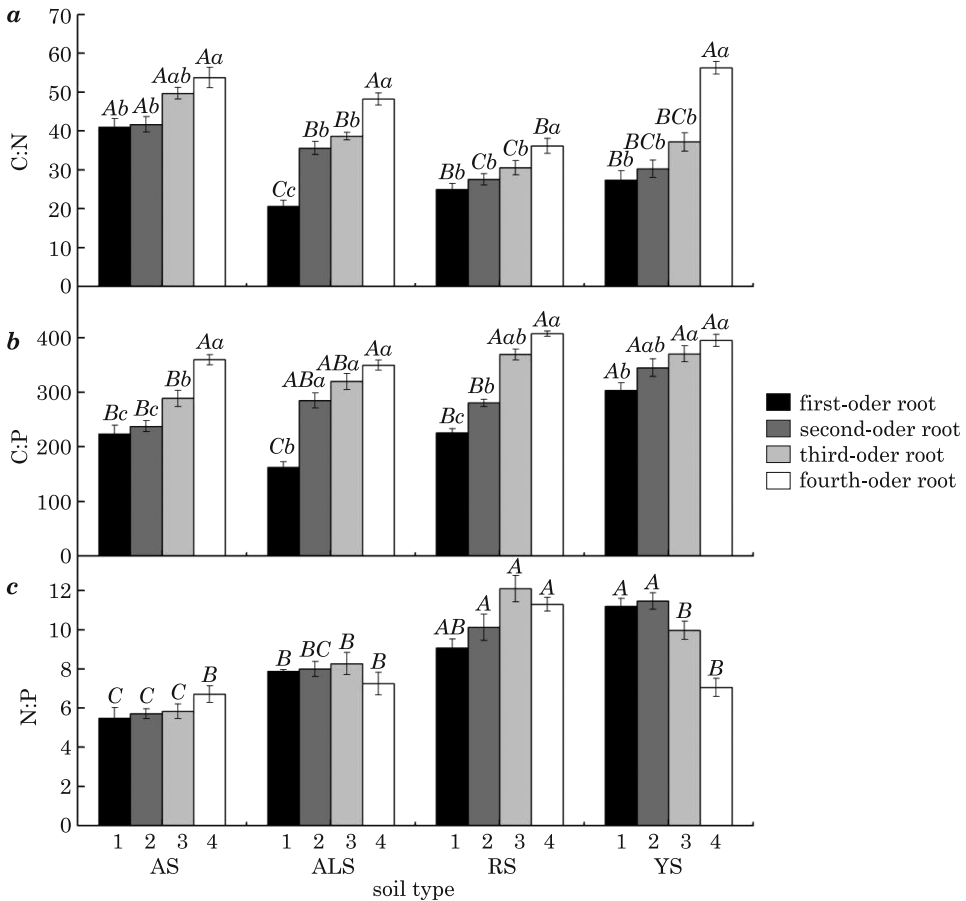


Fig. 4. Effects of soil types on C:N (a), C:P (b), N:P (c) in different root order of *Zanthoxylum armatum*. Data are shown as means \pm SE ($n=5$). Different uppercase letters indicated significant difference in different soil types ($P<0.05$). Different lowercase letters indicated significant difference in different root order ($P<0.05$)

in the same soil showed a relatively consistent change trend compared to those of the C:N and C:P ratios, which all increased with the increase of the root order, and the maximum increment was 57.2% and 53.6%, respectively. Except AS, the N:P ratio first increased and then decreased with the increase of root orders in the other soils. The largest N:P ratio was observed in the third-order root in ALS and RS, in the second-order root of YS, and in the fourth-order root of AS, respectively.

Correlation analysis of biomass and nutrient indices among different root order

As shown in Table 3, the correlations between biomass of the second and third-order roots and nutrient indices were similar, while the correlations

Table 3

Correlation coefficients among biomass, C, N and P content and their stoichiometric ratios in roots of *Zanthoxylum armatum*

Root order	Index	Biomass	C	N	P	C:N	C:P	N:P
First-order root	biomass	1						
	C	-0.688*	1					
	N	0.378	-0.249	1				
	P	0.223	-0.400	-0.646*	1			
	C:N	-0.623*	0.696	-0.863**	0.284	1		
	C:P	-0.497	0.823**	0.286	-0.839**	0.210	1	
	N:P	0.071	0.122	0.898**	-0.899**	-0.596*	0.650*	1
Second-order root	biomass	1						
	C	0.562	1					
	N	0.196	0.189	1				
	P	0.157	0.187	-0.706*	1			
	C:N	0.041	0.331	-0.856**	0.748**	1		
	C:P	0.128	0.391	0.751**	-0.825**	-0.492	1	
	N:P	-0.029	-0.013	0.902**	-0.935**	-0.850**	0.866**	1
Third-order root	biomass	1						
	C	0.501	1					
	N	0.133	-0.418	1				
	P	0.143	0.659*	-0.723**	1			
	C:N	0.004	0.651*	-0.963**	0.817**	1		
	C:P	0.037	-0.314	0.625*	-0.910**	-0.637*	1	
	N:P	-0.108	-0.676*	0.868**	-0.958**	-0.930**	0.837**	1
Fourth-order root	biomass	1						
	C	0.647*	1					
	N	0.012	-0.708**	1				
	P	0.694*	0.715**	-0.343	1			
	C:N	0.155	0.821**	-0.977**	0.459	1		
	C:P	-0.492	-0.353	0.046	-0.898**	-0.137	1	
	N:P	-0.415	-0.890**	0.822**	-0.809**	-0.887**	0.558	1

* significantly correlated at the 0.05 level (bilateral), ** significantly correlated at the 0.01 level (bilateral)

between the first and fourth-order roots were significantly different. There was a significant negative correlation between the nitrogen content and C:N ratio, and a significant positive correlation between the nitrogen content and N:P ratio. The P content was significantly negatively correlated with the C:P and N:P ratios, and positively correlated with the C:N ratio; the C:N ratio

was negatively correlated with the N:P ratio. Moreover, there was a significant negative correlation between the N and P content, and a significant positive correlation between the C:P and N:P ratios in the first three root orders. There was a significant positive correlation between the P content and C:N ratio in the second and third-order roots. Moreover, there was a significant negative correlation between the biomass and C content and C:N ratio in the first-order roots, but there was a significant positive correlation between the biomass and C content and P content in the fourth-order roots, while there was no significant correlation between biomass and nutrient indexes in the second and third-order roots.

Correlation analysis of soil nutrients and root biomass and nutrient indexes

Redundancy analysis (RDA) is a sequencing method combining regression analysis with principal component analysis, which was used here to analyze the effects of soil nutrients on the root biomass, C, N and P content and stoichiometric ratio of *Z. armatum*. As shown in Figure 5, a response variable matrix was established based on the data of the biomass and stoichiometric ratios in roots of *Z. armatum* cultured in different soil types, and

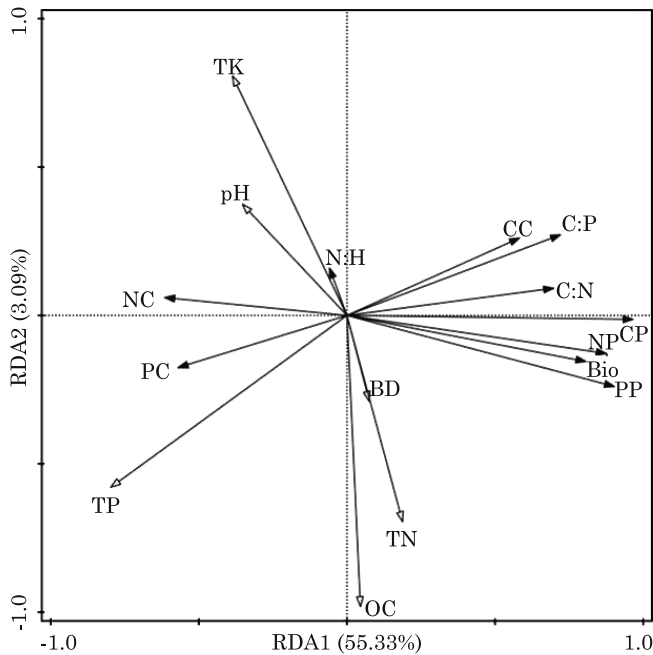


Fig. 5. Redundancy analysis (RDA) of the relationship between soil nutrient factors and biomass, nutrient index in roots of *Zanthoxylum armatum*: OC – organic carbon, BD – bulk density, TN – total nitrogen, TP – total phosphorus, TK – total potassium, Bio – biomass, CC – C content, NC – N content, PC – P content, CP – C percentage, NP – N percentage, PP – P percentage

the environmental factors such as soil nutrients were taken as explanatory variables. The results showed that R^2 was 0.514, and RDA1 axis and RDA2 axis jointly explained 55.33% of the total variance, 52.24% of the total variance was explained by RDA1 axis and 3.09% of the total variance was explained by RDA2 axis. Among the soil nutrient factors, total P ($F=24.0$, $P=0.002$) and organic carbon ($F=18.6$, $P=0.002$) significantly affected the biomass and stoichiometric ratio in roots. The results showed that total P and organic carbon were significantly positively correlated with the P content, but negatively correlated with the C content, C:N, C:P, N:P. The C, N and P proportion and biomass were positively correlated with organic carbon, but negatively correlated with total P, and the N content was positively correlated with total P, but negatively correlated with organic carbon, and total N was positively correlated with C:N and C:P.

DISCUSSION

Soil is an important part of the ecosystem, and the habitat factors provided by soil have a profound influence on the physiological and ecological characteristics, distribution and morphological structure of roots (HE et al. 2017, JIA et al. 2017). For example, *Sophora japonica* and *Gleditsia sinensis* roots showed the greatest biomass accumulation in brown limestone and developmental calcite dolomite, respectively (HU et al. 2008, ZHOU et al. 2010). However, WANG (2018) and ZHI et al. (2020) found that the roots of *Populus × canadensis Moench* and *Firmiana platanifolia* seedling achieved the best growth performance and maximum biomass accumulation in ALS, which was similar to our study. Moreover, some plants also showed a preference for certain soil types or soil attributes, which led to the distribution of some typical plants in some zonal soils. The reason may be the fact that different soil types had different physical and chemical properties and microbial community structure, and soil with good structure, moderate particle size, loose soil quality, good aeration performance, high nutrient content, and strong water and fertility retention encourages the growth and development of plant roots (DAN, BRIX 2017). Our results showed that the bulk density of ALS was the smallest, while the bulk density of YS and RS was larger (Table 1). The two latter soils have fine soil grain composition, and with stagnant water and insufficient soil oxygen supply, plant respiration is reduced while reducing substances increase, and soil can produce certain toxins to roots. ALS was graded as largely loose soil, with good air permeability, in which plant cellular respiration was more intensive, and this soil may provide enough material and energy for the roots. Thus, this plant is conducive to the growth and development of roots. The distribution characteristics of root biomass in different root orders may also directly reflect the adaptation strategies to different soil types (WANG et al. 2018). The root biomass

of *Phoebe Zhennan* S. increased gradually with the increase of root orders, and reached the maximum in the fourth-order root (LI et al. 2016). The root biomass of *Pinus kesiya* var. *Langbianensis* (A. Chev) Gaussen increased with the increase of root orders, and reached the maximum in the fifth-order root (JIA et al. 2017). The present results showed that the root biomass of *Z. armatum* followed the same trend and increased with the increase of root orders of plants cultured in four soil types. This might be due to plants adapting to different growth environments by adjusting the distribution characteristics of root biomass, helping to obtain the nutrients and water.

C, N and P are essential elements for plant growth as building blocks of plant tissues and also playing different functions in plants. The content and circulation of plant and soil nutrients play an important role in the regulation of plant growth and various physiological functions (NIKLAS et al. 2005, REICH et al. 2006). Changes of nutrient levels in tissues and organs of plants in different soil types reflect their growth characteristics and their response and adaptability to different environments. Soil organic matter is the most active component in the soil, which has considerable influence on soil water, fertilizer, etc. Higher content of organic matter and elements may improve soil hydrodynamic conditions and soil temperature, improve the activity of soil microbial community, and change the soil physical and chemical properties. In addition, during the growth process, root growth might provide a suitable place for microorganisms, and chemical substances secreted and released by the root further affect microbial activities (LADANAI et al. 2010). The present results showed that the significant differences in the biomass, C, N, and P content of roots are related with the content of organic matter, nitrogen, phosphorus and potassium of four soil types (Table 1). The content of N and P in the roots of *Larix gmelinii* (Rupr.) Kuzen. and *Fraxinus mandshurica* Rupr. decreased with the increase of root orders (JIA et al. 2010). Different root orders play different functions, and lower roots have active cortical tissue, short life span, vigorous metabolism, low stress resistance, and are more sensitive to environmental changes. However, higher roots have higher lignification degree, forming a cork layer that may block the absorption of water and nutrients (WANG et al. 2017b). Moreover, lower roots are highly sensitive to changes in different soil types, and higher roots provide more efficient nutrient transport by increasing the secondary xylem's density and longevity (JIA et al. 2021). The absorption of N and P elements requires a large amount of carbohydrate consumption, resulting in a lower C content and higher N and P content in lower roots (CHANG, GUO 2008). Our findings showed that soil types had little effect on the distribution of C, N and P elements in roots, but the C and N distribution increased in a positive proportion in subsequent root orders (Table 2). This might be due to the fact that the C content and biomass in older roots increased, while the N content decreased slightly, but the biomass increased more significantly so as to ensure the normal growth of seedlings. Moreover, there may be

a tradeoff between root acquisition and resource maintenance in terms of different soil nutrients.

Ecological stoichiometric characteristics of C, N and P may reflect the relationship between plant nutrient allocation and interaction, and such information has been widely used in the diagnosis of limiting elements and nutrient cycling (SONG et al. 2014). The C:N and C:P ratios represent the ability of plants to absorb nutrients and assimilate carbon elements. The critical value of the N:P ratio may be used as an indicator to judge the status of environmental nutrient supply to plants, and can reflect the limitation of N or P on plants (GUSEWELL 2004). Our findings showed that soil types had a significant effect on the root C:N, C:P and N:P ratios, and these results were closely related to the biomass as well as the accumulation and distribution of C, N and P. Moreover, the C:N, C:P and N:P ratios were different among different root orders (Table 2). The present results are similar to ones reported by TANG et al. (2015), which may be related to the higher N and P content in elementary roots. Moreover, earlier results showed that the thresholds of the N:P ratio in fine roots were 12 and 14 in plants growing in Daqinggou Nature Reserve, Inner Mongolia (CHEN et al. 2011). The present findings showed that the N:P ratio was less than 14 in plants grown in three soil types, indicating that the root growth was limited by nitrogen. The low N and P content might be insufficient during the growth process, and further affect the root growth and development. Thus, the mechanism by which soil types affect the N:P ratio in *Z. armatum* roots is complex and needs further study.

The correlations between the biomass and nutrient indexes of the second and third-order roots of *Z. armatum* showed a similar trend, while the correlations between the first and fourth-order roots were significantly different (Table 3). The results indicated that the second and third-order roots had the same morphological structure and function, while the first and fourth-order roots showed great differences in the morphological structure and function. There was a significant negative correlation between the N content and C:N ratio, and a significant positive correlation between the N content and N:P ratio. The P content was significantly negatively correlated with the C:P and N:P ratios, and positively correlated with the C:N ratio (Table 2). There was a significant positive correlation between the C:P and N:P ratios, indicating that the C:N:P stoichiometric ratio of root orders were significantly affected by the N:P ratio. The N and P content in each root order was significantly negatively correlated, which was different from research results by TANG et al. (2015) and LIU et al. (2020). This may have been caused by the interaction between the unique biological characteristics of *Z. armatum*s and the specific soil environment. There is a significant negative correlation between the C, N and P content in the first-order roots of *Z. armatum*s. Our study is consistent with the research results of ZENG et al. (2016). The root C content of *Larix gmelinii* at different stand ages is significantly negatively

correlated with the N and P content, indicating that the consumption of N and P nutrients promotes the C accumulation so as to ensure normal growth and development. Thus, the biomass, C, N and P content and stoichiometric ratios in different root orders of *Z. armatum*s were strongly correlated, indicating that there was a certain relationship between different nutrient elements in *Z. armatum*s. At the same time, our study also found that there was a significant positive correlation between the total N content in the soil and the N:P ratio, which was consistent with the research results of WANG et al. (2017a). The N content in the soil nutrients had a very significant positive correlation with the root N:P ratio of *Quercus mongolica*. The positive correlation between element content in plant organs and the supply capacity of the element in soil indicates that the growth of the plant is restricted by the element (WANG et al. 2016). Therefore, the root N content of *Z. armatum*s is obviously dependent on the soil N content.

CONCLUSION

The present findings showed that the soil type, root order and their interaction caused significant differences in the biomass, root C and N contents and C:N, C:P ratios of *Z. armatum*s. There was a strong correlation between the biomass and root N and P content and the stoichiometric ratio of C, N and P of root order at all levels, and there was a certain relationship between different nutrient elements in roots. Moreover, the root growth was restricted by nitrogen in AS, ALS and YS, and restricted by both N and P in RS. The current results provide information about the root biomass and the stoichiometric characteristics of C, N and P of *Z. armatum*s seedlings in four soil types, which create a scientific basis for the study of physiological and ecological adaptation strategies and fertilizer management of *Z. armatum*s seedlings under different soil conditions. Further research will aim at the physiological and ecological response characteristics of *Z. armatum*s seedlings in different soil types, so as to systematically reveal the ecological adaptation strategies of *Z. armatum*s seedlings to different soil conditions.

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