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ARTICLE



The Effects of Fertilizers on Rabbiteye Blueberry (Vaccinium ashei Reade.) Root Distribution and Fruit Yield

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ABSTRACT

The root system plays an important role in the growth and development of blueberry. The aim of this study was to assess the impacts of different fertilizers on the root growth and root-yield relationship of blueberry to provide insight into the regulation of root growth and fruit yield by fertilizing from the perspective of the root system. Rabbiteye blueberry variety 'Britewell' as the test material, and six fertilizers, including BF, OR, CF, SF, HF, and RT were used in single-factor fertilization experiments to analyze the effects of different fertilizer treatments on the root morphology, root distribution, and fruit yield of blueberry. Fertilization overall increased the root length density and root surface area in most soil layers, and the RT treatment significantly increased the total root length density and total root surface area 98.6% and 98.5%, respectively, compared with a control lacking fertilizer. In addition, the effect of fertilization on the blueberry root system was mainly observed in the 0–20 cm layer. Fruit yield was positively correlated with total root length density and total root surface area, and negatively correlated with average root diameter. In summary, the SF and RT treatments increased the morphological indexes of the root system, particularly in the shallow soil layers, leading to an increase in blueberry fruit yield.

KEYWORDS

Fertilization; membership function; root morphology; rabbiteye blueberry

1 Introduction

The root system is the primary organ for nutrient absorption in plants that synthesizes and transports physiological activators [1] and also regulates plant growth and development [2,3]. Roots are therefore of great importance for crop growth, and their growth affects shoot growth and crop yield [4,5] as they act as "receptors" for the perception of environmental changes in crop plants. Root growth is sensitive to changes in the soil nutrient environment. For instance, ammonium nutrition inhibits root growth and nitrogen (N) absorption on citrus seedlings [6], while *Arabidopsis thaliana* modulates root N acquisition efficiency in response to the N demands of shoots [5]. One study suggests that fertigation with 50% of the recommended fertilizer could be most effective for enhancing the growth and N use efficiency (NUE) of rabbiteye blueberry [7]. The appropriate amount of P fertilizer application is known as key nutrients for tomato production, and maintaining its adequate levels leads to maximizing tomato yield [8]. By



increasing the amount of P fertilizer, the tomato plant growth was also improved [9]. In addition, N deficiency is more likely to promote water absorption and N accumulation at the same root surface area level, which has been shown to lead to a higher dry mass in maize seedlings [10]. Therefore, fertilization plays an important role in the communication between plant roots, plant growth, and fruit yield. In recent years, most of the studies have focused on the factors affecting root growth and distribution [11,12]. The root–shoot system is affected by irrigation, fertilization, and straw management [13–15], among which fertilization has been regarded as the key factor in root system development.

Rabbiteye blueberries prefer ammonium nitrogen (NH_4^+ -N) over nitrate nitrogen (NO_3^- -N) [16]. N is a key factor in blueberry production, and while high blueberry yields can be acquired via the application of optimal fertilization rates [17], the type of fertilizer that achieves the greatest blueberry fruit yield remains unknown. Insufficient N fertilizer application causes premature senescence, while excessive application results in late ripening and increases environmental pollution. The improper application of phosphorous (P) and potassium (K) fertilizers affect the normal growth of plants [18]. Therefore, selecting the correct fertilizer is of great importance for promoting growth and increasing fruit yield of blueberry plants. Recent studies have measured the impacts of compound fertilizer on blueberry [19–21]. Marty et al. [22] showed that applying chipped ramial wood (CRW) compost to low bush blueberry crops may not only be a mean of increasing growth and fruit production, but also alleviate the need for chemical weed control. In berry crops, N status strongly affects orchard longevity, berry quality and productivity, and root and shoot growth rate [23]. At present, compound fertilizer (N, P, and K) is applied widely. Recent studies suggested that N, P, and K can affect the distribution of roots in the soil in cotton [24] and tomato [25]. The optimal fertilizer application rate might increase root distribution in the layer in which the fertilizer is applied, thus promoting nutrient absorption and increasing the photosynthetic capacity of plants. Phosphate fertilizer was found to induce deeper root growth into the soil within the 0-24 cm layer in maize resulting in a significant increase in the N absorption and yield of the plants. Root growth regulates above ground plant growth, and the impact of fertilization on the vertical distribution of the roots is an important factor influencing the photosynthetic ability of the leaves [13].

The above observations indicated that shoot growth and yield formation are significantly influenced by root growth. Rabbiteye blueberry is an economically important acid-loving plant with a shallow root system [26] and is thus sensitive to fertilizer selection. Plants are primarily dependent on the spatial and temporal distribution of roots for fertilizer absorption [27]. Insufficient or excessive fertilization can affect both blueberry yield and quality [28]. Therefore, it is an urgent need to identify suitable fertilizer regimes for blueberry varieties in Guizhou. As of 2017, Guizhou's blueberry cultivation area was 13,000 ha, and its yield reached 30,000 t, which ranked first in China. At present, most studies were focused on root growth and structure of blueberry [29,30]. However, no report has assessed the spatial distribution of the root system of blueberry. Therefore, the present study was performed to evaluate the effects of different fertilizers on the root distribution characteristics and yield of seven-year-old rabbiteye blueberry 'Britewell' plants in the field. Our findings shall provide insight into a suitable fertilization regime for cultivating a productive root–shoot system in blueberry.

2 Materials and Methods

2.1 Experimental Design

The research site was located in Wuyang Maritt Blueberry Base, Xuanwei Town, Majiang County, Guizhou Province, China (26° 37′–26° 49′ N and 107° 66′–107° 74′ E). The site was located in a subtropical monsoon humid zone with a warm and humid climate throughout the year. The mean annual temperature, rainfall, sunshine hours, and frost-free period were 15°C, 1250 mm, 1200 h, and 310 days, respectively. The thickness of the soil layer exceeds 100 cm. The base soil was acid yellow soil, and the soil pH was 4.5–5.5, which is suitable for the cultivation of rabbiteye blueberry in Guizhou [31]. We

were promised the right to use the plant collection as the cooperation base of the Forestry College of Guizhou University.

In this experiment, 7-year-old rabbiteye 'Britewell' well-grown blueberries were selected as the research objects. The plant row spacing, ground diameter, and tree height were about $1.5 \text{ m} \times 2 \text{ m}$, 18.4-44 mm, and 1.3-1.6 m, respectively. Another field management measures were daily blueberry management. Six types of fertilizers named as blueberry special fertilizer (SF), Haohuahong organic fertilizer (OR), fulvic acid chelated compound fertilizer (HF), soybean meal biological bacterial fertilizer (BF), potassium sulfate compound fertilizer (CF), and Runtian cattle microbial compound fertilizer (RT) were used. The various fertilizer components and related information are shown in Table 1; they were purchased from the market.

Fertilizer name	Nutritional composition and content	Amount of fertilizer application (kg·plant ⁻¹)	Manufacturer		
Special fertilizer for blueberries (SF)	Effective active bacteria count \geq 20 million pieces/g, N + P ₂ O ₅ + K ₂ O \geq 15%, Organic matter \geq 40%, Amino acid \geq 10%, Trace elements \geq 8%, sulfur \geq 10%, Humic acid \geq 10%.	1.0	Beijing Oberton Fertilizer Import and Export Co., Ltd. (Beijing, China)		
Good bonus organic fertilizer (OR)	No effective live bacteria added, $N + P_2O_5 + K_2O \ge 8\%$, Organic matter $\ge 40\%$.	1.5	Guizhou Jiafeng Organic Compound Fertilizer Co., Ltd. (China)		
Fulvic acid chelated compound fertilizer (HF)	No effective live bacteria added, $N + P_2O_5 + K_2O \ge 30\%$, Organic matter $\ge 45\%$, Fulvic Acid \ge 10%.	1.0	Shandong Jiayou Biological Fertilizer Co., Ltd. (Liaocheng, China)		
Soybean meal biological fertilizer (BF)	Effective active bacteria \geq 500 million pieces/g, N + P ₂ O ₅ + K ₂ O \geq 8%, Organic matter \geq 60%, protein \geq 22%, Amino acid \geq 18%, Trace elements \geq 8%, Humic acid \geq 10%.	1.5	Shandong Linyi Hengfeng Fertilizer Co., Ltd. (Linyi, China)		
Potassium sulfate compound fertilizer (CF)	No effective live bacteria added, total nutrient content $\ge 45\%$, including 50% K ₂ O.	1.5	Shandong Risheng Zhongwang Biological Technology Fertilizer Co., Ltd. (Linyi, China)		
Runtian cattle microbial compound fertilizer (RT)	Effective active bacteria count \ge 20 million/g, N + P ₂ O ₅ + K ₂ O \ge 5%, and Organic matter \ge 35%, humic acid \ge 10%.	1.5	Shanghai Runtian Biotechnology Co., Ltd. (Shanghai, China)		
No fertilizer (CK)	0	0	0		

Table 1: Information on the fertilizer composition and application amounts

This experiment included six single-factor treatments, with no fertilizer as the control (CK). A random block design was used. There were 3 replicates for each fertilization treatment, 3 plants for each repetition, and a total of 63 plants, including 9 plants without fertilization (CK). The fertilizer application was consistent with the daily cultivation management practices of the region. The dry ditching fertilization method was used, whereby a ditch of 20 cm width and 20 cm depth was dug around the periphery of the canopy projection; it was covered with soil following fertilization. The fertilizer was applied once every December from 2015 to 2017, with the amount of fertilizer application following the optimal fertilizer quantity recommended by the fertilizer manufacturer. All samples were collected in 2018.

2.2 Sampling

Three bushes demonstrating typical growth were selected from each treatment to sample the root distribution. Sampling was initiated vertically at four diagonal vertices at 20, 40, and 60 cm away from the main stem of the root in a horizontal direction (Fig. 1a). A soil auger of 7 cm core diameter was used. There was no interval between each soil layer. Each point was sampled in layers of 10 cm depth until a depth of 40 cm was reached. For each plant, 12 tubes were sampled from each layer, and 48 tubes were sampled in total from the four layers (Fig. 1b). The roots and soil were placed into plastic bags and transported back to the laboratory for refrigeration after labeling. After sifting most of the soil with a 10-mesh screen in the laboratory, the remaining soil adhering to the root system was rinsed off with running water. The blueberry roots were distinguished from other roots based on characteristics such as shape, color, and elasticity. With the aid of tweezers, the live roots were rinsed in a small plastic basin filled with distilled water and then air-dried. Please clarify how did you separate live from dead roots.



Figure 1: (a) Horizontal sampling legend. Note: **•** Represents the root sampling point. (b) Vertical sampling legend. **•** Represents the sampling point of the soil auger. Note: Each layer of soil was 10 cm in depth, with a total of 40 cm depth

For the determination of the root morphology index, the washed blueberry roots were placed into a clean scan tray and spread out as much as possible. Roots were then scanned with a Microtek ScanMaker i800 Plus equipment (MICROTEK), using a ScanWizard EZ software. Thereafter, the Wanshen LA Root Analysis System was used to analyze root length, root surface area, root volume, average root diameter, and branch number. Root biomass (mg) was obtained using the drying method, whereby roots were dried at 65°C until constant weight (about 48 h). Afterwards, their dry weight was determined. The root parameter calculation formulae were as follows:

Bio mass density
$$(mg/cm^3) = \frac{\text{total root biomass } (mg)}{\text{drill barrel volume } (cm^3) \times \text{barrel number } (No.)}$$
 (1)

Root surface area
$$(cm^2) = \frac{root \ surface \ area \ (cm^2)}{drill \ barrel \ volume \ (cm^3) \ \times drill \ number \ (No.)}$$
 (2)

2

Branching number (No./cm) =
$$\frac{\text{branch number (No.)}}{\text{root length (cm)}}$$
 (3)

Root volume
$$(mm^3/cm^3) = \frac{\text{root sample volume } (mm^3)}{\text{drill tube volume } (cm^3) \div \text{tube number } (No.)}$$
 (4)

Average root diameter (mm) =
$$\frac{\sum V1 + V2 + V3 + \dots Vn}{n (V \text{ for root diameter})}$$
 (5)

Root length density
$$(cm/cm^3) = \frac{\text{root length (cm)}}{\text{drill tube volume (cm^3)} \times \text{tube number (No.)}}$$
 (6)

Specific root length
$$(cm/g) = \frac{\text{root length } (cm)}{\text{root mass } (g)}$$
 (7)

2.3 Statistical Analysis

A one-way analysis of variance (ANOVA) was conducted using SPSS version 19.0 (IBM Corp., Armonk, USA). Post-hoc tests were conducted using Duncan's test, and differences between treatments were considered significant at P < 0.05. Pearson's correlation coefficients were calculated using the correlation procedure in SPSS.

The membership function evaluation method used here was based on the principle of fuzzy mathematics. The membership function is a fuzzy control system that converts clear quantities into fuzzy quantities, and performs fuzzy logic operations. Membership function analysis comprehensively evaluates plant characteristics based on multiple indexes. The membership function calculation formulae were as follows:

Membership value =
$$\frac{X - Xmin}{Xmax - Xmin}$$
 (8)

Anti – membership value =
$$1 - \frac{X - Xmin}{Xmax - Xmin}$$
 (9)

In the formulae, X is the measured value, Xmax is the maximum value, and Xmin is the minimum value. The membership function values are cumulative, and average values were obtained. Treatment efficacy increases with membership value [32].

3 Results

3.1 Impact of Fertilizer on Root Length Density in the Different Soil Layers

The blueberry root system was distributed primarily in the 0–20 cm soil layer. The six fertilizer applications significantly promoted root length density at all soil depths (0–40 cm) compared with CK (Table 2). The application of RT fertilizer treatment resulted in the highest root length density in most soil layers (0–10, 10–20, and 30–40 cm). At soil depths of 0–10, 10–20, and 30–40 cm, the RT fertilizer treatment increased the root length density by 1843%, 1508%, and 848.00%, respectively, compared with controls. At a depth of 20–30 cm, the OR fertilizer treatment increased the root length density by 448.76% compared to CK.

Table 2: Effect of different fertilizers on	the root distribution parameters	s in the different soil layers
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Root Morphology index	Soil Depth	Different fertilizer types						
	(cm)	CK	SF	OR	HF	BF	CF	RT
Root length density (mm/cm ³)	10	0.10g	0.48f	0.59e	0.66d	0.73c	0.92b	1.94a
	20	0.37f	0.84d	1.44b	1.38c	1.41bc	0.70e	1.88a
	30	0.24f	0.70d	1.33a	0.48e	0.86c	0.45e	1.22b
	40	0.17f	0.47c	0.27e	0.37d	0.52b	0.20f	1.02a
Root surface area (mm ² /cm ³)	10	0.57e	1.90d	2.98b	2.50c	2.00d	2.55c	4.99a
	20	1.05f	6.61a	5.16c	4.34d	5.32c	2.79c	6.11b
	30	0.53f	6.58a	5.93b	2.37d	2.48d	1.86e	5.18c
	40	1.15d	2.53a	0.57f	1.12d	1.60c	0.95e	2.41b
Root system volume (mm ³ /cm ³)	10	2.97a	1.05e	2.41b	1.44d	0.65g	0.89f	1.77c
	20	4.85b	11.23a	3.51cd	1.63e	3.77c	1.66e	3.30d
	30	1.71de	14.10a	6.89b	1.94d	1.00f	1.41ef	6.24c
	40	1.41b	3.98a	0.14f	0.38e	0.69cd	0.80c	0.66d
Root biomass density (mg/cm ³)	10	1.11b	0.74d	1.23a	1.02c	0.38e	0.82d	0.79d
	20	1.91b	4.47a	1.62d	1.70c	1.91b	1.09f	1.19e
	30	0.65f	5.73a	2.93b	1.30d	0.58g	0.87e	2.89c
	40	0.68b	1.63a	0.08g	0.25f	0.38d	0.48c	0.27e
Average root diameter (mm)	10	3.51a	1.06bc	1.09b	1.24b	0.89cd	0.87cd	0.73d
	20	1.57b	2.31a	0.95d	0.93d	1.00d	1.09c	1.08c
	30	0.95e	2.68a	1.49b	1.08cd	0.98de	1.11c	1.18c
	40	2.21a	1.19b	0.65d	0.76d	0.99c	1.03c	0.69d
Number of root branching	10	0.75c	0.50e	1.00b	0.44e	1.01b	0.64d	1.20a
(No./cm)	20	0.88bc	0.53d	1.08a	0.37e	0.96b	0.38e	0.84c
	30	0.65c	0.42d	0.88b	0.33e	0.94a	0.32e	0.62c
	40	0.41d	0.37d	0.74a	0.27e	0.65b	0.48c	0.60b

Note: Each soil layer and index was analyzed for each fertilizer treatment. Different letters in a row indicate significant differences at P < 0.05. Control = CK, Blueberry special fertilizer = SF, Haohuahong organic fertilizer = OR, fulvic acid chelated compound fertilizer = HF, soybean meal biological bacterial fertilizer = BF, potassium sulfate compound fertilizer = CF, and Runtian cattle microbial compound fertilizer = RT. The six fertilizer applications significantly promoted the root surface area at most soil depths (0-30 cm) compared to the controls (Table 2). The application of RT fertilizer treatment resulted in the greatest root surface area in the 0–10 cm soil layer. At a soil depth of 0–10 cm, the RT fertilizer treatment increased the root surface area by 774.83% compared with no fertilizer application. At depths of 10–20, 20–30, and 30–40 cm, the SF fertilizer treatment increased the root surface area by 531.36%, 1145.50%, and 120.44%, respectively, compared with CK.

3.3 Impact of Fertilizer on the Root Volume in the Different Soil Layers

The six fertilizer applications significantly decreased the root volume at 0–10 cm soil depth compared with the control (Table 2), with the greatest root volume detected under no fertilizer application. Conversely, at soil depths of 10–20, 20–30, and 30–40 cm, the SF fertilizer treatment increased the root volume by 131.40%, 722.78%, and 181.19%, respectively, compared with CK.

3.4 Impact of Different Fertilizers on the Root Biomass Density in the Different Soil Layers

At a depth of 0–10 cm, OR increased the root biomass density the most, being 10.31% higher than CK and 221.19% higher than BF; the latter obtained the lowest root biomass. At 10–20 cm depth, SF was the most important contributor to root biomass density, and CF showed the lowest biomass at this depth, with SF being 133.56% higher than CK and 308.37% higher than CF. At 20–30 cm depth, SF had the greatest contribution to root biomass density, while BF had the lowest contribution, with SF being 783.85% higher than CK and 890.75% higher than BF. At 30–40 cm depth, the most important contributor to root biomass density was SF, while OR had the lowest contribution. SF was 58.49% higher than CK and 2073.95% higher than OR (Table 2).

3.5 Impact of Fertilizer on the Average Root Diameter in Different Soil Layers

The six fertilizer applications significantly decreased the average diameter of the roots at 0–10 and 30–40 cm soil depths compared to CK (Table 2). At 10–20 and 20–30 cm soil depth, SF had the greatest effect on root diameter, being 47.01% and 180.99% higher than CK, respectively.

3.6 Impact of Fertilizer on Root Branching in the Different Soil Layers

At 0–10 cm depth, RT promoted root branching number the most, being 60.14% higher than in CK and 171.89% higher than in HF. At the depths of 10–20 and 30–40 cm, OR resulted in the greatest increase in root branching number. At 0–40 cm soil depth, HF had the least effect on the number of root branching.

3.7 Impact of Fertilizer on Average Values of Root Distribution Parameters

The total root length density and total root surface area were significantly increased under the six fertilization treatments (Fig. 2). The values from the RT fertilization treatment were the highest among the different treatments and increased the total root length density and total root surface area by 98.6% and 98.5%, respectively, compared with CK. The maximum total root volume and total root biomass values were achieved under SF, being 74.5% and 87.8% higher, respectively, than in CF, which achieved the lowest values. Root biomass was similar between BF and CF. The maximum total root bifurcation number was achieved under OR and BF, while HF showed the lowest. Total root bifurcation number was not significant difference in SF and CF.

3.8 Impact of Fertilizer on Fruit Yield

Fertilization significantly increased fruit yield. The RT treatment had the highest fruit yield. It did not differ significantly from SF, HF, and BF, with an increase of 99.6% compared with CK (Fig. 3).



Figure 2: Bar graphs showing the impact of different fertilizers on total root length density, total root surface area, total root volume, total root biomass, average root diameter, and total number of root branches within the 0–40 cm soil depth. Different letters among treatments indicate significant differences at P < 0.05. Error bars represent the standard deviation. Total: It is the sum of individual values for each depth. Abbreviations for treatments are as described in the material and methods section



Figure 3: Bar chart showing the effect of different fertilizers on fruit yield. Different letters among treatments indicate significant differences at P < 0.05. Error bars represent the standard deviation. Abbreviations for treatments are as described in the material and methods section

3.9 Membership Function Value and Ranking

With the exception of CF and HF, the average values of all treatments were higher than in CK, which indicated that fertilization overall promoted root growth and fruit yield (Fig. 3). Among the treatments, SF had the best effect on promoting root growth and fruit yield, followed by RT. The root systems were found to be largest under the SF treatment (Table 3).

Table 3: Rank of total root morphological index and subordinate function value of fruit yield determined by a membership function evaluation method. Abbreviations for treatments are as described in the material and methods section

	TRL	TRS	TRV	TRS	TAD	TBI	FY	Mean value	Rank
СК	0.00	0.00	0.24	0.12	0.96	0.56	0.00	0.27	6
SF	0.31	0.92	0.99	1.00	0.75	0.21	0.95	0.73	1
OR	0.52	0.73	0.32	0.28	0.13	0.97	0.53	0.50	3
HF	0.38	0.45	0.03	0.11	0.09	0.04	0.81	0.27	5
BF	0.51	0.52	0.05	0.00	0.06	0.92	0.80	0.41	4
CF	0.27	0.31	0.00	0.00	0.11	0.21	0.57	0.21	7
RT	0.99	0.99	0.28	0.20	0.03	0.79	1.00	0.61	2

Note: FY: fruit yield, TRL: total root length density, TRS: total root surface area, TRV: total root volume, TRB: total root biomass, TAD: mean diameter of the total root system, TBI: Total branching number.

3.10 Correlation between Total Root Morphology Index and Fruit Yield

Fruit yield was positively correlated with total root length density and total root surface area, but negatively correlated with average root diameter. Total root length density was positively correlated with total root surface area and total root bifurcation number, and negatively correlated with the average root diameter. Total root surface area was significantly positively correlated with total root biomass density and total root volume. Total root volume was positively correlated with total root biomass density and average root diameter. And total root biomass was significantly positively correlated with mean diameter of the total root system. These results indicate that the root morphological *index* (total root length density, total root surface area and mean diameter of the total root system) influenced each other and significantly affected fruit yield (Table 4).

Table 4:	Correlation	coefficients	between t	total root	morphological	indexes a	na fruit yield	l

	FY	TRL	TRS	TRV	TRB	TAD	TBI
FY	1						
TRL	0.707**	1					
TRS	0.836**	0.776**	1				
TRV	0.265	-0.042	0.545*	1			
TRB	0.357	-0.048	0.570**	0.981**	1		
TAD	-0.532*	-0.689**	-0.339	0.570**	0.493*	1	
TBI	-0.096	0.460*	0.213	-0.113	-0.245	-0.233	1

Note: FY: fruit yield, TRL: total root length density, TRS: total root surface area, TRV: total root volume, TRB: total root biomass, TAD: mean diameter of the total root system, TBI: total branching number. ** Highly significant positive correlation (P < 0.01), * significant positive correlation (P < 0.05).

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4 Discussion

Some researchers believe that the content of fertilizers affects root morphology index. For example, Chen et al. found that the richness of the bacterial community was reduced under both low- and highnitrogen fertilizers compared to the control treatment, and increased in high-N fertilizers plus P or K treatments [33]. Under laboratory conditions, both stimulating and reducing effects of P and K deficiencies on root traits have been reported [34–36]. Therefore, the components of compound fertilizers play a key role in plant growth. This study revealed how different fertilizers affected blueberry root morphology and distribution, as well as how the root system impacted blueberry fruit yield. The root morphology indexes (surface area, volume, biomass density), and average root diameter in most soil layers under the SF treatment were significantly higher than in the control treatment. The root length density in most soil layers was the highest under the RT treatment, and it was also significantly higher than in CK. Fruit yield was positively correlated with total root length density and total root surface area, but negatively correlated with average root diameter. This is consistent with the results of Wang et al. on the morphological development and yield of the peanut root system [37].

Root morphology is the foundation of nutrient and water uptake by plants [38,39]. Root length density and surface area are appropriate indexes for describing a root system [40]. In this study, we found that root length density and surface area generally increased with fertilization and were the highest in the 10–20 cm layer, which is consistent with the findings of Bryla et al. [41] in northern highbush blueberry. The application of fertilizers in this study significantly increased root elongation and resulted in the greatest root length density, surface area, and volume in most soil layers. This root length density finding corroborates the results of Liu et al. [42], who used the Minirhizotron technique to identify root morphological traits in rice under different fertilizer treatments throughout an entire growth period. Similarly, Zhang et al. [24] showed that fertilization increased the root length density of cotton under a drip irrigation system; we observed that the highest root length density, root surface, and fruit yield were obtained in the fertilization treatments. Fertilization deficiency has been shown to lead to low root activity and water consumption [24] as well as a reduction in the production of reactive oxygen species (ROS) in the roots [15], resulting in low root biomass accumulation.

We found that the root system was primarily distributed in the shallow soil layer, with the highest values for root length density, surface area, and biomass density observed in the 0–20 cm layer, accounting for approximately 50% of the corresponding totals. This is in agreement with the results of Min et al. [43]. In this study, relative to the control, the root length density in the OR treatment was lower in the surface and deeper studied soils, but significantly higher in the 10–30 cm soil layer. These results indicate that the fertilization treatment increased the root length distribution in the middle-deep soil layers, which can be expected to promote photosynthesis and water potential in the leaves [44]. In contrast, root biomass density was increased in the surface soil layers but decreased in the deeper ones.

Reports on these areas showed that the growth medium had an effect on plant height, crown diameter, and surface area and biomass densities [45,46]. Total surface area, volume, and biomass density were affected by fertilization, with the greatest values observed in the SF treatment. Blueberry fruit yield was strongly associated with total root length and surface area, illustrating that root morphology significantly affected blueberry fruit yield. Our research results are similar to previous ones. The root morphology indexes were overall highest in the SF and RT treatments, and lowest in the OR and CF treatments. As the biological fertilizer contains microbes, the HF, OR, and CF fertilizers, which are not bio-fertilizers, did not add effective live bacteria. Because of this, they might have had little effects on blueberry root morphology; this is consistent with Yuan et al.'s conclusion that bio-organic fertilizers can promote plant growth [47]. The analysis of the fertilizer components in Table 1 shows that the contents of N, P, K, and organic matter in RT were the lowest among all fertilizers, and the effect of organic fertilizers (RT, OR, BF, SF, HF) was better than that of the inorganic fertilizer (CF) (Table 2). This is not surprising given

that blueberry is an oligotrophic plant. In comparison with other species, rabbiteye blueberry is particularly sensitive to chemical fertilizers [48]. The contents of N, P, calcium, manganese, and K in the leaves of *Vaccinium uliginosum* were previously found to be lower than those of other small berry tree species [49]. In summary, our research results are in line with previous thoughts that fertilizers increase plant growth, biomass, root morphology index and yield [50].

5 Conclusions

The different fertilizers studied significantly affected the root morphology and root distribution of blueberries in the field. The RT treatment improved root growth in each soil layer, and increased the total root length, surface area, volume, fruid yield and biomass. Root morphology and biomass were the most strongly affected by the application of SF and RT in the upper soil layers. These findings indicate that SF and RT can promote root growth, especially for shallower roots (0–20 cm), thereby increasing root biomass and promoting fruit yield.

Conceptualization: Xiaolan Guo: Writing-original draft preparation and Methodology, Chenyan Liu: Formal analysis and investigation, Muhammad Shakeel, Writing-review and editing. Delu Wang: Supervision.

Permission for Plant Material Collection: We have obtained permission from Wuyang Maritt Blueberry Base institute for samples collection. The study was carried out with relevant institutional, national, and international guidelines and legislation.

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