RESEARCH ARTICLE

Effect of slow-release fertilizer application rates on the growth of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings

Manyi Fu[1, 2], Qingbin Zhang[1], Song Geng[3, *]

[1]College of Forestry, Xinyang Agricultural and Forestry University, Xinyang, Henan, China. [2]Xinyang Engineering Technology Centre for Woody Oilseed Crop Cultivation and Development, Xinyang, Henan, China. [3]College of Tianhua, Shanghai Normal University, Shanghai, China.

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***Zanthoxylum armatum* 'Jiuyeqing', an elite cultivar of *Zanthoxylum armatum* DC., has been rapidly adopted in southwestern China due to its high yield, superior quality, drought tolerance, and adaptability to barren soils. However, empirical fertilization practices during seedling cultivation often result in inconsistent fertilizer efficacy and heterogeneous seedling quality, limiting the green and sustainable development of the *Z. armatum* 'Jiuyeqing' industry. This study investigated the effects of different slow-release fertilizer (SRF) application rates on the growth and photosynthetic characteristics of container-grown *Z. armatum* 'Jiuyeqing' seedlings and identified the optimal application level to support precise fertilization practices. A one-way randomized group design was employed with five SRF treatments of 1, 2, 3, 6, and 9 kg/m^3. Growth indices, physiological parameters, and photosynthetic response metrics were measured under each treatment. Seedling quality across treatments was comprehensively evaluated using the affiliation function value method. The results demonstrated that SRF application rates significantly affected the morphological growth, physiological traits, and photosynthetic responses of *Z. armatum* 'Jiuyeqing' container seedlings. Affiliation function analysis indicated that seedlings treated with 6 kg/m^3 fertilizer exhibited the best overall growth performance, achieving an average affiliation function value of 0.85 and ranking the highest among all treatments. Therefore, an application rate of 6 kg/m^3 SRF was recommended for the cultivation of *Z. armatum* 'Jiuyeqing' container seedlings. This study provided a scientific reference for SRF application, offering technical support for optimizing cultivation techniques, improving cost-effective production efficiency, and promoting standardized propagation practices for *Z. armatum* 'Jiuyeqing' container seedlings.**

Keywords: *Zanthoxylum armatum* 'Jiuyeqing'; slow-release fertilizer; container seedling; photosynthetic physiology; growth indexes.

[*]**Corresponding author:** Song Geng, College of Tianhua, Shanghai Normal University, Shanghai 201815, China. Email: gengsong0303@outlook.com.

Introduction

Zanthoxylum armatum 'Jiuyeqing', a variety of *Zanthoxylum armatum* DC. belonging to the genus *Zanthoxylum* L. of the family Rutaceae, is characterized by drought and barren soil resistance, ease of reproduction, rapid growth, and early fruiting. Its fruit is aromatic and possesses a pure, numbing flavor that is highly favored by consumers, resulting in strong market demand and considerable economic value [1]. Container seedling cultivation, a relatively mature propagation technology, has recently gained global attention [2]. This method involves replacing natural soil with specific substrates in containers, providing an ideal growth environment for seedlings and significantly improving their survival rate after transplantation [3]. Advantages of container seedling cultivation include vigorous root development, superior seedling quality, high transplant survival rates, and enhanced stress resistance. In recent years, as *Z. armatum* ‘Jiuyeqing’ has been extensively developed in southern China, the survival rate of seedlings has emerged as a key constraint on industry growth. Therefore, studying container seedling cultivation for this variety is of great importance for addressing challenges in the industry's current development.

Slow-release fertilizer (SRF) is designed to release nutrients gradually according to a controlled release pattern through various regulatory mechanisms, effectively extending the period of nutrient absorption and utilization by plants [4, 5]. The scientific application of SRF plays a critical role in achieving precise fertilization during container seedling cultivation and represents an important direction for advancing seedling production technology. Chen *et al*. demonstrated that SRF application significantly improved the quality of *Pinus massoniana* Lamb. Seedlings [6]. Similarly, Pang *et al*. found that the height, diameter, and biomass of *Aquilaria sinensis* (Lour.) Spreng. container seedlings peaked at an SRF rate of 2.50 kg/m^3, which was significantly higher than other treatments [7]. Pan *et al*. reported that an appropriate SRF application promoted growth, biomass accumulation, and root development in *Carya illinoinensis* (Wangenh.) K. Koch container seedlings, achieving the best seedling quality at an SRF rate of 3.0 kg/m^3 [8]. Additionally, Araújo *et al*. concluded that *Trachycarpus fortunei* (Hook.) H. Wendl. exhibited a high demand for SRF during the seedling stage with optimal seedling quality observed at a fertilizer application rate of 8.0 kg/m^3 [9]. Given these findings, it is of significant theoretical and practical importance to investigate the growth response of *Z. armatum* ‘Jiuyeqing’ container seedlings under varying SRF application rates to achieve high-quality, precise, and efficient seedling cultivation.

Currently, research on *Z. armatum* ‘Jiuyeqing’ primarily focuses on cultivation techniques with limited studies addressing SRF application during container seedling production. Moreover, challenges such as insufficient early-stage nutrient accumulation in container seedlings can negatively impact growth and result in slow post-transplant development. SRF, by providing a long-term nutrient supply, strengthens the seedling nutrient pool, promotes robust growth, and enhances afforestation outcomes. Based on these considerations, this study took *Z. armatum* ‘Jiuyeqing’ container seedlings as the research object and analyzed growth indices, physiological parameters, and photosynthetic responses under different SRF application rates. By evaluating the seedlings' responses to varying SRF levels, the study identified the optimal fertilization rate, thereby provided a scientific reference for the high-quality cultivation of *Z. armatum* ‘Jiuyeqing’ container seedlings.

Materials and methods

Plant materials and growth conditions

The research was conducted at the experimental base of Xinyang Agricultural and Forestry University, Xinyang, Henan, China with an average annual temperature of 15.1 – 15.8°C, 1,900 – 2,000 hours of sunshine, 900 – 1,400 mm of precipitation, and an average annual relative humidity of 77%. This area belongs to the transitional zone between subtropical and warm temperate climates. The *Zanthoxylum armatum* ‘Jiuyeqing’ seedlings were obtained from the Chengxin Nursery Base (Chongqing, China). Planting was conducted using white non-woven seedling bags (Hongda, Tangshan, Hebei, China) measuring 20 cm in diameter and 20 cm in height. The SRF was Asariya 405 resin-coated full-control SRF and obtained from Haifa Group (Haifa, Israel), which contains 14% nitrogen, 14% phosphorus pentoxide, 14% potassium oxide, and 2.5% magnesium oxide. This balancedrelease fertilizer with an effective duration of 8 – 9 months was directly and evenly mixed into the nursery substrate.

Experimental design and methods

The experiment commenced on April 28, 2024. Seedlings of uniform height were selected for transplanting with one seedling planted per non-woven bag at consistent planting depths. The cultivation substrate was prepared by mixing peat soil, vermiculite, and perlite at a 1:1:1 ratio. The SRF application trial followed a one-way randomized block design with five fertilizer gradients of 1 kg/m^3 (T1), 2 kg/m^3 (T2), 3 kg/m^3 (T3), 6 kg/m^3 (T4), and 9 kg/m^3 (T5) along with a control group (CK) receiving no SRF (0 kg/m^3). Each treatment was replicated three times with nine seedlings per replicate. After transplanting, the containerized seedlings were placed on a seedbed under a shade net and thoroughly watered. Subsequent irrigation was performed regularly based on weather conditions and substrate moisture levels. Weeds were promptly removed to prevent competition for nutrients [10].

Measurement of growth and physiological indicators

Seedling height and ground diameter were systematically measured on the 28th of each month from April to September 2024. Plant height was recorded by measuring the distance from the ground surface to the apex of the main stem using a measuring tape, and the average of three readings was used. Ground diameter was measured 5 cm above the soil line with vernier calipers (accuracy: 0.01 mm), also averaging three readings per plant. Due to the non-significant effect of SRF application on simple and compound leaf counts during the early growth stages, leaf numbers were recorded monthly from June to September. On September 28, 2024, the conclusion of the experiment, leaf samples from each treatment group were collected for the determination of soluble sugar, soluble protein, and chlorophyll contents.

Determination of soluble sugar content

Soluble sugar content was determined using a modified anthrone colorimetric method [11]. Approximately 0.2 g of finely ground fresh leaf tissue was placed into a 10 mL stopper-sealed test tube. After adding 10 mL of distilled water, the sample was incubated in a boiling water bath for 20 minutes. The extract was then cooled under running water for 10 minutes and filtered into a 25 mL volumetric flask. A 1 mL aliquot of the supernatant was mixed with 4 mL of anthrone-sulfuric acid reagent. Absorbance was measured at 620 nm using a 722S Visible Spectrophotometer (Jinghua, Shanghai, China). Three biological replicates were analyzed for each treatment, and sugar concentrations were calculated based on a pre-established glucose standard curve.

$$\text{Soluble Sugar (\%)} = \frac{m \cdot V_T \cdot N}{m_0 \cdot V_S \cdot 10^6} \times 100$$

where m was the sugar mass derived from the standard curve (µg). V_T was the total extraction volume (mL). V_S was the sample aliquot volume used for analysis (mL). N was the dilution factor. 10^6 was the gram-to-microgram conversion constant. m_0 was the original sample mass (g).

Determination of soluble protein content

Approximately 0.2 g of finely chopped leaf tissue was homogenized in distilled water using a mortar and pestle. The homogenate was transferred into a centrifuge tube and incubated at room temperature for 1 hour before centrifugation at 4,000 rpm for 20 minutes using MC-12Pro centrifuge (Joanlab, Huzhou, Zhejiang, China). The supernatant was collected and diluted to 10 mL with distilled water in a volumetric flask to prepare the test solution. Protein concentration was determined spectrophotometrically at 595 nm using the Coomassie Brilliant Blue G-250 method (Macklin, Shanghai, China) [12]. Three biological replicates were analyzed for each treatment. Soluble protein content was quantified against a standard calibration curve and calculated as follows.

$$\text{Soluble Protein } \left(\frac{\mathrm{mg}}{\mathrm{g}}\right) = \frac{\mathrm{m} \cdot \mathrm{V_T} \cdot \mathrm{N}}{\mathrm{m_0} \cdot \mathrm{V_S} \cdot 1000}$$

where *m* was the protein quantity derived from the standard curve based on the sample absorbance (mg). V_T was the total volume of the extraction solution (mL). m_0 was the original sample mass (g). V_S was the volume of supernatant used for measurement (mL). *N* was the dilution factor. The coefficient 1,000 was used to convert micrograms to milligrams. After completing the measurements, fresh plant leaves were rinsed with distilled water, and excess water on the leaf surfaces was carefully blotted dry. The midribs were then removed, and the total chlorophyll content, chlorophyll *a* content, and chlorophyll *b* content were determined using the spectrophotometric method as follows.

Chlorophyll a content (mg/g) = $(12.7 \times A_{663} - 2.69 \times A_{645}) \times V \times F \div W \div 1000$

Chlorophyll b content (mg/g) = $(22.9 \times A_{645} - 4.68 \times A_{663}) \times V \times F \div W \div 1000$

Total chlorophyll content (mg/g) = $(20.21 \times A_{645} + 8.02 \times A_{663}) \times V \times F \div W \div 1000$

where V was the volume of extract (10 mL). F was the dilution. W was the mass of sample (g). Gas exchange parameters were measured upon the experiment's completion on September 28, 2024. A CI-340 portable photosynthesis meter (CID, Camas, Washington, USA) was used to determine the net photosynthetic rate (Pn), stomatal conductance (Gs), intercellular CO_2 concentration (Ci), transpiration rate (Tr), and related parameters.

Affiliation function analysis

The affiliation function method based on fuzzy mathematics principles was employed to calculate the affiliation value for each indicator and sum them up for comprehensive comparison. This approach avoided the bias and inaccuracy associated with single-indicator evaluations, thereby making the evaluation results more scientific and reliable as below.

Positive correlation:
$$\mathrm{U(X_j)} = (\mathrm{X_j} - \mathrm{X_{min}})/(\mathrm{X_{max}} - \mathrm{X_{min}})$$

Negative correlation:
$$\mathrm{U(X_j)} = 1 - (\mathrm{X_j} - \mathrm{X_{min}})/(\mathrm{X_{max}} - \mathrm{X_{min}})$$

where $\mathrm{X_j}$ was the value of the j indicator of the treatment. $\mathrm{X_{max}}$ was the maximum value of an indicator in all treatments. $\mathrm{X_{min}}$ was the minimum value of an indicator in all treatments.

Data processing

Microsoft Excel 2019 (Microsoft, Redmond, WA, USA) was used for data organization, while data analysis was performed using ANOVA in SPSS 26.0 (IBM, Armonk, NY, USA). Differences between treatment means were evaluated using the Least Significant Difference (LSD) test.

Results

Effects of different SRF application rates on growth indexes

The results showed that the application of SRF significantly promoted the height growth of *Zanthoxylum armatum* 'Jiuyeqing' seedlings. Differences between treatments in April and May were not obvious, likely due to initial root system damage, during which SRF application did not exert a significant effect. In June, as seedlings entered a rapid growth phase, root absorption capacity recovered, nutrient demand increased, and higher temperatures accelerated the SRF release rate. Consequently, a significant difference in seedling height between the SRF treatment groups and the CK group emerged ($P < 0.05$), although differences among the five SRF application groups remained non-significant (Table 1). As seedlings entered the peak growth period from July to September, significant differences among the five SRF treatment groups became evident ($P < 0.05$). By the end of the experiment, the seedling height increment in the CK group was 15.36 cm, while those in the T1, T2, T3, T4, and T5 groups were 60.35 cm, 78.52 cm, 90.53 cm, 88.52 cm, and 86.00 cm, respectively,

Table 1. Effect of different SRF application rates on seedling height of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings (cm).

Treatment	4/28/2024	5/28/2024	6/28/2024	7/28/2024	8/282024	9/28/2024
CK	11.21 ± 2.06^{a}	10.60 ± 2.06^{a}	11.44 ± 2.46^{b}	13.17 ± 3.06^{d}	22.01 ± 6.18^{d}	26.57 ± 7.41^{d}
T1	11.21 ± 2.06^{a}	16.18 ± 3.88^{a}	27.09 ± 4.55^{a}	42.06 ± 3.90^{c}	62.83 ± 7.00^{c}	71.56 ± 6.98^{c}
T2	11.48 ± 1.27^{a}	16.13 ± 2.62^{a}	30.44 ± 4.52^{a}	51.60 ± 6.09^{b}	75.65 ± 6.36^{b}	90.00 ± 9.14^{b}
T3	10.55 ± 2.48^{a}	14.31 ± 2.04^{a}	31.03 ± 3.99^{a}	61.38 ± 5.42^{a}	90.28 ± 12.15^{a}	101.08 ± 7.28^{a}
T4	11.31 ± 1.41^{a}	14.80 ± 1.33^{a}	29.85 ± 4.49^{a}	58.61 ± 4.07^{a}	82.58 ± 9.98^{ab}	99.83 ± 9.49^{a}
T5	12.97 ± 1.91^{a}	16.34 ± 1.85^{a}	30.50 ± 6.51^{a}	59.38 ± 8.52^{a}	84.32 ± 14.69^{ab}	98.97 ± 9.33^{ab}

Notes: Values in the table were mean ± standard deviation. Different lowercase letters for data in the same column indicated significant differences ($P < 0.05$).

representing increases of 292.30%, 411.20%, 489.39%, 476.30%, and 459.90% over the CK group. These results demonstrated that SRF application significantly promoted height growth with the T3 group performing best. However, differences among T3, T4, and T5 were not significant (Figure 1).

Figure 1. Effects of different SRF application rates on the growth (9/28/2024).

The differences in ground diameter were not significant in April. However, significant differences were observed between the treatment groups and the CK group in May ($P < 0.05$), although the differences among the five treatment groups were still not significant. From June to September, significant differences between the treatment and CK groups persisted ($P < 0.05$). By the end of the experiment, ground diameter increment in the CK group was 1.90 mm, while increments in T1, T2, T3, T4, and T5 groups were 5.90 mm, 6.88 mm, 8.36 mm, 8.13 mm, and 8.04 mm, respectively, representing the increases of 210.53%, 262.11%, 340.00%, 327.89%, and 323.16% over the CK group (Table 2). These results indicated that SRF application significantly promoted ground diameter growth with the T3 group showing the best performance. However, as with height growth, differences among the T3, T4, and T5 groups were not significant. Since the changes in single leaf count were not significant in the initial months, statistical analysis began in June. The significant differences were observed in the number of single leaves between the SRF treatment groups and the CK group starting in June ($P < 0.05$), although no significant differences were found among the SRF treatment groups. As the months progressed, a gradual increase in significant differences among the treatment groups emerged ($P < 0.05$). By the end of the experiment, the increase in single leaves in the CK group was 45.43 leaves, while the increases in the T1, T2, T3, T4, and T5 groups were 106.22, 207.75, 262.15, 336.86, and 362.72 leaves, respectively. These values were 133.81%, 357.3 0%, 477.04%,

Table 2. Effect of different SRF application rates on the ground diameter of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings (mm).

Treatment	4/28/2024	5/28/2024	6/28/2024	7/28/2024	8/28/2024	9/28/2024
CK	1.74 ± 0.24^{a}	1.80 ± 0.33^{b}	1.73 ± 0.27^{c}	1.81 ± 0.30^{e}	3.03 ± 0.85^{c}	3.64 ± 0.82^{d}
T1	1.57 ± 0.11^{a}	2.23 ± 0.22^{a}	3.58 ± 0.44^{b}	5.13 ± 0.28^{d}	6.33 ± 1.33^{b}	7.47 ± 0.77^{c}
T2	1.69 ± 0.27^{a}	2.36 ± 0.49^{a}	4.16 ± 0.55^{a}	6.45 ± 0.66^{c}	7.83 ± 0.55^{a}	8.57 ± 0.42^{b}
T3	1.57 ± 0.23^{a}	2.26 ± 0.25^{a}	4.27 ± 0.61^{a}	7.25 ± 0.63^{bc}	8.79 ± 0.85^{a}	9.93 ± 0.69^{a}
T4	1.78 ± 0.24^{a}	2.23 ± 0.32^{a}	4.45 ± 0.69^{a}	8.01 ± 0.74^{a}	8.75 ± 0.64^{a}	9.91 ± 0.47^{a}
T5	1.82 ± 0.28^{a}	2.25 ± 0.23^{a}	4.42 ± 0.63^{a}	7.03 ± 0.96^{b}	8.94 ± 1.27^{a}	9.86 ± 0.87^{a}

Notes: Values in the table were mean ± standard deviation. Different lowercase letters for data in the same column indicated significant differences ($P < 0.05$).

Table 3. Effect of different SRF application rates on single leaves of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings (piece).

Treatment	6/28/2024	7/28/2024	8/28/2024	9/28/2024
CK	15.00 ± 10.13^{b}	29.57 ± 11.43^{d}	41.43 ± 24.81^{e}	60.43 ± 32.79^{d}
T1	45.67 ± 9.73^{a}	130.22 ± 23.57^{c}	142.11 ± 36.80^{d}	151.89 ± 20.20^{c}
T2	44.75 ± 9.45^{a}	179.13 ± 13.12^{b}	198.25 ± 27.73^{d}	252.50 ± 22.80^{b}
T3	43.71 ± 7.76^{a}	187.86 ± 38.06^{b}	286.28 ± 91.52^{c}	305.86 ± 93.61^{b}
T4	54.43 ± 19.17^{a}	258.29 ± 34.83^{a}	353.57 ± 75.94^{b}	391.29 ± 77.40^{a}
T5	51.71 ± 13.82^{a}	245.00 ± 38.95^{a}	407.57 ± 49.16^{a}	414.43 ± 50.23^{a}

Notes: Values in the table were mean ± standard deviation. Different lowercase letters for data in the same column indicated significant differences ($P < 0.05$).

Table 4. Effect of different SRF application rates on compound leaves of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings (piece).

Treatment	6/28/2024	7/28/2024	8/28/2024	9/28/2024
CK	3.29 ± 1.38^{b}	7.29 ± 3.68^{d}	10.43 ± 6.30^{e}	11.86 ± 8.25^{c}
T1	10.11 ± 3.26^{a}	24.56 ± 6.37^{c}	31.78 ± 8.98^{d}	38.56 ± 5.41^{c}
T2	8.25 ± 2.25^{a}	30.13 ± 3.76^{b}	32.88 ± 3.87^{d}	40.00 ± 4.75^{b}
T3	7.00 ± 1.15^{a}	31.29 ± 7.87^{b}	42.43 ± 12.18^{c}	48.57 ± 13.06^{b}
T4	9.86 ± 4.06^{a}	44.57 ± 4.08^{a}	51.86 ± 10.20^{b}	54.29 ± 15.99^{a}
T5	9.71 ± 2.50^{a}	41.00 ± 5.45^{a}	52.71 ± 9.46^{a}	57.29 ± 8.50^{a}

Notes: Values in the table were mean ± standard deviation. Different lowercase letters for data in the same column indicated significant differences ($P < 0.05$).

641.49%, and 698.42% higher than those in the CK group (Table 3). These results indicated that SRF application significantly promoted the increase in the number of single leaves of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings. A similar trend was observed in the compound leaves. In June, no significant differences were observed between the treatment groups and the CK group. However, as the months progressed, significant differences were observed both between the SRF treatment groups and the CK group, as well as among the different SRF application groups ($P < 0.05$). By the end of the experiment, the increase in compound leaves in the CK group was 8.57 leaves, while the increases in the T1, T2, T3, T4, and T5 groups were 28.45, 31.75, 41.57, 44.43, and 47.58 leaves, respectively. These values were 231.97%, 270.48%, 385.06%, 418.44%, and 455.19% higher than the CK group (Table 4). The test results suggested that SRF application significantly promoted the increase in the number of compound leaves of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings.

Table 5. Effects of different SRF application rates on chlorophyll mass fraction of leaves of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings.

Treatment	Chlorophyll a (mg/g)	Chlorophyll b (mg/g)	Total chlorophyll (mg/g)
CK	0.064 ± 0.004^{d}	0.020 ± 0.001^{e}	0.084 ± 0.004^{e}
T1	0.086 ± 0.003^{c}	0.025 ± 0.002^{d}	0.112 ± 0.005^{d}
T2	0.111 ± 0.003^{b}	0.033 ± 0.002^{c}	0.144 ± 0.005^{c}
T3	0.124 ± 0.004^{b}	0.040 ± 0.001^{b}	0.163 ± 0.004^{b}
T4	0.143 ± 0.014^{a}	0.048 ± 0.003^{a}	0.191 ± 0.017^{a}
T5	0.123 ± 0.002^{b}	0.041 ± 0.002^{b}	0.164 ± 0.004^{b}

Notes: Values in the table were mean ± standard deviation. Different lowercase letters for data in the same column indicated significant differences ($P < 0.05$).

Table 6. Effects of different SRF application rates on soluble sugar and soluble protein amount fractions of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings.

Treatment	Soluble sugar (%)	Soluble protein (mg/g)
CK	2.65 ± 0.27^{f}	1.49 ± 0.11^{e}
T1	3.57 ± 0.34^{e}	2.55 ± 0.29^{d}
T2	4.45 ± 0.27^{d}	3.70 ± 0.28^{c}
T3	5.29 ± 0.21^{c}	4.53 ± 0.32^{b}
T4	7.43 ± 0.23^{a}	4.67 ± 0.43^{a}
T5	6.61 ± 0.37^{b}	4.63 ± 0.21^{b}

Notes: Values in the table were mean ± standard deviation. Different lowercase letters for data in the same column indicated significant differences ($P < 0.05$).

Effects of SRF application rates on physiological indexes of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings

The application of SRF significantly enhanced the chlorophyll content in the leaves of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings. The total chlorophyll mass fraction followed the order of T4 > T5 > T3 > T2 > T1 > CK ($P < 0.05$). A similar trend was observed for chlorophyll a, where the mass fraction decreased in the order of T4 > T3 > T5 > T2 > T1 > CK ($P < 0.05$). For chlorophyll b, the ranking was T4 > T5 > T3 > T2 > T1 > CK ($P < 0.05$) (Table 5). Chlorophyll a, chlorophyll b, and total chlorophyll content in the T4 group were significantly higher than those in the other treatment and CK groups ($P < 0.05$). The results showed a trend of increasing chlorophyll content with the rise in fertilizer application, followed by a decrease beyond the T4 treatment. It was evident that soluble sugar and soluble protein content followed a similar trend, increasing initially and then decreasing with the amount of SRF applied. Both soluble sugar and soluble protein reached their maximum levels under the T4 treatment (6 kg/m^3 SRF). Specifically, soluble sugar content peaked at 7.43%, and soluble protein reached 4.67 mg/g in the T4 group. Compared to the CK group, soluble sugar content in the T1, T2, T3, T4, and T5 groups increased by 34.71%, 67.92%, 99.62%, 180.38%, and 149.43%, respectively ($P < 0.05$). The soluble protein content increased by 1.06, 2.21, 3.04, 3.18, and 3.14 mg/g, representing increases of 71.14%, 148.32%, 204.03%, 213.42%, and 210.74%, respectively ($P < 0.05$) (Table 6). These results demonstrated that SRF application effectively promoted the soluble sugar and soluble protein content in *Zanthoxylum armatum* 'Jiuyeqing' container seedlings.

Effects of SRF application rates on photosynthetic response indexes of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings

The significant differences were found in transpiration rate, stomatal conductance, and net photosynthesis rate between the SRF treatment groups and the CK group ($P < 0.05$).

Table 7. Effects of different SRF application rates on photosynthetic response indexes of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings.

Treatment	Transpiration rate (μmol/m²·s)	Stomatal conductance (mol/m²·s)	Net photosynthetic rate (μmol/m²·s)	Intercellular CO_2 concentration (μmol/m²·s)
CK	436.49 ± 18.81^{e}	15.89 ± 0.33^{f}	3.87 ± 0.59^{f}	290.41 ± 3.45^{a}
T1	465.13 ± 25.95^{d}	17.71 ± 0.16^{e}	5.34 ± 0.21^{e}	276.62 ± 4.43^{b}
T2	507.32 ± 10.79^{c}	18.58 ± 0.30^{d}	6.68 ± 0.72^{d}	251.56 ± 6.46^{c}
T3	571.69 ± 12.28^{b}	19.63 ± 0.15^{c}	8.65 ± 0.41^{c}	230.21 ± 8.93^{d}
T4	632.71 ± 21.46^{a}	21.51 ± 0.12^{a}	11.78 ± 1.15^{a}	188.69 ± 5.71^{e}
T5	578.11 ± 26.50^{b}	20.31 ± 0.38^{b}	9.62 ± 0.39^{b}	205.67 ± 9.24^{f}

Notes: Values in the table were mean ± standard deviation. Different lowercase letters for data in the same column indicated significant differences (P < 0.05).

Table 8. Comprehensive evaluation of the growth of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings.

Treatment	X_1	X_2	X_3	X_4	X_5	X_6	X_7	X_8	X_9	X_{10}	X_{11}	X_{12}	X_{13}	Average Affiliation value	Arrange in order
CK	0.08	0.13	0.08	0.08	0.10	0.03	0.03	0.03	0.03	0.06	0.10	0.05	0.05	0.07	6
T1	0.56	0.58	0.27	0.16	0.26	0.25	0.26	0.18	0.24	0.21	0.22	0.35	0.17	0.29	5
T2	0.75	0.69	0.48	0.39	0.41	0.51	0.51	0.43	0.48	0.34	0.39	0.49	0.39	0.48	4
T3	0.87	0.85	0.59	0.49	0.55	0.69	0.63	0.64	0.63	0.54	0.65	0.67	0.58	0.64	3
T4	0.85	0.82	0.77	0.67	0.90	0.85	0.82	0.89	0.84	0.85	0.90	0.97	0.95	0.85	1
T5	0.83	0.81	0.81	0.71	0.77	0.71	0.63	0.68	0.64	0.63	0.68	0.78	0.79	0.73	2

Notes: X_1: seedling height. X_2: ground diameter. X_3: single leaf. X_4: compound leaves. X_5: soluble protein. X_6: soluble sugar. X_7: chlorophyll a content. X_8: chlorophyll b content. X_9: total chlorophyll content. X_{10}: intercellular carbon dioxide concentration. X_{11}: net photosynthetic rate. X_{12}: stomatal conductance. X_{13}: transpiration rate.

The photosynthetic response parameters showed an initial increase followed by a decrease with the rise in SRF application rate, reaching their maximum values under the T4 treatment. Compared with the CK group, the transpiration rate in the SRF treatment groups increased by 6.56 - 44.95%, stomatal conductance increased by 11.45 - 35.37%, and the net photosynthesis rate increased by 37.98 - 204.39%. Meanwhile, the intercellular CO_2 concentration in the CK group was significantly higher than that in the other treatment groups with a decrease in intercellular CO_2 concentration ranging from 4.99 - 53.91% in the SRF groups. The T4 treatment had the lowest intercellular CO_2 concentration, suggesting an optimal level of SRF application for improving photosynthetic efficiency (Table 7).

Comprehensive analysis of relevant indicators of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings based on the affiliation function method

The growth condition of seedlings is a multifaceted trait, and evaluating seedling development based on a single growth index lacks comprehensiveness and scientific rigor. Therefore, assessing the growth status of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings using multiple indicators provided a more accurate reflection of the effects of various SRF treatments. In this study, the affiliation function method was employed to comprehensively evaluate the growth indexes, physiological indexes, and photosynthetic response indexes of the seedlings under different treatments. The results showed that all treatments were ranked according to the average value of the affiliation function with the order of T4 > T5 > T3 > T2 > T1 > CK (Table 8). This ranking indicated that the SRF treatments had a more beneficial effect on the growth of the seedlings compared to the CK group with the T4 group achieving the highest mean value of the affiliation function at 0.85.

Discussion

The rational use of fertilizer is crucial for plant growth. Reducing fertilizer application while improving fertilizer utilization alongside enhancing the quality of seedling growth is a key focus of current research aimed at reducing costs and increasing production [13-15]. Different tree species have varying fertilizer requirements, and these needs also differ at various stages of seedling growth. Therefore, determining the optimal fertilizer amount for different plants is particularly important [16]. SRFs can synchronize nutrient absorption with plant needs, significantly improving nutrient utilization efficiency. There is a substantial body of research on the use of SRFs in container seedling cultivation including studies on species such as *Cyclobalanopsis gilva* Oerst. [12], *Cunninghamia lanceolata* Hook. [17, 18], *Keteleeria cyclolepis* Flous. [19], *Quercus mongolica* Fisch. [20], *Pistacia chinensis* Bunge. [21], and *Camptotheca acuminata* Decne. [22]. The addition of varying amounts of SRFs to the substrate for container seedlings has been shown to effectively promote seedling growth across these species, indicating differences in the optimal SRF amounts required for different plant species. In container seedling cultivation, nutrient status has a decisive influence on seedling growth. Therefore, using SRFs within a reasonable range can not only meet the nutrient requirements for seedling development but also reduce ineffective nutrient loss and improve nutrient utilization efficiency [23].

In this study, different SRF application rates during April and May had no obvious effects on the height and diameter of *Zanthoxylum armatum* ‘Jiuyeqing’ container seedlings, likely because the root systems were still recovering from transplanting and SRF released nutrients slowly. After June, as SRF nutrient release stabilized and plants entered a rapid growth phase, differences among the various SRF treatments became evident. From June to September, seedling height, diameter, and the number of single and compound leaves in the T1 – T5 groups increased significantly compared to the CK group. By the end of the experiment, the height and diameter were the highest under the T3 treatment, while the number of single and compound leaves was the greatest under T5. The results indicated that SRF application significantly promoted the growth of *Zanthoxylum armatum* ‘Jiuyeqing’ container seedlings. Chlorophyll, the key substance for photosynthesis, plays a critical role in plant growth. Increasing SRF application significantly enhanced the synthesis and accumulation of photosynthetic pigments, specifically chlorophyll a and chlorophyll b, thereby improving the leaves' light absorption capacity and ultimately promoting plant growth [24]. The chlorophyll content in the T1 – T5 treatment groups was significantly higher than that in the CK group with chlorophyll a, chlorophyll b, and total chlorophyll values peaking under T4 treatment. These findings demonstrated that appropriate SRF application promoted chlorophyll synthesis, improved light energy utilization, and enhanced plant growth, consistent with previous research [3, 25, 26]. The accumulation of plant organic matter is an important indicator of growth quality with soluble sugars and soluble proteins serving as key metrics. In this study, SRF application significantly increased soluble sugar and soluble protein contents in *Zanthoxylum armatum* ‘Jiuyeqing’ container seedlings compared to that in the CK group. Both soluble sugar and protein contents peaked under T4 treatment. However, further increases in SRF application led to slight declines, aligning with previous research [3, 27, 28], which suggested that exceeding the plants' critical nutrient threshold could disrupt internal nutrient balance and negatively affect growth. Photosynthesis is the primary energy source for carbon assimilation and material accumulation in plants, making it closely tied to growth status [29]. This study found that stomatal conductance, transpiration rate, and net photosynthetic rate reached maximum values under T4 treatment, while intercellular carbon dioxide concentration was at its lowest. The results suggested that slow-release fertilizer enhanced stomatal conductance, increasing CO_2

fixation, which in turn lowered intercellular CO_2 concentration and boosted photosynthetic and transpiration rates. These results were consistent with previous findings [30]. However, excessive SRF application led to declines in these indicators, warranting further investigation into the underlying causes.

A comprehensive evaluation using the affiliation function method showed that the T4 treatment, corresponding to an SRF application rate of 6 kg/m^3, achieved the highest overall score for *Zanthoxylum armatum* 'Jiuyeqing' container seedlings. Considering the growth, physiological, and photosynthetic response indices, an application of 6 kg/m^3 SRF was recommended to enhance the quality of seedlings during future cultivation of *Zanthoxylum armatum* 'Jiuyeqing' container seedlings.

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