

# FERTILISATION PROMOTES *ZANTHOXYLUM ARMATUM* ‘HANYUAN PUTAO QINGJIAO’ PERICARP YIELD DUE TO SHOOT GROWTH, NUTRIENT STORAGE AND STRESS RESISTANCE

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**Abstract.** In southwest China, the fruit (pericarp) of *Zanthoxylum armatum* ‘Hanyuan Putao Qingjiao’ is picked using the method of “cutting shoots instead of picking fruits” after June. After harvesting, the fruiting branches grow rapidly, accumulate nutrients, and blossom and fruit in the next spring. However, the correlation between fruit yield and shoot growth, nutrient storage and stress resistance is not clear. The effects of different urea (N), superphosphate (P) and potassium (K) fertilization on shoot growth, nutrient content, relative electric conductivity (*Rec*), malondialdehyde (*Mda*) of leaves, and pericarp weight per individual (*Pew*) of *Z. armatum* ‘Hanyuan Putao Qingjiao’ were studied. Fertilisation significantly increased *Pew*, and Treatment 12 (*Tre12*) significantly increased *Pew* by 42.73% compared with the Treatment 1 (*Tre1*,  $p < 0.05$ ). *Pew* was significantly positively correlated with shoot length, shoot number, shoot diameter, branch and leaf dry weight, fruit setting rate, N and P content in shoots and leaves ( $p < 0.01$ ), significantly positively correlated with K content in shoots and leaves ( $p < 0.05$ ), and significantly negatively correlated with leaf *Rec* ( $p < 0.01$ ). *Tre12* (N<sub>2</sub>P<sub>3</sub>K<sub>1</sub>) had a high P content, which was conducive to improving shoot growth and nutrient accumulation in shoots and leaves, protecting the integrity of the leaf membrane and promoting *Pew*.

**Keywords:** Chinese prickly ash, fertilisation, pericarp yield, shoots and leaf growth, nutrients, resistance

## Introduction

Fruit yield has always been one of the main objectives of non-wood forest product management. Analysis of fertiliser demand patterns in plant growth and development and soil fertiliser capacity and the use of fertilisation to improve the fruit yield of the forest is a common and effective method to achieve this objective, and there have been many research reports (Sete et al., 2020; Lo’ay, 2021).

Plants need to consume nutrients during their growth and development. Under limited resources, there are trade-offs in plant resource allocation (Whitehead and Poveda, 2019). If resources are allocated to shoot and root growth and/or nutrient storage, flower bud differentiation is affected, which is not conducive to flowering and fruiting (Louda, 1986), thus affecting yield. However, shoots and roots are important organs for plants to obtain assimilates and mineral nutrients, and their growth potential is closely related to nutrient

supply. Appropriate growth is beneficial for plants in obtaining sufficient nutrients, thus promoting flowering and fruiting performance. Studies have shown that fertilisation can promote both plant growth and fruit yield (El-Gleel Mosa et al., 2018).

Chinese prickly ash is widely distributed in subtropical and middle-eastern warm temperate regions of China (Zhuo et al., 2020), and the pericarp is mainly used as a spice (Luo et al., 2022). It also has health care functions (Singh et al., 2015) and is an important non-wood forest species. At present, fertilisation technology is gradually applied to the cultivation of Chinese prickly ash. In Sichuan Province of China, through fertilisation, 2-3-year-old Chinese prickly ash can bear fruit. However, there are few reports about the effects of fertilisation on flowering and fruiting, shoot and leaf growth, and nutrient storage in this species.

In the process of plant growth, plants often suffer from various environmental stressors, such as drought and low temperatures. Plants need to resist environmental stress to ensure normal flowering and fruiting. Fertilisation can also improve plant stress resistance to the environment (Zhou et al., 2021). In the yearly growth of Chinese prickly ash, late spring cold, which is prone to occur at the leaf-spreading stage (at the end of March), affects the development of plant leaves and young fruit. However, it is not clear what influence fertilisation has on stress resistance while increasing fruit yield.

At present, basic theoretical studies on Chinese prickly ash mainly involve stress tolerance physiology, breeding systems, cultivation techniques, extraction and activity identification of fruit active components, etc. (Wang et al., 2019; Zhou et al., 2020, 2021; Fei et al., 2021; Tian et al., 2021). This study used 3-year-old *Zanthoxylum armatum* 'Hanyuan Putao Qingjiao' as the research material. The effects of fertilisation on shoot and leaf growth, nutrient content (N, P and K) of shoots and leaves, fruit setting, and leaf resistance characteristics were studied, and the relationship between growth, nutrient storage and fruiting were analysed. The main objective of this study was to reveal the yield formation mechanism and provide theoretical support for the high-yielding cultivation of this plant species.

## Materials and methods

### *Experimental site*

The experimental site was located at the Chongzhou Teaching and Research Experimental Site of Sichuan Agricultural University in Sichuan Province, China (E 103°38', N 30°35', elevation of 520 m). The experimental site is characterized by the subtropical humid monsoon climate, with an average annual temperature of 16.2 °C, the average temperature of the hottest month in July and the coldest month in January is 25.3 °C and 5.8 °C, respectively. The annual cumulative sunshine hours are 1161.5 h and the average annual rainfall is 977.8 mm (meteorological data are collected from China Meteorological Data Network: <http://data.cma.cn/>, 1981-2010). The experimental soil is paddy soil, and the basic chemical properties are shown in *Table 1*.

*Table 1. Chemical properties of soil in experimental site*

pH	Soil organic matter (g/kg)	Available nitrogen (mg/kg)	Available phosphorus (mg/kg)	Available potassium (mg/kg)
7.06	18.7	130	4.3	92.8

## Plant materials

The plant material was 3-year-old *Z. armatum* ‘Hanyuan Putao Qingjiao’ with similar growth characteristics. The plant material was obtained from Chinese prickly ash Germplasm Resource Nursery of Sichuan Agricultural University and was identified by Dr. Chen Xiaohong. The row spacing of the plants is 2 m × 3 m, with no interplant.

## Experimental design

### Fertilisation

An orthogonal design of L<sub>16</sub> (3<sup>4</sup>), with 3 replicates in a completely randomised block design (each replicate contained 5 individuals), was used (with 16 treatments in total, Table 2). The nitrogen, phosphorus and potassium fertilizer of each treatment were mixed and applied around the drip line of the tree crown (the fertilization position was about 1.5 m distance from the trunk, Fig. 1A). Fertilizer of each treatment was applied 4 times: (1) 50% of the total fertilizer was used in July 2017, 15 days before fruit collection; (2) 10% of the total fertilizer was used in October 2017; (3) 20% of the total fertilizer was used before flower bud differentiation in February 2018; (4) 20% of the total fertilizer was used after flower bud differentiation in April 2018, and other cultivation techniques refers to the China national standard of LY/T 2042-2012 (The State Forestry Bureau of China, 2012).

**Table 2.** Fertilizer type and dose in each fertilisation treatment for *Zanthoxylum armatum* ‘Hanyuan Putao Qingjiao’

Numbers	Treatments	Content (g/individual)			Fertilizing amount (g/individual)		
		N	P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O	Urea	Superphosphate	Potassium sulfate
Tre 1	N <sub>0</sub> P <sub>0</sub> K <sub>0</sub>	0	0	0	0	0	0
Tre 2	N <sub>0</sub> P <sub>1</sub> K <sub>1</sub>	0	18	45	0	150	83.3
Tre 3	N <sub>0</sub> P <sub>2</sub> K <sub>2</sub>	0	36	90	0	300	166.7
Tre 4	N <sub>0</sub> P <sub>3</sub> K <sub>3</sub>	0	72	180	0	600	333.3
Tre 5	N <sub>1</sub> P <sub>0</sub> K <sub>1</sub>	45	0	45	97.8	0	83.3
Tre 6	N <sub>1</sub> P <sub>1</sub> K <sub>0</sub>	45	18	0	97.8	150	0
Tre 7	N <sub>1</sub> P <sub>2</sub> K <sub>3</sub>	45	36	180	97.8	300	333.3
Tre 8	N <sub>1</sub> P <sub>3</sub> K <sub>2</sub>	45	72	90	97.8	600	166.7
Tre 9	N <sub>2</sub> P <sub>0</sub> K <sub>2</sub>	90	0	90	195.7	0	166.7
Tre 10	N <sub>2</sub> P <sub>1</sub> K <sub>3</sub>	90	18	180	195.7	150	333.3
Tre 11	N <sub>2</sub> P <sub>2</sub> K <sub>0</sub>	90	36	0	195.7	300	0
Tre 12	N <sub>2</sub> P <sub>3</sub> K <sub>1</sub>	90	72	45	195.7	600	83.3
Tre 13	N <sub>3</sub> P <sub>0</sub> K <sub>3</sub>	180	0	180	391.3	0	333.3
Tre 14	N <sub>3</sub> P <sub>1</sub> K <sub>2</sub>	180	18	90	391.3	150	166.7
Tre 15	N <sub>3</sub> P <sub>2</sub> K <sub>1</sub>	180	36	45	391.3	300	83.3
Tre 16	N <sub>3</sub> P <sub>3</sub> K <sub>0</sub>	180	72	0	391.3	600	0

Level 2 (N<sub>2</sub>P<sub>2</sub>K<sub>2</sub>) was an approximation of the local recommended fertilizer application dose

## Sampling and measurements

(1) Shoots growth: In December 2017, the shoot diameter (*Shd*) and shoot length (*Shl*) of each individual were measured using a Vernier calliper and tapeline, and the number of fruit-bearing shoots (*Sha*) on each individual was counted.



**Figure 1.** Fertilizer treatment and harvesting of fruit. A: Fertilizer treatment at the drip line of the tree crown; B: shoots and leaves were heading-back cut together when the fruits were picked

(2) Fruit-setting and pericarp weight: Using the standard branch method, the number of flowers and number of fruit were investigated in March 4<sup>th</sup> and June 27<sup>th</sup> 2018, respectively, and the fruit setting rate (*Fsr*) of *Z. armatum* ‘Hanyuan Putao Qingjiao’ was calculated using the following equation:

$$Fsr = \frac{\text{Number of fruit}}{\text{Number of flowers}} \times 100\% \quad (\text{Eq.1})$$

At the end of June 2018, the fruit of *Z. armatum* ‘Hanyuan Putao Qingjiao’ was collected from each individual, dried and both seeds and impurities were picked out, then the pericarp weight per individual was weighed (*Pew*).

(3) Physiological index: During the leaf-spreading stage (March 29<sup>th</sup>, 2018), nine trees were randomly selected for each treatment, with a total of 144 individuals. The leaves in the middle of shoots were randomly collected from four directions (east, west, north and south). The relative conductivity (*Rec*) was determined with a conductivity metre (DDS-11A, Shanghai Inesa Scientific Instrument Co., Ltd., Shanghai, China; Zhang, 2007), and the malondialdehyde content (*Mda*) was determined with thiobarbituric acid using a spectrophotometer (TU-1810, Beijing Purkinje General Instrument Co., Ltd, Beijing, China), absorbances was read at 532, 600 and 450 nm (Li, 2000).

On July 27<sup>th</sup> 2018, the shoots and leaves were heading-back cut together with fruit when they were picked (Fig. 1B). Both the shoots and leaves were then taken back to the

lab, and the oven-dried weight (*Slw*) and mineral element content (N, P and K) of shoots and leaves were determined. After the samples were treated with sulphuric acid–perchloric acid, the N content was determined using the Kjeldahl analysis. The P content was determined using the molybdenum–antimony anti-spectrophotometric method, and the K content was determined using a TAS-986 atomic spectrophotometer (Beijing Puyan General Instrument Co., Ltd, Beijing, China) (The State Forestry Bureau of China, 1999).

### Data analysis

Statistical analysis of the data was performed using IBM SPSS Statistics 19, and a one-way ANOVA with Duncan's test was used to analyse the significance of differences between fertilisation treatments. Correlations between growth, fruit setting and physiological characteristics were analysed by a two-tailed Pearson test. Meanwhile, principal component analysis (PCA) was used to analyse the possible combinations of growth, fruit setting and physiological characteristics between fertilisation treatments.

## Results

### Shoot growth and shoot and leaf weights

Fertilisation promoted the *Sha*, *Shl* and *Shd* of *Z. armatum* 'Hanyuan Putao Qingjiao' ( $p < 0.05$ , Table 3). The *Sha* in *Tre1* was significantly lower than that of *Tre7–Tre16* ( $P < 0.05$ ), but there were no differences in *Sha* among *Tre7–Tre16* ( $p > 0.05$ ). The *Shl* of *Tre1* was significantly shorter than that of *Tre8–Tre12* and *Tre14–Tre16* ( $p < 0.05$ ), while there were no differences in *Shl* among *Tre8–Tre12* and *Tre14–Tre16* ( $p > 0.05$ ). There was a significant difference in *Shd* between *Tre1* and *Tre10*, as well as *Tre12* ( $p < 0.05$ ), but there was no significant difference between *Tre10* and *Tre12* ( $p > 0.05$ ).

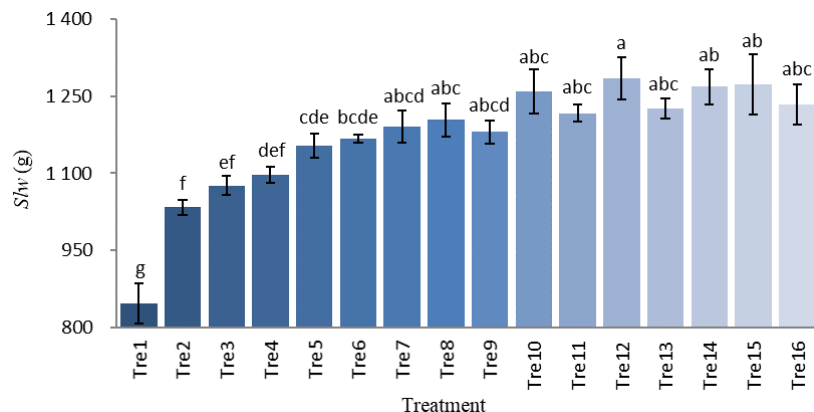
**Table 3.** Growth characteristics of 3-year-old *Zanthoxylum armatum* 'Hanyuan Putao Qingjiao' shoots measured in November

Treatment	Sha (number)	Shl (cm)	Shd (mm)
Tre1	112.44±4.98 d	98.28±4.62 b	8.29±0.48 c
Tre2	116.72±6.61b cd	99.21±6.43 b	8.45±0.41 abc
Tre3	115.89±6.35 cd	99.06±4.80 b	8.34±0.50 bc
Tre4	118.59±6.37 bcd	99.33±5.68 b	8.47±0.47 abc
Tre5	121.56±6.10 bcd	100.21±4.64 ab	8.58±0.41 abc
Tre6	122.67±6.56 bcd	99.67±4.23 b	8.50±0.50 abc
Tre7	125.97±6.33 abc	100.89±5.07 ab	8.67±0.39 abc
Tre8	125.77±6.72 abc	101.50±6.31 a	8.76±0.48 abc
Tre9	125.56±5.83 abc	101.82±4.89 a	8.93±0.47 abc
Tre10	129.00±7.56 ab	104.22±4.61 a	9.27±0.45 a
Tre11	126.44±6.17 abc	103.62±5.86 a	9.03±0.62 abc
Tre12	129.44±7.12 a	104.95±5.73 a	9.23±0.43 ab
Tre13	125.34±6.24 abc	100.94±5.42 ab	8.72±0.37 abc
Tre14	129.22±5.70 a	103.78±5.21 a	9.04±0.43 abc
Tre15	127.56±5.76 abc	103.22±5.52 a	9.00±0.41 abc
Tre16	124.56±5.29 abc	100.52±5.06 a	8.62±0.38 abc

Sha-Shoots amount; Shl-Shoots length; Shd-Shoots diameter. The data are expressed as the means ± SEs, and the different letters indicate a significant difference among the different fertilisation treatments at  $p < 0.05$  level



Fertilisation increased the *Slw* of *Z. armatum* ‘Hanyuan Putao Qingjiao’ ( $p < 0.05$ , Fig. 2). *Tre2* had the smallest increment in *Slw*, which was 22.16% higher than that in *Tre1*, while *Tre12* had the largest increment in *Slw*, which was 51.86% higher than that in *Tre1*.



**Figure 2.** branch and leaf oven-dried weight of *Zanthoxylum armatum* ‘Hanyuan Putao Qingjiao’. *Slw* -Shoots & leaf weight

### Shoot and leaf nutrient content

Different fertilisation treatments had different effects on P, N and K accumulation in shoots (Table 4). The P content of shoots (*Pcs*) in *Tre3*, *Tre4* and *Tre6–Tre16* was significantly higher than that in *Tre1* ( $p < 0.05$ ). *Tre12* presented the highest *Pcs*, which was 1.45 times higher than that presented by *Tre1*. The N content of shoots (*Ncs*) in *Tre4–Tre16* was significantly higher than that in *Tre1* ( $p < 0.05$ ). The highest *Ncs* was recorded in *Tre16*, which was 1.82 times higher than that in *Tre1*. The K content of shoots (*Kcs*) in *Tre3–Tre5* and *Tre7–Tre16* was significantly higher than that in *Tre1* ( $p < 0.05$ ). *Kcs* was the highest in *Tre13*, which was 1.38 times higher than that in *Tre1*.

**Table 4.** Characteristics of nutrient content in shoots

Treatment	<i>Pcs</i> (g/kg <sup>-1</sup> )	<i>Ncs</i> (g/kg <sup>-1</sup> )	<i>Kcs</i> (g/kg <sup>-1</sup> )
Tre1	1.50±0.05 i	6.62±0.18 k	8.63±0.25 h
Tre2	1.59±0.06 hi	7.05±0.21 jk	9.3±0.17 gh
Tre3	1.70±0.06 fgh	7.21±0.22 jk	9.48±0.31 fg
Tre4	1.87±0.04 cdef	7.58±0.28 ij	9.65±0.33 efg
Tre5	1.65±0.05 ghi	8.24±0.27 hi	9.69±0.22 efg
Tre6	1.79±0.04 defg	8.92±0.27 gh	9.29±0.18 gh
Tre7	1.89±0.04 cde	9.4±0.25 fg	10.51±0.21 cde
Tre8	2.08±0.04 ab	9.56±0.34 efg	9.96±0.31 defg
Tre9	1.75±0.06 efgh	10.01±0.34 def	10.72±0.19 cd
Tre10	1.83±0.05 def	10.27±0.23 cdef	11.04±0.26 bc
Tre11	2±0.05 bc	10.39±0.25 cde	9.82±0.27 efg
Tre12	2.17±0.07 a	10.64±0.32 cd	10.21±0.3 cdef
Tre13	1.71±0.05 fgh	11.01±0.38 bc	11.91±0.2 a
Tre14	1.79±0.04 defg	11.59±0.35 ab	11.7±0.36 ab
Tre15	1.95±0.06 bcd	11.87±0.39 ab	11.02±0.37 bc
Tre16	2.07±0.07 ab	12.07±0.34 a	10.53±0.32 cde

*Pcs*-P content of shoots; *Ncs*-N content of shoots; *Kcs*-K content of shoots

There was a significant difference for P, N and K accumulation in leaves under different fertilisation treatments (Table 5). The leaf P content (*Pcl*) of *Tre3*, *Tre4*, *Tre7*, *Tre8*, *Tre10–Tre12*, *Tre15* and *Tre16* was significantly higher than that of *Tre1* ( $p < 0.05$ ), among which *Pcl* of *Tre12* was the highest at 28.57% higher than that in *Tre1*. The leaf N content (*Ncl*) in fertilisation treatments (except for *Tre2–Tre4*) was significantly higher than that in *Tre1* ( $p < 0.05$ ), and *Ncl* in *Tre16* was highest and was 68.43% higher than that in *Tre1*. The leaf K content (*Kcl*) of *Tre4*, *Tre5* and *Tre7–Tre16* was significantly higher than that in *Tre1* ( $p < 0.05$ ), among which the *Kcl* of *Tre13* was the highest and was 41.75% higher than that of *Tre1*.

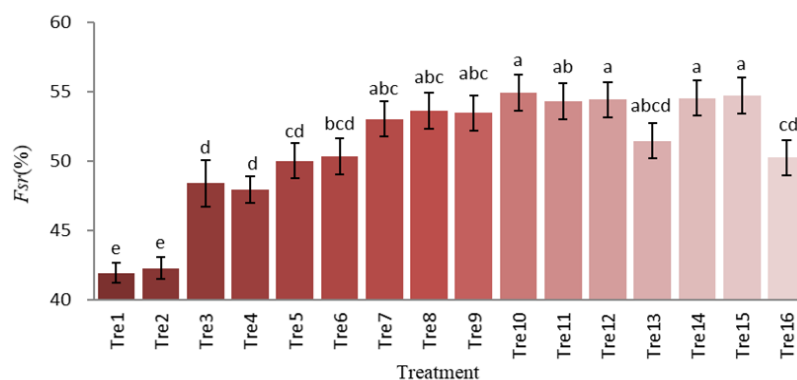
**Table 5.** Characteristics of nutrient content in leaves

Treatment	Pcl (g/kg <sup>-1</sup> )	Ncl (g/kg <sup>-1</sup> )	Kcl (g/kg <sup>-1</sup> )
Tre1	1.75±0.06 g	11.15±0.38 j	10.3±0.19 f
Tre2	1.83±0.05 efg	11.52±0.35 j	10.75±0.35 ef
Tre3	1.97±0.06 cdef	12.1±0.38 ij	11.11±0.21 def
Tre4	2.03±0.07 bcd	12.35±0.4 ij	11.43±0.3 de
Tre5	1.79±0.05 fg	13.27±0.29 hi	11.59±0.35 de
Tre6	1.92±0.05 defg	14.49±0.47 gh	10.82±0.29 ef
Tre7	2±0.06 cde	14.99±0.45 fg	12.08±0.31 cd
Tre8	2.2±0.08 ab	15.27±0.4 efg	11.76±0.23 de
Tre9	1.87±0.06 defg	15.85±0.52 defg	12.88±0.33 bc
Tre10	1.97±0.06 cdef	16.19±0.55 cdef	13.46±0.35 b
Tre11	2.12±0.05 abc	16.59±0.54 bcde	11.72±0.31 de
Tre12	2.25±0.06 a	16.83±0.55 bcd	12.04±0.34 cd
Tre13	1.81±0.04 efg	17.5±0.55 abc	14.6±0.37 a
Tre14	1.93±0.05 cdefg	17.92±0.62 ab	13.92±0.42 ab
Tre15	1.98±0.07 cdef	18.55±0.61 a	13.69±0.47 ab
Tre16	2.12±0.07 abc	18.78±0.6 a	13.16±0.44 b

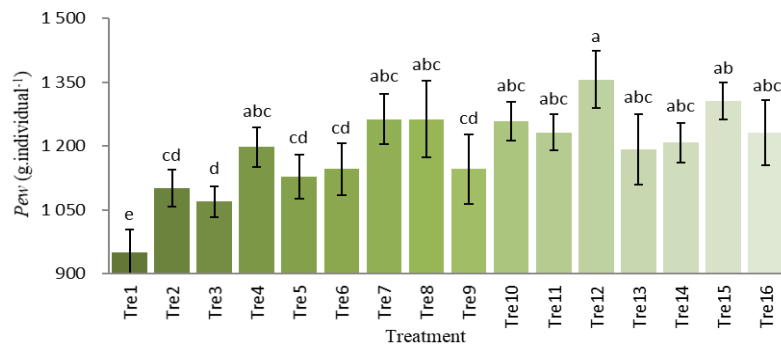
Pcl-P content of leaf; Ncl-N content of leaf; Kcl-K content of leaf

### Fruit setting and pericarp weight

Fertilisation significantly increased the fruit setting rate (*Fsr*), except in *Tre2* ( $p < 0.05$ , Fig. 3). *Tre10* had the largest *Fsr* increment, which was 1.31 times higher than *Tre1*. Fertilisation significantly increased pericarp weight per individual (*Pew*,  $p < 0.05$ , Fig. 4). The *Pew* increment of *Tre3* was the least and was only 12.53% higher than that of *Tre1*. *Tre12* had the largest *Pew* increment and was 42.73% higher than *Tre1*.



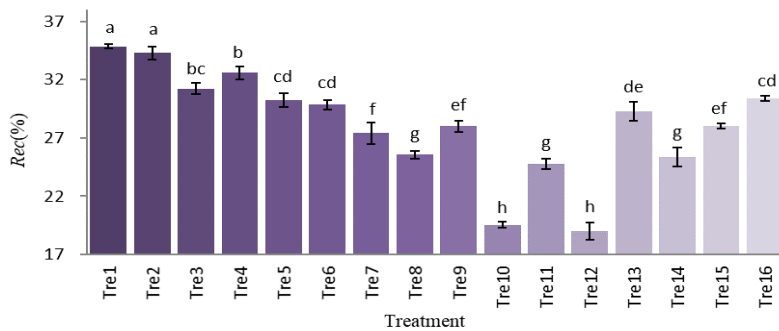
**Figure 3.** Fruit-setting rate of *Zanthoxylum armatum* 'Hanyuan Putao Qingjiao' under different fertilisation treatment. *Fsr*-Fruit setting rate



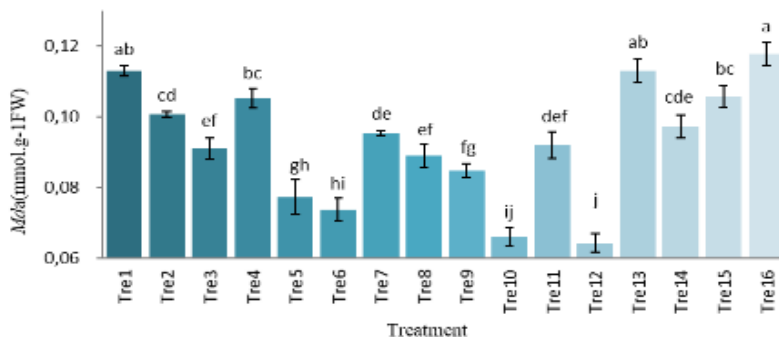
**Figure 4.** Pericarp weight of *Zanthoxylum armatum* 'Hanyuan Putao Qingjiao' under different fertilisation treatment. *Pew*- Pericarp weight

### Peroxidation product of membrane lipids in leaves

Fertilisation significantly reduced the leaf relative electric conductivity (*Rec*) except in *Tre2* ( $p < 0.05$ , Fig. 5). *Tre12* had the greatest *Rec* reduction, which was 45.49% lower than *Tre1*. Fertilisation significantly reduced malondialdehyde content in leaves (*Mda*), except in *Tre4*, *Tre13*, *Tre15* and *Tre16* ( $p < 0.05$ , Fig. 6). *Tre12* had the largest *Mda* reduction, which was 43.36% lower than that in *Tre1*.



**Figure 5.** Relative electric conductivity of leaf under different fertilisation treatment. *Rec*- Relative electric conductivity



**Figure 6.** Malondialdehyde content of leaf under different fertilisation treatment. *Mda*-malondialdehyde



### Correlation between growth, physiological indexes and *Pew*

Correlation analysis between *Pew* and growth, as well as physiological indexes, showed that there were positive correlations at the 0.05 level between *Pew* and *Kcs*, as well as *Kcl*; there were positive correlations at the 0.01 level between *Pew* and *Shl*, *Sha*, *Shd*, *Slw*, *Fsr*, *Pcs*, *Ncs*, *Pcl* and *Ncl*, and there was a negative correlation between the *Pew* and *Rec* ( $p < 0.01$ , Table 6).

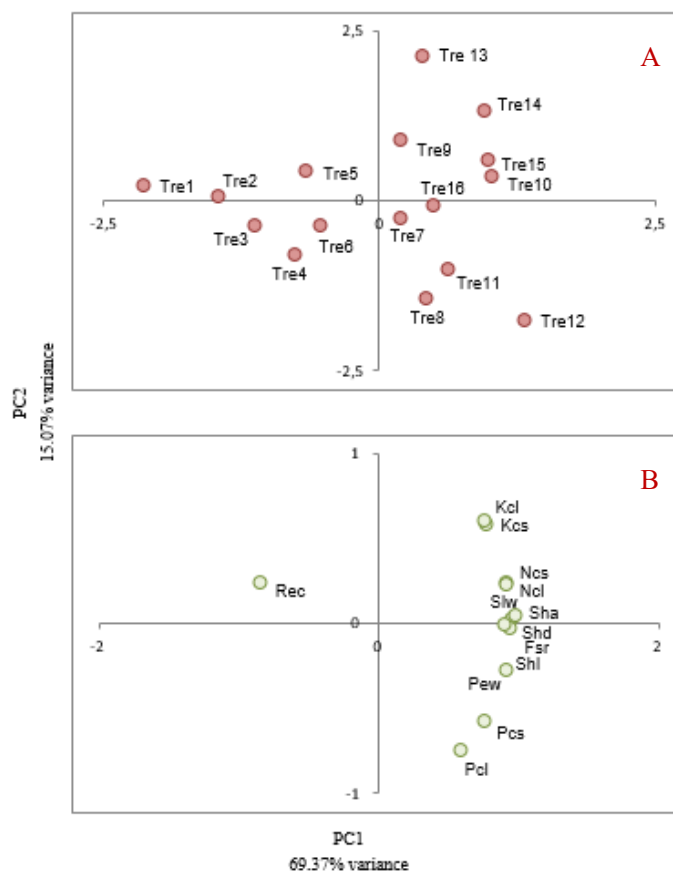
**Table 6.** Correlation between pericarp weight per individual and growth as well as physiological index

Indexes	Shl	Sha	Shd	Slw	Fsr	Pcs	Ncs	Kcs	Pcl	Ncl	Kcl	Rce
r	0.78 <sup>e</sup>	0.87 <sup>e</sup>	0.76 <sup>e</sup>	0.89 <sup>e</sup>	0.81 <sup>e</sup>	0.87 <sup>e</sup>	0.74 <sup>e</sup>	0.57 <sup>s</sup>	0.74 <sup>e</sup>	0.74 <sup>e</sup>	0.54 <sup>s</sup>	-0.76 <sup>e</sup>

<sup>e</sup> and <sup>s</sup> indicates a significance level of  $p < 0.01$  and  $p < 0.05$ , respectively. Slw -Shoots & leaf weight; Fsr-Fruit setting rate; Rec-Relative electric conductivity

### Principal component analysis (PCA)

The PCA results showed the classification of the N, P and K content under the different fertilisation doses (Fig. 7A). The N, P and K content characteristics of *Tre13* (positive score) and *Tre12* (negative score) were distinguished by PC1, and those of *Tre12* (positive score) and *Tre1* (negative score) were distinguished by PC2.



**Figure 7.** Principal component analysis (PCA) of the growth and physiological characteristics of *Zanthoxylum armatum* 'Hanyuan Putao Qingjiao' under different fertilisation treatments

The PCA results of several physiological indexes of *Z. armatum* ‘Hanyuan Putao Qingjiao’ could distinguish fertilisation treatments well (Fig. 7B). *Sha* and *Slw* were associated with positive PC1 scores, whereas *Rec* was associated with negative PC1 scores. *Kcl* and *Kcs* were associated with positive PC2 scores, and *Pcl* and *Pcs* were associated with negative PC2 scores.

## Discussion

Chinese prickly ash is an affinal drug and diet plant. In production, the shoots are cut when fruit is harvested, which is convenient for the harvesting process. In Sichuan Province, China, after the fruit is harvested in early June, the growth of shoots is promoted, and fruit-bearing shoots are formed through fertilisation. From June to December, the shoots continue to grow, while nutrient accumulation and flower bud differentiation occur (flower bud differentiation starts at the end of July and ends in March of the next year). Our results showed that fertilisation significantly promoted shoot growth, nutrient accumulation and stress resistance, which in turn increased *Fsr* and *Pew*.

After the fruit of *Z. armatum* ‘Hanyuan Putao Qingjiao’ was picked using the method of “cutting shoots instead of picking fruits”, the shoots grew rapidly from June to November and developed into fruit-bearing shoots in the coming year. Fertilisation significantly promoted the shoot growth of this plant species ( $p < 0.05$ , Table 3, Fig. 2). *Tre12* had the largest increase in *Sha*, *Shl* and *Slw*, which were 15.12%, 6.79% and 51.86% higher than that in *Tre1*, respectively. *Tre10* had the largest increase in *Shd*, which was 11.82% higher than that in *Tre1*. This is consistent with reports that fertilisation promotes tree growth (Milošević et al., 2013; Guo et al., 2016). It is generally believed that, under limited resources, the increase in plant vegetative growth (such as shoots and leaves) reduces resource allocation in the reproductive (fruiting) process (Hansen et al., 1992), resulting in a decrease in fruit yield. For Chinese prickly ash, flower bud differentiation starts at the end of July, when the shoots grow, and flower bud differentiation plays an important role in fruit setting (Julian et al., 2010). The *Pew* of this plant species was positively correlated with *Shl*, *Sha*, *Shd* and *Slw* ( $P < 0.01$ , Table 6). Furthermore, the *Pew* of *Tre12* was the highest, with  $1356.47 \pm 66.69$  g·individual<sup>-1</sup>, which was 1.43 times higher than that of *Tre1* (Fig. 4). Fertilisation could guarantee the nutrient requirement for both shoot growth and flower bud differentiation of *Z. armatum* ‘Hanyuan Putao Qingjiao,’ and good shoot growth was conducive to improving the *Pew* of this plant species. Under fertilisation, no resource competition occurred between vegetative and reproductive growth in *Z. armatum* ‘Hanyuan Putao Qingjiao.’ Previous studies have also found that good vegetative growth can promote fruit setting and increase fruit yield under fertilisation (Marschner, 2012; Milošević et al., 2013). *Tre12* had a high P level ( $72\text{g P}_2\text{O}_5\cdot\text{individual}^{-1}$ ), and fertilisation with a high P level promoted flower bud differentiation (Eshghi et al., 2007; Zhou et al., 2020), thus increasing the *Pew* of *Z. armatum* ‘Hanyuan Putao Qingjiao’.

The nutrient levels of shoots are important for maintaining plant growth and thus increasing yield (Brinkman and Boerner, 1994). In this study, fertilisation significantly promoted N, P and K accumulation in both shoots and leaves, with the highest P content in shoots and leaves of *Tre12*, the highest N content in shoots and leaves of *Tre16*, and the highest K content in shoots and leaves of *Tre13* (Tables 4 and 5). This was consistent with Milošević et al. (2013), who found that fertilisation significantly promoted the accumulation of mineral nutrients in apricot leaves. The P and N content in shoots and

leaves were significantly positively correlated with  $P_{ew}$  ( $P < 0.01$ ), and the K content in shoots and leaves was significantly positively correlated with  $P_{ew}$  ( $P < 0.05$ , Table 6). Increasing the N, P and K content in shoots and leaves was beneficial in increasing the  $P_{ew}$  of *Z. armatum* 'Hanyuan Putao Qingjiao.' Vajari et al. (2018) showed that the application of urea, zinc sulphate and boric acid increased the N concentration in buds by 33.65% and fruit quantity by 22.21%.

It is generally believed that tree nutrients affect  $F_{sr}$  (Vaughton, 1991; Trueman and Wallace, 1999). Studies have shown that flowering consumes nutrient storage in Litchi trees, resulting in a lower  $F_{sr}$  (Jiang et al., 2012). Fertilisation can significantly improve  $F_{sr}$  (Zubair et al., 2017). In this study, fertilisation significantly promoted the  $F_{sr}$  of *Z. armatum* 'Hanyuan Putao Qingjiao' (Fig. 3) and then increased  $P_{ew}$  (Table 6). In particular, the  $F_{sr}$  of *Tre10* was the highest at 30.92% higher than that of *Tre1* (Fig. 3). In this experiment, fertilisation treatment was applied 4 times, and the explanation that fertilisation increases  $F_{sr}$  of *Z. armatum* 'Hanyuan Putao Qingjiao' may be that fertilisation in February is conducive to promoting flower bud development and nutrient accumulation (Rodrigo et al., 2009; Julian et al., 2010). The nutrients provided by fertilisation in April guarantee young fruit development in this plant species. Fertilisation in July is conducive to the recovery of tree potential, promoting the growth of fruiting shoots and increasing the accumulation of photosynthetic products (Trueman and Wallace, 1999). Fertilisation in October can increase nutrient storage and promote germination and flowering in the spring (Villar-Salvador et al., 2015). As for which period of fertilisation is the most critical to improve the  $F_{sr}$  of *Z. armatum* 'Hanyuan Putao Qingjiao,' it remains to be further studied.

Fertilisation can improve plant stress resistance, such as low temperature and drought stress (Oliet et al., 2013; Faustino et al., 2015), thus ensuring normal growth and development. Compared with the control (*Tre1*), the leaf  $Rec$  and  $Mda$  of *Z. armatum* 'Hanyuan Putao Qingjiao' were significantly reduced by fertilisation ( $p < 0.05$ , Figs. 5 and 6), especially when the application effects of *Tre12* were the best. Fertilisation can reduce membrane lipid peroxidation and protect the integrity of the leaf membrane, which is of great significance for improving the adaptability of Chinese prickly ash to occasional low-temperature stress in March (Islam et al., 2009). There was a negative correlation between  $Rec$  and  $P_{ew}$  ( $p < 0.05$ , Table 6), indicating that fertilisation reduced leaf  $Rec$  and promoted stress resistance, which was beneficial in increasing the fruit yield of *Z. armatum* 'Hanyuan Putao Qingjiao.' Some studies have also shown that fertilisation can significantly reduce electrolyte leakage and improve leaf membrane stability (Kim et al., 2017; Raza et al., 2021).

Principal component analysis of shoot growth, nutrient storage, stress resistance and pericarp yield showed that *Tre12* had the highest PC1 score,  $Sha$  and  $Slw$  and the lowest  $Rec$  values. Meanwhile, *Tre12* had the minimum PC2 scores and the highest  $Pcl$  and  $Pcs$  values (Fig. 7). *Tre12* had the highest pericarp yield (Fig. 4), indicating that *Tre12* significantly promotes shoot and leaf growth and P accumulation and reduces leaf  $Rec$ , which contributes to promoting pericarp yield. A positive correlation between  $P_{ew}$  and  $Sha$ ,  $Shl$ ,  $Slw$ ,  $Pcl$  and  $Pcs$  ( $p < 0.01$ ) and a negative correlation with  $Rec$  also demonstrated these results ( $p < 0.05$ , Table 6).

## Conclusion

Fertilisation treatment significantly affected the pericarp yield of Chinese prickly ash, which was closely related to shoot and leaf growth, nutrient accumulation, and stress resistance, and presented a significant positive correlation between *Pew* and *Shl*, *Sha*, *Shd*, *Slw*, *Fsr*, *Pcs*, *Ncs*, *Pcl*, *Ncl*, *Kcs* and *Kcl* and a significant negative correlation between *Pew* and *Rec* (Table 6). In the process of pericarp yield formation, appropriate N, P and K ratios and fertiliser doses are needed. Our results showed that *Tre12* can significantly promote leaf growth and N, P and K nutrient accumulation in shoots and leaves, reduce *Rec* and *Mda* content, and improve the pericarp yield of *Z. armatum* 'Hanyuan Putao Qingjiao'.

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