

## RESPONSES OF PHYTOHORMONES, CARBON AND NITROGEN STATUS TO THE TRUNK-EXTENSION PRUNING IN THREE-YEAR-OLD *PAULOWNIA* PLANTATION

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### ABSTRACT

Trunk-extension pruning is often used to promote sprouting of latent buds, and therefore extend the vertical growth of the main trunk in young *Paulownia* plantation in China. However, the biological basis for the technique remains unclear. In this study, we applied two pruning treatments and a control to 120 and 108 three-year-old *Paulownia* trees in 2015 and 2016, respectively. We observed the latent buds sprouting in 2015, and found that the number of individuals of first sprouting/total number of individuals and number of sprouted buds/total number of sprouted buds both exceeded 50% from 18 to 24 days after pruning. We collected phloem samples from the top of the stem at 7, 14, 21, and 28 d after the pruning in 2016. We then analyzed phytohormones, total C and N, reduced sugar and free amino acid contents of the samples. Results showed that pruning significantly reduced the contents of abscisic acid (ABA) and indole-3-acetic acid (IAA), especially on day 21 as compared with control. This was coincided with the sprouting of latent buds in the two pruned treatments. Therefore reduced IAA, ABA contents in pruned trees promoted the sprouting of latent buds. Our results provide a biological basis for extending the main trunk height.

**Keywords:** Trunk-extension pruning, Latent buds, Phytohormones, Carbon, Nitrogen, *Paulownia* plantation.

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### INTRODUCTION

*Paulownia* trees are widely cultivated for timber productions in China and several other Asian countries (Brain *et al.*, 1959; Lucas *et al.*, 2011; Smiley, 1961). As the terminal bud of *Paulownia* trees usually dies after leaf shedding in winter and the lateral bud near the top forms a pair of lateral branches, and the growth of the stem exhibits pseudo-dichotomous branching. This significantly inhibits its natural extension, which results in short main trunks, high taper, many knots, and low outturn percentage, therefore low timber yield and poor timber quality. This problem is particularly pronounced in arid and semiarid *Paulownia* cultivation regions in the Huanghuaihai Plain and northwestern China. As found in previous study, more than 80% of *Paulownia* trees have a knot-free main trunk height of only 2-3 m due to poor natural trunk extension, and the outturn percentage is only 40%. This greatly reduces the economic benefit of the *Paulownia* plantations (Wang, 2008).

We previously carried out pruning of lower branches and those at the top of the stem in triennial *Paulownia fortunei* and found that *Paulownia* latent buds sprouted as spindling branches (Wu *et al.*, 2014), resulting in an increase in trunk height of 5.7 m during that year. An 8-year continuous study of the initial main

trunk height, extended trunk height, diameter at breast height, and found that an appropriate intensity of branch pruning can significantly increase the main trunk volume and form factor (Wu *et al.*, 2014). It can be seen that sprouting of latent buds at the top of *Paulownia* stems is key to successful implementation of artificial assisted trunk extension. However, the mechanisms were still not elucidated.

Apical dominance means the inhibitory function of the terminal bud on the sprouting and growth of lateral buds (Cline, 1991). Polar transport of auxin is often used to explain apical dominance. Lateral buds will sprout after pruning of the terminal bud, but when auxin was smeared on the stem of which terminal bud was removed, the sprouting of lateral buds was inhibited by auxin (Leakey and Longman, 1986; Prasad *et al.*, 1993). Previous study showed that six hours after the terminal bud was removed, cytokinin levels in chickpea axillary buds increased 6-fold, and then increased 25-fold twenty-four hours later (Turnbull *et al.*, 1997). This indicates that cytokinins are of vital importance for the sprouting and growth of axillary buds. Hall found that cytokinins even can remove the inhibitory effects of auxins on axillary bud sprouting and growth (Hall, 1973). Buds will enter dormancy when ABA levels are increased, whereas they will exit dormancy when abscisic acid (ABA) levels are

decreased (During and Bachmann, 1975). Salomon found that ABA inhibited the sprouting of adventitious buds after the terminal bud was removed from *Citrus madurensis* seedlings (Salomon, 1976).

Jasmonic acid (JA) has similar physiological functions as ABA as it can also accelerate plant senescence and inhibit bud sprouting. When JA is present at the same time with ABA inside plants, these phytohormones have some synergistic effects (Li *et al.*, 2002). In addition, carbon and nitrogen may be essential to the sprouting of latent buds. Redistribution of carbon and nitrogen inside organs resulted from damage to aboveground organs can affect the sprouting of Chinese fir latent buds (Ye and Jiang, 1989). For instance, removing the terminal bud of *Vernicia fordii* resulted in changing the transport direction of carbon and nitrogen and the loss of apical dominance. This promoted the sprouting and branching of latent buds in the lower parts of the plant (Xie, 1987).

Therefore, we hypothesize that after trunk-extension pruning, changes may occur in phytohormones and the carbon and nitrogen status in the phloem at the top of stems, this will result in the sprouting of latent buds. In this study we observed the sprouting of latent buds and collected phloem samples from the top of the stem after two pruning treatments (low and high pruning intensity) and a Control (no pruning) being applied. Our objective is to investigate the changes in phytohormone and other chemicals contents in the phloem at the top of stems subjected to removal of apical bud and its influence on the sprouting of latent buds. It is expected that results of the research will provide some scientific bases for understanding the biological basis of trunk-extension pruning.

## MATERIALS AND METHODS

**Stress conditions:** The study was conducted at the state-owned Yuanyang Forestry Center in Henan Province, China (34°55.30'–34°56.45' N, 113°46.24'–113°47.59' E). The site is located on the Yellow River alluvial plain, with a continental monsoon climate, distinct four seasons within a year. The mean annual air temperature is 14.0 °C, and with highest temperatures of 29.8 °C in July and lowest temperatures of 0.1 °C in January. The mean annual rainfall is 571.7 mm. The winter and spring seasons are arid, and approximately 70% of the rainfall falling in July, August and September.

**Plant material and treatments:** In late March 2015, we applied three treatments to 120 three-year-old *Paulownia* trees. Then we observed the sprouting of latent buds everyday until almost latent buds sprouted.

The three treatments were as follows: T1: All bifurcation branches at the top and the lower canopy were pruned, only leaving 2-3 branches in the middle of the

canopy; T2: All branches were pruned. Control: Without pruning.

In late March 2016, we applied these three treatments to 108 three-year-old *Paulownia* trees. A hole-punch (diameter 1.5 cm) was used to collect phloem samples from the top of stems on 7, 14, 21, and 28 d after treatments. Three samples were collected from each individual, and samples from 3 individuals were pooled to form 1 sample. Each treatment was repeated in triplicate. After sampling, the samples were wrapped in tinfoil and placed in liquid nitrogen and then placed in a –80°C freezer in the laboratory for analysis.

**Measurement of phytohormones:** High-performance liquid chromatography-electrospray ionization-tandem mass spectrometry (HPLC-ESI-MS/MS) was used to determine the endogenous phytohormone concentration in the samples (Pan *et al.*, 2010; Sun *et al.*, 2017). Extraction and purification for phytohormones were established as below. Firstly, approximately 15 mg dry sample was transferred into a 2 mL centrifuge tube. Secondly, internal standards for the quantification were added to each centrifuge tube, i.e. 2 µL d5-IAA (2 ng/µL) for IAA, 20 µL d6-ABA (0.25 ng/µL) for ABA, 2 µL H2-JA (2 ng/µL) for JA, 20 µL d5-ZT (0.25 ng/µL) for ZT, and 50 µL d2-GA<sub>3</sub> (2 ng/µL) GA<sub>3</sub>. Thirdly, 0.5 mL of the extraction solvent (concentrated HCl : H<sub>2</sub>O : isopropanol = 0.002 : 2 : 1, vol/vol/vol) was added to each tube and put on a shaker with 100 r.p.m for 30 min, and this step should be established at 4 °C. Then, 1 mL of dichloromethane was added to each sample and the mixture was shaken as above. And then, clear phase separation of the mixed solutions was obtained after centrifugation at 1200 r.p.m for 5 min in a refrigerated microcentrifuge. Using a nitrogen evaporator with nitrogen flow, the solvent from the lower phase was collected and concentrated. Lastly, the residues were dissolved in 0.1 mL of methanol solvent (H<sub>2</sub>O : methanol = 1:1, vol/vol) again and centrifuged for 5 min at 1200 r.p.m. After layering, the supernatant were collected for HPLC-ESI-MS/MS analysis.

**Measurement of carbon and nitrogen:** Modified ninhydrin colorimetry colorimetry was used to determine the free amino acid content in plants (Lee and Takahashi, 1966). Fehling colorimetric method was used to determine the reducing sugars content (Scales, 1915). Total carbon and nitrogen content of the samples were determined using the Carlo Erba CHN 1500 elemental analyser (Lamlon and Savidge, 2003).

**Statistical analysis:** The phytohormone, total carbon and nitrogen content in different treatments during different development stages were analyzed. If significant differences among treatments or development stages were tested, Duncan's multiple comparison test was applied subsequently. Dimensionality reduction and classification

were conducted using principal component analysis (Sun *et al.*, 2017).

## RESULTS

**Effect of pruning on lateral bud sprouting:** We found that trees in the Control did not exhibit any bud sprouting while almost all latent buds sprouted in trees in the treatments T1 and T2 during 4–31 d after pruning (Table 1). This period can be uniformly divided into 4 stages: 4–10 d, 11–17 d, 18–24 d, and 25–31 d. Among these stages, the number of individuals of first sprouting/total number of individuals and number of sprouted buds/total number of sprouted buds both exceeded 50% in stage 3, followed by stage 2, where these two variables were approximately 25%.

**Indole-3-Acetic Acid:** For the Control, the IAA content in the phloem at the top of stems increased from 43.98 ng/g to 95.70 ng/g during the first three stages after pruning, then decreased to 50.62 ng/g (Fig. 1A). As compared with the Control, the IAA contents in treatments T1 and T2 remained less than 26 ng/g. The IAA content between treatments T1 and T2 at each of the four stages showed no significant difference.

**Abscisic Acid:** For the Control, the ABA content in the phloem at the top of stems increased from 112.99 ng/g to 433.53 ng/g during the first three stages after pruning, then decreased to 215.29 ng/g (Fig. 1B). The ABA content in treatments T1 and T2 remained less than 127.74 ng/g, as compared with Control. The ABA content between treatments T1 and T2 at each of the four stages showed no significant difference.

**Jasmonic Acid:** The JA content in treatments T1 and T2 decreased in the first three stages, and then increased slightly in stage 4. No significant difference was found in the JA content between treatments T2 and Control in stage 1, stage 2 and stage 4. In stage 3, the JA content in treatments T2 and T1 were significantly less than Control. The JA content in treatment T1 was significantly higher than treatment T2 in stage 1, stage 2 and stage 3. The JA content between treatments T1 and T2 in stage 4 showed no significant difference (Fig. 1C).

**Zeatin:** The highest ZT content of Control occurred in stage 3, i.e., 8.48 ng/g (Fig. 1D). Conversely, the ZT content in the treatments T1 and T2 exhibited an initial decrease followed by an increase, with the lowest content in stage 3, i.e., 2.93 ng/g and 2.01 ng/g, respectively, which was approximately 1/3 and 1/4 that of the Control. The ZT content among treatments T1, T2 and Control at each of the four stages showed no significant difference.

**Gibberellin 3:** The GA<sub>3</sub> content of Control decreased from 7.54 ng/g to 6.53 ng/g in the first two stages, then increased to 6.85 ng/g in stage 3, and then decreased to

6.31 ng/g (Fig. 1E). The GA<sub>3</sub> content among treatments T1, T2 and Control showed no significant difference from stage 2 to stage 4. The GA<sub>3</sub> content in treatments T1 and T2 were significantly less than Control in stage 1.

**ABA/ZT ratio and ZT/IAA ratio:** ABA/ZT ratio and ZT/IAA ratio can be used as another two important indicators of apical dominance change after pruning. For the control, the ABA/ZT ratio showed a gradually increasing trend, which increased from 33.32 to 59.83 in the 4 stages (Fig. 2A). The ABA/ZT ratio in treatment T1 remained quite stable at about 7.48 in the first two stages, then increased to 45.91 ng/g in stage 3, and then decreased to 21.87 in stage 4. The ABA/ZT ratio in treatment T2 increased from 3.17 to 16.30 in the first 3 stages, then decreased to 10.88 in stage 4. The ABA/ZT ratio in these two pruning treatments were significantly less than the Control in stage 1, stage 2 and stage 4. The ABA/ZT ratio between treatments T1 and Control showed no significant difference in stage 3, but they were all significantly higher than Treatment T2.

The ZT/IAA ratio in treatments T1 and T2 was significantly higher than the Control in stage 1 and stage 2 (Fig. 2B) and showed no significant difference between treatments T2 and Control in stage 3, however the ratio in treatment T1 was significantly higher than Control. In stage 4, it turned to be no significant difference in the ZT/IAA ratio between treatment T1 and Control, and the ratio in treatment T2 was significantly higher than treatment Control.

**Total N:** The total nitrogen content in treatment T1 decreased from 0.42% to 0.39% in the first two stages, then increased to 0.41% in stage 3, and then decreased to 0.38% in stage 4. The total nitrogen content in treatment T2 remained quite stable in the four stages at about 0.36%–0.38%. The total nitrogen content among treatments T1, T2 and Control showed no significant difference in the first two stages. No significant difference was also found in the total nitrogen content between Control and treatment T2, and between treatments T1 and T2. However, the total nitrogen content in treatment T1 was significant higher than Control. (Fig. 3A).

**Reducing Sugars:** For the treatment T1, the reducing sugars content decreased from 4.30% to 1.62% during the stage 1 and stage 2, then increased to 2.31% in stage 3, and then decreased to 1.64%. For the treatment T2, the reducing sugars content decreased from 3.31% to 1.17% during the four stages. No significant difference was found among these three treatments in each of the four stages (Fig. 3B).

**Total C:** For the treatment T1, the total C content decreased from 34.12% to 28.79% during the first three stages, then increased to 29.68% in stage 4. For the treatment T2, the reducing sugars content increased from

30.98% to 35.84% during the stage 1 and stage 2, then decreased to 28.08% during the last two stages. The total C content among these three treatments showed no significant difference in stage 1, stage 2 and stage 4. In stage 3, no significant difference was found in the total C content between treatments T1 and T2, but they were all significantly less than Control (Fig. 3C).

**Free Amino Acids:** The free amino acids content in treatment T1 decreased from 0.36% to 0.21% in the first two stages, then increased to 0.31% in stage 3, and then decreased to 0.19% in stage 4. The free amino acid content in treatment T2 decreased from 0.41% to 0.28% in the first two stages, then increased to 0.34% in stage 3, and then decreased to 0.25% in stage 4. The free amino acid content between treatments T1 and T2 showed no significant difference in stage 1, but they were all significantly higher than Control. In stage 2 and stage 3, no significant differences was found in the free amino acid content among these three treatments. In stage 4, no significant difference was found in the free amino acid content between Control and treatment T1 showed no significant difference, but they were all significantly less than treatment T2 (Fig. 3D).

**C/N ratio:** The C/N ratio in treatment T1 remained quite stable at about 82 in stage 1 and stage 2, then decreased to 70.44 in stage 3, and then increase to 78.49 in stage 4. The C/N ratio in treatment T2 increased from 86.75 to 97.99 in the first two stages, then decreased to 73.89 in stage 4. There was no significant differences in the ratio among these three treatments in the first two stages. Then the ratio in the treatments T1 and T2 turned to be significant less than the Control in stage 3 and stage 4 (Fig. 3E).

**Principal Component Analysis:** To identify the different patterns of the responses of different chemicals to pruning, we used principal component analysis (Table 2).

In principal component analysis, the characteristic root and contribution rate are the basis for selecting principal components. Among the five phytohormone indicators and 4 nutrient indicators, we identified the first two most important components that can explain about 37% and 26% of total variance, respectively. Contributions of the other principal components were much smaller (<14%). Therefore, we selected these two most important components for further analysis.

The first most important principal component is dominated by IAA, ABA, total nitrogen and free amino acids, while the second most important principal component 2 is dominated by JA, ZT, total carbon and reducing sugars (see Table 3).

In the space of the two most significant components (see Fig. 4), an increase in vertical direction means that increasing positive influences of IAA and the ABA content, and negative influences of free amino acids and the total nitrogen content. An increase in the horizontal axis means that increasing influences of JA, ZT, reducing sugars, and the total carbon contents. Cluster analysis grouped 3 treatments and 4 times of samples into 5 groups: groups I and II are the samples in the Control collected on days 21 and 28, with higher IAA and ABA contents and a lower total nitrogen and free amino acid contents. Groups IV and IV were day 7 samples that were collected from Control and treatment T1, respectively, with higher JA, total carbon and reducing sugars contents. All the remaining samples are in Group III, with a low IAA and ABA contents, relatively low JA, ZT, total carbon and reducing sugars contents, and a high total nitrogen and free amino acid contents. Most samples from different treatment types at different times at which the latent bud sprouted successfully were almost all clustered in this group, except for the samples taken on day 14 from Control.

**Table 1 Latent bud sprouting status after branch pruning in different treatments\*.**

	4–10 d		11–17 d		18–24 d		25–31 d	
	Number of individuals of first sprouting/total number of individuals	Number of sprouted buds/total number of sprouted buds	Number of individuals of first sprouting/total number of individuals	Number of sprouted buds/total number of sprouted buds	Number of individuals of first sprouting/total number of individuals	Number of sprouted buds/total number of sprouted buds	Number of individuals of first sprouting/total number of individuals	Number of sprouted buds/total number of sprouted buds
T1	10.00%	10.76%	25.83%	24.30%	54.17%	53.43%	10.00%	11.49%
T2	6.50%	7.79%	21.00%	23.14%	58.33%	55.94%	14.17%	13.12%
Control	0	-	0	--	0	--	0	--

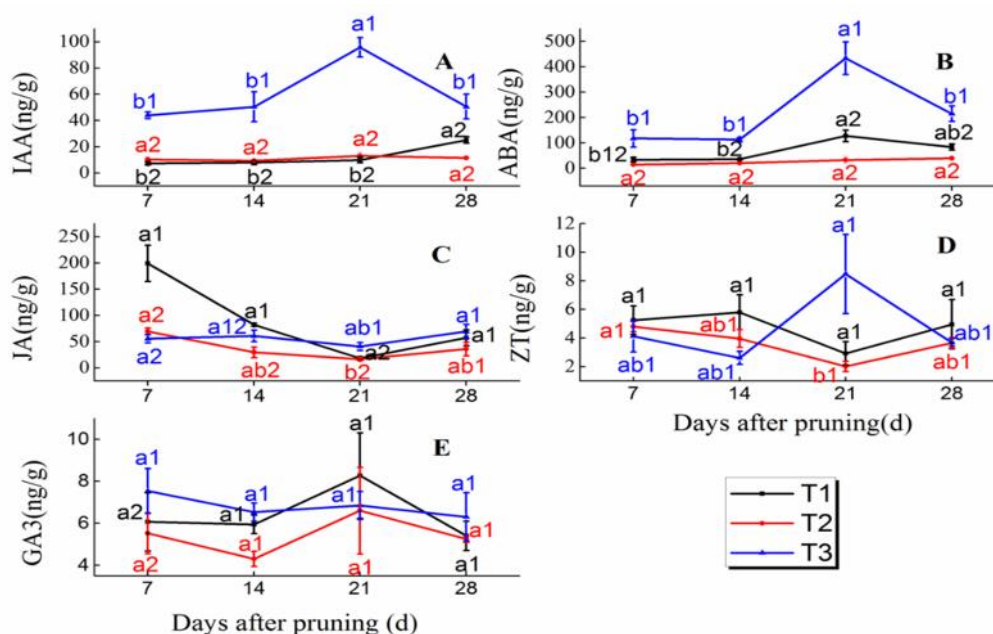
\*T1: All bifurcation branches at the top and the lower canopy were pruned, only leaving 2-3 branches in the middle of the canopy; T2: All branches were pruned. Control: Without pruning. The same below.

**Table 2. Total variance explained.**

Component	Initial Eigenvalues			Extraction Sums of Squared loadings			Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	3.370	37.440	37.440	3.370	37.440	37.440	3.363	37.364	37.364
2	2.304	25.597	63.037	2.304	25.597	63.037	2.311	25.673	63.037
3	1.249	13.875	76.912						
4	.773	8.588	85.500						
5	.558	6.205	91.705						
6	.450	5.002	96.708						
7	.170	1.887	98.594						
8	.093	1.037	99.632						
9	.033	.368	100.000						

**Table 3. Rotated component matrix.**

	Component	
	1	2
IAA	0.929	0.120
ABA	0.926	0.104
JA	-0.247	0.775
ZT	0.491	0.564
GA3	0.284	0.050
Total N	-0.759	0.192
Reducing sugars	-0.192	0.891
Free amino acid	-0.715	0.226
Total C	0.367	0.694

**Fig.1. Phytohormones content at the top of *Paulownia* stems under different treatments and at different development stages.**

**Note:** Different letters on the error bars indicate significant differences within a treatment at different time periods at a significance level of 0.05. Different numbers represent significant differences among treatments within the same stage at a significance level of 0.05. The same applies for the following figures.

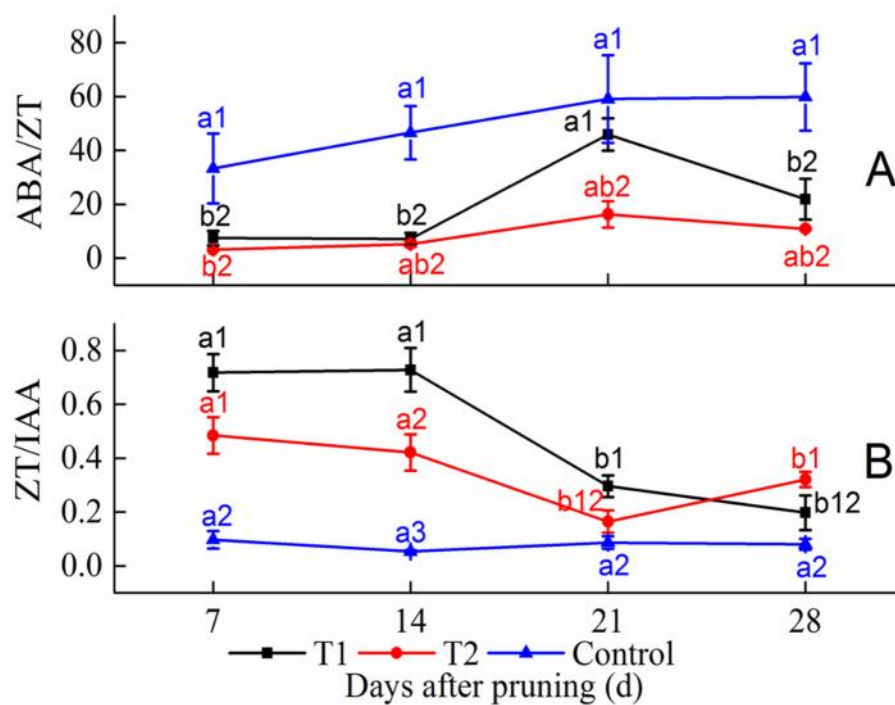


Fig. 2. ABA/ZT, and ZT/IAA ratios in the top of *Paulownia* stems under different treatments and at different development stages.

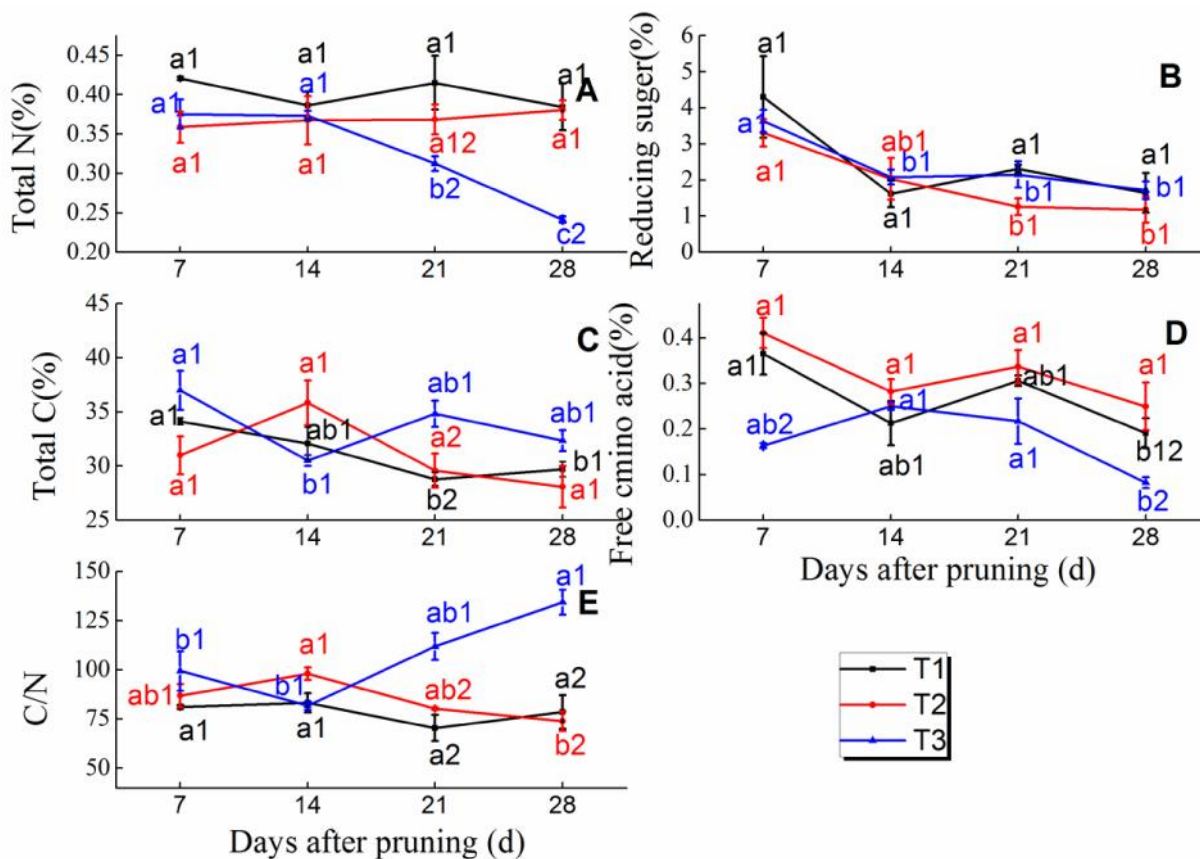


Fig. 3. Carbon and nitrogen content and C/N at the top of *Paulownia* stems under different treatments and at different development stages.



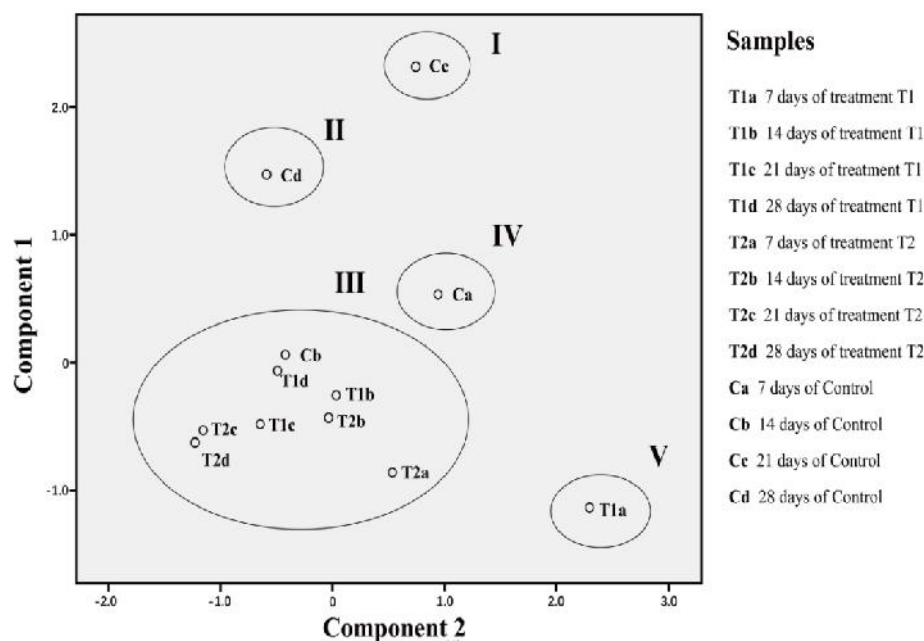


Fig. 4. Scatter plot of the phytohormones carbon and nitrogen nutritional content in the phloem at the top of *Paulownia* stems under different treatments and at development stages.

## DISCUSSION

Phytohormones are chemical messengers that can affect plant morphogenesis (Han *et al.*, 2018). Apical dominance is the primary mechanism that regulates plant sprouting, which is mainly influenced by the levels of auxin and cytokinins (Bangerth, 1994), the ratio of auxin to cytokinin (Shimizu and Mori, 2001; Muradoglu *et al.*, 2009), or the nutritional status of the axillary bud (Cline, 2000). Polar transport of auxin has significant regulating effects on the physiological processes of axillary bud development, and is the basis for the maintenance of apical dominance (Bangerth, 1994). Previous studies showed that IAA levels in the axillary buds rapidly declined, lateral buds sprouted after the removal of the terminal bud (Tsiantis *et al.*, 1999; Prasad *et al.*, 1993). This is consistent with our results in this study. Furthermore we found that the reduced IAA content was coincided with the high sprouting rate of lateral buds in the pruned treatments over the study period. Therefore we conclude that reducing IAA content after pruning stimulated the sprouting and growth of latent buds in *Paulownia* trees in this study. Previous studies found that cytokinin levels increased after the removal of terminal buds (Bergmann, 2003; Turnbull, *et al.*, 1997). This is consistent with our result. We found that the content of ZT, one of cytokinins in plants, in the phloem at the top of stems increased after pruning.

ABA can promote senescence in many plant tissues (Hunter *et al.*, 2004), and ABA contents are intimately associated with axillary bud dormancy. An example is the *eral* mutant that has high sensitivity to

ABA, and a high ABA content can inhibit axillary bud sprouting (Pei *et al.*, 1998). Our study found that the ABA content was significantly reduced by pruning, therefore the sprouting and growth of lateral buds was less suppressed than the Control. Gibberellin treatment can enhance apical dominance (Brain *et al.*, 1959). Response of GA<sub>3</sub> content to pruning was less consistent throughout the four phases. Pruning resulted in significantly lower content of GA<sub>3</sub> than the Control only in stage 1, and that difference became insignificant in the subsequent three stages. We do not yet know the cause for this variable response of GA<sub>3</sub> to pruning over time.

Different plant hormones often work synergistically to regulate plant growth and development at the organ level (Gaspar *et al.*, 1996). Emery *et al.* (1998) found that the growth rate of axillary buds was not significantly correlated with the absolute levels of cytokinins and IAA but was significantly correlated with the ratio of these two hormones. Growth of axillary buds is promoted at high ratio of cytokinin to IAA. Our study found that pruning significantly increased ZT/IAA ratio, therefore the sprouting and growth of latent buds in *Paulownia* trees. Furthermore ZT/IAA ratio in the low pruning intensity treatment (T1) was higher than the high pruning intensity treatment (T2) in the first two stages. Therefore low intensity pruning resulted in earlier sprouting of lateral buds than high-intensity pruning in the young *Paulownia* plantation. This result is consistent with previous observations in the field.

Previous study found that ABA content and bud growth rate were not related in the early bud development, but were negative correlated in the later

stages (Shimizu and Mori, 2001). Results from this study showed that pruning (both low and high intensities) significantly lowered ABA/ZT ratios (except on day 21) in all four stages of lateral bud development, as compared to the Control. Therefore balance of ABA and ZT played an important regulatory role in the growth and development of axillary buds (Hung *et al.*, 2002).

Fife and Nambiar (1984) found that the nitrogen content in the axillary buds increased, which benefited sprouting and growth of lateral buds after the removal of terminal buds. This is consistent with the results of our study. Therefore pruning resulted in the redistribution of nitrogen within the plant and benefited the sprouting of lateral buds and this stimulatory effect may be related to the increased cytokinin content that promoted bud sprouting, as cytokinin contains large amount of nitrogen (Rubinstein and Nagao, 1976).

Our study also found that the treatments and time points at which the latent buds sprouted can almost be grouped together. In addition, these treatments exhibited common patterns, i.e., the maintenance of low levels of IAA and ABA, relatively low levels of JA, ZT, reducing sugars and total carbon, and high levels of total nitrogen and free amino acids. When considering the applying phytohormones, carbon and nitrogen to regulate latent bud sprouting of *Paulownia* trees, we should not only consider the synergistic effect between phytohormones but also these synergistic effects between phytohormones, carbon and nitrogen.

**Conclusions:** This study indicated that the number of individuals of first sprouting/total number of individuals and number of sprouted buds/total number of sprouted buds both exceeded 50% from 18 to 24 days after pruning by removing the terminal buds. Pruning treatments significantly reduced IAA and ABA contents in the phloem at the top of the stems. Seven days after pruning, the IAA content in the two pruned treatments was less than 1/4 and ABA content was less than 1/3 that of the Control, and were maintained throughout the remaining three phases. Pruning also significantly increased ZT/IAA and reduced ABA/ZT and C/N. These reduced contents, ABA/ZT and C/N, increased ZT/IAA resulted in a high sprouting rate for latent buds. In addition, low contents of JA, ZT, reducing sugars and total carbon, and high contents of total nitrogen and free amino acids may be beneficial for the sprouting of latent buds. Our results provide a biological basis for trunk extension that is commonly used for achieving higher and better timber production from *Paulownia* trees.

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