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Effect of pruning intensity and doses of fertilization on content of macronutrients and hormones in leaves of rewatered citrus trees

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Abstract. There are some changes in the rewatered citrus trees, among others are the content of nutrients and hormones. An experiment was conducted to study the effects of pruning and fertilization on the macronutrients and hormones contents in leaves of rewatered citrus trees. The research was conducted in a citrus orchard in Purbalingga, Central Java, Indonesia from June until October 2018. There were two observed factors, namely pruning intensity (0, 5, 10, and 15% of total number of branches each tree) and doses of fertilization (0, 2, and 4 % of weight of harvested fruits at previous season). The research used Randomized Completely Block Design and three replications. Observed variables were content of N, P, K, IAA, GA₃, and C/N ratio. The result of research showed that pruning intensity of 5% gave the highest content of K and IAA, and highest C/N ratio but the lowest content of GA₃. Doses of 2 % fertilization gave the highest content of IAA and the highest C/N ratio but the lowest content of GA₃. High content of K and IAA, and high C/N ratio but lower content of GA₃ were required to induce the flowering of citrus trees.

1. Introduction

Citrus is a very familiar fruit as a good source of vitamin C. As an antioxidant, vitamin C can reduce the risk of cardiovascular and cancer diseases. Vitamin C has an important function in enhancement of the immune system, reduction of cholesterol level, and in formation the of collagen [1]. Citrus contain carotenoids and some secondary metabolite [2]. These beneficial traits imply on the increase of the citrus fruit demand

As other fruit crops in the tropical region, citrus trees need to be exposed to the condition of water stress in a number times to induce flowering [2]. After that, rewatering must be applied to citrus trees to continue the induction of flowering. A few days after rewatering, citrus trees usually started to bear flower. The transition period between rewatering and flowering is an interesting topic to be studied [3]. At that period, there are some changes in the rewatered citrus trees; among others are the content of nutrients and hormones due to the application of treatments like different pruning intensity and doses of fertilization. The content of nutrients and hormones in trees determine the process of flowering of citrus trees.

Pruning the branch aimed to maximize the utilization of light by plant so it induces more flower and promote fruit development by the increasing of carbohydrate synthesis [4]. Pruning the young shoot,



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twigs and leaves that were stricken with diseases gave the highest number of fruits per tree [5]. Application of the appropriate dose of fertilization has been reported to enhance the availability of nutrient in the soil [6]. The improvement of nutrient availability in the soil lead to increase nutrient absorption by plant. Many references reported that content of nutrients in the plant especially in leaves determine the flowering. Vemmos said that C/N influence flowering and fruiting [7]. A low concentration of carbohydrate reduced flower number in *Lantana camara* [8]. The objective of the research is to study the effect of pruning intensity and doses of fertilization on the content of macronutrients and hormones in leaves of rewatered citrus trees.

2. Material and methods

The research used five-years old citrus trees of *Citrus nobilis*. It was carried out from June until October 2018 in an orchard of citrus belongs to farmer located in Purbalingga, Central Java, Indonesia. This orchard is at 60 m above sea level, 7.44°S and 109.43°E. The soil of site research has 0.134% N, 0.082% P, and 0.058% K. There are two seasons, namely dry season which takes place from April until October, whereas rainy season is from October until April. By observing local meteorological station, the average sunshine was seven hours per day, average rainfall was 146 mm per month, and average air temperature was 22-36°C.

This factorial experiment was arranged in Randomized Completely Block Design. There were two factors namely pruning intensity and doses of fertilization. The pruning intensity were 0, 5, 10, and 15% of total number of branches each tree; whereas doses of fertilization were 0, 2, and 4% of weight of harvested fruits at previous season. Each treatment was replicated three times, so there were 36 citrus trees. All of the trees were applied the same cultivation technique.

The observed variables were content N, P, K, C, IAA, and GA₃ in leaves of rewatered citrus trees. The citrus mature leaves as sample were taken at two days after application of rewatering. For determination the content of N, P, K, and C, the leaves were dried by oven at 60°C for 48 h; whereas to determine the content of IAA and GA₃, the leaves were dried by freeze drier. Analysis of N content was conducted by the Kjeldahl method [9]. Content of P was determined by a UV mini-1240 UV-Vis spectrophotometer at $\lambda = 430$ nm (Shimadzu, Kyoto, Japan), K content with a Polarized Zeeman Automatic Absorption spectrophotometer at $\lambda = 768$ nm (Hitachi, Tokyo, Japan). C content was determined by the Walkey & Black method with a UV mini-1240 UV-Vis spectrophotometer at $\lambda = 560$ nm [10]. Content of IAA was determined by HPLC method [11]. The HPLC separations on the ODS-reverse phase column and the SIL-absorption column were made with a Du Pont model 841 high performance liquid chromatograph, which utilized a miniature high pressure pneumatic amplifier pump and either the standard 254 nm UV photo detector and/or the Du Pont model 836 fluorescent detector. Content of GA₃ was determined by HPLC method, used stationary phase of C18, liquid phase of methanol acetate acid, and detector by λ 225 nm. Data were analyzed using analysis of variance (ANOVA) with the statistical program SAS version 9. Following ANOVA, means were separated by Duncan's Multiple Range Test at $p=0.05$.

3. Results and discussion

3.1. Content of some macronutrients in leaves

Table 1 showed that content of K in leaves of rewatered citrus trees was influenced by pruning intensity. Pruning intensity of 5 % gave the highest content of K (1.44%). The optimum content of K in fruit crops promotes to get the high yield [6]. Content of N, P, and C was not influenced by pruning intensity. This result is similar with a claim made by Sakhidin *et al* that the branches bearing different number of fruits per panicle had the same N, P, and C content in the bark [12].

It was suggested that to increase citrus yield, it is necessary to consider in reducing N and P fertilizer input and appropriately increase the proportion of K [13]. K has important role in improving the flowering, fruit set and yield in 'Magallanes' pummelo [14]. K involves the activation of at least 60

different enzymes needed for metabolic processes and catalytic functions. Application of K fertilizer is an efficient way to increase 'Kousui' Japanese pear fruit yield [15]

Table 1. Effect of pruning intensity and doses of fertilization on content of some macronutrients (%)

Treatments	Macronutrients			
	N	P	K	C
Pruning intensity (%)				
0	3.13	0.21	1.36 a	36.06
5	2.99	0.16	1.44 b	37.67
10	2.95	0.09	1.19 a	36.23
15	3.28	0.11	1.20 a	36.25
<i>F value</i>	2.36 ^{ns}	1.00 ^{ns}	3.73*	1.47 ^{ns}
Doses of fertilization (%)				
0	3.08	0.23	1.33	36.93
2	3.02	0.15	1.27	36.39
4	3.17	0.18	1.29	36.34
<i>F value</i>	0.81 ^{ns}	0.46 ^{ns}	0.30 ^{ns}	2.50 ^{ns}

Note : * means are significant at $p = 0.05$; ns = non-significant

K is mobile in plant so this nutrient has important role in synthesis of proteins and carbohydrates, transport of sugars, water and nutrients from the source to the sink. So, the availability of K in the fruits and leaves must be sufficient to promote fruit production [14]

3.2. Content of IAA, GA₃ in leaves and C/N ratio

Content of IAA was influenced by pruning intensity, the highest content of IAA was showed by pruning intensity of 5%. The same pruning intensity also gave the highest C/N ratio but the lowest content of GA₃. Flowering of citrus need the higher content of IAA and C/N ratio but the lower content of GA₃ [16]. It is related with high number of flowers by pruning intensity of 5%. Gibberellin treatment is a common agricultural practice to inhibit flowering in citrus trees. IAA as an auxin promote cell enlargement, endogenous auxins usually increase in developing ovaries [2].

Table 2. Effect of pruning intensity and doses of fertilization on the content of some hormones (%) dan C/N ratio

Treatments	Hormones		C/N ratio
	IAA	GA ₃	
Pruning intensity (%)			
0	0.032 a	0.031 b	11.66 a
5	0.050 b	0.023 a	12.60 b
10	0.047 b	0.027 a	12.29 b
15	0.046 b	0.031 b	11.17 a
<i>F value</i>	49.98**	21.81**	5.43*
Doses of fertilization			
0	0.041 a	0.035 b	12.01
2	0.046 b	0.024 a	12.10
4	0.045 b	0.026 a	11.68
<i>F value</i>	5.34*	59.80**	0.86 ^{ns}

Note: ** means are significant at $p = 0.01$; * means are significant at $p = 0.05$; ns = non-significant

Sakhidin *et al* reported that the higher C/N ratio in branch bark promote the higher number of fruits per panicle of durian [12]. A high C/N ratio was required for floral initiation [17] and differentiation [18] in mango. Flowering requires a large supply of carbohydrate as source of energy [19].

The importance of high C/N for fruiting reported by Thamrin *et al*, increasing C/N ratio by bark strangulation increased fruit set of pummelo (*Citrus grandis* (L) Osbeck) [20]. Carbohydrates serve as

substrates for the synthesis of key metabolites that act alone or work with plant hormones to stimulate the flowering [21].

High C/N or carbohydrates level in leaves supports the flowering processes. However, some references stated that starch concentration may be as limiting factor in flower formation. The effect of lower carbohydrates levels not only in decreasing the number of formed flowers but also in reducing the number of new shoots at next season [16].

4. Conclusion

Pruning intensity of 5% gave the highest content of K and IAA, and highest C/N ratio but the lowest content of GA₃. Doses of fertilization of 2 % gave the highest content of IAA and the highest C/N ratio but the lowest content of GA₃. High content of K and IAA, and high C/N ratio but lower content of GA₃ were required to induce the flowering of citrus trees.

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