

## Growth regulators and carving on breakage of apical dominance in tannia [*Xanthosoma sagittifolium* (L.) Schott] rhizomes

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### Abstract

Specific clones of tannia are commonly used as cooking leaves in some Brazilian states. The crop is propagated exclusively asexually by planting the rhizomes usually after a few years of production. Because of that, there is the need to establish strategies to propagate healthy plantlets with higher sprouting rate for leaf production. This work had the goals to evaluate the influence of growth regulators and carving of the rhizomes sprouting and growth of tannia. Cured rhizomes from the clone 'Caipira' were stored for three months at 5°C and 89% relative humidity. Afterward, the top of half of the rhizomes were carved in a V shape at the top (carving), to stimulate lateral sprouting. The rhizomes were submerged for 30 minutes in solutions containing 6-benzylaminopurine (BAP) and/or 2-chloroethylphosphonic acid (ethephon), and the respective control. The production of new leaves and expansion of leaf area were stimulated by treating the rhizomes with 500 mg L<sup>-1</sup> BAP and 250 mg L<sup>-1</sup> BAP + 250 mg L<sup>-1</sup> ethephon. Regardless the use of growth regulators, the carving induced higher number of new sprouted leaves after 35 days of planting. Similarly, rhizomes treated with 500 mg L<sup>-1</sup> BAP or 250 mg L<sup>-1</sup> BAP + 250 mg L<sup>-1</sup> ethephon had higher number of sprouts after 49 days of planting. Sprouting was anticipated when the carved rhizomes were treated with 250 mg L<sup>-1</sup> BAP + 250 mg L<sup>-1</sup> ethephon.

**Keywords:** *Xanthosoma sagittifolium*, Carving, Ethephon, 6-benzylaminopurine, Sprouting.

**Abbreviations:** ANOVA\_analysis of variance, BAP\_ 6-benzylaminopurine, Ethephon 2-chloroethylphosphonic acid, N:P:K\_ nitrogen:phosphorus:potassium.

### Introduction

The species *Xanthosoma sagittifolium* (L.) Schott, is a popular leaf vegetable consumed by many Brazilians. The rhizomes also can be used as source of starch (Rubatzky and Yamaguchi, 1997). The plant belonging to the family *araceae* is known by the name of 'taioba' being cultivated in tropical and subtropical climate regions of Brazil (Souza et al., 2014). The propagation of tannia is vegetative by using small rhizomes, planted after the cold season is over (Carvalho and Cordeiro, 1990; Filgueira, 2008). But, little is known about the physiology and effect of handling in the rate of sprouting. The lack of propagation organs is a major problem that has hindered the deployment of vegetative propagated crops (Carvalho, 1991; Fogaça et al., 2007). Alternative methods of rapid propagation can solve the problem of this shortage and reduce the time and cost of production. Cytokinins are growth regulators that are active during the process of cell division, mobilization of nutrients, apical dominance and dormancy breakage (Coleman et al., 2001; Garcia et al., 2006; García-Flórez et al., 2009). Among these, control of cell division is of considerable significance in the growth and development of the plant. Cytokinins have an active function on the growth of lateral buds, reducing the apical dominance, and increasing the rate of sprouting (Erig and Schuch, 2006; Vieira et al., 2009). According to García-Flórez et al. (2009) the development of lateral buds are inhibited by the increased concentration of IAA (3-indolyl-acetic acid) on the apical meristem, to act as a drain of

nutrients and cytokinins for the apical bud. Moreover, the high level of auxin in the apical buds aids in maintaining high levels of ABA (abscisic acid) in lateral buds, inhibiting their growth (Taiz and Zeiger, 2002). Thus, removal of the apical bud promote the increase of cytokinins in lateral buds, favoring the development of these. Among the most commonly used cytokinins is 6-benzylaminopurine (BAP) (García-Flórez et al., 2009). The role of 6-benzylaminopurine on the sprouting of tannia rhizomes is not known yet. In addition to that, there is no information available on if by removing the apical dominant bud promotes an increase in the cytokinins in the lateral buds, thereby stimulating their growth. The stimulation of cell division can also be caused by the application of ethylene, resulting in the formation of shoots and adventitious roots (Oliveira et al., 2001). Ethylene is one of the growth regulators commonly used in agriculture, due to the effects of many physiological processes. Due to the high diffusion rate (gas hormone), it becomes difficult to apply in plants, under field conditions (Suttle, 2009). However, this limitation can be overcome by the use of compounds that release ethylene (Coleman et al., 2001). The most widely used is ethephon (2-chloroethylphosphonic acid), commercially known as Ethrel. It is also possible that the exogenous ethylene to stimulate increased synthesis of gibberellins (growth promoter hormone), and may thus increase or decrease the sprouting tubers, depending on the concentration and duration of exposure (Suttle, 1998). In a

previous study on yam tuber of *Dioscorea cayennensis* the use of acetylene induced sprouting and formation of adventitious roots on the sprouting tubers (Oliveira et al., 2001). Thus, ethylene seems to be directly involved in the induction of sprouting and root differentiation. Nevertheless, the influence of ethylene on the sprouting of tannia rhizomes has not yet been determined. The goal of this work was to determine the effect of 6-benzylaminopurine (BAP) and 2-chloroethylphosphonic acid (ethephon), combined with the carving of the apical bud on the sprouting of tannia rhizomes.

## Results

### Growth regulators

Regardless of, if the rhizomes were or were not carved, the growth regulators BAP and ethephon improved most of the measured plant growth characteristics. Applying 500 mg L<sup>-1</sup> BAP or 250 mg L<sup>-1</sup> BAP + 250 mg L<sup>-1</sup> ethephon enhanced the production of leaves compared to control, increasing the number of leaves by 97% and 65.45%, respectively (Table 1). All the other combinations of BAP or ethephon had a similar effect of the number of leaves. Only the 500 mg L<sup>-1</sup> BAP + 500 mg L<sup>-1</sup> ethephon, had an effect on the total leaf area. The treatment of 500 mg L<sup>-1</sup> BAP + 500 mg L<sup>-1</sup> ethephon shortened the height of the plants compared to other plants treated with growth regulators (Table 1). Smaller fresh leaf matter production occurred when the rhizomes were treated with 500 mg L<sup>-1</sup> BAP + 500 mg L<sup>-1</sup> ethephon, but did not affect the total dry production of leaves.

### Carving

Carving the apical bud did not affect the total production of leaves per rhizome (Table 2). But, the addition of BAP and/or ethephon helped the growth of lateral buds (Table 3). The excision of the main bud reduced the final leaf area, length of the aerial part, total fresh and dry matter (Table 2). When the rhizomes were carved, a more uniform sprouting was obtained, which increased the number of sprouting leaves by 16% after 35 days from planting (data not shown). This result showed that the stimulus of lateral buds sprouting was affected by the presence of the main bud. Nevertheless, the total production of fresh and dry matter of leaves at 120 days was lower in the carved rhizomes (Table 2). Irrespective of if the rhizomes were or were not carved, the treatment with 500 mg L<sup>-1</sup> BAP or 250 mg L<sup>-1</sup> BAP + 250 mg L<sup>-1</sup> ethephon resulted in the higher number of new sprouting leaves after 49 days of planting (Table 3). These results show that uniform sprouting requires growth regulator, with the control rhizomes having the lowest sprouting among the treatments (Table 3). Carving was able to improve the final number of sprouted leaves after 120 days after planting (Fig 2).

### Discussion

The highest number of shoots from rhizomes submitted to removal of the apical meristem (carved) due to the strong control that this has on the lateral buds. The carving caused rapid resumption of cell division and the development of lateral meristems. Such control is exercised through some auxin, possibly the 3-indolyl-acetic acid (IAA), synthesized in the apical region and transported to the lateral meristem. Thus, the removal of the apical meristem afforded the increased availability of cytokinins in the lateral meristems (García-Flórez et al., 2009). Cytokinins are also crucial in promoting budding gems in propagative organs (García-Flórez et al., 2009). 6-benzylaminopurine acted on the

control of the hydrolysis of the rhizomes reserves being necessary for induction of  $\alpha$ -amylase to hydrolyze starch. According to Vieira et al. (2008) the development of amylase activity is an important event and can be detected during early germination, and its main function available substrates used for seedling until it becomes photosynthetically self-sufficient. These nutritional reserves, digested by the enzyme accumulated in the form of sugars, amino acids and nucleic acids are then transported to the lateral buds in the rhizomes. According to Ono et al. (2004), breaking the apical dominance can be promoted with synthetic cytokinins, but working with papaya (*Carica papaya* L.) found that cytokinin used isolated, with and without the removal of the apical meristem, does not increase the growth of side shoots. Coelho et al. (2009) studied the growth regulators effect on the propagation of pineapple 'Smooth Cayenne', found higher efficiency of treatment with 6-benzylaminopurine in a concentration of 400 mg L<sup>-1</sup> on the shoot buds. Souza and Finger (2014) reported that pre-treatment of the rhizomes with growth regulators BAP (250 mg L<sup>-1</sup>) + Ethrel (250 mg L<sup>-1</sup>) does not induce greater formation of lateral buds in tannia genotypes Comum, Roxa and Caxixe. The promoting effect of ethylene (Ethrel) is to increase the release and the movement of these hydrolytic enzymes and cause increased breathing and sugar content (Suttle, 2009).

## Materials and Methods

### Preparation and storage of rhizomes

Rhizomes from tannia (*Xanthosoma sagittifolium*) from clone 'Caipira', with medium size (approximately 25g), were harvested at Inhapim, MG, Brazil and transported to the Universidade Federal de Viçosa (UFV), Viçosa, MG. The rhizomes were allowed to cure for 20 days at room temperature in a shaded greenhouse and then, stored in cast boxes of polypropylene, with lid, for three months, at 5°C and 89% relative humidity.

### Treatments and experimental design

The experiment was conducted in a factorial 7 x 2, being composed by combining six concentrations of plant growth regulators, more control with water in rhizomes carved and uncarved in completely randomized design with 6 repetitions. Each experimental unit was composed by one plant. After three months the rhizomes were removed from the cold storage, in half of the rhizomes the apical bud was removed by carving in a "V" shape form (Fig 1). Afterwards, the carved and uncarved rhizomes were dipped for 30 minutes in solutions containing: (1) water (control treatment); (2) 250 mg L<sup>-1</sup> BAP; (3) 500 mg L<sup>-1</sup> BAP; (4) 250 mg L<sup>-1</sup> ethephon; (5) 500 mg L<sup>-1</sup> ethephon; (6) 250 mg L<sup>-1</sup> BAP + 250 mg L<sup>-1</sup> ethephon; (7) 500 mg L<sup>-1</sup> BAP + 500 mg L<sup>-1</sup> ethephon. The rhizomes were allowed to dry for three days over a bench at room temperature and subsequently planted into one liter pots. After three months the plantlets were transplanted to five liter pots filled with 75% soil, 25% organic matter and 30 g of commercial fertilizer N:P:K (4:14:8).

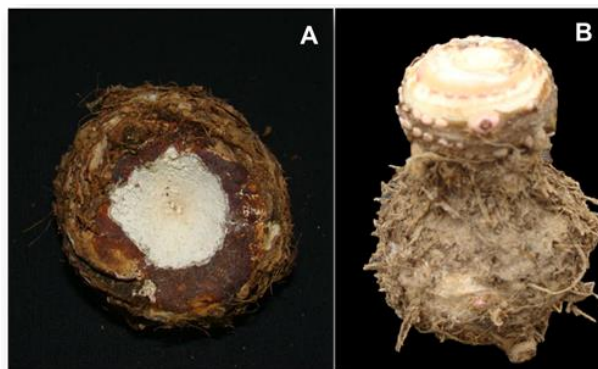
### Traits measured

The number of sprouting leaves was determined weekly and at 120 days after planting the number of leaves, length of aerial portion of the plant, leaf area, fresh and dry matter were also determined. The variable number of shoots, evaluations were made periodically every 7 days, from the time the rhizomes began to sprout. The shoot length was

**Table 1.** Average number of leaves (NL), leaf area (cm<sup>2</sup>) (LA), length of aerial portion of the plant (cm) (LL), fresh matter (g) (FM) and dry matter (g) (DM) of tannia ‘Caipira’, carved or uncarved, after 3 months storage at 5°C following application of different concentrations of growth regulators.

Treatments	Means									
	NL		LA		LL		FM		DM	
Control	6.5	b	435	a	48.7	ab	156.3	ab	9.4	a
250 mg L <sup>-1</sup> ethephon	9.8	ab	419	ab	53.1	a	206.3	ab	11.4	a
500 mg L <sup>-1</sup> ethephon	9.0	ab	440	a	53.5	a	203.5	ab	11.0	a
250 mg L <sup>-1</sup> BAP	8.7	ab	425	ab	50.8	a	187.1	ab	11.5	a
500 mg L <sup>-1</sup> BAP	12.0	a	433	ab	55.5	a	207.2	ab	12.6	a
250 mg L <sup>-1</sup> BAP + 250 mg L <sup>-1</sup> ethephon	10.7	a	438	a	53.5	a	221.8	a	12.6	a
500 mg L <sup>-1</sup> BAP + 500 mg L <sup>-1</sup> ethephon	9.2	ab	397	b	40.2	b	136.7	b	8.8	a

Mean followed by the same letter do not differ by Tukey test at P ≤ 0.05.

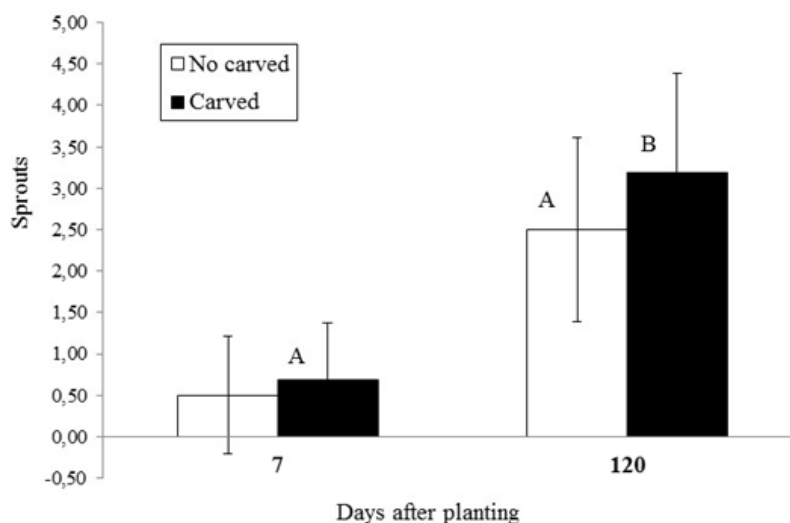


**Fig 1.** Rhizomes of tannia genotype ‘Caipira’ without apical meristem (carved) (A) and with apical meristem (uncarved) (B).

**Table 2.** Average number of leaves (NL), leaf area (cm<sup>2</sup>) (LA), length of aerial portion of the plant (cm) (LL), fresh matter (g) (FM) and dry matter (g) (DM) of tannia ‘Caipira’, carved or uncarved, after 3 months storage at 5°C following application of different concentrations of growth regulators.

Carving	Means									
	NL		LA		LL		FM		DM	
Carved	10.1	a	413	b	47.9	b	175.3	b	10.0	b
Uncarved	8.7	a	440	a	53.6	a	201.5	a	12.1	a

Mean followed by the same letter do not differ by Tukey test at P ≤ 0.05.



**Fig 2.** Number of sprouts of tannia ‘Caipira’ with apical meristem (uncarved) or without apical meristem (carved), after 3 months storage at 5°C. Mean followed by the same letter do not differ by Tukey test at P ≤ 0.05.

**Table 3.** Mean values of sprouting leaves (SL) of tannia ‘Caipira’, carved or uncarved, after 3 months storage at 5°C and application of different concentrations of growth regulators.

Treatment	SL
Control	1.6 <sup>g</sup>
250 mg L <sup>-1</sup> ethephon	2.3 <sup>c</sup>
500 mg L <sup>-1</sup> ethephon	1.9 <sup>e</sup>
250 mg L <sup>-1</sup> BAP	1.8 <sup>f</sup>
500 mg L <sup>-1</sup> BAP	2.3 <sup>b</sup>
250 mg L <sup>-1</sup> BAP + 250 mg L <sup>-1</sup> ethephon	2.4 <sup>a</sup>
500 mg L <sup>-1</sup> BAP + 500 mg L <sup>-1</sup> ethephon	2.0 <sup>d</sup>

Mean followed by the same letter do not differ by Tukey test at  $P \leq 0.05$ .

measured by the distance of the cutting point, just above the base of the petiole to the apex of the leaf blade. The leaf area was determined by measuring the length and width, wherein the length was obtained for the distance between the apex and leaf lamina and the insertion point of the petiole and the width was taken as the sum of the distances between the insertion of petiole and the ends of the two major side ribs (Chapman, 1964). The fresh weight of the aerial part was obtained by weighing on an analytical balance, accurate to three decimal places. The dry matter was determined by drying the plants in an oven with forced ventilation at 70 ° C for 72 hours, where it obtained a constant weight (Seganfredo et al., 2001).

#### Statistical analysis

Data were submitted to analysis of variance ANOVA and the means were compared by Tukey at  $P \leq 0.05$ , using the statistical program SAEG version 9.1 (2007).

#### Conclusions

The production of new leaves and leaf area expansion are stimulated by treatment of tannia rhizomes with 500 mg L<sup>-1</sup> BAP and 250 mg L<sup>-1</sup> BAP + 250 mg L<sup>-1</sup> of ethephon. Carving induces more new sprouting leaves, as determined at 35 days after planting. The treatment with 250 mg L<sup>-1</sup> BAP + 250 mg L<sup>-1</sup> of ethephon in carved rhizomes, provides optimal sprouting of lateral buds in this cultivar.

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