

## Collect Seasons, Indolebutyric Acid and Substrates on Rooting of ‘Bengal’ Lychee Cuttings

Ronan C. Colombo<sup>1</sup>, Adriane M. de Assis<sup>1</sup>, Vanessa Favetta<sup>1</sup>, Lilian Y. Yamamoto<sup>1</sup> & Sergio R. Roberto<sup>1</sup>

<sup>1</sup> Agronomy Department, State University of Londrina, Londrina, PR, Brazil

Correspondence: Ronan C. Colombo, Agronomy Department, State University of Londrina, Rodovia Celso Garcia Cid (PR 445), km 380, P.O. Box 10.011, Zip Code 86057-970 Londrina, PR, Brazil. Tel: 55-43-9836-0480. E-mail: ronancolombo@yahoo.com.br

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### Abstract

Lychee (*Litchi chinensis* Sonn.) propagation by cuttings is a promising alternative in commercial production of seedlings, since it is an easy procedure, in addition to enabling the standardization in the field and the reduction in production costs. Therefore the objective of this work was to evaluate ‘Bengal’ lychee cuttings rooting, collected in spring and summer, using different concentrations of indolebutyric acid (IBA) and substrates. The experimental design was completely randomized, in a  $3 \times 3$  factorial arrangement, with 5 replications and 10 cuttings per plot, and the following factors were adopted: IBA concentrations (0; 1,000 and 2,000 mg L<sup>-1</sup>) and types of substrates (carbonized rice husk, vermiculite medium granules and coconut fiber). At 140 days, the following variables were evaluated: cuttings survival; leaf maintenance; cuttings unrooted with calli; cuttings rooted; roots number per cutting; root length and root dry mass. In addition, the pH, electrical conductivity, density and water retention capacity of the substrates were evaluated. There was no difference in IBA concentrations applied in both evaluated periods. However, in the spring, the carbonized rice husk provided highest mean for the measured variables. On the other hand, in the summer, the rooting percentage presented a lower average in all substrates (< 1%), independent of the IBA concentrations. It was concluded that the ‘Bengal’ lychee propagation by cutting can be carried out in the spring, using carbonized rice husk as substrate, without IBA.

**Keywords:** *Litchi chinensis*, propagation, auxin

### 1. Introduction

The Lychee tree is a fruit species (*Litchi chinensis* Soon.) from China, very appreciated by flavor, peculiar savor and attractive appearance. In Brazil, the fruit is still relatively unknown, it is estimated that less than 10% of the population has already tried lychee, because the availability of the fruit in the market is small and restricted to a few months of the year. However, the potential to become a popular fruit is great (Yamanishi et al. 2010).

Although restricted to small areas, the production of this fruit has aroused the interest of several producers in Brazil, mainly due to the commercial value of the fruits. According to study of Carvalho et al. (2005), the difficulty in the propagation of lychee contributes to the existence of few producers of this fruit in our country, due to the low availability of seedlings.

Layering it is the most used method in the commercial propagation of lychee trees; however, it presents disadvantages such as the depletion of the mother plant when a large number of seedlings are obtained, besides the method being quite laborious, restricting and burdening the seedlings production (Martins et al., 2001, 2005; Lins et al., 2015).

An alternative to produce seedlings of this species is by cuttings, since in addition to allowing the characteristics of the mother plant to be maintained, it is easily executed and provides the greatest seedlings number. However, the main drawback of using this method is related to the difficulty of rooting presented by this species. On the other hand, the promotion of the rooting in cuttings can be stimulated by using growth regulators, which enables the seedlings production by cuttings (Leonel et al., 1995; Carvalho et al., 2005; Bastos et al., 2006; Smarsi et al., 2008; Koyama et al., 2014).

In addition, rooting can be stimulated by environmental conditions, such as the season of collection of cuttings and the substrate, and other elements (Fachinello et al., 2005; Tavares et al., 2012). The substrate, depending on its chemical characteristics, such as pH and electrical conductivity, as well as physical, such as porosity, can limit the availability of water and air in the basal area of the cuttings and interfere in cell metabolism or lead to tissue dehydration. Boechat et al. (2010) reported that a good substrate is that which provides optimum conditions for good root growth, such as porosity and water holding capacity.

Another determining factor it is the season of year which cuttings are collected, since it is related to the stage of the branch and to the level of physiological processes activity in the plants. This explains why some plants are propagated in the growing season and others during dormancy period (Fachinello et al., 2005; Hartmann et al., 2011; Rios et al., 2012).

Thus, the objective of this work was to evaluate the rooting of 'Bengal' lychee cuttings, collected in spring and summer, using three concentrations of indolebutyric acid and three substrates.

## 2. Method

### 2.1 Experimental Design

Experiments were carried out from September 2011 to July 2012. The experimental design was a completely randomized design, with five replications, containing 10 cuttings per plot, in a  $3 \times 3$  factorial arrangement. The studied factors were three substrates (carbonized rice husk, coconut fiber, Amafibra® and vermiculite with medium particles) and three indolebutyric acid (IBA) concentrations ( $0 \text{ mg L}^{-1}$ ;  $1000 \text{ mg L}^{-1}$  and  $2000 \text{ mg L}^{-1}$ ).

### 2.2 Physical and Chemical Characteristics Analysed in the Substrates

Substrates were analyzed to determine their physical characteristics: bulk density ( $\text{g L}^{-1}$ ) and water retention capacity ( $\text{mL L}^{-1}$ ) and chemical characteristics: pH and electrical conductivity ( $\mu\text{S cm}^{-1}$ ), according to the method proposed by Kämpf et al. (2006).

### 2.3 Preparation of the Cuttings

Cuttings of 'Bengal' lychee were collected at morning, in a commercial production area, situated in Uraí, Paraná at 2011 spring (September) and 2012 summer (January).

Herbaceous cuttings with 10-12 cm length were collected and cutting basal leaves were removed; leaving two pairs of leaves and 1/3 of each leaf at the top. Cuttings were submitted at two IBA applications, 1000 or 2000  $\text{mg L}^{-1}$ , by quick-dip (10 seconds), method involves dipping the basal portion of cutting into a concentrated hydroalcoholic solution of IBA, prior to inserting the cutting. A control treatment was performed by immersion of basal cuttings portion in hydroalcoholic solution without IBA.

### 2.4 Conditions of the Experiments

Cuttings were disposed in perforate plastic boxes ( $44 \times 30 \times 7 \text{ cm}$ ) filled with the three substrates and kept in a mist chamber controlled by an intermittent timer and a solenoid valve. The valve was programmed to mist during 10 seconds every three minutes. The nozzle nebulizer employed (Model Mist DanSprinklers, Israel) has a flow of  $35 \text{ L h}^{-1}$ . The mist chamber was maintained in greenhouse covered with transparent polyethylene film and 30% shading. Figure 1 presents daily minimum and maximum temperatures into the mist chamber.

### 2.5 Evaluations

Trials were evaluated at 140 days after the installation, in both seasons. The follows characteristics were assessed: cuttings survive (%); leaves maintenance per cutting (%); cuttings no-rooting with callus (%); cuttings rooting (%); roots number per cutting; roots medium length per cutting (cm); and roots dry mass (g).

### 2.6 Statistics and Data Analysis

All data were analyzed by ANOVA and the treatments averages were compared by Tukey test ( $p < 0.05$ ). Data derived from percentage and counting were transformed to sine-arc  $\sqrt{x/100}$  and  $\sqrt{x+1}$ , respectively.

## 3. Results and Discussion

Significant interaction between substrates and IBA concentrations was not recorded for analyzed characteristic, in both seasons evaluated (Tables 1 and 2).

Table 1. Cuttings survival (CS), leaves maintenance (LM), cuttings non-rooted with calli (CC), rooted cuttings (RC), number of roots (NR), roots medium length (RML) and roots dry mass (RDM) of 'Bengal' lychee (*Litchi chinensis*) cuttings collected in 2011 spring and submitted at indolebutyric acid (IBA) concentrations

IBA (mg L <sup>-1</sup> )	CS (%)	LM (%)	CC (%)	RC (%)	NR	RML (cm)	RDM (g)
0	39.3ns	28.7ns	13.3 a	24.7ns	5.2ns	4.6ns	0.2ns
1000	37.3	28.7	8.7 ab	24.7	5.6	5.7	0.2
2000	44.7	34.3	4.7 b	34.7	5.6	6.8	0.2
Average	40.4	30.5	8.9	28.0	5.5	5.7	0.2
Substrate							
COF	19.3 c	8.7 c	10.0ns	8.7 c	2.1 b	3.9ns	0.1 b
VER	38.7 b	28.7 b	5.3	27.3 b	7.3 a	6.5	0.2 a
CRH	63.3 a	54.3 a	11.3	48.0 a	6.9 a	6.8	0.3 a
Average	40.4	30.5	8.9	28.0	5.5	5.7	0.2
CV (%)	40.3	48.4	75.4	45.7	33.0	64.0	70.0

Note. Different lower case letters in the column for each variable showed a difference in the Tukey test at 5% probability. ns: not significant difference. COF: coconut fiber; VER: vermiculite; CRH: carbonized rice husk.

Table 2. Cuttings survival (CS), leaves maintenance (LM), cuttings non-rooted with calli (CC), rooted cuttings (RC), number of roots (NR), roots medium length (RML) and roots dry mass (RDM) of 'Bengal' lychee (*Litchi chinensis*) cuttings collected in 2012 summer and submitted at indolebutyric acid (IBA) concentrations

IBA (mg L <sup>-1</sup> )	CS (%)	LM (%)	CC (%)	RC (%)	NR	RML (cm)	RDM (g)
0	11.3 ns	11.3 ns	8.7 ns	0.0	0.0	0.0	0.0
1000	12.7	12.1	8.0	0.0	0.0	0.0	0.0
2000	12.0	12.0	9.3	0.0	0.0	0.0	0.0
Average	12.0	11.8	8.7	0.0	0.0	0.0	0.0
Substrate							
COF	6.0 b	6.0 b	3.3 b	0.0	0.0	0.0	0.0
VER	24.0 a	23.4 a	18.7 a	0.0	0.0	0.0	0.0
CRH	6.0 b	6.0 b	4.0 b	0.0	0.0	0.0	0.0
Average	12.0	11.8	8.7	0.0	0.0	0.0	0.0
CV (%)	76.0	77.8	94.3	-	-	-	-

Note. Different lower case letters in the column for each variable showed a difference in the Tukey test at 5% probability. ns: not significant difference. COF: coconut fiber; VER: vermiculite; CRH: carbonized rice husk.

For cuttings collected in spring and summer, significant differences between IBA concentrations was not observed for analyzed characteristics, except for non-rooted cuttings with callus percentage, when collected in spring (Table 1).

Regarding cuttings survive percentage; it is observed in Tables 1 and 2 that IBA concentrations did not influence the cutting survival and leaves maintenance per cutting in both seasons. Similar results are described by Bastos et al. (2006) and Koyama et al. (2014), which studied 'Bengal' lychee trees propagation by cuttings, and reported that the IBA application did not presents influence on cuttings survival percentage.

On the other hand, it can be observed in Tables 1 and 2 that the substrates had a direct influence on these variables, as well as for leaves maintenance. For cuttings collected in spring, highest averages of survival percentage occurred in carbonized rice husk and vermiculite, respectively. However, in summer the highest cuttings survival percentage was recorded in the vermiculite.

As the substrate coconut fiber has low density and high water retention capacity in relation to the other substrates tested (Table 3), it is probable that for the species under study these conditions were unfavorable.

Table 3. pH, electrical conductivity (EC), bulk density (BD) and water retention capacity (WRC) of substrates coconut fiber (COF), vermiculite (VER) and carbonized rice husk (CRH)

Substrates	pH	EC ( $\mu\text{S}\cdot\text{cm}^{-1}$ )	BD ( $\text{kg}\cdot\text{m}^{-3}$ )	WRC ( $\text{mL}\cdot\text{L}^{-1}$ )
COF	6.3	162.7	98.6	756.9
VER	7.1	120.6	211.9	457.5
CRH	6.9	178.1	143.4	569.7

Table 1 shows IBA influence on non-rooted cuttings with callus percentage. Highest average for this characteristic was obtained in non-treated cuttings with plant regulator, but not differing from those treated with  $1000\text{ mg L}^{-1}$ . As the conversion of plant tissues to callus is associated with the hormonal balance between auxins and cytokines in cells, it is likely that the higher concentrations of IBA have caused hormonal imbalance and thus reduced the calogenesis. In contrast, for cuttings collected in summer, IBA application did not promote calogenesis; however, the cuttings kept in vermiculite showed a higher percentage of callus in relation to other substrates.

Smarsi et al. (2008), assessing 'Bengal' lychee tree by layering verified that Plantmax® substrate provided a linear increase in layerings with callus when IBA concentrations increasing too. IBA application ( $4000\text{ mg L}^{-1}$ ) promoted 100% of callus in layering propagation. Calogenesis is an important process in plants propagation by cutting, since callus can differentiate in roots and promote highest rooting percentages.

For the variables such as cuttings rooting percentage, number of roots, root mean length and dry mass of roots per cutting, the IBA application did not result in significant results at both collection times (Tables 1 and 2). These results corroborate those obtained by Carvalho et al. (2005), who did not observe the influence of IBA application in the concentrations of 0 to  $4000\text{ mg L}^{-1}$  in 'Bengal' lychee cuttings, for the same cultivar evaluated in the present study.

Cuttings submitted to different treatments in summer did not rooted, or when rooted, the proportion of rooted cuttings was less than 1%. In this case, the season negatively interfered in cuttings rooting capacity. However, as in some treatments the cuttings presented callus (Table 2), it could be that if the evaluation took place in a later period, the cuttings with calluses would be rooted.

In relation to rooting, Bastos et al. (2006) observed that the cuttings of the same cultivar treated with IBA  $6000\text{ mg L}^{-1}$  presented 50.17% of rooting, whereas in those that did not receive the application of this plant regulator the lowest rooting percentage (13.57%) was obtained. Koyama et al. (2014), which collected litchi cuttings in winter, also obtained low rooting percentages for cuttings treated with IBA  $1000\text{ mg L}^{-1}$  on the substrates carbonized rice husk, coconut fiber and vermiculite (10, 20 and 14% rooting, respectively).

Although the IBA concentrations tested in this study did not stimulate lychee tree cuttings rooting, it was observed a direct influence of the substrates in the rooting of the cuttings collected in the spring. Regardless of the concentration of IBA, the rooting in the substrate carbonized rice hull was superior (48%) to the others. In studies of other fruit tree species, Zietemann and Roberto (2007), testing substrates and IBA on rooting of guava (*Psidium guajava* L.) cuttings, did not observe any difference in the rooting percentage for the Paluma cultivar when was used the substrates carbonized rice husk and vermiculite; and for 'Século XXI', the carbonized rice husk promoted a better rooting of the cuttings.

Similar results can be observed in the study of peach tree (*Prunus persica*) propagation, where the application of IBA  $2000\text{ mg L}^{-1}$  in cuttings wrapped in the substrates carbonized rice husk and vermiculite provided a higher rooting percentage (Cardoso et al., 2011). On the other hand, different from the one found in this study, Pio et al. (2005), studying substrates for the rooting of fig tree (*Ficus carica* L.) herbaceous cuttings, verified that the substrate coconut fiber promoted the highest rooting percentage (86.87%).

For the number and dry mass of roots (Table 1), the highest averages were found on the substrates carbonized rice husk and vermiculite, differing significantly from coconut fiber. Contrasting results were described by Koyama et al. (2014), however, these authors collected cuttings at another season (winter), which can had influenced these results.

Carvalho et al. (2005) observed that the greatest number, length, fresh and dry mass of roots of 'Bengal' lychee cuttings was obtained in cuttings collected in the spring, regardless of the AIB concentrations tested. On the other hand, the same authors obtained a higher percentage of rooted cuttings when cuttings were collected in the summer (48.7%), followed by spring (24.5%) and autumn (8.1%). However, Leonel and Rodrigues (1993)

observed a low survival percentage of 'Bengal' lychee cuttings collected in the summer and Leonel et al. (1995) found that the lychee cuttings in the spring associated with the IBA application ( $5000 \text{ mg L}^{-1}$ ) provided 83% of rooting in vermiculite substrate.

Thus, the cutting collecting season should be determined regionally for each species, since some species rooted better in the beginning of the spring and others between spring and summer (Fachinello et al., 2005). This is related to several factors, such as temperature, solar radiation, nutritional and physiological conditions, and vegetative cycle of the mother plant (Fachinello et al., 2005; Hartmann et al., 2011).

Regarding the temperature inside the greenhouse, averages close to  $30 \text{ }^\circ\text{C}$  were observed during the experimental period, especially between October and February (Figure 1). The temperature may influence rooting, acting mainly on water absorption and metabolism, especially in regions of subtropical climate (Corrêa & Fett-Neto, 2004; Hartmann et al., 2011).

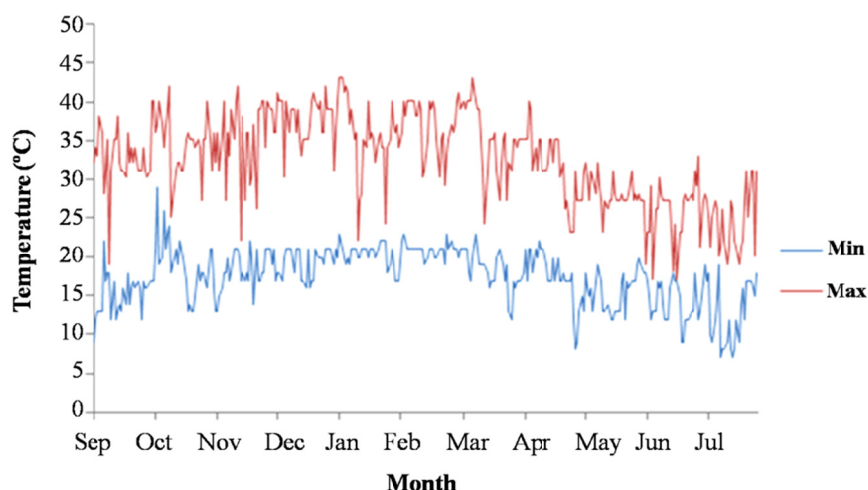


Figure 1. Averages of the minimum (Min) and maximum (Max) temperature ( $^\circ\text{C}$ ) inside the greenhouse during the experiments conduction

Hartmann et al. (2011) reported that cell division is favored by the increase in temperature and, consequently, it helps the formation of roots and the production of shoots. For Hansen (1989) the ideal average temperature associated with these processes is  $24 \text{ }^\circ\text{C}$ . However, excessively high temperatures during the rooting phase should be avoided because they stimulate the development of lateral buds before the appearance of roots. In addition, there is an increase in transpiration and loss of water by the leaves, causing tissue necrosis (Hartmann et al., 2011). Thus, the high temperatures recorded may have interfered in the percentage of rooting of the cuttings, in both seasons of collection. On the other hand, low temperatures decrease the metabolism of the cuttings, leading to less shoot production and longer rooting or, even, do not provide suitable conditions for induction, development and root growth (Xavier, 2002).

In summary, it is verified that the propagation of 'Bengal' lychee tree by cuttings can be viable, through the collection of cuttings in the spring. In addition, carbonized rice husk as a substrate may be a low-cost option, although the availability of the material depends on the region.

## 5. Conclusion

Vegetative propagation of 'Bengal' lychee by cutting can be performed in spring, without applying IBA and using carbonized rice husk as substrate.

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