

**SEASON AND USE OF *Cyperus Rotundus*  
EXTRACT FOR PROPAGATION OF *Cnidoscolus*  
*quercifolius* Pohl BY AIR LAYERING**

**ABSTRACT**

For the forest species, as an example, *Cnidoscolus quercifolius* Pohl. the clonal propagation makes possible greater genetic gains than in the reproduction through seeds in less time. The auxin – based phyto regulators such as Indol Butyric Acid (IBA) and also the aqueous extract of *Cyperus rotundus* L. tubers have been used in plant cloning as rooting promoters an improvement in root quality. The aim of this work was to analyze the efficiency of the cloning technique by air layering in a Caatinga species, *Cnidoscolus quercifolius*, seeking to know the best season, the time requires for rooting and the efficiency of the extract of *C. rotundus*. and IBA. Thus, aqueous extracts of tubers of *C. rotundus*, at concentrations of 0.0% (absolute witness), 10%, 20%, 30% and 40% were also evaluate, also the AIB at the concentration of 6.0 g/L (witness 2). From the results it was verified that the best season for the cloning of *Cnidoscolus quercifolius* by technique of wintering. In general, the use of natural and synthetic auxins positively influenced all the variables analyzed, surpassing the controls. It should be noted that the IBA was statistically superior to the other treatments for the response variables of the treatments applied (notes) and number of roots. It can be concluded that this species may be an advertisement for this technique, since it presented a higher number of rootes weeds in a shorter time, when compared to other forest species.

*Keywords: clonal propagation, natural auxin, vegetative propagation.*

**1. INTRODUCTION**

The seedling production of tree species has been carried out using the sexed and asexual method. The first one refers to the production of seedlings through seeds and the second one through cloning (XAVIER et al., 2009). Among the advantages of cloning is the fact that heterozygous material can be perpetuated with the integral maintenance of genetic information, as well as independence of seed availability, elimination of seed dormancy problems, reduction of juvenile stage and uniformity in clonal plantations.

For forest species, cloning allows greater genetic gains than in seed reproduction, in less time. Contrary to agricultural species, the forest usually presents a prolonged juvenile phase before reaching flowering and maturity (NEVES et al., 2006). In particular for the faveleira, this shows several vatangens.

The species *Cnidoscolus quercifolius* Pohl. (faveleira) is a pioneer plant. It is a species of the family Euphorbiaceae, with high willingness to withstand the drought, rustic and fast growing. It can be used in the restoration of areas in degradation, destined to the composition of reforestation, in medicine, animal and human feeding, energy, sawing, biodiesel, among others. It presents a considerable concentration of phosphorus enitrogen, indicating a potential in the feeding of bovine herds, sheep goats (LORENZI, 2002).

The use of the waterproofing is one of the traditional techniques of plant cloning that connotes in the removal of a piece of the circumference from the bark of branches, so as to expose a ring of the inner tissue, onto which is added a moist substrate, covered by film plastic. Due to the removal of a part of the bark, co-factors of rooting, auxins and photoassimilates accumulate, which are associated with the exogenous use of growth regulators, such as Indol Butyric Acid (IBA), which are important elements to promote adventitious rooting. This technique stands out for the easy application, greater success in species of difficult rooting, besides not needing of infrastructure for seedlings production, like greenhouse.

In the literature it is known the use of aqueous extracts of tubers of *Cyperus rotundus* L. as root promoters and potentiator in roots formed. Lorenzi (2000) says that Indol Butyric Acid (IBA) is present in high concentrations, which is a specific phytohormone that will induce the formation of rooting in plants. The use of this extract can cause a decrease in the cost of seedlings production through the porporquia, in addition the *C. rotundus* occurs with abundance in all regions of the world.

Cloning has some advantages, highlighting the fact that the heterozygous material will be perpetuated with the integral maintenance of the genetic information, thus generating more uniform clonal plantations with similar characteristics and quality. In addition, the independence of seed availability, contours problems with seeds that present difficulties to break dormancy and decrease the time in the juvenile phase. In view of this, it is known that *Cnidocolus quercifolius* presents a problem in the collection of seeds because its dispersion is autocoric, throwing its seeds at a certain distance.

The exploitation of native species has contributed to reduce the genetic variability of multiple forest species. One of the measures to mitigate this destruction in native areas is the implantation of orchards with plants of interest, so that species of the natural forests of the region are conserved. In planting these areas, there is a need for seedling formation, and for this, techniques such as cloning are used. The importance of cloning in native species such as *Cnidocolus quercifolius* is evidenced by the value and multiple uses of these specie for the region, and the scarcity of clonal studies, mainly in the Northeast region of Brazil.

However, the objective of the present work was to analyze the efficiency of the cloning technique by air layering in a caatinga species, *Cnidocolus quercifolius*, seeking to know the best season, the time necessary for rooting and the efficiency of the extract of tera and the IBA.

## **2. Material and methods**

### **2.1 Experiment installation locations**

The research area is located at the Federal University of Campina Grande, Campus Patos - PB, located at coordinates 7 ° 03'30 " S and 37 ° 16'30 " W; the other experimental area is located at Farm NUPEARIDO (Nucleus of Research for the Semi-Arid Tropic), of the Rural Health and Technology Center (CSTR), distanced about 6 km from the first area in the Patos Campus, located at coordinates 07 ° 05 '13' 'S and 37 ° 15'40 "W. The species *C. quercifolius* which grows naturally in this area was used. According to Köppen, the climate of the region is characterized by the type Bsh, considered hot and dry with two well-defined seasons, one dry and one rainy, relative air humidity around 55%, average temperature of 30 ° C and annual average rainfall of 700 mm.

## **2.2 Preparation of the concentrations of aqueous extracts of *Cyperus rotundus* L. for cloning of the *Cnidocolus quercifolius* Pohl.**

Extracts of *C. rotundus* L. tubers were prepared at concentrations of 0% (100% distilled water, absolute witness), 10%, 20%, 30% and 40%, using additionally indole butyric acid auxin, at the concentration of 6.0 g/l, as the showed the best result in the induction of rooting in *Cnidocolus quercifolius* (CAMPOS, 2010). Then routed to the laboratory of plant physiology, then washed and dried using paper towel.

First, 100g of tubers were placed in a becker then distilled water was added to make up the 250ml volume, this material being milled in a blender and sieved to give the 40% concentration. Subsequently, the extracts were prepared at 10% concentrations (37.5 ml distilled water + 12.5 ml extract); 20% (25 ml distilled water + 25 ml extract) and 30% (12.5 ml distilled water + 37.5 ml extract). The extracts were conditioned in a refrigerator for application to the air layering in the morning of the following day.

## **2.3 Conduction and Installation of Experiment**

Two experiments were installed. The first experiment was installed in the dry period (August to January) in the year 2012 and the second in the rainy season (February to July) in 2013. The air layering were randomly installed in the plant using six branches per tree, to form a repetition of the experiment. Both experiments remained in the field for 90 days.

In each period, juvenile stage matrices with healthy and vigorous leafy branches were chosen. Not finding total healthy branches available in a single matrix, for allocation of one repetition, another similar plant was used to complete said repetition. The branches were specially arranged in the quadrants of the plant.

For the preparation of the air layering, branches with diameter between 1 and 2 cm were ringed with a pocketknife, and the bark was completely removed, forming a ringing approximately 1.5 cm wide and 60 cm below the tip. Soon afterwards, the synthetic or natural IBA solution was introduced into the ring with the aid of a brush, at the pre-established concentrations.

Then the twig was covered with a transparent plastic film with the two open ends, having dimensions of 360 x 250 x 0.15 mm in length, width and thickness, respectively. One end of the film was tied. Subsequently, the substrate was vermiculite of medium granulometry and the amount of water preset for the wetting of the skin. The other end of the plastic film was tied, thus creating a dark and moist environment over the girdle.

It was used in each chamber 600 cm<sup>3</sup> of vermiculite. To determine the amount of water to be used to place on the substrates, a water retention capacity test was performed using three replicates. In each, a total of 500 ml of water was added in 600 cm<sup>3</sup> of vermiculite and the amount of water retained was calculated. After this result, it was determined that a volume of 120 ml of water in each layer, obeyed 70% of the substrate field capacity, remaining 30% of the pores of the free substrates for aeration.

Water was added to the substrates with the use of a graduated plastic syringe, in a quantity pre-established in the water holding capacity test. This process provided a humid environment around the incision, to facilitate and promote the appearance and development of the roots in the air layering.

## 2.4 Data Collection

In this part of the methodology, similar activities were carried out in the work of (PIMENTA et al., 2014; FARIAS JR, 2011; CAMPOS, 2010). Weekly observations were made of the surface of the substrate to analyze the degree of humidity of the air layering and the appearance of roots within the plastic film until the removal of the air layering. The sand was moistened whenever necessary.

Finally, the weedy branches were removed with the aid of pruning shears and sent to the laboratory of Plant Physiology of the CSTR / UFCG, where the plastic films were removed and the roots separated from the substrate by washing them. Soon after, the information for the evaluations was collected.

The variables analyzed were: presence of air layering with callus (formation of undifferentiated cell mass in the ring region); presence of air layering with root primordia; presence of rooted air layering and percentage of them. In rooted sandworms, we analyzed: number of roots, length of largest root (cm); fresh mass and dry mass of roots (g).

The variables: presence of air layering with calluses; presence of air layering with root primordia; presence of rooted air layering and length (cm) of the largest root for air layering were evaluated through the allocation of notes to air layering (farias junior, 2011). The grades were assigned on a scale of 0 to 4 according to the following criteria: 0 = no-rooting; 1 = with callus formation; 2 = with root primordia; 3 = with roots up to 4 cm and 4 = with roots greater than 4 cm.

The determinations of the fresh mass and dry mass values of roots were made after counting and measuring the roots. For fresh mass, the roots of the air layering were extracted and immediately obtained in semianalytic scale, noting the respective value (g). Then, the roots were packed in paper bags and placed in an oven at 65 °c for approximately 3 days until reaching a constant mass to obtain the dry mass.

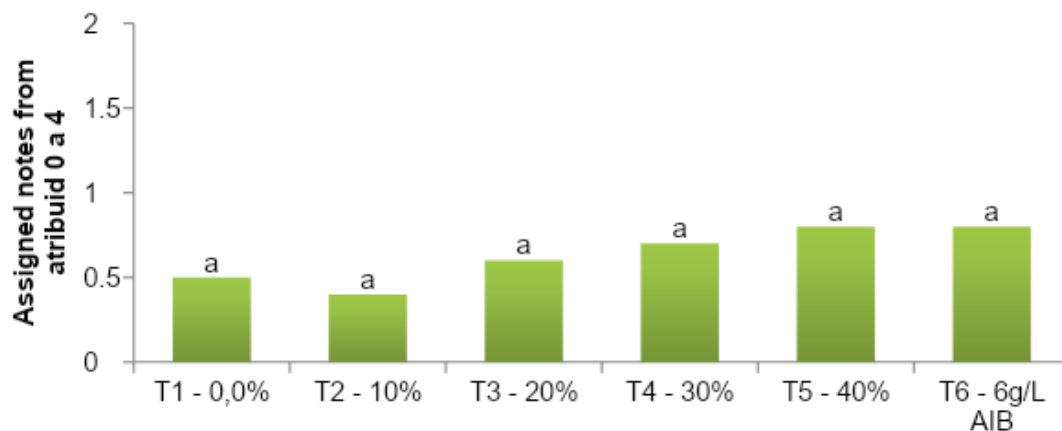
## 2.5 Experimental design

The experiments were installed in a completely randomized block design (dbc) (BANZATTO and KRONKA, 2006). The experiments with the faveleira were used six treatments and eight replications, where each plot constituted a hut, totaling forty-eight plots.

Because the data did not meet the requirements of normality and homoscedasticity, even after the data transformation, the nonparametric kruskal-wallis test was applied. The analyzes were performed using the statistical package action version 2.5 (ESTATCAMP, 2013), at a significance level of 5%.

## 3. RESULTS AND DISCUSSION

In the dry period no rooting was observed in any air layering, only the presence of root primordia. Therefore, it was possible to evaluate only the response of the treatments to the treatments applied, by assigning notes (Figure 1). Although the differences are not significant, it is verified that higher values were observed in the treatments with greater concentration of the extract of *Cyperus rotundus* L. and with the use of synthetic auxin.



**Figure 1.** Response of the air layering to rooting (Notes 0 to 4), at 90 days after the performance of the aliquots in *Cnidoscopus quercifolius* dry period. \* T1 to T5: 0.0% to 40% of *C. rotundus* extract; T6: 6.0 g / l AIB. \*\* Averages followed by the same letter do not differ from each other by the Kruskal-Wallis test, at a significance level of 5% ( $P > 0.05$ ).

It is observed that, the number of air layering that emitted roots according to the treatments and time after the performance of the alporquias (Table 1). The appearance of adventitious roots on the surface of the substrate was observed in the T3 treatment (20%) at 42 days after installation and the highest number of rooted air layering occurred in the T6 treatment (6g / l AIB). These results are consistent with Pasqual et al. (2001), when they say that the use of auxins allows an increase in the percentage of rooting and the acceleration of the formation of roots.

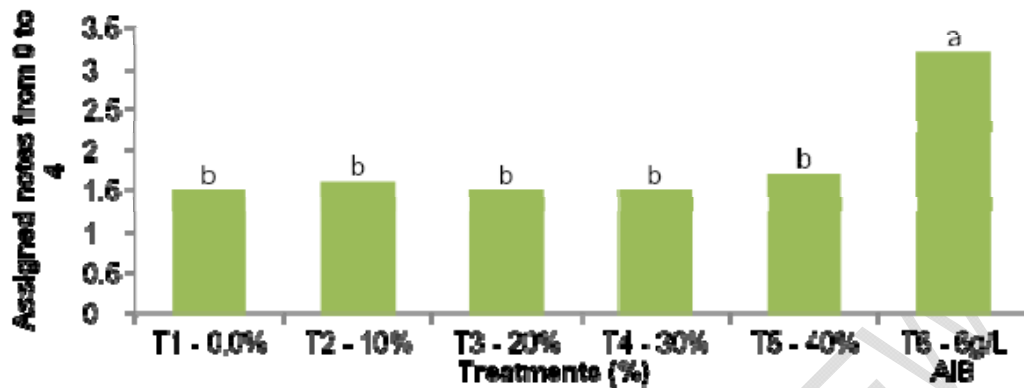
**Table 1.** Cumulative values of rooted *Cnidoscopus quercifolius* air layering, as a function of the application of *C. rotundus* and AIB extract concentrations.

Treatments	Time after the performance of the alporquias (days)									
	28	35	42	49	56	63	70	77	84	90
T1 (0%)	-	2	2	3	3	3	3	3	3	3
T2 (10%)	-	1	1	3	3	3	3	3	3	3
T3 (20%)	-	-	1	1	1	1	1	1	1	1
T4 (30%)	1	2	2	2	2	2	2	2	2	2
T5 (40%)	1	3	3	3	3	3	3	3	3	3
T6 (6,0 g/L)	1	2	4	4	5	6	6	6	6	6

**Source** - Pimentel (2014).

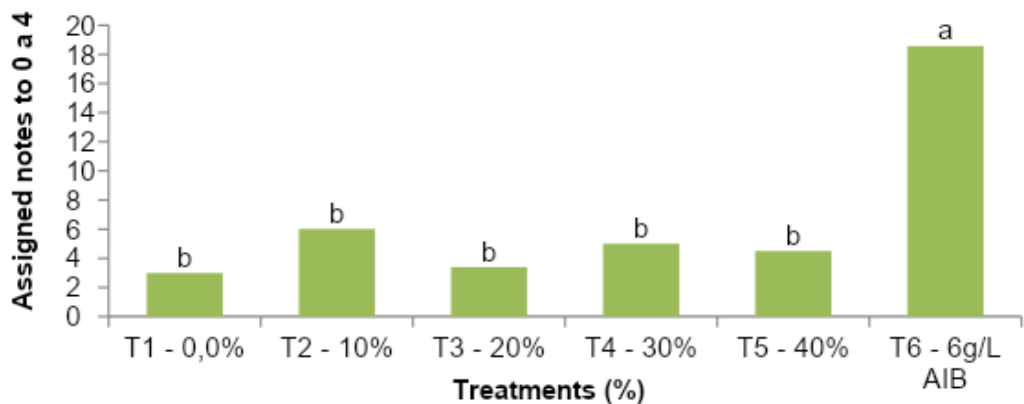
Figure 2 shows the results of the response of the treatments applied to the rainy season. As mentioned in the methodology, the variables: air layering without rooting; with callus formation; with root primordia; with roots up to 4 cm, and with roots larger than 4 cm; were assessed by assigning grades to the air layering (grades attributed on a scale of 0 to 4). The note number corresponds to the response of the branches to the treatments applied. T6 (synthetic auxin) presented better results than the other treatments with natural auxin. Comparing the notes from this rainy period with the dry period, there was an expressive increase in all treatments, in which only a significant difference was observed between the

T6 and the others in the rainy season.



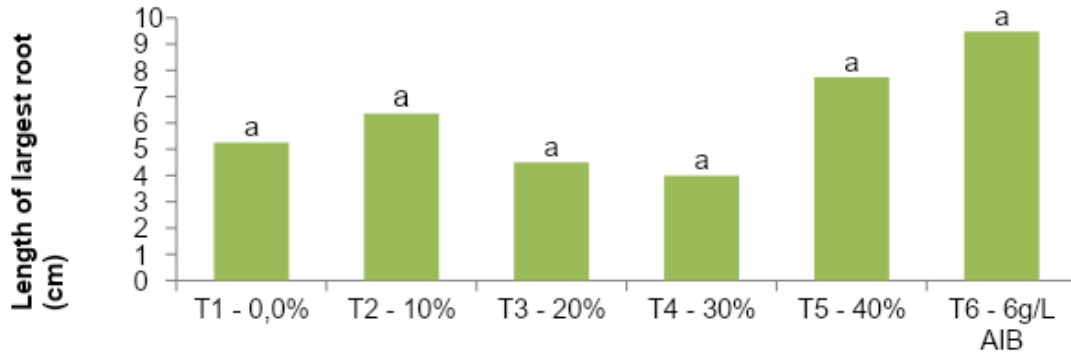
**Figure 2.** Response of air layering to rooting (Assigned notes 0 to 4), at 90 days after the performance of the alnacquies in *Cnidocolus quercifolius*. \* T1 to T5: 0.0% to 40% of *C. rotundus* extract; T6: 6.0 g / l AIB. \*\* Averages followed by the same letter, do not differ by the Kruskal-Wallis test, ( $P > 0.05$ ).

According to Figure 3, the T6 treatment (6g / l AIB) obtained on average the highest number of roots, evidencing an expressive difference between the T6 (AIB) and the other treatments. Pimenta et al (2014) working with the same species and concentration found that it was superior to the control (0% AIB).



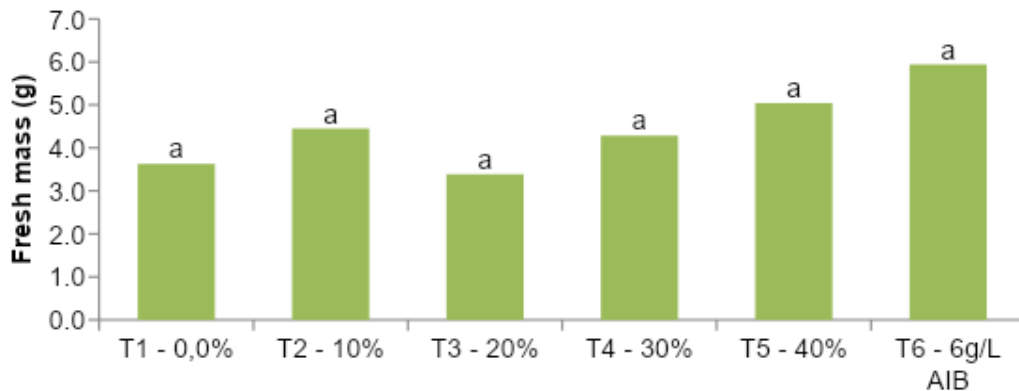
**Figure 3.** Number of roots observed at 90 days after reaching the aliquots in *Cnidocolus quercifolius*, Patos-PB, 2013. \* T1 to T5: 0.0% to 40% of *C. rotundus* extract; T6: 6.0 g / l AIB. \*\* Averages followed by the same letter, do not differ by the Kruskal-Wallis test, ( $P > 0.05$ ).

It is observed in figure 4 that the T5 (40%) and T6 (AIB) treatments obtained the highest average of the Length of the largest root (cm), showing that for this parameter the synthetic auxin was relatively similar to natural auxin since the average did not present any significant difference between these treatments.

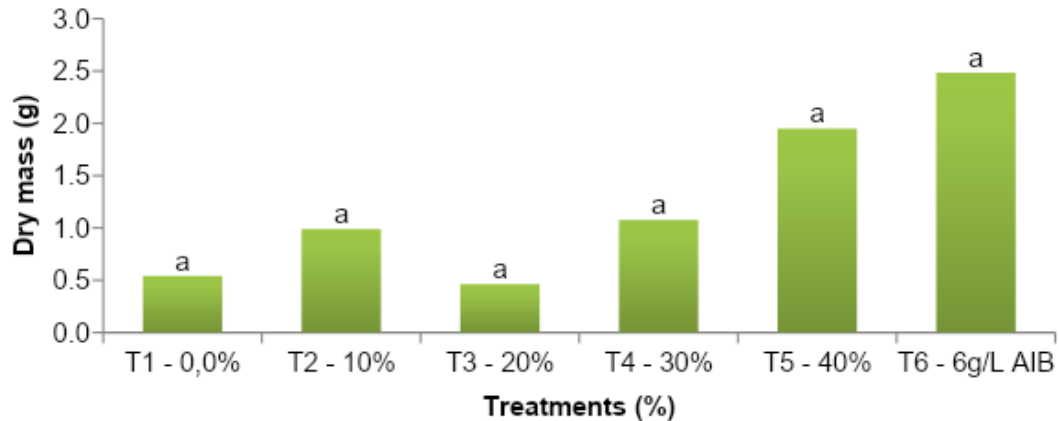


**Figure 4.** Length of the largest root (cm) observed, at 90 days after the performance of the alnacus in *Cnidocolus quercifolius*. \* T1 to T5: 0.0% to 40% of *C. rotundus* extract; T6: 6.0 g / l AIB. \*\* Averages followed by the same letter, do not differ by the Kruskal-Wallis test ( $P > 0.05$ ).

The fresh mass (Fig. 5) and dry mass (Fig. 6) had similar behavior to the length of the largest root (cm). The highest mean values were obtained in T5 (40%) and T6 (AIB) treatments. Lucena et al. (2014), using the same IBA dosage, in the same species, found higher values for root dry mass in relation to control (0%).



**Figure 5.** Fresh mass (g) observed at 90 days after the performance of the alnacus in *Cnidocolus quercifolius*. \* T1 to T5: 0.0% to 40% of *C. rotundus* extract; T6: 6.0 g / l AIB. The means are not comparable to the Kruskal-Wallis test, ( $P > 0.05$ ).



**Figure 6.** Dry mass (g) observed, at 90 days after the performance of the alporquia in *Cnidocolus quercifolius*. \* T1 to T5: 0.0% to 40% of *C. rotundus* extract; T6: 6.0 g / l AIB. The means are not comparable to the Kruskal-Wallis test, ( $P > 0.05$ ).

Analyzing all the results obtained, it is generally observed that the use of natural auxin positively influenced all the analyzed variables, however, the synthetic auxin was superior to the natural auxin in all analyzed variables, being statistically superior for the response variables of the air layering, where grades are given on a scale of 0 to 4 and for the number of roots ( $P < 0.05$ ).

It is important to note that *C. rotundus* L. is a very common species in nurseries, gardens, orchards, vegetable gardens and crops. The species is considered one of the most important weeds in the world due to its wide distribution, competitive capacity and aggressiveness, as well as the difficulty of control and eradication. Thus, the use of the aqueous extract of tereza is feasible for cloning of faveleira by the watering method, minimizing the costs and environmental impacts of the synthetic hormone, since the former is obtained naturally. However, it should be considered that the IBA performance was higher, suggesting the study of other factors, which may improve the efficiency of auxin extracted from the tuber of *C. rotundus* L.

Campos (2010), working with *Cnidocolus quercifolius* using 6.0 g L<sup>-1</sup>, also by the alporquia technique found that the season with the highest rooting index and, less time occurred in the rainy season, where the appearance of roots on the surface of the substrates occurred two months after the performance of the air layering, with the use of AIB in concentrations similar to those used in this research.



#### 4. CONCLUSION

The best time for the cloning of *Cnidoscopus quercifolius* seedlings by the watering technique is the rainy season. In general, it was noted that the use of exogenous auxin positively influenced all the analyzed variables, surpassing the absolute control.

For *C. quercifolius* the aib was statistically superior to the other treatments for the variables response of the air layering to rooting (notes) and number of roots, in which the highest number of rooted air layering was found in the T6 treatment (AIB). It is noteworthy that the root growth was observed first in the two treatments with the highest concentration of extract *Cyperus rotundus* L.-and IBA.

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