Evaluation of the productivity and rooting of cuttings of ten *Eucalyptus* clones

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ABSTRACT

Objective: evaluating the productivity of mother plants and the rooting percentage of cuttings of *Eucalyptus grandis*, *Eucalyptus urophylla* and *Eucalyptus urograndis*.

Design: Ten clones of each species were evaluated in a clonal miniature garden in a gutter system fitted with drip irrigation. **Results**: The assessed number of sprouts and viable cuttings variables registered high significant differences (P<0.0001) between clones: UP1 (17,947 shoots and 34.05 viable cuttings), UG2 (12,120 shoots and 22.96 viable cuttings) and G2 (10,254 shoots and 23.96 viable cuttings). Regard the measured parameters in the irrigation water, the average values of EC 0.5 mS/cm and pH 5 to 7 were obtained. The stem cuttings established in the module had high significant differences (P<0.0001) in terms of the rooting percentage. Three clones were superior, the best of which, clone G2, reports 78.58% rooting, which developed in environmental conditions of relative humidity greater than 80% and temperature between 25-30 °C.

Findings/conclusions: With the obtained values from the measured variables, a rooting productivity projection was carried out considering a 54 mother plants/m² density, resulting in a minimum average annual production of viable cuttings from 10,000 to 27,000 depending on the clone, with 2,000 to 18,000 cuttings with rooting possibilities per square meter.

Keywords: Asexual propagation, Clonal miniature garden, plant growth regulators.

INTRODUCTION

The Eucalyptus genus has a diversity of approximately 700 species. It is native to Australia and nearby islands and has industrial importance for cellulose production, lumber milling and chipboards. Their first forest plantations in Mexico date back to 1950, with less than 7,000 hectares planted with broadleaf species, mainly eucalyptus. The "Pulsar" company had a eucalyptus plantation with a 300,000 hectares area at Emiliano Zapata, Tabasco, Mexico (Martínez *et al.*, 2006). The objective of this work was to evaluate the productivity of mother plants and rooting percentage of cuttings of *Eucalyptus grandis*, *Eucalyptus urophylla* and *Eucalyptus urograndis*.

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MATERIALS AND METHODS

Study Area Location

The study was developed in a nursery of the Proplanse S.A. de CV company, located at km 12.5 of the Baláncan-El Triunfo highway, Balancán, Tabasco, Mexico (UTM coordinates, zone 15 Q 659601 m E and 1976492 m N) where a clonal miniature garden with mother plants of the *Eucalyptus* genus was established.

Background of the evaluated clones

In this research, ten different clones of *Eucalyptus grandis* (1), *Eucalyptus urophylla* (8) and *Eucalyptus urograndis* (1) obtained from selected trees in operational plantations and trials because of their outstanding productivity and economic interest, the characteristics of diameter and height of these trees are summarized in Table 1.

The clone capture process consisted of knocking down selected trees to promote sprouts emission, from these,

vegetative material was collected. The cuttings were rooted and multiplied to obtain mother plants of the clonal miniature garden. A total sample population of n=130 individuals as an experimental unit (Table 2) was evaluated.

An analysis of variance (ANOVA) was performed on the assessed variables, the number of regrowths, number of viable cuttings and survival percentage, the value of the F statistics and its significance are shown. With mean comparison analysis with 5% TUKEY test to determine homogeneous or heterogeneous groups. For this, the STATISTIX 10 statistical software was used.

Mother plant productivity

The mother plants productivity data recollection consisted of counting the sprouts obtained per vegetative unit sampled from each clone, later the viable cuttings made from these sprouts were counted.

Table 1 . Summary of the characteristics of the mother trees of <i>Eucalyptus</i> Labill.					
Number	Material	Species	Source	Diameter (cm)	Height (m)
1	UP1	E. grandis	Arboretum, Balancán	47	31
2	G2	E. urophylla	Arboretum, Balancán	57	35
3	UG2	E. urograndis	Arboretum, Balancán	53	36
4	PC11	E. urophylla	V. Carranza, Huimanguillo	53	41
5	PC12	E. urophylla	V. Carranza, Huimanguillo	60	39
6	PC21	E. urophylla	V. Carranza, Huimanguillo	60	42
7	PC16	E. urophylla	V. Carranza, Huimanguillo	59	38
8	PC30	E. urophylla	V. Carranza, Huimanguillo	54	41
9	PC37	E. urophylla	V. Carranza, Huimanguillo	53	52
10	PC31	E. urophylla	V. Carranza, Huimanguillo	53	39

Table 2. Sample size of the experimental unit per evaluated clone.					
Number	Clone	Species	Source	Ν	(n)
1	UP1	E. grandis	Arboretum, Balancán	38	15
2	G2	E. urophylla	Arboretum, Balancán	36	15
3	UG2	E. urograndis	Arboretum, Balancán	66	15
4	PC11	E. urophylla	V. Carranza, Huimanguillo	180	15
5	PC12	E. urophylla	V. Carranza, Huimanguillo	44	15
6	PC21	E. urophylla	V. Carranza, Huimanguillo	48	15
7	PC16	E. urophylla	V. Carranza, Huimanguillo	78	15
8	PC30	E. urophylla	V. Carranza, Huimanguillo	5	5
9	PC37	E. urophylla	V. Carranza, Huimanguillo	5	5
10	PC31	E. urophylla	V. Carranza, Huimanguillo	341	15
Total				841	130

Table 3. Days elapsed from cutting to next harvest.						
Evaluation						
Clone	I	Ш	Ш	IV	V	Average
UP1	19	21	21	20	21	20
G2	20	17	22	20	20	20
UG2	21	20	22	17	42	24
PC11	31	21	19	16	40	25
PC12	21	21	23	20	36	24
PC21	22	21	22	20	37	24
PC16	18	21	24	24	28	23
PC30	17	22	24	24	29	23
PC37	17	22	24	24	26	23
PC31	19	21	26	22	24	22
General average					23	

Table 3 shows the number of days elapsed between each harvest per clone, obtaining a 23 d general average.

Growth regulator application and establishment of stakes

For the stimulation of adventitious roots in the cuttings made from the clonal miniature garden, a growth regulator treatment was carried out using exogenous auxins, in this case, AIB (Indole-3-butyric acid) in its RADIX[™] 1500 commercial presentation, formulated as an impregnable powder with 0.15% as the active ingredient.

Irrigation Water Characterization

The management area for cuttings production has evolved in such a way that the spacing of the mother plants has been reduced. Higashi *et al.* (2004) mention the types of production systems and the distances between plants: field clonal garden ($0.5 \times 0.5 \text{ m}$), clonal miniature garden in gutters on a sand substrate ($0.1 \times 0.1 \text{ m}$) with drip irrigation system, clonal garden in 8 L plastic containers and mini clonal fiberglass garden ($0.1 \times 0.1 \text{ m}$). Higashi *et al.* (2002) point out that there is no nutrient solution for all plant species under growing conditions, the nutrients necessary for development are the same, but the amounts extracted differ between and within species. One of the factors determining the success of a clonal propagation operational program employing cuttings is the used nutrient solution in the miniature

Table 4. Average values of the water quality evaluation obtained from the irrigation of a clonal miniature garden.

	Productivity		pH water		C.E. (mS/cm)	
Evaluation	Sprouts	Viable cuttings	average	Deviation standard	average	Deviation standard
I	8	18	5.48	0.709	0.009	0.008
	7	15	4.61	0.942	1.092	0.927
	11	21	4.26	0.849	0.636	0.420
IV	12	23	3.81	0.641	0.55	0.393
V	14	20	3.71	0.362	0.63	0.147

garden; the chemical properties of irrigation water (pH and Electrical Conductivity) influence the usage of nutritive elements. Silveira *et al.* (1999) cited by Higashi *et al.* (2002) recommend that the electrical conductivity should be in the range of 1.25 to 2.3 mS/cm for a hydroponic clonal miniature garden system and the pH between 5.8 and 6.0. However, positive results have been obtained with EC and pH outside these recommended ranges.

RESULTS AND DISCUSSION

During the evaluation period of the clonal miniature garden, the irrigation water parameters were taken, the values behaving differently in each evaluation (Table 4.).

Table 5 shows the number of viable shoots and cuttings produced by each mother plant.

Number of sprouts

During the trial period, with respect to the variable number of regrowth, an increasing trend in the amount of regrowth per individual of each clone was noted according to the age of the mother plant: in the evaluation I, the UP1 clone produced an average of 11 regrowths per individual, 21 regrowths produced on average in the intermediate evaluation (III) and the V evaluation with 28 regrowths on average per individual (Figure 1).

A possible response to this effect of increasing the number of regrowth was the nutrition applied in the clonal miniature garden and the effect of the type of pruning carried out in the regrowth crop. From the analysis of variance summarized in Table 5, three homogeneous groups were formed where the best group was constituted by the UP1 clone **Table 5**. Analysis of variance of the measured variables in a miniature garden and means comparison per clone via the Tukey statistical test. Degree of significance: highly significant (<0.0001), averages with a common letter are not significantly different (p>0.05).

ClonE	Sprouts number	Viable cuttings number
UP1	17.947 ^A	34.05 ^A
G2	10.254 ^{BC}	23.29 ^B
UG2	12.120 ^B	22.96 ^B
PC11	7.929 ^{CD}	12.69 ^D
PC12	9.296 ^{BCD}	15.80 ^{CD}
PC21	9.581 ^{BCD}	16.59 ^{CD}
PC16	10.173 ^{BC}	18.83 ^{BC}
PC30	8.095 ^{BCD}	16.43 ^{BCD}
PC37	9.760 ^{BCD}	18.72 ^{BCD}
PC31	6.625 ^D	15.29 ^{CD}
F	19.78	22.00
P-VALUE	<0.0001	<0.0001

(A) with an average of 17.95 sprouts, followed by the UG2 clone with 12.12 sprouts (B) and group D constituted For clone PC31 with only 6.36 sprouts, the other clones were heterogeneous between B, C and D. Figure 2 shows the measured variables, observing clones such as G2 (median 10 sprouts), PC12 (median 9), PC21 (median 9) and PC31 (median 8), the lower whisker (0) indicate that during the evaluation period there were biases in some individuals and no regrowth was obtained.

Viable stem cuttings

Overall, obtaining viable stem cutting during the productivity evaluation behaved as follows: In the first evaluation, 18 viable stem cutting were obtained on average, 15 stem cutting in the second evaluation, and 21 viable stem cutting in the third evaluation and 23 viable stem cutting on average in the fourth evaluation which ended in December with 20 viable stem cutting. Figure 3 shows the behavior and trend of the number of regrowth variable of each clone according to the progress of the evaluation.

Three homogeneous groups formed in which the best group was the UP1 clone with an average of 34.05 viable cuttings (A), followed by the UG2 clone with average 22.96 viable cuttings (B) and group C by the PC12 clone (14.96 viable cuttings), PC31 (14.68 viable cuttings), PC30 (13.80 viable cuttings) and PC11 (11.84 viable cuttings); clones

PC16, PC37, G2 and PC21 were heterogeneous (Table 6). Figure 4 summarizes the analyzed data by clone, the central tendency, and its means.

Wendling *et al.* (2003) obtained 5 to 7 cuttings per mini stain for each collection in higher production, and 1.2 to 3 in a lower production using four *Eucalyptus grandis* clones. Analysis of variance percentage of rooting

The results of the analysis of variance for the rooting percentage of the cuttings made of the mini garden are shown in Table 6, the values of F and their significance degree, with P<0.0001 showing highly significant differences. For verification, a Tukey's multiple comparison test was carried out (Table 6. Analysis of variance). Two homogeneous groups superior to the others were formed, where clone G2 reported 78.58%

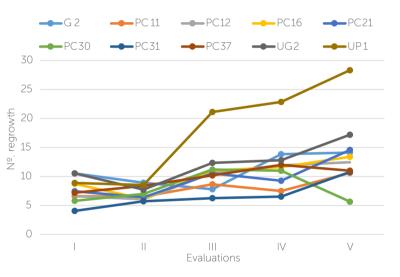


Figure 1. Average behavior of sprouts per clone according to the data collected during the evaluation period.

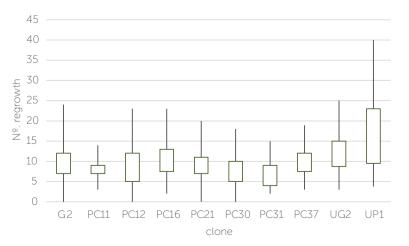


Figure 2. Number of sprouts per clone. Outliers represented by asterisks (*)

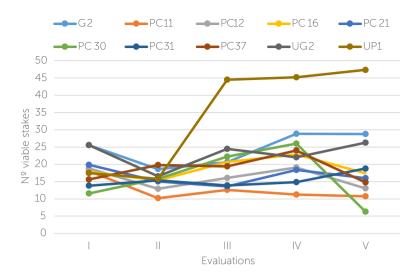
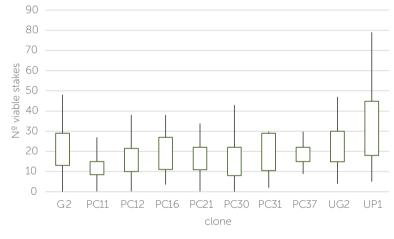


Figure 3. Behavior of the variable per clone for each evaluation.





rooting (A), followed by clone PC37 (30.36%), PC30 (19.58%) and PC16 (19.34), in the heterogeneous group (A, B and C) constituted by clones UP1, UG2, PC11, PC31, PC12, PC21 with only 68.75, 68.00, 46.44, 42.76, 34.39 and 33.32 percent rooting.

According to Arnold *et al.* (1991) and Leakey (1985) the second preselection level occurs in clonal propagation, using only those clones with greater than 70% rooting capacities and according to the data obtained, the clones G2, UG2 and UP1 would pass (Figure 5).

Monitoring of pH and the irrigation water C.E. allows to assess their values and, where appropriate, correct them to adapt them to 0.5 mS/cm EC and pH 5 to 7, due to the relation with the mother plants intake, reflected in the number

Classe	Module				
Clone	% rooting	CV (%)			
UP1	68.75 ^{AB}	22.16			
G2	78.58 ^A	13.44			
UG2	68.00 ^{AB}	29.70			
PC11	46.44 ^{ABC}	43.57			
PC12	34.39 ^{BC}	41.85			
PC21	33.32 ^{BC}	56.67			
PC16	19.34 ^C	70.15			
PC30	15.67 ^C	78.53			
PC37	30.36 ^C	44.05			
PC31	42.76 ^{ABC}	59.59			

7.96

< 0.0001

Table 6. Analysis of variance and Tukey's multiple comparison test ($p \le 0.05$) of the rooting percentage.

of sprouts and rooted cuttings. According to the obtained results and the criteria of a clonal operative program, clone G2 reported the adequate rooting percentage; However, UP1 and UG2 clones present an average close to the adequate, therefore, these three clones are suitable to start the clonal operating program of the Proplanse company; the values of the variables in the rooting phase are within the recommended range for clonal propagation utilizing cuttings.

CONCLUSIONS

F

p-value

The implementation of a clonal *Eucalyptus* miniature garden system with high cuttings productivity and

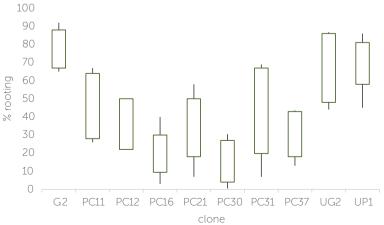


Figure 5. Rooting Percentage per clone, obtaining three highly significant clones; UG, UG2 and UP1.

good rooting capacity for seedling production is feasible. According to the analysis of the parameters measured in the clonal miniature garden, there is a relationship between viable cuttings and the number of shoots according to the clone, in such a way that three superior clones were identified (UP1 (17,947 shoots and 34.05 viable cuttings), UG2 (12,120 shoots and 22.96 viable cuttings) and G2 (10,254 shoots and 23.96 viable cuttings).

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