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Initial growth of eucalyptus plants treated with gibberellin

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This study was aimed at studying the effect of gibberellin (GA_3) on the growth of Eucalyptus plants. The experiment was conducted with 90-day-old seedlings on full sun bench at Goiás State University, Brazil, following the completely randomized design in a 5 × 2 factorial arrangement (five *eucalyptus* samples and two treatments with gibberellins). The application of gibberellin to eucalyptus is a promising practice, as it significantly affects the vegetative growth and increases the stem biomass accumulation.

Key words: Hormone, silviculture, early growth.

INTRODUCTION

The ongoing increase in economic value and the scarcity of hardwoods have boosted the diversified use of eucalyptus (Souza et al., 2012). The productivity of plantations in Brazilian territory is superior to that of traditional countries such as Australia (center of the species origin), and its current situation provides opportunities for the consolidation of Brazil as one of the major powers in the world's forest-based industry. The forestry sector accounts for 3.5% of the country's Gross Domestic Product (GDP) and generates 4.7 million direct and indirect jobs (Abraf, 2013).

Notwithstanding Brazil's high potential in the forestry sector, the country can further expand production and transfer wealth to other segments of economy. And in order to remain competitive, carrying on this process in a consistent and sustainable manner, it requires the development of research aimed to increase productivity, which will open new markets, increase exports and simultaneously modernize and ensure high social and environmental standards to forestry activities (Ferreira et al., 2012).

Reducing the cutting age and increasing the biomass accumulation of eucalyptus forests will contribute to the sector development and to the growth of Brazil's participation in the world market. However, the production of trees suitable for short-time harvesting will certainly depend on physiological changes in the species and/or adoption of new management techniques.

The use of plant growth regulators in agriculture has been an important management technique, as it has

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increased the productivity of several species, still its application is not yet a common practice in cultures that have not reached high technological level.

There is great interest in understanding the interference of plant growth regulators in lignification, wood density, early growth and yield (Pereira et al., 2011), since their performance does not depend solely on chemical composition, but also on how they are "perceived" by target tissues. In other words, the same substance can cause different effects depending on the tissue or organ upon which it is acting, on hormone concentration, and on the time of development of the same tissue (Wei et al., 2012). The use of growth regulators to improve wood quality, induce flowering, slow down or accelerate the vegetative growth of several species has been a constant issue in scientific research (Doorn et al., 2011; Kiba and Sakakibara, 2010; Xiong et al., 2009; Pereira et al., 2011).

The current world scenario is characterized by increasing environmental pressure towards the exploitation of renewable energy sources rather than fossil fuels. And in this direction, scientific understanding of how physiological aspects guide the growth of the species will contribute to greater production of early growth plants. However, little is known about the biochemical action and physiology of growth regulators in eucalyptus. Therefore, the present study was designed to investigate the effect of gibberellin (GA_3) on eucalyptus plant growth, with a view to reducing the cutting age and accelerating biomass accumulation.

MATERIALS AND METHODS

Experimental design

The work was accomplished at Goiás State University, Ipameri Unit (17º43'19"S, 48º09'35"W, Alt. 773 m), located in the municipality of Ipameri, Goiás, a region with predominant Aw climate according to the Köppen classification. The experiment was carried out on a bench exposed to full sun, following the completely randomized design in a 5 × 2 factorial arrangement (five eucalyptus samples, including three clones from the crossing of *Eucalyptus grandis* x *Eucalyptus urophylla* "*Eucalyptus urograndis* GG100", "*Eucalyptus urograndis* H13" and "*Eucalyptus urograndis* Super Clone"; one clone derived from the crossing between *Eucalyptus urophylla* x *Eucalyptus camaldulensis* "*Eucalyptus urocan* 58", and one sample from seeds of *Eucalyptus citriodora* x two treatments with 100 and 0 mg L^{-1} of gibberellins) and three replications. The eucalyptus seedlings were irrigated daily to maintain soil moisture near field capacity. At 90 days after germination the seedlings were transplanted and cultivated in six-liter pots with soil, sand and manure at 3:1:0.5 proportion, respectively. After substrate analysis, pH correction to 6.0 and fertilization were carried out. The foliar applications of GA_3 at 100 mg L^{-1} concentration and 50 ml per plant were accomplished on April 28 and 30 and May 28 and 30, 2012 at 100, 101, 129 and 130 days after germination. The plants that did not receive gibberellin were sprayed with deionized water in similar amount to that used for the regulator application. Maximum uniformity was sought in the application of gibberellin via spraying on all leaf area using a metering valve attached to a backpack sprayer. At 160 days after emergence the plants were analyzed.

Growth variables

The number of leaves and the stem length and diameter were measured between 8 and 10 am using a graduated ruler and calipers. The leaves, roots and stems were removed and put to dry in an oven at 72°C until steady dry mass state, and then weighed separately. Based on the dry mass data, the leaf mass ratio (LMR), root mass ratio (RMR), stem mass ratio (SMR), shoot/root system ratio (S/RS) and total biomass (TB) were calculated.

Fully expanded leaves were used to obtain the specific leaf area (SLA) and the leaf area (LA). Six discs with 14-mm diameter and known area were collected from each fully expanded leaf and dried at 70°C for 72 h, after which mass and SLA were measured. The LA was obtained by measuring the length and diameter.

Photosynthetic pigments

In order to determine the total chlorophyll and carotenoid concentration values, leaf discs with known area were removed and placed in glass containing dimethyl sulfoxide (DMSO). Subsequently, extraction was accomplished in water bath at 65°C for one hour. Aliquots were extracted for spectrophotometric reading at 480, 649.1 and 665 nm. The contents of chlorophyll a (Cl *a*), chlorophyll b (Cl *b*) and carotenoids were determined according to the equation proposed by Wellburn (1994).

Statistical procedures

The experiment was set up following the completely randomized design in a 5 × 2 factorial arrangement with three replications, and the data was submitted to variance analysis and Newman Keuls test for multiple comparisons of treatment averages using SISVAR 5.3 software (Ferreira, 2011).

RESULTS

Statistical analysis showed significant differences between treatments and samples for all variables (Tables 1 and 2). Only for the reason of shoot/root system (S/RS) and because of the root mass was no interaction treatment x samples (Tables 3 and 4). The average leaf number values varied significantly among the samples, where the *Eucalyptus citriodora* species had the lowest number (55.16) compared to that of the other samples, which were similar. However, the *Eucalyptus urocan* 58 species presented a greater number of leaves (138.50) compared to the others.

There was statistical difference in the number of branches, where the *E. citriodora* sample had a smaller number (5.16) compared to the others, which had similar amounts. However, the *Eucalyptus urograndis* GG100 sample stood out (14.00) among the others, presenting the greatest numerical amount of branches. Regarding the plant height, all samples showed high growth, highlighting the *E. urograndis* GG100 hybrid, which differed significantly from the others, with 113.83-cm height. The samples treated with GA_3 were statistically superior in height (104.46) compared to those not treated with gibberellin (Table 1).

The samples did not show any significant differences in

Table 1. Summary of the variance analysis and mean square test for number of leaves, branches, plant height, stem diameter (Ø), leaf area (LA) and specific leaf area (SLA) of different eucalyptus samples treated with GA_3.

Source of variation	DOF	Mean square					
		No. of leaves	No. of branches	Height cm)	Ø (mm)	LA (cm^2)	SLA (m^2kg^{-1})
Sample	4	6335*	70*	1879*	3.3	465*	53
Treatment	1	710	0.30	13525*	21*	0.96	147
Sample treatment*	4	555	13	222	0.46	44	20
Error	20	551	6.5	104	1.2	101	47
CV (%)		20	23	12	14	33	27
Treatment				**Averages**			
Without GA_3		107.3^A	11.00^A	62.0^B	6.8^B	30.3^A	23.2^A
With GA_3		107.1^A	11.2^A	104.5^A	8.5^A	30.6^A	27.6^A
Sample				**Averages**			
E.citriodora		55.2^B	5.2^B	68.3^B	6.6^A	38.9^A	22.2^A
E.urograndis GG100		116.0^A	14.0^A	113.8^A	7.5^A	24.2^{AB}	25.5^A
E.urocan 58		138.5^A	12.3^A	75.6^B	7.4^A	20.3^B	27.4^A
E.urograndis H13		129.6^A	12.3^A	80.6^B	8.3^A	28.6^{AB}	29.3^A
E.urograndis super clone		127.6^A	11.6^A	77.6^B	8.5^A	40.1^A	29.3^A

Values represent the arithmetic average. Averages followed by the same letter within each column do not differ among themselves, at 5% probability by Newman Keuls test. * = Significant.

Table 2. Summary of variance analysis and average test for shoot/root system ratio (S/RS), total biomass (TB), leaf mass ratio (LMR), stem (SMR) and root (RMR), leaf carotenoid concentration (Car) and total chlorophyll (Chl *a*+*b*) of different *eucalyptus* samples treated with GA_3.

Source of variation	DOF	Mean square						
		S/RS	TB (g)	LMR	SMR	RMR	Car ($g\ kg^{-1}$)	Chl *a*+*b* ($g\ kg^{-1}$)
Sample	4	1.6	141*	0.004*	0.006	0.007*	0.34*	1.4
Treatment	1	0.05	51	0.15*	0.14*	0.0007	0.08	2.0
Sample treatment*	4	3.2*	19	0.002	0.006	0.009*	0.17	0.5
Error	20	0.95	39	0.0008	0.0012	0.002	0.12	1.1
CV (%)		28	26	7	10	17	20	14
Treatments					**Averages**			
Without GA_3		3.4^A	22.0^A	0.48^A	0.28^B	0.23^A	1.57^A	7.53^A
With GA_3		3.5^A	24.6^A	0.34^A	0.41^A	0.24^A	1.68^A	7.02^A
Sample					**Averages**			
E. citriodora		3.9^A	15.0^A	0.45^A	0.34^B	0.21^B	1.4^B	7.6^A
E. urograndis GG100		3.9^A	25.8^A	0.39^B	0.40^A	0.21^B	2.0^A	7.6^A
E. urocan 58		2.9^A	23.0^A	0.39^B	0.32^B	0.29^A	1.6^{AB}	7.6^A
E. urograndis H13		3.3^A	26.9^A	0.41^B	0.36^B	0.23^{AB}	1.7^{AB}	6.6^A
E. urograndis Super Clone		2.9^A	25.7^A	0.41^B	0.33^B	0.26^{AB}	1.4^{AB}	6.9^A

Values represent the arithmetic average. Averages followed by the same letter within each column do not differ among themselves at 5% probability by Newman Keuls test. * Significant.

the stem diameter (Table 1), however, there was a numerical variation of 1.82 mm between the *E. urograndis* Super Clone and the *E. citriodora* species samples. Regardless of the sample, the plants treated with GA_3 showed longer diameter (8.51 mm), with 1.68-mm increase compared to untreated plants (6.83 mm). The leaf area varied among the different samples, highlighting larger leaf area in *E. urograndis* Super Clone (40.16) and smaller leaf area in *E. citriodora* (20.37). However, the treated and untreated plants had almost the

Table 3. Breaking down of S/RS of different eucalyptus samples treated with GA_3.

Sample	Without GA_3 spraying	With GA_3 spraying
E. citriodora	3.30^{aA}	4.67^{aA}
E. urograndis GG100	3.30^{aA}	4.54^{aA}
E. urocan 58	4.01^{aA}	1.81^{bB}
E. urograndis H13	3.46^{aA}	3.17^{abA}
E. urograndis Super Clone	2.79^{aA}	3.08^{abA}
CV (%)	28.51	

Different lowercase letters indicate significant differences between the samples; different capital letters indicate significant differences between treatments in each sample by Newman Keuls test at 5% significance level.

Table 4. Breaking down of RMR of different eucalyptus samples treated with GA_3.

Sample	Without GA_3 spraying	With GA_3 spraying
E. citriodora	0.230^{aA}	0.183^{aB}
E. urograndis GG100	0.216^{bA}	0.356^{aA}
E. urocan 58	0.233^{aA}	0.190^{aB}
E. urograndis H13	0.223^{aA}	0.240^{aB}
E. urograndis Super Clone	0.266^{aA}	0.250^{aB}
CV (%)	17.55	

same leaf area, and they did not differ statistically. Regarding the specific leaf area there was no difference between the samples and treatments with GA_3 (Table 1).

The variance analysis and average test show interaction between the shoot/root system ratio (S/RS) factors, as shown in Table 2. In numerical values, the highest ratio was obtained in *Eucalyptus citriodora* (4.67) treated with GA_3, and the lowest ratio was recorded in *Eucalyptus urocan* 58 (1.81), and even with this difference in average values, there was no statistical difference between samples. However, *Eucalyptus urocan* 58 had the lowest values among the samples treated with GA_3.

The total biomass of samples differed statistically, and *E. citriodora* had the lowest total biomass (15.03). The leaf mass ratio was higher in plants not sprayed with GA_3, however, the plants treated with GA_3 showed higher stem mass ratio, with *E. urograndis* GG100 showing the highest numerical value. The root mass ratio showed slight variation between samples, with *Eucalyptus urocan* 58 presenting the highest numerical value in relation to the other samples, but without statistical difference between plants treated with GA_3 and untreated ones.

The samples differed statistically in carotenoid concentration, where the *E. urograndis*. GG100 sample presented higher average (1.975) than the others. There was no significant difference between samples treated with g GA_3 and untreated ones.

When the sample treatment results are broken down, it is noted that regardless of the eucalyptus sample, the highest numerical values were obtained in plants treated with GA_3 (Table 4).

Different lowercase letters indicate significant differences between the samples; different capital letters indicate significant differences between treatments in each sample by Newman Keuls test at 5% significance.

DISCUSSION

Physiological analysis of the plants at an early stage helps to identify promising species with high productivity potential and contributes to the choice for suitable materials for the prevailing conditions (Peixoto et al., 2006). Originally the hybrid clones and *E. citriodora* showed marked differences in vegetative growth. The variations recorded in the samples growth and development are determined by genetic characteristics, however, the GA_3 applications intensified the vegetative growth and promoted significant changes in the allocation of assimilates and biomass partition, as discussed below.

The application of GA_3 did not cause significant changes in the vegetative growth of the studied samples. The similarity in the number of leaves, branches, leaf area, specific leaf area, biomass, leaf and root mass ratio, chlorophyll and total carotenoids between plants treated with GA_3 and untreated ones indicate that the time between regulator application and evaluations was short for vegetative growth and changes in anatomical variables, such as specific leaf area. Despite the lack of statistical difference, the trend of specific leaf area increase in plants treated with GA_3 is possibly related to higher leaf transmittance of solar radiation in the canopy. Increased leaf transmittance allows greater solar radiation distribution in the canopy, providing light energy to lower leaves, contributing to increased photosynthetic activity and biomass accumulation over time. Increases in the specific leaf area, and reductions in leaf thickness decrease absorption and increase leaf transmittance, changing the solar radiation distribution pattern in the canopy (Borges et al., 2014). Greater biomass in plants treated with GA_3, even with no statistical difference, is an indication that a longer time interval between application and evaluation is necessary to record any difference.

The notable changes in vegetative growth are due to the differences among the samples used. *E. urograndis* Super Clone stood out among the samples for presenting high number of leaves, large leaf area and specific leaf area, and great biomass. However, *E. urograndis* GG100 was the sample with the most desirable features for forestry due to its higher biomass distribution to the stem resulting in greater height and large stem diameter. Larger stem diameter in *E. urograndis* GG100 clones has

been observed in experiments and appears to be a significant variable of the sample (Pinto et al., 2011).

Greater plant height, stem diameter and stem mass ratio in plants treated with GA_3 indicate that the growth regulator caused changes in biomass distribution with greater assimilates allocation to the stem to the detriment of the root system and mainly of the leaves. The high stem growth in height and diameter is of great interest to forestry as it directly affects the eucalyptus early growth and wood quality and productivity. The application of GA_3 changes the vegetative growth, intensifying the development of shoots (Sponsel, 2006). The absence of difference in photosynthetic pigment concentration and leaf area is evidence that there was no change in the photosynthetic mechanism to maximize photosynthesis, the existing differences being related to changes in biomass distribution. These changes may be associated to the fact that hormones generally act upon cellular communication, since they are defined as “chemical messengers produced in a cell or tissue that modulate cellular processes in other cells, by interacting with specific proteins called receptors”, contributing to metabolic processes that allow the maintenance and growth of primary organs (Taiz and Zeiger, 2013).

Higher S/RS ratio in plants treated with gibberellin is because they allocate greater biomass to the stem, thus increasing the shoot mass. Lower RMR in treated plants is due to the same fact, as larger investment in the shoot took place at the expense of smaller biomass distribution to the root. Greater translocation of assimilates to the shoot can be beneficial under adequate soil fertility conditions because of higher stem and leaf development.

Biomass production is determined by the species genetic characteristics (Santana et al., 2002). The remarkable differences in vegetative growth indicate that the species have genetic variability. Regardless of the treatment with GA_3, the *E. citriodora* species showed less vigorous vegetative growth than the hybrids. The *E. urograndis* GG100 clone showed high biomass distribution to the stem, proving to be a promising material for future studies. The application of GA_3 in eucalyptus plants is a promising practice, as it significantly affects vegetative growth and enhances stem biomass accumulation (the element of economic interest).

Conclusions

1. The application of GA_3 in *Eucalyptus* seedlings accelerates vegetative growth, changes biomass distribution and possibly interferes with the early growth and wood productivity.
2. E. citriodora was the sample with the least vigorous vegetative growth.
3. The *E. urograndis* GG 100 hybrid clone stood out from the others for showing the desirable features for forestry, such as greater stem mass ratio, height and stem diameter.

Conflict of Interest

The authors have not declared any conflict of interest.

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REFERENCES

Associação Brasileira de Produtores de Florestas Plantadas (ABRAF) (2013). Available at: www.abraflor.org.br Acessed on: 16 May.
Borges LP, Torres Jr HD, Neves TG, Cruvinel CKL, Santos PGF, Matos FS (2014). Does Benzyladenine Application Increase Soybean Productivity. Afr. J. Agric. Res. 9:2799-2804.
Doorn WGV, Perik RRJ, Abadie P, Harkema HA (2011). treatment to improve the vase life of cut tulips: Effects on tepal senescence, tepal abscission, leaf yellowing and stem elongation. Postharv. Biol. Technol. 61:56–63.
Ferreira SM, Petrauski C, Marques GM, Silva ML, Cordeiro SA, Soares NS (2012). Competitividade do Brasil no mercado internacional de madeira serrada. Cerne 18:99-104.
Ferreira DF (2011). Sisvar: a computer statistical analysis system. Ciênc. Agrotecnol. 35:1039-1042.
Kiba T, Sakakibara H (2010). Role of cytokinin in the regulation of plant development. In: PUA, E.C, DAVEY, M. R. Plant developmental biology-biotechnological perspectives. Springer, New York pp. 237-254.
Peixoto CP, Cerqueira EC, Soares Filho WS, Castro Neto MT, Ledo CAS, Matos FS, Oliveira JG (2006). Análise de crescimento de diferentes genótipos de citros cultivados sob déficit hídrico. Rev. Bras. Frutic. 28:439-443.
Pereira RPW, Abreu HS, Monteiros BO, Souza ND (2011). Variação ligno-anatômica em mudas de *eucalyptus grandis* tratadas com reguladores de crescimento. Cerne 17:369-375.
Pinto SIC, Furtini Neto AE, Neves JCL, Faquin V, Moretti BS (2011). Eficiência nutricional de clones de eucalipto na fase de mudas cultivados em solução nutritiva. Rev. Bras. Ciênc. Solo 35:523-533.
Santana RC, Barros NF, Neves JCL (2002). Eficiência de utilização de nutrientes e sustentabilidade da produção em procedências de *Eucalyptus grandis* e *Eucalyptus saligna* em sítios florestais do Estado de São Paulo. Rev. Árvore 26:447-457.
Souza JT, TrevisaN R, Denardi L, Stangerlin DM, Vivian MA, Haselein CR, Santini JE (2012). Qualidade da madeira serrada provenientes de árvores dominantes e média de *Eucalyptus grandis* submetidos a secagem. Cerne 18:167-174.
Sponsel V (2006). Gibberellins: Regulators of plant height and seed germination. Plant Physiol. 4:509-538.
Taiz L, Zeiger E (2013). Fisiologia vegetal. Porto Alegre: Artmed. 954p.
Wei ZZ, Luo LB, Zhang HL, Xiong M, Wang X (2012). Identification and characterization of 43 novel polymorphic EST-SSR markers for arum lily, *Zantedeschia aethiopica* (Araceae). Am. J. Bot. 99:493–497.
Wellburn AR (1994). The spectral determination of chlorophylls a and b, as well as total carotenoids, using various solvents with spectrophotometers of different resolution. J. Plant Physiol. 144:307-313.
Xiong G, Li J, Wang Y (2009). Advances in the regulation and crosstalks of phytohormones. Chin. Sci. Bull. 54:2718–2733.