EFFECT OF GIBBERELLIC ACID AND THE BIOSTIMULANT STIMULATE® ON THE INITIAL GROWTH OF TAMARIND[1]

ANA CRISTINA VELLO LOYOLA DANTAS[2], JOÃO MARIANO DE OLIVEIRA QUEIROZ[3], ELVIS LIMA VIEIRA[2], VANESSA DE OLIVEIRA ALMEIDA[4]

ABSTRACT- Plant growth regulators and biostimulants have been used as an agronomic technique to optimize the production of seedlings in various crops. This study aimed to evaluate the influence of gibberellic acid and the biostimulant Stimulate® on the initial growth of tamarind (*Tamarindus indica* L.). The experiments were conducted in a nursery with 50% shading, in a randomized block design with five replications and five plants per plot. Thirty eight days after sowing, the leaves were sprayed seven times a day with 0.0 (control), 0.8, 1.6, 2.4 and 3.2 mL of gibberellic acid L^{-1} aqueous solution and with 0.0 (control), 6.0,12.0, 18.0, and 24.0 mL Stimulate® L^{-1} aqueous solution. Stem diameter (SD), plant height (PH), longest root length (LRL), shoot dry mass (SDM), root dry mass (RDM) and RDM:SDM ratio were evaluated ninety days after sowing. Variance and regression analysis showed that GA_3 at 4% promoted plant growth (height), but had no significant effect on stem diameter, longest root length, shoot and root dry mass and the RDM:SDM ratio. On the other hand, all concentrations of Stimulate® significantly increased plant height and shoot and root dry mass of tamarind seedlings.

Index terms: *Tamarindus indica L.*, plant growth regulators, seedling production.

INFLUÊNCIA DO ÁCIDO GIBERÉLICO E DO BIOESTIMULANTE STIMULATE® NO CRESCIMENTO INICIAL DE TAMARINDEIRO

RESUMO- O emprego de reguladores e bioestimulantes vegetais como técnica agronômica para se otimizar a produção de mudas tem sido utilizado em diversas culturas. Este trabalho teve como objetivo, avaliar a influência do ácido giberélico e do bioestimulante Stimulate®, isoladamente, no crescimento inicial de plantas de tamarindeiro (*Tamarindus indica* L.). Os experimentos foram conduzidos em viveiro telado com 50% de sombreamento, em delineamento em blocos casualizados, com cinco repetições e cinco plantas por parcela. As pulverizações foram iniciadas aos 38 dias após a semeadura (DAS), e cada produto foi ministrado em sete pulverizações foliares diárias, nas concentrações de 0,0 (controle - água), 0,8; 1,6;2,4 e 3,2 mL de ácido giberélico L^{-1} de solução aquosa e 0,0 (controle - água), 6,0; 12,0; 18,0 e 24,0 mL de Stimulate® L^{-1} de solução aquosa. Aos 90 DAS, foram avaliados: o diâmetro do caule, altura da planta, comprimento da maior raiz, massa seca da parte aérea (MSPA) e da raiz (MSR) e a relação MSR/MSPA. Os dados foram submetidos à análise de variância e análise de regressão para os tratamentos, em isolado, GA_3 4% e Stimulate®. A pulverização foliar com ácido giberélico (GA_3 4%) é eficiente para promover o crescimento em altura de plantas de tamarindeiro, no entanto não promove efeitos significativos sobre o diâmetro do caule, crescimento da maior raiz, massa seca da parte aérea, raiz e na relação MSR/MSPA. Todas as concentrações utilizadas de Stimulate®, via pulverização foliar, promovem o incremento na altura de planta, massa seca da parte aérea e da raiz de plantas de tamarindeiro.

Termos para indexação: *Tamarindus indica*, reguladores vegetais, produção de muda.

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[2]Agr. Eng. Dr., Centro de Ciências Agrárias, Ambientais e Biológicas, Universidade Federal do Recôncavo da Bahia (CCAAB/UFRB). Câmpus Universitário. CEP: 44380-000, Cruz das Almas-BA. E-mails: acloyola@ufrb.edu.br; elvieira@ufrb.edu.br
[3]Biologist. MSc. in Agricultural Sciences/UFRB. Prof. Instituto Federal de Educação, Ciência e Tecnologia Baiano, Câmpus Catu. Rua Barão de Camaçari, 118. CEP: 48110-000, Catu-BA. E-mail: joaomqueiroz@ig.com.br
[4]MSc in Plant Genetic Resources. CCAAB/UFRB. Câmpus Universitário. CEP:44380-000, Cruz das Almas-BA. E-mail: voagro@click21.com.br

INTRODUCTION

The tamarind (*Tamarindus indica* L.) belongs to the Dicotyledonous family Leguminosae native to Equatorial Africa, India and Southeast Asia. It grows in tropical and subtropical regions, but does not survive at low temperatures (PATHAK et al., 1991). The tree provides wood, fruits, seeds, forage, medicinal compounds and has industrial potential. It can be grown commercially or used in agroforestry systems, but its growth is slow (PEREIRA et al., 2007).

Seedling production is the first stage to obtain commercially feasible cultures. Thus the importance of research that aims to increase plant initial growth rate. Souza et al. (2007) studied the effect of different nitrogen and phosphorus concentrations on the production of tamarind seedlings, and Silva et al. (2011) used chemical treatments to increase seed germination and initial growth rate, but their results were not satisfactory in terms of aerial part length.

Plant growth regulators have been steadily used in various cultures as an agronomic technique to increase production. Research on plant growth regulators focuses on mastering and controlling physiological processes and the results have been surprising (DAVIES, 2004).

Plant growth regulators are organic compounds, such as auxins, cytokines, gibberellins and ethylene, produced by plants (SILVA;DONADIO, 1997). In low concentration they promote, inhibit or modify physiological and morphological processes of growth and development (VIEIRA; CASTRO, 2001).

Gibberellins also affect plant growth when applied exogenously, alone or associated with other plant growth regulators such as auxins and cytokines which affect stem growth through cell elongation and division, as well as fruit development (DAVIES, 2004).

Relevant physiological effects of exogenous biostimulants such as Stimulate® present cytokinin, auxin and gibbrellin in its composition. Biostimulants such as Stimulate® may, depending on its composition, concentration and proportion of compounds, increases plant growth and development by enhancing cell division, differentiation and enlargement. It can also improve nutrient uptake and utilization. Stimulate® is especially effective when applied with foliar fertilizers, and it is also compatible with pesticides (STOLLER DO BRASIL, 1998).

Plant hormones are involved in every aspect of plant growth and development. Natural or synthetic hormones can be applied on leaves, fruits and seeds to promote physiological changes in seed germination, seedling vigor, growth and development of roots and leaves as well as an increase in organic matter (VIEIRA; CASTRO, 2004). The application of growth regulators during the initial stages of plant development promotes root growth; allows rapid recovery after hydric stress; increases resistance to insects, pests, diseases and nematodes, and promotes rapid and uniform plant establishment which improves nutrient absorption and yield.

Tamarind is well adapted in the Northeast of Brazil, but further research is required to improve its commercial potential. The success of a commercial culture depends on the amount of seedlings that can be produced in a short period of time. Thus, the aim of this study was to evaluate the influence of gibberellic acid and bioestimulant Stimulate® spray on the initial growth of tamarind plants.

MATERIAL AND METHODS

Experiments were conducted at the Universidade Federal do Recôncavo da Bahia, (12°40’0” S and 39°06’0” W, 200 m above sea level) (SEI, 2010). The climate, according to Koppen’s classification system, is tropical humid (Aw and Am).

Plants were grown in a nursery with 50% of shading, from February to April, 2009. Seeds were extracted manually from ripe fruits produced by a 25 year-old tree. The seeds were washed with tap water until complete endocarp removal and dried for 24 hours in the shade. Previous to sowing, selected seeds were soaked in water for 2 hours following Pereira et al. (2009). Seeds were sowed in black polyethylene bags (32 cm x 20 cm x 0.15 mm) that contained soil and manure (3:1) with the following chemical composition: pH 6.5; P 42.0 mg dm^{-3}; K^+ 1.54 mg dm^{-3}; Ca + Mg 2.9 $cmol_c$ dm^{-3}; Ca^{2+} 1.8 $cmol_c$ dm^{-3}; Mg^{2+} 1.1 $cmol_c$ dm^{-3}; Al^{3+} 0.0 $cmol_c$ dm^{-3}; Na 0.14 $cmol_c$ dm^{-3}; H+Al 2.20 $cmol_c$ dm^{-3}; S 4.58 $cmol_c$ dm^{-3}; CTC 6.78 $cmol_c$ dm^{-3}; V 68.0%; and organic matter 15.03 g kg^{-1}. Three seeds were planted per bag, and after thirty days just one plant per bag was left.

Two experiments were performed to evaluate the isolated effect of gibberellic acid (GA_3 4%) and Stimulate®, both applied as foliar spray. GA_3 4% is a liquid plant growth regulator that contains gibberellic acid. Stimulate® composition includes 0.009% of kinetin; 0.005% of gibberellic acid; 0.005% of indolbutyric acid and 99.98% of inactive ingredients (STOLLER DO BRASIL, 1998).

Thirty eight days after sowing (DAS), the plants with 6 to 8 leaves were distributed in 25 blocks to facilitate foliar-directed spraying during the early hours of the day (between 6:00 and 7:00 am). Spraying was performed with a manual sprayer and no vegetal oil was added to the solutions. Seven sprayings were applied on the whole plants every 24 hours. Each block received approximately one liter of the corresponding solution. After an eight hours period, the plants were watered in order to keep the substrate wet.

After spraying, the plants were placed in a randomized block design with five replications, five plants per plot and five treatments per experiment, as follows: experiment 1 – five concentrations of GA_3 at 4%: T1 – 0.0 (control - water), T2 – 0.8 (32 mg of GA_3 L^{-1}); T3 – 1.6 (64 mg of GA_3 L^{-1}); T4 – 2.4 (96 mg of GA_3 L^{-1}) and T5 – 3.2 mL (128 mg of GA_3 L^{-1}) in aqueous solution; experiment 2 – five concentrations of Stimulate®: T1 – 0.0 (control - water); T2 – 6.0; T3 – 12.0; T4 – 18.0, and T5 – 24.0 mL in aqueous solution.

Stem diameter (SD) at the hypocotyl; plant height (PH) measured from the base of the stem to the last leaf insertion; longest root length (LRL), from the calyptra to the base of the stem; shoot and root dry mass (SDM, RDM) and RDM:SDM ratio were determined 90 days after sowing. Dry mass was obtained by drying the material in a forced-draft oven for 72 hours at 65 ºC ± 5 ºC (BENINCASA, 1988) and then weighing it in a four-digit scale.

Analysis of variance (F test) and polynomial regression were used to compare data. Statistical analysis was performed by using SISVAR (FERREIRA, 2003).

RESULTS S AND DISCUSSION

Experiment 1 – Gibberellic acid (GA_3 at 4%)

Gibberellic acid sprayed on tamarind seedlings had a positive effect on plant height (Table 1), but did not significantly affect the other traits analyzed.

The positive effect of gibberellic acid on plant growth has been reported by several authors, especially when applied during the initial developmental stages (COELHO et al., 1983; MODESTO et al., 1996; LEITE et al., 2003; LEONEL; PEDROSO, 2005). As shown in Figure 1, gibberellic acid, in all concentrations tested, promoted an increase in tamarind plant height. These results confirm the active role in cell division and elongation of this plant growth regulator (HIGASHI et al., 2002) which promotes cell wall extensibility (RAVEN et al., 2001).

The regression $\hat{Y} = -3.3246x^2 + 16.978x + 31.556$ (Figure 1) has an optimal correlation coefficient (R^2 = 91.4%) and indicates that the maximum plant height (53.22 cm) was obtained in the concentration of 2.60 mL gibberellic acid L^{-1} in aqueous solution (104 mg of GA_3 L^{-1}). This increase represents 12,8% (35,6 cm) in relation to the control (31.6 cm). When considering the highest concentration applied (3.2 mL of gibberellic acid L^{-1} in aqueous solution, 128 mg of GA_3 L^{-1}), the height increase was of 64,0% (20,2 cm) when compared to the control.

In terms of stem diameter (Table 1), the effect of gibberellic acid is not clear. Modesto et al. (1996), Ferreira et al. (2002) and Leonel and Pedroso (2005) reported that the plant growth regulator promoted an increase in stem diameter in *Passiflora alata*, *Citrus limonia* and *Annona squamosa*, respectively. On the other hand, Modesto et al. 1999 did not find increase of stem diameter when 150 mg of gibberellic acid L^{-1} in aqueous solution was applied to 'Cleopatra' tangerine. The authors attribute these results to the fact that the variety studied has a slow development. Tamarind is also a slow developing tree and presented a small increase in diameter, which was in average of 2.3 cm after 90 days.

Root length and root dry mass of cherimoya and sweet passiflora were not affected by GA_3. The growth reported by several authors such as Castro and Vieira (2001); Barros (2004); Santos and Vieira, (2005); Ferreira et al. (2007) and Prado Neto et al. (2007) was observed when gibberellic acid was applied on the seeds only, or on the seeds and leaves, which indicates the need of supplementary application.

According to Cato (2006), in order to promote an increase of the root system that causes the root:shoot ratio to differ from the control, it is necessary that the three bio-regulators (gibberellic acid, indolbutyric acid and kinetin) act synergistically. The experiments involved just one plant growth regulator which explains why the other variables did not respond to the presence of GA_3 at 4%.

Experiment 2 – Effect of Stimulate®

Analysis of variance showed that Stimulate® significantly affected plant height and shoot and root dry mass (Table 2). No significant increase in stem diameter and longest root length was observed.

In terms of plant height, the square regression equation (Figure 2) $\hat{Y} = 0.0057x^2 + 0.0172x + 34.277$ and the correlation coefficient (R^2) = 83.4% indicate that the maximum plant height (37,98 cm) was obtained in the concentration of 24 mL of Stimulate® L^{-1} in aqueous solution. The increase represents 11,7% (3,70 cm) when compared to the control (34,28 cm). Stimulate® increased plant height in all concentrations tested (Figure 2). Vieira et al. (2007)

and Almeida and Vieira (2009) also observed an increase in plant height when applying Stimulate® to *Nicotiana tabacum*. On the other hand, Albuquerque et al. (2004) and Echer et al. (2006) working with castor oil plant and yellow passiflora, respectively, found that Stimulate® did not affect plant height.

The linear equation for shoot dry mass (Figure 3) was $\hat{Y} = 0.0172x + 1.546$, ($R^2$= 88,2%), which indicates that higher concentrations should be used to obtain maximum values for that trait. All concentrations applied resulted in an increase of shoot dry mass when compared to the control. Similar results were obtained by Milléo et al. (2000) when using Stimulate® on maize cultures and by Santos (2004), who studied cotton plants.

Figure 4 shows the linear equation for root dry mass $\hat{Y} = 0.0033x + 0.314$, ($R^2$= 78.1%). The highest concentration of Stimulate® (24 mL L^{-1}) resulted in a dry mass increase of 20.1% when compared to the control. Our results are in agreement with Santos (2004) who observed that Stimulate® significantly increased root dry mass production of cotton plants.

The results indicate that Stimulate® improves physiological processes and promotes initial growth. Due to the balanced presence of plant growth regulators, Stimulate® has a synergistic effect which could be responsible for the increase in plant height, shoot dry mass and root dry mass observed in the experiments, indicating the effectiveness of the bio-stimulant. Stimulate® promotes growing and development by influencing division, differentiation and elongation of cells. It also increases nutrient absorption and use, and is especially efficient when applied with foliar fertilizers (CASTRO et al., 1998)**.** Notwithstanding**,** Stimulate® had no significant effect on stem diameter and longer root length.

It is important to observe that plant growth regulators may have different effects on plant organs. For instance, the same concentration of a certain hormone can cause stem growth and at the same time inhibit root growth (CASTRO;VIEIRA 2001). Hence, further research should be undertaken to determine the concentration of Stimulate® that promotes maximum growth of young tamarind plants in order to obtain high quality seedlings in a short period of time.

TABLE 1 - Analysis of variance comparing stem diameter (SD), plant height (PH), longest root length (LRL), shoot dry mass (SDM), root dry mass (RDM) and RDM:SDM ratio in tamarind (*Tamarindus indica* L.) sprayed with gibberellic acid (GA_3 at 4%).Cruz das Almas – BA, 2009.

Treatments	DF	Square Means					
		SD	PH	LRL	SDM	RDM	RDM/SDM
Block	4	0.014^{ns}	29.008^{ns}	20.164^{ns}	0.293^{ns}	0.001^{ns}	0.019^{ns}
Treatment	4	0.098^{ns}	438.297**	31.176^{ns}	0.302^{ns}	0.009^{ns}	0.007^{ns}
Error	16	0.039	11.864	23.121	0.108	0.005	0.007
CV (%)		8.35	7.50	13.49	24.72	23.88	34.02
Means		2.380	45.956	35.641	1.331	0.310	0.252

**Significant at 1% probability; ^{ns} not significant

TABLE 2 - Analysis of variance comparing stem diameter (SD), plant height (PH), longest root length (LRL), shoot dry mass (SDM), root dry mass (RDM) and RDM/SDM ratio in tamarind (*Tamarindus indica* L.) sprayed with Stimulate®. Cruz das Almas – BA, 2009.

Variation source	DF	Square Means					
		SD	PH	LRL	SDM	RDM	RDM/SDM
Block	4	0.022^{ns}	17.339^{ns}	42.825^{ns}	0.088^{ns}	0.001^{ns}	0.0007^{ns}
Treatment	4	0.088^{ns}	39.227*	47.177^{ns}	0.391*	0.023**	0.0010^{ns}
Error	16	0.030	1.373	24.518	0.105	0.005	0.001
CV (%)		7.33	9.02	13.36	19.38	21.64	17.28
Means		2.39	35.701	37.07	1.675	0.350	0.211

**Significant at 1% probability; *significant at 5% probability; $^{n/s}$not significant.

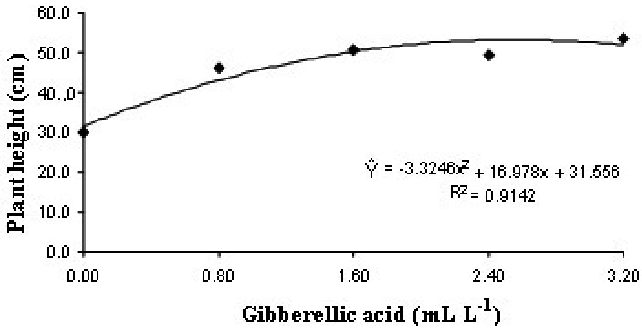

FIGURE 1 -Tamarind (*Tamarindus indica* L.) plant height sprayed with gibberellic acid.

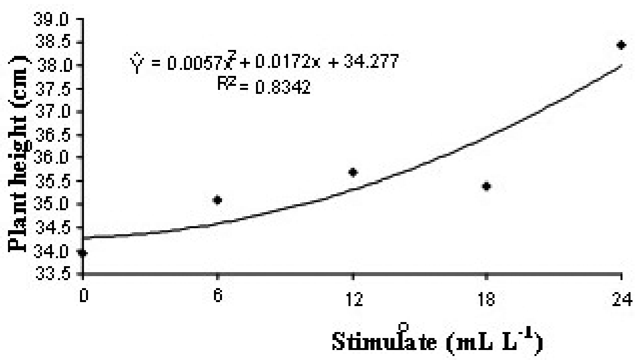

FIGURE 2 - Tamarind (*Tamarindus indica* L.) plant height sprayed with Stimulate®.

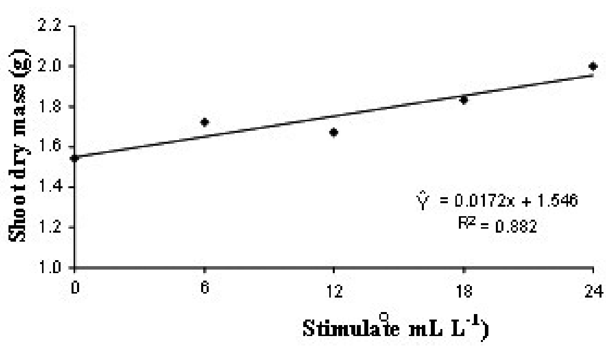

FIGURE 3 - Shoot dry mass of tamarind (*Tamarindus indica* L.) sprayed with Stimulate®.

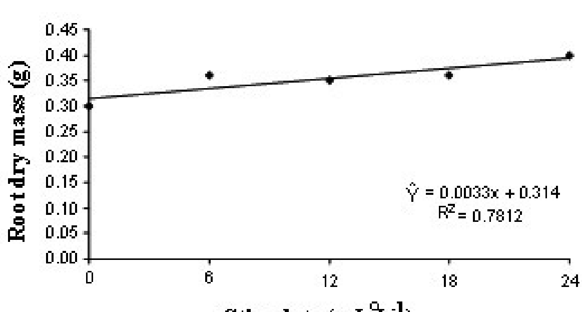

FIGURE 4 - Root dry mass of tamarind (*Tamarindus indica* L.) sprayed with Stimulate®.

CONCLUSIONS

1-Foliar spraying of young tamarind plants with gibberellic acid (GA_3 at 4%) promotes plant height increase.

2-Foliar spraying of young tamarind plants with gibberellic acid has no effect on shoot diameter, longest root length, shoot and root dry mass and root:shoot ratio.

3-Foliar spraying of young tamarind plants with biostimulant Stimulate® in all concentrations tested promotes an increase in plant height and of shoot and root dry mass.

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