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EFFECT OF GIBBERELLIC ACID ON "*IN VITRO*" GERMINATION OF RED STRAWBERRY GUAVAS (*Psidium cattleianum Sabine*)

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ABSTRACT
Red strawberry guava (*Psidium cattleianum* S.), Myrtaceae, is a southern Brazil native fruit specie. The fruits are mostly destined for fresh consumption. The species can be also used in environmental remediation activities of their intensive fruit and seed production. One of the main constraints for commercial production of red strawberry guava fruits is the obtaining of plants with desirable fruit characteristics and stabilized agronomical traits. So far, high heterogeneity in seed emergency has been observed increasing the production costs and time to establish orchards. The purpose of the study was to test the effect of gibberellic acid (Progibb) in the percentage of emergency and speed index (ESI) of "in vitro" strawberry guava seeds. Seeds were selected from the central area of the fruit and sown in LP 50% medium, with sucrose (10 g/L) and agar (0,8%) were added. The pH was adjusted to ±5.8 before autoclaving and then followed by incubation at 25°C ± 4° under 12 hours photoperiod. The isolated seeds were soaked for 18 hours in 0, 2, 10, 20 or 50 mg L-1 Exponential of the active ingredient in the solution. The experimental design was completely randomized and each treatment consisted of ten replications, and the experimental units consisted of one Petri dish containing five seeds each one. Averages were submitted to ANOVA and regression analysis and the statistical differences determined via Duncan test. ($p<5\%$). The red strawberry guava seeds had a emergency percentage of approximately 64%, delayed effects in E.S.I was observed at the concentration above of 10 mg.L-1 a.i, however, further studies are in need do confirm these preliminary results.

KEYWORDS: Plant growth regulator, seed germination, physiology, native fruits.

INTRODUCTION

The red strawberry guava tree (*Psidium cattleianum* Sabine) is a native fruit plant found in the south of Brazil, which is used in programs of environmental recuperation and reforestation. It doesn´t only serve as an important source of food for the fauna with which it is associated, but also for medicinal use (Crivelaro *et al*., 2010; Brighenti, *et al*., 2008, Jeong, et al, 2011) and natural consumption (EMBRAPA, 1984, Glufke, 1999). The possibility of maintaining the interesting characteristics of this specie generates the needs of developing techniques not only for macro propagation but also for propagation in vitro by the advantages that this last one can offer. The vegetative propagation by cutting or grafting, has not been successful for this specie (Donadio *et al*., 2004), however, the use of green cuttings showed itself high and efficient for its reproduction by a percentage on the obtained roots (Altoé *et al*., 2011). By its part, propagation *in vitro*, as basic tool for conservation, cloning, and another kind of studies with this specie, has not been studied to a great magnitude.

The obtaining of explants for use in tissue culture for this specie is affected mainly by fenolic oxidation and contamination associated with endogenous and exogenous microorganisms when the explant is obtained of an adult plant. In this case, one of the possibilities for preliminary studies is the obtaining of in vitro seedlings; but according with preliminary observations that specie presents heterogeneity in the emergency, attributable possibly to endogenous and/or exogenous mechanisms for survival. Between the endogenous conditions there is a possibility of finding the genetics, biological, physical and chemist-physiological as the presence of inhibitors alkaloids, essential oils, and unsaturated acids. Among the exogenous it will find the physics, chemicals and/or mechanics (Souza, 2001; Mendes, 2009). Beyond the previous one, in the case of positive photoblastics seeds as red strawberry guava, the germination can be affected by the absence of the *phytochrome* light stimulus, one of the intracellular proteins associated with the photo dependent mechanisms (Taiz and Zaiger, 2004, Franflin, 2009). In the context of seed physiology, gibberellins have been proposals exercising two main functions in the germination process: first, promoting gene expression that encode for hydrolytic enzymes of the endosperm and second stimulating directly the embryo growth. (Brady & McCourt, 2003). These molecules act like stimulus in germination by the induction of hydrolytic enzymes that break the tissues barriers, such as the endosperm and the testa seed, promoting in this way the breakdown and mobilization of reserve, beyond, the expansion of the embryo (Taiz & Zeiger, 2004; Henderson *et al*., 2004). However, its use still generates controversy, therefore studies in *Arabidopsis sp*., has shown that its isolated use does not provide similar results in all species of this genus (There-Rachedi *et al*., 2004; Finkelstn *et al*., 2004). Looking at viable alternatives for the commercial seedlings development of this specie, the goal of this

research was to test the red strawberry guava seed emergency on *in vitro* conditions submerged in different concentrations of giberellic acid (Progibb®).

MATERIAL AND METHODS

The experiment began in November of 2011 in the Biotechnology Laboratory in the department of Horticulture and Forestry at the Faculty of Agricultural science, Federal University of Rio Grande do Sul (UFRGS). The fruits from which the seeds were extracted were collected from a single tree, in the metropolitan region of Porto Alegre in October, 2011.

The seeds were collected from the equatorial region of fruits removing the mucilage that would re-covered them by abrasion with limestone and subsequently rinse for ten minutes followed by submersion in a solution of NaOCl (1%). Seeds were subsequently dried in the shade and then stored in a freezer (4ºC) for approximately one month (Figure 1).

FIGURE 1. Aspect of fruit and seeds of *P. cattleianum* used to *in vitro* establishment, Porto Alegre, 2013.

The *in vitro* establishment of the seeds was done in sterile laminated flux, where after disinfection of the seeds for ten minutes in immersion under NaOCl (2%) and triple washed with sterile unionized water, were incubated in test tubes containing 10mL of medium. Was adopted LPm medium (50%) (Von Arnold and Eriksson, 1981), increased with sucrose (10 g.L-1) and agar (0,8%) and the pH adjusted to ±5.8 before autoclaving under 1.5 ATM 15 min. After three days, the seeds were removed from the tubes and were submerged for 18 hours on concentrations of giberellic acid evaluated (0; 2; 10; 20 and 50 mg L-1) that gave origin to the treatments, the source being the commercial product Progibb®.

The material remained in incubating room at ± 25° C, photo period of 16 hours (light lamp OSRAM™ 40W). To each four days the emergency was evaluated and, after 64 days, they were evaluated percentage of emergency (% E), and the emergency´s speed index (E.S.I), was calculated using the formula I.V.E = G1/N1 + G2/N2 +... + Gn/Nn; where G1, G2, Gn = number of seedlings computed in the first , second and last count; and N1, N2, Nn = number of days of sowing to the first, second and last count (Maguire, 1962).

The experimental design was completely randomized, with the experimental unit composed by a Petri dish with five seeds, having ten repetitions in each treatment. The percentage measurements of emergency were transformed ($\sqrt{x+1}$) and analyzed by the Duncan test ($P \leq 0,05$), in the E.S.I, the results were analyzed by the Dunkan test ($P \leq 0,05$), beyond the analyzes of regression in function of the concentrations tested.

RESULTS AND DISCUSSION

In the giberellic acid concentrations evaluated, was not observed significant influence of the emergency´s final percentage, staying on average with 64%. Immersion of the seeds in giberellic acid provoked a delay in the emergency (Figure 2). The seeds that did not pass for immersion in giberellic acid began it emerge from 33 days after the incubation reached 50% of emergency after 48 days, being the most superior of the treatments.

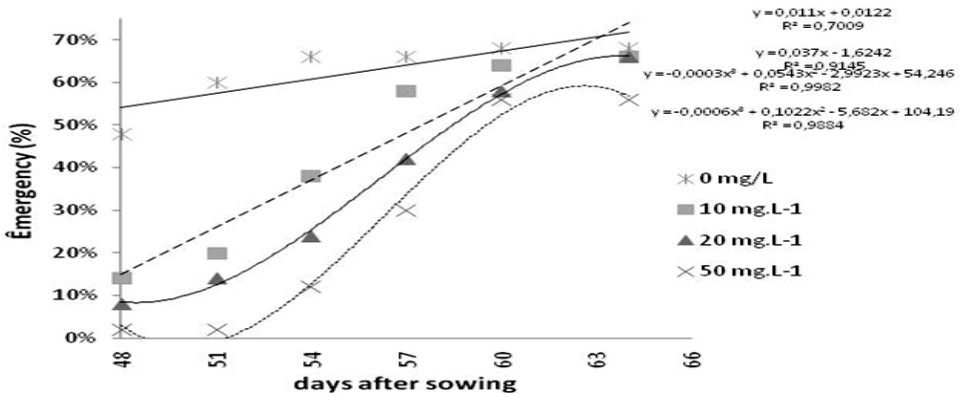

FIGURE 2. Dispersion emergence over time strawberry guava seeds in response to the giberellin concentration. Porto Alegre, RS 2013.

In case of Emergency Speed Index - E.S.I that evaluates the emergency in function of the number days necessary for its occurrence, it was observed that concentrations above 10 mg.L-1 of giberellic acid were harmful having difference significant statistically (Figure 2) among the treatments. In this sense although have shown the emergency accumulated in a short time, the use of the phytoregulator showed inhibitory in the condition tested.

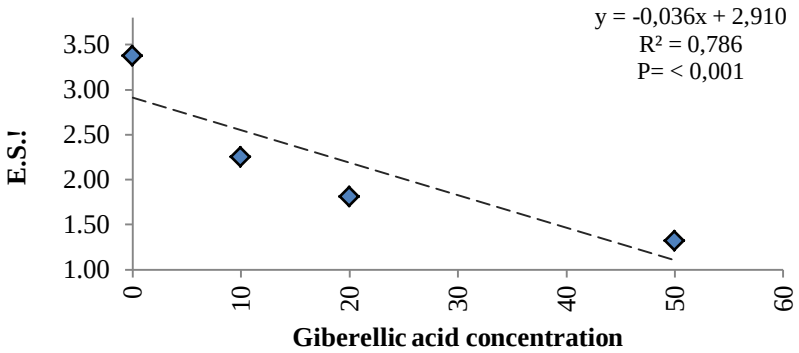

FIGURE 3. Average emergency speed índex (E.S.I) of strawberry guava seeds depending on the concentration of giberellic acid, Porto Alegre, RS 2013.

High concentrations of phytoregulators generally are related as inhibitors for germination, fact for which must be to research the ideal concentration in function of the application method and which product should be used. In germination of Atemóia seeds (*Anona cherimola* M), imbibition for 36 hours in solution of 500 mg.L-1 of gibberellins promoted the greatest percentage of germination (Oliveira, 2010). With the present study, the negative action of the giberellic acid in concentrations considered lows, in case of from 10 mg L-1 inhibitory effect was verified.

This difference in response to different hormone concentrations may be due among other reasons, the positive photoblastic behavior of P. *cattleianum*, a condition in which the seed needs light to germinate of stimulation (Santos *et al.*, 2004).

Particularly known is the output of the quiescent state or dormancy induced by lack of cold, dehydration or light can be overcome by gibberellins. Due to the narrow relation between the phytochrome, that in an operational sense acts like responsive intracellular protein to the light, and gibberellins in the action of the light in the stimulus of the germination, can be measured by the endogenous increase of gibberellins. (Taylorson & Hendricks, 1997).

Understanding that the germination process responds to the interaction between gibberellins and other growth regulators such as abscisic acid, cytokinins and brassinosteroid, one might think that the low percentage of emergence has relation to both the dose used, the immersion time as with the content of endogenous hormone own seed. According Taylorson & Hendricks (1997), applied cytokinins usually have little influence in dormancy and germination controlling in comparison with GA and ABA, but its activity is more pronounced when combined with another promoter, such as gibberellin, light, or ethylene.

CONCLUSIONS

The red strawberry guava seeds, in the tested conditions present a medium emergency percentage of approximately 64%. The red strawberry guava seeds emergency is delayed by the imbibitions on giberellic acid in the tested solutions varying between 10 mgL^{-1} and 50 mgL^{-1} for 18 hours.

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