

# Photosynthetic Efficiency in Aroeira and Cedro After the Application of Salicylic Acid

Maria Eunice Lima Rocha<sup>1</sup>, Fernanda Ludmyla Barbosa de Souza<sup>1</sup>, João Alexandre Lopes Dranski<sup>1</sup>,  
Mayra Taniely Ribeiro Abade<sup>1</sup>, Pablo Wenderson Ribeiro Coutinho<sup>1</sup>, Tatiane Priscila Chiapetti<sup>1</sup>,  
Luanne Karoline Rinaldi<sup>2</sup>, Marlison Tavares Ávila<sup>1</sup>, Kildemir da Costa Milomes Junior<sup>1</sup>, Hannah Braz<sup>1</sup>,  
Marlene de Matos Malavasi<sup>1</sup> & Ubirajara Contro Malavasi<sup>1</sup>

<sup>1</sup> Universidade Estadual do Oeste do Paraná, Paraná, Brazil

<sup>2</sup> Universidade Estadual de Maringá, Paraná, Brazil

Correspondence: Maria Eunice Lima Rocha, Universidade Estadual do Oeste do Paraná, Paraná, Brazil. E-mail: eunice\_agronomia@yahoo.com.br

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## Abstract

The objective of this research was to quantify the photosynthetic efficiency of *Cedrela fissilis* and *Schinus terebinthifolius* seedlings after application of salicylic acid for eight weeks. The experimental design was completely randomized, composed of four treatments, with five replicates of 20 seedlings each. The treatments consisted of increasing doses of the salicylic acid solution: 0, 100, 200, 300 mg L<sup>-1</sup>. The solution was composed of salicylic acid, deionized water and adjuvant and applied with hand sprayer, weekly for 2 months. The parameters evaluated after the application of the acid consisted of the rate of CO<sub>2</sub> assimilation, leaf transpiration, stomatal conductance and internal CO<sub>2</sub> concentration, as well as water use efficiency, intrinsic water use efficiency and instantaneous carboxylation efficiency. In seedlings of *Schinus terebinthifolius* there was a reduction of the photosynthetic parameters and consequently a better use of water as the doses of the plant growth regulator were increased. In the seedlings of *Cedrela fissilis* the dose of 200 mg L<sup>-1</sup> resulted in greater CO<sub>2</sub> fixation per molecule of water lost and in this sense, it can be inferred that the increase in the doses of salicylic acid improved the photosynthetic efficiency, but the use of water was lower. Thus, for both species, lower doses are more recommended because there must be a balance between the fixed amount of CO<sub>2</sub>, the plant generated from photoassimilates and the amount of water lost, in order not to generate a negative potential in the plant metabolism.

**Keywords:** defense, native, photosynthetic efficiency

## 1. Introduction

The use of native wood species adds great commercial and social value to the environment in which they are inserted. In this sense, it is of great interest to increase the production of large scale and better quality seedlings. Thus, studies focusing on the potential, uses, establishment, development, practices and management in the nursery and in the field should be developed (Gonçalves et al., 2012).

The *Schinus terebinthifolius* Raddi. is a native tree, not endemic to Brazil belonging to the family of Anacardiaceae. Among the characteristics of the species, it is considered a pioneer species with rusticity and great phenotypic plasticity, and therefore has great potential of use in programs of environmental reforestation and revegetation of degraded areas (Carvalho et al., 2013).

Aroeira is used for fodder of bees and goats, as raw material for fences and as an ornamental plant, as well as cosmetic, food and habitat restoration (Carvalho et al., 2015; Nickerson & Flory, 2015).

In addition, there are reports that the species also has a larvicidal, insecticidal, antidepressant, hepatoprotective and photoprotective effect (Abdou et al., 2015; Bulla et al., 2015; Piccinelli et al., 2015).

*Cedrela fissilis* Vellozo is a native tree species which can reach from 20 to 35 m in height and a diameter of 60 to 90 cm widely used for reforestation programs. The species is described as deciduous and occurs generally in moist, deep, and well drained soils (Caires et al., 2011). Cedar is found from the Amazon to the Atlantic Forest, present in both primary forest and secondary forests, but its occurrence is higher between Rio Grande do Sul and

Minas Gerais (Biernaski et al., 2012). Studies have highlighted the potential of the species for remediation of soils contaminated by heavy metals (Sakuragui et al., 2013).

Due to the quality of its wood, cedar has been progressively explored in places of occurrence and for this reason, for several years, it has been listed as a threatened species deserving preservation (Santos et al., 2009).

Plants have substances which manifested in response to the most diverse stressors resulting in physiological, morphological, biochemical, cellular and even molecular changes. Some of these compounds, known as plant hormones, will act as flags, affecting plant development and growth (Lopez et al., 2008).

Knowing the dynamics of growth regulators is extremely important, since it can contribute to the management of forests and the quality of the final product, as these will act directly in the formation of wood and in the attenuation of biological or non-biological stresses (Pereira et al., 2011; Asgher et al., 2015).

Recent research has given great importance to the endogenous application of salicylic acid in order to act in the development, growth and establishment of the seedlings in the field, besides promoting greater resistance to possible environment stresses.

Salicylic acid (SA) is an endogenous growth regulator, considered a signaling molecule, belonging to the group of phenolic compounds. The molecule consists of a hydroxyl group attached to an aromatic ring and participates in several biological processes of the plants, activating metabolic routes linked to plant defense (Hayat et al., 2010; Khan et al., 2010).

Some benefits can be observed when exogenous application of SA. Among those, an increase in germination, in growth and development of plants and an increase in the content of photosynthetic pigments (Hayat et al., 2010; Kabiri et al., 2012; Kang et al., 2014). In addition, recent studies have highlighted the potential of salicylic acid as an important regulator of photosynthetic activities, acting on photosystem I (PSII), photosynthetic pigments and the activity of enzymes linked to the primary and secondary metabolism of plants, such as rubisco and anidrase (Noriega et al., 2012; Zhang et al., 2015).

The concentration of these compounds in the plant can be influenced by many factors such as soil conditions, climate and plant development. The objective of this research was to quantify photosynthetic efficiency of *Cedrela fissilis* and *Schinus terebinthifolius* seedlings after application of SA for 8 weeks.

## 2. Material and Methods

The research was conducted in the western region of the State of Paraná, latitude 24°33'24" S, longitude 54°05'67" W and altitude of 420 m. According to IAPAR and the classification of Koppen, the climate of the region is Cfa subtropical, maintaining the average annual temperature between 22 and 23 °C, with rains well distributed during the year and hot summers (Caviglione et al., 2001). Relative humidity and air temperature during the application of the treatments (Figure 1) were obtained with a data logger (Klima Logg Smart Model).

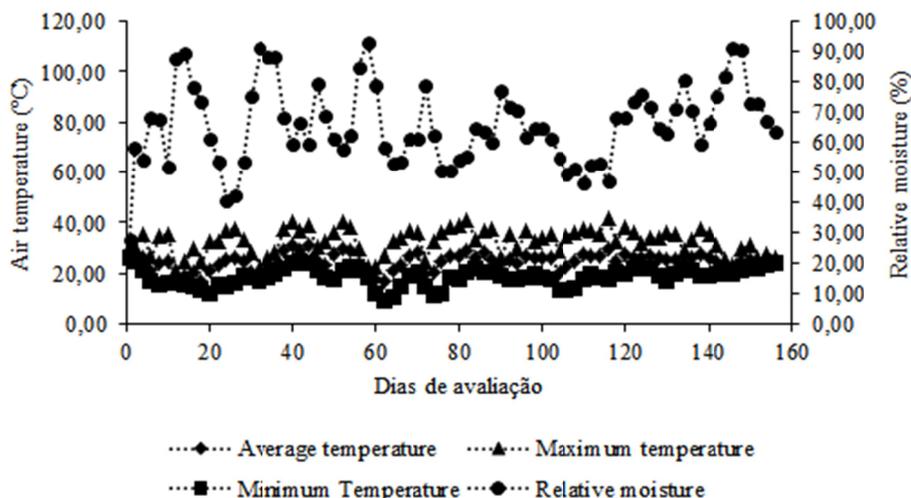


Figure 1. Minimum, average and maximum values of the temperature and average of the relative humidity of the air during the experiment with cedar and aroeira in greenhouse

We used 400 seedlings of *Schinus terebinthifolius* (aroeira) and 400 seedlings of *Cedrela fissilis* (Cedro) obtained from Itaipu Binacional and the Environmental Institute of Paraná (EIP). The three month seedlings were propagated in 120 cm<sup>3</sup> plugs filled with a mixture of local soil and humus (NITOSOLO RED Eutroferic of very clayey texture), in a ratio of 3:1.

Prior to the treatments, seedlings underwent acclimatization in a protected shadehouse for approximately 30 days between August and September. During that period, the seedlings were fertilized with 3 mL of nutrient solution in order to provide macro and micronutrients required in the growing phase (Table 1).

Table 1. Composition of the nutritive solution for seedling fertilization

Nutritive solution					
KH <sub>2</sub> PO <sub>4</sub>	MgSO <sub>4</sub>	KNO <sub>3</sub>	Ca (NO <sub>3</sub> ) <sub>2</sub> 4H <sub>2</sub> O	Micronutrients complete	Fe-EDTA
----- mL L <sup>-1</sup> -----					
1.0	2.0	5.0	5.0	1.0	1.0

Seedlings were measured (height, diameter and number of leaves) before treatments. Aroeira seedlings showed averages (n = 400) of 14.60 cm height, 2.99 mm diameter and 13.34 leaves whereas for cedar seedlings (n = 400) showed averages of 8.09 height, 2.51 mm diameter and 6.62 leaves.

The experimental design was a completely randomized composed of 4 treatments, 5 replicates with 20 seedlings in each, totaling 400 experimental units (seedlings) for each species. The treatments consisted of increasing doses of SA:

- T1: 100 mg L<sup>-1</sup> of salicylic acid;
- T2: 100 mg L<sup>-1</sup> of salicylic acid;
- T3: 200 mg L<sup>-1</sup> of salicylic acid;
- T4: 300 mg L<sup>-1</sup> of salicylic acid.

Treatments were applied weekly for 2 months from September 26 until November 14. The solution consisted of SA, deionized water and adjuvant (Agral®), 30 mL to 100 L<sup>-1</sup> of water. We used a hand sprayer between 6 and 8 am. Irrigation consisted of three shifts of 15 minutes each in addition to weeding.

Measurement included photosynthetic potential, measured by gas exchange, CO<sub>2</sub> assimilation rate (A), leaf transpiration (E), stomatal conductance (g<sub>s</sub>) and internal CO<sub>2</sub> concentration (C<sub>i</sub>) in leaves fully expanded and photosynthetically active using a IRGA (Li-6400 XT). Measurements were carried out on cloudy days from 9 to 11 a.m, in order to obtain greater uniformity in relation to the climatic conditions for the analyzes. With artificial saturated light of 1,200 μmol m<sup>-2</sup> s<sup>-1</sup> and CO<sub>2</sub> concentration at 380 μmol, under a constant temperature of 25 °C.

At the end of the test, the chlorophyll content was determined by the Arnon method (1949), in leaves located in the middle third of the plants (photosynthetically active), 14.0 cm<sup>2</sup> of leaf area were removed and packed in Falcon tubes of 15.0 cm<sup>3</sup> precoated with foil and filled with 10 ml of 80% acetone. The samples were incubated at 25 °C for 48 h.

$$\text{Chlorophyll } a = 12.7 A_{663} - 2.69 A_{645} \quad (1)$$

$$\text{Chlorophyll } b = 22.9 A_{645} - 4.68 A_{663} \quad (2)$$

$$\text{Total Chlorophyll} = \text{Chlorophyll } a + \text{Chlorophyll } b \quad (3)$$

The results were submitted to the Bartlett and Shapiro-Wilk tests in order to test the homogeneity and normality followed by the analysis of variance. When appropriated we tested treatment means using Dunnett test and comparison between SA doses the control treatment (0 mg L<sup>-1</sup>).

### 3. Results and Discussion

SA may induce morphological, biochemical or physiological alterations such as the increase in the content of photosynthetic pigments under normal or stressful conditions in plants (Singh & Usha, 2003; Khodary, 2004). Several studies have demonstrated the importance of exogenous application of SA to various metabolic functions in plants, whether physiological, ecological or biochemical. These responses have not been fully clarified, mainly because both the characteristics of the species and the climatic factors can influence the responses to the use of this inducer (Pál et al., 2014).

Results from seedlings of *S. terebinthifolius* did not show difference ( $P > 0.05$ ) with application of SA for chlorophyll a, b and total. On the other hand, seedling of *C. fissilis* treatments 1 and 4 did not differ from each other, but differed from treatments 2 and 3, which showed a reduction in chloroplastidic pigments (Figure 2).

At the 300 mg L<sup>-1</sup> dose of SA, the existing reduction was lower than detected from the other treatments. This treatment did not vary from the control seedlings because the responses to exogenous application of the growth regulator may vary according to the dose, time of application, physiology and age of the vegetables, besides varying according to the species and time of exposure to the inducer (Picolotto et al., 2009).

Chlorophyll b in seedlings of *C. fissilis*, was not influenced by ( $P > 0.05$ ) with the increase in SA doses.

In *C. fissilis* seedlings there was a decrease in the concentration of total chlorophylls resulting by reduction of the number of leaves.

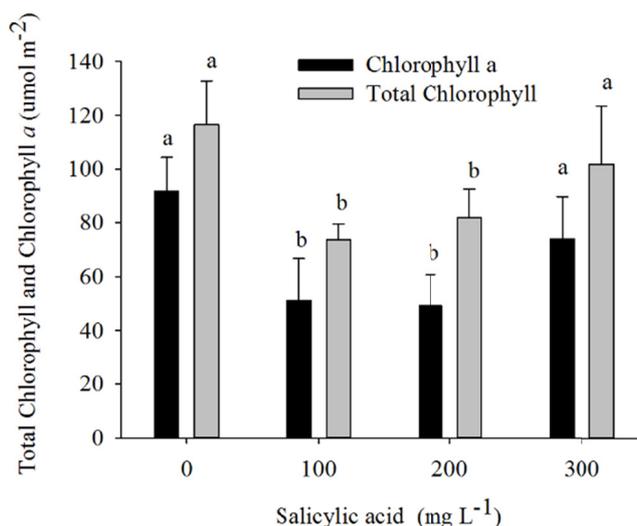


Figure 2. Chlorophyll a and total chlorophyll content in *Cedrela fissilis* seedlings after application of salicylic acid

*Note.* The averages followed by the same letter do not differ statistically from each other by the Dunnett test at the 5% probability level.

Measurement of net CO<sub>2</sub> assimilation showed a significant difference ( $P > 0.05$ ) as SA doses increased in seedlings of both species. Mean values of the treatments were equal to 5.54  $\mu\text{mol m}^{-2} \text{s}^{-1}$  in *C. fissilis* seedlings and for *S. terebinthifolius* the averages ranged from 4.07 to 4.49  $\mu\text{mol m}^{-2} \text{s}^{-1}$ . Similar results were obtained by Mazzuchelli, Souza and Pacheco. (2014) where the application of salicylic acid did not result in significant statistical difference ( $P > 0.05$ ) for the photosynthetic determinations in *Eucalyptus urophylla* x *Eucalyptus grandis* (clone H13) plants submitted to irrigation regimes.

Temperatures between 30 and 35 °C can promote inhibition of CO<sub>2</sub> assimilation in C<sub>3</sub> plants and impair activation of Rubisco, inhibiting the enzyme linked to its conversion (Rubisco activase), resulting in stomatal closure and reduction of photosynthetic activity (Oliveira et al., 2005).

The internal CO<sub>2</sub> concentration is an important parameter, since it allows direct evaluation of plant productivity from the determination of intercepted solar energy and the amount of CO<sub>2</sub> in non-stressing edapho climatic conditions, high CO<sub>2</sub> content will result in high rates of photosynthesis (Taiz & Zeiger, 2017).

In *S. terebinthifolius* seedlings, the internal CO<sub>2</sub> concentration did not show difference ( $P > 0.05$ ) between treatments 1, 2 and 3. With the dose of 300 mg L<sup>-1</sup>, however, the lowest mean was observed, equivalent to 254.46  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (Figure 3).

The Ci presented reduction in seedlings submitted to the control treatment for the maximum dose of SA. This is related to the stomatal conductance, which was also lower in this treatment. Thus, the lower the conductance the lower the flow of gases, water and luminosity that flow through the stomata.

The lower the gas exchange between plants and the lower atmosphere the assimilation of CO<sub>2</sub> and the less the conversion of light energy into photoassimilates, causing the plant to have fewer reserves and become more susceptible to adverse conditions.

SA is required in small amounts to activate or inhibit routes in plant metabolism. High doses of SA can affect photosynthesis, osmotic adjustment and antioxidant metabolism, not promoting tolerance in plants, on the contrary, triggering a high level of stress in the plants (Hayat et al., 2010). In this sense, *S. terebinthifolius* seedlings responded negatively to the maximum dose of SA demonstrating that excess chemical stimulation could impair essential plant activities (Figure 3). The reduction of the maximum value obtained with 100 mg L<sup>-1</sup> compared to 300 mg L<sup>-1</sup> was 15.75% in the internal CO<sub>2</sub> concentration.

In controversy Nivedithadevi et al. (2012) reinforced that the use of SA at appropriate concentrations may improve photosynthetic rate and consequently the growth and productivity of the species, but the results will vary according to the intrinsic and extrinsic conditions of the species.

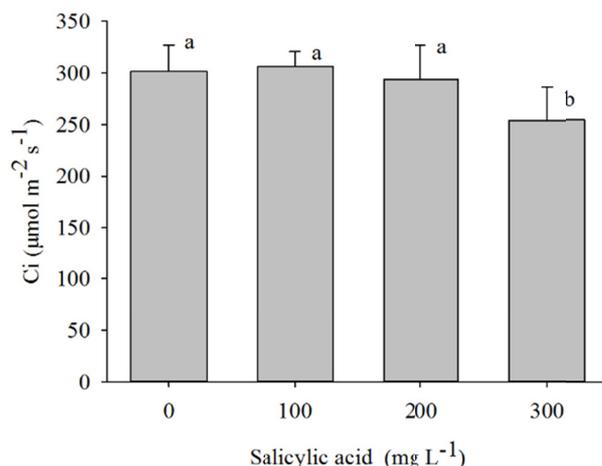


Figure 3. Internal concentration of CO<sub>2</sub> (Ci) in seedlings of *Schinus terebinthifolius* after application of salicylic acid

*Note.* The averages followed by the same letter do not differ statistically from each other by the Dunnett test at the 5% probability level.

In *C. fissilis* seedlings the internal CO<sub>2</sub> concentration were not affected ( $P > 0.05$ ) by SA and the averages ranged from 212.63 to 288.45 µmol m<sup>-2</sup> s<sup>-1</sup>.

Responses to exogenous application of SA will not always be noticeable, since the effect is dependent on the stage of development of the plants, applied amount of the hormone, concentration, mode of application, edaphic and climatic conditions, besides factors related to physiology and morphology of the seedlings (Horváth et al., 2007).

The rate of assimilation and the concentration of CO<sub>2</sub> are correlated variables, since the higher CO<sub>2</sub> concentration in the leaves is the result of the greater assimilation in the plants, but if any structure or physiological activity is impaired, this will trigger a series of problems (Jadoski et al., 2005). In this research, for example, stomatal conductivity was low, which could result in low assimilation and carbon sequestration, lower energy production, ceasing of essential plant activities, leading to the death of cells and plants. Therefore, it is interesting that salicylic acid acts in these processes, conferring tolerance to the vegetables under stressful conditions.

When analyzing the instantaneous efficiency of carboxylation in seedlings of *S. terebinthifolius* and *C. fissilis* there was no influence ( $P > 0.05$ ) from doses of SA and the average across treatments was 0.021 and 0.018, respectively.

High concentrations of SA may negatively affect the physiology of seedlings, promoting the decrease of carbon fixation as a consequence of Rubisco inhibition. As a result, the main activities of the plants can be impaired due to the interruption in the Calvin cycle, mainly the photosynthesis and respiration of the plants.

The instantaneous efficiency of carboxylation depends directly on the availability of CO<sub>2</sub> present inside the leaves, amount of radiation, temperature and enzymatic activity, so when these factors are affected the carboxylation efficiency is impaired, leading to the paralysis of activities essential for the normal functioning of the vegetable.

Moreover, if the amount of CO<sub>2</sub> is low, it will hinder the entry of this gas into the cells, affecting the carbon input and energy production, interrupting the entire vegetative cycle of plants (Taiz & Zeiger, 2017).

Seedlings when exposed to some nursery practices alter their physiological characteristics. Among those, the photosynthetic rates, which, in general, are low due to the low stomatal conductance when the stress conditions are exposed. Therefore, practice can alter the perspiration, promoting the reduction of the same and promoting the osmotic regulation of the cells.

In seedlings of *S. terebinthifolius*, stomatal conductance was reduced with increased doses of SA. For the dose of 300 mg L<sup>-1</sup>, the average stomatal conductance was 0.05 μmol m<sup>-2</sup> s<sup>-1</sup>, which may have occurred because at the maximum dose there was a lower flow of water and gases in the stomata (Figure 4). With the decrease of the conductivity there is less contribution and transport of energy from the environment to the cells, interrupting the flow soil-plant and atmosphere. This interaction is complex because it depends on the environmental variables, the characteristics of the species and the growth inducer. Additionally, depending on those conditions, the application of exogenous hormonal sources can bring some harm to the general metabolism of plants. Melotto et al. (2006) reported that 0.4 mM of SA induced stomatal closure and reduced stoma media mediated by stomata in *Arabidopsis thaliana*.

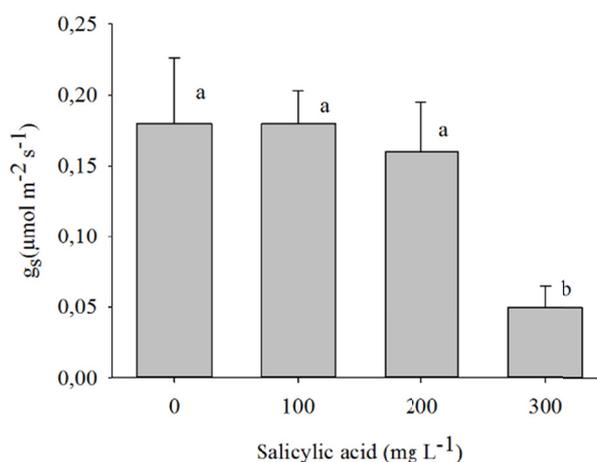


Figure 4. Stomatal conductance (g<sub>s</sub>) in seedlings of *Schinus terebinthifolius* after application of salicylic acid

*Note.* The averages followed by the same letter do not differ statistically from each other by the Dunnett test at the 5% probability level.

In addition to this justification, factors such as temperature and humidity may have influenced this response in the seedlings. Situations of light or water stress may have been signaled the root to produce abscisic acid and promote stomatal closure, thus reducing water flow and CO<sub>2</sub>.

As can be observed in Figure 1, there was great oscillation of air temperature and relative air humidity in the months when the seedlings were in the field, mainly in the final phase, where the quantification of gas exchanges (between 140 and 160 days). The humidity reached approximately 91% and the maximum and minimum temperature ranged from 17.3 to 37.4 °C, which may justify the physiological changes observed in both species.

According to Oliveira et al. (2002), as the stomatal conductance is a point measure and will determine the number of stomata that are open at the time of reading. If the plant is exposed to a momentary stress situation the response will present a reduction, agreeing with the results obtained in research.

In seedlings of *C. fissilis* the stomatal conductance did not vary according to the application of SA and the average reading was 0.097 μmol m<sup>-2</sup> s<sup>-1</sup>.

The plants present different responses even when exposed to the same conditions. So it is interesting to highlight these characteristics and to study in detail what changes, whether these are morphological, physiological or biochemical or even characteristics of the environment that will affect the growth and survival of the seedlings, both initially development phases.

Studies have highlighted the potential of hormones such as auxin, cytokinins, ethylene, brassinosteroids, jasmonate and, more recently, salicylic acid in stomatal regulation (Haubrick et al., 2006).

Under stress conditions, the stomata, which are pores of the epidermis, can be affected and thus activate responses in the plants, promoting the increase of AS, this in turn, along with other hormones may promote stomatal closure.

These responses may be reflected in the generation of reactive oxygen species and accumulation of calcium and hydroxyl, neutralizing the environment and stimulating the closure of the pores of plants (Melotto et al., 2006; Khokon et al., 2011).

Inversely proportional to the use of water, transpiration was reduced in *S. terebinthifolius* seedlings and the means were 3.15; 3.14; 3.01 and 1.57  $\mu\text{mol m}^{-2} \text{s}^{-1}$  with increasing doses of SA and reduction of treatment 1 to 4 was 50.16% (Figure 5).

Both carbon assimilation and transpiration occurs preferentially by the stomatal route. In *S. terebinthifolius* the  $\text{CO}_2$  contribution decreased when SA doses increased due to the stomatal closure (less stomatal conductance) and seedlings lost less water in the form of vapor, reduced the acquisition of photoassimilates, but presented a better use of water.

Marenco et al. (2014) pointed out that the water potential of the leaf does not remain constant throughout the day. This occurs basically, because leaf transpiration is different from the rate of absorption by the roots, generating electrochemical potential gradient. Therefore, there is a water movement from the root system to the aerial tissues promoting the continuous soil-plant-atmosphere. The same authors further argued that sap flow will increase during the first hours of the day because of irradiance and the stomata will open up, increasing stomatal conductance and increasing transpiration.

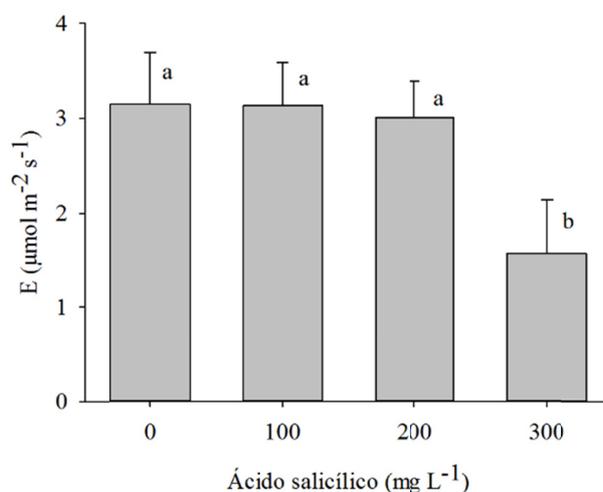


Figure 5. Transpiration (E) in seedlings of *Schinus terebinthifolius* after application of salicylic acid

*Note.* The averages followed by the same letter do not differ statistically from each other by the Dunnett test at the 5% probability level.

On the other hand, seedlings of *C. fissilis* did not show a difference ( $P > 0.05$ ) for transpiration after application of SA with values ranging from 2.28 to 3.03  $\mu\text{mol m}^{-2} \text{s}^{-1}$ .

Values (Figure 6) of water use efficiency in *S. terebinthifolius* seedlings submitted to treatments 2 (1.74) and 3 (2.75) were statistically similar to the control treatment (2.01) but differed from seedlings that received the maximum tested dose of the regulator (3.34). With increased doses of SA there was a better use of water by seedlings, revealing that SA may have favored and improved metabolic activities associated with water use.

Under field conditions, this characteristic can be very important, since it may reduce seedling and decrease the loss of water, attenuating post-planting shock (Jacobs & Landis, 2009).

The greatest use of water was obtained with the dose of 300 mg L<sup>-1</sup> due to the lower transpiration. However, at the same dose, there was smaller stomatal conductance and consequently photosynthesis.

In summary, the photosynthetic efficiency also decreases and although the seedling is losing less water, it is also producing fewer photoassimilates and consequently reducing plant biomass.

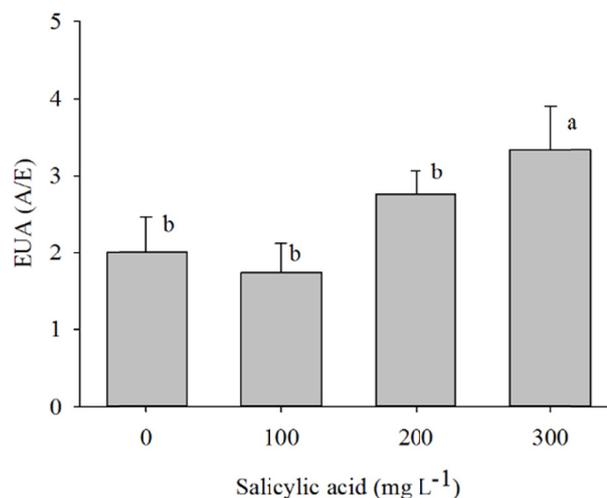


Figure 6. Efficiency of water use (EUA) on seedlings of *Schinus terebinthifolius* after application of salicylic acid

*Note.* The averages followed by the same letter do not differ statistically from each other by the Dunnett test at the 5% probability level.

The efficiency of water use in *C. fissilis* seedlings showed no difference ( $P > 0.05$ ) with the application of SA. In adverse situations, seedlings may present changes in their photosynthetic activities, such as stomatal conductance, transpiration and the efficiency of water use, but this is dependent upon factors linked to the species or the environment (Ferraz et al., 2012).

Scalon et al. (2011) with *Guazuma ulmifolia* Lam. seedlings reported that the water use efficiency did not show difference ( $P > 0.05$ ) when exposed to the water stress condition. It should be emphasized that those variables are dependent on internal and external factors of the plants, so, in a certain condition they will express different responses.

With seedlings of *S. terebinthifolius* subjected to SA the intrinsic efficiency of water use resulted in differences ( $P < 0.05$ ) only for the treatment equivalent to the 300 mg L<sup>-1</sup> with a value of 84.68 (Figure 7).

The relationship between photosynthesis and transpiration indicates the instantaneous efficiency of water use, demonstrating the amount of carbon the plant attaches to each molecule of water lost (Jaimez et al., 2005).

Thus, when there was an efficient use of water, there was a reduction in the internal concentration of CO<sub>2</sub> and its assimilation, demonstrating that the dose may not be adequate when assessing all the physiological characteristics.

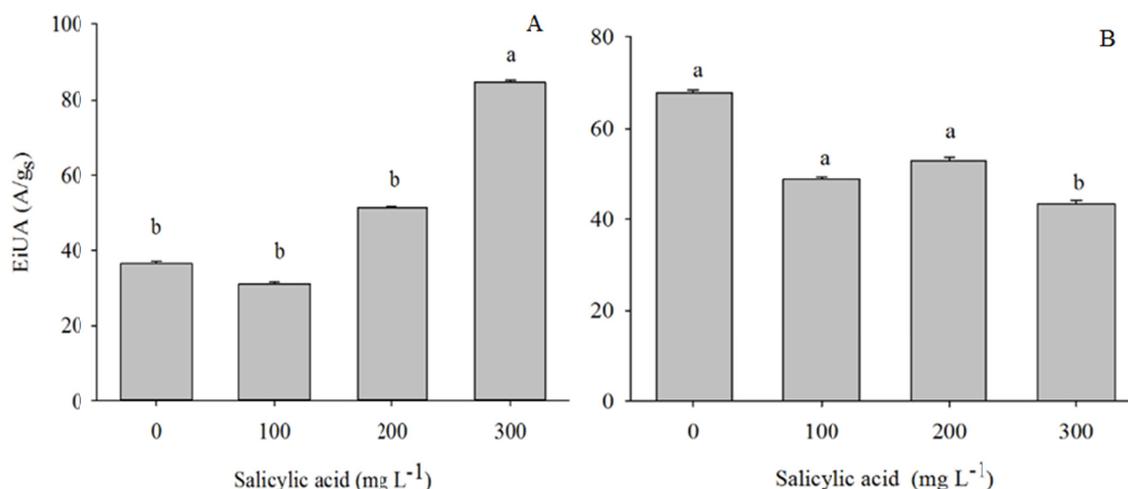


Figure 7. Intrinsic water use efficiency (EiUA) in seedlings of *Schinus terebinthifolius* (A) and *Cedrela fissilis* (B) after application of salicylic acid

*Note.* The averages followed by the same letter do not differ statistically from each other by the Dunnett test at the 5% probability level.

The intrinsic water use efficiency use in *Cedrela fissilis* seedlings after application of 300 mg L<sup>-1</sup> of AS resulted in the lowest mean, equal to 43.27 that differed statistically from the other treatments (Figure 7).

Plant hormones are required in small amounts and the excess can generate numerous stresses on plants, altering physiological, biochemical and morphological activities, as well as stimulating the production of secondary metabolites.

In addition, terrestrial plants have specific signs that can interrupt some metabolic activities under adverse conditions in order to reduce energy expenditure by redirecting them to produce protective substances.

Thus, the parameters studied are very important when choosing the most appropriate species for each purpose and environment, since they are important indicators of the development of the plants under specific conditions.

#### 4. Conclusion

Seedlings of *S. terebinthifolius* receiving SA for eight weeks resulted in a reduction in internal CO<sub>2</sub> concentration, stomatal conductance and transpiration, but increased water use efficiency and the intrinsic efficiency of water use, demonstrating that the doses tested favored the use of water, but reduced assimilation and production of CO<sub>2</sub> and, consequently, the contribution of photoassimilates.

However, it is interesting to note that photosynthetic reduction is a strategy of the terrestrial plants to avoid stressful conditions and may be advantageous provided that this situation occurs quickly, not significantly affecting the normal metabolism of the plants.

The maximum tested dose of the plant regulator induced stress in seedlings and therefore there was a reduction of the photosynthetic parameters, which may have contributed to the increase of resistance.

In seedlings of *C. fissilis* there was a reduction in the intrinsic efficiency of water use and the dose of 200 mg L<sup>-1</sup> reflected in the greater loss of water and higher carbon fixation. Chlorophyll a and total contents decreased as the SA doses increased, but the 300 mg L<sup>-1</sup> dose did not differ from the control seedlings. Different from results obtained with *S. terebinthifolius* seedlings, the dose of 200 mg L<sup>-1</sup> resulted in greater CO<sub>2</sub> fixation per molecule of water lost. Therefore, it can be inferred that the increase in the doses of salicylic acid improved the photosynthetic efficiency.

For seedlings of both species lower doses of SA are more recommended in order not to generate a negative potential in the plant metabolism.

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