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GAS EXCHANGE IN *Annona emarginata* (SCHLTDL.) H. RAINER SUBJECTED TO SALT STRESS AND APPLICATION OF PLANT GROWTH REGULATOR

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ABSTRACT - The aim of this study was to evaluate the effects of the application of plant growth regulator on gas exchange in *A. emarginata* seedlings subjected to different NaCl concentrations. We evaluated the gas exchange in *A. emarginata* seedlings subject to the presence and absence of plant growth regulator consisting of kinetin, gibberellic acid, and indolbutyric acid and three concentrations of NaCl (0 mM, 50 mM, and 100 mM). When assessing these factors individually, it was observed that in the presence of plant growth regulators, there was a reduction in stomatal conductance and transpiration, and NaCl concentration (50 mM and 100 mM) caused a reduction in CO_2 assimilation, stomatal conductance, and transpiration. There was no significant interaction between plant growth regulators and salt stress. Therefore, gas exchange in *A. emarginata* seedlings when under salt stress is not influenced by the prior application of plant growth regulators. However, individually, both the implementation of the regulators and the salt stress led to a reduction of gas exchange in *A. emarginata* seedlings.

Key words: auxin, cytokinin, gibberellin, photosynthesis, sodium chloride.

TROCAS GASOSAS EM Annona emarginata (SCHLTDL.) H. RAINER SUBMETIDA A ESTRESSE SALINO E APLICAÇÃO DE REGULADOR VEGETAL

RESUMO - O objetivo deste estudo foi avaliar os efeitos da aplicação de regulador vegetal nas trocas gasosas de mudas de *A. emarginata* submetidas a diferentes concentrações de NaCl. Foram avaliadas as trocas gasosas nas mudas submetidas à presença e ausência de regulador vegetal constituído por cinetina, giberelina A_3 e ácido indolbutírico, e três concentrações de NaCl (0 mM, 50 mM e 100 mM). Ao se avaliar os fatores isoladamente, foi observado que na presença de regulador houve redução na condutância estomática e transpiração e a concentração de NaCl (50 mM e 100 mM) provocou redução na assimilação de CO_2, na condutância estomática e transpiração. Não foi encontrada interação significativa entre o regulador vegetal e o estresse salino. Portanto, as trocas gasosas de *A. emarginata* sob estresse salino não são influenciadas pela aplicação prévia do regulador vegetal. Porém, isoladamente, tanto a aplicação do regulador como o estresse salino levaram à redução das trocas gasosas de *A. emarginata*.

Palavras-chave: auxina, citocinina, cloreto de sódio, fotossíntese, giberelina.

INTRODUCTION

According to the SSSA (2008), salt-affected soils are those that are modified due to excess soluble salts, exchangeable sodium, or both to adversely affect the growth and development of plants. The high concentration of salts, such as NaCl, decreases water absorption by the roots and interferes with the entry of other ions (HAJIBAGHERI et al., 1989; LAMBERS, 2003).

Worldwide, about 20% of cultivated areas are under the influence of saline soils. Most of these soils are found in Asia (ASHRAF e FOOLAD 2007); however, according Pessarakli; Szabolcs (1999), all continents have this problem. Saline soils occur in arid and semi-arid regions due to low precipitation and high evaporation.

These conditions do not allow leaching of the salts present in the soil, resulting in the accumulation of salt levels that are toxic to plants (FAGERIA et al., 2010). Improper use of fertilizers can also lead to increased salt concentrations, even in high rainfall conditions (FAGERIA et al., 2011).

The plant response to salt stress is modulated by hormonal signals that regulate development and defense mechanisms (POSPÍŠILOVÁ, 2003). For example, the plant hormone ABA acts by closing the stomata in plants under salt stress (RYU e CHO, 2015), leading to less water loss through transpiration (CHAVES et al., 2009).

However, decreased stomatal conductance can decrease the diffusion of CO_2, which affects the carbon assimilation rate. Moreover, salt stress also leads to

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changes in the photochemical processes and enzymes responsible for CO_2 assimilation (LAWLOR; CORNIC, 2002; MUNNS, 2002; KEYS et al., 2003; FLEXAS et al., 2004; GALMÉS et al., 2007). Therefore, one of the metabolic processes most affected by salinity is photosynthesis (MUNNS et al., 2006).

According to Chaves et al. (2009) salt stress and water deficit cause some similar photosynthetic metabolic responses. Reports from farmers indicate that the native brazilian species *Annona emarginata* (Schltdl.) H. Rainer which is used as rootstock for producing atemoya seedlings (*Annona squamosa* L. x *A. cherimoya* Mill.) and to recover degraded areas displays tolerance to soil water deficit (TOKUNAGA, 2000).

Mantoan et al. (2015) demonstrated that *Annona emarginata* presents photochemical apparatus tolerant to water deficit. According to Saimbhi (1993), the application of plant growth regulators can minimize the physiological damage caused by salt stress.

Studies involving the application of plant growth regulators such as gibberellins (AFZAL et al., 2005), auxins (KHAN et al., 2004) and cytokinins (DHINGRA e VARGHESE, 1985) have demonstrated beneficial, stress-relieving effects (SAIMBHI, 1993). Thus, the aim of this study was to evaluate the effects of the application of plant growth regulators on gas exchange in *Annona emarginata* plants subjected to different NaCl concentrations.

MATERIALS AND METHODS

Two-year-old *Annona emarginata* seedlings were purchased from a commercial nursery and transplanted into 2 L polyethylene bags containing vermiculite of medium texture.

The plants were then kept in greenhouse shading with 30% shade for 50 days for acclimatization. During acclimatization, the plants were irrigated every other day with water until percolation; they were also irrigated once a week with Hoagland; Arnon (1950) solution diluted to 75% of ionic strength for the fertilizer.

Before the start of the salt treatments, half of the pots received foliar application of a mixture of spreader-sticker (mineral oil) 0.5% v/v with a growth regulator composed of 0.09 g L^{-1} kinetin, 0.05 g L^{-1} gibberellin (GA_3), and 0.05 g L^{-1} indolbutyric acid (IBA - Stimulate®). The other half of the seedlings did not receive the application of the regulator.

Approximately 24 h after the application of Stimulate® the pots were irrigated every two days with three sodium chloride solutions (NaCl) at concentrations of 0.50, and 100 mM (0.036, 5.59, and 11.11 dS m^{-1} electrical conductivity, respectively) until percolation.

The experimental design was a randomized block in double factorial (presence and absence of plant growth regulator x three concentrations of NaCl) with four replications of one plant per pot.

Gas exchange of the first fully expanded leaf of each *Annona emarginata* seedling was evaluated at 12 days after the start of treatment (DAST). For this, we used an infrared gas analyzer (LI-6400, LI-COR, Lincoln, NE, USA). The variables analyzed were net CO_2 assimilation (A_{Net}; µmol CO_2 m^{-2} s^{-1}), stomatal conductance (g_S; mol H_2O m^{-2} s^{-1}), transpiration (E; mmol H_2O m^{-2} s^{-1}), carboxylation efficiency (*ANet/Ci*), and water use efficiency (A_{Net}/E; µmol CO_2 mmol H_2O^{-1}).

Data were submitted to analysis of variance (ANOVA) of two factors (presence and absence of plant growth regulator x three concentrations of NaCl) followed by a Tukey test ($p<0.05$) using the SigmaPlot program, version 12.

RESULTS AND DISCUSSION

Differences were observed in the gas exchange of *Annona emarginata* seedlings subjected to different NaCl concentrations and in the absence and presence of the growth regulator (Table 1). However, there was no interaction between these factors, showing that, in terms of the effect on gas exchange at 12 DAST, there was no link between the implementation of the regulator at a 0.5 L ha^{-1} and salt stress.

As observed in this study, Oliveira et al. (2013) did not observe beneficial effects of the application of Stimulate® in the concentration of 2 L ha^{-1} combined with salt stress (5 dS m^{-1}) to develop *Vigna unguiculata*.

Considering only the concentration of NaCl, the plants treated with concentrations of 50 and 100 mM showed significant reduction in A_{Net} in g_S and E compared the plants under 0 mM. This reduction in g_S shows one of the first signs of salt stress (TEZARA et al., 2003; FLEXAS et al., 2004; RODRÍGUEZ et al., 2005; RAMANI et al., 2006; CHAVES et al., 2009; CHA-UM e KIRDMANEE, 2009), which can also reduce the CO_2 input, resulting in lower A_{Net} (CHAVES et al., 2009).

Limiting A_{Net} can also be influenced by the reduction of rubisco enzyme activity and photochemical processes (SILVA e ARRABACA, 2004; XU et al., 2009), which was not observed in this study, since there were no differences in A_{Net}/Ci shown among the three concentrations of NaCl.

In relation to E, the observed reduction may have been influenced by low g_S, which was observed at NaCl concentrations of both 50 and 100 mM. According Mohammadkhani; Heidari (2008), the reduction of g_S can lead to lower E and prevent dehydration of the tissue generated by saline stress. Although low g_S can limit the A_{Net}, the A_{Net}/E may increase (BOGEAT-TRIBOULOT et al., 2007; GALLÉ et al., 2007; GALMÉS et al., 2007). This is in agreement with what was observed in this study; the A_{Net}/E did not differ among the concentrations of NaCl.

TABLE 1 - CO_2 assimilation (μmol CO_2 m^{-2} s^{-1}), stomatal condutance (mol H_2O m^{-2} s^{-1}), transpiration (mmol H_2O m^{-2} s^{-1}), carboxylation efficiency and water use efficiency (μmol CO_2 mmol H_2O^{-1}) in *A. emarginata* seedlings subjected to different NaCl concentrations with and without the application of plant growth regulators evaluated at 12 DAST. UNESP, *Campus* Botucatu, SP. 2018.

	CO_2*	SC	TR
Treatments	A_{Net}	g_S	E
NaCl conc.			
0 mM	3.38 a	0.030 a	1.19 a
50 mM	1.92 b	0.016 b	0.65 b
100 mM	1.38 b	0.011 b	0.46 b
PGR			
Without app.	2.60 a	0.023 a	0.93 a
With app.	1.85 a	0.015 b	0.61 b
Variance analysis			
NaCl con.	$p<0.01$	$p<0.001$	$p<0.001$
PGR	n.s.	$p<0.05$	$p<0.05$
NaCl con. x PGR	n.s.	n.s.	n.s.

	CE*	WUE
Treatments	*ANet/Ci*	A_{Net}/E
NaCl con.		
0 mM	0.02 a	2.86 a
50 mM	0.01 a	2.91 a
100 mM	0.01 a	3.13 a
PGR		
Without app.	0.01 a	2.97 a
With app.	0.01 a	2.96 a
Variance analysis		
NaCl con.	n.s.	n.s.
PGR	n.s.	n.s.
NaCl con. x PGR	n.s.	n.s.

*NaCl con. = NaCl concentrations, without app. = without application, with app. = with application, PGR = plant growth regulator, CO_2 = CO_2 assimilation, SC = stomatal condutance, TR = transpiration, CE = carboxylation efficiency, WUE = water use efficiency.

Although the presence of the growth regulator reduced g_S and E, there were no changes in the A_{Net}, A_{Net}/Ci, and A_{Net}/E of *A. emarginata*. According to Pospísilová (2003), cytokinins have an antagonistic effect on ABA, which reduces salt tolerance (JAVID et al., 2011). However, the application of auxins and gibberellins can reduce g_S (POSPÍŠILOVÁ, 2003). This could explain the reduction of g_S and E observed in this experiment.

CONCLUSIONS

Gas exchange in *Annona emarginata* seedlings under salt stress is not influenced by the prior application of growth regulators.

However, individually, both the implementation of a regulator and salt stress led to a reduction of gas exchange in *A. emarginata.*

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