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Productive and physiological responses of jambu (*Acmella oleracea*) under nutrient concentrations in nutrient solution

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ABSTRACT

In the last years, jambu has become popular and greatly appreciated, due to its remarkable taste. Thus, hydroponically cultivated jambu is promising, since it achieves better yield and production quality. The aim of this study was to evaluate the effect of ionic concentration in nutrient solution on growth, productivity and gas exchange of jambu. The experimental design was completely randomized, with five treatments and four replicates. The treatments consisted of variations of ionic concentration using the nutrient solution proposed by Hoagland & Arnon (25, 50, 75, 100 and 125%). The length of the main stem, stem diameter, number of inflorescence, leaf area, fresh and dry biomass (shoot, root and inflorescence), photosynthesis, stomatal conductance, transpiration, internal CO_2 concentration, *Ci/Ca* ratio and instant carboxylation efficiency were evaluated. Ionic concentrations significantly affected the studied variables, except the stem diameter, the internal CO_2 concentration and the *Ci/Ca* ratio. The number of inflorescences and the leaf area grew linearly with maximum values (37.8 units plant^{-1} and 1650.8 cm^2 plant^{-1}, respectively) obtained in ionic concentration of 125%. Maximum responses were observed for shoot fresh and dry mass (63.9 and 6.9 g plant^{-1}), root fresh and dry mass (16.7 and 2.0 g plant^{-1}) inflorescence fresh and dry mass (11.0 and 1.8 g plant^{-1}), respectively, at ionic concentration of 125%. Liquid photosynthesis, stomatal conductance, transpiration and instant carboxylation efficiency achieved maximum responses of 17.9 µmol CO_2 m^{-2} s^{-1}, 0.3 mol H_2O m^{-2} s^{-1}, 6.3 mmol m^{-2} s^{-1} and 0.06 with estimated concentrations of 84, 70, 80 and 83% of ionic strength, respectively. Thus, we concluded that the ionic concentration of 125% is indicated to obtain a greater biomass accumulation.

Keywords: *Acmella oleracea*, ionic concentration, fresh and dry mass.

RESUMO

Respostas produtivas e fisiológicas do jambu (*Acmella oleracea*) sob concentrações de nutrientes na solução nutritiva

Nos últimos anos o jambu tem apresentado crescente valorização e popularidade devido ao seu sabor e paladar marcantes. Assim, o cultivo hidropônico da espécie torna-se promissor, uma vez que alcança melhor rendimento e qualidade de produção. Objetivou-se com esse estudo avaliar o efeito da concentração iônica em solução nutritiva no crescimento, produtividade e trocas gasosas do jambu. O experimento foi conduzido no delineamento inteiramente casualizado com cinco tratamentos e quatro repetições. Os tratamentos foram constituídos de variações da concentração iônica a partir da solução nutritiva proposta por Hoagland & Arnon (25, 50, 75, 100 e 125%). Foram avaliados o comprimento da haste principal, diâmetro do caule, número de inflorescências, área foliar, biomassa fresca e seca (parte aérea, raiz e inflorescência), fotossíntese, condutância estomática, transpiração, concentração interna de CO_2, relação *Ci/Ca* e eficiência instantânea de carboxilação. As concentrações iônicas afetaram significativamente as variáveis estudadas, exceto o diâmetro do caule, a concentração interna de CO_2 e a relação *Ci/Ca*. O número de inflorescência e a área foliar cresceram linearmente com máximos valores (37,8 unidades planta^{-1} e 1.650,8 cm² planta^{-1}, respectivamente) obtidos na concentração iônica de 125%. Observou-se máximas respostas para massa fresca e seca da parte aérea (63,9 e 6,9 g planta^{-1}), massa fresca e seca da raiz (16,7 e 2,0 g planta^{-1}) e massa fresca e seca da inflorescência (11,0 e 1,8 g planta^{-1}), respectivamente, na concentração iônica de 125%. A fotossíntese líquida, a condutância estomática, a transpiração e a eficiência instantânea de carboxilação obtiveram máximas respostas de 17,9 µmol CO_2 m^{-2} s^{-1}, 0,3 mol H_2O m^{-2} s^{-1}, 6,3 mmol m^{-2} s^{-1} e 0,06 com as concentrações estimadas de 84, 70, 80 e 83% da força iônica, respectivamente. Assim, conclui-se que para obtenção de maior acúmulo de biomassa, indica-se a concentração iônica de 125%.

Palavras-chave: *Acmella oleracea*, concentração iônica, massa fresca e seca.

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Acmella oleracea, popularly known in Brazil as "jambu", is a condiment plant from the Amazon of great importance for gastronomical and medicinal purposes, mainly in Para State (Sampaio *et al*., 2019). This plant belongs to *Asteraceae* family, and it is also known as "agrião-do-Pará", "agrião-do-norte", "agrião-do-Brasil", "abecedaria" and "jambuaçu". It is a small, semi-erect growth habit plant, measuring 30-60 cm tall, with cylindrical fleshy stem and decumbent branches. Flowers are arranged in capitula (inflorescence) which give rise to achene fruits (Gusmão & Gusmão, 2013). This plant has attracted the interest of pharmaceutical and cosmetics industries, due to spilantol, as numerous tests have proven the anti-inflammatory, analgesic and anesthetic action of this compound (Sampaio *et al*., 2020).

However, the production for commercialization is in small properties in the municipalities near Belém-PA (Gusmão & Gusmão, 2013). Besides, this crop faces several technical problems which make the productive process rustic and inefficient, generating low productivity and lack of standardization and plant quality (Sampaio *et al*., 2018). Also, seasonality is common, with a greater offer in the second semester due to the festivities in the northern region of Brazil (Gusmão & Gusmão, 2013).

Thus, aiming quality production in sufficient amount, jambu cultivation under hydroponic system can be promising, since this system promotes greater input allocative efficiency, higher productivity and quality of products (Portela *et al*., 2012). Hydroponic system is a technique for growing plants without soil as a source of nutrients, so that these nutrients are available to the crop through a balanced nutrient solution (known as standard solution) which promotes growth and development of the cultivated plants. So, in this system, the nutrient solution constitutes one of the most important aspects in obtaining high quality vegetable products (Sambo *et al*., 2019).

Several formulations of standard nutrient solutions for leafy vegetables and fruits can be found in literature (Furlani, 1997; Furlani *et al*., 1999). However, it is worth mentioning that the formulations, as well as their ionic concentrations, can vary even within the plant species, since the absorption of nutrients varies with the cultivar, the developmental stage, the hydroponic system and the climatic conditions in which the crop is submitted (Portela *et al*., 2012).

No suggestion for nutrient solution formulation for jambu crop can be found in literature, though. In general, it is emphasized that the formulation proposed by Hoagland & Arnon (1950) is considered the common standard solution in which variations in relation to macro and micronutrients can be verified among them (Furlani *et al*., 1999; Cometti *et al*., 2008). Thus, further studies on nutrient solution parameters in productive performance of jambu are necessary.

Considering the nutrient solution, electrical conductivity (EC) stands out, since, it provides information on the nutrient concentration in the nutrient solution (Sambo *et al*., 2019). Thus, significant effects of EC variation on productivity, quality and physiological aspects in several crops can be noticed in literature (Luz *et al*., 2012; Portela *et al*., 2012; Baron *et al*., 2015). Portela *et al*. (2012), evaluating the effect of EC of the nutrient solution on growth, productivity and quality of the strawberry crop observed that the increase of EC favored the growth and production of plants, as well as increased total soluble solid contents, anthocyanins, L-ascorbic acid and total phenols in fruits. In a study, carried out by Baron *et al*. (2015), on production of *Annona muricata* seedlings, the results showed that EC variations of nutrient solution promoted differences in gas exchange and seedling biomass accumulation.

Given the above, and due to a small number of studies on jambu performance under hydroponic system, this study aimed to evaluate the effect of ionic concentrations, using the nutrient solution proposed by Hoagland & Arnon, (1950) on growth, productivity and gas exchange of jambu plants, searching to define the concentration which promotes higher yield.

MATERIAL AND METHODS

The experiment was conducted in a greenhouse from September 1 to December 21, 2019, at Federal Rural University of Amazon (UFRA), located in the municipality of Belém-PA (1°28'S, 48°30'W, 9 m altitude). According to Köppen, the climate is '*Afi*', 26ºC average temperature (Alvares *et al*., 2013).

The experiment was carried out in a completely randomized design with five treatments and four replicates. The treatments consisted of variations of ionic concentration, using Hoagland & Arnon's nutrient solution (1950), showing the following concentration (macronutrients in mmol L^{-1} and micronutrients in μmol L^{-1}): 15.0 N; 1.0 P; 6.0 K; 5.0 Ca; 2.0 Mg; 2.0 S; 90.0 Fe; 46.3 B; 18.30 Cl; 9.10 Mn; 0.8 Zn; 0.3 Cu and 0.1 Mo. Based on this standard solution, we used ionic concentrations (25%, 50%, 75%, 100% and 125%), and the electrical conductivity was 0.7, 1.3, 2.0, 2.8 and 3.3 dS m^{-1}, respectively. For standard solution composition, we used pure reagents for analysis, being the nutrient sources: $NH_4H_2PO_4$, KNO_3, $Ca(NO_3)_2$, $MgSO_4$, H_3BO_3, $CuSO_4.5H_2O$, $FeCl_3$ + Na_2.EDTA, $MnSO_4.H_2O$, $H_2MoO_4.H_2O$ and $ZnSO_4.7H_2O$.

Plants were grown under substrate system using sterilized ground silica as a substrate. The authors used 2 L plastic pots filled with substrate and coated with aluminum foil in order to minimize the sunlight incidence (increase in solution temperature), also avoiding the proliferation of algae inside these containers. The collecting containers of nutrient solution were painted with aluminum metallic paint. Each pot containing one jambu plant represented one experimental unit.

The solution was supplied manually in the pots, with daily frequency being placed in the morning and drained in the late afternoon. Whenever necessary, the water, lost through evapotranspiration, was replaced with distilled water. The solution was renewed weekly, and pH was checked daily with the aid of a portable peagameter (HANNA®) and, when necessary, correction was performed, using NaOH solution or citric acid ($C_6H_8O_7$) 1N, keeping it within the range from 5.5 to 6.5.

Seedlings were produced in expanded polystyrene trays of 128 cells filled with coconut fiber substrate. Afterwards, seeds were sown (the achenes) at a density of six seeds per cell. After germination, trays were transferred to benches, being kept under subsurface fertigation system, using

Hoagland & Arnon (1950) formulation at an ionic concentration of 25%. Seven days after germination, seedlings were thinned, and just one seedling per cell was kept. Seedlings were transplanted at 21 days after germination. Harvest was done at 56 days after germination.

Before harvesting, between 9 and 11 am, the authors analyzed the gas exchanges in the second pair of leaves of the main stem tip of the plants with the aid of a infrared gas analyzer (IRGA), model LI-6400XT (LI-COR, Linconl, NE). The physiological variables evaluated were: a) CO_2 concentration in the substomatic chamber (*Ci*, µmol mol^{-1}); b) stomatal conductance (*gs*, mol H_2O m^{-2} s^{-1}); c) liquid photosynthesis (*A,* µmol CO_2 m^{-2} s^{-1}); d) ratio between the concentration of CO_2 in the substomatic chamber and the concentration of CO_2 in the environment (*Ci/Ca*); e) transpiration (*E*, mmol m^{-2} s^{-1}); and, f) carboxylation efficiency (*A*/*Ci*), obtained from the ratio between liquid photosynthesis and CO_2 concentration in the substomatic chamber.

After harvest, the following traits were evaluated: a) main stem length (cm), determined with the aid of a graduated scale; b) stalk diameter, measured with the aid of a digital caliper; c) shoot fresh and dry mass (g $plant^{-1}$); d) root fresh and dry mass (g $plant^{-1}$); e) inflorescense fresh and dry mass (g $plant^{-1}$); f) number of inflorescence (unit $plant^{-1}$), through simple counting; and g) leaf area (cm^2 $plant^{-1}$), determined with the aid of an area integrating device, LICOR® model LI-3100.

Fresh and dry masses were determined after separating each part, and being weighed using a precision scale (0.001 g). In order to determine dry mass, the samples of each part were kept in paper bags and taken to an oven, temperature 65ºC for 72 hours until reaching constant weight.

The results obtained in this study were submitted to variance analysis and, when significant, regression analysis was carried out. Then, the models and coefficients were tested by t test. The model was chosen based on the significance showed and superior determination coefficient (R^2) (>0.7). To analyze the data obtained in the experiment, we used statistic software Sisvar version 5.6 (Ferreira, 2011).

RESULTS AND DISCUSSION

Ionic concentrations of nutrient solutions affected growth, productivity and gas exchanges; the concentrations did not affect the stem diameter, internal CO_2 concentration and *Ci/Ca* ratio, though.

An increase of nutrient solution concentration did not promote significant increases in stem diameter, which varied from 5.0 to 5.4 mm, considering the smallest (25%) and the greatest (125%) tested ionic concentrations, respectively (Figure 1A). Similarly, in lettuce plants (*Lactuca sativa*) variety Romana grown under hydroponics, Cunha-Chiamolera *et al*. (2017) verified that an increase of ionic concentration of nutrient solution did not affect the stem diameter of the plant.

For jambu, the authors observed that the stem diameter is a trait which is little affected by nutrient availability in the nutrient solution, since both in ionic concentration of 25% (5.0 mm) and of 125% (5.4 mm) no evidence for growth trend related to this variable was noticed (Figure 1A). Besides, the fact that jambu is a rustic plant should be considered (Gusmão & Gusmão, 2013), since it can explain the ability to tolerate extreme variations of nutrient solutions tested in this study. The authors also highlight that even in low concentration of nutrients, no characteristic symptoms of nutritional deficiencies were observed.

In relation to main stem length, the authors observed that an increase of the ionic concentration promoted positive responses up to a certain concentration, which was explained by a quadratic model (Figure 1B). The maximum estimated growth was 26.7 cm at a concentration of 90.2%; from this concentration, the authors noticed a decrease in this variable, which can be explained by a possible nutritional disorder, due to a high concentration of salts in the solution, which resulted in plant morphological changes (Fontes, 2016). For cucumber crop (*Cucumis sativus*), Diniz *et al.* (2015), evaluating the effect of different nutrient proportions (12,5, 17, 25, 50 and 100%) on nutrient solution proposed by Furlani *et al*. (1999), observed higher plants grown in more diluted solutions, where they also verified that, as the concentration of nutrients in the solution increased, plant length decreased. According to Fontes (2016), excess of nutrients can cause changes in cellular and biochemical level of the plants, which results in changes in photoassimilate partitioning in different plant organs.

For number of produced inflorescences (Figure 1C), the authors observed positive linear response in relation to an increase of ionic concentration, production of 37.8 inflorescences $plant^{-1}$ at the highest concentration (125%), corresponding to an increase of approximately 165% in relation to the smallest tested concentration (25%). For this crop, Rodrigues *et al*. (2014) observed good response for inflorescence production in relation to nutrient availability in the soil, also being represented by a positive linear function, proving the influence of the availability of nutrients for jambu flowering.

Similarly to what was observed for inflorescence number, ionic concentrations increased linearly for leaf area, showing leaf expansion of 1650.8 cm^2 $plant^{-1}$ at the highest ionic concentration (125%). Andrade (2019), evaluating the influence of ionic concentration on lettuce cultivars under hydroponic NFT system, also observed an increase in leaf area of plants as the concentration of the solution was increased, which was represented by a positive linear function. Silva *et al.* (2019) observed that an increase in ionic concentration of the solution promoted a linear increase in leaf area of the lettuce.

We noticed that the production of leaves and inflorescences of jambu under hydroponics system is responsive to an increased nutrient availability in the nutrient solution due to its linear responses, showing that the plant can also express higher responses when using higher ionic concentrations. Thus, concerning productivity, these results are interesting for the crop, since both leaves and inflorescences of jambu are commercialized (Sampaio *et al.*, 2018).

Both shoot dry and fresh mass (Figures 2A and 2B) increased proportionally to the increase of ionic concentration of the nutrient solution, being higher responses observed in plants grown in a solution at 125%, 63.9 and 6.9 g plant^{-1}, respectively, representing an increase of 184% for fresh mass and 83% for dry mass comparing with values obtained in ionic concentration at 25% (22.5 and 3.8 g plant^{-1}, respectively).

Positive effect on production considering an increase of ionic concentration in hydroponics system had already been demonstrated for several vegetable species (Cometti *et al.*, 2008; Genuncio *et al.*, 2012; Portela *et al.*, 2012). Luz *et al.* (2012), evaluating the effect of different ionic concentrations (50, 75, 100 and 125%) of the formulation recommended by Furlani *et al.* (1999), observed an increase in shoot dry and fresh mass accumulation in parsley (*Petroselinum crispum*) and coriander (*Coriandrum sativum*). Evaluating productive responses of lettuce cultivars under hydroponics system under different ionic concentrations (50, 75 and 100%), Genuncio *et al.* (2012) observed a positive effect with a nutrient availability increase, being the highest response achieved at 100% of ionic strength.

Based on these results, studies relating the effect of nutrient solution formulations and their concentrations on vegetative and reproductive stages for each species show high relevance. Thus, using these studies, it is possible to indicate different moments regarding periodic nutrient replacement, establish nutritional requirements for each species, as well as reduce the nutrient concentration in the solution without yield loss, thus making it possible to reduce production costs in order to increase the nutrient use efficiency by crops (Cometti *et al.*, 2008).

For root fresh and dry mass (Figures 2C and 2D), the authors noticed that an increase in concentration promoted linear increases, and the maximum responses (16.7 and 2.0 g plant^{-1}, respectively) at 125% concentration, corresponding to increases of 242.5 and 71.4%, respectively, when compared with values obtained at 25% concentration (4.9 and 1.2 g plant^{-1}). For jambu, despite the increase in salinity due to the increase of the ionic concentration of the nutrient solution, the crop did not show a reduction in biomass productivity and/or characteristic symptoms of plants under salt stress (Negrão *et al.*, 2017). Similarly, for lettuce crop, Cometti *et al.* (2008) also verified variations in root dry mass accumulation as the ionic concentration of Furlani (1997) nutrient solution increased, observing that more diluted solutions obtained less dry mass accumulations than concentrated solutions.

In relation to jambu reproductive performance, the authors verified that fresh and dry mass of the inflorescence were influenced by the increase of ionic concentration, being represented by increasing linear functions (Figure 2E and 2F), the ionic concentration at 125% stood out, showing the highest responses observed in this study (11.0 and 1.8 g plant^{-1}, respectively). Phenological stages directly influenced on nutritional demand of plants, being flowering and fruiting the periods of greatest demand for nutrients, since inflorescences and fruits represent drain organs with high demand for photoassimilates to reach their maturation (Marschner, 2012; Taiz *et al.*, 2017). This behavior can explain the greatest flowering of jambu obtained at a concentration of 125%, since in this concentration larger availability of nutrients for growth and development of a plant can be verified, when comparing with other tested concentration.

Therefore, the electrical conductivity adjustment (ionic concentration) for hydroponics cultivation of jambu is a central parameter for obtaining greater yields both for fresh mass of shoot and inflorescence. Thus, the results obtained in this study justify EC adjustments in other commercial standard solutions used in vegetable production under hydroponic systems (Furlani *et al.*, 1999), based on EC around 3.3 dS m^{-1}, since this value corresponds to 125% ionic strength of Hoagland & Arnon standard solution (1950). In general, it is important to highlight that the nutrient solutions of current standards nutritional

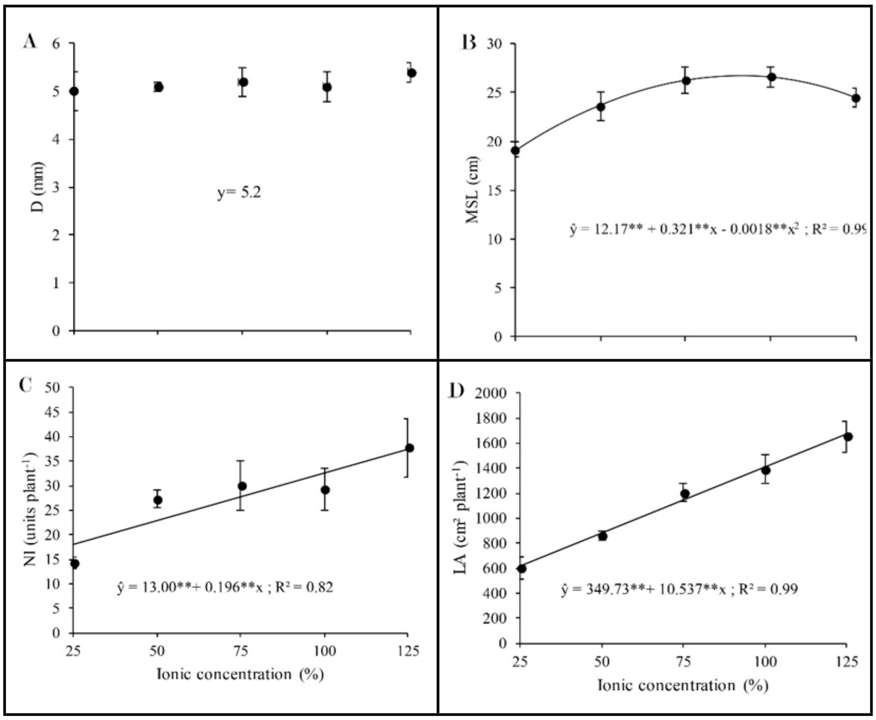

Figure 1. Stem diameter (D), main stem length (MSL), number of inflorescences (NI) and leaf area (LA) of jambu plants in relation to the variation in ionic concentration in the nutritive solution. **significant at 1% probability; *significant at 5% probability by t test. Belém, UFRA, 2020.

solutions have as a common ancestor the solution proposed by Hoagland & Arnon, (1950), with slight variations in macro and micronutrient levels between them (Cometti *et al*., 2008; Furlani *et al*., 1999), thus making the adjustment for other formulations interesting.

Gas exchanges were influenced by variations of ionic concentration in nutrient solution, as observed for growth and production, except of *Ci* and *Ci/Ca* ratio which obtained averages of 283.4 µmol mol^{-1} and 0.73, respectively (Figure 3).

For *A*, *gs*, *E* and *A/Ci*, the authors observed quadratic effects of ionic concentration in nutrient solution. Thus, an increase of ionic concentration promoted positive responses for these traits up to reaching maximum value of 84, 70, 80 and 83%, respectively.

Thus, the authors observed that as the photosynthetic rate increased, a similar increase in stomatal conductance, which is related to stomatal opening for CO_2 entry and transpiration was noticed, suggesting that a reduction in both *gs* and *E* is associated with a decrease in *A*. This is because, although a decrease in the opening of the stomatal pore was verified, no changes in *Ci* and in *Ci/Ca* ratio were noticed, which indicates that the reductions observed for liquid photosynthesis from the estimated concentration of 84% of the ionic concentration are not of stomatal order, but due to a biochemical limitation of the process. So, even with the stomatal closure, the photosynthetic machinery in the chloroplast not being compromised, CO_2 will continue to be fixed, thus promoting a reduction in the amount of internal CO_2 (Lemos Neto *et al*., 2020). However, this effect was not observed in this study, since no significant variations of *Ci* (Figure 3D), as well as *Ci/Ca* ratio were observed (Figure 3E).

In addition, even with CO_2 availability, reductions in the efficiency of carboxylation from 83% ionic strength could be noticed (Figure 3F), indicating that no CO_2 assimilation for synthesis of organic compounds was observed. This fact further reinforces the possible biochemical limitation throughout the photosynthesis process. According to Marschner (2012), the increase in EC of the nutrient solution can cause changes of the photosynthetic electron transport chain and/or the enzyme CO_2 fixation system, with negative effects on the photosynthetic rate (Baron *et al*., 2015).

Andrade (2019), evaluating physiological responses of lettuce cultivars in hydroponics system under ionic concentration variations, verified reductions in *A*, *gs*, *E* and *A/Ci* in more concentrated solutions, being represented by quadratic functions. We highlight that, as well as in the present study, the mentioned author did not observe significant effect of *Ci* and *Ci/Ca* ratio with an increase of ionic concentration of the solution. Thus, the loss of photosynthetic efficiency observed in this study was also related to possible non-stomatic limitations, mainly due to differential rates in the absorption of nutrients such as Ca and Mg, which had their levels reduced as the electrical conductivity of the solution increased

Baron *et al*. (2015), evaluating the influence of nutrient availability of Hoagland & Arnon nutrient solution 2 (1950) on gas exchanges, nutritional state and leaf biomass production of star fruit (*Annona emarginata*) seedlings, observed that the plants grown under 100% ionic concentration showed lower contents of Mg, Ca and S in their leaves in relation to other concentrations. Besides, these authors also observed reduction of liquid photosynthesis and instant carboxylation efficiency at 100% ionic concentration, which indicates that the changes in mineral composition

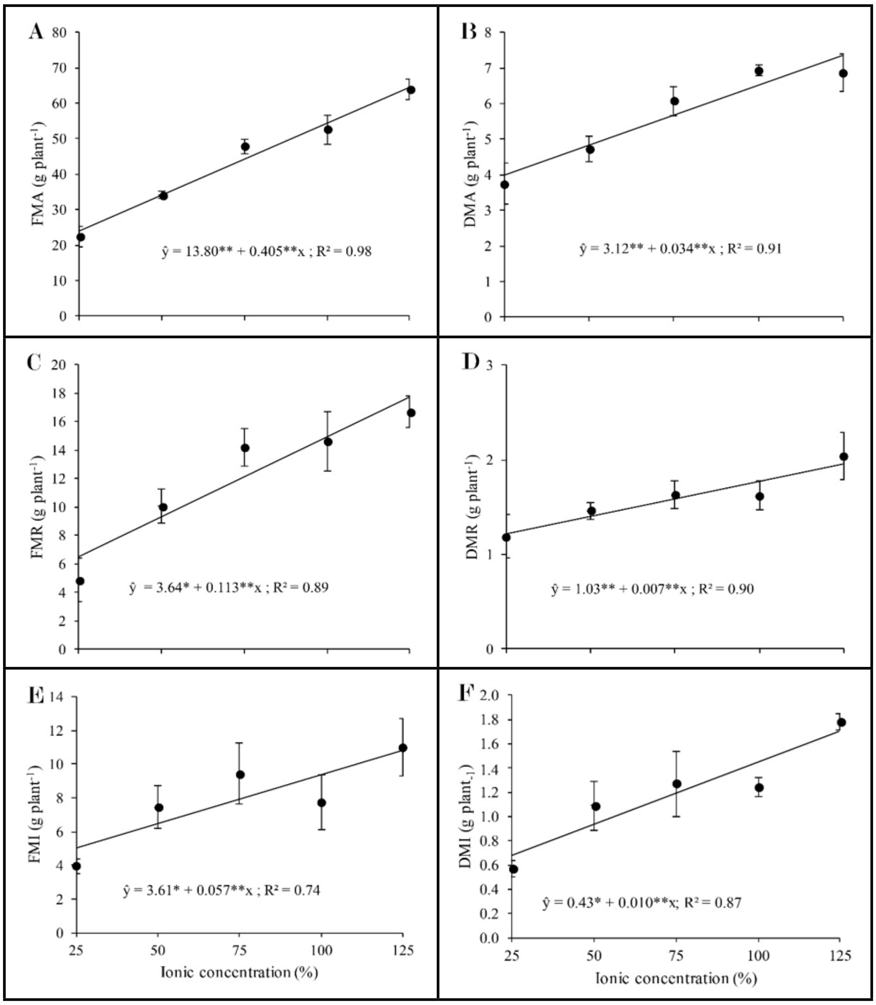

Figure 2. Shoot fresh mass (FMA), shoot dry mass (DMA), root fresh mass (FMR), root dry mass (DMR), fresh mass of inflorescence (FMI) and dry mass of inflorescence (DMI) of jambu plants in relation to the variation of ionic concentration in the nutrient solution. **significant at 1% probability; *significant at 5% probability by t test. Belém, UFRA, 2020.

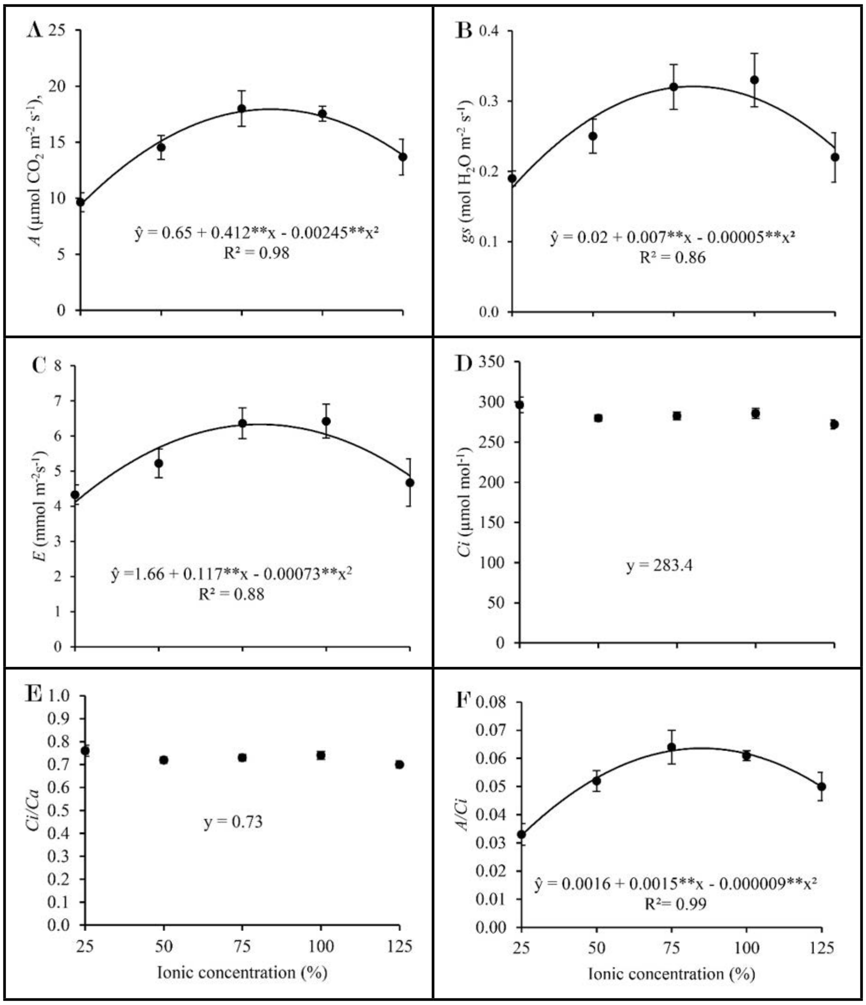

Figure 3. Liquid photosynthesis (A), stomatic conductance (gs), transpiration (E), internal CO2 concentration (Ci), Ci/Ca ratio (Ci/Ca) and instantaneous carboxylation efficiency (A/Ci) of jambu plants in relation to ionic concentration variation in nutrient solution. **significant at 1% probability; *significant at 5% probability by t test. Belém, UFRA, 2020.

affect photosynthetic performance of the crop.

Growth and productive responses observed in jambu as the concentration of nutrients in the nutrient solution increased may be related to the increase in physiological traits. However, we highlight that reductions in *A* (Figure 3A) and *A/Ci* (Figure 3F) in higher nutrient concentrations were noticed, which corroborate most of the results obtained for other variables. We verified that jambu throughout its vegetative and reproductive cycle adapted to this condition of loss of photosynthetic efficiency, though. This is because a linear increase in growth (Figure 1) and productivity of the crop was noticed (Figure 2) with the increase of ionic concentration in the nutrient solution, mainly using the concentration of 125%.

Thus, the authors concluded that ionic concentrations in the nutrient solution influenced on growth, production and physiological traits of jambu, considering that to obtain greater biomass accumulation, we suggest the ionic concentration of 125% of Hoagland & Arnon standard solution (1950), which corresponds to an electrical conductivity of 3.3 dS m^{-1}.

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