GAS EXCHANGE, PHISIOLOGICAL INDEXES AND IONIC ACCUMULATION IN *Annona emarginata* (Schltdl.) H. Rainer SEEDLINGS IN NUTRIENTS SOLUTION¹

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ABSTRACT - "Araticum-de-terra-fria" (Annona emarginata (Schltdl.) H. Rainer) has been consider a good alternative in rootstock production for the main commercial Annonaceae species. Although this species develops in different soil and climate conditions, there is no understanding by the physiological responses of this species at different nutritional levels. Thus, the objective of this study was to evaluate the influence of different ionic strengths on development of vegetative species known as "Araticum-de-terra-fria". It was evaluated in seedlings grown in different ionic strengths (25% I, 50% I, 75% I and 100% I) of the complete nutrient solution Hoagland and Arnon (1950) nº 2, for 140 days, the following characteristics: Gas Exchange (CO, assimilation rate, stomatal conductance, internal CO, concentration, transpiration rate, water use efficiency, Rubisco carboxylation efficiency); Vegetative growth characteristics (diameter, leaf number, dry matter); Physiological Indexes (leaf area ratio, specific leaf area, relative growth rate, net assimilation rate, leaf weight ratio) and Ionic Accumulation (nutrients leaf analysis). Seedlings grown under 50% I showed the highest values of Leaf CO, assimilation rate, water use efficiency, carboxylation efficiency, growth, relative growth rate, net assimilation rate and ionic accumulation in the total dry matter. So it is concluded that "Araticum-de-terra-fria" seedlings grown under intermediate nutrient concentrations of complete nutrient solution Hoagland and Arnon (1950) no 2, explored more adequately their physiological potential that justify their adaptation in different nutritional conditions and allow reducing the amount of mineral nutrition of seedlings production.

Index terms: Annonaceae, Araticum-de-terra-fria, seedlings, rootstock, macronutrients.

TROCAS GASOSAS, ÍNDICES FISIOLÓGICOS E ACÚMULO IÔNICO EM PLANTAS JOVENS DE *Annona emarginata* (Schltdl.) H. Rainer EM SOLUÇÃO NUTRITIVA

RESUMO - "Araticum-de-terra-fria" (Annona emarginata (Schltdl.) H. Rainer) tem sido considerada uma boa alternativa na produção de porta-enxertos para as principais espécies comerciais da família botânica Annonaceae, tais como atemoia, fruta-do-conde (ou pinha) e cherimólia. Embora esta espécie se desenvolva em diferentes tipos de solos e condições climáticas, não está elucidado como ela responde fisiologicamente sob diferentes disponibilidades nutricionais. Desta maneira, o objetivo deste estudo foi avaliar como diferentes forças iônicas interferem no desenvolvimento de plantas jovens desta espécie, também conhecida como "Araticum-de-terra-fria". Avaliaram-se, em plantas jovens de "araticum-de-terra-fria", cultivadas em diferentes forças iônicas (25% FI, 50% FI, 75% FI e 100% FI) da solução nutritiva completa nº 2 de Hoagland e Arnon (1950), por 140 dias, as seguintes características: Trocas Gasosas (assimilação de carbono, condutância estomática, concentração de carbono interno, transpiração, eficiência do uso da água e eficiência de carboxilação da enzima Rubisco); Crescimento Vegetativo (diâmetro de caule, número de folhas, altura e massa de matéria seca); Índices Fisiológicos (razão de área foliar, área foliar específica, taxa de crescimento relativo, taxa de assimilação líquida de carbono, razão de massa foliar e Acúmulo Iônico (análise foliar). As plantas cultivadas em 50% de força iônica foram aquelas com os maiores valores de assimilação de carbono, eficiência do uso da água, eficiência da enzima Rubisco, taxa de crescimento relativo, taxa de assimilação líquida de carbono e acúmulo iônico das folhas. Conclui-se que plantas jovens de "araticum-de-terra-fria", mantidas em concentrações intermediárias de nutrientes da solução nutritiva completa nº 2 Hoagland and Arnon (1950), exploraram seu potencial fisiológico, o que justifica sua adaptação em diferentes condições nutricionais e permite reduzir a quantidade de elementos minerais na produção de mudas.

Termos para indexação: Annonaceae, Araticum-de-terra-fria, produção de mudas, porta-enxerto.

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INTRODUCTION

The genus *Annona* includes several economically important subtropical and tropical fruit tree species including cherimoya (*A. cherimola* Mill.), soursop (*A. muricata* L.), sugar apple (*A. squamosa* L.), and atemoya (A. squamosa L. × A. cherimola Mill). The species of the genus Annona can present importance in folk medicine as Anticarcinogenic treatment, antipyretic effects, cramps, diarrhea and digestive problems; action in agriculture as a natural insecticide; ecological and ornamental, ecological recovery of degraded areas (*Annona rugulosa* Schltdl.) and power base of animals belonging to local wildlife (MITTAL et al., 2010; OKOYE et al., 2012).

The production of high-quality seedlings requires a rootstock compatible with the canopy and resistant or tolerant to pathogens (PADILLA and ENCINA, 2011). Thus, the species Annona emarginata (Schltdl.) H. Rainer variety "terrafria" has been successfully evaluated, although empirically, as an alternative for the production of atemoya, soursop and cherimoya seedlings (BETTIOL NETO et al., 2006). In addition, the normal nutrient patterns in leaf dry matter over the cultivation time must be known since they provide highly important information for a nutritional management plan aimed at meeting the demand for nutrients (FRANCO and PRADO, 2008) and assuring the advantages of clonal propagation and consequently productivity.

However, since *A. emarginata* species is native from the South American continent and present natural occurrence in nutritionally poor soil regions, this species still little studied, its physiological behavior must be assessed, especially at the initial development stage, in order to provide information to safely enhance the production of high-quality seedlings, assuring the increasing demand of the international market for Annonaceae fruits (BETTIOL NETO et al., 2006).

Besides, several nutrient solutions have already been proposed to fruits (Fig plants), vegetable crops (lettuce plants) and, in some cases, there are remarkable and excessive differences in the employed macro and micronutrient concentrations (SEVIL KILINC et al., 2007; HELBEL JUNIOR et al., 2008). Although the nutritional solutions super estimate the quantities of nutrients offered the plants these are important for the study of the nutritional needs of plant species, in which the variation of the concentrations of nutrients in these solutions may reflect the real needs of the vegetal (RESH, 2001).

As, however, there are no studies that evaluate the needs of the species Araticum-de-terra-fria, a possible starting point is the variation of nutrient concentration in nutrient solution no. 2 of Hoagland and Arnon (1950).

Photosynthetic and developmental patterns are affected by variations in the availability of mineral elements due to the direct effect of the last on several metabolic steps; however, in our literature review, descriptions of how these mechanisms are set for Araticum-de-terra-fria were not found. In addition, nutrient solutions have been widely used in studies on plant physiology, especially those related to mechanisms coordinating the plant growth (LARCHER, 2006; MARSCHNER, 2012).

In this context, different ionic strengths of nutrient solution do not affect the development of Araticum-de-terra-fria plantlets. So, the objective of this study was to evaluate the influence of different ionic strengths of Hoagland and Arnon (1950) solution on some physiological and developmental of *Annona emarginata* (Schltdl.) H. Rainer variety "terra-fria".

MATERIAL AND METHODS

Experimental local and plant material. The experiment was carried out in a greenhouse in Botucatu Municipality, State of São Paulo, Brazil, between July 2008 and February 2009. This locality presents the following geographic coordinates: 48° 24' 35"W, 22° 49'10"S and 850m mean altitude above the sea level. Annona emarginata (Schltdl.) H. Rainer variety "terra-fria" seeds were subjected to phytosanitary treatment with sodium hypochlorite (1%) and fungicide CAPTAN® at 0.2% active ingredient. After this treatment, sowing was done on polystyrene trays filled with expanded vermiculite. When seedlings presented 7-10cm (height) mean size, they were transplanted to pots of around 1.5L volume filled with continuously aerated nutrient solution.

Treatments. Treatments consisted of varying ionic concentrations (ionic strength, I) of complete nutrient solution Hoagland and Arnon (1950) n° 2 which had a concentration of 100% and was thus considered 100% I, electrical conductivity (2.0 \pm 0.02 mS cm⁻¹ EC). From the initial concentration, subsequent dilutions were done in demineralized water up to a concentration of 75%, i.e. 75% I (1.50 \pm 0.2 mS cm⁻¹ EC), and successively to 50% I (1.00 \pm 0.2 mS cm⁻¹ EC) and 25% I (0.5 \pm 0.02 mS cm⁻¹ EC). The volume of PA salts (pure for analysis) used as macronutrient sources [NH₄H₂PO₄, KNO₃,

 $Ca(NO_3)_3.4H_3O$ and $MgSO_4.7H_3O$] were 0.50, 1.50, 1.0 and 0.5 mL L⁻¹, respectively, in the Treatment 25% I; 1.0, 3.0, 2.0 and 1.0 mL L⁻¹ in the Treatment 50% I; 1.5, 4.50, 3.0 and 1.50 mL L⁻¹ in the Treatment 75% I; and 2.0, 6.0, 4.0 and 2.0 mL L⁻¹ in the Treatment 100% I (control). For micronutrient sources (H₂BO₂; MnCl₂.4H₂O; ZnSO₄.7H₂O; CuSO₄.5H₂O; H₂MoO₄. H₂O; Fe-EDTA and FeSO₄.7H₂O), 0.25 mL L⁻¹ of each salt was used in the Treatment 25% I; 0.50 mL L⁻¹ in the Treatment 50% I; 0.75 mL L⁻¹ in the Treatment 75% I; and 1.0 mL L⁻¹ in the Treatment 100% I (Table 1). The electrical conductivity (EC) of the nutrient solution was daily monitored by using a laboratory bench EC meter (DIGIMED®, CD-21 model), and pH was daily adjusted to 6.0 ± 0.5 by using a laboratory bench pH meter (DIGIMED®, DMPH-3 model). The nutrient solution was renewed weekly or whenever safety intervals changed for each Treatment (± 0.2 mS cm⁻¹ relative to the original value used in each Treatment), and demineralized water was employed to replace the evapotranspirated water. The control of pests in "Araticum-de-terrafria" seedlings was done through manual cleaning (cochineals) and by using tobacco syrup, known as natural insecticides which represent low risk to the environment.

Gas exchange. It was used the open system Infra-Red Gas Analyzer (IRGA, LI-6400 model, LI-COR) with CO, and water vapor measured through infrared radiation. These measurements were performed at 63, 98 and 126 DAT (Days After Transplant) from 9 to 11 a.m., on a sunny day, by selecting 5 plants (replicates) per Treatment, choosing and standardizing their second or third fully expanded leaf. The evaluated Gas Exchange features were: net CO₂ assimilation rate $(A_{net}, \mu mol)$ CO, $m^{-2}s^{-1}$), stomatal conductance (g_s , mol $m^{-2}s^{-1}$), internal CO_2 concentration $(C_i, \mu mol CO_2 mol$ ¹air) and transpiration rate (E, mmol water vapor m⁻²s⁻¹). Water use efficiency (WUE, μmol CO, (mmol H₂O)⁻¹) was determined through the ratio between CO₂ assimilation and transpiration rate (A_{pet}/E) and the Rubisco (Ribulose 1,5-Bisphosphate Carboxylase/Oxygenase, EC 4.1.1.39) instantaneous carboxylation efficiency $(A_{\rm nef}/C_i)$.

Ionic accumulation. The levels of nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and sulfur (S) in the leaves were quantified at 28, 56, 112 and 140 DAT in the Laboratory of Plant Mineral Nutrition, Department of Natural Resources/Soil Sciences, School of Agronomical Sciences (FCA), São Paulo State University "Júlio de Mesquita Filho" (UNESP), Botucatu *campus*, State of São Paulo, Brazil. Based

on these results, calculations for unit adequacy were done, i.e. conversion of g kg⁻¹ into g plant⁻¹, for each mineral element accumulated in the plant.

Vegetative Growth. At 28, 56, 84, 112 and 140 DAT, the following characteristics were measured: stem diameter (mm); height (cm); leaf number (units); and leaf area (cm²) with an area meter (LI-COR®, LI-3100 model). The plant material was washed in demineralized water and dried in a forced aeration oven at 65°C until constant weight in order to obtain leaf, stem, root and total dry matter (g), measured with an analytical balance of up to 0.001g sensitivity.

Physiological Indexes. Harvests were performed at 28, 56, 84, 112 and 140 DAT, using 5 plants (replicates) per Treatment in each evaluation period. The seedlings were separated into leaf, stem and root and leaf with results expressed as square decimeters (dm²). After quantification of leaf area and total and leaf dry matter at every 28 days until the final experimental period (140 DAT), the physiological indexes RGR (Relative Growth Rate), NAR (Net Assimilation Rate), LAR (Leaf Area Rate), SLA (Specific Leaf Area) and LWR (Leaf Weight Rate) were obtained through their respective mathematical formulas, according to the recommendation in several papers directed to quantitative growth analysis. Thus, the total dry matter, leaf blade and leaf area plants, submitted to the different Treatments, were adjusted as a function in the time, considering the version ln $y = a + bt + ct^2$ to dry matter > 0, according to the equations: $DM(y) = a.e (bt + ct^2)$ and LA(y1) = a.e $(b_1t + c_1t^2)$, where DM, dry matter and LA, leaf area (RADFORD, 1967). The physiological indexes RGR and NAR were calculated by solving the integrals through numerical methods for each replicate of all Treatments by using the software SAS. To calculate RGR, in g g⁻¹day⁻¹, it is assumed that new growth is simply a function of the existent dry matter and this index is a function of the initial size, i.e. the increase in grams. It was calculated by the equation: **RGR** = 1/(TDM). dTDM/dt, where d/dt = (log TDM); TDM represents the total dry matter. NAR, in g dm⁻² day⁻¹, expresses the net photosynthesis rate, in terms of produced dry matter, in grams, per square decimeter of leaf area, per time unit: NAR = 1/(LA)dTDM/dt, where: d/dt = (log TDM); TDM represents the total dry matter. LAR, in dm² g⁻¹, expresses the useful leaf area for photosynthesis and was obtained from the instantaneous values of leaf area (LA), responsible for the interception of light energy and CO₂, and total dry matter (TDM), resultant from photosynthesis. LAR is considered a measurement of the photosynthetic capacity of a plant according

to the equation: **LAR** = **LA/TDM**. SLA, in dm² g⁻¹, indicates the inverse of leaf thickness and was obtained through the ratio between leaf area (LA) and leaf dry matter (LDM): **SLA** = **LA/LDM**. LWR is expressed as g/g and defined as the ratio between leaf dry matter (LDM) and total dry matter (TDM): **LWR** = **LDM/TDM**.

Experimental Delineament and Statistical Analysis. The plants were conducted and distributed into randomized blocks with four Treatments and five replicates, each one composed of one plant. Five data collections were done to measure plant growth and calculate the physiological indexes, in addition to three collections to evaluate leaf gas exchange, and four collections to analyze leaf nutrients. Data on plant growth variables were subjected to analysis of variance (ANOVA), and means compared by Tukey's Test at 5% significance ($P \le 0.05$). Statistical analyses were done by using the software SAS, version 9.0 (LITTELL et al., 2002).

RESULTS AND DISCUSSION

The humidity and temperature data recorded over the cultivation period for "Araticum-de-terrafria" seedlings in greenhouse are described in Figure 1. The Treatments resulted in changes in both variables of gas exchange and ionic accumulation which also resulted in changes in the patterns of development.

Gas Exchange. As regards photosynthetic responses, it must be emphasized that the access of the atmospheric CO₂ to the photosynthetic cells from the mesophyll occurs through stomatal opening and, therefore, variations in the stomatal conductance (g_e) directly affect A and E (ALVAREZ et al., 2011). In addition, g_s is influenced by the environment (CAMPOSTRINI et al., 2010), abscisic acid (MAHOUACHI et al., 2006) and mineral nutrition (MARSCHNER, 2012). In plants grown under 100% I, transpiration rate (E) decreased (Figure 3C), whereas A values were stable up to 98 DAT and decreased up to 126 DAT, similarly to $A_{\rm rad}/C_{\rm i}$ (Figure 3B), which resulted in CO₂ accumulation in the leaves (Figure 3F). Thus, there was higher WUE (Figure 4E) even with lower Rubisco activity since E values decreased over time. The reduction in Aand E values observed for plants kept under 100% I, reaching the lowest values at 126 DAT (Figure 3A and 3B), can be explain by increased EC in the nutrient solution may lead to reduced vegetative development for either the whole plant or individual parts due to ion accumulation (EVETT et al., 2012), directly affecting photosynthesis (CHATZISSAVVIDIS and

THERIOS, 2010).

Araticum-de-terra-fria seedlings kept in nutrient solution of 25% I had low net CO, assimilation rate (A_{net}) (Figure 3A), lower instantaneous carboxylation efficiency (A_{net}/C_i) (Figure 3B), and increased transpiration (E) (Figure 3C), which resulted in lower water use efficiency (WUE) (Figure 3D). It must be emphasized that the increased conductance (g_s) (Figure 3E) from 98 DAT did not lead to a higher net CO, assimilation rate; there was also higher internal CO, concentration (C_i) (Figure 3F). Furthermore, the highest C_i values were detected under 25% I, especially up to 98 DAT (Figure 3C), which is a result of the low efficiency of Rubisco in the carboxylation (Figure 3F). Thus, even with an increasing E in this period (Figure 3D) and a tendency toward increased stomatal conductance (Figure 3B), there was not CO₂ influx into the leaf due to the failure caused in the photosynthetic apparatus by the low ion availability in the nutrient solution, preventing the chemical potential reduction of the internal CO, and consequently a higher CO, influx into the leaf. A lower Rubisco activity leads to a lower organic synthesis, and since E is high, there is a decrease in WUE. Plants kept under 25% I had the lowest WUE values (Figure 3D), which may have directly influenced the stomatal conductance of seedlings and subsequently the enzymatic activities (BENLLOCH-GONZÁLEZ et al., 2010), affecting the use of mineral nutrients (WANG et al., 2010). The dependence of WUE on water and nutrient supply for C₂ plants was demonstrated in several studies, including that of Kumar and Dey (2011) with strawberry, De Barros et al. (2007) with corn, peanut and Sun et al. (2006) with wheat plants.

Plants kept under 50% I presented higher $A_{\rm net}$ values, relative to those under the remaining ionic strengths, from the first evaluation, even with patterns of g_{c} close to those under more elevated ionic strengths up to 98 DAT. On the other hand, A_{net} and $A_{\rm per}/C_{\rm i}$ values decreased up to 98 DAT, which resulted in CO_2 accumulation. The lower A_{net}/C_1 values may be due to the increased mean air temperature (Figure 1), since this species presents C₃ metabolism, the ideal temperature of which ranges from 25 to 30°C. However, the plants rapidly adapted, increasing A_{net} values. The higher A_{net} values for plants kept under 50% I, relative to the remaining Treatments (Figure 3A), may be due to the better utilization of the provided mineral elements, which resulted in increased photosynthetic rates for mature leaves or even higher photosynthetic capacity of the leaves (TUCCI et al., 2011). Thus, $A_{\rm net}$ and $A_{\rm net}$ /C $_{\rm i}$ values detected for plants grown under 50% I were higher in general (Figure 3A and 3F), relative to those observed for the remaining Treatments. Besides, those plants had high E values are related to the high rates of CO, assimilation and probably high water consumption, implying an enhanced development of the species (THONGBAI et al., 2010). The decreased E and increased A_{net} values, mediated by Rubisco activity, resulted in higher WUE for such plants (Figure 3F). It should be highlighted, therefore, that CO₂ diffusion and water loss through transpiration are independent processes (LARCHER, 2006). In addition, transpiration can be considered a "necessary cost" associated with stomatal opening to allow CO₂ diffusion from the air to photosynthesis (AFROUSHEH et al., 2010), besides allowing the mass flow of the mineral nutrients absorbed in the roots to the shoot (MARSCHNER, 2012); however, A_{net} and E are not always dependent variables, as observed in the present study. Among the factors that affect photosynthesis are the mineral elements (TAIZ and ZEIGER, 2010) the different ionic strength affect photosynthesis differently, as can be seen by the results. Considering therefore that ionic strength altering gas exchange, it must be determined how this 'pool' of elements were accumulated causing such changes.

Ionic Accumulation. The analysis of significant interactions between the evaluation periods and the different ionic strengths indicated that 25% I led to the lowest accumulation of all elements, which had significantly lower levels over time, differing from the results for 50% I, which led to the highest accumulation, especially at 112 and 140 DAT (Figure 4), and did not differ from 75 and 100% I in some evaluations. Thus, significant accumulations of N, P, Ca, Mg and S in the total dry matter were detected from 112 DAT, whereas K had a singular behavior, with accumulations from 56 DAT and no difference from 140 DAT. The specific observation of each element showed that N had higher accumulation in the plants kept under 50 and 75% I at 112 DAT (0.12 g plant⁻¹) and under 50% I at 140 DAT (0.13 g plant -1) (Figure 4A). Phosphorus and potassium presented significant accumulation at 112 and 140 DAT in the plants grown under 50% I [Figure 4B (0.01 g plant⁻¹) and 4C (0.06 g plant⁻¹), respectively]. Calcium (Figure 4D) and sulfur (Figure 4E) had a similar behavior, with higher values under 50% I at 112 DAT and no difference from 75 and 100% I at 140 DAT; however, Mg was significantly lower for plants kept under 100% I (Figure 4F) in the last evaluation.

Thus, the ionic strength of 50% *I* was observed efficiency maximum of mineral elements

with operations in photosynthesis, since the nitrogen, which is essential for the biochemical composition of several non-protein compounds such as coenzymes, secondary metabolites, polyamines, photosynthetic pigments (BENLLOCH-GONZÁLEZ et al., 2010) and fundamental role in the activity or quantity of Rubisco (URBAN, 2012). Thus, $A_{\rm ret}$ is closely related to the leaf N level (BAR-YOSEF, 2008; BAR-YOSEF et al., 2009), corroborating the present results, since the highest accumulation of this element was observed at the end of the experiment (Figure 4A), and the highest A_{net} values were detected for plants kept under 50% I (Figure 3A). Phosphorus is another element of great importance; it is considered crucial to keep phosphorylation reactions during CO, assimilation (WARD et al., 2011) besides positively helping other photosynthetic parameters such as g_a and WUE (NAEEM et al., 2010), which were observed in the present study when plants were grown under 50% I. Phosphorus levels below those ideal for plants result in decreased A_{net} values since many intermediate steps of CO, fixation involve phosphate sugars (MAATHUIS, 2009). Potassium is closely related to stomatal regulation, transpiration and osmoregulation (MARSCHNER, 2012). The present results showed that A_{net} values decreased as potassium availability was lower (FAN et al., 2011). Calcium plays a structural role in the cells and signalizes some plant responses to the environment, e.g. stomatal regulation (MCAINSH and PITTMAN, 2009). The apparent absence of effect on photosynthesis is due to the small quantities of this element needed for this metabolic activity (TAN et al., 2011). Magnesium, accumulated in plants kept under 50% I from 112 DAT, plays a fundamental role in photosynthesis, specially inducing light reactions in the stroma (DINAKAR et al., 2012), and is required for thylakoid stacking, enzymes involved in CO₂ assimilation and partition of assimilates between starch and sugars (DING et al., 2008). Sulfur, also first accumulated in plants kept under 50% I at 112 DAT, is responsible for the formation of sulfolipids, which are normally found at small proportions in the thylakoids (EPSTEIN and BLOOM 2006; IQBAL et al., 2012). Some plant proteins contain sulfur, such as ferredoxins, which participates in the photosynthetic process such as chlorophyll formation and electron transfer reactions (GILL et al., 2012). Thus, the suitable supply of mineral elements group led to a successful photosynthetic process, which resulted in higher plant development according to vegetative growth variables and physiological indexes (Table and Figure 2).

Vegetative Growth and Physiological Indexes.

D. BARON et al.

"Araticum-de-terra-fria" seedlings had significant differences in growth among Treatments over time, especially from 112 DAT, always with lower values for the plants grown under 25% I (Table 2). Significant differences were observed for diameter, height, leaf area, and leaf, stem and total dry matter from 112 DAT, and the plants grown under 50% presented the highest values relative to plants kept under 25%, although 50% did not differ from plants kept under 75 and 100% at 112 and 140 DAT. The exception includes plants grown under 100% I, the total dry matter of which decreased at 112 DAT (Table 2). As regards leaf number, the lowest values were obtained for plants kept under 25% I, also from 112 DAT, relative to the remaining Treatments, which were not significantly different (Table 2). However, significant differences in root dry matter were only observed at 140 DAT, with the highest values detected for plants kept under 50% I, significantly different from those kept under 25% I.

Leaf area ratio (LAR) of plants grown under different ionic strengths decreased over the cultivation period (Figure 2A) and was not significantly different among Treatments (Table 3). Conversely, specific leaf area (SLA) values increased over time (Figure 2B), but without significant differences, similarly to LAR (Table 3). Specific leaf area (SLA) did not present a constant behavior over the studied period, independently of the evaluated Treatment. The adopted Treatments led to lower RGR for plants kept under 50, 75 and 100% I over time, unlike those kept under 25% I (Figure 2C). However, plants grown under 50% I had the highest mean RGR values, differing from those grown under 25% but not from those kept under 75 and 100% *I* (Table 3). There was higher dry matter accumulation relative to the pre-existing material (RGR) for plants kept under 50, 75 and 100% I, with significant differences between mean values obtained under 50 and 25% I (Table 3). Thus, RGR showed a similar behavior to that described in the literature, with high values at the first intervals and subsequent decreases at the final intervals (DAVID et al., 2007). As regards net assimilation rate (NAR), there was a decrease over time for all Treatments, with the lowest values observed for plants under 25% I (Figure 2D), which differed from those under 50%, but both Treatments were not different from 75 and 100%. Leaf weight ratio (LWR) had the highest values over time under 100% I and the lowest values under 25% I (Figure 2E); however, there were not significant differences in the mean total values among the different ionic strengths (Table 3). Net assimilation rate (NAR) is resultant of the interaction between variations

in environmental factors (light, CO₂, temperature, water and nutrient availability, leaf age etc.) and specific differences of each species, according to their genotype (LAMBERS et al., 2008). NAR indicates the efficiency of the assimilatory system involved in dry matter production, estimating thus the net photosynthesis in this case, the highest NAR value was detected in plants kept under 50% *I* (Table 3), due to the better use of the supplied mineral elements. This results agree with those in studies developed in hydroponic systems with varying elements in the nutrient solution, including the studies of David et al. (2007), Alves (2008) and David and Boaro (2009), emphasizing that this comparison was done among plants of different species.

The Araticum-de-terra-fria seedlings responded to the different ionic strengths, with higher growth values (RGR and NAR) for plants kept under 50 and 75% I, lower values for those kept under 25% I, and intermediate values for those under 100% I. We can observed that drastic reductions in the supply of mineral elements (25% I), significantly decrease the plant development, similarly to the results obtained by Batista et al. (2003) to soursop seedlings in different fertilizations levels. However, under 100% I, which represents a "standard" solution for plants, photosynthesis and accumulation of mineral elements were lower, which shows that this species does not require large quantities of mineral elements to express its development potential. This plants seems to be similar a others wild plants adapted to nutrientpoor soils, that present a growth strategy named type I (slow growth), that are not more efficient in nutrient acquisition than cultivated or invasive species (type II, fast growth) (CHAPIN, 1980), besides low rate of maximum potential growth and low efficient use of nutrients when high strength ionic availability.

In this context, it can be concluded that, the use of 50% I was sufficient for an effective action of mineral elements on the different physiological processes, resulting in higher photosynthetic efficiency and consequently higher plant development that explain their rusticity and adaptation in different nutritional conditions.

TABLE 1- Nutrient composition of different ionic strengths (*I*) of complete nutrient solution n° 2 of Hoagland and Arnon (1950) used for the cultivation of "Araticum-de-terra-fria" seedlings.

Used compounds	Stock Solution (g L ⁻¹)	Treatments ionic strength (<i>I</i>) (mM L ⁻¹)			
Macronutrients (M) (1)		25%	50%	75%	100%
$\mathrm{NH_4H_2PO_4}$	115.0	0.12	0.24	0.47	0.63
KNO_3	101.1	0.84	1.68	2.52	3.36
$Ca(NO_3)_2.4H_2O$	236.1	0.97	1.95	3.90	5.19
$MgSO_4.7H_2O$	246.5	0.51	1.02	2.03	2.71
Micronutrients		25%	50%	75%	100%
$^{(1)} H_3 BO_3$	2.86	2.95	5.90	11.8	15.7
MnCl ₂ .4H ₂ O	1.81	1.86	3.71	7.42	9.90
$ZnSO_4.7H_2O$	0.22	0.22	0.45	0.91	1.20
CuSO ₄ .5H ₂ O	0.08	0.08	0.16	0.33	0.44
$H_2MoO_4.H_2O$	0.02	0.02	0.04	0.08	0.11
Fe Solution ⁽²⁾		25%	50%	75%	100%
Fe-EDTA	26.1	0.07	0.05	0.11	0.14
FeSO ₄ .7H ₂ O	24.9	0.06	0.05	0.10	0.14

(M) = Molar; (1) = dilution in distilled water and volume completed to 1 L; (2) = dilution in 700 mL distilled water containing 268 mL NaOH (40g/L) and volume completed to 1 L.

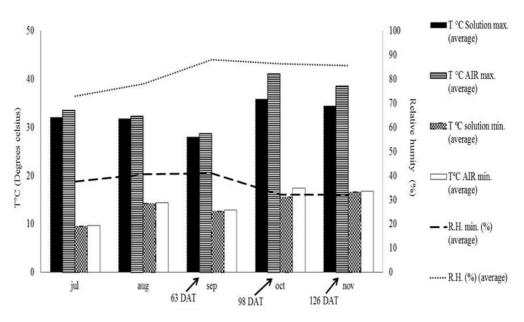
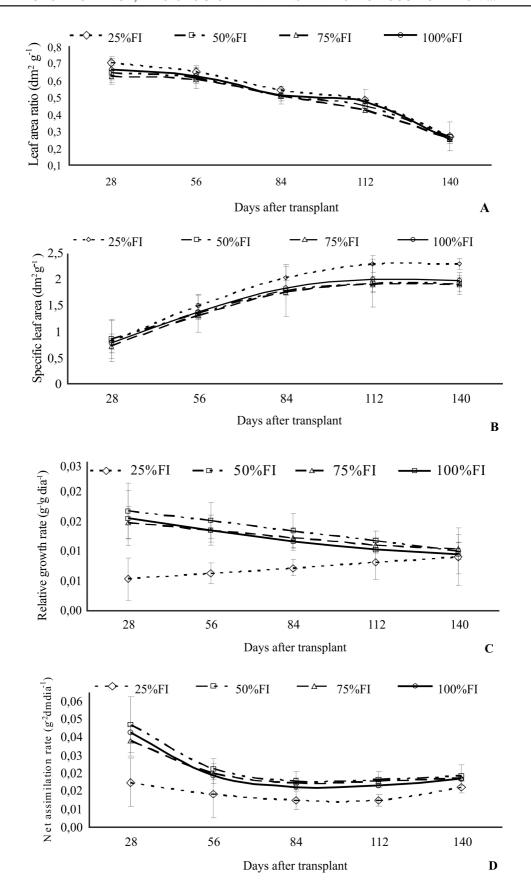


FIGURE 1 - Humidity and temperatures data of nutrient solutions in greenhouse recorded over the cultivation period for "Araticum-de-terra-fria" seedlings under different ionic strengths of complete nutrient solution no 2 of Hoagland and Arnon (1950). Arrows indicate the dates when leaf gas exchange was evaluated. DAT (Days after transplanting).

TABLE 2- Growth of "Araticum-de-terra-fria" seedlings under different ionic strengths (*I*) of complete nutrient solution n° 2 of Hoagland and Arnon (1950) over time. Results are the means and each value was obtained from 5 plants per Treatment (*n*=5)Uppercase letters represent the comparison of a same ionic strength at different harvests and Lowercase letters represent the comparison of the different each harvest; distinct letters indicate significant differences according to Tukev's Test at 5% (*P*<0.05). DAT. Days After Transplanting.

	according to Tukey's Test at 5% ($P \le 0.05$). DAT, Days After Transplanting.							
	Leaf number (units)				Diameter (mm)			
Treat/ DAT	25% I	50% I	75% I	100% I	25% I	50% I	75% I	100% I
28	4.8 Ac	6.0 Ac	5.6 Ac	5.4 Ab	2.1 Ad	2.3 Ac	2.2 Ad	2.18 Ad
56	9.2 Bbc	14.6 Abc	10 Bbc	9.4 Bb	2.9 Ac	3.1 Abc	3.1 Acd	3.08 Ac
84	12.8 Bb	15.6 ABb	16 Ab	15 Bb	3.3 Abc	3.9 Ab	3.7 Ac	3.60 Ac
112	20.4 Ba	28.4 a	28 Aa	27.6 Aa	3.7 Bb	5.2 Aa	4.9 Ab	4.82 Ab
140	24.4 Ba	28.6 Aa	29.2 Aa	28.4 Aa	5.466 Ba	6.058Aa	5.984 Aa	5.952 Aa
	Height (cm)				Stem dry matter (g)			
Treat/ DAT	25% I	50% I	75% I	100% I	25% I	50% I	75% I	100% I
28	7.08 Ab	8.3 Ad	8.20 Ad	7.50 Ac	0.17 Aa	0.19 Ab	0.19 Ab	0.19 Ab
56	14 Aa	13.9 Ac	13.42 Ac	13.18 Ab	0.19 Aa	0.21 Ab	0.19 Ab	0.19 Ab
84	15.6 Aa	19 Ab	18.66 Ab	17.98 Aab	0.3 Aab	0.32 Ab	0.32 Ab	0.32 Ab
112	18.46 Ba	24.7 Aa	22.50 ABa	a20.32 ABa	0.4 Bb	0.71 Aa	0.65 Aa	0.36 Ab
140	18.60 Ba	25.4 Aa	25.04 ABa	121.36 ABa	0.44 Bb	0.8 Aa	0.66 ABa	0.60 Aa
	Leaf area (cm²)				Root dry matter (g)			
Treat/ DAT	25% I	50% I	75% I	100% I	25% I	50% I	75% I	100% I
28	16 Ac	16.3 Ac	16 Ac	16.1 Ac	0.24 Aa	0.25 Ab	0.25 Ab	0.25 Ab
56	33.9 Abc	37.5 Ac	35.9 Ac	35.7 Abc	0.26 Aa	0.3 Ab	0.30 Ab	0.27 2Ab
84	58.8 Aab	61.4 Abc	71.9 Ab	70.7 Aab	0.31 Aa	0.33 Ab	0.33 Ab	0.32 Ab
112	81.6 Ba	121 Aab	107 Aa	105.9 Aba	0.42 Aa	0.83 Aa	0.75Aa	0.66 Aa
140	90.2 Ba	158.2 Aa	112.8 ABa	110.4 ABa	0.43 Ba	0.85 Aa	0.79 ABa	0.63 ABa
-	Leaf dry matter (g)				Total dry matter (g)			
Treat/ DAT	25% I	50% I	75% I	100% I	25% I	50% I	75% I	100% I
28	0.21 Aa	0.22 Ac	0.22 Ab	0.21 Ab	0.61 Ab	0.67 Ab	0.65 Ac	0.64 Ac
56	0.28 Aa	0.29 Abc	0.28 Ab	0.29 Ab	0.74 Ab	0.79 Ab	0.77 bc	0.75 Ac
84	0.32 Aa	0.35 Abc	0.35 Ab	0.33 Ab	0.93 Aab	1.02 Ab	1.03 Ac	0.98 Ac
112	0.44 Ba	1.01 Aab	0.7 Aa	0.69 Aa	1.27 Ca	2.41 Aa	2.11 Aa	1.71 Bb
140	0.448 Ba	1.014 Aa	0.873 Aa	0.831 Aa	1.32 Ba	2.6 Aa	2.23 ABa	2.12 ABa



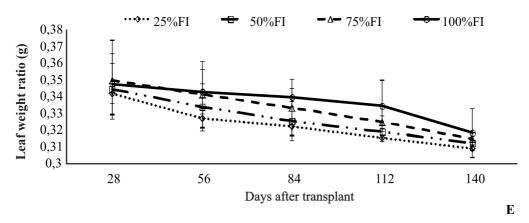
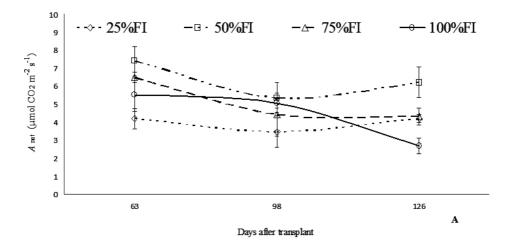
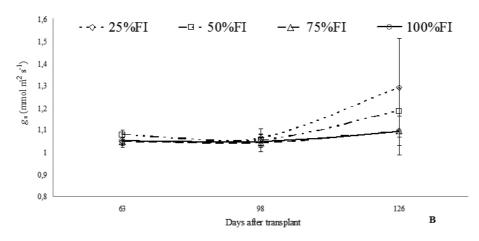


FIGURE 2- Physiological indexes for "Araticum-de-terra-fria" seedlings grown under different ionic strengths (I) of complete nutrient solution n° 2 of Hoagland and Arnon (1950). Results are the means (± S.E.) and each value was obtained from 5 plants per Treatment (n=5) in 5 evaluation periods (28, 56, 84, 112 and 140 Days After Transplanting). A – Leaf Area Ratio (LAR); B – Specific Leaf Area (SLA); C – Relative Growth Rate (RGR); D – Net Assimilation Rate (NAR); E – Leaf Weight Ratio (LWR). Values adjusted by the exponential quadratic equation.





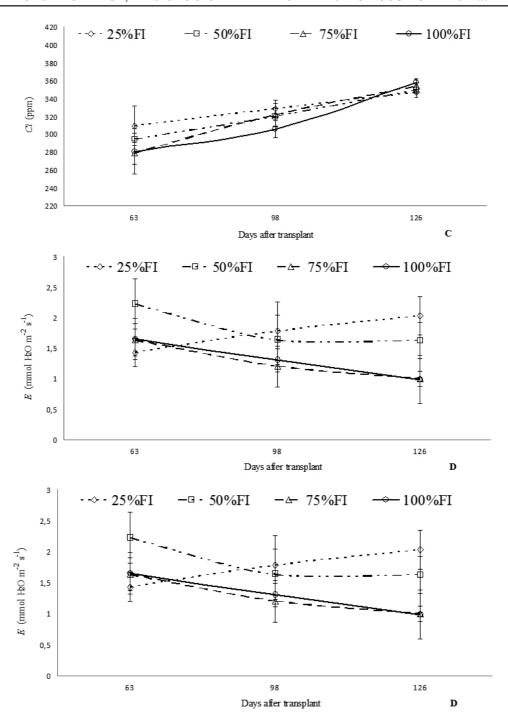
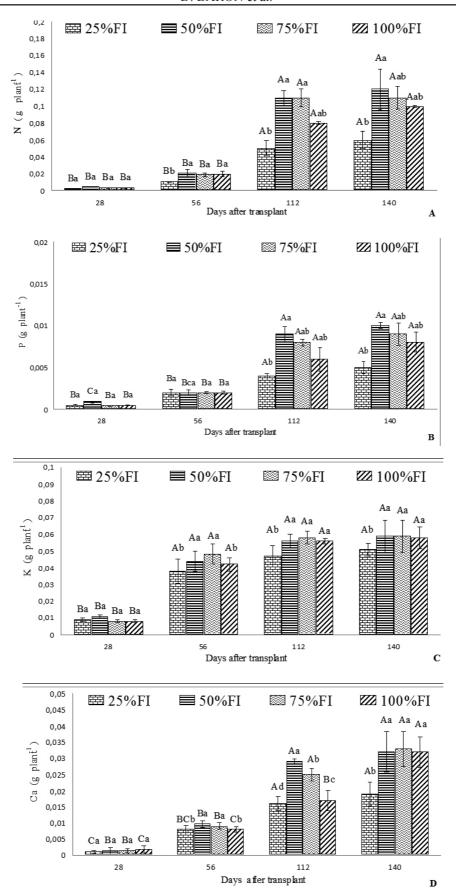


FIGURE 3-Leaf gas exchange for "Araticum-de-terra-fria" seedlings grown under four ionic strengths (*I*) of complete nutrient solution n° 2 of Hoagland and Arnon (1950) in 3 periods (63, 98 and 126 Days After Transplanting). Results are the means (\pm S.E.) and each value was obtained from 4 plants per Treatment (n=4) in 4 different evaluation periods. A – Net CO₂ assimilation rate (A_{net}); B - Stomatal conductance (g_s); C - Internal carbon concentration (C_i); D - Transpiration rate (E); E - Water use efficiency (WUE); F - Instantaneous carboxylation efficiency (A_{net}/C_i).



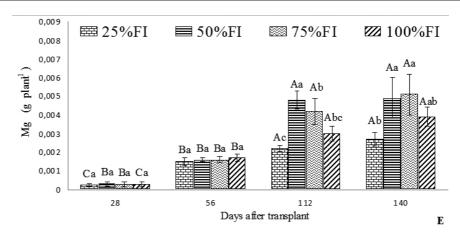


FIGURE 4-Leaf ion concentration as g plant¹¹ DM (dry matter) determined for "Araticum-de-terra-fria" seedlings grown under different ionic strengths (*I*) of complete nutrient solution n° 2 of Hoagland and Arnon (1950) in 4 evaluation periods (28, 56, 112 and 140 Days After Transplanting). Results are the means (± S.E.) and each value was obtained from four plants per Treatment (*n*=4). Uppercase letters represent the comparison of a same ionic strength at different harvests; distinct letters indicate significant differences according to Tukey's Test at 5% (*P*≤0.05). A – Nitrogen (N); B – Phosphorus (P); C – Potassium (K); D – Calcium (Ca); E – Sulfur (S); F – Magnesium (Mg). Lowercase letters represent the comparison of different ionic strengths at each harvest; distinct letters indicate significant differences according to Tukey's Test at 5% (*P*≤0.05).

TABLE 3 - Mean values of the physiological indexes LAR (Leaf Area Ration), SLA (Specific Leaf Area), RGR (Relative Growth Rate), NAR (Net Assimilation Rate) and LWR (Leaf Weight Rate) for "Araticum-de-terra-fria" seedlings grown under different ionic strengths (I) of nutritive solution n° 2 of Hoagland and Arnon (1950) over the cultivation period. Results are the means and each value was obtained from 5 plants per Treatment (n=5). For each Physiological Index, means followed by distinct letters significantly differed according to Tukey's Test at 5% ($P \le 0.05$).

Treatment / P.I.	LAR	SLA	RGR	NAR	LWR
25% I	0.5272a	1.788a	0.0072b	0.0153b	0.3228a
50% I	0.4949a	1.568a	0.0134a	0.0273a	0.3306a
75% I	0.4822a	1.519a	0.0124ab	0.0245ab	0.3371a
100%~I	0.5040a	1.598a	0.0121ab	0.0246ab	0.3410a
\overline{F}	0.72	0.32	6.53	4.48	0.22
P > F	0.5569	0.8128	0.0043	0.0182	0.879
<i>CV</i> (%)	11.47	19.44	21.58	25	10.78

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