Biofortification of Chia Genotypes with Lithium Hydroxide

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Abstract

Lithium (Li) is an important alkali metal that exists in the elemental form of nature. Thus, the objective was to evaluate the effect of foliar fertilization with doses of lithium hydroxide on the development and productivity of two chia genotypes, in the south of the state. The experiment was carried out at the Federal University of Tocantins, Campus de Gurupi-TO, in the agricultural year 2017/18 in pots of 8 dm^{-3} containing red-yellow dystrophic oxisol, deep and clayey texture, in a randomized block design, under a factorial 5x2 scheme, with four replications. The characteristics of plant height, upper stem height, stem diameter, bunch length, number of bunches, liquid photosynthesis, transpiration, stomach conductance and, after harvest (145 days), thousand grain mass, grain yield and Lí content in the grains were evaluated leaf and soil. The highest concentrations of lithium in the chia grains are obtained with the *application of 24.6 g ha^{-1} and 18.5 g ha^{-1} of LiOH for the genotypes originating in Paraguay and Argentina, respectively. The best responses in height, length of bunch, number of bunches, lithium content in the grain, mass of a thousand grains, liquid photosynthesis, transpiration and stomach conductance were obtained with the genotype from Paraguay.*

Keywords: *Salvia hispanica* L; lithium accumulation; development; dose.

Introduction

According to the essentiality criteria Li is not considered essential for plant development, however, there are reports that relatively low concentrations stimulate growth and accumulate this element in parts interesting for human consumption, however, high concentrations can result in negative effects on development and cause leaf necrosis (JIANG et al., 2014, HAWRYLAK-NOWAK et al., 2012; SHAHZAD et al., 2016).

On the other hand, the Li is an essential trace element of great importance in the human diet. Studies prove that intake at optimal levels in the body can influence metabolism and contribute to the prevention of diseases such as Alzheimer's, schizophrenia, phobic anxiety, depression, and suicide prevention (CIPRIANI et al., 2005; MATSUNAGA et al., 2015; MARTINSSON et al., 2013). Based on a review of Li food intake in different countries, a daily dose of 1.0 mg/day was proposed for a 70 kg person (GALLICCHIO, 2011; SCHÄFER, 2012).

The main sources of Li in the diet are grains, vegetables, dairy products, meat, and drinking water, however, Li levels in water and food in some parts of the world are below the recommendation of daily intake (NORRA et al., 2010; LIAUGAUDAITE et al., 2019). So far no reference values for Li in food have been established, although some research has already been conducted (EKMEKCIOGLU & MARKTL, 2006).

There is little research on the content of Li in crops, however, there is great evidence of deficiency of this element which makes it necessary to find strategies to increase its content in food and maintain adequate levels of Li in the human body (GONÇALVES et al. 2015; SILVA et al., 2019). An interesting strategy would be to consider adding Li through agronomic biofortification.

Biofortification is an agronomic practice that consists of increasing the concentration of one or more essential nutrients in edible parts of the plant during cultivation by handling fertilization via the soil or foliar without compromising crop yields (WHITE & BROADLEY, 2009). The agronomic practice via foliar fertilization is more effective mainly when the study involves a trace element, of the order of g ha$^{-1,}$ seeking a rapid response of the crop, to improve the distribution of the product applied and, at the same time, low production costs (EL-AAL et al., 2010; SILVA et al., 2019; ZODAPE et al., 2011).

Due to the importance of studies with biofortification of Li cultures, associated with the scarcity of information on the subject and the benefits that this element in optimal levels provides, this work aimed to evaluate the effect of fertilization via foliar doses of lithium hydroxide, in the development and productivity of two genotypes of chia, cultivated in the south of the state of Tocantins.

Material and methods

The experiment was conducted in the experimental area of the Federal University of Tocantins (UFT), University Campus of Gurupi, located in the southern region of the state of Tocantins, at an altitude of 280 m, at 11°43'45" latitude and 49°04'07" longitude. According to the Köppen classification, the climate of Tocantins is of the Aw type, since it has an average annual temperature of 24.9 °C, average annual rainfall of 1850 mm, with concentrations of rainfall in summer and with a dry winter, having the humid climatic type (B1) (ROLDÃO & FERREIRA, 2019). The data regarding precipitation, temperature, and relative humidity during the experiment were collected at the Gurupi Campus weather station, and are presented in Figure 1.

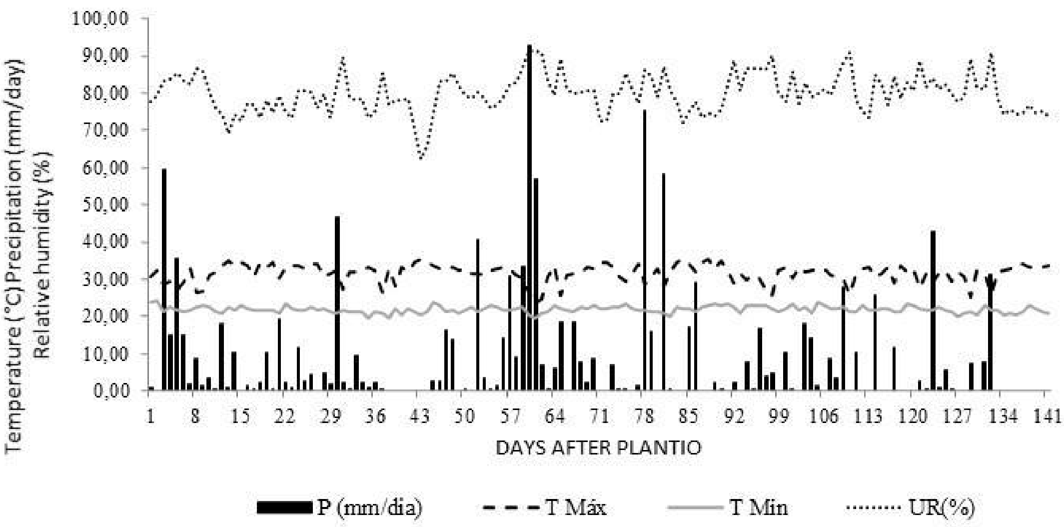

Figure 1 - Daily variation of air relative humidity (%), temperatures (°C), and rainfall (mm day) from December 2017 to April 2018, UFT, Gurupi – TO (Source: Weather Station of Gurupi - TO).

The experiment was installed in randomized block delineation in a 5x2 factorial arrangement scheme, with four repetitions. The first factor was constituted by five lithium doses (0, 10, 20, 30, and 40 g ha^{-1}) using as source the lithium hydroxide (LiOH, sigma-Aldrich, Duque de Caxias - RJ) and the second factor by two chia genotypes, being one from Paraguay and the other from Argentina (Argentina).

For better germination uniformity, chia seedlings were produced in polystyrene trays with 128 cells, using the commercial Nutrimax® substrate. During the period between sowing and transplanting, the trays remained inside an arc-covered vegetation house, 20 m long, 6 m wide, 1.75 m high. The side and front walls are made of 50% shaded canvas, the roof is covered with 150 microns of polyethylene plastic film, without climate control, and the trays are placed on the benches. The water was supplied to the seedlings through irrigations made twice a day manually, to keep the moisture content of the substrate always high. After 15 days the seedlings were transferred to pots with a volume of 8L to the open field, gluing 4 seedlings per pot. At 20 days after transplanting, thinning was performed, keeping one plant per pot spaced 0.50 m between blocks and 0.40 m inside the blocks.

The soil used was collected in the 0-20 cm layer of the Baobá farm in São Desidério (S 12°34.555' W 46°08, 405'), extreme West of Bahia, classified with dystrophic red-yellow latossolo, deep and clayey texture (EMBRAPA, 2013). To avoid the presence of harmful pathogens the soil was placed in raw cotton bags with dimensions of 50 x 70 cm (width and length) and inserted in a vertical analog autoclave (CS/Prismatec) with 300 Liter pedal, safety valve, and pressure regulation system at 120 °C for 2 hours for soil sterilization by wet heat.

The result of the physical-chemical soil analysis was: pH in $CaCl_2$ = 5.9; M.O (%) = 1.5; P (Honey) = 92.1 mg dm^{-3}; K = 90 mg dm^{-3}; Ca+Mg = 3.4 cmolc dm^{-3}; H+Al = 1.6 cmolc dm^{-3}; Al = 0.0 cmolc dm^{-3}; SB= 3.63 cmolc dm^{-3}; V = 69%; 450 g kg^{-1} of sand; 100 g kg^{-1} of silt and 450 g kg^{-1} of clay. The fertilization was performed according to the soil analysis, applying 60 kg ha^{-1} of P_2O_5 in the form of superphospatotriple (36%) and 60 kg ha^{-1} of K_2O in the form of potassium chloride (60%), mixed in the soil preparation in a homogeneous way. 125 kg ha^{-1} of nitrogen (46%) was applied in the form of urea in coverage at 30, 60, and 90 days after transplantation. During the conduct of the experiment, the soil was kept moist through individual daily irrigations in each experimental unit.

The culture was shown to be sensitive to common pests of other species, with two applications of insecticide at 80 and 90 days after transplantation for cow control (*Macrodactylus pumilio*) and whitefly (*Bemisia tabaci*) being used Alfacipermetrine (0.5 g i.a. ha^{-1}), for cow control, and Acetamiprid (75 g i.a. ha^{-1}) for whitefly control. No fungi or viruses were observed in any of the phenological phases of the plants.

The lithium concentrations were supplied twice via foliar, in the morning (between 8:00 and 9:00) being half applied at 70 days (before the plants' bloom), in the initial phase of bunches emission and, another half at 100 days, at the beginning of the grain filling after transplanting. To facilitate the application a stock solution was prepared weighing 0.5g (500 mg) of lithium diluted in 2 liters (2000 ml), obtaining standard LiOH solution (0.25 mg mL^{-1}). For example, 1.65 mL of stock solution was used for the treatment of 10 g ha^{-1} of Li. These doses of the standard solution were completed with 700 mL of water and applied evenly to the plants of each experimental unit. The doses were converted to hectare considering the population of 200,000.00 plants, exporting the doses per pot to the hectare. A high-pressure manual sprayer (Guaranyind) with a maximum pressure of 2070 KPa (300 PSI), a flow rate of 1.1 L/min, and an adjustable tip was used for the application. To avoid drift, a 1.0 m high plastic canvas was installed around the experimental unit.

Harvesting was done manually between 125 and 130 days after transplanting when the plants reached 80% of yellow leaves, or dark-colored, dried or dead (MIRANDA, 2012).

The characteristics evaluated were:

Height of plants (AP, cm) - obtained by measuring the length between the neck of the plants to the highest end of the bunches, using trend graduated in cm;

Height of the upper stem (HS, cm) - obtained by measuring the length between the neck of the plants to the highest end of the last stem, using trend graduated in cm;

Stem diameter (DC, mm) - obtained by measuring the central part of the plant, using a 150mm/6" stainless steel digital caliper (MTX);

Bunch length (CC, cm) - obtained by measuring the main bunch, using a graduated ruler in cm;

Number of bunches (NC) - determined through direct counting in the sampled plants;

Productivity (PD, g $vaso^{-1}$) - after threshing the bunches the grains were placed in paper bags measuring 7 x 11 cm (length x width) and inserted in an oven with forced air circulation (Solab, model SL-102) at 60 ºC until they reached a constant weight. Later the samples are weighed on a 0.01 g precision scale (Gehaka BK4000);

Mass of one thousand grains (MMG, g) - determined after the drying process, by counting one thousand grains with the help of histological tweezers of fine point (10 cm), weighed later on a precision scale of 0.01 g (Gehaka BK4000);

Li content (mg kg^{-1}) - after the drying process in an oven with forced air circulation (Solab, model SL-102) at 60 ºC, the samples of grains and leaves were finely crushed separately with the aid of a pestle with a kneader, according to the methodology described by Falco et al. (2017). The collection of the soil samples to determine the Li content was performed according to EMBRAPA (1997). Then the materials were submitted to wet digestion using sulfuric acid, perchloric acid, and catalytic mixture, composed of anhydrous sodium sulfate (Na_2SO_4) and copper sulfate pentahydrate ($CuSO_4.5H_2O$). The Li content was determined in the strata acquired, according to the methodology described by Malavolta et al. (1997). The reading was carried out in a Quimis (Q398M2) flame photometer, using a 10% (v/v) diluted standard solution to calibrate the apparatus, with a measuring range in clinical analyses for lithium (Li) from 0.0 to

1.5 mmol/L. To facilitate the measurement the digester was diluted in the proportion of one part to five of deionized water (5 ml + 25 ml) and transferred to a 50 ml container before reading.

The gas exchanges were measured in completely expanded leaves, located in the intermediate part of the plant, from 8:00 a.m. to 11:00 a.m. on a sunny day. The rate of liquid photosynthesis (A) (µmol of CO_2 m^{-2} s^{-1}), stomach conductance (GS) (mol of CO_2 m^{-2} s^{-1}), and perspiration (E) (mmol of H_2O m^{-2} s^{-1}) were determined using open system equipment of photosynthesis with CO_2 and water vapor analyzer by infrared radiation (*Infra-Red Gas Analyser* - IRGA, LCiSD model, from ADC System, UK).

The data obtained were submitted to analysis of variance using the F test, adopting 1 and 5% probability. Then they were submitted to regression analysis, evaluating the significance of betas and the coefficients of determination using the Sisvar program version 5.6 (FERREIRA, 2008). The graphs of the regressions were plotted using the statistical program SigmaPlot version 12.0®.

Result

The summary of the analysis of variance with the mean square, as well as the respective mean values and coefficients of variation for all agronomic characteristics, Li content in grain, leaf, and soil are shown in Table 1. It is observed that the sources of variation genotype and dose alone had a greater influence on the agronomic characteristics. It is also observed the significance of the interaction only for several clusters and Li content in the grain. Thus, a linear or quadratic response was calculated by providing the regression equations and the coefficient of determination (R2) when necessary.

Table 1. Summary of the analysis of variance characteristics plant height (PH), upper stem height (USH), stem diameter (SD), bunch length (BL), grain yield (GY), number of bunches (NB), lithium content in the grain (LiCG), Li content in the soil (LiTS), Li content in the leaves (LiCL), a mass of one thousand grains (MTG), liquid photosynthesis (A), stomach conductance (SC) and perspiration (E) of *Salvia hispanica* L., cultivated in the south of the state of Tocantins as a function of five doses (0, 10, 20, 30, 40 g ha^{-1}) of lithium hydroxide (LiOH) and two genotypes (Argentina and Paraguay).

	Source of variation					Media General	C.V. (%)
	Block	Genotype (G)	Dose (D)	Interaction GxD	Error		
	Degree of freedom						
	3	1	4	12	40		
PH	35.76	180.63*	139.34ns	81.81ns	31.76	135.28	4.17
USH	29.96	225.63*	100.63ns	106.63ns	46.11	116.63	5.82
SD	0.16	0.06ns	3.14**	1.34ns	0.54	8.93	8.21
BL	2.83	18.23**	8.84**	2.79ns	2.14	12.02	12.16
GY	369	2187.74*	6676.66**	137.01ns	258.10	72.95	22.02
NB	38.49	50.63*	36.19**	82.06*	9.97	31.88	9.91
LíCG	1.40	954.53**	4.74**	6.62**	1.03	14.05	7.22
LiTS	12.85	13.81ns	103.33ns	37.49ns	16.24	26.50	15.21
LiCL	16.39	9.40ns	31.09ns	20.46ns	17.93	32.27	13.12
MTG	0.011	0.024**	0.015ns	0.009ns	0.005	1.094	5.19
A	47.34	60.57*	9.02ns	8.81ns	8.11	8.29	24.36
SC	0.002	0.011*	0.016**	0.024ns	0.002	0.147	21.32
E	1.13	6.15*	11.76**	2.59ns	1.02	4.42	22.85

C.V.: Coefficient of Variation. **:significant at the 1% probability level ($p \leq 0.01$); *: significant at the 5% probability level ($p \leq 0.05$); ns: non-significant ($p > 0.05$) by F test and regression analysis.

The plants of the Paraguay genotype showed greater height (Fig. 1A), superior stem (Fig. 1B), bunch length (Fig. 1C), and grain yield (Fig. 1D) showing the superiority of approximately 3.0, 4.0, 7.7, and 22.9 percent, respectively, compared to the general average of the Argentine genotype.

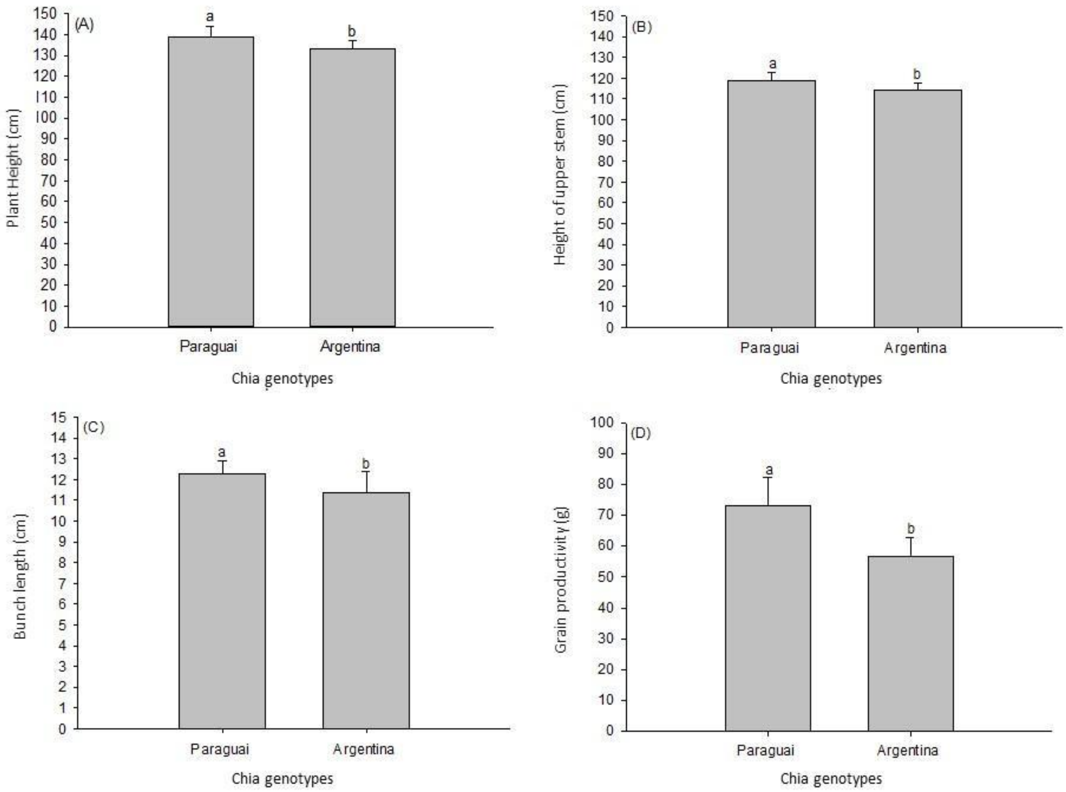

Table 2. Plant height (A), upper stem (B), bunch length (C), and grain yield (D) of two chia genotypes (Argentina and Paraguay) as a function of the application via LiOH foliar. Averages followed by the same lowercase letter between the genotypes do not differ by the Tukey test ($p \leq 0.05$).

For the stem diameter, the increased doses of LiOH showed to be able to promote positive responses for the chia genotypes up to the dose of 21.7 g ha^{-1} increasing by approximately 14.2% (Figure 2A). For the length of the cluster, they showed positive influence until the dose 9.1 g ha^{-1} increasing by approximately 6.8% (Figure 2B), however, in these characteristics (DM and CC) the greater dose of LiOH (40 g ha^{-1}) was reduced 11.9 and 12.2%, respectively, when compared with the control treatment.

On the other hand, for grain productivity according to the derivation of the quadratic regression equation, the application of LiOH influenced negatively showing a reduction in productivity as the dose of LiOH via foliar (Figure 2C) increases, being maximum in the dose of 40 g ha^{-1} with a reduction of 32.6% about the control treatment.

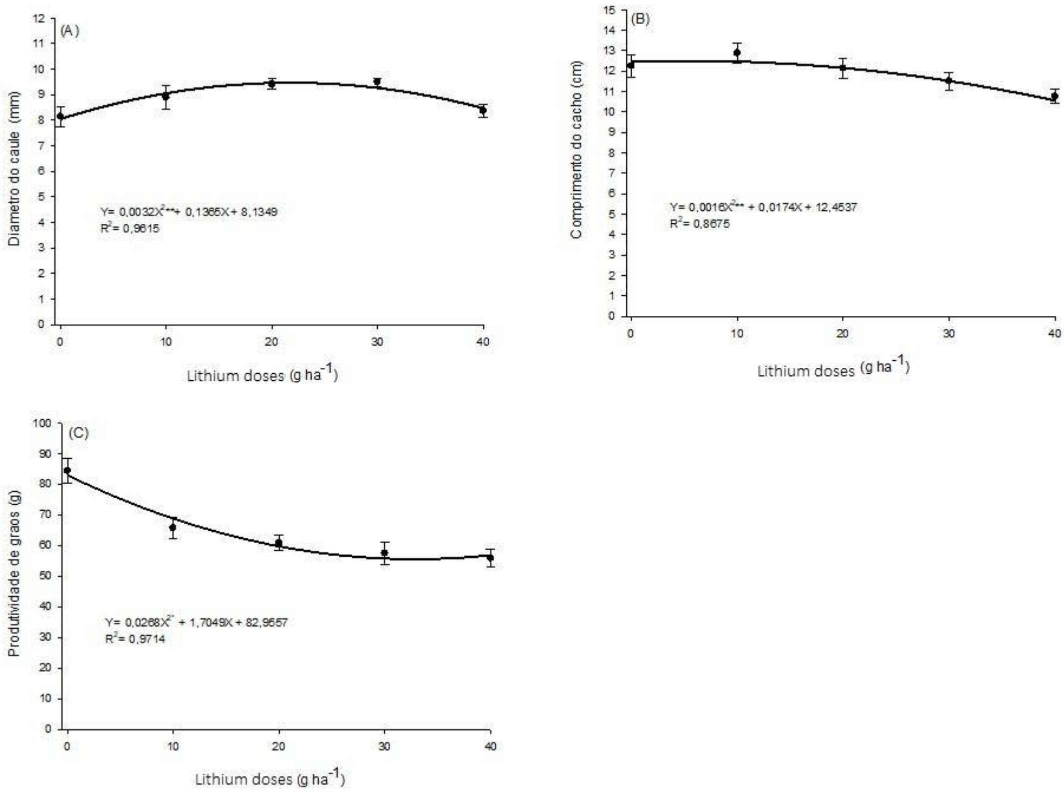

Figure 2. Stem diameter (A), bunch length (B), and grain yield (C) of chia genotypes as a function of five doses (0, 10, 20, 30, 40 g ha^{-1}) of lithium hydroxide (LiOH) foliar application.

Plants of the Paraguay genotype showed an increase in the number of bunches (29.2 percent) up to the dose of 20.3 g ha^{-1}, while the Argentine genotype provided an increase (6.5 percent) up to the dose of 9.3 g ha^{-1} (Figure 3A). Subsequently, there was a decrease in the averages, being maximum in the dose of 40 g ha^{-1} with a reduction of 24.6% for the Paraguay genotype and 21.1% for the Argentina genotype, when compared to the control treatment. The chia genotypes presented differentiated behavior for this characteristic, it was observed superiority of the Paraguayan genotype in the doses (10, 20, and 30 g ha^{-1}) and a greater number of bunches in the Argentina genotype without LiOH application.

The foliar application of LiOH doses results in increases in the lithium content of chia grains for both genotypes. The genotype Paraguay showed an increase in lithium concentration up to a dose of 24.6 g ha^{-1}, providing a maximum content of 21.9 mg kg^{-1} (Figure 3B). For the Argentine genotype, there was an increase in the concentration of the element up to the dose of 18.5 g ha^{-1}, providing a maximum content of 11.2 mg kg^{-1}. The results indicate the content of lithium in the grain superiority of the Paraguay genotype when compared to the Li content of the Argentina genotype in all applied doses of LiOH (0, 10, 20. 30, and 40 g ha^{-1}). The success of Li biofortification through foliar fertilization is thus evidenced, with an increase of 24.7 and 19.3% in the chia grain, for the genotype Paraguay and Argentina, respectively with the control treatment.

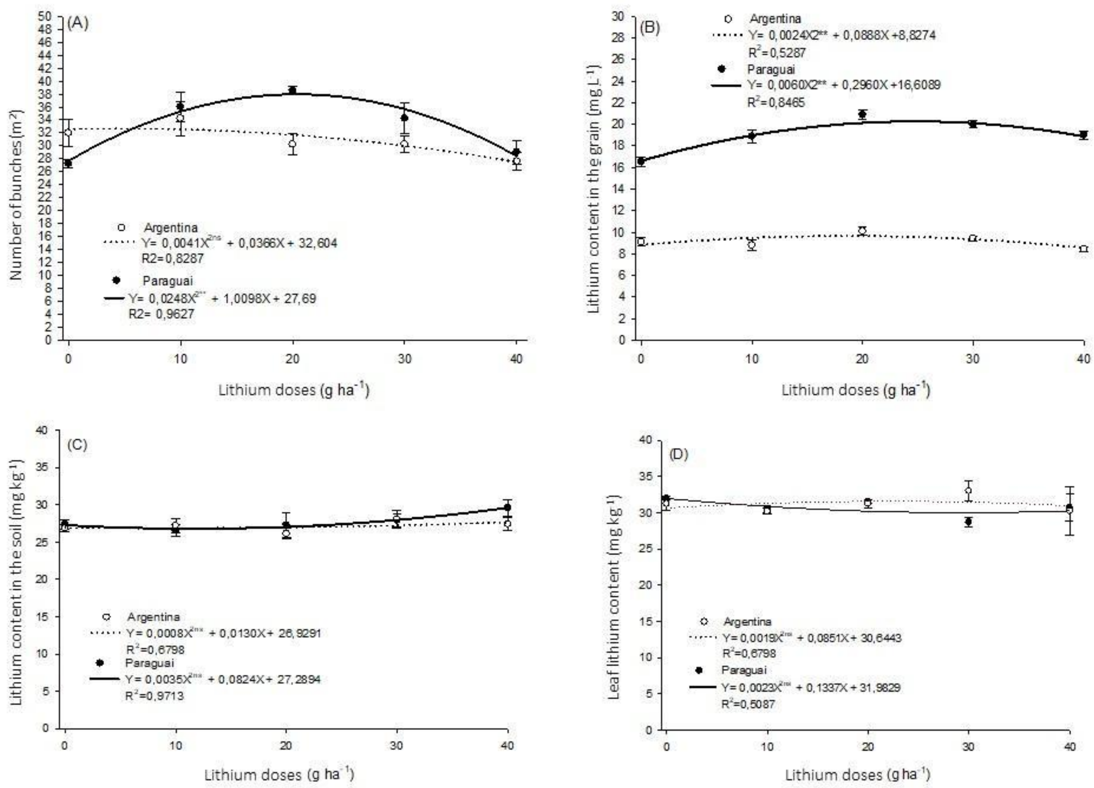

Figure 3. The number of clusters (A), lithium content in grain (B), lithium content in soil (C), and lithium content in leaf (D) of two chia genotypes (Paraguay and Argentina) as a function of foliar application of five doses (0, 10, 20, 30, 40 g ha^{-1}) of lithium hydroxide (LiOH).

For the characteristics of Li content in the leaf and soil, although there is no significant difference between the chia genotypes and applied LiOH doses (Figures 3C and D), Li concentrations ranging from 26.1 to 29.6 mg kg^{-1} were observed in the soil, showing slight superiority of the Paraguay genotype with additions up to the maximum dose (40 g ha^{-1}) when compared to the Argentina genotype (Figure 3C). The leaves of the chia genotypes varied from 28.7 to 33.2 mg kg^{-1}, showing a slight superiority of the genotype Argentina with additions up to the maximum dose (22.3 g ha^{-1}) and later decreases (Figure 3D). It should be noted that in this study, no necrotic or chlorotic stains were observed in the plants, regardless of their maturation stage, even at the maximum dose applied (40 g ha^{-1}).

The Paraguay genotype showed a greater mass of one thousand grains (Figure 4A), liquid photosynthesis (Figure 4B), stomach conductance (Figure 4C), and perspiration (Figure 4D) showing the superiority of approximately 7.4; 18.4; 23.5 and 29.3 percent when compared to the Argentina genotype (Figure 4), respectively.

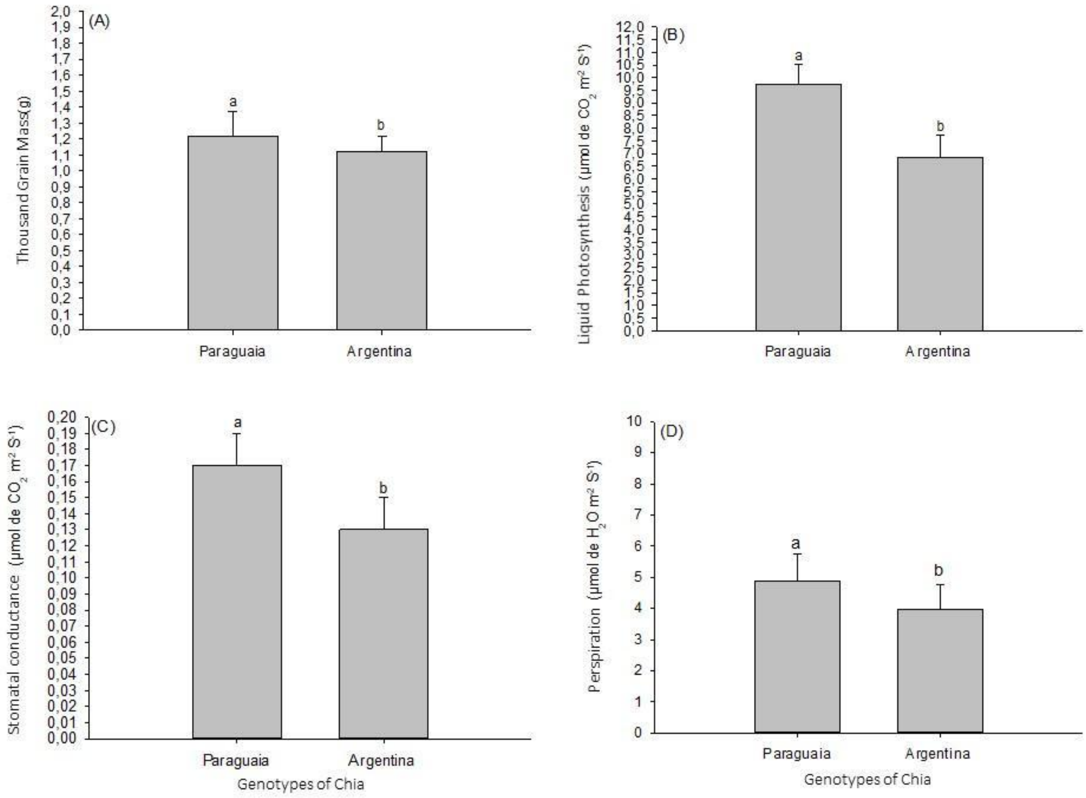

Table 4: Mass of one thousand grains (A) liquid photosynthesis (B), stomach conductance (C), and perspiration (D) of two chia genotypes (Argentina and Paraguay) as a function of the application via LiOH foliar. Averages followed by the same lowercase letter between the genotypes do not differ by the Tukey test ($p \leq 0.05$).

However, when plants of chia genotypes were exposed to different doses of LiOH, they showed an increase in stomach conductance (58.8%) up to a dose of 17.2 g ha^{-1} (Figure 5A). Similarly, the increase in doses resulted in increases in perspiration (62.3%) of the plants up to the dose of 23.6 g ha^{-1} (Figure 5B).

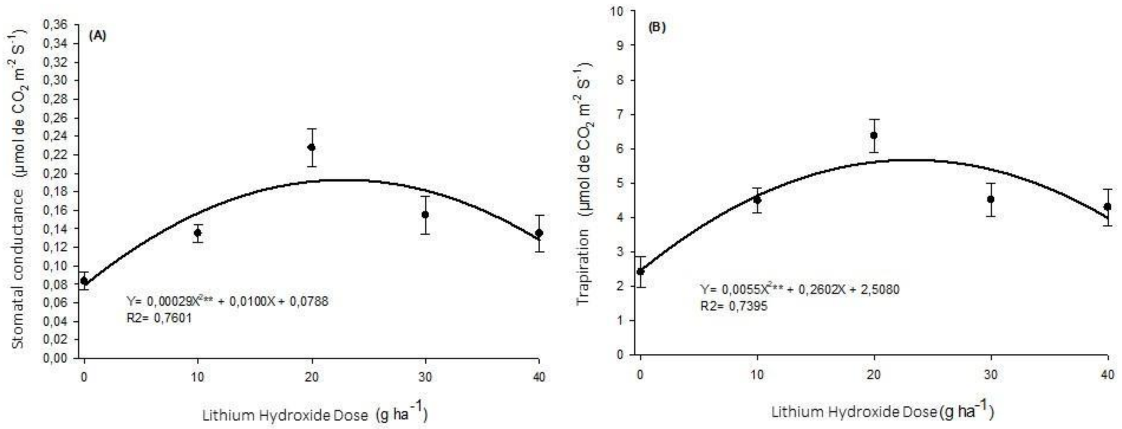

Figure 5: Stomach conductance (A) and perspiration (B) of two chia genotypes (Paraguay and Argentina) as a function of foliar application of five doses (0, 10, 20, 30, 40 g ha^{-1}) of lithium hydroxide (LiOH).

Discussion

Although Li appears to be a non-essential, non-physiological metallic ion in plant diets, there is evidence that different plant species can absorb a considerable concentration of Li (JIANG et al., 2014; KALINOWSKA et al., 2013). However, high concentrations of Li deserve due attention as they can cause toxicity and decrease in plant development, as observed for soybean (SANTOS et al., 2019), maize (ANTONKIEWICZ et al., 2017), *Brassica juncea* (MAKUS & ZIBILSKE, 2015) and sunflower seedlings (STOLARZ et al., 2015).

The results obtained in this study demonstrated that the application of increasing doses of LiOH by foliar fertilization on chia genotypes does not result in or reduction of plant height and upper stem height. A probable explanation of the lack of response may be because the doses of foliar fertilization in chia culture were applied in the phases of flowering and grain filling when these morphological characteristics were already fully developed and LiOH could not act on the plants.

However, low doses of LIOH applied via foliar fertilization in chia culture, resulted in the addition of the DC, CC, and NC averages (Figures 2A, 2B, and 3A), allowing a favorable development process. Recent studies evaluating the effects of Li on the agronomic characteristics of lettuce have reported that applications of low concentrations of lithium sulfate (7.5 mg dm^{-3}) and lithium hydroxide (22 mg dm^{-3}), provide significant increases in the development of the culture (SILVA et al., 2019). Bakhat et al. (2019) also described that the low concentration of Li (20 mg kg^{-1}) resulted in higher fresh biomass in spinach compared to the control treatment. According to Sobolev et al. (2019) in small amounts, Li has a positive effect on plant metabolism, increases the photochemical activity of chloroplasts, translocation of sugars, activates enzymatic processes, regulates biosynthesis and accumulation of alkaloids, increases the intensity of respiration, absorption, and accumulation of mineral elements, favoring plant growth and development. However, high concentrations have an inverse effect, studies show that this element in high concentrations has induced some metabolic changes that play a vital role in some biological functions of plants, i.e., they deregulate several responses dependent on Ca signaling, nucleic acids, DNA biosynthesis and metabolic processes of the plasmatic membrane H+ ATPase interrupting the stretching and cell differentiation (BAKHAT et al., 2019; SHAHZAD et al., 2016; TANVEER et al. 2019).

In this study, a negative response was found in the maximum applied dose of LiOH (40 g ha^{-1}), presenting a reduction in the averages of all evaluation characteristics. Negative responses of similar Li ions were documented by Santos et al. (2019) when they observed that doses of lithium hydroxide higher than 60 mg dm^{-3}, caused symptoms of phytotoxicity and decreased soybean yield.

Generally, high concentrations of Li increase ethylene production, inhibiting plant growth (MARTINES et al. 2018). This is possible because lithium can interact with inositol triphosphate and inhibit the activity of myo-inositol-1-phosphate and consequently can affect the induction of the gene related to the production of the enzyme 1-aminocyclopropane-1carboxylic synthase (ACC synthase), which regulates the ethylene production (HARWOOD 2005; LIANG et al. 1996; NARANJO et al., 2003). High concentrations of this hormone can cause a reduction in plant size, reduction in the growth of leaves, stems, and roots, epinasty, senescence, and leaf abscission, increase in the thickness of the kaolin base, reduction in photosynthesis, contributing to less development (DANGL et al., 2000; DUTRA et al., 2012).

For the productivity characteristic, the addition of Li via foliar in the form of LiOH exerted decreasing effects on the yield with the increase of doses (Figure 2C). These reductions can be attributed to the fact that high doses of Li are harmful in the bloom phase, promoting changes in pollen or interfering in important enzymatic activities (GUMBER et al., 1984). Besides, the accumulation of Li in the cytosol can interact with proteins and modulate various biochemical processes, affecting plant yield and development (BAKHAT et al., 2019; HARWOOD, 2005). Therefore, it can be speculated that high doses of lithium may interfere with the development and productivity of chia culture because this element has induced some metabolic changes of vital importance in plants. However, the effect of lithium in low and high concentrations on the morphogenesis of the chia plant has not yet been fully clarified.

In this study the foliar application of doses of LiOH promoted an increase of Li contents in the chia grain, which according to the point of maximum efficiency evaluated, occurred positively until the application of 18.5 and 24.6 g ha^{-1}, in the genotypes Argentina and Paraguay, respectively (Figure 3B), demonstrating that doses of LiOH applied via foliar fertilization resulted in the biofortification in the chia grain, and the best averages were observed in the genotype Paraguay.

The experiments performed by Robinson et al. (2018), reported that the effect of increased Li concentration on plants is positively correlated with Ca, Fe, K, and Zn, which indicates that Li can be transferred mainly in the xylem and accumulate in the roots, leaves, seeds or fruit. On the other hand, Seregin & Kozhevnikova (2020) reported that low molecular weight binders (nicotianamine, histidine, phytochelatins, phytosiderophores, and organic acids) play an important role also in the transport and concentration of large amounts of metals in plants.

However, according to Shahzad et al. (2016), the accumulation of Li by different plant species can also be partially explained by different experimental conditions, age of plants, sampling date, and exposure. Li concentration is expected to be higher in dicotyledons (e.g., *Arabidopsis thaliana, Helianthus annuus*, and *Brassica napus*) than in monocotyledons (e.g., *Zea mays)* (HAWRYLAK-NOWAK et al., 2012; MARTINEZ et al., 2018). Chia is a plant belonging to the dicotyledonous class (DI SAPIO et al., 2012) so it can be expected that plants can easily absorb this element. It is important to mention that the doses of lithium have been plotted in two seasons of an application being the first before flowering and the second applied in the filling of the grain.

The specific ability to accumulate different amounts of Li in different parts of the plant also depends on the species, dose, source applied, adaptation, and tolerance to this element, with some species acting as hyper-accumulators while others are sensitive to the toxicity of Li (QIAO et al., 2018; FRANZARING et al., 2016). Concerning the culture of chia, there is no information published so far about concentration, adaptation, toxicity, or distribution of Li in plant tissues, so more research is needed.

Li is found in small quantities in all soils, mainly in the clay fraction and to a lesser extent in the organic fraction of the soil. Normally this element enters the environment employing weathering processes, once in the environment, it is easily transported by the xylem to the vegetal biomass, by potassium carriers of high affinity, therefore, the plants will easily absorb Li in the leaves (ARAL & VECCHIO-SADUS, 2008; EVANGELOU et al., 2016; MARTINEZ et al., 2018). Tanveer et al. (2019) also reported that lithium can enter the xylem via HKT1 or HKT2 (monovalent cation carriers that play a very important role in stress tolerance), replacing K+ and then can be transported to the leaves. In this study, the accumulation of Li in the leaves varied from 28.7 to 33.2 mg kg^{-1} of biomass, while in the soil, from 5 26.1 to 29.6 mg kg^{-1}, this according to the averages observed, but without showing significant effect (Figures 3C and 3D). The concentration of Li detected in the soil may be a probable explanation for the presence of Li in control plants. Furthermore, another possible explanation is the fact that Li is a natural element of drinking water, being found mainly in ionic form in water (ARAL & VECCHIO-SADUS, 2008). According to the literature, surface water contains Li in levels between 1 and 10 μg/L (SCHRAUZER, 2002)

Physiological adaptations allow some species to tolerate and resist adverse ionic conditions that are toxic to most other plants (JIANG et al., 2019). In this study, the physiological characteristics of stomach conductance and perspiration were also positively influenced by low doses of lithium hydroxide applied via foliar fertilization (Figures 5A and 5B). According to Wehr et al. (2017), the increase in perspiration (GS) also promotes the increase in stomach conductance (E) of plants, a fact observed in this work. In low concentrations, one of the important specific characteristics of the physiological role of Li is its ability to regulate the biosynthesis and accumulation of alkaloids (KASHIN, 2019). However, the maximum dose applied (40 g ha^{-1}) was influenced negatively, independent of the chia genotype. Jiang et al. (2014), reported a significant reduction in stomach conductance in wild plants (*Apocynum venetum*) submitted to the application of 400 mg Li dm^{-3} of soil. Lithium is potentially toxic in increased concentrations.

It is known that higher plants complete their life cycle without Li, but the intensification of Li stress in plants can significantly influence the physiology or mediate some biochemical processes, for example, it replaces Mg in chlorophyll, reduces stomach conductance, being possible an imbalance between the biochemical and photochemical phases of photosynthesis, breakage of plastoquinone, carotenoids and inhibits the Calvin cycle decreasing the generation of ATP (RZYMSKI et al, 2017; SHAHZAD et al., 2016). It is known that the generation of ATP in plant tissues, is the main source of energy for various metabolic processes in plant tissues. According to Tanveer et al. (2019), the decrease of ATP production from plants under Li stress may cease essential cellular functions that can affect cellular physiology. Information on the physiological functions and distribution patterns of Li in plants is limited or relatively little recognized (KALINOWSKA et al., 2013). However, further studies are still needed.

In this study, the differences in morphological and physiological characteristics (Figures 1 and 4), observed between the genotypes of chia may be due to different responses of adaptation to the Cerrado environment. According to Furquim et al. (2018), cultures in the Cerrado Biome have developed several morphological and phenological adaptations for survival in adverse conditions. The superiority of the Paraguay genotype may be due to an improvement in adaptability in the Cerrado Biome compared to the Argentine genotype that is recently adapting to this region.

According to Fidelis et al. (2019), the Paraguay genotype is rustic and is adapting to the edaphoclimatic conditions of the Cerrado. Chia genotypes cultivated under different environmental and cultural conditions showed variation in productivity, growth, and composition of the chia seed (AYERZA & COATES, 2009, AYERZA, 2019). The transport and dispersion of chia genotypes to new environments in the cerrado may be a possible explanation of the morphological differences between the genotypes documented in this research. In this study, the difference between chia genotypes for plant height, the upper stem was only 4.89 and 5.99 cm respectively (Figures 1A and 1B).

As for productivity characteristics, the Paraguay genotype showed longer bunch length (12.30 cm), some bunches (33 bunches/plant), and mass of one thousand grains (1.21 g) resulting in higher yield (Figures 1C, 1D, 3A, and 4A). However, it is important to note that the averages of plant height, upper stem height, bunch length, number of bunches, and mass of one thousand grains obtained in this study are close to the averages obtained in other recent studies evaluating doses of fertilization (NPK) in chia culture (FIDELIS et al., 2019; GRIMES et al. 2019; SOUZA & CHAVES et al., 2017). As for the physiological characteristics, the genotype Paraguay also showed greater liquid photosynthesis (9.71 µmol of CO_2 m^{-2} S^{-1}) stomach conductance (0.17 µmol of H_2O m^{-2} S^{-1}) and perspiration (4.87 µmol of CO_2 m^{-2} S^{-1}) (Figures 4A, 4C, and 4D).

Depending on the genotype, doses, source, and phenological phase of application it is possible to achieve biofortification with Li thus increasing the levels of this element in vegetables and cereals. Młyniec et al. (2014) and Vita et al. (2015) report that Li can be beneficial in stabilizing anxiety, mood and reducing suicides in the population caused by low consumption of this element.

The recommended minimum daily intake of Li is 1 mg for an adult of 70 kg per day (SCHRAUZER, 2002; MARSHALL, 2015). Considering that the minimum recommended daily intake of chia is 25 g (CHICCO et al., 2009; JIN et al., 2012), it is possible to estimate that the consumption of 25 g of chia cultivated with an application of 24.6 g ha^{-1} (21.9 mg kg^{-1}) in the Paraguay genotype or 18.5 g ha^{-1} (11.2 mg kg^{-1}) in the Argentina genotype using LiOH as the source, would represent 54.95 and 28.2% of the recommended level, respectively. However, these calculations do not include the loss of the absorbed lithium content in the grain that may occur during processing; thus, they also do not include the availability of this metal in the body.

Biofortification is seen as a sustainable strategy to provide nutritious food to the population, but further studies are still needed on the agronomic biofortification with lithium for the adequacy of concentrations to be applied (BOLDRIM et al., 2012; SZKLARSKAE & RZYMSKI, 2019).

Conclusion

The highest concentrations of lithium in the chia grains are obtained with the application of 24.6 g ha^{-1} and 18.5 g ha^{-1} of LiOH for the genotypes Paraguay and Argentina, respectively.

The best responses in height, bunch length, number of bunches, lithium content in the grain, the mass of one thousand grains, photosynthesis, perspiration, and stomach conductance were obtained with the Paraguay genotype.

Regardless of genotype, LiOH doses above 30 and 40 g ha^{-1} promote reduction on morphological and physiological characteristics in chia culture.

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References

ALLENDER, W. J.; CRESSWELL, G. C.; KALDOR, J.; KENNEDY, I. R. Effect of lithium and lanthanum on herbicide induced hormesis in hydroponically-grown cotton and corn. **Journal of Plant Nutrition**, v. 20, n. 1, p. 81-95, 1997. DOI: 10.1080/01904169709365235.

ANJUM, N. A.; ADAM, V.; KIZEK, R.; DUARTE, A. C.; PEREIRA, E.; IQBAL, M.; AHMAD, I. Nanoscale copper in the soil-plant system-toxicity and underlying potential mechanisms. **Environmental research**, v. 138, n. 1, p. 306-325, 2015. DOI: doi.org/10.1016/j.envres.2015.02.019.

ANJUM, S. A.; ASHRAF, U.; KHAN, I.; TANVEER, M.; ALI, M.; HUSSAIN, I.; WANG, L. C. Chromium and aluminum phytotoxicity in maize: morpho-physiological responses and metal uptake. **CLEAN-Soil, Air, Water,** v. 44, n. 8, p. 1075-1084, 2016. DOI: doi.org/10.1002/clen.201500532.

ANTONKIEWICZ, J.; JASIEWICZ, C.; KONCEWICZ-BARAN, M.; BĄCZEK-KWINTA, R. Determination of lithium bioretention by maize under hydroponic conditions. **Archives of environmental protection**, v. 43, n. 4, p. 94-104, 2017. DOI 10.1515/aep-2017-0036.

ARAL, H.; VECCHIO-SADUS, A. Toxicity of lithium to humans and the environment a literature review. **Ecotoxicology and Environmental Safety**, v. 70, n. 3, p. 349-356, 2008. DOI: https://doi.org/10.1016/j.ecoenv.2008.02.026.

AYERZA, R. Antioxidants, protein, oil content and fatty acids profiles of chia seeds (*Salvia hispanica* L.) genotype Tzotzol growing in three tropical ecosystems of Bolivia, Ecuador and Paraguay. **International Journal of Agriculture Environment and Food Sciences**, v. 3, n. 3, p. 191-196, 2019. DOI: 10.31015/jaefs.2019.3.11.

AYERZA R., R.; COATES, W. Influence of environment on growing period and yield, protein, oil and alfa-linolenic content of three chia (*Salvia hispanica* L.) selections. **Industrial Crops and Products**, v. 30, n. 2, p. 321-324, 2009. DOI: https://doi.org/10.1016/j.indcrop.2009.03.009.

BAKHAT, H. F.; RASUL, K.; FAROOQ, A. B. U.; ZIA, Z.; FAHAD, S.; ABBAS, S.; HAMMAD, H. M. Growth and physiological response of spinach to various lithium concentrations in soil. **Environmental Science and Pollution Research**, v. 124, n. 1 p. 1-9, 2019. DOI: 10.1007/s11356-019-06877-2.

BARTOLO, M. E.; CARTER, J. V. Lithium decreases cold-induced microtubule depolymerization in mesophyll cells of spinach. **Plant physiology**, v. 99, n. 4, p. 1716-1718, 1992. DOI: https://doi.org/10.1104/pp.99.4.1716.

BOLDRIN, P. F.; FAQUIN, V.; RAMOS, S. J.; GUILHERME, L. R. G.; BASTOS, C. E. A.; CARVALHO, G. S.; COSTA, E. T. de S. Selenate and selenite in the production and agronomic biofortification with selenium in rice. **Pesquisa Agropecuária Brasileira**, v. 47, n. 6, p. 831-837, 2012. DOI: 10.1590/S0100-204X2012000600014.

BUESO, E.; ALEJANDRO, S.; CARBONELL, P.; PEREZ-AMADOR, M.A.; FAYOS, J.;BELLES, J.M.; RODRIGUES, P.L.; SERRANO, R. The lithium tolerance of the Arabidopsis cat2 mutant reveals a cross-talk between oxidative stress and ethylene. **The**

Plant Journal, v. 52, n. 6, p. 1052-1065, 2007. DOI: https://doi.org/10.1111/j.1365-313X.2007.03305.x

CHICCO, A. G.; D'ALESSANDRO, M. E.; HEIN, G. J.; OLIVA, M. E.; LOMBARDO, Y. B. Dietary chia seed (*Salvia hispanica* L.) rich in alpha-linolenic acid improves adiposity and normalises hypertriacylglycerolaemia and insulin resistance in dyslipaemic rats. **British Journal of Nutrition**, v. 101, n. 1, p. 41-50, 2009. DOI: 10.1017/S000711450899053X.

CIPRIANI, A.; PRETTY, H.; HAWTON, K.; GEDDES, J. R. Lithium in the prevention of suicidal behavior and all-cause mortality in patients with mood disorders: a systematic review of randomized trials. **American Journal of Psychiatry**, v. 162, n. 10, p. 1805-1819, 2005. DOI: 10.1176/appi.ajp.162.10.1805.

DANGL, J.; COOK, D.; HASELKORN, R.; LAST, R.L.; MARTIENSSEN, R.; MCCOUCH, S.; RETZEL, E.F.; SOMERVILLE, C.R.; WESSLER, S.; YATES, J. The national plant genomics initiative: objectives for 2003-2008. **Plant Physiology**, v. 130, n. 1, p. 1741-1744, 2002. DOI: https://doi.org/10.1104/pp.900055.

DI SAPIO, O.; BUENO; M.; BUSILACHI; H.; QUIROGA; M.; SEVERIN; C. Marofoanatomic characterization of leaf, stem, fruit and seed of *Salvia hispanica L.* (Lamiaceae). **Latin American and Caribbean Bulletin of Medicinal and Aromatic Plants**, v. 11, n. 3, p. 249-268, 2012. Available at: http:*//www.redalyc.org/articulo.oa?id=85622739007.*

DUTRA, C. C.; do PRADO E. A. F.; PAIM, L. R.; SCALON, S. D. P. Q. Development of sunflower plants under different water supply conditions. Semina: **Agricultural Sciences**, v. 33, n. 1, p. 2657-2667, 2012. DOI: 10.5433/1679-0359.2012v33Supl1p2657.

EKMEKCIOGLU, C.; MARKTL W. **Essential trace elements: clinical and nutritional medicine**. 1 online resource, Berlin: Springer, 2006, p. 173-177. DOI: doi.org/10.1007/3-211-35107-8.

EL-AAL, F. S. A.; SHAHEEN, A. M.; AHMED, A. A.; MAHMOUD, A. R. Effect of foliar application of urea and amino acids mixtures as antioxidants on growth, yield and characteristics of squash. **Research Journal of Agriculture and Biological Sciences**, v. 6, n. 5, p. 583-588, 2010. Disponível em: https://www.cabdirect.org/cabdirect/abstract/20103372601.

EMBRAPA. National ground research center. **Brazilian soil classification system**. 3. ed. Brasília: Centro Nacional de Pesquisa de Solos, 2013, p. 353.

EMBRAPA. Brazilian Agricultural Research Company National Soil Research Center. Manual of soil analysis methods. 2. ed. Rio de Janeiro: Embrapa, 1997.

EVANGELOU, M. W. H.; A. BÜRGI, B. H.; ROBINSON, M. S.; GUNTHARDT-GOERG, M.; SCHÖNGENS R. S. New method to determine the concentrations of elements in the foliage of poplar and willow cuttings . **International Journal of Phytoremediation**, v. 18, n. 9, p. 943-948, 2016. DOI: 10.1080/ 15226514.2015.1131234.

FALCO, B.; INCERTI, G.; BOCHICCHIO, R.; PHILLIPS, T. D.; Amato, M.; Lanzotti, V. Metabolomic analysis of *Salvia hispanica* seeds using NMR spectroscopy and multivariate data analysis. **Industrial crops and products**, v. 99, n. 1, p. 86-96, 2017. DOI: https://doi.org/10.1016/j.indcrop.2017.01.019.

FERREIRA, D. F. SISVAR: a program for analysis and teaching of statistics. **Revista Symposium**, Lavras, v. 6, n. 1, p. 36-41, 2008.

FIDELIS R. R.; CHAN G. A. H.; RAUBER, W. A.; TAVARES, T. C. O. LOPES, M. B. S.; MARQUES, K. R.; OSORIO, P. R. A.; BURIN, L. X.; VELOSO, D. A.; OLIVEIRA L. B.; FERNANDES, P. S M. DIAS, M. A. R. SANTOS, M. M.; AGUIAR R. W. S. Response curve of *Salvia hispanica* L. to different dosages of phosphorus in soils of the cerrado. **International Journal of Advanced Engineering Research and Science**, v. 6, n. 8, p. 2349-6495, 2019. DOI: 10.22161/ijaers.68.4.
FORBES, V. E. Is hormesis an evolutionary expectation. **Functional Ecology**, v. 14, n. 1, p. 14-24, 2000. DOI:10.1046/j.1365-2435.2000.00392.x.

FRANZARING, J.; SCHLOSSER, S.; DAMSOHN, W.; FANGMEIER, A. Regional differences in plant levels and investigations on the phytotoxicity of lithium. **Environmental pollution**, v. 216, n. 2, p. 858-865, 2016. DOI:10.1016/j.envpol.2016.06.059.

FURQUIM, L. C.; DOS SANTOS, M. P.; DE ANDRADE, C. A. O.; DE OLIVEIRA, L. A.; EVANGELISTA, A. W. P. Relationship between native plants of the cerrado and water. **Scientific@-Multidisciplinary Journal**, v. 5, n. 2, p. 146-156, 2018. DOI: https://doi.org/10.29247/2358-260X.2018v5i2.p146-156.

GALLICCHIO, V. S. Lithium-still interesting after all these years. **Trace Elements & Electrolytes, v.** 28, n. 1, p. 56-69, 2011. DOI: 10.4103/0019-5545.49460.

GONÇALVES, A. S. F.; GONÇALVES, W. M.; SILVA, K. M. J.; OLIVEIRA, R. M. The biofortification vegetable use: The review. **Cerrado Agrociências,** v. 6, n. 1, p. 75-87, 2015. Available at: https://chesci.com/?s=biofortification.

GRIMES, S. J.; PHILLIPS T. D.; CAPEZZONE F.; GRAEFF-HÖNNINGER S. Impact of Row Spacing, Sowing Density and Nitrogen Fertilization on Yield and Quality Traits of chia (*Salvia hispanica* L.) Cultivated in southwestern Germany. **Agronomy,** v. 9, n. 3 p. 1-21, 2019. DOI: 10.3390/agronomy9030136.

GUMBER, S. C.; LOEWUS, M. W.; LOEWUS, F. A. Further studies on myo-inositol-1-phosphatase from the pollen of Lilium longiflorum Thunb. **Plant physiology**, v. 76, n. 1, p. 40-44, 1984. DOI: https://doi.org/10.1104/pp.76.1.40.

HARWOOD, A. J. Lithium and bipolar mood disorder: the inositol-depletion hypothesis revisited. **Molecular psychiatry**, v. 10, n. 1, p. 117-126, 2005. DOI: 10.1038/sj.mp.4001618.

HAWRYLAK-NOWAK, B.; KALINOWSKA M.; SZYMAŃSKA M. A study on selected physiological parameters of plants grown under lithium supplementation. **Biological Trace Element Research**, v. 149, n. 3, p. 425-430. 2012. DOI: 10.1007/s12011-012-9435-4.

JIANG, L.; WANG, L.; MU, S. Y.; TIAN, C. Y. Apocynum venetum: A newly found lithium accumulator. **Flora-Morphology, Distribution, Functional Ecology of Plants**, v. 209, n. 5-6, p. 285-289, 2014. DOI: http://dx.doi.org/10.1016/j.flora.2014.03.007.

JIANG, L.; WANG, L.; TANVEER M.; TIAN C. Lithium biofortification of medicinal tea Apocynum venetum. **Scientific Reports**, v. 9, n. 8182, p. 1-8, 2019. DOI: 10.1038/s41598-019-44623-3.

JIN, F.; NIEMAN, D. C.; SHA, W.; XIE, G.; QIU, Y.; JIA, W. Suplementation of Milled Chia Seeds Increases Plasma ALA and EPA in Postmenopausal Women. **Plasts Food for Human Nutrition**, v. 67, n. 2, p. 105-110, 2012. DOI: 10.1007 / s11130-012-0286-0.

JURKOWSKA, H.; ROGÓŻ, A.; WOJCIECHOWICZ, T. Comparison of lithium toxic influence on some cultivars of oats, maize and spinach. **Acta Agraria et Silvestria**. **Series Agraria**, v. 36, n. 1, p. 37-42, 1998. Disponível em: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3624008.

KALINOWSKA, M.; HAWRYLAK-NOWAK, B.; SZYMAŃSKA, B. The infl uence of two lithium forms on the growth, L-Ascorbic acid content and lithium accumulation in lettuce plants. **Biological trace element research**, v. 152, n. 2, p. 251-257, 2013. DOI: 10.1007/s12011-013-9606-y.

KASHIN, V. K. Lithium in Soils and Plants of Western Transbaikalia. **Eurasian Soil Science**, v. 52, n. 4, p. 359-369, 2019. DOI: 10.1134/S1064229319040094.

KAVANAGH, L.; KEOHANE, J.; CABELLOS, G. G. "Induced Plant Accumulation of Lithium". **Geosciences**, v. 8, n. 2, p. 56-73, 2018. DOI: 10.3390/geosciences8020056.

LANZA, M. G. D. B.; SILVA, V. M.; MONTANHA, G. S.; LAVRES, J.; de CARVALHO, H. W. P.; DOS REIS, A. R. Assessment of selenium spatial distribution using µ-XFR in cowpea (*Vigna unguiculata* (L.) Walp.) plants: Integration of physiological and biochemical responses**. Ecotoxicology and Environmental Safety**, v. 207, p. 111216, 2020. DOI: https://doi.org/10.1016/j.ecoenv.2020.111216.

LIANG, X.; SHEN, N. F.; Theologis A. Li+-regulated 1-aminocyclopropane-1-carboxylate synthase gene expression in *Arabidopsis thaliana*. **The Plant Journal**, v. 10, n. 6, p. 1027-1036, 1996. DOI: https://doi.org/10.1046/j.1365-313X.1996.10061027.x.

LIAUGAUDAITE, V.; NAGINIENE, R.; RASKAUSKIENE, N.; MICKUVIENE, N.; BUNEVICIUS, A.; SHER, L. Relationship between lithium levels in drinking water and suicide rates: a nationwide study in Lithuania. **Archives of suicide research**, v.1, s/n, p. 1-13, 2019. DOI: https://doi.org/10.1080/13811118.2019.1674226.

LI, X.; GAO, P.; GJETVAJ, B.; WESTCOTT, N.; GRUBER, M. Y. Analysis of the metabolome and transcriptome of Brassica carinata seedlings after lithium chloride exposure. **Plant Science**. v. 177, n. 1, p. 68-80, 2009. DOI: 10.1016/j.plantsci.2009.03.013.

MAKUS, D. J.; ZIBILSKE, L.; LESTER, G. Effect of light intensity, soil type, and lithium addition on spinach and mustard greens leaf constituents. **Subtropical Plant Science**, v. 58, n. 1, p, 35-41, 2015. DOI: https://pubag.nal.usda.gov/catalog/57340.

MALAVOLTA, E; VITTI, G. C.; OLIVE TREE, S. A. **Evaluation of the nutritional state of the plants**. Principles and applications. 2nd ed. Piracicaba : POTAFOS, 1997, 319p.

MARSHALL, T. M. Lithium as a nutrient. Journal of American. **Physicians and** Surgeons, v. 20, n. 4, p. 104-109, 2015. Disponivel em: https://www.jpands.org/jpands2004.htm.

MARTINEZ, N. E.; SHARP, J. L.; JOHNSON, T. E.; KUHNE, W. W.; STAFFORD, C. T.; DUFF, M. C. Reflectance-Based Vegetation Index Assessment of Four Plant Species Exposed to Lithium Chloride. **Sensors**, v. 18, n. 9, p. 2750, 2018. DOI: https://doi.org/10.3390/s18092750.

MARTINSSON, L. Y.; WEI, D. Xu.; MELAS, P. A.; MATHÉ, A. A.; SCHALLING, M.; LAVEBRATT, C.; BACKLUND, L. Long-term lithium treatment in bipolar disorder is associated with longer leukocyte telomeres. **Translational Psychiatry**, v. 3, n. 5, p. 261-267, 2013. DOI: 10.1038/tp.2013.37.

MATSUNAGA, S.; KISHI, T.; ANNAS, P.; BASUN, H.; HAMPEL, H.; IWATA, N. Lithium as a treatment for Alzheimer's disease: a systematic review and meta-analysis. **Journal of Alzheimer's Disease**, v. 48, n. 2, p. 403-410, 2015. DOI: 10.3233/JAD-150437.

MIRANDA, F. **Technical guide for the management of Chia (*Salvia hispanica* L.) cultivation in Nicaragua** Sébaco: Central de Cooperativas de Servicios Multiples Exportacion e Importacion Del Norte (Cecoopsemein RL.), 2012. 14p. Available at: http://cecoopsemein.com/Manual_de_poduccion_de_CHIA_SALVIA_HISPANICA.pdf. Access: 18 Aug. 2019.

MLECZEK, M.; SIWULSKI, M.; RZYMSKI, P.; BUDZYŃSKA, S.; GĄSECKA, M.; KALAČ, P.; NIEDZIELSKI, P. Cultivation of mushrooms for production of food biofortified with lithium. **European Food Research and Technology** v. 243, n. 6, p. 1097-1104, 2017. DOI: 10.1007/s00217-016-2823-9.

MŁYNIEC K; DAVIES C. L.; SÁNCHEZ I. G. A.; PYTKA K, BUDZISZEWSKA B, NOWAK G. Essential elements in depression and anxiety. **Pharmacol Rep**. v. 66, n. 2, p. 534-544, 2014. DOI: 10.1016 / j.pharep.2014.03.001.

NARANJO, M.A.; ROMERO, C.; BELLES, J.M.; MONTESINOS, C.; VICENTE, O..; SERRANO, R. Lithium treatment induces a hypersensitive-like response in tobacco. **Plant**, v. 217, n. 3, p. 417-424, 2003. DOI: 10.1007/s00425-003-1017-4.

NORRA, C.; FEILHAUER, J.; WIESMÜLLER, G. A.; KUNERT, H. J. Differential effects of endogenous lithium on neurobehavioural functioning: a study on auditory evoked potentials. **Psychiatry research**, v. 178, n. 1, p. 176-181, 2010. DOI: https://doi.org/10.1016/j.psychres.2009.04.021.

QIAO, L.; TANVEER, M.; WANG, L.; TIAN, C. Subcellular distribution and chemical forms of lithium in Li-accumulator Apocynum venetum. **Plant physiology and biochemistry,** v. 132, n. 1, p. 341-344, 2018. DOI: 10.1016/j.plaphy.2018.09.022.

ROBINSON, B. H.; YALAMANCHALI, R.; REISER, R.; DICKINSON, N. M. Lithium as an emerging environmental contaminant: Mobility in the soil-plant system. **Chemosphere**, v. 197, n. 1, p. 1-6, 2018. DOI: https://doi.org/10.1016/j.chemosphere.2018.01.012.

ROLDÃO, F. A.; FERREIRA, O. V. Climatologia do Estado do Tocantins-Brasil. Caderno de Geografia, v. 29, n. 59, p. 1161-1181, 2019. DOI: https://doi.org/10.5752/P.2318-2962.2019v29n59p1161.

RZYMSKI, P.; NIEDZIELSKI, P.; SIWULSKI, M.; MLECZEK, M.; BUDZYŃSKA, S.; GĄSECKA, M.; PONIEDZIAŁEK, B. Piotr et al. Lithium biofortification of medicinal mushrooms Agrocybe cylindracea and Hericium erinaceus. Journal of food science and technology, v. 54, n. 8, p. 2387-2393, 2017. DOI: 10.1007/s13197-017-2679-4.

SANTOS, A. C. M.; MARQUES, K. R.; RODRIGUES, L. U.; FARIA, A. J. G.; NASCIMENTO, V. L.; FIDÉLIS R. R. Biofortification of soybean grains with foliar application of Li sources. **Journal of Plant Nutrition,** v. 42, n. 19, p. 1532-4087, 2019. DOI: 10.1080/01904167.2019.1659339.

SEREGIN, I. V.; KOZHEVNIKOVA, A. D. Low-molecular-weight ligands in plants: role in metal homeostasis and hyperaccumulation. **Photosynthesis Research**, v.1, p. 1-46, 2020. DOI: 10.1007/s11120-020-00768-1

SCHÄFER, U. Evaluation of beneficial and adverse effects on plants and animals following lithium deficiency and supplementation, and on humans following lithium treatment of mood disorders. **Trace Elements & Electrolytes, v.** 29, n. 2, p. 91-112, 2012. DOI: 10.5414/TEX01222.

SCHRAUZER, G. N. Lithium: Occurrence, dietary intakes, nutritional essentiality. **Journal of the American College of Nutrition,** v. 21, n. 1, p. 14-21, 2002. DOI: 10.1080/07315724.2002.10719188.

SHAHZAD, B.; TANVEER, M.; HASSAN, W.; SHAH, A. N.; ANJUM, S. A.; SARDAR ALAM CHEEMA, S. A.; ALI, I. Lithium toxicity in plants: Reasons, mechanisms and remediation possibilities e a review. **Plant physiology and biochemistry**, v. 107, n. 1, p. 104-115, 2016. DOI: 10.1016/j.plaphy.2016.05.034.

SILVA R. R.; FARIA A. J. G.; ALEXANDRINO, G. C.; RIBEIRO E. A.; SANTOS, A. C. M.; DEUSDARA, T. T. Enrichment of lithium in lettuce plants through agronomic biofortification. **Journal of Plant Nutrition**, v. 32, n. 17, p. 2102-2113, 2019. DOI: 10.1080/01904167.2019.1648671.

SOBOLEV, O. I.; GUTYJ, B. V.; DARMOHRAY, L. M.; SOBOLIEVA, S. V.; IVANINA, V. V.; KUZMENKO, O. A.; KARKACH P. M.; FESENKO V. F.; BILKEVYCH V. V.; MASHKIN Y. O.; TROFYMCHUK A. M.; STAVETSKA R. V.; TKACHENKO S. V.; BABENKO O. I.; KLOPENKO N. I.; CHERNYUK S. V. Lithium in the natural environment and its migration in the trophic chain. **Ukrainian Journal of Ecology**, v. 9, n. 2, p. 195-203, 2019. Disponivel em: http://rep.btsau.edu.ua/handle/BNAU/2489.

SOUZA, R. S.; CHAVES, L. H. G. Initial growth of chia (*Salvia hispanica* L.) submitted to nitrogen, phosphorus and potassium fertilization. **Australian Journal of Crop Science**, v. 11, n. 5, p. 610-615, 2017. Disponivel em: https://search.informit.com.au/documentSummary;dn=957807698316448;res=IELHS.

STOLARZ, M.; KRÓL, E.; DZIUBIŃSKA, H. Lithium distinguishes between growth and circumnutation and augments glutamate-induced excitation of Helianthus annuus seedlings. **Acta physiologiae plantarum**, v. 37, n. 4, p. 69, 2015. DOI: 10.1007/s11738-015-1814-y.

SZKLARSKA, D.; RZYMSKI, P. Is Lithium a Micronutrient? From biological activity and epidemiological observation to food fortification. **Biological trace element research**, v. 189, n. 1, p. 18-27, 2019. DOI: https://doi.org/10.1007/s12011-018-1455-2.

TANVEER, M.; HASANUZZAMAN, M.; WANG, L. Lithium in Environment and Potential Targets to Reduce Lithium Toxicity in Plants. **Journal of Plant Growth Regulation,** v. 38, n. 4, p. 1574-1586, 2019. Disponível em: https://pubag.nal.usda.gov/catalog/6763117.

VETTER, J. Lithium content of some common edible wild-growing mushrooms. **Food Chemistry**, v. 90, n. 1-2, p. 31-37, 2005. DOI: https://doi.org/10.1016/j.foodchem.2004.03.019.

VITA A.; PERI L.; SACCHETTI E. Lithium in drinking water and suicide prevention: a review of evidence. **Int Clin Psychopharmacol**, v. 30, n. 1, p. 1-5, 2015. DOI: 10.1097 / YIC.000000000048.

WEHR, R.; COMMANE, R.; MUNGER, W. J.; MCMANUS, B. J.; NELSON, D. D.; ZAHNISER, M. S. Dynamics of canopy stomatal conductance, transpiration, and evaporation in a temperate deciduous forest, validated by carbonyl sulfide uptake. **Biogeosciences,** v. 14, n. 1, p. 389-401, 2017. DOI: 10.5194/bg-14-389-2017.

WHITE, P. J.; BROADLEY, M. R. Biofortification of crops with seven mineral elements of ten lacking in human diets iron, zinc, copper, calcium, magnesium, selenium and iodine. **New Phytologist**, v. 182, n. 1, p, 49-84, 2009. DOI: 10.1111/j.1469-8137.2008.02738.x.

ZODAPE, S. T.; GUPTA, S. A.; BHANDARI, U. S.; RAWAT, D. R.; CHAUDHRY, K. ESWARAN J.; CHIKARA. Foliar application of seaweed sap as biostimulant for enhancement of yield and quality of tomato (*Lycopersicon esculentum* Mill.), **Journal of Scientific & Industrial Research, v.**70, n. 3, p.215-219, 2011.