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Lithium chloride in seed germination and initial growth of Guajillo chili seedlings

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ABSTRACT

Objective: To evaluate the effects of lithium (Li) supplied as lithium chloride (LiCl) on the emergence of seeds and initial growth of chili (*Capsicum annuum* L.) var. Guajillo seedlings.

Design/methodology/approach: Guajillo variety chili seeds were treated with five doses of Li chloride (0, 20, 50, 75, and 100 μM) during the germination phase. The treatments lasted 20 days. In this period, parameters related to seed germination and initial growth of seedlings were evaluated. With the data obtained, analysis of variance and comparison of means test (Duncan) were carried out with the SAS software.

Results: The doses of LiCl evaluated did not affect the percentages of germination and relative germination, the coefficient of velocity of germination, the average velocity of germination, and the weights of fresh and dry seedling biomass. Doses of 25 and 50 μM LiCl favored the germination index and the seed vigor index. Likewise, they significantly increased the height of the stem. On the contrary, the 100 μM Li dose significantly reduced the relative radicle growth, the germination index and the stem height.

Limitations on study/implications: This study used only a single Li source, so the effects that accompanying anions have, are unknown.

Findings/conclusions: Low doses of LiCl have positive effects on shoot growth in the initial phase of seedling growth, without affecting germination parameters.

Keywords: Non-essential elements, toxicity, stimulation, growth promotion.

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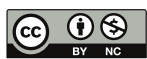
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INTRODUCTION

Lithium (Li) is an alkaline element of group IA in the periodic table. It is the most electropositive (-3.04 V), the lightest (6.94 g mol⁻¹), and the least dense ($\rho=0.53$ g cm⁻³) metal that exists. These specific properties place it in a strategic position in the world economy (Tarascon, 2010).

Lithium is widely distributed on the planet; its abundance in the lithosphere is 20 mg kg⁻¹, in soils from 7 to 200 mg kg⁻¹, in surface water from 1-10 g L⁻¹, in seawater of 0.18 g L⁻¹, and in mineral waters the levels can reach 100 g Li L⁻¹ (Pais & Jones, 1997; Tanveer *et al.*, 2019).

In higher plants, Li is not an essential element, while its content depends on the levels



Image by Michael Myers at Pixabay

present in the soil, generally trace amounts that vary from 0.002 to 63 mg kg⁻¹ (Aral *et al.*, 2011; Shahzad *et al.*, 2016; Baran, 2019). Due to its chemical similarity with sodium (Na⁺) and potassium (K⁺), the Li⁺ ion is considered an antagonist for both essential nutrients (Pais & Jones, 1997). Its positive effects on higher plants can occur in very small concentration ranges in which it promotes growth. On the contrary, high concentrations negatively interfere with plant metabolism (Aral *et al.*, 2011; Baran, 2019).

Non-essential metals such as Li are becoming an environmental and public health problem in various regions of the world. Its imminent arrival to agricultural soils through irrigation with contaminated water can cause serious problems in crop production and yield (Tanveer *et al.*, 2019).

Knowledge of the impact of Li is essential in crop production systems. To date, there is still little information about its effects on processes as specific as germination and plant growth. Therefore, the objective of this research was to study the effects of Li supplied as lithium chloride (LiCl), on the emergence of seeds and initial growth of chili (*Capsicum annuum* L.) var. Guajillo seedlings.

MATERIALS AND METHODS

Seed disinfestation

Chili seeds of the Guajillo variety were disinfested in a 2% sodium hypochlorite solution for 15 min, then five rinses were performed with sterile distilled water.

Treatments and experimental design

Five concentrations of LiCl were evaluated: 0, 25, 50, 75, and 100 μ M (LiCl, CAS-No: 7447-41-8, Sigma Aldrich, St. Louis, MO, USA). Each treatment had three replicates. The experimental unit consisted of a Petri dish with 12 seeds, distributed in a completely randomized design within a germination chamber (Thermo Scientific, model 310M, Waltham, MA, USA) at 32 °C. The seeds were kept hydrated with the Li solutions with different concentrations for 20 days.

Evaluated variables

Every 48 h, the number of germinated seeds was recorded for the variables germination percentage (GP), relative germination percentage (RGP), likewise, from germination, the radicle length (RL) was measured. As soon as the shoot emerged, stem height (SH) was measured every 48 h. With the data obtained, the coefficient of velocity of germination (CVG), average velocity of germination (AVG), relative radicle growth (RRG), germination index (GI), and the seed vigor index (SVI) were estimated. After 20 days of treatment, the fresh and dry weights of the seedlings were evaluated. The aforementioned measurements were carried out in accordance with the methodologies described by Anjum *et al.* (2005) and Buendía-Valverde *et al.* (2018).

Data analysis

An analysis of variance and comparison of means were performed with the data using the Duncan test ($P \leq 0.05$), using the SAS software (SAS, 2011).

RESULTS AND DISCUSSION

Germination percentage, relative germination percentage, coefficient of velocity of germination, and average velocity of germination

The highest germination percentages occurred in seeds treated with 25 and 50 μM LiCl (84.4%); however, they were not different from those of the control treatment (77.8%). Likewise, although there are no statistical differences in the relative percentage of germination, it can be seen that doses 25 and 50 μM LiCl increased this variable by more than 8% (Table 1).

The coefficient of velocity of germination indicates the speed of germination. Its value increases when the number of germinated seeds increases and the time for germination is reduced (Talská *et al.*, 2020). In this study, it is observed that treatments with some dose of Li present slightly higher values than the control in this variable; however, with no statistical differences (Table 1).

Average velocity of germination was not influenced by the treatments; however, the lowest value was recorded in the control without Li (Table 1).

During the germination process, it has been reported that Li can have positive or negative effects, because the response depends on the concentration applied (Baran, 2019). In this research, germination parameters were generally negatively affected by the 100 μM LiCl dose. On the contrary, the 25 and 50 μM LiCl treatments stimulate parameters related to germination and radicle and shoot growth.

Relative radicle growth, germination index, and seed vigor index

The relative radicle growth was reduced by almost 36% with the 100 μM LiCl treatment compared to the control. Slight, non-significant increases compared to the control were recorded with doses 25 and 50 μM LiCl (Figure 1). Likewise, the germination index decreased by 41.6% with the 100 μM LiCl dose, compared to the control (Figure 2). Regarding the seed vigor index, it was recorded that the 25 and 50 μM LiCl doses exceeded the value obtained with the 100 μM LiCl concentration by 36.1% (Figure 3).

The negative effects of high doses of lithium chloride on germination parameters of Guajillo chili seeds coincide with what was observed in the germination of amaranth (*Amaranthus viridis* L.) seeds treated with 25 to 100 mg $\text{Li}_2\text{SO}_4 \text{ kg}^{-1}$. In such study, doses

Table 1. Germination percentage (GP), relative germination percentage (RGP), Coefficient of velocity of germination (CVG), and average velocity of germination (AVG) of Guajillo chili seeds treated with lithium chloride (LiCl).

LiCl (μM)	GP	RGP	CVG	AVG
0	77.78 \pm 7.70a	100.00 \pm 9.90a	9.39 \pm 0.90a	0.49 \pm 0.21a
25	84.44 \pm 10.72a	108.57 \pm 13.78a	10.25 \pm 0.20a	0.90 \pm 0.14a
50	84.44 \pm 6.94a	108.57 \pm 8.92a	11.41 \pm 0.40a	0.84 \pm 0.07a
75	73.33 \pm 8.82a	94.29 \pm 11.34a	10.79 \pm 0.69a	0.73 \pm 0.05a
100	68.89 \pm 7.70a	88.57 \pm 9.90a	10.40 \pm 0.41a	0.65 \pm 0.05a

Means \pm SD with the same letter in each column indicate that there are no statistical differences (Duncan, $P \leq 0.05$).

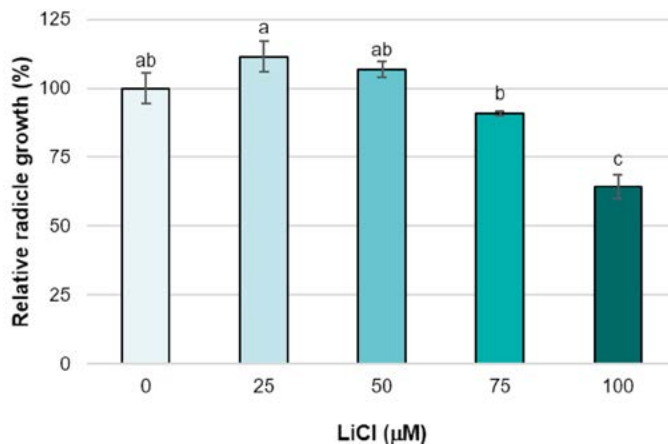


Figure 1. Relative radicle growth of chili seedlings from the treatment of Guajillo chili seeds with lithium chloride (LiCl) during the germination phase. Means \pm SD with different letters indicate statistical differences (Duncan, $P \leq 0.05$).

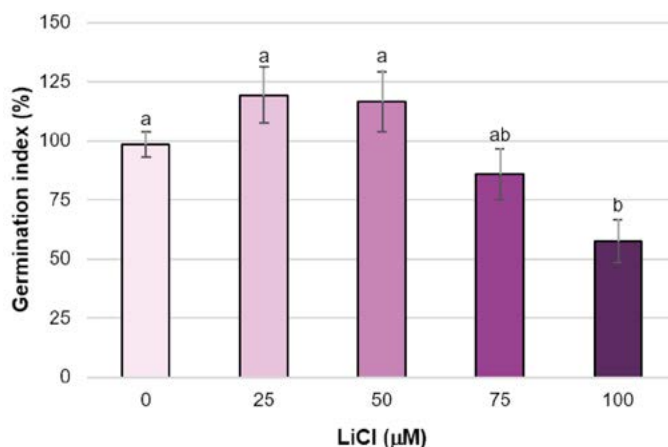


Figure 2. Germination index in chili seeds treated with lithium chloride (LiCl). Means \pm SD with different letters indicate statistical differences (Duncan, $P \leq 0.05$).

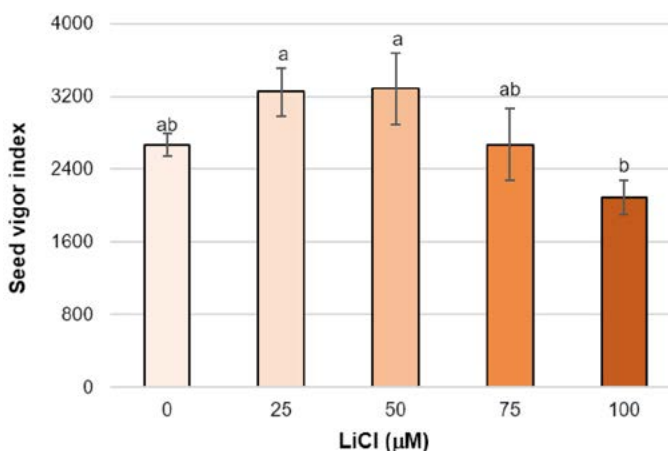


Figure 3. Seed vigor index of Guajillo chili treated with lithium chloride (LiCl). Means \pm SD with different letters indicate statistical differences (Duncan, $P \leq 0.05$).

of 50 and 75 mg $\text{Li}_2\text{SO}_4 \text{ kg}^{-1}$ reduced the germination coefficient rate by 73 and 57%; while, the 75 and 100 mg $\text{Li}_2\text{SO}_4 \text{ kg}^{-1}$ treatments decreased the germination percentage by 57 and 41%. The germination velocity was reduced with all doses of Li_2SO_4 , with the 50 mg $\text{Li}_2\text{SO}_4 \text{ kg}^{-1}$ concentration being the one that reduced it the most compared to the control (Gayathri *et al.*, 2022). Li *et al.* (2009) observed a significant reduction in the germination rate of Abyssinian mustard (*Brassica carinata*) seeds after exposure to LiCl in a concentration range of 60-180 μM LiCl.

Radicle length and shoot height

After 13, 15, and 17 days from the start of the test, it was observed that the length of the radicle increased with the 50 μM LiCl dose by 26.7, 31.1, and 26.6%, respectively, compared to the control. In the same way, the 25 μM LiCl dose increased radicle length by 32.3 and 31% after 13 and 15 days, compared to the control. On the contrary, only in the sampling carried out 9 days after the start of the test, did the 100 μM LiCl dose reduce the radicle length by 35.7% with respect to the control (Figure 4).

The stem growth was greater than the control with the 25 μM LiCl dose by 77.1, 67.1, 53.6, 40.8, and 26.4% after 9, 11, 13, 15, and 17 d from the start of the test. Likewise, the 50 μM LiCl concentration increased the stem height by 68, 48.3, and 31.8% after 11, 13, and 15 days after the start of treatments, with respect to the control. On the contrary, the highest evaluated dose (100 μM LiCl) significantly reduced shoot growth in all evaluations carried out compared to the control. This negative effect decreased as time passed, with values of 100, 83.7, 65.4, 44.7, 38.8, and 35.7% on days 9, 11, 13, 15, 17, and 18, respectively (Figure 5).

In amaranth plants, where treatments with doses ranging from 10 to 100 mg $\text{Li}_2\text{SO}_4 \text{ kg}^{-1}$ increased root length; on the contrary, those same concentrations decreased stem length when treating the seedlings for 21 days (Gayathri *et al.*, 2022). Kalinowska *et al.* (2013) studied the effects of LiCl and LiOH on the growth of butterhead lettuce (*Lactuca*

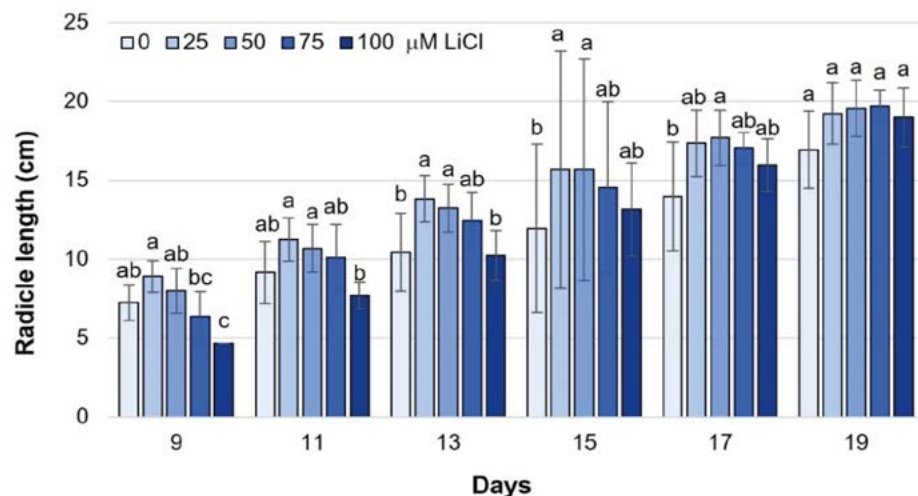


Figure 4. Radicle length of chili seedlings from seeds treated with lithium chloride (LiCl). Means \pm SD with different letters indicate statistical differences (Duncan, $P \leq 0.05$).

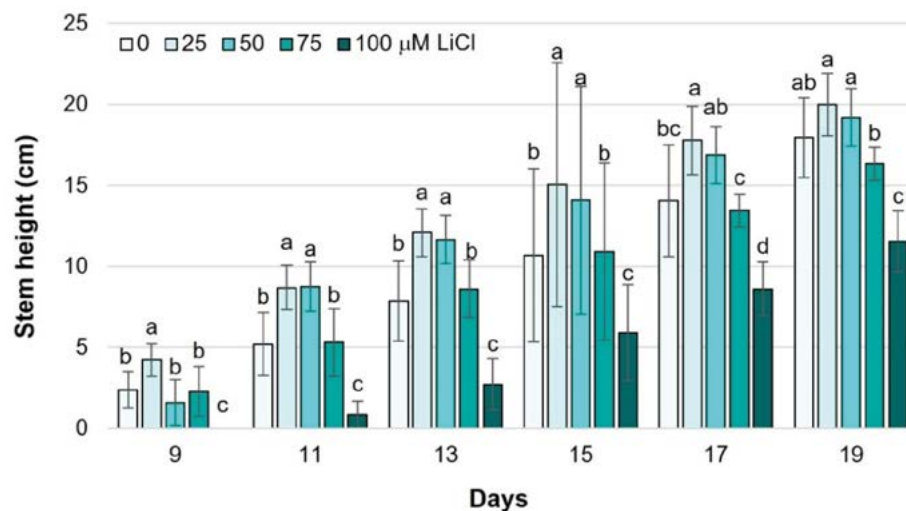


Figure 5. Stem height of Guajillo chili seedlings from seeds treated with lithium chloride (LiCl). Means \pm SD with different letters indicate statistical differences (Duncan, $P \leq 0.05$).

sativa var. *capitata*); their results revealed that the reduction in shoot growth is correlated with increasing concentrations from 20 to 100 mg Li dm⁻³ with both Li compounds, with a higher inhibition rate in LiOH than in LiCl. Similarly, previous studies in Abyssinian mustard seedlings demonstrated that 60-120 μ M treatments reduced root growth after 10 d of LiCl exposure (Li *et al.*, 2009). In soybean (*Glycine max*) plants grown in soil, the application of 25 mg Li kg⁻¹ for 28 d increased plant height by 10% compared to the control. On the contrary, when applying 100 and 200 mg Li kg⁻¹ the significant reduction of 5 to 55% in plant height was imminent (Shakoor *et al.*, 2023).

The inhibition of growth at high Li concentrations suggests the increase in the production of reactive oxygen species (ROS) (Iannilli *et al.*, 2024). In soybean plants exposed to the Li⁺ ion, an increase in the activity of the enzymes superoxide dismutase (SOD) and catalase (CAT) was observed, which protect structures at the subcellular level and prevent oxidative damage caused by ROS (Shakoor *et al.*, 2023). In spinach (*Spinacia oleracea*) leaves, concentrations of 20 to 80 mg Li kg⁻¹ were observed to cause lipid peroxidation and significant increases in the production of H₂O₂, SOD, CAT, and ascorbate peroxidase (APX) (Bakhat *et al.*, 2020).

Fresh and dry biomass of seedlings

The LiCl concentrations evaluated did not significantly affect the fresh and dry biomass of Guajillo chili seedlings; however, decreases are observed in both variables with the 100 μ M LiCl dose (Table 2).

In amaranth seedlings treated for 21 days with Li₂SO₄, concentrations of 50 to 100 mg kg⁻¹ increased the total biomass. The application of 10 to 100 mg kg⁻¹ inhibited root biomass, and treatments with 10, 50, 75, and 100 mg kg⁻¹ increased shoot biomass (Gayathri *et al.*, 2022). In maize (*Zea mays*) plants grown in Hoagland nutrient solution, there have been significant increases with doses 16 and 32 mg Li dm⁻³ in stem biomass,

Table 2. Fresh and dry biomass of Guajillo chili seedlings from seeds treated with lithium chloride (LiCl).

LiCl (μM)	Fresh seedling biomass (mg)	Dry seedling biomass (mg)
0	34.778 \pm 0.210a	3.556 \pm 0.509a
25	30.667 \pm 1.528a	3.500 \pm 0.220a
50	32.333 \pm 2.028a	3.167 \pm 0.083a
75	33.889 \pm 5.394a	3.278 \pm 0.241a
100	26.444 \pm 3.025a	2.722 \pm 0.268a

Means \pm SD with the same letter in each column indicate that there are no statistical differences (Duncan, $P \leq 0.05$).

while, with the range of 1 to 32 mg Li dm⁻³, the biomass of leaves, roots, and total increased (Antonkiewicz *et al.*, 2017). Negative effects of Li on biomass have also been reported. In butterhead lettuce, treatment with two sources of Li (LiCl and LiOH) at concentrations of 20, 50, and 100 mg dm⁻³ decreased the fresh weight of the shoot. The concentration range of 50 to 100 mg dm⁻³ decreased the fresh root biomass; however, the doses of 2.5 and 20 mg dm⁻³ significantly increased the fresh root weight, suggesting beneficial effects of Li in lettuce growth at low doses (Kalinowska *et al.*, 2013). Likewise, LiCl caused significant reductions in the fresh weight of Abyssinian mustard seedlings in the concentration range of 30 to 120 μM after exposure for 10 d (Li *et al.*, 2009).

CONCLUSIONS

Doses of 25 to 50 μM improve germination and seed vigor indices, in addition to promoting stem growth. It is concluded that doses less than 50 μM LiCl are positive in the germination and initial growth of Guajillo chili seedlings.

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