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DOSES FOLIARES DE ZINCO NA BIOFORTIFICAÇÃO O DO JAMBU (Acmella Oleracea L.)

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DOSIS FOLIARES DE ZINC EN LA BIOFORTIFICACIÓN DEL JAMBÚ (Acmella Oleracea L.)

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RESUMO

Este estudo visou avaliar qual dose de zinco foliar é a mais adequada para a biofortificação do jambu. Foi conduzido no IFPA, campus Belém, o experimento com cinco doses foliares

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de zinco (0, 10, 20, 30 e 40 mg mL-1, fonte de Zn (sulfato de zinco) num sistema de cultivo hidropônico. Após cinco dias das mudas serem transplantadas, foi dado início ao experimento de pulverização foliar de zinco, que se estendeu durante 21 dias. Os resultados obtidos após à análise de variância e o teste de Tukey mostraram que a aplicação de 10 mg/L-1 foi a mais adequada para a biofortificação do jambu, logo, as aplicações superiores a 10 mg/L-1 influenciaram negativamente as plantas de jambu, apresentando sintomas característicos de fitotoxidez. As desordens apresentadas na planta foram as raízes menores e escurecidas, clorose das folhas e folhas deformadas. Portanto, no presente trabalho 10mg/L-1 foi a dosagem ótima para a biofortificação do jambu.

Palavras-chave: Saúde. Hidroponia. Hortaliça. Fome oculta.

ABSTRACT

The aim of this study was to assess which dose of foliar zinc is most suitable for biofortifying jambu. The experiment was conducted at IFPA, Belém campus, with five foliar doses of zinc (0, 10, 20, 30 and 40 mg mL-1, Zn source (zinc sulphate) in a hydroponic cultivation system. Five days after the seedlings were transplanted, the foliar zinc spraying experiment began and lasted 21 days. The results obtained after the analysis of variance and Tukey's test showed that the application of 10 mg/L-1 was the most suitable for the biofortification of jambu; therefore, applications of more than 10 mg/L-1 negatively influenced the jambu plants, showing characteristic symptoms of phytotoxicity. The plant disorders were smaller and darkened roots, leaf chlorosis and deformed leaves. Therefore, in this study 10mg/L-1 was the optimum dosage for the biofortification of jambu.

Keywords: Health. Hydroponics. Vegetable. Hidden hunger.

RESUMEN

El objetivo de este estudio fue evaluar qué dosis de zinc foliar es la más adecuada para biofortificar el jambu. El experimento se realizó en el IFPA, campus de Belém, con cinco dosis foliares de zinc (0, 10, 20, 30 y 40 mg mL-1, fuente de Zn (sulfato de zinc) en un sistema de cultivo hidropónico. Cinco días después del trasplante de las plántulas, se inició el experimento de pulverización foliar de zinc, que duró 21 días. Los resultados obtenidos tras el análisis de la varianza y la prueba de Tukey mostraron que una aplicación de 10 mg/L-1 fue la más adecuada para la biofortificación del jambu, mientras que aplicaciones superiores a 10 mg/L-1 tuvieron un efecto negativo sobre las plantas de jambu, mostrando síntomas característicos de fitotoxicidad. Los trastornos de las plantas fueron raíces más pequeñas y ennegrecidas, clorosis foliar y hojas deformadas. Por lo tanto, en este estudio 10mg/L-1 fue la dosis óptima para la biofortificación del jambu.

Palabras clave: Salud. Hidroponía. Vegetal. Hambre oculta.

INTRODUCTION

Agronomic biofortification consists of enriching foods with minerals, especially Fe and Zn, through differentiated cultural treatments (VERGÜTZ *et al*., 2016). This biofortification can be done through some techniques, such as soil fertilization, seed treatment and foliar application, which are characterized as lower cost, more accessible and quick result techniques, as it only influences fertilization. Biofortification presents itself as an innovative and efficient tactic to combat malnutrition and food insecurity, particularly in areas where micronutrient shortages are frequent (LOUREIRO *et al*., 2018). The effects of this strategy have gone beyond personal health, as they foster the resilience of rural communities, stimulating crop diversity and adaptation to climate change. In addition, biofortification has the potential to reduce the need for food supplements and external measures, reinforcing the independence of the population in vulnerable situations (DE CARVALHO; NUTTI, 2012).

Jambu (*Acmella oleracea L.*), belonging to the Asteraceae family, is a plant native to the Amazon Basin widely used in cooking. In Pará, the leaves are widely consumed in typical regional dishes such as duck in tucupi, moqueca, caldeirada and tacacá, as well as salads (HOMMA et al., 2014). The largest production of jambu occurs in the northern region of Brazil, which results in financial profitability for the population of this area. However, there are no official market statistics on the prices and consumption of jambu, it is known that the sale value in the northern region of Brazil fluctuates according to the time of year, close to festive events, the price reaches high values in the market, since it is often used in typical dishes of the region (BRASIL, 1997; HOMMA et al. 2014; MEDEIROS, 2014).

The consumption of jambu not only enriches the diet, but also promotes the appreciation of biodiversity and the food culture of the Amazon. Incorporating it into traditional recipes is a way to take advantage of its nutritional advantages and unique flavors. In jambu, there are several types of minerals, including macrominerals such as nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), and sulfur (S), as well as trace minerals such as boron (B), copper (Cu), iron (Fe), and zinc (Zn). The levels of these minerals are found in the largest leaves of the jambu (BORGES; GOTO; LIMA, 2013). Regarding health benefits, jambu has an extensive variety of uses, such as: vasodilator and antioxidant activity (WONGSAWATKUL et al., 2008), anesthetic effect (FREITAS-BLANCO et al., 2016), anti-inflammatory and antimicrobial action (GUPTA; PATEL; RAVINDRA et al., 2012), neuroprotective (SUWANJANG et al., 2017), gastroprotective and antinociceptive (NASCIMENTO et al., 2013; NOMURA et al., 2013).

Spirantol, a metabolite much researched in jambu for its biological properties, its concentration is higher in jambu flowers, but it is also found in the leaves and stem, in smaller quantities (DIAS et al., 2012). The concentration of spirantol is highest in the floral capitula of jambu. In addition, it is also found in the leaves and stem, although in smaller quantities. Spirantol has properties that fight inflammation, pain and antimicrobial action. It has the ability to relieve pain, lower fever, and fight infections. In addition, it is recognized for promoting blood circulation and can have benefits for digestion (MBEUNKUI et al., 2011; FAVORETO, R. GILBERT, B., 2010).

Folk medicine employs jambu, a plant with a high content of spirantol, recognized for its anesthetic and anti-inflammatory properties. Jambu is often used in teas and typical meals, highlighting its cultural and therapeutic relevance, particularly in the interval of pain and in strengthening the immune system. Spirantol, found in the leaves and flowers of jambu, is the component that causes the tingling sensation and numbness in the mouth, making this plant a traditional remedy for toothaches and throats (BALIEIRO et al., 2020; SEKO et al., 2020).

Zinc is an essential nutrient for human health, performing several structural, biochemical, and regulatory functions. Zinc is, after iron, the most widely distributed trace mineral in the human body, being present in large quantities in all tissues. It performs functions involved in cell division, metabolism, sexual development, immunity and cognitive capacity, performing significant structural, catalytic and regulatory actions (PEREIRA, 2009; MACÊDO, 2010; CHASAPIS, 2012; PEDRAZA, 2013).

The lack of zinc is seen as a global issue, especially in underdeveloped nations and in areas of poverty in large metropolises. The consequences can be serious, resulting in physical and intellectual damage. The relative lack of zinc should be taken into account in phases of accelerated growth, where the need for the mineral increases, such as in childhood, adolescence, pregnancy and breastfeeding (PERSON, *et al.*, 2006).

Hidden hunger, also known as hidden malnutrition, refers to a deficiency of essential micronutrients. According to the World Health Organization (WHO), poor diet, which encompasses not only malnutrition, but also specific deficiencies and overnutrition, still persists globally at the beginning of this millennium. More than 2 billion people have nutritional deficiencies and 800 million individuals do not meet their fundamental needs for energy and protein, another 600 million are affected by the consequences of an improper and unbalanced diet (JUNQUEIRA & PEETZ, 2001).

Hidden hunger affects both built and developing nations, although in different hidden ways (HORTON; ROSS, 2003). In advanced countries, malnutrition can arise in particular situations, while in developing countries, stagnant food and poverty intensify the problem. Hidden hunger affects a significant part of the populations in several countries, including those considered developed, such as the United States. It may be more common among vulnerable groups, such as the elderly and low-income individuals, who do not provide access to nutritious food (VILAS BOAS, 2013). In developing nations, scarcity of food variety and food uncertainty are crucial elements that exacerbate nutritional deficiencies. In addition, undisclosed hunger can result in long-term health problems, such as chronic illness and impaired child growth, impacting the productivity and well-being of communities (BRAVO, 2007).

In 2008, global food prices reached astronomical levels, remaining unchanged to this day. Therefore, during a crisis, a significant amount of capital from impacted sectors is directed to agribusiness, whether for the production of food commodities or not, with a focus on the production of agrofuels (GUEDES, 2011; MARQUES, 2011).

The biofortification strategy aims to increase the concentration of micronutrients, such as zinc, in food, contributing to the fight against hidden hunger. This strategy, which enriches crops with nutritious nutrients, can improve nutrition and reduce nutritional deficiencies in vulnerable groups (LOUREIRO *et al*., 2018). Jambu is a vegetable increasingly present in several dishes in Pará cuisine, in addition to being easy to prepare and low cost, any strategy aimed at improving agronomic biofortification can result in several health benefits. Therefore, the present study aims to evaluate the effect of the application of foliar Zn doses on the production and accumulation of the nutrient in jambu in hydroponic cultivation.

METHODOLOGY

PLACE OF STUDY

The experiment was conducted in a protected environment in a hydroponic system located in the experimental area of the Plant Biology Sector, at the Federal Institute of Education, Science and Technology of Pará – IFPA, Belém campus, which is located at coordinates 1º 17' 42" South Latitude and 47º 55' 00" Longitude WGr, at an altitude of 50 m. by the Köppen classification, it is of the Af type. And the average temperature is 26.5 °C. The average annual rainfall is 2432 mm (EMBRAPA, 1993).

EXPERIMENTAL PROCEDURE

The jambu seedlings were donated by Hidroponia Maria Sena after 15 days of sowing. Subsequently, they were transplanted to the hydroponic cultivation profiles, installed within a protected environment. It remained for another 27 days and then they were harvested for foliar analysis of zinc content. The total time of experiment days including transplanting, zinc application, harvesting and foliar zinc analysis was 42 days.

The hydroponic cultivation profiles were 75 mm, with each bench consisting of 16 profiles (for one treatment), three reservoirs, with a capacity of 310 L, totaling 768 plants, with 4 replications. The nutrient solution (SN) used for the production of jambu was developed and recommended by Furlani (1997) for most vegetables in the hydroponic cultivation system. For the solution to remain balanced, it is necessary to take into account the EC (Electrical Conductivity) and the pH, with their respective values suitable for a good production, the concentration of SN varies according to the species of the plant and the type and size of the crop. According to Martinez (1997), the pH of the nutrient solution with CaCO3 is between 5.5 and 6.5, which is suitable for hydroponics. For EC, we observed in real time the adequate means, 1500 to 1700 μS/cm, values below 1000 μS/cm, there was nutrient replacement in the solution. Based on the concentration of Zn in Zinc Sulfate (ZnSO4.7H2O), which is 20%:Zn, concentrations of 0, 10, 20, 30 and 40 mg mL-1 of zinc were obtained for each treatment.

The EC and pH of the reservoirs of each treatment were checked daily. For nutrient replacement, a stock solution was used, i.e., a concentrated nutrient solution, for each treatment with their respective concentrations.

The foliar zinc spraying was done at three different times, the first application was on 09/25, the second on 10/01 and the third, on 10/08.

EVALUATED CHARACTERISTICS

Jambu was harvested 30 days after transplanting (DAT). 8 plants were evaluated centered to avoid the edge effect of each replication for agronomic characteristics and zinc leaf contents. For agronomic characteristics: The aerial part and the root of the jambu. They were weighed separately on a 0.01g precision scale of model S2202H 2200g, to obtain fresh mass. In the aerial part, leaves and stem of the jambu plants were considered. After this step, the shoot and roots were placed in paper bags, and taken to the drying oven with forced air circulation at a temperature of 65 ºC until a constant mass was obtained and thus the dry mass was obtained. For the purpose of comparison, the zinc contents in the aerial part were purchased bundles of jambu in traditional cultivation at two fairs in the metropolitan region of Belém. And analyses were also carried out of the essential oils present in the area of the treatment with 10mg of foliar zinc and in the packs purchased at the fairs. The dry mass of the aerial part was sent to the Chemistry Analysis Laboratory of the Belém campus, where the samples were crushed in a Whiley mill to determine the zinc content, by means of perchloric nitric digestion.

STATISTICAL ANALYSIS

The data obtained were submitted to analysis of variance (ANOVA). For the variables fresh matter and leaf zinc contents, Tukey's test was used, and for both analyses the Agroestat Software was used and for the preparation of tables and graphs the office package, Excel 2016, was used.

RESULTS AND DISCUSSION

The weights of fresh matter of the shoot, root and total were influenced by the foliar zinc doses. (Table 1). It is observed that there was a significant difference between the treatment with 10 mg. $^{L-1}$ of foliar zinc in relation to the other treatments. When compared to treatment with 40 mg. L-1, the total fresh matter weight (TFW), was more than 50% higher. And in relation to the control treatment, the increase was 39%. This demonstrates that treatment with the dosage of 10 mg of zinc positively influences the production of fresh matter. And that from this dosage there is a decrease in the production of fresh matter, with a negative effect on the production of fresh matter both in the shoot and in the roots. This can be considered as not only zinc saturation, but also competition with other mineral nutrients, such as potassium, calcium and magnesium, which can be impacted by high zinc concentrations (KHURANA & CHATTERJEE, 2001).

Table 1. Production of fresh mass (g/plant) of shoots (PMFA), roots (PMFR) and total (PMFT) of jambu plants as a function of treatments.

Treatment	PMFA	PMFR	PMFT
Dose 10 mg de Zn	218.66a	50.33a	269.00a
Dose 20 mg de Zn	115.00b	33.33b	148.33b
Dose 30 mg de Zn	111.66 b	21.00b	132.66 b
Dose 40 mg de Zn	111.00b	20,00b	131.00b
Control	143.00ab	22.66b	165.66 b
Test F	4,95**	19,27**	8,80**
CV (%)	25,61	17,17	21,63

Averages followed by the same letter vertically do not differ at 5% probability by Scott Knott's test. CV-Coefficient of variation.

The table above shows the results obtained from the analysis of the production of the plant's fresh mass, considering the aerial part, the roots and the total part. Where in the treatment with a dose of 10 mg of Zinc (Zn), the Production of Fresh Mass of the Aerial Part (PMFA) was 218.66 g, Production of Fresh Mass of the Roots (PMFR) with 50.33 g, and Production of Total Fresh Mass (PMFT), with 269 g. The second treatment, with 20 mg of

Zn, obtained a value of 115.00 for PMFA, 33, 33 for the PMFA and 148.33 for the PMFT. In the treatment of 30 mg of Zn, the value for PMFA was 111.66, for PMFA was 21.00, and for PMFT, 132.66. In the treatment with the dosage of 40 mg of Zn, the values for PMFA, PMFR and PMFT were, respectively: 111, 20 and 131 g. The control treatment, which was the plants that did not have foliar application of zinc, obtained the following averages: PMFA- 143.00; PMFR- 22.66 and PMFT- 165.66 g.

The increase in total fresh matter weight in relation to other treatments may be related to the fact that zinc favors cell division and cell development (MACÊDO, 2010). On the other hand, the reduction of total fresh matter at concentrations of 20, 30 and 40 mg/L is the result of the impact of excess zinc on plants in these quantities (KHURANA & CHATTERJEE, 2001).

The first dose (10 mg) showed a higher zinc absorption value compared to the other concentrations, as shown in Figure 1. It was also noted that the other concentrations showed results of zinc phytotoxicity in the leaves and roots of jambu, causing visual symptoms in the leaves and roots, due to the fact that the excess of the nutrient is harmful, causing visual symptoms of toxicity, affecting the production of fresh matter, as illustrated in the images of figures 2 and 3.

Zn stimulated different responses in some parameters: plant production and growth, zinc uptake and accumulation, as well as that for other parameters the trends of genetic materials were similar. In biomass, except for the Production of Shoot Fresh Mass (MFPA), the other data did not show differences in production between the varieties in all variables. "The leaf zinc content of curly lettuce was influenced by the cultivar and doses; however, no significant interaction between these factors was verified. Higher zinc application rates promote linear increases in leaf zinc content, with an increment rate of 0.049 mg kg-1 of foliar zinc for each gram of zinc (g ha-1) applied". The data of GRACIANO et al. are related to the research carried out in this article, since the different doses of foliar zinc application impacted the weights of fresh matter of the shoot, roots and total. However, no significant differences were noted between the other treatments. The analysis of the treatment that received 10 mg of zinc compared to that that received 40 mg varied a variation of more than 50% in total fresh matter weight (TFW). Compared to the control treatment, there was a reduction of 39%. Showing that the administration of 10 mg of zinc positively affects the production of fresh matter. Therefore, from this dose, the production of fresh matter decreases, resulting in a negative impact on both shoot and root production.

The total fresh mass value for Zn doses was adjusted to the second-degree equation, with the highest value being found at the 10mg dose. L-1 with an R2 of 0.96, demonstrating the beneficial effect of zinc on the jambu plant. This behavior of the jambu plant observed in the graph indicates that with the increase in dosages from 10 mg. L-1 occurs a decrease in total fresh matter due to the decrease in leaf area, potassium, iron and phosphorus uptake (PRADO, 2008). Increased zinc levels can affect plant physiology, resulting in a reduction in total fresh matter. This effect can be attributed to factors such as zinc toxicity at high concentrations, which impairs growth and nutrient absorption, leading to slower biomass development (Gupta et al. 2016).

The dynamics shown in image 1, which shows a drop in total fresh mass as doses exceed 10 mg. L−1, can be justified by several physiological effects. With the increase in zinc doses, the plant may begin to present phytotoxicity, which presents itself through symptoms such as decreased leaf area. The decrease in leaf area impairs the plant's photosynthesis capacity, leading to reduced energy production and, consequently, a decrease in biomass. such findings highlight the relevance of providing the correct amount of zinc to maximize the advantages without causing side effects (Prado, 2017).

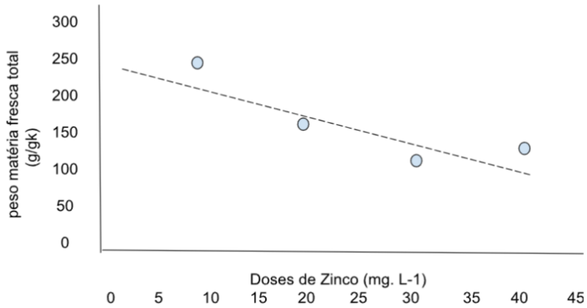

Figure 1: Total Fresh Matter Weight as a Function of Zinc Doses

Zinc phytotoxicity in plants occurs when there is an excessive amount of this micronutrient, resulting in several symptoms that affect plant development. The most frequent signs include yellowing of the leaves, especially at the ends, which can develop into necrosis. The leaves have spots or burns on the edges, and root growth can be impaired, leading to a smaller root system that is less effective at absorbing water and nutrients. In more serious situations, toxicity can culminate in the death of the plant. Therefore, it is essential to conduct close monitoring of zinc levels in the soil to prevent these negative effects and ensure crop health. The symptoms of zinc phytotoxicity in the jambu plant (figures 2 and 3) were due to the high concentrations of the nutrient, being, respectively: 20 mg/L-1, 30 mg/L-1 and 40 mg/L -1, the visible symptoms were: chlorosis at the beginning of the leaves, deformed leaves and then necrosis of the leaves, are similar to what Prado 2007, characterized in maize seedlings with high doses of zinc in the seeds, attributing them to low photosynthetic rate. E Sousa Soares *et al* 2001, *E. maculata and E.*

urophylla were sensitive to increasing doses of Zn in solution, exhibiting symptoms of phytotoxicity, manifested mainly as chlorosis, root browning and inhibition of plant growth. The manifestation of zinc phytotoxide in the roots of jambu occurs through a series of visual symptoms that reflect the physiological stress of the plant, impairing its growth and evolution. It is essential to consider these symptoms in advance to implement corrective actions that can mitigate the negative impacts of excess zinc in planting. In the roots of the jambu, the phytotoxide resulting from zinc can be detected through a variety of visual signs that signal stress in the plants, such as: the roots may exhibit a dark or greenish shade, causing the presence of cell lesions. In addition, root development can be impaired, leading to decreased development and a slimmer, more susceptible structure.

Table 2. Leaf contents of macro and micronutrients in the leaves of jambu plants as a function of the treatments.

Treatments	**N**	**P**	**K**	**Ca**	**Mg**	**S**
	Content (g/kg)					
10 mg. L-1	34.74a	7.64a	41.90c	16.71ab	9.60a	4.98a
20 mg. L-1	34.39a	6.69ab	40.50c	17.45ab	8.98a	4.19a
30 mg. L-1	37.24a	6.40b	32.05d	16.04ab	9.50A	4.61a
40 mg. L-1	40,13a	7.23ab	41.17c	15.07ab	8.97a	4.36a
Control	41.00a	7.55ab	49.85 b	19.04a	9.17a	4.31a
Fair	41.02a	7.64a	68.35a	12.98ab	5.77b	4.98a
Test F	3.87 NS	5.90*	101,67**	4.38NS	16,68**	1.64NS
CV (%)	5,92		3,83	8,64	5,44	10,46

Treatments	**Ass**	**Fe**	**Mn**	**Zn**
	Content (mg/kg)			
10 mg. L-1	7.42ab	98.60a	38.26a	9.6a
20 mg. L-1	7.87ab	106.61a	44.63a	8.98a
30 mg. L-1	5.44b	110.48a	44.63a	9.5A
40 mg. L-1	6.60ab	113.35a	40.66a	8.97a
Control	7.86ab	112.34a	66.71a	9.17a
Fair	8.72a	134.69a	34.38a	5.77b
Test F	5,44*	0.54NS	2.60NS	18,68**
CV (%)	9,52	20,53	22,35	5,44

Means followed by the same letter vertically do not differ at 1% probability by the Scott Knott test, NS- Not significant, CV- Coefficient of variation.

Figure 2. Visual symptoms of zinc phytotoxicity in jambu leaves

Figure 3. Visual symptoms of zinc phytotoxicity in jambu roots

Table 1. Essential oils analyzed in the aerial part of the jambu plant

Treatment with 10mg of zinc	Bought at the fair
Elemene <delta-> = 2.58	Elemene <delta-> = 1.88
Elemene<beta-> = 2.4	Elemene<beta-> = 1.45
Caryophyllene <(E)- = 21.58	Caryophyllene <(E)-> = 23.99
Elemene <gamma-> = 3.39	Elemene <gamma-> = 3.12
Isogermacrene D = 0.84	Humulene <alpha-> = 2.11
Humulene <alpha-> = 1.64	**Germacrene D = 25.98**
Germacrene D = 29.91	Zingiberene <alpha-> = 1.04
Zingiberene <alpha-> = 1	1-Pentadecene = 2.87
1-Pentadecene = 4.01	Farnesene<(E,E)-alpha-> = 2.23
Farnesene<(E,E)-alpha-> = 2.2	Kessane = 2.45
Kessane = 2.33	Caryophyllene oxide = 2.55
Caryophyllene oxide = 1.78	Oplopenone<beta-> = 0.64
Cadinol<alpha-> = 1.13	Cadinol<alpha-> = 1.08
Spirantol = 1.3	**Spirantol = 1.29**
Phytol = 6.63	**Phytol = 6.54**

In chart 1, it can be seen that there is an increase in the levels of essential oils, especially Germacrene D, Espilantol and Phitol when comparing the treatment with 10 mg of zinc in hydroponic cultivation in relation to common cultivation. Therefore, it reveals the possibility of advancing in new studies that can test these effects of zinc dosages on the levels of these essential oils. In particular, Spirantol is known to have a proven pharmacological effect (GUPTA; PATEL; RAVINDRA et al., 2012; NASCIMENTO et al., 2013; NOMURA et al., 2013; FREITAS-BLANCO et al., 2016; SUWANJANG et al., 2017), and in the present research, it was found only in the aerial part, jambu flowers were not used for the analysis of essential oils, where it is most found (Jacobson, 1957).

CONCLUSION

Foliar applications of zinc above 10 mg. L-1 negatively influenced the jambu plants, presenting characteristic symptoms of phytotoxicity;

The foliar application of 10 mg. L-1 of zinc was the one in which the jambu plant accumulated the most zinc without showing symptoms of phytotoxicity. Therefore, in the present study it was the optimal dosage for the biofortification of jambu;

The presented disorders of phytotoxicity in the plant were smaller and darkened roots, chlorosis of the leaves and deformed leaves.

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