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Nutritional contents of young cupuaçu (Theobroma grandiflorum) plants subjected to increasing copper doses

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Abstract

The cupuaçu tree (Theobroma grandiflorum), primarily cultivated in the North and Northeast regions of Brazil, has significant economic importance, especially for small producers. With the increasing demand, research on genetic improvement and agricultural practices has intensified, and cupuaçu cultivation has expanded with the use of modern technologies. The study conducted at Embrapa Amazonia Oriental in Belém-PA evaluated the effects of increasing doses of copper (Cu) on two cupuaçu tree progenies. The experiment was conducted in a greenhouse with five copper doses: 0, 15, 30, 60, and 120 mg/kg of soil, analyzing variables such as vegetative development and dry biomass. Copper is essential for photosynthesis and cellular respiration, but in excess, it can be toxic to plants. Progeny 63 showed an increase in Cu levels up to a critical dose of 15 mg/kg, followed by a decrease, indicating regulatory mechanisms against toxicity. Progeny 61, on the other hand, showed greater stability, with a gradual increase in Cu levels in the leaves and roots at higher doses, suggesting greater efficiency in utilizing and tolerating excess copper. Progeny 63 had difficulties translocating copper to the stem at high doses, while progeny 61 showed a more dynamic physiological response, allowing Cu redistribution throughout the plant parts. These results indicate that progeny 61 is more adaptive and resistant to the stress caused by Cu accumulation, while progeny 63 is more sensitive to toxicity, which may hinder its long-term development. At excessive doses, copper can affect stem lignification, causing damage to growth and plant stability.

Keywords: copper toxicity, cupuaçu tree, physiological response, plant tolerance, progenies

Introduction

The cupuaçu tree (Theobroma grandiflorum), a member of the Malvaceae family, is primarily grown in the Northern and Northeastern regions of Brazil. It plays a significant economic role for small-scale producers and traditional communities, contributing to their income and sustainability. The fruits of the cupuaçu tree are widely used in the food industry, mainly for producing juices, pulps, sweets, and jellies. With growing economic interest in this crop, research focused on genetic improvement and the development of more efficient agricultural practices has become increasingly important.

Historically, cupuaçu cultivation was limited to small orchards and home gardens, with farming techniques passed down through generations. However, as demand for cupuaçu has risen both domestically and internationally, it became essential to improve agricultural practices to meet this growing demand for quality products. The adoption of modern agricultural technologies, in combination with traditional knowledge from producers, has increased production and facilitated market expansion, particularly on the international stage.

Adequate mineral nutrition is vital for agricultural productivity, with micronutrients playing key roles in plant development. Copper (Cu), although required in small amounts, is essential for various physiological processes, including photosynthesis, cellular respiration, electron transport, and the formation of cell walls. However, when present in excessive amounts, copper can be toxic to plants, causing damage to root growth and impairing the absorption of other essential nutrients. Excess copper can also induce oxidative stress and reduce the plant's ability to absorb water and nutrients, which negatively impacts overall plant development.

Research highlights the importance of proper copper management in agricultural crops to improve

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productivity, but excess copper can lead to significant physiological damage. Studies on various crops, including Theobroma grandiflorum, indicate that plants may accumulate copper in their roots as a storage mechanism. However, high levels of copper accumulation can disrupt root growth and affect nutrient absorption.

The ideal copper dose for crops remains a topic of debate, particularly considering the potential negative effects of copper overload in plants. A thorough understanding of how copper interacts with the physiological mechanisms of the cupuaçu tree is critical for developing more effective management strategies.

In this context, the aim of this study is to analyze the effects of increasing copper doses on the development and nutrition of young cupuaçu plants. The study will focus on quantifying nutrient levels in the leaves, stems, and roots, contributing valuable insights into the optimal copper management for cupuaçu cultivation. This research will help improve agricultural practices and provide guidance for maximizing the productivity and sustainability of this important crop.

Material And Methods

The experiment was conducted at the physical base of Embrapa Amazônia Oriental, in the municipality of Belém-PA, in a greenhouse with an area of approximately 100 m². The structure is composed of glass roofing and a lantern-type opening, along with metal mesh sidewalls that protect against pests without compromising air circulation. A shade cloth was installed under the roof to reduce light entry by 50%. According to the Köppen climate classification, the region has an Afi climate, with an average annual temperature of 26°C (BASTOS et al., 2002).

The materials used in the research were progenies 61 and 63, from the genetic improvement program of the cupuaçu tree at Embrapa Amazônia Oriental. The experimental design was completely randomized, with a 5 × 2 factorial arrangement, totaling 10 treatments. The factors analyzed were five copper (Cu) doses applied to two progenies, with five replications per treatment, resulting in 50 experimental units (5 treatments × 5 replications).

The macronutrient solutions were prepared with the following dosages: 78 g of urea, 169 g of dicalcium phosphate, 58 g of potassium chloride, 207 g of magnesium sulfate, 87 g of calcium sulfate, and 27 g of sodium sulfate. Each compound was separately dissolved in 2,020 mL of distilled water, with 20 mL of the solution applied to each seedling. For micronutrients, solutions were prepared containing 6.2 g of manganese sulfate, 3.5 g of boric acid, and 9 g of zinc sulfate, dissolved separately in 820 mL of distilled water, with 20 mL applied per seedling.

Copper doses were applied in increasing amounts across the following treatments: T1 (0 mg/kg of soil), T2 (15 mg/kg of soil, with 9.7 g of copper sulfate), T3 (30 mg/kg of soil, with 19.4 g of copper sulfate), T4 (60 mg/kg of soil, with 38.8 g of copper sulfate), and T5 (120 mg/kg of soil, with 77.6 g of copper sulfate). All solutions were separately diluted in 820 mL of distilled water. Fertilizer weighing was done using a digital scale, and the volume of distilled water was measured using a 1,000 mL graduated cylinder. Cultural practices included controlled irrigation (300 mL of water per seedling), manual pest control, and weed removal. At 85 days after planting, treatments with macro and micronutrients were applied. The seedlings, grown in 4 kg polyethylene bags and organized by progeny on benches, received nutrients in liquid form, applied with a syringe to ensure precise dosages. Analysis was carried out monthly over 171 days. In the control treatment, seedlings were irrigated daily with 300 mL of water to maintain field capacity.

At the end of the experiment, 256 days after planting, roots, stems, and leaves were collected and dried in an oven at 70°C for 72 hours. The dried material was then weighed using a digital precision scale to determine dry biomass. Subsequently, the samples were ground in an electric grinder, identified by plant organ and progeny, and sent for nutritional analysis in a specialized laboratory. The variables analyzed included vegetative development, height, stem diameter, leaf area, number of leaves, dry mass of leaves, dry mass of stems, dry mass of roots, and total dry mass. The results were subjected to analysis of variance and compared using the Sisvar mean test, considering a significance level of 5%.

Results And Discussion

The analysis of variance (**Table 1**) revealed that both the main effects of the factors Progeny and doses, as well as their interaction, were statistically significant for the analyzed variables. These results indicate that the response to different doses varies depending on the progeny evaluated. Thus, it is possible to identify, for each response variable, which progenies showed the best performance based on the applied doses (**Table 2**).

Copper Content in Leaves

The results presented in **Figure 1** show differentiated responses of cupuaçu progenies 61 and 63 to increasing doses of applied copper (Cu). The regression equations fitted to the quadratic polynomial model indicate that Cu

Table 1 - Summary of the analysis of variance for the applied treatments and evaluated variables

fv	gl	leaf	d. stem	ms root
progeny	1	**	**	**
doses	4	**	**	**
progeny*doses	4	*	**	**
cv (%)		11,12	10,31	14,4

FV: Source of Variation; GL: Degrees of Freedom; CV: Coefficient of Variation; D. Stem: Stem Diameter; MS Root: Root Dry Mass; **,*: Significant at the 1% and 5% levels, respectively, by the SNK test ($p < 0.05$). Source: Author (2025).

Table 2 - Soil Analysis

parameters		units	values
depth		(cm)	0 - 20
c		g/kg	11,4
mo		g/kg	19,7
n		%	0,079
n		g/kg	0,79
relation		c/n	14,56
P		mg/dm³	424
K			748
Na			191
Al		cmolc/dm³	0,1
Ca			5,0
Ca+Mg			6,7
pH		water	5,4
H+Al		cmolc/dm³	3,30
CTC	total	cmolc/dm³	12,75
	effective		9,55
saturation	base	v%	74,11
	al	m%	1,05

Source: Author (2025).

absorption and translocation varied between progenies, suggesting distinct physiological adaptation mechanisms to micronutrient supply.

In progeny 63, there was a sharp increase in leaf Cu content up to the dose of 15 mg kg^{-1}, followed by a tendency toward stabilization or a slight decline at higher doses. This behavior suggests that progeny 63 has a high initial capacity for micronutrient uptake but may exhibit homeostatic regulatory mechanisms that limit excessive accumulation in the leaves.

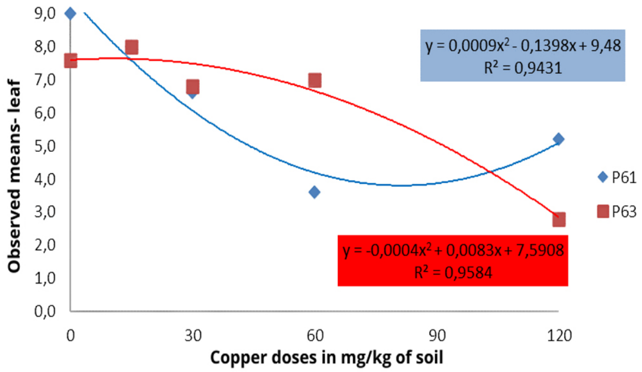

Figure 1 – Regression equations for leaf Cu content in cupuaçu progenies 61 and 63 as a function of applied Cu doses. Source: Author (2025).

On the other hand, progeny 61 showed a more stable response to increasing Cu doses, with a less pronounced progressive accumulation. This may indicate greater physiological efficiency in Cu utilization, optimizing its role in essential metabolic processes such as the activation of enzymes involved in photosynthesis, lignification, and electron transport (PRADO, 2020). Furthermore, the lower variation in leaf Cu content in progeny 61 may suggest better regulation of micronutrient redistribution within the plant, reducing the risk of toxicity at higher doses.

The results indicate that, from a critical Cu dose onward, leaf Cu content began to decrease, suggesting the activation of physiological regulatory mechanisms and/or possible toxic effects due to excess micronutrient. This decline may be related to limitations in Cu uptake by the roots due to ionic competition with other cations or to the precipitation of the element into less available forms in the soil (MARSCHNER, 2012). In addition, plants exposed to high Cu concentrations may activate tolerance strategies such as metal sequestration in vacuoles or complexation with phytochelatins and metallothioneins, reducing its translocation to the leaves (PRADO, 2020).

The differential response between progenies 61 and 63 reinforces the existence of genetic variability in Cu tolerance among cupuaçu genotypes. Progeny 63 exhibited a classic toxicity response pattern, reaching a maximum Cu content in the leaves at a dose of 15 mg kg^{-1}, followed by a progressive decline at higher doses, suggesting the activation of defense mechanisms to prevent excessive metal accumulation in plant tissues.

In contrast, progeny 61 showed a distinct behavior. Although it also initially exhibited a reduction in leaf Cu content with increasing doses, this trend was reversed from the 60 mg kg^{-1} dose onward, resulting in a renewed increase in micronutrient concentration in the leaves. This pattern suggests that progeny 61 has a more dynamic regulatory mechanism, allowing for adjustments in Cu uptake and redistribution under high availability conditions. This behavior may be related to greater physiological plasticity, granting it greater tolerance to environments with elevated levels of the element.

Copper Content in the Stem

The results presented in **Figure 2** indicate that Cu content in the stems of cupuaçu progenies 61 and 63 decreased in response to increasing doses of the micronutrient, following a quadratic polynomial model.

In progeny 63, a continuous reduction in stem Cu content was observed as the applied doses increased. This behavior suggests that this progeny exhibits limitations

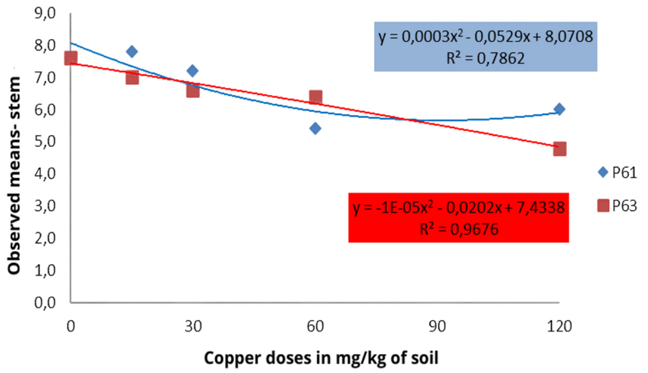

Figure 2 – Regression equations for Cu levels in the stem of cupuaçu progenies 61 and 63 as a function of the applied Cu doses. Source: Author (2025).

in Cu translocation to the stem, possibly due to retention of the element in the roots or activation of exclusion mechanisms that restrict its movement to aerial tissues. Such a mechanism may act as a defense strategy against toxicity, minimizing the exposure of stem tissues to excess Cu.

On the other hand, progeny 61 displayed a different behavior. Initially, stem Cu levels decreased with increasing Cu doses, following a trend similar to that of progeny 63. However, at higher doses, this trend was reversed, with an increase in Cu content in the stem. This pattern may indicate a lower restriction in Cu translocation or a physiological adjustment that favors metal transport to aerial parts after reaching a certain absorption threshold.

The slight recovery in stem Cu content at higher doses in progeny 61 may be associated with a more efficient physiological response to excess micronutrient, allowing for Cu redistribution across different plant organs. This behavior may reflect a greater adaptive capacity in this progeny, granting it higher tolerance to stress caused by Cu accumulation. In contrast, progeny 63 appears to experience more pronounced toxicity, resulting in continuous reduction in Cu translocation to the stem, which may compromise long-term plant development.

Previous studies, such as that of Silva & Corrêa (2021), have shown that high Cu doses can negatively affect the initial growth of *Theobroma grandiflorum* seedlings, including reduced stem development. This effect was evident in progeny 63, in which increasing Cu doses led to a progressive reduction in stem Cu content, reinforcing the hypothesis that this progeny is more sensitive to excess metal.

Moreover, copper plays an essential role in stem lignification, acting as a cofactor in lignin biosynthesis enzymes (Yruela, 2009). At adequate doses, this process contributes to the structural strengthening of the plant, increasing its mechanical resistance. However, in excess, Cu can induce excessive production of reactive oxygen species (ROS), leading to oxidative damage in stem tissues through lipid peroxidation and cellular degradation (Lombardi & Sebastiani, 2005). This effect may explain the drastic reduction in translocation and stem growth observed in progeny 63 at high Cu doses.

Reduced stem development can significantly impact cupuaçu productivity, as it affects plant stability and its ability to support the canopy. Additionally, weakened stems may be more susceptible to infection by fungal and bacterial pathogens, which tend to proliferate in plants under heavy metal stress (Gomes et al., 2017).

Copper levels in the root

The results presented in **Figure 3** indicate that the Cu levels in the stem of progenies 61 and 63 of cupuaçu trees decreased in response to the increased doses of the micronutrient, fitting a quadratic polynomial model.

In progeny 63, a continuous reduction in Cu levels in the stem was observed as the applied doses increased. This behavior suggests that this progeny has a limitation in Cu translocation to the stem, possibly due to retention of the element in the roots or the activation of physiological exclusion mechanisms that restrict its movement to aerial tissues. This mechanism may be a defense strategy against toxicity, minimizing exposure of stem tissues to excess Cu.

On the other hand, progeny 61 exhibited a distinct behavior. Initially, Cu levels in the stem decreased with increasing applied doses, following a similar trend observed in progeny 63. However, at higher doses, this trend was reversed, with an increase in Cu levels in the stem. This pattern may indicate a lower restriction in Cu translocation or a physiological adjustment that favors metal transport to the leaf tissues after a certain

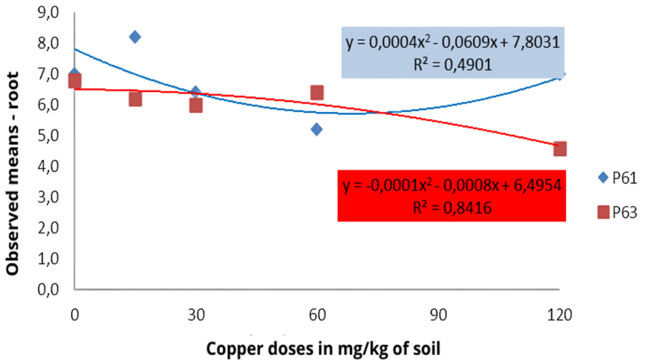

Figure 3 – Regression equations for Cu levels in the roots of cupuaçu progenies 61 and 63 as a function of the applied Cu doses. Source: Author (2025).

absorption threshold.

The slight recovery of Cu levels in the stem at high doses in progeny 61 may be associated with a more efficient physiological response to cope with excess micronutrient, allowing redistribution of Cu among different plant organs. This behavior may reflect a greater adaptive capacity of this progeny, conferring higher tolerance to stress caused by metal accumulation. In contrast, in progeny 63, toxicity appears more pronounced, resulting in a continuous reduction in Cu translocation to the stem, which may compromise plant development in the long term.

Previous studies, such as Silva and Corrêa (2021), have demonstrated that the application of high doses of Cu can negatively affect the initial growth of Theobroma grandiflorum seedlings, including reduction in stem development. This effect was evident in progeny 63, where increased Cu doses resulted in a progressive reduction of Cu levels in the stem, reinforcing the hypothesis that this progeny is more sensitive to metal excess.

Furthermore, copper plays an essential role in stem lignification, acting as an enzymatic cofactor in lignin biosynthesis (Yruela, 2009). At adequate doses, this process contributes to the structural strengthening of the plant, increasing its mechanical resistance. However, in excess, Cu can induce exacerbated production of reactive oxygen species (ROS), triggering oxidative damage in stem tissues through lipid peroxidation and cellular degradation (Lombardi & Sebastiani, 2005). This effect may explain the drastic reduction in Cu translocation and stem growth observed in progeny 63 at high Cu doses.

The reduction in stem development can significantly compromise cupuaçu production, as it affects plant stability and its capacity to support the canopy. Additionally, weakened stems may be more susceptible to fungal and bacterial pathogen infections, which tend to proliferate in plants under heavy metal stress (Gomes et al., 2017).

Conclusion

The evaluation of the two cupuaçu progenies (P61 and P63) in relation to the studied Cu doses indicated greater accumulation in progeny P63 up to a critical dose, followed by a reduction in levels, suggesting regulatory mechanisms against toxicity.

Progeny P61 showed a greater adaptive capacity, with increased Cu levels in the leaves and roots after higher doses, indicating tolerance to the metal.

References

Alves, E.O., Freitas, C.B.S., Oliveira, M.G., Silva, M.S. 2024. Genetic parameters and dissimilarity between progenies of ‘cupuaçu’ tree (*Theobroma grandiflorum*, Malvaceae) from free pollination in southeastern Pará, Brazil. *Revista Verde de Agroecologia e Desenvolvimento Sustentável* 19: 30-36.

Clement, C.R., Alves, R.M., Vicentini, A., Balée, W., Epps, P., Magalhães, M.P., Alves-Pereira, A., Carvalho, J.E.U., Ramirez, H. 2024. Finding the origin of domestication of cupuaçu requires more than genomics. https://preprints.scielo.org/index.php/scielo/preprint/view/8304.

Cunha, E., Lima, G., Sales, J., Oliveira, E. 2021. Determination of thermophysical properties of cupuassu (*Theobroma grandiflorum*) dry almonds. *The Journal of Engineering and Exact Sciences* 7: 11955-01-12E.

Cunha-Junior, J.B.D., Tavares, L.B. 2020. *Fenologia do cupuaçuzeiro (Theobroma grandiflorum (Willd. ex Spreng.) Schum.) em um sistema agroflorestal, submetido à diferentes lâminas de irrigação no município de Castanhal, PA*. (Trabalho de Conclusão de Curso) – Universidade Federal Rural da Amazônia, Belém, Brasil.

Ferraz dos Santos, L.; Santana Silva, R.J.; Falcão, L.L.; Alves, R.M.; Marcellino, L.H.; Micheli, F. 2022. Cupuassu (*Theobroma grandiflorum* [Willd. ex Sprengel] Schumann) fruit development: key genes involved in primary metabolism and stress response. *Agronomy* 12: 763.

Ferraz, I.D.K., Calvi, G.P. 2011. Teste de germinação. In: Lima Júnior, M.J.V. (ed.) *Manual de procedimentos de análise de sementes florestais*. Abrates, Londrina, Brasil. p. 5.1-5.36.

Franklin, B., Nascimento, F.D.C.A. 2020. Plants for the future: data compilation ofnutritional composition of guavaboi, burity, cupuaçu, murici and peach palm. Brazilian Journal of Development 6: 10174- 10189.

Genovese, M.I., Lannes, S.C.D.S. 2009. Comparison of total phenolic content and antiradical capacity of powders and “chocolates” from cocoa and cupuassu. *Food Science and Technology* 29: 810-814.

Gomes, M.P., Marques, D.M., Carneiro, M.M.L.C., Soares, Â.M. 2017. Effects of copper on plants: a toxicological approach. *Environmental Toxicology and Chemistry* 36: 1345-1355.

Jatav, H.S., 2020. Micronutrientes essenciais para o desenvolvimento das plantas e seu impacto na agricultura. *Journal of Agricultural Science* 8: 305-314.

Lim, T.K. 2012. *Theobroma grandiflorum*. In: *Edible medicinal and non medicinal plants*. Springer, Dordrecht, Netherlands.

Lombardi, L., Sebastiani, L. 2005. Copper toxicity in *Prunus cerasifera*: growth and antioxidant enzymes responses. *Tree Physiology* 25: 1273-1280.

Lombardi, M.P., Sebastiani, L. 2005. A toxicidade do cobre nas plantas: efeitos sobre o crescimento radicular e a absorção de nutrientes. *Plant and Soil* 267: 155-163.

Pereira, A., Abreu, V., Rodrigues, S. 2018. Cupuassu—*Theobroma grandiflorum*. In: *Exotic fruits*. Academic Press, Amsterdam, Netherlands. p. 159-162.

Ramos, S. Salazar, M. Nascimento, L. Carazzolle, M. Pereira, G. Delforno, T. Nascimento, M. Aleluia, T. Celeghini, R. Efraim, P. 2020. Influence of pulp on the microbial diversity during cupuassu fermentation. *International Journal of Food Microbiology* 318: 108465.

Silva da Costa, R. Pinheiro, W. B.S. Arruda, M.S.P. Costa, C.E.F. Converti, A. Costa , R.M.R. Silva Júnior, J.O.C. 2022. Thermoanalytical and phytochemical study of the cupuassu (*Theobroma grandiflorum* Schum.) seed by-product in different processing stages. *Journal of Thermal Analysis and Calorimetry* 147: 275-284.

Silva, A.S., Silva, A.L.S., Silva, L.F., Silva, A.L.S. 2023. Tolerância de plantas de *Cordia americana* expostas ao excesso de cobre. *Ciência Florestal* 33: 1-14.

Silva, E.M.F., Corrêa, V.R.C. 2021. *Theobroma grandiflorum: os efeitos do cobre, gessagem e correção do solo em fase inicial de desenvolvimento*. Universidade Federal Rural da Amazônia, Belém, Brasil. p. 40-51

Souza, C.A. 2016. Impactos do excesso de cobre e zinco no crescimento de plantas de videira. *Revista Brasileira de Ciências Agrárias* 11: 45-53.

Souza, C.R.B., Brunetto, G., Couto, E.G., Santos, D.R. 2016. Impacto do excesso de cobre e zinco no solo sobre videiras e plantas de cobertura. Embrapa Uva e Vinho, Bento Gonçalves, Brasil. p. 91-101.

Tavanti, G. 2021. O papel do cobre na fisiologia das plantas e sua importância na fotossíntese e metabolismo celular. *Revista Brasileira de Fisiologia Vegetal* 33: 145-157.

Vieira, M.E., Alves, R.M., Freitas, M.S.M., Viégas, I.J.M., Chaves, S.F.S., Peçanha, D.A., Vivas. M. 2022). Seleção de clones de cupuaçuzeiro visando qualidade de fruto. *Revista Brasileira de Ciências Agrárias* 17: 1-9.

Yruela, I. 2009. Copper in plants: acquisition, transport and interactions. *Functional Plant Biology* 36: 409-430.

Yruela, I. 2009. Efeitos da toxicidade do cobre nas plantas: mecanismos fisiológicos e respostas a estresses. *Environmental and Experimental Botany* 68: 11-19.

Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.