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Micronutrient deficiency affects the development of the seedlings of the cagaita, a *Myrtaceae* typical of the Brazilian Cerrado

A deficiência de micronutrientes afeta o desenvolvimento de mudas de cagaita, uma *Myrtaceae* típica do Cerrado brasileiro

La deficiencia de micronutrientes afecta el desarrollo de las plántulas de la cagaita, una *Myrtaceae* típica del Cerrado brasileño

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Resumo

As plantas de cagaita (*Eugenia dysenterica* DC.) são altamente dependentes do micronutriente ferro durante seus estágios iniciais de desenvolvimento. Diante disso, testamos a hipótese de que o desenvolvimento inicial das mudas é afetado pela falta de micronutrientes específicos. Cultivamos mudas dessa espécie em solução hidropônica completa e em tratamentos individuais, nos quais B, Cu, Fe, Mn, Zn ou Mo foram omitidos. O aparecimento de sintomas de deficiência de micronutrientes em resposta às diferentes soluções de tratamento foi monitorado visualmente por 120 dias após o transplantio, sendo determinados os parâmetros de crescimento e o conteúdo de diferentes nutrientes nos tecidos vegetais. A deficiência do micronutriente Mo foi o tratamento que mais afetou o no crescimento das mudas. Os sintomas mais comumente observados de deficiência de nutrientes foram clorose e necrose (privação de Cu e Zn) e comprometimento do desenvolvimento dos pêlos radiculares (privação de Fe e

Cu). De maneira geral, o Fe foi o micronutriente que mais se acumulou no caule e nas raízes das mudas expostas à solução nutritiva padrão, o que confirma a importância da disponibilidade desse nutriente nos estágios iniciais de desenvolvimento das mudas para a produção de plantas saudáveis de cagaita, com maior viabilidade.

Palavras-chave: *Eugenia dysenterica* DC.; Nutriente; Hidroponia; Crescimento; Estresse nutricional.

Abstract

Cagaita (*Eugenia dysenterica* DC.) plants are highly dependent on the micronutrient iron during their early development stages. Given this, we tested the hypothesis that initial seedling development is affected by a lack of specific micronutrients. We planted seedlings of this species in standard hydroponic solution, and in individual treatments, in which B, Cu, Fe, Mn, Zn or Mo were omitted. The development of symptoms of micronutrient deficiency in response to the different treatment solutions was monitored visually for 120 days after transplantation, and growth parameters and the content of different nutrients in the plant tissue were determined. The deficiency of the micronutrient Mo was the treatment with the greatest effect on seedling growth. The most commonly observed symptoms of nutrient deficiency were chlorosis and necrosis (Cu and Zn deprivation) and impaired root hair development (Fe and Cu deprivation). Overall, Fe was the micronutrient that accumulated most in the stems and roots of the seedlings exposed to the standard nutrient solution, which confirms the importance of the availability of this nutrient in the initial stages of seedling development for the production of healthy cagaita plants, with greater viability.

Keywords: *Eugenia dysenterica* DC.; Nutrient; Hydroponics; Growth; Nutrient stress.

Resumen

Las plantas de Cagaita (*Eugenia dysenterica* DC.) Dependen en gran medida del hierro micronutriente durante sus primeras etapas de desarrollo. Ante esto, probamos la hipótesis de que el desarrollo inicial de las plántulas se ve afectado por la falta de micronutrientes específicos. Sembraron plántulas de esta especie en solución hidropónica estándar y en tratamientos individuales, en los que se omitieron B, Cu, Fe, Mn, Zn o Mo. El desarrollo de síntomas de deficiencia de micronutrientes en respuesta a las diferentes soluciones de tratamiento se monitoreó visualmente durante 120 días después del trasplante, y se determinaron los parámetros de crecimiento y el contenido de diferentes nutrientes en el tejido vegetal. La deficiencia del micronutriente Mo fue el tratamiento con el mayor efecto en el crecimiento de las plántulas. Los síntomas más comúnmente observados de deficiencia de nutrientes fueron clorosis y necrosis (privación de Cu y Zn) y el desarrollo del vello radicular deteriorado (privación de Fe y Cu). En general, el Fe fue el micronutriente que más se acumuló en los tallos y raíces de las plántulas expuestas a la solución nutritiva estándar, lo que confirma la importancia de la disponibilidad de este nutriente en las etapas iniciales del desarrollo de las plántulas para la producción de plantas de cagaita saludables.

Palabras clave: *Eugenia dysenterica* DC.; Nutriente; Hidropónico; Crecimiento; Estrés de nutrientes.

1. Introduction

The cagaita (*Eugenia dysenterica* DC.) is a slow-growing, medium-sized fruiting tree native to the Brazilian Cerrado savanna biome. The consumer market for cagaita fruit, which has an acid pH, low titratable acidity, and high humidity (Camilo et al., 2014), is expanding rapidly (Reis and Schmiele, 2019), given that it can be used for the production of a range of processed foods, in particular drinks (Martins et al., 2017). However, the lack of reliable data on the nutritional requirements of this plant is a factor limiting the large-scale production of seedlings necessary for not only the commercial production of the fruit, but also reforestation and the recuperation of degraded habitats in the Cerrado (Bessa et al., 2016). Up to now, the only data relevant to the mass production of the seedlings of this species refer to macronutrients (Bessa et al., 2019).

Micronutrients are normally required by plants in extremely small quantities, and can be toxic at high concentrations. Given this, any deviation from the optimal concentrations of these essential nutrients in the soil may result in modifications of the structure, physiology or nutritional balance of the plant, although different species appear to respond differentially to the lack of micronutrients (e.g. Viégas et al., 2018; Araújo et al., 2016; Souza et al., 2016).

The homeostasis of micronutrients in the cell is achieved through the strict control of the acquisition, efflux, and storage of these elements. In the case of boron, for example, the cell equilibrium is regulated primarily by influx and efflux. When there is a deficiency of B, transporters, such as NIP5, may be activated (Takano et al., 2006), and when there is an excess of B, exporters may become active. By contrast, the homeostasis of iron requires the regulation of the genes related to the acquisition of this nutrient through the rhizophere, and the subsequent remobilisation of this element to the vacuole (Giehl et al., 2009). Any alteration that affects cell homeostasis will provoke biochemical intracellular imbalances that will have a negative impact on the development of the plant and the formation of its leaves. These alterations may be clearly observed, manifesting themselves in the form of visible symptoms of nutritional deficiency.

The rapid identification of changes in growth patterns and leaf morphology may contribute to the successful cultivation of seedlings, especially those of perennial species, by permitting the prediction of nutritional deficiency before the plant reaches an advanced stage of malnutrition, which would threaten its survival. The accurate determination of a plant's nutritional status may not only help prevent losses, but may also provide a baseline for the rational application of nutritional supplements, supported by the principles of precision agriculture (Barbedo et al., 2019). In this case, a visual diagnosis, which indicates the complete absence of micronutrients will be essential for the understanding of the plant's real requirements of these essential nutrients, although few insights are available for the native plants of the Cerrado. At the present time, all that is known of the development of the cagaita is that Fe is the most important nutrient for the initial development stage of the plant (Bessa et al., 2016).

In this context, the present study adopted a hydroponic approach, which permits the exact control of the components of the nutrient solution, to evaluate the hypothesis that the initial growth of *E. dysenterica* seedlings may be affected by the lack of specific micronutrients. The study tested the effects of the lack of micronutrients on the growth and nutrient balance of *E. dysenterica* seedlings, and recorded the visual symptoms provoked by the conditions of deficiency.

2. Materials and Methods

Extraction of the fruit and production of the seedlings

The cagaita fruit were obtained from wild plants growing on the Fazenda Gameleira, a private property in the municipality of Montes de Claros de Goiás, in Goiás, Brazil, in October 2018. These fruits were de-pulped manually for the extraction of the seeds, which were scarified manually to accelerate germination. Approximately 80 scarified seeds were sown in plastic trays containing autoclaved sand as the substrate. Thirty days after sowing, when the seedlings had 3–4 leaves, they were transplanted to the hydroponic cultivation system, using containers with a capacity of 8 litres, in a greenhouse, under a mean irradiance of 250 µmol $m^2.s^{-1}$.

To adapt the seedlings, they were initially kept in the hydroponic system for 30 days, using half of the ionic power of Hoagland and Arnon's (1950) solution with treatments that lacked one micronutrient (B, Fe, Cu, Zn, Mn or Mo), and the standard (complete) solution. At the end of this period, the seedlings were exposed to the treatment solutions.

Hydroponic cultivation of the seedlings, growth biometry, and the determination of the nutritional parameters

The pH of the nutrient solutions lacking the different micronutrients was adjusted daily to 5.5±0.5 using HCl and NaOH. The solutions were changed every time they reached 30% of the initial electrical conductivity.

Growth was evaluated based on four biometric parameters: length of the stem and the root, diameter of the stem, and the number of leaves. The seedlings were evaluated at 30, 60, 90, and 120 DAT (days after transplantation). At the final evaluation, the seedlings were separated into root, stem, and leaves for drying in a forced air circulation oven at 65ºC until reaching a constant weight, in order to determine the dry mass. Once dried, the plants were ground in a Wiley-type mill equipped with a sieve (20 mesh) to determine the micronutrient (B, Cu, Fe, Mn, Zn, and Mo) content of the root and the aerial part of the plant, using the procedure of Malavolta (1997).

The visual symptoms of micronutrient deficiency were evaluated and photographed digitally at 15-day intervals between 30 DAT and 120 DAT. A qualitative approach was used to select the photographs that best represented the visible effects of the nutritional disorders.

Statistical analysis

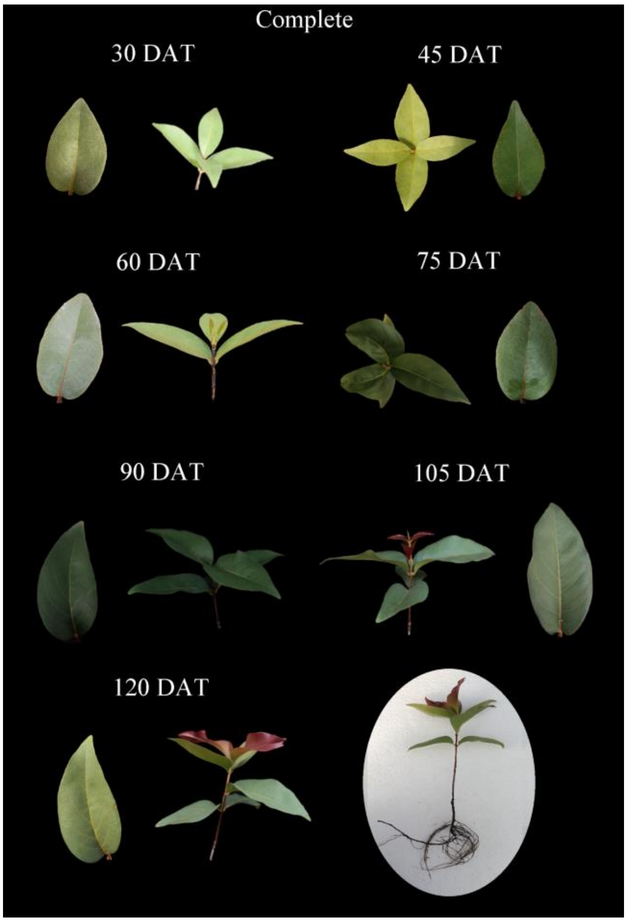
The experiment had a double factorial delineation (6 nutrient deficiencies x 4 evaluation times), arranged in random blocks. The treatments involved deficiencies of the B, Cu, Fe, Mn, Zn, and Mo, with the standard nutrient solution being used as the control. The four measures of seedling development were obtained at 30, 60, 90, and 120 DAT, with four repetitions per treatment (each repetition consisted of a pot with two seedlings). The mean values of the four parameters were evaluated using an Analysis of Variance, with Tukey's *post hoc* test, considering a probability of 5%. This analysis was run in the R 3.4.4 platform (R Core Team, 2018).

3. Results

Symptoms of deficiency and nutrient content

During all assessment stages, from 30 to 120 DAT, the *E. dysenterica* seedlings grown in standard nutrient solution (containing all macro- and micronutrients) developed healthy leaves with no symptoms of malnutrition. The roots of these plants also developed normally, indicating effective nutrient uptake from the solution (Figure 1).

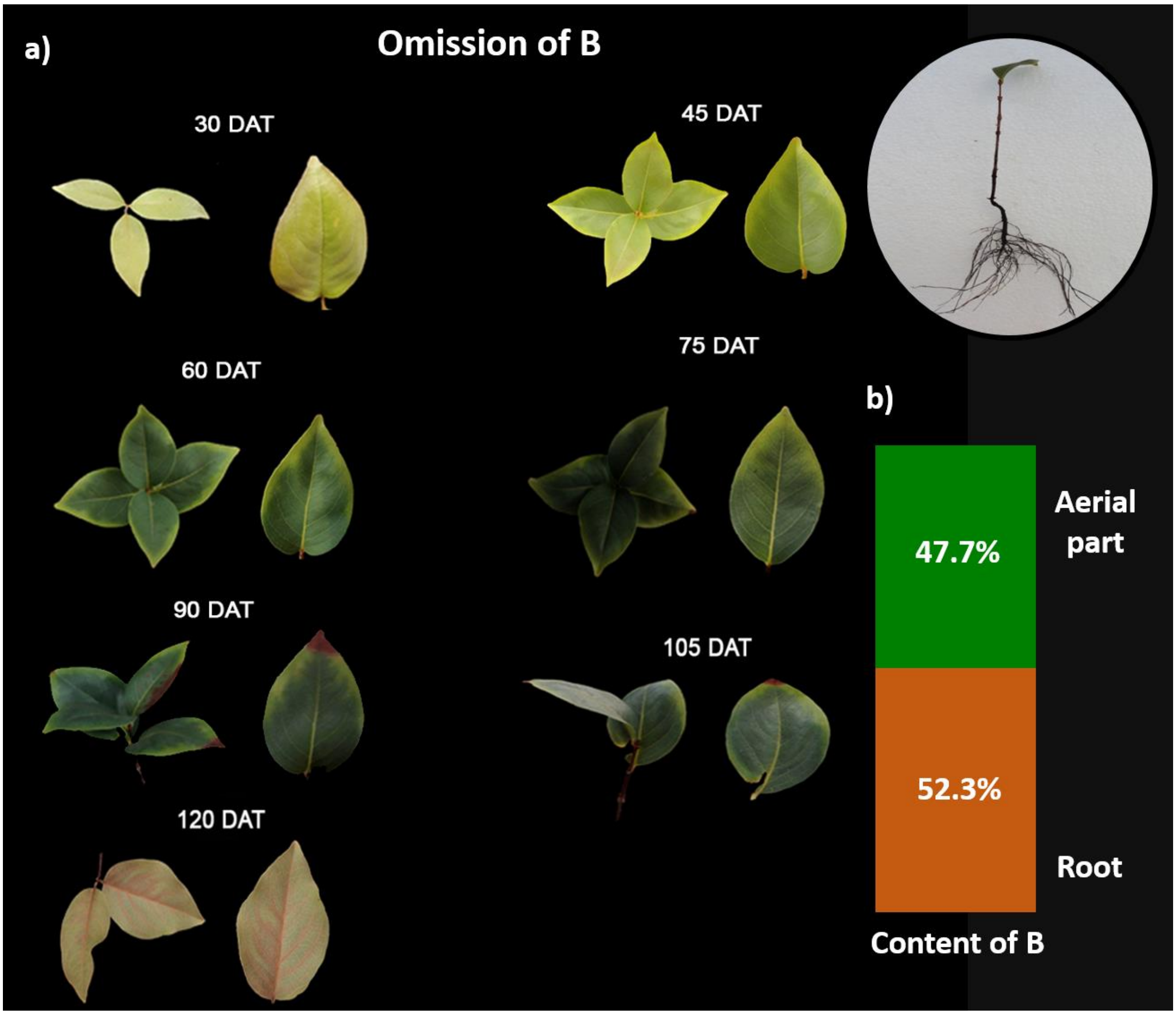
Figure 1 – Appearance of the leaves and shoots of *Eugenia dysenterica* DC. seedlings grown in a hydroponic system using a standard nutrient solution (including B, Fe, Mn, Cu, Zn, and Mo). The records were acquired at 30, 45, 60, 75, 90, 105 and 120 days after transplantation (DAT) to the nutrient solution.



Source: Authors.

Symptoms of B deficiency were observed from 30 DAT onward, with chlorosis occurring in the younger leaves, starting at the margins and progressing toward the central parts of the blade. Marginal necrosis was observed starting at 60 DAT. Another symptom of B deficiency was the loss of apical dominance, observed at 90 DAT. The roots presented no decrease in secondary growth or hair formation, in contrast with all the other treatments (Figure 2a). The accumulation of B was slightly greater in the roots (52.3%) than in the shoots (47.7%) of the plants of the B treatment (Figure 2b).

Figure 2 – Appearance of the symptoms of B deficiency in the leaves and shoots of the *Eugenia dysenterica* DC. seedlings grown in a hydroponic system in a nutrient solution lacking B. Records were acquired at 30, 45, 60, 75, 90, 105, and 120 days after transplantation (DAT) to the nutrient solution (a). Partitioning of B in the shoots and roots of the plants treated with nutrient solution lacking B (b).



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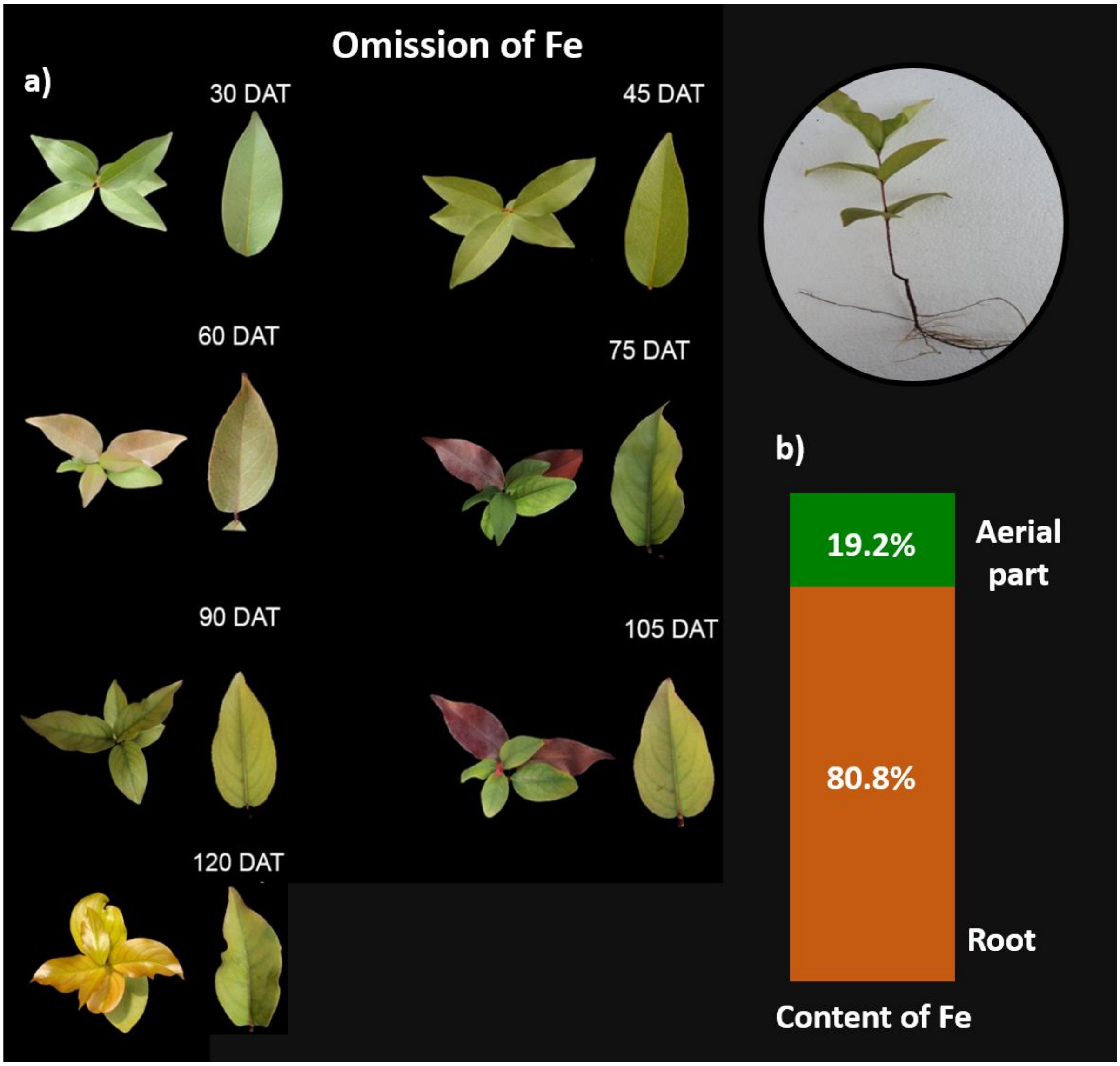
Symptoms of Fe deficiency were identified starting at 60 DAT, when chlorosis began to develop in the younger leaves. Chlorosis started at the base of the petiole and quickly spread to the blade. At 75 DAT, chlorosis had already reached the entire area of the blade of the younger leaves, which had a direct impact on the development of these leaves. The roots were dark with few hairs, especially at 105 DAT (Figure 3a). The plants in this treatment allocated Fe primarily(80.8%; Figure 3b) to the root tissue.

Figure 3 – Appearance of the symptoms of Fe deficiency in the leaves and shoots of the *Eugenia dysenterica* DC. seedlings grown in a hydroponic system in a nutrient solution lacking B. Records were acquired at 30, 45, 60, 75, 90, 105, and 120 days after transplantation (DAT) to the nutrient solution (a). Partitioning of Fe in the shoots and roots of the plants treated with nutrient solution lacking Fe (b).



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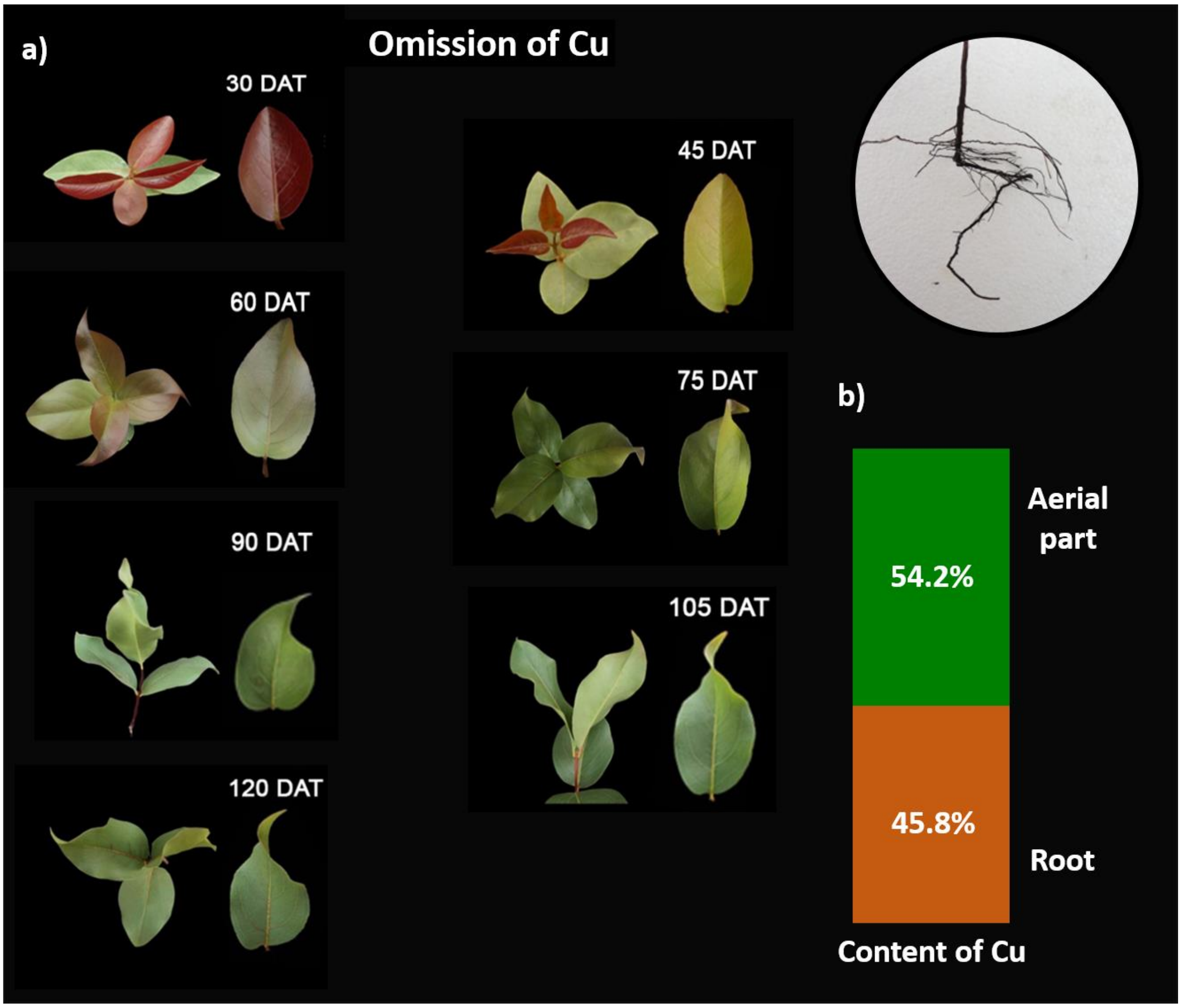
From 45 DAT, symptoms of Cu deficiency became visible with the onset of necrotic spots and purpling in the younger leaves. This purpling started at the leaf apex (farthest from the petiole) and progressed toward the central area of the blade. Starting at 75 DAT, the leaves were also wrinkled (Figure 4a). The roots were dark with few hairs, further decreasing with the intensification of the deficiency from 90 DAT. The Cu content of the roots and shoots of these plants were broadly similar, albeit slightly higher in the root (54.2%; Figure 4b).

Figure 4 – Appearance of the symptoms of Cu deficiency in the leaves and shoots of the *Eugenia dysenterica* DC. seedlings grown in a hydroponic system in a nutrient solution lacking B. Records were acquired at 30, 45, 60, 75, 90, 105, and 120 days after transplantation (DAT) to the nutrient solution (a). Partitioning of Cu in the shoots and roots of the plants treated with nutrient solution lacking Cu (b).



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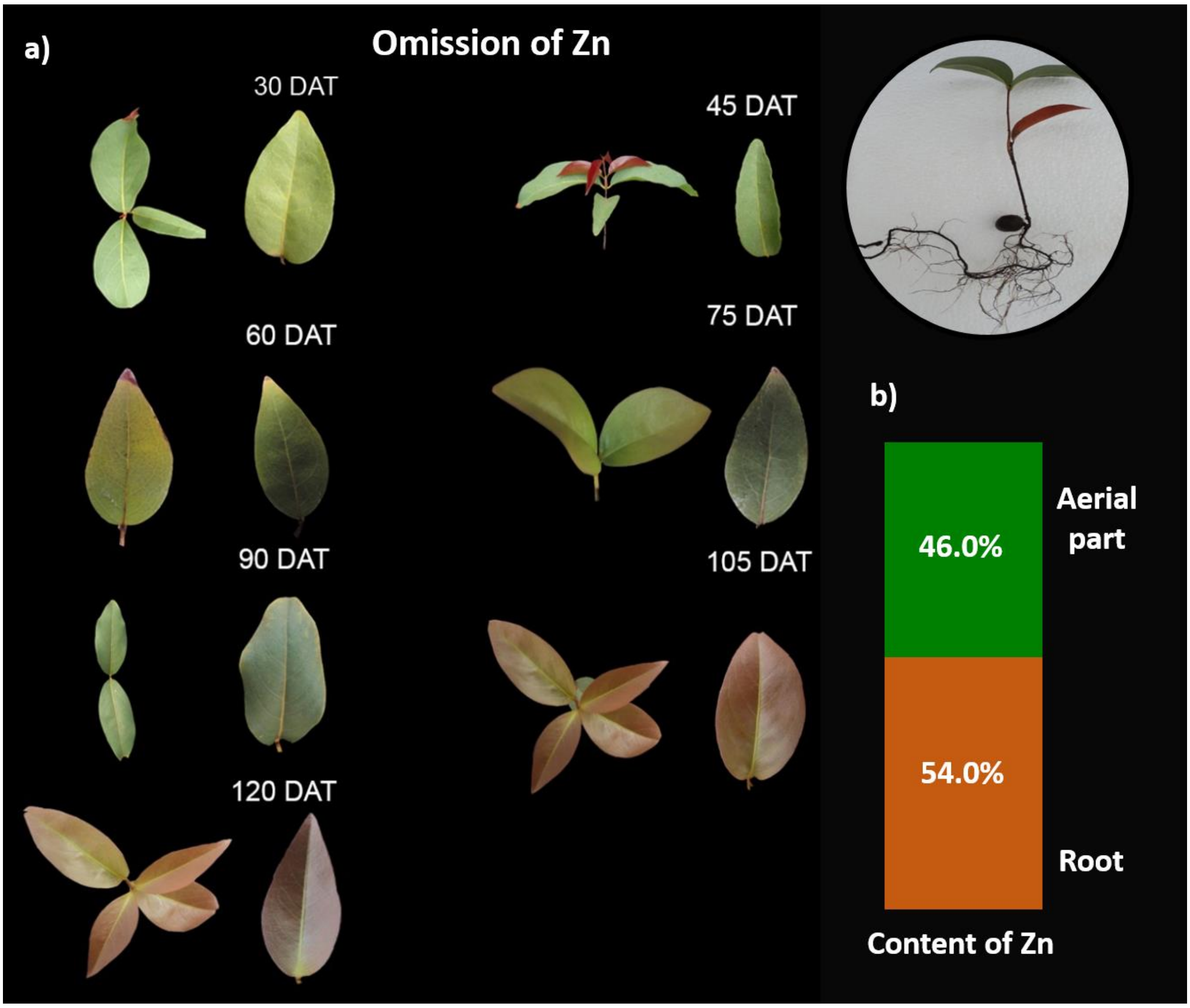
The omission of Zn induced detectable symptoms, specifically chlorosis, starting at 60 DAT. Younger leaves were also less well developed, and a clear difference in the size of younger and older leaves was visible at 90 DAT. At 105 DAT, the leaf chlorosis progressed to purpling (Figure 5a). Root development was not impaired in this treatment, and the roots developed a considerable number of hairs. In this treatment, the Zn accumulated in the tissues was also very similar, with 54.0% in the roots and 46.0% in the shoots (Figure 5b).

Figure 5 – Appearance of the symptoms of Zn deficiency in the leaves and shoots of the *Eugenia dysenterica* DC. seedlings grown in a hydroponic system in a nutrient solution lacking B. Records were acquired at 30, 45, 60, 75, 90, 105, and 120 days after transplantation (DAT) to the nutrient solution (a). Partitioning of Zn in the shoots and roots of the plants treated with nutrient solution lacking Zn (b).



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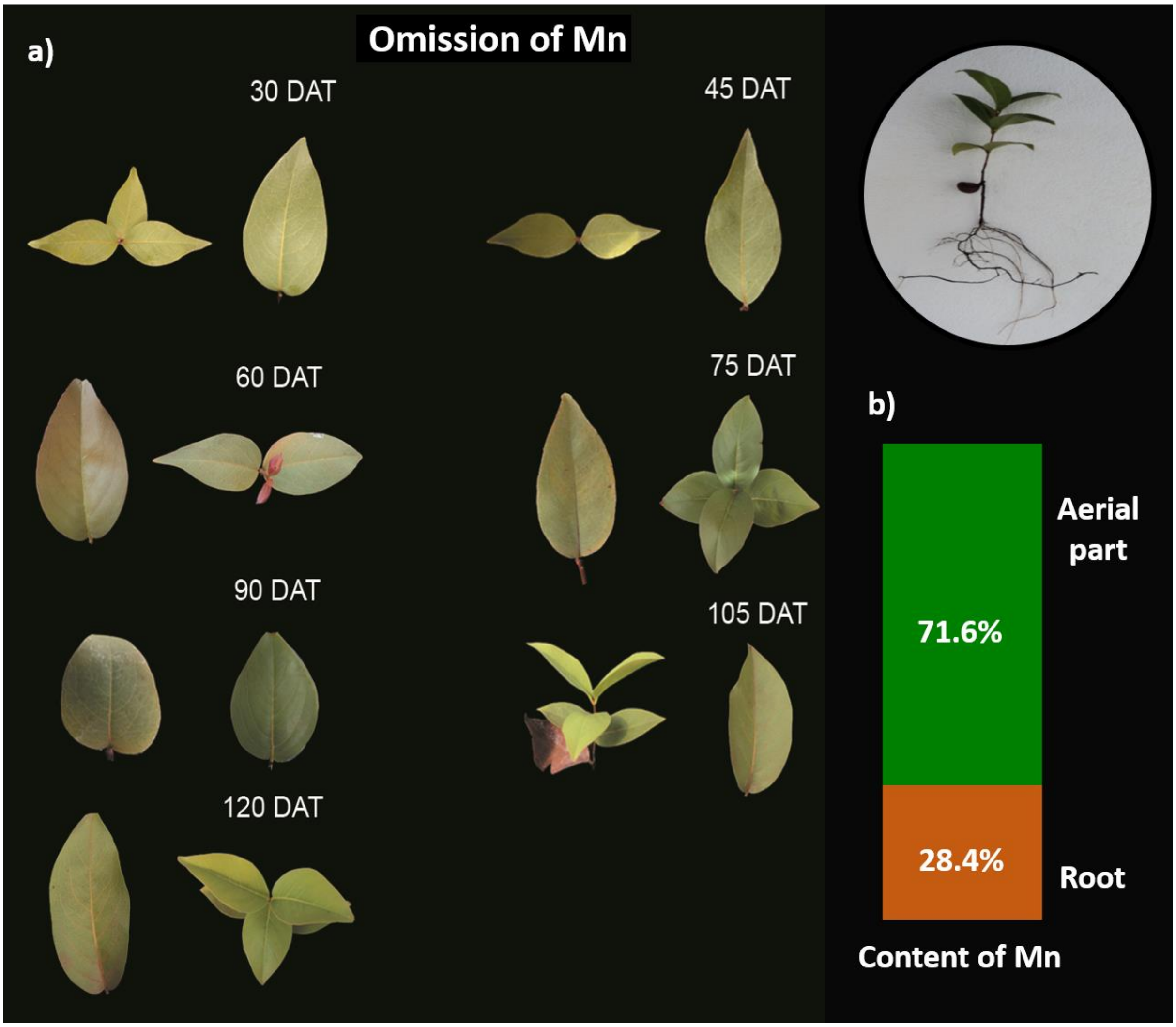
Symptoms of Mn deficiency appeared at 60 DAT, with widespread chlorosis in all new leaves, establishing a type of chlorotic network (Figure 6a). At 90 DAT, this chlorosis progressed to some points of leaf necrosis. The roots were nevertheless similar to those of the plants growing in standard nutrient solution and thus appeared to be unaffected by the lack of Mn. This element accumulated more in the root tissue (71.6%) than the shoots (Figure 6b).

Figure 6 – Appearance of the symptoms of Mn deficiency in the leaves and shoots of the *Eugenia dysenterica* DC. seedlings grown in a hydroponic system in a nutrient solution lacking B. Records were acquired at 30, 45, 60, 75, 90, 105, and 120 days after transplantation (DAT) to the nutrient solution (a). Partitioning of Mn in the shoots and roots of the plants treated with nutrient solution lacking Mn (b).



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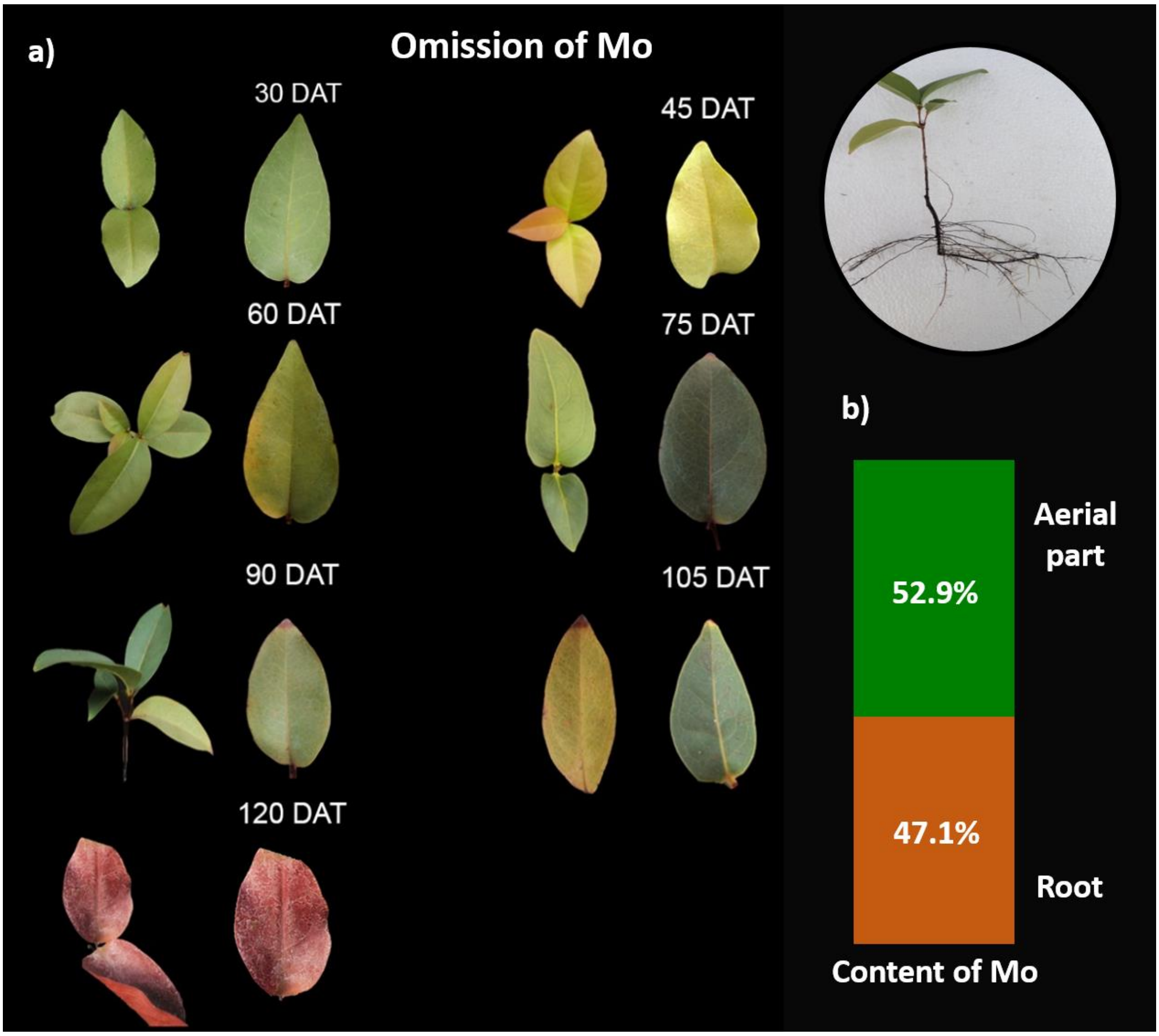
The effects of Mo deficiency became noticeable in the seedlings at 45 DAT, when the leaves, in particular the older ones, were distinctly yellow. As the deficiency progressed, some isolated necrotic points were observed in the leaves (Figure 7a). The development of the roots was not impacted, and there was no change in color, with the aspect being similar to the seedlings cultivated in standard nutrient solution. The plants in this treatment had a similar Mo content in the shoots (52.9%) and roots (47.1%; Figure 7b).

Figure 7 – Appearance of the symptoms of Mo deficiency in the leaves and shoots of the *Eugenia dysenterica* DC. seedlings grown in a hydroponic system in a nutrient solution lacking B. Records were acquired at 30, 45, 60, 75, 90, 105, and 120 days after transplantation (DAT) to the nutrient solution (a). Partitioning of Mo in the shoots and roots of the plants treated with nutrient solution lacking Mo (b).



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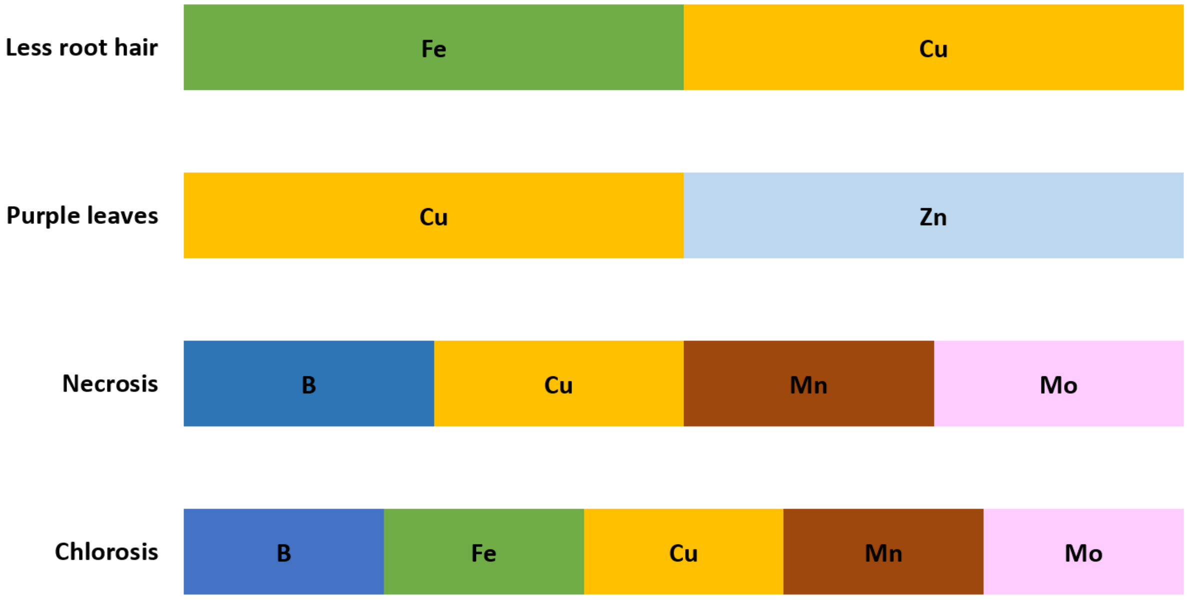
The symptoms of deficiency observed most commonly in the *E. dysenterica* seedlings were chlorosis and necrosis (Figure 8), which were each observed in plants from five of the six treatment groups. Purple leaves were observed only in the plants in the Cu and Zn treatments, while the development of root hairs was affected only by the lack of Fe and Cu.

Figure 8 – Principal symptoms of nutritional deficiency observed in the *Eugenia dysenterica* DC. seedlings grown in a hydroponic system using nutrient solutions lacking B, Cu, Fe, Mn, Zn, and Mo. The tables outline the nutrient deficiency treatment in which the symptoms were assessed during seedling development.



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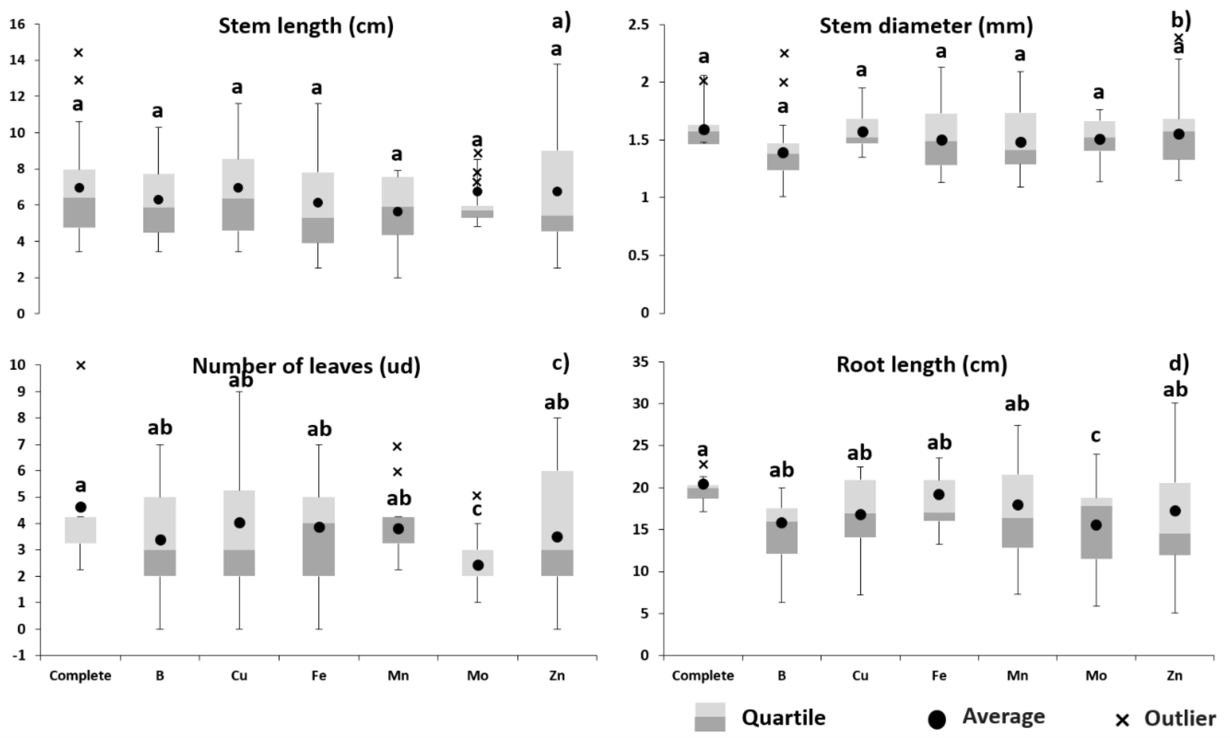
Plant growth

The lack of micronutrients in the nutrient solution had no effect on the stem development of the *E. dysenterica* seedlings. At 120 DAT, the plants in the different treatments had a mean stem length and diameter similar to the means observed in plants grown in the standard nutrient solution (Figures 9a and b). However, the number of leaves and root length were affected by nutrient deficiency. The largest mean number of leaves (4.6 units) was observed in plants grown in the standard nutrient solution, and the Mo treatment had the strongest effect on leaf development, with a mean of only 2.4 leaf units (Figure 9c). Root length presented similar results. The shortest mean root length (15.54 cm) was recorded in the plants in the Mo treatment, while plants grown in the standard nutrient solution had a mean root length of 20.45 cm (Figure 9d).

Figure 9 – Effects of micronutrient deficiency on stem length (cm) and diameter (mm), the number of leaves and the root length (cm) of *Eugenia dysenterica* DC. seedlings grown in a hydroponic system with standard nutrient solution or in treatment solutions lacking B, Cu, Fe, Mn, Mo or Zn. Data collected at 120 days after transplantation (DAT). *Different letters above the black dots indicate significant differences ($p < 0.05$) between treatments, based on the Tukey test.



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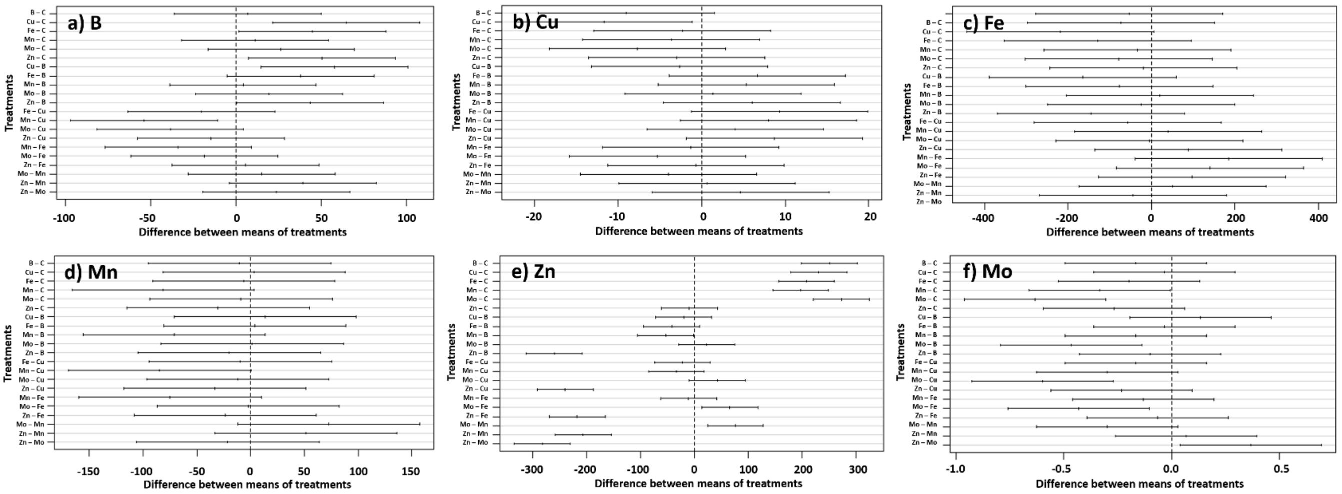
Micronutrient contente

Shoots

The highest B content (170.00 mg.kg^{-1}) was recorded in the shoots of the plants in the Cu treatment, and this value exceeded significantly that recorded in plants grown in both the standard nutrient solution (105.00 mg.kg^{-1}; p = 0.0021) and in the B treatment (112.00 mg.kg^{-1}, p = 0.0058; Figure 10a). The Cu content of the shoots was affected significantly by the standard nutrient solution ($p < 0.001$). The mean Cu content recorded in this treatment (32.00 mg.kg^{-1}) differed significantly from the mean value of the plants in the Cu treatment (21.00

mg.kg^{-1}; p = 0.0261; Figure 10b). The treatments had no effect on the concentrations of Fe and Mn in the *E. dysenterica* seedlings in the hydroponic cultures sampled at 120 DAT (Figure 10c and d). However, the concentration of Zn in the shoots was affected significantly by the treatments ($p < 0.001$). The highest mean concentration of Zn was observed in the Mo treatment (309.00 mg.kg^{-1}). This mean was significantly higher than those recorded in the plants grown in the standard nutrient solution and in the solutions without Zn, Mn or Fe, with means of 36.33 mg.kg^{-1} ($p < 0.0001$), 26.33 mg.kg^{-1} ($p < 0.0001$), 233.00 mg.kg^{-1} ($p < 0.0028$), and 244.00 mg.kg^{-1} ($p < 0.0105$), respectively (Figure 10e). The treatments also had a effect on the Mo content of the shoots ($p < 0.001$). The highest mean Mo content (0.90 mg.kg^{-1}) was found in the seedlings raised with standard nutrient solution, which was significantly higher than the means recorded in Mn (0.56 mg.kg^{-1}; p = 0.0449) and Mo treatments (0.26 mg.kg^{-1}, p = 0.0001; Figure 10f). The micronutrients accumulated in the seedling shoots in the following (decreasing) order: Fe>Mn>B>Zn>Cu>Mo.

Figure 10 – Results of the Tukey test ($p < 0.05$) comparing the micronutrient content (B, Cu, Fe, Mn, Zn, and Mo) of the shoots of *Eugenia dysenterica* DC. seedlings grown in a hydroponic system in either standard nutrient solution or treatment solutions lacking one micronutrient (B, Cu, Fe, Mn, Zn or Mo). Data collected at 120 days after transplantation (DAT).



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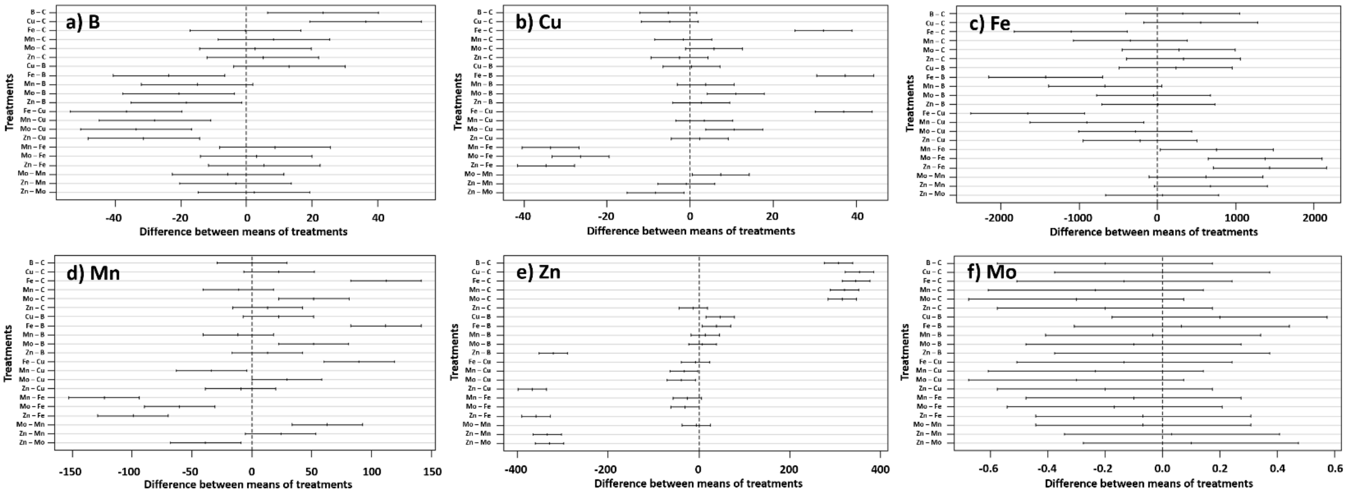
Roots

Significant variation was found in mean root B content in the *E. dysenterica* seedlings sampled at 120 DAT ($p < 0.001$). The highest mean concentration of this nutrient in the roots was observed in the Cu treatment (151.33 mg.kg^{-1}), and was significantly higher than the B root concentrations in all other treatments, except for the B treatment (138.30 mg.kg^{-1}; $p = 0.1934$; Figure 11a). The root Cu content was also significantly affected by the different treatments ($p < 0.001$). The highest mean root Cu content (59 mg.kg^{-1}) was observed in the plants of the Fe treatment, and this value was significantly higher than those recorded in all other treatments (Figure 11b).

Figure 11 – Results of the Tukey test ($p < 0.05$) comparing the micronutrient content (B, Cu, Fe, Mn, Zn, and Mo) of the roots of *Eugenia dysenterica* DC. seedlings grown in a hydroponic system in either standard nutrient solution or treatment solutions lacking one micronutrient (B, Cu, Fe, Mn, Zn or Mo). Data collected at 120 days after transplantation (DAT).



Source: Authors.

The root Fe concentration was also affected significantly by the treatments ($p < 0.001$). As expected, the lowest Fe content was observed in the plants of the Fe treatment (472.33 mg.kg^{-1}), and this value was lower than those recorded in all other treatments (Figure 11c). The Mn content was also affected significantly by the treatments ($p < 0.001$). The highest mean concentration (163.66 mg.kg^{-1}) of this nutrient was observed in the Fe treatment, and this Mn content was higher than those recorded in all the other treatments (Figure 11d).

The treatments also significantly affected the root Zn content ($p < 0.001$). The highest mean value of this nutrient was found in the plants of the Cu treatment (396.66 mg.kg^{-1}), although it was not significantly different from the value recorded in the Fe treatment (163.66

$mg.kg^{-1}$, p = 0.9703; Figure 11e). By contrast, very low Zn values were recorded in the plants grown in the standard nutrient solution (42.66 $mg.kg^{-1}$) and in the Zn treatment (30.00 $mg.kg^{-1}$), which did not differ significantly (p = 0.7988). These values were significantly lower than those recorded in the other treatments, however. No significant variation was found in the root Mo content of the plants in the different treatments (Figure 11f). Micronutrients accumulated in the seedling roots in the following (decreasing) order: Fe>B>Mn>Zn>Cu>Mo.

4. Discussion

The present study provides the first systematic insights into some of the visual characteristics that may be used to detect nutrient deficiencies in *E. dysenterica* seedlings. Purple leaves, for example, appear to indicate a deficiency of Cu or Zn. The accumulation of flavonoids, in particular red/purple anthocyanins, in leaf tissue can be used as an indicator of changes in plant metabolism derived from nutrient deficiency (Stewart et al., 2001). Pourcel et al. (2013) concluded that anthocyanins are key regulators of stress responses and plant development, and Asad et al. (2015) demonstrated that the accumulation of anthocyanin in the leaves of *Noccaea caerulescens* plants decreases with increasing Zn availability.

The results of the present study indicated that the growth parameters of *E. dysenterica* plants are affected primarily by limitations in the Mo content. This micronutrient affected both the number of leaves and root length, which may be explained by the relationship between this micronutrient and enzymes of the N uptake pathway. Molybdenum is present in the structure of the enzymes nitrate reductase and nitrogenase. Given this, a deficiency of Mo reduces the biological fixation of N and, therefore, overall plant productivity (Moraes et al., 2008). Molybdenum is a cofactor in the active centre of the enzymes that catalyse steps of the metabolism of not only N, but also C and S. This micronutrient is thus essential for plant growth under varying environmental conditions (Tejada-Jimenez et al., 2018). In *Hancornia speciosa*, another fruiting tree species native to the Cerrado, a lack of Mo was also found to be critical for growth, and contributed to early leaf abscission (Bessa et al., 2013a).

The elements B, Cu, Zn, and Mo accumulated differentially in the *E. dysenterica* shoots, most likely due to competition or inhibition effects between these nutrients because the presence of one may affect the uptake of the others by the plant, due to the competition for the same absorption site (Epstein and Bloom, 2006). In many cases, the concentrations of the nutrient in the shoots and roots were higher in the treatment plants than those of the control, indicating the adjustment of the metabolism of the plant to adapt to the stress caused by the deficiency of the nutrient. Plants may develop physiological and even molecular responses to adapt to the lack of a nutrient (Kulcheski et al., 2015), increasing the uptake of one ion in the absence of another. The understanding of these adaptive mechanisms should support the development of effective tools for the improvement of seedling cultivation practices.

The micronutrient that accumulated most in both shoots and roots was Fe, which is an essential element for plant production. Vert et al. (2002) found genetic evidence that the transporter IRT1 is essential for the uptake of Fe from the soil in *Arabidopsis*. A knockout mutant of this transporter was chlorotic, leading to serious growth problems. Along with other metals, such as Mg and Mn, Fe plays a key role in CO_2 fixation through photosynthetic activity, and is an important factor ensuring electron flow in the PSII/PSI proteins (Briat et al., 2015; Connorton et al., 2017). In a study of *E. dysenterica* seedlings, Bessa et al. (2016) reported that this micronutrient had accumulated most in the plant tissues at 180 DAT, as observed by Bessa et al. (2013b) in *Anacardium othonianum* Rizz.

The highest mean concentration of boron was recorded in the plants grown in the nutrient solution that lacked Cu. This may have resulted from the attempt of the plants to ensure homeostasis in the concentration of metals in their tissues. A number of different cellular organelles, such as the chloroplasts and mitochondria, depend on Fe, Cu, Mn, and Zn for their functioning, and deficiencies of these minerals, and in particular, toxic concentrations, may disrupt significantly these functions, ultimately hindering plant growth and development (Bashir et al., 2011; Duy et al., 2011; Vigani et al., 2013; Bashir et al., 2016). Given this, many plants attempt to prevent metal toxicity by acquiring other micronutrients.

At 180 DAT, Bessa et al. (2016) observed a negative correlation between the B content and the stem length and number of leaves in *E. dysenterica* seedlings. In the present study, however, the absence of this micronutrient had no effect on any of the growth parameters analysed. Boron appears to be toxic in this species, even at low concentrations. Notwithstanding, further studies are necessary to improve the understanding of the effects of different B concentrations on *E. dysenterica* growth and metabolism.

5. Final Considerations

To produce healthier *E. dysenterica* seedlings, producers should focus primarily on the supply of boron, given that this plant is poorly tolerant of deficiencies in this micronutrient. Symptoms of chlorosis and necrosis are are commonly observed in plants of this species under micronutrient deficiency, and these features can be used by the growers of seedlings as a reliable indicator of reduced access to nutrients. Producers should also consider the supply of iron, given that the seedlings raised on the standard nutrient solution accumulated the highest concentrations of this micronutrient, in both the roots and the aerial parts. This emphasises the importance of the availability of Fe in the initial stages of development of the *E. dysenterica* seedlings, and indicates that supplementation of this nutrient may be an effective strategy for the improvement of productivity.

This work brings the perspective that future works, focused on the mineral nutrition of *E. dysenterica*, are mainly concerned with elucidating which are the ideal concentrations of B and Fe necessary for the development of seedlings of this species. In this way, unnecessary expenses with fertilizers, as well as environmental damage caused by the excessive application of nutrients in the culture of the cagaita can be avoided.

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