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Manganese and fluorine suppress bacterial blight on coffee seedlings grown in a nutrient solution

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ABSTRACT

The use of manganese (Mn) and fluorine (F) in the management of bacterial blight were evaluated in coffee seedlings grown in a nutrient solution. The experiment was carried out with the cultivar Catuaí Vermelho IAC 99. The treatments consisted of the combination of five doses of Mn with five of F, applied via leaf, using Mn sulfate and sodium fluoride, in a 5x5 factorial scheme. The plants were inoculated with bacterial suspension seven days after foliar application of F and Mn doses. The incidence and severity assessments were performed at an interval of 24 hours for 10 days. Photosynthetic activity was assessed using the infrared gas analyzer. Stomatal conductance, photosynthesis, transpiration, PAR radiation and internal CO_2 were analyzed. The chlorophyll content was calculated indirectly. The leaf analysis was performed by digestion in HNO_3 to determine the levels of Mn. Variables such as Area Under Incidence Disease Progress (AUIDP), Area Under Severity Disease Progress (AUSDP), chlorophyll a, b, and total concentrations and photosynthesis were submitted to the Shapiro-Wilk test. The treatment means were subjected to linear regression analysis. Data were analyzed using software R. There was a significant interaction ($P < 0.05$) between the concentrations of Mn and F for the AUIDP and (AUSDP). Doses between 0.7 and 1.4 g L^{-1} of Mn combined with doses of 0.10 to 0.12 g L^{-1} of F were more effective in suppressing the bacterial blight, after analysis for both variables. The increase in Mn concentrations in leaves reduced liquid photosynthesis. The interaction between Mn and F suppressed the bacterial blight intensity of the coffee plants in nutrient solution.

Key words: *Coffea arabica* L.; Mineral nutrition; Epidemiology; Superphosphate simple; Micronutrients.

1 INTRODUCTION

Coffee cultivation, processing, marketing, transportation and market value have propelled business growth across the world, besides offering upwards of 25 million jobs globally. Brazil alone accounted for 32.11% of the 169,9 million bags produced worldwide in 2020, (Conselho dos exportadores de café do Brasil, Cecafe 2021).

Bacterial blight caused by *Pseudomonas syringae* pv. *garcae* (Young et al., 1978) is one of the main bacterial diseases of coffee. It occurs mainly in crops implanted in regions of altitudes above 1000 meters, exposed to cold winds (Ito et al., 2008). Symptoms in these conditions can be necrotic leaf spots, with a large yellow halo, hence the name of the disease. In addition, death of plagiotropic branches and even the orthotropic, necrotic flowers and new fruits, causing great losses in productivity (Pozza; Carvalho; Chalfoun, 2010). In the nursery, due to sprinkler irrigation, constant nitrogen fertilization, temperatures between 18 and 24ºC and density of seedlings can be more intense, reaching up to 100% of seedlings (Belan et al., 2016).

Due to these losses, producers, from the nursery to the field, mainly use spraying cupric products and antibiotics to control the bacterial blight (Zoccoli; Takatsu; Uesugi, 2011); Rodrigues et al. (2013). However, mainly, the use of antibiotics, with specific mode of action, must be cautious, to avoid the selection of resistant populations (Pozza; Pozza, 2017). As an alternative measure, cultural management through balanced mineral nutrition stands out (Elmer; Datnoff, 2014). According to Marschner (2012), mineral nutrients influence the anatomy and chemical composition of plant cells by acting directly on the biosynthesis of phenols, lignins, quinones, tannins, flavonoids, among other compounds related to plant defense, which can increase or reduce disease resistance.

The use of macronutrients in reducing coffee diseases intensity has already been observed for cercosporiosis or brown eye spot (Garcia Júnior et al., 2003); Pozza et al. (2001), phoma leaf spot (Lima et al., 2010) and rust (Santos et al., 2008; Talamini et al., 2003). For the bacterial blight, Pérez et al. (2017) found a reduction in the area under the severity progress curve supplying nitrogen doses between 7 to 13 mmol L^{-1} in nutrient solution. However, the use of micronutrients for this pathosystem is still unknown.

Manganese (Mn) is one of the most studied micronutrients, due to its importance in the metabolic routes related to the resistance of plants to pathogens, both in the leaf and in the root system (Thompson; Huber, 2007). Mn regulates the biosynthesis of lignin and suberin through enzymatic

activation in the route of shikimic acid and phenylpropanoids (Xu et al., 2012). Both compounds are phenolic polymers resistant to enzymatic degradation, and therefore, considered physical barriers against the infection of pathogens. Dordas (2008) highlighted the contribution of these polymers to the reduction of downy mildew and take-all of wheat caused by *Gaeumanomyces graminis* (Sacc.). The authors also verified the effect of Mn in increasing resistance to diseases caused by *Fusarium* spp. and *Sclerotinia sclerotiorum* (Lib. De Bary) in potatoes and cotton, respectively (Dordas, 2008).

On the other hand, fluoride is not considered essential or beneficial to plants. However, the element can act as a bactericidal agent, as has been used to control dental caries caused by *Streptococcus mutans* (Clarke), among other pathogens of bacterial etiology (Kanduti; Sterbenk; Artnik, 2016). According to Breaker (2012), fluoride concentrations between 1,000 and 10,000 mg L^{-1} are potentially toxic to bacteria. This dose inhibits the phosphoryl transferase enzyme, essential for producing energy and synthesizing nucleic acids. The effect of fluoride on host-pathogen relationships remains largely untapped. Some plant species can accumulate fluoride in concentrations closer to 4.000 mg L^{-1} without showing symptoms of toxicity like *Eurya emarginata* (Thunb) while others are extremely sensitive to doses under 20 mg L^{-1} like *Tibouchina urvilleana* (DC) (Jha; Nayak; Sharma, 2009). Thus, the use of fluoride may be a viable alternative in the management of bacterial diseases in plants (Mikkonen et al., 2018).

In view of the above and due to the worldwide need for environmentally correct management measures, to reduce diseases in crops, it is necessary to use alternative methods aiming at the sustainability of coffee cultivation. Thus, the objective was to evaluate combined increasing doses of Mn and F in a foliar spray for the management of bacterial blight spots in coffee seedlings grown in a nutrient solution.

2 MATERIAL AND METHODS

2.1 Experimental site

The present study was carried out at Phytopathology Department (Departamento de Fitopatologia - DFP) at the Federal University of Lavras (Lavras, Minas Gerais (MG), Brazil).

2.2 Experimental design and treatments

The experiment was carried out in a nutritive solution, repeated twice, in a plant growth chamber under controlled conditions, at a temperature of 22 °C ± 2 °C, a photoperiod of 12 hours, maintained with fluorescent lamps of 40 watts and relative humidity above 70%, using 2-liter nebulizers, with an evaporation rate of 25 m $hour^{-1}$. The maximum, average and minimum temperature and relative humidity were monitored by an HT-500 Instrutherm® datalogger, located close to seedlings canopy.

Treatments consisted of five Mn doses 0.7; 1.4; 2.8; and 5.6 g L^{-1} combined with five F doses 0; 0.01875; 0.0375; 0.075; and 0.15 g L^{-1} sprayed on the leaves, in a 5 x 5 factorial scheme of analysis of variance, totaling 25 treatments. The experimental design was in randomized blocks, with four replicates, each repetition consisting of a 2.6 liter pot containing two seedlings.

The nutrient solutions were prepared with the basic solution of Hoagland and Arnon (1950) and the doses of Mn and F applied on the leaves, using Mn sulfate ($MnSO_4$) and sodium fluoride (NaF) as sources of these elements. The sources of macronutrients used were NH_4NO_3, $Ca(NO_3)_2.4H_2O$, KNO_3, KCl, KH_2PO_4, $MgSO_4.7H_2O$ and $CaCl_2.6H_2O$. The micronutrients used to compose the stock solution were $ZnSO_4.7H_2O$ (0,22 mL L^{-1}), $MnSO_4.4H_2O$ (3,0 mL L^{-1}), $CuSO_4.5H_2O$ (0,08 mL L^{-1}), $H_2MoO_4.H_2O$ (0,02 mL L^{-1}) e 1,0 mL L^{-1} of Fe-EDTA Solution. Two treatments were applied via foliar spray using a hand spray, with an interval of seven days between applications at the seedling stage with 3 pairs of leaves.

Aeration of the solution in the vessels was provided continuously by an air compressor connected to the 2.6-liter pots through hoses. The pH of the solution was maintained between 5.5 and 6.0 with the addition of 0.1 mol L^{-1} HCl or 0.1 mol L^{-1} NaOH, monitoring it weekly with a pocket digital pH meter. When necessary, the volume of water in the vessels was supplemented with deionized water.

2.3 Obtaining seedlings and acclimatization

Coffee seeds of the cultivar Catuaí Vermelho IAC 99, susceptible to *Pseudomonas syringae* pv. *garcae* (Sera; Sera; Fazuoli, 2017), were washed in water, disinfected with 50% alcohol for 50 seconds and 1% sodium hypochlorite for one minute and rinsed with sterile distilled water. Subsequently, they were sown in plastic trays (0.50 x 0.35 x 0.15 m) containing washed sand. Then, these trays were taken to a plant growth chamber with a temperature of 28 °C ± 2 ºC and a 12-hour photoperiod. After the emission of the pair of cotyledon leaves, the seedlings were watered with a basic solution by Hoagland and Arnon (1950) at 20% of the ionic strength. This solution was applied every ten days until the seedlings emitted the first pair of definitive leaves.

After 25 days, seedlings were transferred to 2.6-liter pots containing Hoagland's basic solution at 50% ionic strength to acclimatize in aqueous conditions. Seedlings remained in this solution for another 20 days. After that period 200 seedlings were selected according to size uniformity and the solution was replaced to 100% of the ionic strength.

Ion depletion of the nutrient solution was monitored weekly with the compaction meter for K^+ (Horiba-CARDY®). When depletion reached 30% of the initial value of K^+ ions, the nutrient solution was changed in the different treatments.

2.4 Obtaining inoculum and inoculation

The reference isolate CFPB1634 of *P. syringae* pv. *garcae* souche pathotype was used to prepare the bacterial suspension. Thus, colonies of the bacterium were grown in test tubes containing MB1 medium. After 48 hours, the bacterial suspension was prepared in saline (0.85% NaCl), sterilized and adjusted to 5.1 x 109 CFU mL^{-1} (absorbance 0.8) in a spectrophotometer at 600 nm (OD600), as described by Oliveira and Romeiro (1990).

Plants were inoculated seven days after foliar application of F and Mn doses. To increase the probability of successful inoculation, plants were previously kept in a humid chamber for 72 hours. For this, transparent plastic bags were sprayed inside with distilled water and moistened cotton balls were placed over the lids in each 2.6-liter pot and kept in a plant growth chamber as described above.

A bacterial suspension was sprayed on the leaf's abaxial surface and after 72 hours in a humid chamber, kept in the same conditions described above, the plastic bags were removed.

2.5 Disease assessments

When the first symptoms of bacterial blight were observed, incidence and severity assessments were carried out every 24 hours for 10 days on the first and second pairs of newer fully developed leaves, totaling four leaves per seedling and eight leaves per pot. Incidence was evaluated according to the percentage of infected leaves (Equation 1) and the severity using the scale proposed by Belan et al. (2014), being: 1 - from 0 to 0.99% severity; 2 - from 1.0 to 2.0% severity; 3 - from 2.01 to 4.0% severity; 4 - from 4.01 to 8.0% severity; 5 - from 8.01 to 16.0% severity; 6 - from 16.01 to 25.0% severity; 7 - from 25.01 to 45.0% severity and 8 -> 45.1%.

$$I(\%) = \left(\frac{Nf}{Nt}\right) * 100 \quad (1)$$

On what:
Nf: number of damaged leaves
Nt: total number of leaves evaluated from the host

Then, the mean incidence and severity of the disease per plot was obtained. And plots of disease incidence and severity over time were plotted.

2.6 Area under the disease progress curve

The area under disease progress curve (AUDPC), for incidence (AUIPC), and severity (AUSPC) was calculated according to Equation 2 (Shanner; Finney, 1977)

$$AUDPC = \sum_{i=1}^{n-1} \frac{(Y_i + Y_{i+1})}{2} * (T_{i-1} - T_i) \quad (2)$$

On what:
AUDPC = area under the disease progress curve for incidence severity (AUSPC) or (AUIPC) ;
Yi = proportion of disease in the i-th observation;
Ti = time, in days, in the i-th observation;
n = total number of observations

2.6 Analysis of leaf manganese, photosynthesis and chlorophyll content

Photosynthetic activity was evaluated on two fully expanded leaves from the first definitive pair, to obtain averages, using the infrared gas analyzer (IRGA) (LI-6400XT Portable Photosynthesis System, LI-COR, Lincoln, USA) at 15 days after inoculation. The readings were made with a photosynthetically active radiation source (PAR) in a closed chamber fixed in 600 µmol of photons m^{-2} s^{-1} (Blue $^+$ Red LED LI-6400-02B, LI-COR, Lincoln, USA). The analyzed variables were stomatal conductance, photosynthesis, transpiration, PAR radiation and internal CO_2.

The chlorophyll content was calculated indirectly, from the reading made with the SPAD-502® portable chlorophyll meter (Matsumoto et al., 2008) performed 3 days after inoculation, on the four leaves, with four measurements per leaf. The average per plant of these 16 evaluations was used to perform the statistical analysis.

After ending of the evaluations, the plants were cut and the leaves, stems and roots were washed with distilled water, packed in paper bags and dried in an oven, at 60 ºC, until they reached constant weight. The leaves of each treatment were ground and taken for leaf analysis by digestion in HNO_3 to determine the levels of Mne.

2.7 Statistical analysis

Experiments were compared by joint analysis of the data over time. The variables area under incidence disease progress, area under severity disease progress, the levels of chlorophyll a, b and variables analyzed for photosynthesis were submitted to the Shapiro-Wilk test to verify the assumptions of analysis of variance (ANOVA) For AUSDP, the transformation to log10 (Y + 1) was necessary in order to stabilize the variance of the residues. Treatment means were compared by F test ($p < 0.05$) and the significant quantitative variables were subjected to adjust of linear models by regression analysis. All data were

analyzed using the software R (R DEVELOPMENT CORE TEAM, 2020).

3 RESULTS

There was no difference between the experiments conducted over time in the joint analysis of the data of the analyzed variables ($p > 0.05$). Therefore, the data below refer to the average of these variables in the two experiments.

3.1 Progress in the incidence of bacterial spot

The first symptoms of bacterial blight were observed five days after inoculation, with an increase in the incidence of the disease over time (Figure 1). The lowest incidence (3.12%) was observed when the highest dose, 5.6 g L^{-1} of Mn was applied, combined with 0.15 g L^{-1} of F. In contrast, the highest incidence value (46.87%), occurred in the combination of doses of 2.8 g L^{-1} of Mn and 0 g L^{-1} of F at 11 days after inoculation, remaining constant until the last day of evaluation (Figure 1).

3.2 The area under the incidence progress curve (AUIPC)

There was significant interaction ($p < 0.05$) between the doses of Mn and F for the AUIPC. The lowest AUIPC, of 26.3, was observed between 0.7 to 1.4 g L^{-1} of Mn and 0.10 to 0.12 g L^{-1} of F. The region represented by dark blue is highlighted, ranging from 0 to 100 as having the lowest AUIPC (Figure 2). Thus, in any combination of doses of Mn and F within that region, the lowest values for this variable are found. However, the dose previously mentioned with the lowest AUIPC, of 26.3, was established because it is located in a region far from the areas of greatest risk of greater AUIDP and also because they present a lower expenditure of products for the halo blight management (Figure 2).

3.3 The progress curve of the severity of the bacterial spot

Progress in the severity of the disease was observed over time. The lowest value of severity, 0.05% was observed with the combination of 0.7 g L^{-1} of Mn with 0.15 g L^{-1} of F. The highest severity, of 4.12%, was observed at 17 days after inoculation in the combination of doses of 0 g L^{-1} of Mn and 0.01875 g L^{-1} of F (Figure 3).

3.4 The area under the severity progress curve (AUSPC)

There was a significant interaction ($p < 0.01$) between the concentrations of Mn and F for the variable AUSPC, and the behavior was similar to AUIPC. The doses of Mn between 0.7 and 1.4 g L^{-1} combined with the concentration of 0.08 to 0.12 g L^{-1} of F showed less AUSPC (Figure 4). The highest AUSPC was observed for Mn concentrations between 2.8 and 4 g L^{-1} combined with 0 to 0.02 g L^{-1} of F. The mentioned doses were established following the same criterion for AUIPC.

3.5 Manganese content in the leaves, photosynthesis, and chlorophylls

There was a linear increase ($p < 0.05$) of Mn in the tissues of the aerial part only with the increase of the doses of Mn sprayed through the leaves (Figure 5).

There was no significant interaction ($p > 0.05$) between the doses of Mn and F in liquid photosynthesis and chlorophylls in leaves of coffee seedlings. However, the increase in Mn doses reduced linearly the photosynthesis ($p < 0.01$). The reduction was from 9.59 to 9.23 µmol m^{-2} s^{-1} corresponding to the increase in Mn doses from 0 to 5.6 g L^{-1}, respectively (Figure 6).

4 DISCUSSION

Nutrition for coffee is necessary for healthy growth and plant resistance to pathogens. The adequate nutritional balance should always be the first line of defense against plant diseases (Pozza et al., 2015; Pérez et al., 2020). During infection by pathogens, plants produce various defense compounds to form physical and chemical resistance barriers (Agrios, 2005). In the present study, the interaction of Mn and F in reducing the bacterial blight intensity was verified. As the behavior of AUIPC and AUSPC were similar in doses between 0.7 and 1.4 g L^{-1} of Mn combined with the concentration of 0.08 to 0.12 g L^{-1} of F, they were suggested as the most effective in decrease the disease intensity.

Silva et al. 2019 evaluated the relationship between brown eye spot space-time progress and soil fertility and mineral nutrition in irrigated coffee crops and observed a correlation ($p < 0.01$) between the incidence of the disease and leaf nutrients (B, K, and P) varying between 0 to 23% and 0 to 25% depending on the irrigation method of the coffee tree. The authors concluded that the levels of nutrients in the soil and the leaves should be monitored throughout the crop cycle since the variation of the leaf content in the area can directly interfere in the productivity and intensity of the disease, requiring the replacement of plants at specific points.

The effect of the isolated application of Mn doses to reduce diseases in coffee in nutrient solution was found by Pérez et al. (2020). The intermediate dose of 0.50 g L^{-1} of Mn reduced the area under the rust severity progress curve (*Hemileia vastatrix*) by 50% compared to the highest dose of 4.00 g L^{-1} evaluated in the study by these authors. This

intermediate dose of 0.50 g L^{-1} was lower than the range of 0.7 to 1.4 g L^{-1} of the present study, in which there was a greater reduction in the bacterial blight. However, the combined effect should also be considered with doses of F belonging to the evaluated treatment. For other bacterial diseases, in rice (*Xanthomonas oryzae*) (Thompson; Huber, 2007), soybeans (*Pseudomonas syringae* pv. *glycinea*) (Ersek et al., 1985) and tomatoes (*P. syringae*), (Conlin; McCarter, 1983), the beneficial effect of Mn in reducing the diseases severity by these causal agents has also been reported when plants were grown in places with greater micronutrient availability (Huber; Wilhelm, 1988).

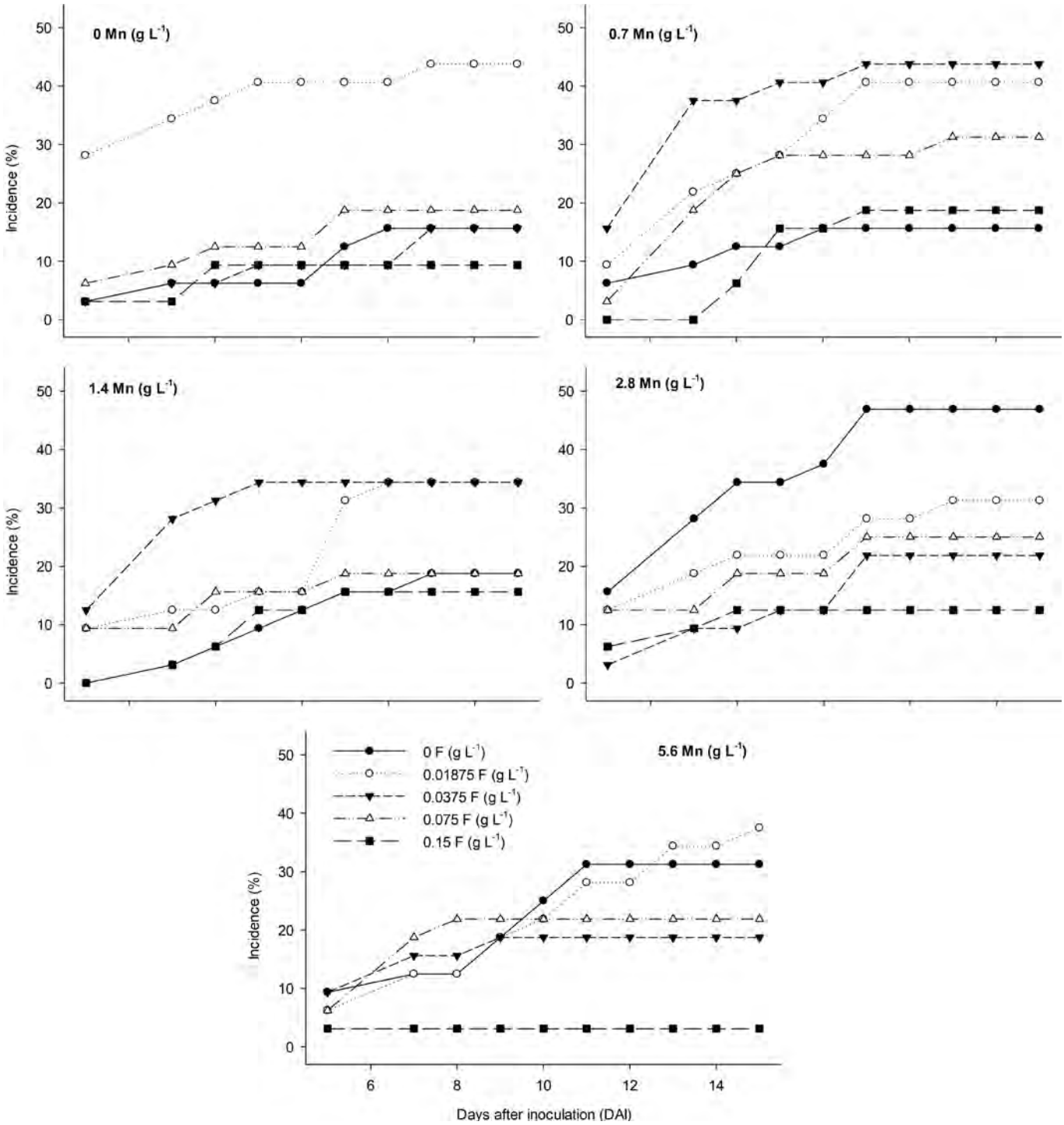

Figure 1: Bacterial blight incidence progress curve (%) of (*Pseudomonas syringae* pv. *garcae*) on coffee (*Coffea arabica*) seedlings leaves, depending on the concentrations of manganese (Mn) and fluorine (F) sprayed on the seedlings.

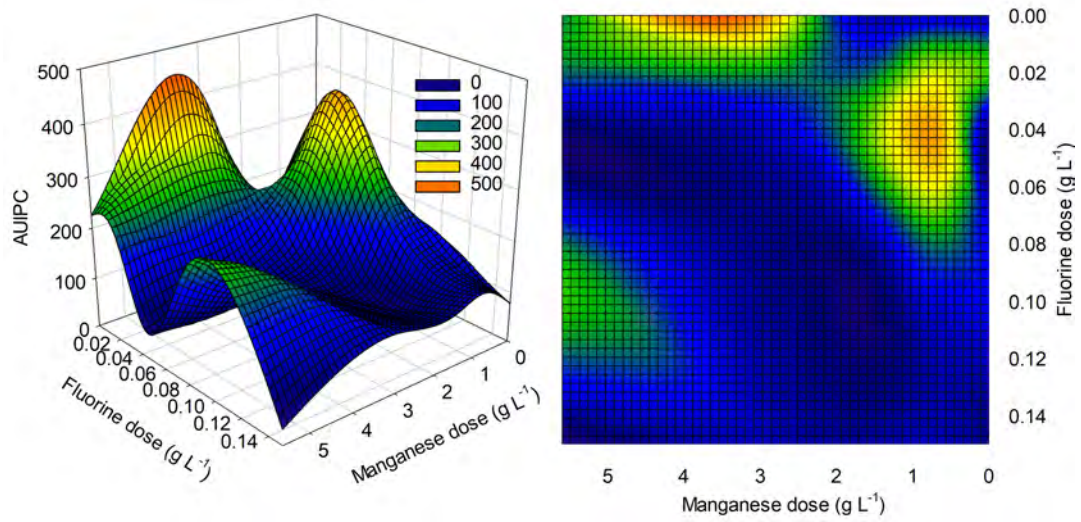

Figure 2: Area under the incidence progress curve (AUIPC) of bacterial blight (*Pseudomonas syringae* pv. *garcae*) on coffee (*Coffea arabica*) seedling leaves, depending on the concentrations of manganese (Mn) and fluorine (F) sprayed on the seedlings.

On the other hand, the intensity of "Brittle leaf disease - BLD" in date palms (*Phoenix dactylifera* L.) is related to Mn deficiency. The influence of this nutrient on BLD was studied by Saidi et al. (2012). According to the authors, Mn concentrations decreased significantly from 1.8 to 1.26g kg^{-1} in leaves and from 2.627 to 2.1g kg^{-1} in dried date palm roots with increased disease severity. Another study showed that the BLD has also been associated with the accumulation of phenolic compounds and a significant reduction in cell wall lignification in infected leaves (Rahmania et al., 2011).

Mn affects different biochemical processes, acting as a cofactor for at least 35 enzymes (Saidi et al., 2013). The influence of this micronutrient on disease intensity has been observed when applied in deficient or excess doses depending on the culture. The results obtained in the present study can be explained by either direct or indirect effects of Mn. Directly, when it regulates the expression of genes related to physical barriers in cells, bacterial infection is consequently limited, or indirectly, by acting on the plant's biochemical defense responses against bacterial tissue colonization, in order to reduce the incidence and severity of the bacterial blight.

Regarding the Mn content found in the leaves, according to the results obtained in this study, it was possible to verify the nutrient absorption in the leaves of the coffee trees proportionally according to the different doses applied, making the ions available in the internal organs of the plants, which are necessary for the biosynthesis of essential molecules and also to activate defense mechanisms against *P. syringae* pv. *garcae* (Graham; Webb, 1991). Mn, as well as Cu, Fe and B are involved in the metabolic route for the biosynthesis of phenolic compounds, including quinones, tannins and flavonoids (Pozza et al., 2001; Taiz; Zeiger, 2004; Silva et al., 2019).

The increase of Mn in plant tissues and its relationship with photosynthesis in several plant species was reported by Millaleo et al. (2013). According to these authors, the decline of photosynthesis is considered as one of the main mechanisms that constitute the toxic effects of Mn excess and is proposed as an early indicator for the toxicity of this micronutrient in tobacco (Nable; Houtz; Cheniae, 1988) and rice (Lidon; Barreiro; Ramalho, 2004). In the present study, there was a reduction in photosynthesis in the leaves of coffee seedlings in response to the increase in doses of Mn from the dose added to the nutrient solution $MnSO_4.4H_2O$ 3,0 µL L^{-1}) by Hoagland and Arnon (1950).

The reduction of photosynthesis influenced by excess of Mn^{+2} ions can be explained due to the mediation in the transfer of energy in the fundamental ATP or ADP molecules in several processes synthesized in the cytoplasm. Mn can replace Mg as a cofactor for most phosphorylative enzymes, forming the bridge between ATP or ADP pyrophosphate and phosphorylase, however, its efficiency is less than that of Mg. As a result of this substitution, the lack of Mg inhibits CO_2 fixation although the concentration of chlorophyll molecules is sufficient. Magnesium is required in photophosphorylation and phosphorylation reactions, limiting the regeneration of rubisco, an enzyme that captures atmospheric carbon dioxide of the air (Nelson, 2018).

Regarding fluoride, the toxic mechanisms of F in different bacterial species and some fungi are very complex and have not been fully understood (Martinez, 2012, Fina, 2016). Marquis et al. (2003) reported that fluoride more efficiently enters bacterial cells at acidic pH values as HF and dissociates when exposed to the more neutral intracellular pH.

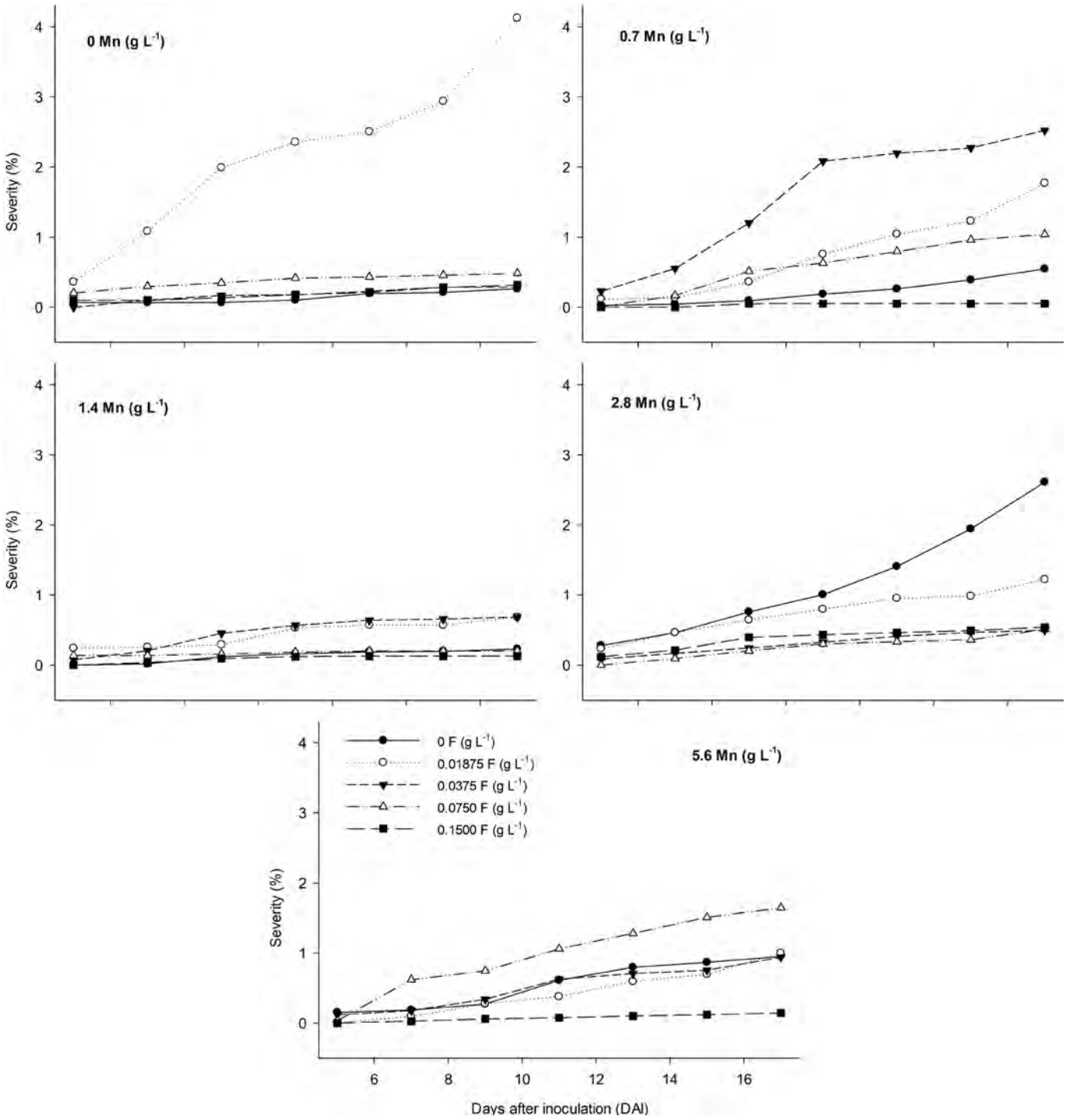

Figure 3: Progress curve of the severity of bacterial blight (*Pseudomonas syringae* pv. *garcae*) on coffee seedlings (*Coffea arabica*) leaves, depending on the concentrations of manganese (Mn) and fluorine (F) sprayed on the seedlings.

For dentistry, the impact of fluoride on oral bacteria is well documented in the literature (Marquis et al., 2003; Wiegand et al., 2007). Oral bacteria are inhibited by fluorine at a concentration in the range of 10–1600 mg L^{-1}. Other studies have reported the inhibition of soil microorganisms by inorganic fluoride, making it difficult to increase the accumulation of organic matter in the soil (Tscherko; Kandeler, 2015). This authors observed that microbial biomass and enzyme activity in the soil decreased substantially at water-extractable fluoride concentrations that exceeded 20 mg g^{-1} of soil. These findings suggest that, if present in sufficient concentration, fluoride can have a negative impact on biological wastewater treatment systems.

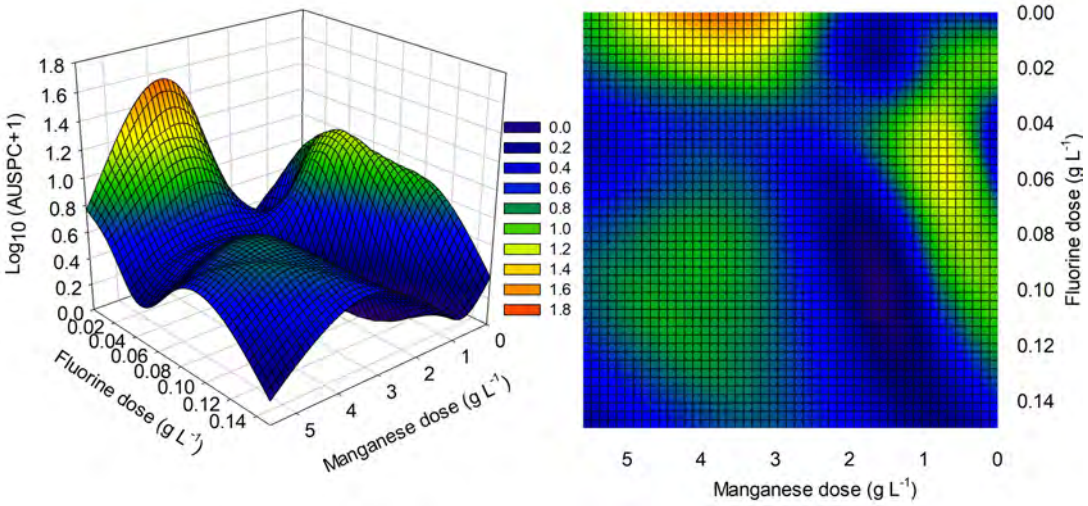

Figure 4: Area under the severity progress curve (AUSPC) of bacterial blight (*Pseudomonas syringae* pv. *garcae*) in coffee (*Coffea arabica*) seedlings leaves, depending on the concentrations of manganese (Mn) and fluorine (F) sprayed on the seedlings.
* Data transformed by log10 (AUSPC + 1).

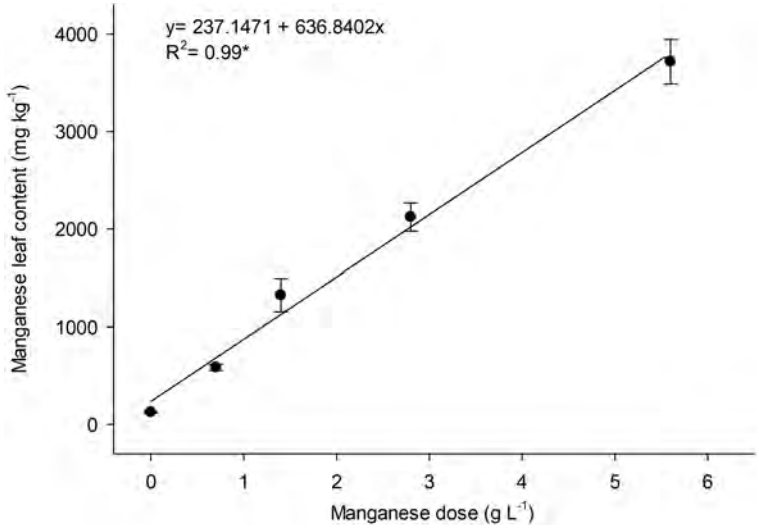

Figure 5: Manganese content (Mn) in the leaves of coffee seedlings (*Coffea arabica)* with bacterial blight (*Pseudomonas syringae* pv. *garcae*) symptons as a function of increasing doses of manganese (Mn) sprayed on the leaves.

In contrast, the toxicity of F in pathogens can be explained by this element interfering in the storage and extracellular synthesis of polysaccharides and by inhibiting the activity of enolase, which in the presence of F forms the magnesium-fluorine-phosphate complex, leading to destabilization in the cellular system environment (Li et al., 2013). The effect of F on translocation and interaction with cations such as Mg^{+2} and Ca^{+2} is also known to be indispensable in the formation of chlorophyll molecules and for the maintenance and stability of the cell wall, respectively (Ren; Rajashankar; Patel, 2012).

According to the results of this research, the correct and balanced application of Mn with F is suggested as a possible alternative management method to bacterial blight in order to reduce dependence on the use of pesticides in coffee growing.

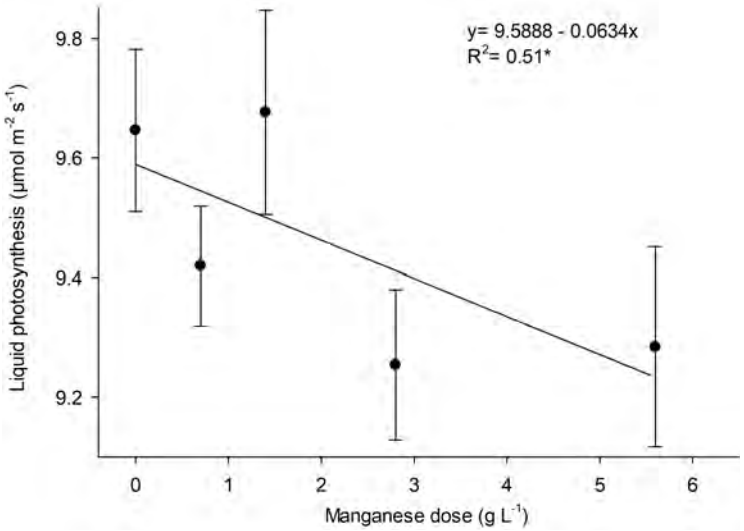

Figure 6: Liquid photosynthesis in coffee seedlings (*Coffea arabica*) with symptoms of bacterial blight (*Pseudomonas syringae* pv. *garcae*) and submitted to different concentrations of manganese (Mn) sprayed on the seedlings.

6 CONCLUSIONS

The interaction between Mn and F suppress the bacterial blight intensity of the coffee plants in nutrient solution. The greatest reduction in both AUIPC and AUSPC was observed in the combination of doses from 0,7 to 1,4 g L^{-1} of Mn and 0,10 to 0,12 g L^{-1} of F, while AUDCP was 5,6 g L^{-1} of Mn and 0,15 g L^{-1}. There was a linear increase in leaf Mn with the increase of this nutrient applied via leaf. However, the increase in Mn doses (from 0 to 5.6 g L^{-1}) linearly reduced liquid photosynthesis.

7 ACKNOWLEDGEMENTS

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