



ISSN: 2230-9926

Available online at <http://www.journalijdr.com>

IJDR

International Journal of Development Research

Vol. 10, Issue, 05, pp. 35995-36000, May, 2020

<https://doi.org/10.37118/ijdr.18929.05.2020>



RESEARCH ARTICLE

OPEN ACCESS

PHYTOMASS AND IRON ACCUMULATION IN GREEN CORN FERTILIZED WITH FERROUS SULFATE

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ARTICLE INFO

Article History:

Rec
Rec
28th
Accepted 11th April, 2020
Published online 30th May, 2020

Key Words:

Foliar fertilization, Iron biofortification, Nutritional quality, *Zea mays* L.

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Jackson de Mesquita Alves,

ABSTRACT

The low availability of cationic micronutrients such as iron (Fe) in alkaline soils can limit the production of : the
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design, with five Fe concentration (0, 1, 2, 3 and 4 g L⁻¹) and four blocks totaling 20 experimental plots. At 78 days after planting, the dry mass of leaves, stems, straw, ears and green grains, as well as Fe concentration and accumulation in leaf, green grains tissues were evaluated. The fertilization of corn with ferrous sulfate via leaf in alkaline soil increased the production of dry green corn phytomass, but did not interfere in the dry mass of grains. Iron doses increased Fe concentration and accumulation in leaves and grains of green corn with gains of up to 43.8 and 42.0%, respectively.

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Citation: Flávio Sarmento de Oliveira, Jackson de Mesquita Alves, Francisco de Assis Freitas, 2020. "Phytomass and iron accumulation in green corn fertilized with ferrous sulfate", *International Journal of Development Research*, 10, (05), 35995-36000.

INTRODUCTION

Corn is one of the main cereals currently cultivated worldwide, serving for human food, animal feed and raw materials for industry, standing out for its nutritional qualities (Santos *et al.*, 2020). In Brazil and in the world, one of the challenges in maize culture is to increase not only productivity, but also the nutritional quality of the harvested product, especially for human consumption (Loureiro *et al.*, 2018). Several soils where corn is grown can show an alkaline reaction (pH > 7.0), due to parent material (limestone) and, or to the low degree of weathering (Oliveira *et al.*, 2018). One of the consequences of soil alkalinity is a decrease in the availability of cationic micronutrients such as iron, due to the chemical precipitation reactions that produce stable forms of iron, such as oxides (Mielk *et al.*, 2016) causing deficiency and limiting corn production. Improving the nutritional quality of the harvested product, called biofortification, can be done through

techniques such as genetic improvement and gene manipulation (Garg *et al.*, 2018; Kumar *et al.*, 2019) or through fertilization, called agronomic biofortification (Connorton and Balk, 2019). Agronomic biofortification via soil, seed treatment and foliar application are characterized as techniques of lower cost, more accessible and quick results, as it influences only fertilization and therefore has been the subject of some research (Loureiro *et al.*, 2018; Nikhil, 2018; Connorton and Balk, 2019). In several publications there are reports of the positive effect of fertilizing corn with iron sources in the production of phytomass and grain yield (Mielki *et al.*, 2016; Rui *et al.*, 2016; Yongli *et al.*, 2018; Nikhil, 2018; Filipek-Mazur *et al.*, 2019) and some of them about the use of new sources like iron oxide nanoparticles (Elanchezhiana *et al.*, 2017; Yoon *et al.*, 2019). In soils with alkaline reaction, the use of ferrous sulfate as an iron source can be advantageous due to the oxidation process of Fe²⁺ to Fe³⁺ to generate acidity in the soil (Annisa and Nursyamsi, 2016) and

thus increase the availability of iron and other micronutrients such as zinc and manganese for plants. Researches carried out on semi-arid and alkaline soils is scarce, a fact that justifies the absence of official recommendations for fertilization with micronutrients such as iron, especially for the production of green corn. In this way, the present work is a contribution to the generation of information so that the farmers can produce green corn as a higher nutritional quality in relation to the iron content in the grains. The objective was to evaluate the influence of iron doses via leaf on the production of dry phytomass, the levels and accumulations of iron in green grains and in the leaf tissues of green corn irrigated in alkaline soil.

MATERIAL AND METHODS

An experiment was carried out in a field condition, in a commercial production area, in the municipality of Vieiropolis-Paraíba (PB), region of hinterland, located 06° 31' 47,91" S and 38° 14' 29,42" W, with altitude of 324 m. According to the Classification Köppen (1956), the climate is hot and dry semi-arid, with high temperatures during the day, softening the night, presenting annual variations within a range of 23 to 30° C, with possible higher peaks, mainly during the dry season period. The rainfall regime, in addition to being low, is irregular, with annual averages around 900 mm per year CPRM (2005). The soil of the experimental area was classified as Luvisolo Chromic (Santos *et al.*, 2013). Before the installation of the experiment, the soil was sampled in the 0 - 0.20 m layer for its chemical and physical characterization, according to the methodology contained in Embrapa (1997). The soil analysis results were: $\text{pH}_{\text{H}_2\text{O}} = 7.23$, $\text{Ca}^{2+} = 7.6 \text{ cmol}_c \text{ dm}^{-3}$, $\text{Mg}^{2+} = 3.06 \text{ cmol}_c \text{ dm}^{-3}$, $\text{K}^+ = 0.60 \text{ cmol}_c \text{ dm}^{-3}$, $\text{Na}^+ = 0.70 \text{ cmol}_c \text{ dm}^{-3}$, $\text{Al}^{3+} = 0.00 \text{ cmol}_c \text{ dm}^{-3}$, $\text{H}^+ \text{ Al} = 0.00$, organic matter = 39.50, sand = 567 g kg^{-1} , silt = 166 g kg^{-1} and clay = 267 g kg^{-1} . The experiment was carried out in a randomized block design, with five treatments corresponding to five doses of Fe (0, 1, 2, 3 and 4 g L^{-1}), with four blocks. The experimental plots consisted of three double rows of planting with 5.0 m in length, totaling approximately 100 plants in 30 m^2 of each plot. The useful plots were constituted by the central double row, with 0.5 m of each end neglected. The soil preparation of the experimental area was done through a plowing at 0.20 m depth. The hybrid corn, AG 1051®, was sown directly in the field, under a double row planting system, with 0.40 m spacing within the double row, 0.30 m between plants in the planting line and 2.0 m between the centers of each double row, obtaining an estimated population density of 33,33 plants ha^{-1} .

Sowing was carried out on March 28, 2018, at an average depth of 0.03 m, with two seeds per hole in the planting line, in which, subsequently, thinning was carried out in the vegetative phase "V3" (three leaves definitive), with only a single seedling remaining per hole. Low salinity water (0.1 dS m^{-1}) was used for irrigation of the cultivation area, using a localized irrigation system, pressurized by three-phase electrode, using a single drip tape, model Taldrip®, Naan Dan brand Jain®, nominal diameter of 17 mm, emitters spaced at 0.3 m and flow rate of 1.0 L h^{-1} at 100 KPa of nominal pressure, in the center of each double row. For foliar fertilization, referring to each Fe concentration, commercial ferrous sulfate (20% Fe and 11% S) was used. The applications were made using a manual applicator with 20 L of syrup capacity at a constant pressure of 2.7 kgf.cm^{-2} and a syrup volume of 200 L ha^{-1} in the

phenological phase V6, that is, with six leaves definitive. The planting fertilization consisted of the application of 20 kg of N in the form of urea (45% of N) and 70 kg ha^{-1} of P_2O_5 in the form of simple superphosphate (18% of P_2O_5). The cover fertilization with nitrogen and potassium was carried out following the recommendation of Freire *et al.* (1999), using urea (45% of N) and potassium sulfate (48% of K_2O and 17% of S), respectively, dissolved in irrigation water (fertigation) and injected into the system using venturi-type equipment, totaling 120 kg ha^{-1} of K_2O and 180 kg ha^{-1} of N. The irrigation depth was determined based on the reference evapotranspiration (ET_0) estimates by the method of Hargreaves and Samani (Hargreaves, 1974), with the aid of the system software for estimating evapotranspiration (SEVAP) (Silva *et al.*, 2005). For this purpose, an analog dry bulb thermometer was installed, next to the cultivation area, to record the maximum and minimum daily temperatures during the conduct of the experiment, with the readings obtained at 9:00 a.m. The control of weeds in the experimental area was done within the critical competition period, comprised between the vegetative phases "V3" and "V12" (third and twelfth final leaf) of the corn crop, following the recommendation of Vargas *et al.* (2006). For this purpose, spraying was carried out with 20 L spray equipment at constant pressure, based on the systemic action selective herbicide, Atrazine Nortox500 SC®, with 6-chloro-N²-ethyl-N⁴-isopropyl-1,3,5-triazine-2,4-diamine (Atrazine), 500 g L^{-1} , as active ingredient, in the dosage of 5.0 L ha^{-1} of commercial product (2.5 L ha^{-1} of Atrazine).

The control of the cartridge caterpillar (*Spodoptera frugiperda* JE Smith) was carried out whenever the presence of the first insects in the area was verified, with sprays from the vegetative phase "V3", based on the insecticide Lannate® BR, having the active ingredient S-methyl N-methylcarbamoyloxy thioacetimidate (Metomil), 215 g L^{-1} , in the dosage of 0.6 L ha^{-1} of commercial product (129 g ha^{-1} of Metomil). At 78 days after sowing, when harvesting the ears of green corn, five plants from the useful portion of each experimental unit were cut close to the soil, separated into stem, leaves, straw, cob and green grains subsequently, the materials of the stem, leaves, and straw of the ear of each repetition were crushed in a forage machine, model DPM 2 Nogueira, to reduce the size of the parts and consequent improvement in the drying process. Then, all parts of the corn plant were dried in an oven with forced air circulation, at ± 65 °C, until constant mass, to determine the dry phytomass of stem, leaves, straw of ear, cob and grains and total. The values of green and dry phytomass of each plant part of the corn were estimated for kg ha^{-1} , considering the density of 33.33 plants ha^{-1} used in this experiment. Samples of dry material from leaves and grains were crushed in a Willey mill to determine the levels of Fe in the extract resulting from the nitric-perchloric digestion of the tissues, through atomic absorption spectrometry, according to the methodology described in Malavolta *et al.* (1997) based on the levels of Fe in the leaves and grains and in the dry phytomass produced, the accumulations for the nutrient in each part were estimated, according to the expression: $\text{mg planta}^{-1} \text{ nutrient} = (\text{nutrient content in dry phytomass (mg kg}^{-1}) \times \text{dry phytomass of the part (g plant}^{-1})) / 1000$. At the end of the experiment, soil samples, in the 0 - 0.20 m depth layer, were collected in the useful area of each experimental plot to determine Fe levels, according to the methodology contained in Embrapa (1997). The data were submitted to analysis of variance (ANOVA) by the F test and regression analysis. All tests were

carried out at the level of 5% probability using the SISVAR® software (Ferreira, 2011).

RESULTS

The dry mass of leaves (Figure 1A), stem (Figure 1B) and straw (Figure 1C) were affected quadratically by Fe doses, reaching maximum values in doses of 2.6 g L⁻¹, 0.8 g L⁻¹ and 2.1 g L⁻¹, respectively. The dry mass of grains (Figure 1E), cob (Figure 1D) and ear (Figure 1F) were not influenced by Fe doses. Fe doses positively influenced the levels and accumulated amounts of Fe in corn leaves and grains.

grains was 1.8 g L⁻¹ and represents an increase of 45.3% in relation to the zero dose.

DISCUSSION

The increase in dry mass of leaves, stems and straw of green corn as a function of Fe doses, is partly justified by the lack of this micronutrient in the soil and its plant functions. The functions of Fe in plants are well documented, such as their participation in the composition of a heme prosthetic group, essential for the activity of several enzymes, cofactor of oxidative stress attenuating enzymes (SOD and peroxidases)

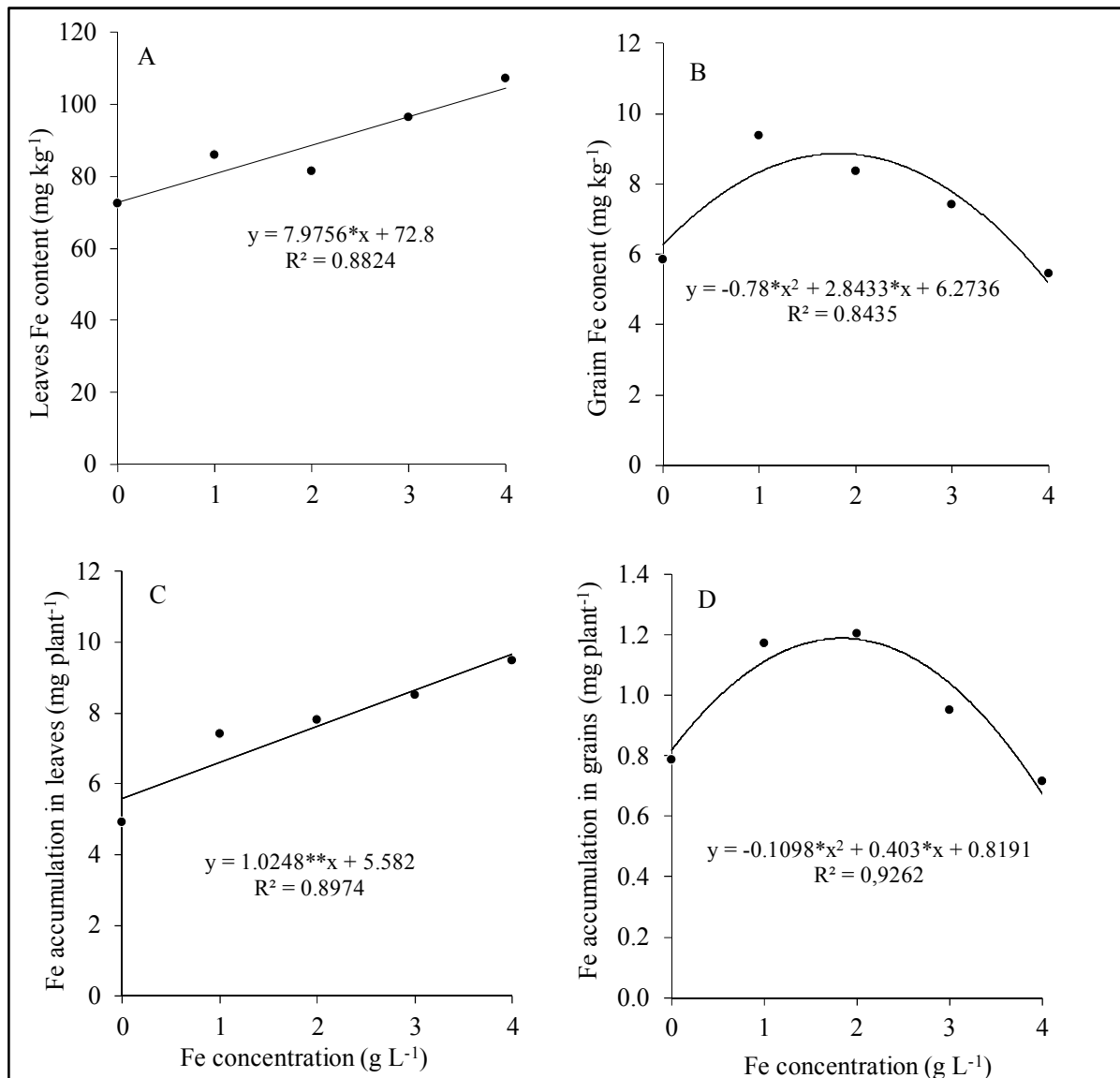


Figure 1. Dry mass of leaves (A), stem (B), straw (C), cob (D), grains (E) and ears (F) of AG 1051 green corn as a function of iron doses. *and** significant at 5% and 1% by the F test

There was a linear increase in leaf Fe contents (Figure 2A), with values ranging from 72.8 to 104.7 mg kg⁻¹ and an increase of 43.8% in the highest Fe dose (Figure 2B). In the grains the maximum content of 8.9 mg kg⁻¹ was obtained in the dose of 1.8 g L⁻¹ of Fe, representing an increase of 42.0% in relation to the zero dose. The accumulated amounts of Fe in the leaves (Figure 2C) and in the grains (Figure 2D) followed the same behavior observed for the respective levels. The maximum accumulated amount (1.19 mg plant⁻¹) of Fe in the

and chlorophyll synthesis (Elanchezhiana *et al.*, 2017). These functions, directly or indirectly, are related to the photosynthetic rate and protein synthesis and, therefore, to the growth and production of phytomass (Rui *et al.*, 2016; Askary *et al.*, 2017). Fe doses increased especially the production of dry leaves, which is probably due to the role of Fe in the synthesis of chlorophyll, which is found in a greater proportion in the leaves (Elanchezhiana *et al.*, 2017). The increase in the percentage values of dry mass of leaves was accompanied by a decrease in the percentages of dry mass of stem.

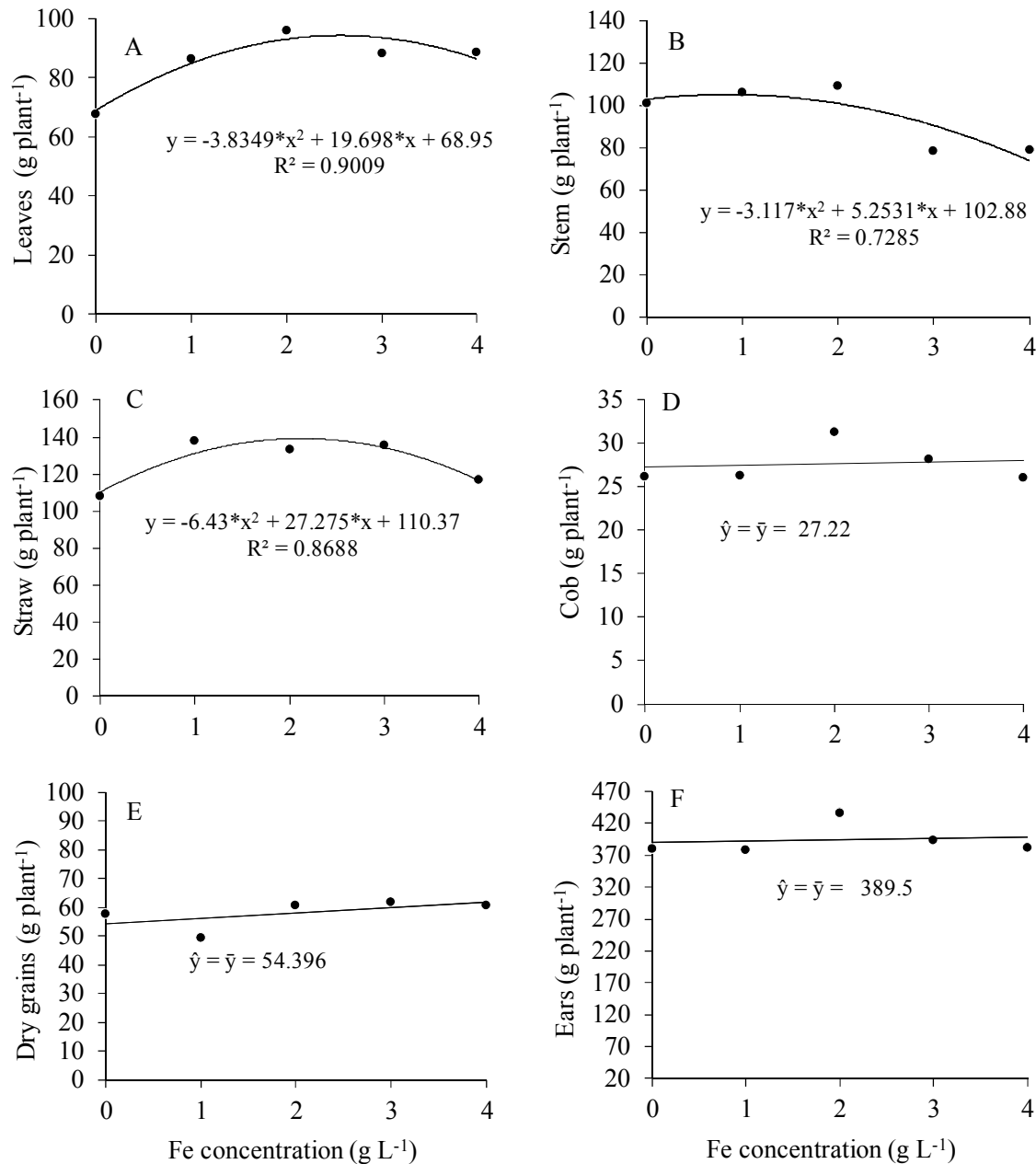


Figure 2. Iron content in leaves (A) and grains (B) and iron accumulation in leaves (C) and grains (D) of green corn AG 1051 as a function of iron doses. *and** significant at 5% and 1% by F test

These results indicate that fertilization with Fe favored the production of leaves to the detriment of stems and demonstrates the importance of fertilizing corn with Fe, when it is desired to increase the production of corn for forage. In addition, the leaves are the most active organ in the plant (Marschner, 2012; Taiz *et al.*, 2017) which can positively affect the several key metabolic processes for the growth and development of corn. Variations in the levels of Fe in the dry mass of leaf tissues from 72.8 to 104.7 mg kg⁻¹, in Fe doses of zero and 4 g L⁻¹, respectively are suitable for corn whose values must be 50 at 250 mg kg⁻¹ for maize (Gott *et al.*, 2014; Malavolta *et al.*, 1997). In the milky grains (80% moisture) the maximum Fe content obtained at the dose of 1.8 g L⁻¹ was 8.9 mg kg⁻¹. This content is equivalent to 27.91 mg kg⁻¹ considering dry grains with 12% moisture. Queiroz *et al.* (2011) observed that the levels of Fe in grains from 23 maize strains ranged from 13.4 to 35.6 mg kg⁻¹. These values are also consistent with those obtained in other studies (Queiroz *et al.*, 2011).

Connorton and Balk (2019) cite that the typical levels of Fe in corn grains are around 30 mg kg⁻¹, and vary between 11 to 60 mg kg⁻¹. The accumulated amounts of Fe in the dry mass of leaves and grains, reflect the combined effect of the levels and the respective dry mass production and due to this, the linear increase of Fe accumulation in the grains as a function of Fe doses is due practically just to the increase in the production of its dry mass (Marschner, 2012). In grains, the maximum Fe accumulation estimated at 1.19 mg per plant (or ear), represents up to 59.5% of the daily Fe requirements for adolescents aged 12 to 16 years for example (Abbaspour *et al.*, 2014). On the other hand, in terms of nutritional quality, the bioavailability of Fe, that is, the iron content contained in compounds such as phytates, ferritin, may be more important than the total content (Glahn *et al.*, 2019). In this study, the results clearly elucidated the need to fertilize green corn with Fe in order to increase the production of vegetative phytomass and the production of grains with higher Fe content in alkaline soil (Ahmad *et al.*, 2014; Durgude *et al.*, 2014; Saleem *et al.*, 2016). Although Fe can be supplied through other sources and

forms of application (Dhaliwal *et al.*, 2019; Elanchezhiana *et al.*, 2017; Yoon *et al.*, 2019), foliar fertilization may be more advantageous in alkaline soils (Ahmad *et al.*, 2014; Durgude *et al.*, 2014; Saleem *et al.*, 2016), for being easier to operate, and for avoiding precipitation of Fe in the form of insoluble oxides (Dhaliwal *et al.*, 2019; Rengel, 2015).

Conclusions

The fertilization of corn with ferrous sulfate via leaf in alkaline soil increased the production of dry green corn phytomass, but did not interfere in the dry mass of grains. Iron doses increased Fe concentration and accumulation in leaves and grains of green corn with gains of up to 43.8 and 42.0%, respectively.

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