



Influence of copper sources on the management of leaf rust, growth and crop yield of Arabica coffee

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Abstract

The fertilization with micronutrients in coffee plantations is a widespread practice; however, the benefits provided by these elements, as well as their sources, requires broader and deeper studies. Therefore, the objective of this study was to evaluate the influence of different sources of copper (Cu) on the growth, crop yield and intensity of leaf rust on Arabica coffee plants. The experiment was conducted at the “Eloy Carlos Heringer” Centre for Coffee Research (Centro de Pesquisas Cafeeiras “Eloy Carlos Heringer” – CEPEC) in municipality of Martins Soares, Minas Gerais State, Brazil. The experimental design used was a randomised block, with treatments consisting of seven copper sources: COC = copper carbonate; HOC_p = copper hydroxide (powder); HOC_l = copper hydroxide (liquid); OCC = copper oxychloride; SCO = copper sulphate; SCO + CAL = copper sulphate + lime, and OXC = cuprous oxide, with three replications. The treatments were applied in three sprayings, spaced at 30-day intervals. Before the first application and 30 days after each spraying, leaves were collected for nutritional analysis. Periodically, morphological and agrochemical parameters were assessed, where the count of vegetative internodes, number of leaves, and incidence and severity of rust were performed. At harvest, the crop yield, weight of unripe, dry and ripe fruits, percentage of floating fruits, and sieve classification were evaluated. For the studied conditions, the treatments with OCC and HOC_p provided greater reductions in the intensity of rust; treatments with HOC_p, HOC_l and OCC promoted greater increases in crop yield; and HOC_l promoted higher uniformity of ripening.

Key words: Growth, nutrition, phytosanitary, productivity, sustainability.

Introduction

Coffee is cultivated in more than 50 countries and is consumed in dozens of other nations, being currently the world's second largest commodity by market value, only behind oil. It is among the products that most contributed to the expansion of the world economy ¹. Given the great diversity of the different coffee-growing regions, various factors influence the success of the cultivation, particularly the technological level, production cost, occurrence of weather adversities (frost, drought, insolation), and attack of pests and diseases ^{2,3}, implying that an increase in research is needed to develop effective and sustainable management alternatives.

One of the major limitations of coffee production is the incidence of leaf rust, a plant disease caused by the fungus *Hemileia vastatrix* Berk. & Br. and considered the main disease that affects coffee plants. It causes numerous direct and indirect types of damage, such as defoliation (early leaf fall), fall of flowers, death of plagiotropic branches (potentially compromising production by up to 50%), and limitation of subsequent harvests ^{4,5}. To avoid or mitigate such damage, many producers use synthetic fungicides in excess, impacting the environment, altering the ecological balance and even poisoning living beings.

As an alternative to this problem, special attention is given to copper-based fungicides, which have a broad spectrum of activity, reduced risk of selection of resistant pathogen strains, and are less problematic to the environment ⁶. When applied to the leaf surface, the copper fungicide works as defence against the leaf rust infection, forming a barrier capable of preventing the germination and penetration of urediniospores into the leaf tissues³.

Copper has received special attention due to its ability to combine the nutritional need (triggering a series of benefits to the plant) with the potential for disease management, as well as the proportional tonic effect, influencing the vigour and retention of leaves on the plants ⁷. These effects combine to increase the overall crop yield.

The main copper-based compounds used as spray solutions on the coffee plants are found in commercial formulations under four categories: copper hydroxides, oxychlorides, oxides, and sulphates. The difference in activity of these compounds depends on the chemical nature of the copper salts, primary particle size, dispersibility, tenacity, and adherence of these particles ⁸; however, little is known regarding the specific effects of these different

compounds. The objective of this study was to evaluate the influence of different copper sources on growth and yield, in addition to their fungicidal action against leaf rust, in productive plants of Arabica coffee.

Materials and Methods

Description of the local and plant material: The experiment was conducted in the Eloy Carlos Heringer Centre for Coffee Research (Centro de Pesquisas Cafeeiras Eloy Carlos Heringer - CEPEC), located in the municipality of Martins Soares, MG (20°14'45" S and 41°50'47" W, elevation of 763 m). The climate, according to the Köppen classification, is Cwa, mesothermal with humid summers. The soil of the area was characterised as dystrophic Red-Yellow Latosol⁹, with the chemical characteristics presented in Table 1.

In the recommended phenological stages, samples of the soil and leaves were collected and analysed to manage the fertilising and liming, aiming to supply the nutritional demand of the coffee plants, according to the recommendations¹⁰. Climatological data were collected daily at the CEPEC weather station, located 100 m from the experiment, thus obtaining the monthly averages for temperature and precipitation during the period in which the experiment was conducted (Fig. 1). The experiment started at February 2012 and was conducted and evaluated until September 2013. The plants were of the cultivar Catuaí Vermelho IAC 44, spaced at 2.5 m x 0.8 m, 15 years of age, pruned in September 2010. Before implementation of the experiment, an analysis was performed to assess the nutritional levels of the leaf tissues of the plants utilised to evaluate their nutritional state and make recommendations for fertilisation (Table 2).

Table 1. Chemical attributes of the dystrophic Red-Yellow Latosol, in August 2011, prior to the experiment (CEPEC, Martins Soares, MG, Brazil).

Attributes	Values
pH (H ₂ O)	4.99
P (mg dm ⁻³)	22.60
K (mg dm ⁻³)	48.00
Ca (cmol _c dm ⁻³)	2.24
Mg (cmol _c dm ⁻³)	0.55
Al (cmol _c dm ⁻³)	0.24
H + Al (cmol _c dm ⁻³)	9.80
BS (cmol _c dm ⁻³)	2.93
CEC (t) (cmol _c dm ⁻³)	3.17
CEC (T) (cmol _c dm ⁻³)	12.73
V (%)	23.00
m (%)	7.60
OM (dag kg ⁻¹)	3.70
P-rem (mg L ⁻¹)	19.61
Zn (mg dm ⁻³)	5.80
Fe (mg dm ⁻³)	28.60
Mn (mg dm ⁻³)	5.50
Cu (mg dm ⁻³)	0.20
B (mg dm ⁻³)	0.19
S (mg dm ⁻³)	28.18

Attributes determined following the methodology of Guimarães¹⁰.

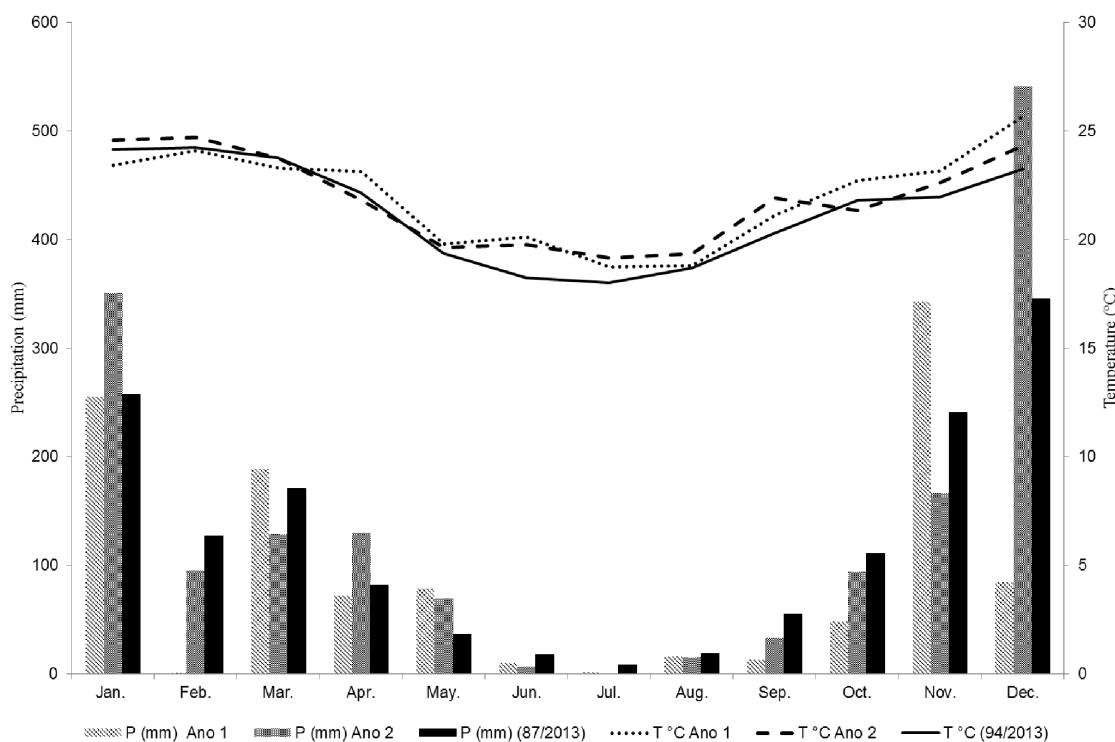


Figure 1. Monthly values of accumulated rainfall (mm month⁻¹) and average temperature (°C month⁻¹) observed in the experimental area during the two years evaluated and the local historical series (values observed in the period of 1987 to 2013, at CEPEC, Martins Soares, MG, Brazil).

Table 2. Analysis of the nutritional levels in leaf tissues for the coffee plants prior to the copper fertilization (December 2011, CEPEC, Martins Soares, MG, Brazil).

N	P	K	Ca	Mg	S	Zn	Fe	Mn	Cu	B
----- dag kg ⁻¹ -----					----- mg kg ⁻¹ -----					
2.91	0.16	2.45	0.83	0.21	0.23	7.60	91.41	32.70	14.75	33.00

Experimental design: The experiment followed a randomised block design, with split plot scheme, with plots subdivided in time, with four replications. The seven copper-based treatments composed the plots, and the influence of the experimental year was assessed (first and second reproductive cycles) composed the subplots. In this scheme, the following variables were studied: number of new vegetative nodes per plagiotropic branch (NVNPB), number of new leaves per plagiotropic branch (NLPB), copper content in the green leaf tissues, area under the disease incidence progress curve (AUDIPC) and crop yield. For the variables related to rust severity (AUDSPC) and quality of the coffee fruits (unripe, dry and ripe fruits, average percentage of floating fruits, and sieves 17, 16, 14, and lower), a randomised block design was used with three replications and the same seven treatments with copper sources. Each experimental plot consisted of four rows of 10 plants, considering the useful area to be the two central rows and six plants of each row, totalling 12 plants per plot, fully surrounded by protective plant borders.

Copper sources: The treatments consisted of seven copper sources: COC, HOC_p, HOC_l, OCC, SCO, SCO + CAL, and OXC (Table 3). The treatments were applied over the leaves in three parcels, spaced 30 days apart, from February to April. This period is based on the local history of early occurrence of rust, obtained by monthly monitoring from the years to 1994 to 2013 of experimental crops of the CEPEC, without fungicide application (Fig. 2). The applications were performed by the same operator, using the same sprayer, in the morning. The copper doses applied were based on the recommendation¹⁰ of 600 g ha⁻¹. Thus, based on the concentration of each product, the same copper dose was applied in all treatments. A “blank test” was performed before each application to guarantee the uniformity of the solution. The sprayer used (PJH, Jacto) has a 20 L plastic tank with a metal base and internal components and a piston-pressure chamber in brass. A cone spray nozzle (JD-10 A, disc 1.0 mm, Jacto) was used with the shape of an empty cone, stainless steel disc, small drops, angle of 80 degrees at 60 psi.

Table 3. Composition of the products utilised in the experiment.

Copper source	Active ingredient	Concentration (% Cu)
COC	Carbonate of copper	48
HOC _p	Copper hydroxide (powder)	35
HOC _l	Copper hydroxide (liquid)	35
OCC	Oxychloride of copper	50
SCO	Copper sulphate	25
SCO + CAL	Copper sulphate and lime	25
OXC	Cuprous oxide	50

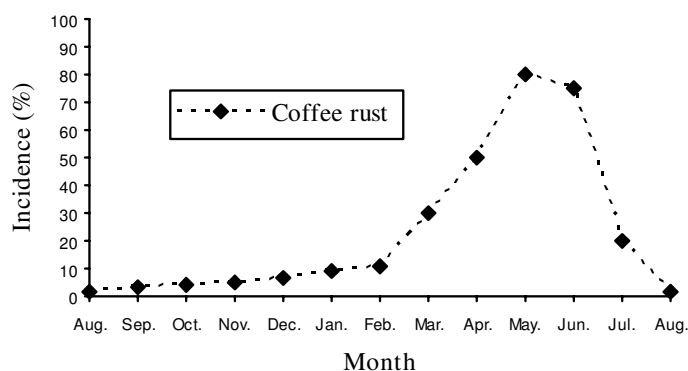


Figure 2. Historical series of rust incidence observed in the CEPEC for the years 1994 to 2013 (CEPEC, Martins Soares, MG, Brazil).

Evaluation of the study: The number of new vegetative nodes on the plagiotropic branches (NVNPB) and new leaves (NLPB) was monitored monthly during the period from February 2012 to September 2013. Two plagiotropic branches were evaluated per plant, in the middle section of the plants, on opposite sides, previously identified in February 2012. The leaf copper content, was evaluation before the first spraying and 30 days after each spraying, leaves were collected; they were packaged in paper bags and dried in laboratory oven, with forced air circulation, at 60°C until reaching constant mass. Subsequently, the leaves were ground in a Wiley mill and passed through a 20-mesh sieve (0.841 mm). A portion of the samples was then subjected to nitric-perchloric digestion for determination of the copper concentrations by flame atomic absorption spectrophotometry¹¹ in the Laboratory of Plant Mineral Nutrition of the Centro de Ciências Agrárias, Universidade Federal do Espírito Santo (CCA-UFES). The evaluations of rust incidence on the leaves of coffee plants were performed at intervals of 30 days, from February 2012 to September 2013, referring to the two harvests of the agricultural years 2011/2012 (year 1) and 2012/2013 (year 2). A non-destructive method of evaluation was chosen, based on evaluating leaves from the middle third, between the 3rd and 4th pair of leaves from two plagiotropic branches per plant, previously marked in February 2012 with a ribbon tied to the first vegetative internode. The incidence of disease was determined according to the percentage of leaves infected with the leaf rust in a sampling of 100 leaves. The severity of leaf rust were evaluated monthly, on the same period and branches described for the intensity, using 12 plants per plot, totalling 24 leaves per plot. Rust severity was assessed using the scale¹², that consists of three diagrams of coffee leaves with 30, 50, and 70% of the areas marked indicating severity, where in each leaf a known quantity of the area (1, 3, 5, 7, and 10%) is occupied by pustules of the leaf rust. The data of rust incidence and severity in coffee leaves observed during the evaluation period were transformed into area under the disease progress curve (AUDPC)¹³. Harvest was performed in the month of June (for both years) when, on average, 90% of the fruits were fully mature.

Fruits were harvested from all plants in the plot and used to estimate the productivity of each plot, in bags of processed coffee per hectare (bags ha⁻¹). In the 2013 harvest, coffee fruits were removed from four plagiotropic branches per plot and washed and separated into four groups: floating, dry, unripe and ripe, quantifying the weight of each group. The fruits were then dried in an oven with forced circulation at a temperature of 60 °C until the grains reached 12% moisture. After drying, the samples were weighed, and the grains were separated from the parchments and hulls. The grains were weighed to determine the yield of dry processed grains from each plot. Samples of flat grains were classified using number 14, 16, and 17 sieves, where the percentage of grains retained on each individual sieve was evaluated, including smaller grains that passed through all the sieves and were deposited on the bottom, representing the grains with size smaller than 14.

Statistical analysis: Data were subjected to analysis of variance, and in the presence of significant differences, the treatments were differentiated and studied with appropriate statistical techniques, using the statistical analysis software SISVAR¹⁴. The Tukey test ($p \leq 0.05$) was used to determine if there was differentiation between productive cycles and the Scott-Knott criteria ($p \leq 0.05$) was used to group the copper source regarding the similarity of their effects.

Results

Vegetative growth and copper content of leaf tissue: Based on analysing the number of vegetative nodes and number of leaves on plagiotropic branches (Table 4) for the two years of cultivation, it was verified that there was no difference among the copper sources, and for both characteristics, there was greater vegetative growth in the first year. The compound OXC, followed by HOC_p, SCO + CAL, and OCC, provided greater increases of Cu content in the leaf tissues of the coffee plant during the first cycle of cultivation (Table 5); in the second cycle, the OXC and HOC₁ compounds resulted in the highest level of Cu in the leaf tissue. The values of Cu observed in leaf tissues resulting from OXC and HOC_p in the first year were greater than in the second year, where HOC₁ promoted higher values in the second year than in the first (Table 5).

Table 4. Means of number of new vegetative nodes (NVNPB) and number of new leaves on plagiotropic branches (NLPB) of plants of Arabica coffee sprayed with different sources of copper, during two productive cycles, from February 2012 to September 2013 (CEPEC, Martins Soares, MG, Brazil).

Copper source	NVNPB		NLPB	
	First cycle	Second cycle	First cycle	Second cycle
COC	11 aA	7 aB	25 aA	8 aB
HOC _p	11 aA	7 aB	26 aA	7 aB
HOC ₁	11 aA	8 aB	27 aA	9 aB
OCC	11 aA	7 aB	26 aA	9 aB
SCO	10 aA	7 aB	24 aA	9 aB
SCO + CAL	10 aA	7 aB	24 aA	9 aB
OXC	11 aA	7 aB	25 aA	8 aB

Means followed by the same lower-case letter in the column do not differ by the Scott-Knott test ($p \leq 0.05$), and means followed by the same upper-case letter in the line do not differ by the Tukey test ($p \leq 0.05$).

Table 5. Means of copper content (mg kg⁻¹) in leaf tissues of Arabica coffee sprayed with different sources of copper, during two productive cycles, from February 2012 to September 2013 (CEPEC, Martins Soares, MG, Brazil).

Copper source	Productive cycle	
	First cycle	Second cycle
COC	20.93 dA	25.20 cA
HOC _p	41.11 bA	24.93 cB
HOC ₁	28.43 cB	41.77 aA
OCC	37.82 bA	31.00 bA
SCO	26.50 cA	26.03 cA
SCO + CAL	37.63 bA	34.51 bA
OXC	68.13 aA	45.03 aB

Means followed by the same lower-case letter in the column do not differ by the Scott-Knott test ($p \leq 0.05$), and means followed by the same upper-case letter in the line do not differ by the Tukey test ($p \leq 0.05$).

Intensity of the coffee leaf rust: In the first year, it was observed that the compound OCC resulted in the greatest reduction in the area under the disease progress curve based on the incidence of leaf rust, an area reduction of 88.5%. The compound OXC was least effective in rust management, providing the lowest reduction (59.1%) (Table 6). In the second year, better results were observed in relation to rust management for the compounds HOC_p and OCC, which resulted in reductions of 35.7 and 29.1% of the AUDIPC, respectively, demonstrating superiority over the others regarding the fungicidal effect. In the first year, the area was lower, indicating that the disease was less severe (Table 6). For the severity of leaf rust, the compounds COC and HOC_p showed greater reduction of the AUDSPC, and the compound OXC resulted in the lowest reduction, presenting the highest AUDSPC (954.33), thus indicating greater disease intensity (Table 6).

Crop yield: In the first evaluated harvest, the application of copper sources did not result in different productivities (Table 7). In the second cycle, however, there was a differential response in the crop yield of coffee plants, with the results indicating higher productivity of plants treated with HOC_p, HOC₁, and OCC. The application of SCO resulted in smaller proportion of unripe and dry fruits (Table 8); however, the compound HOC₁ resulted in the highest proportion of ripe fruits and lowest percentage of floating fruits, which is desirable because it reflects the better quality of the grains. In general, the application of HOC_p and HOC₁ sources resulted in an increase in grain size, as indicated by the increase in the average grains retained in sieve number 17 and reduction in the average grains retained in the sieve number 14 (Table 9).

Discussion

Effect of copper sources on the coffee plant: All copper sources promoted leaf growth compared to the average values before application of the treatments. The compound OXC promoted the greatest increase in the copper content of leaves, with values above the range considered ideal, namely, between 7 and 50 mg kg⁻¹¹¹; other compounds presented values within this range. With greater reduction of AUDIPC and AUDSPC, the compounds OCC and HOC_p showed the best results and thus greater efficiency for the management of coffee leaf rust. This result is most likely

Table 6. Means of area under the disease incidence progress curve (AUDIPC) and severity progress curve (AUDSPC) for leaf rust (*Hemileia vastatrix* Berk. & Broome) of Arabica coffee, with their respective percentages of reduction of area, sprayed with different sources of copper, during two productive cycles, from February 2012 to September 2013 (CEPEC, Martins Soares, MG, Brazil).

Copper source	AUDIPC		AUDSPC
	First cycle	Second cycle	First cycle
COC	197.85 cB (77.2%)	14,111.47 bA (17.8%)	406.65 a (70.0%)
HOC _p	156.90 bB (81.9%)	11,029.05 aA (35.7%)	447.60 a (67.0%)
HOC ₁	156.00 bB (82.0%)	14,027.48 bA (18.3%)	526.23 b (61.2%)
OCC	99.15 aB (88.5%)	12,166.42 aA (29.1%)	500.25 b (63.1%)
SCO	166.05 bB (80.8%)	14,118.67 bA (17.8%)	728.85 c (46.2%)
SCO + CAL	218.25 dB (74.8%)	15,072.60 bA (12.2%)	548.17 b (59.6%)
OXC	354.75 eB (59.1%)	14,228.25 bA (17.1%)	954.33 d (29.6%)

Means followed by the same lower-case letter in the column do not differ by the Scott-Knott test ($p \leq 0.05$), and means followed by the same upper-case letter in the line do not differ by the Tukey test ($p \leq 0.05$).

Table 7. Means of crop yield (bag ha⁻¹) of Arabica coffee sprayed with different sources of copper, during two productive cycles, from February 2012 to September 2013 (CEPEC, Martins Soares, MG, Brazil).

Copper sources	Crop yield (bag ha ⁻¹)	
	First cycle	Second cycle
COC	7.04 aB	97.33 bA
HOC _p	6.91 aB	106.33 aA
HOC ₁	8.46 aB	107.33 aA
OCC	7.16 aB	102.67 aA
SCO	7.04 aB	97.67 bA
SCO + CAL	8.41 aB	96.00 bA
OXC	7.51 aB	93.33 bA

Means followed by the same lower-case letter in the column do not differ by the Scott-Knott test ($p \leq 0.05$), and means followed by the same upper-case letter in the line do not differ by the Tukey test ($p \leq 0.05$).

Table 8. Means of unripe fruit mass (UFM), dry fruit mass (DFM), ripe fruit mass (RFM) and percentage of floating fruits of Arabica coffee sprayed with different sources of copper, during two productive cycles, from February 2012 to September 2013 (CEPEC, Martins Soares, MG, Brazil).

Copper source	UFM (g)	DFM (g)	RFM (g)	Floating fruits (%)
COC	50.67 c	35.33 b	371.00 f	12.00 a
HOC _p	51.00 c	49.00 a	365.33 g	12.00 a
HOC ₁	33.33 d	15.00 e	440.00 a	7.00 c
OCC	77.33 b	11.67 f	419.33 b	7.33 c
SCO	31.33 d	8.67 g	400.67 c	8.00 b
SCO + CAL	47.00 c	21.00 d	374.33 e	8.33 b
OXC	113.00 a	28.67 c	384.67 d	6.33 c

Means followed by the same lower-case letter in the column do not differ by the Scott-Knott test ($p \leq 0.05$).

Table 9. Means of percentage of grains retained in sieves 17, 16, 14 and lower of Arabica coffee sprayed with different sources of copper, obtained in the 2013 harvest (CEPEC, Martins Soares, MG, Brazil).

Copper source	Sieve 17 (%)	Sieve 16 (%)	Sieve 14 (%)	Lower
COC	35.33 d	32.67 a	22.33 b	9.00 a
HOC _p	46.00 a	29.67 b	17.67 c	9.67 a
HOC ₁	47.67 a	26.33 b	13.67 d	12.00 a
OCC	40.67 c	30.00 b	19.00 c	11.33 a
SCO	39.67 c	35.00 a	18.33 c	9.67 a
SCO + CAL	43.00 b	27.33 b	17.67 c	9.33 a
OXC	36.67 d	31.67 a	25.67 a	9.67 a

Means followed by the same lower-case letter in the column do not differ by the Scott-Knott test ($p \leq 0.05$).

because these compounds promote a more gradual release to the plant¹⁵, and it is believed that the element remains on the leaf surface for longer periods, promoting a better and more long-lasting fungicide action.

Overall, the HOC_p, HOC₁, and OCC compounds resulted in higher crop yield, greater uniformity of ripening, and production of bigger grains. This result may be related to these compounds allowing a better utilization of copper by the plant, resulting in increased photosynthetic activity, as more than 50% of the copper content in the leaves is present in chloroplasts, in the form of plastocyanin, a protein that participates in the electron flow of the photosynthesis¹⁶. There is, consequently, a higher biomass accumulation, resulting in better supply for the demands of the reproductive organs, allowing

the better development of fruits and promoting the ripening uniformity. Another factor that may have contributed to the increased productivity and grain size provided by the compounds HOC_p, HOC₁, and OCC is that these compounds have been superior for the leaf rust management, which resulted in more healthy and vigorous leaves, increasing their photosynthetic potential.

Responses of the coffee plants along two years of treatment with copper sources:

The greater vegetative growth observed in the first year resulted in higher productivity in the second evaluated cycle. In the first year, coffee yield was low, a characteristic of bienniality, a phenomenon of the physiological nature of Arabica coffee plants, which requires its growth in one year to support the fruit production in the following year¹⁷. This process is related to the direction of the photosynthates produced by the plant, where in years of high hanging fruit load, they are destined for the formation and growth of fruits, and in years of low hanging fruit loads, they are directed to the formation of new plant buds that result in new branches¹⁸. Thus, plant growth varies in two-year cycles¹⁹, and consequently, so does the number of leaves, as shown in Table 4.

Another factor influenced by the bienniality process was the content of copper; in general, the values in the first year were higher than in the second one, where there was greater remobilisation of this nutrient due to greater production of fruits. Based on this result, the importance of copper supply to coffee is reinforced, especially during periods of high fruit production.

In the second productive cycle evaluated in this experiment, there was a greater incidence of leaf rust in the plants, shown by the greater AUDIPC, which is caused by the elevated hanging

load that occurred in this year. Greater disease intensity is observed in years of high fruit production, which may be related to the drainage of photoassimilates from leaves to fruits^{5,20}. This drainage alters the resistance of the plant due to nutritional imbalance, based on the assumption that any change in leaf nutrient levels in addition to carbohydrates of the coffee plant in production may make the plant more susceptible to diseases.

Additional factors that may have caused the higher AUDIPC in the second year was the high temperature from November (first year) to March (second year), as well as the high precipitation in November (first year) and January (second year) (Fig. 1), which create favourable conditions for the fungus development.

Adverse climatic conditions may cause the inoculum to increase earlier in the season, decreasing the efficacy of copper-based compounds, as its main effect in the management of leaf rust is preventive³. The vigorous vegetative growth together with the lower intensity of leaf rust in the first year allowed an increased metabolism investment in fruit production, resulting in a higher productivity in the second year evaluated in this experiment.

Conclusions

Given the conditions studied, the different copper sources used in the cultivation of Arabica coffee promote different responses in the leaf rust management, crop yield, ripening and size of grains. The treatment with OXC provided greater accumulation of copper in leaf tissues; however, the treatments with OCC and HOC_p resulted in greater reductions in leaf rust intensity. Treatments with HOC_p, HOC_i, and OCC promoted greater increases in crop yield, and HOC_i resulted in a higher ripening uniformity.

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