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Export of macronutrients for coffee fruits submitted to different doses of formulation 20-00-20

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ABSTRACT

The objective of this study was to quantify the export of macronutrients by coffee fruits *Coffea canephora* "Conilon - BRS Ouro Preto," under different doses of the fertilizer formulation 20-00-20 under rainfed conditions, in the Western Amazon. The treatments were six doses of fertilizer 20-00-20, in the proportions: 0; 250; 500; 1,000; 2,000; 3,000 kg ha⁻¹ year⁻¹ and the experimental design was completely randomized with 15 replications. There was a difference for the accumulation of dry mass, content and export of nutrients when comparing straw and grain. The export of nutrients by the grain follows a decreasing order: N > K > Mg > P > Ca > S and the export by straw follows the order: K > N > Ca > Mg > S > P. The doses of the formulation not influence the levels of nutrients in the fruits but promoted an exponential increase in dry mass of grains and straw, with a maximum accumulation of 6,413 kg ha⁻¹ of dry mass of grain and 5,455 kg ha⁻¹ in straw at a dose of 3000 kg ha⁻¹. It is concluded that the doses of fertilizer formulation 20-00-20 do not promote changes in the concentrations of nutrients N, P, K, Ca, Mg, and S in the fruits of the coffee tree "Conilon - BRS Ouro Preto." However, it promotes greater accumulation of dry mass in both grains and straw and greater export of nutrients by the fruits.

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Introduction

Clonal crops of *Coffea canephora* formed from varieties adapted to the edaphoclimatic conditions of a given region can produce more than 7000 kg of processed grains per hectare (120 bags ha⁻¹) (Bornomo et al. 2017), if conducted according to the recommendation's techniques for culture. Among these recommendations, the correct use of fertilizers (Prezotti and Bragança 2013) stands out, which are essential to replace nutrients exported annually by fruits.

Coffee trees with high productive potential also have high nutritional requirements, accumulating large amounts of nutrients in their tissues (Serrano, Da Silva, and Formentini 2011). However, the amount of nutrients retained in the coffee tissue varies according to the location and time of year, in addition to the tissue and organ of the same plant (Bragança et al. 2007) and may also vary with the genotype (Partelli et al. 2014; Marré et al. 2015; Covre et al. 2016) and the environmental conditions of the cultivation site (Laviola et al. 2008).

Regarding the genotype, there is great genetic variability of the species *Coffea canephora* distributed around the world (Montagnon, Cubry, and Leroy 2012) as well as, within Brazil (Ferrão et al. 2013; Souza et al. 2013). This genetic difference influences both the absorption and translocation of nutrients from the root system to the fruit (Amaral et al. 2011), as well as, in its maturation cycle, which results in different nutrient accumulation curves during the filling phase of fruits (Partelli et al. 2014; Marré et al. 2015).

Climatic conditions, such as temperatures and precipitation, are also important factors that influence the absorption of nutrients and are closely linked to the growth and fruiting of the coffee tree, which may delay or advance the formation of fruits. High temperatures and water supply increase sweat flow rates and mineral capture and translocation (Laviola et al. 2009; Martins et al. 2011), while mild temperatures reduce sweating and absorption speed of nutrients (Ramalho et al. 2014b). In addition, the reduction of soil water availability limits the access of roots to nutrients, as well as causes limitation or interruption of transpiration flow by closing the stomata (Carvalho et al. 2011).

For the conditions of the Brazilian Amazon, in particular, the occurrence of high temperatures, associated with low values of relative humidity of the air during the dry season, from June to September (Alvares et al. 2013), results in high rates evapotranspiration (Martins et al. 2011) which, in the absence of supplementary irrigation, can cause coffee growth to stall (Araújo et al. 2011) or even the loss of dry matter due to leaf fall.

Due to the specificities of the climatic conditions in the Amazon, the Brazilian Agricultural Research Corporation (Embrapa), developed a variety of clonal coffee trees, called “Conilon - BRS Ouro Preto” adapted to the cultivation conditions in the Brazilian Western Amazon (Ramalho et al. 2014a). However, information on the responses of this variety to the supply of nutrients is still limited, which hinders the correct nutritional management of crops. Therefore, the objective in this study was to quantify the export of macronutrients by coffee fruits *C. canephora* “Conilon - BRS Ouro Preto,” under different doses of the fertilizer formulation 20-00-20, under rainfed conditions, in the Western Amazon.

Material and methods

The experiment was carried out at an experimental field of the *Embrapa Rondônia*, in the city of Porto Velho (RO) Brazil (8°53'20" S and 63°06'40" W), during 2014/2015 harvest. According to Köppen, the local climate is classified as *Am* type, which stands for rainy tropical with rainy summers (October to May) and dry winters (June to September). Average temperatures range from 26.3 °C in the summer to 25.9 °C in the winter. Average annual rainfall is 2,200 mm (Alvares et al. 2013). Data on rainfall, temperature, and air relative humidity during the experimental period were obtained from a weather station of the Rondônia State Secretariat of Environmental Development (Sedam 2017) (Figure 1).

Coffea canephora plants of the clonal variety “Conilon - BRS Ouro Preto,” which is composed of 15 genotypes (clones) of intermediate ripening (May), were grown in the experimental area. These genotypes show typical phenotypic characteristics of plants of the Conilon botanical group. The variety was developed under rainfed conditions (Ramalho et al. 2014a) and has been recommended for cultivation in the Southwestern Amazon.

The coffee trees were planted in December 2008 and received fertilizers for the production of 70 bags per hectare during the years/harvests of 2009, 2010, 2011 and 2012 according to the recommendations of Marcolan et al. (2009). In September 2012, the plants received drastic pruning of the orthotropic stems to standardize the crown. After pruning, coffee trees had six stems per plant remaining (9996 stems per hectare). From September 2012 to September 2013, the period prior to the installation of the experiment, the plants received 50 kg hectare⁻¹ of P₂O₅, in the form of triple superphosphate, 50 kg of nitrogen in the form of urea, 100 kg of nitrogen in the

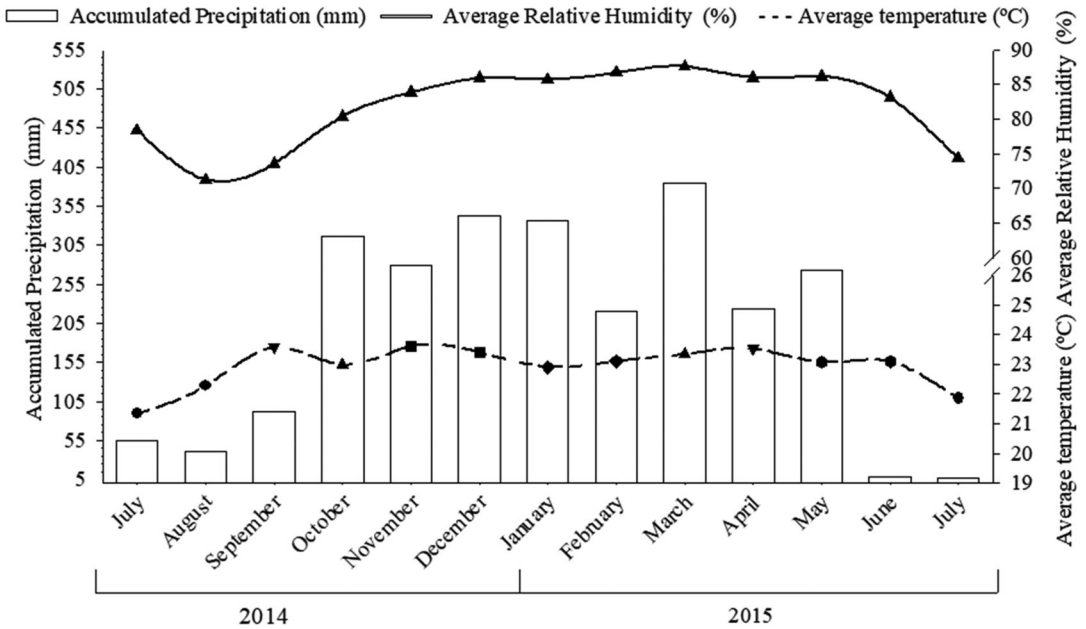


Figure 1. Accumulated Precipitation, Average Relative Humidity (%) and Average Temperatures (°C) for the 2014/2015 harvest. Source: Sedam (2017). Porto Velho, RO, 2017.

Table 1. Chemical and physical attributes of a Red-Yellow Soil at the Embrapa Experimental Station, in the 0-10, 10-20 and 20-40 cm layers. Porto Velho, 2014.

Layers (cm)	pH H ₂ O	P mg dm ⁻³	K	Ca	Mg	Al + H cmol _c dm ⁻³	Al	CTC	MO g kg ⁻¹	m	V %
00-10	5.6	24.83	0.53	4.87	4.67	11.88	0.28	21.55	43.30	3	44
10-20	5.3	18.38	0.39	3.21	3.18	12.70	0.66	19.47	37.16	9	34
20-40	5.0	9.33	0.26	1.99	1.86	11.96	0.91	16.12	31.15	19	25
Density (Visible soil) gcm ³	Particle(Real)	Porosity Micro m ³ m ⁻³	Macro	Total	Water content (m ³ m ⁻³) 6 10 30 ²	Tensions (kPa) 0.42 0.41	0.37	100	1500 ³		
1.03	2.33	0.42	0.08	0.50				0.34		0.27	

pH in water 1: 2.5, M.O. by wet digestion, P and K determined by the Mehlich I, Ca, Mg and Al exchangeable method extracted with 1 mol KCl.

form of sulfate ammonium, 60 kg of K₂O in the form of potassium chloride and 30 kg hectare⁻¹ of the complexed fertilizer FTE BR 12®. The other cultural treatments were carried out following the technical recommendations for culture in the region (Marcolan and Espindula 2015). From October 2013 until March 2015, the plants started to receive different doses of fertilizer, according to the pre-established treatments and design.

The soil in the area was classified as a Red-Yellow Latosol (Oxisol) with a clay texture (Santos et al. 2018), and its chemical and physical attributes were determined in the layers of 0-10, 10-20 and 20-40 cm, in July 2014 (Table 1). In September 2014, before the start of the 2014/2015 harvest, two tons of dolomitic limestone (with 76% PRNT) were applied on the surface, without incorporation, to increase the basis saturation from 44 to 60% in the layer 0-10 cm.

The treatments were six doses of the fertilizer formulation 20-00-20, in the following proportions: 0; 250; 500; 1,000; 2,000; 3,000 kg ha⁻¹. The experimental design was completely randomized with 15 replications, each replication consisting of a clone of the variety “Conilon - BRS Ouro Preto.” The experimental plot consisted of 10 plants with a spacing of 3 m between rows by 2 m between plants, totaling 1,666 plants ha⁻¹.

The treatments (doses of fertilizer) were applied in installments during the rainy seasons of each year, from October 2013 to March 2015, totaling two fertilization cycles. For each year, doses were divided into four applications: October, December, January and March. The crop was maintained in rainfed conditions, without supplementary irrigation and the cultivation was carried out following the recommendation for culture in Amazon (Marcolan et al. 2009). The fruits were harvested from 15 May to 15 June 2015.

For the supply of phosphorus, 100 kg hectare⁻¹ of P₂O₅ were applied annually in 2013/2014 and 2014/2015 divided into two applications, beginning of October and beginning of March, in the form of simple superphosphate. In October, 50 kg hectare⁻¹ of complexed fertilizer FTE BR 12[®] was also applied to supply micronutrients. In November and March of each year, 20 kg of zinc sulfate, 15 kg of boric acid and 20 kg of magnesium sulfate were also applied (Marcolan et al. 2009).

For chemical analysis, two liters of fruit were removed from 8 plants of the useful plot and subjected to natural drying in a covered terrace. Subsequently, the samples were packed in paper bags and taken to dry in an oven with forced air circulation at 65 °C, until constant mass. Then the straw was separated from the grain, by means of a manual peeler and ground in a stainless-steel Wiley mill for chemical analysis.

The contents of macronutrients, accumulation of dry mass and export of nutrients by grains and straw were determined. To quantify the levels (g kg⁻¹) of Nitrogen (N), Phosphorus (P), Potassium (K), Magnesium (Mg), Calcium (Ca) and Sulfur (S), the plant material was submitted to sulfuric digestion and nitro-perchloric. The digested samples, obtained by sulfuric digestion, were used to determine N. The samples processed by nitro-perchloric digestion were used to determine the levels of P, K, Mg, Ca and S, following the methodology described by Embrapa (2009).

The accumulation of dry mass was determined by the production of grains or straw, given in kg ha⁻¹, corrected to zero% of humidity by the equation.

$$\text{Dry mass of grain or straw (kg ha}^{-1}\text{)} = \text{Wet mass in kg ha}^{-1} - [(\text{wet mass}) * (\text{humidity}/100)] \quad (1)$$

The export of macronutrients (kg ha⁻¹) in grain and straw was calculated as follows:

$$\text{Export of macronutrients} = \text{DM of the grain or straw} * \text{Nutrient content (g kg}^{-1}\text{)} \quad (2)$$

The data were submitted to analysis of variance. To compare the dry mass accumulation, contents and export of nutrients in the grain or in the straw, the Tukey test was applied using the program Genes Version 2016.6.9 (Cruz 2017). For dose purposes, regression curves were adjusted using SigmaPlot[®] software version 10 (SYSTAT SOFTWARE Inc, 2006). The mathematical models were chosen taking into account the highest values of the determination coefficients (R²), the significance of the regression coefficients (β_i) and the regression F test, both up to 5% probability, as well as the behavior of the biological phenomenon.

Results

Nutrient content

For all evaluated nutrients, the interaction of fertilizers dose with the fruit tissue was not significant, indicating that the levels of nutrients in grains and straw responded similarly to the increase in fertilizer doses. However, when comparing these tissues of the fruit (grain and straw), it was possible to observe differences in the content of macronutrients regardless of the fertilizer dose used. Therefore, the result of the comparison test of means was presented only for the average nutrient content (Table 2).

Table 2. Content of macronutrients (g kg^{-1}) in grains and straw of *Coffea canephora* "Conilon - BRS Ouro Preto" as a function of six doses of fertilizer 20-00-20. Porto Velho, 2015.

Nutrient	Tissue	Dosage						Averages	CV%
		0	250	500	1,000 g kg^{-1}	2,000	3,000		
Nitrogen	Grain	26.73	25.66	27.45	26.66	28.65	27.15	27.05 a	17
	Straw	18.78	18.61	20.77	19.04	19.70	19.58	19.17 b	
Phosphorus	Grain	1.59	1.68	1.60	1.35	1.45	1.35	1.50 a	30
	Straw	1.42	1.17	1.20	0.95	1.09	1.03	1.15 b	
Potassium	Grain	21.26	21.34	21.28	21.62	20.72	18.22	20.76 b	14
	Straw	31.56	31.76	32.10	30.10	31.92	30.28	31.28 a	
Calcium	Grain	1.29	1.36	1.04	1.19	1.32	1.55	1.28 b	24
	Straw	4.82	5.25	5.16	5.10	5.54	4.70	5.10 a	
Magnesium	Grain	2.43	2.43	2.24	2.15	2.19	2.20	2.28 a	16
	Straw	1.78	1.66	1.74	1.52	1.61	1.59	1.65 b	
Sulfur	Grain	0.85	0.79	0.76	0.85	0.89	1.01	0.85 b	19
	Straw	1.48	1.48	1.37	1.40	1.46	1.33	1.42 a	

Means followed by the same lowercase letter, in the column, within the same nutrient, do not differ by Tukey's test at (5%) probability.

When comparing the tissues of the fruit, the grains showed higher concentrations of nitrogen, phosphorus and magnesium than straw. On the other hand, straw had a higher concentration of potassium, calcium and sulfur (Table 2). Also, regardless of the fertilizer dose used, it was possible to order in decreasing order the nutrients that appear in higher concentrations in the grains $\text{N} > \text{K} > \text{Mg} > \text{P} > \text{Ca} > \text{S}$ and, in the straw, $\text{K} > \text{N} > \text{Ca} > \text{Mg} > \text{S} > \text{P}$ (Table 2).

Dry mass accumulation and nutrient exports

The increase in the fertilizer doses promoted an exponential accumulation of dry mass, both in the grains and in the straw, with a trend of more pronounced increase under the smaller doses and less expressive gains under the high doses (Figure 2). However, when grains and straws were compared to each other, it was observed that there was no difference in the accumulation of dry mass (Table 3).

For the export of macronutrients by grain and straw, all nutrients studied showed exponential behavior, to the point of maximum, with a rapid increase under the lowest doses and with a tendency to stabilize under the highest doses, with the exception of Calcium (Ca) and Sulfur (S) that showed positive linear behavior (Figure 3).

Coffee trees exported higher amounts of nitrogen (N), phosphorus (P), magnesium (Mg) per grain, and higher amounts of potassium (K), calcium (Ca) and sulfur (S) through straw, regardless of the dose used, with exception of the zero dose. At this dose, there was no difference between grains and straw for the export of N, P, K, Mg and S. Regardless of the dose used, nutrient exports followed the decreasing order $\text{N} > \text{K} > \text{Mg} > \text{P} > \text{Ca} > \text{S}$ for grain and $\text{K} > \text{N} > \text{Ca} > \text{Mg} > \text{S} > \text{P}$ for straw (Table 3).

Discussion

Nutrient content

The lack of effect of fertilizer doses on nutrient levels in grains and straw (Figure 2) is related to the dilution effect of these macronutrients for the production of dry fruit mass (Covre et al. 2018). This is because the accumulation of biomass and the accumulation of nutrients are dependent factors (Prezotti and Bragança 2013).

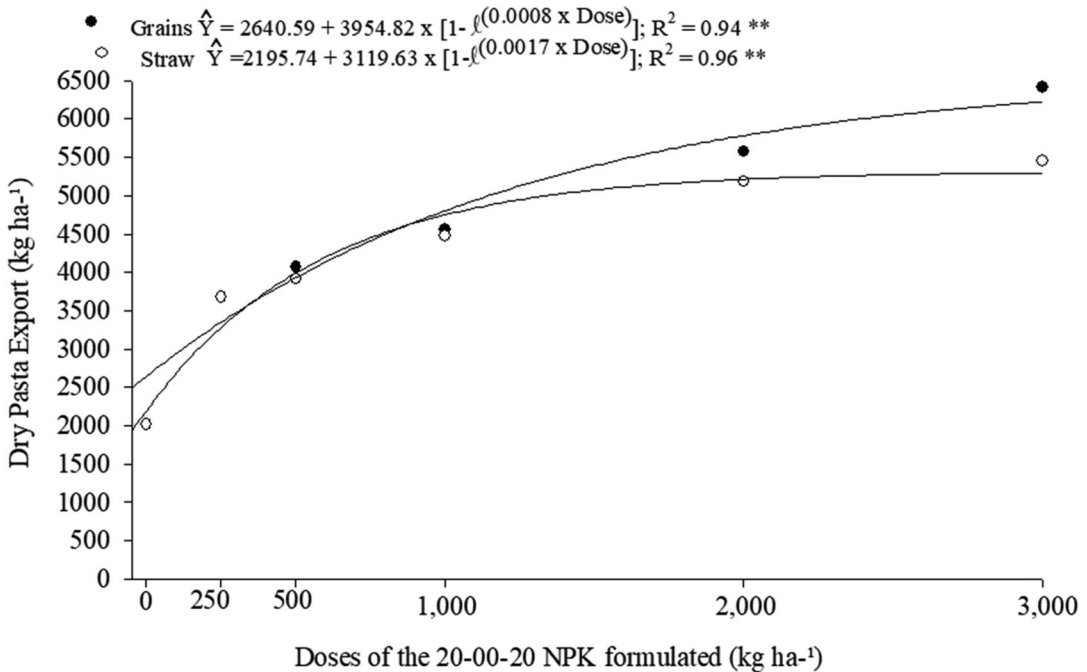


Figure 2. Accumulation of dry mass (kg hectare^{-1}) by grains and straw of *Coffea canephora* "Conilon - BRS Ouro Preto" as a function of six doses of the formulation 20-00-20. Porto Velho, 2015.

Table 3. Accumulation of dry mass and export of macronutrients (kg ha^{-1}), for the grains and straw of *Coffea canephora* "Conilon - BRS Ouro Preto," according to six doses of the formulation 20-00-20. Porto Velho, 2015.

Nutrient	Tissue	Dosage						Averages	CV%
		0	250	500	1,000	2,000	3,000		
		kg ha^{-1}							
Dry mass	Grain	2,286a	3,837a	4,066a	4,557a	5,571a	6,413a	4,455a	34
	Straw	2,019a	3,673a	3,912a	4,477a	5,182a	5,455a	4,120a	
Nitrogen	Grain	57.30a	98.48a	111.65a	121.53a	15967a	172.27a	121.73a	68
	Straw	37.94a	68.40b	81.30b	85.27b	102.13b	106.83b	79.30b	
Phosphorus	Grain	3.64a	3.45a	6.53a	6.17a	8.12a	11.66a	6.74a	50
	Straw	2.88a	4.33b	4.72b	4.28b	5.64a	5.66a	4.56b	
Potassium	Grain	48.62a	81.89b	86.52b	98.52b	115.44b	116.88b	91.31b	45
	Straw	63.73a	116.7a	125.60a	134.78a	165.41a	165.19a	130.04a	
Calcium	Grain	3.03b	4.98b	4.19b	5.55b	7.37b	8.95b	5.70b	59
	Straw	9.75a	17.23a	19.21a	20.36a	28.72a	32.37a	21.21a	
Magnesium	Grain	5.57a	9.33a	9.14a	9.83a	12.23a	14.11a	10.06a	45
	Straw	3.60a	6.11b	6.83b	7.43b	8.35b	8.67b	6.74b	
Sulfur	Grain	1.94a	3.04b	3.10b	3.90b	4.97b	6.52a	3.86b	48
	Straw	2.99a	5.44a	5.38a	6.30a	7.60a	7.28a	5.78a	

Averages followed by the same lowercase letter in the column, within each nutrient, do not differ by Tukey's test at (5%) probability.

For nutrients P, Ca, Mg and S the absence of an increase in the levels, due to the doses of the formulation 20-00-20, is justified by the absence of these nutrients in the fertilizer composition. On the other hand, a decrease in the concentration of these nutrients could be expected due to the increase in dry mass and, consequently, a dilution effect, which did not occur, indicating that the levels of these nutrients in the soil (Table 2) were sufficient to supply the demand of coffee trees.

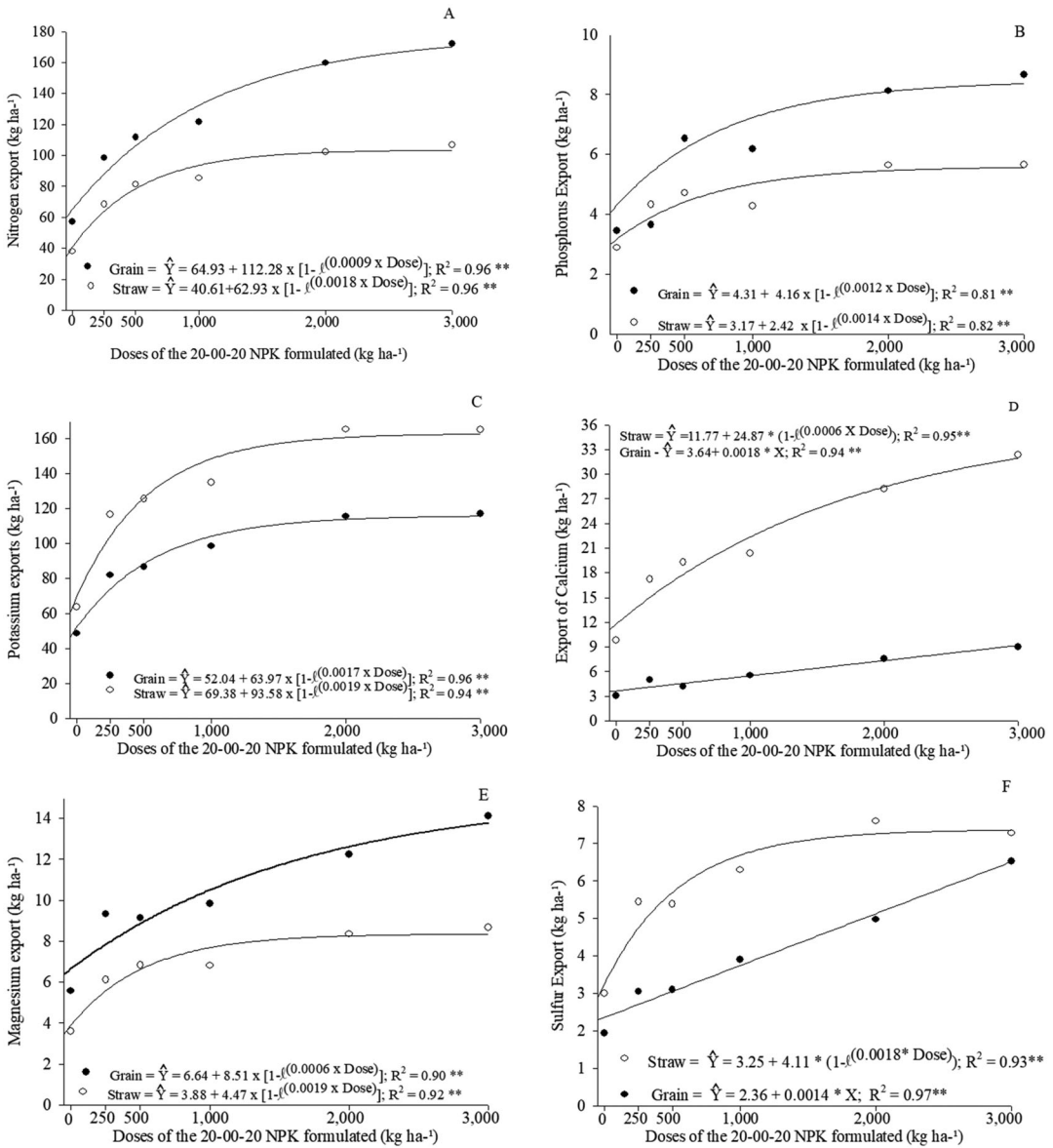


Figure 3. Exports of N, P, K, Ca, Mg and S by *Coffea canephora* “Conilon - BRS Ouro Preto” coffee beans and straw based on six doses of the formulation 20-00-20. Porto Velho, 2015.

The decreasing order of the concentration of nutrients in the grains N > K > Mg > P > Ca > S and, in the straw, K > N > Ca > Mg > S > P (Table 2) was similar to that observed by Covre et al. (2016) and is related to the metabolic functions of these macronutrients in each tissue.

N is essential for the plant because it participates in the constitution of membranes, in the processes of ionic absorption, photosynthesis, respiration, multiplication and cell differentiation (Taiz et al. 2017), presenting significant increases between the stages of rapid expansion, filling and maturation of conilon coffee fruits (Partelli et al. 2014; Covre et al. 2018). In addition, nitrogen is an element that makes up the structure of 11S globulin proteins, glyceraldehyde-3-phosphate dehydrogenase, LEA Dehydrin (Livramento et al. 2017), present in abundance in coffee

beans. Therefore, it is expected that with increasing doses of nitrogen fertilizers, the levels of these proteins in the grains will increase, as reported for *C. arabica* coffee (Reis et al. 2011) and *C. canephora* coffee (Perrois et al. 2015).

K is the second most required nutrient in the fruit formation process. It participates in important metabolic processes such as osmotic balance and metabolic reactions, activating various enzymes such as pyruvate kinase, phosphofructokinase and ADP-glucose. These enzymes, in turn, act in energy transport, protein synthesis and starch production (Pereira and Peres 2016), being freely present and with high mobility in tissues (K^+ ion) (Taiz et al. 2017). Its role is fundamental during the reproductive phase of the coffee tree, because during the formation of the fruits there is a decrease in the concentration of starch in the branches and leaves (Rena and Carvalho 2003), due to the intense drain to the fruits. This correlation between starch concentration and production shows the importance of K in coffee physiology (Bragança et al. 2007), especially for the activation of enzymes that are essential for the synthesis of organic compounds, during fruit ripening (Laviola et al. 2008).

P participates in the composition of carbohydrates, phospholipids that make up membranes, coenzymes, nucleoproteins and nucleic acids, ensuring membrane stability. In addition, it acts on vital plant processes such as photosynthesis and respiration due to the storage and transfer of energy in the form of ATP (adenosinatriphosphat) and in the molecular formation of DNA and RNA (Taiz et al. 2017). Thus, it is expected that this macronutrient will be highly required for the processes of cell differentiation, division and multiplication, as in the initial phase of rapid expansion in the coffee bean pellet stage (Cunha and Volpe 2011).

Ca is a macronutrient that structurally composes compounds such as pectate, carbonate, oxalate, phosphate, calmodulins and annexins; it acts in the enzymatic regulation of ATPase, alpha amylase, phospholipase, nucleases and polygalacturonase, as well as, it in the functioning of membranes, in the transport of auxins, abscission and senescence, among other metabolic processes (Li et al. 2009). Acquired initially by the roots, most of the Ca is transported in the xylem, although a small part can be transported through the phloem. When there are several drains involved, the redistribution of Ca in the plant becomes reduced due to its mobility (Bragança et al. 2007), however, the leaves have proportionally greater surface of exposure, thus, the Ca goes, preferably, to these organs, to the detriment of the fruits.

Mg has a fundamental role in photosynthesis, by modulating the activity of the enzyme RuBP carboxylase, which catalyzes carbon fixation. This element also plays a role in the structural stabilization of membranes and as a connecting element for the aggregation of ribosome subunits (Taiz et al. 2017).

S is a constituent of the amino acids cysteine and methionine, coenzymes, such as thiamin and biotin, proteins such as ferredoxins and thioredoxins, and sulfolipids, being essential for membrane activity and protein synthesis (Taiz et al. 2017).

Dry mass accumulation and nutrient exports

The exponential behavior observed, both for the accumulation of dry mass of grains and straw (Figure 2) suggests that the genetic differences of the genotypes that make up the variety “Conilon BRS - Ouro Preto” (Ramalho et al. 2014a) influence the response from coffee to fertilizer application. This is because genotypes that are part of the same variety may exhibit different behaviors as to the absorption and accumulation of nutrients in their tissues (Marré et al. 2015), which can guarantee greater production stability by area, in the different growing environments of the Amazon. However, the limited potential for individual response of some genotypes can compromise the response of the variety to the increase in fertilizer quantities.

In addition to plant genotypic differences, another factor that may have influenced the coffee plant's responses to fertilizer application is the spacing used between plants (Bragança et al. 2009). In this research, the spacing used for planting $3\text{ m} \times 2\text{ m}$, resulted in a density of only

1,666 plants, reaching a maximum production of 6,413 kg ha⁻¹ of dry mass for the grain and 5,455 kg ha⁻¹ of dry mass for the straw (Table 3).

For *C. canephora*, the spacing that promotes greater productivity is 2 × 1 (5,000 plants ha⁻¹) and 3 × 1 (3,333 plants ha⁻¹) (Verdin Filho et al. 2014). This is because, in dense crops, the increase in root density per area leads to a higher rate of nutrient recovery, especially for those with less mobility in the soil. In addition, the shading provided by the density promotes an increase in the water content in the soil, which contributes to more favorable dynamics of macro-nutrient transport in the soil, especially in the reproductive phase (Covre et al. 2018).

As for the increase in nutrient exports due to the doses of fertilizers, the exponential or linear behavior was related to the increase of dry mass accumulation, since the concentration of nutrients was not influenced by the doses of 20-00-20.

Following the decreasing sequence of nutrient exports by N > K > Mg > P > Ca > S grains and, by straw, K > N > Ca > Mg > S > P (Table 3), it is observed that the incorporation of straw in the soil would constitute an important organic source of some minerals, with emphasis on N and K. The use of straw can reduce the need for mineral nutrients in the field (Fernandes et al. 2013), thus reducing the production costs.

Conclusions

The increase in doses of the fertilizer formulation 20-00-20 does not promote changes in the concentrations of nutrients N, P, K, Ca, Mg and S in coffee fruits “Conilon - BRS Ouro Preto.” However, it promotes greater accumulation of dry mass, both in grains and straw and, consequently, greater export of nutrients by the fruits.

“Conilon - BRS Ouro Preto” coffee plants export more N, P and Mg for grains and more K, Ca and S for straw, regardless of the dose of fertilizer 20-00-20 used.

The macronutrients most exported by coffee fruits “Conilon - BRS Ouro Preto” are N > K > Mg > P > Ca > S, for grains and K > N > Ca > Mg > S > P for straw.

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