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# Variability and nutritional balance among genotypes of *Coffea canephora* (Rubiaceae) in drought versus adequate water supply

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**ABSTRACT.** We examined the effect of water availability on the nutritional balance of 15 genotypes of the clonal cultivar “Conilon BRS Ouro Preto” of *Coffea canephora* grown in two contrasting environments in terms of water availability. Biomass production and nutritional balance parameters, based on the deviance from the standard ratio among nutrients for the species, were estimated after 170 days of cultivation in these contrasting environments. The variability of responses among genotypes indicated a favorable for identifying diversity among these genotypes and for selection aiming to explore their nutritional parameters, especially for the concentration of phosphorus and magnesium in green tissues. Cultivation in the environments with low water supply caused losses up to 29% in the biomass production of the young plants (with most severe losses observed for the aerial part); the magnitude of these losses varied among genotypes. Genotypes 125 and 155 accumulated significantly higher amounts of biomass when compared to the others, regardless of the water supply. Overall, the nutritional indexes of the tested genotypes showed greater metabolic inflexibility towards water stress than what is reported for other cultivars of Robusta coffee.

**Key words**: Coffee; mineral nutrition; water deficit; diversity

## INTRODUCTION

Agriculture is constantly challenged by abiotic and biotic stresses that often negatively influence crops, generally causing unwanted modifications in plant growth, metabolism and crop yield (Pinto, et al., 2008). Among these stresses, the losses caused to water deficit have been the main challenge, since water is one of the most limiting inputs for plant growth and development worldwide (Cavatte et al., 2011).

The magnitude of the changes caused by water deficit in plant metabolism and growth is a result of complex interactions between several factors, including genotype, the duration and severity of the deficit, and the phenological stage of the plant (Santos and Carlesso, 1998).

Among the several improved genotypes of *Coffea canephora* that are currently recommended and cultivated, there are some genetic materials that are able to survive and yield satisfactorily even under moderate water stress conditions (Ferrão et al., 2000; DaMatta, 2004). Tolerance to water stress naturally occurs in almost all species, but its extent varies from species to species and even within species (Markesteijn, 2010). This tolerance usually results from integrated factors at all organizational levels: anatomical, morphological, cellular, biochemical and molecular, such as inhibition of leaf expansion, foliar abscission, enhanced root growth and stomatal regulation (Taiz and Zeiger, 2013).

Global climate change and the increased rate of events of climatic extremes such as prolongated droughts(Assad et al., 2004; Bunn et al., 2015),has increased the importance of efforts to find genotypes that are more likely to tolerate water stress. The development of improved cultivars with tolerance to water deficit should be a priority for coffee breeding and research (Marraccini et al., 2012).

Availability of water is a major factor in the nutritional balance in plants. Water is required for the normal course of all metabolic processes in plants and regulates the absorption of mineral nutrients from the soil, translocation among the organs, gas exchange and metabolic activity (Martins et al., 2014). Therefore, changes in the water supply can modulate the acquisition and use of nutrients, having potential effects on the overall nutritional balance of the plant.

In the coming years, studies aimed at optimizing water use will be of utmost importance for the maintenance of cultivation sustainability. Along this line, this experiment aimed to examine how water supply affects nutritional balance in various genotypes of *C. canephora*.

## MATERIAL AND METHODS

### Local setup and experimental design

The experiment was developed in a greenhouse installed in the municipality of Ouro Preto do Oeste, Rondônia State, in northern Brazil. This site has an elevation of 256 m above sea level; the average air temperature of the region ranges from 24°C minimum to 32°C maximum; the average air humidity is 85% and the annual accumulated rainfall is 2,250 mm. The climate is classified as equatorial tropical (warm and humid), being mostly rainy, with a dry season from June to September.

Plastic pots of 18 liters volume were prepared to receive the plants in the greenhouse and were filled with 10 kg of soil. The soil was collected from a field covered by native vegetation, at a depth of 10-20 cm, dried in the shade, homogenized with a 4.0 mm mesh sieve and separated in samples of 10 kg to fill each pot. A representative sample of this soil was analyzed to determine its physical-chemical attributes (Embrapa, 1997), which was classified as an Oxisol (red-yellow latosol) and medium texture (33% clay, 15% silt and 52% sand). The soil presented 1.21 g $cm^{-3}$ of density (graduated cylinder method), pH 5.9 (water, 1:2.5 ratio), 4.00 mg $dm^{-3}$ phosphorus (extracted by Mehlich-1), 120.90mg $dm^{-3}$potassium (extracted by Mehlich-1), 4.25 $cmol_c$ $dm^{-3}$ calcium (extracted by potassium chloride at 1 mol $L^{-1}$), 1.33 $cmol_c$ $dm^{-3}$ f magnesium (extracted by potassium chloride at 1.0 mol $L^{-1}$) and 3.30$cmol_c$ $dm^{-3}$ potential acidity (extracted by calcium acetate at 0.5 mol $L^{-1}$, ph 7.0).

The experiment followed a factorial scheme 15x2, studying 15 improved genotypes of *C. canephora* and two modified environments for water supply levels. It followed a completely randomized design, with four repetitions and one plant per pot as the experimental parcel. The two modified environments were prepared to configure different levels of water availability for the plants.

## Selected genotypes

The 15 genotypes used in the experiment (referred to as 56, 57, 61, 73, 88, 89, 120, 125, 130, 155, 160, 184, 189, 199 and 203) compose the entire group of genotypes from the clonal cultivar “Conilon BRS Ouro Preto” (SNPC Certification number: 20130061), which is recommended for planting in Rondônia State.

The genotypes were asexually multiplied using cuttings from matrix plants, which were cultivated in a multiplication field, being similar in age, growth, nutritional and phytosanitary status. The cuttings were prepared and cultivated in a nursery until the stage of four fully developed pairs of leaves. The grown plantlets were then moved to the prepared pots to compose the experimental plots.

## Water supply

A hydro-physical analysis was made of the soil, according to the methodology proposed by Embrapa (1997), which established the soil moisture at the field capacity (tension of 10 kPa) as 23.73% and at permanent wilting point (tension of 1,500 kPa) as 15.74%. These results were used to determine the total available water in the soil and to manage the irrigation, following the methodology described by Bernardo et al. (2008).

The two levels of water supply used in the experiment were established aiming to allow the plants to have access to contrasting conditions of water availability. The level of 100% of available water in the soil was used as the standard reference. The depletion of water in each pot was monitored daily by weighing the pots with an analytical scale (BPW Platform, precision: 1 g), allowing this depletion to reach different levels in accordance with the level established for the treatments. The plants selected to be kept in conditions of higher water supply were irrigated daily to return the water availability in the soil to 100% constantly. The soil from the parcels selected to be cultivated with low water supply was allowed to deplete down to the level of 25% of water availability before the irrigation returned it to the reference level of 100%.These conditions for irrigation started at the 50th day after transplanting the coffee plantlets to the pots and were maintained until the 170th day.

## Cultivation and nutritional management

The physical-chemical analyses were used to establish fertilization management. The fertility of the soil was previously adjusted according to adequate levels of each nutrient for the early growth of coffee plants; nitrogen was supplied in four parcels (every 25 days after the first month); phosphorus and potassium were supplied in a single application before transplanting the plantlets to the pots. The nutritional levels of the fertilization were established based on the recommendation for controlled environments (Novais et al., 1991).

The surface of the soil of each pot was protected and covered with an expanded polystyrene layer in order to avoid water losses by evaporation. The plants were cultivated using the common practices established in accordance with their eventual need and following current recommendations for the cultivation of Conilon coffee in Brazil (Ferrão et al., 2017).

## Biomass and nutritional parameters

After 170 days of cultivation, the plant organs (leaves, stems and roots) were cut and separated in paper bags, which were then dried in a laboratory oven (STF SP-102/2000 CIR, with forced air circulation at 65°C), until their masses achieved a constant weight, monitored using a laboratory scale (SHIMADZU AUW-220D; precision: 0.00001 g). The final weight was used to determine the biomass of each organ, expressed as dry matter of roots, leaves, aerial part (steam + leaves) and total (aerial part + roots).

After drying, the dry matter of leaves was triturated (CIENLAB EC-430, 8 blades, 1,725 rpm, 20 mesh) to obtain a homogeneous powder. Triplicate samples of the powder were used to determine the concentration of each nutrient in the leaf tissues.

To quantify the N content, 0.5 g (+/-0.001g) of the powder were transferred to Taylor tubes (25 mm x 200 mm) and submitted to the stages of sulfuric digestion ($H_2SO_4$), distillation (NaOH 40%) and titration (NaOH 0.02 mol $L^{-1}$) of nitrogen in a distiller (Marconi MA-036), according to the Kjeldahl method (Ma and Zuazaga, 1942).

To quantify P, K, Ca, Mg and S contents, 0.5 g (+/-0.001g) of the powder was transferred to Taylor tubes (25 mm x 200 mm) and submitted to the stages of nitric-percloric acid digestion ($HNO_3$, 65%and $HClO_4$, 70%) in a digestion block (Tecnal, TE-007D) at 180-190°C for 3 hours; then, 3 mL of ascorbic acid ($C_6H_8O_6$, 0.87M) was added and the determination was made by spectrophotometry (Femto, 700 Plus) (Embrapa, 1997).

The results of biomass and nutritional contents were used to establish nutritional indexes, calculated based on the methods proposed by Martins et al. (2014). The ratios among nutrients were compared with standards to examine the nutritional balance (Malavolta, 1996; Bragança et al., 2007).

## Data analyses

The data were subjected to analysis of variance, using the F-test to identify the interactions and differences among levels of each factor. The genetic parameters for each variable were estimated according to the methodology described by Cruz and Carneiro (2003). The model used was:

$$Y_{ijk}=\mu+G_i+E_j+GE_{ij}+\varepsilon_{ijk} \qquad \text{(Equation 1)}$$

where $Y_{ijk}$ represents the phenotypic value of the ijk$^{th}$ observation, μ is the general mean, $G_i$ is the fixed effect of the i$^{th}$ genotype, $E_j$ is the random effect of the j$^{th}$ environment (available water in the soil), $GE_{ij}$ is the effect of the interaction between the i$^{th}$ genotype and the j$^{th}$ environment, and $\varepsilon_{ij}$ is the random error related to the ij$^{th}$ observation.

The means of genotypes were analyzed using the Scott-Knott criterium and the means of water supplies were studied using the Tukey criterium (both at 5% probability). The genetic parameters for each variable were estimated using the methods described by Cruz and Carneiro (2003), whereby the values for quadratic component associated with the expressed phenotype as a result of the interaction genotype-environment ($\hat{\sigma}^2_{ge}$), the environmental variance component ($\hat{\sigma}^2_e$), the genotypic variance component ($\hat{\phi}_g$), the genotypic determination coefficient ($H^2$), the genetic variation coefficient ($CV_g$), and the variation index ($CV_g/CV$) were estimated. The analyses were performed using the statistical software GENES (Cruz, 2013).

# RESULTS

## Genetic parameters

Significance of interactions between the effects of genotypes and water supply were examined by the F test for all traits. The estimated genetic parameters are presented in Table 1.

**Table 1.** Genetic parameter estimates for nutritional indexes of genotypes of *Coffea canephora* grown with different levels of water supply.

| Parameter | N | P | K | Ca | Mg | S | TDM |
|---|---|---|---|---|---|---|---|
| $\hat{\phi}_g$ | 4.36 | 0.14 | 5.37 | 1.66 | 0.59 | 0.18 | 52.47 |
| $\hat{\sigma}^2_{ge}$ | 0.43 | 0.03 | 2.86 | 0.49 | 0.22 | 3.54 | 86.59 |
| $\hat{\sigma}^2_e$ | 15.18 | 0.19 | 11.54 | 3.78 | 0.82 | 4.33 | 283.40 |
| $H^2$ | 67.16 | 78.26 | 64.33 | 69.33 | 72.76 | 7.04 | 39.07 |
| $CV_g$ | 7.61 | 16.66 | 7.39 | 9.73 | 12.90 | 2.75 | 5.40 |
| $CV_g/CV$ | 0.54 | 0.85 | 0.68 | 0.66 | 0.85 | 0.21 | 0.43 |

$\hat{\phi}_g$: genotypic quadratic component; $\hat{\sigma}^2_{ge}$: variance component associated to interaction between genotype and environment; $\hat{\sigma}^2_e$: environmental variance component; $H^2$:coefficient of genotypic determination (%); $CV_g$: coefficient of genetic variation; $CV_g/CV$: variation index TDM: total dry matter.

The estimated values of genotypic quadratic components ($\hat{\phi}_g$) surpassed the estimated values of the component associated with the interaction between genotype and environment ($\hat{\sigma}^2_{ge}$) for most variables, which only was not observed for sulfur content and the accumulation of dry matter. The higher environmental effect in the determination of these variables can also be seen in the lower values of coefficient of genotypic determination ($H^2$). For N, P, K, Ca and Mg, the coefficients of genotypic determination were higher than 64%, which combined with the estimated values of variation indexes (ranging from 0.54 to 0.85), indicate a favorable condition for identifying diversity among these genotypes and for a possible selection aiming to explore their nutritional status, especially for phosphorus and magnesium.

## Biomass production

Different means for both accumulation and allocation of biomass among the plant organs were observed for the genotypes, as well as significant differences caused by the changes in the water supply (Table 2).

**Table 2.** Dry matter accumulated in roots (RDM), leaves (LDM), aerial part (ADM) and total (TDM) of 15 genotypes of *Coffea canephora* (cultivar "Conilon – BRS Ouro Preto"), after 170 days of cultivation with different levels of water supply (100% and 25% of the available water in the soil, referred to as $AW_{100\%}$ and $AW_{20\%}$, respectively).

| **Genotype** | **RDM (g)** | | **LDM (g)** | | **ADM (g)** | | **TDM (g)** | |
|---|---|---|---|---|---|---|---|---|
| | **$AW_{100\%}$** | **$AW_{25\%}$** | **$AW_{100\%}$** | **$AW_{25\%}$** | **$AW_{100\%}$** | **$AW_{25\%}$** | **$AW_{100\%}$** | **$AW_{25\%}$** |
| 56 | 37.93 dA | 38.47 cA | 53.70 bA | 40.63 bB | 90.95 bA | 73.77 bB | 128.91 cA | 112.23 cB |
| 57 | 46.38 cA | 42.81 cA | 62.92 aA | 50.40 aB | 104.53 aA | 79.30 bB | 148.06 bA | 125.06 bB |
| 61 | 51.08 bA | 58.76 aA | 48.33 cA | 45.17 bA | 99.86 aA | 86.10 aB | 134.06 cA | 141.44 aA |
| 73 | 64.73 aA | 40.95 cB | 48.27 cA | 43.25 bB | 100.57 aA | 79.45 bB | 166.96 aA | 117.67 cB |
| 88 | 36.70 dA | 30.32 dA | 55.80 bA | 48.24 aB | 98.90 aA | 85.30 aB | 134.03 cA | 115.79 cB |
| 89 | 50.63 bA | 46.08 bA | 51.57 cA | 51.93 aA | 92.23 bA | 93.75 aA | 138.46 cA | 145.82 aA |
| 120 | 53.33 bA | 47.74 bA | 57.65 bA | 48.03 aB | 108.78 aA | 90.43 aB | 158.24 bA | 138.18 aB |
| 125 | 60.95 aA | 56.23 aA | 55.83 bA | 45.73 bB | 104.03 aA | 88.51 aB | 167.54 aA | 144.74 aB |
| 130 | 54.37 bA | 48.90 bA | 55.65 bA | 48.00 aB | 95.95 aA | 87.33 aB | 151.53 bA | 140.53 aA |
| 155 | 62.55 aA | 48.69 bB | 49.07 cA | 49.27 aA | 86.83 bA | 81.67 bA | 164.49 aA | 132.35 aB |
| 160 | 62.59 aA | 53.11 aA | 54.10 bA | 42.30 bB | 100.70 aA | 80.45 bB | 160.62 aA | 126.48 bB |
| 184 | 58.43 aA | 45.22 bB | 34.55 dA | 39.62 bA | 70.65 cA | 71.43 bA | 131.30 cA | 119.79 cB |
| 189 | 49.74 bA | 45.80 bA | 49.63 cA | 48.30 aA | 82.48 bA | 84.03 aA | 136.23 cA | 128.61 bA |
| 199 | 41.05 dA | 42.01 cA | 51.43 cA | 43.37 bB | 89.05 bA | 78.02 bB | 131.09 cA | 118.61 cB |
| 203 | 54.36 bA | 46.68 bB | 60.05 aA | 50.17 aB | 96.18 aA | 87.84 aA | 154.85 bA | 136.78 aB |

Means followed by the same uppercase letter in the row or lowercase in the column do not differ by Tukey or Scott-Knott tests, respectively, at 5% probability.

For most genotypes, the plants from the environment with decreased water supply ($AW_{25\%}$) presented lower growth rates and therefore lower means of accumulation of biomass. However, this limitation in growth was less pronounced for root biomass, for which only genotypes 73, 155, 184 and 203 differed from the environment with high water supply ($AW_{100\%}$), presenting losses of approximately 37%, 22%, 23% and 14%, respectively.

Regarding the differences in the accumulation of biomass in the root system, it was possible to identify four homogeneous groups of genotypes for both conditions of water supply. Genotypes 125 and 160 presented higher means of root biomass regardless of the level of available water in the soil, while genotype 88 presented the lowest means for root dry matter under both conditions (Table 2).

For leaf dry matter, a negative influence of restriction in the water supply was observed in most genotypes, resulting in smaller leaves for treatments with depletion of water to 25% of available water in the soil, with losses of between 10% and 24%. Only genotypes 61, 89, 155, 184 and 189 maintained similar leafiness regardless of the water supply.

The limitation of water supply decreased differentiation among genotypes; while it was possible to identify four different groups of homogeneous genotypes under the condition of higher supply, it was possible to observe only two groups when the soil was allowed to deplete to 25% of available water. Genotypes 57 and 203 stood out for developing the highest leaf biomass regardless of the availability of water, while genotype 184 presented lower means under both conditions (Table 2).

Accumulation of biomass in the aerial part of genotypes 89, 155, 184, 189 and 203 did not differ with the change in water supply. For the others, the lower water supply caused losses between 9 and 24% in the accumulation of biomass in the aerial part. Similar to leaf biomass, the lower water supply also caused a decrease in the observable differences among genotypes for ADM. While three different groups were observed under the condition of higher water supply, only two were differentiable with low water supply. Genotype 184 also presented the lowest accumulation of biomass in the aerial part under both conditions of water supply; while genotypes 61, 88, 120, 125, 130 and 203 grouped together with higher shoot biomass regardless of the level of water supply (Table 2). Overall, the limitation caused by the restriction in water supply caused losses between 8% and 29%in total dry matter; this negative effect was only not observed for genotypes 61, 89, 130 and 189.

Three homogeneous groups of genotypes were observed in both environments. Genotypes 125 and 155 stood out in terms of accumulation of high total biomass under both conditions of water supply, while genotypes 56, 88, 184 and 199 presented lower overall biomass production regardless of the level of available water in the soil (Table 2). Genotype 73 was highly responsive to water supply, since although it belongs to the group with the lowest biomass accumulation when cultivated with low water supply, this genotype responded to the higher water supple with a gain of 42% in its total biomass.

## Nutritional status

There were significant differences in the concentration of primary macronutrients in the green tissues among genotypes and between conditions of water supply. The interaction between the effects of these factors was unfolded and is presented in Table 3.

Nitrogen concentration in leaf tissues was not influenced by alteration of the water supply for most genotypes; however, a higher concentration was observed for the conditions of lower water supply for genotypes 73 and 203, which presented gains of 26 and 34% of their nitrogen content, respectively. Limitation of the water supply decreased the differentiation among genotypes; while it was possible to identify two homogeneous groups in the environment with high supply, there was no differentiation among genotypes in the environment with low supply. Genotypes 56, 61, 88, 155, 184, 189 and 199 stood out for having the highest concentrations of nitrogen in their green tissues when the available water was near 100% (Table 3).

For phosphorus, limitation in the water supply only caused a negative effect in the concentration of this nutrient in the leaves of genotypes 88 and 160, with losses ranging from 30 to 35%. It was possible to differentiate two homogeneous groups of genotypes in each environment for phosphorus content. Genotypes 120, 160 and 199 presented the highest concentrations of phosphorus in their tissues under both conditions of water supply (Table 3).

**Table 3.** Concentration of primary macronutrients in leaf tissues of 15 genotypes of *Coffea canephora* (cultivar "Conilon – BRS Ouro Preto"), after 170 days of cultivation with different levels of water supply (depletion to 100% and 25% of the available water in the soil, referred to as $AW_{100\%}$ and $AW_{20\%}$, respectively).

| Genotype | N (g kg$^{-1}$) | | P (g kg$^{-1}$) | | K (g kg$^{-1}$) | |
|---|---|---|---|---|---|---|
| | $AW_{100\%}$ | $AW_{25\%}$ | $AW_{100\%}$ | $AW_{25\%}$ | $AW_{100\%}$ | $AW_{25\%}$ |
| 56 | 28.57 aA | 31.35 aA | 1.54 bA | 2.14 bA | 25.05 cB | 30.67 aA |
| 57 | 24.70 bA | 29.39 aA | 1.94 bA | 1.65 bA | 29.91 cA | 27.04 bA |
| 61 | 30.85 aA | 27.66 aA | 2.01 bA | 2.10 bA | 32.25 bA | 25.76 bB |
| 73 | 25.15 bB | 31.81 aA | 2.45 bA | 2.58 aA | 29.47 cA | 26.03 bA |
| 88 | 27.89 aA | 27.02 aA | 2.34 bA | 1.53 bB | 34.57 bA | 29.77 aA |
| 89 | 24.79 bA | 25.56 aA | 2.08 bA | 2.10 bA | 34.10 bA | 32.10 aA |
| 120 | 23.46 bA | 27.89 aA | 2.99 aA | 2.71 aA | 33.56 bA | 30.31 aA |
| 125 | 21.32 bA | 23.42 aA | 2.53 bA | 2.49 aA | 32.10 bA | 29.22 aA |
| 130 | 23.74 bA | 27.71 aA | 2.36 bA | 1.98 bA | 31.05 cA | 28.19 bA |
| 155 | 28.48 aA | 28.80 aA | 1.98 bA | 2.04 bA | 32.86 bA | 31.51 aA |
| 160 | 22.81 bA | 27.16 aA | 3.49 aA | 2.43 aB | 34.46 bA | 30.58 aA |
| 184 | 28.16 aA | 28.16 aA | 2.44 bA | 2.05 bA | 30.32 cA | 30.31 aA |
| 189 | 30.21 aA | 30.53 aA | 1.88 bA | 1.78 bA | 32.79 bA | 30.75 aA |
| 199 | 31.08 aA | 33.54 aA | 3.23 aA | 2.93 aA | 44.57 aA | 33.70 aB |
| 203 | 22.05 bB | 29.58 aA | 2.04 bA | 1.95 bA | 34.99 bA | 32.77 aA |

Means followed by the same uppercase letter in the row or lowercase in the column do not differ by Tukey or Scott-Knott tests, respectively, at 5% probability.

Losses were also observed for potassium content in genotypes 61 and 199, which had concentrations 20-24% lower when cultivated with low water supply. For most other genotypes, the potassium content was not modified by the change in the water supply. However, genotype 56 presented a gain of 22% in the potassium concentration with the restriction in available water. Three homogeneous groups were differentiated for potassium content in the environment with high water supply, while only two were observed for the plants subjected to low supply. Genotype 199 stood out in both environments, presenting a higher concentration of potassium in their leaves (Table 3). The concentration of secondary macronutrients seemed to be less affected by the change in the water supply, as most genotypes presented similar contents for calcium, magnesium and sulfur in both environments (Table 4).

The condition of low water supply altered the concentration of calcium only for genotypes 88 and 184, causing losses of 22 and 25%, respectively. Two groups of homogeneous means were identified for both environments; however, the genotypes that formed these groups differed depending on the water supply condition. Genotypes 89, 184 and 189 stood out for being clustered in the groups of higher content under both conditions (Table 4).

Genotypes 56, 88 and 155 presented changes in the magnesium content depending on the conditions of water depletion, with a gain of 32% for genotype 88 and losses of 25-28% for genotypes 56 and 155 with increased water supply. The genotypes were clustered in three groups in the environment with high water supply and two when cultivated with low water supply. Genotypes 56, 61, 130, 135, 184 and 203 were grouped in the lowest groups of concentration of this nutrient, regardless of the water supply; while genotype 125 presented higher contents under both conditions (Table 4).

**Table 4.** Concentration of secondary macronutrients in leaf tissues of 15 genotypes of *Coffea canephora* (cultivar "Conilon – BRS Ouro Preto"), after 170 days of cultivation with different levels of water supply (depletion to 100% and 25% of the available water in the soil, referred to as $AW_{100\%}$ and $AW_{20\%}$, respectively).

| Genotype | Ca (g kg$^{-1}$) | | Mg (g kg$^{-1}$) | | S (g kg$^{-1}$) | |
|---|---|---|---|---|---|---|
| | $AW_{100\%}$ | $AW_{25\%}$ | $AW_{100\%}$ | $AW_{25\%}$ | $AW_{100\%}$ | $AW_{25\%}$ |
| 56 | 10.50 bA | 12.82 bA | 5.02 cB | 6.42 bA | 13.03 bB | 18.65 aA |
| 57 | 13.36 aA | 13.33 bA | 6.14 bA | 5.35 bA | 14.09 bA | 12.55 cA |
| 61 | 12.43 bA | 11.85 bA | 4.36 cA | 5.20 bA | 18.97 aA | 13.91 cB |
| 73 | 11.45 bA | 12.89 bA | 6.35 bA | 6.06 bA | 13.46 bA | 12.61 cA |
| 88 | 14.35 aA | 11.25 bB | 7.42 aA | 5.56 bB | 13.75 bB | 18.86 aA |
| 89 | 14.91 aA | 16.16 aA | 6.05 bA | 5.70 bA | 15.15 bA | 14.22 cA |
| 120 | 12.16 bA | 12.46 bA | 6.07 bA | 5.30 bA | 14.54 bB | 17.72 aA |
| 125 | 13.61 aA | 12.99 bA | 8.66 aA | 8.58 aA | 16.11 bA | 13.56 cA |
| 130 | 11.62 bA | 11.34 bA | 4.76 cA | 5.08 bA | 13.75 bA | 15.74 bA |
| 155 | 11.84 bA | 10.82 bA | 4.49 cB | 5.93 bA | 14.46 bA | 16.21 bA |
| 160 | 15.50 aA | 14.02 bA | 5.98 bA | 6.80 bA | 14.20 bB | 20.45 aA |
| 184 | 13.76 aB | 17.23 aA | 5.58 cA | 6.72 bA | 14.62 bA | 15.97 bA |
| 189 | 15.67 aA | 15.52 aA | 6.40 bA | 6.18 bA | 16.85 aA | 17.70 aA |
| 199 | 15.84 aA | 13.29 bA | 6.35 bA | 5.60 bA | 19.39 aA | 19.47 aA |
| 203 | 12.25 bA | 12.29 bA | 5.11 cA | 5.53 bA | 13.83 bA | 15.74 bA |

Means followed by the same uppercase letter in the row or lowercase in the column do not differ by Tukey or Scott-Knott tests, respectively, at 5% probability.

Sulfur concentration in the leaf tissues was only affected by the restriction of water supply for genotypes 56, 61, 88 and 160. Among these, genotypes 56, 88 and 160 presented gains in the nutritional content from 27 to 44%, while genotype 61 presented a loss of 27% when the water supply was increased. Two homogeneous groups were identified for the environment with high water supply and three for the environment with low supply. Genotypes189 and 199 were in the groups of higher sulfur concentration under both conditions of available water (Table 4).

## Nutritional balance

There was a high degree of similarity among genotypes regarding the direction of the effect of water supply on the nutritional balance (Table 5). The only divergent effects were observed for N/P ratio of genotypes 56, 57, 88 and 189; and for K/Mg ratio of genotypes 61, 130, 155 and 189.

In genotypes 57, 88 and 189, there was a negative imbalance of the N/P ratio under adequate water supply and a positive imbalance under water deficit. For the genotype 56, the N/P ratio suffered a negative imbalance in response to water deficit, and a similar behavior was observed for genotypes 61, 130, 155 and 189 for the K/Mg ratio.

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**Table 5.** Differences in the ratio between the nutritional contents of reference plants and the contents in leaf tissues of 15 genotypes of *Coffea canephora* (cultivar "Conilon – BRS Ouro Preto"), after 170 days of cultivation with different levels of water supply (depletion to 100% and 25% of the available water in the soil, referred to as AW$_{100\%}$ and AW$_{20\%}$, respectively).

| **Geno-type** | **N/P** | | **N/K** | | **N/S** | | **K/Ca** | | **K/Mg** | | **Ca/Mg** | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **AW$_{100\%}$** | **AW$_{25\%}$** | **AW$_{100\%}$** | **AW$_{25\%}$** | **AW$_{100}$ %** | **AW$_{25}$ %** | **AW$_{100}$ %** | **AW$_{25}$ %** | **AW$_{100}$ %** | **AW$_{25}$ %** | **AW$_{100}$ %** | **AW$_{25}$ %** |
| 56 | 1.56 | -2.35 | -0.21 | -0.33 | -14.81 | -15.32 | 0.48 | 0.49 | -0.39 | -2.14 | -4.96 | -5.05 |
| 57 | -4.29 | 0.80 | -0.52 | -0.26 | -15.25 | -14.66 | 0.34 | 0.13 | -1.10 | -1.53 | -4.87 | -4.56 |
| 61 | -1.66 | -3.80 | -0.39 | -0.28 | -15.37 | -15.01 | 0.69 | 0.27 | 0.41 | -1.21 | -4.20 | -4.77 |
| 73 | -6.73 | -4.33 | -0.50 | -0.17 | -15.13 | -14.51 | 0.67 | 0.07 | -0.91 | -1.55 | -5.25 | -4.86 |
| 88 | -5.09 | 0.69 | -0.54 | -0.44 | -14.97 | -15.57 | 0.51 | 0.75 | -1.75 | -0.57 | -5.11 | -5.03 |
| 89 | -5.11 | -4.80 | -0.62 | -0.55 | -15.36 | -15.20 | 0.39 | 0.09 | -0.80 | -1.03 | -4.58 | -4.21 |
| 120 | -9.16 | -6.72 | -0.65 | -0.43 | -15.39 | -15.43 | 0.86 | 0.53 | -1.06 | -0.84 | -5.05 | -4.70 |
| 125 | -8.57 | -7.59 | -0.69 | -0.55 | -15.68 | -15.27 | 0.46 | 0.35 | -2.77 | -3.06 | -5.48 | -5.54 |
| 130 | -6.94 | -2.99 | -0.59 | -0.37 | -15.27 | -15.24 | 0.77 | 0.59 | 0.56 | -0.15 | -4.61 | -4.82 |
| 155 | -2.64 | -2.88 | -0.48 | -0.44 | -15.03 | -15.22 | 0.88 | 1.01 | 1.33 | -1.20 | -4.41 | -5.23 |
| 160 | -10.47 | -5.81 | -0.69 | -0.46 | -15.39 | -15.67 | 0.32 | 0.28 | -1.28 | -1.89 | -4.46 | -4.99 |
| 184 | -5.44 | -3.26 | -0.42 | -0.42 | -15.07 | -15.24 | 0.30 | 0.14 | -0.47 | -1.77 | -4.58 | -4.49 |
| 189 | -0.94 | 0.18 | -0.43 | -0.36 | -15.21 | -15.27 | 0.19 | 0.08 | 0.62 | -0.90 | -4.60 | -4.54 |
| 199 | -7.38 | -5.56 | -0.65 | -0.35 | -15.40 | -15.28 | 0.91 | 0.64 | -0.84 | -0.50 | -4.56 | -4.68 |
| 203 | -6.22 | -1.83 | -0.72 | -0.45 | -15.41 | -15.12 | 0.96 | 0.77 | -0.35 | -6.35 | -4.65 | -4.83 |

Approximate values considered globally adequate for coffee (Malavolta, 1996; Bragança et al., 2007).

## DISCUSSION

High phenotypic and genotypic variability in populations of *C. canephora* are commonly observed (Fonseca et al., 2006; Ferrão et al., 2008; Rodrigues et al., 2012), due to its natural mechanism of self-incompatibility (Lashermes et al., 1996). Various studies show high genetic diversity for growth, biomass and mineral nutrition among improved genotypes of *C. canephora*, with different magnitudes of expression in environments with different levels of nutritional stresses (Colodetti et al., 2014; Martins et al., 2013; 2016).

The efficiency of the morphological and physiological mechanisms that determine the absorption and utilization of nutrients by plants is influenced by the expression of genetically controlled traits, and heterogenic behavior among genotypes of coffee is widely reported (Carelli et al., 2006; Martins et al., 2013; 2015a; 2015b; Rodrigues et al., 2015).

The significant differences observed among genotypes exposed to the same level of water availability in the soil, or even the same genotype under different water supplies, can be a function of the high genetic diversity observed among populations of *C. canephora*; for example, some genotypes may have alleles linked to drought tolerance (Souza et al., 2015).

Considering that tolerance to water stress was not the main selection criterion of genotypes to compose the clonal cultivar "Conilon BRS Ouro Preto", the existence of different behaviors among the genotypes is expected. The expression of different levels of tolerance, which is a result of intrinsic physiological responses of these genotypes, could be related to different stomatal control, photosynthetic rates, changes in leafiness, canopy architecture, or even the efficiency to absorb and use nutrients, among other traits (DaMatta, 2004).

There was lower allocation of biomass in the root system compared to the aerial part and, for most genotypes, the effects of the restriction of the water supply caused greater losses in the biomass of leaves and steams. The continued growth of root system combined with the reduced growth of leaves is a strategy to decrease the water loss by transpiration, which can be developed under conditions of water deficit; this has been observed in several coffee genotypes (DaMatta et al., 2004; Dominghetti et al., 2016).

There were significant changes in the concentration of nutrients in the biomass of many genotypes caused by the imposition of different water availabilities. Since water is a major factor in the absorption, transport and use of nutrients, this is a common relation that has been described in several papers involving interactions between the availability of water and mineral nutrition (Rezende et al., 2010; Dominghetti et al., 2014).

The environment with low water supply even promoted a higher concentration of N, K, Ca, Mg and Sin the leaves of some genotypes (56, 73, 88, 155, 160, 184 and 203). However, these genotypes also presented a decrease in total dry matter production (Table 2). This increase in nutritional content may be related to a concentration of the nutrient in the leaves (Maia et al., 2005; Carmo et al., 2011), as small amounts of the nutrient being translocated to the leaves (stronger metabolic sink) may cause a higher final concentration in the smaller leaves (lower biomass).

In other cases, the low water supply had the opposite effect, decreasing the nutritional content of P, K, Ca, Mg and S of some genotypes (61, 88, 160 and 199). The fact that some nutrients in the soil are mainly transported towards the roots by means of mass flow or arrive through a diffusion process makes the phenomenon entirely dependent on a liquid medium for the movement to occur, which is the water present in the soil (Barber, 1995; Matiello et al., 2009).

Overall, most nutritional content ratios for the genotypes of the cultivar "Conilon BRS Ouro Preto" presented negative values, which indicates, for example (considering the N/P, N/K and N/S ratios), that it is necessary to absorb larger amounts of N to fix adequate amounts of P, K and S; as well as to absorb higher amounts of K to fix Ca and Mg, also implying a modification of the Ca/Mg ratio.

The observed imbalance in nutritional ratios indicates that the genotypes of this cultivar may be more nutritionally demanding than other cultivars of Robusta coffee (traditionally cultivated and used as standards for nutritional parameters). Also, these imbalances imply that water deficit only occasionally was able to cause changes in the nutritional balance of this cultivar, demonstrating a greater metabolic inflexibility towards water stress (Table 5) than other cultivars or genotypes of Robusta coffee (Martins et al., 2014).

## CONCLUSIONS

There was considerable variability in biomass accumulation and nutritional content among the genotypes of the clonal cultivar “Conilon BRS Ouro Preto”, with different patterns of responses towards water availability in the soil.

Cultivation in the environments with low water supply caused losses of up to 29% in the biomass production of the young plants (with most severe losses observed in the aerial parts), but the magnitude of these losses varied with genotype. Genotypes 125 and 155 accumulated higher amounts of biomass when compared to the others, regardless of the water supply. Overall, the nutritional indexes of the genotypes show higher metabolic inflexibility towards water stress than what is reported for other cultivars of Robusta coffee.

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## CONFLICTS OF INTERESTS

The authors declare no conflict of interest.

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