Plant Physiology/ Original Article

Morphological, physiological, and agronomic traits of crossings of 'Icatu' x 'Catimor' coffee tree subjected to water deficit

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Abstract – The objective of this work was to select genotypes of *Coffea arabica* with good yield and potential tolerance to water deficit, as well as to try to understand the physiological and anatomical mechanisms involved in the adaptability of these genotypes to water stress. The physiological, anatomical, and agronomic traits of 19 genotypes of *C. arabica* were evaluated under the two following water conditions: regular irrigation and no irrigation (soil water deficit). The 'IPR 100', 2, 5, and 7 genotypes showed agronomic, physiological, and anatomical traits that contributed to a better water status maintenance in the initial development of coffee plants. Based on these results, these genotypes are potentially tolerant to water deficit. The 4, 10, 11, 14, 15, and 'Bourbon Amarelo IAC J10' genotypes show a lower adaptability of the anatomical structures under soil-water deficit conditions. The coffee tree genotypes display leaf plasticity, such as the thickness of palisade and spongy parenchyma, and the number, position, dimensions, and mobility of stomata under water deficit conditions.

Index terms: *Coffea arabica*, hydric stress, leaf anatomy.

Características morfológicas, fisiológicas e agronômicas de cruzamentos de cafeeiro 'Icatu' x 'Catimor' submetidos a deficit hídrico

Resumo – O objetivo deste trabalho foi selecionar genótipos de *Coffea arabica* com boa produtividade e potencial de tolerância ao deficit hídrico, além de tentar compreender os mecanismos fisiológicos e anatômicos envolvidos na adaptabilidade desses genótipos ao estresse hídrico. Avaliaram-se características fisiológicas, anatômicas e agronômicas de 19 genótipos de *C. arabica* sob as duas seguintes condições hídricas: irrigação regular e sem irrigação (deficit hídrico no solo). Os genótipos 'IPR 100', 2, 5 e 7 apresentaram características agronômicas, fisiológicas e anatômicas que contribuíram para melhor manutenção do estado hídrico no desenvolvimento inicial do cafeeiro. Com base nesses resultados, esses genótipos são potencialmente tolerantes ao deficit hídrico. Os genótipos 4, 10, 11, 14, 15 e 'Bourbon Amarelo IAC J10' apresentam menor adaptabilidade das estruturas anatômicas em condições de deficit hídrico no solo. Os genótipos de cafeeiro apresentam plasticidade foliar, como a espessura do parênquima paliçádico e esponjoso, e o número, posição, dimensões e mobilidade dos estômatos em condições de deficiência hídrica.

Termos para indexação: *Coffea arabica*, estresse hídrico, anatomia foliar.

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Introduction

Coffee (*Coffea arabica* L.) is one of the largest globally traded commodities, and it represents a significant income source for farmers in Latin American, African, and Asian countries. Environmental stresses caused by extreme weather concerning temperature and drought, either alone or combined, represent the most limiting factors for agriculture yield worldwide. Water determines the success of coffee farming, since it influences the phenology of the plant (Silva et al., 2019). Longer droughts and, sometimes, excess rainfall are expected to threaten the sustainability of agricultural production on a global scale, with consequences for the quantity and quality of coffee crop (DaMatta et al., 2018).

Water deficit is one of the most constraining factors to the primary production of ecosystems and crop yields, mainly due to restrictions imposed on photosynthetic carbon fixation (León-Burgos et al., 2022). Since rainfed cultivation is currently dominant in small arabica coffee-growing systems, the selection and characterization of drought-tolerant progenies can lead to new cultivars, which can ensure lower risk production and higher income for small coffee growers (Santos et al., 2021).

Regarding plant physiology, plants show complex physiological responses, showing different alterations in their mechanisms under soil-water deficit. Previous reports have investigated the physiological responses of *C. arabica* plants under water deficit conditions (Avila et al., 2020; Souza et al., 2020; Semedo et al., 2021). Evaluations for foliar physiological traits have been of great importance for the rapid identification of genotypes with differential tolerance to water, thermal, or other stresses (Brodribb & McAdam, 2017). Moreover, some other mechanisms of plants are able to reduce the effects of soil water deficit such as the increase of the root system, reduction of leaf area, osmotic adjustment, stomatal closure, and others (Fahad et al., 2017). In-depth scientific knowledge is crucial to assure the sustainability of the coffee production chain in constantly changing weather (DaMatta et al., 2019).

Agronomic and anatomical changes are dependent on plant physiological responses, thus the joint analysis of these traits can carry relevant information to understand the coffee tree tolerance mechanisms to water deficit, and the positive consequences for coffee breeding. So far, no study has covered analyses of physiological, anatomical, and agronomic traits in *C. arabica* genotypes under soil-water deficit, thus, the information presented in this work can help coffee breeders to enhance the Brazilian coffee-growing systems and to achieve good yield, even under unfavorable weather conditions, enabling the launch of new cultivars.

The objective of this work was to select genotypes of *C. arabica* with good yield and potential tolerance to water deficit, as well as to try to understand the physiological and anatomical mechanisms involved in the adaptability of these genotypes to water stress.

Materials and Methods

The experiment was carried out in a randomized complete block design with four replicates. The factorial arrangement in 19x2 consisted of 19 genotypes (15 progenies and 4 cultivars) and two soil-water conditions. The experimental plot was composed of one plant.

The chosen progenies were from the coffee breeding program conducted and coordinated by the Empresa de Pesquisa Agropecuária de Minas Gerais (Epamig), with the participation of the Universidade Federal of Lavras (UFLA) and the Universidade Federal de Viçosa (UFV). Fifteen F6 progenies from the crossings of 'Icatu Vermelho IAC 3851-2' × 'Catimor UFV 1602' were chosen, with basis on their productivity and agronomic vigor, from a field experiment conducted in the Epamig, in the municipality of São Sebastião do Paraíso, in the state of Minas Gerais (MG), Brazil, during 2013, 2014, and 2015 – a period of intense water deficit in the region. Plants were scored according to a 10- point arbitrary scale, by which score 1 corresponds to plants with the poorest vegetative vigor and marked depletion symptoms. The score 10 indicates plants with excellent vigor, more leafy, and with marked vegetative growth of the productive branches. Productivity was measured by weighing fruit immediately after harvest. Then, a sample of about 3 L from each plot was weighed and placed for drying in the sun. After drying, coffee fruit were weighed, processed, and weighed again to calculate productivity in processed bags (bag ha^{-1}). The cultivars 'Catuaí Vermelho IAC 144' (Grisi et al., 2008) and 'Bourbon Amarelo IAC J10' (Ronchi et al., 2015) were used as non-tolerant controls; and 'Siriema'

(Melo et al., 2014) and 'IPR 100' (Carvalho et al., 2017) were used as tolerant controls.

The experiment was carried out in a greenhouse, in the experimental field of Epamig Lavras, in the municipality of Lavras, MG, Brazil. The climate of Lavras, according to the Köppen-Geiger's classification, is of type Cwb – mesothermic climate, with mild and rainy summers. During the experimental period, minimum, average, and maximum of air temperature, and relative humidity were respectively 17°C, 26°C, 35°C, 39%, 63%, and 87%, in the greenhouse with free exchange of air. The seedlings were grown in 20 L polyethylene pots, with a substrate consisting of a mixture of subsoil soil, sand, and cattle manure (3:1:1, v/v/v), and they were kept in a free exchange air greenhouse for eight months, to produce sufficient leaves for the physiological analysis. Fertilization was carried out according to the substrate analysis, following the recommendations for the culture (Guimarães et al., 1999). Highly incident pests and diseases in the region were preventively controlled by regular phytosanitary treatments, using the active ingredient chlorpyrifos, for the curative control of leaf miner, and copper hydroxide, as a preventive *Cercospora coffeicola.* Until the day before being subjected to water treatments, all seedling were well-watered in the pots, to maintain their substrate at pot capacity. A moisture sensor (ML2X Thetaprode Delta-T Devices, Cambridge, England) calibrated according to the respective soil tension was used to control the amount of water applied to each pot.

On October 30, 2016, the plants were divided into two groups to begin the experiment. One group continued to receive regular irrigation at pot capacity (control plants), while the other group, consisting of the same genotypic material, went through total suspension of irrigation (plants subjected to drought stress). Physiological traits were evaluated immediately before the treatments and after 7, 14, 21, 28, 35 and 42 days of treatments.

For the agronomic characterization, measurements were performed for the initial stem diameter (ISD), leaf area (LA), and initial plant height (IPH) in all plants right before the treatments. At 42 days after the water treatments, all plants were re-evaluated again for the dimension of final stem diameter (FSD) and final plant height (FPH), and evaluated for the leaf dry matter (LDM), root dry matter (RDM), and total dry matter (TDM = LDM + RDM). Values of ISD and FSD (mm) were taken from the plant root collars with a digital caliper, whereas the values of LA (cm^2) were measured using the leaf dimension method, described by Barros et al. (1973). The plant measurements of IPH and FPH (cm) were obtained on the orthotropic branches measured in the root collar, as far as in the insertion of the last node. We reckoned the DCP measures by multiplying by two the length of the longest plagiotropic branch in each plant. At the end of the experiment, both the shoot and root systems were washed, told apart, and dried in an oven with air circulation, at 70°C for 72 hours, to obtain the respective dry matter (g).

Physiological characterization: leaf water potential (Ψ_W) analyses were performed in fully expanded leaves of the third or fourth apex proximal pair of plagiotropic branches, in the middle section of plants without any apparent damage caused by pests or diseases. The Ψw was evaluated in individual leaves, before dawn, using a Scholander type pressure pump (PMS Instruments Plant Moisture-Model 1000, Albany, OR, USA).

For the anatomic characterization, leaves collected at the end of the experiment had their central portion first fixed in FAA (formaldehyde, glacial acetic acid, and ethanol at 50%, 1:1:9, v/v) for 48 hours and, then, preserved in ethanol at 70% (Johansen, 1940) to preserve the material. The cross-sections obtained with the help of a table microtome type LPC were clarified in 50% sodium hypochlorite (v/v), washed three times in distilled water, stained with *safrablau* dye (0.1% Astra blue and 1% safranin, 7:3 proportion), and finally assembled on semi-permanent slides containing 50% glycerol (v/v) (Kraus & Arduin, 1997). The slides were observed and photographed using an optical microscope. The variables phloem thickness (PT, μm), palisade parenchyma thickness (PPT, μm), and stomatal density (SD, number of stomata mm^{-2}) were measured using the ImageTool software (Sousa et al., 2012).

Data analysis considered a randomized complete block design for the agronomic and anatomical variables and a split-plot design subdivided in time for the physiological ones. Variance analyses and model parameters checked by Shapiro's and Bartlett's tests were performed using the F-test, with subsequent application of Scott-Knott's test, at 5% probability, for grouping the means using the statistical program Genes (Cruz, 2013).

Results and Discussion

At 5% probability, by Scott-Knott's test, the 2, 5, 7, 12, 13, and 'IPR 100' genotypes showed smaller initial leaf area (LA of 3001, 3230, 2986, 3187, 3007 and 2970 cm², respectively), which may favor the water status of coffee trees under water restriction conditions, since plants transpire at lower proportion to maintain the water supply in the soil, providing a defense mechanism against water stress (Ribeiro et al., 2017). Genotypes 4, 14, and 15 had a higher LA of 5,016, 4,781, and 4,518 cm², respectively.

Genotype 7 showed the greatest difference between FSD and ISD in both water conditions (Table 1).

Table 1. Increase of diameter (D) and height (H) considering differences between the means of both the initial and final values of stem diameter and height, initial stem diameter (ISD), final stem diameter (FSD), and percentage of diameter growth (DFD); and initial stem height (ISH), final stem height (FSH), and percentage of height growth (DFH) of *Coffea arabica* genotypes under irrigated (I) and water deficit (WD) treatments (WT).

Genotype	Stem diameter (mm)					Genotype	Stem height (cm)					
	WT	ISD	FSD	Difference	DFD (%)			WT	ISH	FSH	Difference	DFH (%)
7	I	9.8	13.0	3.3	33	*	11	I	44.5	58.9	14.4	32	*
IAC 144	I	9.2	12.2	3.0	32	*	14	I	49.5	64.3	14.8	30	*
8	I	9.1	12.0	2.9	32	*	2	I	47.4	61.0	13.6	29	*
1	I	10.3	13.4	3.1	30	*	10	I	51.3	65.6	14.4	28	*
13	I	8.9	11.6	2.7	30	*	6	I	44.8	57.0	12.3	27	*
15	I	10.6	13.7	3.1	29	*	4	I	49.3	60.5	11.3	23	*
4	I	10.6	13.6	3.1	29	*	3	I	48.1	59.1	11.0	23	*
6	I	10.1	13.1	3.0	29	*	8	I	47.3	58.3	11.0	23	*
2	I	10.6	13.5	2.9	28	*	13	I	38.9	47.8	8.9	23	*
14	I	10.8	13.7	3.0	27	*	IAC 144	I	45.5	55.4	9.9	22	*
B.A. IAC J10[1]	I	9.9	12.5	2.6	27	*	5	I	43.8	53.5	9.8	22	*
12	I	9.6	12.2	2.6	27	*	15	I	46.8	56.3	9.5	20	*
Siriema	I	10.0	12.6	2.6	26	*	Bourbon Amarelo IAC J10	I	70.1	83.5	13.4	19	*
11	I	9.9	12.4	2.5	26	*	7	I	56.6	67.4	10.8	19	*
3	I	10.7	13.4	2.7	25	*	9	I	50.6	60.1	9.5	19	*
10	I	10.4	12.7	2.3	22	*	12	I	48.3	57.5	9.3	19	*
5	I	9.6	11.8	2.2	22	*	1	I	43.9	52.0	8.1	19	*
IPR 100	I	9.2	11.2	2.0	22	*	5	WD	43.5	50.8	7.3	17	*
9	I	11.3	13.6	2.3	20	*	IPR 100	I	42.1	49.1	7.0	17	*
7	WD	8.2	9.4	1.1	14	*	Siriema	I	50.6	57.4	6.8	13	*
IPR 100	WD	9.6	10.8	1.1	12	*	3	WD	46.1	52.1	6.0	13	*
5	WD	9.5	10.4	0.9	10	*	10	WD	48.8	54.5	5.8	12	*
8	WD	10.3	11.1	0.8	8	*	1	WD	46.6	52.3	5.6	12	*
3	WD	10.5	11.2	0.8	7	*	2	WD	44.4	49.9	5.5	12	*
15	WD	10.7	11.3	0.7	6	*	7	WD	43.6	48.9	5.3	12	*
1	WD	10.5	11.1	0.7	6	*	15	WD	50.0	55.6	5.6	11	*
9	WD	10.0	10.6	0.6	6	*	11	WD	46.8	52.0	5.3	11	*
13	WD	10.0	10.3	0.3	3	ns	4	WD	48.8	53.9	5.1	11	*
12	WD	9.8	10.1	0.3	3	ns	9	WD	46.3	51.1	4.9	11	*
4	WD	11.2	11.5	0.3	3	ns	6	WD	48.1	52.9	4.8	10	*
6	WD	10.1	10.4	0.3	3	ns	12	WD	47.3	51.5	4.3	9	*
B.A. IAC J10[1]	WD	10.2	10.4	0.2	2	ns	14	WD	48.0	51.9	3.9	8	*
IAC 144	WD	9.3	9.4	0.1	1	ns	IPR 100	WD	48.5	52.3	3.8	8	*
2	WD	9.0	9.1	0.1	1	ns	Bourbon Amarelo IAC J10	WD	72.4	77.3	4.9	7	*
Siriema	WD	10.4	10.4	0.0	0	ns	Siriema	WD	53.8	57.8	4.0	7	*
14	WD	10.5	10.3	0.0	0	ns	IAC 144	WD	45.4	48.6	3.3	7	*
10	WD	10.5	10.1	0.0	0	ns	13	WD	43.5	46.8	3.3	7	*
11	WD	11.2	10.7	0.0	0	ns	8	WD	50.6	53.6	3.0	6	*

*Significant by the t-test, at 5% probability. nsNonsignificant. [1]Bourbon Amarelo IAC J10.

This behavior evidences a highly flexible genotype, very responsive to irrigation, since when receiving irrigation, its development was greater among the genotypes under study. Genotype 5, even under water deficit, showed similar stem height growth to the 'IPR 100' and 'Siriema' in the irrigated plants, displaying greater capacity to maintain its well-developed agronomic characteristics under stress, when in the initial stage of plant development.

Differences between initial and final stem diameter varied from 0 to 33% (Table 1), with significant growth for all treatments under irrigation. However, eight genotypes showed a significant difference between FSD and ISD under stress; genotypes 3, 5, 7, 8, and 'IPR 100' displayed the greatest differences between the initial and final stem diameter when subjected to water deficit.

In the water deficit condition, the 1, 2, 3, 5, 7, and 10 genotypes had the highest growth rates concerning the plant height. Under irrigation conditions, genotypes 11, 14, 2, 10, and 6 showed the highest growth rates, varying from 32 to 27%, from the initial height of the plants (Table 1).

Starting the irrigation suspension process caused a reduction of the values of predawn water potential of the plants (Table 2). This physiological impact results from the gradual increase of water deficit to which plants were subjected. However, some genotypes showed distinct physiological and anatomical behaviors, evidencing different levels of tolerance.

Physiological evaluations displayed differences regarding water potential (Ψ_W) after the twenty-first day of the application of water treatments, when genotype 4 and 'Siriema' started showing Ψ_W values below -3.0 MPa (Table 2).

The genotypes of *C. arabica* and *C. canephora* show variability regarding drought tolerance. In general, *C. canephora* tolerates longer periods of drought than *C. arabica* (DaMatta et al., 2018). Stomatal regulation is a plant functionality that occurs to maintain a constant difference between leaf-water potential and soil-water potential, in the period of lower water availability (Kumagai et al., 2015). Genotypes 4 and 'Siriema' showed a greater sensitivity to water scarcity, in a short period of deficit (21 days), since these genotypes' predawn water potentials were

Table 2. Means of the water potential (Ψ_W) of *Coffea arabica* genotypes under either irrigation (I) or water deficit (WD) conditions, over seven evaluation periods, totaling 42 days[1].

Genotype	T1 (0 day)		T2 (7 days)		T3 (14 days)		T4 (21days)		T5 (28 days)		T6 (35 days)		T7 (42 days)	
	I	WD	I	WD	I	WD	I	WD	I	WD	I	WD	I	WD
1	-0.34a	-0.41a	-0.34a	-0.58a	-0.43a	-0.55a	-0.49a	-0.89a	-0.40a	-3.32c	-0.35a	-5.37d	-0.40a	-6.07b
2	-0.50a	-0.36a	-0.41a	-0.43a	-0.45a	-0.58a	-0.40a	-0.64a	-0.41a	-1.16a	-0.35a	-1.33a	-0.38a	-4.07a
3	-0.45a	-0.44a	-0.35a	-0.51a	-0.36a	-0.45a	-0.45a	-1.20a	-0.53a	-2.88c	-0.31a	-4.80d	-0.40a	-5.53b
4	-0.54a	-0.64a	-0.31a	-0.50a	-0.30a	-0.47a	-0.58a	-3.84b	-0.56a	-4.70d	-0.34a	-5.40d	-0.35a	-5.23b
5	-0.31a	-0.44a	-0.35a	-0.58a	-0.38a	-0.49a	-0.45a	-0.71a	-0.48a	-0.98a	-0.38a	-1.63a	-0.64a	-4.90a
6	-0.28a	-0.36a	-0.44a	-0.53a	-0.36a	-0.66a	-0.51a	-0.66a	-0.36a	-1.40a	-0.30a	-3.69c	-0.34a	-5.53b
7	-0.28a	-0.31a	-0.34a	-0.55a	-0.29a	-0.48a	-0.55a	-1.03a	-0.51a	-1.25a	-0.46a	-4.66d	-0.43a	-5.36b
8	-0.35a	-0.54a	-0.31a	-0.53a	-0.33a	-0.74a	-0.50a	-1.31a	-0.49a	-3.80c	-0.39a	-4.78d	-0.33a	-5.23b
9	-0.56a	-0.60a	-0.33a	-0.73a	-0.29a	-0.75a	-0.49a	-1.09a	-0.48a	-2.06b	-0.34a	-4.04c	-0.30a	-5.38b
10	-0.50a	-0.48a	-0.34a	-0.46a	-0.29a	-0.65a	-0.31a	-0.93a	-0.33a	-3.81c	-0.33a	-4.26c	-0.31a	-5.53b
11	-0.49a	-0.56a	-0.44a	-0.55a	-0.33a	-0.47a	-0.44a	-0.72a	-0.48a	-1.57a	-0.39a	-4.54d	-0.41a	-6.03b
12	-0.55a	-0.49a	-0.38a	-0.63a	-0.40a	-0.55a	-0.34a	-0.81a	-0.59a	-1.05a	-0.36a	-2.83b	-0.41a	-4.42a
13	-0.46a	-0.40a	-0.34a	-0.51a	-0.39a	-0.53a	-0.46a	-0.75a	-0.43a	-1.10a	-0.35a	-4.64d	-0.36a	-5.60b
14	-0.53a	-0.43a	-0.39a	-0.51a	-0.28a	-0.41a	-0.53a	-0.87a	-0.48a	-2.51b	-0.35a	-4.40c	-0.41a	-5.01a
15	-0.56a	-0.44a	-0.38a	-0.55a	-0.41a	-0.64a	-0.45a	-1.86a	-0.50a	-5.23d	-0.36a	-5.88d	-0.36a	-5.88b
B.A. IAC J10[2]	-0.49a	-0.41a	-0.36a	-0.34a	-0.41a	-0.59a	-0.53a	-0.81a	-0.61a	-1.74b	-0.36a	-3.64c	-0.46a	-5.00a
IAC 144	-0.51a	-0.46a	-0.45a	-0.49a	-0.28a	-0.45a	-0.46a	-0.91a	-0.53a	-2.13b	-0.34a	-4.86d	-0.36a	-4.84a
IPR 100	-0.51a	-0.41a	-0.50a	-0.46a	-0.46a	-0.55a	-0.50a	-0.76a	-0.66a	-1.29a	-0.36a	-2.22b	-0.54a	-4.58a
Siriema	-0.48a	-0.44a	-0.31a	-0.55a	-0.64a	-0.66a	-0.46a	-3.07b	-0.51a	-4.01c	-0.35a	-4.88d	-0.33a	-5.13a

[1]Means followed by equal letters, in the columns, do not differ, by Scott-Knott's test, at 5% probability. Number of days after suspension of irrigation: T1, 0; T2, 7; T3, 14; T4, 21; T5, 28; T6, 35; and T7, 42. [2]Bourbon Amarelo IAC J10.

significantly reduced in comparison with those of the others. However, under water deficit, there is damage to the photosynthetic apparatus, such as a reduction of the ATP synthesis, decrease of the activity of ribulose 1.5 bisphosphate carboxylase/oxygenase, or even decreases of the rate of these molecules regeneration step, which causes an overall impairment in the carbon metabolism, including respiration.

The decrease of stomatal conductance occurs as the evaporative demand increases. This process reduces the transpiration rate and, consequently, contributes to the increase or maintenance of Ψ_W within limits that allow of the maintenance of plant growth (León-Burgos et al., 2022). This is a fundamental mechanism for adaptation to stress conditions, which can significantly contribute to the selection process of materials that are tolerant to water deficit.

Because Ψ_W has a negative correlation with leaf area, genotypes 2, 5, 6, 7, 11, 12, 13, and 'IPR 100' showed higher Ψ_W values on the twenty-eighth day of evaluation, and genotypes 4 and 15 (larger leaf areas) rapidly reduced their Ψ_W, after the water deficit treatment. Regarding genotypes 2, 5, 12, and 13, we can associate their larger Ψ_W to the smaller leaf area. In a similar work, the authors evaluated and classified 'IPR 100' with traits of tolerance to water deficit (Carvalho et al., 2017).

At 35 days after the treatment application, genotypes 2 and 5 showed greater Ψ_W than the others (-1.33 MPa and -1.63 MPa, respectively). In turn, 'IPR 100' showed -2.22 MPa Ψ_W, and 52.6% genotypes – 1, 3, 4, 7, 8, 11, 13, 15, 'IAC 144', and 'Siriema' – presented values lower than -0.45 MPa (Table 2).

Within 42 days of water deficit, genotypes 2, 5, 12, 14, 'Bourbon Amarelo IAC J10', 'IAC 144', 'Siriema', and 'IPR 100' had higher water potential than the others. Nevertheless, every genotype was under extremely severe water deficit conditions with reduced gas exchanges.

The parameters evaluated for leaf anatomy show that the imposition of water deficit on coffee trees triggered direct responses, due to the low availability of water, providing leaf structure changes, such as increased stomatal density, reduction of phloem thickness, and alterations of palisade parenchyma thickness and number of xylem vessels. Leaf anatomy varies among genotypes and the identification of resistant plants to certain environmental conditions is useful. The

coffee tree genotypes display leaf plasticity, such as the thickness of palisade and spongy parenchyma, the number, position, dimensions, and mobility of stomata to face water deficit (Rodrigues et al., 2016).

Regarding the phloem (Figure 1) and palisade parenchyma (Figure 2) thickness, the genotype 2 behaved differently from the other genotypes by increasing the values of these parameters under water deficit conditions. Such increase of the phloem thickness can allow of a greater flow of carbohydrates from the shoots to the roots, as well as the translocation of nutrients necessary for a better development of the plant (Castro et al., 2009; Ribeiro et al., 2012); this anatomical change can lead to an improved production of photoassimilates that will allow of a better root growth and maintenance of water supply to the shoots. Similarly, Viana et al. (2018) found greater palisade parenchyma thickness for a five genotypes resistant to rust, of the germoplasm bank from the state of Minas Gerais, among the 15 studied. This fact may be closely linked to the photosynthetic rate, since the palisade parenchyma thickness is related to the CO_2 fixation by the plant, eventually contributing to the unchanged stomatal density of the genotype under both irrigation and water deficit (Figure 3).

For phloem thickness under water deficit conditions, the genotypes showed a significant difference, and were separated into two groups, those with higher values, consisting of genotypes 1 and 2 (Figure 4), and 6, 7, 9, 14, 15, 'IAC 144', and 'IPR 100'. The small values were observed for genotypes 5, 12, 13, 'Siriema', and 'Bourbon' (Figure 1).

The highest values for the palisade parenchyma thickness were observed for genotypes 2, 5, 7, and 14 which preserved their values under water deficit conditions, while the remaining genotypes significantly reduced their parenchyma thickness at 42 days of water deficit (Figure 2).

The stomatal density values were significantly altered with the application of water deficit and increased in almost all genotypes, except for genotype 2 (Figure 3), whereas under irrigation conditions, no difference was observed between the genotypes regarding the number of stomata mm^{-2}. Under water deficit, the genotype 11 had the highest number of stomata mm^{-2}, followed by genotypes 1, 6, 8, 9, 15, 'Siriema', and 'IAC 144'. Therefore, this knowledge can be used to select genetic material with larger stomatal densities to provide a better capture and fixation of

atmospheric CO_2, to improve photosynthetic efficiency and photoassimilate production (Batista et al., 2010; Gama et al., 2017; Dittberner et al., 2018).

Stomatal density is an important ecophysiological parameter which directly affects gas exchange and photosynthesis. Usually, genotypes which are resistant to water stress have a higher stomatal density (Grisi et al., 2008; Batista et al., 2010; Queiroz-Voltan et al., 2014; Gama et al., 2017). The increase of stomatal density observed for genotypes 1, 6, 8, 9, 11, 15, 'Siriema',
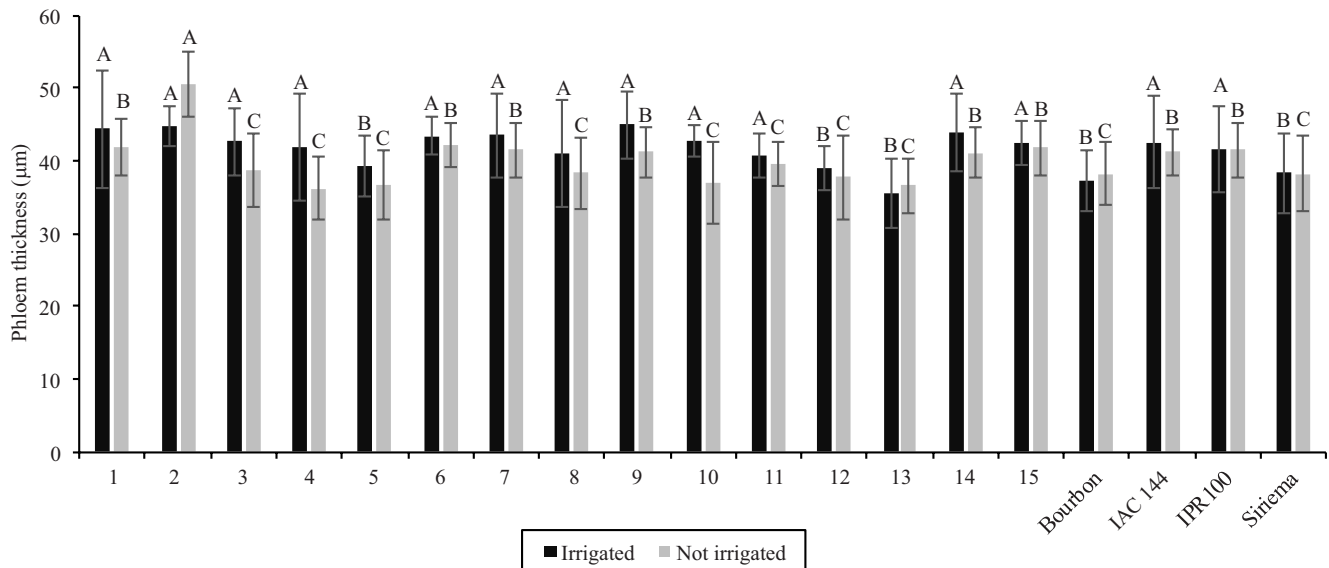
Figure 1. Phloem thickness of *Coffea arabica* genotypes evaluated under either irrigation or water deficit conditions, at 42 days after the application of water treatments. Means followed by equal uppercase letters for each water treatment do not differ by Scott-Knott's test, at 5% probability.
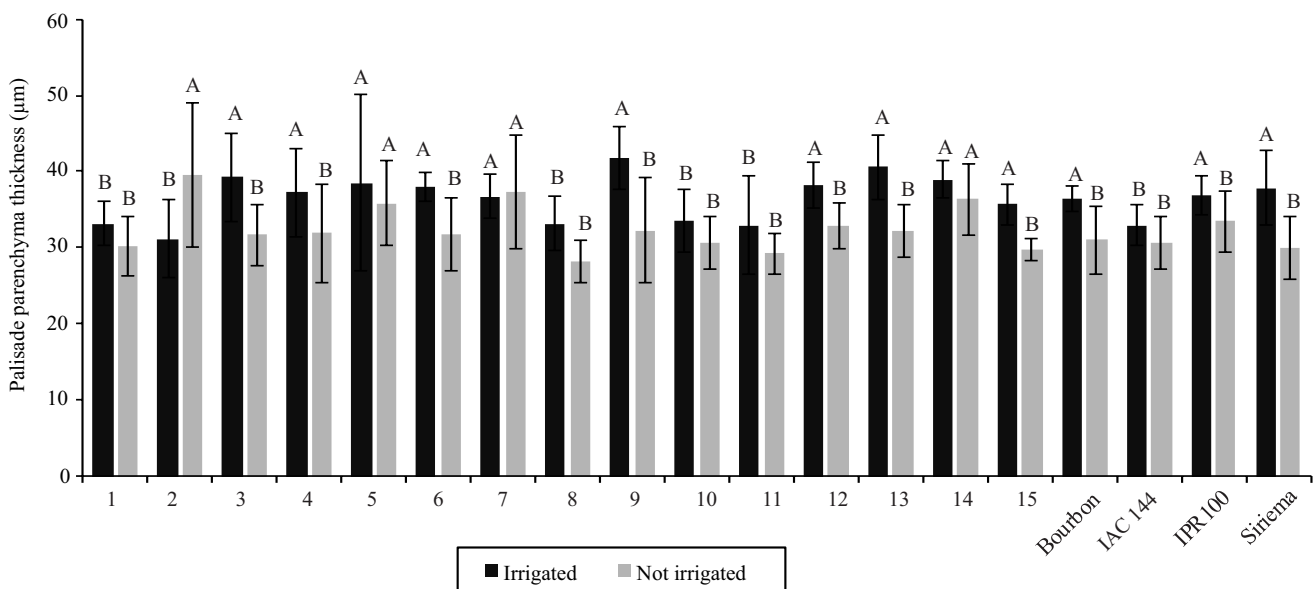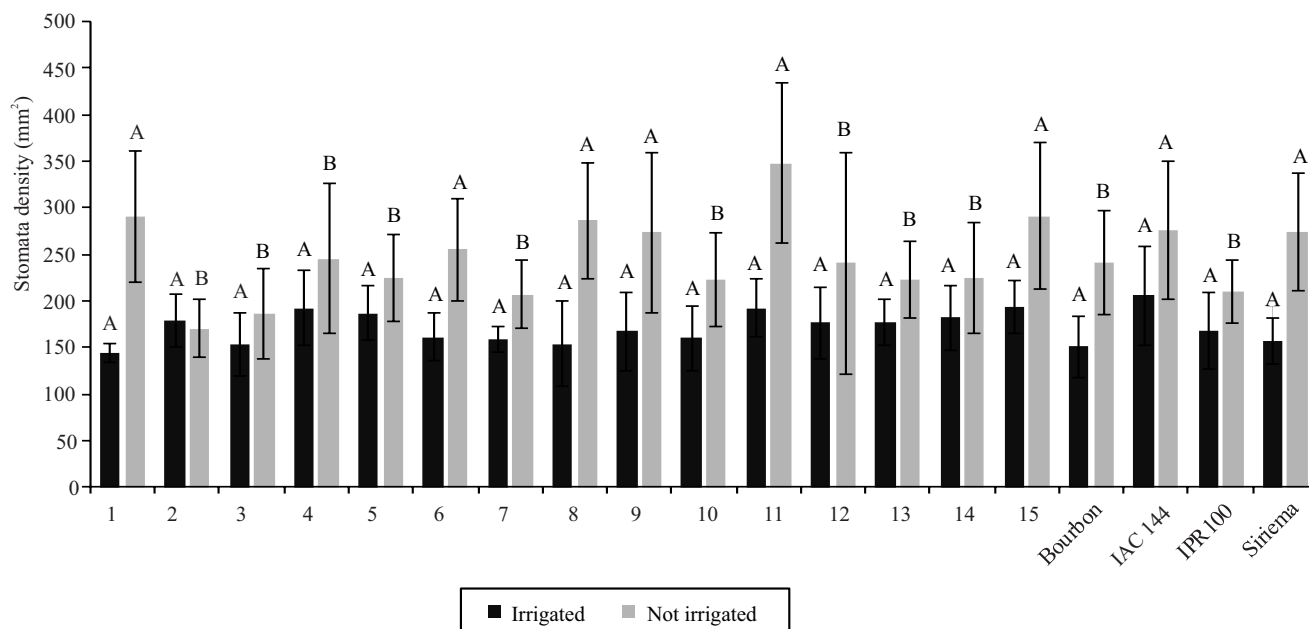
Figure 2. Palisade parenchyma thickness (µm) of *Coffea arabica* genotypes evaluated under either irrigation or water deficit conditions, 42 days after the application of water treatments. Means followed by equal uppercase letters for each water treatment do not differ by Scott-Knott's test, at 5% probability.

Figure 3. Stomata density of *Coffea arabica* genotypes evaluated under either irrigation or water deficit conditions, at 42 days after the application of water treatments. Means followed by equal uppercase letters for each water treatment do not differ by Scott-Knott's test, at 5% probability.
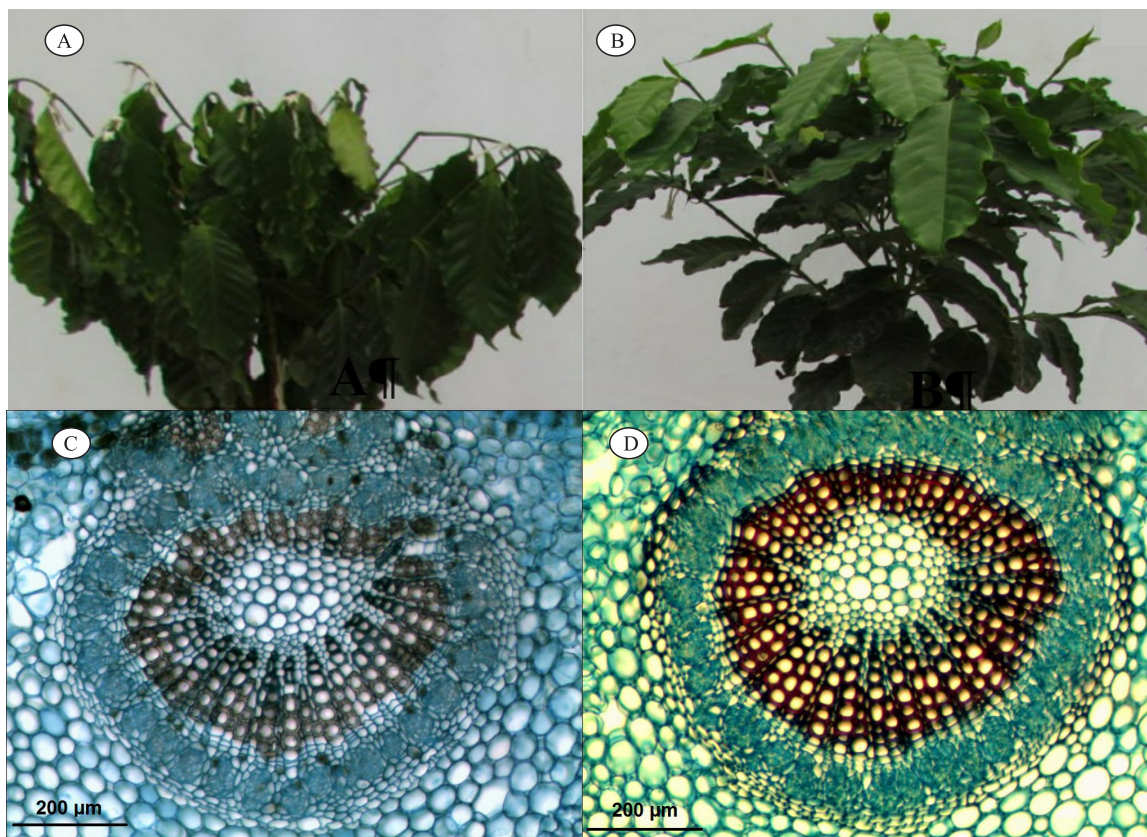
Figure 4. Phenotypical image of *Coffea arabica* genotype 2 and anatomical cross-sections of leaves, at the end of the experiment: A, plant subjected to water deficit conditions; B, plant subjected to irrigation; C, cross-sections of leaves subjected to water deficit conditions; and D, cross-sections of leaves subjected to irrigation. Photos by Larissa Sousa Coelho.

and 'IAC 144' reflects their greater ability to capture CO_2, which can improve the photosynthetic efficiency (Castro et al., 2009; Batista et al., 2010), since larger stomatal density implies faster stomatal movements, in shorter periods, allowing of an adequate gas exchange, consequently improving the plant adaptability to water deficit (Oliveira & Miglioranza, 2013).

Conclusions

1. *Coffea arabica* genotypes 2, 5, 7, and 'IPR 100' showed agronomic, physiological, and anatomical traits that contribute to a better water status maintenance in the initial development of coffee plants, and, therefore, are potentially tolerant to water deficit.

2. The 4, 10, 11, 14 , 15, and 'Bourbon Amarelo IAC J10' genotypes show a lower adaptability of anatomical structures under soil-water deficit conditions.

3. The coffee tree genotypes display leaf plasticity, such as the thickness of palisade and spongy parenchyma, and the number, position, dimensions, and mobility of stomata under water deficit conditions.

Acknowledgments

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