ANATOMICAL CHARACTERISTICS AND WATER RELATIONS IN Coffea Canephora L.

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Abstract

Coffee cultivation emerges as a more viable and commercially sustainable planting alternative for areas degraded by sugar cane monoculture. Therefore, studies with coffee cultivars seek for plants that are more adapted to the environmental of Brazilian northeast. Thus, it is possible to obtain a better performance in the water consumption and maintenance of the high productivity levels in the field. Regarding this, studies of the morphological characteristics and mechanisms of physiological response of the plant, which control the loss of water by the leaves, are of extreme importance. So, they allow the adaptive evaluation of the variety to the region of Zona da Mata of Pernambuco, aiming an better adequate use of the species under study. Therefore, this work, which was carried out in the Department of Nuclear Energy of the Universidade Federal de Pernambuco (UFPE) and in the Instituto Agronômico de Pernambucano (IPA) Experimental Unit, consisted of the characterization and morpho-anatomical analysis of Coffea canephora L. (Early Vitória variety 2 INCAPER). Then were performed the following procedure: selection of clones in the experimental field, collection of botanical material, characterization of hydraulic properties, hydraulic functionality and analysis of experimental data. In general, it was concluded that the clone of the early Vitória variety 2 did not show large phenotypic variations. Indicating that these plants are exhibiting the same adaptive response in the Brazilian environment.

Key-words: conilon coffee, adaptative behavior, phenotypic analyses

1. Introduction

Coffea arabica (Arabica coffee) and Coffea canephora Pierre exFroehner (Conilon coffee) are the most economically important coffee species on the world market. These are the most used in the preparation of the commercial blend of roasted coffee. Currently, Brazil is one of the largest producers and exporters of coffee, besides that the country has a strong consumer market for such product.

The C. canephora presents a quality of the produced beverage slightly inferior to that of C. arabica. Because it has a caffeine presence of only about 2.38%. However, the conilon coffee is a strong competitor in the international market because it has a lower price and allows a high extraction of soluble solids (Andreoli et al., 1993).

The market has been demanding an especial attention to aspects related to the environmental responsibility of the enterprises. In this sense, there is a great concern for the efficiency of water use and maintenance of the high productivity levels of coffee plantations. Besides that, interactions of genotypes versus locations are determinant in the evaluation of adaptability and phenotypic stability of the varieties. Thus, aiming the recommendation of a specific species for a given region.

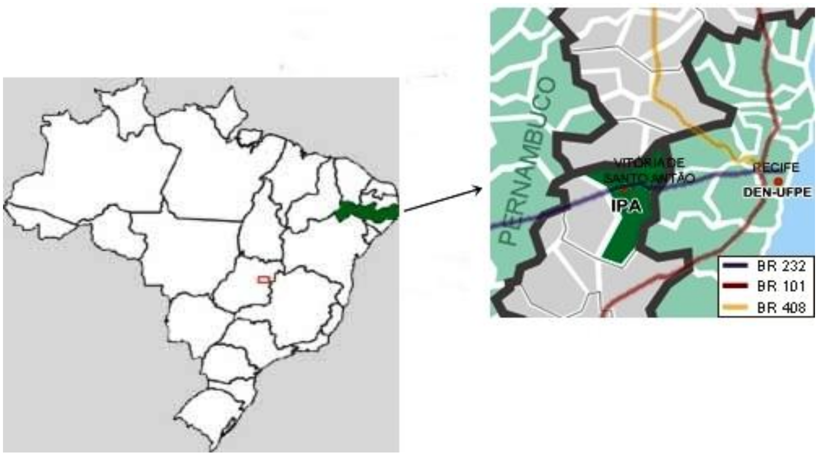
In order to obtain a better knowledge about the conilon coffee species, the Government of Pernambuco State has been carrying out researches with the introduction and selection of Conilon coffee (Coffea canephora L.) clones in the Zona da Mata. Therefore, the present work had the purpose of characterizing the structure and hydraulic functionality of samples collected in the experimental field and the adaptive evaluation of the variety to the region.

2. Methodology

2.1. Experimentation site

The experiments were conducted at the IPA Unit located at the Experimental Station Luiz Jorge da Gama Wanderley, which belongs to the Instituto Agronômico de Pernambuco (IPA), in Vitória de Santo Antão, South Zona da Mata of Pernambuco (Figure 1). The geographic coordinates of the location are 08º 08’ 00’’ South latitude; 35º 22’ 00’’ longitude and altitude 146m. The climate of the region, according to the Thornthwaith & Mather (1955) climate classification, is of type C1S2A'a'. That implies in a subhumid climate, with small water surplus during the rainy season, mega thermal and with vegetation all year long (IPA, 2010). The region has an annual average temperature of 25.4ºC, a rainfall of 1025mm / year, potential evapotranspiration of 1344mm / year and the predominant soils are LVA14 and PE39 - Dystrophic Red-Yellow Latosol and Dystrophic Red-Yellow Argisol and eutrophic equivalent, respectively.

Figure 1 - Location of the experiments.



Source: PROMATA (2010).

The demonstrative unit (UD) has 7200 m2, soil type Red-Yellow Latosol, slightly acid and with good natural fertility. The planting was carried out in April 2006 with 4 plots, spacing of 2,0 m x 1,5 m and size of plots of 1800 m^2, totaling 1660 plants. The materials used came from vegetative propagation by cuttings from Instituto Capixaba de Pesquisas Agrícolas (INCAPER). The plots are Clone 2 / Var. Vitória 12; Clone 2 / Var. Vitória 2; Clone 3 / Early and Clone 7 / Var. 8121 / Clone 3 / Var. 8141 Robustão. The irrigation used was micro sprinkler, Figure 2.

Figure 2 - Demonstrate unit of conilon coffee cultivated at the Experimental Station Luiz Jorge da Gama Wanderley (IPA), Vitória de Santo Antão, Pernambuco.

Source: Own Author

The Clone Incaper Variety Vitória 2 presented the best visual adaptive responses in relation to the other clones cultivated in the U.D. of the Experimental Station "Luiz Jorge da Gama Wanderley" - IPA. Therefore the coffee conilon variety 2 early was the plant selected for this study.

Thus, in the experimental unit (IPA -Vitória de Santo Antão), nine healthy individuals of C. canephora Early Vitoria variety 2 were selected and marked. This analysis was performed according to the visual similarity of the plant height, crown volume, and number of stems. The purpose of this was to obtain measures related to the characterization and hydraulic functionality of the plants.

2.2. Hydraulic Relations

The leaves of the conilon coffee tree maintain a high relative water content, even under potentially negative water potentials (Damatta et al., 1993; Pinheiro et al., 2005). That is probably a way it has to avoid desiccation, rather than tolerate (Damatta et al., 1993). Therefore, visible symptoms of wilt on foliage are rare, except when the soil moisture is very low (Ferrão et al., 2007).

The main components of the differential adaptation to drought among conilon clones seem to be behavioral, probably governed by water use rates and/or soil water extraction efficiency (Damatta et al., 2000; Pinheiro et al. Al., 2005; Damatta & Ramalho, 2006). Physiological evaluations suggest that clones with relatively high production under dry conditions, do so by maintaining adequate water status, a combination of deep root systems and satisfactory stomatal control of transpiration, and maintenance of leaf area with leaves presenting a more vertical orientation of leaf fronds (Damatta & Ramalho , 2006).

2.3. Plant Measurements

A research report from the Experimental Station "Luiz Jorge da Gama Wanderley" (IPA, 2010) analyzed the values of plant heights and crown diameters of the various plots. On average, Clone Vitória Variety Vitória 2 Early presented higher height and crown diameter than the others. It has also overcome the other Clones / Var. in productivity. It has then reached a relative increase of 50% in relation to the Victory Clone 12, the least productive. Thus, it indicates that the coffee clone conilon variety 2 is a precocious plant with high phenotypic strength, Table 1.

Table 1 - Agronomic Characteristics of the clone variety Incaper Early Vitória 2.

Form of propagation	***Asexual (Clonal)***
Number of clones	1
Planting form	In line
Visual avaluation index	7.85 (Scale from 0 to 10)
Vegetative Strength	high
Average productivity	40 sc. Benef./ha
Average plant height	2.00 m
Cup diameter médium	1.75 m
Plant architecture	Normal, suitable for density from 2.3 to 3.3 thousand plants / ha
Type of tillage	Semi-thickened
Fruit maturation	Disuniform
Months of maturation	March to May
Study area	Zona da Mata of Pernambuco

Source: Own Author

2.4. Leaf Area Estimation

A main sample composed of 180 leaves of coffee variety Vitória 2 Precoce / INCAPER was used to measure the area of the leaf. These leaves were randomly collected from all the faces and positions of the crown, from 9 similar plants (20 leaves/plants). These samples were scanned with a table scanner and afterward were determined by image analysis the length of the central rib (cm) and area of each leaf (cm2). For this purpose was used the Image Tool program (Wilcox et al., 2002), Figure 3 and Table 2.

Figure 3 - Exemplary of coffee leaves that were scanned: a) calculation of the area; B) calculation of the length of the central vein.

Source: Own Author

Table 2 - Average of the central vein length (CNC) and leaf area in nine individuals of Coffea canephora cultivated in an experimental field in the Southern Forest of the State of Pernambuco, Brazil.

Plants	*CNC (cm)*	*Area (cm^2)*
P1	12.20	43.30
P2	13.07	45.24
P3	12.17	39.50
P4	13.47	55.96
P5	12.36	39.03
P6	12.57	50.33
P7	12.57	44.28
P8	12.86	49.85
P9	11.43	39.31
Average	12.52	45.20
SD	1.23	9.33
CV (%)	9.83	20.63

Source: Own Author

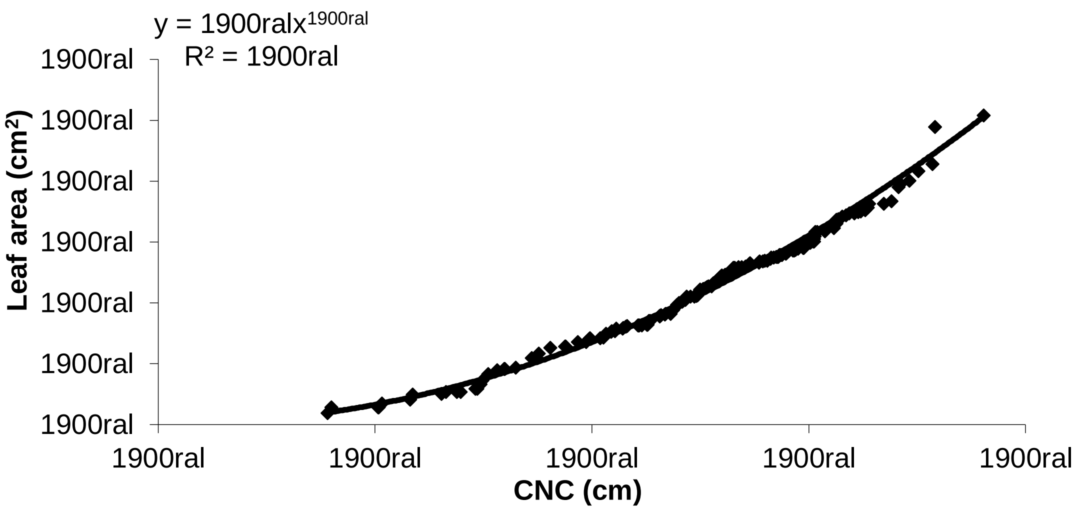
Based on the length relationships of the central rib and leaf area (Partelli et al., 2006) a power equation was fitted, Equation [1]:

$$Y = 0,2448 \cdot CNC^{2,0444} \quad [1]$$

Where Y is the estimated value for the leaf area (cm²); CNC is the length of the central rib (cm); 0.2448 and 2.0444 are adjusted regression coefficients and R^2 = 0.99.

In Figure 4 It can be observed the power equation adjusted for the experimental data. That is the relation between the length of the central rib and the leaf area of the conilon coffee.

Figure 4 - Area of the leaf in function of the central vein length (CNC) for the conilon coffee variety EarlyVitória 2/ INCAPER.



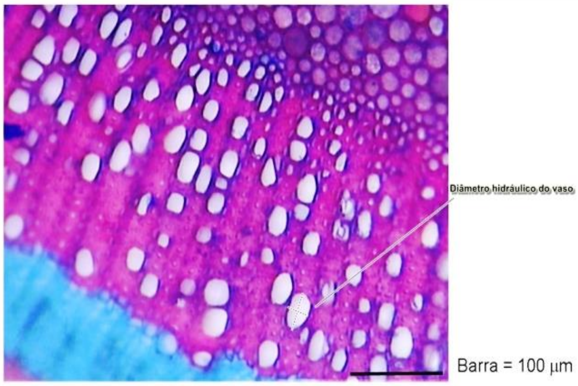
Source: Own Author

2.5. *Xylem Specific Conductivity*

The hydraulic character measurements of water transport elements (xylem) were obtained from freehand cross sections of herbaceous parts of branches, stained with safranin and astra blue (Krauter, 1985). The best sections were examined in digital images obtained under coupled optical microscope with digital camera in magnification of 400x and 100x. The cross-sectional area of the stem, the cross-sectional area occupied by the vascular bundle (mm²) and the xylem conduction vessel density (n .mm-²) were obtained in three images.

In 10 cells per vascular bundle were measured the largest and smallest perpendicular length of the lumen (space limited by the cell wall) and of the xylem cells was measured (Sellin et al., 2008). As well as the wall thickness of these vessels (μm), Figure 5.

Figure 5 - Measures of the largest and smallest (μm) perpendicular length of the lumen and xylem cells.



Source: Own Author

The average of the vessels diameter (D) was calculated by Equation [2].

$$D = \frac{\sum_{i=1}^{n} \sqrt{a_i b_i}}{n} \qquad [2]$$

Where a_i is the smallest and b_i the largest perpendicular diameter of the xylem vessel (Sallin et al., 2008). The hydraulic diameter (D_h) was calculated through Equation [3] (Sperry et al., 1994; Kolb and Sperry 1999, Sellin, 2008).

$$Dh = \frac{\sum_{i=1}^{n} (\sqrt{a_i b_i})^5}{\sum_{i=1}^{n} (\sqrt{(a_i b_i})^4} \qquad [3]$$

The theoretical specific conductivity of the xylem (K_t) relates the xylem anatomy with the ideal efficiency in the water flow by the Hagen Poiseuille equation, which was modified for elliptic cross-sectional area (Calkin et al., 1986). This has undergone an addition of the following parameters: water density and cross-sectional area occupied by the xylem (Sellin et al., 2008). It was obtained by the Equation [4].

$$Kt = \frac{\pi\rho}{64\eta Ax} \sum_{i=1}^{n} \frac{a^3{}_i b^3{}_i}{a^2{}_i + b^2{}_i} \qquad [4]$$

Where ρ is the water density (kg m^{-3}), η is the dynamic viscosity of the water (both for water at 35 °C) and A_x is the transverse area occupied by the xylem (μm^2). The theoretical leaf specific conductivity (LSC) (LSC_t; $kgm^{-1}s^{-1}MPa^{-1}$), was calculated as a product between K_t and the Huber value (VH = xylem area/leaf area) (Sellin et al., 2008).

3. *Results and Discursion*

3.1. Characterization of Hydraulic Properties and Functionalities

The leaves of the nine individuals of the Early Vitória 2 (INCAPER) variety of C. canephora that were evaluated showed variations in the petiole average length. This presented as oscillating between 0.79 and 1.13 cm and being statistically equal in plants eight (P8), four (P4) and two (P2). These measures were similar among individuals P4, P3 and P5. The other individuals were similar among themselves, Table 3. The mean value of the leaf blade length ranged from 10.85 to 13.81 cm, without presenting statistically significant variations. The mean value of the leaf blade width ranged from 4.00 to 5.37 cm. Where the individual P4 presented the highest values for leaf width, being statistically different only from individuals P5 and P9. The mean values of leaf index ranged from 2.50 to 2.80 for all individuals analyzed. Because they are larger than 1, the leaf blade is longer in length than in width, Table 3.

Table 3 - Morphological characteristics of nine individuals of Coffea canephora variety Early Victoria 2 (INCAPER) cultivated in an experimental field in the Southern Forest of the State of Pernambuco, Brazil. (LI = Leaf Index)

Plants	***Petiole***	***Leaf blade***		
	Length	**Length**		**LI**
	cm	**cm**		**cm**
P1	0.82 ± 0.18 c	12.19 ± 2.86 a	P1	0.82 ± 0.18 c
P2	0.97 ± 0.26 ac	12.96 ± 3.51 a	P2	0.97 ± 0.26 ac
P3	0.91 ± 0.19 bc	12.57 ± 2.07 a	P3	0.91 ± 0.19 bc
P4	1.12 ± 0.35 ab	13.81 ± 3.03 a	P4	1.12 ± 0.35 ab
P5	0.91 ± 0.18 bc	12.37 ± 2.82 a	P5	0.91 ± 0.18 bc
P6	0.88 ± 0.14 c	12.52 ± 2.43 a	P6	0.88 ± 0.14 c
P7	0.79 ± 0.19 c	12.40 ± 3.36 a	P7	0.79 ± 0.19 c
P8	1.13 ± 0.22 a	12.82 ± 2.72 a	P8	1.13 ± 0.22 a
P9	0.84 ± 0.20 c	10.85 ± 3.06 a	P9	0.84 ± 0.20 c

Different letters in the same column indicate significant differences between individuals ($P < 0.05$).

Source: Own Author

The mean leaf area ranged from 36.27 to 58.18 cm, being higher in P4. The leaf epidermis of all the analyzed plants presented variations in the number of stomata per leaf area and in terms of polar length of the stomata. The individual P7 presented the highest value of stomas density per leaf area, followed by P3. The individuals P1 and P4 presented the lowest values. The individuals P2, P5, P6, P8 and P9 were significantly equal to P1, P3 and P4. P2, P4, P5 and P6 presented the highest polar length values of the stomas, while the lowest values were found in P1, P3, P7, P8 and P9, Table 4.

Results found for the stomatal density and polar length parameters of the stomata agree with the data obtained by Voltan (1992) with coffee of the Coffea canephora species.

According to the obtained results, the small variations found would be due to the position of the first adult leaf of the branch that was evaluated.

Table 4 - Variations of epidermal characters of the leaf of nine individuals of Coffea canephora variety Early Vitoria 2 (INCAPER) cultivated in an experimental field in the Southern Forest of the State of Pernambuco, Brazil.

Ind.	***Leaf area***	***Stomas thickness***	***Stomas polar length***
	cm^2	n. mm^{-2}	µm
P1	43.57 ± 17.75ab	251.22 ± 16.35 c	21.23 ± 1.47 b
P2	44.48 ± 19.18ab	261.79 ± 17.78 bc	22.05 ± 1.34 ab
P3	41.58 ± 9.53ab	304.07 ± 35.29 b	21.08 ± 1.20 b
P4	58.18 ± 25.36a	255.76 ± 24.47 c	21.60 ± 0.73 ab
P5	58.18 ± 25.36b	276.79 ± 15.66 bc	22.68 ± 1.24 a
P6	51.11 ± 17.78ab	282.48 ± 38.27 bc	21.90 ± 1.61 ab
P7	43.18 ± 19.14ab	352.39 ± 85.44 a	21.08 ± 1.06 b
P8	49.30 ± 18.24ab	285.89 ± 38.39 bc	21.05 ± 0.82 b
P9	36.27 ± 14.26b	271.11 ± 16.24 bc	21.00 ± 0.78 b

Different letters in the same column indicate significant differences between individuals ($P < 0.05$).

Source: Own Author

The cross-sectional area of the branch presented values ranging up to 1.54 of difference between the highest and lowest values. For the xylem cross-sectional area in these branches, the variation of the values was 0.81. Despite these results, the proportion of areas occupied by the xylem in the total cross-sectional area of the branches varied between 3.2 and 3.6. However, P4 was an exception because it presented a ratio of 2.9 between these areas. This individual stood out for having a higher value in proportion (34.96%) of the area occupied by the xylem in the branch, Table 5.

The individual P4 stood out because it presents the highest values for the parameters related to the total transverse area of the stem, the transversal area occupied by the xylem in the stem and the percentage of this area, Table 5. The lowest value for the total cross-sectional area of the branch was presented by individual P3, being significantly the same for all other individuals. The transverse area occupied by the xylem at P4 (34.96% of the branch) was significantly equal to that found at P2 and P8, and different from the others, Table 5.

The individuals P4, P8 and P2, in this sequence, presented the highest values of the total transverse area of the branch, the transversal area occupied by the xylem and percentage value of area occupied by the xylem in the branch. The individual P5 was the one that presented the fourth greater value only for the total transverse area of the branch and transversal area occupied by the xylem. The fourth highest value for the percentage area occupied by the xylem in the stem was presented by individual P3, which had the lowest values for the total transverse area of the branch and transversal area occupied by the xylem, Table 5.

Table 5 - Variations in the hydraulic characteristics of nine individuals of Coffea canephora variety Early Vitória 2 (INCAPER) cultivated in an experimental field in the Southern Brazil of Pernambuco, Brazil.

Ind.	***Total transverse area of the branch***	***Transversal area occupied by the xylem***	***Area of the area occupied by the xylem in the branch***
	mm^2	mm^2	%
P4	6.35 ± 1.07 a	2.22 ± 0.80 a	34.96
P8	5.76 ± 1.07 ab	1.78 ± 0.58 ab	30.90
P2	5.56 ± 1.07 ab	1.63 ± 0.51 ab	29.31
P5	5.44 ± 1.07 ab	1.56 ± 0.48 b	28.67
P1	5.35 ± 1.07 ab	1.49 ± 0.44 b	27.85
P6	5.35 ± 1.07 ab	1.49 ± 0.44 b	27.85
P7	5.37 ± 1.06 ab	1.51 ± 0.46 b	28.11
P9	5.39 ± 1.04 ab	1.51 ± 0.46 b	28.01
P3	4.81 ± 1.65 b	1.41 ± 0.41 b	29.31

Different letters in the same column indicate significant differences between individuals (P <0.05).

Source: Own Author

The wall thickness of the xylem conduction vessels was higher in P1 and significantly different from all other individuals. The lowest mean values for this parameter were found in P4, P5, P6, P8 and P9, Table 6. Among the individuals analyzed, the largest hydraulic diameter was found in P2 and the lowest in P1, Table 6.

Table 6 - Variation in the hydraulic characteristics of the stem of nine individuals of Coffea canephora variety Early Vitoria 2 (INCAPER) cultivated in an experimental field in the Southern Forest of the State of Pernambuco, Brazil.

Plants	***Wall Thickness***	***Hydraulic diameter of the vessel***
	(μm)	**(μm)**
P1	2.77 ± 0.36 a	17.82 ± 1.66 e
P2	1.43 ± 0.17 b	31.45 ± 2.17 a
P3	1.22 ± 0.17 bc	30.12 ± 2.48 ab
P4	1.09 ± 0.15 c	28.19 ± 0.74 bc

P5	1.03 ± 0.24 c	26.93 ± 2.70 c
P6	1.12 ± 0.17 c	29.67 ± 2.33 ab
P7	1.22 ± 0.33 bc	27.02 ± 1.81 c
P8	1.05 ± 0.10 c	28.51 ± 0.87 bc
P9	1.15 ± 0.16 c	23.13 ± 1.82 d

Different letters in the same column indicate significant differences between individuals (P <0.05).

Source: Own Author

For the nine evaluated individuals of Coffea canephora L. (Early Vitória variety 2 INCAPER) the estimated hydraulic conductivity (K_t) was higher in P2. This plant was statistically equal to P3, P4, P5, P6, P7 and P8. For this parameter the lowest value was observed in P1, being this plant statistically equal to P4, P5, P7, P8 and P9 (Table 7). Results obtained for the estimated hydraulic conductivity parameter (K_t) agree with the data obtained by (Dauzat, 2001) with coffee of the species Coffea arabica. The estimated specific conductivity of the stem (CEC_t) showed similar behavior to that observed in the hydraulic conductivity, for all plants analyzed. It can be seen in Table 7.

Table 7 - Variation in the estimated hydraulic conductivity (K_t) and estimated specific stem conductivity (CEC_t) for nine individuals of Coffea canephora cultivated in an experimental field in the Mata Sul of Pernambuco state, Brazil.

Plants	***K_t***	***CEC_t***
	$x10^{-4}$ kg s^{-1} m^{-2} MPa^{-1}	**kg $m^{-1}s^{-1}MPa^{-1}$**
P1	10.80 ± 9.06 c	152,941 ± 128,300 c
P2	84.23 ± 31.83 a	3,757,835 ± 1,420,163 a
P3	79.13 ± 34.65 ab	3,131,713 ± 1,371,602 ab
P4	45.85 ± 27.02 abc	1,814,533 ± 1,069,453 abc
P5	51.68 ± 32.24 abc	2,045,278 ± 1,276,102 abc
P6	68.79 ± 19.00 ab	2,722,464 ± 0,752,237 ab
P7	49.66 ± 32.49 abc	1,965,274 ± 1,285,819 abc
P8	56.22 ± 31.93 abc	2,225,136 ± 1,263,716 abc
P9	23.42 ± 11.45 bc	926,836 ± 453,143 bc
Average	**52.20 ± 24.06**	**2,082,445.6 ± 1,088,759.91**

Different letters in the same column indicate significant differences between individuals (P <0.05).

Source: Own Author

4. Conclusion

According to the obtained results, we can affirm that the Premature Vitória 2 variety presents a good adaptive response to the region. Thus, it can be a good option for the development of the culture commercially. This happened due to its rusticity, productivity, and lower water consumption when related to other similar crops. It is also an alternative for the recovery of degraded and abandoned areas. Which worn because of the intensive cultivation of sugarcane monoculture in this region over decades. Note that the knowledge about the physiological mechanisms of conilon coffee is still scarce in the state of Pernambuco, which is summarized to observational aspects. Therefore, still a need for further studies on this variety.

5. Acknowledgements

The authors acknowledge the Fundação de Amparo à Ciência e Tecnologia do Estado de Pernambuco (FACEPE) for the M.Sc and Ph.D. scholarships and the Instituto de Pesquisas Agronômicas (IPA) for the support on the project.

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