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PHYSICAL AND ANATOMICAL PROPERTIES OF *Hevea brasiliensis* CLONES

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ABSTRACT

Our goal was to determine physical properties and anatomical features in 33-year-old *Hevea brasiliensis* clones. We cut wood samples from clones LCB510, RRIM600, IAN873, IAN717 and GT1 planted in Selvíria, Mato Grosso do Sul, Brazil. We used standard techniques in wood studies. We found that clones differ in basic density, volumetric shrinkage and anatomical features, with the exception of ray width. Basic density, volumetric shrinkage, fiber length, fiber wall thickness, vessel element length and vessel diameter tended to increase from pith to bark, while vessel frequency propended to decrease. We conclude that wood of the studied clones has potential for industrial use.

Keywords: Basic density, cell dimensions, radial variation rubber tree, volumetric shrinkage.

INTRODUCTION

Currently, in Brazil, plantations of *Hevea brasiliensis* (rubber tree) occupy 218307 hectares (IBÁ 2019). Between 2016 and 2021 the global production of rubber extraction for industrial purposes was expected to reach 52 million m³ of wood (Dhamodaram 2008). In Brazil, after the latex production cycle (25-30 years), *Hevea* is commonly used as firewood and charcoal (Lara Palma 2010). However, logs with a diameter above, or equal to, 15 cm could be used for the production of sawn boards and panels, and logs smaller than 15 cm in diameter and larger than 5 cm could be used in the production of bioenergy (Dhamodaram 2008).

Hevea brasiliensis wood is considered light and soft with low natural durability and indistinct sapwood (Lorenzi 2002). The apparent density ranges from 0,56 g/cm^3 to 0,65 g/cm^3, and freshly cut moisture in wood is approximately 60 %, which can be reduced to 15 % when air-dried over a period of at least 10 days of exposure under these conditions (May and Gonçalves 2018). It is well known that the behavior of *H. brasiliensis* wood during the drying process is a precondition for determining its industrial use. For example, volumetric shrinkage, a physical property, indicates how much wood dimensions change according to variation of humidity in the environment. This, in turn, will determine if cracks will occur when, for example, using this wood for the manufacture of doors and windows (Rubber Board 2002). Rubber tree wood is also technically feasible for wood cement board manufacture; sheets for the production of vertically laminated veneer lumber panels (LVL); floors; wood beams, stairway steps (Okino *et al.* 2004, Faria *et al.* 2019a).

However, fast-growing species, such as rubber trees, show frequent problems inherent in wood quality, such as a high percentage of sapwood, which results in less resistance to deterioration, less dimensional stability and low physical-mechanical resistance (Shukla and Sharma 2018). A major problem with the use of products derived from rubber wood arises from the high susceptibility to attacks by xylophagous agents, mainly fungi and insects. This occurs from the indistinctness of heartwood and the high content of starch and sugars present in wood (Peries 1980). Therefore, prophylactic treatment is recommended immediately after cutting the wood, *i.e.*, less than 24 hours (Prakash 1990).

The physical, mechanical and anatomical properties of rubber wood must be studied in order to properly assess its quality before determining its end use after extracting latex from the tree (Naji *et al.* 2012). Not many studies in the literature have reported on radial variation of *H. brasiliensis* wood properties (Leonello *et al.* 2012). The lack of research focused on the quality and varied uses of rubber wood in Brazil calls for more efforts to better characterize the potential industrial use of *Hevea* wood. Therefore, our goal was to determine physical properties and anatomical features in 33-year-old *Hevea brasiliensis* clones (LCB510, RRIM600, IAN873, IAN717 and GT1) in the context of industrial use after the extraction of latex.

MATERIALS AND METHODS

Location and sampling

Wood samples from *H. brasiliensis* were obtained from five clones (LCB510, RRIM600, IAN873, IAN717and GT1) from plantations located in the municipality of Selvíria, Mato Grosso do Sul State, Brazil. The locations have a mean annual precipitation of 1440 mm/yr and an annual average temperature of 23 ºC (Flores *et al.* 2016). The soil is classified as dystrophic Red Latosol (LVd) according to Santos *et al.* (2018).

The northward position of each selected tree was identified to standardize the collection of wood samples. Then, we felled five randomly selected 33-year-old trees per clone and cut discs 10 cm in thickness from each tree at breast height (D, 1.3 m from the ground). Tree height and DBH of selected trees are shown in Table 1. In each disc, we cut five samples in each strip from pith to bark**.** For a total of 25 samples per clone: 0 % (close to the pith), 25 %, 50 %, 75 %, and 100 % (close to the bark).

Table 1: Mean and standard deviation of DBH and tree height (HT) of *Hevea brasiliensis*.

Clone	DBH (cm)	HT (m)
LCB510	25,20 (0,57)	16,94 (2,97)
RRIM600	24,10 (0,55)	18,76 (1,10)
IAN873	23,30 (1,20)	17,52 (1,74)
IAN717	27,00 (2,03)	20,94 (1,52)
GT1	25,00 (1,00)	18,76 (2,42)
Values in parentheses are standard deviation		

Basic density (Db)

Basic density was determined by the method of maximum moisture content NBR (ABNT 2003). Samples of 2 cm x 2 cm x 3 cm were saturated by treatment with a vacuum system for 72 h to obtain saturated volume of wood. In sequence, the samples were dried in a laboratory kiln to determine the oven-dried mass at 103 ºC ± 2 °C.

Volumetric shrinkage (ε_v)

Volumetric shrinkage was determined according to the NBR (ABNT 1997). The samples were saturated in water, measured with a caliper, and oven-dried at 103 ºC ± 2°C. The dry volume of each sample was then determined. The difference in percentage between the two measurements is the volumetric shrinkage.

Anatomical features

Wood samples (2 cm^3) were softened by cooking in water and glycerin in a proportion of (4:1) until they presented ideal conditions for sectioning. Histological sections 20 µm in thickness were obtained using Leitz 1208 and Zeiss-Hyrax S50 slide microtomes. Sections were clarified by washing in 60 % sodium hypochlorite to remove cell contents; stained with safranin; Provisional slides were mounted in 60 % glycerin for measurements (Johansen 1940).

In addition to the histological sections, dissociated wood was prepared according to Franklin method (Berlyn and Miksche 1976). Thin sticks were cut and placed in wheaton containers, containing 100 volume hydrogen peroxide solution and glacial acetic acid (1:1). The containers were sealed with adhesive tape and remained 48 hours in an oven at 60 ºC. Subsequently, the material was washed with running water and stained with 1 % alcoholic safranin. The terminology and characterization of wood followed the IAWA list (IAWA 1989). All anatomical measurements were obtained with a microscope Olympus CX 31 equipped with a camera Olympus Evolt E330 and a computer with image analyzer software Image-Pro 6.3. (Software Media Cybernetics 2021).

Statistical analyses

To evaluate the effect of clone x radial variation within the tree on physical and anatomical properties, the variance homogeneity test was initially performed through Hartley's test and later the F test of variance analysis according to the experimental design of randomized blocks. The F test was applied (P > 0,05), and the means were compared using Tukey's test. The relationship between variables was evaluated using Pearson's correlation. Statistical analyses were conducted using the SAS statistical program (SAS 1999).

RESULTS AND DISCUSSION

Table 2 shows the results of analysis of variance. No statistically significant difference was observed in ray width for clones. No statistically significant difference was observed in basic density, ray width or ray frequency for radial position. Furthermore, no significant interaction was noted between clones x radial position for all properties, demonstrating no dependence among these variables.

Physical Properties

The average value for basic density was 0,58 g/cm^3 (Table 2). Our results showed that *H. brasiliensis* clones are considered moderately heavy, ranking it in the class C20 by NBR (ABNT 1997). Wood basic density varied according to clone type from 0,56 g/cm^3 at GT1 to 0,60 g/cm^3 at LCB510 (Table 3). The wood density averages (Table 3) are higher than those calculated by

Chukwuemeka (2016). The basic density value obtained for clone RRIM600 is similar to that found by Raia *et al.* (2018). However, Santana *et al.* (2001) reported lower values for 40-year-old *H. brasiliensis* clones. These differences between density values, when compared to the literature, can be explained by such factors as genetics, different tree ages, and/or the local characteristics of each plantation (Chaendaekattu and Mydin 2018; Rungwattana *et al.* 2018).

Table 2: Analysis of variance of basic density (BD), volumetric shrinkage (ε_v), fiber length (FL), fiber wall thickness (FWT), vessel element length (VEL), vessel diameter (VD), vessel frequency (VF), ray width (RW), ray height (RH) and ray frequency (RF) of 33-year-old *Hevea brasiliensis*.

		Mean squares									
Causes of variation	GL	BD (g/cm^3)	ε_v (%)	FL (µm)	FWT (µm)	VEL (µm)	VD (µm)	VF (n/mm^2)	RW (µm)	RH (µm)	RF (n/mm^1)
Clone (C)	4	0,0053**	30,04**	1578281**	2,56**	85342**	3679 **.	5,42 **.	225 n.s.	44270**.	22,91**
Radial Position (RP)	4	0,0016 n.s.	12,90**	433454**	1,69 **	69727 **	6759 **	15,55 **	240 n.s.	12314 **	0,44 n.s.
(C) x (RP)	16	0,0005 n.s.	1,98 n.s.	17880 n.s.	0,24 n.s.	3549 n.s.	519 n.s.	0,48 n.s.	207 n.s.	12314 n.s.	0,48 n.s.
Residual	100	0,0008	2,27	21584	0,18	4838	337	0,64	222	2926	0,78
Mean		0,58	8,32	1244	4,48	758	182	3,14	46	413	11,87
Standard Deviation		0,03	1,86	197	0,57	96,76	26	1,18	14,88	65,51	1,20
CV_e (%)		5,02	18,14	11,80	9,4	9,17	10,05	25,47	32	13,07	7,43

** significant at 1% level of significance; n.s. = not significant and CV_e= coefficient of experimental variation

The average value obtained for volumetric shrinkage of 8,32 % (Table 2) is considered low for rubber trees (Mainieri and Chimelo 1989). These values are lower than those verified by Raia *et al.* (2018).

Clones IAN717 and GT1 had the smallest volumetric shrinkage, and clone RRIM600 had the highest (Table 3). Our results differ from those of Santana *et al.* (2001), who reported lower values for volumetric shrinkage of clones IAN717 and GT1 compared to our study. The volumetric shrinkage values of clone RRIM600 are similar to those obtained by Lara Palma (2010).

Anisotropy values found for rubber trees are usually high; therefore, this species is considered very unstable for production of wooden furniture (Raia *et al.* 2018). However, some *Eucalyptus* species, which are already being used commercially, also present values of anisotropy very close to that of the rubber tree and, hence, may not be an obstacle to the use of this wood for certain purposes (Batista *et al.* 2010; Santana *et al.* 2001). Minimal variations in shrinkage and swelling along the stem in rubber wood were reported by Owoyemi *et al.* (2018). Rubber wood to be used in internal flooring must undergo thermal modification processes to improve dimensional stability (Emmerich and Militz 2020).

Table 3: Average of basic density (BD), volumetric shrinkage (ε_v), fiber length (FL), fiber wall thickness (FWT), vessel element length (VEL), vessel diameter (VD), vessel frequency (VF), ray width (RW), ray height (RH) and ray frequency (RF) of 33-years-old *Hevea brasiliensis*.

Treatment	**BD (g/cm^3)**	**(ε_v) (%)**	**FL (µm)**	**FWT (µm)**	**VEL (µm)**	**VD (µm)**	**VF (nº/mm^2)**	**RA (µm)**	**RW (µm)**	**RF (nº/mm^1)**
LCB510	0,60 ^{a} (0,01)	8,11 ^{bc} (1,82)	1247 ^{b} (204)	4,89 ^{a} (0,68)	769 ^{b} (89)	179 ^{b} (26)	3,00 ^{b} (1,05)	468 ^{a} (62)	44 ^{a} (5,43)	10,63 ^{c} (1,22)
RRIM600	0,59 ^{ab} (0,03)	9,69 ^{a} (1,69)	1187 ^{b} (207)	4,65 ^{ab} (0,56)	731 ^{bc} (90)	184 ^{ab} (26)	3,01 ^{b} (1,15)	366 ^{a} (37)	46 ^{a} (4,28)	11,95 ^{b} (0,75)
IAN873	0,57 ^{b} (0,03)	9,18 ^{ab} (1,75)	1174 ^{b} (172)	4,05 ^{d} (0,37)	699 ^{c} (70)	164 ^{c} (26)	3,89 ^{a} (1,40)	378 ^{cd} (61)	42 ^{a} (3,84)	13,31 ^{a} (0,92)
IAN717	0,57 ^{b} (0,03)	7,54 ^{c} (1,61)	1239 ^{b} (203)	4,50 ^{bc} (0,48)	741 ^{bc} (75)	188 ^{ab} (19)	2,62 ^{b} (0,65)	418 ^{bc} (59)	45 ^{a} (6,56)	11,71 ^{b} (0,44)
GT1	0,56 ^{b} (0,02)	7,07 ^{c} (1,06)	1374 ^{a} (136)	4,31 ^{cd} (0,32)	853 ^{a} (87)	197 ^{a} (21)	3,18 ^{a} (1,20)	437 ^{ab} (48)	50 ^{a} (32)	11,70 ^{b} (0,73)
Radial Position	0,57 ^{a} (0,03)	7,31 ^{c} (2,02)	1083 ^{d} (1,92)	4,11 ^{d} (0,35)	695 ^{d} (77)	162 ^{c} (23)	4,02 ^{a} (1,28)	395 ^{b} (63)	42 ^{a} (5,29)	11,74 ^{a} (1,35)
Radial Position	0,57 ^{a} (0,04)	7,81 ^{bc} (1,66)	1149 ^{cd} (162)	4,33 ^{cd} (0,61)	725 ^{cd} (80)	170 ^{c} (23)	3,86 ^{a} (1,27)	408 ^{ab} (66)	49 ^{a} (32)	11,76 ^{a} (1,04)
Radial Position	0,58 ^{a} (0,03)	8,73 ^{ab} (1,99)	1247 ^{bc} (156)	4,46 ^{bc} (0,42)	757 ^{bc} (93)	185 ^{b} (22)	3,05 ^{b} (0,96)	394 ^{b} (66)	44 ^{a} (4,50)	12,01 ^{a} (1,18)
Radial Position	0,58 ^{a} (0,03)	8,99 ^{a} (1,37)	1348 ^{ab} (117)	4,71 ^{ab} (0,55)	788 ^{ab} (83)	196 ^{ab} (18)	2,42 ^{bc} (0,41)	425 ^{ab} (59)	47 ^{a} (5,49)	12,00 ^{a} (1,27)
Radial Position	0,59 ^{a} (0,02)	8,74 ^{ab} (1,77)	1397 ^{a} (165)	4,80 ^{a} (0,60)	829 ^{a} (94)	200 ^{a} (23)	2,33 ^{c} (0,45)	447 ^{a} (63)	48 ^{a} (4,45)	11,80 ^{a} (1,20)

Values in parentheses are standard deviation. Means followed by different letters on the same column indicate different mean values for the Tukey test (at 5 % level of significance).

Anatomical features

Only ray width did not differ significantly among the clones (Table 2). Fiber length of clone RRIM600 (Table 3) is shorter than that obtained by Suhaimi and Sahri (2003).

Clones LCB510 and RRIM600 showed the highest values of fiber wall thickness, which differed statistically from that of clone IAN873, which had the lowest value (Table 3). Fiber wall thickness (Table 3) was lesser than that found by Teoh *et al.* (2011) and Norul Izani and Sahari (2008) in their studies.

In general, clones in the present study showed fiber dimension values below the average found in the literature. However, for fiber length and fiber wall thickness, similar values were obtained by Ramos *et al.* (2018). In contrast, longer fibers with thicker walls were found in 20-year-old *Hevea brasiliensis* studied by Faria *et al.* (2019b) who reported that the rubber tree has potential for use in cellulose and paper production. However, wall fraction index was high, and flexibility coefficient was low which could drastically interfere in cellulose and paper production.

If there is a negative correlation between growth and fiber length according to Chaendaekattu and Mydin (2018), it may not be possible to simultaneously attain vigorous growth and longer fibers. Clone RRIM2020, which grew in a wider spacing, had shorter fiber length than trees growing in higher population density (Saffian *et al.* 2014). This growth relationship was tested with 100% radial position anatomy results and no significant relationship for our data was found. The population density of trees in our study was 500 tree/ha, and this density is the most used in *H. brasiliensis* planting in Brazil.

Vessel element length differed among clones, with clone LCB510 having the highest mean (769 µm) and clone IAN873 the lowest (670 µm) (Table 3). Vessel diameter was the same in clones LCB510 and RRIM600, but narrower in clone IAN873 (Table 3). These values are within the standard (70 µm – 224 µm) for rubber tree (Reghu 2002). *Hevea brasiliensis* wood usually presents large vessel diameter values (Schoch *et al.* 2004).

Vessel frequency was higher for clone IAN873 and lower for clone IAN717 (Table 3). Ray width did not differ among clones (Table 3). Ray height was taller in clone LCB510 and shorter in clone RRIM600, while ray frequency was higher in clone IAN873 and lower in clone LCB510 (Table 3). Values of 47 (µm), 523 (µm) and 8 (nº/mm^{1}), respectively, for width, height and ray frequency for

H. brasiliensis wood submitted to latex exploration were obtained by Ramos *et al.* (2016). These values are, on average, higher than those observed in our study.

In general, we observed that clone LCB510 showed higher values for basic density and fiber wall thickness, but lower ray frequency. These values suggest high resistance in comparison to the other clones in this study. On the other hand, clone IAN873 had lower basic density and fiber wall thickness and higher vessel frequency and ray frequency, making it a material with less strong wood features. Thicker wall fibers and higher wood density strike a positive correlation previously found in *Pittosporum undulatum* wood by Longui *et al.* (2011). To explain, fiber cells are more frequent than other wood cells; thus, fibers are positively correlated to higher density owing to mass increase (Fujiwara *et al.* 1991).

Pith-bark Variation

Basic density and volumetric shrinkage tend to increase from pith to bark (Table 3). While this variation was not enough for significant differentiation to occur for basic density, it did occur for volumetric shrinkage (Table 3). No significant difference was noted between juvenile and adult wood for basic density by the presence of high levels of extractives in juvenile wood, which is usually closer to the pith (Severo *et al.* 2013).

Most tropical species have a tendency to present smaller cellular dimensions in the pith region compared to wood close to the bark. The same pattern occurs in rubber wood, with the exception of vessel frequency, where the reverse phenomenon normally occurs (Table 3). However, this trend was not significant for ray width and ray frequency. This same trend also occurred in *Astronium lecointei* (Melo *et al.* 2013).

Narrower and fewer vessels are observed in the pith region, while wider and fewer vessels are found in the region close to the bark (Figures 1A and 1B). The pattern of variation in cell dimensions was very similar to that found by Lima *et al.* (2011) in *Cariniana legalis* and Melo *et al.* (2013) in *Astronium lecointei*.

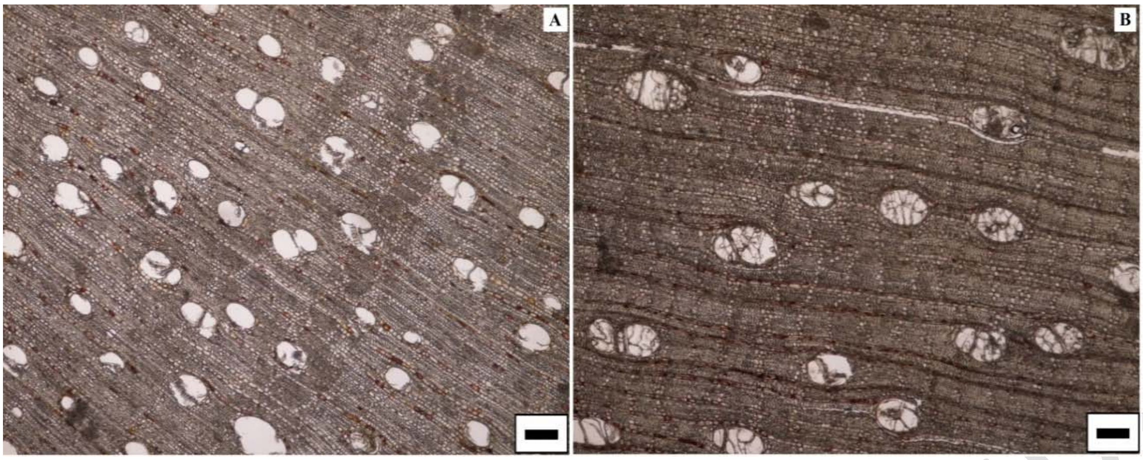

Figure 1: Photomicrographs of *Hevea brasiliensis* wood. A and B – Transversal sections – A: from the pith region and B: from the bark region. Note the smaller diameter and higher frequency of vessel (A) and the larger diameter and lower vessel frequency (B). Scale bar = 100 µm.

To better explain the relationships among basic density, volumetric shrinkage and cell dimensions, relative to radial position, Pearson's correlation analyses were performed (Table 4). Only ray dimensions failed to show a significant relationship with radial position. In addition, vessel diameter had a strong negative relationship with radial position (Table 4).

Based on these significant correlations, we performed regression analyses to verify the best models to explain these relationships. Almost all regression models showed positive correlations with radial position, except volumetric shrinkage. This model represented only 72 % of correlations among these variables (Figures 2 and 3).

Table 4: Pearson's Correlation Coefficient (PCC) obtained for correlations among the variables studied and radial position.

	BD	**ε_v**	**FL**	**FWT**	**VL**	**VD**	**VF**	**RH**	**RW**	**RF**
Radius (cm)	$0,94^{**}$	$0,89^{*}$	$0,99^{**}$	$0,99^{**}$	$0,99^{**}$	$-0,98^{**}$	$0,96^{**}$	$0,85^{n,s,}$	$0,57^{n,s,}$	$0,41^{n,s,}$

** significant at level 1% of significance; * significant at level 5% of significance and n.s = not significant.

Basic density, fiber length, volumetric shrinkage and fiber wall thickness had very similar behavior, i.e., they showed a tendency of stabilization at a distance of 9 cm from the log radius. This indicates the occurrence of a transition between juvenile and adult wood (Figures 2A, 2B, 2C and 2D).

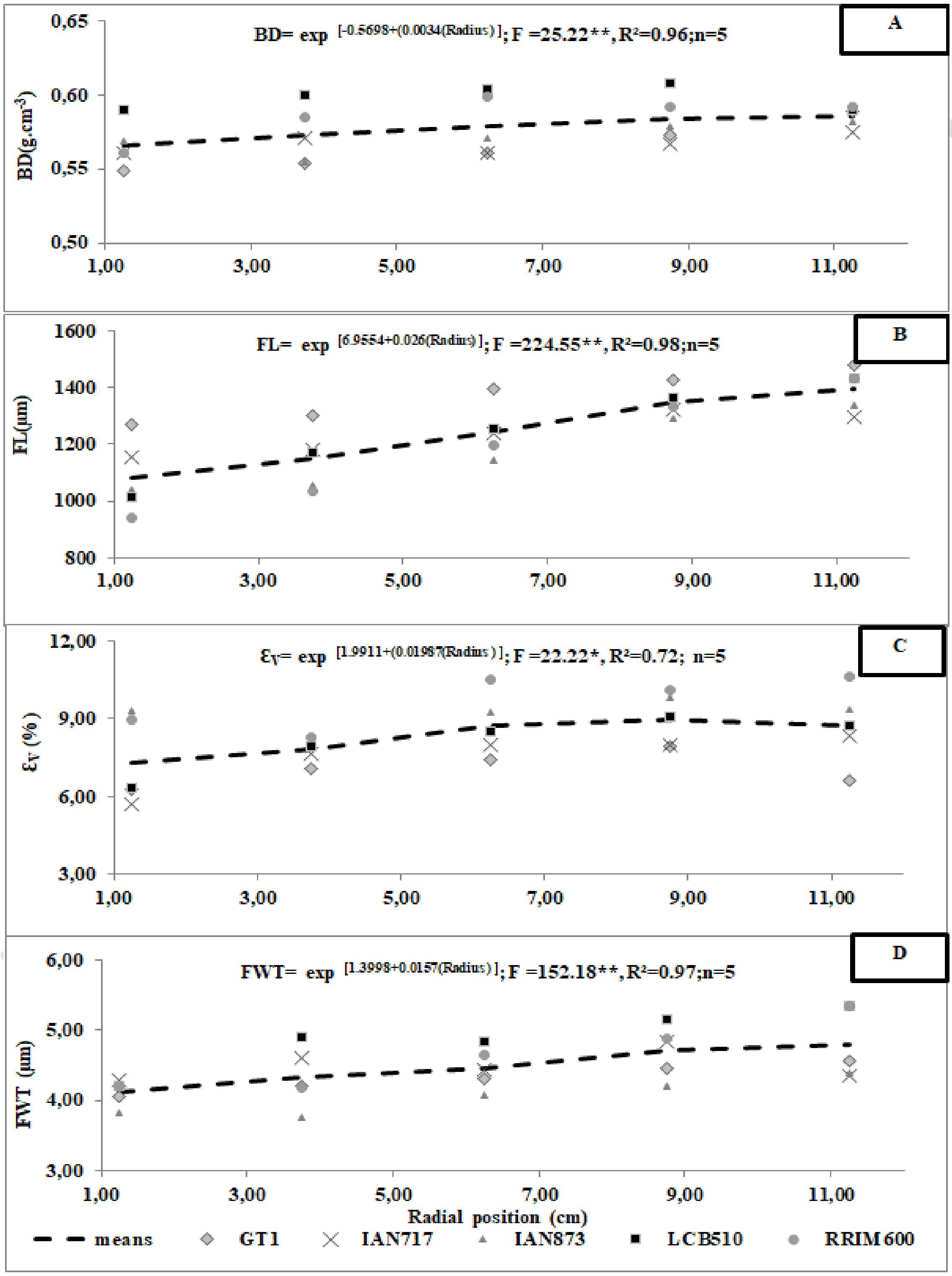

Figure 2: Correlation between basic density and radial position (A), fiber length and radial position (B), volumetric shrinkage and radial position (C), and fiber wall thickness and radial position (D) of 33-year-old *Hevea brasiliensis*.

Ferreira *et al.* (2011) found the stabilization point around 4,0 cm – 5,5 cm in the log radius when analyzing fiber length of 50-year-old *H. brasiliensis*. This may have been the result of age difference among the analyzed trees since the presence of cells with larger dimensions close to cambium is related to tree aging (Wilkes 1988). Wood density of *H. brasiliensis* do not have any significant correlation with tree growth and fiber length, as these properties have strong genetic control (Chaendaekattu and Mydin 2018, Rungwattana *et al.* 2018).

Vessel element length and vessel diameter progressively increased toward the bark, while the inverse occurred for vessel frequency (Figures 3A,3B and 3C). This occurred because juvenile wood in the pith region exhibits greater physiological activity, thereby producing a greater number of narrower vessels.

Consequently, narrower vessels in juvenile wood are related to tradeoff lower hydraulic conductivity and higher embolism resistance, since that ability of vessels to conduct water increases proportionally with diameter, but large vessels in adult wood, which results in higher hydraulic conductivity can be submitted vessel to embolism under high water potentials (Wheeler *et al.* 2005; Lachenbruch and McCulloh 2014, Santiago *et al.* 2018, Simioni *et al.* 2020).

Also, as the transition from juvenile to adult wood occurs, the bark region presents fewer wide vessels (Wilkes 1988, Melo *et al.* 2013). However, according to Santos *et al.* (2019), cell dimensions of rubber wood can vary, depending on the different types of wood (tension, reaction and normal) in each radial position sampled.

Based on the results, the evaluated rubber tree clones have the necessary potential for use in some industrial activities that do not require much physical resistance in civil construction or the manufacture of different types of decorative objects and handcrafts. This was also the conclusion of Raia *et al.* (2018) for *H. brasiliensis* wood based on the homogeneity of its physical properties along tree height. They also found that the rubber tree presents characteristics similar to those of other species already used commercially.

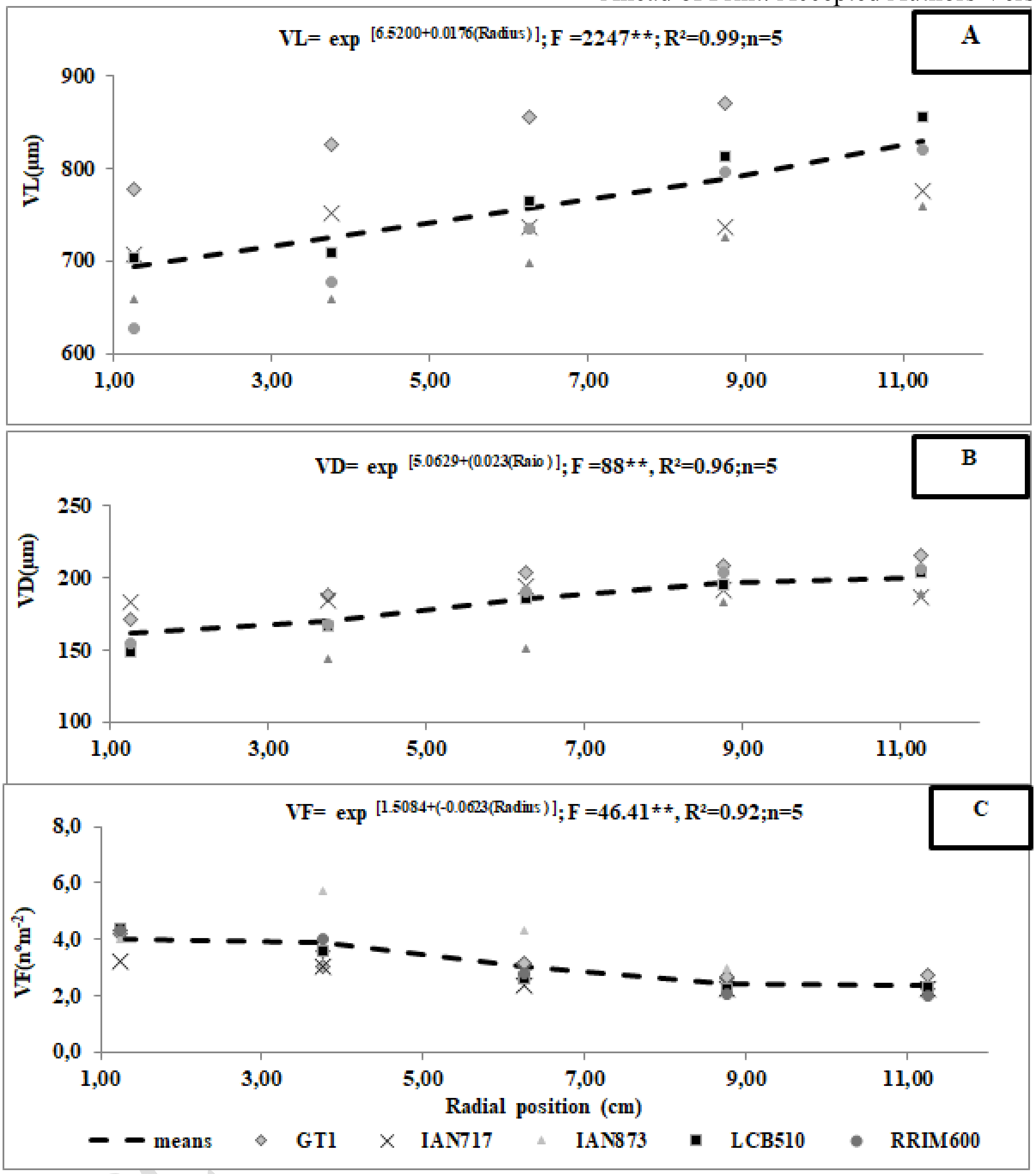

Figure 3: Correlation between vessel length and radial position (A), vessel diameter and radial position (B), and vessel frequency and radial position (C) of 33-year-old *Hevea brasiliensis*.

CONCLUSIONS

Hevea brasiliensis clones differ from each other with respect to basic density and volumetric shrinkage, as well as almost all anatomical dimensions, with the exception of ray width. The interaction between clones x radial position was not significant, demonstrating no dependence among these variables. Basic density, ray width and ray frequency do not differ with respect to radial position. It appears that height, width and ray frequency have no significant correlation with radial position. Basic density, volumetric shrinkage, fiber length, fiber wall thickness, vessel element length and vessel diameter tend to increase towards bark. Vessel frequency has a tendency to decrease toward the bark. In general, we can consider that these clones have potential for use in civil construction in light structures and the manufacture of furniture and different types of decorative objects.

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