

ANATOMICAL, ULTRASTRUCTURAL, PHYSICAL AND MECHANICAL WOOD PROPERTIES OF TWO-YEAR-OLD *Eucalyptus grandis* × *Eucalyptus urophylla* CLONES¹

Antonio José Vinha Zanuncio^{2*}, Amélia Guimarães Carvalho², Angélica de Cassia Oliveira Carneiro³, Mario Tomazello Filho⁴, Paulina Valenzuela⁵, William Gacitúa⁵ and Jorge Luiz Colodette³

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² Universidade Federal de Uberlândia, Instituto de Ciências Agrárias, Monte Carmelo, MG-Brasil. E-mail: <ajvzanuncio@yahoo.com.br> and <ameliagcarvalho@gmail.com>.

³ Universidade Federal de Viçosa, Departamento de Engenharia Florestal, Viçosa, Mg-Brasil. E-mail: <cassiacarneiro1@gmail.com> and <colodett@ufv.br>.

⁴ Universidade de São Paulo, Departamento de Ciências Florestal, Piracicaba, SP-Brasil. E-mail: <mtomazel@usp.br>.

⁵ Universidad del BioBio, Centro de Biomateriales e ingeniería, Chile. E-mail: <pnvalenzu@ubiobio.bl> and <gacitua@ubiobio.bl>.

*Corresponding author.

ABSTRACT – *Eucalyptus* wood from adult trees is used for several purposes; however, the wood of younger trees has limited use. This study aims to characterize and propose uses of two-year-old eucalyptus wood. Six two-year-old *Eucalyptus grandis* × *Eucalyptus urophylla* clones have been selected and their anatomical, ultrastructural, physical and mechanical wood characteristics evaluated. The wood of Clone A shows more robust fibers with better microfibril arrangement, resulting in better mechanical properties, and therefore, a better performance for structural use. Clone F showed a low variation of wood basic density in the radial direction, facilitating its machinability, and with the Clone B, showed a lower anisotropy, and therefore, the wood is recommended for locations with high variations of humidity. The heterogeneity of the wood characteristics of the evaluated clones confirms the need for further studies, to choose those most adequate to each use.

Keywords: *Fiber; Nanoindentation; X-ray densitometry*

CARACTERÍSTICAS ANATÔMICAS, ULTRAESTRUTURAIS, FÍSICAS E MECÂNICAS DA MADEIRA DE CLONES DE *Eucalyptus grandis* × *Eucalyptus urophylla* COM DOIS ANOS DE IDADE

RESUMO – A madeira de eucalipto de árvores adultas é utilizada para os mais diversos fins, entretanto, a utilização desta madeira de árvores de idade precoce é limitada. O objetivo foi caracterizar e propor utilizações para madeira de eucalipto com dois anos de idade. Seis clones de *Eucalyptus grandis* × *Eucalyptus urophylla*, com dois anos de idade, foram selecionados e suas características anatômicas, ultraestruturais, físicas e mecânicas avaliadas. A madeira do clone A apresentou fibras mais robustas e de melhor arranjo microfibrilar, resultando em melhores propriedades mecânicas e, por isto, melhor desempenho para uso estrutural. O clone F apresentou madeira com pouca variação da densidade básica no sentido radial, facilitando sua trabalhabilidade e, com o clone B, apresentou menor coeficiente de anisotropia e, por isto, a madeira é recomendada para locais com alta variação de umidade. A heterogeneidade das características da madeira dos clones avaliados corrobora a necessidade estudos por clone para uma sua melhor utilização.

Palavras-Chave: *Fiber; Nanoindentação; Densitometria de raio-X*



1. INTRODUCTION

The forest sector is very important for the Brazilian economy (IBA, 2015; FAO, 2015). *Eucalyptus* plantations aim to produce wood for various purposes, such as panels (Bal and Bekta^o, 2014; Castro et al., 2014), cellulose (Gomes et al., 2014; Carvalho et al., 2015), energy (Zanuncio et al., 2013a, Zanuncio et al., 2014a), and lumber (Ananias et al., 2014).

Wood is a heterogeneous material, making its use difficult (Kollmann and Côté, 1968). The anatomical and ultrastructural characteristics reflect the physical and mechanical behavior of wood (Muñoz et al., 2012; Longui et al., 2014), and therefore, its use depends on a complete survey of these features.

Woods with higher basic density have higher mechanical strength and higher volume variation due to air relative moisture changes (Hein et al., 2013; Schulgasser and Witztum, 2015). Furthermore, woods with low microfibril angle and higher cell wall fraction also tend to be more resistant (Hein et al., 2013; Longui et al., 2014).

Silvicultural practices such as thinning and natural phenomena such as wind damages, can induce wood harvest when the trees are young. The lack of alternatives for this material leads to its use for energy (Guerra et al., 2014). However, its use in the production of small objects (Vieira et al., 2010) and in the furniture industry (Lopes et al., 2011) is unexplored and can add value to this wood type.

The wood from two-years-old trees should be used to increase the gain of investments in the forestry sector. The use of this wood depends on a complete study of their anatomy, ultrastructure, physics and mechanics. Therefore, the aim of this study was to characterize the anatomical, ultrastructural, physical and mechanical wood properties of two-year-old *Eucalyptus grandis* × *Eucalyptus urophylla* clones and suggest uses for this material.

2. MATERIAL AND METHODS

2.1. Biological Material

Three two-year-old trees were selected from each of the six *Eucalyptus grandis* × *Eucalyptus urophylla* clones from Belo Oriente, Minas Gerais State, Brazil, 42°22'30" South longitude" and 19°15'00" West latitude",

the height and diameter of trees represented the average of settlement, the diameter varied between 10.5 and 11.4 cm and the height varied between 15.4 and 17.5 m among clones. In each tree, three 5 cm thickness disks were removed at 1.3 m above the ground level to determine the wood basic density, anatomy and ultrastructure. A three-meter log was removed in each tree from just above this position and a central plank was obtained to make the samples for the mechanical characterization and evaluation of the dimensional and volumetric variation of the wood.

2.2. Wood anatomical characterization

A wood sample was obtained from an intermediate position from pith to bark, in one of the 5 cm disks that was removed from a tree trunk at 1.3 m above the ground level. Histological slides (Johansen, 1940) and macerated materials were prepared (Franklin, 1945). The length and width of the fiber, lumen diameter, diameter and frequency of the vessels, and height and width of the rays were measured using optical microscope and Axio Vision LE Rel. 4.3 program. Each anatomical parameter evaluated was measured 30 times in each sample per tree, totaling 90 measurements per clone. The cell wall thickness of the fiber was obtained by the difference between the width of the fiber and the lumen diameter, divided by two. The cell wall fraction was calculated using the equation: $C.W.F. = ((2 \times C.W.T.) / F.W.) \times 100$, where, C.W.F. = Cell wall fraction (%); F.W. = fiber width (µm); C.W.T. = cell wall thickness (µm).

2.3. Microfibril angle measurement

The microfibril angle of the S2 layer was measured in the wood specimens used in the anatomical characterization. After saturation, the wood blocks were cut with a microtome in the tangential plane in 10 µm thick sections and macerated with hydrogen peroxide solution and glacial acetic acid in the ratio 2:1 at 55°C for 24 hours to prepare temporary slides (Leney, 1981). The measurement of the microfibril angle was performed by polarized light microscopy with an Olympus BX51 microscope adapted with a rotary stage, graduated from 0° to 360°. The microfibril angle was measured on 30 fibers in each tree, totaling 90 measurements per clone

2.4. Nanoindentation

A wood sample was removed in intermediate position from pith to bark. A 3 × 3 × 3 mm specimen was made from this sample and embedded in epoxy resin solution to determine the modulus of elasticity and hardness

of the S2 layer and the middle lamella. The nanoindentation was performed in a TriboIndenter Hysitron TI-900®. The maximum applied load was 100 μN for 60 seconds, with discharge performed in 20 $\mu\text{N/s}$. The modulus of elasticity of the fiber was determined according to the equation: $E = (1 - \nu_m^2) \times (1/E_r - (1 - \nu_i^2/E_i))^{-1}$, where: E = Modulus of elasticity (MOE) in GPa; following manufacturer's instructions, $\nu_i = 0.07$; $\nu_m = 0.35$ and $E_i = 1140$ GPa. The reduced modulus (E_r) was obtained from the load-displacement curve, particularly from the initial slope when elastic response was generated (Muñoz et al., 2012).

The fiber hardness was determined by the maximum load supported by the specimen divided by the contact area, according to the equation: $H = P_{\max}/A$, where: H = Hardness (GPa); P_{\max} = maximum load of indenter penetration; A = projected contact areas at maximum load.

These procedure was performed in 30 fibers per tree, resulting in 90 measurements per clone

2.5. Characterization of the wood physical properties

The wood basic density was determined by the ratio between the dry mass and green volume of wood in one of the 5 cm disks removed from 1.3 m above the ground level, according to NBR1194: 2003 (ABNT, 2003).

The wood samples were subjected to X-ray densitometry, to determine their apparent density variation in the radial direction. Diametral samples were obtained in one of the disks removed at 1.3 m above the ground. These sections were conditioned at 23°C and 50% relative humidity, after this period, the samples showed 10% moisture on the dry basis. The analysis was performed using the TRQ-01XTree-Ring Analyzer equipment.

Thirty samples ($2 \times 2 \times 4$ cm) per clone were saturated with water, the volume were recorded by immersion in water and the radial and tangential dimensions were measured with a caliper. Then, the samples were dried at 103°C and the volume and dimensions were recorded again. The volumetric swelling of the wood was determined using the equation: $VS(\%) = (V_s - V_d) \times 100/V_d$, where: $VS(\%)$ = volumetric swelling; V_s = volume of saturated wood and V_d = volume of dry wood. The radial swelling was determined using the equation: $RS(\%) = (RL_s - RL_d) \times 100/RL_d$, where: $RS(\%)$ = radial swelling; RL_s = radial length of the saturated

wood and RL_d = radial length of dry wood. The tangential swelling was calculated according to the equation: $TS(\%) = (TL_s - TL_d) \times 100/TL_d$, where: $TS(\%)$ = tangential swelling; TL_s = tangential length of saturated wood and TL_d = tangential length of dry wood. Finally, the anisotropy was determined by the ratio between the tangential and radial swellings.

The dry wood mass was obtained from thirty samples ($2 \times 2 \times 4$ cm), dried at 103°C and placed in a climatic chamber at 23°C and 50% relative humidity for 15 days. The equilibrium moisture content was calculated using the equation: $EMC(\%) = (WM - DM) \times 100/DM$, in which: EMC = equilibrium moisture content; WM = wet mass and DM = dry mass.

2.6. Wood mechanical characterization

Only the static bending and parallel compression were performed, due to the low diameter of the trees, the preparation of specimens for other tests became infeasible. The wood samples were conditioned at 23°C and 50% relative humidity to stabilize their mass. The compression parallel to the grain was determined from the samples with $2 \times 2 \times 4$ cm, and the modulus of elasticity (MOE) and rupture (MOR) from samples with $2 \times 2 \times 30$ cm, in a procedure adapted from D 143–94: 1997 (ASTM, 1997). Thirty wood samples were used per clone.

2.7. Statistical analysis

The variance homogeneity (Bartlett's test at 5% significance) and normality test were performed (Shapiro-Wilk test at 5% significance). The means per parameter of each clone were compared with the Scott-Knott test at 5% probability.

3.RESULTS

3.1. Wood anatomical and ultra-structural characterization

The wood anatomical parameters evaluated varied among the clones (Table 1). The height and width of the rays showed higher coefficients of variation in the classification of histological sections of *Eucalyptus* wood. The lumen diameter, cell wall thickness and cell wall fraction showed higher values for this parameter in the fiber classification, indicating constituents with higher wood variation. All parameters in the evaluation of the wood ultrastructure had coefficient of variation below 10%.

Table 1 – Wood anatomical and ultrastructural characterization of two-year-old *Eucalyptus grandis* × *Eucalyptus urophylla* clones.**Tabela 1** – Caracterização anatômica e ultraestrutural da madeira dos clones de *Eucalyptus grandis* × *Eucalyptus urophylla* com dois anos de idade.

| Cl. | F.L. (mm) | F.W. (µm) | L.D. (µm) | C.W.T. (µm) | C.W.F. (%) |
|-----|---------------------------|--------------------------------|----------------------------|--------------------------|---------------------------|
| A | 0.971 ^(15.2) c | 18.5 ^(13.3) b | 9.8 ^(17.3) b | 4.36 ^(16.8) c | 47.3 ^(15.2) c |
| B | 0.822 ^(14.7) a | 17.6 ^(13.2) b | 10.4 ^(16.4) c | 3.57 ^(17.3) a | 40.9 ^(16.3) a |
| C | 0.929 ^(14.6) c | 20.1 ^(12.1) c | 12.1 ^(16.4) d | 3.98 ^(17.1) b | 39.7 ^(17.2) a |
| D | 0.873 ^(14.6) b | 18.0 ^(12.4) b | 10.2 ^(16.8) c | 3.92 ^(17.2) b | 43.7 ^(16.2) b |
| E | 0.880 ^(15.7) b | 17.9 ^(13.7) b | 10.2 ^(16.9) c | 3.80 ^(17.7) b | 42.5 ^(17.5) b |
| F | 0.794 ^(12.8) a | 15.7 ^(13.4) a | 8.9 ^(16.4) a | 3.38 ^(17.3) a | 43.2 ^(16.2) b |
| | Ves. Diam. (µm) | Freq. (pores/mm ²) | Ray height (µm) | Ray width (µm) | |
| A | 90.1 ^(13.6) a | 10.1 ^(11.5) b | 239.3 ^(18.5) a | 6.84 ^(17.9) b | |
| B | 109.6 ^(14.5) b | 11.1 ^(12.5) b | 306.4 ^(18.1) c | 7.64 ^(16.8) c | |
| C | 100.1 ^(12.1) a | 10.4 ^(10.8) b | 217.8 ^(18.3) a | 8.48 ^(13.5) d | |
| D | 113.2 ^(13.5) b | 8.8 ^(12.1) a | 226.3 ^(17.2) a | 6.28 ^(15.4) a | |
| E | 112.5 ^(14.3) b | 10.7 ^(12.8) b | 278.2 ^(18.2) b | 6.13 ^(16.2) a | |
| F | 106.6 ^(13.2) b | 13.9 ^(11.8) c | 207.3 ^(17.8) a | 5.80 ^(17.5) a | |
| CL. | Microfibril angle (°) | MOE of S2 layer (GPa) | Hardness of S2 layer (GPa) | MOE of m.l. (GPa) | Hard. of m.l. (GPa) |
| A | 9.2 ^(9.8) a | 15.5 ^(6.2) d | 0.289 ^(7.02) b | 9.97 ^(7.23) c | 0.335 ^(6.97) c |
| B | 10.9 ^(6.6) d | 11.6 ^(7.0) a | 0.266 ^(7.34) a | 7.13 ^(7.23) a | 0.299 ^(6.89) b |
| C | 10.4 ^(8.2) c | 12.6 ^(7.1) b | 0.269 ^(7.32) a | 7.95 ^(6.96) b | 0.298 ^(7.07) b |
| D | 9.16 ^(7.2) a | 14.1 ^(7.6) c | 0.278 ^(7.19) b | 8.47 ^(7.55) b | 0.302 ^(7.34) b |
| E | 9.8 ^(7.3) b | 13.2 ^(7.2) b | 0.262 ^(7.12) a | 8.67 ^(7.18) b | 0.309 ^(7.04) b |
| F | 10.5 ^(6.2) c | 13.6 ^(6.4) b | 0.278 ^(6.63) b | 7.15 ^(6.81) a | 0.274 ^(7.21) a |

CL.= *Eucalyptus grandis* × *Eucalyptus urophylla* clone; F.L.=Fiber length; F.W.= fiber width; L.D.=Lumen diameter; C.W.T.= Cell wall thickness; C.W.F.=Cell Wall Fraction; Ves. Diam.= Vessel diameter; Freq.=Vessel Frequency; Moe of m.l.= Moe of middle lamella; Hard. of m.l.= Hardness of middle lamella. Means followed by the same letter vertically per parameter does not differ by the Scott-Knott test at 5% probability. Values in superscript represent the coefficient of variation.

3.2. Characterization of the wood physical properties

The physical behavior of wood differed between the *Eucalyptus* clones, but with less variability for equilibrium moisture and basic density (Table 2).

The wood X-ray densitometry showed that even clones with similar density, may have different density patterns along the radial direction (Figure 1).

3.3. Wood mechanical characterization

The wood mechanical properties varied between clones, with the highest values for the modulus of elasticity and the least for the compression parallel to the grain (Table 3).

4. DISCUSSION

4.1. Wood anatomical and ultrastructural characterization

Clone A had a greater cell wall thickness and a smaller lumen diameter, with a reverse tendency for Clone C. Thus, Clone A showed the largest cell wall fraction, while Clone C the lowest (Table 1). The fibers are the main components of hardwood (Panshin and

De Zeew, 1980), and therefore, a high cell wall fraction ensures better mechanical properties to the timber, as reported *Eucalyptus propinqua* wood (Longui et al., 2014). Oliveira et al. (2012) found values between 11.05 and 12.09µm for the lumen diameter, and between 2.94 and 3.88 for the wall thickness in *Eucalyptus grandis* W. Hill ex-Maiden.

The frequency and the average diameter of the pores, which vary between clones (Table 1), are hollow structures that increase the wood permeability (Panshin and De Zeew, 1980). Thus, a higher frequency and diameter of these structures result in a good response to the drying (Shahverdi et al., 2012) and preservative treatments (Taghiyari, 2012). The ray cells are fragile because of their thin cell walls (Gricar and Eler, 2015), thus, materials with a higher height and width of the rays may have inferior mechanical properties, limiting their use for structural purposes.

Clones A and D had the lowest microfibril angle values (Table 1). The arrangement of the cellulose chains in the cell wall was fundamental to its strength (Donaldson, 2008; Hein et al., 2013). Combining the fact that Clone A showed larger cell wall fraction, the

Table 2 – Wood basic density, equilibrium moisture content, volumetric swelling, radial swelling, tangential swelling and anisotropy in six two-year-old *Eucalyptus grandis* × *Eucalyptus urophylla* clones

Tabela 2 – A densidade básica, umidade de equilíbrio higroscópico, inchamento volumétrico, inchamento radial, inchamento tangencial e coeficiente de anisotropia da madeira de clones de *Eucalyptus grandis* × *Eucalyptus urophylla* com dois anos.

| Clone | Basic density) (g/cm ³) | Equi. mois. cont. (%) | Vol. swell. (%) |
|-------|---|--------------------------|--------------------------|
| A | 0.421 ^(4.2) d | 11.23 ^(2.1) a | 18.24 ^(7.3) b |
| B | 0.370 ^(3.6) a | 10.58 ^(2.5) b | 16.43 ^(7.2) a |
| C | 0.372 ^(4.0) a | 10.55 ^(1.9) b | 16.62 ^(6.3) a |
| D | 0.423 ^(3.7) d | 11.32 ^(1.6) a | 20.99 ^(7.3) c |
| E | 0.387 ^(3.2) b | 10.41 ^(1.8) b | 16.53 ^(6.5) a |
| F | 0.412 ^(3.1) c | 10.56 ^(2.1) b | 16.91 ^(6.7) a |
| | Rad. swell. (%) | Tang. swell. (%) | Anisotropy |
| A | 5.89 ^(7.7) b | 10.23 ^(7.9) c | 1.76 ^(7.5) b |
| B | 5.24 ^(7.4) a | 8.86 ^(8.3) a | 1.66 ^(7.6) a |
| C | 5.68 ^(6.9) b | 9.78 ^(7.8) b | 1.77 ^(7.2) b |
| D | 6.21 ^(7.6) a | 11.54 ^(8.3) d | 1.81 ^(7.4) c |
| E | 5.12 ^(6.1) a | 9.21 ^(8.9) a | 1.74 ^(7.4) b |
| F | 6.28 ^(6.2) c | 9.67 ^(8.9) b | 1.62 ^(7.9) a |

Equi. mois. cont.=Equilibrium moisture content; Vol. swell.=Volumetric swelling; Rad. swell.=Radial swelling; Tang. swell. = Tangential swelling. Means followed by the same letter vertically per parameter does not differ by the Scott-Knott test at 5% probability. Values in superscript represent the coefficient of variation.

fibers of this material showed higher hardness and modulus of elasticity. The opposite happened for clone B, which, due to its high microfibril angle and low cell wall fraction, showed the lowest values for the mechanical properties of fibers.

4.2. Characterization of the wood physical properties

The wood basic density, equilibrium moisture content, volumetric, radial, and tangential swelling were higher for Clone D and lower for Clone B (Table 2). Materials with a higher wood basic density have a higher mass per unit volume, resulting in a higher

adsorption of humidity and increasing the and linear and volumetric swelling (Schulgasser and Witztum, 2015; Rouco and Muñoz, 2015).

All materials had a lower radial swelling than tangential swelling (Table 2). The reason for this is still debated, but this may be because of the orientation of the ray cells, which provided their microfibrils in the radial direction, offering greater resistance to compression (Kollmann and Côté, 1968; Glass and Zelinka, 2010). This was also reported for *Corymbia citriodora*,

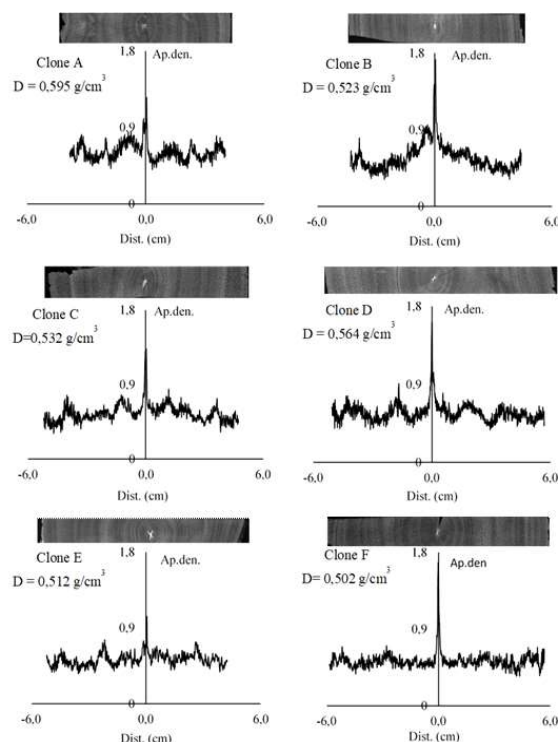


Figure 1– Wood X-ray densitometry of the most representative samples per clone. Dist = distance from pith (cm.); Ap. den. = Apparent density (g/cm³); D = average apparent density of the sample.

Figura 1– Densitometria de raio-X das amostras de madeira mais representativas por clone. Dist. = Distância da medula (cm); Ap den. = Densidade aparente (g/cm³); D= densidade aparente média da amostra.

Table 3 – Modulus of rupture (MOR) and elasticity (MOE) and compression parallel to the grain in six two-year-old *Eucalyptus grandis* × *Eucalyptus urophylla* clones

Tabela 3 – Módulo de ruptura (MOR) e elasticidade (MOE), resistência a compressão paralela às fibras em seis clones de *Eucalyptus grandis* × *Eucalyptus urophylla* com dois anos

| Clones | MOR (MPa) | MOE (MPa) | Comp. par. (MPa) |
|--------|--------------------------|--------------------------|--------------------------|
| A | 74.6 ^(11.2) d | 7038 ^(12.4) d | 40.8 ^(9.5) c |
| B | 51.8 ^(9.4) a | 4527 ^(11.2) a | 33.9 ^(10.5) a |
| C | 50.7 ^(11.3) a | 5423 ^(10.2) b | 32.9 ^(10.2) a |
| D | 62.8 ^(10.5) c | 5643 ^(11.3) b | 34.1 ^(10.1) a |
| E | 57.6 ^(10.1) b | 5247 ^(10.5) b | 32.7 ^(9.5) a |
| F | 70.2 ^(10.4) d | 6236 ^(9.3) c | 36.1 ^(11.2) b |

Comp. par.= Compression parallel to the grain; Means followed by the same letter vertically per parameter does not differ by the Scott-Knott test at 5% probability. Values in superscript represent the coefficient of variation.

Eucalyptus grandis, *Eucalyptus saligna* and *Pinus elliottii* (Pelozzi et al., 2012; Menezes et al., 2014).

The anisotropy did not show any relationship with the wood basic density, even being the ratio of the tangential and radial swelling, showing Pearson correlation coefficient of 0,2235 between these two variables. The highest value for the anisotropy in clone D indicates that its wood has restricted use in places with high humidity variation. However, treatments such as acetylation (Xie et al., 2013; Himmel et al., 2015) and heat treatment (Korkut, 2012; Zanuncio et al., 2014b) can reduce the variation in the wood dimensions and allow its use in such places.

The wood apparent density of the samples was higher in the pith region corresponding to the parenchymal cell deposits such as crystals and starch granules, which interacted strongly with the X-rays, resulting in high apparent density (Figure 1) (Panshin and De Zeew, 1980; Belini et al., 2011). The region corresponding to these parenchyma cells was not accounted for the average apparent density, because it was not considered as wood.

Sample F showed less variation in the wood apparent density from pith to bark, while Clone A showed the highest variation (Figure 1). The wood density has relation with drying (Zanuncio et al., 2013b; Zanuncio et al., 2015) and wood machinability (Moura et al., 2011). Thus, materials with a homogeneous wood density along the radial direction have a more uniform behavior, facilitating its use.

4.3. Wood mechanical characterization

The wood of Clone A had a higher mechanical strength (Table 3), suggesting it is more suitable for structural use, like beams and trusses, or furniture manufacture subjected to mechanical stress, such as bookcases and chairs (Lopes et al., 2011). Müller et al. (2014) found values of 83.53 and 9754.67 MPa for modulus of rupture and modulus of elasticity in *Eucalyptus benthamii* wood, values higher than those found in this study .

Materials with greater cell wall fraction showed higher basic density, reducing the dimensional stability, and with the lower microfibril angle, improved the mechanical properties of the fibers and consequently the wood as a whole. This demonstrated how the anatomical and ultrastructural characteristics affected the physical and mechanical characteristics of the wood, and consequently, its use.

5. CONCLUSION

The evaluated wood parameters varied among the *Eucalyptus* clones. The materials with greater cell wall fraction resulted in higher dimensional instability and those with the lowest microfibril angle indicated better mechanical properties. The wood of Clone A showed a low microfibril angle, high cell wall fraction, and better mechanical properties and was suitable for structural use. Clones B and F showed a wood with low anisotropy and were suitable for use in locations with high humidity variations. The heterogeneity of the material revealed the importance of a comprehensive study of each clone, to define the best use of its wood.

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