THE WIDE VARIATION OF AMAZONIAN STOCKED HARDWOODS AFFECTING NATURAL RESISTANCE TO ARBOREAL TERMITES OVER TIME

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ABSTRACT

After forest suppression in mining areas, deterioration of stocked tropical hardwoods occurs at different rates and levels. Prioritizing logs to avoid wastage is challenging because the wide interspecific variability of wood traits makes natural resistance unpredictable. This work aimed to compare the biodeterioration of four Amazonian hardwoods from a mining stockyard to arboreal termite attacks over six weeks under laboratory conditions. The woods' chemical composition, anatomy, basic density, and crystallinity were determined. Mass loss and visual diagnosis of the specimens supported the biodegradability analyses. The species showed a wide range of mass loss after six weeks: *Jacaranda copaia* (pará-pará) - 15,2 %; *Pseudopiptadenia suaveolens* (timborana) - 0,8 %; *Aegiphila integrifolia* (tamanqueira) - 5,2 %; and *Euxylophora paraensis* (pau amarelo) - 0,5 %. Biodegradation did not stabilize over time for the lowest-density species. The crystallinity indicated the initial degradation of amorphous wood components, followed by a non-selective feeding mechanism. Gathering all species, increased extractive and lignin levels, fiber wall thickness, and basic density favored

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the natural wood resistance, oppositely to large vessel elements. The much lower basic density of *J. copaia* explains its lowest natural resistance. Wall thickness, pore diameter, and density overcame chemical composition on wood biodegradability. Low-density hardwoods stocked in mining yards are a priority for proper destinations.

Keywords: Amazonian species, arboreal termites, basic density, biodeterioration, crystallinity, mass loss, organic extractives, tropical hardwood, xylophagous, natural resistance.

INTRODUCTION

Brazil is the world's fourth-largest producer of bauxite after China (3rd), Guinea (2nd), and Australia (1st). In 2020, national production earned R$ 4,5 billion (Brazilian currency - reals) and reached 32897,8 million tons (IBRAM 2021, ABAL 2023a, ABAL 2023b). Despite financial benefits, environmental impacts from mining are significant drawbacks, and the ecosystem landscapes are entirely modified (Jesus *et al.* 2016). Complete deforestation is the first mining activity, followed by removing the organic soil and sterile soil layers to reach the mineral as allowed by the National Normative Instruction n° 07 (SEMAS 2015). Consequently, the company's stock many harvested logs in the explored areas. Pará, a federal state in the Amazonia region, comprises over 91% of the country's production (ABAL 2017); hence, it currently faces the challenge of finding feasible and legal destinations for the many harvested logs.

The state government allows to solve the problem through the Ordinary Law n° 6958 (SEMAS 2007), regulating wood harvested from areas licensed to explore deposits and mines within Pará. It only allows three wood destinations, all through donations: construction of popular housing, schools, or chemical-dependence treatment institutes. Nevertheless, the donation beneficiaries are fully responsible for processing the wood. Such demand discourages potential receptors that do not possess wood sawing facilities or qualified labor. Therefore, the mining companies are still looking for solutions because the stocked logs hinder the recovery of degraded areas and naturally degrade, releasing CO_2 into the environment. While the legal solutions are debated, timeless stocking raises the damage extension decay causes to valuable tropical hardwoods.

In the stockyard, the logs undergo both abiotic and biotic deterioration (Thaler *et al.* 2014). At least partly, the rain leaches extractives from the wood, likely changing relevant biodegradability properties (Brischke *et al.* 2014, Thaler *et al.* 2014). Regarding biological activities, certain agents can naturally decay wood if the environment is favorable (Karim *et al.* 2016). Fungi (Karim *et al.* 2016), bacteria (Klaassen 2014), marine borers (Palanti *et al.* 2015), and insects (Gascón-Garrido *et al.* 2013) attack wood and consume its cell wall components. Nevertheless, many kinds of wood have their own natural defense mechanisms against xylophagous agents developed while the trees evolved (Arango *et al.* 2006). Natural resistance is essential for selecting woods for specific uses (Jacobs *et al.* 2019). It guarantees a long-term use of wood products while causing minimum environmental impacts by avoiding harmful pesticides (Arango *et al.* 2006, Thaler *et al.* 2014). In the stockyards, knowledge of natural resistance may support decision-making on which logs need urgent removal and destination and selecting the most resistant ones for donations.

Tropical forests host the highest diversity of termites among biomes because of their favorable habitat, altitude, and climate (Eggleton 2000). Arboreal-nesting termites exhibit a wide adaptation to environmental conditions (Novita *et al.* 2020) and may become aggressive wood pests (Diniz *et al.* 2020). Their ability to masticate, digest, and assimilate wood determines its consumption (Vasconcellos and Moura 2010). The species from *Nasutitermes* widely occur in Brazil, attacking wood in the natural forest, crops, and urban environments (Vasconcellos and Moura 2010, Paes *et al.* 2016). It consumes wood of various qualities, regardless of the drying stage (wet or dried in-service) or being partially deteriorated (Scheffrahn *et al.* 2005). Therefore, in direct contact with the ground of degraded areas and surrounded by primary forests, stocked mining logs are especially attractive to *Nasutitermes* arboreal termites.

The wide variability of wood's chemical, anatomical, and physical attributes determines the variations of natural resistance to xylophages among species or samples within the tree, e.g., heartwood and sapwood (França *et al.* 2016). Moreover, wood quality affects each xylophagous agent consumption differently. For instance, almécega (*Protium heptaphyllum* (Aubl.) Marchand) wood was more resistant to termite *Nasutitermes corniger* than *Cecropia* sp. wood. However, *Nasutitermes macrocephalus* termite degraded more almécega (*Protium heptaphyllum* (Aubl.) Marchand) wood than *Cecropia* sp. wood (Vasconcellos and Moura 2010).

Wood natural resistance is generally measured by percentage mass loss when submitted to a xylophagous agent attack, such as fungi, termites, or bores (Benítez *et al.* 2021, Baufleur *et al.* 2022). Thus, density is intrinsically an essential trait to determine the mass loss rate of different woods, i.e., a higher-density wood would lose less percentage mass than a lower-density wood if both had the same mass consumption by a xylophagous agent. Consequently, literature often reports the general trend of denser woods with higher natural durability (Arango *et al.* 2006, França *et al.* 2016, Owoyemi *et al.* 2017, Deklerck *et al.* 2020). Nevertheless, high-density species with low resistance (Romano and Acda 2017), low-density species with high resistance (Ribeiro *et al.* 2014, Palanti *et al.* 2015), or non-correlation between density and resistance at all (Thaler *et al.* 2014) are also quite frequent.

When the relationship between density and wood mass loss through decaying is unclear, microstructural tissue proportion and wood chemical composition are the key traits to explain natural wood resistance (Dadzie 2019). The role of the type and proportion of organic extractives in wood durability is widely investigated. For instance, chestnut wood is rich in tannin, known for its anti-xylophagous properties. However, leaching extractives from wood with methanol or water significantly removed 11 % and 15 % of its total mass, but durability only slightly decreased (Thaler *et al.* 2014). Oppositely, nearly all eight wood species attacked by termites or fungi exhibited higher mass loss when extractives were removed. Indeed, extracted samples had mass losses comparable to the non-durable controls (Kirker *et al.* 2013). The activity of extractives from three Brazilian hardwoods timber known for their high resistance to biodegradation, sucupira (*Bowdichia virgili-oides* Kunth), *branco* (*Anadenanthera colubrina* (Vell.) Brenan), and jatoba (*Hymenaea stigonocarpa* (Mart. ex Hayne), evaluated in the laboratory tested positive against termites *Nasutitermes corniger* (Santana *et al.* 2010).

The Amazon hardwoods will probably show a complex combination of traits that will confer more or less natural resistance to arboreal termite attack. Laboratory assays under controlled environments may reveal which logs are more susceptible to rapidly degrading and disappearing, releasing CO_2 into the environment instead of being properly used. Moreover, the knowledge of the natural resistance of Amazon hardwoods is limited compared to Amazonia's extensive biodiversity (Reis *et al.* 2017).

Therefore, this work aimed to compare the biodegradation of four Amazonian hardwoods from mining stockyard to arboreal termite attacks over six weeks under laboratory conditions and to identify the most critical traits that favor natural resistance. The assumption that lower-density woods will deteriorate faster and more intensely than high-density woods is challenged when Amazonian woods are investigated in depth. In addition, evaluating biodegradation over time increases knowledge about the resistance of species, indicating the best way to proceed with logs stored in mining yards in the Amazon.

Materials and methods

Sampling, collecting, and termite bioassay of the hardwoods from a bauxite mining stockyard

The samples were collected in the log yard of Mineração Paragominas SA, a Hydro group company located near Paragominas, northeast of the Pará state. The trees were harvested from a primary rainforest in 2017 from the Miltônia III plateau located at 3º 30' and 3º 45' south latitude and 48º 30' and 48º 45' longitude west of Greenwich to allow bauxite mining. They remained in the stockyard from 2017 to 2018. The company does not measure the trees because they cannot be marketed, but the disks sampled had diameters ranging from ≈ 60 - 75 cm.

The collected species were timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes), pau amarelo (*Euxylophora paraensis* Huber), tamanqueira (*Aegiphila integrifolia* (Jacq.) Moldenke) and pará-pará (*Jacaranda copaia* (Aubl.) D. Don), identified according to the index number (NIDX) 04/2019 by the Embrapa Amazônia Oriental xylotec. The selection was primarily based on wood density. Two species with sensitively high densities and two with low densities were chosen. The second criterion was color. Within a density class, woods of distinctive color were selected, considering the possibility of being related to extractives of different natures at different levels (Lima *et al.* 2021).

Three logs (L_I, L_{II}, and L_{III}) of each species from each log pile (P_I, P_{II}, and P_{III}) were selected. A 7 cm-thick disc was extracted from the extremity of each log. From the disc, an 8 cm-width central piece containing the heartwood and sapwood intact was obtained and later subdivided into fractions of 2 cm by 1 cm in cross-sectional view, measured from the medulla. The length of the specimens (axial direction) was adjusted to 5 cm. One specimen per sampled log was randomly selected for the anatomical analysis, totaling three samples per species. Basic density and moisture content were measured in five specimens of each log, totaling fifteen

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samples per species. The biodegradation test was carried out with ten samples per log, totaling thirty samples per species. The same samples that underwent the bioassay were removed and processed at the end of each analysis, forming composed samples by species for X-ray analysis. All specimens were removed from the discs randomly. The two slabs of the three disks of each species were used for chemical analyses (Figure 1).

The termite attack bioassay followed the ASTM D3345-22 (2022) standard. Flasks of 500 mL were filled with 200 g of sand, containing moisture of 80 % of the holding capacity by adding 30 mL of distilled water. A wood specimen and $1 \pm 0,05$ g of arboreal termites of the genus *Nasutitermes* sp., collected in a natural environment in Belém, Pará, Brazil, were added to each flask. The flasks were covered with fabric elastic sealed, allowing air circulation while preventing escape and letting termites breathe. Five replicates were set up for each wood species and weekly evaluation time.

The above-mentioned standard establishes 28 days under room conditions for the biodegradation assay, followed by the analysis of the wood mass loss in the end. In this investigation, replicates were added to allow dissembling and mass loss determination every 7 days to provide the decay behavior over time. Moreover, the assay was extended to 42 days to increase the possibility of finding significant wood decaying for species with higher wood density. Therefore, every week (7 days), five samples of each species were taken to measure mass loss and visual degradation analysis. At the end of the experiment, the specimens were removed, and the termites were separated.

The local climate is classified as humid tropical in the Am climate zone (humid with low duration dry season) according to Kôppen-Geiger's classification, with an average annual temperature of 27 °C and an average relative humidity above 80 %.

Wood characterization descriptions

Anatomical analysis

The wood maceration was performed to measure the anatomical traits according to the method of Franklin (Franklin 1945). A Motic optical microscope (BA310Elite) was used to take photos of the temporary slides. A 10x magnifying lens was coupled to measure the length and diameter of the vessel elements. The fiber lengths were obtained with a 4x lens, and the lumen and wall thicknesses with a 40x lens. Fifty measurements were performed for each anatomical trait as established by the International Association of Wood Anatomists (IAWA 1989). All variables were measured using the Motic Images Plus 3.0 program Motic software (Motic China Group Co. Ltd 2022).

Chemical analysis

The content of total extractives was determined according to ABNT NBR 14853:2010 (2010). Hot and cold-water-soluble extractives were determined according to ABNT NBR 14577:2017 (2017). Ashes were determined in a muffle furnace Lab1000® at 525 °C, according to ABNT NBR 13999:2017 (2017). The insoluble lignin content was quantified according to ABNT NBR 7989:2010 (2010). The percentage of holocellulose (cellulose + hemicelluloses) was determined, as described by Browning (1963). The cellulose content was calculated using the method of Kennedy *et al.* (1987). The content of hemicelluloses was derived from the difference between the values of holocellulose and cellulose. All analyses were carried out in triplicate.

Basic density analysis

The basic density of the specimen was determined as the ratio between the anhydrous mass (moisture content = 0 %) and its saturated (green) volume according to the procedure described in ABNT NBR 11941:2003 (2003).

Bioassay: Natural resistance to *Nasutitermes* sp. termites

The two biodegradation variables for each specimen were percentage mass loss degradation and level score by visual examination according to ASTM D3345-22 (2022) which classifies and scores wear as follows: healthy, with superficial scarifications (10); superficial attack (9); moderate attack, with penetration (7); intense attack (4); failure with rupture of the specimen (0).

For percentage mass loss determination, the specimens were weighed before and after the termite bioassay for the percentage mass loss. Previously, the dry base moisture of the woods was deter-

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mined in five specimens per species (see the flowchart). This value was subsequently used to estimate an equivalent initial mass without the moisture of the specimens selected for the termite attack. After the attack, they were dried at 100 ± 5 °C for approximately 24 h up to a 0 % moisture content and weighed. All masses were obtained with a 0,0001 g precision. Percentage mass loss was calculated by Equation 1.

$$ML = \left(\frac{IM_{eq} - M_f}{IM_{eq}} \right) x\ 100 \qquad (1)$$

Where: *ML* is the mass loss of the wood specimens after biodegradation (%); IM_{eq} is equivalent to 0 % moisture content initial mass of the wood specimens (g); M_f is the final mass of the wood specimens at 0 % moisture content (g) after 7, 14, 21, 28, 35, and 42 days.

X-ray diffraction (XRD) analysis

The crystalline structure of the woods after termite degradation was analyzed using a diffractometer (Model D2 Phaser, Bruker®, Germany) with Cu-Kα radiation ($Λ = 1,540$ Å) and nickel filter, operating at 30 kV and 10 mA. The samples were evaluated with θ ranging from 5° to 55°.

The samples' crystalline index (CI) was obtained using Magic Plot Student after filtering raw data to determine the crystalline and amorphous fractions by area calculations. The CI was calculated using Equation 2, proposed by Segal *et al.* (1959).

$$CI = \left(\frac{1 - I_{am}}{I_{200}} \right) x\ 100 \qquad (2)$$

Where: *CI* is the crystalline index (%); I_{200} is the maximum intensity of the main diffraction peak (2θ = 22,7° for cellulose I and 2θ = 21,7° for cellulose II); and I_{am} is the intensity of diffraction corresponding to only the amorphous material (2θ = 18° for cellulose I, and 2θ = 16° for cellulose II).
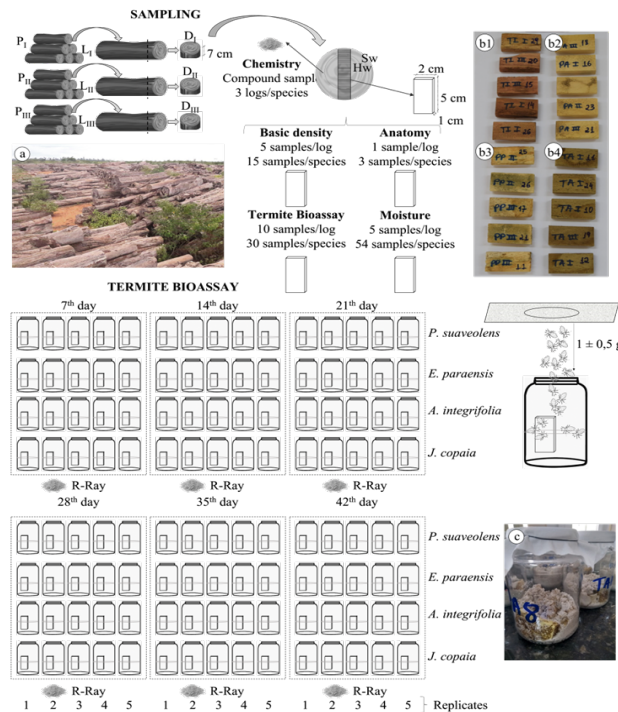

Figure 1: Collecting, sampling, and analyses. Piles in the stockyard (P_I, P_{II}, and P_{III}); logs from a given species (L_I, L_{II}, and L_{III}); Wood disks (D_I, D_{II}, and D_{III}); the heartwood (cr) and the sapwood (sp) of the sample disk; R-ray diffraction analysis (R-XD); (a) logs in the mining stockyard; (b1) *P. suaveolens*; (b2) *E. paraensis*; (b3) *J. copaia*; (b4) *A. integrifolia*; (c) termite attack set.

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Statistical analysis

The parameters were logarithmized and analyzed by generalized linear models (GLMs), followed by analysis of variance to verify statistical differences among species. The averages were compared through the post-hoc test, and the test predictions function from the ggeffects package in R was used.

Results and discussion

Pau amarelo *(Euxylophora paraensis* Huber) and pará-pará (*Jacaranda copaia* (Aubl.) D.Don) showed similar and higher fiber lengths than timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W.Grimes) and tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks), which also differed. Pau amarelo *(Euxylophora paraensis* Huber) showed the thickest fiber walls, followed by timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W.Grimes) and pará-pará (*Jacaranda copaia* (Aubl.) D.Don) with similar values. Meanwhile, tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) fibers showed the thinnest fiber walls. Fiber lumen diameter differed among the four species. Pau amarelo *(Euxylophora paraensis* Huber) had the longest vase elements, followed by pará-pará (*Jacaranda copaia* (Aubl.) D.Don)*,* with a higher value than timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W.Grimes) and tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) pará-pará (*Jacaranda copaia* (Aubl.) D.Don) depicted a bigger vase element diameter than pau amarelo *(Euxylophora paraensis* Huber)*;* both diameters were higher than timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W.Grimes) and tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) diameters (Table 1).

Table 1: Biometry of the cellular structures of the hardwoods.

Species	Fiber (µm)			Vase element/Pore (µm)	
	Length	Wall thickness	Lumen diameter	Length	Diameter
E. paraensis	$1159,9^{(195,9)}$a	$5,8^{(1,4)}$a	$6,2^{(2,1)}$a	$768,7^{(416,1)}$a	$170,3^{(90,2)}$b
J. copaia	$1132,5^{(166,2)}$a	$4,0^{(0,8)}$b	$21,6^{(4,8)}$d	$487,9^{(117,3)}$b	$255,7^{(74,5)}$a
P. suaveolens	$1018,0^{(152,1)}$c	$4,2^{(1,0)}$b	$8,9^{(3,9)}$c	$327,5^{(89,1)}$c	$148,2^{(33,6)}$b
A. integrifolia	$1066,7^{(185,7)}$b	$3,5^{(1,0)}$c	$14,3^{(4,6)}$b	$347,4^{(84,6)}$c	$135,7^{(38,0)}$c
X^2	0,53	22,08	144,0	56,529	34,53

Average [(Standard Deviation)]; X^2 = species deviance has a highly significant effect if p-value < 0,001. Averages followed by the same letter in the column did not differ by the post-hoc test at 5% significance.

The parenchyma, fibers, and vase elements comprise the xylem of hardwoods; hence, their variable proportions and biometry primarily influence wood's overall properties (Zhao *et al.* 2019). Fibers are elongated cell structures with a high length/diameter ratio containing a primary wall and a thicker secondary wall with variable lignification degrees, and that usually lose their protoplasm, forming a lumen. They confer mechanical support and strength to the wood (Evert 2013). Even though this study included only four Amazon hardwoods, they revealed a wide variability of dimensions of the anatomical structures. Fiber biometry greatly influences basic density since it is a primary determinant of the void/mass ratio in wood (Nazari *et al.* 2020). Pau amarelo *(Euxylophora paraensis* Huber) fiber' biometry with the thickest wall and smallest lumen diameter favors increased density the most. pará-pará (*Jacaranda copaia* (Aubl.) D. Don) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) wall thicknesses would confer similar basic densities, but the former had a far bigger lumen diameter meaning more void proportion within a wood volume. tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) combined the thinnest wall with the second-biggest lumen diameter, favoring decreased wood density. Other work revealed positive correlations between density and fiber wall thickness, unlike fiber length (Carrillo *et al.* 2015).

Vessel elements (or pores in wood's transversal face) are also abundant and highly variable (Zhao *et al.* 2019). As tube-like structures that aim to transport water (Evert 2013), their diameter, length, and frequency will also alter the void/mass ratio in the wood. timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) and tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) showed vase elements with close and

shorter lengths and smaller diameters, which favor wood density increase. Both traits differed for pará-pará (*Jacaranda copaia* (Aubl.) D. Don) and pau amarelo *(Euxylophora paraensis* Huber) woods, with the latter showing longer vase elements but with smaller diameters.

Pau amarelo (*Euxylophora paraensis* Huber) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) showed the highest and statistically equal basic densities. This trait was statistically higher in tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) than in pará-pará (*Jacaranda copaia* (Aubl.) D. Don). The marked differences among fibers and vase elements' biometry help elucidate basic density differences. The thickest fiber wall of pau amarelo (*Euxylophora paraensis* Huber) stood out, as much as the widest fiber lume of pará-pará (*Jacaranda copaia* (Aubl.) D. Don) (Figure 2).
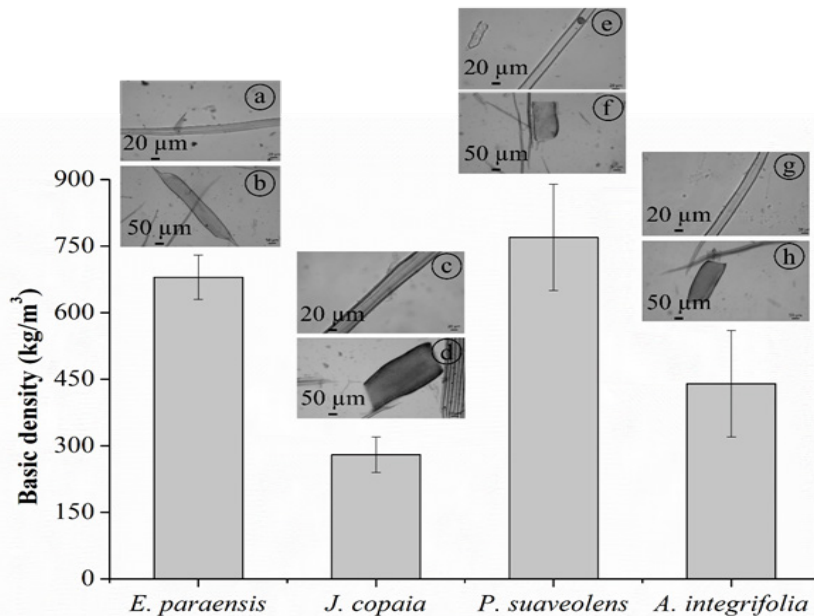

Figure 2: Contrast between cell biometry and the basic density of the hardwoods: (a) macerates of *E. paraensis* - fiber and (b) vessel element; (c) *J. copaia* - fiber and (d) vessel element; (e) *P. suaveolens* - fiber and (f) vessel element; and (g) *A. integrifolia* - fiber and (h) vessel element. X^2 (species deviance) = 1,927 (p-value < 0,001 has a highly significant effect). Averages followed by the same letter did not differ by the post-hoc test at 5 % significance.

In hardwoods, the anatomical structure, including cell wall thickness, fiber width, vessel width and frequency, parenchyma proportion, and chemical composition, determine wood's basic density. Thus, species with similar density can differ markedly in fiber properties and cell dimensions (Carrillo *et al.* 2015). Pará-pará (*Jacaranda copaia* (Aubl.) D. Don) outstanding low basic density was conferred by a very large fiber lumen diameter overcoming an average fiber wall thickness, besides the big diameter of the pores. The density similarity of pau amarelo (*Euxylophora paraensis* Huber) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) is explained by the first's fiber biometry balanced by the former's vase element biometry, both favoring increased wood density. The fiber lumen diameter of tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D. Jacks) primarily explains its intermediate density besides the thinnest fiber wall.

Based on average densities, Csanády *et al.* (2015) classification ranks pará-pará (*Jacaranda copaia* (Aubl.) D. Don)*,* tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks)*,* pau amarelo (*Euxylophora paraensis* Huber), and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) as very-low, low, medium, and high-density woods, respectively. The wood constitution corresponds mainly to the cell wall, which is the target of decay and termite feeding. Regarding any wood biodegradation, higher densities lead to a lower percentage of mass loss for a given degraded mass in grams. According to Stallbaun *et al.* (2017), high-density woods tend to decrease the fragmentation ability of termites, thus inhibiting their consumption. Therefore, cell biometry and bulk density are closely related to mass loss rate.

The chemical composition of the wood is subdivided into non-structural and structural components. Organic total extractives belong to the first class. Their content widely ranged from 5,1 and 12,1 % and statistically varied among the four hardwoods. timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes)

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stood out with the highest extractive content. Inorganics are also non-structural components. Although the levels were higher for two species, very low amounts were found, reaching 0,07 % in tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks). The tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) showed no statistical difference for this trait, which was below the contents found for pau amarelo (*Euxylophora paraensis* Huber) and pará-pará (*Jacaranda copaia* (Aubl.) D. Don) (Figure 3a).

There was a statistical difference in hot water-soluble extractives among the four species. However, timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) had the highest content of cold water-soluble extractives, but the values were similar among the remaining species. Besides the highest total extractive content (polar and non-polar), timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) stood out with the highest hot-water and cold-water-soluble extractive content, but they corresponded to a portion of total extractives ($\approx$8 % of 12 %). Pau amarelo (*Euxylophora paraensis* Huber) had the second-highest total extractive content, but the lowest contents of both cold and hot water-soluble extractives, which were similar. tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) was ranked third for total extractive content and showed very similar hot-water soluble extractive content, but lower cold-water extractive content. Cold and hot water removed extractive contents were close to each other and similar to the content of total extractives for pará-pará (*Jacaranda copaia* (Aubl.) D. Don) (Figure 3b).
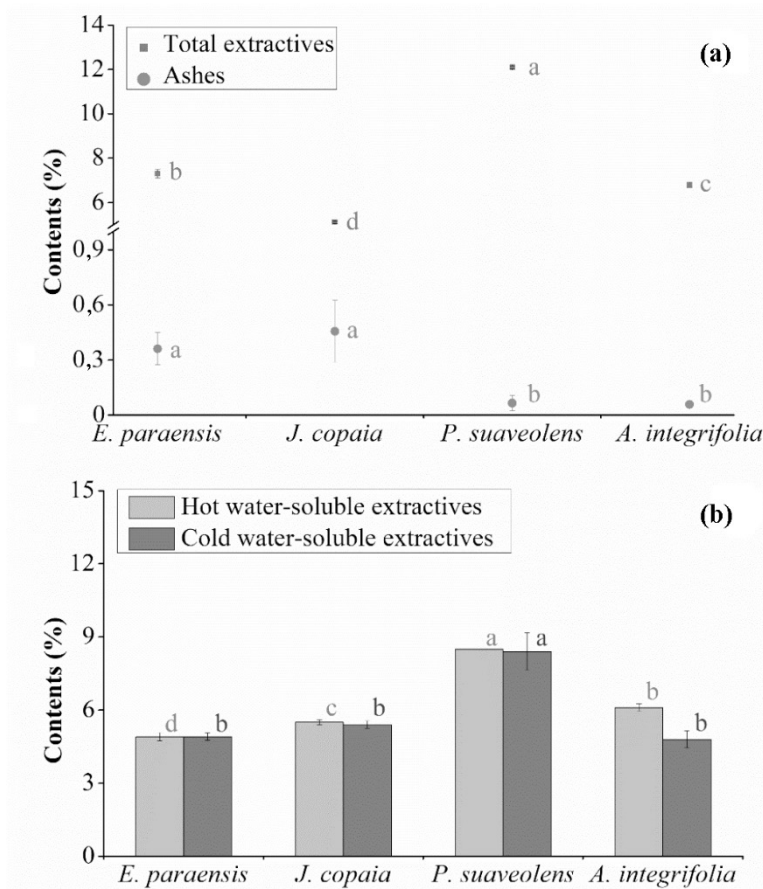

Figure 3: Secondary chemical composition (based on total wood mass): total extractives and ashes (a); water-soluble extractives (b). X^2 (species deviance has a highly significant effect if p-value < 0,001) = 1,183 for total extractives, 1,367 for ashes, 0,528 for hot water-soluble extractives, and 0,605 for cold water-soluble extractives. Averages followed by the same letter for each secondary component did not differ by the post-hoc test at 5 % significance.

Organic extractives widely vary among wood species (Lima *et al.* 2021). They do not structure the cell wall but occur within the cell lumen, on the cell wall surface, or in the parenchyma cells (when in the sapwood in the last case). Such non-structural components are removable by organic solvents and water. The cold-wa-

ter procedure removes inorganic compounds, tannins, gums, sugars, and coloring matter, while the hot-water procedure additionally removes starches (Tofanica *et al.* 2011). Comparing the levels of water-soluble and total extractives indicates how much of the total extractives are polar and non-polar. Based on hot-water soluble extractives, such ratios were 7/5 % for pau amarelo (*Euxylophora paraensis* Huber), 5/5 % for pará-pará (*Jacaranda copaia* (Aubl.) D. Don), 12/8 % for timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes), and 7/6 % for tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks). Therefore, the majority or almost all extractives of the woods are polar. timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) showed more difference between the extractives of different natures. tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) showed the most meaningful difference between hot and cold-water extractives, suggesting starch presence, making it more susceptible to xylophagous (Melo *et al.* 2015).

Extractives may confer the wood's natural resistance depending on their nature and amount. They are classified into aliphatic and alicyclic compounds (i.e., terpenoids and terpenes) and phenolic compounds, such as flavonoids and tannins (Broda 2020). Gums, tyloses, and other phenolic compounds can act as slow termite poisons (Nicholson and Hammerschmid 1992, Dahali *et al.* 2023). Wood-extracted tannin showed an antifeedant effect over the subterranean termite *Reticulitermes speratus* (Ismayati *et al.* 2018). The high extractive content of red cedar (*Toona ciliata* M.Roem.) was the most probable explanation for its high natural resistance despite the low basic density of 320 kg/m³ (Ribeiro *et al.* 2014). The rise in extractive level because of the decomposition of the primary components of the cell wall also indicates the extractive's protective potential (Gallio *et al.* 2020). The low durability of turkey oak (*Quercus cerris* L.) was attributed to its low extractive and tannin contents (Bajraktari *et al.* 2018). Moreover, extractives that are impregnated in the cell wall may increase durability by delaying lignin degradation (Füchtner and Thygesen 2023). The secondary wood chemical components also include ashes. All four Amazonian hardwoods had remarkably low ash contents; hence, a meaningful protective effect is unlikely.

Differences occurred among the lignin content of the four species, being remarkably high in timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) and much lower in pau amarelo (*Euxylophora paraensis* Huber). Oppositely, there were no statistical differences among the holocellulose content of the species, except between tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes). On the other hand, when holocellulose was subdivided into cellulose and hemicelluloses, the species differed. Pau amarelo (*Euxylophora paraensis* Huber) and tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) showed no statistical difference between the cellulose contents, which were lower than the contents of pará-pará (*Jacaranda copaia* (Aubl.) D. Don) and tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks). The hemicelluloses' levels differed among the hardwoods, with the highest difference between tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) (Table 2).

Table 2: Primary chemical composition of the hardwoods (based on extractive-free wood mass)

Species	Lignin (%)	Holocellulose (%)	Cellulose (%)	Hemicellulose (%)
E. paraensis	$25,1^{(0,64)}$d	$80,6^{(1,27)}$ab	$48,9^{(0,77)}$b	$31,8^{(0,77)}$b
J. copaia	$27,7^{(0,68)}$b	$80,6^{(0,37)}$ab	$50,1^{(0,24)}$a	$30,5^{(0,24)}$c
P. suaveolens	$29,4^{(0,43)}$a	$78,5^{(1,66)}$b	$50,8^{(0,57)}$a	$27,6^{(0,57)}$d
A. integrifolia	$26,3^{(0,13)}$c	$81,2^{(1,90)}$a	$48,0^{(0,06)}$b	$33,2^{(0,06)}$a
X^2	0,041	0,002	0,006	0,055

Average [(Standard Deviation)]; X^2 = species deviance has a highly significant effect if p-value < 0,001.
Averages followed by the same letter did not differ by the post-hoc test at 5 % significance.

Although not so wide, the variation in primary chemical composition is a key explanation for hardwood's natural resistance. The termites consume the cellular wall but can digest and are fed from polymeric carbohydrates, cellulose, and hemicelluloses. However, lignin encrusts the polysaccharides in the wood cell wall providing mechanical strength, imperviousness, and resistance to pathogens (Boerjan *et al.* 2003). Lignin is a heterogeneous phenylpropanoid polymer derived primarily from oxidative couplings of p-hydroxycinnamyl alcohols (Boerjan *et al.* 2003). Due to its chemical complexity and lack of vulnerable linkages, lignin confers

lignocellulosics' high resistance to most microbial attacks. Its deconstruction is a critical step in lignocellulose digestion. It first enables the dissociation of the recalcitrant lignin polymers from the cellulose and hemicelluloses in which they are embedded, making the polysaccharides available for assimilation and energy (Tarmadi *et al.* 2018).

The wood cell wall digestion is a highly coordinated process achieved by the termite's gut-resident microbial symbionts. The flagellated protists in the hindguts of the so-called lower termites effectively degrade cellulose by various cellulolytic enzymes. The fecal pellets of the *Cycnotrachelodes formosanus* termite revealed selective digestion of cellulose over hemicelluloses and lignins (Tarmadi *et al.* 2018). Despite the lowest cellulose content (27,6 %) of tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks), the difference from the other species' contents (≈ 49-51 %) was low. The levels of hemicelluloses had a wider variation among the hardwoods (≈ 28-33 %), but unlike lignin, they are not feed-protective; hence differences in wood chemical resistance to termites are likely dependent on lignin content. This macromolecule structure must be deconstructed to make cellulose and hemicelluloses available (Tarmadi *et al.* 2018). Nevertheless, despite the highest lignin content, timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) had the lowest basic density, while pau amarelo (*Euxylophora paraensis* Huber) had the opposite results.

The hardwoods showed different behaviors of mass loss percentage over time (Figure 4). Pará-pará (*Jacaranda copaia* (Aubl.) D. Don) biodegradation sharply increased and mass loss was statistically above the other species on all evaluation days (Figure 4a). Meanwhile, the species showed similar mass loss up to the 28th day, when a statistical difference between pau amarelo (*Euxylophora paraensis* Huber) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W.Grimes) occurred. However, on the 35th day, the mass loss of tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) remarkably increased concerning the other two species and reached its maximum on the 42nd day. pau amarelo (*Euxylophora paraensis* Huber) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) showed lower and more stable degradation levels over time (Figure 4b).
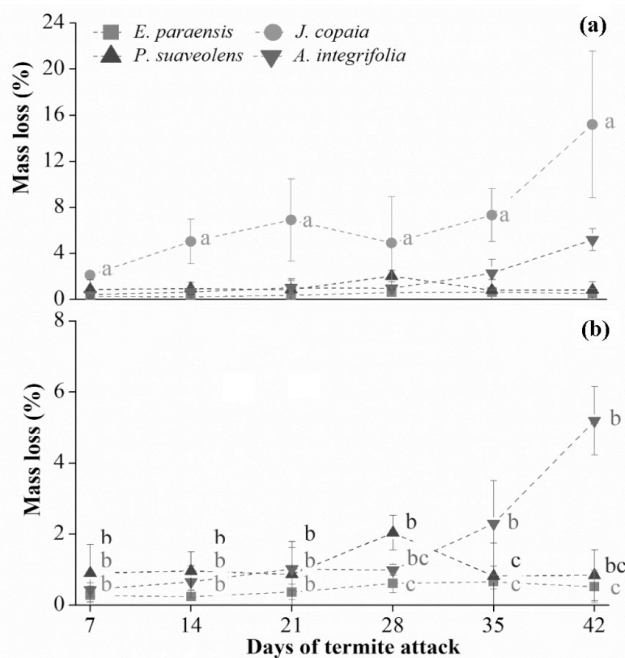

Figure 4: Percentage mass loss of the hardwoods attacked by arboreal termites over time: (a) all species, and (b) magnification of the scale for improved view of *A. integrifolia*, *P. suaveolens*, and *E. paraensis*. X^2 (species deviance has a highly significant effect if p-value < 0,001) for each day = 2,452 for 7th 6,847 for 14th 8,462 for 21st 3,725 for 28th 8,523 for 35th, and 17,505 for 42nd. Averages followed by the same letter did for each day not differ by the post-hoc test at 5 % significance.

Wear level (visual scores) differed among all species ranking pau amarelo (*Euxylophora paraensis* Huber) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) as the most and equally resistant ones for most of the evaluation days. Nevertheless, despite the statistical similarity, pau amarelo (*Euxylophora paraensis* Huber) only had full 10's; hence, observers could not detect a single sign of biodeterioration. The grades for wear level were statistically lower for tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) and

pará-pará (*Jacaranda copaia* (Aubl.) D. Don). The second showed the most degraded appearance for most of the evaluation days, following the mass loss pattern (Table 3).

Table 3: Wear-level evolution of the hardwoods over time and final appearance.

Species	7th	14th	21th	28th	35th	42th
J. copaia	$9,0^{(0,0)}$b	$8,2^{(1,1)}$b	$7,0^{(0,0)}$c	$8,2^{(1,1)}$b	$7,8^{(1,1)}$c	$7,4^{(0,9)}$b
A. integrifolia	$8,6^{(0,9)}$b	$8,8^{(1,1)}$ab	$9,0^{(1,1)}$b	$9,4^{(0,5)}$a	$7,8^{(1,1)}$c	$9,2^{(0,4)}$a
E. paraensis	$10,0^{(0,0)}$a	$10,0^{(0,0)}$ab	$10,0^{(0,0)}$a	$10,0^{(0,0)}$a	$10,0^{(0,0)}$a	$10,0^{(0,0)}$a
P. suaveolens	$9,6^{(0,5)}$a	$8,8^{(1,1)}$ a	$9,2^{(0,4)}$b	$9,6^{(0,5)}$a	$9,8^{(0,4)}$a	$9,4^{(0,5)}$a
X^2	0,070	0,110	0,354	0,119	0,303	0,267

Average [(Standard Deviation)]; X^2 = species deviance has a highly significant effect if p-value < 0,001 and p<0,05.Averages followed by the same letter did not differ by the post-hoc test at 5 % significance.

The fastest and most intense degradation over time of the lowest-density hardwood analyzed herein, pará-pará (*Jacaranda copaia* (Aubl.) D. Don), followed by the second-most intense degradation of tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks), confirmed the relation between density and degradation susceptibility. However, a high-density wood would not necessarily be as much degraded as a low-density one, but the same wear would require a longer attack. Unlike pará-pará (*Jacaranda copaia* (Aubl.) D. Don), tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) remarkable degradation occurred only on the 35th day, equalizing pará-pará (*Jacaranda copaia* (Aubl.) D. Don) degradation on the 7th day. tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) cell wall was thinner than pará-pará (*Jacaranda copaia* (Aubl.) D. Don). Still, its smaller pore diameter possibly explains its long time to reach a noticeable degradation by hindering termites' entrance into the wood. The other two hardwood species, with the highest bulk densities, had much lower and more stable degradation. Besides decreasing density and accelerating biodegradation, bigger pores possibly favor termite establishment within the wood for more efficient feeding, explaining the positive correlation between the variables.

Despite usual, higher density does not always guarantee a higher natural resistance. For instance, Costa *et al.* (2019) found that *Dipteryx odorata* had the highest density (1101 kg/m³) among seven species and showed a mass loss of 1,61 % when attacked by termite *Nasutitermes octopilis*. Still, *Cariocar villosum*, with the lowest density (490 kg/m³), had a mass loss of only 1,45 %. The most degraded wood was *Astronium lecointei,* with a mass loss of 12,6 % and an average density of 610 kg/m³. Another work showed that, despite the very low density of 320 kg/m³, red cedar (*Toona ciliata* M.Roem.) hardwood had a far lower mass loss percentage (1,7 %) than ocote (*Pinus oocarpa* Schiede ex Schltdl.) softwood (6,6 %) with a density of 360 kg/m³ (Ribeiro *et al.* 2014). Only wood chemistry can explain such discrepancies in the literature because it distinguishes the palatability of the several hardwoods.

The X-ray diffractograms of the hardwoods kept their typical cellulose pattern over time despite the termite attacks, but the species depicted different crystalline indexes. Moreover, the modification behavior of the crystalline index was not the same for the four species. Over time, a wider range of this attribute occurred for pará-pará (*Jacaranda copaia* (Aubl.) D. Don), while it was slightly more stable in timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes). It increased from the 7th to the 21st day for pau amarelo (*Euxylophora paraensis* Huber) and pará-pará (*Jacaranda copaia* (Aubl.) D. Don) but decreased again, becoming similar to the start point. On the other hand, the crystalline index of tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.

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Jacks) wood oscillated over time but, in the end, reached the same value as the beginning (Figure 5).
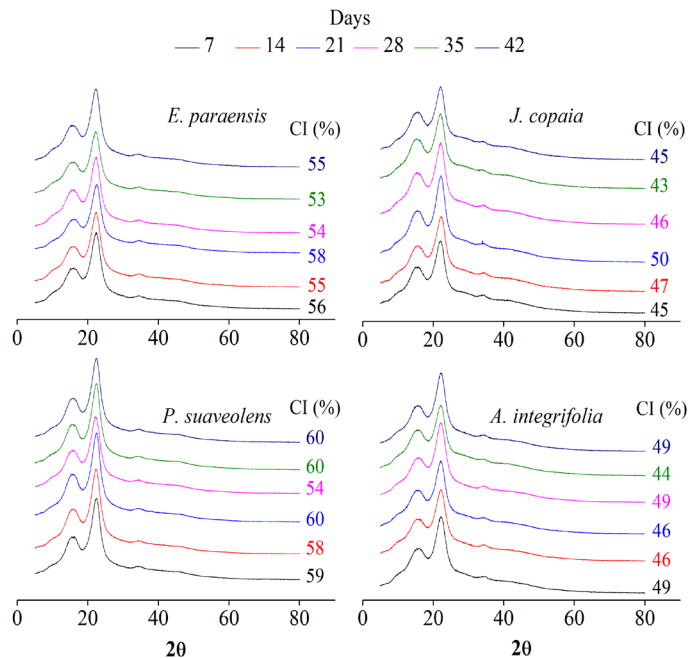

Figure 5: X-ray diffraction and crystalline indexes (CI) of the hardwoods attacked by arboreal termites over time.

Cellulose occurs in the cell wall as semi-crystalline microfibrils, the target of termite feeding. All samples exhibited typical cellulose Iβ diffractograms, revealing two fundamental peaks ascribed to the crystallographic planes (101) at 15,5°, (101) at 16,5° and (002) at 22,6°. Additionally, a weak peak appeared at ~34° in all samples, assigned to the (040) crystallographic plane, which reflects a mixture of crystalline celluloses I, II, and Iβ. Xylan and lignin do not display any diffraction peaks but diffuse scattering halos ranging from 12° to 27°, overlapping with the crystalline diffraction peak positions (Bouramdane *et al.* 2022). The crystalline regions of the cellulose are more recalcitrant to microbial and enzymatic attack than the amorphous domains, influencing the enzymatic hydrolysis rate (Zhao *et al.* 2012). Pau amarelo (*Euxylophora paraensis* Huber) and timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) species had higher crystalline indexes than tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D. Jacks) and pará-pará (*Jacaranda copaia* (Aubl.) D. Don), possibly contributing to their enhanced resistance to termite attack.

The hydrolysis of the amorphous regions of cellulose by easy enzymatic digestion (Kafle *et al.* 2015), leaving behind the crystalline portions (Dar *et al.* 2018), possibly explains the increased crystallinity that appeared for timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes), pará-pará (*Jacaranda copaia* (Aubl.) D. Don), and pau amarelo (*Euxylophora paraensis* Huber) at different evaluation days. The evolution of broad signals towards sharp ones that appeared in the diffractograms possibly is caused by small crystallite sizes. The biodegradation could induce defects in the crystal network, decreasing the organized portions responsible for more diffused crystalline peaks (Ling *et al.* 2019). However, the crystalline indexes of pau amarelo (*Euxylophora paraensis* Huber) and pará-pará (*Jacaranda copaia* (Aubl.) D. Don) decreased again over time, approaching the indexes of the first evaluation day. This pattern suggests that several chemical types of degradation mechanisms, such as hydrolysis and oxidation, occur with a similar facility in the cellulose's amorphous and crystalline regions (Bouramdane *et al.* 2022). Therefore, this work allows inferring degradation may eventually start on the amorphous fractions of the cell wall but later become less selective. Previous work also reported homogeneous deterioration of the main components of the deteriorated woods Fourier Transform Infrared Spectroscopy (Gallio *et al.* 2020).

The wear level, average mass loss percentage, and final mass loss percentage ultimately reveal the combined effect of the hardwoods' chemistry, anatomy, and basic density on their natural resistance to termites. Besides the lowest lignin and average extractive contents, pau amarelo (*Euxylophora paraensis* Huber) was by

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far the most resistant hardwood among the four, proving its anatomy and basic density overcame its chemistry when arboreal termites attacked it. Pará-pará (*Jacaranda copaia* (Aubl.) D. Don) lowest density made it the least resistant among the four hardwoods. It is also possible that its lowest level of organic extractives unfavoured its natural resistance, although this effect is not easily proven. A study showed that varied wood species exhibited higher weight loss by termite and fungi feeding when extractives were removed before the attack (Kirker *et al.* 2013). A similar bioassay could review how protective the extractives of the hardwoods studied herein actually are. Compared to pará-pará (*Jacaranda copaia* (Aubl.) D. Don), tamanqueira (*Aegiphila integrifolia* (Jacq.) B.D.Jacks) intermediate density delays but does not prevent its biodegradation, which seems to relate mostly to its cells' biometry. Timborana (*Pseudopiptadenia suaveolens* (Miq.) J.W. Grimes) showed slight evidence of chemistry capability to protect from termite feeding. This wood had fibers with thinner cell walls and larger lumens than pau amarelo (*Euxylophora paraensis* Huber), but their densities were similar. The small pore diameters explained its density, but it also had the highest extractive and lignin contents, possibly favoring its increased resistance.

The benefits or drawbacks of faster or slower species-dependent biodegradation in mining stockyards depend on the point of view. Although this work limits its evaluation to only four hardwoods and one type of degradation in laboratory facilities, their unique anatomy and chemistry provided useful insights into how susceptible the different species are when left unprotected in a stockyard. If the company can donate such logs for useful applications, this work revealed that wood density remains the most direct criterion and low-density species should be a priority for protection. On the other hand, if the companies cannot decide on practical applications, slower degradation of higher-density species is a drawback because they will remain in the environment longer.

CONCLUSIONS

The anatomical traits and chemical compositions of four tropical hardwoods stocked in a bauxite mine in the Amazon differed markedly. The anatomical traits that favored increased density also favored increased resistance to the arboreal termite attack. Thicker cell walls of the fibers and smaller diameters of the pores were the most relevant ones. The capability of the chemical composition to provide more or less naturally resistant hardwoods was unclear in this work because of the direct effect of anatomical biometry and basic density on wood mass loss and visual wear. Nevertheless, combined with the lowest density, *J. copaia* had the unfavorable lowest extractive level. Besides its thinnest fiber cell wall, *A. integrifolia* was not so degraded. The highest lignin and organic extractive levels possibly favored this species.

The behavior of degradation varied among the hardwood species. The two denser species (*E. paraensis* and *P. suaveolens*) were degraded slower and without sharp increase over time. The one with the lower density (*J. copaia*) depicted a consistent and sharp increasing degradation over time. The one with the average basic density (*A. integrifolia*) started degrading meaningfully after 35 days of arboreal termite attack. Thus, its basic intermediate density delayed but did not prevent degradation. The changes in the crystalline indexes over time suggested that termite feeding mechanisms may be more or less selective depending on degradation stages and species. The denser and most resistant species also had the highest crystalline indexes.

Authorship contribution

A. K. S. S. C.: Data curation, formal analysis, investigation, methodology, software, and writing - original draft. M. C. B. D.: Data curation, formal analysis, investigation, methodology, software, and writing - original draft. I. V.: Data curation, formal analysis, investigation, methodology, software, and writing - original draft. T. M. S.: data curation, funding acquisition, and writing - review & editing. M. T-F.: Funding acquisition, resources, and visualization. J. L. L. A. Formal analysis, and methodology. L. M. M.: Visualization, resources, writing

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- review & editing. T. P. P.: funding acquisition, methodology, resources, visualization, and writing - review & editing. G. C. F.: Funding acquisition, project administration, and supervision. M. G. S.: Conceptualization, funding acquisition, project administration, and supervision. C. F. O. N.: Conceptualization, project administration, resources, and supervision. L. B.: Conceptualization, funding acquisition, investigation, project administration, resources, supervision, writing - original draft, and writing - review & editing.

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