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# Selectivity of soil constituents by termites in the construction of Brazilian termite mounds

Tatiele Fruett<sup>1</sup>, Alberto Vasconcellos Inda<sup>1</sup>, Vidal Barrón<sup>2</sup>, Enilson Luiz Saccol de Sá<sup>1</sup>, Keyrauan Taha<sup>1</sup>, Antonia Finkler Dias Fernandes<sup>1</sup>

<sup>1</sup>Universidade Federal do Rio Grande do Sul – Depto. de Solos, Av. Bento Gonçalves, 7712 – 91540-000 – Porto Alegre, RS – Brasil.

<sup>2</sup>Universidad de Córdoba – Departamento de Agronomía, Campus de Rabanales, Edificio C4 – 14071 – Córdoba – España

\*Corresponding author <alberto.inda@ufrgs.br>

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Received July 26, 2022 Accepted November 23, 2022 **ABSTRACT**: Termites can create structures that alter the physical and chemical properties of soils. In this process, termites are selective about the soil constituents they will use to construct their mounds. Considering the common occurrence of termite mounds in Brazilian soils, this study aimed to investigate the selective action of termites in the mound building process. Samples were collected from six termite mounds and control soils (at a distance of 15 to 30 m from the termite mound) in different regions in Brazil to analyze the fine earth fraction. The content of clay fraction, organic C and Fe in pedogenic iron oxides increased in the mounds resulting in specific surface area increments. X-ray diffraction indicated a selectivity of termites by clay-sized particles such as kaolinite, gibbsite and iron oxides (hematite and goethite) rather than larger particles such as quartz. The proportion of low-crystalline iron oxides and the maghemite amount decreased in the mounds. The change of color parameters in the termite mounds was due to a combination of increase in clay fraction, organic carbon and iron oxides. The techniques used were sensitive, indicating changes and similarities between the control soils and the termite mounds.

**Keywords**: X-ray diffraction, VIS-MIR spectra, tropical soils, macrofauna, bioturbation

### Introduction

Termites (Blattodea: Termitoidae) are invertebrates in the soil macrofauna that feed on soil and plant matter or only wood and grass (Brune, 2014), and create structures that facilitate their locomotion and survival in the soil (Jouquet et al., 2006; Ferreira et al., 2011; Jouquet et al., 2016b). These organisms are more adapted to dry and warm pedoenvironments, where they can supplant the function that earthworms play (Evans et al., 2011; Jouquet et al., 2016a). However, as regards the action of termites in soil formation, several questions remain to be investigated (Lobry de Bruyn and Conacher, 1990; Jouquet et al., 2015; Jouquet et al., 2016a).

In the process of building their mounds, which involves the formation of tunnels and chambers, termites concentrate organic material -deposits of feces resulting from feeding activity-, clay sized particles and nutrients compared to the control soils (Donovan et al., 2001; Jouquet et al., 2002; Rückamp et al., 2012; Sarcinelli et al., 2013; Wang et al., 2015; Jembere et al., 2017; Traoré et al., 2019; Jouquet et al., 2020). The extent of this selectivity and its differential effects depend on the properties of the soils and their environments (Jouquet et al., 2016b), the termite species and their feeding strategies, as well as available ecological resources (Kaiser et al., 2017; Shanbhag et al., 2019).

Studies on the selectivity of soil constituents by termites in constructing their mounds are numerous on the Asian and African continents. On the other hand, unlike those continents, fungus growing termites are not found on the American continent, and few studies have been carried out in South America on the topic of particle

selectivity by termites (Kaschuk et al., 2006; Rückamp et al., 2012; Sarcinelli et al., 2013). The present study presupposes that termites change the soil characteristics during the construction of their mounds and that these changes in the soils vary according to termite species and environmental conditions. Thus, the morphological, physical, chemical, and mineralogical characteristics of soil and termite mounds in the different regions of Brazil were investigated. In addition to the traditional physical and chemical analysis, this study seeks to characterize the mineralogical compositions of control soils and termite mound materials and elucidate mineralogical changes by X-ray diffraction and magnetic susceptibility analysis. Furthermore, color change differences between control soil and termite mound materials were investigated by diffuse reflectance spectroscopy.

#### **Materials and Methods**

This study was carried out on samples collected in Rio Grande do Sul (RS-JC), Rio Grande do Sul (RS-ES), Minas Gerais (MG), Piauí (PI), Mato Grosso (MT) and Pará (PA) environments located in five states in Brazil (Figure 1; Table 1). Control soil samples were collected under natural vegetation at soil depths of 0.00-0.20, 0.20-0.40 m (RS-JC, RS-ES, MG and PI) and 0.00-0.40 m (MT and PA) in areas close to the termite mounds (distance 15 to 30 m from termite mound) with no visible action of the termites. Single-layer collection in the MT and PA environments was due to the expressive similarity of the soil in this layer. Termite mound samples were collected in triplicate in the top and middle positions of the active epigeal mounds (Figure 2). Samples were collected from



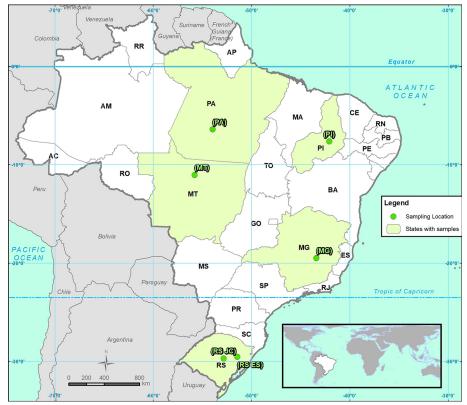
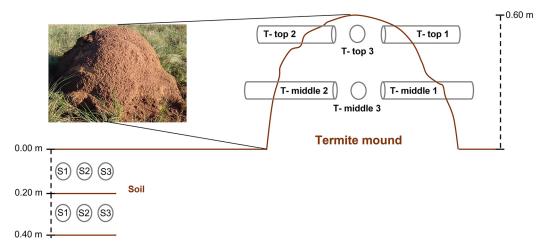


Figure 1 – Location (environments) of the studied soils and termite mounds.



**Figure 2** – Details where samples were collected from the termite mounds.

the external wall to the center of the mounds and termite mounds with active colonies and representative sizes were chosen from all the environments. The samples were air dried, ground and passed through 2 mm mesh sieves to obtain the fine earth fraction.

# Physical and chemical analysis

Particle size distribution analysis was carried out using

the pipette method (Teixeira et al., 2017). The specific surface area (SSA) of the fine earth was estimated by the method of water adsorption in an atmosphere where relative humidity (UR) = 20 % (Quirk, 1955). For the calculation of the SSA, it was assumed that a water molecule covers an area of 0.108 nm². Magnetic susceptibility ( $\chi$ ) was measured at 0.47 kHz ( $\chi_{\rm H}$ ) in the fine earth fraction using a Bartington magnetometer with a dual frequency sensor (System MS2).

Sample code	Sample code Minicipality/state Lithology Climate <sup>3</sup>	l ithology	Climate <sup>3</sup>	, o i	7 tun 4	Altitude	Soil classification	fication	Vegetation	Height	Termite specimen Feed habit		Gengraphic location
odilibic code	mailleibailig/ state	LIUNIUS	OIII IN C	χtς	х ррг	יונוממכ	OOII CIRSSI	וכמווסוו	* Cectation	ııcıgıır	icillice apecillicil		acogi apliic iocation
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RS-JC	Júlio de Castilhos/ Rio Grande do Sul	basalt/ sandstone	Cfa	18.0	1,575	513	Latossolo <sup>1</sup> Vermelho	Rhodic <sup>2</sup> Ferralsol	Pampa (grasslands)	0.75	Cortaritermes	grass	29°6′1.52″ S 53°37′19.36″ W
RS-ES	Eldorado do Sul/ Rio Grande do Sul	granite	Cfa	19.5	1,309	96	Argissolo Vermelho	Rhodic Acrisol	Pampa (grasslands)	09:0	Cornitermes	litter and soil	30°6′10.84″ S 51°40′33.80″ W
MG	Lavras/ Minas Gerais	gabbro	Cwa	19.0	1,530	936	Latossolo Vermelho	Rhodic Ferralsol	Cerrado (woody-grasslands)	1.05	Cornitermes	litter and soil	21°12′18.03″ S 44°59′36.95″ W
MT	Canabrava do Norte/recent Mato Grosso sediment	recent sediments	Am	27.0	1,578	202	Neossolo Quartzarênico	Gleyic Arenosol	Cerrado (woody-grasslands)	1.65	Cornitermes	litter and soil	11°0′39.66″ S 51°38′7.33″ W
PA	Santana do Araguaia/ Pará	granite	Am	27.5	1,919	199	Argissolo Vermelho- Amarelo	Haplic Acrisol	Amazônia (ombrophilous forest)	1.50	Labiotermes	soil	9°41'36.52" S 50°57'31.11" W
础	Cristino Castro/ Piauí	sandstone	Aw	26.7	849	345	Latossolo Amarelo	Xanthic Ferralsol	caatinga-cerrado contact	0.95	Syntermes	litter and soil	8°51'13.30" S 44°17'36.07" W
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Brazilian Soil Classification System (Santos et al., 2018); <sup>2</sup>WRB (IUSS Working Group WRB, 2015); <sup>3</sup>Mean annual temperature; <sup>4</sup>Mean annual precipitation; Cfa = temperate without dry season; Cwa = temperate w winter; Am = tropical monsoon; Aw = tropical savannah.

Total organic carbon (TOC) of the soil was determined in the fine earth fraction after grinding the samples in agate grains. Approximately 1 g of the sample was subjected to dry combustion in a SHIMADZU VCSH carbon analyzer. Pedogenic iron oxides (Fed) were extracted in the fine earth fraction using two successive extractions with sodium dithionite-citrate-bicarbonate at 80 °C (Mehra and Jackson, 1960), and poorly crystallised ones (Feo) were extracted by 0.2 mol L<sup>-1</sup> ammonium oxalate at pH 3 in the dark (Teixeira et al., 2017).

# Mineralogical analysis

The minerals present in the fine earth of soils and termite mounds were identified and characterized by X-ray diffraction (XRD) in Bruker-D2-Phaser equipment equipped with a fast linear detector (type LYNXEYETM) and analysis software (DIFFRAC. SUITE™). The equipment used CuK $\alpha$  radiation ( $\lambda = 1.5418 \text{ Å}$ ) with an Ni filter, 10 mA current, and 30 kVA voltage. To verify possible mineralogical changes between soil and termite mound materials, samples of the same weight (600 mg) of both materials were finely ground in an agate mortar and analyzed as randomly oriented powders in the 2-40° 2θ range with a scanning speed of 1° min<sup>-1</sup>. A semiquantitative comparison between the minerals present in the soil samples and the thermite mound samples was made based on the intensity of the reflections of the minerals in the XRD. The XRD data (d-spacings and the intensities of the reflections) were obtained using the Eva Diffract Suite program. The identification of the minerals in the samples was based on the d-spacings after searching the d-spacing tables for CuKα radiation contained in Brown and Brindley (1980).

# **Color parameters**

Samples of the fine earth of soils and termite mounds were finely ground in an agate mortar and analyzed using a diffuse reflectance spectrophotometer. The spectra were recorded in the wavelength range from 380 to 800 nm with 0.5 nm intervals using a UV-Visible-NIR CARY 5000 instrument with a 110 mm diameter coupled integrating sphere. For calibration, maximum reflectance (100 % T) was obtained with polytetrafluoroethylene (PTFE) - Teflon, and minimum reflectance (0 % T) was obtained by placing a black stripe at the entrance of the light beam. The values of X, Y and Z were obtained from the spectrum and applied in the Munsell Conversion program, which provides hue (H), value (V) and chrome (C), RGB data, among other color parameters. The redness rating (Torrent and Barrón, 1993) was calculated using the formula RRf (hm) = (10 - H)\*(C/V). The hematite / (hematite + goethite) ratio (Hm / (Hm + Gt)) was estimated by the relative intensities of the bands between + 410 and 445 nm (IGt) and between + 530 and 580 nm (IHm) of the spectrum using the second derivative of the Kubelka-Munk function (Torrent and Barrón, 2008).

A Tensor 27 MIR spectrophotometer from Optics-Bruker equipped with a Pike EasiDiff Diffuse reflection hemisphere was used to scan the wavelength range from 4,000 to  $600~{\rm cm}^{-1}$  (2,500 to  $16,666~{\rm nm}$ ) at intervals of 8 to 1.64 times per second. Potassium bromide (KBr) was used as a spectral reference for the MIR band.

#### Identification of termite species

The termite specimens collected from the mounds in each environment were stored in 70 % alcohol before identification. The illustrated key for the identification of termite genera (Insecta: Isoptera) that are found in Brazil and the Catalog of the termites of the New World (Insecta: Isoptera) (Constantino, 1998) were used for specimen identification. Photos for identification were taken at ZEISS Stemi 508 Stereo Microscope using an Axiocam ICC5 with specimens in preservation solution (alcohol 70 % + glycerol 1 %).

Termite specimens collected in six environments are shown in Figure 3. In the RS-ES, MG and MT environments, the specimens were classified according to the genus as Cornitermes. The other genus were Cortaritermes in RS-JC, Syntermes in PI, and Labiotermes in PA. As regards typical feeding habits,

the Cornitermes and Syntermes genus feed on litter (light soil organic matter) and soil (minerals and humus) [litter-soil-feeders], while Labiotermes feed only on soil (minerals and humus) [soil-feeders] and Cortaritermes feed only on grasses [grass-feeders] (Donovan et al., 2001; Lima and Costa-Leonardo, 2007). In the litter-soil-feeders and soil-feeders feeding habits, the term "soil" includes minerals and humus materials.

The mounds of Cornitermes cumulans, a very common termite in South American grasslands, display morphological transformations during the colony development. Young colonies inhabit small subterranean mounds that develop into large, conspicuous, epigean mounds inhabited by populous colonies. The walls of large mounds are composed of a porous mass of sands densely cemented with organic matter and clay in the mound, and a compact mass of the same components in the floor (Cosarinsky, 2011). Species from the Cornitermes genus build sub-conical or globular mounds that can reach 2 m in height but usually have a diameter of approximately 1 m (Cosarinsky, 2011). The termite species Cortaritermes fulviceps, in the northeast of Argentina, constructs mounds as smaller earthy domes, 30-40 cm high, with no carton hive in its base inhabited by the larvae (Cosarinsky, 2004a, b). Species

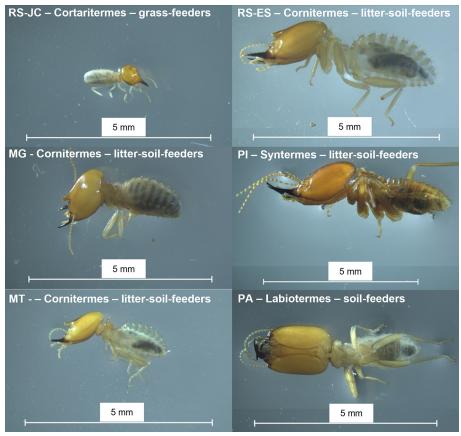


Figure 3 – Photos of the termite specimens collected in the Rio Grande do Sul (RS-JC), Rio Grande do Sul (RS-ES), Minas Gerais (MG), Piauí (PI), Mato Grosso (MT) and Pará (PA) environments.

of the Syntermes genus construct their mounds with 2-m-high conical shape and large diameters at the base (Constantino, 1995). In the lowlands of the Amazonian rainforest, the termites of the Labiotermes labralis species feed on soil (minerals and humus) and are common nest-building species and their colonies build single spheroidal arboreal mounds close to the ground (Pequeno et al., 2013).

# Data analysis

The statistical analysis of the evaluated parameters was determined using the PAST Statistics Software® (Hammer et al., 2001), where averages were compared using the Tukey test at the 5 % level of significance (p < 0.05). The data obtained in the soils were compared to the data from the termite mounds only within each collection site (environment).

#### **Results and Discussion**

#### Physical and chemical characterization

The clay content in the soils varied between 5 % (Mato Grosso – 0-0.40 m) and 48 % (Minas Gerais – 0.20-0.40 m), and the content in the termite mounds varied between 14 % (Mato Grosso – top) and 68 % (Minas Gerais – top)

(Table 2). The clay content increased with depth in three environments where it was possible to collect the two soil layers (0.00-0.20 and 0.20-0.40). In the termite mounds, the clay content was similar between the top and middle positions. In all environments, the clay contents were higher in the termite mounds than in the control soil, mainly in the 0.00-0.20 m layer of the soils, agreeing with the feeding habit of most of the species under study, in which soil particles are included (Cornitermes, Syntermes and Labiotermes), and indicating selectivity on the part of the termites for particles smaller than  $2 \mu m$ . The smallest increase in the clay content in the mound compared to the soil (T/S ratio) was found in the Rio Grande do Sul (RS-JC and RS-ES) environments, where the termites were classified, respectively, as grass-feeders (Cortaritermes), which feed mainly on dry leaves found in the undergrowth; and litter-soil-feeders (Cornitermes), which feed predominantly on light soil organic matter mixed with soil. Similarly, in a field study in southwestern Ethiopia, Jembere et al. (2017) found that the clay content decreased as the distance from the termite mounds increased. However, our results contrast with a study in southern Brazil, where no differences were found in clay content between adjacent soils at both the bottom and the top of mounds (Kaschuk et al., 2006).

The amounts of total organic carbon (TOC) in soils ranged from 0.50 % (Piauí – 0.20-0.40 m) to

Table 2 – Physical attributes and iron content relative to pedogenic iron oxides (Fed) and poorly crystalline iron oxides (Feo) in the soil (S) and termite mound (T) materials in the Rio Grande do Sul (RS-JC), Rio Grande do Sul (RS-ES), Minas Gerais (MG), Piauí (PI), Mato Grosso (MT) and Pará (PA) environments.

Environment/ specimen	Sample	Clay	Silt	Sand	TOC1	Fed	Feo	Feo/Fed	$\chi_{lf}^{\ 2}$	SSA <sup>3</sup>
•				%		g kį	g <sup>-1</sup>		10 <sup>-7</sup> m <sup>3</sup> kg <sup>-1</sup>	m² g-1
	S 0.00-0.20	26	8	66	1.00 c	14.23 b	0.71 b	0.05	73.7 c	31.5 a
RS-JC	S 0.20-0.40	33	10	57	0.89 d	16.94 a	0.68 b	0.04	79.1 a	32.0 a
Cortaritermes	T middle	33	11	56	2.40 a	16.33 a	0.69 b	0.04	59.3 d	34.7 a
	T top	37	10	53	2.25 b	16.03 b	1.01 a	0.06	77.0 b	34.9 a
	S 0.00-0.20	26	17	57	0.99 c	8.84 b	1.06 a	0.12	10.4 d	33.5 b
RS-ES	S 0.20-0.40	33	17	50	0.96 c	11.23ab	0.51 c	0.05	29.0 a	31.7 b
Cornitermes	T middle	36	19	45	1.76 a	12.55 a	0.95 ab	0.08	14.4 c	41.9 a
	T top	36	18	46	1.69 b	12.32 a	0.81 b	0.07	16.1 b	42.2 a
	S 0.00-0.20	46	42	12	2.60 c	84.83 a	1.39 a	0.02	896.8 b	45.9 a
MG	S 0.20-0.40	48	42	10	1.64 d	83.28 a	1.28 a	0.02	944.3 a	43.8 a
Cornitermes	T middle	51	39	10	3.98 b	86.26 a	0.96 a	0.01	815.0 d	49.9 a
	T top	68	17	15	4.58 a	86.87 a	1.24 a	0.01	859.8 c	51.2 a
LAT.	S 0.00-0.40	5	2	93	0.58 c	3.58 c	0.97 b	0.27	0.6 c	12.5 c
MT Cornitermes	T middle	16	4	80	1.69 b	16.88 a	0.93 b	0.06	1.6 b	19.4 b
	T top	14	4	82	2.37 a	12.50 b	1.44 a	0.12	2.6 a	27.4 a
PA Labiotermes	S 0.00-0.40	17	7	76	1.31 b	6.75 a	0.86 a	0.13	2.3 b	20.2 b
	T middle	34	9	57	1.13 c	7.14 a	0.25 c	0.03	2.0 c	34.2 a
	T top	34	9	57	1.98 a	7.37 a	0.46 b	0.06	2.6 a	28.8 b
	S 0.00-0.20	14	3	83	0.60 c	2.77 d	0.15 c	0.05	0.6 с	16.9 b
Pl	S 0.20-0.40	28	6	66	0.50 d	7.79 a	0.17 bc	0.02	1.1 a	19.3 a
Syntermes	T middle	29	6	65	1.52 a	6.47 b	0.36 a	0.06	0.8 bc	21.5 a
	T top	27	6	67	1.11 b	5.73 c	0.23 b	0.04	0.9 ab	17.5 b

Means followed by the same letter do not differ by Tukey's test at the p < 0.05 probability level; <sup>1</sup>Total organic carbon; <sup>2</sup>Low frequency magnetic susceptibility; <sup>3</sup>SSA = specific surface area.

2.60 % (Minas Gerais - 0-0.20 m) and amounts in termite mounds ranged from 1.11 % (Piauí - top) to 4.58 % (Minas Gerais - top) (Table 2), with higher values in termite mounds than in the control soils in all evaluated environments. The lowest increment of C in the termite mound was found in the Pará environment. The termite specimens were classified as soil-feeders (Labiotermes), which feed on humified organic matter associated with soil minerals. Considering the low soil C content, soilfeeder termites need to ingest a larger amount of soil to ingest enough humus, as shown by the increment in clay in the termite mound in this environment (Table 2). On the other hand, in soils with higher humified organic matter contents, soil-feeder termites can concentrate high C contents in the mounds. This C accumulation is due not only to how termites build their mounds but also to recycling nutrients and decomposition from their food base composed of minerals and organic matter (litter or humus) (Kaschuk et al., 2006). These results corroborate the organic C enrichment factors in mounds from the Brazilian Cerrado, in the order of 1.4 to 2.0, compared to the control soil (Rückamp et al., 2012), with the concentration of organic C 11.9 times higher in mounds formed from sandy soils in the Bahia Atlantic Forest in Brazil (Sarcinelli et al., 2013).

The amounts of Fe relative to pedogenic iron oxides (Fed) in soils varied between 2.8 g kg-1 (Piauí -0-0.20 m) and 84.8 g kg<sup>-1</sup> (Minas Gerais - 0-0.20 m), and the amounts in termite mounds varied between 5.7 g kg<sup>-1</sup> (Piauí - top) and 86.8 g kg<sup>-1</sup> (Minas Gerais - top) (Table 2). The low Feo/Fed ratio values, mostly below 0.15, indicated a predominance of crystalline iron oxides (hematite, goethite and maghemite) compared to poorly crystallised iron oxides such as ferrihydrite (Bigham et al., 2002). In four environments (Rio Grande do Sul (JC and ES), Piauí and Mato Grosso), the amounts of Fed were higher in termite mounds than in the 0-0.20 m layer of soils. In addition, although not statistically significant, an increasing trend in Fed was verified in the mound materials in the Minas Gerais environment. Considering that pedogenic iron oxides are constituents of the clay fraction, these results corroborate the enrichment of this fraction in the termite mounds. Individually in the environments, it was verified that the increase in clay in the termite mound increased the Fed amount, regardless of feeding habit. Analyzing termites as bioindicators in the Caatinga in northeastern Brazil, Alves et al. (2011) found that even grass-feeder termites have clay-sized particles in their intestines.

In the Mato Grosso and Pará environments, the increase in the degree of crystallinity of iron oxides in the mounds, expressed by a decrease in the Feo/Fed ratio, may be related to the preferential dissolution of poorly crystallised iron oxides. This mineralogical alteration process may involve protonic dissolution reactions due to the decomposition of the organisms and the production of acids; basic reactions due to the high pH (pH > 8.0) of the termites' intestinal tract;

and complexative/protonic reactions by the production of organic ligands and acids during litter/wood/leaf decomposition. Another hypothesis for increasing iron oxide crystallinity in the termite mounds would be due to the transportation of soil from reductive (soils) to oxidative (mounds) environments (Abe and Wakatsuki, 2010). However, further studies are needed for a better understanding of this.

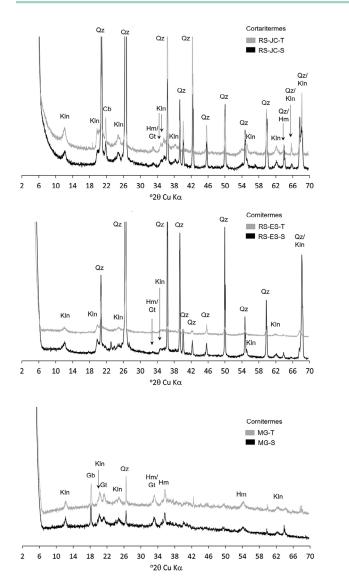
The increases in the clay and Fed in the mounds did not reflect increases in  $\chi$ , indicating the occurrence of magnetic iron oxides (magnetite and/or maghemite). In the environments where the materials presented the greatest magnetic susceptibilities (Rio Grande do Sul (JC and ES) and Minas Gerais), there was also a reduction in the  $\chi$  value in the mounds (Table 2). This reduction may result from the termite selectivity for the clay fraction, where the magnetite is not found. It may also suggest a selective dissolution of maghemite crystals which are less stable than hematite and goethite (Inda et al., 2013).

The specific surface area (SSA) in the soils varied between 12.5 m² g⁻¹ (Mato Grosso – 0-0.40 m) and 45.9 m² g⁻¹ (Minas Gerais – 0-0.20 m), and that in the termite mounds varied between 17.5 m² g⁻¹ (Piauí – top) and 51.2 m² g⁻¹ (Minas Gerais – top) (Table 2). In the RS-ES, MT and PA environments, the increase in SSA in termite mounds compared to control soils was significant, while in the other environments, only trends of SSA increase were observed.

#### X-ray diffraction (XRD)

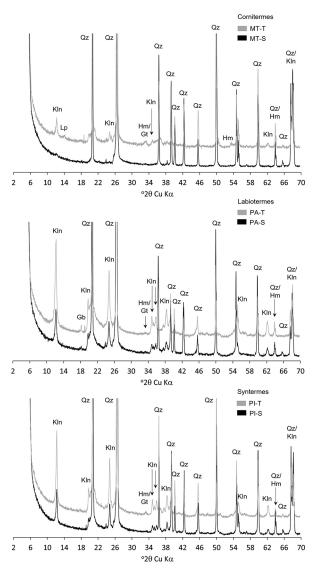
The mineralogical compositions identified in the XRD analyses were representative of intensely weathered soils from tropical and subtropical regions (Oliveira et al., 2020; Schaefer et al., 2008), formed by different proportions of kaolinite, gibbsite, iron oxides (hematite, goethite) and quartz (Figures 4 and 5). Considering that the analyzed samples were of equal weight and that the intensities of the reflections were not altered in order to construct Figures 4 and 5, the soils and termite mound materials in the Rio Grande do Sul (JC and ES) and Mato Grosso environments were considered quartz-kaolinitics; in the Piauí and Pará environments, the materials were kaolinitic-quartz; and in the Minas Gerais environment, the materials were oxidic (gibbsite, hematite, goethite, quartz)-kaolinitic.

In no environment did the analyses of the fine earth fraction ( $\varnothing$  < 2 µm) indicate a change in mineralogical bulk compositions between soils and termite mounds. However, as all analyzed samples had the same weight, considerable changes were in fact observed in the intensities of the reflections between the diffractograms of the soils and those of the termite mounds (Figures 4 and 5). In four environments (Rio Grande do Sul-JC, Piauí, Mato Grosso and Pará), the intensity of the quartz reflections decreased in the diffractograms of the termite mounds compared to those of the soils (layer 0.00-0.20 m), while the intensity of the



**Figure 4** – X-ray diffraction of powder samples of fine earth fraction of materials from the 0.00-0.20 m layer of soil (S) and the termite mound-middle portion (T) in the Rio Grande do Sul (RS-JC), Rio Grande do Sul (RS-ES) and Minas Gerais (MG) environments. KIn = kaolinite; Gb = gibbsite; Cb = cristobalite; Qz = quartz; Hm = hematite; Gt = goethite.

kaolinite reflections increased in the diffractograms of the termite mounds. In the case of iron and aluminum oxides, the intensity of reflections of hematite and goethite increased in the Rio Grande do Sul-JC, Piauí and Mato Grosso termite mounds; those of lepidocrocite increased in the Mato Grosso termite mound; and those of gibbsite increased in the Pará termite mound. These results corroborate the termite selectivity for minerals that are components of the clay fraction ( $\varnothing$  < 2 µm) in the construction of its mounds to the detriment of the quartz that predominates in the silt and sand fractions as shown by Donovan et al. (2001).



**Figure 5** – X-ray diffraction of powder samples of fine -earth fraction of materials from the 0.00-0.20 m and 0.00-0.40 m layer of soil (S) and the termite mound-middle portion (T) in the Piaui (PI), Mato Grosso (MT) and Pará (PA) environments. KIn = kaolinite; Gb = gibbsite; Cb = cristobalite; Qz = quartz; Hm = hematite; Gt = goethite; Lp = lepidocrocite.

A good example of this selectivity was observed in the diffractogram of the termite mound material from the Rio Grande do Sul-ES environment (Figure 4), where the quartz reflections were much lower than those in the soil material, possibly due to the size of the quartz crystals, considering that the soil was developed from granite. On the other hand, in the Minas Gerais environment, the diffractograms of the soil and the termite mound were similar in not suggesting selectivity on the part of the termites, possibly due to the soil being a very clayey soil developed from basalt. Similarly, Sarcinelli et al. (2013) concluded that the intensity of the selection of materials

and the changes caused by termites in constructing their mounds would be greater in more sandy and less fertile soils.

# Visible (VIS-NIR) and mid-infrared (MIR) spectra

The VIS and MIR spectra of the soils and termite mound materials in two of the environments are shown in Figure 6. In the VIS spectra, the positions of the bands relative to iron oxides goethite and hematite are shown, while in the MIR spectra, the positions of the bands of the kaolinite and quartz (Nguyen et al., 1991; Stenberg et al., 2010) are exhibited. The spectra obtained in the Minas Gerais environment exemplify a similarity between the materials of the control soil and those of the termite mound in the two analyzed spectra (Figure 6, top), as already verified through XRD. The spectra of the materials obtained in the Mato Grosso environment suggest possible differences between the materials (Figure 6, bottom). An analysis used to express the similarity between soil and termite mound materials in each environment was the correlation between the spectral data (soil versus termite mound) obtained in the two analyzed bands (VIS and MIR). The determination coefficients obtained for the visible range (380 to 730 nm) were greater than 0.96, with most correlations showing  $R^2 \ge 0.99$  (Table 3). On the other hand, the  $R^2$ values of the correlations between the data obtained in the mid-infrared range (300 to 4,000 cm<sup>-1</sup>) indicated

**Table 3** – Coefficients of determination (R<sup>2</sup>) relative to the correlations between visible (VIS) and mid-infrared (MIR) spectra data obtained in the soil (S) and termite mound (T) materials in the Rio Grande do Sul (RS-JC), Rio Grande do Sul (RS-ES), Minas Gerais (MG), Piauí (PI), Mato Grosso (MT) and Pará (PA) environments.

Environment/ specimen	Correlation	VIS (R²)	MIR (R²)
	S 0.00-0.20 versus T middle	0.99	0.78
RS-JC	S 0.20-0.40 versus T middle	0.99	0.90
Cortaritermes	S 0.00-0.20 versus T top	0.99	0.79
	S 0.20-0.40 versus T top	0.98	0.92
	S 0.00-0.20 versus T middle	0.99	0.97
RS-ES	S 0.20-0.40 versus T middle	0.99	0.91
Cornitermes	S 0.00-0.20 versus T top	0.99	0.95
	S 0.20-0.40 versus T top	0.99	0.93
	S 0.00-0.20 versus T middle	0.99	0.98
MG	S 0.20-0.40 versus T middle	0.99	0.97
Cornitermes	S 0.00-0.20 versus T top	0.99	0.93
	S 0.20-0.40 versus T top	0.99	0.97
MT	S 0.00-0.40 versus T middle	0.97	0.69
Cornitermes	S 0.00-0.40 versus T top	0.99	0.81
PA	S 0.00-0.40 versus T middle	0.98	0.87
Labiotermes	S 0.00-0.40 versus T top	0.99	0.86
	S 0.00-0.20 versus T middle	0.99	0.96
PI	S 0.20-0.40 versus T middle	0.99	0.93
Syntermes	S 0.00-0.20 versus T top	0.96	0.95
	S 0.20-0.40 versus T top	0.98	0.94

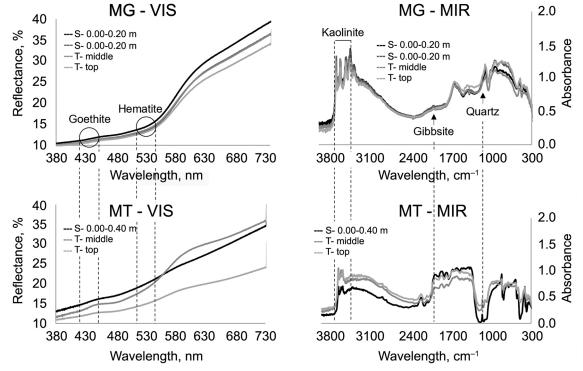


Figure 6 – Visible (VIS) and mid-infrared (MIR) spectra of fine-earth fraction of soil (S) and termite mound (T) materials in the Minas Gerais (MG-Cornitermes) and Mato Grosso (MT-Cornitermes) environments.

possible changes in the materials of the termite mound compared to the soil materials, mainly in the Mato Grosso, Rio Grande do Sul-JC and Pará environments (Table 3).

The analysis of the proportion between hematite and goethite [Hm/(Hm+Gt)] showed an expressive range of values, varying between 0.07 in the soil of the Piauí environment and 0.69 in the soil of the Minas Gerais environment (Table 4). The soil classification varied between goethitic soils [Hm/(Hm+Gt) < 25 %] and hematitic-goethitic soils [50 < Hm/(Hm+Gt) ≤ 75 %] (Ramos et al., 2020), showing the pedoenvironmental diversity between soils in different locations (Bigham et al., 2002). However, there were no significant changes in the values of the Hm/(Hm+Gt) ratio between soils and termite mounds in different environments, which are, therefore, in agreement with the high values of R<sup>2</sup> obtained in the correlations of the visible band (where this relationship is determined) indicating that the proportion of these iron oxides was not affected by the passage in the digestive tract of the termites.

The parameters hue (H), value (V) and chroma (C); the redness rating (RR); and the RGB combination indicated different intensities of color change between the control soils and the termite mounds (Table 5). Based on these parameters, the soil and termite mound materials in the Rio Grande do Sul-ES and Minas Gerais environments, were similar, possibly because they were more clayey soils (Sarcinelli et al., 2013), causing color

**Table 4** – Ratio hematite/(hematite+goethite) (Hm/(Hm+Gt)) and classification of the sample according to this proportion in the soil (S) and termite mound (T) materials in the Rio Grande do Sul (RS-JC), Rio Grande do Sul (RS-ES), Minas Gerais (MG), Piauí (PI), Mato Grosso (MT) and Pará (PA) environments.

Environment/ specimen	Sample	Hm/(Hm+Gt)	Classification <sup>1</sup>		
	S 0.00-0.20	0.55			
RS-JC	S 0.20-0.40	0.53	hematitic – goethitic		
Cortaritermes	T middle	0.59	$[50 < Hm/(Hm + Gt) \le 75 \%$		
	T top	0.59			
	S 0.00-0.20	0.48			
RS-ES	S 0.20-0.40	0.52	goethitic – hematitic		
Cornitermes	T middle	0.49	[25 < Hm/(Hm + Gt) ≤ 50 %		
	T top	0.49			
	S 0.00-0.20	0.67			
MG	S 0.20-0.40	0.69	hematitic – goethitic		
Cornitermes	T middle	0.67	$[50 < Hm/(Hm + Gt) \le 75 \%$		
	T top	0.67			
	S 0.00-0.40	0.28	11.50		
MT Cornitermes	T middle	0.30	goethitic – hematitic [25 < Hm/(Hm + Gt) ≤ 50 %		
Corniterines	T top	0.34	[23 < HIII/(HIII + GL) ≤ 30 %		
	S 0.00-0.40	0.12			
PA Labiotormos	T middle	middle 0.14 goethitic			
Labiotermes	T top	0.14	[Hm/(Hm + Gt) ≤ 25 %]		
	S 0.00-0.20	0.07			
PI	S 0.20-0.40	0.09	goethitic		
Syntermes	T middle	0.08	[Hm/(Hm + Gt) ≤ 25 %]		
	T top	0.08			

<sup>&</sup>lt;sup>1</sup>Ramos et al. (2020).

**Table 5** – Color parameters of soil (S) and termite mound (T) materials in the Rio Grande do Sul (RS-JC), Rio Grande do Sul (RS-ES), Minas Gerais (MG), Piauí (PI), Mato Grosso (MT) and Pará (PA) environments.

Environment/ specimen	Sample	Hue	Value	Chroma	$RR^1$	Combination RGB
	S 0.00-0.20	5.72YR	4.96	3.66	3.16	
RS-JC	S 0.20-0.40	5.78YR	5.02	3.77	3.17	
Cortaritermes	T middle	5.88YR	4.82	3.50	2.99	
	T top	6.56YR	4.59	2.90	2.17	
	S 0.00-0.20	7.08YR	5.22	3.78	2.11	
RS-ES	S 0.20-0.40	6.49YR	4.94	3.67	2.61	
Cornitermes	T middle	6.66YR	5.12	4.01	2.62	
	T top	6.69YR	5.07	3.91	2.55	
	S 0.00-0.20	3.69YR	4.96	4.81	6.12	
MG	S 0.20-0.40	3.55YR	4.80	4.57	6.14	
Cornitermes	T middle	3.83YR	4.82	4.51	5.77	
	T top	3.87YR	4.71	4.28	5.57	
LAT	S 0.00-0.40	8.38YR	5.26	3.09	0.95	
MT Cornitermes	T middle	6.72YR	5.29	4.02	2.49	
	T top	8.05YR	4.62	2.54	1.07	
PA Labiotermes	S 0.00-0.40	8.83YR	5.93	3.10	0.61	
	T middle	8.55YR	6.72	3.66	0.79	
	T top	8.89YR	6.13	3.17	0.57	
	S 0.00-0.20	8.86YR	6.11	3.53	0.66	
PI	S 0.20-0.40	8.98YR	6.52	4.21	0.66	
Syntermes	T middle	8.90YR	5.92	3.36	0.62	
	T top	8.85YR	6.09	3.78	0.71	

<sup>&</sup>lt;sup>1</sup>RR = Redness rating.

saturation. In the Piauí environment, small changes in the chroma indicated similarities between the materials of the soil at 0.00-0.20 m and the middle portion of the mound and between the materials of the soil at 0.20-0.40 m and the top of the mound. The similarity between the colors of the materials (soil and termite mound) within these three environments was corroborated by the high coefficients of determination of the correlations performed (Table 3).

In the Rio Grande do Sul-JC environment, the material at the top of the mound showed an increase in hue and a decrease in the value, chroma and redness rating compared to the soil materials and the middle portion of the mound. In the Mato Grosso environment, the termite mound materials showed altered color parameters compared to the control soil material, emphasizing the middle portion, where the hue decreased considerably while the chroma and redness rating increased. In the Pará environment, only the material in the middle of the mound differed in color from the material in the soil, with increases in the value, chroma and redness rating. In these last three environments, the changes observed in the color of the termite mounds in relation to the control soils were also indicated by the lower coefficients of determination of the correlations performed (Table 3).

In each environment alone, the color changes observed between the soils and the termite mounds resulted from combinations, mainly between the increase in clay fraction, organic carbon and iron oxides verified in the termite mounds, which decrease the hue and the value and increase the chroma and the redness rating. In addition, it should be considered that changes in color between the portions of the termite mounds can also be a consequence of the difference in exposure to the weather.

The results reinforce the importance of termites in soil formation, in the process of vertical and lateral bioturbation of organic and mineral particles, in nutrient cycling, the accumulation of organic matter, and in constructing structures that positively affect soil porosity and water and gas flows.

#### **Conclusions**

Termites are selective when obtaining materials from adjacent soils during the construction of mounds. This selectivity is expressed in increases in the contents of clay, total organic carbon and pedogenic iron oxides such as hematite, goethite and lepidocrocite. The selectivity for organic and mineral components of clay size increases the specific surface area of the termite mounds. For the construction of termite mounds, termites select clay-sized particles such as kaolinite, iron oxides (hematite, goethite, lepidocrocite) and aluminum (gibbsite) rather than larger particles such as quartz. In this selective process, the contents of crystalline iron oxides (hematite and goethite) increases in termite mounds, while

the magnetic susceptibility indicates a reduction in maghemite contents. No significant changes exist in the proportions of hematite and goethite (ratio Hm/(Hm+Gt)) between soils and termite mounds materials. The parameters of color hue, value and chroma; the redness rating; and the RGB combination are also sensitive to different intensities of color change between the soil and the termite nest in different environments. The change in these parameters in specific environments results from combinations of increases in clay, organic carbon and iron oxides in termite mounds.

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# **Authors' Contributions**

Conceptualization: Inda, A.V.; Barrón, V.; Fruett, T. Formal analysis: Inda, A.V.; Barrón, V.; Fruett, T.; De Sá, E.L.S.; Taha, K.; Fernandes, A.F.D. Funding acquisition: Inda, A.V.; Barrón, V. Investigation: Fruett, T.; Inda, A.V.; Barrón, V.; De Sá, E.L.S.; Taha, K.; Fernandes, A.F.D. Supervision: Inda, A.V.; Barrón, V.; Fruett, T. Writing-original draft: Inda, A.V.; Barrón, V.; Fruett, T.; De Sá, E.L.S.; Taha, K.; Fernandes, A.F.D. Writing-review & editing: Inda, A.V.; Barrón, V.; Fruett, T.

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