

## Depth dependence of black carbon structure, elemental and microbiological composition in anthropic Amazonian dark soil



Marcela C. Pagano<sup>a,\*</sup>, Jenaina Ribeiro-Soares<sup>a</sup>, Luiz G. Cançado<sup>a</sup>, Newton P.S. Falcão<sup>b</sup>,  
Vívian N. Gonçalves<sup>c</sup>, Luiz H. Rosa<sup>c</sup>, Jacqueline A. Takahashi<sup>d</sup>, Carlos A. Achete<sup>e</sup>,  
Ado Jorio<sup>a,\*</sup>

<sup>a</sup> Departamento de Física, ICEx, Universidade Federal de Minas Gerais, Avenida Antônio Carlos, 6627, Pampulha, 31.270-901 Belo Horizonte, MG, Brazil

<sup>b</sup> Departamento de Ciências Agronômicas, Instituto Nacional de Pesquisas da Amazônia, Manaus, 69011-970 AM, Brazil

<sup>c</sup> Departamento de Microbiologia, ICB, Universidade Federal de Minas Gerais, Avenida Antônio Carlos, 6627, Pampulha, 31.270-901, Belo Horizonte, MG, Brazil

<sup>d</sup> Departamento de Química, ICEx, Universidade Federal de Minas Gerais, Avenida Antônio Carlos, 6627, Pampulha, 31.270-901, Belo Horizonte, MG, Brazil

<sup>e</sup> INMETRO, Xerém, Duque de Caxias, RJ 25250-020, Brazil

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### ABSTRACT

“Terras Pretas de Índio” are anthropic Amazonian soils rich in pyrogenic black carbon, which might be responsible for the soil long-term stability and high fertility. This black carbon, produced by the Indians while handling their residues, became a model material for agriculture and environment. The key question to answer for artificially reproducing the desired agricultural properties of the *Terra Preta de Índio* is whether the black carbon structure found today in these soils is the same as produced by the ancient Indians, or whether its structure results from long-term complex physical, chemical and biological activities in the soil. To address this question, this work investigates the depth dependence of the properties from a soil collected from the Balbina site, in Presidente Figueiredo, Amazonas State, Brazil. The black carbon structure and the soil composition are investigated, with special emphasis on the poorly studied microbiological composition (fungi, bacteria, arbuscular mycorrhizas). The comparative analysis between the properties from shallower (newer) and deeper (older) soil strata indicates that, while soil composition exhibits depth dependence, the pyrogenic black carbon structure does not. This finding suggests that this model material should be reproducible by repeating the pyrolysis conditions utilized in their production.

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## 1. Introduction

Understanding the characteristics of the Amazonian Dark soils “Terras Pretas de Índio” (TPIs) is of ecological importance, because the TPI soils represent a residue-based model for tropical sustainable agriculture (Sombroek et al., 2003; Neves et al., 2003; Cohen-Ofri et al., 2006, 2007; Falcão et al., 2003; Glaser, 2007). The TPI sites are identified by the presence of ceramics and by their deep black horizons, generally down to 1 m in depth (Fraser et al., 2011). The dark color comes from the high content of black carbon (BC), which are here defined as stable charcoal particles present in the terrestrial ecosystems (Lian et al., 2006; Liang et al., 2008). In the case of the TPI soil, the black carbon is pyrogenic,

produced by the Indians when burning residues. Almost no black carbon or ceramics are detectable in immediately surrounding soils, already below 20 cm in soil depth (Glaser et al., 2001). The TPI soils sequester up to 70 times more carbon than the surrounding soils, and it is chemically and microbially stable (Glaser et al., 2001; Steiner et al., 2004). Consequently, the microbial community in the TPI is different from those in the surrounding soils (Grossman et al., 2010; Glaser and Birk, 2012).

The TPI pyrogenic black carbon (TPI-BC) is made of sp<sup>2</sup>-ordered carbon nanocrystallites with lateral dimensions of  $L_a \sim 3\text{--}8\text{ nm}$  (Jorio et al., 2012; Ribeiro-Soares et al., 2013). Despite a structural complexity, the TPI-BC dimensionality ( $L_a$ ) has been investigated as a critical parameter defining the stability vs. reactivity properties of the soil (Jorio et al., 2012; Ribeiro-Soares et al., 2013; Archanjo et al., 2014, 2015). To be able to reproduce charcoal structures similar to the TPI-BC, for the use as a soil conditioner, it is necessary to understand whether the TPI-BC structure (mostly  $L_a$ ) found today was generated by the char and burning, or whether the time

\* Corresponding authors. Fax: +55 31 3409 5600.

E-mail addresses: [marpagano@gmail.com](mailto:marpagano@gmail.com) (M.C. Pagano), [adojorio@fisica.ufmg.br](mailto:adojorio@fisica.ufmg.br) (A. Jorio).

exposure to complex physical, chemical and biological degradation played a role. Some authors are discussing how the microbial communities inhabiting the soil may influence its structure (Kabir et al., 1998; Oehl et al., 2005; Talbot et al., 2008; Eilers et al., 2012). While the elemental composition of the TPI has been largely studied (Glaser and Birk, 2012; Schaefer et al., 2004; Kern and Kampf, 2005; Cunha et al., 2009), few works addressed the microbiological composition (Grossman et al., 2010; Glaser and Birk, 2012).

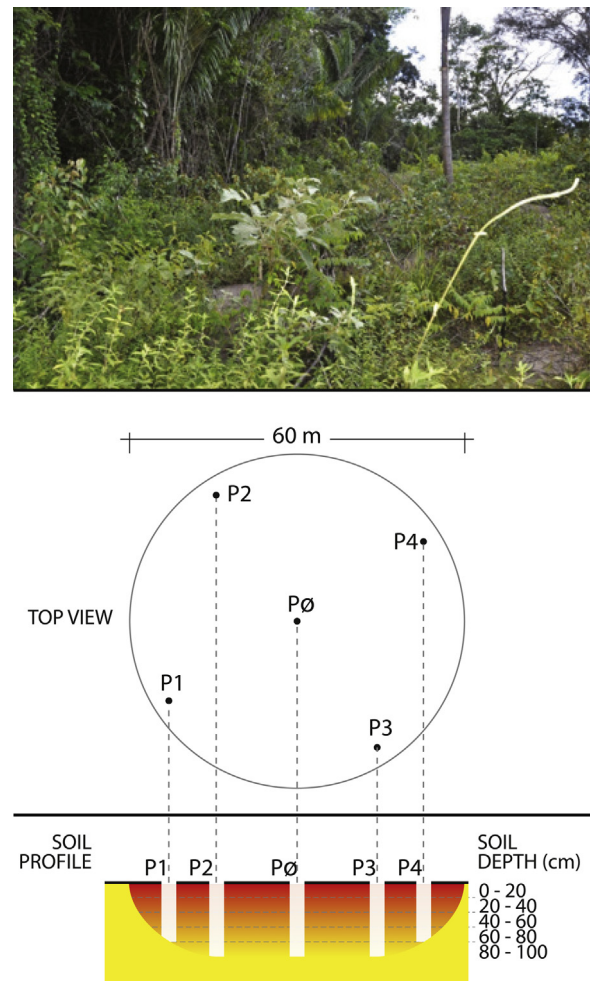
The primary objective of this work is to detect changes in the critical parameter  $L_a$  of TPI-BC as a function of soil depth, crossing this information with the elemental and microbiological composition, from the surface down to 100 cm in depth. Estimates point to a TPI formation rate of 1 cm for every 10 years of Indian occupation (Smith, 1980). The comparative analysis among the black carbon properties from shallower (newer) and deeper (older) soil strata should provide information about the stability of their structures. The elemental composition of the soils has been measured and the microbial occurrence evaluated to test the influence of chemical elements and microorganisms as potential charcoal degradation agents. Due to the lack of information about the microbiological composition of the TPI soils, this part is discussed here in more details. The types of microorganisms present in TPI and the abundance of the associated fungi in the surface and deeper soil strata are determined. The central hypothesis of this work is that, despite differences in (1) the time permanence in the soil, (2) the elemental and (3) the microbiological soil compositions, the as produced TPI-BC maintained their basic structural properties, represented by a unique distribution of  $L_a$  values.

## 2. Material and methods

### 2.1. Samples

Individual TPI soil samples of about 0.5 kg were collected from the Balbina site, in Presidente Figueiredo (Lat. 2°09'39"S, Long 60°00'W), altitude 60 m, at 180 km from Manaus, Amazonas State, Brazil. The samples were collected in October 2011, and preserved in plastic sleeves until processed and subsampled for various biological, physical and chemical analyses. Five points within the 60 m diameter circular TPI site were selected for sampling the soil composition, as sketched in Fig. 1—four points located near the site boundaries, and one point at the central region. After removal of the litter layer, these points were probed at various depths: 0–20 cm, 20–40 cm, 40–60 cm, 60–80 cm for all sites, plus a 80–100 cm stratum for points P0, P2 and P3. Textural composition of the TPI Balbina soils is sandy, with texture independent of soil depth. The structural and microbiological analyses were performed on each sample separately, although the information presented here will be a statistical mixture among the points with same depth. For elemental analysis, however, the samples from the same depth were mixed because the amount of required material is larger for such analysis.

Control soil samples (about 0.5 kg) were collected in surrounding soils, which are predominant in the region: (i) Ultisol (USDA), under secondary rain forest vegetation or "capoeira", 30 years old, with sandy texture; (2) the immediately surrounding Oxisol, under primary forest, with a clayey texture. The ultisol and oxisol were collected at 0–20 cm and 20–40 cm strata depths only, since black carbon is not found for deeper strata. Other types of samples were analyzed for comparison, originating from: (i) native forest soil (riparian site, 0–20 cm depth) from southeastern Brazil (Sabará, Minas Gerais State, 735 m a.s.l.); (ii) turf (accumulation of decayed vegetation and other organic material), from a farm in Minas Gerais State, Brazil, from 0 to 20 cm soil depth; (iii) compost (leaves, stems, grasses and animal feces), collected from the Federal



**Fig. 1.** Relative locations of the samples within the site of "Terra Preta de Índio", in Balbina, Presidente Figueiredo, Manaus State, Brazil. Five points were sampled, one in the center (P0) and four at the boundaries (P1–P4). For each point, samples were taken every 20 cm with a Dutch auger, down to 80 cm in depth for P1 and P4, and down to 100 cm in depth for the other points.

University of Minas Gerais-Campus, after 6 months of maturity; (iv) peat samples collected from the Federal University of Minas Gerais-Campus, at 7 m soil depth, during a building excavation; (v) activated charcoal (Zajac and Groszek, 1997; Suhas and Ribeiro, 2007; Qiu et al., 2008; Nabais et al., 2010), and (vi) synthetic vegetal charcoal (Schulz and Glaser, 2012), obtained from Synth<sup>®</sup> (São Paulo, Brazil).

### 2.2. Characterization methods

#### 2.2.1. Elemental analysis

For elemental characterization, soil composition was obtained at Minimax<sup>®</sup> Agropecuary Laboratory, Belo Horizonte, Brazil. Phosphorus and potassium were measured according to the Mehlich I method. Organic carbon content (colorimetric method) was measured according to (EMBRAPA, 1997).

#### 2.2.2. Structural analysis

Structural characterization of the pyrogenic TPI-BC was based on Raman spectroscopy, as discussed in Ribeiro-Soares et al. (2013). The micro-Raman scattering experiments were performed with an Andor<sup>®</sup> Technology-Sharmrock sr-303i spectrometer (2 cm<sup>-1</sup> spectral resolution), coupled to a charge-coupled device (CCD) detector. The backscattering configuration was used, with a

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