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Soil biota shift with land use change from pristine rainforest and Savannah (Cerrado) to agriculture in southern Amazonia

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Abstract

Southern Amazonia is currently experiencing extensive land use change from forests to agriculture caused by increased local and global demand for agricultural products. However, little is known about the impacts of deforestation and land use change on soil biota. We investigated two regions in southern Amazonia (rainforest and Savannah/Cerrado biomes), analysing soil biota community turnover based on 16S (Archaea and Bacteria) and 18S rRNA genes (Eukaryotes, including Fungi, Protists and Animalia) and correlating them with soil chemistry and land use intensity. We found that soil biota community structure is driven by land use change in both Cerrado and rainforest. Crop fields approximatively doubled the richness of soil Archaea, Bacteria and Protists. We propose that crop systems not only increase soil pH and fertility, but also create continued disturbance (crop seasons) that stimulates soil diversity, as predicted by the dynamic equilibrium model (DEM) and the intermediate disturbance hypothesis (IDH). Even though agricultural fields had higher soil biota richness, some taxa were suppressed by agriculture (6/31 operational taxonomic units of Archaea, 245/1790 of Bacteria, 12/74 of Animalia, 20/144 of Fungi and 25/310 of Protists). Consequently, land use change in this region should proceed with caution. In the southern Amazonia region of Brazil, current laws require farmers to keep 20%–80% pristine vegetation areas on their property. Our data support the relevance of this law: since there are unique soil taxa under native vegetation, keeping these pristine areas adjacent to the agricultural fields should maximize soil biodiversity protection in these regions.

KEYWORDS
agriculture, Amazonia, bacteria, land use change, microbial community structure, Protists

1 | INTRODUCTION

Land use change from native vegetation to agriculture is an important concern in a globally changing world. Land use change originated with the transition from nomadic human populations to modern societies, with this shift pointing to the beginning of the Anthropocene (Huston, 2005; Pongratz et al., 2008). Today, global population growth and concentrations in big cities and densely populated areas have stimulated agricultural expansion worldwide to meet an increasing demand for food. Several countries worldwide cannot support their own needs for food and depend on imports, especially in Asia and Europe (Schramski et al., 2019). Population growth and land

use change have created great challenges, from impacts on biodiversity to greenhouse gas emissions, including those stemming from transport of agricultural products and their supply chain (Huston, 2005; Pongratz et al., 2008). Ecological theories reflect how human activities alter plant and animal communities, giving rise to concepts such as habitat fragmentation, extinction debt and disturbance (Miller et al., 2011; Veresoglou et al., 2015; Wearn et al., 2012).

However, these theories are challenging to apply to soil microbial communities (Thakur et al., 2020). Soil microorganisms not only face high-frequency fluctuations in environmental settings, such as temperature, pH, moisture, and availability of nutrients and energy, but also in the types of resources they consume (Fierer et al., 2009; Lammel et al., 2015; Thakur et al., 2020). Microorganisms live in very heterogeneous habitats, at a scale of pores that can vary from <1 μm to >1 mm, and at a scale of micro- (<250 μm) and macro-aggregates (>250 μm). Consequently, soil structure also affects the microorganisms' habitats at fine scale; for example, micropores can protect microorganisms from predation by larger organisms (Baveye et al., 2018). In addition, environmental changes at this microscale can be more drastic than at the macroscale (landscape). While diurnal and daily variations in water and temperature at the soil surface might have minimal impacts on plants, they could drastically affect microbial microhabitats near the soil surface and consequently impact soil microbial communities (Lammel et al., 2015; Nacke et al., 2014).

Methodologically, all these changes are usually analysed at the soil core sampling scale, rarely at the soil aggregate or microscale, thus generally assuming that: (i) the soil core scale represents the heterogeneity of the microscale (mainly because soil biota dispersion is expected to be high at the core scale); (ii) the core scale survey is representative of the landscape scale; and (iii) the survey represents the viable soil biota community at that sampling time assuming a reduced potential bias of dormant and dead taxa and extracellular DNA (Thakur et al., 2020).

Consequently, even though deforestation causes habitat fragmentation at the landscape scale by creating plant cover patches, the conversion to agriculture may cause habitat homogenization at the microscale by homogenizing the soil matrix (Rodrigues et al., 2013). Despite the loss of native plant cover, soil microhabitats remain, but are physically and chemically homogenized by agricultural practices. While under native vegetation the heterogeneous plant cover increases spatial variation in soil nutrient, water and energy fluxes, in agricultural systems the plant cover is more homogeneous and should increase homogenization of resource inputs for soil biota (Lammel, Nüsslein, et al., 2015; Rodrigues et al., 2013). Harvest events, in particular, can increase below-ground productivity, by adding massive amounts of litter, especially related to root decomposition (Johnson et al., 2006). Environmental fluctuations are more frequent in agricultural fields because of farming practices (disturbances) such as sowing, crop rotation, use of fertilizers and pesticides, harvesting and the associated decomposition of the above-ground litter and the roots, as well as animal traffic and manure in pastures (Lammel, Nüsslein, et al., 2015; Smith et al., 2016). Deforestation also changes the exposure of the soil surface to solar radiation, and thus shifts soil temperature and water content, due to reduction of the plant canopy (Lammel, Nüsslein, et al., 2015; Nacke et al., 2014). Therefore, land use change from native vegetation to agriculture simultaneously affects several factors that could influence soil biota community turnover, including addition/shift of resources (litter, especially from crop roots, and fertilizers) and disturbances (agricultural management practices, such as sowing, tillage and harvest events, pesticide application and crop rotation).

The dynamic equilibrium model (DEM) predicts that certain combinations of productivity and disturbance maximize species diversity, including low productivity and low disturbance, intermediate productivity and intermediate disturbance, and high productivity and high disturbance (Huston, 2014). A vast literature indicates that an intermediate disturbance and productivity maximizes species diversity and abundances, which is summarized by the intermediate disturbance hypothesis (IDH) (Connell, 1978; Huston, 2014; Miller et al., 2011). The IDH has been recently used to explain community assemblages of microorganisms at the microcosm scale, suggesting that extreme disturbances favour deterministic processes, while intermediate disturbances favour stochastic processes and higher diversity (Santillan et al., 2019). A main challenge of both the DEM and IDH is that they are difficult to test in the field, because it is hard to quantify disturbance and also because disturbance gradients are often confounded with parallel productivity gradients; thus, some researchers contend that this hypothesis may be untestable (e.g., discussion between Fox, 2013 and Huston, 2014). However, recent studies have suggested that the role of environmental fluctuation, including changes in resources (soil fertility) and disturbance (agricultural practices) within the IDH framework, should be taken into account to better understand the impacts of land use change on soil microbial community structure (Brinkmann et al., 2019; George et al., 2019).

In recent decades, the region of southern Amazonia has been the most dynamic agricultural frontier worldwide. This region is located in the transition zone from the Savannah biome (Cerrado is a biodiversity hotspot) to the rainforest biome (for details, see "Study area description"). Since the 1980s, Brazilian laws in this region have required farmers to preserve 20%–80% of the native vegetation on their farms (details in FAOLEX, 2019), allowing a unique opportunity to study soils from agricultural fields in parallel with the adjacent native vegetation. Previous studies in this region showed that soil microbial community structure shifted with land use change and was largely correlated with soil fertility changes (pH and nutrients, such as N, P, K, Ca, Mg and trace elements). Native vegetation on Oxisols (Ferralsols Latosols) in this region (the predominant soil type in southern Amazonia) is characterized by usually homogenous low soil pH and low fertility, while agricultural fields always have increased soil pH and fertility (Carvalho et al., 2007; Lammel, Feigl, et al., 2015; Lammel, Nüsslein, et al., 2015; Merloti et al., 2019; Pedrinho et al., 2019). Therefore, all correlations of soil biota comparing native vegetation soils to the very distinct agricultural soils will generally yield statistically significant correlations that should be interpreted with caution, because in this case, soil fertility covaries with other parallel

changes caused by land use change (Lammel et al., 2018; Lammel, Nüsslein, et al., 2015). Consequently, we suggest that ecological theories other than just deterministic processes caused by soil fertility should be taken into account to explain the microbial community shifts in this region, which we examine here in relation to the DEM and IDH.

We use here two different soil surveys in the southern Amazonia region to assess soil biota community structure (Archaea, Bacteria, Fungi, Protists and Animalia) and to relate these communities to soil chemistry, land use types and intensity (native vegetation, pasture and crops). One survey focused on the effects of land use change on soil biota, comparing rainforest and Savannah (Cerrado) biomes. The other survey investigated seasonal changes in the rainforest region. Based on the DEM and IDH, we hypothesized that in agricultural soils disturbances coupled with increased resource availability increase soil biotic diversity. We assumed that land use change can be disadvantageous for some soil species that were abundant under native vegetation and allows rare species to thrive in the new environment. Furthermore, we hypothesized that land use intensity is the most important driver of soil diversity in two different contexts: across spatial and short-term scales in South Amazonia. Namely, we tested if land use intensity was the most important driver for the first soil survey comparing the rainforest and Cerrado regions. In the second survey we investigated both land use intensity and seasonal change as drivers of soil biotic structure.

2 | MATERIAL AND METHODS

2.1 | Study area description and design

Land use change in the Southern Amazonia region occurs predominantly from native vegetation to extensive pastures or to crop fields (Galford et al., 2010). In the last three decades, land use change in this region has transformed the Brazilian state of Mato Grosso into the largest agricultural area in Brazil. In this state, native rainforest is estimated at 350,000 km^2 and savannah at 210,000 km^2, while pastures occupy 190,000 km^2 and crop fields cover 110,000 km^2 (Galford et al.,2010). Two agricultural hot spots in Mato Grosso state were previously chosen for soil surveys: one in the ecoregion of the Cerrado in the municipality of Campo Verde, and another in the rainforest in the municipality of Sinop (Lammel, Feigl, et al., 2015; Lammel, Nüsslein, et al., 2015) (Figure S1).

Three major changes occur simultaneously with land use change by deforestation from native vegetation to agriculture, namely shifts in plant cover, in soil chemistry and in the intensity of disturbance. In pastures, the natural vegetation is mainly replaced by the grass *Urochloa brizantha* (Hochst., syn. *Brachiaria brizantha* A. Rich.), a dominant species that covers the soil perennially. For crop fields, no-till systems and double cropping are commonly used. In double cropping, two crops are sown and harvested in succession within the same agricultural year, usually first soybean is cultivated followed by different crop grasses such as corn, sorghum or millet (Carvalho et al., 2007; Lammel, Nüsslein, et al., 2015). In terms of soil fertility, the major changes are increases in pH (from 3.8 to 5.5–6) and in mineral nutrient concentrations (mainly increases in P, Ca, Mg, K and micronutrients). Soybean annual fertilization requires around 250 kg ha^{-1} of N–P_2O_5–K_2O 00–20–20 (triple superphosphate 33%, single superphosphate 33%, and KCl 34%, w/w), inoculation with *Bradyrhizobium* for N-fixation, and periodic liming (Ca and Mg carbonates) to elevate the soil pH to around 5.5. Pastures rarely receive annual fertilization (usually single superphosphate) or liming (for more information, see Lammel et al., 2017; Lammel, Feigl, et al., 2015; Lammel, Nüsslein, et al., 2015). Concerning disturbances in relation to the native soils, pastures experience greater heating at the soil surface and the impacts of cattle such as soil compaction, intensive grazing and manure (Lammel, Feigl, et al., 2015). Crop fields have the highest intensity of disturbance owing to their dynamic management with double cropping and crop rotation, resulting in increased decomposition of plant harvest residues and roots, and because they receive applications of herbicides, insecticides and fungicides (Carvalho et al., 2007; Lammel, Nüsslein, et al., 2015).

In terms of the DEM/IDH, we assumed that it is reasonable to consider all these disturbance factors together, forming an overall gradient of low disturbance context in pristine vegetation and increased disturbances in agricultural land uses, being intermediate in pastures and highest in the intensive double-crop systems.

2.2 | Survey 1—Land use change from rainforest or Cerrado to agricultural use

This soil survey was carried out in January 2009 (midsummer season) in both biomes: tropical rainforest (UTM S120553.3 W552846.0) and Cerrado (UTM S151588.8 W550700.0) (Lammel, Nüsslein, et al., 2015). Within the Savannah and rainforest biomes, two vegetation types are predominant in this region, the "Cerrado *sensu stricto*" (Savannah biome) and the "Semideciduous forest" (rainforest biome). Even though both regions have a tropical climate (Aw, in Köppen classification) with high annual precipitation (~2,000 mm), the dry winter and fire occurrences are more accentuated in the savannah region, resulting in very distinct habitats (Sanches et al., 2008; Simon et al., 2009). The Cerrado has a less diverse vegetation and lower litter deposition, but with higher abundance of recalcitrant compounds, such as lignin (Lammel, Nüsslein, et al., 2015). The semideciduous rainforest has higher plant diversity, including species that occur in the Amazon rainforest, and higher litter deposition (Sanches et al., 2008). Despite the differences in litter deposition, the soil carbon content and overall soil fertility are very similar in both vegetation types (Lammel, Nüsslein, et al., 2015). It is assumed that soil organic matter (SOM) turnover is more intense in the forest region than in the Cerrado region because of higher litter deposition rates combined with higher decomposition rates (Lammel, Nüsslein, et al., 2015).

Soil samples were taken from three replicated sites in each of the three different land use types: native vegetation, crop fields/

soybeans and pastures located in the two biomes (Figure S1). The agricultural sites were established by deforestation more than 20 years before the survey (Lammel, Nüsslein, et al., 2015 and Lammel, Feigl, et al., 2015).

The sampling design was completely randomized, with three replicate sites for each of the three land use types in each region. Replicate sites were chosen to have comparable topographic and edaphic properties. Soils were identified as Red Oxisols with clay texture (49%–53% of clay) in both biome-regions. For each of the three replicate sites, five soil samples were randomly collected within an area covering 1 ha. The replicate sites were hundreds of metres distant from one another. Sampling areas characterizing a particular land use type were adjacent within hundreds of metres to pristine rainforest, and also close to each other in the Cerrado biome (<2 km). Individual soil samples were taken from 0–17 cm soil depth using 5-cm-diameter PVC tubes (Lammel, Nüsslein, et al., 2015). All sampling equipment was disinfected with 70% ethanol, and care was taken to avoid contamination from surrounding materials. Soil cores were frozen on the day of collection and stored at −20°C. Later, DNA was extracted from each soil sample, which included five samples in three replicate sites, for a total of 15 extractions per land use in each biome. DNA samples were pooled in equal volumes for each replicated site.

2.3 | Survey 2—Temporal survey (rainforest only)

Soil samples from the rainforest area (described in Survey 1) were sampled in two different seasons (Lammel, Feigl, et al., 2015). The first sampling was in the first week of November 2010 at the beginning of the rainy season (spring) and just prior to soybean sowing. Samples were taken a second time in January 2011 in the middle of the rainy season (summer) and immediately after soybean flowering, termed stage R3 (Lammel, Feigl, et al., 2015). In addition to the sampling sites described in Soil Survey 1, in this survey there was an additional new crop site established after deforestation only 3 years before the soil survey (first year cultivated with rice, followed by 2 years of soybean; Lammel, Feigl, et al., 2015). Soil was sampled from two to three replicated sites for each land use type. In each of the replicated sites, five soil samples were taken and DNA was extracted from each individual sample. DNA samples were pooled in equal volumes for each replicated site.

2.4 | Soil analysis and amplicon sequencing

Soil and litter chemical data were obtained from previous studies by the authors using the same fields and sampling time (Lammel, Nüsslein, et al., 2015 and Lammel, Feigl, et al., 2015). Litter data are only available for the 2009 survey (Survey 1) and soil microelements only for the 2010/11 survey (Survey 2) (Table S1).

Soil DNA was extracted from 0.25 g from each sample using the PowerSoil DNA Isolation Kit (Mobio). Polymerase chain reaction (PCR) amplification and microbial diversity analysis were performed based on the Earth Microbiome guidelines for 16S and 18S rRNA genes (www.earthmicrobiome.org/protocols-and-standards), with the following modifications: (i) we reduced the number of cycles to 25 to reduce the potential formation of chimeras, (ii) we used the proofreading Kapa Hi Fi Polymerase (Kapa) instead of the Platinum mix (Thermo Scientific), and (iii) we used a dual PCR indexing protocol based on Illumina P5/P7 adapters to index the samples during sequencing (Illumina). No amplification was observed in the negative controls (see also Appendix S1 "Quality Control").

Following PCR amplification, the samples were purified with the magnetic beads kit NucleoMag NGS kit, and subsequently subjected to PCR indexing. The samples were then quantified using PicoGreen, pooled in equal masses of DNA, and submitted to sequencing. The 16S rRNA genes were sequenced with Illumina MiSeq kit V2 (2× 150 bp) and Illumina MiSeq HiSeq X (2× 150 bp), and the 18S rRNA genes were sequenced with Illumina MiSeq kit V3 (2× 300 bp). The 16S rRNA fastq files generated by the Illumina software were assembled using the FLASH software (Magoč & Salzberg, 2011) and 18S rRNA fastq files were directly exported for further analysis. 16S rRNA gene sequencing yielded 600–246,333 sequences per sample and 18S rRNA gene sequencing yielded 24,459–117,308 sequences per sample. The sequences were deposited in the NCBI bioproject PRJNA587110.

The primers used in this study are known to comprehensively access Bacteria and Protists, while also offering significant insights for Archaea, Fungi and Animalia (Amaral-Zettler et al., 2009; Caporaso et al., 2012).

2.5 | Sequence analysis

Sequence files were analysed with the QIIME2 pipeline (Bokulich et al., 2018). First, sequences were quality filtered and assigned to amplicon sequence variants (ASVs) using DADA2 (Bokulich et al., 2018). The ASVs were further clustered with a cut-off of 98% to avoid overestimating operational taxonomic unit (OTU) richness, since a single cell of an organism can contain rRNA copy variants (Parks et al., 2019) and to mitigate potential errors due to the sequencing process (details in Appendix S1 "Quality Control"). Later, taxonomy of the OTUs was assigned based on the Naïve-Bayes machine learning algorithm using the standard setup in QIIME2 and the Silva 132 database (Quast et al., 2013). The OTU tables were then aggregated (merged) according to the Silva taxonomy in level D7 (species) for 16S rRNA data and in level D14 (species) for the 18S rRNA data and exported for further analysis in R. Samples that had fewer than 1,000 reads were filtered out from future analysis. The 16S rRNA data were filtered and nonprokaryotic, chloroplast and mitochondrial sequences were removed from the OTU table. The 18S rRNA data were filtered by removing prokaryote and plant sequences from the table. To avoid the overestimation of richness due variation in library size, data were then normalized by resampling with replacement of the OTU from the first quartile of the read numbers, following a previously described approach (Veresoglou et al., 2019). For the 16S rRNA primer,

sequences were resampled to 34,310 reads per sample, and for the 18S rRNA primer to 12,400 reads per sample.

2.6 | Statistical analysis

Statistical analysis was performed in R (R Core Project, 2019) and multivariate analyses were performed with the R package VEGAN (Oksanen et al., 2019). Soil chemical data were analysed by analysis of variance (ANOVA) and Tukey's *post hoc* test to test differences between the soil parameters among land use types and by a principal components analysis (PCA) (Figure S2). We generated a transformation-based redundancy analysis (tb-RDA) for each taxonomic group based on Hellinger-transformed data (to reduce the impact of zero and rare OTUs) and constrained the tb-RDA to land use type and soil parameters which were the two first axes of the soil PCA, and region for survey 1, or sampling time for survey 2 (Legendre & Gallagher, 2001). The results were tested for correlation with individual soil chemistry parameters, litter composition, and land use intensity with the VEGAN-ENVFIT function (Oksanen et al., 2019). Land use intensity was considered as an ordinal variable with the values "1" to Forest, "2" to Pasture and "3" to crop fields (following the rationale in section 2.1). We further used permutational multivariate analysis of variance (perMANOVA) to test the effect of land use types and regions on microbial community structures. Hellinger transformation and variation partitioning (VEGAN "varpart") was applied to estimate the effect of the environmental parameters on the soil biota community compositions (Legendre, 2008).

Later, due to the compositional characteristics of the abundance data, the number of OTUs was constrained to the number of sequences obtained, and OTU tables were centred log-ratio (clr) transformed and plotted following the conversions of native vegetation to agriculture (Fernandes et al., 2014). We then tested the shift of each OTU abundance for each conversion using the compositional approach ALDEX2 in R, which was set based on the clr transformation, and used a generalized linear model (glm) with Gaussian family and further correction of the *p*-values by the Benjamini–Hochberg false discovery rate (FDR) (Fernandes et al., 2014). We also generated a heatmap with those 100 OTUs that had the highest variation of abundance between native and agricultural sites across all samples.

3 | RESULTS

Land use change from native vegetation to agriculture clearly affected soil chemistry (Table S1, Figure S2) and microbial community structure (Figures 1–4). Native vegetation had a low pH (3.8–3.9) and low fertility (P 2–4 mg dm^{-3}, Ca+Mg+K < 9 mmol c. dm^{-3}), while agricultural soils always had increased pH values (4.5–5.2) and increased fertility conditions (P 2–55 mg dm^{-3}, Ca+Mg+K 21–43 mmol c. dm^{-3}) (Table S1). Microbial community structure was mainly driven by land use types (Figures 1–3 and Table 1) and will be described below (also detailed in Figures S3–S9).

3.1 | Survey 1—Land use change in rainforest and Cerrado to agriculture

In the first survey, we compared land use change from native vegetation (rainforest or Cerrado) to pasture and crop fields. In the tbRDA, the prokaryotic community structure was clustered according to its soil origin (biome) and also according to land use (Figure 1a). This result was in agreement with the perMANOVA results which yielded strong support for land use type (Tables 1, $R^2 = .60$, $p < .01$) and weaker support for region ($R^2 = .08$, $p < .01$). In the tbRDA for prokaryotes the native vegetation samples from rainforest and Cerrado clustered in distinct quadrants, while pasture and crop samples clustered together in distinct quadrants for each biome (Figure 1a). For the eukaryotic communities, native vegetation soils also clustered separately from agricultural soils (Figure 1b), a result that was supported by the perMANOVA test for land use type ($R^2 = .38$, $p < .01$) but less so for sampling time ($R^2 = .09$, $p < .01$). The biome region also affected clustering of the crop fields and they did not cluster together, but did by biome. On the other hand, the biome region had no effect on differentiating the native soils or the pasture fields which clustered together by land use type, but not by biome region. Both microbial communities, prokaryotic or eukaryotic, were correlated with the distinct soil chemistry of the land uses (Table 1). Variation partitioning yielded similar results, indicating land use type to be an important factor discriminating the biota communities (Table 1; Figure S3). The variables with the highest correlations with the microbial community structure were land use intensity ($R^2 = .90$ for eukaryotes and $R^2 = .91$ for prokaryotes), pH ($R^2 = .83$ for eukaryotes and $R^2 = .93$ for prokaryotes), and soluble Al ($R^2 = .80$ for eukaryotes and $R^2 = .90$ for prokaryotes), which is an indirect effect of pH, since Al solubility is pH-dependent and the agricultural soils received lime (Ca and Mg carbonates), precipitating the Al (Lammel et al., 2018), and Ca, which is also a covariable with pH and a consequence of the lime application ($R^2 = .83$ for eukaryotes and $R^2 = .77$ for prokaryotes).

We further investigated the effects of land use on the differential abundance of microbial community compositions, also termed microbial community turnover. The conversion from forest to pasture statistically affected 102 (out of 844, 12%) OTUs, the conversion from forest to crops affected 164 (out of 1,393, 12%) OTUs, the conversion from Cerrado to pasture affected 229 (out of 1,112, 21%) OTUs, while Cerrado to crops affected 305 (out of 1,165, 26%) OTUs (Figure 1c1-2 and d1-2). Note that OTUs plotted with maximum negative values on the clr scale indicate that these OTUs fall within the range of our detection limit (Figure 1).

3.2 | Survey 2—Temporal survey in the land uses in rainforest

In the second soil survey we analysed temporal surveys in the rainforest area, and analysed shifts in microbial community structures in response to land use change from native rainforest to pasture

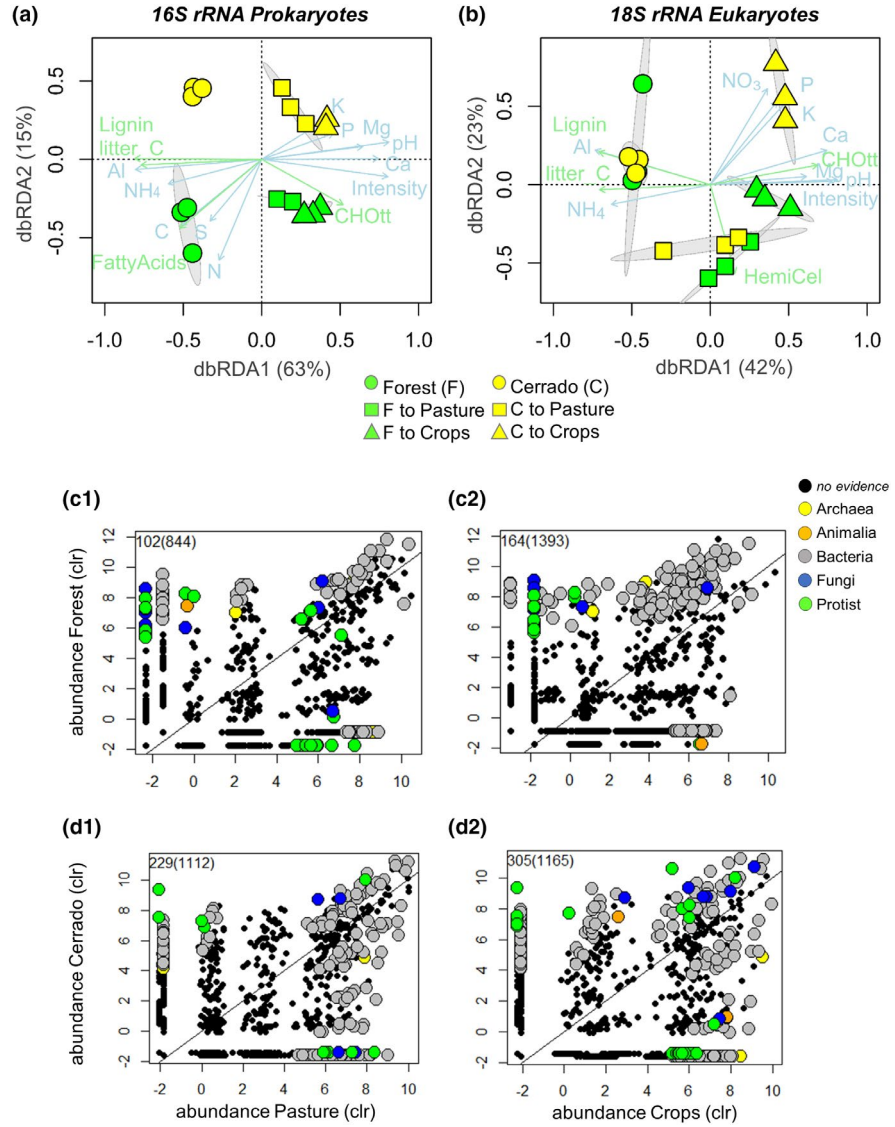

FIGURE 1 Effect of the land use change from native vegetation to agriculture in two regions (rainforest and Cerrado) on the soil biota (Survey 1). The graphs on the top indicate the changes in the community structures assessed by a transformation-based redundancy analysis (tbRDA) for the genes (a) 16S rRNA (Prokaryotes) and (b) 18S rRNA (Eukaryotes) (additional details by taxonomic groups in Figures S2–S6). Ellipses surround data for microbial communities that did not differ from each other (represent 99% confidence intervals, *SE*). The graphs below represent the community shifts from native forest to (c1) pasture and (c2) crops; and from native Cerrado to (d1) pasture and (d2) crops (each dot represents an OTU and significant changes were accessed by ALDEX2, $p < .05$, and coloured according to the taxa; the abundance data were centred log-ratio transformed, clr). Abbreviations in the tbRDA: *rCN*, CN ratio; *FA*, fatty acids; *IN*, litter N; *IC*, litter C; *Intensity*, land use intensity; *HCel*, hemicellulose; and *CHOtt*, carbohydrates (details in Table 1)

and crop fields. In the tbRDA, the prokaryotic community structures clustered according to both sampling time (perMANOVA $R^2 = .11$, $p < .01$) and land use ($R^2 = .66$, $p < .01$) (Figure 2a, Table 1). For the eukaryotic communities, a similar pattern was observed, and community structure was affected by sampling time (perMANOVA $R^2 = .16$, $p < .01$) and also by land use ($R^2 = .34$, $p < .01$) (Figure 2a, Table 1). Both microbial communities, prokaryotic and eukaryotic, were correlated with the distinct soil chemistry of the land uses (Table 1) and overall similar patterns to the first survey were observed. The variables with the highest correlation ($p < .01$) to microbial community structures were land use intensity ($R^2 = .56$ for eukaryotes and $R^2 = .95$ for prokaryotes), N ($R^2 = .68$ for eukaryotes and $R^2 = .83$ for prokaryotes) and pH ($R^2 = .74$ for eukaryotes and $R^2 = .94$ for prokaryotes). Other correlations were related to indirect effects (covariables) of pH (Lammel et al., 2018), including soluble Al ($R^2 = .62$ for eukaryotes and $R^2 = .94$ for prokaryotes), Fe ($R^2 = .72$ for eukaryotes and $R^2 = .96$ for prokaryotes), Mn ($R^2 = .83$ for eukaryotes and $R^2 = .86$ for prokaryotes) and Ca ($R^2 = .91$ for prokaryotes).

We further investigated the effects of land use on microbial community turnover. In spring 2010/11, the conversion from forest to pastures affected 24 (out of 1,339, 2%) OTUs, the conversion from forest to crops (soy) 77 (out of 1,386, 6%) OTUs, and the conversion to a new soy field (soy 2y) 33 (out of 1,186, 3%) OTUs (Figure 2c1–3). In summer 2010/11, the conversion from forest to pastures affected 183 (out of 1,054, 17%) OTUs, the conversion from forest to crops (soy) 216 (from 1,276, 17%) OTUs, and the conversion to a new soy field (soy 2y) 181 (from 1,232, 15%) OTUs (Figure 2d1–3).

3.3 | Overall community changes

When both surveys were analysed according to land use types, an increase in richness (approximately double) was observed for both Prokaryotes and Eukaryotes in agricultural soils (Figure 3; Figures S4–S9). Most of the dominant high-order taxonomic groups did not have statistically significant changes; however, we detected changes (ANOVA, $p < .05$) in the Protist subgroups Apicomplexa and Cercozoa,

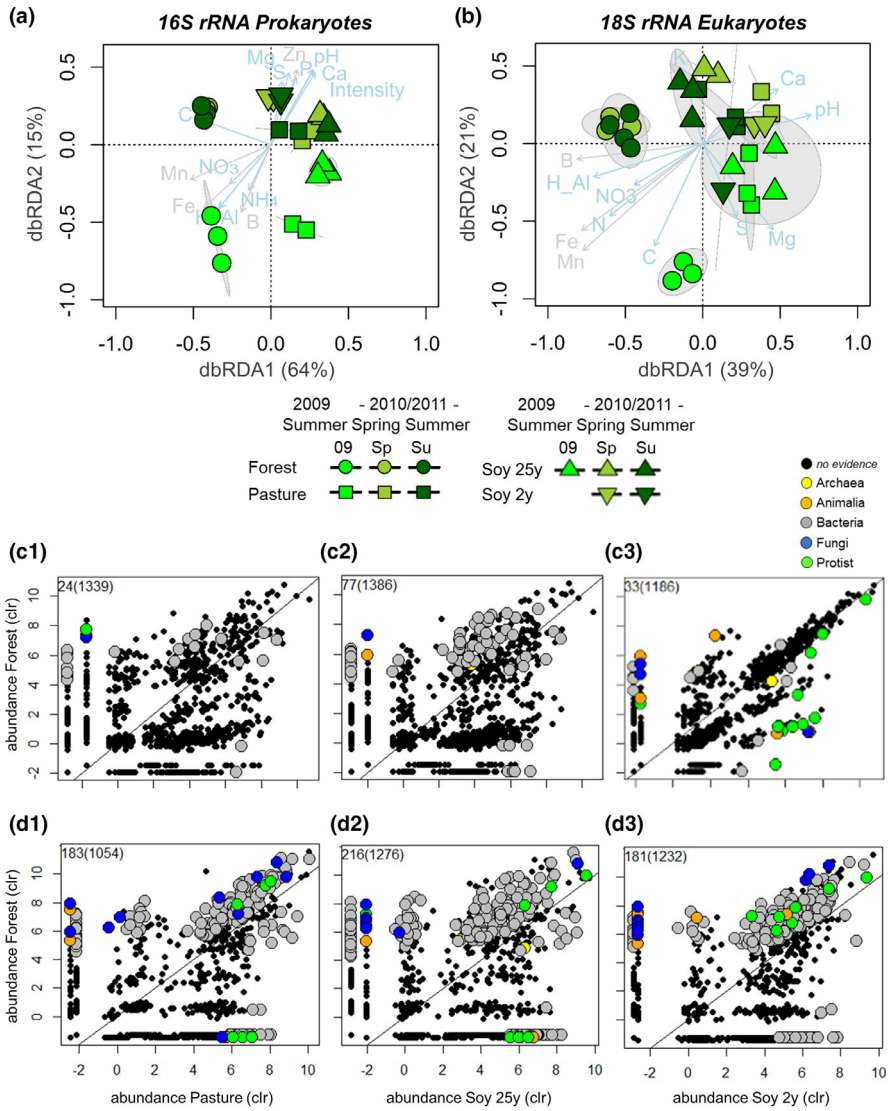

FIGURE 2 Effect of the land use change from rainforest to agriculture on the soil biota at different sampling times (Survey 2). The graphs on the top indicate the changes in the community structures assessed by a transformation-based redundancy analysis (tbRDA) for the genes (a) 16S rRNA (Prokaryotes) and (b) 18S rRNA (Eukaryotes) (additional details by taxonomic groups in Figures S2–S6). Ellipses surround data for microbial communities that did not differ from each other (represent 99% confidence intervals, *SE*). The graphs below represent the community shifts from native forest in spring 2010/11 to (c1) pasture, (c2) soy and (c3) soy 2y; and from native forest in summer 2010/11 to (d1) pasture, (d2) soy and (d3) soy 2y (each dot represents an OTU and significant changes were accessed by ALDEX2, $p < .05$, and coloured according to the taxa; the abundance data were centred log-ratio transformed, clr)

in Nematoda (Animalia), in Ascomycota and Basidiomycota (Fungi), in Nitrososphaeria (Archaea), and in Actinobacteria (Bacteria).

Furthermore, we correlated the changes in soil biota richness from native vegetation to pasture and crop fields to the size of organisms (Figure 4). For microbiota from crop sites, there is a trend towards smaller groups (e.g., Bacteria and Archaea) to be most strongly affected, but this is less evident for pasture sites. The hypervariable group Protist is an exception to this trend (Figure 4). Unfortunately, the lack of tables available for soil Protists that detail their size to allow for better taxonomic resolution prevented further analysis (more details in Discussion).

Lastly, soil biota shifts were more evident at fine taxonomic resolution, but most of the taxa could not be identified at the genus/species level (Figure 4). The heatmap shows an overall shift pattern of the 100 taxa most affected by land use change from native vegetation to agriculture (aldex2, FDR-corrected). Native vegetation samples primarily clustered together with each other, according to biome and sampling time, and secondarily clustered with the newly established crop fields (s2). The older agricultural fields of pasture (P) and soybean sites (S) formed a distinct cluster away from the native vegetation and clustered secondarily by sampling time (Figure 4). Even if overall the soil biota diversity increased in the agricultural sites, agriculture decreased the relative abundance of some taxa, including 6/31 OTUs of Archaea, 245/1,790 of Bacteria, 12/74 of Animalia, 20/144 of Fungi and 25/310 of Protists (Figures 1 and 2).

4 | DISCUSSION

We investigated two distinct soil surveys in southern Amazonia, focusing analysis on the impact of land use change from native vegetation to agriculture. First, we provided further support that microbial community structures shift with land use and are highly correlated with soil chemical changes, such as pH and fertility (Brinkmann et al., 2019; Jesus et al., 2009; Goss-Souza et al., 2017; Lammel, Feigl, et al., 2015; Lammel, Nüsslein, et al., 2015; Mendes et al., 2015; Merloti et al., 2019; Pedrinho et al., 2019). Second, we also found that general soil biota diversity is enhanced in the agricultural soils (Jesus et al., 2009; Mendes et al., 2015; Rodrigues et al., 2013) and we

TABLE 1 Permutational multivariate analysis of variance (perMANOVA), variation partitioning (*varpart*) and correlation (*envfit*) among soil biota community structures across land use types and explanatory variables

	Survey 1					Survey 2			
	Eukaryotes		Prokaryotes			Eukaryotes		Prokaryotes	
Variable	R^2	*p*	R^2	*p*	Variable	R^2	*p*	R^2	*p*
perMANOVA									
Land use (LU)	**.38**	*.001*	**.60**	*.001*	Land use	**.34**	*.001*	**.65**	*.002*
Region	**.09**	*.013*	**.08**	*.001*	Time	**.16**	*.001*	**.11**	*.001*
LU:Region	**.13**	*.011*	**.14**	*.004*	LU:Time	**.19**	*.002*	**.10**	*.020*
Variation partitioning [a]	*Variation*	*p*	*Variation*	*p*		*Variation*	*p*	*Variation*	*p*
Land use	**.06**	*.001*	**.12**	*.001*	Land use	**.15**	*.001*	**.19**	*.001*
Region	**.03**	*.013*	**.10**	*.007*	TIME	**.12**	*.001*	**.12**	*.001*
Soil (PCA axes)	**.05**	*.002*	**.04**	*.013*	Soil (PCA axes)	**.04**	*.014*	**.03**	*.012*
Residual	**.65**		**.37**		*Residual*	**.66**		**.30**	
Envfit correlation	R^2	*p*	R^2	*p*		R^2	*p*	R^2	*p*
LU intensity (LUI)	**.90**	***.001***	**.91**	***.001***	LUI	**.56**	***.001***	**.95**	***.001***
NH_4	.56	*.002*	.53	*.010*	NH_4	.24	*.061*	.32	*.016*
NO_3	.70	*.001*	.14	*.383*	NO_3	.31	*.028*	.41	*.009*
pH	**.83**	***.001***	**.93**	***.001***	pH	**.62**	***.001***	**.94**	***.001***
Al	.80	*.001*	.90	*.001*	Al	.64	*.001*	.87	*.001*
N	*.04*	*.701*	.67	*.001*	N	.68	*.001*	.83	*.001*
C	.31	*.061*	.62	*.002*	C	.63	*.005*	.80	*.001*
P	.82	*.001*	.33	*.044*	P	*.11*	*.345*	.66	*.001*
S	*.29*	*.018*	.36	*.035*	S	.32	*.030*	.53	*.001*
K	.64	*.003*	.39	*.036*	K	*.29*	*.040*	*.12*	*.360*
Ca	.83	*.001*	.77	*.003*	Ca	.42	*.009*	.91	*.001*
Mg	.52	*.004*	.58	*.007*	Mg	.60	*.001*	.70	*.001*
Litter hemi-cellulose	*.39*	*.005*	*.13*	*.381*	*B*	.52	*.001*	.60	*.003*
Litter cellulose	*.11*	*.258*	*.01*	*.930*	*Cu*	*.19*	*.156*	*.26*	*.098*
Litter lignin	.62	*.002*	.76	*.001*	*Fe*	.72	*.001*	.96	*.001*
Litter fatty acids	.32	*.020*	.54	*.004*	*Mn*	.83	*.001*	.86	*.001*
Litter carbohydrates	.55	*.004*	.41	*.038*	*Zn*	.22	*.100*	.71	*.001*
Litter C	.56	*.005*	.69	*.003*					
Litter N	*.17*	*.195*	.35	*.065*					
Litter CN	*.19*	*.175*	*.26*	*.133*					

[a]Details of variation partitioning in Figure S3.

[b]The main variables discussed in the text are highlighted in bold.

suggest that these observations fit the predictions of the DEM and IDH (Houston, 2014; Brinkmann et al., 2019; George et al., 2019).

Our results are in line with previous studies that reported soil characteristics as the main drivers of microbial community changes across land uses in southern Amazonia (Lammel, Feigl, et al., 2015; Lammel, Nüsslein, et al., 2015; Mendes et al., 2015; Merloti et al., 2019; Pedrinho et al., 2019). However, such observations should be interpreted with caution, because soil chemistry in native vegetation on Oxisols (Ferralsols/Latosols) is usually very stable, as demonstrated by a large survey in Brazil made by the RADAM project (Cooper et al., 2005). In southern Amazonia, pH of Oxisols is commonly 3.8–4.0 in $CaCl_2$ or 4.4–4.6 in H_2O, and usually demonstrates low fertility (Cooper et al., 2005; Quesada et al., 2011). In agricultural soils, however, soil pH and fertility (mainly macronutrients, such as P, K, Ca and Mg) are always improved, which otherwise would not permit farming, as most crops are highly demanding in nutrients (Carvalho et al., 2007; Lammel, Nüsslein, et al., 2015; Lammel et al., 2015b and Lammel et al., 2017). This creates two distinct extremes making land use intrinsically connected with soil chemistry and potentially blurring correlations of soil fertility values with microbial

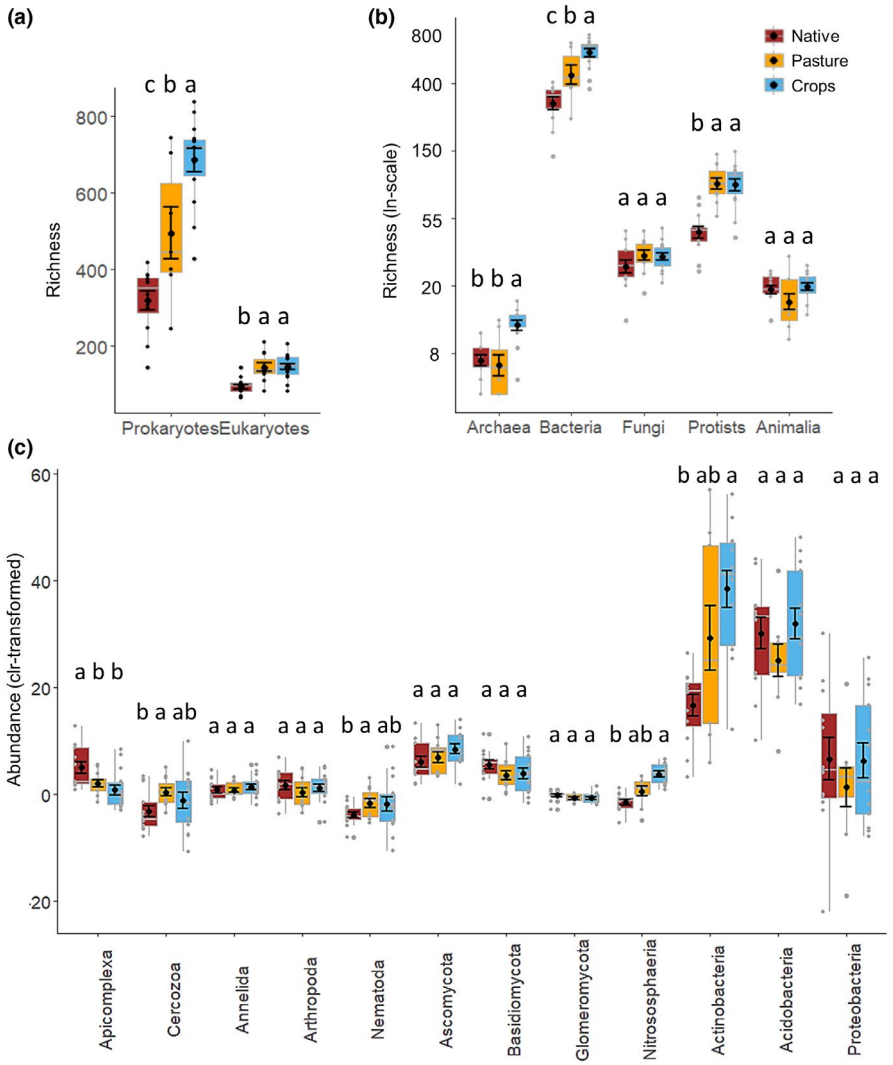

FIGURE 3 Distribution of soil taxa according to land use: native vegetation, pasture, and crops. (a) Richness of OTUs for Prokaryotes (gene 16S rRNA) and Eukaryotes (gene 18S rRNA); (b) richness according to high-order taxonomic groups (Archaea, Bacteria, Fungi, Protist and Animalia), natural logarithm (ln) scale; (c) abundances of the most abundant taxa (centred log ratio transformed, clr) across each high-order taxonomic group (details in Figures S2–S5). Land uses with the same letter for each taxonomic group do not differ by the Tukey's *post hoc* test, $p < .05$. Classical ANOVA/Tukey's *post hoc* was also used to rank the relative abundance data, because there is no test yet available for ranking compositional data

community structure (Lammel et al., 2015a, 2018). This also implies that any correlation of pH and fertility with soil biota with the native vegetation should be interpreted with caution, as pH, which also covaries with Ca and Al, is usually very homogeneous across the native vegetation soils in this region (Carvalho et al., 2007). Thus, soil pH and its covariables should not be suggested as the sole cause of microbial changes (Fierer et al., 2009; Lammel et al., 2018), but rather pH can be regarded as an indicator of the changes, as changes of soil fertility overlap with other alterations encapsulated in land use change (Lammel, Nüsslein, et al., 2015; Mendes et al., 2015).

This strong connection between land use types and soil chemistry leads to our second point. As described above, the overall changes in the soil habitat may also impact the soil microbial communities. As previously demonstrated, habitat changes caused by land use change are, beyond soil fertility, also related to other parameters. These parameters include changes in resource quantity and quality caused by changes in plant cover, as well as disturbances caused by tillage and pesticide application (Mendes et al., 2014; Nivelle et al., 2016; Zhang et al., 2018). In addition, there are also soil pollutants and other factors that are rarely quantified, such as heavy metals that can contaminate fertilizers, adjuvants of pesticides (e.g., tensioactive substances and conservants) and residuals from the machines and other supplies (e.g., oil and plastic). While soil microbial community structure adapted over centuries to the stable plant cover in native vegetation with little anthropogenic input (despite atmospheric depositions), agriculture creates several disturbances in soil communities. These disturbances can happen on a cyclical basis such as the presence of animals and grazing in pastures or crop rotations and chemical applications in crop fields. Even though this complex mix of factors is experimentally hard to control or disentangle, farmers would benefit from keeping soils at intermediate disturbance levels, as extreme disturbances can also affect crop yield and jeopardize the viability of their farms. Because the South Amazonia region is a new agricultural region, we could not find any extreme disturbance situation that could repress the soil community to levels lower than the native vegetation (e.g., extremely degraded soils). Thus, while none of these disturbances are as drastic as other human activities such as mining or construction (Joyner et al., 2019; Lima et al., 2016;

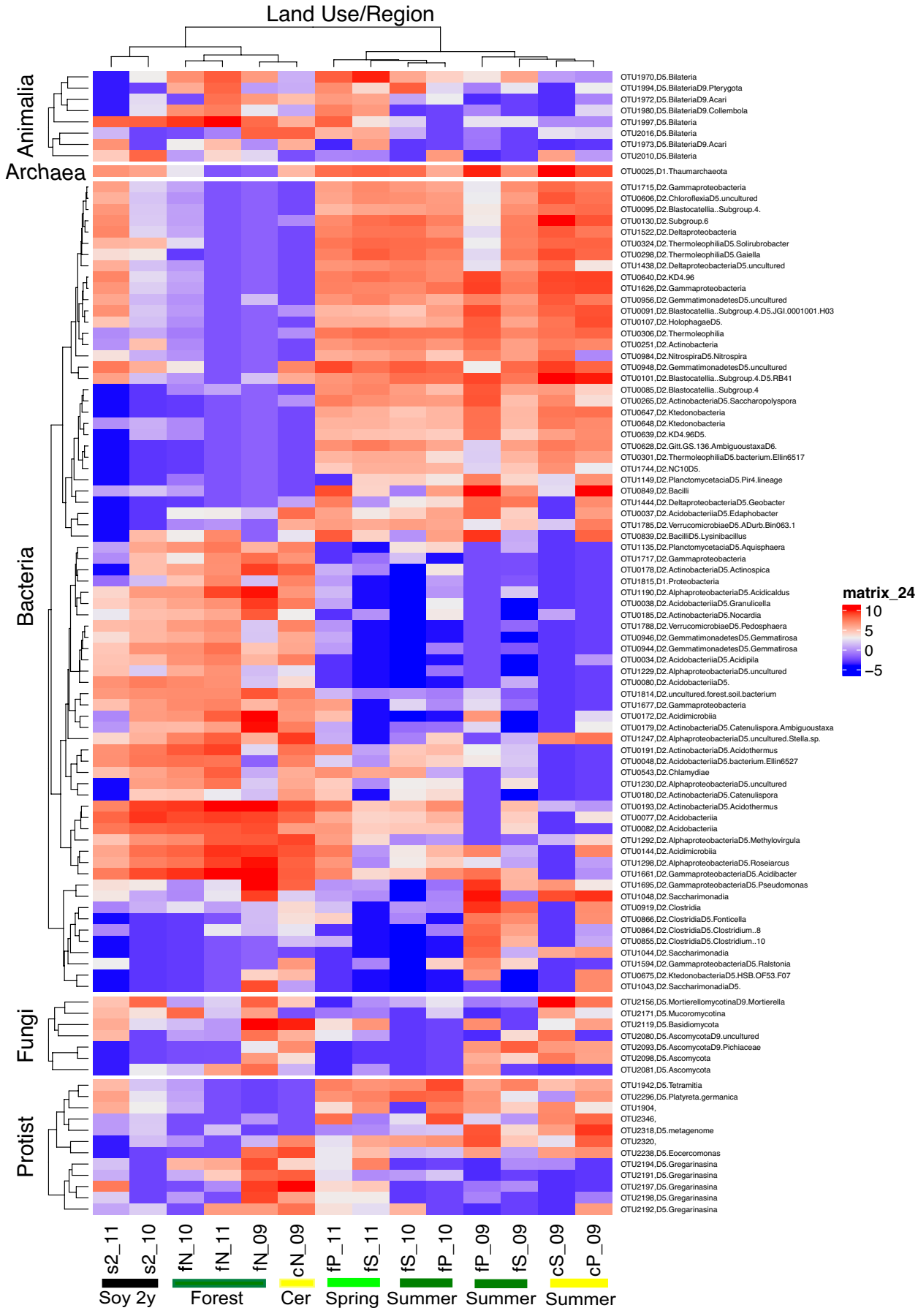
Land Use/Region
Animalia
Archaea
Bacteria
Fungi
Protist
OTU1970,D5.Bilateria
OTU1994,D5.BilateriaD9.Pterygota
OTU1972,D5.BilateriaD9.Acari
OTU1980,D5.BilateriaD9.Collembola
OTU1997,D5.Bilateria
OTU2016,D5.Bilateria
OTU1973,D5.BilateriaD9.Acari
OTU2010,D5.Bilateria
OTU0025,D1.Thaumarchaeota
OTU1715,D2.Gammaproteobacteria
OTU0606,D2.ChloroflexiaD5.uncultured
OTU0095,D2.Blastocatellia..Subgroup.4.
OTU0130,D2.Subgroup.6
OTU1522,D2.Deltaproteobacteria
OTU0324,D2.ThermoleophiliaD5.Solirubrobacter
OTU0298,D2.ThermoleophiliaD5.Gaiella
OTU1438,D2.DeltaproteobacteriaD5.uncultured
OTU0640,D2.KD4.96
OTU1626,D2.Gammaproteobacteria
OTU0956,D2.GemmatimonadetesD5.uncultured
OTU0091,D2.Blastocatellia..Subgroup.4.D5.JGI.0001001.H03
OTU0107,D2.HolophagaeD5.
OTU0306,D2.Thermoleophilia
OTU0251,D2.Actinobacteria
OTU0984,D2.NitrospiraD5.Nitrospira
OTU0948,D2.GemmatimonadetesD5.uncultured
OTU0101,D2.Blastocatellia..Subgroup.4.D5.RB41
OTU0085,D2.Blastocatellia..Subgroup.4
OTU0265,D2.ActinobacteriaD5.Saccharopolyspora
OTU0647,D2.Ktedonobacteria
OTU0648,D2.Ktedonobacteria
OTU0639,D2.KD4.96D5.
OTU0628,D2.Gitt.GS.136.AmbiguoustaxaD6.
OTU0301,D2.ThermoleophiliaD5.bacterium.Ellin6517
OTU1744,D2.NC10D5.
OTU1149,D2.PlanctomycetaciaD5.Pir4.lineage
OTU0849,D2.Bacilli
OTU1444,D2.DeltaproteobacteriaD5.Geobacter
OTU0037,D2.AcidobacteriiaD5.Edaphobacter
OTU1785,D2.VerrucomicrobiaeD5.ADurb.Bin063.1
OTU0839,D2.BacilliD5.Lysinibacillus
OTU1135,D2.PlanctomycetaciaD5.Aquisphaera
OTU1717,D2.Gammaproteobacteria
OTU0178,D2.ActinobacteriaD5.Actinospica
OTU1815,D1.Proteobacteria
OTU1190,D2.AlphaproteobacteriaD5.Acidicaldus
OTU0038,D2.AcidobacteriiaD5.Granulicella
OTU0185,D2.ActinobacteriaD5.Nocardia
OTU1788,D2.VerrucomicrobiaeD5.Pedosphaera
OTU0946,D2.GemmatimonadetesD5.Gemmatirosa
OTU0944,D2.GemmatimonadetesD5.Gemmatirosa
OTU0034,D2.AcidobacteriiaD5.Acidipila
OTU1229,D2.AlphaproteobacteriaD5.uncultured
OTU0080,D2.AcidobacteriiaD5.
OTU1814,D2.uncultured.forest.soil.bacterium
OTU1677,D2.Gammaproteobacteria
OTU0172,D2.Acidimicrobiia
OTU0179,D2.ActinobacteriaD5.Catenulispora.Ambiguoustaxa
OTU1247,D2.AlphaproteobacteriaD5.uncultured.Stella.sp.
OTU0191,D2.ActinobacteriaD5.Acidothermus
OTU0048,D2.AcidobacteriiaD5.bacterium.Ellin6527
OTU0543,D2.Chlamydiae
OTU1230,D2.AlphaproteobacteriaD5.uncultured
OTU0180,D2.ActinobacteriaD5.Catenulispora
OTU0193,D2.ActinobacteriaD5.Acidothermus
OTU0077,D2.Acidobacteriia
OTU0082,D2.Acidobacteriia
OTU1292,D2.AlphaproteobacteriaD5.Methylovirgula
OTU0144,D2.Acidimicrobiia
OTU1298,D2.AlphaproteobacteriaD5.Roseiarcus
OTU1661,D2.GammaproteobacteriaD5.Acidibacter
OTU1695,D2.GammaproteobacteriaD5.Pseudomonas
OTU1048,D2.Saccharimonadia
OTU0919,D2.Clostridia
OTU0866,D2.ClostridiaD5.Fonticella
OTU0864,D2.ClostridiaD5.Clostridium..8
OTU0855,D2.ClostridiaD5.Clostridium..10
OTU1044,D2.Saccharimonadia
OTU1594,D2.GammaproteobacteriaD5.Ralstonia
OTU0675,D2.KtedonobacteriaD5.HSB.OF53.F07
OTU1043,D2.SaccharimonadiaD5.
OTU2156,D5.MortierellomycotinaD9.Mortierella
OTU2171,D5.Mucoromycotina
OTU2119,D5.Basidiomycota
OTU2080,D5.AscomycotaD9.uncultured
OTU2093,D5.AscomycotaD9.Pichiaceae
OTU2098,D5.Ascomycota
OTU2081,D5.Ascomycota
OTU1942,D5.Tetramitia
OTU2296,D5.Platyreta.germanica
OTU1904,
OTU2346,
OTU2318,D5.metagenome
OTU2320,
OTU2238,D5.Eocercomonas
OTU2194,D5.Gregarinasina
OTU2191,D5.Gregarinasina
OTU2197,D5.Gregarinasina
OTU2198,D5.Gregarinasina
OTU2192,D5.Gregarinasina
matrix_24
10
5
0
−5
s2_11
s2_10
fN_10
fN_11
fN_09
cN_09
fP_11
fS_11
fS_10
fP_10
fP_09
fS_09
cS_09
cP_09
Soy 2y
Forest
Cer
Spring
Summer
Summer
Summer

FIGURE 4 Heatmap of those 100 abundant and prevalent OTUs that differed the most in abundance between native vegetation and agricultural fields ($p > .05$). The legend at the bottom encodes with the first letter the biome Cerrado (c) or forest (f), the second letter Native (N), Soy (S) or Soy 2 years (S2), or Pasture (P), followed by the year of the survey: 09, summer 2009; 10, spring 2010; and 11, summer 2011. Note that spring and summer in Brazil occur during the transition of the years (10/11)

Wang et al., 2016), our results suggest that productive agricultural systems, such as commercial pastures and crops, create intermediate and constant levels of disturbance.

Thus, we suggest that in the agricultural sites, the shift in microbial community structure is related to both disturbances and soil chemical changes (results that were consistent in both spatial and temporal surveys, Table 1). We suggest that these factors drive reductions in the abundance of previously dominant groups in native soil, allowing rare groups to thrive (Figures 1 and 2). This is especially evident in the heatmap analysis, which shows a clear transition from native vegetation to new agricultural fields and then to old fields (Figure 4). In addition, based on our temporal survey across seasons (survey 2), shifts in microbial communities were also affected by seasonal and yearly changes (Figures 2 and 4; Table 1), as the different temporal soil collections yielded slightly different communities. These changes are probably related to seasonal climatic variations that affect soil moisture and temperature (Bell et al., 2009; Mendes et al., 2015). However, land use was still a stronger factor than spatial or temporal factors (Table 1).

Interestingly, overall prokaryotes were more strongly influenced by the land use types than eukaryotes (Table 1). We considered that prokaryotes were more strongly influenced by land use than other soil biota because organism size is roughly correlated with generation time and metabolic status, and is indicative of trophic level in the food web (Zinger et al., 2019). Thus, smaller organisms, such as bacteria, are expected to adapt/shift faster to land use change than eukaryotes (Veresoglou et al., 2015). We plotted the diversity data according to organism size, and the shift in richness with organism size was evident for most taxa, but not for Protists (Figure S10). Attempts to correlate soil organism size with sequencing data are very recent (Zinger et al., 2019), and there are limited data on size distribution of organisms. Additionally, we suggest that the dispersal of prokaryotes may be more intense at the scale of the sample core, while eukaryotes are able to disperse to longer distances (e.g., between land uses) (Thakur et al., 2020). Among eukaryotes, most fungi produce spores and animals have a higher range of movement, while prokaryotes are dependent on passive transport by, for example, air and animals to reach longer distances (Bahram et al., 2015; Richter-Heitmann et al., 2020; Thakur et al., 2020). A previous study suggested that dispersal of microbial taxa between different land uses in the tropics is moderate (Goss-Souza et al., 2017), and then deterministic and stochastic processes should be the main drivers of assemblage of the soil biota communities. Thus, land use change could affect soil biota community assemblage by combining deterministic processes (niche changes due to soil physical–chemical changes and plant cover changes) and stochastic processes (constant stimulation by disturbance in agricultural systems) (George et al., 2019; Goss-Souza et al., 2017; Mendes et al., 2014).

Recent studies have shown that soil microbial richness is usually enhanced by the conversion of native vegetation to agriculture, and the community structure turnover is intense (Brinkmann et al., 2019; Jesus et al., 2009; George et al., 2019; Pedrinho et al., 2019; Rodrigues et al., 2013). Furthermore, some studies suggest that the frequency and intensity of disturbance increased the diversity of soil biota communities (Brinkmann et al., 2019; George et al., 2019; Santillan et al., 2019). We suggest that the DEM/IDH framework should be taken into account for a better understanding of land use change effects on soil biota communities. In the context of the DEM framework (Houston, 2014), the increased amount of disturbance in terms of frequency and intensity (e.g., double cropping, completely changing plant cover twice a year) on the crop fields could be an equalizing process, slowing down the process of competitive exclusion by routinely inducing a stochastic process of re-assemblage of the communities at every sowing–harvest cycle. Also, the disturbance caused by the application of pesticides and by changing crop type could act as a stabilizing process, by limiting the population size of some taxa more than others.

The current biggest challenge in using the DEM/IDH to explain soil biota across land uses is to establish measurable indices to quantify disturbance and productivity in the below-ground context. Within the framework of the DEM, it is not clear whether the changes from native to agricultural soils better fit a shift to intermediate productivity and intermediate disturbance (IDH), or to high productivity and high mortality rate (Huston, 2014). We clearly need further studies to systematically include disturbance and productivity measurements as explanatory variables for diversity in soil across land use types. However, our study suggests that the impact of land use change on soil biota should be seen as more than deterministic processes caused by soil pH and fertility. Even though soil fertility parameters are relatively easy to quantify, correlation with soil biota is confounded by other covariables, as described before. Therefore, we agree with recent papers calling for the consideration of disturbances when analysing the effects of land use change on soil biota (Brinkmann et al., 2019; George et al., 2019).

Relating to changes of specific taxa, our study is in accordance with previous studies indicating that Bacteria were the most diverse microbial group in the soil samples (Pedrinho et al., 2019; Zinger et al., 2019). Bacteria also presented the highest turnover with the change from native vegetation to agriculture. Shifts were observed in all major taxonomic groups throughout the tree of life; however, the changes are more evident at the finest taxonomic resolution of OTU levels (Figure 4). Most of the affected taxa are related to uncultured microorganisms, so future improvements in methods and taxonomic databases will allow us to better understand the impacts on individual taxa. Some patterns of shifting community compositions such as the increase of Actinobacteria in

agricultural sites were also observed in previous studies (George et al., 2019; Pedrinho et al., 2019). Even though there was an overall increase of richness in agricultural soils (Figure 3), in a previous study we detected a significant decrease in fungal abundance in crop fields during the summer season (Lammel et al., 2017; Lammel, Nüsslein, et al., 2015). This suggests that the group Fungi should be examined with more specific primers and with greater detail in further studies. Previous studies in the same sampling area detected a remarkable increase in the abundance of Archaea in crop fields (Lammel et al., 2015a, 2017). We made a similar observation here (Figure 3), especially for members of the class Nitrososphaeria. This class is known to contain ammonia-oxidizing microorganisms and it is very likely that agricultural management increases nitrogen availability and thus favoured this particular group (George et al., 2019; Hamaoui et al., 2016).

Furthermore, we detected in agricultural soils an increased richness of the most abundant groups of Protist (Figure S11), and also an increased relative abundance of some potential bacterivores, such as Tetramitia and Platyreta (Figure 4). It is possible that bacterivores benefited from the increased richness of Bacteria at this sites (Oliverio et al., 2020). We also detected a decreased relative abundance of the Protist group Apicomplexa (Figure 3). Apicomplexa are usually related to parasitic organisms, such as Gregarinasina (Figure 4), a parasite of invertebrates (Oliverio et al., 2020). We speculate that shifts in the Animalia community structure could have impacted this Protist group (Figure 4). In addition, Cercozoa, a major group of Protists, and Nematoda were altered by the conversion to agricultural soils (George et al., 2019). Agricultural perturbation, such as pesticides, possibly also affected some of these organism groups in the soil (Geisen et al., 2018).

5 | CONCLUSIONS

We found that microbial communities were strongly affected by land use change in both the Cerrado and rainforest biomes in southern Amazonia. The soils converted from native vegetation to crop fields had overall the highest richness of soil biota. We propose that dynamic cropping systems not only altered the soil chemistry by increasing soil pH and fertility, but also created continued disturbances in the form of crop rotations that stimulated soil diversity, in line with the DEM and the IDH. Even though agricultural fields had higher microbial richness, we also detected intensive microbial community turnover with the conversion from native vegetation to agricultural fields, indicating that several soil taxa are suppressed by agriculture. Consequently, land use change in this region should proceed with caution. In the southern Amazonia region of Brazil, current laws require farmers to keep 20%–80% pristine native vegetation areas on their property. Our data support the merit of upholding this law: because there are unique soil biota taxa in native vegetation soil, keeping these areas will contribute to maximizing soil biodiversity protection in farm soils.

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CONFLICT OF INTEREST

The authors declare no competing interests.

AUTHOR CONTRIBUTION

D.R.L., C.E.P.C. and K.N. conceived the study. D.R.L. performed the experiments and bioinformatics and statistical analyses. D.R.L. wrote the manuscript with input from all co-authors.

DATA AVAILABILITY STATEMENT

Sequencing data and metadata are available at the NCBI Bioproject PRJNA587110.

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SUPPORTING INFORMATION

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