Soil microbes under threat in the Amazon Rainforest

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Abstract

Soil microorganisms are sensitive indicators of land-use and climate change in the Amazon, revealing shifts in important processes such as greenhouse gas production, but have been underutilized in conservation and management initiatives. Integrating soil biodiversity with other disciplines while expanding sampling efforts and targeted microbial groups is crucially needed.

Keywords: land-use change; biodiversity conservation; microbial ecology; soil microbial communities; soil ecosystem services; methane cycle; climate change.

Current trends in land-use change in the Brazilian Amazon

The Amazon rainforest is facing unparalleled threats, especially in Brazil. Approximately 60% of the Amazon Basin is within Brazil's borders, and 20% of this area has been deforested so far [1]. Compared to the year with the lowest deforestation ever recorded (2012), clearing rates of the Brazilian Amazon increased by 185% in 2021, reaching the highest values of the decade (see: http://terrabrasilis.dpi.inpe.br/app/dashboard/deforestation/biomes/legal_amazon/rates). This process is largely driven by agriculture, with cattle pastures (87%) and soybean fields (9%) as the main anthropogenic land uses (see: https://plataforma.brasil.mapbiomas.org). Even what we recognize as forest is not an entirely pristine environment – besides **secondary forests** (Glossary), large areas have been affected by selective logging or understory fires [2].

While much of Amazonian conservation efforts focus on above-ground species and habitats, soils provide indispensable ecosystem services, such as carbon storage, greenhouse gas (GHG) regulation, nutrient cycling, and disease and pest control [3]. In the Amazon, agricultural and pasture lands are usually established through forest clearing and burning. This process alters the natural characteristics of soils, causing initial increases in soil pH, nutrient content and availability, and **soil structure** degradation [4]. In the long run, converting rainforests to pastures may result in nutrient depletion [4] and, not surprisingly, more than half of Amazonian pastures (56%) are now moderately or severely degraded (see: https://plataforma.brasil.mapbiomas.org). These impacts have negatively affected below-ground biodiversity, including microbes (Figure 1).

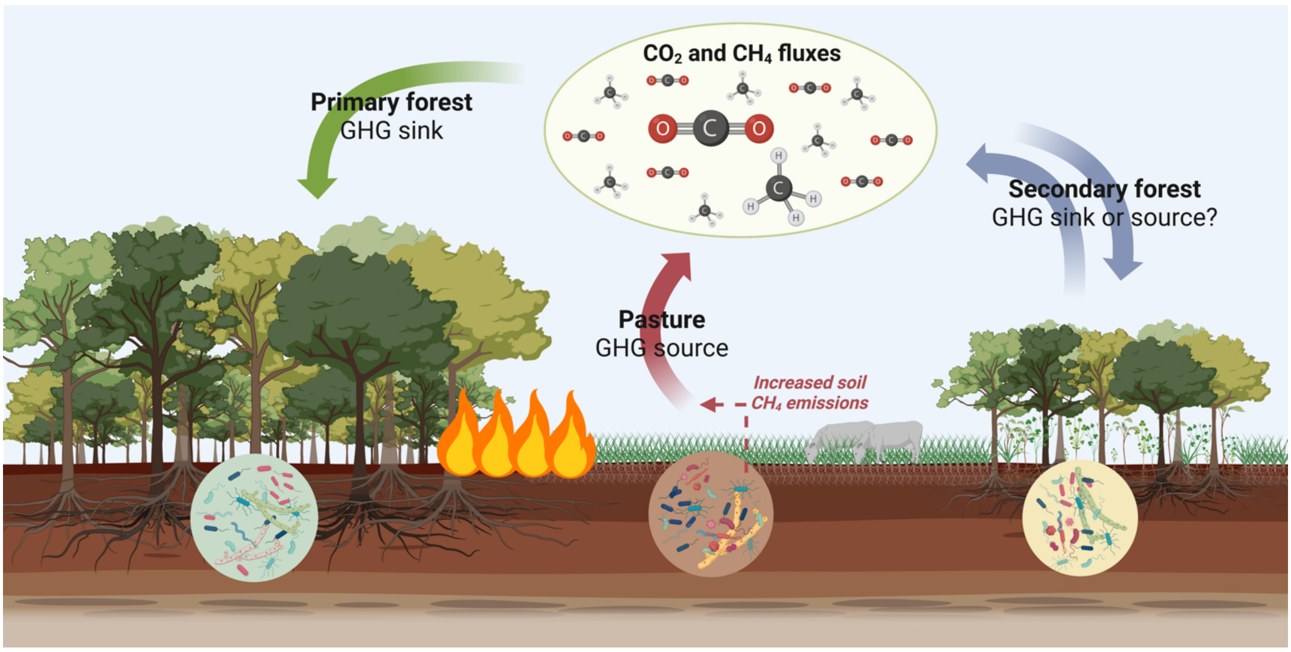

Figure 1. Impacts of forest-to-pasture conversion in the Brazilian Amazon. Three typical Amazonian landscapes (L-R): primary forest, pasture (the most common anthropogenic land use of the region), and secondary forest (originated from abandoned deforested areas). Pasture establishment, through vegetation clearing and burning, alters above-ground biodiversity and releases large greenhouse gas (GHG) emissions, while primary forests are sinks of carbon dioxide

(CO_2) and methane (CH_4). In the short-term, total GHG emissions from deforestation cannot be entirely offset by secondary forest growth. For belowground, these processes are even less understood. Forest-to-pasture conversion impacts soil properties, microbial communities, and their ecosystem services, leading to a CH_4 sink-to-source shift from soils due to changes in the number of CH_4-producing and -consuming microorganisms. However, it is still unclear if soil microbial communities from secondary forests and their roles, including CH_4 regulation, can be completely recovered. Created with BioRender.com.

Land-use impact patterns on soil microbes and their functions

Almost all soil chemical transformations depend on microorganisms [3], and with the advancement and availability of molecular and sequencing techniques (e.g., **amplicon sequencing** and **shotgun metagenomic sequencing)**, high-resolution studies on Amazonian microbes have become possible. Such studies have identified consistent forest-to-pasture conversion impacts on microbial communities, including responses in composition and structure [5]. One unexpected finding was that pastures have higher alpha (local) taxonomic richness and **phylogenetic diversity** of soil bacteria than forests, meaning a greater number of species, which are also more distantly related to each other [6]. However, for both indexes, pastures exhibit lower beta diversity (landscape) compared to forests, which indicates that their soil bacterial communities are more similar across space, while forest communities become increasingly dissimilar as the distance between samples grows [6]. This *biotic homogenization* process can be associated with the increase in the ranges of a few species and the loss of endemic forest taxa [6].

In general, if an environment loses microbial species and becomes more similar across space, its **microbial core functions** may not be strongly affected; the greatest impact will likely be observed on specialized processes restricted to particular taxa, for which there may be little **functional redundancy** [7]. The alteration of these processes can change the overall **functional profile** of soil microbial communities, and recent studies have provided evidence of such effects in the Amazon following the conversion of forest to pasture [5], with alarming impacts on crucial microbial functions.

For example, microorganisms can regulate the GHG exchange between the soil and the atmosphere. This is particularly relevant for methane (CH_4) due to its high **global warming potential**. Converting Amazonian forests into pastures has changed the number of methane-producing archaea (**methanogens**) and -consuming bacteria (**methanotrophs**) in soils, as well as the balance between these groups. Well-drained forest soils typically act as methane sinks, but when converted, they result in pastures with more methanogens and **methanogenesis**-related genes and, thus, larger gas emissions [e.g., 8, 9]. This *sink-to-source shift* is intensified by removing soil cover and **liming** [8], showing that not only the land use itself matters but also the management employed.

Another serious concern about land-use changes are the possible microbe-related impacts on human and animal health. For example, through metagenomics, Amazonian deforestation was linked to changes in the structure of the soil **resistome** and increased amount and diversity of antibiotic-resistance genes [10]. Land-use and climatic stressors on soils can also be associated with increasing exposure and emergence of infectious diseases [11].

Scientific challenges and main gaps in the field

Even in the **omics** era, it is still not a trivial task to understand the composition, distribution, and functional roles of soil microbial communities. However, this information is critical if we want to evaluate the impacts of anthropogenic changes on microbial-mediated ecological processes and interactions with above- and below-ground organisms. Inter- and transdisciplinary approaches are needed to help us understand, predict, and mitigate the complex responses of Amazonian forest ecosystems to deforestation and land conversion.

Consider the response of methane in Amazonian pastures as a case study. The importance of this GHG source – little discussed in national GHG budget assessments – and its microbial causes could be explored through the combination of molecular (e.g., gene abundances) and environmental measurements (e.g., gas fluxes). If linked to isotopic data, the metabolic pathways of soil methane production could be further identified, leading to a deeper understanding of this microbial response to land-use change. The organisms driving it could be explored through the combination of environmental genomics and bioinformatics approaches. These have already allowed the recovery of **metagenome-assembled genomes** from potentially novel Amazonian soil microorganisms involved in the methane cycle [12]. Machine-learning methods could also be used to predict emissions based on meteorological, soil, and microbial properties, helping us to create measures to avoid further emissions, besides planning restoration and management strategies.

Despite these areas of progress, fundamental questions remain, and we outline the main gaps that can serve as a reference for future research (Box 1). While land use and management practices affect soil microorganisms, what are the best practices that could be adopted in deforested areas to encourage the recovery of a healthy soil microbiome? In addition, secondary forests show some signs of taxonomic and functional recovery [5], but it is unclear if microbial communities and the processes they mediate can completely return to their original states (and how long this recovery might take). Furthermore, how are microbes affected by selective logging or understory fires, given the 1 M km^2 of degraded forests [2]?

As if this scenario is not challenging enough, the Amazon is also suffering from climatic changes, including increases in temperature (~0.7 °C over the last four decades) and the frequency and intensity of extreme events (floods and droughts), as well as changes in precipitation [13]. Specific studies of climate-related impacts on Amazonian soil microbes are scarce, and there is little information on whether land use-climate change interactions are altering ecosystem services provided by the soil microbiome. For the methane cycle, our previous results revealed that, under adverse events of increased soil moisture, land-use impacts could be intensified, leading to further emissions from Amazonian pasture soils [9].

Another key constraint in the field is that most of our knowledge comes from bacterial studies; however, bacteria are only one component of the soil microbiota. Much less information is available about other environmentally essential Amazonian soil microbes, such as fungi or archaea (the least-understood branch of microbial life). Fungi are known to play essential roles in soils, including complex associations with plants and the production of nitrous oxide (N_2O), but their overall responses to forest-to-pasture conversion are inconsistent across studies [6]. Besides, to our knowledge, only one study has evaluated the Amazonian soil **virome** and its response to environmental change [14], which can also influence soil biogeochemical cycles.

Box 1. Current scientific gaps in the field.

Land-use and climate change

- *Responses to forest degradation and forest-to-cropland conversion of soil microbes and their functions, in addition to the inclusion of other (not-yet evaluated) soil microbial-mediated processes.*
- *Are land-use impacts on soil microbes consistent across the Amazon and its rainfall gradients?*
- *Relationship between land-use impacts on soil microbes and above-ground taxa. Are they in the same direction and magnitude?*
- *Effects of climate change and its interactions with land use on soil microbes and functions.*

Neglected organisms

- *Impacts of land-use change on soil fungal communities, particularly at a functional level.*
- *Are there impacts on other microeukaryotes and the soil virome?*

Deforested areas and secondary forests

- *Role of sustainable practices to help recover a healthy soil microbiome in pastures and croplands.*
- *Can soil microbial communities and their ecosystem services return to their original states after forest regeneration compared to primary forests? How long would this process take?*
- *Can plant-soil-microbial dynamics help recover abandoned areas?*

Ecosystem services and public policies

- *Prediction of soil microbial ecosystem services and their changes.*
- *Contribution of microbial analyses to environmental and soil health reports and application in public policies.*

Towards a microbial framework in a changing forest

A bigger understanding of the tiniest forms of life in the Amazon rainforest is needed now more than ever, which can help us predict and mitigate the impacts of environmental change on ecosystem processes. However, the importance of this complex field is often underestimated and poorly understood. Considering the challenges described here and the fact that large areas of the Amazon have never been microbiologically investigated [15], we urgently need to increase sampling efforts across the basin, following ethical procedures and best practices for data and metadata collection, storage, and, especially, sharing.

Anthropogenic impact studies on microbes in the Brazilian Amazon are predominantly clustered in six geographic locations [5], with very few considering different soil types, deeper soil layers, and across time. Establishing or working in cooperation with long-term plot studies could help improve this scenario. For example, in the past 20 years, the Forest Plots project, a network of hundreds of permanent plots distributed across the basin, has contributed to our understanding of the taxonomic and functional diversity of the Amazonian flora (see: https://forestplots.net/). Introducing below-ground long-term monitoring in these plots could increase our knowledge of the above-ground relationships of microbial communities.

As exemplified for the methane cycle, studies can benefit from the inclusion of different dimensions of biodiversity (i.e., taxonomic, phylogenetic, and functional) and their integration with other disciplines – bringing together soil microbial ecologists with expertise in different taxa, as well as researchers from other fields (e.g., botany, landscape and restoration ecology, biogeochemistry, and data science). Inter- and transdisciplinary approaches, including methods to predict the influence of microbial communities on ecosystem functions, are crucial to improve the translation of our discoveries into policy. For the Amazon region in particular, special care must also be taken to ensure equal research collaboration efforts between and within countries, avoiding colonial practices often seen in ecological studies across the region.

Finally, as advocated by Averill [15] for the terrestrial microbiome, we urgently need to integrate Amazonian soil microbial diversity into conservation, restoration, and management efforts, creating practices to scale the science. We should hear and engage with local voices, considering their relationships with the land and sustainable practices, and our studies must communicate with other scientific and non-scientific audiences and stakeholders, highlighting the importance of preserving and recovering these soils. In our experience working in the region, we also believe that our research efforts should be used to co-create scientific dissemination projects and inclusive learning opportunities to foster young scientists in Amazonia and throughout Latin America, democratizing access to training in cutting-edge approaches to environmental science.

Competing interests

The authors declare no conflict of interest.

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Author contributions

A.M.V. wrote the manuscript with contributions from J.B.G., J.A.M., E.B., K.G.P., S.M.T., and B.J.M.B. J.B.G. produced the figure with contributions from A.M.V. and J.A.M. All authors critically revised the manuscript.

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Glossary

Amplicon sequencing
targeted sequencing method to analyze the genetic variation in specific genes and/or regions.

Functional profile
functional traits (metabolic potential) of a microbial community and their distribution patterns.

Functional redundancy
coexistence of distinct taxa capable of carrying out the same function.

Global warming potential (GWP)
Cumulative radiative forcing over a particular time horizon resulting from the emission of a unit mass of a specified greenhouse gas relative to that of carbon dioxide (CO_2). Methane (CH_4) has a GWP_{100} of 28.

Liming
application of calcium- and magnesium-rich materials into the soil, reacting as a base in acidic soils to neutralize their acidity and enable plant development.

Metagenome-assembled genomes (MAGs)
draft or complete genomes recovered from metagenomic data.

Methanogenesis
form of anaerobic respiration (organic matter degradation) that generates the gas methane as final product.

Methanogens
microorganisms capable of producing methane. Most are anaerobic Archaea that produce methane using carbon dioxide and hydrogen, acetate, and/or methylated compounds as main substrates.

Methanotrophs
microorganisms capable of oxidizing methane. Most are aerobic Bacteria that use methane as their sole source of carbon and energy.

Microbial core functions
common microbial functions that can be performed by several microorganisms (see [7] for more information).

Omics
refers to the study of biological systems at different levels, including genes (“genomics”), transcripts (“transcriptomics”), proteins (“proteomics”), and metabolites (“metabolomics”).

Phylogenetic diversity
measure of biodiversity that incorporates the phylogenetic difference between species.

Resistome
set of all antibiotic-resistance genes in a given sample.

Secondary forests
forests that have been cleared at least once. In the Amazon, secondary forests are typically generated from the abandonment of deforested areas.

Shotgun metagenomic sequencing
direct DNA sequencing from the organisms in a given sample.

Soil structure
refers to the arrangement of soil particles and associated pores.

Virome
the entire community of viruses in a given sample.