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Microbial bioindicators of soil functioning after disturbance: The case of gold mining in tropical rainforests of French Guiana

Heidy Schimann^{a,*}, Caroline Petit-Jean^b, Stéphane Guitet^c, Tatiane Reis^d, Anne Marie Domenach^e, Jean-Christophe Roggy^a

^a INRA-Joint Research Unit Ecology of Guiana Forests (Ecofog), campus agronomique, BP 709, 97387 Kourou Cedex, France

^b CNRS-Joint Research Unit Ecology of Guiana Forests (Ecofog), campus agronomique, BP 16, 97389 Kourou Cedex, France

^c ONF-National Office of Forest, Réserve de Montabo, BP 7002, 97307 Cayenne Cedex, France

^d University of West Indies and French Guiana (UAG) – Ecofog, campus agronomique, BP 16, 97389 Kourou Cedex, France

^e SOLICAZ – campus agronomique, BP 76, 97389 Kourou Cedex, France

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ABSTRACT

In the context of an ongoing monitoring study on the impacts of gold-mining activities on critical ecosystem processes, we explored the use of soil Denitrifying Enzyme Activity (DEA) and Substrate Induced Respiration (SIR) as ecosystem indicators in tropical rainforests of French Guiana. We also propose DEA/SIR ratio as ecosystem attribute able to describe the state of an ecosystem and to reflect changes in ecological processes. With this purpose, we measured SIR, DEA and DEA/SIR ratio in five gold-mining areas and five surrounding natural reference rainforests. We also measured indicators in two conditions of spontaneous regeneration of vegetation (stratified or not) and two conditions of soil rehabilitation (prior preparation of soils or not). We showed a high variability of DEA, SIR and DEA/SIR ratio in the natural reference forests. This pointed out the necessity to identify relevant reference systems - *i.e.* proving a close match in all relevant ecological dimensions - to compare with closed perturbed systems in order to assess the levels of alterations after disturbances. Results showed a high impact of gold mine on microbial processes with a strong decrease of DEA (10-fold lower), SIR (2-fold lower) and DEA/SIR ratio (8-fold lower) in perturbed areas in comparison with natural reference forests. The type of spontaneous vegetation (stratified or not) influenced the values of indicators as well as prior rehabilitation of soils, demonstrating the capacity of DEA, SIR and DEA/SIR ratio to respond in proportion to the perturbation (robustness) and to the different levels of restoration (sensitivity). The systematic decrease of the ratio DEA/SIR observed in the studied perturbed situations demonstrates clearly that the structure of microbial communities has been also modified. The ratio DEA/SIR proved to be robust and sensitive, and able to describe in fairly fine way changes of soil microbial communities in terms of structure and function in gold mine areas and during processes of restoration. We propose to use DEA, SIR and DEA/SIR ratio as bioindicators of both structural and functional aspects of C and N cycling in soils. Together with others bioindicators based on key supporting functions in soils, these indicators should accurately evaluate the ecological potential of natural ecosystems and the levels of degradation in case of land-use changes.

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1. Introduction

Nowadays, increase of anthropogenic degradation of ecosystems lead to an unsustainable intensification of the use of natural resources (Smil, 1997; Sala et al., 2000). Strong public policies are now needed to give a framework of sustainable management, which requires an accurate knowledge of ecosystems responses to anthropogenic modifications (Stokstad, 2005). The concept of ecosystem services has become an important model to link functioning of ecosystems and human well-being (MIllenium Ecosystem Assessment, 2005; Vihervaara et al., 2010); and their evaluation serve now as a criterion of decision in conservation policies (Kinzig et al., 2007). According to the integrative typology proposed by MEA (2005), these services rely on a relatively limited number of ecosystem functions (nutrients cycles, soils formations, *etc.*) that support them; and it seems more appropriate to evaluate ecosystems according to their ecological potential estimated from the analyses of their supporting functions rather than according to

^{*} Corresponding author. Tel.: +594 594 32 92 92; fax: +594 594 32 43 02. *E-mail address:* heidy.schimann@ecofog.gf(H. Schimann).

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the services they provide at given time (Bastian et al., 2011). Indeed, these supporting functions are more integrative, since a more limited number of functions can relate the extreme complexity of the structures and on-going processes exclusive to the systems. From a scientific perspective, studying perturbations of the ecosystems services or of their supporting functions meets with several difficulties. The major one lies in the fact that most ecosystems have already been altered by humans, who have left a strong impact on the current functioning (Dambrine et al., 2007). The most difficult task is therefore to identify relevant reference systems - i.e. proving a close match in all relevant ecological dimensions - to compare with perturbed systems, in order to assess the level of alteration of the services or of the functions. In this general context, tropical rainforests of French Guiana are a model of choice with reference non anthropogenic (primary forest) vs perturbed situations that can be used to measure parameters characterizing the ecological potential of the reference (Bastian et al., 2011), and to evaluate the level of degradation of these parameters in disturbed systems. It is now well recognized that direct measurements of parameters that support full suite of ecosystem services, or surrogate measurements representing the functions that support suite of services, are the only way to establish baselines from which to assess the current state of the perturbed situations (Hoeltje and Cole, 2007; Palmer and Filoso, 2009; Paetzold et al., 2010). The major difficulty lies therefore in the identification of integrative parameters that could reflect the capacity of the ecosystem to ensure its services as a whole. According to Anderson (2003), these key parameters are those related to soil functioning and more particularly to soil quality, also referred to as "soil natural capital" (Dominati et al., 2010). Indeed, many of the supporting functions such as soil formation, nutrient cycling, and primary production are all dependent on soil processes and indicate the centrality of soils in the provision of ecosystem services (Bennett et al., 2010; Palm et al., 2007). The concept of soil natural capital refers to the physical, chemical and biological properties that contribute to soil function and thus to the maintenance of ecosystem services as a whole (Knoepp et al., 2000). Specifically, soil natural capital should be characterized through a number of complementary indicators that relate mainly to their capacity in defining ecosystem processes and for which the biotic-abiotic interlinkages would find their expression (Anderson, 2003). Nevertheless, the complexity of the soil system makes difficult the choice of these indicators that would fulfill all criteria to be accurate (Dale and Beyeler, 2001): (i) easily measurable, (ii) sensitivity to variation in soil management and climate, (iii) predictive response to stress, (iv) low variability in response, and (v) accessibility and utility to stakeholders and decision makers. Measurements of soil microbial processes meet criteria for useful indicators of soil natural capital (see Bastida et al., 2008 for review; Harris, 2009; Ritz et al., 2009). Indeed, they are of crucial importance in biogeochemical cycling and have critical roles in plant interactions (Schimann et al., 2008; Wardle et al., 2004). Moreover, by virtue of their involving complex adaptive systems (i.e. the biota) they integrate multi-dimensional phenomena in ways that other indicators do not (Francaviglia, 2008). Moreover, they constitute the most integrative value of soil natural capital among its different levels of organization (Mausbach and Tugel, 1997). Also, soil microorganisms offer a huge metabolic diversity that cannot be taken into account in an exhaustive manner. It is then necessary to choose microbial activities that reflect the global functioning of soils and meet two types of selection criteria: (i) the ecological importance of the fluxes they generated, (ii) the representativeness according to the entire soil microbial community (Nannipieri et al., 2003). In this context, Schloter et al. (2003) consider that valuable bioindicators are those referring to the 'driving forces' for C and N cycling in soils. Among these functions, key processes such as soil respiration, nitrification and denitrification activities represent relevant 'target bioindicators' (Harris, 2009; Nannipieri et al., 2002).

The objectives of this study were to test the usefulness of two soil microbial functions (denitrification DEA, respiration SIR) as 'ecosystems indicators' according to Heink and Kowarik (2010) definition in the particular context of tropical rainforest: attributes with the capacity to describe the state of an ecosystem and to reflect changes in ecological processes. In particular, we needed to test (i) their sensitivity (i.e. their capacity to respond in proportion to different degrees of disturbances and/or of restoration) and (ii) their robustness (i.e. their capacity to display a predictive response to a given perturbation - Dale and Beyeler, 2001; Holt and Miller, 2011). Sensitivity and robustness have been tested on one type of disturbance, gold mining, with 3 levels of natural or human-induced restoration. Moreover, we propose a new bioindicator, the ratio DEA/SIR that we think can describe in a fairly fine way changes in the functioning of soil microbial communities in perturbed systems and during processes of restoration. Measuring the variations of these bioindicators in perturbed systems could thus accurately describe their status compared to undisturbed reference systems (Schimann et al., 2007). The overall aim is to develop a panel of easily measurable bioindicators to evaluate the ecological potential of natural ecosystems and the level of degradation of this potential in disturbed systems.

2. Materials and methods

2.1. Experimental sites

The work was undertaken in French Guiana, South America $(2-6^{\circ}N; 51^{\circ}50'-54^{\circ}50'W)$. Annual rainfall ranges from 2 to 4 m and the monthly temperatures vary slightly around 26 °C. The climate belongs to the seasonal equatorial type. The experimental sites were located in eastern French Guiana and consisted in five open-cast gold mines, unexploited since 5–7 years: Boulanger, Georgeon, Grand Chardy, Yaoni and La Boue.

On each gold-mine, we selected two modalities of natural restoration (without soil rehabilitation) defined by the type of the spontaneous vegetation: (i) Non Regenerated and Non Rehabilitated (NRNR) dominated by herbaceous species and (ii) Regenerated and Non Rehabilitated (RNR) characterized by herbaceous species, shrubby and pioneer tree species covering from 5 to 25% of the soil surface (Table 1). In Georgeon, soils in some areas have also been rehabilitated (filling in of the excavations by bulldozers). We selected 2 new modalities which allow testing the effects of both type of vegetation and soil rehabilitation: Regenerated Rehabilitated (RR), and Non Regenerated Rehabilitated (NRR) (Table 1).

In each site, we also sampled the surrounding natural rainforests (F) that we considered as reference ecosystems because they represent a pre-disturbance condition. These reference forests represent the target that the ecological restoration is designed to attain and a baseline for evaluation of the restoration.

All sites displayed very different soil contents in C (between 0.34 and 5.36% C) and N (between 0.10 and 0.39% N). In particular, %C means in undisturbed forests (F), Regenerated and Non Rehabilitated areas (RNR) and in Non Regenerated and Non Rehabilitated areas (NRNR) were significantly different $(3.47 \pm 0.08, 1.25 \pm 0.05)$ and 0.84 ± 0.04 respectively). We observed the same trend for %N means with 0.27 ± 0.006 , 0.09 ± 0.003 and 0.05 ± 0.002 respectively. There were lower variations in C/N (between 15.40 and 12.30) (Table 1). C/N means in undisturbed forests (F) and mine areas (M) were not significantly different (13.6 ± 0.2) and 14.2 ± 0.1 respectively.

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