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Microbial activities response to As exposure in soil and sediments surrounding a gold mining area at Paracatu—Brazil

E. Sabadini-Santos & E.D. Bidone
Curso de Pós-graduação em Geoquímica Ambiental, Universidade Federal Fluminense (UFF), Brazil

Z.C. Castilhos
Centro de Tecnologia Mineral, Ministério da Ciência, Tecnologia e Inovação (CETEM/MCTI), Brazil

M.A.C. Crapez
Curso de Pós-graduação em Biologia Marinha, Universidade Federal Fluminense (UFF), Brazil

ABSTRACT: Bacterial community exposed to high As concentrations deviate energy from growth to cell maintenance modifying enzymatic activity and increasing its extracellular polymeric substances. Even if the number of bacterial cells presented in soil and sediment samples were in the same order of 10^7 cell cm^{-3}, they declined in soil samples closer to mining area. Dehydrogenase activity (0.6–70.0 μg INT-F g^{-1}) showed the same trend, suggesting inhibition by toxic effect of metals. Opposite behavior was observed to esterase activities (2.2–7.9 μg FDA h^{-1} g^{-1}) that are representative of increasing energy demand by the community under environmental stress. The labile organic matter was mainly carbohydrate (1.4–10.7 mg C g^{-1}) of allochthonous source and its higher concentration in sediment from station 9 explains the rise of esterase activities at that site.

1 INTRODUCTION

Due to their function and ubiquitous presence, bacterial community can act as an environmentally very relevant indicator of pollution. A clear negative influence of metals on microbiological mediated processes and on the structure and diversity of microbial community have been reported worldwide (Beelen & Doelman, 1997; Harrison, 2007). Our objective is to understand the microbial community response to increasing As concentrations, searching for alterations of enzymatic activities and biomass, that should vary according to the development of resistance. This study is part of the environmental and health assessment conducted by Brazilian research institutions under the general coordination of CETEM, at Paracatu municipality, where the largest gold mine in Brazil ("Morro do Ouro") is located.

2 METHODS

2.1 *Sampling and As determination*

The sediments and soils samples were collected during the dry season along the Córrego Rico watershed—its spring is located inside the open pit mine site and at its medium segment is located the Paracatu city: St2 output of the mine area, St1 urban area and St9 downstream of the urban area. Arsenic determinations by ICP-MS (Model 42 IC-MS, Perkin Elmer MS) were done. The detection limit was 0.5 mg kg^{-1}.

2.2 *Microbial analyses*

The total number of bacterial cells (CELL) was quantified by epifluorescent microscopy (Kepner & Pratt, 1994). The activity of dehydrogenase enzymes was measured by the reduction of INT (iodonitrotetrazolium chloride) to INT—formazan (INT-F) in a spectrophotometer (Trevors 1984). Esterase enzymes activities (EST) were analyzed according to Stubberfield & Shaw (1990) based on fluorogenic compounds (fluorescein diacetate—FDA). Concentrations of total biopolymers (carbohydrate, lipids and proteins) were determined by spectrophotometric methods. Carbohydrates (CHO) were quantified according Gerchacov and Hatcher (1972). Lipids (LIP) were analyzed according to Marsh & Wenstein (1966) and proteins (PRT), according to Hartree (1972). The concentrations of CHO, PRT and LIP were expressed in carbon equivalent (mg C. g^{-1}) using the conversion factors 0.40, 0.49 and 0.75 respectively (Fabiano *et al.*, 1995). All determinations

described above were done in triplicates of 1 g of sediment samples.

3 RESULTS AND DISCUSSION

Microorganisms are present in sediments in numbers about 10^{10} cells g^{-1} *d.w.* (Meyer-Reil & Köster, 2000). Their biomass are greater than all other benthic organisms and through their organization in multispecies biofilms, bacteria develop physicochemical gradients where organic matter can be metabolized by hydrolytic enzymes (Flemming & Wingender, 2010). Even if the number of CELL presented in soil and sediment samples were in the order of magnitude of 10^7 cell cm^{-3}, they declined smoothly in soil samples closer to mining area (Figure 1). Bacterial community exposed to high As concentrations deviates energy from growth to cell maintenance functions (Beelen & Doelman, 1997; Harrison, 2007).

DHA shows opposite trend to As concentration in soil and sediment (Figure 1). The activity of dehydrogenase enzymes is based on the fact that INT acts specifically as an artificial electron acceptor, when the succinate dehydrogenase complex in the electron transport chain is reoxidized (Stubberfield & Shaw, 1990). It allows the generation of energy (adenosine triphosphate—ATP), which is only made by viable cells. Consequently, DHA activity declines as the number of bacterial cells reduces or if its activity is inhibited by the toxic effect of metals including As (Beelen & Doelman, 1997; Harrison, 2007).

In contaminated environments, bacteria need more energy to survive, and thus they might be used as sensitive indicators of pollution. This assumption is supported by the increasing trend of EST of the community under high concentrations of As (Figure 1). EST act on biopolymers and transform them into low-molecular-weight organic carbon that could be degraded intracellular. Its increasing trend with arsenic concentration reflects the energy demand rising of the community. The EST activity did not decline at St 9 because of the large amount of organic matter.

Biopolymers present higher concentrations at sediments from St 9. PRT and LIP were lower than CHO, ranging from 0.02 to 0.37 and 0.12 to 0.85 mg C g^{-1}, respectively. CHO varied from 1.35 to 10.65 mg C g^{-1} and represent more than 90% of the biopolymeric carbon (BPC), the sum of PRT, LIP and CHO that represented the amount of bioavailable carbon present in samples (Figure 1). It suggests that this organic matter consist of an allochthonous rather than autochthonous origin and has low nutritional value to local biota.

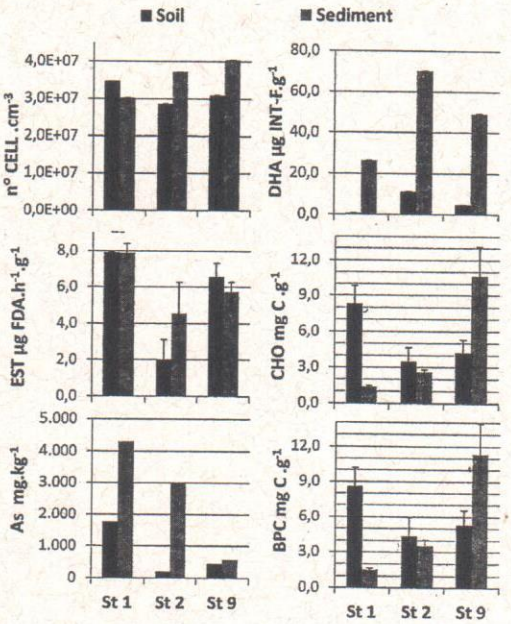

Figure 1. Distribution of bacterial cells, DHA activity, EST activities, As, CHO and CBP concentrations in samples of soil and sediments.

4 CONCLUSIONS

The number of bacterial cells and DHA activity showed a smooth decline tendency in soil samples closer to mining area, suggesting activity inhibition by toxic effect of As. Opposite behavior was observed to EST activities that are representative of increasing energy demand by the community to its maintenance under environmental stress. The labile organic matter was mainly CHO of allochthonous source and its higher concentration in sediment from downstream of the urban area explains the rise of EST activities at that site.

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