

Characterization of redox-related soil microbial communities along a river floodplain continuum by fatty acid methyl ester (FAME) and 16S rRNA genes

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ABSTRACT

Redox states affect substrate availability and energy transformation, and, thus, play a crucial role in regulating soil microbial abundance, diversity, and community structure. We evaluated microbial communities in soils under oxic, intermittent, and anoxic conditions along a river floodplain continuum using fatty acid methyl ester (FAME) and 16S rRNA genes-based terminal-restriction fragment length polymorphism (T-RFLP) bacterial fingerprints. In all the soils tested, microbial communities clustered according to soil redox state by both evaluation techniques. Bacteria were dominant components of soil microbial communities, while mycorrhizal fungi composed about 12% of the microbial community in the oxic soils. Gram-positive bacteria consisted >10% of the community in all soils tested and their abundance increased with increasing soil depth when shifting from oxic to anoxic conditions. In the anoxic soils, Gram-positive bacteria composed about 16% of the total community, suggesting that their growth and maintenance were not as sensitive to oxygen supply as for other microbes. In general, microorganisms were more abundant and diverse, and distributed more evenly in the oxic layers than the anoxic layers. The decrease in abundance with increasing oxygen and substrate limitation, however, was considerably more drastic than the decrease in diversity, suggesting that growth of soil microorganisms is more energy demanding than maintenance. The lower diversity in the anoxic than the oxic soils was attributed primarily to the differences in oxygen availability in these soils.

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1. Introduction

It has long been recognized that redox states play a crucial role in regulating microbial activity and community structure (Lü[demann et al., 2000\)](#page--1-0). Shifting redox states from the oxic surface layers to anoxic subsurface zones leads to development of microhabitats and microbial communities stratifying along the soil profile. Microbial species in the subsurface soils could be strikingly different from those in the surface soil [\(Crocker et al., 2000; Fritze et al., 2000; Blume et al., 2002](#page--1-0)).

Following characterization of 40 strains of G+, aerobic, heterotrophic bacteria isolated from saturated subsurface lacustrine, paleosol and fluvial sediments, [Crocker et al. \(2000\)](#page--1-0) found that most of the subsurface isolates did not cluster with previously established surface soil species in phylogenetic analyses of 16S rRNA gene sequences or with hierarchical cluster analyses of cellular fatty acid profiles.

Although extensive studies have been conducted to reveal microbial communities in surface soils or sediments [\(Kieft](#page--1-0) [et al., 1997; Sahm et al., 1999; Urakawa et al., 2000; Agnelli et al.,](#page--1-0)

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[2004; Sun et al., 2004](#page--1-0);), few have been directed to characterize microbial communities in the transitional redox zones that encounter alternating oxidation–reduction. Such soil ecosystems are prominent in floodplains. As the transformer, sink, source of nutrients, floodplains provide a unique set of various habitats for diverse biota [\(Tockner et al., 1999](#page--1-0)). Periodic erosion and sedimentation processes often lead to remobilization of nutrients, heavy metals, pesticides, and other potential environmental contaminants [\(Walling et al., 1996;](#page--1-0) Krü[ger et al., 2005](#page--1-0)). Some of these processes can have adverse effects on water quality, aquatic habitats, and the agricultural use of the floodplain area (Krü[ger et al., 2005; Noe and Hup,](#page--1-0) [2005\)](#page--1-0).

The floodplain ecosystem of Tisza River in Hungary offers a unique opportunity in the evaluation of redox-related soil microbial communities. Tisza River has a history of periodically flooding. In 2000, 100,000 m^3 of wastewater containing cyanide and heavy metals from an upstream broken tailing dam were released to the river ecosystem [\(Fleit and Lakatos,](#page--1-0) [2003\)](#page--1-0), jeopardizing the sustainability of the ecosystem as the home for many unique florae and faunae in Europe. Subsequently, considerable international attention has been directed to evaluate the Tisza River ecosystem health and function.

In another related study (Unpublished data), we found that the overall changes in ecosystem health and function are gradual, making it challenging to reveal changes that were directly induced by metal contaminations even though the major accidental contamination in 2000 resulted in dead fish floating in the river. Over the years, contaminants were suspended, settled, resuspended, and resettled along the river channel and floodplain. Metal concentrations in the floodplain soils taken in 2003 were relatively low compare to other reported metal contaminated soils [\(Pennanen et al., 1996;](#page--1-0) Konstantinidis et al., 2003; Krüger et al., 2005). For this study, the main goal was to determine impact of redox states on microbial community diversity and structure throughout the soil profile using fatty acid methyl ester (FAME) analysis and

16S rRNA-based terminal-restriction fragment length polymorphism (T-RFLP) fingerprinting. In addition, variations in microbial communities between surface soils and between subsurface soils were also examined.

2. Materials and methods

2.1. Soils

The Tisza river region is located in central Europe with an annual precipitation of 640 mm and temperatures varying from -28 to 22 °C (10–11 °C by average). Soil samples were taken along the Tisza River in October 2003 from two locations, upstream (U, $48^{\circ}13'N$ 22 $^{\circ}33E$ for US1) and downstream (D, 48°11'N 21°44E for DS1), which represent two landscape replications. Soil profiles from both locations show hydromorphic properties and these soils are classified as Meadow-Chernozems [\(FAO-ISRIC-ISSS, 2006](#page--1-0)). At each location, four sampling sites (S) within a radius of approximately 1 km were identified. Sites 1–4 were defined from the river channel towards the floodplain, with their flooding recurrence interval ranging from 1 year to 100 years. At each site, a soil core (5 cm diameter and about 280 cm long) was obtained and was segmented into three samples based on redox states, including oxic (rarely waterlogged), intermittent (seasonally waterlogged), and anoxic (permanently waterlogged) layers. The redox state of the soil was determined based on redoximorphic features [\(Schoeneberger et al., 2002\)](#page--1-0). Generally speaking, the redox zones had two chroma matrix colors; while the reduced zones showed gray depletions in sand and had 0 chroma color in clay. The anoxic layer for soil US3 was not reached following sampling to 281 cm. Field-moist soil samples were stored and transported on ice. Samples were sieved (<2 mm) and mixed thoroughly, and stored in sealed plastic bags at 4 °C. A portion of each sample was air-dried and another portion was freeze-dried. Field-moist soils were used

^a D-Downstream location, U-Upstream location, S-site, Int-Intermittent, and ND-not determined.

^b DS1 was a pasture, located in the midway between the river and the boundary to the first terrace. It is in the local low point of the floodplain. DS2 was a cultivated field located on natural levee along Tisza River. DS3 was a pasture, located near an oxbow that lies at the boundary between the floodplain and first terrace. DS4 was a pasture located on first terrace above the floodplain that has a flooding recurrence interval of 100 years. US1 was a pasture located along an oxbow drainageway near the boundary between the floodplain and first terrace. US2 was planted with hardwood trees and located on a high area of the general landscape in the floodplain. US3 was a cultivated field located about 100 m from Tisza Rive channel, not far from US1 and US2. US4 was a apple orchard located on first terrace above the floodplain that has a flooding recurrence interval of 100 years.

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