



Exploring Microbial Iron Oxidation in Wetland Soils

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Iron is one of the most abundant elements on earth and is essential for life. Because of its importance, iron cycling and its interaction with other chemical and microbial processes has been the focus of many studies. Iron-oxidizing bacteria (FeOB) have been detected in a wide variety of environments. Among those is the rhizosphere of wetland plants roots which release oxygen into the soil creating suboxic conditions required by these organisms. It has been reported that in these rhizosphere microbial iron oxidation proceeds up to four orders of magnitude faster than strictly abiotic oxidation. On the roots of these wetland plants iron plaques are formed by microbial iron oxidation which are involved in the sequestering of heavy metals as well organic pollutants, which of great environmental significance. Despite their important role being catalysts of iron-cycling in wetland environments, little is known about the diversity and distribution of iron-oxidizing bacteria in various environments.

This study aimed at developing a PCR-DGGE assay enabling the detection of iron oxidizers in wetland habitats. Gradient tubes were used to enrich iron-oxidizing bacteria. From these enrichments, a clone library was established based on the almost complete 16s rRNA gene using the universal bacterial primers 27f and 1492r. This clone library consisted of mainly α - and β -Proteobacteria, among which two major clusters were closely related to *Gallionella* spp. Specific probes and primers were developed on the basis of this 16S rRNA gene clone library. The newly designed *Gallionella*-specific 16S rRNA gene primer set 122f/998r was applied to community DNA obtained from three contrasting wetland environments, and the PCR products were used in denaturing gradient gel electrophoresis (DGGE) analysis. A second 16S rRNA gene clone library was constructed using the PCR products from one of our sampling sites amplified with the newly developed primer set 122f/998r. The cloned 16S rRNA gene sequences all represented novel culturable iron oxidizers most closely related to *Gallionella* spp. Based on their nucleotide sequences four groups could be identified, which were comparable to the DGGE banding pattern obtained before with the gradient tubes enrichments.

The above mentioned nested PCR-DGGE method was used to study the distribution and community composition of *Gallionella*-like iron-oxidizing bacteria under the influence of plants species, soil depth, as well as season. Soil samples from Appels, Belgium, an intertidal, freshwater marsh known to hold intensive iron cycling, were taken from 5 different vegetation types in April, July and October 2007. Soil cores were sliced at 1-cm intervals and subjected to chemical and molecular analyses. The DGGE patterns showed that the community of iron-oxidizing bacteria differed with vegetation type, and sediment depth. Samples taken in autumn held lower diversity in *Gallionella*-related iron oxidizers than those sampled in spring and summer.