

Genome Sequence of the *Verrucomicrobium Opitutus terrae* PB90-1, an Abundant Inhabitant of Rice Paddy Soil Ecosystems[∇]

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Bacteria of the deeply branching phylum *Verrucomicrobia* are rarely cultured yet commonly detected in metagenomic libraries from aquatic, terrestrial, and intestinal environments. We have sequenced the genome of *Opitutus terrae* PB90-1, a fermentative anaerobe within this phylum, isolated from rice paddy soil and capable of propionate production from plant-derived polysaccharides.

The *Verrucomicrobia* comprise an excellent example of a bacterial phylum that is hardly detected by conventional cultivation-based approaches. However, these bacteria are frequently encountered in 16S rRNA gene libraries from a wide variety of ecosystems, including aquatic and terrestrial environments (20), as well as the mammalian gastrointestinal tract (5, 19). During the past 5 years, improved cultivation techniques have resulted in the isolation of a range of members of this phylum (8). This resulted in the observation that different lineages of the *Verrucomicrobia* have a compartmentalized cell organization, similar to that of the *Planctomycetes* (14, 15). More recently, it has been suggested that the *Planctomycetes-Verrucomicrobia-Chlamydiae* superphylum may have contributed significantly to eukaryogenesis (18), an innovation of considerable interest in biological evolution.

The availability of a comprehensive set of verrucomicrobial genomes will allow comparative genomic analyses of members from this phylum, providing relevant information in the fields of taxonomy and evolution. It will expand the phylogenetic coverage for the comparison of the metabolic capacities of aquatic, terrestrial, and intestinal populations of verrucomicrobia and their interactions with their environments. Recently, a species of the *Verrucomicrobia*, *Verrucomicrobium spinosum*, was shown to be amenable to transformation (6), paving the way for the development of genetic tools in other members of this phylum.

Opitutus terrae PB90-1 is an obligatory anaerobic member of the phylum *Verrucomicrobia*. It was isolated from rice paddy

soil (4), along with several closely related strains (2, 11). The metabolism of *Opitutus* spp. seems suited for growth on plant-derived (poly)saccharides in this soil and for interaction with methanogens in response to local hydrogen partial pressures (3, 10).

The genome was sequenced using the GS20 454 pyrosequencing platform in combination with Sanger sequencing (7.53× coverage for pyrosequencing, 12.0× coverage for Sanger). A hybrid 454/Sanger assembly was made using the Arachne assembler. Possible misassemblies were corrected, and gaps between contigs were closed by editing in Consed by custom primer walks from subclones or PCR products.

Coding sequences (CDSs) were predicted using Prodigal (9). GenePrimp (17) was used to manually curate start sites and analyze intergenic regions for missed CDSs. Protein descriptions were predicted using Interpro and BlastP. Noncoding genes and miscellaneous features were predicted using tRNAscan-SE (16), RNAMMer (13), Rfam (7), TMHMM (12), and signalP (1).

The genome of *Opitutus terrae* PB90-1 contains a single circular chromosome of 5,957,605 bp. The overall G+C content of the chromosome is 65.3%, less than the 74% predicted by Chin et al. (4). The chromosome contains 4,632 protein-coding genes (CDSs), 65 tRNA genes, and 3 rRNA genes. A putative function could be predicted for 3,102 (67.0%), and signal peptides are predicted in 1,428 (30.8%) protein coding genes.

Nucleotide sequence accession number. The genome sequence of the type strain *Opitutus terrae* PB90-1 is deposited in GenBank under accession number CP001032.

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