The Genome of the Plant Growth-Promoting Rhizobacterium

*Paenibacillus polymyxa* M-1 Contains Nine Sites Dedicated to Nonribosomal Synthesis of Lipopeptides and Polyketides

Ben Niu,1,2 Christian Rueckert,3 Jochen Blom,3 Qi Wang,1,* and Rainer Borriss2*

MOA Key Laboratory of Plant Pathology, Department of Plant Pathology, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, People’s Republic of China;1 Bakteriengenetik, Institut für Biologie, Humboldt-Universität Berlin, Chausseestrasse 117, 10115 Berlin, Germany;2 and Computational Genomics, Center for Biotechnology (CeBiTec), Universität Bielefeld, D-33594 Bielefeld, Germany3

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The complete genome sequence of M-1 consisted of a 5.8-Mb chromosome and a 360-kb plasmid. Nine sites were dedicated to nonribosomal synthesis of lipopeptides and polyketides. Eight of them were located at the chromosome, while one gene cluster predicted to encode an unknown secondary metabolite was present on the plasmid. Plant growth-promoting rhizobacteria (PGPR) have been applied as environmentally friendly alternatives to agrochemicals to improve crop yield and quality (16). The PGPR strains belonging to *Paenibacillus polymyxa* (1) promote plant growth by producing indole-3-acetic acid (IAA) (9) and volatile compounds (14). They are also known to suppress fungal phytopathogens (2, 5–7, 13, 31, 34) and plant-parasitic nematodes (12, 29). Due to its action as a biocontrol agent, *P. polymyxa* produces several peptide antibiotics (3, 8, 10, 11, 15, 18, 20, 22–24, 26–28, 32) which might be important in control of plant pathogens (25).

Strain M-1, isolated from surface-sterilized wheat root tissues, was identified by 16S rRNA gene sequencing and by physiological and biochemical analysis as being *P. polymyxa* (33). It is capable of colonizing root surfaces of wheat, promoting wheat growth, and suppressing wheat sharp eyespot disease. In addition, it acts antagonistically against several phytopathogens in *vitro* by producing antibiotics, including fusaricidin and polymyxin, and by secreting hydrolytic enzymes, for example, endo-β-1,3-glucanase.

Genomic DNA prepared from M-1 was used for construction of a 3-kb-long paired-end library with a GS FLX library preparation kit in combination with GS FLX paired-end adaptors (both from Roche, Mannheim, Germany) according to the manufacturer’s protocol. The reads were assembled using the GS *De Novo* Assembler software program, and the resulting scaffolds were oriented based on the occurrence of unique single nucleotide polymorphisms (SNPs) in the repetitive rRNA (RRN) contigs. In total, 869,907 reads, including 312,451 paired reads, were assembled with a total of 185,008,620 bp. Utilization of the paired-end information allowed scaffolding of the 55 contigs larger than 500 bp into 16 scaffolds containing 45 contigs. Gap closure was done by long-range PCR (using Phusion polymerase; New England BioLabs, Frankfurt [Main], Germany) and subsequent Sanger sequencing (ITB Biotech, Bielefeld, Germany). Prediction of protein-encoding sequences was initially accomplished with the REGANOR server (17). Manual and automatic annotation was done using the annotation software program GenDB 2.4 (19).

The complete genome sequence of M-1 consisted of a circular 5,864,546-bp chromosome and a 366,576-bp plasmid, with G+C values of 54.58% and 37.61%, respectively. Five thousand sixty-one genes (CDS), 14 rRNA operons, and 110 tRNAs resided in the chromosome, while 345 genes were located on the plasmid. Many important genes were found to be plasmid linked, such as those encoding ribosomal proteins and genes involved in replication, repair and methylation, transcription, translation initiation, metabolism of amino acids and carbohydrates, transport, and drug resistance. In addition, several genes related to transposases, phage proteins, and conjugation indicated events of horizontal gene transfer in the plasmids (4).

Nine sites involved in nonribosomal synthesis of secondary metabolites were identified. One gene cluster, 38 kb in size, resided on the plasmid, while the others were present in the chromosome. About 4.5% of the whole M-1 genome was devoted to nonribosomal synthesis of secondary metabolites, including polymyxin and fusaricidin. This is similar to findings for *Bacillus subtilis* (30) but lower than the percentages for *Bacillus amyloliquefaciens* FZB42 and *Streptomyces avermitilis*, reported as being 8.5% and 6.4%, respectively (4, 21).

**Nucleotide sequence accession numbers.** The complete sequences of the *Paenibacillus polymyxa* M-1 main chromosome and of the *Paenibacillus polymyxa* M-1 plasmid pPPM1a have been deposited in EMBL (accession numbers HE577054 and HE577055, respectively).
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REFERENCES