

Genome Sequence of *Bacillus pumilus* S-1, an Efficient Isoeugenol-Utilizing Producer for Natural Vanillin

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***Bacillus pumilus* S-1 is an efficient isoeugenol-utilizing producer of natural vanillin. The genome of *B. pumilus* S-1 contains the epoxide hydrolase and six candidate monooxygenases that make it possible to explore the mechanism involved in conversion of isoeugenol to vanillin in the *B. pumilus* strain.**

Vanillin (4-hydroxy-3-methoxybenzaldehyde) is one of the most important substances widely used in foods, beverages, perfumes, and pharmaceuticals (8). Natural vanillin is extracted from vanilla beans and is much costlier than the chemically synthesized product. Combined with the increasing customer-led demand for natural flavors that are consistent with the attributes “natural” and “healthy,” production of vanillin by extraction cannot meet the growing worldwide market (2). Therefore, growing interest has been focused on producing natural vanillin from other cheap natural sources, such as isoeugenol and ferulic acid, by biotransformation (5, 10, 11).

Bacillus pumilus S-1, which is an aerobic, Gram-positive, spore-forming rod bacterium, is capable of efficiently transforming isoeugenol to vanillin. In a growing culture of *B. pumilus* S-1, 10 g liter⁻¹ isoeugenol was converted to 3.8 g liter⁻¹ vanillin in 150 h, with a molar yield of 40.5% (4). Although some enzymes catalyzing the conversion from isoeugenol to vanillin in the Gram-negative strains were previously identified and purified (11), the enzymes catalyzing this conversion in Gram-positive strains such as *B. pumilus* have not been identified.

Here, we present the draft genome sequence of *B. pumilus* S-1, which was obtained using the Illumina Genome Analyzer IIx system at the Shenzhen Huada Genomics Institute (China) with a paired-end library. A total of 1.75×10^8 filtered reads were assembled using Velvet software (12), resulting in a 3.69-Mb draft genome sequence involved in 144 scaffolds. The open reading frames (ORFs) were predicted using the RAST server (1) and Glimmer 3.0 software (3). The ORFs was analyzed using BLAST to search the nonredundant protein database, and functional determinations were performed using the Clusters of Orthologous Genes (COGs) (9) and Kyoto Encyclopedia of Genes and Genomes (KEGG) databases. Genes encoding tRNA and rRNA

were identified using tRNAscan-SE (7) and RNAmmer (6) software, respectively.

The draft genome sequence of *B. pumilus* S-1 comprises 3,692,744 bp, with a G+C content of 41.26%. There are 3,886 ORFs predicted in the genome, among which 2,877 ORFs have positive biology functions. Strain S-1 has complete Embden-Myerhoff-Parnas, pentose phosphate, and tricarboxylic acid (TCA) cycle pathways. In addition, strain S-1 contains an abundance of predicted transport systems, particularly those associated with various phosphotransferase systems and ABC transporters, suggesting that strain S-1 has the ability to use a variety of carbohydrates. Monooxygenases and epoxide hydrolases were considered to be the most probable enzymes capable of converting isoeugenol to vanillin (4). In the genome of strain S-1, we found the ORFs encoding epoxide hydrolase and six candidate monooxygenases, enabling exploration of the mechanism of isoeugenol-vanillin conversion in the strain.

Besides its versatile characteristics with respect to carbohydrate metabolism, the *B. pumilus* S-1 genome includes a large number of genes relevant to DNA repair and adaptation to stress conditions such as oxidative stress and heat. Further research may explain how the strain obtains these genes and how the genes allow it to adapt to various unfavorable conditions.

Nucleotide sequence accession number. The sequence determined in this whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under accession no. AGBY00000000.

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