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## Complete genome sequence and comparative genome analysis of the *Paenibacillus mucilaginosus* KO2



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#### ABSTRACT

*Aim: Paenibacillus mucilaginosus (P. mucilaginosus)* K02 is implicated in mineral weathering. However, relevant molecular mechanisms remain obscure. The study aims to uncover the bacterium's physiological processes using genomic approaches.

Methods and results: Genomic DNA from *P. mucilaginosus* KO2 was sequenced using high-throughput Solexa sequencing technology and then conducted for Clusters of Orthologous Group (COG) annotation. Thereafter, genome sequences of KO2 were compared with two strains, 3016 and KNP414. Mummer was applied for collinearity analysis of three *P. mucilaginosus* genomes. BLAST was used to identify pan and core genes in these strains. Finally, a phylogenetic tree was constructed using the maximum likelihood method by TreeBeST. Complete genome sequence of *P. mucilaginosus* KO2 indicated the strain comprises one circular chromosome with 8,819,200 bases containing 58.3% GC content and 84.75% coding regions. A total of 7299 predicted ORFs were identified in the genome, among them, several genes were related to carbonic anhydrase (CA), and exopolysaccharide biosynthesis and secretion. Moreover, proteins of the predicted genes were annotated in COG categories such as "Carbohydrate transport and metabolism" and "Inorganic ion transport and metabolism." In comparison with KNP414 and 3016, KO2 exhibited chromosomal recombination or transposition. A total of 6662 core genes were identified among three *P. mucilaginosus* strains. The phylogenomic study indicated that *P. mucilaginosus* KO2 was clustered with *P. mucilaginosus* strains 3016 and KNP414.

Conclusions: In *P. mucilaginosus* KO2, genes related to CA and exopolysaccharide biosynthesis and secretion, and that involved in metabolism-related processes might play significant roles in mineral weathering.

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#### 1. Introduction

Paenibacillus mucilaginosus (synonym Bacillus mucilaginosus, P. mucilaginosus) is characterized as a Gram positive and facultatively anaerobic bacterium [1]. With the ability to fix nitrogen, degrade insoluble soil minerals and release available nutritional

ions, *P. mucilaginosus* has successfully captured researchers' attentions [2,3].

It is commonly known that bio-weathering process is essential for the release of nutrients from rocks [4], and *P. mucilaginosus* is one of the soil bacteria that has been investigated with regard to silicate mineral weathering [5]. Liu et al. isolate one strain of *Paenibacillus*, KT, and confirm this strain has great potential to release potassium from silicate minerals, even from potassium-bearing rock [6]. In addition, Hu et al. identify another two *P. mucilaginosus* strains, KNP413 and KNP414, both of which facilitate to the dissolution of mineral phosphate and potassium [7]. Recently, another study isolate a novel *P. mucilaginosus* strain, M327 (with the proposed name of *Paenibacillus* susongensis sp.

*List of abbreviations:* COG, Clusters of Orthologous Group; NCBI, National Center of Biotechnology Information; PGAAP, Prokaryotic Genomes Automatic Annotation Pipeline; ORF, Open Reading Frame.

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**Table 1**Statistical information of sequences assemble.

	Scaffold	Contig
Total num (#)	89	676
Total length (bp)	9,066,847	8,854,367
N50 (bp)	1,915,011	37,116
N90 (bp)	226,134	6997
Max length (bp)	3,038,726	253,487
Min length (bp)	511	47
GC content (%)	58.18%	58.18%

strain, it has not been well-characterized as the two strains. Although several studies have demonstrated that it facilitates enhance of silicate mineral weathering [12–14], potential mechanisms from gene level or protein level remain obscure. Therefore, in the present study, we performed the full genome sequence of strain KO2, and further investigated the corresponding protein sequences, followed by annotation analysis via alignment against the Clusters of Orthologous Group (COG) database. Moreover, we also conducted a comparative genome analysis on three *P. mucilaginosus* 

#### **COG** function classification

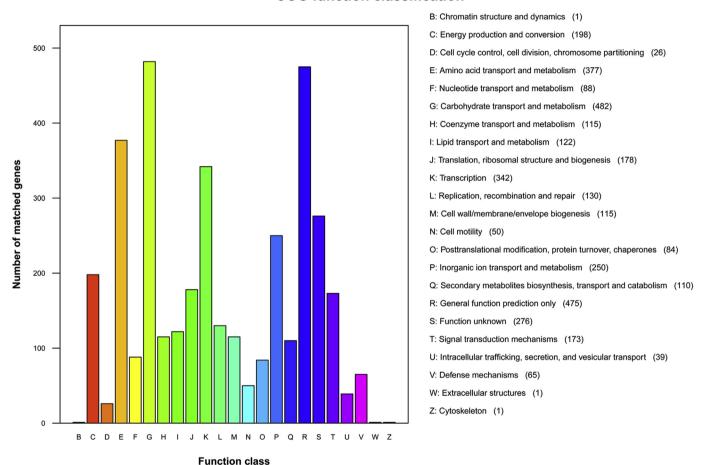


Fig. 1. Clusters of Orthologous Group annotations for the Paenibacillus mucilaginosus KO2 genome.

nov.), which is also a mineral weathering bacterium [8]. However, molecular mechanisms of the related metabolic processes during weathering have rarely been reported, partly due to poor genetic information.

Genomics provides an effective resource for better understanding the *P. mucilaginosus* biological mineral weathering process [9]. Currently, genome sequences of several *P. mucilaginosus* strains have completed, such as 3016 (GenBank accession no. NC\_016935.1), KNP414 (accession no. NC\_015690.1) and K02 (accession no. NC\_017672) [10,11]. Complete genome sequence of 3016 suggests this strain is a circular chromosome, containing a set of crucial genes which are involved in nutrients metabolism-related processes [11], while KNP414 strain harbors eight genes associated with nitrogen assimilation [10]. With regard to K02

strains, 3016, KNP414 and K02, to identify common genes in these strains. With the comprehensive analyses, the study aims to reveal underlying molecular mechanisms on silicate mineral weathering process in K02, and lay a foundation of using this strain as a fertilizer in agriculture.

#### 2. Materials and methods

#### 2.1. Bacterial strain

*P. mucilaginosus* KO2 was deposited in the Environmental Biological Science and Technology Research Center, Institute of Geochemistry, Chinese Academy of Sciences and College of Life Science, Nanjing Normal University. *P. mucilaginosus* KO2 was

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