Complete genome of *Brachybacterium* sp. P6-10-X1 isolated from deep-sea sediments of the Southern Ocean

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**A B S T R A C T**

*Brachybacterium* sp. P6-10-X1 is a rare actinobacterium isolated from deep-sea sediments in the Southern Ocean. To explore the potential of natural product biosynthesis, the genome was completely sequenced. It contained a circular chromosome of 4,385,603 bp with an average GC content of 70.9%. Genome mining revealed four biosynthetic gene clusters potentially producing new natural products.

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**1. Introduction**

Actinobacteria are a group of economically and biotechnologically priceless bacteria, due to the abilities to produce natural products of pharmaceutical interests and enzymes of industrial applications. Since known natural products have been frequently re-isolated from the mostly investigated genus *Streptomyces*, other actinobacterial lineages (referred to as rare actinobacteria) attract increasing attention for discovering novel compounds (Azman et al., 2015). The genus *Brachybacterium* is one of the rare actinobacterial lineages. *Brachybacterium* species have been isolated from various environments including oil-contaminated coastal sand (Chou et al., 2007), salt-fermented seafood (Park et al., 2011), lake sediment (Liu et al., 2014) and even medieval wall paintings (Heyrman et al., 2002). In addition, *Brachybacterium* contained unique polyketide synthase (PKS) genes (Selvin et al., 2016), and showed antimicrobial activities (Undabarrena et al., 2016). It suggests that *Brachybacterium* is an unexplored resource for discovering new natural products.

However, the genus has been largely understudied. To date, only one strain, *Brachybacterium faecium* DSM 4810, had its genome completely sequenced (Lapidus et al., 2009). In addition, there were 8 draft genomes available in the GenBank database. Considering the importance and potential biotechnological applications of the genus, more representatives should be investigated, especially the ones from extreme environments. The Southern Ocean represents the most extreme marine environments, expected to host microorganisms with special adaptation abilities and genetic resources producing novel bioactive compounds. Here, we present the genome sequencing and data mining of *Brachybacterium* sp. P6-10-X1, isolated from marine sediments at water depth of 465 m in the Southern Ocean.

**2. Data description**

The strain P6-10-X1 was isolated by the standard dilution plating technique on marine agar 2216 from sediments collected by a box-corer during the 29th Chinese Antarctic Scientific Expedition. The strain can grow at temperatures from 4 °C to 37 °C with the salinity from 0 to 3‰. It formed round and pale yellow colony on Marine Agar 2216 plates (Table 1). The most closest type strain was *Brachybacterium fresconis* LMG 20336\(^{T}\), sharing 99.45% 16S rRNA gene sequence identity. Genomic DNA was extracted using QIAamp® DNA Mini kit (QIAGEN). The genome was sequenced on both the Illumina HiSeq 4000 and the PacBio RSII sequencing platforms (Table 1). The most closest type strain was *Brachybacterium fresconis* LMG 20336\(^{T}\), sharing 99.45% 16S rRNA gene sequence identity. Genomic DNA was extracted using QIAamp® DNA Mini kit (QIAGEN). The genome was sequenced on both the Illumina HiSeq 4000 and the PacBio RSII sequencing platforms (Table 1). DNA was randomly interrupted into fragments of approximately 600 bp and 10 kb to construct libraries for the Illumina paired-end sequencing and PacBio RSII sequencing platforms (Table 1). DNA was randomly interrupted into fragments of approximately 600 bp and 10 kb to construct libraries for the Illumina paired-end sequencing and PacBio sequencing, respectively. Up to 670 Mb and 761 Mb clean data from the Illumina and the PacBio sequencing were assembled using SOAPdenovo software (Luo et al., 2012), and RS_HGAP assembly version 3 [https://github.com/PacificBiosciences/SMRT-Analysis](https://github.com/PacificBiosciences/SMRT-Analysis), respectively. The assembled genome contained a circular chromosome of 4,385,603 bp, with an average GC content of 70.9% (Fig. 1 and Table 2). A total of 4045 genes were predicted by Glimmer 3.0.2 (Delcher et al., 2007).

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hypothetical proteins. In addition, a total of 2467 (61.0%), 2622 (64.8%), 3187 (78.8%), 2238 (55.3%), and 174 (4.3%) genes were annotated by querying the Gene Ontology, KEGG, Cluster of Orthologous Groups (COGs), Swiss-Prot and the CAZy databases, respectively. There were 3 rrn operons and 50 tRNAs responsible for 21 amino acids, predicted by rRNAmmer (Lagesen et al., 2007) and tRNAscan (Lowe and Eddy, 1997), respectively.

The strain has complete central metabolism pathways for glycolysis, the TCA cycle, fatty acid biosynthesis and metabolism, and pentose phosphate pathway etc., which can provide precursors such as amino acids, acetyl-CoA and malonyl-CoA for secondary metabolite biosynthesis. Four putative secondary metabolite biosynthetic gene clusters were predicted by the antiSMASH program (Supplementary Table 1) (Blin et al., 2013). The cluster 1 belonged to siderophore type, existing in the aerobic and facultative anaerobic bacteria and fungi (Neilands, 1995). It matched the desferrioxamine B biosynthetic gene cluster from Strep- tomyces griseus subsp. griseus NBRC 13350, sharing three similar genes encoding monoxygenase (BH708_07410), pyridoxal-dependent decarboxylase (BH708_07415) and siderophore biosynthetic protein (BH708_07405). The amino acid sequence similarities were between 47 and 52%. Siderophores have great potential in clinical and agricultural applications, in addition to beneficial iron transport (Neilands, 1995). For example, the mesylate salt of desferrioxamine B is marketed as medicine Desferal for treatment of iron overload in human (Go’mez et al., 2004). Therefore, Brachybacterium sp. P6-10-X1 has the potential to produce novel siderophores for pharmaceutical applications.

The cluster 2 was predicted to produce ectoine, which was first discovered in an extremely halophilic phototrophic bacterium, Halorhodospira halochloris, isolated from Wadi Natrun, Egypt (Lentzen and Schwarz, 2006). The cluster 2 shared three similar genes with ectoine biosynthetic gene cluster from Streptomyces chrysomallus, including genes encoding diaminobutyrate acetyltransferase (BH708_08975), aminotransferase class-III (BH708_08980), and L-ectoine synthase (BH708_08985). The amino acid sequence similarities were between 50 and 63%. Ectoine is a kind of compatible solute protecting cells under high osmotic pressure, and has been used as a moisturizer in cosmetics (Motitschke et al., 2000). Therefore, the biosynthesis and applications of ectoine from the Antarctic Brachybacterium are worthy of further research.

The cluster 3 belonged to terpene type, the largest group of natural products. Terpenes have potential applications in cosmetic, pharmaceutical and biotechnology industries (Gershenzon and Dudareva, 2007). The cluster 3 matched the carotenoid biosynthetic gene cluster from...
Brevibacterium linens, sharing five similar genes encoding hypothetical protein (BH708_13335), betacarotene desaturase (BH708_13350), lycopene cyclase (BH708_13360), lycopene cyclase (BH708_13365), and cytochrome P450 (BH708_13345). The amino acid sequence similarities were between 52 and 65%. Further experiments are encouraged to verify whether the cluster 3 synthesizes carotenoid-like terpenes. In addition, the cluster 4 was predicted to encode type III PKS. However, no known gene clusters were found to be similar with this cluster, indicating its novelty.

In summary, Brachybacterium sp. P6-10-X1 represents the first completely sequenced Brachybacterium isolated from the Southern Ocean. It shows promising genetic abilities to produce new natural products of potential biotechnological applications. The genomic data mining helps further biosynthetic studies to explore the predicted natural products from the Antarctic rare actinobacterium.

Culture deposition and nucleotide sequence accession number

Brachybacterium sp. P6-10-X1 was deposited to the China Center for type Culture Collection (CCTCC) with the accession number CCTCC AB 2017016. The complete genome sequence of Brachybacterium sp. P6-10-X1 has been deposited in the GenBank under accession number CP017297.

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.margen.2017.04.001.

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