



# Draft Genome Sequences of Sporulation-Impaired *Bacillus pumilus* Strain NRS576 and Its Native Plasmid p576

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**ABSTRACT** *Bacillus pumilus* spores can cause foodborne poisonings. *B. pumilus* strain NRS576 forms spores with a very reduced efficiency due to the presence of a plasmid, named p576. Here, we report the genome sequence of strain *B. pumilus* NRS576 and its plasmid p576.

Organisms in the *Bacillus* genus are Gram-positive bacteria that can form spores resistant to radiation, heat, and chemicals. *Bacillus pumilus* is present in soil samples, but some strains are associated with food poisoning, and spore formation is relevant to its pathogenicity (1–3). *B. pumilus* NRS576 forms spores with low efficiency due to a plasmid (4), p576, which we have sequenced previously (5). Here, we report the chromosomal and plasmid sequences of strain NRS576 obtained from the *Bacillus* Genetic Stock Center. A total DNA sample (6), obtained from cells growing in LB medium at 37°C, was used for sequence determination with the Illumina MiSeq platform. DNA libraries were prepared with the NEBNext DNA library prep kit for Illumina (New England Biolabs). Briefly, 1 µg DNA was sonicated, and fragments of ~675 or 1,075 bp were selected. DNA ends were repaired, A-tailed, and ligated to adapters. Next, fragments were PCR amplified (8 cycles) and quality checked on a DNA 7500 chip on a 2100 Bioanalyzer (Agilent). Library sizes of ~800 and 1,200 bp were isolated, validated (Bioanalyzer), and titrated with quantitative PCR (qPCR). After denaturation, the libraries were seeded on a flow cell (MiSeq v2, 2 × 150 bp) at a density of 16 pM. The sequencing rendered 3,963,212 (800-bp library) and 2,015,074 (1,200-bp library) paired-end reads. Data processing was done with default parameters. Adapters (Cutadapt [7]) and low-quality sequences (Sickle 1.33 [8]) were removed. After verifying the quality of the processed data (FastQC [9]), *de novo* assembly was performed (SPAdes v3.9.1 [10–13]). Three apparent extrachromosomal elements were detected with plasmidSPAdes (13), (i) plasmid p576, (ii) bacteriophage phiX174 (added as a control for amplification and sequencing [14]), and (iii) sequences similar to part of the *Brevibacillus laterosporus* DSM25 *bogC* gene cluster. PhiX174 sequences and contigs smaller than 250 bp were removed, and sequences similar to *bogC* were considered genomic DNA. General statistics obtained with the bioinformatic tool Quast (15) gave an  $N_{50}$  value of 313,965 bp and an  $L_{50}$  value of 5.

One 43,328-bp contig corresponded to p576, which is 106 bp smaller than previously determined (5). The plasmid p576 contains direct-repeat sequences. The apparent deletions correspond to these repeats. Otherwise, the p576 sequences differ in one single base pair (gene 64 codon 492 [GAA462GGA]). We sequenced p576 and genomic DNA with mean coverages of about 70 and 30, respectively, and this implies that p576 has a copy number of two. The previously published p576 nomenclature (5) was respected. Sanger (5) and next-generation sequencing (NGS)-determined p576 sequences are publicly available.

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NRS576 genomic sequences were located on 32 contigs with a total length of 3,675,031 bp and 41.6% GC content. Based on our annotation with PROKKA (16), the genome contains 3,811 putative genes, distributed as 3,641 coding DNA sequences (CDS), 86 noncoding RNAs (ncRNA), 73 tRNAs, 10 rRNAs, and 1 transfer-messenger RNA (tmRNA).

**Data availability.** This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under accession number [UWJF00000000](https://www.ncbi.nlm.nih.gov/assembly/GCA014481.1). The version described here is the first version. Raw sequence reads have been submitted to the Sequence Read Archive (SRA) (accession numbers [ERR2811649](https://www.ncbi.nlm.nih.gov/sra/ERR2811649) and [ERR2811650](https://www.ncbi.nlm.nih.gov/sra/ERR2811650), corresponding to  $2 \times 150$ -bp paired-end sequences of 800- and 1,200-bp fragments, respectively). The p576 sequences are available under DDBJ/ENA/GenBank accession numbers [LR026976](https://www.ncbi.nlm.nih.gov/nuccore/LR026976) (Sanger) and [LR026977](https://www.ncbi.nlm.nih.gov/nuccore/LR026977) (NGS).

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We declare no conflict of interest.

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