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Draft Genome Sequences of Four Thermophilic Spore Formers Isolated from a Dairy-Processing Environment

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Spores of thermophilic spore-forming bacteria are a common cause of contamination in dairy products. Here, we report draft genome sequences of four thermophilic strains from a milk-processing plant or standard milk, namely, a *Geobacillus thermoglucosidans* isolate (TNO-09.023), *Geobacillus stearothermophilus* TNO-09.027, and two *Anoxybacillus flavithermus* isolates (TNO-09.014 and TNO-09.016).

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One of the regular problems in the production of dairy concentrates is contamination by heat-resistant spores from thermophilic bacteria of the genera *Anoxybacillus* and *Geobacillus* (1). Genome sequences of two thermophilic strains previously isolated from fouling samples from two dairy-production plants (1) and with relatively strong biofilm-forming capacities, namely, *Geobacillus thermoglucosidans* TNO-09.020 and *Anoxybacillus flavithermus* TNO-09.006, were reported previously (2, 3). Here, we publish the draft genome sequences of an additional set of four dairy thermophiles from a published study (1).

G. thermoglucosidans TNO-09.023, *G. stearothermophilus* TNO-09.027, *A. flavithermus* TNO-09.014, and *A. flavithermus* TNO-09.016 were pregrown on tryptic soy agar (TSA) plates (at 55°C overnight, in plastic bags with wet tissues to prevent evaporation). Genomic DNA was isolated from pelleted cell material of freshly grown flask cultures in tryptic soy broth (TSB) (at 55°C, with optical density at 600 nm [OD_{600}] of 0.8 to 1.0), inoculated from a single representative freshly grown colony from a TSA plate. The method involved a mild degradation step of cell walls by lysozyme at 37°C, followed by lysis of cells by the addition of sarcosyl. Proteins were removed by extraction with phenol-chloroform, RNA was degraded using RNase, and the resulting DNA was precipitated and washed with isopropanol (50% [vol/vol]) and 70% (vol/vol) ethanol, respectively (4). DNA was dissolved in water (1 to 9 μg/μl; size on 0.8% agarose gel, ≥20 kb) and used for sequencing.

The isolated DNA was sheared to 250- to 350-bp fragments and paired-end sequenced on an Illumina HiSeq 2000 outsourced to BaseClear (Leiden, The Netherlands). Ray 2.3.1 (5) was used for assembly. The RAST server (6) was used to annotate the genomes.

Accession number(s). The genome sequences of the four strains have been deposited at DDBJ/EMBL/GenBank under the accession numbers listed in Table 1. The version described in this paper is the first version.

TABLE 1 Sequenced strains and their sources

Species	Strain	Source of isolation	Accession no.
Geobacillus thermoglucosidans	TNO-09.023	Casein pipeline	LUCT00000000
Geobacillus stearothermophilus	TNO-09.027	Casein pipeline	LUCR00000000
Anoxybacillus flavithermus	TNO-09.014	Standard milk	LUFB00000000
Anoxybacillus flavithermus	TNO-09.016	Evaporator	LUCQ00000000

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