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# Draft Genome Sequence of the Thermoalkaliphilic *Caldalkalibacillus thermarum* Strain TA2.A1[▽]

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**The genes and molecular machines that allow for a thermoalkaliphilic lifestyle have not been defined. To address this goal, we report on the improved high-quality draft genome sequence of *Caldalkalibacillus thermarum* strain TA2.A1, an obligately aerobic bacterium that grows optimally at pH 9.5 and 65 to 70°C on a wide variety of carbon and energy sources.**

Thermoalkaliphilic microorganisms are poorly understood as a group, and there is very little information available on the genes and molecular machines that allow for a thermoalkaliphilic lifestyle. To live at high pH and temperature, akin to a proton desert, these bacteria must overcome a number of thermodynamic challenges. These include capturing and retaining protons from an alkaline environment (pH 9.5) to drive endothermic reactions in the cell membrane (2), increased membrane permeability to protons at high temperature (16), and a low protonmotive force that appears to be suboptimal for growth (12). Studies of the membrane-bound $F_1F_o$-ATP synthase of *Caldalkalibacillus thermarum* strain TA2.A1 (formerly *Bacillus* sp. strain TA2.A1) have revealed a number of unique adaptations that enable this enzyme to function at extremes of pH and temperature by using protons as coupling ions (2, 8, 9, 11, 13–15). Additionally, thermoalkaliphilic bacteria with a respiratory metabolism have an obligate growth requirement for iron (10). At alkaline pH, the solubility constant for iron decreases far below the requirement for living cells, and the concentration of bioavailable iron is estimated to be approximately $10^{-23}$ M at pH 10 (3), suggesting that thermoalka-liphilic bacteria must possess powerful, yet undiscovered, mechanisms to sequester iron. Despite these apparent thermodynamic problems and the challenge of severe iron limitation, thermoalkaliphilic bacteria grow rapidly (doubling time $< 60$ min) under these conditions, demonstrating that these bacteria are superbly adapted to combat these challenges.

The sequencing of genomes from representative thermoalkaliphilic bacteria is required to identify and characterize genomic, biochemical, metabolic, and physiological properties responsible for microbial growth at high temperature and pH. To address this objective, we sequenced the genome of the thermoalkaliphilic aerobic bacterium *Caldalkalibacillus thermarum* strain TA2.A1. *C. thermarum* strain TA2.A1 grows optimally at temperatures of 65 to 70°C at pH 9.5 and was isolated from an alkaline thermal bore at Mount Te Aroha, New Zealand. *C. thermarum* strain TA2.A1 grows on sucrose, common $C_4$-dicarboxylates, glutamate, pyruvate, and trehalose; however, glucose and fructose fail to support robust growth (12–13).

Genomic DNA of *C. thermarum* strain TA2.A1 was isolated as previously described (6). The genome size is ~2.986 Mb, which is based upon an average of ~18× genome coverage for 454 GS FLX shotgun data and ~216× coverage for 36-bp Illumina paired-end data from a library with an average insert size of 350 bp. It is defined as a "high-quality draft," which is a high-quality assembly with automated improvements (1), and production methods have been described previously (4, 7).

The *C. thermarum* strain TA2.A1 draft genome consists of 247 contigs, with an average G+C content of 47.5%. A total of 3,105 protein-coding genes were predicted by the Prodigal algorithm (5). The TA2.A1 genome sequence will allow the genes and molecular machines of this thermoalkaliphile and others to be studied in greater detail.

**Nucleotide sequence accession number.** The whole-genome shotgun project has been deposited in DDBJ/EMBL/GenBank under accession number AFCE00000000. The nucleotide and protein sequences, the annotated genome sequence, a metabolic reconstruction, and various query tools such as BLAST are available at the Computational Biology at Oak Ridge National Laboratory (ORNL) website (http://genome.ornl.gov/microbial/guest/TA2_3/).

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