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GENOME ANNOUNCEMENT

Complete Genome Sequence of the Beer Spoilage Organism *Pediococcus claussenii* ATCC BAA-344^T

Vanessa Pittet,[a] Teju Abegunde,[b] Travis Marfleet,[a] Monique Haakensen,[a]* Kendra Morrow,[a] Teenus Jayaprakash,[b] Kristen Schroeder,[a] Brett Trost,[b] Sydney Byrns,[a] Jordyn Bergsveinson,[a] Anthony Kusalik,[b] and Barry Ziola[a]

Department of Pathology and Laboratory Medicine[a] and Department of Computer Science,[b] University of Saskatchewan, Saskatoon, Saskatchewan, Canada

***Pediococcus claussenii* is a common brewery contaminant. We have sequenced the chromosome and plasmids of the type strain *P. claussenii* ATCC BAA-344. A ropy variant was chosen for sequencing to obtain genetic information related to growth in beer, as well as exopolysaccharide and possibly biofilm formation by this organism.**

Lactobacilli and pediococcal isolates are frequently found as brewery contaminants. Growth of these bacteria in beer results in a turbid and unpalatable (i.e., spoiled) product, causing economic losses for brewers. As the ability to grow in beer is not a species-conserved trait (7), identification of beer-spoiling isolates is difficult. An understanding of the genetics that allow given isolates of a species to grow in beer is therefore needed to accurately predict the spoilage potential of these organisms when found in a brewery.

Pediococcus claussenii is a common beer spoilage organism (2). The type strain (ATCC BAA-344), which we sequenced, produces exopolysaccharide (referred to as rope, or slime), which may be involved in biofilm formation and persistence in a brewery. This bacterium spoils beer in 7 to 8 days and contains the known beer spoilage-associated gene *horA* (7). Ethanol tolerance for lactic acid bacteria is midrange (9), and hops resistance is high (V. Pittet, B. R. Bushell, and B. Ziola, unpublished data).

Sequencing was done using ABI 3730xl (6× coverage) and Illumina Solexa 1G (250× coverage) sequencing platforms. Sixteen million paired Illumina reads were assembled using Velvet (13). The resulting 1,757 contigs were combined with 1,500 fosmid and 14,600 plasmid Sanger reads (paired), and assembled using Phred/Phrap/Consed (3, 4, 5). The resulting 56 contigs were then validated and visualized using amosvalidate and Hawkeye, respectively (8, 12). Read pair data were used to order and orient contigs, and gaps were closed by ABI 3730xl sequencing of PCR amplicons. The genome consists of the bacterial chromosome (1,829,111 bp) and eight plasmids (pPECL-1 [1,815 bp], pPECL-2 [2,450 bp], pPECL-3 [17,830 bp], pPECL-4 [23,136 bp], pPECL-5 [36,338 bp], pPECL-6 [18,800 bp], pPECL-7 [16,067 bp], and pPECL-8 [currently 20,815 bp]). The sixth and seventh plasmids were not circularized due to repetitive transposon regions found in both plasmids, making PCR-based gap closing very difficult. Efforts to complete the eighth plasmid are ongoing. The overall G+C content of the genome is 36.8%, whereas that of the plasmids ranges from 34.9% to 42.5%.

Annotation was done using combined results from RAST (1) and the Institute for Genome Sciences Annotation Engine and manual annotation tool Manatee. The results predicted 1,740 coding sequences (CDS), 57 tRNA genes, and 4 rRNA operons in the chromosome and 129 CDS in the plasmids (2, 2, 20, 16, 35, 21, 15, and currently 18 CDS for plasmids 1 to 8, respectively). Approximately 30 genes are related to mobile elements (e.g., phage and transposons); however, no CRISPR locations were predicted (6). Interestingly, as in *Lactobacillus reuteri*, all genes required for synthesis of vitamin B_{12} are present (11). From the point of view of beer spoilage, several genes are of interest, including the already-known hops resistance gene *horA* (10), found on pPECL_8. As additional genome sequences from beer-spoiling bacteria become available, genomic comparisons will help elucidate the mechanisms by which these organisms grow in beer, with the end application for breweries being the rapid detection of contaminants.

Nucleotide sequence accession numbers. The *P. claussenii* ATCC BAA-344^T sequences were deposited in GenBank under accession numbers CP003137, CP003138, CP003139, CP003140, CP003141, CP003142, CP003143, CP003144, and CP003145 for the chromosome and plasmids 1 to 8, respectively.

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Address correspondence to Barry Ziola, b.ziola@usask.ca.

* Present address: Contango Strategies, Saskatoon, Saskatchewan, Canada.

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