

Research team first to fully sequence bacterial genome important to fuel and chemical production

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Sagar Utturkar, coauthor of high-rating biotechnology paper, in the team's Oak Ridge National Laboratory lab.

Researchers at the Department of Energy's Oak Ridge National Laboratory are the first team to sequence the entire genome of the *Clostridium autoethanogenum* bacterium, which is used to sustainably

produce fuel and chemicals from a range of raw materials, including gases derived from biomass and industrial wastes.

The ORNL work was funded by LanzaTech, a biotechnology company based in Illinois with an innovative carbon recycling process.

LanzaTech's gas fermentation platform uses proprietary microbes for efficiently converting carbon-rich waste gases and residues into useful fuels and chemicals.

Successfully sequencing *Clostridium autoethanogenum*—classified as a complex, class III microbe because of its many repeating units of DNA bases—has been of significant interest to the biotechnology industry. A Biotechnology for Biofuels paper co-authored by ORNL's Steve Brown and Miriam Land, University of Tennessee doctoral student Sagar Utturkar and collaborating LanzaTech researchers generated a top-5-percent rating from Altmetric, an online rating system that measures the volume and value of recognition an article receives from research communities and media outlets.

"With the complete genomic sequence, we will have a better understanding of the microbe's metabolism and mutations that will enable LanzaTech to make modifications to the wild-type, or naturally occurring, strain for optimizing the conversion of waste into fuel," Brown said. "Our ORNL lab has a lot of experience sequencing genomes, and we have the analytic capability to tackle this project."

The research team sequenced the more than 4.3 million base pairs of DNA that make up the organism's genome using RS-II long-read sequencing technology developed by Pacific Biosciences (PacBio).

Although long-read sequencing technologies still struggle with high error rates, they promise to advance the [biotechnology industry](#) by making it possible to sequence microorganisms with many repeating sequences,

such as *Clostridium autoethanogenum*, within a reasonable amount of time at reasonable cost. The ORNL team performed a greater number of reads and used data algorithms to correct for errors associated with the long-read technology. The team also compared the RS-II long-read results to two short-read technologies, concluding the short-read technologies were unable to sequence the entire genome because of the bacterium's repetitive sequences, as expected.

"In our paper we compared three generations of sequencing technologies and explained why the long-read technology was able to finish the genome," Brown said. "Now, ORNL is independently looking at six different organisms using PacBio to compare and contrast experiences using this technology."

The project also revealed information about the genetic history of *Clostridium autoethanogenum* through short DNA sequences known as CRISPR systems, which retain genetic mutations such as those created during a viral infection that are subsequently passed on to future generations of a microbe. CRISPR systems are important indicators of strengths and vulnerabilities that biotechnology companies like LanzaTech look for when genetically modifying a microbe.

More information: "Comparison of single-molecule sequencing and hybrid approaches for finishing the genome of *Clostridium autoethanogenum* and analysis of CRISPR systems in industrial relevant Clostridia," Steven D Brown et al. *Biotechnology for Biofuels* 2014, 7:40 [DOI: 10.1186/1754-6834-7-40](https://doi.org/10.1186/1754-6834-7-40)

Provided by Oak Ridge National Laboratory

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