Title
Development of a High Efficiency Anaerobic Electroporation Apparatus for Biofuel Generation from Waste Gases

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Cooperative Research and Development Agreement (CRADA) Final Report

Report Date: 02/15/2022

In accordance with Requirements set forth in the terms of the CRADA, this document is the CRADA Final Report, including a list of Subject Inventions. It is to be forwarded to the DOE Office of Scientific and Technical Information upon completion or termination of the CRADA, as part of the commitment to the public to demonstrate results of federally funded research.

Parties to the Agreement: LanzaTech Inc and Berkeley Lab

CRADA number: FP00007280

CRADA Title: TCF High Efficiency Anaerobic Electroporation

Responsible Technical Contact at Berkeley Lab: Nathan Hillson

Name and Email Address of POC at Partner Company(ies): Michael Koepke, Michael.Koepke@lanzatech.com

Sponsoring DOE Program Office(s): Office of Energy Efficiency and Renewable Energy

LBNL Report Number: LBNL-2001449

OSTI Number: [SPO to complete]

Joint Work Statement Funding Table showing DOE funding commitment:

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
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<tbody>
<tr>
<td>DOE Funding to LBNL</td>
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<tr>
<td>Participant Funding to LBNL</td>
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<td>Participant In-Kind Contribution Value</td>
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<td>Total of all Contributions</td>
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</tr>
</tbody>
</table>

Provide a list of publications, conference papers, or other public releases of results, developed under this CRADA: (Publications must include journal name, volume, issue, Digital Object Identifier)

None.

Provide a detailed list of all subject inventions, to include patent applications, copyrights, and trademarks:
Executive Summary of CRADA Work:
The Joint BioEnergy Institute (JBEI) researchers were co-inventors of the technology that will be used on this project and have developed a more current version of the chip and controller. JBEI will also assist with the design of the pathways and implementation of pathways on the chip. LanzaTech has developed novel gas fermentation technology that captures and utilizes greenhouse gases for production of fuels and chemicals. In contrast to traditional fermentation that uses sugars as substrate (and releases CO2 as a byproduct), gas fermentation utilizes C1 substrates carbon monoxide (CO) or CO2. This enables a diverse range of feedstock options including waste gases from industrial sources (e.g., steel mills and processing plants) or syngas generated from any biomass resource (e.g., agricultural waste, municipal solid waste, or organic industrial waste). Biomass is then gasified, allowing for maximum yields and complete carbon utilization including the recalcitrant lignocellulosic fraction that cannot be utilized in traditional sugar fermentation.

To maximize the value that can be added to the array of gas resources that the LanzaTech process can use as an input, LanzaTech has pioneered genetic modification of acetogens and developed a comprehensive set of genetic tools to perform routine strain modification, including genome editing tools as CRISPR/Cas9 and libraries of validated genetic parts as promoters and terminators. Using this platform, production of over 50 new molecules have been demonstrated directly from gas. For a few selected molecules production rates and yields have been optimized and surpass production of native producers and engineered E. coli or yeast strains, but a higher throughput approach for combinatorial optimization of pathways is required to further advance synthesis of additional products in parallel.

Summary of Research Results:

1. Matured the technology through testing by a commercial partner to get it closer to commercialization.
2. Developed a fully-integrated system that can be used for optimization of engineering of anaerobic organisms.
APPENDIX A (Reference Only)

This appendix has been developed by DOE to assist DOE Labs in drafting the **Executive Summary** and **Summary of Research Results** sections of the CRADA Final Report.

**Executive Summary of CRADA Work:**

Include a discussion of 1) how the research adds to the understanding of the area investigated; 2) the technical effectiveness of the materials, methods or techniques investigated or demonstrated, and their economic feasibility, if known; and 3) how the project is otherwise of benefit to the public. The discussion should be a minimum of one paragraph and written in terms understandable by an educated layman.

**Summary of Research Results:**

- **INCLUDE, IF APPLICABLE:** "This product contains Protected CRADA Information, which was produced on [DATE] under CRADA No. [###-###] and is not to be further disclosed for a period of [up to and not to exceed] five (5) years from the date it was produced except as expressly provided for in the CRADA."
- Summarize project activities for the entire period of performance, including original hypotheses, approaches used, problems encountered, any departure from planned methodology, and an assessment of their impact on the project results. Incorporate technical data, e.g., facts, figures, analyses, and assumptions used during the life of the project to support the technical conclusions of the work. It is acceptable to incorporate the technical data by reference to other publicly available sources, such as any publications or other reports, but not websites. Provide a comparison of the actual accomplishments with the goals and objectives of the project. Where possible, the summary should cover each task listed in the Statement of Work (SOW) and should note any deviations from the project plan, or lack of technical data.
- Identify products, potential applications, and technology transfer activities developed under the CRADA, including those completed and anticipated at the time of the report. These include, but are not limited to: 1) networks or collaborations fostered; 2) technologies/techniques/methodologies; 3) other products that reflect the results of the project, such as commercial products, internet sites, data or databases, physical collections, audio or video, software, models, educational aid or curricula, and instruments or equipment.

Note: Recommended characteristics of Scientific and Technical Information reports can be found at https://www.osti.gov/stip/attributes