U.S. Department of Energy (DOE)
Bioenergy Technologies Office (BETO)
2017 Project Peer Review

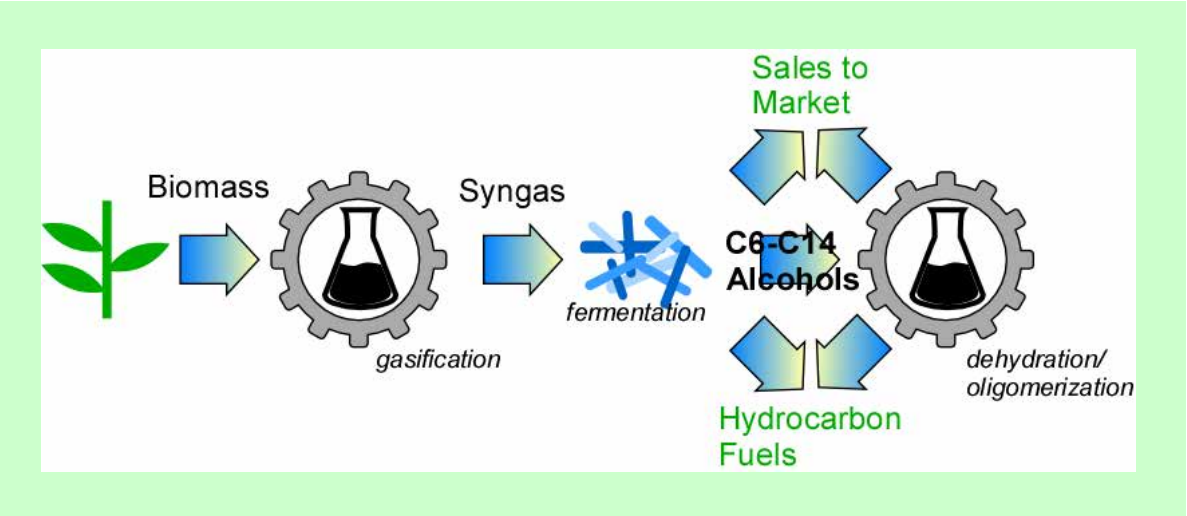
Bio-syngas to Fatty Alcohols (C6-14) as a Pathway to Fuels

3/9/17
Biochemical Conversion

Principal Investigator: Devon C. Rosenfeld
The Dow Chemical Company

Project Goal

Develop a new bio-syngas fermentation process using engineered bacteria for production of intermediate (C6-C14) fatty alcohols leveraging robust chemical markets enabling scaling to biofuels production at < $3/gge.



Aligned with Conversion R&D strategic goal of developing commercially viable technologies for converting biomass feedstocks via biological and chemical routes into energy dense, fungible, finished liquid transportation fuels and well as chemical intermediates.

Quad Chart Overview

Timeline

- Project start: 10/1/2016
- Project end: 12/31/2018
- 10% complete (Validation)

Budget

	FY 16 Costs	Total Planned Funding (FY 17-Project End Date)
DOE Funded	$0	$1,988,639
Project Cost Share (Dow)	$0	$883,929
Project Cost Share (LanzaTech)	$0	$329,688
Project Cost Share (Northwestern U.)	$0	$34,233

*If there are multiple cost-share partners, separate rows should be used.

Partners

- The Dow Chemical Company
  - Analytical
  - Process separations
- LanzaTech, Inc.
  - Strain optimization
  - Fermentation optimization
- Northwestern University
  - Computational modeling
- Project led by Dow with collaboration across partners key to success

Quad Chart Overview-Con't

Conversion R&D Technical Barriers Addressed

- Ct-H: Efficient catalytic upgrading of gaseous intermediates to fuels and chemicals
- Ct-A: Feedstock Variability
- St-E: Best practices and systems for sustainable bioenergy production

Aligned with MYPP Technical Targets

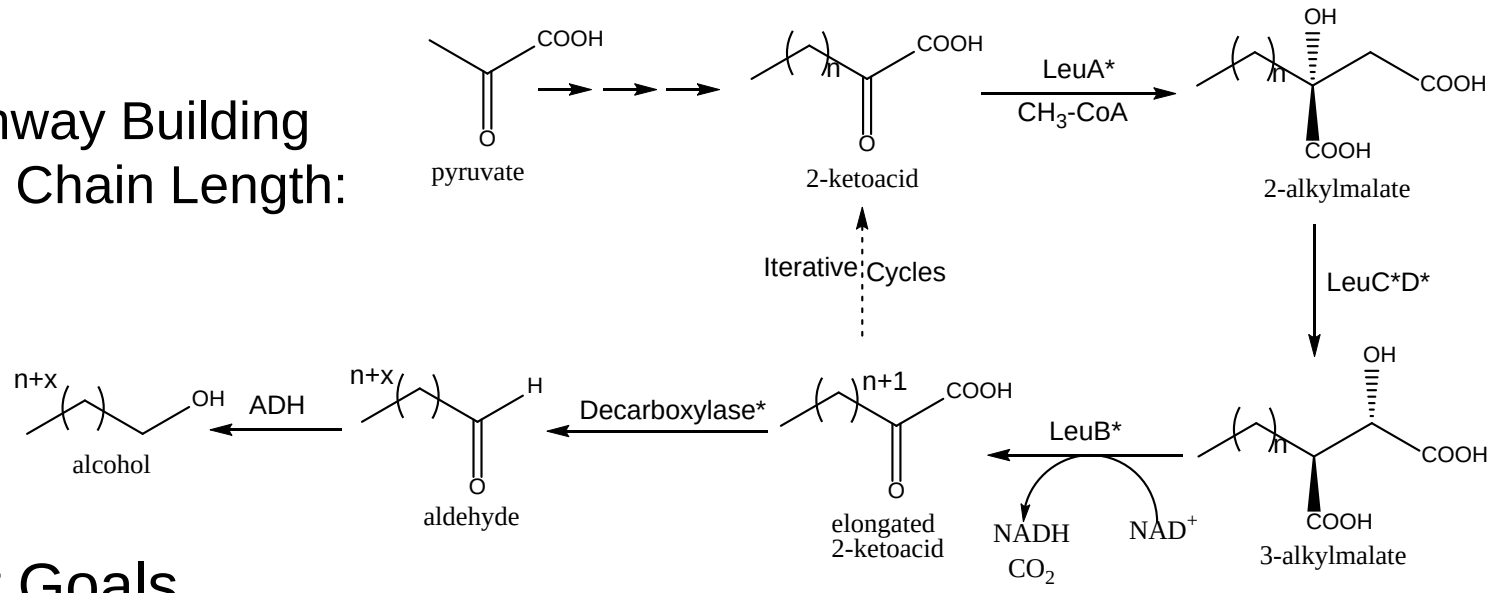
- By 2020, provide enabling capabilities in synthetic biology for industrially relevant, optimized chassis microorganisms and Design-Built-Test-Learn cycles for fuel and chemical production that reduces time-to scale-up by at least 50% compared to the current average of ~10 years.

- By 2021, complete R&D necessary to set the stage for a 2022 verification that produces both fuels and high-value chemicals to enable a biorefinery to achieve a positive return on investment.

Project Overview

- Dow and LanzaTech previously validated syngas conversion to fatty alcohols via the +1 pathway in *Clostridium*

+1 Pathway Building Carbon Chain Length:



- Project Goals
  - Resolve challenges discovered during validation research that limited yield
  - Optimize the strain and the syngas fermentation to deliver titer and productivity targets
  - Characterize and quantify the fermentation products
  - Develop a conceptual flowsheet for separating and purifying products

Management Approach

- Dow is the project lead, coordinating participant activities.
- Teleconference every two weeks and semiannual in person team meetings to monitor work progress ensure timely milestone completion, and address any issues that may arise.
- Project teams organized according to alignment of core expertise and capabilities to delivery for specific project task.

Project Lead	Tasks	Description
Dow	4, 6, 10	product characterization, design conceptual flow sheet for separations and purification, project management
LanzaTech	1-3, 7-9	BETO validation, strain and fermentation optimization
Northwestern	5	Computational modeling for pathway development

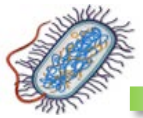
- Successful task delivery through cross partner collaboration
- Go/No-go milestones with fatty alcohol titer target at the end of each budget period to assess progress and update techno-economic assessment (TEA)

Technical Approach

Strain engineering to resolve previously discovered challenges and eliminate unproductive pathways

Fermentation optimization to maximize fatty alcohol yield

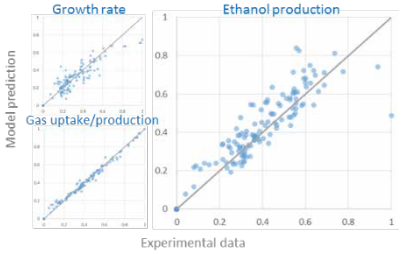
Biomass Syngas



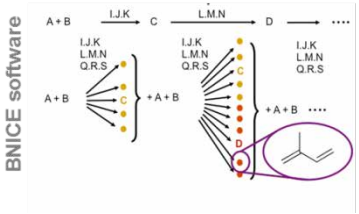
Proprietary Engineered Microbe

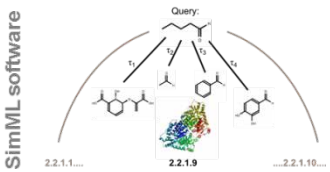
C6-C14 fatty alcohols

Genome scale modeling and TEA



Computational modeling to identify promiscuous enzymes





Product characterization for carbon balance and design of separations flow sheet

Relevance

Develop a new bio-syngas fermentation process using engineered bacteria for the production of intermediate (C6-C14) fatty alcohols robust chemical markets enabling scaling to biofuels production for sale at < $3/gge

- Our project is aligned with BETO's vision and mission statements and strategic goals.
- Our technology will impact the industry and deliver BETO goals by:
  1. Creating a disruptive bioconversion technology leveraging robust chemical markets to traverse the "valley of death" of biofuels scaling
  2. Enabling feedstock versatility and decoupling raw materials from food crops.
  3. Displacing petroleum derived fuels and chemicals with domestically produced, cost competitive bio-renewables with improved infrastructure compatibility.
  4. Exceeding advanced biofuels GHG reduction target of >50% versus conventional

- Our technology has the versatility to potentially function as a front end upgrading process within an integrated biorefinery or to bolt in within conventional conversion infrastructure such as an FT refinery.

Future Work

- Project kicked off 1/2017 after successfully passing BETO validation Go/No-Go milestone
- Key research activities and milestones of budget period 2 (1/17-12/17)

  Strain optimization resolving challenges discovered in prior validation

  – Computational predictions of alternative and potentially promiscuous enzymes

  – Build strain library with improved enzymes and validate improvement *in vivo*

  Fermentation optimization to drive pathway titer

  – Test top performing strains from strain optimization studies in syngas fermentation and optimize conditions for syngas conversion and alcohol titer

  Characterize fermentation products
- Go/No-Go milestone at the end of month 15 (12/31/17)

  C6-C14 alcohol from syngas demonstrated at a total titer of 50% and a productivity of 5% of our final milestone metrics in 2-L-CSTR bioreactor, GEM and TEA updated

Future Work

- Key research activities and milestones of budget period 3 (1/18-12/18)

  Characterize fermentation products

  – Identify all fermentation products through analysis of components in all phases

  – Develop methods to quantify fermentation products

  Develop conceptual flow sheet for separation and purification

  – Employ process simulations tools to identify potential separation schemes

  – Generate conceptual block flow diagram

  Strain optimization

  – Strain construction to balance expression to maximize pathway flux

  – Computational predictions for direct intermediate conversion to fuels

  Fermentation optimization

  – Test top performing strains from strain optimization studies in syngas fermentation and optimize conditions for syngas conversion and alcohol titer

- Go/No-Go milestone at the end of month 27 (12/31/18)

  C6-C14 alcohols from syngas demonstrated at a total titer, productivity and selectivity meeting TEA metrics to produce biofuels at price of < $3/gge. Update GEM and TEA model.

Summary

Goal: Develop a new bio-syngas fermentation process using engineered bacteria for production of intermediate (C6-C14) fatty alcohols leveraging robust chemical markets enabling scaling to biofuels production at < $3/gge.

- *Overview:* Rooted in prior research by Dow and LanzaTech validating syngas fermentation to fatty alcohols via the +1 pathway with yield limiting challenges
- *Approach:* Deploy strengths of three partners to resolve challenges, maximize fatty alcohol titer and devise purification scheme
- *Relevance*: Aligned with BETO's long term vision with potential to deliver at least four goals from the MYPP
- *Future Work:*
  - BP2 (M4-M15): resolve pathway challenges through strain engineering, fermentation optimization and computational pathway modeling
  - BP3 (M16-M27): maximize fatty alcohol titer through strain and fermentation optimization, full product characterization for designing separation and purification flow-sheet

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- *You may refer to them during the Q&A period if they are helpful to you in explaining certain points.*

Responses to Previous Reviewers' Comments

- If your project is an on-going project that was reviewed previously, address 1-3 significant questions/criticisms from the previous reviewers' comments (refer to the 2015 Peer Review Report, see notes section below)
- Also provide highlights from any Go/No-Go Reviews

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Publications, Patents, Presentations, Awards, and Commercialization

- List any publications, patents, awards, and presentations that have resulted from work on this project
- Use at least 12 point font
- Describe the status of any technology transfer or commercialization efforts

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