Integrating Chemical Catalysis and Biological Conversion of Carbon Intermediates for Deriving Value Added Products from Carbon Dioxide

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Project Overview

- CO₂ capture is increasingly viewed as an important technology towards meeting climate goals.
- Electrocatalytic systems excel at converting CO₂ into C₁ and C₂ compounds, but are challenged in forming additional carbon bonds.
- Biological systems can build complex carbon compounds from simple carbon compounds, but are less efficient at upcycling CO₂ due to gas diffusion limitations and limited conversion to long carbon chain biomolecules.
- Project goal: Develop an integrated platform that takes advantage of the strengths of electrocatalysis and bioconversion to convert CO₂ → C₁ and C₂ intermediates → bioproducts.

Abbreviations: polyhydroxybutyrate (PHB), tricarboxylic acid cycle (TCA)
## Timeline
- **Start:** 01 October 2018
- **End:** 01 September 2022

<table>
<thead>
<tr>
<th>FY20 Costed</th>
<th>Total Award</th>
</tr>
</thead>
<tbody>
<tr>
<td>DOE Funding</td>
<td>$472,106</td>
</tr>
<tr>
<td>Project Cost Share</td>
<td>$66,420</td>
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</tbody>
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## Project Goal
Develop a two-stage process integrating electrocatalysis and bioconversion to upcycle CO₂ to intermediates to polyhydroxybutyrate and biomass, which will inform both techno-economic and life cycle analyses of the complete system.

### End of Project Milestone
Achieve a 37% process carbon conversion efficiency from an input CO₂ stream to polyhydroxybutyrate and biomass for biocrude using microbial bioconversion; develop an accompanying techno-economic and life-cycle analysis to assess barriers to cost-competitive product generation and sensitivity of the system to market dynamics.

## Project Partners
- Johns Hopkins University
- Pacific Northwest National Laboratory
- San Diego State University

## Funding Mechanism
DE-FOA-0001916: Bioenergy Engineering for Products Synthesis (BEEPS)
**Topic area 5:** Rewiring carbon utilization
**Year:** 2018
Structure and Team

Dr. Chao Wang (JHU)

Dr. Marina Kalyuzhnaya (SDSU)

Dr. Alex Beliaev Dr. Pavlo Bohutskyi (PNNL)

Dr. Michael Betenbaugh (JHU)

Dr. Sarah Jordaan (JHU)

Electrocatalysis
Optimization of electrocatalytic conversion of CO₂ to reduced carbon compounds

Biosynthesis
Engineering of *M. alcaliphilum* 20Z⁰ to convert reduced carbon intermediates to PHB

Optimization
Characterization of engineered *M. alcaliphilum* 20Z⁰ and optimization of the biological production of PHB

Integration
Integration of electrocatalytic reduction and bioconversion processes

TEA/LCA
Techno-economic and life cycle analysis of complete process to assess commercial viability

Overview Management Approach Impact Progress & Outcomes Summary

Abbreviations: Johns Hopkins University (JHU), San Diego State University (SDSU), Pacific Northwest National Laboratory (PNNL), polyhydroxybutyrate (PHB), techno-economic and life cycle analysis (TEA/LCA)
Structure and Team

**Risk:** low CCE toward methanol  
**Mitigation:** co-product production (e.g., formate, acetate)

**Risk:** low conversion of substrates to product  
**Mitigation:** consider variable inputs and engineer metabolic pathways

**Risk:** growth varies with operating conditions  
**Mitigation:** study range of conditions to find optimum

**Risk:** potential toxic compounds in electrochemical effluent  
**Mitigation:** adjust chemical feedstocks for biological use

**Risk:** economic viability is tied to commodity markets  
**Mitigation:** apply TEA/LCA to identify best targets and opportunities

Abbreviations: Johns Hopkins University (JHU), San Diego State University (SDSU), Pacific Northwest National Laboratory (PNNL), polyhydroxybutyrate (PHB), techno-economic and life cycle analysis (TEA/LCA)
Background

- Few studies on integrated electrocatalytic-biocatalytic systems
- Limited work on selective electroreduction of CO\textsubscript{2} to methanol
- Anaerobes can capture industrial off-gases and upcycle them to short carbon chain chemicals/fuels

Technical Approach

- Two-step process
  1. Electrocatalysis: use of electricity and inorganic catalysts to convert chemicals
  2. Bioconversion: use of bacteria to convert chemicals

CO\textsubscript{2} \rightarrow \text{methanol, acetate, formate} \rightarrow \text{PHB, biomass}

Challenges

- Electrocatalysis: efficient conversion of CO\textsubscript{2} into methanol
- Bioconversion: efficient growth and channelling of non-ideal substrates into valuable products

Abbreviations: polyhydroxybutyrate (PHB)
## Go/No-Go Decision Points

<table>
<thead>
<tr>
<th>Budget Period 1</th>
<th>Budget Period 2</th>
<th>Budget Period 3</th>
</tr>
</thead>
</table>
| **Demonstrate potential to achieve** CCE > 37% from CO₂ to fuel or product | **Demonstrate** \( \text{FE} \geq 70\% \) and \( \text{CCE} \geq 80\% \) toward formate platform products | **Electrocatalysis:** \( \text{FE} \geq 50\% \), \( \text{CCE} \geq 50\% \) toward methanol platform products  
**Bioconversion:** \( \text{CCE} \geq 40\% \), yield \( \geq 0.3 \) on methanol |
| Critical because: it will enable PHB and biomass production at proof-of-concept CCE | Critical because: it will produce enough substrate for integrated process | Critical because: it will enable efficient biomass and PHB production |

Abbreviations: Faradaic efficiency (FE), carbon conversion efficiency (CCE), polyhydroxybutyrate (PHB)
## Key Metrics

<table>
<thead>
<tr>
<th>Team</th>
<th>Metric</th>
<th>Assumptions</th>
<th>Analyses</th>
</tr>
</thead>
<tbody>
<tr>
<td>Electrocat.</td>
<td>Faradaic efficiency</td>
<td>Electrocatalyst design can improve FE</td>
<td>GC-MS, NMR</td>
</tr>
<tr>
<td></td>
<td>Carbon conversion efficiency</td>
<td>Co-products will improve CCE</td>
<td>GC-MS, NMR, flow control</td>
</tr>
<tr>
<td></td>
<td>Product concentration and rate</td>
<td>Recirculation can improve concentration</td>
<td>NMR, flow control (gas, liquid)</td>
</tr>
<tr>
<td>Biosynthesis</td>
<td>Recombinant protein expression</td>
<td>Proteins are expressed &amp; active</td>
<td>SDS-PAGE, enzyme activities</td>
</tr>
<tr>
<td></td>
<td>Yield (g PHB/g cell dry weight)</td>
<td>Engineering can improve yield</td>
<td>Extraction; cell dry weight</td>
</tr>
<tr>
<td></td>
<td>Carbon distribution</td>
<td>Metabolic tuning can improve</td>
<td>Metabolomics, NMR, GC-MS</td>
</tr>
<tr>
<td>Optimization</td>
<td>Yield (g biomass/g substrate)</td>
<td>Dependent on dilution rate</td>
<td>Extraction; cell dry weight</td>
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<tr>
<td></td>
<td>Carbon conversion efficiency</td>
<td>Dependent on media composition</td>
<td>NMR, HPLC, GC</td>
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<tr>
<td>Integration</td>
<td>Operating mode (batch, semibatch)</td>
<td>Culture behavior is consistent</td>
<td>Reactor design</td>
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<tr>
<td></td>
<td>Yield (g biomass+PHB/g substrate)</td>
<td>Dependent on methanol</td>
<td>Extraction; cell dry weight</td>
</tr>
<tr>
<td></td>
<td>Carbon conversion efficiency</td>
<td>Substrate utilization pathways</td>
<td>NMR, HPLC, GC</td>
</tr>
<tr>
<td>TEA/LCA</td>
<td>Net present value</td>
<td>Material costs, selling prices</td>
<td>Aspen process simulation</td>
</tr>
<tr>
<td></td>
<td>CO₂ emissions</td>
<td>Process parameters</td>
<td>Emissions modeling</td>
</tr>
</tbody>
</table>

Abbreviations: techno-economic and life cycle analysis (TEA/LCA), sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE), nuclear magnetic resonance (NMR), high performance liquid chromatography (HPLC), gas chromatography (GC), gas chromatography-mass spectrometry (GC-MS), Faradaic efficiency (FE)
Technological Impact

• New electrochemical reduction process and catalyst for high efficiency energy conversion and chemical transformation
• Efficient microbial catalyst for upcycling carbon intermediates
• Integrated platform for CO₂ electroreduction and bioconversion

Abbreviations: coordinated organic framework (COF), multi-wall carbon nanotubes (MWCNT)
Potential Industrial Impact

- Companies working to improve CO₂ capture, electroreduction
- Project will demonstrate potential to electroreduce CO₂ into valuable products
- TEA will identify key areas for growth to ensure competitiveness
- Project will demonstrate feasibility of linking electrochemical and biological processes to generate useful products

Abbreviations: techno-economic analysis (TEA)
Academic Impact

• Conferences

• Publications
  • Ruttinger, A. W., Tavakkoli, S., and Jordaan, S. M. Evaluating Technology and Market Scenarios for the Deployment of a Profitable Carbon Capture and Utilization Process. [In Submission]

Applied Energy Symposium
## Key Milestones (Budget Period 2)

<table>
<thead>
<tr>
<th>Team</th>
<th>Key Milestone</th>
<th>Current State</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Electrocatlysis</td>
<td>1 mM formate at 2 ml/min, ≥60% FE, ≥70% CCE</td>
<td>21 mM, &gt;90% FE at 100 mA/cm², &gt;90% CCE</td>
<td>&gt;100%</td>
</tr>
<tr>
<td></td>
<td>1 mM methanol and other C₁ and C₂⁺ metabolites at 2 ml/min, ≥40% FE, ≥40% CCE</td>
<td>C1 Pathway (Methanol) 55 mM, 20% FE at 100 mA/cm², 20% CCE</td>
<td>90%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>C1/C2 Pathway (Methanol+Acetate) 55 mM methanol, 19 mM acetate, 70% FE, &gt;50% CCE</td>
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<tr>
<td>Bioconversion</td>
<td>M. alcaliphilum 20Z expressing PHB biosynthesis pathway</td>
<td>Individual PHB pathway genes into biocatalyst; enzyme activities verified; integration of pathway under assessment</td>
<td>75%</td>
</tr>
<tr>
<td>Optimization</td>
<td>Experimental biocatalyst CCE (%) and yield (g biomass per g substrate)</td>
<td>Highest biomass yield: 0.11-0.13 g DW per g MeOH Highest CCE to biomass: &gt;20% (with MeOH as a substrate)</td>
<td>100%</td>
</tr>
<tr>
<td>Integration</td>
<td>Batch-to-batch operating mode, total CCE≥25% (toward biomass), yield ≥0.20 g of biomass per g CO₂</td>
<td>Integrated batch-to-batch growth characterization Total CCE: ~12% Total yield: ~0.07 g biomass/g CO₂</td>
<td>50%</td>
</tr>
<tr>
<td>TEA/LCA</td>
<td>Complete Aspen model, according to integrated technology for all cases.</td>
<td>Aspen process model integrated for all cases proposed TEA completed for all cases proposed Acetate scenarios and emissions analysis pending</td>
<td>80%</td>
</tr>
</tbody>
</table>

Abbreviations: techno-economic and life cycle analysis (TEA/LCA), Faradaic efficiency (FE), carbon conversion efficiency (CCE), polyhydroxybutyrate (PHB), dry weight (DW)
Technical Achievements: Electrocatalysis

- Methanol (CH$_3$OH)
  - 55 mM methanol
  - 20% FE toward methanol
  - 20% CCE toward methanol

- Methanol + Acetate (C$_2$H$_3$O$^-$)
  - 55 mM methanol
  - 19 mM acetate
  - 70% FE toward methanol/acetate
  - >50% CCE toward methanol/acetate

Abbreviations: Faradaic efficiency (FE), carbon conversion efficiency (CCE)
Technical Achievements: Biosynthesis

- Completed construction of glyoxylate shunt
- Improving formate utilization
- Expression of PHB pathway with active enzymes

Abbreviations: polyhydroxybutyrate (PHB), methenyl-cyclohydrolase and formaldehyde-activating enzyme (mch/fae), malyl-CoA lyase (mcl), pyruvate dehydrogenase (pdh), phosphoketolase 1, 2, and 3 (xfp1, xfp2, xfp3), 3-ketothiolase (phaA), acetoacetyl coenzyme A reductase (phaB), polyhydroxyalkanoate synthase (phaC), adenosine triphosphate (ATP), nicotinamide adenine dinucleotide (NAD), fructose 6-phosphate (F6P), ribulose 5-phosphate (Ru5P), coenzyme A (CoA), methanol, acetate, 3 CH$_2$O, 5 C$_2$H$_3$O$_2$. 

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Technical Achievements: Optimization

- >20% baseline CCE to biomass on methanol
- 0.13 baseline yield (g dry weight/g methanol)
- Effect of dilution rate on growth characteristics in continuous culture

Abbreviations: polyhydroxybutyrate (PHB), carbon conversion efficiency (CCE)
Technical Achievements: Integration

- CCE: ~12%
- Yield: ~0.07 g biomass/g CO₂
- Demonstration of biomass production on electrocatalysis product

Cell growth on methanol/acetate (25 mM)

0.04 M Carbonate (Low Control)
Electrocatalysis Product

0.5 M Carbonate (High Control)

Abbreviations: polyhydroxybutyrate (PHB), carbon conversion efficiency (CCE)
Technical Achievements: TEA/LCA

- ASPEN model developed
- TEA completed for proposed scenarios
- Currently analyzing impact of acetate production
- Evaluating emissions implications

CO₂ → Methanol, Acetate → Biomass, PHB

Abbreviations: polyhydroxybutyrate (PHB), techno-economic analysis and life cycle analysis (TEA/LCA)
Summary

• **Overview:** Project seeks to develop an efficient and integrated electrocatalytic-bioconversion process to upgrade CO₂

• **Management:** Process development and risk mitigation are split by topic expertise

• **Approach:** Electroreduction of CO₂ to methanol and engineered microbial growth on non-ideal substrates are both the project’s biggest challenges and innovations

• **Impact:** Project success will have substantial impact as the world continues to adopt CO₂ upgrading technologies

• **Progress & Outcomes:** Each team has made significant progress toward their milestones goals and are addressing risks appropriately
Additional Slides

For DOE evaluation
Responses to Previous Reviewers’ Comments

Not applicable; project has not been previously peer-reviewed.
Publications, Patents, Presentations, Awards, and Commercialization

• Conferences

• Publications
  • Ruttinger, A. W., Tavakkoli, S., and Jordaan, S. M. Evaluating Technology and Market Scenarios for the Deployment of a Profitable Carbon Capture and Utilization Process. [In Submission]

• Current technology transfer or commercialization efforts awaiting proof-of-concept