Goal Statement

The goal is 

- to develop *improved strains and cultivation methods* to increase the algal biofuel intermediate yield by at least 40% and
- to develop *new harvest and dewatering technology* to reduce the energy for downstream processing by at least 88%

in an *integrated outdoor system that reduces the projected minimum selling price* (MSP) of algae biomass by 58%

**The project outcome:** Exceeded the goal and technology targets

**Technologies demonstrated in an integrated large-scale facility that operated with power plant CO$_2$ and full media recycle**

- A breakthrough cultivation method increased the growth phase productivity by 80% with 1/3 of the original energy input
- A breakthrough harvesting and dewatering technology that is commercially available now
Quad Chart Overview

**Timeline**
- **Validation:** 10/13 - 10/14
- **Project work:** 11/14 - 6/16
- **% complete:** 99%

**Budget**

<table>
<thead>
<tr>
<th></th>
<th>FY 12 – FY 14 Costs ($000)</th>
<th>FY 15 Costs ($000)</th>
<th>FY 16 Costs ($000)</th>
<th>Total Planned FY 17 + ($000)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>DOE Funded</strong></td>
<td>159</td>
<td>2838</td>
<td>1997</td>
<td>3</td>
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<tr>
<td><strong>Cost Share</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TSD</td>
<td>40</td>
<td>709</td>
<td>499</td>
<td>1</td>
</tr>
<tr>
<td>UCSD</td>
<td>9</td>
<td>45</td>
<td>49</td>
<td>-</td>
</tr>
<tr>
<td>Evodos</td>
<td>-</td>
<td>22</td>
<td>20</td>
<td>-</td>
</tr>
<tr>
<td>GE</td>
<td>18</td>
<td>415</td>
<td>90</td>
<td>-</td>
</tr>
<tr>
<td>Kuehnle</td>
<td>-</td>
<td>125</td>
<td>93</td>
<td>-</td>
</tr>
<tr>
<td>Other</td>
<td>2</td>
<td>49</td>
<td>31</td>
<td>-</td>
</tr>
</tbody>
</table>

**Barriers**
- Aft-B. Sustainable Algae Production
- Aft-D. Sustainable Harvesting
- Aft-H. Overall Integration and Scale-Up

**MYPP targets addressed:**
- ✓ 2018 - algae yield of 2500 gal/ac-yr
- ✓ 2020 - algae yield of 3700 gal/ac-yr
- ✓ 2022 – 407 kWh/ton energy for farm
- ✓ 2022 - $494/ton algae for HTL pathway
- ✓ 2022 – $4.72/GGE for lipid pathway

**Partners**

**Strain Improvement**
- University of California, San Diego
- Kuehnle AgroSystems
- Hamilton Robotics

**Harvesting, dewatering & extraction**
- TSD Management Associates
- General Electric, Evodos
- Texas A&M, Crown Iron works
- PNNL
1 - Project Overview History

Kauai Algae Facility
Integrated from inoculation through harvesting
All CO$_2$ from adjacent power plant flue gas
Demonstrated Contamination control
Full cultivation media recycle
Advanced raceway design

Algal Biomass Yield Phase 1
Biofuel intermediate yield: 1360 to 1900 gal / ac-yr
Pre-processing energy (% of biofuel): 87% to 10%
Demonstrate in an Integrated outdoor system
Limited to economically viable technology
# Project Overview Summary

<table>
<thead>
<tr>
<th>Area</th>
<th>Baseline</th>
<th>Phase 1 Goals</th>
<th>Lipid Pathway</th>
<th>HTL Pathway</th>
<th>MYPP 2022</th>
</tr>
</thead>
<tbody>
<tr>
<td>Productivity: (gal oil/acre-year)</td>
<td>1360</td>
<td>1900</td>
<td>2200</td>
<td>4200</td>
<td>5000</td>
</tr>
<tr>
<td>Pre-processing: (% of the biofuel energy)</td>
<td>87%</td>
<td>10%</td>
<td><strong>9.6%</strong></td>
<td><strong>11.1%</strong></td>
<td>10%</td>
</tr>
<tr>
<td>Integration: algae paste MSP ($/mt AFDW)</td>
<td>$ 1536</td>
<td>$ 900</td>
<td>$ 597</td>
<td>$ 437</td>
<td>$ 494</td>
</tr>
<tr>
<td>Integration: Protein (% protein in algae meal)</td>
<td>15%</td>
<td>40%</td>
<td><strong>48%</strong></td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Integration: Farm energy (kwh/mt AFDW)</td>
<td>860&lt;sup&gt;a,b&lt;/sup&gt;</td>
<td>NA</td>
<td>270</td>
<td>160</td>
<td>407</td>
</tr>
<tr>
<td>5. Integration: MFSP ($/GGE)</td>
<td>$ 17.69&lt;sup&gt;a&lt;/sup&gt;</td>
<td>NA</td>
<td>$ 3.33&lt;sup&gt;c&lt;/sup&gt;</td>
<td>$ 5.37&lt;sup&gt;d&lt;/sup&gt;</td>
<td>L: $ 5.90&lt;sup&gt;a&lt;/sup&gt; H: $ 4.72&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup> From 2016 MYPP  
<sup>b</sup> From 2016 MYPP, our model projected 1810  
<sup>c</sup> Assumes $500/mt for the co-product algae meal  
<sup>d</sup> Conversion cost from PNNL/NREL 2014 design report
2 – Approach (Management)

All technologies filtered through comprehensive cost model

- Economically viable
- Integration impacts and opportunities

Technology development map

- Prioritize research
- Many options
- Quick advancement/early risk retirement
- Synergistic projects or opportunities

<table>
<thead>
<tr>
<th>Technology</th>
<th>Yield</th>
<th>Cost</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adv. cultivation</td>
<td>70%</td>
<td>($4.70)</td>
</tr>
<tr>
<td>Fast lipid accum.</td>
<td>50%</td>
<td>($3.70)</td>
</tr>
<tr>
<td>O₂ Tolerance</td>
<td>25%</td>
<td>($0.90)</td>
</tr>
<tr>
<td>Constitutive lipid</td>
<td>87%</td>
<td>($4.30)</td>
</tr>
</tbody>
</table>

Frequent telecoms to discuss results and opportunities

- Rapid communication
- Synergistic projects and opportunities
- Cost and technology status/potential transparent to team
2 – Approach (Technical)

**Biofuel Intermediate Yield**
- Strain Improvement
  - Proven outdoor strains
  - 2 labs, multiple green and diatom strains
  - Non-GMO lipid & growth improvements
  - Integral growth requirement
- Cultivation
  - Proven contamination control
  - Advanced cultivation methods
  - Control optimization

**Preprocessing Energy**
- Harvesting
  - Membrane filter
  - Clarifier with chemical or bio floc
- Dewater
  - Improved centrifuges
  - Belt press or wicking belt
- Extraction
  - Thermal or acid lysis
  - Mechanical or solvent separation

**Top Challenges**
- Complexity of abiotic and biotic variation
- Translating lab to large-scale outdoor cultivation
- Inability to achieve early risk retirement for strain optimization
- Producing sufficient material for downstream processing work
Technical Accomplishments, Progress and Results
Advanced cultivation methods
Strain improvement - Hildebrand lab

Novel mutagenesis/high throughput fluorescent activated cell sorting:

- 3,600x's more efficient in viable mutants
- Applied to GAI-229, Nitzschia amphibia
- Nine new strain lines generated with improved lipid accumulation


Native sequence genetic manipulation

- Avoids GMO classification
- Demonstrated antisense knockdown of CGI-58 lipase improved TAG content

Reduced photosystem antenna size

- New strain lines started
- Reduced pigmentation lines on left
Zobi™ provides a solution to harvesting and dewatering issues

<table>
<thead>
<tr>
<th>Options Tested by Global Algae Innovations</th>
<th>High efficiency</th>
<th>Low Energy</th>
<th>Low Cost</th>
<th>No Flocculent</th>
<th>Universal</th>
<th>High Conc.</th>
<th>Scalable</th>
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<tbody>
<tr>
<td>Centrifuge</td>
<td>NO</td>
<td>NO</td>
<td>NO</td>
<td>YES</td>
<td>YES</td>
<td>YES</td>
<td>NO</td>
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<tr>
<td>Dissolved air floatation</td>
<td>NO</td>
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<td>YES</td>
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<td>Clarifier</td>
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<td>YES</td>
<td>NO</td>
<td>NO</td>
<td>NO</td>
<td>YES</td>
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<tr>
<td>Wicking belt</td>
<td>NO</td>
<td>YES</td>
<td>NO</td>
<td>NO</td>
<td>NO</td>
<td>YES</td>
<td>NO</td>
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<tr>
<td>Belt press</td>
<td>NO</td>
<td>YES</td>
<td>YES</td>
<td>NO</td>
<td>NO</td>
<td>YES</td>
<td>YES</td>
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<tr>
<td>Cross-flow filtration</td>
<td>YES</td>
<td>NO</td>
<td>NO</td>
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<td>VSEP membrane</td>
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<td>NO</td>
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<td>YES</td>
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<td>Other novel membrane</td>
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<td>NO</td>
<td>YES</td>
<td>YES</td>
<td>YES</td>
<td>NO</td>
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<tr>
<td>Zobi Harvester™</td>
<td>YES</td>
<td>YES</td>
<td>YES</td>
<td>YES</td>
<td>YES</td>
<td>YES</td>
<td>YES</td>
</tr>
</tbody>
</table>
Zobi Harvester™ - 100% harvest efficiency

Crystal Clear Permeate
- Less than 0.1 NTU
- No algae, bacteria or predators

5 m³/hr system

Permeate  Feed  Retentate
Zobi Harvester™ - 15 to 20% algal slurry

Eliminates need for secondary dewatering
Zobi Harvester™ - very low energy use
Zobi Harvester™ - universally applicable

- Many green algae
- Many diatoms
- Six cyanobacteria
- One red algae

No flocculent

20 m³/hr system
Collets enable use of standard Crown Iron Works solvent extractors

Collets

Crown Iron Works Model IV immersion extractor

Crown utilized our pilot-scale data to develop a quote and utility requirements for commercial scale facilities
4 – Relevance

Higher yield, lower energy use, lower cost algae biofuel intermediate production in a large-scale integrated outdoor facility

Directly Supports the BETO mission to “Develop and demonstrate transformative and revolutionary bioenergy technologies for a sustainable nation.”

By achieving three major Algal R&D targets:
• Achieved BETO MYPP 2020 yield target
• Achieved BETO MYPP 2022 energy use target
• Achieved BETO MYPP 2022 algal biomass and fuel cost targets

State of the art advancements
• Zobi Harvester™: 1/10th to 1/150th energy use, 100% harvest, 15-20% solids
• Advanced cultivation: 80% higher productivity than prior sloped raceways

Tech Transfer/marketability
• Zobi Harvester™ commercially available and in commercial algae operations
• Advanced cultivation
  - included on 8 teams for ABY2 and PEAK FOAs
  - incorporated in a recently awarded integrated biorefinery scale-up project
  - being tested for high value product applications.
5 – Future Work

• Close out the project
• Remaining budget is sufficient
Summary

1. **Overview:** Addressed key BETO targets - yield, energy use, cost

2. **Approach:** Comprehensive cost model
   Early risk retirement with multiple technology options
   Fully integrated large-scale outdoor operations

3. **Technical Accomplishments/Progress/Results:**
   - Harvesting: more than order of magnitude improvement
   - Cultivation: advanced methods – nearly doubled productivity
   - Strain: cutting edge tools developed for vital improvements
   - Exceeded yield, energy use & cost goals as well as MYPP future targets

4. **Relevance**
   - Harvesting technology is commercially available product and in use
   - Cultivation methods incorporated into a new IBR scale-up project and made available to multiple R&D teams bidding on BETO FOA’s
Additional Slides
# Patents & Commercialization

<table>
<thead>
<tr>
<th>Patent Area</th>
<th># of Patents</th>
<th>Anticipated Divisional patents</th>
<th>Application dates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zobi harvester</td>
<td>3 US and 1 PCT</td>
<td>20 US patents</td>
<td>5/9/16 to 9/22/16</td>
</tr>
<tr>
<td>Advanced Cultivation</td>
<td>4 US provisional</td>
<td>6 US patents</td>
<td>5/9/16</td>
</tr>
</tbody>
</table>

Zobi Harvester™ in production for commercial sale and in use

In discussions on use of cultivation technology for high value products

Integrated biorefinery project to scale-up to 160 acres for biofuels and poly-ols
Zobi™ harvester incorporates GE’s scalable membrane technology

- Over 6 billion gallons per day installed capacity
- Plant sizes up to 228 million gallons per day