

Marine AlGae Industrialization Consortium (MAGIC)



Zackary Johnson

March 2021

Algae Platform Review

DOE Bioenergy Technologies Office (BETO)
2021 Project Peer Review

This presentation does not contain any proprietary, confidential, or otherwise restricted information



TRUCENT



ADM



Bucknell
UNIVERSITY

WBS 1.3.5.310



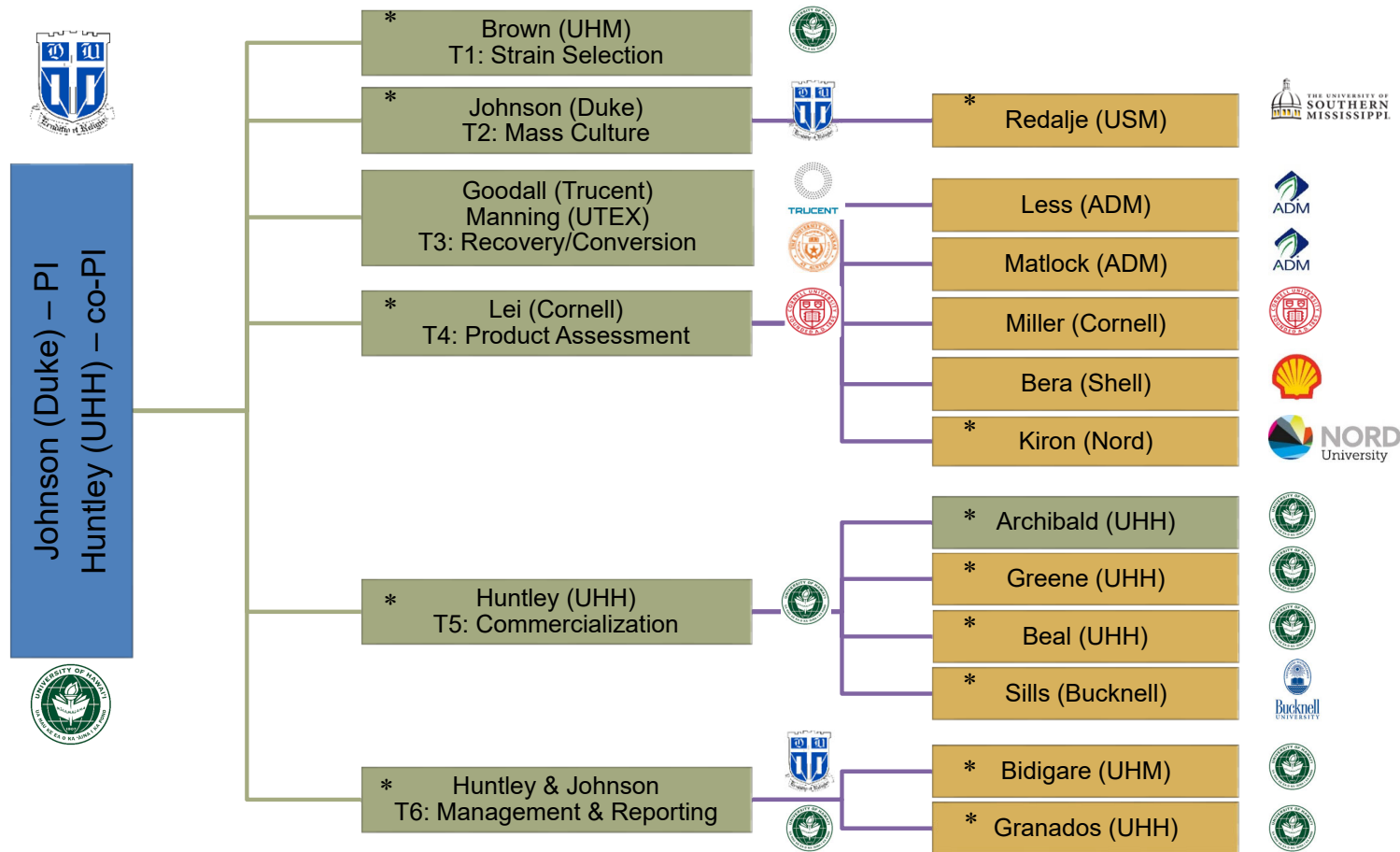
NICHOLAS SCHOOL OF THE
ENVIRONMENT

forging a sustainable future

1 - Project Overview

- The Consortium began in 2008, funded by Shell, built a 6-acre demonstration facility (Cellana), and funded 4 years of commercial R&D, ended 2012 – developed platform technology
- With support from DOE & USDA (2010-2015) we demonstrated the feasibility of the production of commercially viable, sustainable biofuels and animal feed co-products from marine algae
- Based on this success, the Duke Consortium was developed to demonstrate algae biofuel (oil) AND high value co-products from residuals (oil extract algae - LEA) across multiple algae strains:
 - *Are there co-products that lead to increased LEA value, while maintaining biofuel production, to drive down the overall cost of biofuel?*
- Achieved through marine algae strain selection, production of biomass, evaluation of different separation technologies, testing of multiple algae products (experimentally evaluating biofuel, animal/aquafeeds) and integrative TEA/LCA

1 – Management / Structure



Green = executive management team; * = long-time Consortium member
 Management Tools: Website, Database, Project Management Software, Remote/In-person meetings (group/sub-groups)

2 - Approach – Major Tasks

Integrated Process (not all activities are co-sited)

- 1) **Strain development** will deliver new strains to meet product specifications for biofuel and animal feed applications for
- 2) **Mass culture** using an innovative hybrid system of PBRs and open ponds to produce ~40 kg ash-free dry weight for multiple strains
- 3) **Recovery and conversion** of algal feedstock to refined biofuels and food and feed ingredients – by two pathways - to be used in
- 4) **Product demonstrations** to experimentally assess product efficacy and value, and
- 5) **Commercialization analyses** of relevant scale facilities based on demonstrated results using an iterative TEA/LCA process

Unique features: marine algae, PBR/pond hybrid technology, co-products

Top challenges: co-product value, LCA, EROI, productivity (challenging temperate environment)

Critical success factors: production, processing, product viability

3- Goals and Impacts

- **Our Project Goal**

Demonstrate and validate high-value co-products – *drive down the cost of biofuel by increasing the value of algae “co-products”*

Achieved through downstream unit testing AND multiple product testing

- **BETO MYPP Goals (2)***

- Model the sustainable supply of 1 million metric tonnes ash free dry weight (AFDW) cultivated algal biomass (2017)
- Demonstrate valuable co-products produced along with biofuel intermediates to increase value of algal biomass by 30% (2019)

- **Relevance**

Increased selling price for total algae biomass is one of the key drivers of economics and adoption

- **Outcome**

- A clear pathway to economically competitive, sustainable biofuels at scale
- Results disseminated through peer-reviewed publications

*goals when project selected

4- Progress and Outcomes - Task 1: *Strain development*

Subtask Summary: Strains selected from our collection of >600 strains, cultivated at bench-scale, and their growth characteristics and biochemical profiles compared to explicit product specifications. The 10 best-performing strains were selected for Mass Culture.

Initial Key Variables for Strain Selection

- Growth Rate (d^{-1})
- Sinking Index upon harvest (Note: sinking \neq amenable to centrifuging)
- % Ash upon Harvest
- Lipid Proxy - Nile Red:AFDW
- Lipid Proxy at Harvest and Assessment
- % Protein (Bio-rad assay and C:N)
- % Protein at Harvest and Assessment

Assessment = replete growth
Harvest = nutrient deplete (cells stressed)



Strain	Growth rate (d^{-1})
C649	1.42
H1117	0.99
C959	0.94
C954	0.92
C930	0.89
C1041	0.80
D046	0.77
C920	0.77
CHLOC01	0.75
C417	0.70
C1000	0.69
C985	0.66
C782	0.62
BORAD02	0.62
C046 max	0.51
C046 average	0.48

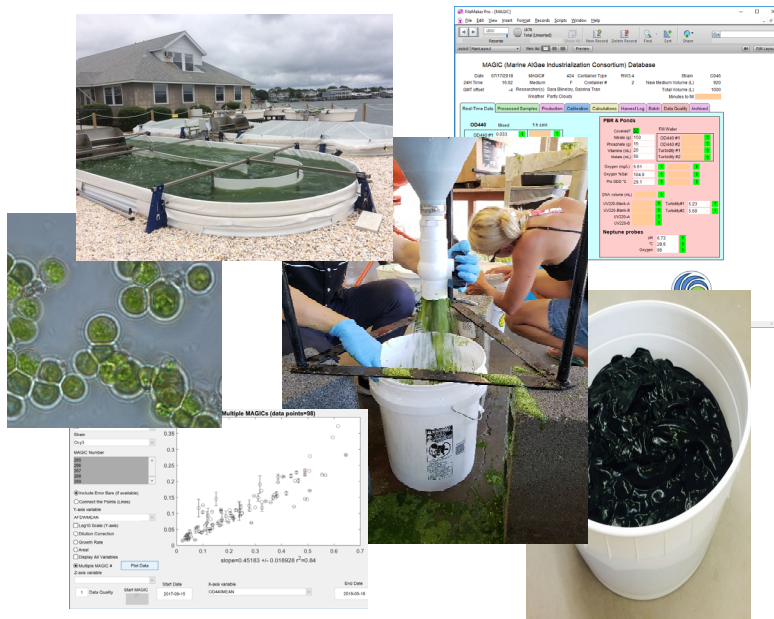
For each of these variables, strains were ranked against a **baseline strain** (C046) to determine top 10 candidates

Task 2: *Cultivation*

Task Summary: *Mass culture* will produce algae feedstock (10-30% total suspended solids, 25 to 50 kg per strain) for ten strains identified by Strain Validation (Task 1). All mass culture will be done using a hybrid cultivation system and following key operating parameters specified in the TEA/LCA and described in a cultivation design analysis.

M2.1 Deliver a Cultivation Plan for *mass culture* by the Consortium – **Report Delivered**

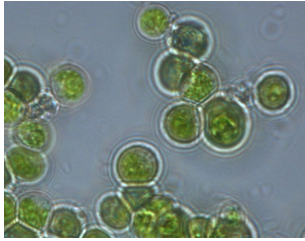
M2.2 (DP) Deliver feedstock for processing – **Biomass Produced / Reports Delivered**



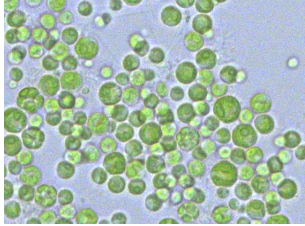
Major Milestones

- 7 strains grown at scale ~5000 L / 29 m²
- 4 suitable for downstream testing (ash, harvestability)
- >250 kg produced
- Production database with >5500 entries, >325 fields
- Harvest database with >250 entries, >110 fields
- Matlab analysis GUI
- Dozens of MAGIC SOPs (wiki)

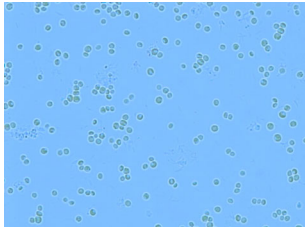
Task 2: *Cultivation (cont.)*



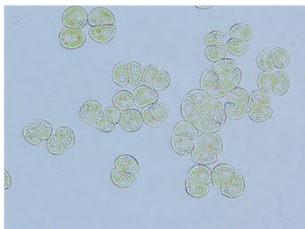
Desmodesmus sp.
69 production runs
~40 kg AFDW harvested
'type' strain



Chlorella sp.
48+ production runs
~40 kg AFDW harvested
high oil (constitutive)



Nanochloropsis sp.
57 production runs
~44 kg AFDW harvested
small size, high omega-3



Tetraselmis sp.
56 production runs
~50 kg AFDW harvested
large size, high omega-3

Challenges

- steep learning curve for new strains
- changing environments

Opportunities for future

- Other strains (of course!)
- Operational: harvesting
- Operational: stocking density
- Operational: water quality / reuse of water



Task 3: *Recovery and Conversion*

Task Summary: This task will use two (three) methods to process 25 to 50 kg dry weight (DW) per strain with algae from Task 2, yielding 1) oil for hydroprocessing, 2) whole algae and lipid-extracted algae meal for feed trials.

Methods: 1) **Trucent** - hexane solvent extraction; 2) **UT** – membrane oil separation; and 3) **MATRIC** – hexane solvent extraction (replaces Trucent)

M3.1 Integrated operational process - **DONE: 2018**

M3.2 (DP) Process 4 strains of feedstock and deliver products – **IN PROGRESS**

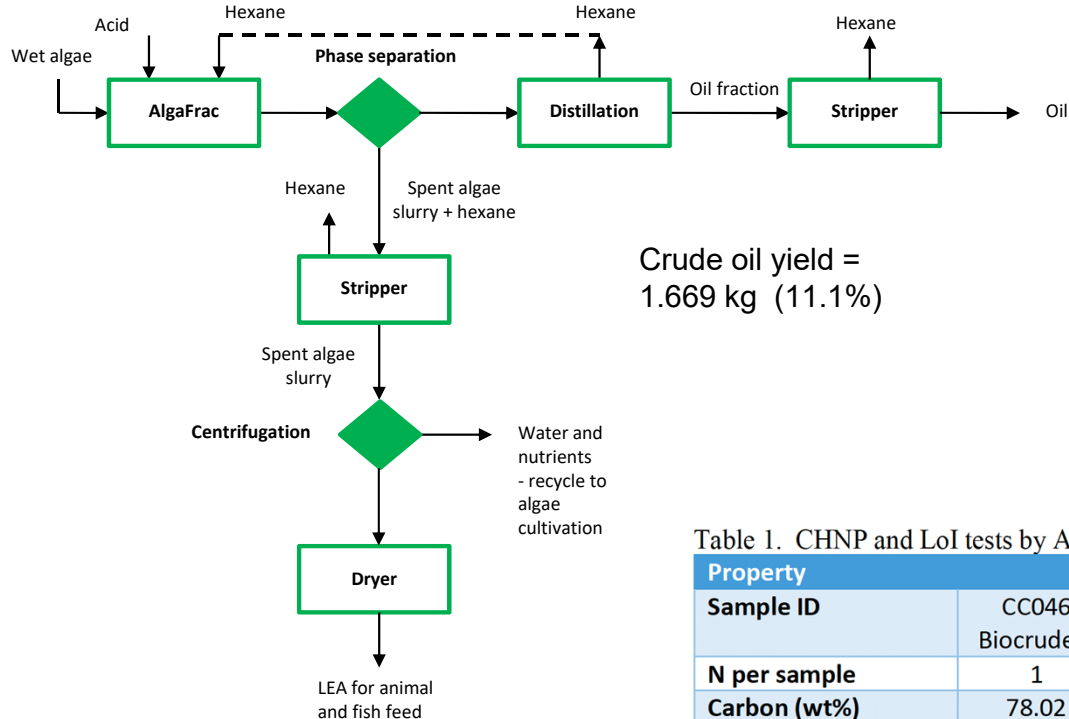
- 1) **Trucent** - hexane solvent extraction successful for pilot strain (*Desmodesmus*).
- 2) **UT/B&D** – membrane oil separation of 5 strains, lab-scale and field testing, unsuccessful
- 3) **MATRIC** – hexane solvent extraction of 3 strains, pending DOE paperwork

Summary:

- Trucent successfully recovered 1.6 L biocrude and 14 kg LEA from C046
- C046 oil was hydroprocessed by Emerging Fuel Technologies into diesel prototype
- MATRIC was hired to replace Trucent, but large-scale extractions are pending
- UT/B&D tried low-cost methods for 5 strains, lab-scale, but oil extraction was low



Hexane Extraction of *Desmodesmus* (C046)



Crude oil yield =
1.669 kg (11.1%)

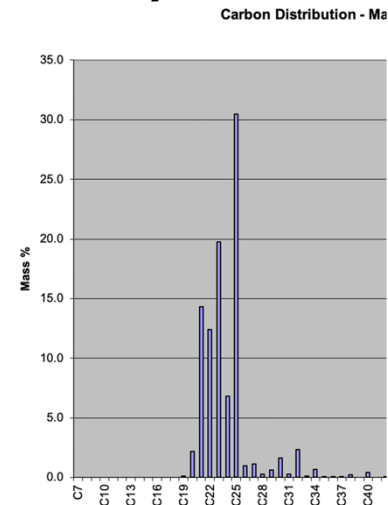


Table 1. CHNP and LoI tests by ALS Environmental on C046 biocrude

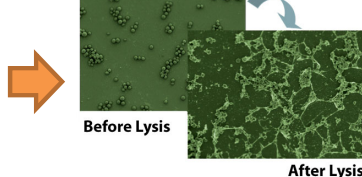
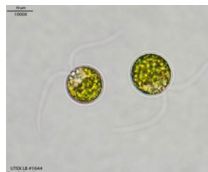
Property				
Sample ID	CC046 Biocrude 1	CC046 Biocrude 2	C046 Biocrude 3	Mean± Stdev
N per sample	1	1	1	
Carbon (wt%)	78.02	78.11	76.42	77.52±0.95
Hydrogen (wt%)	11.41	11.49	11.16	11.35±0.17
Nitrogen (wt%)	0.09	0.09	<0.05	0.06±0.05
Phosphorus (mg/kg)	99	94	87	93±6
Loss on Ignition (moisture free wt%)	99.95	99.94	100.05	99.98±0.06

LEA yield:
~14 kg for
feed trials



UT/B&D (OpenAlgae) Membrane Extraction

Pulsed-Electric Field
Lysis Unit



Membrane Oil
Recovery Skid



Lyser replaced with
acid pretreatment

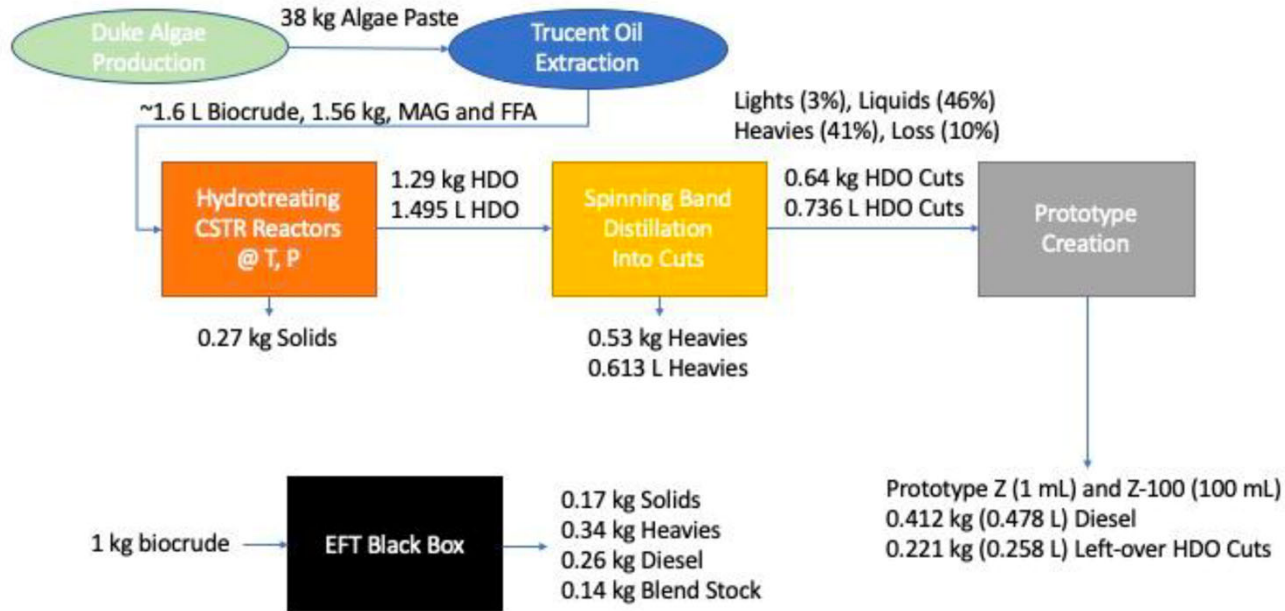
Membrane operated
without solvent

Summary:

Budget prevented installation of high-voltage lyser and explosion proof solvent extraction unit
Experimented with low-cost processes at lab-scale for 5 strains (below) - low oil recovery
Algae that was not processed provided to feed trials as whole meal

OCY3	6/8/18	OK	Frozen	~150 L	No	~3%	High ash, clogging, 295 mL naphtha/oil in 4 jars, dried for feed trials
S002	7/14/18	OK	Frozen	~200 L	No	~5%	High ash, clogging, 20 mL naphtha/oil, dried for feed trials
C046	10/4/18	OK	Frz Dr	771 g	Yes	~1%	Less clogging, 1.6 L naphtha/oil, algae discarded
C046X	12/5/18	OK	Frozen	18 L	Yes	<1%	Alternate naphtha flush trials, algae discarded
C018A	8/7/19	NC	Fresh	75 L	Yes	NA	Did not clog, 380 mL naphtha/oil, algae discarded
C018B	8/7/19	NC	Fresh	75 L	No	~2%	Did not clog, 350 mL naphtha/oil, algae discarded
C985	2/25/20	NC	Fresh	75 L	Yes	NA	Breakthrough occurred on membranes A&B, experiment failed

Task 4: *Product Assessment*: Hydroprocessing of C046 oil



**Successful
production of a
fuel product
from algae**

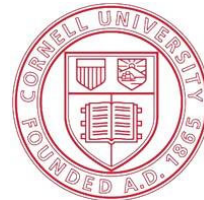


Task 4: ***Product Assessment: Poultry Feed***

Study 1: Effects of supplemental dietary full-fatted and defatted *Desmodesmus sp.* microalgae on growth performance, gut health, and excreta hydrothermal liquefaction of broiler chicks

Tao Sun, Kui Wang, Benjamin Wyman, Hanifrahmawan Sudiby, Guanchen Liu, Colin Beal, Schonna Manning, Zackary I. Johnson, Tolunay B. Aydemir, Jefferson W. Tester, and Xin Gen Lei

Manuscript accepted by Algal Research



Duke

NICHOLAS SCHOOL OF THE ENVIRONMENT



forging a sustainable future

Results of Study #1

- Supplemental algae and LEA improved growth by 11-40%
- Both types of biomass altered gene expression of inflammation in the duodenum and liver (17% to 2.2-fold)
- Both types of biomass up-regulated the intestinal tight junction protein (5-34%)
- Heating values of excreta from the C046 and LEA-fed chicks were 16% greater than the controls (average of 34 vs. 29 MJ/kg).
- Implication
 - **14% improvement of feed use efficiency = 24 mt feed saving = \$16 billion**

Next steps: test remaining algae (underway)

Task 4: ***Product Assessment: Aquafeeds Feed***

Develop and demonstrate a high-value salmon feed ingredient that is rich in protein, pigments and omega-3 fatty acids and is price competitive.

Target algae: C046 (*Desmodesmus* sp.)

Objective: To replace fishmeal in low-fishmeal feeds with algal products

Two feeding trials :

- Lab-scale trial with whole and lipid-extracted algae
- Farm trial with lipid-extracted algae



Trial with salmon smolts – laboratory scale

Results –Growth, Feed performance

Other key findings

Parameters	CD	LD	WD
Final weight (g)	689 ± 17	651 ± 17	699 ± 13
Condition factor	1.39 ± 0.01	1.37 ± 0.02	1.37 ± 0.02
Specific Growth Rate (% day ⁻¹)	0.88 ± 0.04	0.81 ± 0.03	0.90 ± 0.02
Thermal Growth Coefficient	3.17 ± 0.14 ^a	2.88 ± 0.13 ^b	3.25 ± 0.09 ^a
Feed Conversion Ratio	0.71 ± 0.01 ^a	0.96 ± 0.02 ^c	0.83 ± 0.01 ^b
Protein Efficiency Ratio	2.93 ± 0.04	2.18 ± 0.05	2.50 ± 0.04

Data presented as mean ± sem; n = 6 replicate tanks.

Different superscript letters indicate significant differences (P<0.05) in a row.

- Protein & energy digestibility of CD & LD were similar and significantly higher than of WD
- Fillet ∑ EPA & DHA was slightly higher for CD, while EPA content was slightly higher for WD&LD
- Expression of antioxidant, anti-inflammatory, immune-related and amino acid transport genes were higher in the alga-fed fish, particularly WD

SGR almost similar for CD and WD, and lowest for LD; these differences were more evident in TGC. FCR and PER best for CD followed by WD.

Trial with salmon – farm scale

Results –Growth, Feed performance

Parameters	CDF	LDF
Final weight (kg)	4.22 ± 0.11	4.10 ± 0.08
Specific Growth Rate (% day ⁻¹)	0.42 ± 0.01	0.41 ± 0.01
Thermal Growth Coefficient	3.75 ± 0.12	3.59 ± 0.11
Feed Conversion Ratio	1.13 ± 0.01^a	1.20 ± 0.00 ^b

Data presented as mean ± sem; n = 3 replicate sea cages
Different superscript letters indicate significant differences (P<0.05) in a row.

SGR and TGC were almost same for the two groups, despite the LDF feed being lower in protein and energy content compared to CDF feed. However, FCR was better for the CDF feed.

Other key findings

- The body proximate composition and energy contents did not exhibit any significant differences. However, the protein content was lower for LDF, reflecting the feed protein content, while the energy content was higher for the LDF, aided by the higher body lipid content.
- The flesh pigmentation was nearly the same for the two groups of fish.

- Lipid extracted C046 can effectively replace a portion of fishmeal in the feeds of both Atlantic salmon smolt and market size fish.
- The low FCR observed could be improved by optimizing the feed formulation.



Task 5: *Commercialization Analysis (TEA/LCA)*

Task 5.1 *Techno-Economic Analysis and Life Cycle Assessment*

Task Summary: Use TEA/LCA as a reiterative design tool to guide product development. Consider markets, competitors, and distribution.

M5.1: Deliver revised TEA/LCA for each product based on updated product specifications from the Target Product Workshop (M6) – **DONE - 2017**

M5.2 Updated TEA/LCA based on results of *Strain development* and initial *Recovery and Conversion* analyses (M18) – **DONE - 2018**

M5.3 Updated TEA/LCA based on final results of *Product Assessment* (M33) – **PENDING**

Summary:

- 1) TEA/LCA model¹ has been implemented with consideration for yields, strain composition, and target products
- 2) TEA/LCA will be conducted when oil extraction, oil hydroprocessing, and feed trials have been completed
- 3) End-product valuation strategies are being developed

1 - (Beal 2015, Gerber 2016, Beal 2018, Sills 2020)

End Product Valuation Strategies

End-Products:

- 1) Diesel blend stocks produced from hydroprocessed biocrude
- 2) Broiler chicken feed ingredients: whole algae and lipid-extract algae (LEA)
- 3) Salmon feed ingredients: whole algae and lipid-extract algae (LEA)
- 4) Human food ingredients: whole algae and lipid-extract algae (LEA)

Valuation Methods:

- 1) Replacement Value: Algae selling price equals value of ingredients it replaces

$$X_a = \frac{\sum_i^n X_i \cdot m_i}{\sum_i^n m_i} \left[\frac{\$}{t} \right]$$

- 2) Omega-3 Fatty Acid Added Value: Algae selling price based on protein + omegas

$$X_a = X_{a_base} + X_{\omega 3}$$

- 3) Consumer Values Marketing Added Value: Premium based on protein + marketing for consumer values, such as vegan, fisheries-friendly, USA-grown, pigmentation, etc.

$$X_a = X_{a_base} + X_{CV}$$

- 4) Improved Animal Health Added Value: Algae earn a premium for improving gut health, immunity, etc.

$$X_a = X_{a_base} + X_{AH}$$

MAGIC - Quad Chart Overview

Timeline

- October 1, 2015
- Sept 30, 2021

	FY20 Costed	Total Award
DOE Funding	1,000,524	\$5,240,313
Project Cost Share	35,292*	\$1,315,853 (20.1%)

Project Partners

Partners: ADM (5%), Bentley (2%), Bucknell (1%), Cornell (8%), Nord (8%), UTEX (8%), Shell (2%), UHH (23%), UHM (7%), USM (2%), Valicor (8%), Duke (26%)

- all project cost share has been met

Project Goals

Sustainable Algae Production: Demonstrate sustainable biorefinery systems via TEA and LCA

Algal Biomass Characterization, Quality, and Monitoring: Quantify efficacy of biofuel intermediates and co-products for multiple strains produced at ≥ 40 kg scale

Overall Integration & Scale-up: Show that “integrated” unit operations deliver sustainable production of biofuel intermediates and co-products

End of Project Milestone

Demonstrate a combined product value of $> \$1,000/\text{MT}$ that yields a biofuel intermediate that exceeds the RFS for advanced biofuels, $\text{EROI} > 3$, and sells for $< \$5/\text{gge}$. This will primarily be achieved by enhancing the value of the co-products (i.e. LEA)

Funding Mechanism

DE-FOA-0001162, TARGETED ALGAL BIOFUELS AND BIOPRODUCTS (TABB), 2014



MAGIC Summary

Overview This Consortium has demonstrated a fully “integrated” process flow for the production of biofuels and high-value bioproducts at a relevant scale.

Approach Demonstrate and validate high-value co-products – *drive down the cost of biofuel by increasing the value of algae “co-products”*

Technical Accomplishments/Progress/Results

- Demonstration of each project component
- Demonstration of overall integration
- Successful production of ‘finished’ fuel product from algae
- Successful demonstration of enhanced algae/LEA value for poultry and aquafeeds
- 35+ Peer-reviewed publications since project approval

Relevance Results address central BETO MYPP 2017 (and out year) goals. Rigorous demonstration and enhancement of co-product value, based on an integrated production process and efficacy trials are expected to increase revenues. Global impacts are significant.

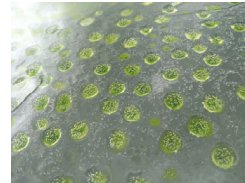
Thank you



Zackary Johnson: zij@duke.edu

<http://www.duke.edu/~zij>

<http://www.ml.duke.edu/webcam/algae/>



U.S. DEPARTMENT OF
ENERGY

EERE #DE-EE0007091

It's a team effort...Thanks!



Duke | NICHOLAS SCHOOL OF THE ENVIRONMENT
forging a sustainable future



Additional Slides

Task 1 – supplementary slide

Clarity on product specifications → biochemical characterization (AA/FA) of top strains to refine selection

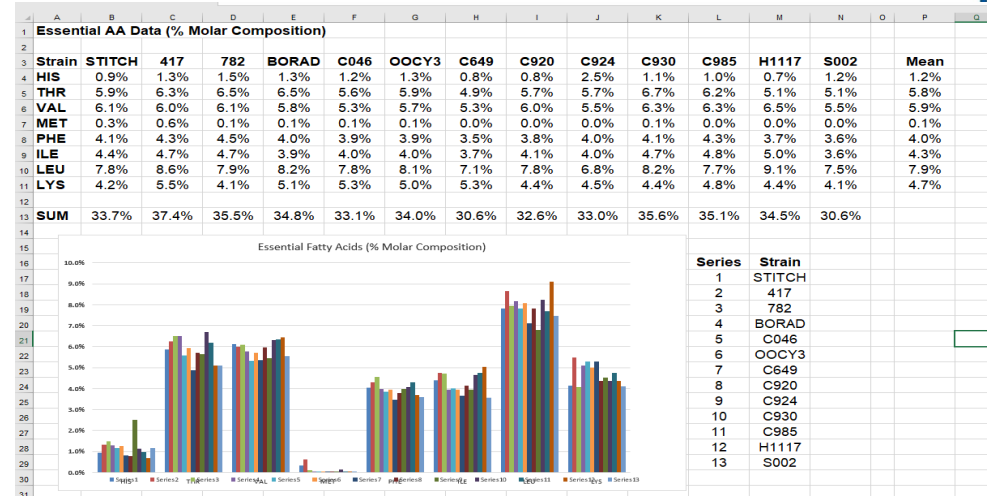
A. Food-quality Strain Rankings - Based on EPA Content

Strain	Genus	Rank	%EPA	%Lipid	%Ash
417	<i>Nannochloropsis oceanica</i>	1	9.4	44.5	8.4
930	<i>Pavlova</i> sp.	2	7.2	28.4	19.6
782	<i>Tetraselmis</i> sp.	3	6.2	32.3	7.2
985	<i>Tetraselmis</i> sp.	4	5.1	28.4	7.0
924	<i>Tetraselmis</i> sp.	5	4.0	21.7	15.4

B. Fuel-quality Strain Rankings - Based on FAME Content

Strain	Genus	Rank	%FAME	%Lipid	%Ash
417	<i>Nannochloropsis oceanica</i>	1	33.9	44.5	8.4
BORAD	<i>Scotiellopsis</i> sp.	2	31.2	37.2	4.0
920	<i>Chlorella</i> sp.	3	25.5	32.6	19.2
930	<i>Pavlova</i> sp.	4	24.5	28.4	19.6
649	<i>Chlorella</i> sp.	5	21.4	25.8	12.2

The distribution of essential amino acids (% molar composition) was conserved among the thirteen strains of microalgae



Summary:

Successfully delivered 10 strains for mass culture.
Growth at process development scale further determined strains for feedstock.



Need to know in future projects:

- Defined product specifications (chicken / egg)
- Seasonal temperatures for outdoor growth
- Planned methods of harvesting
- Nutrient status at harvest at scale

Task 4.2 supplementary slide: Protocol of Study 1

- **2 Expt, 396 chicks (12 cages/treatment, 5-6 chicks/cage)**
- **Microalgae:**
 - *Desmodesmus sp*
- **Corn and soybean meal basal diet**
- **Treatments: 0, 5% C046, and 5% LEA**
- **Free access to feed and water**
- **Duration: 2 weeks (starter period)**
- **Growth performance of chicks**
- **Blood, liver and duodenum: biochemical analysis**
- **Excreta: hydrothermal liquefaction (300° C, 60 min)**

Task 4.2 supplementary slide 2: Next Steps of the Poultry Research Task

- **Study 2: Effects of feeding EPA-rich *N. oceanica* on enrichments of n-3 fatty acids and 25 (OH) D3 in chicken meat:**
 - 180 chicks, 5 treatments, 6 cages/treatment, 5 chicks/cage, and 6-week feeding
 - Concentrations of n-3 fatty acids and 25(OH)D3 in tissues
- **Study 3: Effects of feeding microalgal DHA oil, EPA-rich *N. oceanica*, and 25(OH)D3 on leaky gut of broiler chickens:**
 - Dextran sodium sulfate was orally administered to induce leaky gut at weeks 3 and 6 (1 day before sampling and 1 chick/cage)
 - Fluorescein isothiocyanate (FITC-d) was given 2.5 hours before sampling to measure penetration (leaky) of gut
 - Concentrations of FITC-d in the blood and intestinal morphology
 - Tissue gene expression and protein production

Task 4.3 – supplementary slide #1 – Trial with salmon smolts – laboratory scale

Study Design

Fish and experimental groups in the feeding trial

Trial responsible	Nord University (NU)
Location	NU Research Station, Bodø, Norway
Fish; size at start	Atlantic salmon smolt; 349g
Experimental groups	Control (CD), Lipid extracted alga (LD), Whole alga (WD)
Tanks; replicates	800L seawater flow-through system (7°C); 6 replicate tanks/feed group
Feeding duration	77 days

Ingredient composition and analytical information of the experimental feeds

Key ingredients	Control (CD)	LE alga alga (LD)	Whole alga (WD)
Fish meal	10	5	5
Plant ingredients	64	64	64
Oils	18.5	18.4	17.8
C046	-	10	10

Proximate composition (%) and gross energy (kJ⁻¹)

Protein	47.9	47.9	47.9
Lipid	40.4	42.0	41.9
Ash	5.7	6.5	6.7
Energy	23.2	23.6	23.0

Task 4.3 – supplementary slide #2: Trial with salmon – farm scale

Study Design

Fish and experimental groups in the feeding trial

Trial responsible	Nord University (NU)
Location	GIFAS, Inndyr, Norway
Fish; size at start	Atlantic salmon adult; 1.83kg
Experimental groups	Control (CDF), Lipid extracted alga (LDF)
Cages; replicates	5m ³ sea cages; 3 replicate cages/feed group
Feeding duration	199 days

Ingredient composition and analytical information of the feeds

Key ingredients	Control (CDF)	LE alga alga (LDF)
Fish meal	15	10
Plant ingredients	64.3	61.7
Oils	23	22.9
C046	-	10

Proximate composition (%) and gross energy (kJ⁻¹)

Protein	49.2	43.8
Lipid	23.3	22.6
Ash	9.8	11.5
Energy	23.3	22.8

Task 4.3 – supplemental slide #3: Planned trials with salmon smolt – laboratory scale

Whole biomass of C018, C985, H1117 – March–September 2021

- Incorporating up to 5% of the algae (depending on the biomass available)
- Evaluating them as fishmeal/oil replacers and as functional feed ingredient through challenge studies.

Lipid extracted biomass of C018, C985, H1117 – June-December 2021*

- Incorporating up to 5% of the algae (depending on the biomass available)
- Evaluating them as fishmeal/plant protein replacers and as functional feed ingredient through challenge studies.

* The trial will commence when the LEA is made available; 6-8 months are required to complete the proposed tasks.



Task 5 supplementary slide: TEA/LCA Model Publications

Publications using TEA/LCA model:

Beal et al., Algal Research, 2015 – 100 ha algae production facility in Texas and Hawaii locations

Gerber et al., ES&T, 2016 – Target cultivation and financing parameters to achieve sustainable production

Walsh et al., Env Res Lett, 2016 – Integrated assessment model to evaluate GHG, land, and water impacts of global-scale algae production

Greene et al., Oceanography, 2016 and Greene et al., Earth's Future, 2017 – Evaluates pathways for algae to contribute to global sustainability

Beal et al., Earth's Future, 2018 – Using BECCS to produce CO₂, heat, and electricity to run algae production (algae is food, NOT the fuel)

Beal et al., Scientific Reports, 2018 – Algae production in Thailand for shrimp feed

Sills et al., Algal Research, 2020 – LCA methods for functional unit and allocation for algal biorefinery

Beal et al., Biomass and Bioenergy, 2021 – Sustainability assessment of alternative jet fuel for US DoD including algal pathways

Publications (35+)

Gerber LN, Tester JW, Beal CM, Huntley ME, Sills DL (2016). Target Cultivation and Financing Parameters for Sustainable Production of Fuel and Feed from Microalgae. Environmental Science & Technology.

<http://doi.org/10.1021/acs.est.5b05381>

Greene C, Huntley M, Archibald I, Gerber L, Sills D, Granados J, Tester J, Beal C, Walsh M, Bidigare R, Brown S, Cochlan W, Johnson Z, Lei X, Machesky S, Redalje D, Richardson R, Kiron V, Corless V (2016). Marine microalgae: Climate, energy, and food security from the sea. Oceanography 29: 10-15.

<http://doi.org/10.5670/oceanog.2016.91>

Greene CH, Huntley ME, Archibald I, Gerber LN, Sills DL, Granados J, Beal CM, Walsh MJ (2017). Geoengineering, marine microalgae, and climate stabilization in the 21st century. Earth's Future 5: 278-284.

<http://doi.org/10.1002/2016EF000486>

Hulatt CJ, Berecz O, Egeland ES, Wijffels RH, Kiron V (2017a). Polar snow algae as a valuable source of lipids? Bioresource Technology 235: 338-347. <http://doi.org/10.1016/j.biortech.2017.03.130>

Hulatt CJ, Wijffels RH, Bolla S, Kiron V (2017b). Production of Fatty Acids and Protein by Nannochloropsis in Flat-Plate Photobioreactors. PLOS ONE 12: e0170440. <http://doi.org/10.1371/journal.pone.0170440>



Publications (cont.)

Johnson ZI, Bidigare RR, Blinebry SK, Brown SL, Cullen JJ, Loftus SE, Redalje DG, Swink C, Van Mooy BAS (2017). Screening for Lipids From Marine Microalgae Using Nile Red. In: Lee SY (ed). Consequences of Microbial Interactions with Hydrocarbons, Oils, and Lipids: Production of Fuels and Chemicals. Springer International Publishing: Cham. pp 1-22.
http://doi.org/10.1007/978-3-319-31421-1_382-1

Kim J, Barcus M, Magnuson A, Tao L, Lei XG (2016a). Supplemental defatted microalgae affects egg and tissue fatty acid composition differently in laying hens fed diets containing corn and flaxseed oil. The Journal of Applied Poultry Research 25: 528-538.
<http://doi.org/10.3382/japr/pfw034>

Kim J, Magnuson A, Tao L, Barcus M, Lei XG (2016b). Potential of combining flaxseed oil and microalgal biomass in producing eggs-enriched with n – 3 fatty acids for meeting human needs. Algal Research 17: 31-37. <http://doi.org/10.1016/j.algal.2016.04.005>

Kiron V, Sørensen M, Huntley M, Vasanth GK, Gong Y, Dahle D, Palihawadana AM (2016). Defatted biomass of the microalga, *Desmodesmus* sp., can replace fishmeal in the feeds for Atlantic salmon. Frontiers in Marine Science 3. <http://doi.org/10.3389/fmars.2016.00067>



Publications (cont.)

Loftus SE, Johnson ZI (2017). Cross-study analysis of factors affecting algae cultivation in recycled medium for biofuel production. *Algal Research* 24, Part A: 154-166.

<http://doi.org/10.1016/j.algal.2017.03.007>

Walsh MJ, Gerber Van-Doren L, Sills DL, Archibald I, Beal CM, Lei XG, Huntley ME, Johnson Z, Greene CH (2016). Algal food and fuel coproduction can mitigate greenhouse gas emissions while improving land and water-use efficiency. *Environmental Research Letters* 11: 114006.

<http://doi.org/10.1088/1748-9326/11/11/114006>

Sørensen M, Gong Y, Bjarnason F, Vasanth GK, Dahle D, Huntley M, Kiron V.
(2017) *Nannochloropsis oceanica*-derived defatted meal as an alternative to fishmeal in Atlantic salmon feeds. *PLoS ONE*, 12(7): e0179907.

<https://doi.org/10.1371/journal.pone.0179907>

Gatrell S, Derksen T, O'Neil E, Lei XG (2017). A new type of defatted green microalgae exerts dose-dependent nutritional, metabolic, and environmental impacts in broiler chicks. *J. Appl. Poult. Res.*, 26(3): 358-366. <https://doi.org/10.3382/japr/pfx003>



Publications (cont.)

- Manor ML, Kim JG, Derksen TJ, Schwartz RL, Roneker CA, Bhatnagar RS, Lei XG (2017). Defatted microalgae serve as a dual dietary source of highly bioavailable iron and protein in an anemic pig model. *Algal Res.* 26: 409-414. <https://doi.org/10.1016/j.algal.2017.07.018>
- Walsh MJ (2017). Product-Focused Innovation and Value Creation are Needed to Drive Commodity-Scale Algae Production. Policy Commentary. *Industrial Biotechnology* 13(5): 223-227. <https://doi.org/10.1089/ind.2017.29097.mjw>
- Walsh, MJ, L Gerber Van Dorren, N Shete, A Prakash, U Salim (2018). Financial tradeoffs of energy and food uses of algal biomass under stochastic conditions. *Applied Energy* 210: 591-603. <https://doi.org/10.1016/j.apenergy.2017.08.060>
- Gong Y, Guterres HADS, Huntley M, Sørensen M, Kiron V (2018). Digestibility of the defatted microalgae *Nannochloropsis* sp. and *Desmodesmus* sp. when fed to Atlantic salmon, *Salmo salar*. *Aquaculture Nutrition* 24(1): 56-64. <http://dx.doi.org/10.1111/anu.12533>
- Beal CM, Archibald I, Huntley M, Greene CH, Johnson ZI (2018). Integrating Algae with Bioenergy Carbon Capture and Storage (ABECCS) Increases Sustainability. *Earth's Future*, 6: 524–542. <https://doi.org/10.1002/2017EF000704>



Publications (cont.)

Gatrell SK, Magnuson AD, Barcus M, Lei XG (2018). Graded levels of a defatted green microalgae inclusion in diets for broiler chicks led to moderate up-regulation of protein synthesis pathway in the muscle and liver. *Algal Res.* 29:290-296.

<https://doi.org/10.1016/j.algal.2017.11.039>

Lei XG (2018). Invited: Sustaining the future of animal feed protein. *Ind. Biotechnol.* 14(2).

<https://doi.org/10.1089/ind.2018.29120.xgl>

Sun T, Yin R, Magnuson AD, Tolba SA, Liu GC, Lei XG (2018). Dose-Dependent Enrichments and Improved Redox Status in Tissues of Broiler Chicks under Heat Stress by Dietary Supplemental Microalgal Astaxanthin. *J. Agri. Food Chem.* 66:5521-5530.

<https://doi.org/10.1021/acs.jafc.8b00860>

Magnuson AD, Sun T, Yin R, Liu G, Tolba SA, Shinde S, Lei XG (2018). Supplemental microalgal astaxanthin produced coordinated changes in intrinsic antioxidant systems of layer hens exposed to heat stress. *Algal Res.* 33:84-90. <https://doi.org/10.1016/j.algal.2018.04.031>

Beal CM, Gerber LN, Thongrod S, Phromkhunthong W, Kiron V, Granados J, Archibald I, Greene CH, Huntley ME (2018). Marine microalgae commercial production improves sustainability of global fisheries and aquaculture. *Scientific Reports.* 8:15064.

<https://doi.org/10.1038/s41598-018-33504-w>



Publications (cont.)

In Press

Tao L, Sun T, Magnuson AD, Qamar TR, Lei XG (2018). Defatted microalgae-mediated enrichment of n-3 polyunsaturated fatty acids in chicken muscle is not affected by dietary selenium, vitamin E, or corn oil1. *In Press at J. Nutr.*



Patents, Awards, and Commercialization

No patents have been applied for based on the work supported by DOE.

No special awards have been received.

All primary results from this project are being published in the open, peer-reviewed literature. The publications from this project – cited above – provide a comprehensive and detailed analysis of commercialization potential. This information will be available to anyone with access to the open literature.

