Bioconversion of Thermochemical Intermediates

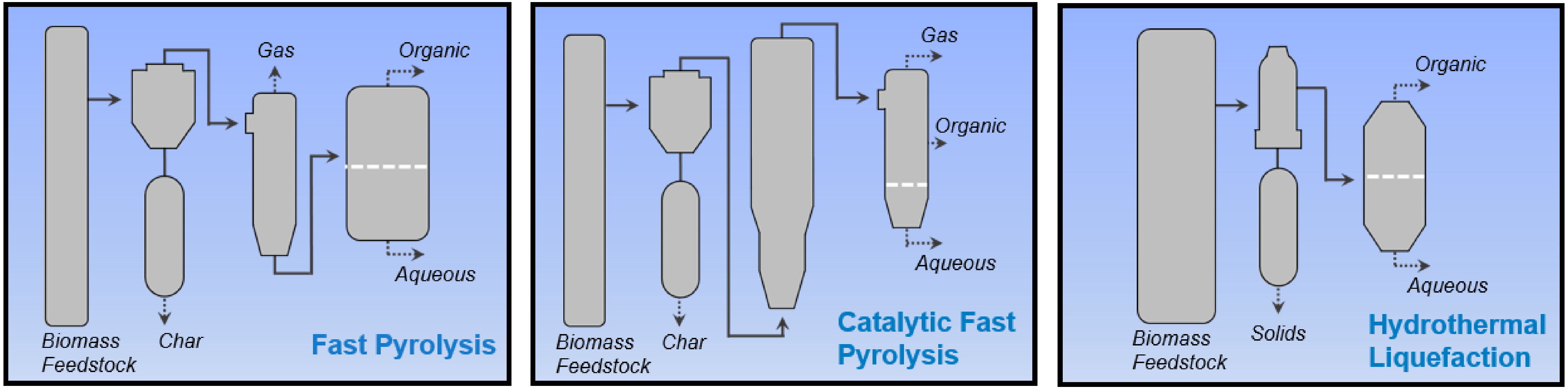
Technology Session Review Area: Biochemical Conversion & Lignin Utilization

PI: Gregg T. Beckham, National Renewable Energy Laboratory

Project overview

Goal: Adapt bio-funneling concept to waste carbon valorization in thermochemical (TC) processes

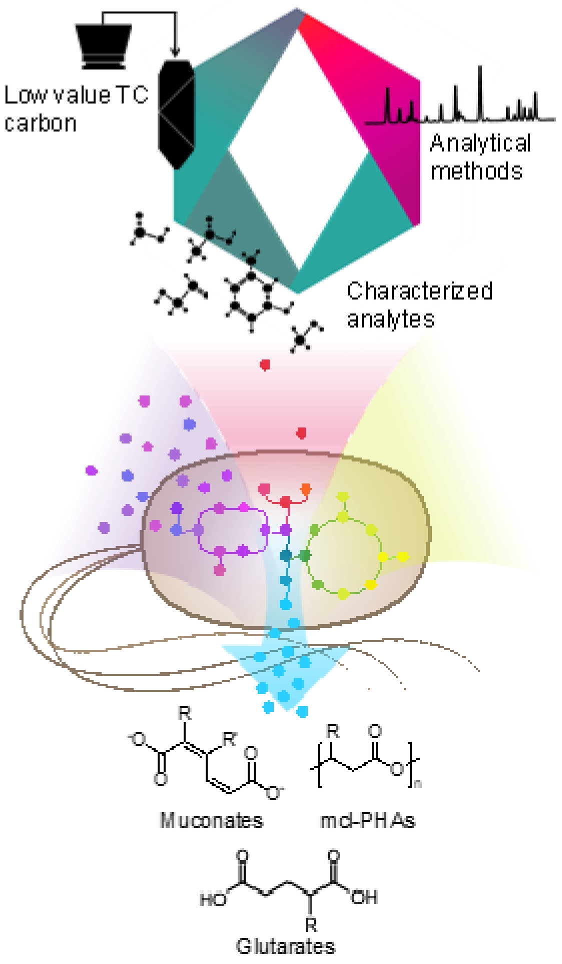
- Focus to date mostly on catalytic fast pyrolysis (CFP); applicable to hydrothermal liquefaction (HTL)
- Collaboration with Catalytic Upgrading of Pyrolysis Products (CUPP) and TC Analysis projects



Heilmeier Catechism:

- **Goal**: Biologically valorize low-value carbon from TC processes
- **Today**: Aqueous carbon **(~50-100 g/L)** lost via WWT (often up to 10% carbon loss)
- **Important**: WWT today is $0.24/gge cost to CFP processes
- **Risks**: Substrate toxicity and heterogeneity are main challenges

Management



Task 1: Analytics

- Led by analytical chemistry expert (LC-MS, GC-MS), B. Black
- Milestones: rigorous stream characterization and method development

Task 2: Bioconversion

- Staffed with strain engineering experts, A. Borchert, W.R. Henson
- Milestones: substrate catabolism and toxicity tolerance gains
- Collaborate with CUPP on substrates, performance-advantaged project on products

Project organization:

- Meetings: monthly for project, 1-on-1 with PI-task leads, PI-postdocs, *ad hoc* with CUPP and TC Analysis
- Coordinate samples and analytics data for TEA via single point of contact (B. Black)
- Ops & Project Mgrs – lab space, equipment, reporting, finances

Risks:

- Substrates change as upstream process conditions change – bioconversion efforts must be agile
- Substrate toxicity is substantial – tackled through pretreatment, strain engineering, and bioprocess development

Approach

Overall approach:

- Work with TC Analysis project to identify TEA and LCA drivers
- Conduct analytics method development and deployment for bench and pilot-scale streams
- Use genome-reduced *Pseudomonas putida* EM42 as our primary chassis for strain engineering
- Access substrates from the CUPP project and other BETO-funded TC efforts

Challenges:

- Overcoming substrate toxicity
- Maintaining sufficient agility to rapidly analyze and pivot to new substrates in emerging waste streams
- Project resources limited for bioprocess development

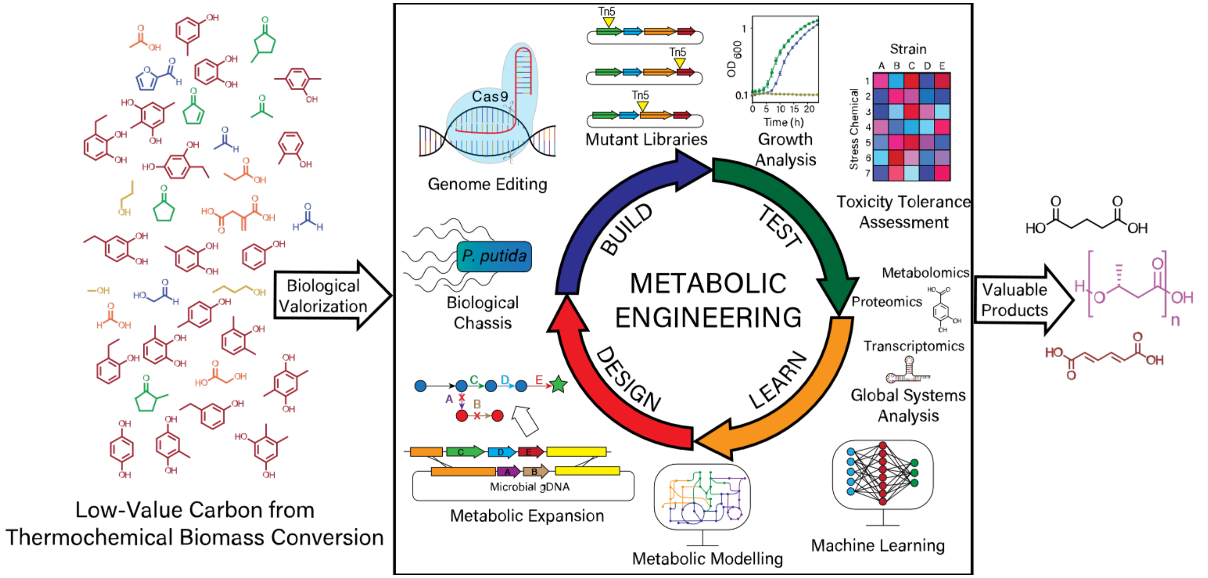


Image adapted from Borchert, Henson, Beckham, ***Curr. Opin. Biotech.***, in review

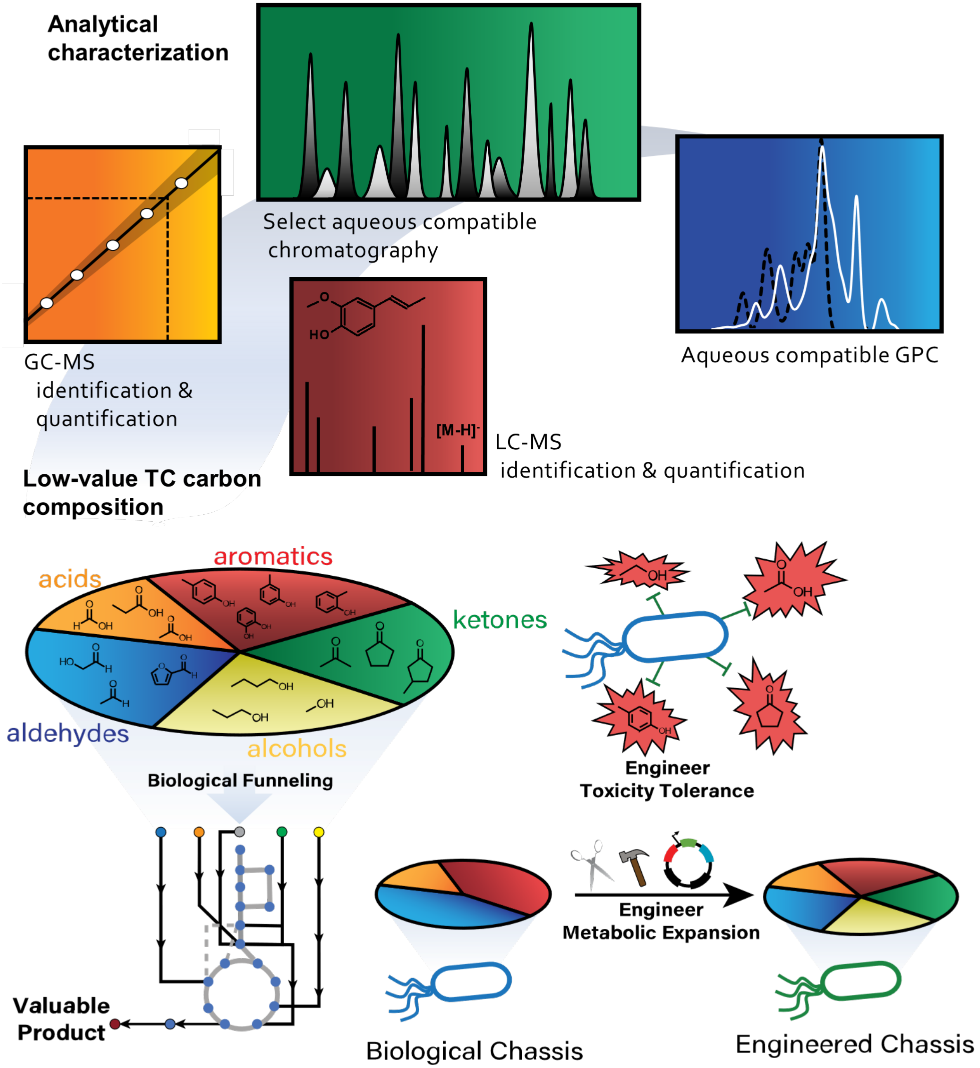
Major milestones, Go/No-Go Decisions:

- State-of-the-art is mostly unknown when we began
- FY20: 70% conversion of cyclopentanone (identified from recent CFP catalyst developments)
- FY21: ≥50% conversion of process-relevant/state-of-technology CFP wastewater stream
- FY22: ≥75% conversion at ≥75% selectivity with a \$0.50/gge contribution to MFSP

Impact

Scientific:

- Non-conventional, heterogeneous waste streams are prime for bioconversion & analytics
- High-impact papers on using microbes to valorize unconventional substrates (*EES, Nature Comm, PNAS,* etc.)
- New enzymes and toxicity tolerance gains broadly useful



Industrial:

- Re-engaging with RTI for CFP WW
- Collaborations with other industries on strains, enzymes, analytics

RTI INTERNATIONAL

Overall:

Bioconversion can reduce WWT costs and improve carbon conversion efficiency (up to 10%) in TC biomass conversion and improve biofuels MFSP (up to $1/gge)

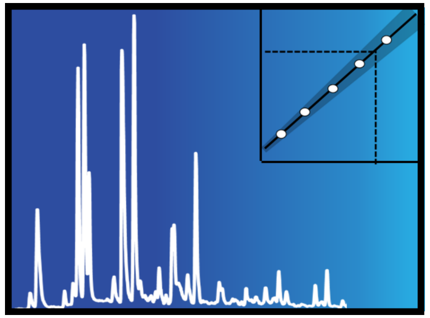
Outline of Progress and Outcomes

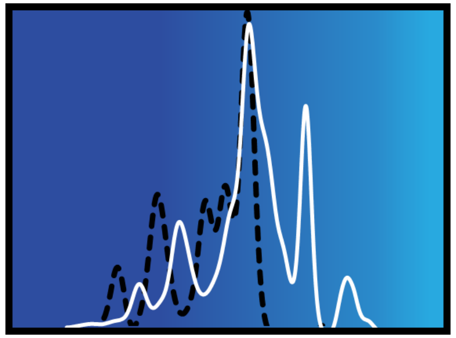
Analytics

- Aqueous-compatible analytical method development
- Aqueous stream composition survey across various upstream CFP catalytic conditions
- Ongoing analysis of new *ex situ* CFP stream

Bioconversion

- Expanding *P. putida* catabolism for CFP WW
- Engineering *P. putida* for methyl-muconate production for polymer applications
- Production and utilization of cyclic ketones
- Improving toxicity tolerance with detailed membrane characterization efforts





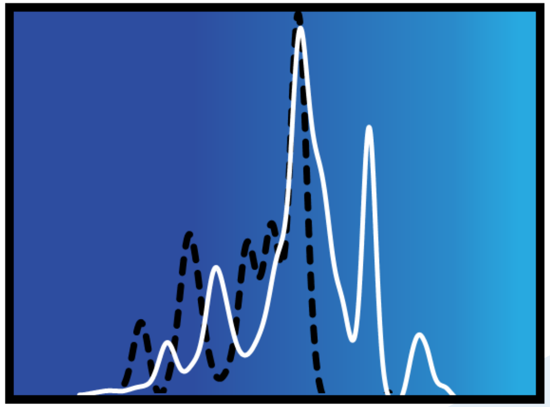
Substrate	g/kg	Native	Engineered
Acetic acid	4.14	✓	✓
Formic acid	0.98	✓	✓
Propanoic acid	0.1	✓	✓
2-Itaconic acid	0.65		
2-Hydroxyacetic acid	0.43	✓	✓
Acetaldehyde	0.51	✓	✓
2-Hydroxyacetaldehyde	0.16	✓	✓
Formaldehyde	1.07	✓	✓
Furfural	1.39		✓
Acetone	2.06		✓
Cyclopent-2-en-1-one	0.19		
Catechol	4.57	✓	✓
Phenol	2.94		✓
o-cresol	0.92		✓
m-cresol	2.14		✓
p-cresol	0.72		✓
3-Methyl catechol	1.14		✓
4-Methyl catechol	0.96		✓
2,5-Dimethylphenol	0.75		
2,6-Dimethoxyphenol	0.24		
2-Ethylphenol	0.28		✓
3-Ethylphenol	0.03		✓
Hydroquinone	0.7		
2-Methylbenzene-1,4-diol	0.22		
2,5-Dimethylbenzene-1,4-diol	0.29		
4,5-Dimethylbenzene-1,3-diol	0.09		
3-Ethyl catechol	0.1		✓
4-Ethyl catechol	0.46		
4-Ethylbenzene-1,3-diol	0.33		
Methanol	1.14	✓	✓
Propan-1-ol	0.25	✓	✓
Butan-1-ol	0.29	✓	✓
Total mass (g/kg)	**30.24**	13.64	24.96
Consumable carbon		45.11%	89.35%

Analytics: Method development for aqueous TC streams

Identification methods

understand compounds present

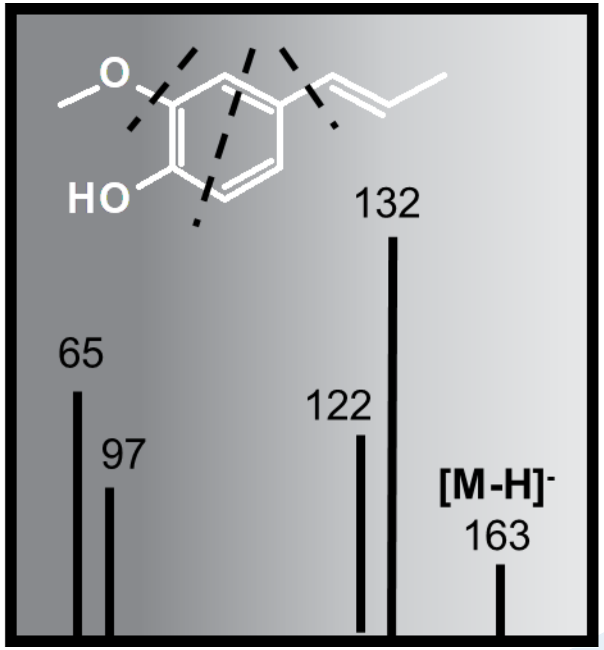
$_{aq}$GPC



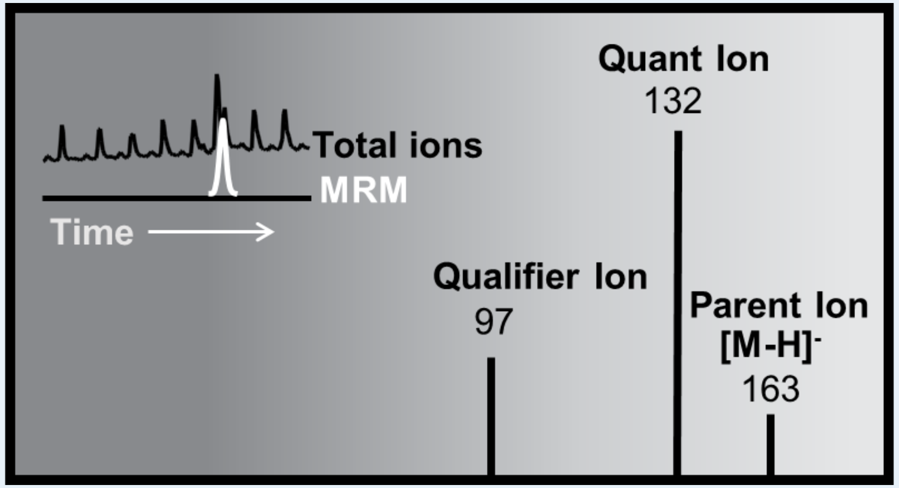
Analysis of aqueous TC streams is generally limited to GC and GC-MS

Combined with state-of-the-art LC and LC-MS techniques, upwards of 200 unique compounds can be characterized

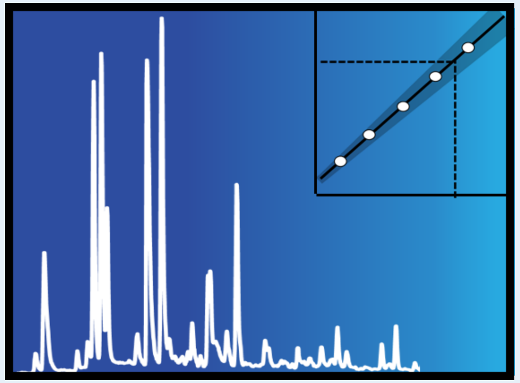
LC-MS$_{qTOF}$



LC-MS$_{triplequad}$



GC$_{wax}$-MS



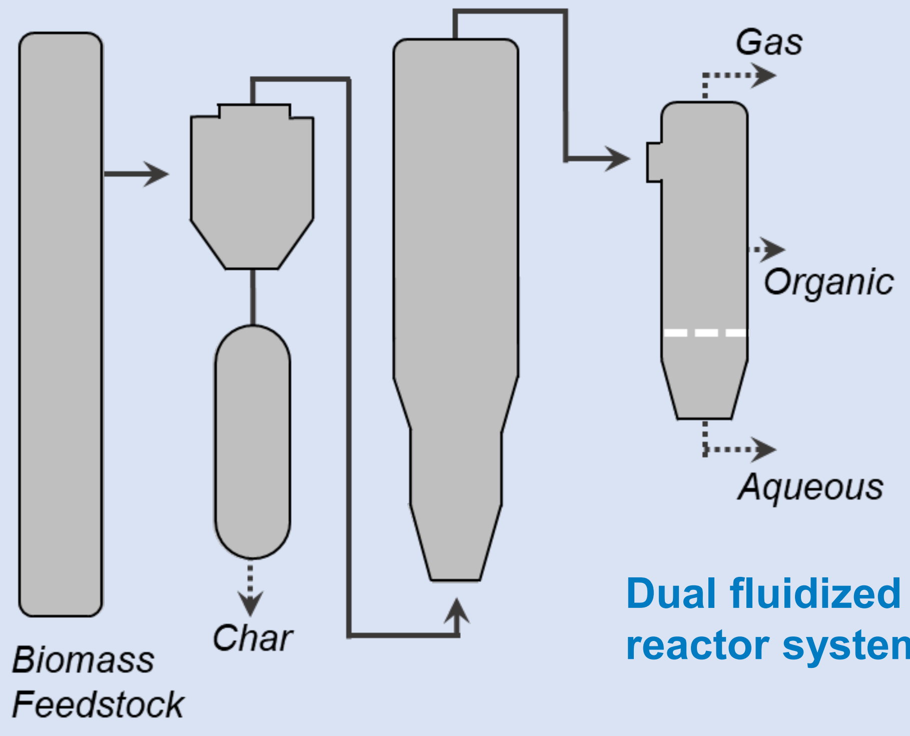
Upstream impact – direct feedback for process efficiency & optimization
Downstream impact – targeted valorization

Quantification methods

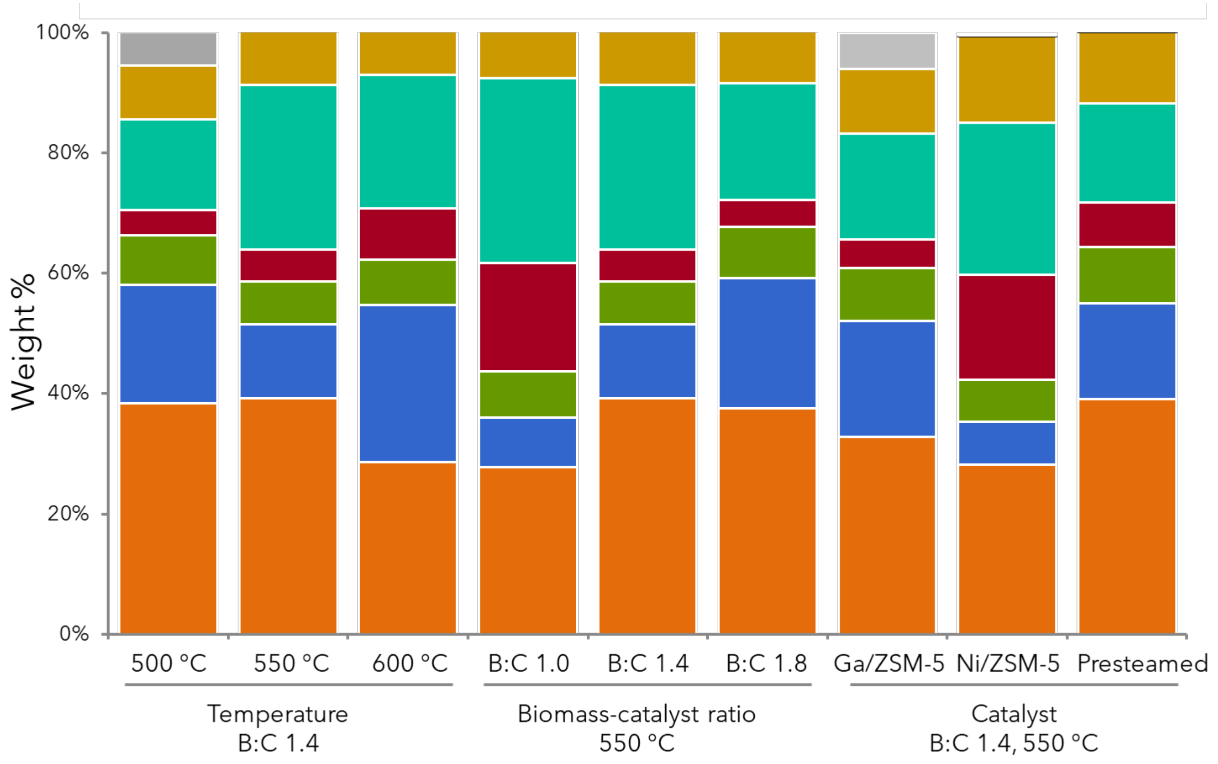
define amount of each compound present

Black et al., *ACS Sustain. Chem. Eng.* 2016 & Wilson et al., *Green Chem.* 2019

Analytics: *ex-situ* CFP aqueous stream – examining CFP conditions



Dual fluidized bed reactor system



100+ unique compounds characterized from the aqueous phase across multiple catalytic process conditions – to achieve near 100% carbon closure

Analytical Techniques used

- $GC_{DB\text{-}5}$-MS, $GC_{HP\text{-}1}$-MS GC_{wax}-MS
- $LC\text{-}MS_{ion\text{-}trap}$, $LC\text{-}MS_{qTOF}$, $LC\text{-}MS_{triplequad}$
- aqGPC, LC_{H+}-RID, LC_{Zn2+}-ELSD
- Ultimate and Proximate analyses

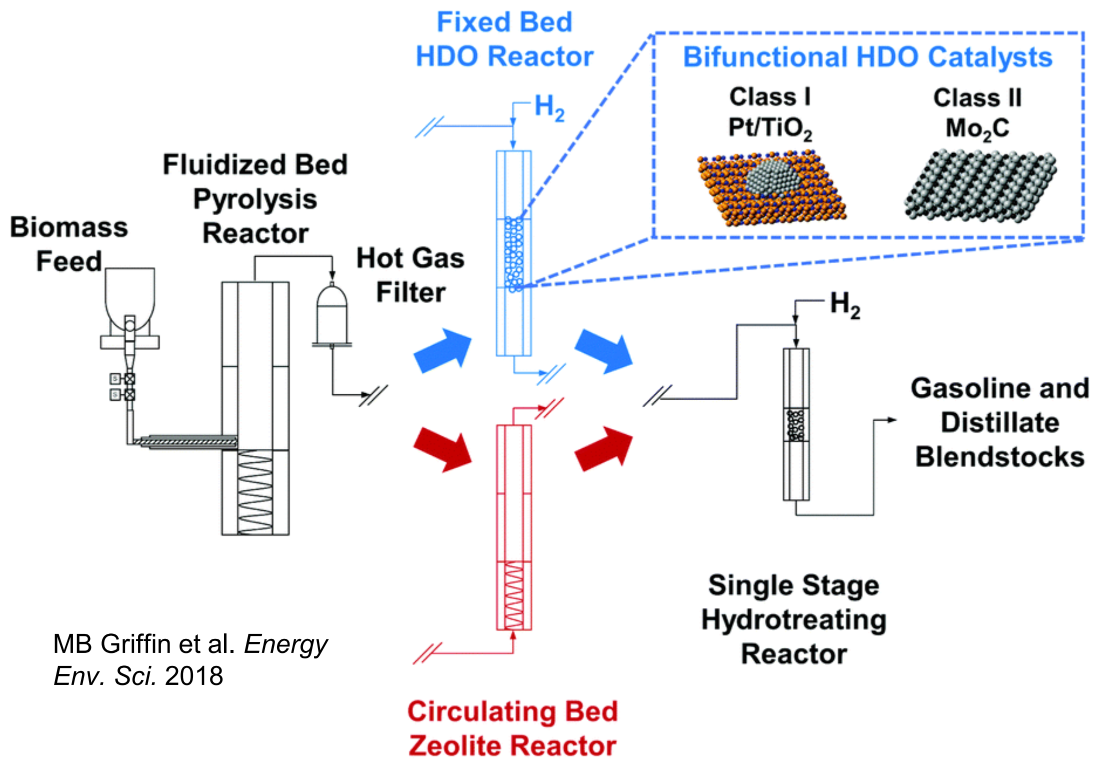
Black et al., in preparation

Analytics: Ongoing work on *ex-situ* CFP Pt/TiO_2 aqueous waste streams

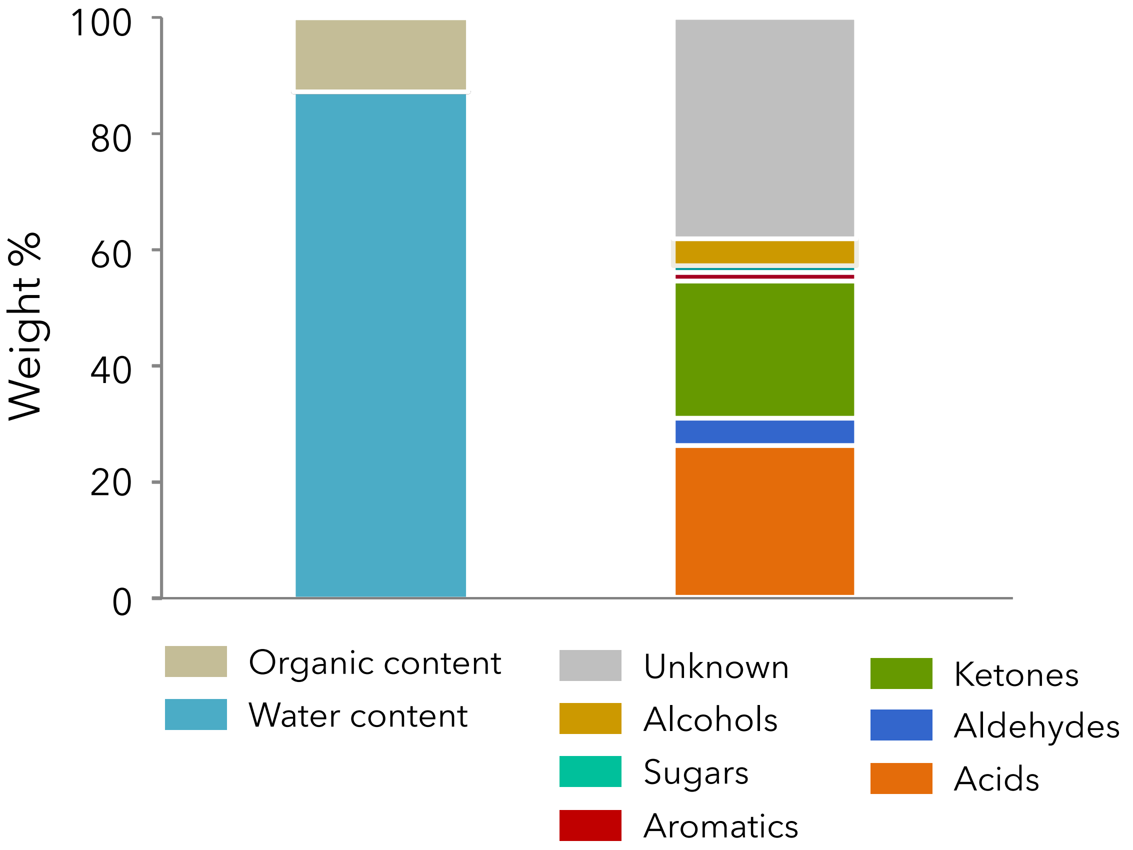
Goal: Upstream catalyst changes require characterization of new CFP aqueous phase

Approach: Use methods previously developed for CFP aqueous stream characterization

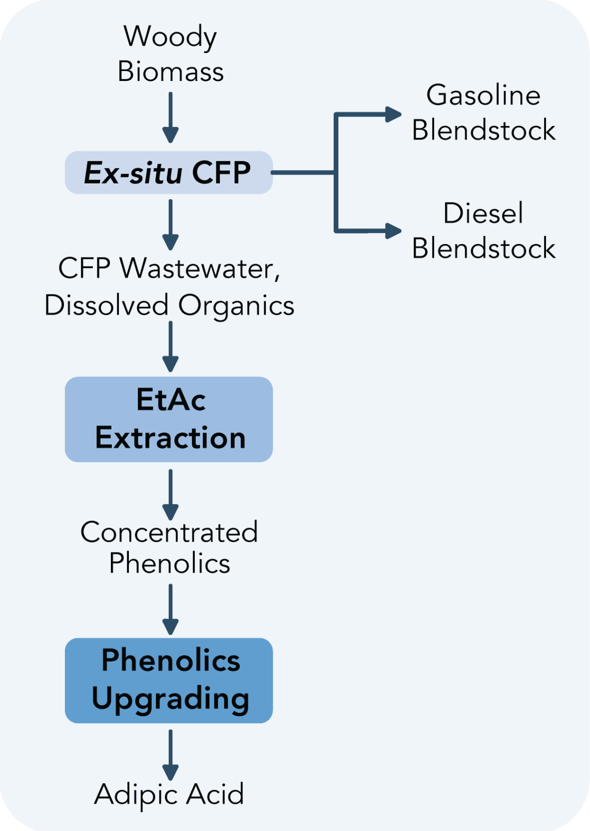
Current work: Adjusting methods to capture volatiles; efforts ongoing to close carbon balance (~40% remaining)

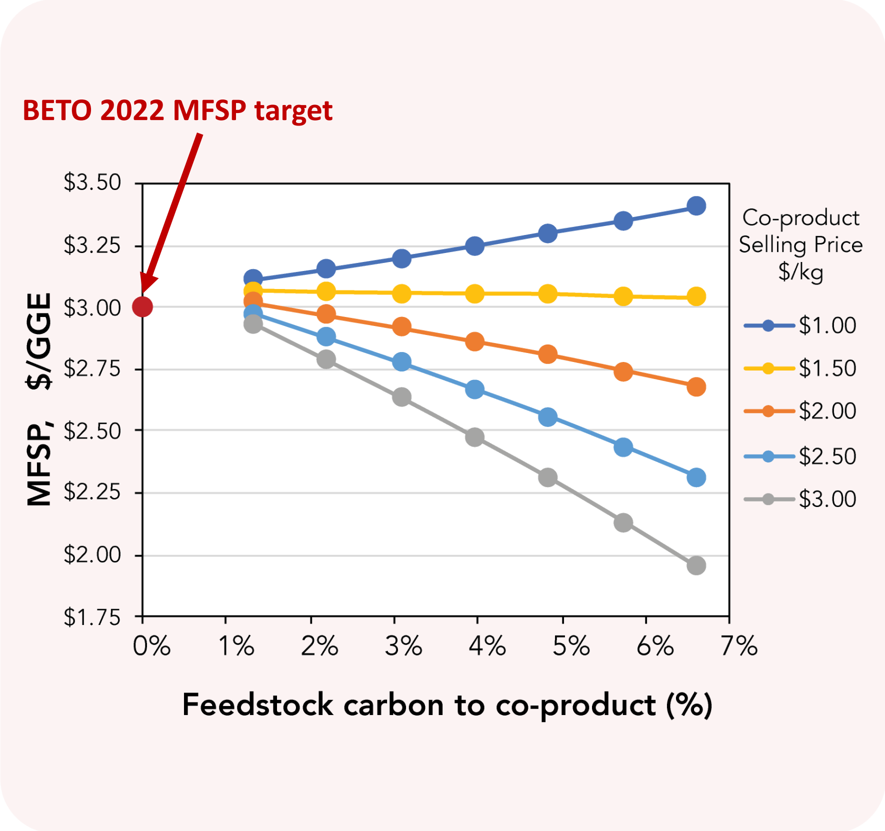


MB Griffin et al. *Energy Env. Sci.* 2018



TEA of biological upgrading approach





Henson, Meyers et al., in preparation

Work conducted in collaboration with Thermochemical Conversion Analysis project at NREL

WWT costs from Dutta et al. 2020. *Ex Situ Catalytic Fast Pyrolysis of Lignocellulosic Biomass to Hydrocarbon Fuels: 2019 State of Technology and Future Research*

MFSP reductions are possible through conversion of carbon in aqueous-phase to co-products

- Current design: treatment of CFP aqueous product stream costs $0.24/gge
- Proposed design: target performance-advantaged bioproducts (PABP) that command high selling prices
- Sensitivity analysis demonstrates co-product selling price and percent feedstock carbon diverted to co-products are significant drivers of MFSP reduction

CFP wastewater bioconversion: (alkyl)phenols

Substrate	g/kg	Native	Engineered
Acetic acid	4.14	✓	✓
Formic acid	0.98	✓	✓
Propanoic acid	0.1	✓	✓
2-Itaconic acid	0.65		
2-Hydroxyacetic acid	0.43	✓	✓
Acetaldehyde	0.51	✓	✓
2-Hydroxyacetaldehyde	0.16	✓	✓
Formaldehyde	1.07	✓	✓
Furfural	1.39		✓
Acetone	2.06		✓
Cyclopent-2-en-1-one	0.19		
Catechol	4.57	✓	✓
Phenol	2.94		✓
o-cresol	0.92		✓
m-cresol	2.14		✓
p-cresol	0.72		✓
3-Methyl catechol	1.14		✓
4-Methyl catechol	0.96		✓
2,5-Dimethylphenol	0.75		
2,6-Dimethoxyphenol	0.24		
2-Ethylphenol	0.28		✓
3-Ethylphenol	0.03		✓
Hydroquinone	0.7		
2-Methylbenzene-1,4-diol	0.22		
2,5-Dimethylbenzene-1,4-diol	0.29		
4,5-Dimethylbenzene-1,3-diol	0.09		
3-Ethyl catechol	0.1		✓
4-Ethyl catechol	0.46		
4-Ethylbenzene-1,3-diol	0.33		
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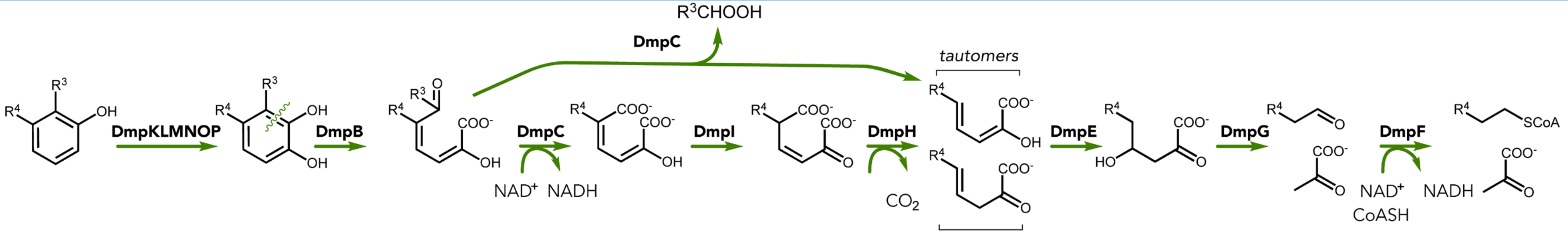

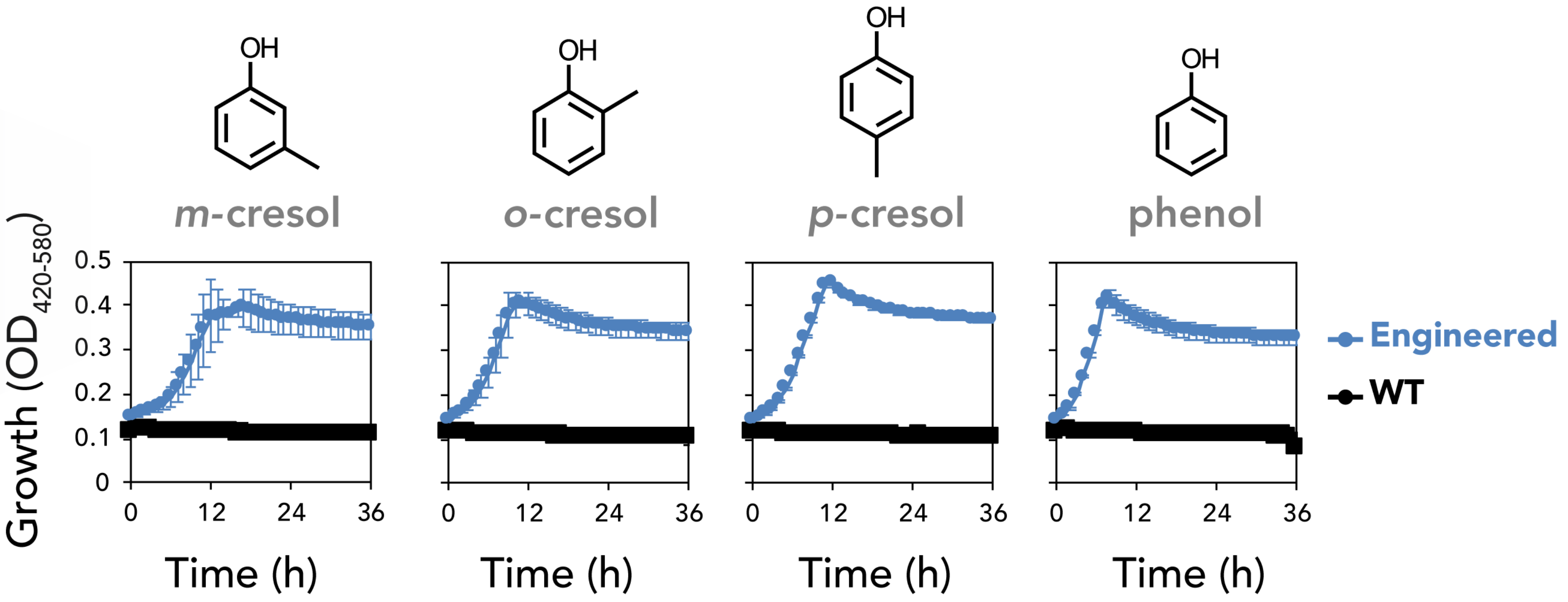

Strain engineering improves *P. putida* catabolic potential to ~76% wastewater carbon

- Engineered strain consumes methylphenols and its methylcatechol intermediates

Henson, Meyers et al., in preparation

CFP wastewater bioconversion: aldehydes, alcohols, ketones

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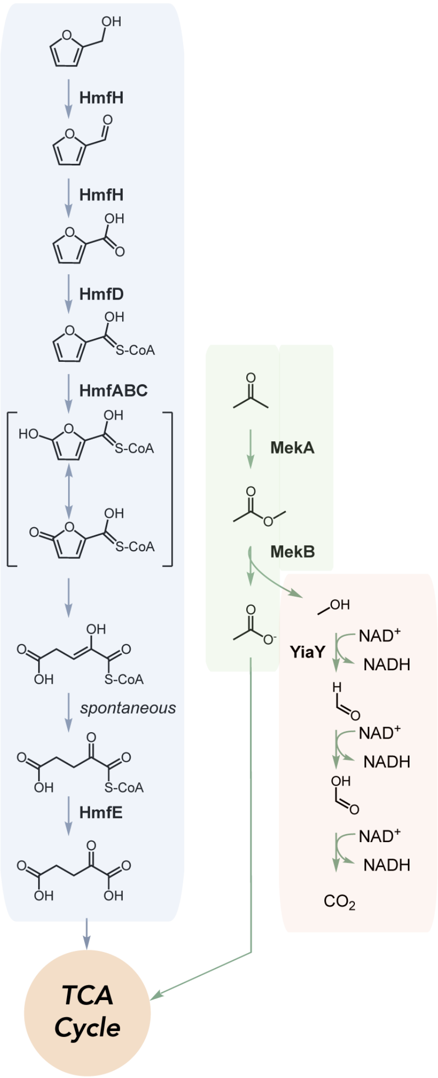

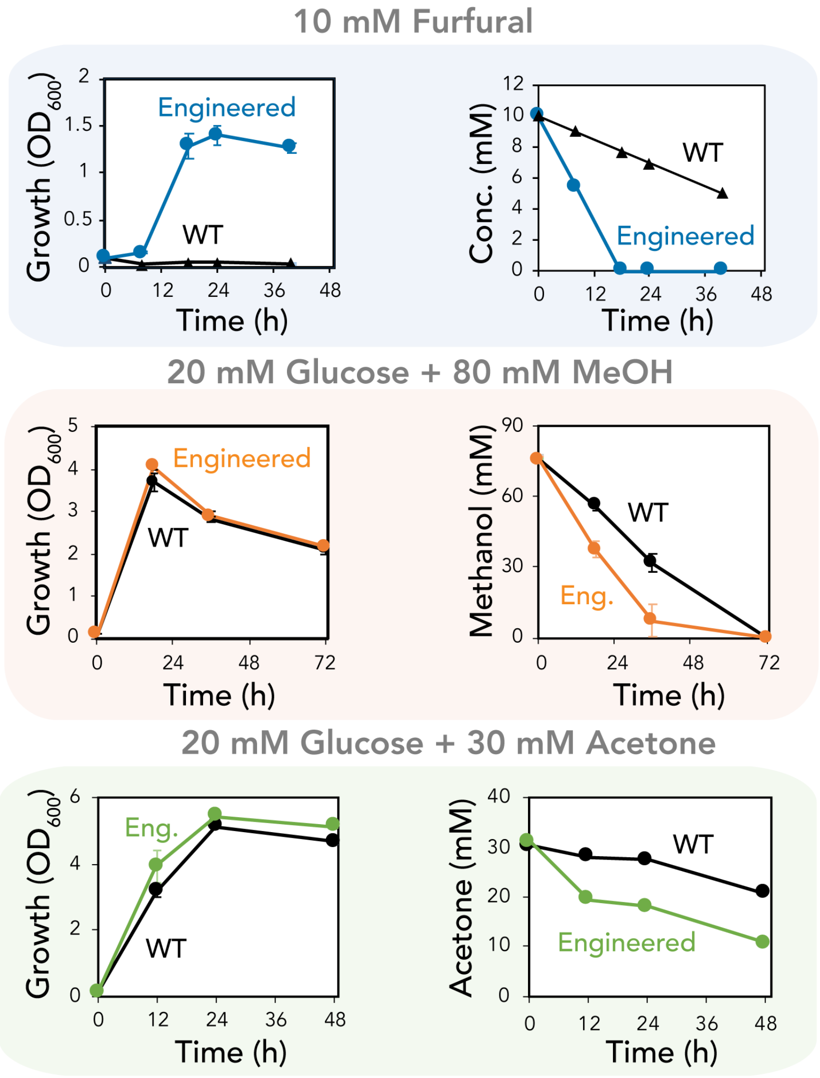

Henson, Meyers et al., in preparation

Engineering three additional pathways enables utilization of ~90% CFP wastewater carbon

- MekAB has broad substrate specificity for catabolism of other CFP wastewater compounds

CFP wastewater bioconversion: combining all pathways

CFP wastewater

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Mock CFP wastewater

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Acetaldehyde	0.51	✓	✓
Formaldehyde	1.07	✓	✓
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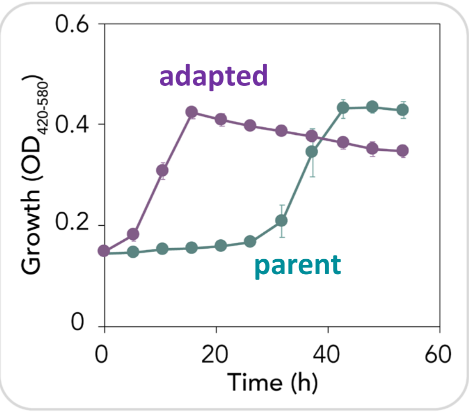
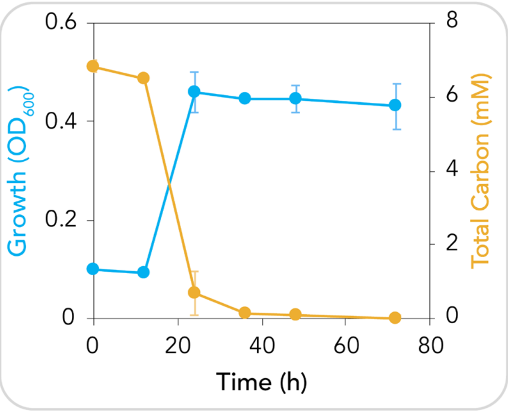
3-Methyl catechol	1.14		✓
4-Methyl catechol	0.96		✓
Methanol	1.14	✓	✓
Propan-1-ol	0.25	✓	✓
Butan-1-ol	0.29	✓	✓
Total mass (g/kg)	25.22	12.95	25.22
Consumable carbon		51.35%	100.00%

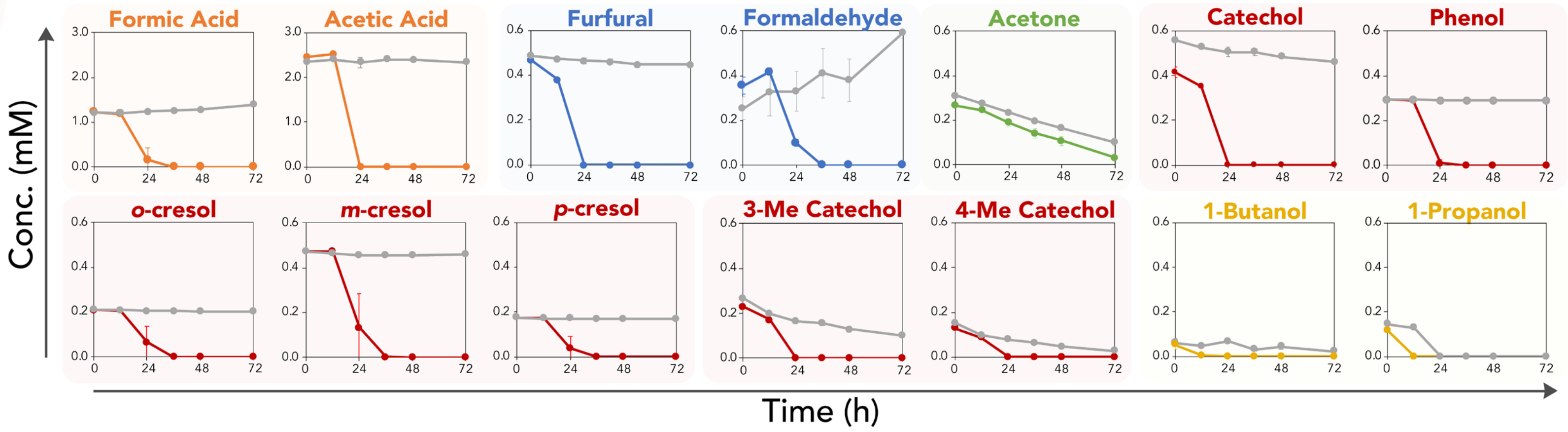
ALE reduces lag phase



Growth using mock CFP stream



Utilization of mock CFP stream components



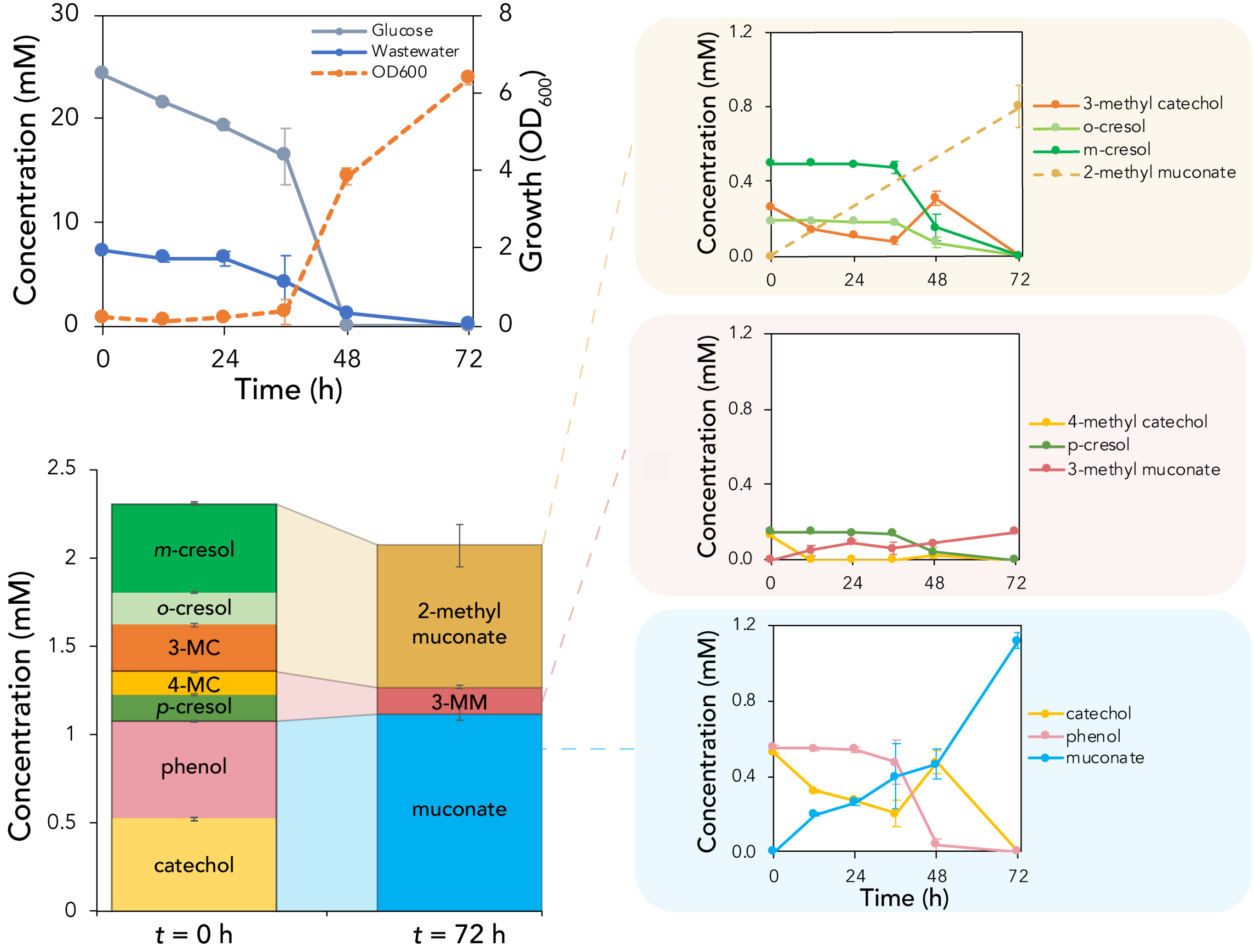
~30 kb of engineered pathways enable 100% conversion of mock CFP wastewater stream

- Adaptive laboratory evolution (ALE) improved lag phase of strains using mock CFP stream
- Whole genome sequencing underway to identify causative mutations

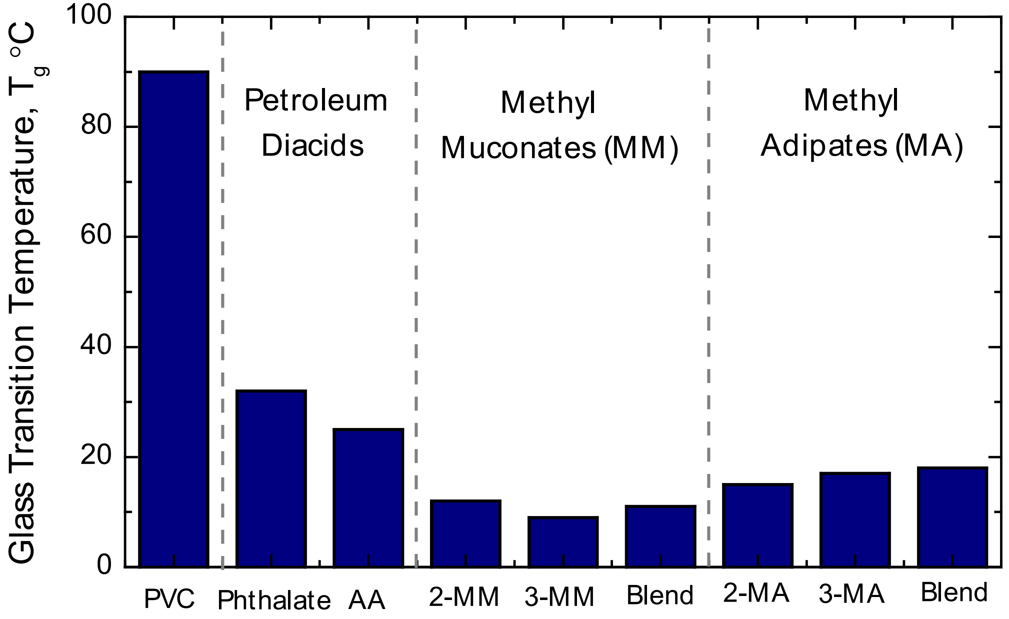
Henson, Meyers et al., in preparation

CFP wastewater bioconversion: Methyl-muconate products

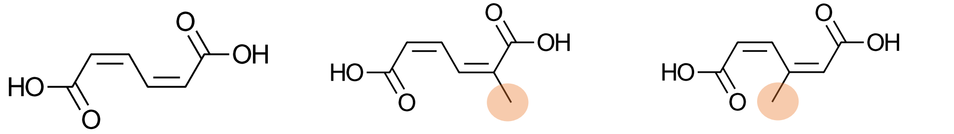
Conversion of mock wastewater to methyl-muconates



Methyl muconates as potent plasticizers



muconic acid **2-methyl muconic acid** **3-methyl muconic acid**



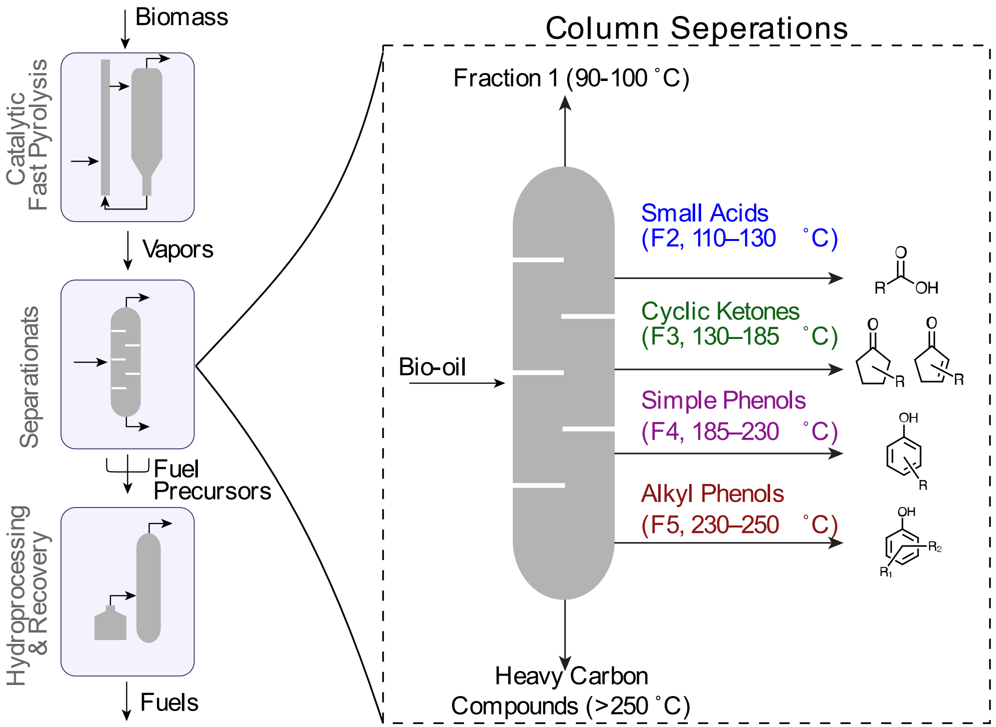
Methyl group provides unique properties vs. muconic acid

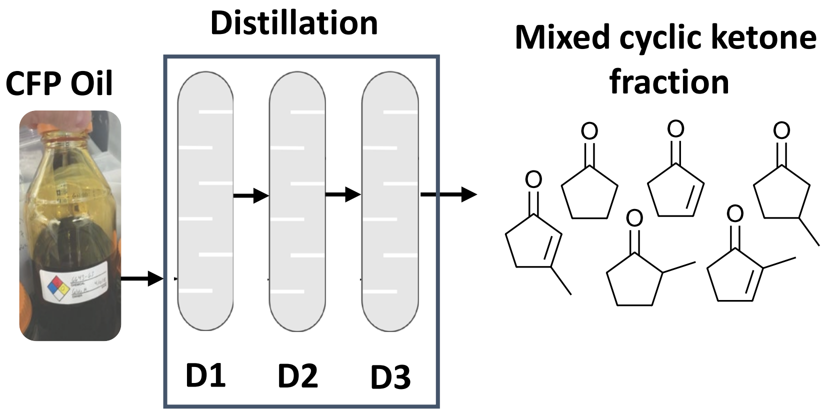
- Collaboration with Synthesis and Analysis of Performance-Advantaged Bioproducts project

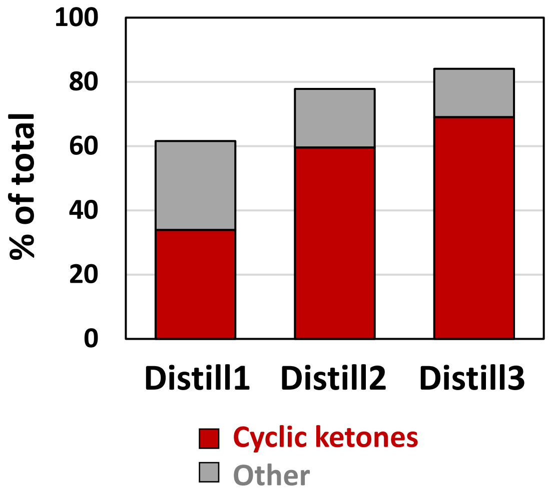
- **Mock CFP wastewater converted to methyl-muconate blend at ~90% yield**
- **Methyl muconate blend offers performance advantage as a plasticizer**

Henson, Meyers et al., in preparation

Cyclic ketones are also a major pyrolysis product

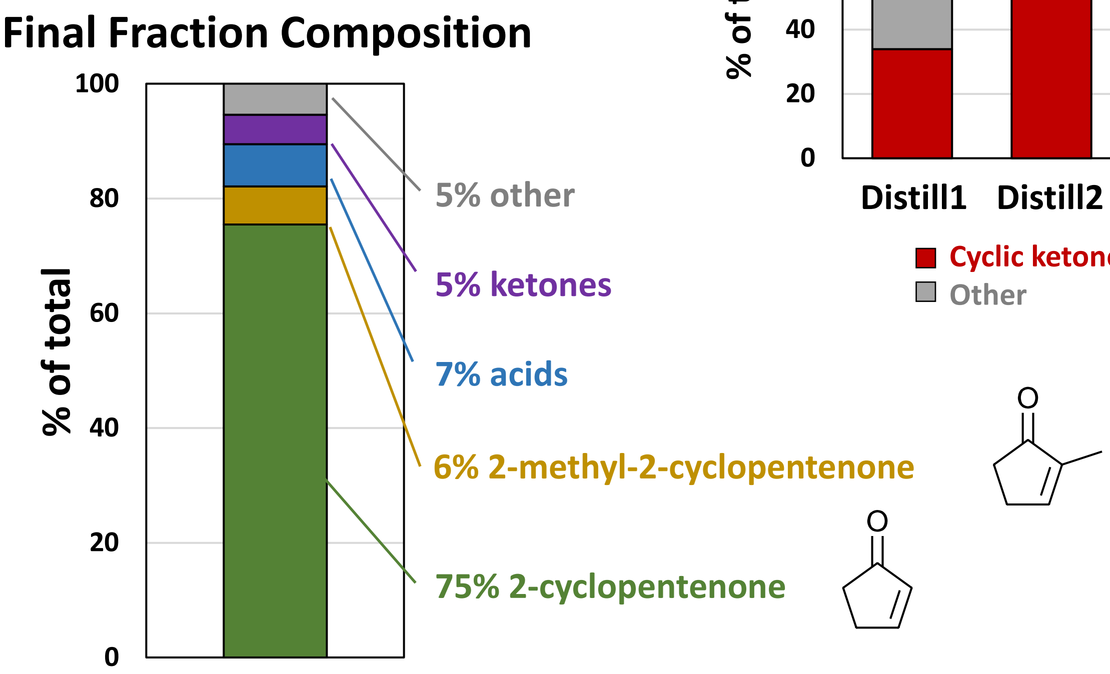






Cyclic ketones comprise 6.2% of total carbon from distillate fraction of Pt/TiO_2 CFP stream – not currently targeted/useful for fuels

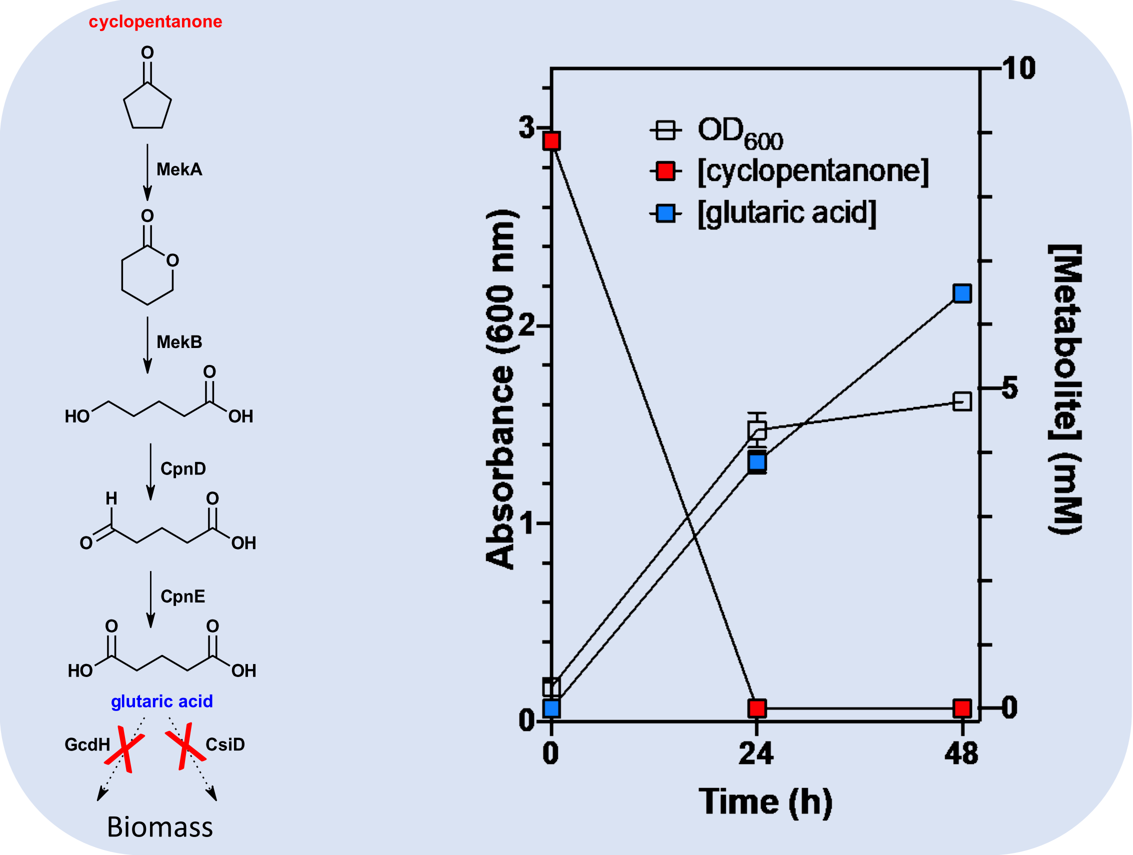
- Consecutive distillations of Fraction 3 achieves streams with mixed cyclic ketones at 69% purity
- Demonstrates responsiveness to new substrates
- Targeted for bioconversion



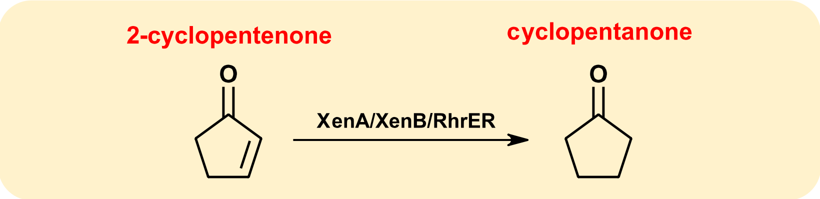
Wilson et al., in preparation

Engineering utilization of cyclic ketones

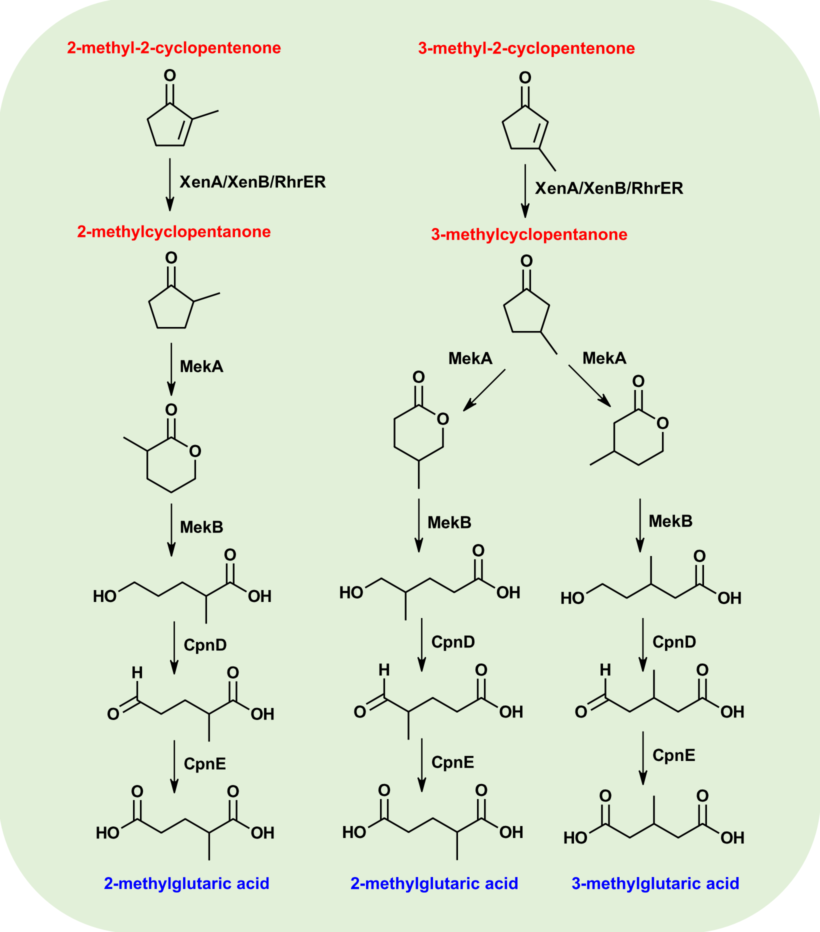
Conversion of cyclopentanone to glutaric acid



Conversion of 2-cyclopentenone to cyclopentanone



Create methyl-glutarates from methyl-cyclopentanones

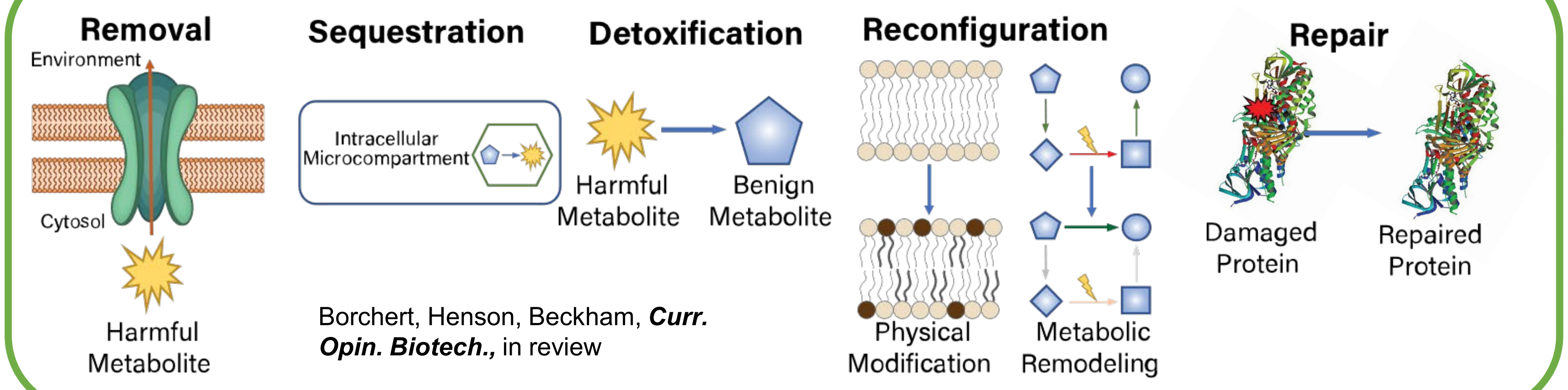


Expressed heterologous MekAB and CpnDE to enable conversion of cyclopentanone to glutaric acid (> 70% yield)

- Inducing expression of alkene reductases, XenA, XenB, or RhrER to reduce 2-cyclopentenone to cyclopentanone
- Assessing promiscuity of involved enzymes to produce methyl-glutaric acids from methylated cyclic ketones

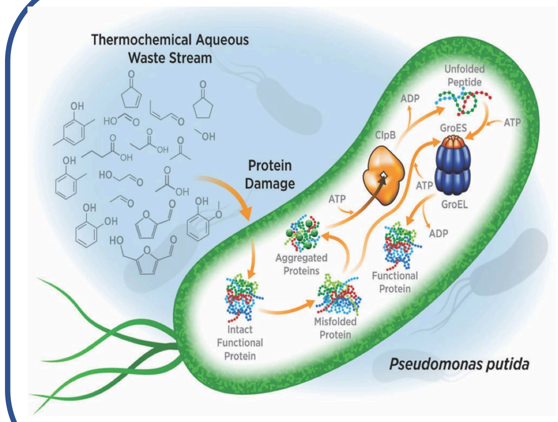
Strategies for improving metabolite stress tolerance

Many strategies for improved stress tolerance:



Borchert, Henson, Beckham, ***Curr. Opin. Biotech.,*** in review

Chaperone over-expression improves TC WW tolerance



Jayakody et al. *EES* 2018

TC WW exposure causes induction of MexEF:OprN efflux pump: Over-expression ongoing

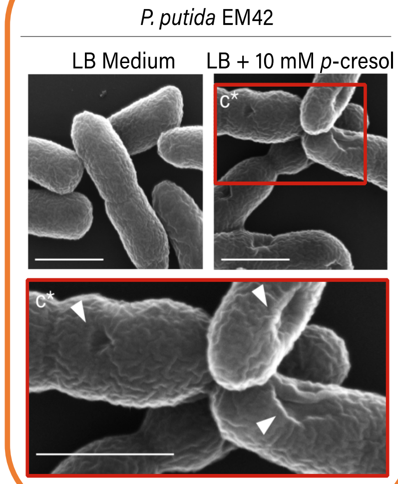
RNA and Protein Expression Following treatment with TC Wastewater

Gene	Description	RNA-seq	Proteomics
PP_3425	multidrug efflux RND membrane fusion protein MexE		
PP_3426	multidrug efflux RND transporter MexF		
PP_3427	multidrug efflux RND outer membrane protein OprN		

log2: 3, 2, 1, 0, -1, -2, -3

Data from Jayakody et al. EES 2018, ongoing work

***p*-cresol from TC WW perturbs membrane**

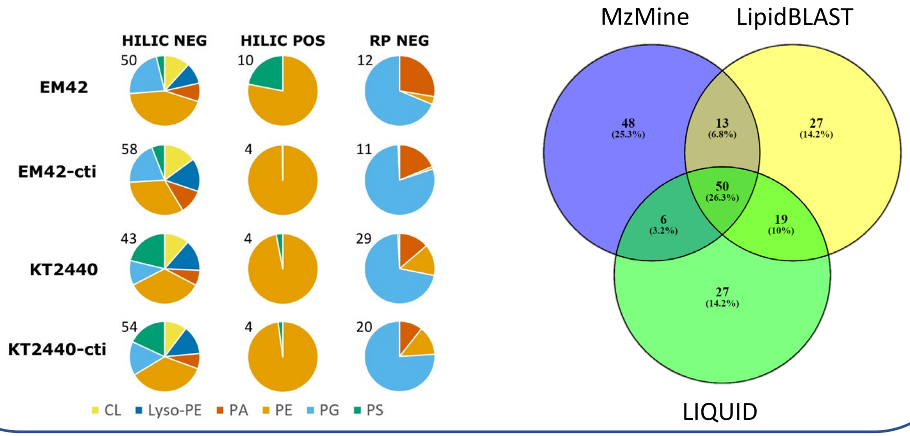


Ongoing: improve TC wastewater tolerance via membrane engineering and efflux pump expression

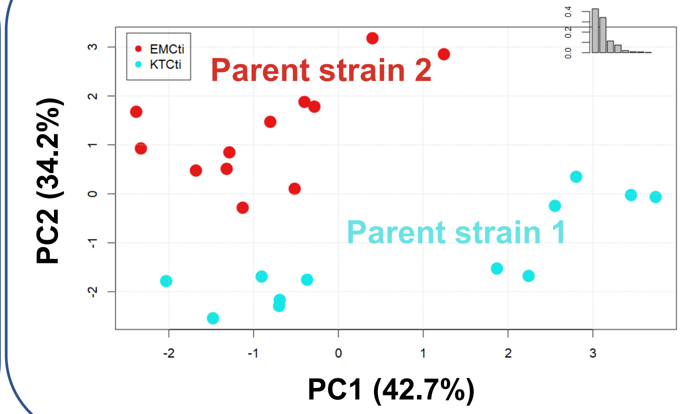
Membrane characterization & engineering for toxicity tolerance

Lipidomics platform development

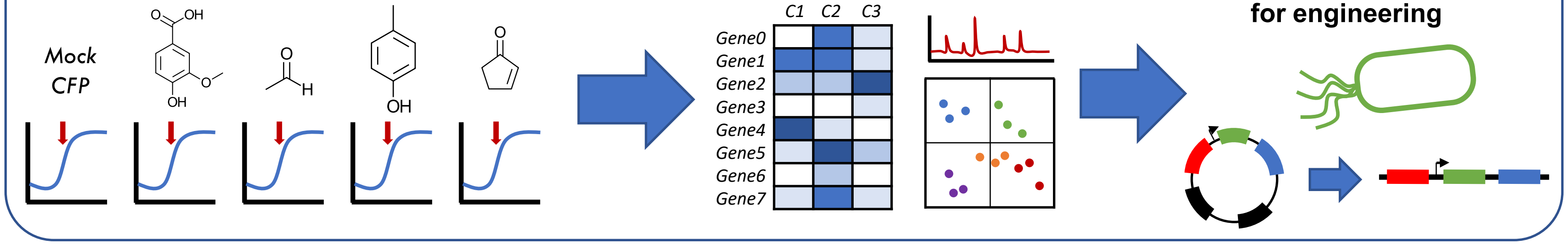
6+ lipid classes, 60+ species identified



Comparison between different strains



Multi-omics for toxicity tolerance



Detailed membrane characterization using multiple –omics for improving toxicity tolerance

- Developed lipidomics platform for high-throughput characterization of *P. putida* lipidome (with ORNL)
- Implementing platform with other -omics to identify additional native tolerance mechanisms

Reeves, Henson, et al., ASMS Meeting June 2020. Henson, Reeves, Borchert et al. in preparation.

Summary

Overview

- Aim to use biology and analytics to convert waste carbon in TC processes

Management

- Agile project that works closely with upstream and downstream projects to maximize impact

Approach

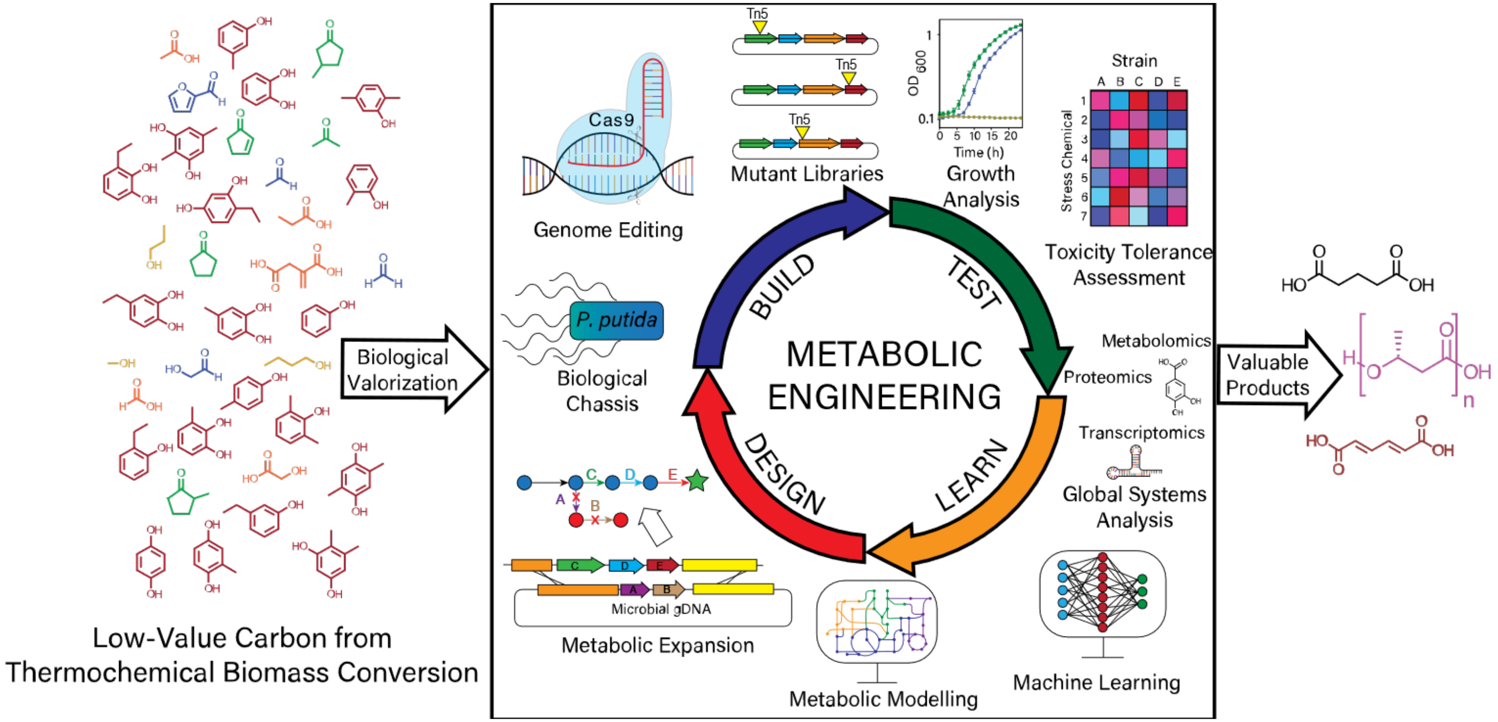
- Analysis and analytics-guided efforts directly inform bioconversion efforts

Impact

- Learnings here are applicable well beyond pyrolysis and HTL

Progress and Outcomes

- Achieved ~100% carbon closure and ~90% conversion for key CFP streams



Quad charts

Timeline

- Active Project Duration: 10/1/2019 – 9/30/2022
- Total Project Duration: 10/1/2014 – 9/30/2022

	FY20	Active Project (FY20-22)
DOE Funding	$400,000	$1,200,000

Project Partners

BETO projects: Catalytic Upgrading of Pyrolysis Products (2.3.1.314) and Thermochemical Platform Analysis (2.1.0.302), and Synthesis and Analysis of Performance-Advantaged Bioproducts (2.3.4.501)

Barriers addressed

- Ct-I Development of process capable of processing high moisture feedstocks in addition to conventional AD
- Ct-J Identification and evaluation of potential co-products

Project Goal

Adapt bio-funneling concept to waste carbon valorization in thermochemical (TC) processes

End of Project Milestone

Demonstrate the biological conversion of realistic CFP fractions (and HTL fractions, bandwidth permitting) to enable ≥75% conversion extents with ≥75% selectivity to the target products in a bioreactor system at the 0.5-2 L scale. The end-of-project milestone metrics should be in line with a projected ≥$0.50/gge contribution to the MFSP.

Funding Mechanism

Bioenergy Technologies Office FY20 AOP Lab Call (DE-LC-000L071) – 2019

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Collaborators:
David Dayton (RTI), Robert Hettich (ORNL), Nick Wierckx (Jülich)

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Additional Slides

Responses to previous reviewer comments

- This is another P. putida platform development program that focuses on aqueous thermochemical process waste streams that may have up to 10% waste carbon. Valorizing this level of waste to a useful coproduct, as well as cleaning up the wastewater stream itself, appears to be a worthy target, although it depends on the concentration (a well-developed TEA will bear out the cost-benefit). Strain tolerance is a key issue, and the discovery of a misalignment of substrate composition from TC processing at different scales is key, and presents a challenge. New strategies, strain engineering methods, and potential PABPs have all resulted from this project so far, which is good progress.
- The PIs have presented a scientifically compelling approach to conversion of carbon that would otherwise be lost. The products are reasonable and recognized in the chemical industry. While the specific application of this work to waste streams faces considerable challenges, the broader applicability to lignin conversion is promising, as it is trying to address lignin's structural heterogeneity.
- This project tackles the difficult task to biologically transform low levels of carbon-containing compounds that can be recovered from thermochemical effluents into valuable biochemicals. Although difficult, this objective can reduce wastewater treatment costs incurred by biorefineries, while generating valuable coproducts. This project has made excellent progress. For example, the team has uncovered important differences in aqueous stream compositions as a function of pilot versus bench-scale TC processes. They also made significant improvements to strain tolerance. Codigestion of thermochemical effluents with other industrial process streams represents an important opportunity for future investigation.
- The team has done well coordinating across two very different work programs from analytics to bioengineering and extending the funnel of carbon utilization. Also commended for their ability to work well across different disciplines from process, catalyst team, analytics, and bio team all coordinating well, which speaks to good team management. The project is a success overall, though there is a question as to whether the value return to fuel GGE is sufficient to justify further work, albeit this can contribute learning to lignin bioconversion. Also, there is an interesting development for methyl branched mucconates, which perhaps can be reapplied to lignin programs. Excellent utilization of the waste carbon.
- Overall this is a strong project that has made a number of good technical achievements. I would like to see more explicit statements of the translation of the accomplishments in the basic science, which seem to be numerous, into practical results. For example, what level of carbon utilization has been achieved in bioprocessing of either mock or real catalytic fast pyrolysis wastewater streams, and what is the yield of useful products? How does the performance in these type of tests compare with the necessary targets for commercial viability, and what are the key remaining challenges to be overcome? Maybe all of that is part of future work. This team has done some very solid work to bring the underlying technology along quite far, so it would be good to see their vision as to how that gets translated to a viable, ready-to-deploy technology.
  - We have achieved 89% carbon utilization of a pyrolysis wastewater stream. The product yield to muconates is approximately >50% of the carbon. The product yield to PHAs, inherently, will be approximately 30%–40% of the carbon. In terms of developing a bioprocess towards a TEA model, we have just developed a rigorous and comprehensive TEA model, and we are baselining strain performance and bioprocess performance against these targets now.

Publications, presentation, awards, and commercialization

In preparation

Brenna A. Black, William E. Michener, Michelle L. Reed, Kristiina Iisa, and Gregg T. Beckham, *Ex-situ* catalytic fast pyrolysis aqueous stream composition and the influence of process parameters for valorization, In preparation.

William R. Henson‡, Alex W. Meyers‡, Lahiru N. Jayakody, Annette DeCapite, Christopher Kinchin, Brenna A. Black, William E. Michener, Christopher W. Johnson, and Gregg T. Beckham. Enabling biological conversion of pyrolysis-derived wastewater using modular catabolic pathways in Pseudomonas putida. In preparation.

William R. Henson‡, Alex W. Meyers‡, Nicholas Rorrer‡, Caroline Hoyt, Todd Vander Wall, Rui Katahira, Jared J. Anderson, Brenna A. Black, William E. Michener, Lahiru Jayakody, Davinia Salvachúa, Christopher W. Johnson, Marc A. Hillmyer, and Gregg T. Beckham. Methyl muconic acids as a new biomass-derived plasticizer: from cresols to polymer incorporation via biological upgrading. In preparation

William R. Henson‡, Andrew J. Borchert‡, William E. Michener, Alexander W. Meyers, Brenna E. Black, and Gregg T. Beckham. Biological upgrading of purified cyclic ketone aqueous waste streams to C5 diacids in Pseudomonas putida KT2440. in preparation

William R. Henson‡, David T. Reeves‡, Andrew J. Borchert‡, Lahiru N. Jayakody, Alexander W. Meyers, Robert L. Hettich, and Gregg T. Beckham. Omics driven membrane engineering of Pseudomonas putida for improved thermochemical aqueous stream tolerance. In preparation.

In review or revision

Andrew J. Borchert, William R. Henson, Gregg T. Beckham*, Challenges and opportunities in biological funneling of heterogeneous and toxic substrates beyond lignin, in review at *Curr. Opin. Biotech*.

In print

Morgan M. Fetherolf, David J. Levy-Booth, Laura E Navas, Jie Liu, Jason C Grigg, Andrew Wilson, Rui Katahira, Gregg T. Beckham, William M. Mohn, Lindsay D. Eltis*, Characterization of alkylguaiacol-degrading cytochromes P450 for the biocatalytic valorization of lignin, *PNAS* (2020), 117, 25771-25778.

Nolan Wilson, Abhijit Dutta, Brenna A. Black, Calvin Mukarakate, Kim Magrini, Joshua A. Schaidle, William E. Michener, Gregg T. Beckham, Mark R. Nimlos*, Valorization of aqueous waste streams from thermochemical biorefineries, Green Chem. (2019) 21, 4217-4230.

Kirsten Davis, Laura R. Jarboe, Davinia Salvachua, Gregg T. Beckham, Zhiyou Wen, Robert C. Brown, Ryan G. Smith, Marjorie R. Rover*, Promoting microbial utilization of phenolic substrates from bio-oil, *J. Ind. Microbiol. Biotech.* (2019) 46, 1531-1545.

Publications, presentation, awards, and commercialization

In print

Melodie M. Machovina‡, Sam J.B. Mallinson‡, Brandon C. Knott‡, Alexander W. Meyers‡, Marc Garcia-Borràs‡, Lintao Bu, Japheth E. Gado, April Oliver, Graham P. Schmidt, Daniel Hinchen, Michael F. Crowley, Christopher W. Johnson, Ellen L. Neidle, Christina M. Payne, Kendall N. Houk*, Gregg T. Beckham*, John E. McGeehan*, Jennifer L. DuBois*, Enabling microbial syringol utilization via structure-guided protein engineering, *PNAS* (2019) 116, 13970-13976.

Wing-Jin Li, Lahiru N. Jayakody, Mary Ann Franden, Matthias Wehrmann, Tristan Daun, Bernhard Hauer, Lars M. Blank, Gregg T. Beckham*, Janosch Klebensberger*, Nick Wierckx*, Laboratory evolution reveals the metabolic and regulatory basis of ethylene glycol metabolism by *Pseudomonas putida* KT2440, *Environ. Microbiol.* (2019), 21, 3669-3682.

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Anne Starace, Brenna A. Black, David D. Lee, Elizabeth C. Palmiotti, William E Michener, Jeroen ten Dam, Michael J. Watson, Gregg T. Beckham, Kimberly A. Magrini, and Calvin Mukarakate, Characterization and catalytic upgrading of aqueous stream carbon from catalytic fast pyrolysis of biomass. *ACS Sustainable Chemistry and Engineering* (2017) 5, 11761-11769.

Michael T. Guarnieri, Mary Ann Franden, Christopher W. Johnson, Gregg T. Beckham. Conversion and assimilation of furfural and 5-(hydroxymethyl)furfural by Pseudomonas putida KT2440. Metabolic Engineering Communications (2017) 4, 22-28

Jeffrey G Linger, Sarah E Hobdey, Mary Ann Franden, Emily M Fulk, Gregg T Beckham. Conversion of levoglucosan and cellobiosan by *Pseudomonas putida* KT2440. Metabolic Engineering Communications (2016) 3, 24-29

Brenna A. Black, William E. Michener, Kelsey J. Ramirez, Mary J. Biddy, Brandon C. Knott, Mark W. Jarvis, Jessica Olstad, Ofei D. Mante, David C Dayton, and Gregg T Beckham, Aqueous stream characterization from biomass pyrolysis. *ACS Sustainable Chemistry and Engineering* (2016) 4, 6815-6827.

Presentations

“For the characterization of strain-resolved *Pseudomonas putida* lipidomes.” American Society for Mass Spectrometry Annual Meeting, June 2020.

Patents and Patent Applications

Patent applications

Thermochemical wastewater valorization via enhanced microbial toxicity tolerance using cis trans isomerase (Cti) : ROI-19-74, pending

Issued patents (cumulative)

Enzymatic Cleavage of Cellobiosan: ROI-14-71

Engineered strain of *p.putida* to enable consumption of ethylene glycol during production of PET: ROI 17-26

Biocatalysts for conversion of thermochemical waste streams: 18-36