DOE Bioenergy Technologies Office (BETO) 2017 Project Peer Review

Upgrading Biorefinery Waste for Bioplastics

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Biochemical Conversion
Biorefinery Upgrading

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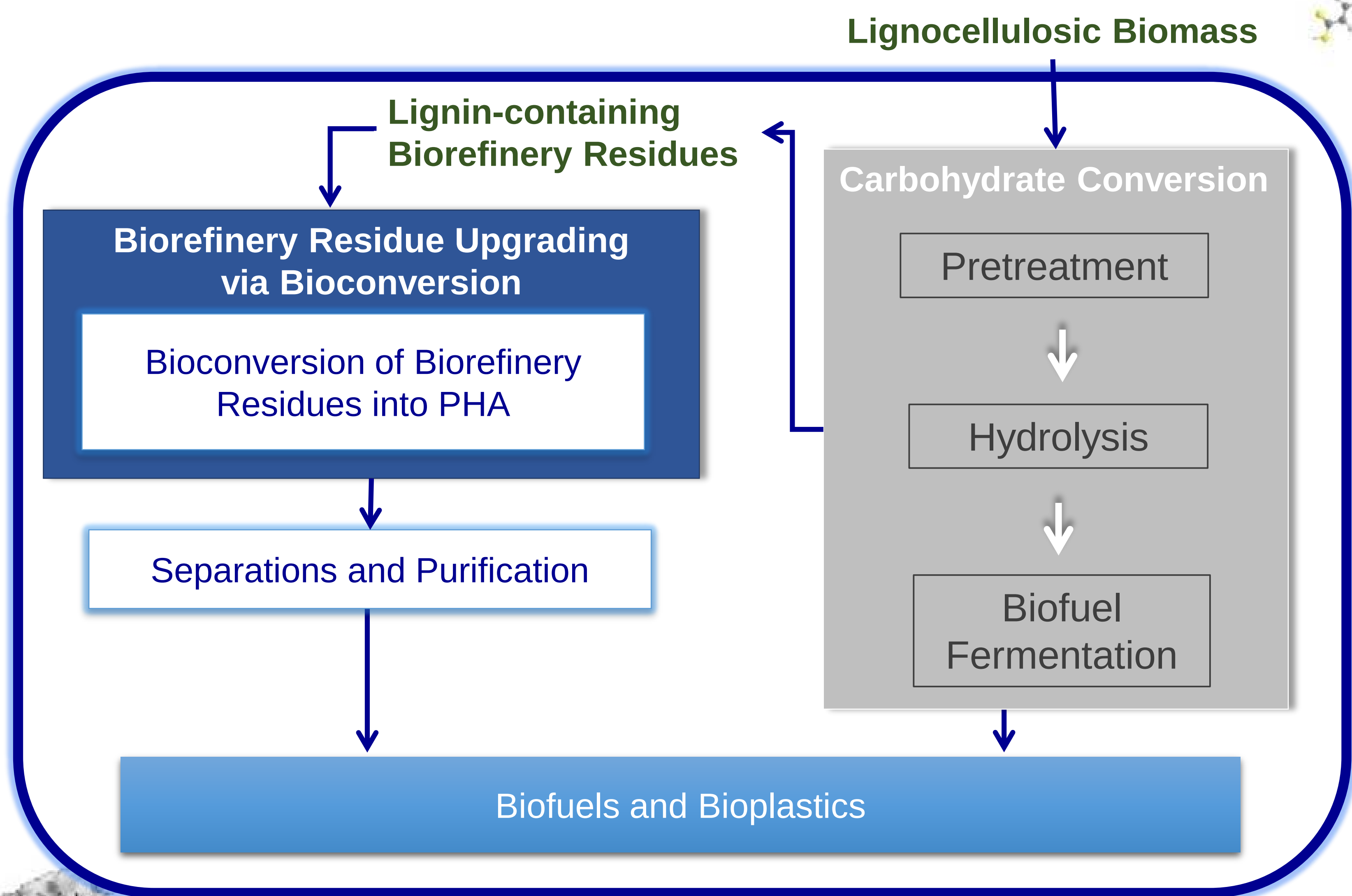
Project Goal: Upgrading Biorefinery Waste to Fungible Products

- Address one of the most challenging issues in biofuel production: upgrading the lignin-containing biorefinery residues to fungible bioproducts.
- Develop a viable bioprocess to convert biorefinery waste to bioplastics at less than $5 dollar/Kg. -- Project Outcome
- Overcome the key challenges for biorefinery cost-effectiveness and sustainability as laid out by BETO MYPP; Bring down the biofuel cost toward $3/GGE.

- BETO Missions:
  - Manage biorefinery waste
  - Reduce carbon emission by complete biomass usage
  - Improve biorefinery economics and sustainability

Project Goal: Upgrading Biorefinery Waste for Fungible Products



Quad Chart Overview

Timeline

- Project start date: 07/01/2016
- Project end date: 06/30/2019
- Percent complete: 10%

Barriers

- Barriers addressed
  - Ct-G. Efficient Intermediate Cleanup
  - Ct-J. Process Integration
  - Ct-L. Aqueous Phase Utilization

Budget

	FY 16 Costs	Total Planned Funding (FY 17-End)
DOE Funded	$42,367	$2,457,626
Project Cost Share (Comp.)*	$0.00	$785,071

Partners

- Partners
  - University of Tennessee, Knoxville/ Oak Ridge National Laboratory
  - Washington State University
  - ICM inc.
- Commercial Partners and Relevance
  - **ICM inc. to scale up the technology in 50 Liter reaction and achieve the on-site integration with biorefinery.**

Project Overview

FOA Topic: Process development and optimization of a single unit operation for the upgrading of chemically or biologically derived intermediates to fuels and products.

Project Title:

Upgrading lignin-containing biorefinery waste to bioplastics

Objectives:

This project uniquely addresses BETO's mission and FOA's goals through process enablement, development, and optimization for the bioconversion of lignin-containing biorefinery residues into bioplastics.

(1) Process enablement by screening and engineering microorganisms to convert biorefinery waste streams to PHA for bioplastics;

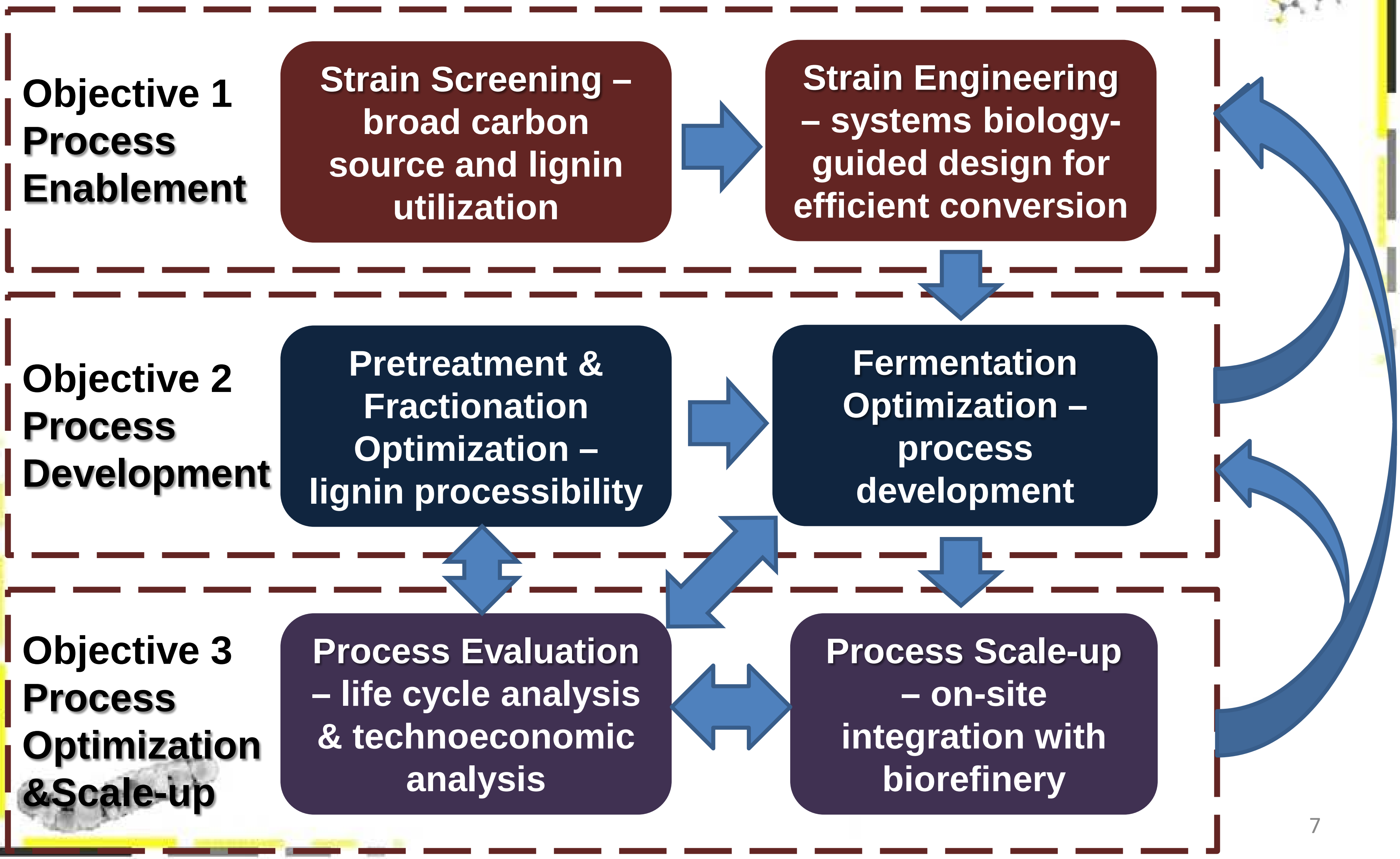
(2) Process development by characterizing biorefinery residues, optimizing pretreatment and lignin fractionation, enhancing fermentation, and designing the novel bioprocess;

(3) Process integration and optimization by biorefinery on-site scale-up, technoeconomic and life cycle analysis for the lignin-to-PHA up-grading process.

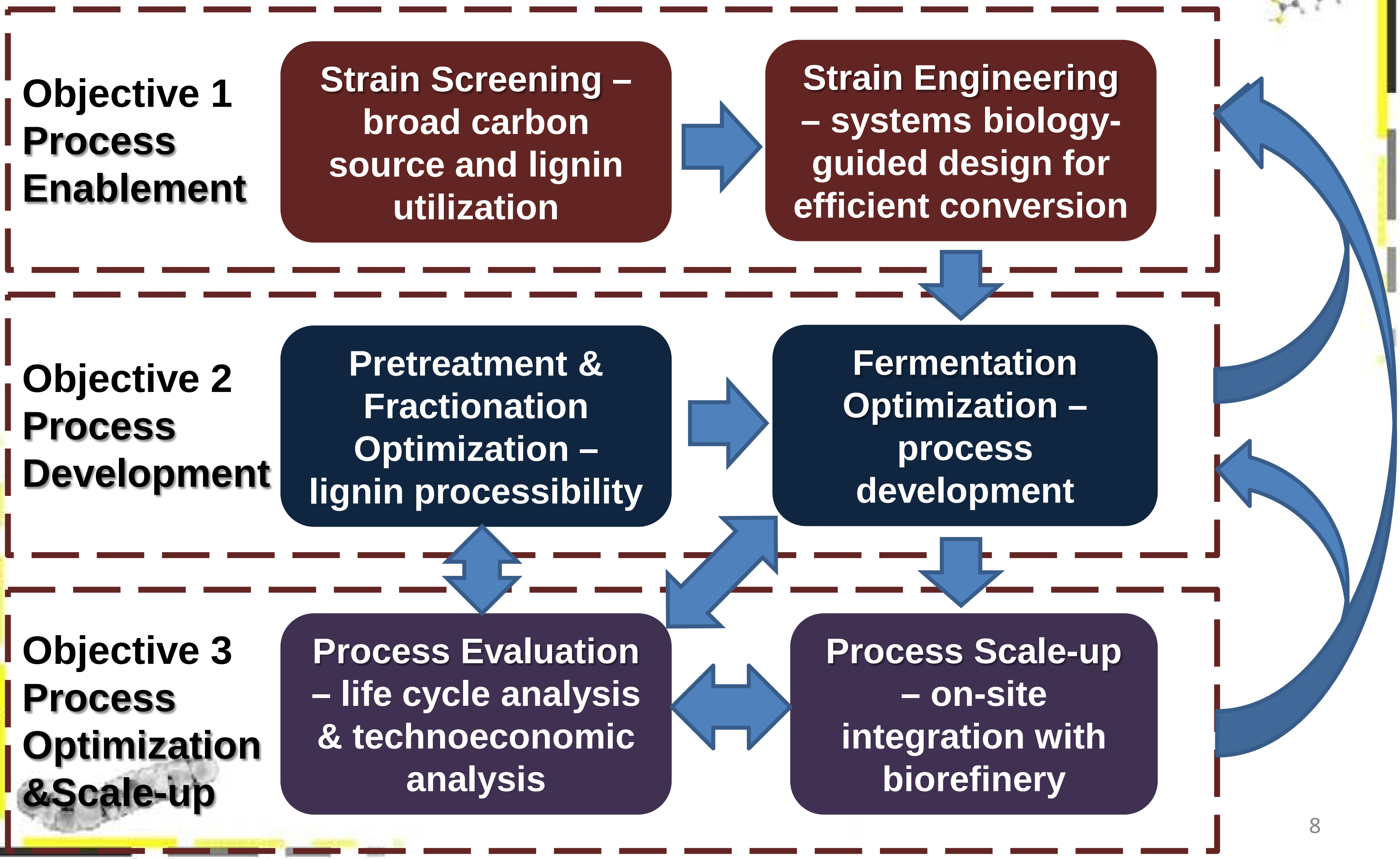
Management Approach

- Defined and measurable milestones were laid out for technology development and commercialization.
- Go/No-Go milestones were set at the end of each year and each of the two budget periods. BP1 ends at 24 months.
- Monthly group teleconferences and semi-annual group meetings were implemented to evaluate the progresses against milestones.
- Regular teleconferences between the PI and the program management are implemented to evaluate progresses, mitigate risks, and address management issues.
- Engage industrial partners including ICM inc. for deliverables relevant to EERE MYPP.
- Integrate TEA and LCA throughout the project to ensure the relevance of the project outcome.

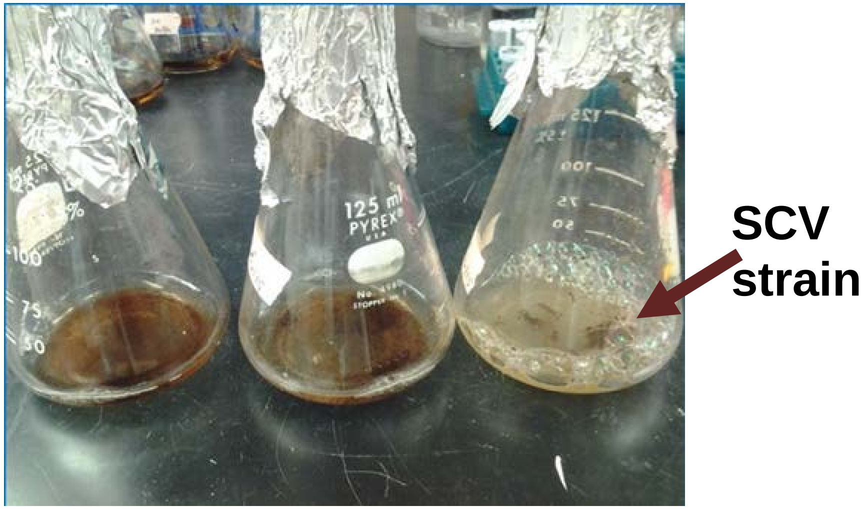
Technical Approach
Target: Titer 8.4g/L, Efficiency 30%

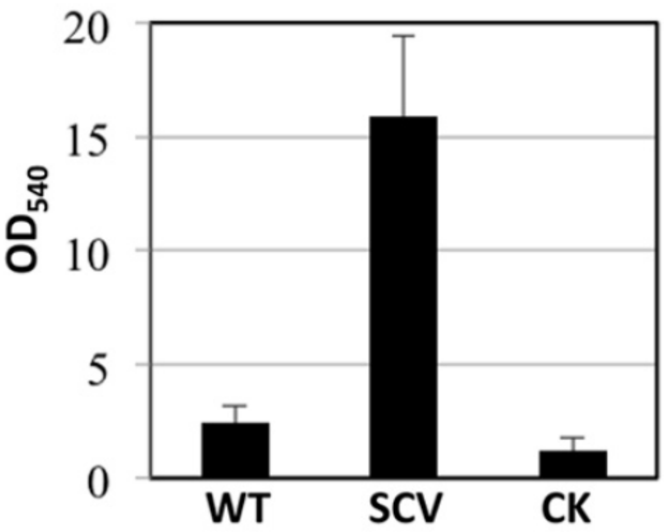


Technical Approach & Accomplishment



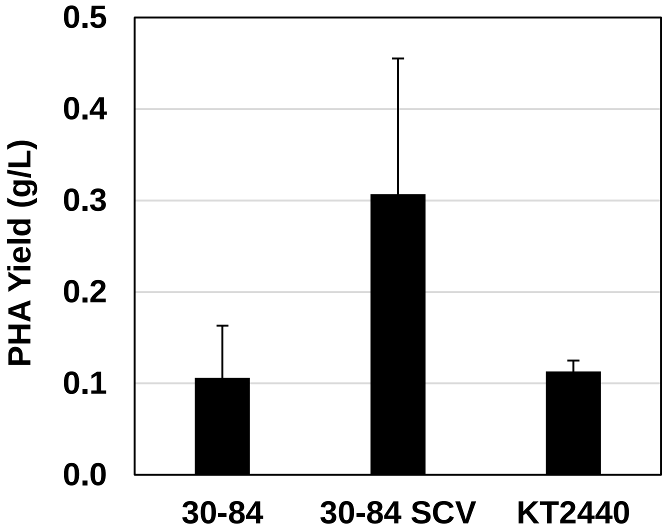
Strain Screening – scv Strain for High Productivity on Biorefinery Waste



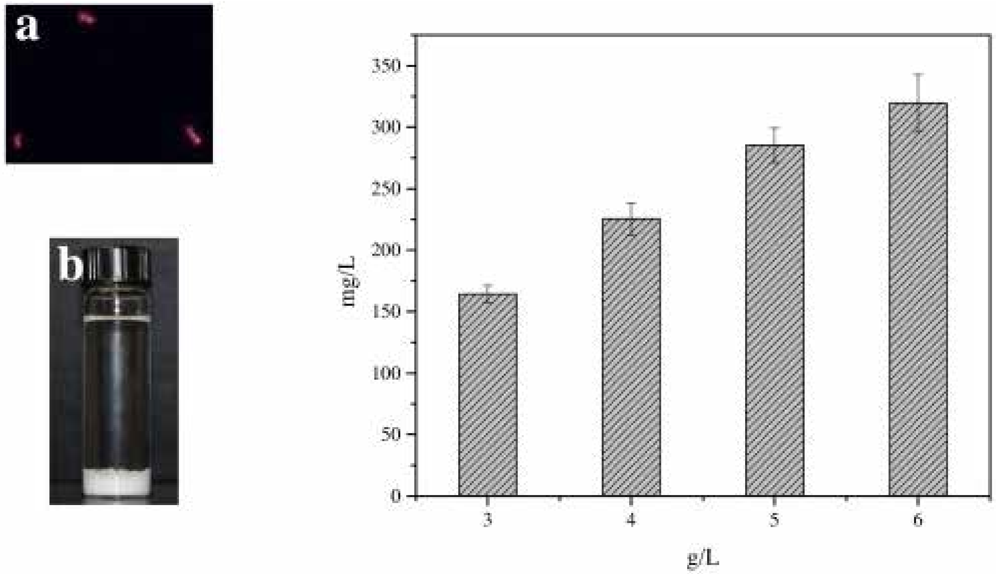


Strain screening has identified a Small Colony Variant (SCV) *pseudomonas* strain:

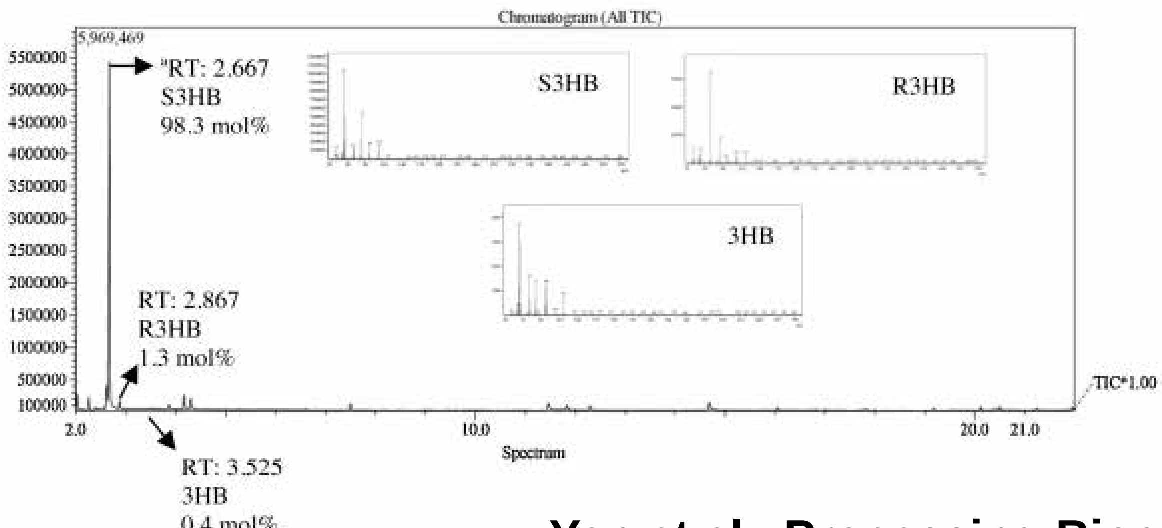
- Aggregates on lignin-containing substrates
- Grows to a higher concentration on biorefinery waste
- Produces much more PHA on biorefinery waste



Strain Screening – *Cupriavidus basilensis* B-8 as an Effective Strain for Lignin Conversion

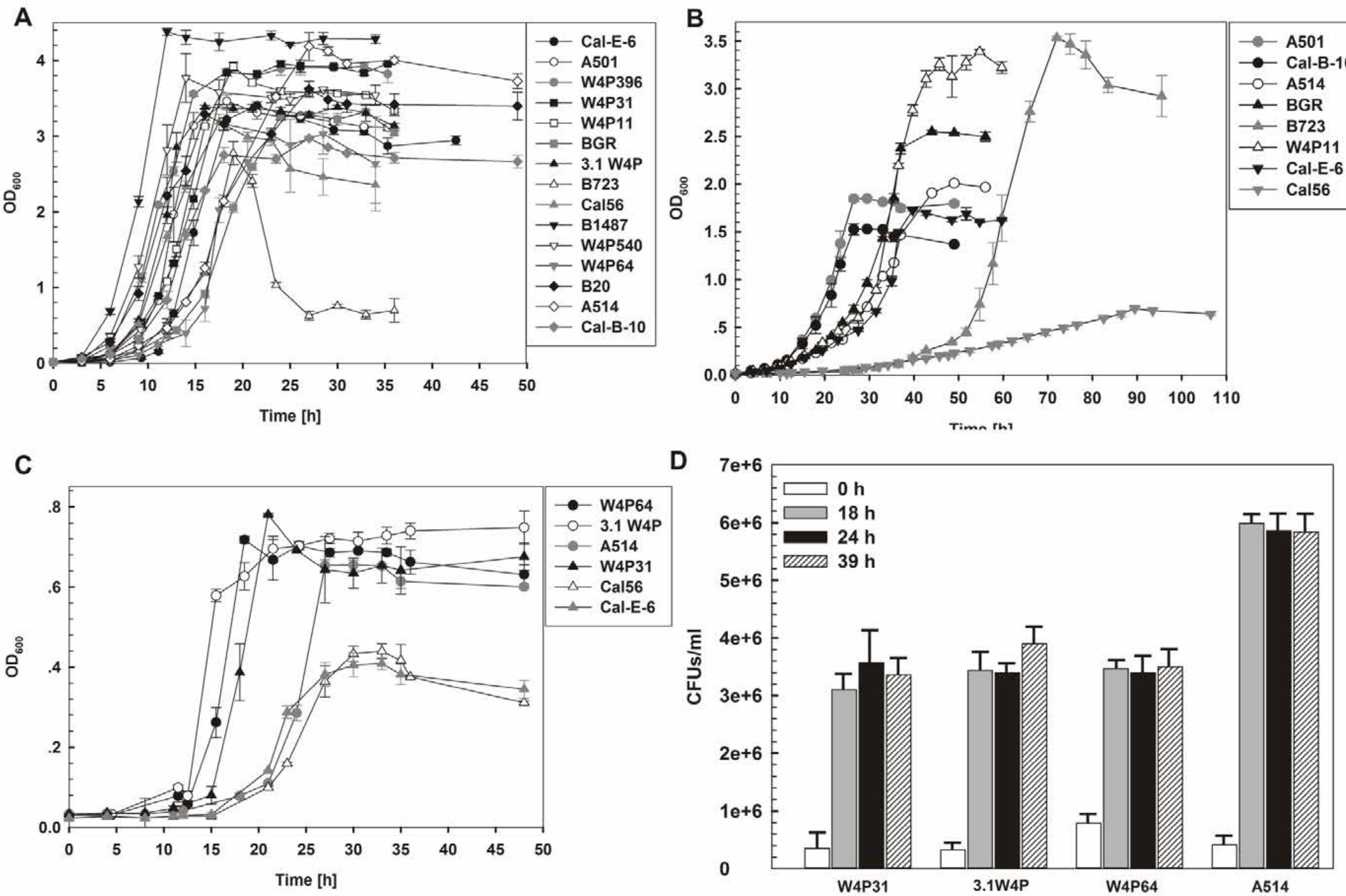


Cupriavidus basilensis **B-8 can convert lignin to bioplastics at over 300mg/L.**



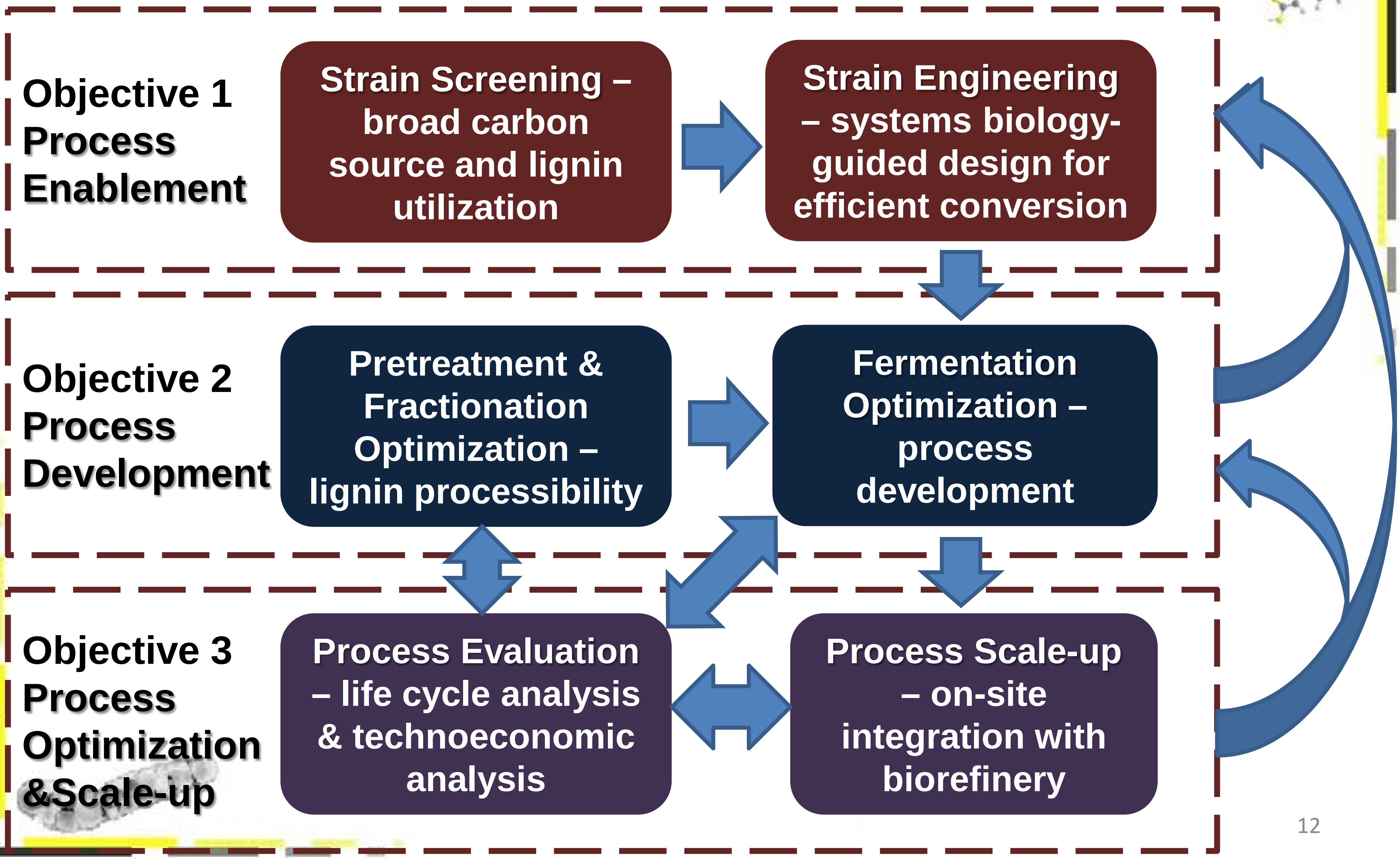
Yan et al., Processing Biochemistry 2017, 52, 238–242

Discovering a Lignin-Utilization *P. putida* Strain

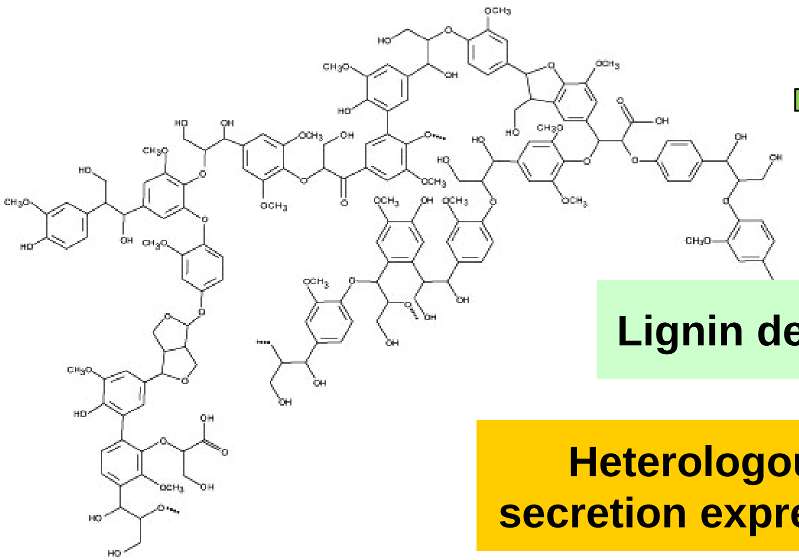


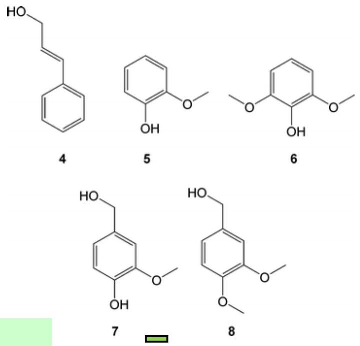
Lin, et al. Green Chemistry 18 (20), 5536-5547

Technical Approach& Accomplishment



Technical Approach – Strain Engineering

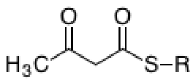




Lignin depolymerization

Aromatic compound utilizition capacity

Heterologous secretion expression



Systems biology-Guided Design

Carbon flux redirection

PHA production

Critical Factors for Success:

(1) Titer: >8.4g/L PHA

(2) Reaction Time: <72 hour

(3) Feedstock Load: >30g/L

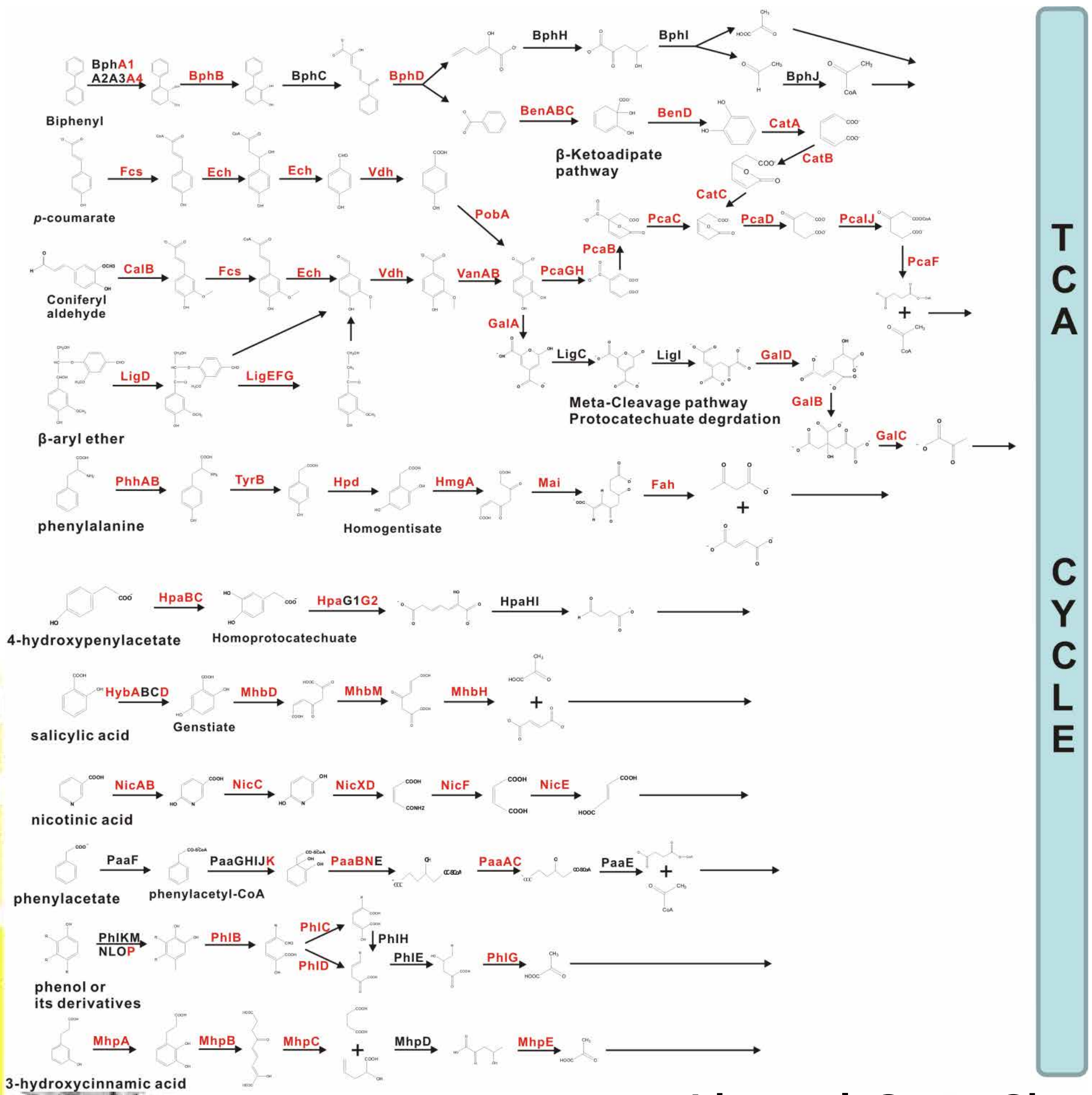
Major Challenges:

(1) Lignin Processibility

(2) Lignin Depolymerization

(3) Carbon Flux Optimization for Higher Targeted Products

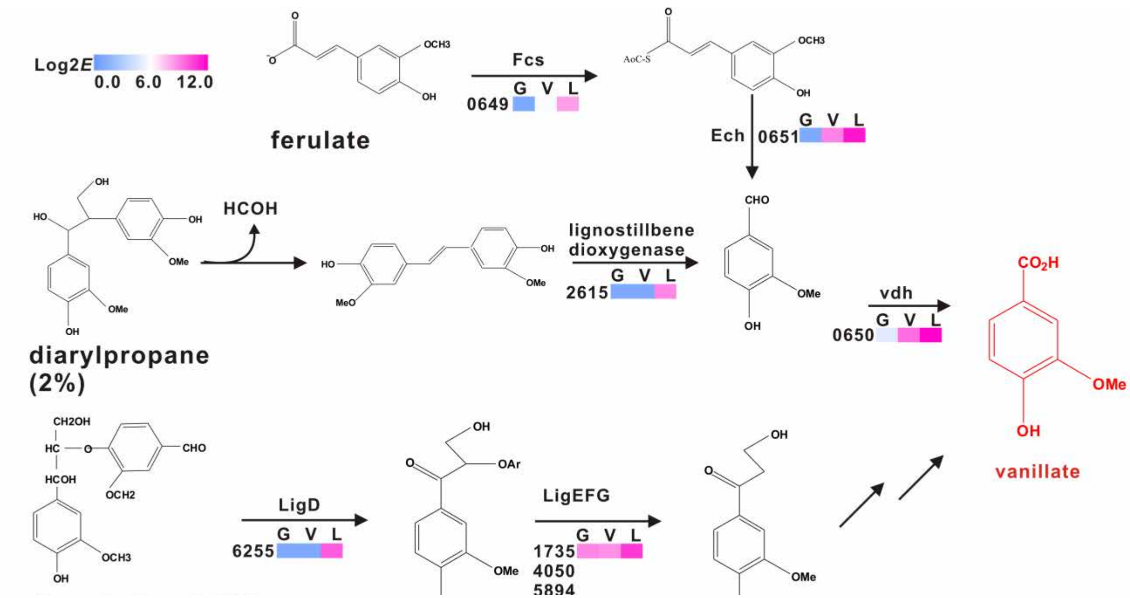
Lin, et al. Green Chemistry 18 (20), 5536-5547

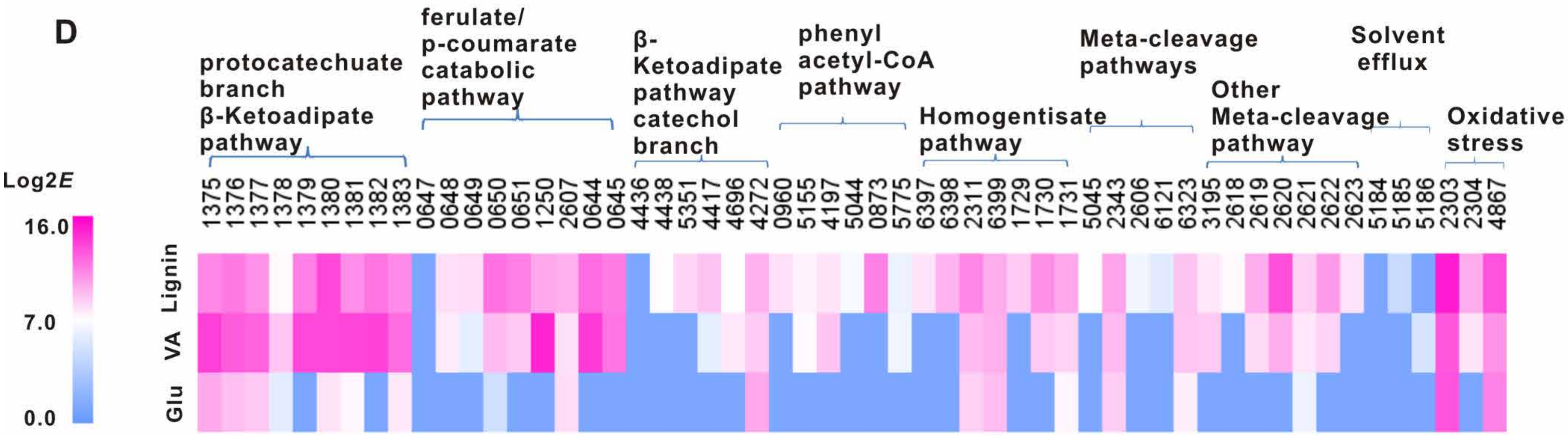


Broad Aromatic Compound Degradation Pathway as Revealed by Genome Sequencing

Lin, et al. Green Chemistry 18 (20), 5536-5547

Overview of Proteomics-based Systems Biology Study





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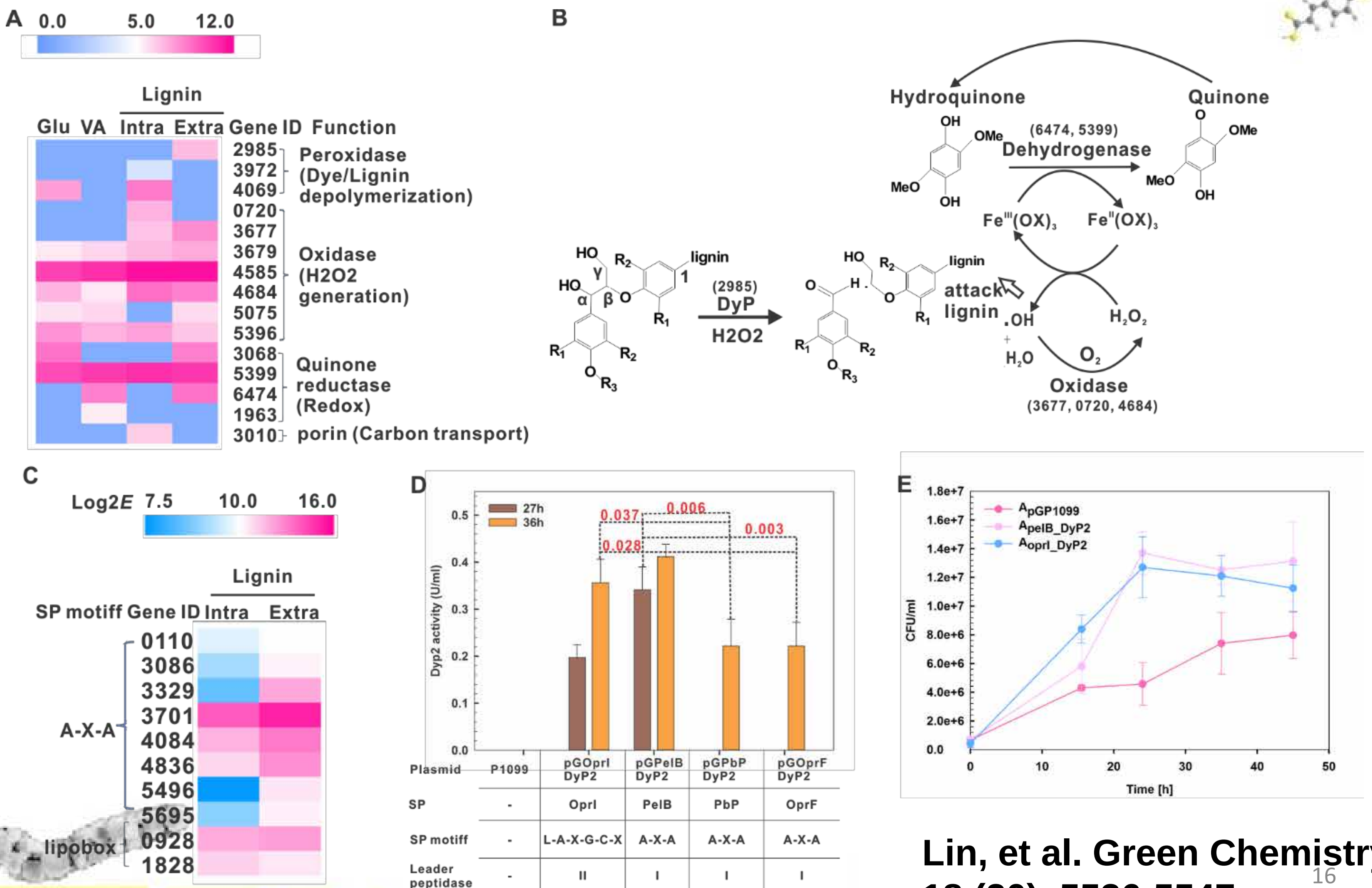
Design of Lignin Depolymerization Modules



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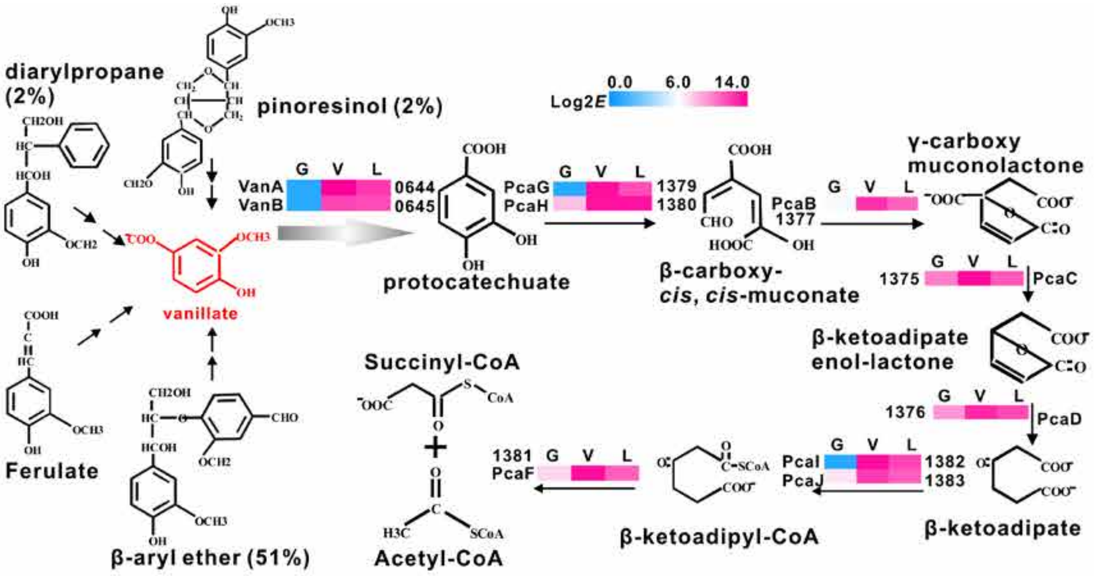
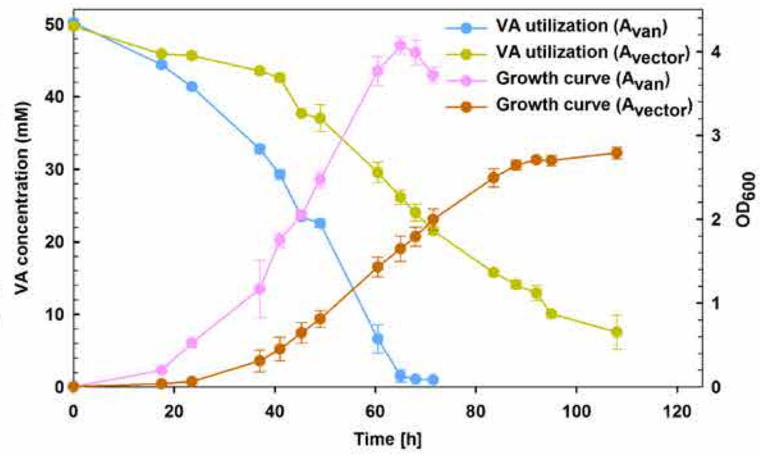
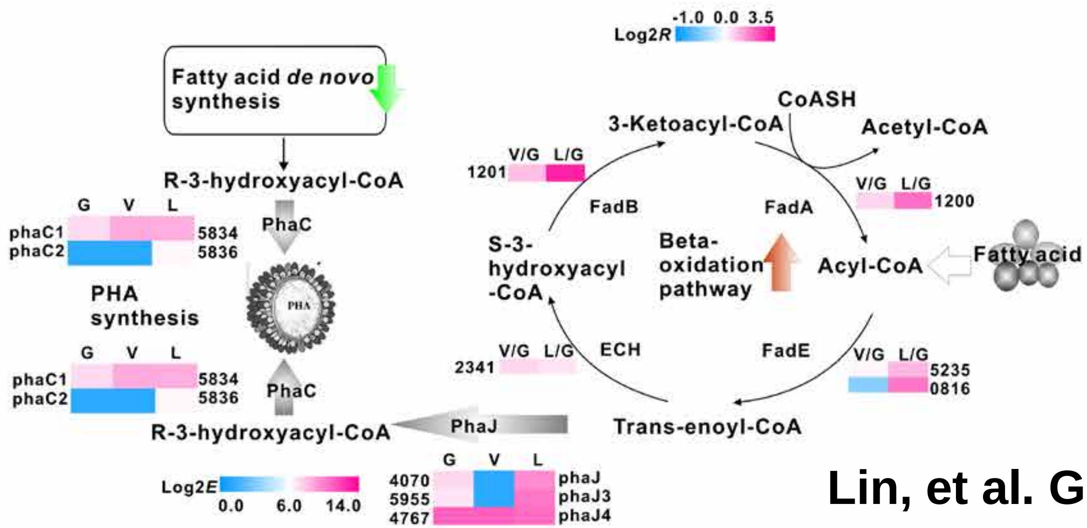
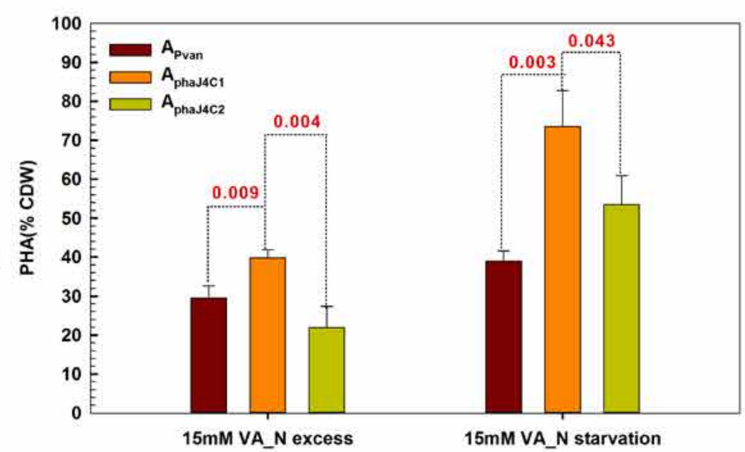
Design of Aromatic Compound Catabolism and PHA Biosythesis Modules to Maximize Carbon Flux









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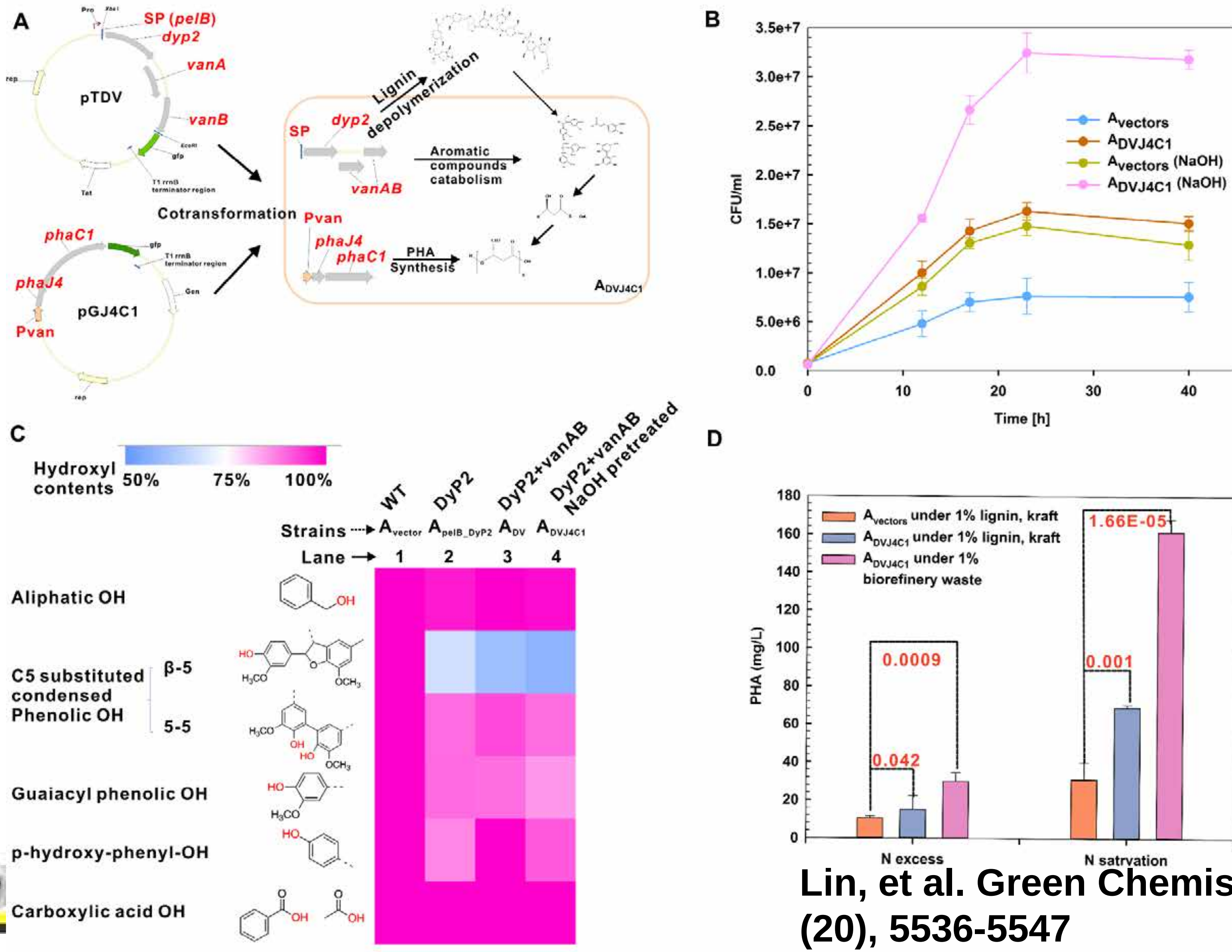
Multiple Module Integration for Biorefinery Residue Upgrading to PHA



Lin, et al. Green Chemistry 18 (20), 5536-5547

Strain Screening and Engineering for Biorefinery Residue Upgrading

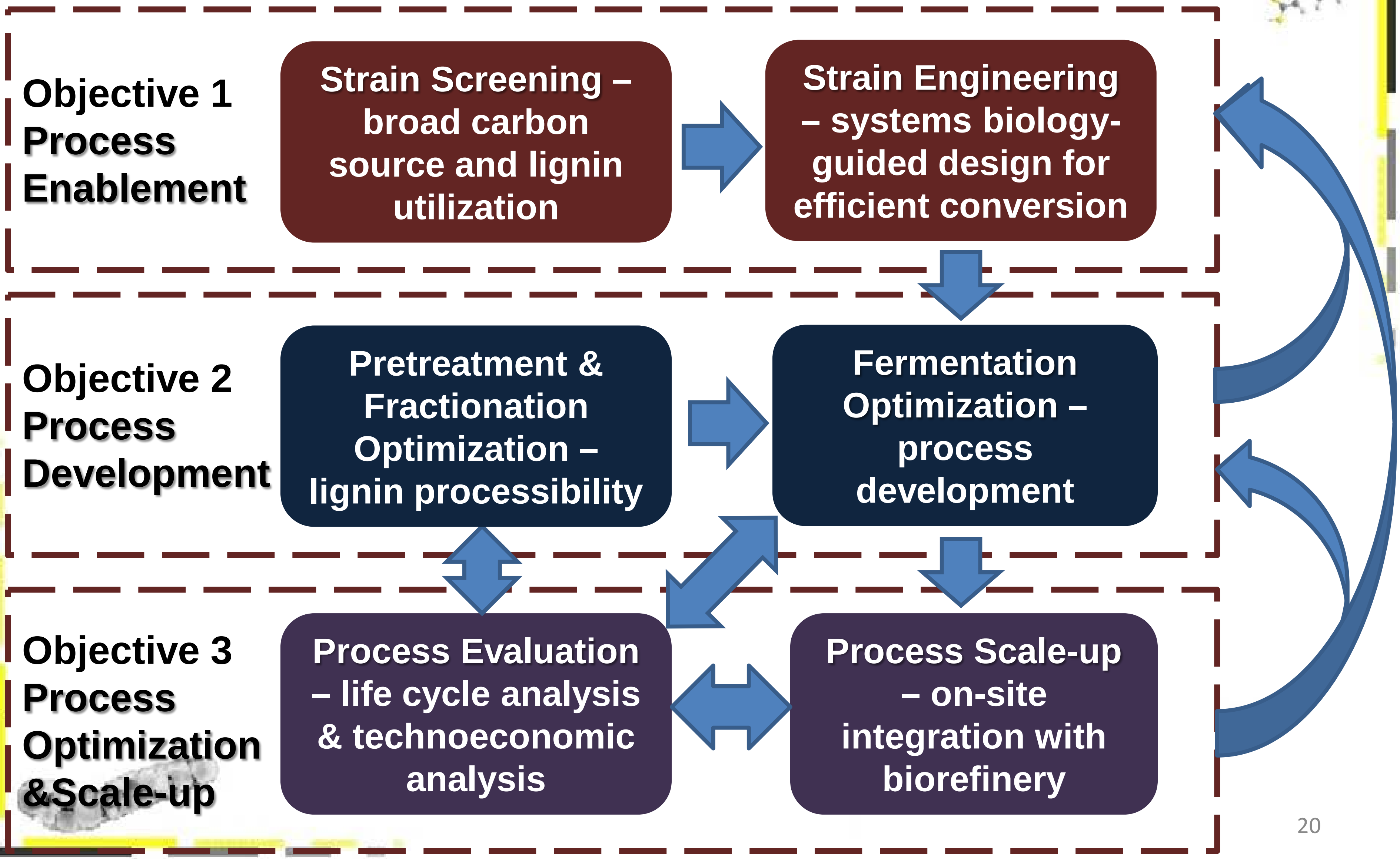
1. *Pseudomonas putida* strain with strong aromatic compound and lignin degradation capacity has been identified.

2. Comparative genomics is an effective approach to reveal lignin and aromatic compound degradation mechanisms – coordinative pathways.

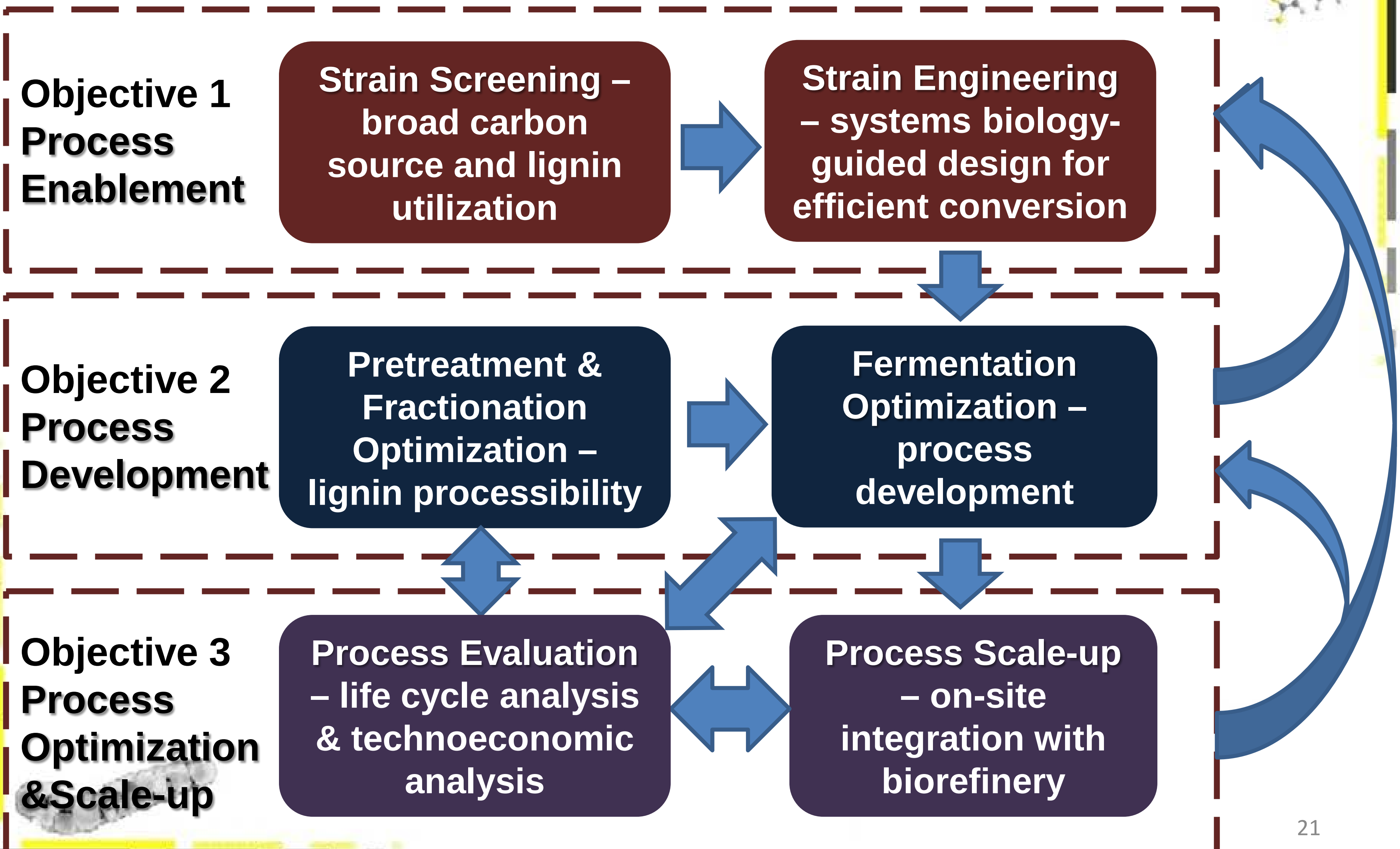
3. Systems biology-guided strain engineering is effective in guiding the design of three functional modules to enhance the upgrading of biorefinery waste to PHA.

4. The strain screening and engineering can be integrated to enable the process for upgrading biorefinery waste to PHA.

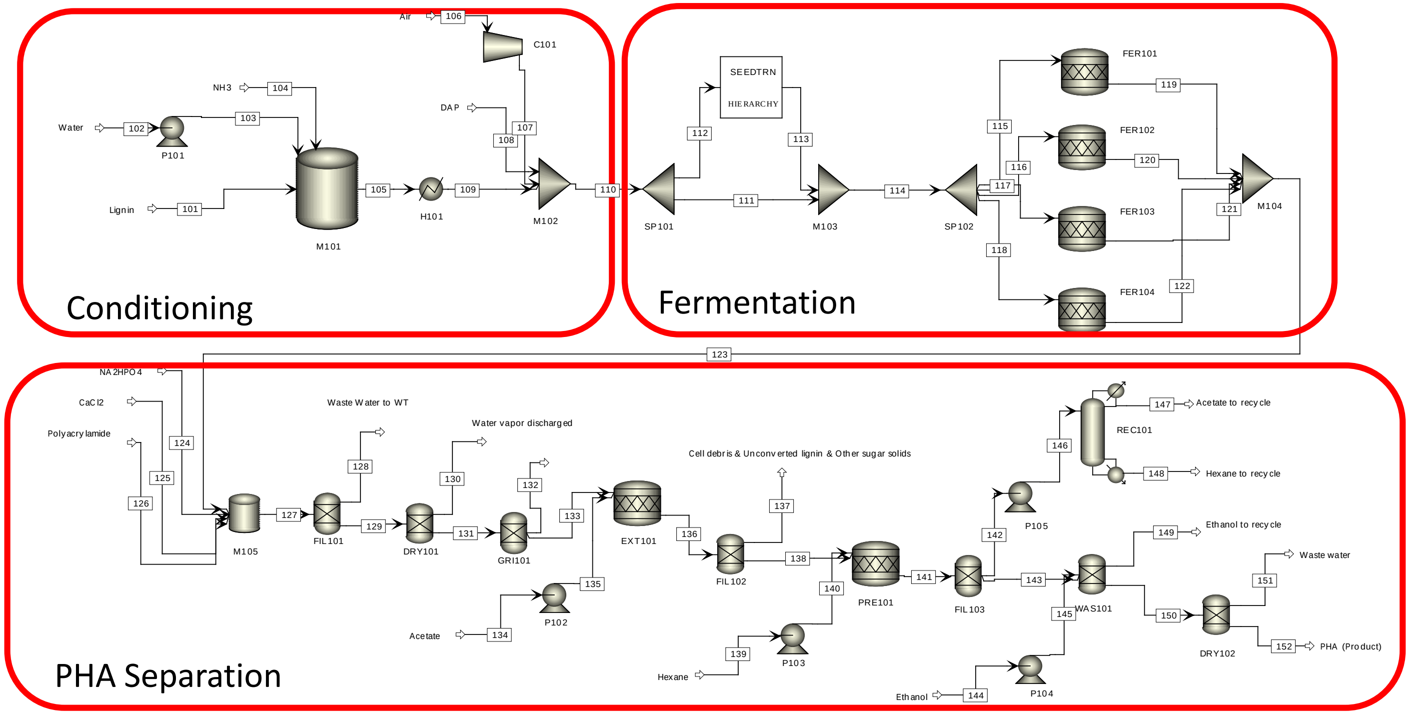
Technical Approach



Technical Approach Toward Future Work



Aspen Plus Model to Evaluate the Route for Biorefinery Residue to Bioplastics



Revenue Potentials for the Biorefinery Residue to PHA Route

	Lignin Utilization	PHA conversion	Impact on energy resource distribution	
			Available lignin for energy generation (kg/hr)	Potential revenue from energy generation (MM$/yr)
Case 1	10%	10%	12,103	18.57
Case 2	40%	30%	10,758	17.46
Case 3	60%	40%	9,291	16.26
Case 4	60%	60%	7,824	15.46

The NREL ethanol plant electricity cost is about 13.71 MM$/yr

Techno-Economic Analysis of Up-grading of Biorefinery Residues to PHA

	UNITS	CASE 1	CASE 2	CASE 3	CASE 4
Annual production	MMkg	0.5	5.96	11.9	17.84
Total Capital Cost	MM$	24.2	61.2	86.5	87.3
Total Operating Cost	MM$/yr	7.0	12.8	16.6	16.7
Raw Material	MM$/yr	1.1	4.2	6.3	6.3
Utilities	MM$/yr	0.12	0.25	0.33	0.35
Unit cost	$/kg	23.82	3.51	2.28	1.57
Rate of return	%	10	10	10	10
Minimum selling price	$/kg	28.49	4.36	2.84	2.06

Case1: 10% lignin flowrate to PHA process; 10% conversion of lignin to PHA
Case2: 40% lignin flowrate to PHA process; 30% conversion of lignin to PHA
Case3: 60% lignin flowrate to PHA process; 40% conversion of lignin to PHA
Case4: 60% lignin flowrate to PHA process; 60% conversion of lignin to PHA

Conclusion for Approach – Future Work

- Strain screening will be used to identify 1-2 most effective strain for converting biorefinery residues to bioplastics.
- Systems biology-guided strain engineering will deliver strain with better lignin depolymerization capacity and more carbon flux from lignin to PHA. The strain will have a higher conversion rate and more percentage PHA in cell dry weight.
- Pretreatment and fractionation optimization will be carried out to maximize both sugar and lignin bioproduct yield from biorefinery.
- Fermentation optimization will be carried out to improve titer and PHA conversion efficiency for the bioconversion process.
- TEA and LCA will be carried out to evaluate how the advances of technologies contribute to the cost-effectiveness and sustainability of biorefinery.
- We will work with commercial partner to scale up the process to 50L scale.

Future Work

Strain Screening

Strain Engineering

Pretreatment & Fractionation Optimization

Fermentation Optimization

We will integrate the outcome from strain screening and engineering with the optimization of pretreatment/fractionation and fermentation to deliver an efficient process for upgrading biorefinery waste to PHA. The process design and research endeavor will be evaluated and guided by TEA and LCA.

Process Integration for Biorefinery Residue Upgrading to PHA

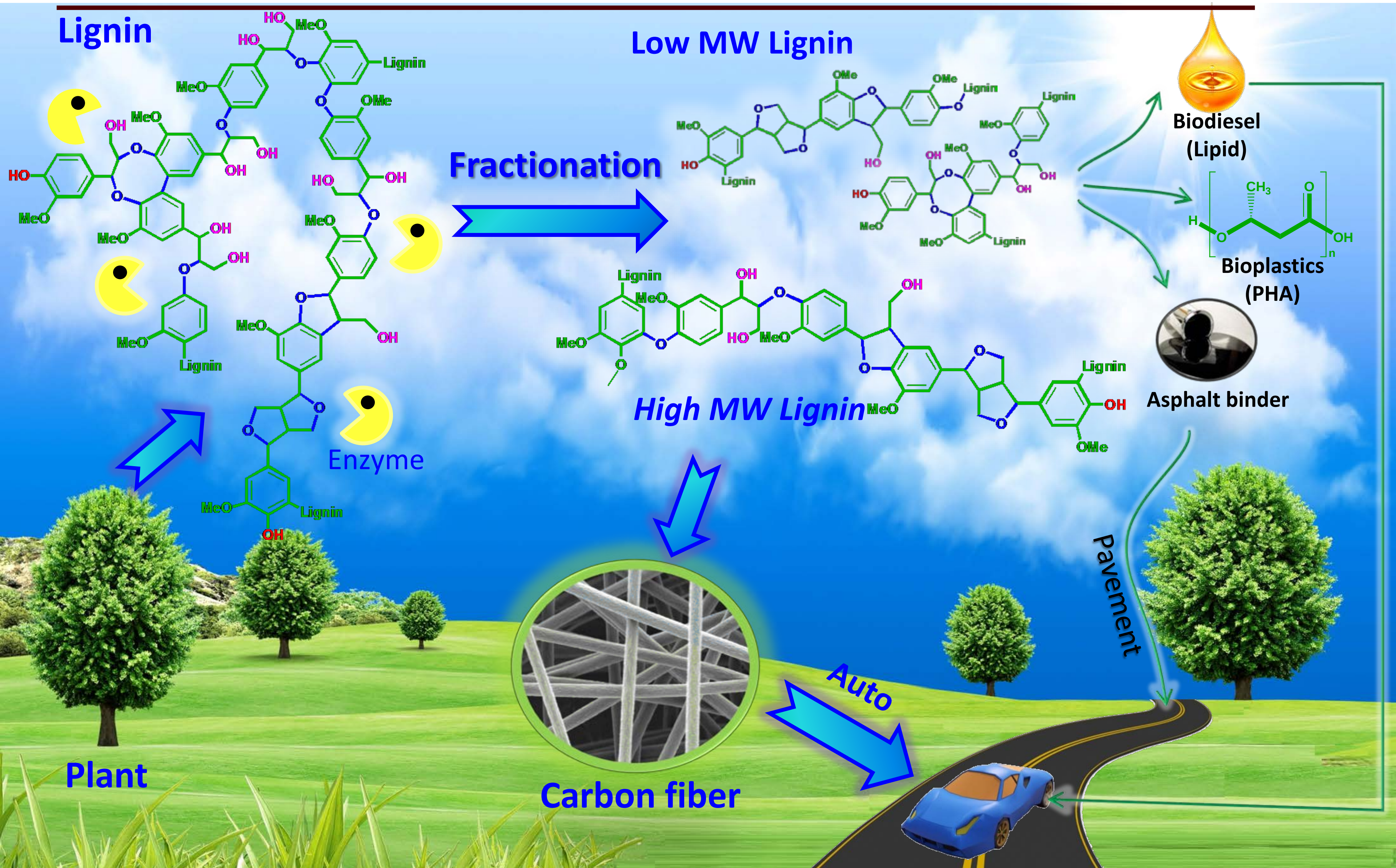
Process Evaluation – LCA and TEA

Process Scale-up for Biorefinery Integration

Relevance – Direct Relevance to MYPP and Challenges in Biorefinery

- The project will deliver a bioprocess to convert biorefinery residues to PHA at less than $5 per kg. The new biorefinery stream will add significant value to the lignocellulosic biofinery.
- The project will enable the multi-stream integrated biorefinery (MIBR) to maximize the yield of both carbohydrate-based biofuels and lignin-based bioproducts, which will turn biorefinery into biomanufacturing facility.
- With the multiple product stream and the maximized yield for both biofuels and bioproducts, the project well address the MYPP goal to achieve $3/GGE fuels. The project will improve the overall cost-effectiveness of biorefinery and reduce the fuel production cost.
- With more complete utilization of biorefinery residues, the project will address the mission of BETO, the MYPP goals, and the challenges in biofuel industry by improving biorefinery efficiency and sustainability.
- The research will significantly advance the current state-of-the-art in biorefinery residue upgrading. The technical breakthrough can be integrated with different platforms to produce valuable compounds from waste stream. Certain part of the technologies were licensed and we are working with commercial partner for scale up and commercialization.

Relevance – Enabled Multistream Integrated Biorefinery (MIBR)



Published Articles

1. Shangxian Xie; Qining Sun, Yunqiao Pu, Furong Lin, Su Sun, Xin Wang, Arthur J. Ragauskas, ***Joshua S. Yuan****, Advanced chemical design for efficient lignin bioconversion, *ACS Sustainable Chemistry & Engineering*, In press.
2. Yan Shi, Qiang Li, Xin Wang, Shangxian Xie, Liyuan Cai, ***Joshua S. Yuan****, Directed bioconversion of Kraft lignin to polyhydroxyalkanoate by *Cupriavidus basilensis* B-8 without any pretreatment, *Process Biochemistry*, In press.
3. Lu Lin, Yanbing Cheng, Yunqiao Pu, Su Sun, Xiao Li, Mingjie Jin, Elizabeth A. Pierson, Dennis Gross, Bruce E. Dale, Susie Y. Dai, Arthur J. Ragauskas, ***Joshua S. Yuan****, Systems biology-guided biodesign of consolidated lignin conversion, *Green Chemistry*, 2016, 18, 5536-5547.
4. Qiang Li, Shangxian Xie, Wilson K. Serem, Mandar T. Naik, Li Liu, Joshua S. Yuan*, Quality Carbon Fiber from Fractionated Lignin, Green Chemistry, In Press.

A PCT patent filing has been carried out.

Acknowledgement

Project Management:

Jay Fitzgerald Joshua Messner

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Dr. Jeremy Javers Dr. Susie Y. Dai

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