IOWA STATE UNIVERSITY

University Honors Program

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Sustainability Review of Modified *Escherichia coli* Lignocellulose Biomass Component for Biofuel Production

INTRODUCTION

- Biofuels are used as a more sustainable alternative to gasoline with renewable feedstock.
- Lignocellulose biomass sources are commonly used to create biofuels through a fermentation step.
- *Escherichia coli* is a common additive in the fermentation step since it can utilize both pentose and hexose sugars.
- Bacteriophage infections (*Figure 1*) are common in this process and lead to loss of product. Thermal tolerance also poses high process costs.
- Genetic modification can overcome these issues and more.

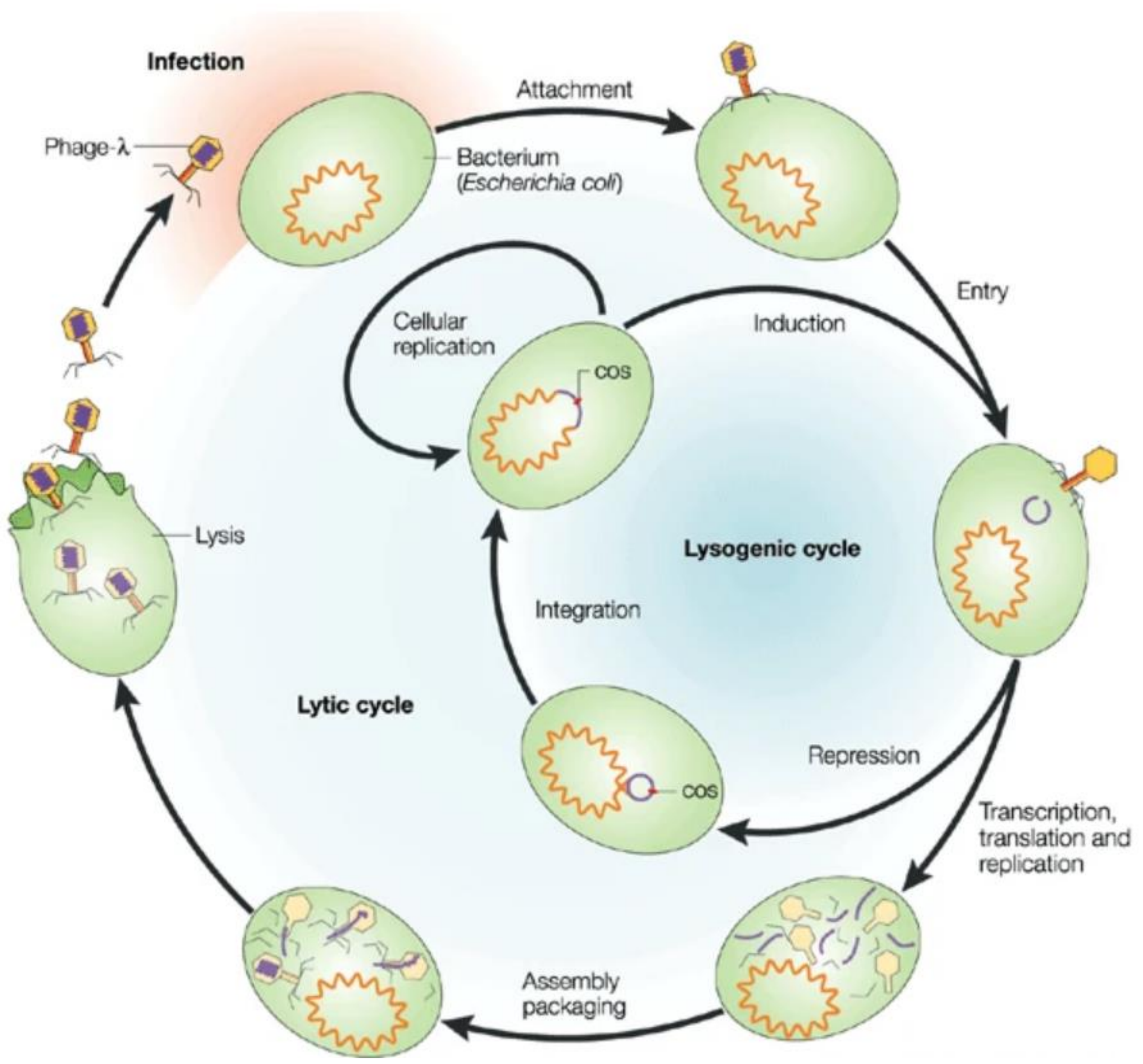

Figure 1: Schematic of lytic infection of a bacteria by bacteriophage [1].

MODIFIED *E. coli* STRAINS

- CRISPR/Cas9 is a genetic modification pathway that can be used to speed up modifications. It is more efficient than alternative modification methods.
- Proteins that spread lysate or enzymes that control thermal tolerance can be modified to produce the desired effect on the strain.
- CRISPR/Cas9 cuts DNA for gene knockouts using RNA guides to target base pairs.
- Biocontainment, non-pathogenic strains, standard lab safety practices reduce environmental and human risk of working with modified bacteria strains.

RESOURCES

- Lignocellulose (*Figure 2*) biomass sources come from woody waste of agricultural products. 40 million tons of byproduct produced per year.
- Hemicellulose component is hydrolyzed into hexose and pentose sugars. Enzymes and acids are needed for cell wall degradation and hydrolyzation.
- *E. coli* is abundant and used in processes. Its ability to utilize hexose and pentose sugars make it more robust than alternatives.

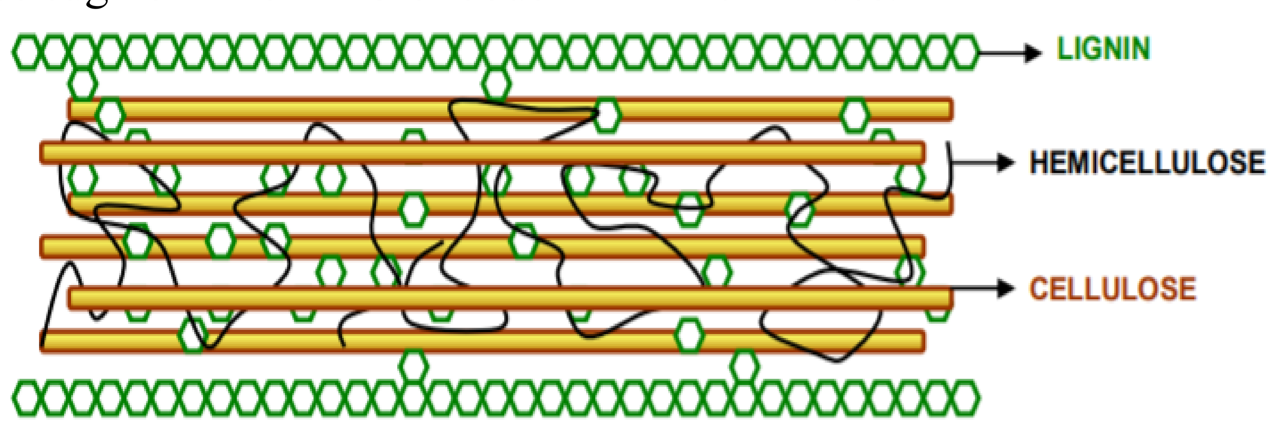

Figure 2: Depiction of the structure of lignocellulose components [2].

PROCESSING

- The fermentation of sugars (glucose) to biofuel (ethanol) pathway in *E. coli* (*Figure 3*) requires energy input.
- A separation process is needed to purify the biofuel product.
- 10-20 kW/ m^3 of heat is generated through the fermentation process with a power input of 2-5 kW/m^3. Operating temperatures within *E. coli* growth range of 30-37°C
- High operating costs from cooling (exothermic fermentation), agitation (constant mixing), aeration (1.33 kg O_2/kg biomass), and single-use supplies (filters, resins, testing materials).

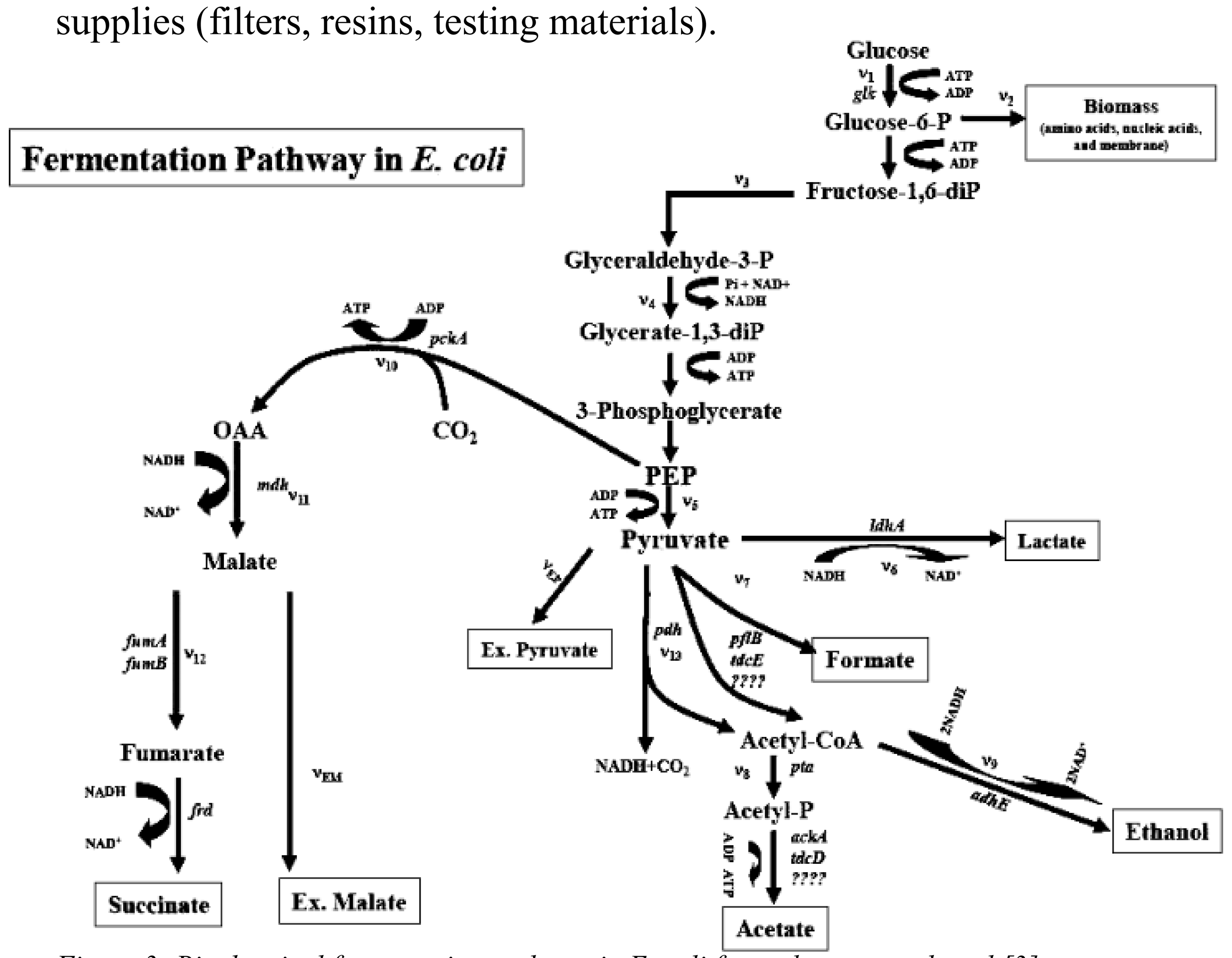

Figure 3: Biochemical fermentation pathway in E. coli from glucose to ethanol [3].

ALTERNATIVES

- Other common biomass sources include agricultural, municipal, and industrial wastes.
- Energy density (*Figure 4*) for 26 biomass sources show lignocellulose (willow, logan, sawdust, bamboo) mid-level energy density.
- Non-woody agricultural waste has lowest energy density and can take resources from other processes, specifically livestock feed.
- Industrial wastes (coals) have high energy density, but production processes pose environmental risks and have high ignition temperatures.
- Municipal wastes have high energy density and are otherwise unusable byproducts.

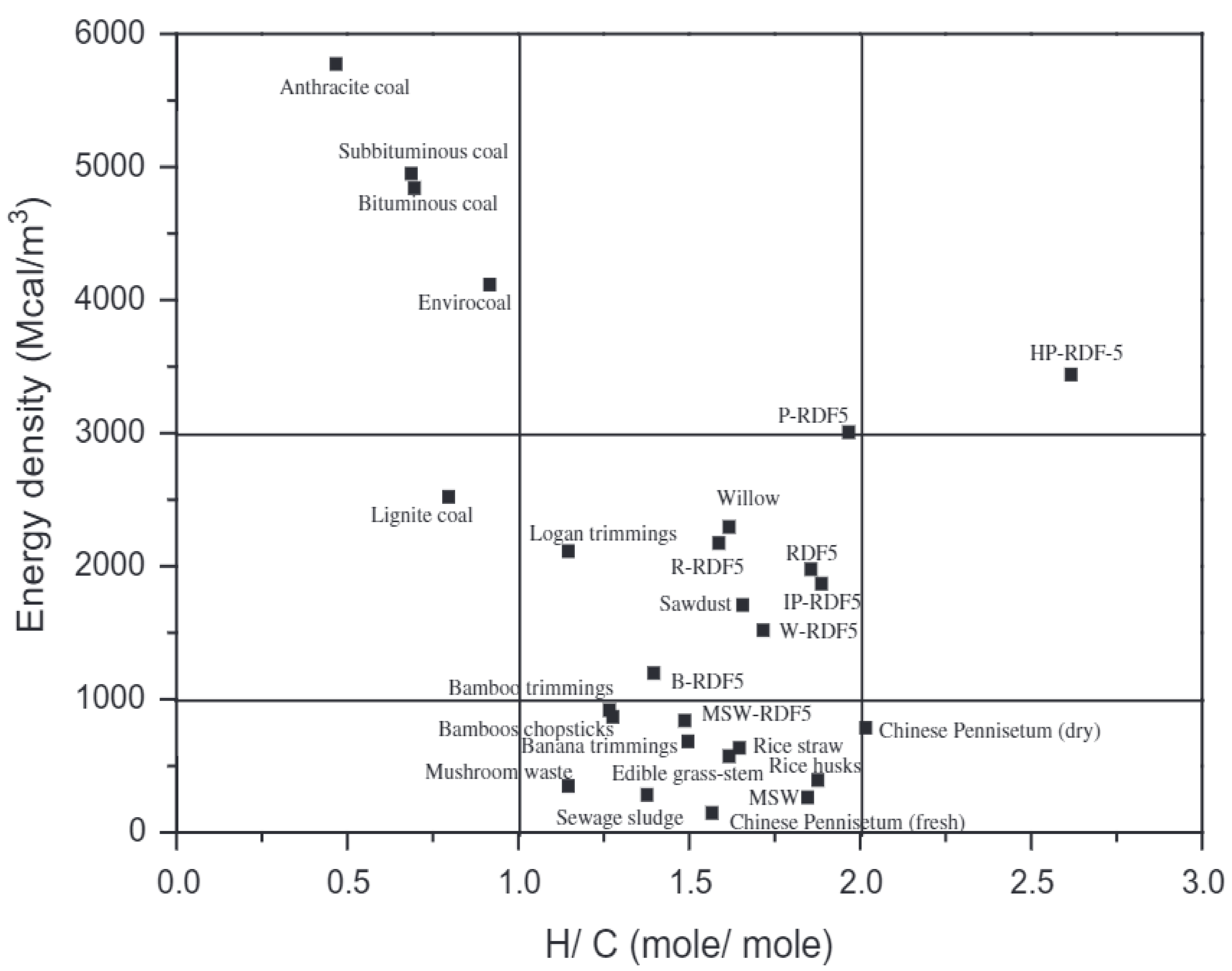

Figure 4: Energy density (Mcal/m^3) for 26 biomass sources [4].

ON POSTER REFERENCES

[1] B. Orzechowska, M. Mohammed, "The War Between Bacteria and Bacteriophages," *Intech*, vol. 32, pp. 137–144, 2019,

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[4] K. Y. Chiang, K. L. Chien, and C. H. Lu, "Characterization and comparison of biomass produced from various sources: Suggestions for selection of pretreatment technologies in biomass-to-energy," *Appl. Energy*, vol. 100, pp. 164–171, 2012.