



ENGINEERING *ESCHERICHIA COLI* FOR BIOFUEL PRODUCTION

Metabolic Flux Analysis of the Propanogenic *E. coli*

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Problem Statement

After reading the case study “Engineering *Escherichia coli* for Biofuel Production” [1], use Metabolic Flux Analysis (MFA) to analyze the metabolic network by answering the following questions:

1. Draw a simplified network by keeping the bare essentials (use principles of MFA).
2. Calculate the maximum theoretical yield of 1-propanol from glycerol considering that ATP and NADH are only produced via the description in the map. How does this change your maximum theoretical yield? Why?
3. Using the simplified map, go through the following MFA process:
 - i. Write down the reactions.
 - ii. Do balances on all components in the simplified map (except for ATP).
 - iii. Identify the intracellular compounds.
 - iv. Apply pseudo-steady state.
 - v. Look at measured compounds (Table 1) and decide if you can solve for all rates.

Table 1 – Culture performance of batch cultivation in a bioreactor for CPC-PROH3 using glycerol as the major carbon source

	Glycerol	Dry Cell Weight (Biomass)	Succinate	Acetate	Propionate	Ethanol	1-Propanol
Initial Concentration (g/L)	30.73	2.638	0.318	0.259	0.370	0.363	0
Final Concentration (g/L)	0	5.474	0.906	4.297	1.152	9.897	2.438

References

- [1] Kajan Srirangan, Lamees Akawi, Lyndia Stacey, Cheryl Newton, Perry Chou and Marc Aucoin, Module 01. “Engineering *Escherichia coli* for Biofuel Production”. Waterloo Cases in Design Engineering (WCDE), University of Waterloo.