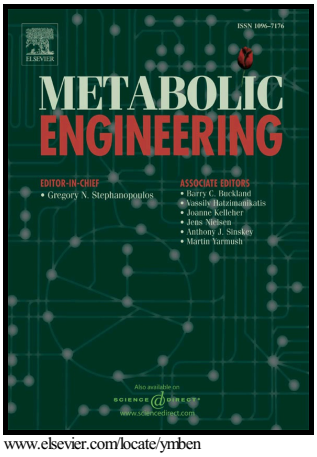

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PII: S1096-7176(18)30053-3
DOI: https://doi.org/10.1016/j.ymben.2018.04.018
Reference: YMBEN1393

To appear in: *Metabolic Engineering*

Received date: 1 February 2018
Revised date: 4 April 2018
Accepted date: 23 April 2018

Cite this article as: I-Ting Tseng, Yi-Ling Chen, Ching-Hsun Chen, Zhi-Xuan Shen, Cheng-Han Yang and Si-Yu Li, Exceeding the theoretical fermentation yield in mixotrophic Rubisco-based engineered *Escherichia coli*, *Metabolic Engineering,* https://doi.org/10.1016/j.ymben.2018.04.018

Exceeding the theoretical fermentation yield in mixotrophic Rubisco-based engineered *Escherichia coli*

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Abstract

Rubisco-based engineered *Escherichia coli* MZLFB (*E. coli* BL21(DE3) Δ*zwf*, Δ*ldh*, Δ*frd*) containing heterologous phosphoribulokinase (Prk) and Ribulose-1,5- bisphosphate carboxylase/oxygenase (Rubisco) was constructed for the mixotrophic growth. However, *in situ* CO_2 recycling was hindered by clogs of pyruvate during glucose metabolism, which consequently resulted in an insufficient regeneration of NAD^+ through the *pflB*-mediated ethanol production. Recombinant plasmid pLOI295 (encodes pyruvate