Title: Multifaceted Adaptive Laboratory Evolution Approaches for Improving Host Phenotypes

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Project Goals: Establish the scientific knowledge and new technologies to transform the maximum amount of carbon available in bioenergy crops into biofuels and bioproducts.

Abstract Text:

Natural microorganisms are not optimized for industry-relevant conditions and therefore often show unsatisfactory performance. In this study, multifaceted Adaptive Laboratory Evolution (ALE) approaches were applied for *Pseudomonas putida* KT2440 to improve its phenotypes to achieve efficient biochemical production. Firstly, we constructed efficient xylose and galactose catabolism by continuously evolving engineered KT2440 stains in a constant or weaning-off condition. Secondly, we significantly improve tolerance against isoprenol by growing cells with gradually increased isoprenol concentrations. With these multiple ALE strategies successfully generated strains with improved phenotypes. Whole-genome sequencing, transcriptome sequencing, and reverse engineering unveiled mutational mechanisms, enabling the improved phenotypes. Collectively, ALE proved to be an effective method for strain optimization and knowledge generation which can be utilized to develop efficient microbial bioprocesses.

References/Publications

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2. Lim et al. (2020) Generation of Ionic Liquid Tolerant *Pseudomonas putida* KT2440 Strains via Adaptive Laboratory Evolution. *Green Chem*. 22(17), 5677–5690.
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Funding Statement: This work conducted by the Joint BioEnergy Institute was supported by the Office of Science, Office of Biological and Environmental Research, of the U.S. Department of Energy under Contract No. DE-AC02-05CH11231.