Harnessing bacterial metabolism for biochemical production from lignin streams

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Channeling waste- and biomass-derived resources through bacterial metabolism to produce target chemicals provides a critical step for valorizing complex feedstocks in circular bioeconomy systems. Soil bacteria, such as *Pseudomonas putida,* have broad metabolic capabilities that can be exploited to produce commodity chemicals, such as *cis,cis*-muconate and β-ketoadipate, which act as precursors for direct-replacement or performance-advantaged bioproducts. Taking advantage of feedstocks derived from plant biomass for sustainable bioproduction, *P. putida* can funnel aromatic compounds derived from lignin to atom-efficient products while sugars can be utilized for cellular growth and energy. However, to reach process-relevant titers, rates, and yields of bioproducts, optimization of bacterial metabolism is necessary.

The work presented here integrates systems and synthetic biology to elucidate carbon metabolism, energy and co-factor balances, and stress responses during biochemical production. Employing genetic engineering strategies, key bottlenecks in the muconate production pathway were alleviated, improving titers by 3.5-fold and initial production rates by up to 2-fold. Additional work assessing bacterial metabolism in fed-batch bioreactors identified growth-dependent stress responses and metabolic shifts during production of β-ketoadipate. Overall, this understanding of cellular metabolism was used to identify engineering targets and bioprocess improvements to mitigate metabolic bottlenecks and stressors, generating more robust bacterial platforms for improved biochemical production.