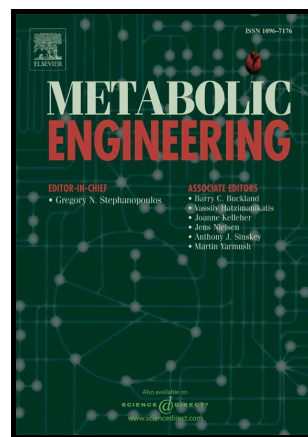


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Metabolic Engineering of *Pseudomonas taiwanensis* VLB120 with Minimal Genomic Modifications for High-Yield Phenol Production

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Abstract

Aromatic chemicals are important building blocks for the production of a multitude of everyday commodities. Currently, aromatics production relies almost exclusively on petrochemical processes. To achieve sustainability, alternative synthesis methods need to be developed. Here, we strived for an efficient production of phenol, a model aromatic compound of industrial relevance, from renewable carbon sources using the solvent-tolerant biocatalyst *Pseudomonas taiwanensis* VLB120. First, multiple catabolic routes for the degradation of aromatics and related compounds were inactivated, thereby obtaining the chassis strain *P. taiwanensis* VLB120Δ5 incapable of growing on 4-hydroxybenzoate (Δ*pobA*), tyrosine (Δ*hpd*), and quinate (Δ*quiC*, Δ*quiC1*, Δ*quiC2*). In this context, a novel gene contributing to the quinate catabolism was identified (*quiC2*). Second, we employed a combination of reverse- and forward engineering to increase metabolic flux towards the product, using leads obtained from the analysis of aromatics producing *Pseudomonas putida* strains previously generated by mutagenesis. Phenol production was enabled by the heterologous expression of a codon-optimized and chromosomally integrated tyrosine phenol-lyase encoding gene from *Pantoea agglomerans* AJ2985 (*PaTPL2*). The genomic modification of endogenous genes encoding TrpE^{P290S},

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