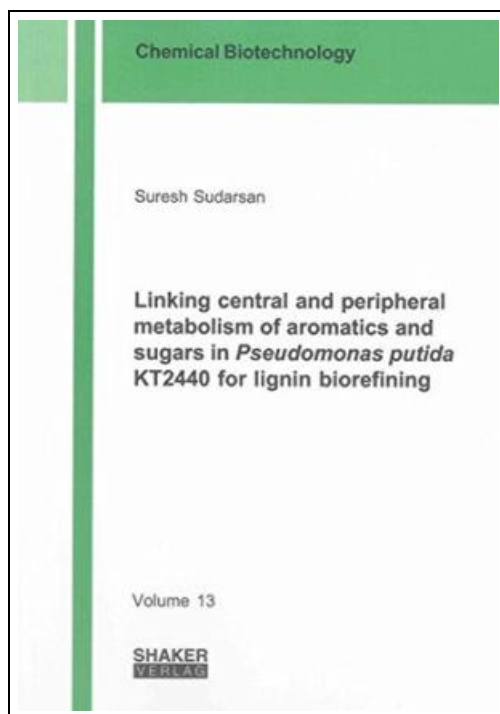


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LINKING CENTRAL AND PERIPHERAL METABOLISM OF AROMATICS AND SUGARS IN PSEUDOMONAS PUTIDA KT2440 FOR LIGNIN BIOREFINING



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Shaker Verlag Feb 2014, 2014. Buch. Condition: Neu. Neuware - One common pressing need in the field of metabolic engineering is to uncover the metabolic principles by which a microorganism operates. Metabolic principles serve as a scaffold to address the increasing awareness of metabolic complexity in various microbial processes. Dedicated attempts to develop biocatalysts for the use of lignin as a carbon source are challenging due to its heterogenic nature, but to a large extent on our lack of knowledge on how lignin monomer metabolism is linked to the fluxes of central metabolic pathways. This is especially true for aromatic compounds. Here in this study, we investigated and addressed the major limitations, especially the knowledge gaps engrossed in using a versatile microbe such as *Pseudomonas putida* towards lignin based biorefining. First, the functional organization and operation of bacterial networks as a 'bow-tie' was verified experimentally in the KT2440 strain of *Pseudomonas*, and was indispensable to identify the regulation hierarchies of carbon metabolism. Our findings invite reformulation of rational strain engineering approaches, with an ultimate target towards transcriptional modifications for using alternate carbon sources (including lignin monomers), and the needful metabolic modifications required to increase the rate of central metabolic reactions. Second, the metabolic and regulatory interactions of the aromatic metabolism was understood by in vivo kinetic analysis of benzoate metabolism via the - keto adipate (ortho-cleavage) pathway. The developed kinetic model was characterized with quantitative metabolic data from dynamic benzoate pulse experiments. Additionally, the steady state metabolome data was checked for thermodynamic consistency with network embedded thermodynamic (NET) analysis using the genome scale metabolic model iJN746 of *P. putida*. NET analysis unraveled those reactions of benzoate metabolism which are subjected to active genetic and allosteric regulation. Implications of which will be useful for extending the framework of mechanistic modeling of...

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