METABOLIC ENGINEERING OF *SACCHAROMYCES CEREVISIAE* FOR THE PRODUCTION OF BENZYLISOQUINOLINE ALKALOIDS

Thesis by
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ABSTRACT

The engineering of synthetic metabolic pathways in microbial hosts holds much promise for the synthesis of new chemicals and materials, including a variety of natural and non-natural products. The benzylisoquinoline alkaloids (BIAs) represent a large and structurally diverse class of plant secondary metabolites that exhibit a broad range of pharmacological activities. The reconstitution of a BIA biosynthetic pathway in an engineered microbial host offers several advantages over isolation from plants, including the targeted production of key intermediate molecules, rapid biomass accumulation, ease of purification, and the availability of genetic tools for strain engineering and pathway optimization.

Here we describe the development of a synthetic BIA pathway in an engineered yeast host which incorporates heterologous enzymes from a variety of organisms. The BIA backbone is derived from two molecules of tyrosine and is assembled through a heterologous pathway comprising enzymatic activities from plants, bacteria, and humans. Simultaneous efforts have focused on the downstream portion of this pathway to convert a commercially available substrate to the major branch point intermediate reticuline. By synthesizing both stereoisomers of reticuline from a racemic substrate, we have demonstrated production of BIA metabolites along the diversified sanguinarine/berberine and morphinan branches. Further optimization, scale-up, and a combination of bioconversions and chemical synthesis will potentially revolutionize drug discovery and manufacturing of these compounds.

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