Metagenomic Screening for Novel Cytochrome P450 Monooxygenase

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Abstract

Metagenomic DNA of a population of uncultured organisms was extracted from activated sludge, and by using polymerase chain reaction(PCR) method, the gene encoding cytochrome P450(CYP) domain was recovered. Degenerate PCR primers based on published sequence information gave internal gene fragments homologous to known P450. This gene was cloned and expressed in *Escherichia coli*. Reduction of the protein with dithionite and treatment with carbon monoxide resulted in the formation of a carbon monoxide difference spectra with the typical 450 nm absorbance of a P450. The purified gene product catalyzed the oxidation of 7-Ethoxycourmin to 7-Hydroxycourmarin.

Reference

 Tsuneo Omura and Ryo Sato, The carbon monooxide-binding pigment of liver microsomes, The Journal of Biological Chemistry (1964), 239, 2370-2378.