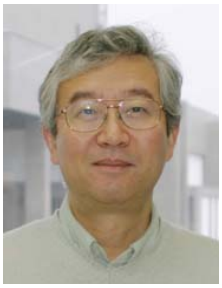
【Grant-in-Aid for Scientific Research(S)】
Science and Engineering (Chemistry)



Title of Project： Molecular Design of Biocatalysts for Hydroxylation of Small Alkanes

Yoshihito Watanabe
(Nagoya University, Research Center for Materials Science,
Professor)

Research Area：Bioinorganic Chemistry
Keyword：Enzyme, Cytochrome P450, Gaseous Alkane, Catalyst, Hydroxylation reaction

【Purpose and Background of the Research】

Methane and ethane, the major components of natural gas, have been regarded as one of the most promising resources as alternatives of oil. However, due to their inertness, it is necessary to transform them into their derivatives to utilize them for synthetic chemistry. Although conventional hydroxylation reactions can convert them to the corresponding alcohols, it requires extremely harsh reaction conditions. In this research project, we intended to develop biocatalytic systems that can proceed hydroxylation of methane as well as ethane under mild conditions.

【Research Methods】

Cytochrome P450s (P450s) are ubiquitous enzymes that comprise a superfamily of heme-containing monooxygenases. P450s isolated from bacteria have been regarded as attractive candidates for oxidation catalysts because of their high catalytic activity for direct oxygen insertion into unactivated C-H bonds. However, the substrate specificity of bacterial P450s is very high and thus their use in organic synthesis has been limited.

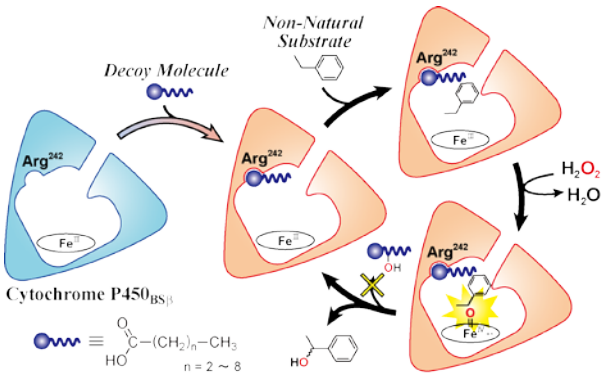


Figure 1. Schematic representation of the reaction system using decoy molecules.

We have demonstrated that the substrate specificity of hydrogen peroxide-dependent P450$_{BS\beta}$ can be altered by inducing its substrate misrecognition using a decoy molecule, which is recognized by P450$_{BS\beta}$ as a substrate due to its structural similality. In the presence of decoy molecules, a wide variety of nonnatural substrates could be oxidized by the H_2O_2/P450$_{BS\beta}$ system (Fig 1). Recently, we have expanded this strategy to other P450s such as P450$_{SP\alpha}$ and P450$_{BM3}$. For example, we have reported that the addition of perfluorocarboxylic acids as decoy molecules to P450$_{BM3}$ resultded in the hydroxylation of alkanes to the corresponding alcohols. In this research project, we intend to construct a reaction system by developping decoy molecules that strongly accerarate the hydroxylation of methane.

【Expected Research Achievements and Scientific Significance】

Controlling the substrate specificity of enzymes is one of the key research topics in the field of biocatalytic transformation of organic compounds. The present proposed strategy using decoy molecules is a novel approach to change substrate specificity of enzymes without any mutagenesis. The biocatalytic systems using decoy molecules developed in our laboratory would provide alternatives to conventional chemical processes for small alkane hydroxylation and for valuable product formations such as fine chemicals and drugs.

【Publications Relevant to the Project】

[1] N. Kawakami, O. Shoji, Y. Watanabe, "Use of Perfluorocarboxylic Acids To Trick Cytochrome P450BM3 into Initiating the Hydroxylation of Gaseous Alkanes" *Angew. Chem. Int. Ed.* **2011**, *50*, 5315-5318.
[2] O. Shoji, T. Fujishiro, H. Nakajima, M. Kim, S. Nagano, Y. Shiro, Y. Watanabe, "Hydrogen Peroxide Dependent Monooxygenations by Tricking the Substrate Recognition of Cytochrome P450$_{BS\beta}$" *Angew. Chem. Int. Ed.* **2007**, *46*, 3656-3659.

【Term of Project】FY2012-2016
【Budget Allocation】171,100 Thousand Yen

【Homepage Address and Other Contact Information】

http://bioinorg.chem.nagoya-u.ac.jp/
E-mail: yoshi@nucc.cc.nagoya-u.ac.jp