**Poster Presentation**

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## *Structure of hydrogen sulfide-producing enzyme from a periodontal pathogen*

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Hydrogen sulfide ($H_2S$) is one of the predominant volatile sulfur compounds that are primarily responsible for oral malodor and contribute to the progress of periodontal disease. $H_2S$ in the human oral cavity is generally produced by enzymatic actions of oral bacteria. *Fusobacterium nucleatum*, a Gram negative periodontal pathogen, is known to be one of the heaviest $H_2S$ producers [1]. For now, four genes (*fn0625*, *fn1055*, *fn1220*, and *fn1419*) encoding pyridoxal-5'-phosphate (PLP)-dependent $H_2S$-producing enzymes have been identified and characterized in *F. nucleatum* ATCC 25586. Of the four enzymes, Fn1055 protein is a unique $H_2S$-producing enzyme, which produces $H_2S$ and L-serine from L-cysteine [2]. Therefore, Fn1055 might play important roles in L-serine biosynthesis in addition to $H_2S$ production in this periodontal pathogen.

Crystal structures of recombinant Fn1055 and its site-directed mutant complex with L-cysteine (a substrate) were determined at 2.1 Å resolution. The enzyme forms a homodimer whose subunits are related by a two-fold axis. The subunit is composed of two domains with α/β structure. The PLP cofactor forms a covalent internal aldimine linkage with the ε-amino group of Lys46 at the bottom of active site cleft between the domains, in the absence of substrate. On the other hand, in the cocrystal of mutant with L-cysteine, the introduced L-cysteine was found to be covalently bound to PLP, instead of Lys46. This covalent intermediate was identified as an α-aminoacrylate, which is the key species of PLP-dependent-enzyme catalysis, by spectrophotometric measurement. Along with the intermediate formation, closure of active site cleft was also observed. Furthermore, we found an amino acid residue acting as a base and confirmed its indispensability for catalysis by enzymatic analyses. These results support that $H_2S$ production by Fn1055 proceeds through the β-elimination of L-cysteine, and enable us to propose a detailed catalytic mechanism of Fn1055.

***[1]*** *S. Persson et al., Oral Microbiol. Immunol., 1990, 5, 195-201,* ***[2]*** *Y. Yoshida et al., Microbiology, 2011, 157, 2164-2171*

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