DEFLUORINATION OF BENZOTRIFLUORIDE BY *Rhodococcus* sp. 065240

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Introduction. Fluorinated compounds are wildly used in the fields of pharmacology, agrochemistry and materials chemistry. The unique properties of fluorine such as strong electronegativity and strong carbon-fluorine bond may confer high stability on fluorinated compounds. Because of their unusual stability, problems of residual fluorinated compounds in the environment have been increasing. Fluorinated compounds are made from the natural inorganic resource, fluorite, i.e., calcium fluoride. Since fluorite-producing countries are very limited, recycling system of fluorine also has lately attracted considerable attention.
In order to find a clue to the solution of these problems, we have screened bacterial strains which catalyze defluorination of fluorinated compounds. In this study, we analyzed a mechanism of bacterial defluorination.

Methods. Benzotrifluoride (BTF) was used as a model compound. BTF has a trifluoromethyl group connected to a benzene ring. No bacteria have not been found which can catalyze defluorination from the trifluoromethyl group. Actinomycetes were isolated from soil samples on humic acid-vitamin agar plates and their ability of defluorination of BTF was assayed by measuring concentrations of free fluoride ion of the cultures by a fluoride ion-selective electrode. Proteomic analysis was carried out by 2D-gel electrophoresis and MALDI-TOF MS. Metabolites were analyzed by ^{19}F NMR.

Results. Several actinomycete strains were found to catalyze defluorination of BTF (1). One of them, 065240 strain, showed the highest ability of defluorination (Fig. 1). 16S rRNA analysis revealed that the strain belongs to the *Rhodococcus* group, and therefore named *Rhodococcus* sp. 065240. In order to investigate a mechanism of defluorination, proteomic analysis of 065240 cells grown with or without BTF was carried out. As a result, it was found that several proteins including isopropylbenzene degradation enzymes were induced by BTF. Metabolites in the culture supernatants were analyzed by ^{19}F NMR. Several fluorinated compounds including trifluorobutyric acid (TFBA) and trifluoroacetic acid (TFA) were detected (Fig. 2). This suggests that BTF was degraded via these compounds as intermediates.

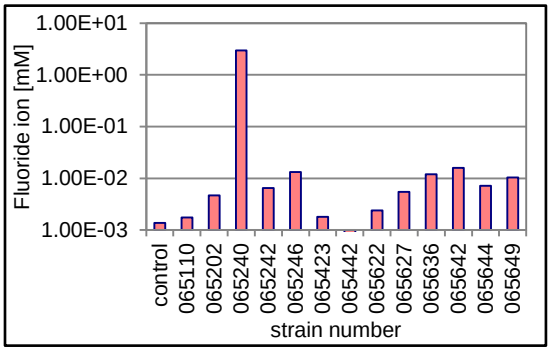

Fig.1 Screening for BTF-degrading bacteria. Concentrations of F^- ion of culture supernatants were analyzed by a fluoride ion-selective electrode.

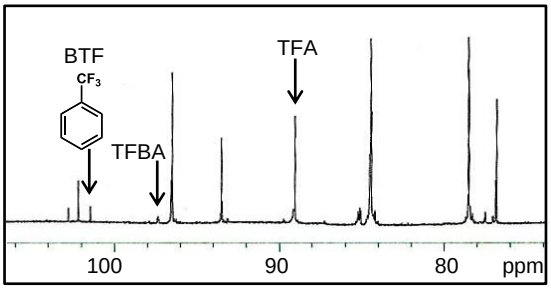

Fig.2 ^{19}F NMR spectrum of the culture supernatant of *Rhodococcus* sp. 065240 grown in the presence of BTF.

Conclusions. *Rhodococcus* sp. 065240 strain, which catalyzed defluorination from BTF, was isolated. Protemomic analysis suggested that isopropylbenzene degradation pathway is involved in BTF degradation.

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References.

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