Division of Environmental Chemistry
– Molecular Microbial Science –

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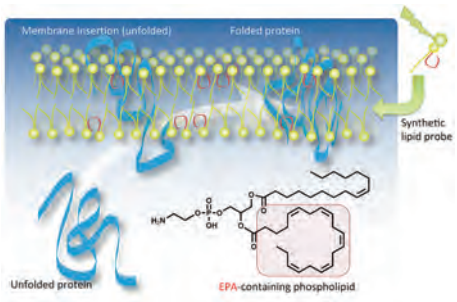
Scope of Research

Microorganisms are found almost anywhere on Earth. They have a great diversity of capacities to adapt to various environments including chemically and physically unusual environments. Our main subject is to clarify the molecular basis of environmental adaptation of microorganisms and their application. Specific functions of proteins and lipids that play essential roles in environmental adaptation of extremophilic microorganisms are of our particular interest. Mechanistic analysis of microbial enzymes, in particular those involved in unique metabolic pathways, and their application are also undertaken.



KEYWORDS
Molecular Microbial Science
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Selected Publications

Dai, X.-Z.; Kawamoto, J.; Sato, S. B.; Esaki, N.; Kurihara, T., Eicosapentaenoic Acid Facilitates the Folding of an Outer Membrane Protein of the Psychrotrophic Bacterium, *Shewanella livingstonensis* Ac10, *Biochem. Biophys. Res. Commun.*, **425,** 363-367 (2012).
Sato, S.; Kawamoto, J.; Sato, S. B.; Watanabe, B.; Hiratake, J.; Esaki, N.; Kurihara, T., Occurrence of a Bacterial Membrane Microdomain at the Cell Division Site Enriched in Phospholipids with Polyunsaturated Hydrocarbon Chains, *J. Biol. Chem.*, **287,** 24113-24121 (2012).
Fukuyama, S.; Mihara, H.; Miyake, R.; Ueda, M.; Esaki, N.; Kurihara, T., Characterization of a Thermostable 2,4-Diaminopentanoate Dehydrogenase from *Fervidobacterium nodosum* Rt17-B1, *J. Biosci. Bioeng.*, (in press) (2013).
Sato, S. B.; Park, J.; Kawamoto, J.; Sato, S.; Kurihara, T., Inhibition of Constitutive Akt (PKB) Phosphorylation by Docosahexaenoic Acid in the Human Breast Cancer Cell Line MDA-MB-453, *Biochim. Biophys. Acta.*, **1831,** 306-313 (2013).
Siwek, A.; Omi, R.; Hirotsu, K.; Jitsumori, K.; Esaki, N.; Kurihara, T.; Paneth, P., Binding Modes of DL-2-Haloacid Dehalogenase Revealed by Crystallography, Modeling and Isotope Effects Studies, *Arch. Biochem. Biophys.*, **540,** 26-32 (2013).
Yoshimune, K.; Kawamoto, J.; Kurihara, T., Proteins Involved in Cold Adaptation Cold-Adapted Microorganisms, *Cold-Adapted Microorganisms*, 97-110 (2013).

Elucidation of Metal-reducing System of a Psychrotrophic Bacterium, *Shewanella livingstonensis* Ac10

A cold-adapted microorganism, *Shewanella livingstonensis* Ac10, isolated from Antarctic seawater has more than 40 kinds of cytochrome-related genes possibly involved in respiratory-linked metal reduction. In the presence of trivalent iron, ferric citrate, as a final electron acceptor for bacterial iron respiration, *S. livingstonensis* Ac10 inducibly produces eight proteins including a homolog of a phosphate-selective porin, PhoE, of *Escherichia coil*. PhoE has positively charged residues at its membrane-spanning region and acts as a channel protein to facilitate the phosphate uptake. However, the role of PhoE in the bacterial iron respiration is still unknown. In this study, we focused on a physiological role of PhoE in the iron-respiration mechanism of *S. livingstonensis* Ac10 and generated the gene-deletion mutant, Δ*phoE*, by two-step single crossover homologous recombination. When fumarate was used as a final electron receptor, Δ*phoE* grew normally. On the other hand, in the presence of ferric citrate, Δ*phoE* showed the growth retardation, and the generation of divalent iron was significantly reduced (Figure 1). Introduction of a *phoE*-expression vector suppressed the growth retardation, indicating that PhoE is required for the iron respiration of this strain. We also examined the growth of Δ*phoE* in the presence of insoluble iron oxide (III), and demonstrated that the growth of Δ*phoE* and the parent strain was almost similar. These results suggest that *S. livignstoneneis* Ac10 inducibly produces PhoE to facilitate iron uptake through membrane in the presence of soluble trivalent ferric ion.

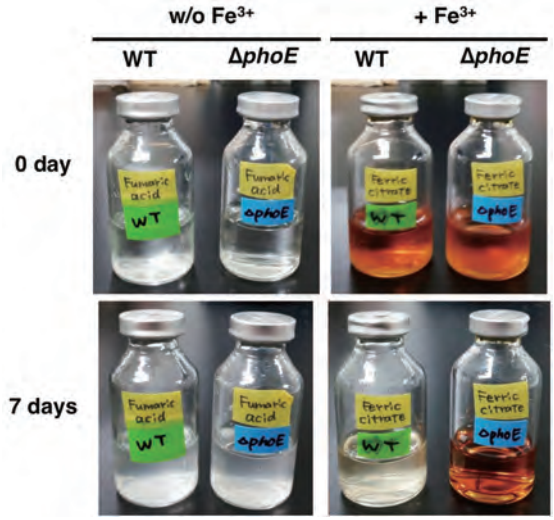

Figure 1. Respiratory iron reduction of *Shewanella livingstonensis* Ac10 and *phoE*-disrupted mutant.

Subcellular-localized 1-Acylglycerol-phosphate Acyltransferase Regulates the Cell Division of a Psychrotrophic Bacterium, *Shewanella livingstonesis* Ac10

Shewanella livingstonensis Ac10 isolated from Antarctic seawater produces a kind of polyunsaturated fatty acids, eicosapentaenoic acid (EPA), as the membrane phospholipids, which alters the physicochemical properties of cell membrane and facilitates the function of cell-division proteins and the folding of membrane proteins at low temperatures. EPA is exclusively restricted to the *sn*-2 position of phospholipids. 1-Acyl-*sn*-glycerol-3-phosphate acyltransferase (PlsC) catalyzes the acylation at the *sn*-2 position of 1-acyl-*sn*-glycerol-3-phosphate (LPA) to form phosphatidic acid (PA), suggesting that the PlsC is involved in the synthesis of EPA-containing phospholipids. *S. livingstonensis* Ac10 has five putative candidate genes coding for PlsC, PlsC1 to PlsC5. The gene-disrupted mutant of PlsC1 (Δ*plsC1*) showed the growth retardation and filamentous cells at low temperatures, and the amount of EPA-containing phospholipids was significantly decreased, indicating that PlsC1 is a key enzyme in the biogenesis of EPA-containing phospholipids. When His-tagged PlsC1 was expressed in Δ*plsC1*, the growth retardation and filamentous cell formation were suppressed, and His-tagged PlsC1 was localized at mid-cell region of *S. livingstonensis* Ac10. On the other hand, *E. coli* PlsC also catalyzes the acylation of LPA from EPA-coenzyme A similar to PlsC1 *in vitro*. Immunofluorescence microscopy was also demonstrated that His-tagged PlsC expressed in Δ*plsc1* was widely distributed in cell. These results suggest that the biogenesis of EPA-containing phospholipids synthesized by the activity of PlsC1 at mid-cell region is required for the appropriate cell division of this strain.

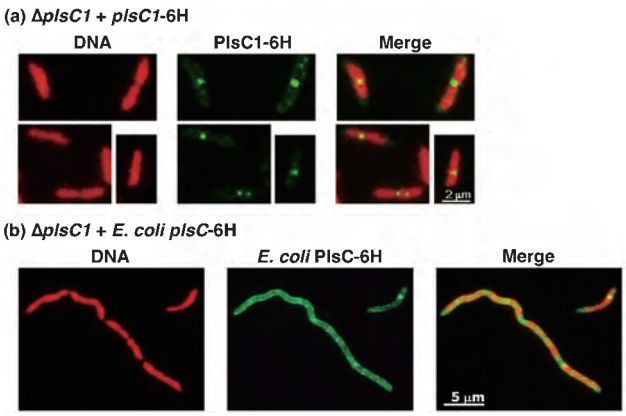

Figure 2. Subcellular localization of PlsC1 (a) and *E. coli* PlsC (b) in the *plsC1*-disrupted strain of *Shewanella livingstonensis* Ac10.