Division of Environmental Chemistry
– Molecular Microbial Science –

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Prof
KURIHARA, Tatsuo
(D Eng)

Assist Prof
KAWAMOTO, Jun
(D Agr)

Assist Res Staff
TANAKA, Yumi

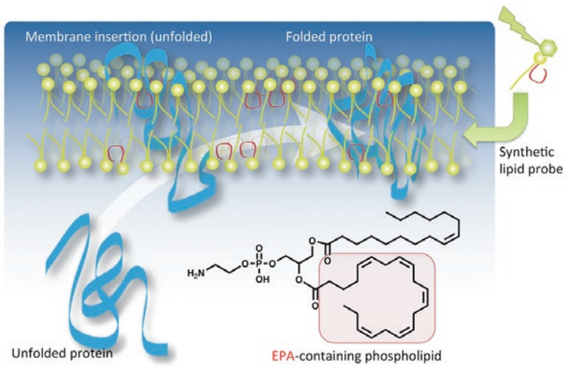
Res Support Staff
UTSUNOMIYA, Machiko

Students

CHO, Hyun-Num (D3)
GONG, Chunjie (D3)
IMAI, Takeshi (D3)
EIKYU, Yurie (M2)
MIZUTANI, Ayano (M2)
SUGIURA, Miwa (M2)
UENO, Genjiro (M2)
KAWAI, Soichiro (M1)
MORIMOTO, Ryohei (M1)
YAMAURA, Takayuki (M1)
SUMIDA, Yukie (RS)

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
Biochemistry
Psychrotroph
Polyunsaturated Fatty Acid
Bioengineering

Selected Publications

Sato, S.; Kawamoto, J.; Sato, S. B.; Watanabe, B.; Hiratake, J.; Esaki, N.; Kurihara, T., Occurrence of Bacterial Membrane Microdomain at the Cell Division Site Enriched in Phospholipids with Polyunsaturated Hydrocarbon Chain, *Journal of Biological Chemistry*, **287,** 24113-24121 (2012).
Park, J.; Kawamoto, J.; Esaki, N.; Kurihara, T., Identification of Cold-inducible Inner Membrane Proteins of the Psychrotrophic Bacterium, *Shewanella livingstonensis* Ac10, by Proteomic Analysis, *Extremophiles*, **16,** 227-236 (2012).
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, *Biochemical and Biophysical Research Communications*, **425,** 363-367 (2012).
Nakayama, T.; Kamachi, T.; Jitsumori, K.; Omi, R.; Hirotsu, K.; Esaki, N.; Kurihara, T.; Yoshizawa, K., Substrate Specificity of Fluoroacetate Dehalogenase: An Insight from Crystallographic Analysis, Fluorescence Spectroscopy, and Theoretical Computations, *Chemistry*, **27,** 8392-8402 (2012).
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, *Biochimica et Biophysica Acta*, **1831,** 306-313 (2013).

Characterization of 1-Acyl-*sn*-glycerol-3-phosphate Acyltransferase from a Polyunsaturated Fatty Acid-producing Bacterium, *Shewanella Livingstonensis* Ac10

Shewanella livingstonensis Ac10, a psychrotrophic bacterium, produces the omega-3 polyunsaturated fatty acid eicosapentaenoic acid (EPA), as a fatty acyl chain of phospholipids at low temperatures. EPA is incorporated into 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 to form phosphatidic acid (PA). We found that 5 genes code for proteins homologous to *Escherichia coli* PlsC (named PlsC1 through PlsC5), suggesting that these PlsCs are involved in the synthesis of EPA-containing phospholipids. To examine the role of these putative PlsCs, we constructed the knockout mutants of each plsC gene (ΔplsC1 to ΔplsC5). In the mutant ΔplsC1, the amount of phospholipids containing EPA was less. Functional expression studies in a temperature-sensitive mutant of PlsC, *E. coli* JC201, showed that PlsC1 has a PlsC activity with a broad acyl-coenzyme A (acyl-CoA) specificity including EPA-CoA. These results indicate that PlsC1 is a key enzyme in the synthesis of EPA-containing PA in *S. livingstonensis* Ac10.

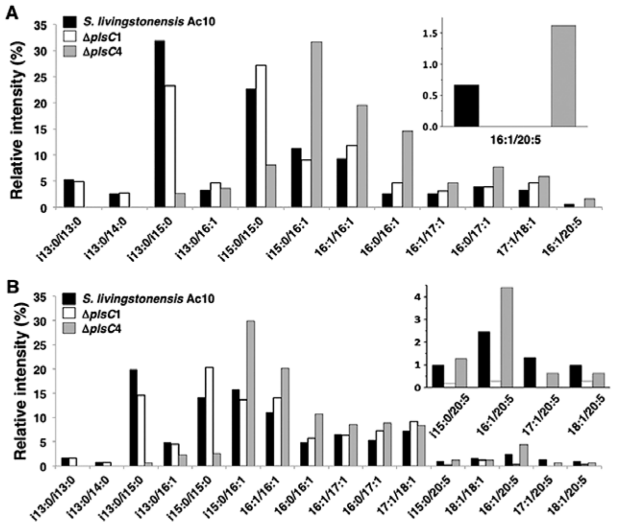


Figure 1. Composition of phosphatidylethanolamine (A) and phosfatidylglicerol (B) of *S. livingstonensis* Ac10 cultivated at 4°C.

Eicosapentaenoic Acid Regulates the Membrane Localization of Cell Division Proteins of a Psychrotrophic Bacterium, *Shewanella Livingstonensis* Ac10

A cold-adapted microorganism, *Shewanella livingstonensis* Ac10 isolated from Antarctic seawater, produces eicosapentaenoic acid (EPA) as an acyl chain of its membrane phospholipids at 4˚C. When EPA-biosynthesis genes were disrupted, the EPA-lacking mutant showed the growth retardation and filamentous cells at 4˚C, but not at 18˚C, suggesting that EPA-containing phospholipids have an important role in the cell division of this bacterium at low temperatures. We also found that, in the absence of EPA, the membrane localization of a cell division-related protein, FtsE, was changed, and supplementation of EPA-containing phospholipids suppressed the defect of membrane localized FtsE. To confirm the involvement of EPA with FtsE and its partner protein, FtsX, *in vivo* localization of these proteins was analyzed. In the wild type, FtsE was localized at cell division site. On the other hand, spiral formed FtsE was observed only from the EPA-less mutant (Figure 2). These results indicate that EPA-containing phospholipids regulate the membrane localization of FtsE and the assembly of FtsEX complex during its cell division at low temperatures.

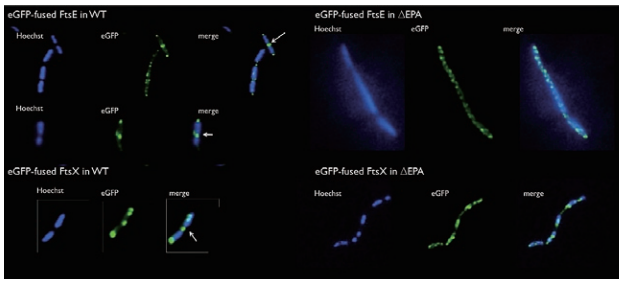


Figure 2. Localization of eGFP-fused FtsE and FtsX in *Shewanella livingstonensis* Ac10 and the EPA-less mutant.