

ENZYMES INVOLVED IN POLYUNSATURATED FATTY ACID SATURATION METABOLISM IN LACTIC ACID BACTERIA AND ITS APPLICATION FOR FUNCTIONAL LIPID SYNTHESIS

Jun Ogawa, Div. Appl. Life Sci., Grad. Sch. Agric., Kyoto Univ. Japan
ogawa@kais.kyoto-u.ac.jp

Michiki Takeuchi, Lab. Ind. Microbiol., Grad. Sch. Agric., Kyoto Univ. Japan
Shigenobu Kishino, Div. Appl. Life Sci., Grad. Sch. Agric., Kyoto Univ. Japan
Yasunori Yonejima, Nitto Pharmaceutical Industries, LTD, Japan

Key Words: fatty acid saturation, fatty acid hydratase, dehydrogenase, isomerase, enone reductase.

Polyunsaturated fatty acids and probiotic lactic acid bacteria are reported to be effective to prevent metabolic syndrome. The mechanism, however, was not clear yet. We revealed the polyunsaturated fatty acid saturation metabolism in *Lactobacillus plantarum* AKU 1009a, which converted linoleic acid into conjugated linoleic acid (CLA)¹⁾.

The enzyme system for this saturation metabolism was found to consist of four enzymes (hydratase²⁾, dehydrogenase³⁾, isomerase, enone reductase⁴⁾) and generate hydroxy fatty acids, oxo fatty acids, and conjugated fatty acids as intermediates. The homologous genes encoding these four enzymes were found in genome sequences of many gut microorganisms. Therefore, acting in concert, gut microbiota may mediate the unsaturated fatty acid saturation metabolism in gastrointestinal tract.

Furthermore, we confirmed the existence of these fatty acids in host tissues depending on the existence of gut microbes using specific pathogen free (SPF) mouse and germ free mouse¹⁾. Successive analysis revealed health promoting activity of these hydroxy and oxo fatty acids, i.e., intestinal epithelial barrier protection⁵⁾, anti-obesity⁶⁾, and anti-diabetic activity⁷⁾, etc. Therefore, we developed novel production system for these fatty acid metabolites using the enzymes from probiotic lactic acid bacteria^{8,9,10)}.

10-hydroxy-*cis*-12-octadecenoic acid (HYA), an initial intermediate of linoleic acid saturation, has immunomodulatory activity and ameliorates intestinal epithelial barrier impairment, etc⁵⁾. HYA was found in foods such as cheese, bacon, milk and vegetable pickles, but at low level. We developed hydroxy fatty acid production process using fatty acid hydratase in probiotic lactic acid bacteria. HYA was produced from safflower oil rich in linoleic acid (approximately 75%) with high conversion rate of approximately 50% with *Lactobacillus plantarum*. We achieved the industrial scale production using 2,000 L fermenter and 500 L reactor. Other C18 Δ 9 unsaturated fatty acids such as oleic acid, α -linolenic acid, and γ -linolenic acid were also converted to corresponding 10-hydroxy fatty acids. The various hydroxy fatty acids provided by this technology using fatty acid hydratase in probiotic lactic acid bacteria are promising as novel functional fatty acids. These studies could open a new application of the enzymes involved in polyunsaturated fatty acid saturation in lactic acid bacteria to novel functional lipid production.

- 1) S. Kishino, et al. : Proc. Natl. Acad. Sci. USA, 110, 17808 (2013).
- 2) M. Takeuchi, et al. : J. Biosci. Bioeng. 119, 636 (2015).
- 3) M. Takeuchi, et al. : J. Mol. Catal., B Enzym. 117, 7 (2015).
- 4) H. Feng, et al. : FEBS Journal, 282, 1526-1537 (2015).
- 5) J. Miyamoto, et al. : J. Biol. Chem., 290, 2902 (2015).
- 6) T. Nanthirudjanar, et al. : Lipids, 50, 1093-1102 (2015).
- 7) T. Goto, et al. : Biochem. Biophys. Res. Commun., 459, 597 (2015).
- 8) H. Sakurama, et al. : J. Lipid Res., 55, 1855 (2014).
- 9) M. Takeuchi, et al. : J. Appl. Microbiol., 120, 1282-1288 (2016).
- 10) M. Takeuchi, et al. : Biosci. Biotechnol. Biochem., 80, 2132-2137 (2016) .