2012 Master Thesis

Enzymatic Characterization of Human Soluble Epoxide Hydrolase

Kwansei Gakuin University, School of Science and Technology
Department of Bioscience, Imaoka Laboratory, Elsa Angelin Leuhery

Soluble Epoxide Hydrolase (sEH) is an enzyme which has a C-terminal epoxide hydrolase and N-terminal phosphatase activities. The endogenous substrates for epoxide hydrolase (EH) activity are epoxyeicosatrienoic acids (EETs), which have biological functions such as vasodilation, anti-inflammation, and angiogenesis. In previous study in our laboratory, it was found that the substrates for phosphatase domain are lysophosphatidic acids (LPAs), which have biological roles in cell proliferation and motility. It was also found that overexpression of phosphatase activity of sEH suppressed VEGF expression and cell growth. In human, sEH has six genetic variants of polymorphism (K55R, R103C, C154Y, R287Q, V422A, and E470G). It is reported that K55R increases the risk of ischemic stroke, and R287Q enhances insulin resistance in type II diabetes patients. However, the activities of these variants toward endogenous substrates including EETs and LPAs are not well known. In this study, the effects of sEH polymorphism on EH and phosphatase activities toward synthetic substrates, PHOME and 4-methylumbelliferyl phosphate (4-MUP) were investigated. We also investigated EH activity toward EETs and phosphatase activity toward LPAs.EH activities of all variants toward PHOME were almost same compared with wild type (WT) sEH. Phosphatase activities toward 4-MUP of all variants except E470G were lower than WT, but some variants had higher phosphatase activity toward LPAs than WT did. These results suggest that the effects of substitution of amino acid in sEH polymorphism on phosphatase activities toward endogenous and synthetic substrates are different. The changes in phosphatase activity of sEH polymorphism may affect the physiological functions of LPAs.