[S5-4] [4/18/2003(Fri) 15:50-16:30/Maple Hall]

New Insights in Arachidonate Cascade: Biochemical Characterization and Biological Significance of Three Distinct Prostaglandin E Synthases

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Biosynthsis of prostaglandin E2 (PGE2), the most common prostanoid with potent and diverse bio-activities, is regulated by three sequential enzymatic steps composed of phospholipase A2, cyclooxygenase (COX), and prostaglandin E synthase (PGES). Recently, three distinct PGESs have been identified; two of them are membrane-bound enzymes, mPGES-1 and mPGES-2, and the third one is a cytosolic enzyme, cPGES. Biochemical properties and biological significance of each enzyme will be discussed.

(1) mPGES-1, a glutathione (GSH)-requiring perinuclear protein, is induced by proinflammatory stimuli, is down-regulated by anti-inflammatory glucocorticoids, and is functionally coupled with COX-2 in marked preference to COX-1. Since Cox-2-derived PGE2 is implicated in various patho-physiological events, such as rheumatoid arthritis, febrile response, bone metabolism, and tumorigenesis, we examined the potential involvement of mPGES-1 in these phenomena using biochemical techniques. Gene disruption study of mPGEs-1 was also carried out.

(2) cPGES is a GSH-requiring enzyme and is expressed constitutievly. This enzyme is functionally coupled with COX-1, not COX-2, to promote immediate PGE2 production. Since the enzymatic activity of recombinant cPGES prepared from bacterial expression system was rather low, we hypothesized that mammalian cells may contain some accessory components that enhance cPGES activity. Indeed, we identified heat shock protein (HSP) 90, a molecular chaperon that forms a stable complex with cPGES in cells, to be one of

such factors. In addition, several lines of evidence suggest that cPGES was directly phosphorylated by casein kinase II (CK-II), one of the client proteins of HSP 90. The phosphorylation of cPGES was accompanied by a marked increase in its enzymatic activity. This is the first evidence that the eicosanoid-biosynthesis is under the control of the molecular chaperon and its client protein.

(3) mPGES-2 was originally purified from bovine heart and has a thioredoxicin-like domain. Its *in vitro* activity did not strictly show GSH-dependence but was activated by various thiol reagents. This enzyme existed mostly as an N-terminally truncated protein in a wide variety of cells and tissues constitutively, and was functionally coupled with both COX-1 and COX-2.

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