Expression of Cyclooxygenase-2 via NF-κB in Cultured Human Breast Epithelial Cells

Kim Jung-Hwan°, Surh Young-Joon

College of Pharmacy, Seoul National University, Seoul, 151-742, Korea.

The inducible form of cyclooxygenase-2 (COX-2) has been often observed in various types of cancerous and transformed cells. In this study, we examined molecular mechanism underlying regulation of COX-2 expression through of the eukaryotic transcription factor NF-κB. Cyclooxygenase expression induced by 12-O-tetradecanoylphorbol-13-actate (TPA) in human breast epithelial cells (MCF10A) were inhibited by specific mitogen-activated protein kinase (MAPK) inhibitors, such as SB 203580 (p38 MAPK inhibitor) and PD 98059 (ERK inhibitor) and also by dominant negative (DN) MAP kinase expressing vectors (pCMV5-p38 MAPK DN mutant or pCEP4-pERK2 DN mutant vector). In the luciferase reporter gene assay, NF-κB transcriptional activities were suppressed by dominant negative MAPKs mutant vectors. In anther study, we assessed the COX-2 expression and NF-κB activation in activated H-ras oncogene transformed MCF10A cells, but we did not found any distinct differences between MCF10-H-ras and its parental cell line in terms of COX-2 and NF-κB activation.
These result suggest that ras activation alone is not sufficient to induce COX-2 expression in human breast epithelial cells and activation of other pathways including p38 MAPK may be required for up-regulation of COX-2 in these cells.

[PC1-38] [10/20/2000 (Fri) 15:30 - 16:30 / [Hall B]]

Synthesis and Characterization of DDT Immunogens

Hong JY, Kim SH, Choi MJ

Life Sciences Division, Korea Institute of Science and Technology, Seoul, Korea

For development of the immunodetection method of DDT family (4,4'-dichlorodipheny-2,2,2-trichloroethane, a persistent and broad toxic organochlorine pesticide), various DDT derivatives were synthesized and characterized for the use of immunogen and the coating ligand of the antibody evaluation. The appropriate lengths of linkers were introduced to investigate more efficient DDT derivatives. Carboxylic acid group was chosen as a functional group for coupling with carrier protein at the terminal position of linker. DDA was readily obtained from the oxidation reation of DDT. DDHP having three carbon linker was directly prepared from the reaction of glutaric anhydride with 4-chlorophenylmagnesium bromide, subsequently chlorination of hydroxyl group at C-1 position gave DDCP. Other derivatives with long chain linker and amide group in linker were prepared through similar reaction. The detail synthetic method for DDA, DDAAP, DDHP DDCP, DDHH, DDCH, DDHHAP, DDCHAP will be discussed.
Among these hapten derivatives, DDA, DDHP and DDCP were conjugated with keyhole limpet hemocyanin for the use of immunogen to produce antibodies. The BSA conjugates of these derivatives were prepared as a coating ligand for the antibody screening. Several monoclonal antibody clones were screened using these probes.

[PC1-39] [10/20/2000 (Fri) 15:30 - 16:30 / [Hall B]]

Effects of endocrine disruptors in whole-organ culture of mouse mammary glands

Je KH°, *Cho MH, and Mar WC

Natural Products Research Institute Seoul National University, Seoul 110-460, *College of Veterinary Medicine, Seoul National University, Suwon 441-744

Estrogens are potent mitogens in a number of target tissue including the mammary gland where they play a pivotal role in the development and progression of mammary carcinoma. Many endocrine disruptors (EDs) show the estrogenic effect. As the effects of EDs are reported to be main causes of hormone-related cancers such as breast cancer among women, we studied the effects of EDs using mouse mammary gland organ culture (MMOC) model. Also, the expression of estrogen receptor and p53 in the preneoplastic lesion was measured by using the flow cytometry. The research on more parameters related to the breast cancer will help in proving the mechanism of preneoplastic lesion by EDs. Moreover, it will help develop the antiestrogenic agent to inhibit the EDs activity.

[PC1-40] [10/20/2000 (Fri) 15:30 - 16:30 / [Hall B]]

Effects of PCBs on human mast cell line HMC-1.

Kwon OK°, Moon TC, Chang HW

College of Pharmacy, Yeungnam University, Gyongsan 712-749, Korea

Polychlorinated biphenyls (PCBs) are widely spread environmental contaminants consisting of chemical mixtures containing many of the 209 possible congeners. The potential immunomodulatory properties of PCBs have been the subject of extensive experimental investigations. The available evidence indicates that the immune system is a target for PCBs and is perhaps one of the most sensitive indicators for adverse PCB induced health effects.
Mast cells are the primary effector cells of immediate hypersensitivity reactions in humans and their numbers are increased in a broad spectrum of pathologic conditions.
We have examined effects of PCBs on human mast cell line HMC-1. In this study, expressions of xenobiotic responsive genes were analyzed to examine their molecular mechanisms in 2,2',4,4',5,5'-hexachlorobiphenyl (2,2',4,4',5,5'-hexaCB)-treated HMC-1. Reverse transcriptase-polymerase chain reaction (RT-PCR) and immunoblot analysis were performed to detect altered expressions of genes associated with 2,2',4,4',5,5'-hexaCB responses. The RT-PCR analysis showed that interleukin-6 (IL-6) and cyclooxygenase-2 (COX-2) genes were well expressed. Whereas interleukin-1β (IL-1β) and interleukin-4 (IL-4) did not expressed. In case of tumor necrosis factor-α (TNF-α) and aromatic hydrocarbon receptor (AhR), gene expressions were decreased by dose dose- and time-dependent manner. However transcription levels of AhR nuclear translocator (ARNT) were not changed.

[PC1-41] [10/20/2000 (Fri) 15:30 - 16:30 / [Hall B]]

Molecular Mechanism of Dioxin-induced Endocrine Disruption through Induction of Oxidative Estrogen Metabolism

Hurh Yeon-Jin°[1], Cho Myung-Haing[2] and Surh Young-Joon[1]

[1]College of Pharmacy, Seoul National University, Seoul 151-742, [2]College of Veterinary Medicine, Seoul National University, Suwon 441-744

2,3,7,8-Tetrachlorodibenzo-para-dioxin (TCDD; dioxin), the prototype agonist of the aromatic hydrocarbon (Ah) receptor, has a marked effect on estrogen metabolism in MCF10A cells by induction of human cytochrome P1A1(CYP1A1) and P450 1B1 (CYP1B1), which are responsible for hydroxylation of 17beta-estradiol (E_2) at C-2 and C-4 positions, respectively. The resulting catechol estragens, 2-hydroxyestradiol ($2OHE_2$) and 4-hydroxyestradiol ($4OHE_2$) have been