COX-2 expression. Second, E2 potentiated BaP-induced nuclear factor-kB (NF-kB) activation, which regulates COX-2 expression. Third, although the aryl hydrocarbon receptor (AhR) did not play a role in BaP-induced COX-2 expression, the potentiation effect of E2 itself was AhR dependent. We further demonstrated that BaP induced the production of genotoxic E2 metabolites (2- and 4- hydroxyestradiols) via AhR-up-regulated cytochromes P450 1A1 and 1B1. These metabolites could directly activate NF-kB to further promote COX-2 mRNA expression in human lung epithelial cells. These findings were further supported by increased PGE2 secretion in rat lung slice cultures. Our findings that the BaP-VE2 interaction enhanced COX-2 expression and hydroxyestradiol accumulation in the media of cultivated lung cells and tissues provide the needed scientific basis for higher risk of BaP-associated lung cancer in females.

P2-118 **BSTB: Tumor and Cell Biology Posters, Tue, Sept 4**

Five-bromodeoxyuridine induces differentiation of a human small cell lung cancer cell line is associated with alteration of gene expression

Chen, Yuan; Pacyna-Gengelbach, Manuela; Deutschmann, Nicole; Ye, Fei; Petersen, Iver

Institute of Pathology, University Hospital Charite, Berlin, Germany

Small cell lung carcinoma (SCLC) is a highly metastatic disease with a poor prognosis due to its resistance to current modes of therapy. SCLC appears to arise from neuroendocrine cells with the potential to differentiate into a variety of lung epithelial cell lineages. In order to investigate molecular events underlying the cell type transition in SCLC, we established a differentiation cell model by modification of a SCLC cell line H526 with a differentiation inducing agent 5-bromodeoxyuridine (BrdU). The BrdU treatment led to a dramatic conversion from a suspension cell line H526 to an adherent cell line variant H526B exhibiting an epithelioid phenotype. DNA fingerprinting by random amplified polymorphic DNA (RAPD) method showed an identical DNA binding pattern between H526 and H526B, indicating that the H526B subpopulation arose from the original cell line H526 and the contamination from other cell types can be ruled out. The BrdU modified cells H526B remarkably reduced the ability of colony formation in soft agar and suppressed the tumor growth rate in immune-deficient nude mice. The phenotypic transition was consistent with upregulation of several lung cancer differentiation markers such as surfactant protein C (SFTPC), thyroid transcription factor 1 (TTF-1), Connexin 26 (Cx26), insulin-like growth factor binding protein-related protein 1 (IGFBP-rP1), as well as homeobox genes like LAGY, PITX1 and HOXB2. Our data suggest that BrdU induced cell differentiation could be linked to the development of a less aggressively phenotype in small cell lung cancer.

P2-119 **BSTB: Tumor and Cell Biology Posters, Tue, Sept 4**

All-trans retinoic acid can restore transforming growth factor-β tolerance in non-small cell lung cancer cell lines through inhibition of Smad1/5/8

Choi, Hyo-Seon[1] Young Mi, Whang[1] Kim, Tae Woo[2] Kim, Seok Jin[3] Kim, Yeul Hong[4]

[1] Genomic Research Center for Lung and Breast/Ovarian Cancers, Seoul, Korea [2] Graduate School of Medicine, Korea University, Seoul, Korea [3] Department of Internal Medicine, Korea University College of Medicine, Seoul, Korea [4] Genomic Research Center for Lung and Breast/Ovarian cancers, Department of Internal Medicine, Korea University College of Medicine, Brain Korea 21 Program for Biomedical Sciences, Seoul, Korea

Transforming growth factor-β (TGF-β) is an essential regulator of cellular physiological processes including proliferation, differentiation and angiogenesis.

TGF-β signaling is mediated through a heterotetrameric cell-surface receptor complex of type II receptor (TβII) and type I receptor (TβRI); type I receptor. Type I receptor has 2 subtypes known as activin receptor-like kinase 5 (ALK5: known as TβI) and activin receptor-like kinase 1 (ALK1). TβII/ALK5 complex activates Smad2/3 pathway and TβRII/ALK1 complex activates Smad 1/5 pathway. These pathways have been known to be associated with cellular growth inhibition through induction of phosphorylated Smad2; whereas cellular proliferation through induction of phosphorylated Sma1/5 in cancer cells.

Retinoids have been shown to possess differentiation, antiproliferative and apoptosis-inducing properties in cancer cells.

We have studied the effects of all-trans retinoic acid (ATRA) after treated with different levels of TGF-β1 in non-small cell lung cancer cell lines (NSCLCs). In addition, we examed whether ATRA effects in signal pathway of NSCLCs.

We analyzed expression of TGF-β receptor I and II (TGF-β I, TGF-β RII) by RT-PCR and treated with ATRA and a variety of concentrations of TGF-β1 for different durations in NSCLCs. In addition, the time-dependent cytotoxic effects of ATRA alone, TGF-β1 alone and combination were studied by the MTT and cell counting assays. We carried out western blot analysis to evaluate whether combined treatment affects the expression of Smads or TGF-β RI (ALK5, ALK1). Finally, we carried out migration assay to evaluate whether the combined treatment of ATRA and TGF-β1 affects the migration of NSCLCs.

The results showed that TGF-β1 alone increased cellular growth of NSCLCs, and the combined treatment of ATRA and TGF-β1 inhibited the growth of NSCLCs. The combined treatment of ATRA and TGF-β1 reduced TGF-β1-induced phosphorylated Smad1/5/8. However, the total protein levels of ALK5 and ALK1 were not changed by the combined treatment of ATRA and TGF-β1. In addition, the combined treatment of ATRA and TGF-β1 reduced migration compared to the treatment with TGF-β1 in A549 and H1299 cell lines.

Therefore, these findings suggest that treatment with ATRA reduced the growth and migration of NSCLCs by inhibition of TGF-β1-induced phosphorylated-Smad1/5/8 which caused growth progression and migration. ATRA may have a potential therapeutic role in the treatment of lung cancer patients with increased levels of TGF-β1.

This study was supported by a grant of the Korea Health 21 R&D project, the Ministry of Health & Welfare, Republic of Korea (A010250).