[P-62]

SAFETY EVALUATION OF ADENOVIRUS-MEDIATED P16 GENE TRANSFER BY USING MICROARRAY AND 2D/MALDI-TOF

Misun Park, Hoil Kang, Jaehee Pyo, Sinae Lim, Seungwan Jee, Miok Eom, Taikyung Ryeom and Okhee Kim

Division of Genetic Toxicology, National Institute of Toxicological Research, KFDA, Seoul

p16INK4a tumor suppressor gene transfer in the non-small cell lung cancer cells by transduction of recombinant adenovirus (Ad5CMV-p16) resulted in significant inhibition of cancer cell growth (Anticancer Res., 1998, 18:3257-3261). As a safety concern, we have investigated gene and protein expression after transduction of adenoviral vector (Ad5CMV-p16) in human non-small cell lung cancer (A549) cells by using microarray and 2D gel electrophoresis/ MALDI-TOF. For the detection of oncogenes or unexpected gene expressions, we compared the differential gene expression level in the A549 cells treated with Ad5CMV (null type) and Ad5CMV-p16 virus, respectively, by using oligonucleotide chip which contains oligonucleotides related with signal transduction pathways, cell cycle regulations, oncogenes and tumor suppressor genes. We found that 27 genes were down-regulated among 1200 genes on oligonucleotide chip (Mergen), and several genes including N-ras related gene were up-regulated in the Ad5CMV-p16 virus treated cells compare with Ad5CMV, two fold or more. We confirmed the microarray data by using RT-PCR. We have conducted 2D gel electrophoresis to detect any unexpected protein expression by transduction of AdCMV-16 in A549 cells. We found that the expression of several proteins were changed to 3 fold or more by using PDQuest program. To characterize proteins in more detail, we identified proteins by MALDI-TOF analysis. Currently, we are confirming these results in nude mice lung cancer model. These results suggest that we have to consider the potential effects of the other gene and protein expressions except therapeutic gene on the host cells in gene transfer.

Keyword : afety evaluation, adenoviral vector, microarray, 2D/MALDI-TOF