115 Gene Expression in Mouse Lymph Node Cells irradiated by Ultraviolet-Rays

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Mouse lymph node cell (M10) is highly sensitive against X-ray irradiation. Growth of this cell depended on the UV-rays irradiation. The cell growth was repressed in proportion to the irradiation time. Dynamic changes of 597 genes after the UV-B (310 nm) irradiation were investigated on DNA array membranes (Atlas #7741-1, CLONTECH) using P-33 labeling probe. Radioactivities obtained from imaging analyzer (BAS1500, Fuji film co. ltd.) were analyzed using software of ArrayGauge. The expression of mRNA was further analyzed by EX-ARRAY program. Seven housekeeping genes were used as standards. Irradiations for 10-second, 20-second and 30-second were done, and changes in the gene expression after the cultivation for 2 h were compared with cells without irradiation. As for a group of activated genes, UV-rays reached more than 80 kinds in 597 genes for 10-second irradiation, while 110 kinds as for a group of repression genes. Activated genes decreased due to the irradiation time, and it became clear that repression genes increased in proportion to the irradiation time.

116 Induction of Metallothionein mRNA in Mouse Lens Epithelial Cell by UV Irradiation

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The cataract is promoted by UV irradiation. It is well known that the main causes of UV induced cataract are reactive oxygen species which are generated by UV irradiation. Metallothionein regulates detoxification from heavy metals, and free radical scavenging in living tissues. In this study, we analyzed the expression and induction of Metallothionein-I (MT-I) by UV irradiation using mouse lens epithelial cell line (alphaTN 4-1). AlphaTN 4-1 was irradiated by UV with various doses, and then the cells were incubated for several hours. After incubation each cells were collected and the expression of MT-I mRNA was analyzed by the quantitative RT-PCR method. The relative amounts of MT-I mRNA were normalized against GAPDH mRNA. MT-I was constantly expressed in alphaTN 4-1 and induced by UV irradiation. The amounts of MT-I mRNA changed with increasing dose of UV irradiation. These results suggest that MT-I is activated by UV irradiation and can function as a radical scavenger in lens.

117 Effects of UVB Irradiation on Telomere and Telomerase in Murine Cells

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Telomeres at terminal regions of linear chromosomes in eukaryotes play important roles not only in maintaining the chromosome integrity but also in the exercise of normal cellular functions. Here we studied how UVB irradiation affects telomere and telomerase in cultured murine cells. A TRF method and a TRAP assay measured the telomere length and telomerase activity, respectively. Immediately after UVB irradiation no change in telomere length was shown, but an apparent shortening of telomere was observed markedly in normal BALB cells compared with SCID cells 24 h after UVB irradiation. Telomere activity was strikingly enhanced in a dose dependent fashion 1 h after UVB irradiation: up to 20 times in SCID cells and 10 times in BALB cells as high as non-irradiated cells. Nine-fold high activity was still held in SCID cells 24 h after UVB irradiation. These results suggest that UVB-induced high telomerase expression may repair UVB-damaged telomeres. It is also suggested that DNA-PKcs, deficient in SCID cells, may be involved in the repair process of UVB-damaged telomeres.

118 Responses of cultured cells to chronic low-dose rate UV-B irradiation

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In an attempt to understand the biological effect of physiological and chronic low-dose rate UV-B irradiation, cultured human cells were irradiated with low-dose rate UV-B in situ using a CO_2 incubator equipped with a UV-B tube. The dose-dependent formation of cyclobutane pyrimidine dimers and (6-4) photoproducts in normal human fibroblastic cells, XP 2OS cells, and human keratinocytes (HaCaT) were observed during the chronic irradiation. Synthesis of DNA and RNA in the cells were substantially reduced during the chronic exposure associated with the accumulation of G_1/S phase cells, indicating that DNA replication and transcription were immediately blocked. The intracellular oxidative level elevated continuously with increasing irradiation time. Furthermore, all of the signaling molecules examined, ERK, JNK, p38, and p53, were kept phosphorylated during the irradiation. These observations suggest that DNA damage-dependent S-phase block and p53 phosphorylation, as well as continuous generation of reactive oxygen species followed by MAPK phosphorylation, were coincidentally taking place during the chronic exposure.