The Japan Radiation Research Society

151 Mechanism of delayed mutagenesis by radiation-induced genomic instability

Hiroko YAMAJI[1], Keiji SUZUKI[1], Seiji KODAMA[1], Masami WATANABE[1], [1]Lab. Radiat. Life Sci., Sch. Pharm. Sci., Nagasaki Univ.

Genomic instability is induced in the progeny of surviving cells, which is manifested by the expression of various phenotypes, such as delayed reproductive death, delayed chromosomal instability, and delayed mutagenesis. In the present study, we examined the molecular mechanism of delayed mutagenesis in CHO cells, that harbor the reporter plasmid containing the fusion gene of *LacZ* gene and Zeocin resistance gene. After X-irradiation, white colonies appeared among the blue-stained colonies (LacZ+) in the presence of X-gal as a substrate, indicating that X-irradiation caused the *LacZ* gene mutation. We collected the primary colonies, and performed the secondary colony formation in the absence of Zeocin. The frequency of white colonies was significantly higher than the control colonies. Using PCR, the *LacZ* gene was recovered in approximately 60% of the primary white colonies, but not in the secondary white colonies. These results indicate that deletion mutation occurs predominantly in the progeny of surviving cells. It is suggested that recombination may be involved in delayed mutagenesis.

152 Hypersensitivity of Bloom's syndrome fibroblasts to cell growth inhibition by hydroxyurea

Takayuki KURIHARA[1], Masao INOUE[2], Kuniyosi IWABUCHI[3], Kouichi TATSUMI[4], [1]Div. Basic Sci. Med. Res. Inst. Kanazawa Med. Univ. [2]Div. Core Facility. Med. Res. Inst. Kanazawa Med. Univ. [3]Biochem. Kanazawa Med. Univ. [4]Div. Biol. Oncol., Natl. Inst. Radiol. Sci.

To examine the function of BLM helicase in human cells, BS and normal cells were treated with hydroxyurea (HU), and then cell growth, bromodeoxyuridine incorporation and Chk2 phosphorylation after release from the inhibition by HU were compared between the treated and untreated cells. The hypersensitivity of BS cells to HU for cell growth inhibition was clearer at low concentrations of HU and for a shorter exposure time (1–3 hr), but it disappeared for the extended exposure time (6–9 hr). HU treatment retarded the progression of DNA replication in both BS and normal cells. However, normal cells recovered the progression of DNA replication immediately after release from HU inhibition, while BS fibroblasts did so far behind. Chk2 proteins were phosphorylated by exposure to x-ray in normal cells, and the replication block by short exposure to HU (2 hr) did not have much effect on the activation of Chk2 in both BS and normal cells. The hypersensitivity of BS cells to HU seems to result from the retarded recovery of DNA replication following release from the inhibition by HU, and the cause of the hypersensitivity may be independent of the activation of Chk2 by HU.

153 Strain Differences and Effects of *Scid*-gene on the Genomic Instability in Radiation-induced Thymic Lymphoma in Mice

Yuko YOSHINAGA[1], Hiroshi ISHIHARA[2], Yoshiya SHIMADA[2], Izumi TANAKA[2], Junya NAGAI[2], Toshihiko SADO[1], [1]Oita Univ. Nurs. Health Sci. [2]Natl. Inst. Radiol. Sci.

Since mutation spectra observed in thymic lymphomas (TLs) were known to differ among mouse strains or carcinogens, it has been suggested that the underlying mechanism of carcinogenesis might differ. Thus, we investigated the incidence of genomic instability as expressed by microsatellite instability, IAP retrotransposition, mutation of K-*ras*, LOH in the region containing *Ikaros*, a tumor suppressor gene in TLs, using (1) TLs induced in B6C3F1 mice by radiation or ENU, (2) radiation-induced TLs induced in TL-induction sensitive C57BL/6, resistant C3H, and the hybrid B6C3F1. Using Scid mice, we also studied (3) the effects of genetic background on the IAP retrotransposition of spontaneous and radiation-induced TL. As a result, (1) in B6C3F1 mice, TL-specific microsatellite instability and IAP retrotransposition was rarely observed. ENU-induced TL showed high mutation rates in K-*ras*, while radiation-induced TL manifested high incidence of LOH containing *Ikaros* . (2) As high as 20% of TLs induced in C3H mice had TL-specific IAP transposon. F1 manifested characteristics similar to one parental strain. (3) Microsatellite instability was not observed in Scid mice. TLs induced in both C3H and C.B.-17 strains manifested an increased expression of IAP transposition by radiation regardless of whether they had a *Scid* gene or not.

NII-Electronic Library Service