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"Relationship between spontaneous γH2AX foci formation and progenitor functions in circulating hematopoietic stem and progenitor cells among atomic-bomb survivors"

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Study Findings

The self-renewability of hematopoietic stem cells has been suggested to decline in atomic bomb (A-bomb) survivors exposed to higher radiation doses and who had more DNA damage in their hematopoietic stem and progenitor cells (HSPCs), which are responsible for the production of red, white, and other blood cells.

Explanation

1. Study Purpose

Accumulation of DNA damage in hematopoietic stem cells is known to be one factor in the functional decline of human hematopoiesis associated with aging. Not known, however, is whether an association exists between radiation exposure and hematopoietic decline due to accumulated DNA damage. This study examined the association between radiation exposure and hematopoietic function by measuring DNA damage frequency in the circulating HSPCs of A-bomb survivors.

2. Study Methods

Using peripheral blood collected from 229 consenting A-bomb survivors in Hiroshima who participated in the Adult Health Study (AHS) from 2011 to 2013, we evaluated DNA damage frequency using γH2AX foci formation* as a marker. Also assessed was hematopoietic function including self-renewability of HSPCs, which can be detected by the cobblestone area-forming cell** assay in CD34-positive/lineage marker-negative (CD34+Lin-) HPSCs. In addition, we measured the length of granulocyte telomeres*** as a marker for the assessment of hematopoietic system aging.

***γH2AX foci formation**: In the presence of DNA damage such as DNA double-strand breaks, γH2AX and other DNA repair proteins form a mass called a "focus" at the break. By detecting such foci and ascertaining their numbers, DNA damage frequency can be determined.

****Cobblestone area-forming cell**: When a cell-culture environment is created *in vitro* similar to one in which hematopoietic stem cells self-renew in the bone marrow (culturing HSPCs in the absence of hematopoietic factors and in the presence of stromal cells), hematopoietic stem cells form an undifferentiated cobblestone-like colony, allowing assessment of how many hematopoietic stem cells have self-renewability.

*****Length of granulocyte telomeres**: Cellular telomere length is a marker used to assess aging because length of a telomere, a repeat base sequence located at the end of a chromosome, is shortened at each cell division. The length of granulocyte telomeres is a marker to assess aging of the entire hematopoietic system, because granulocytes have a short lifecycle of a few days *in vivo* and are continuously produced from hematopoietic progenitor cells.

3. Study Results

(1) Frequency of DNA damage in HSPCs in association with radiation dose

DNA damage frequency (γH2AX foci formation) gradually decreased with increasing radiation dose up to 1.5 Gy. At higher doses, an increase in foci was observed. Because DNA damage detected on the basis of γH2AX foci can be repaired within a few days, detected DNA damage may have been induced by oxidative stress *in vivo* over the period of a few days before blood drawing rather than by A-bomb radiation. The association observed in this study with radiation dose may indicate lower levels of DNA damage stress or more effective DNA repair among A-bomb survivors exposed to 1.5 Gy or less. Such details are unclear at this point in time.

(2) Frequency of DNA damage in HSPCs in association with granulocyte telomere length

Frequency of γH2AX foci formation increased with decreasing length of granulocyte telomeres, which suggests an association between DNA damage in HSPCs and aging of the hematopoietic system. A similar association has been reported by other studies. In this study, the association with aging was also confirmed in A-bomb survivors.

(3) An interaction effect between radiation dose and frequency of DNA damage

A negative interaction effect between radiation dose and γH2AX foci formation frequency was indicated in the cobblestone formation assay. This result suggests that the self-renewability of hematopoietic stem cells may have decreased among survivors

who had a higher radiation dose and more DNA damage.

Study Significance

Many years after radiation exposure and as the A-bomb survivors continue to age, their hematopoietic functions as a group show normal aging, but this study suggests that among A-bomb survivors with large quantities of accumulated DNA damage in HPSCs, reduced self-renewability of hematopoietic stem cells due to aging may be modified by A-bomb radiation exposure.

The Radiation Effects Research Foundation has studied A-bomb survivors and their offspring in Hiroshima and Nagasaki for nearly 70 years. RERF's research achievements are considered the principal scientific basis for radiation risk assessment by the United Nations Scientific Committee on the Effects of Atomic Radiation (UNSCEAR) and for recommendations regarding radiation protection standards by the International Commission on Radiological Protection (ICRP). RERF expresses its profound gratitude to the A-bomb survivors and survivors' offspring for their cooperation in our studies.

§***Mutation Research-Genetic Toxicology and Environmental Mutagenesis*** is a section of *Mutation Research* (peer-reviewed journal that publishes research papers in the area of mutation research with a focus on fundamental mechanisms underlying the phenotypic and genotypic expression of genetic damage). This section publishes papers in the field of genetic toxicology, focusing on genotoxicity testing of specific agents, *in vitro* or *in vivo,* and assessment of health effects resulting from genotoxic exposures in human populations. (Impact factor in 2015: 1.83)