Paper published in *Radiation Research*

"Irradiation at different fetal stages results in different translocation frequencies in adult mouse thyroid cells"

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Radiat Res 2016 (October); 186(4):360-6
(doi: 10.1667/RR14385.1)

Study Findings

We examined chromosome aberration frequencies in thyroid tissues of adult mice irradiated during the fetal stage and found that radiation effects varied depending on the fetal stage at the time of irradiation. While mice irradiated as 6.5-day fetuses (before organogenesis) had a relatively low chromosome aberration frequency (5.8×10^{-3}), adult mice irradiated as 15.5-day fetuses (during the fetal stage) showed frequencies that were nearly the same as that of their mothers' (25.3×10^{-3}). Taking into account findings from previous studies, we interpreted these results as an indication that fetal tissue stem cells record radiation damage primarily when exposed after locating in their niche*; before locating in their niche, however, radiation-damaged cells may undergo negative selection and be unable to locate in the niche, and therefore the effects of radiation damage may not remain.

Niche*: Microenvironment in which tissue stem cells can function.

Explanations

Based on findings from epidemiological studies (case-control studies) starting in the 1950s, fetuses have been considered highly sensitive to cancer risks from radiation. However, RERF's study using chromosome aberration frequency in lymphocytes as a marker revealed that few effects of radiation were observed among in-utero survivors. The present study investigating chromosome aberrations in tissues of mice irradiated as fetuses may provide a clue for understanding these apparently contradictory observation results.

1. Study Purpose

To verify differences in fetal irradiation-related chromosome aberrations among tissues of different types, we previously studied hematopoietic cells in mice and mammary gland epithelial cells in rats. In this study, we examined chromosome aberration frequencies in mouse thyroid epithelial cells. We also conducted experiments using irradiation at different stages of fetal development to determine whether or not chromosome aberration frequencies following fetal irradiation differ by when irradiation occurs.

2. Study Methods

Pregnant female mice were irradiated with 2 Gy of X-rays at day 6.5 or 15.5 after conception. After the neonatal mice reached adulthood (at least eight weeks of age), their thyroid glands and those of their mothers were removed. We cultured thyroid epithelial cells for five or six days and prepared chromosome samples using conventional methods. To detect stable chromosome aberrations, such as translocation, occurring in thyroid cells due to fetal irradiation, we employed fluorescence in situ hybridization (FISH),* which labels chromosomes 1 and 3 in green and red, respectively. We then observed slides of labeled chromosomes and counted the number of translocations and other stable chromosome aberrations. In a portion of the mice, spleen T lymphocytes were also cultured and assessed for chromosome analysis.

*FISH (fluorescence in situ hybridization) is a staining technique. To determine the presence or absence of structural abnormality in a target chromosome (chromosomes 1 and 3 in this study), DNA is extracted from a library of the target chromosome and labeled with fluorescence to prepare a probe. (A library contains DNA extracted from individual chromosomes and amplified in Escherichia coli.) Following preparation of single-strand DNA from double-strand DNA of the target chromosome sample (denaturalization), a reaction is prompted with the aforementioned probe (hybridization), making it possible to label a specific DNA sequence artificially.

3. Study Results

(1) Adult mice irradiated with 2 Gy of X-rays as 15.5-day fetuses had a higher translocation frequency (30/1155 or 25.3×10^{-3}) in thyroid cells than did non-irradiated adult controls (0/1007 or 0.1×10^{-3}), a figure that was nearly the same as that experienced by irradiated mothers and other adult females (43/1244 or 33.6×10^{-3}). These results are consistent with previously observed results in fetally irradiated rat mammary cells.

(2) When fetuses were exposed to 2 Gy at an earlier stage of development (day 6.5) before thyroid organogenesis, however, the resulting translocation frequency was much lower (3/502 or 5.8×10^{-3}) than that in the mice irradiated at day 15.5 (30/1155 or 25.3×10^{-3}). These results reveal that fetal radiation sensitivity differs by timing of irradiation, and that the effects of thyroid irradiation prior to thyroid organogenesis are insignificant. On the other hand, few chromosome aberrations were observed in hematopoietic tissues regardless of the timing of irradiation (it is understood that hematopoietic stem cells locate in their niches only after birth).

Study Significance

This study revealed that chromosome aberration frequencies in mouse thyroid cells due to fetal irradiation differed by timing of irradiation (prior to organogenesis/during the fetal stage). Such differences may be partly attributed to whether or not irradiated tissue stem cells have located in their niche. If tissue stem cells are irradiated before locating in this way, aberrant cells cannot locate in their niche; when such cells are irradiated after locating in their niches, however, stem cells may repair DNA damage with the aim of survival, and thus cause mis-repairs of DNA that lead to chromosome aberrations. This study's results will contribute to understanding of the mechanisms behind cancer risks due to fetal radiation exposure.

The Radiation Effects Research Foundation has studied A-bomb survivors and their offspring in Hiroshima and Nagasaki for around 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.

§***Radiation Research***, which is an official monthly journal of the Radiation Research Society, publishes original peer-reviewed papers and review articles on radiation effects and related issues in the fields of physics, chemistry, biology, and medicine. (Impact factor in 2015: 2.67)