Analysis of Unstable Chromosome Aberrations for Partial Body Exposures with Gamma Rays

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Introduction

Scoring of unstable chromosome aberrations (dicentrics, rings and fragments) in circulating lymphocytes are the most extensively studied biological means for estimating individual exposure to ionizing radiation [1]. The effects of radiation, however, can be significantly different when only portions of the body or an individual organ system are irradiated, such as might occur during the use of radiation for medical treatment [2], where some problems can arise especially to quantify the fraction of body irradiated. After partial-body exposure, peripheral blood samples contain a mixture of exposed and unexposed lymphocytes, which render the interpretation of the overall aberration frequencies more difficult.

In this experiment, peripheral blood samples from four volunteers (two Indonesian and two Korean) were irradiated with 2.0 Gy of gamma rays for simulation of partial-body exposure by mixing irradiated and non-irradiated blood from the same volunteers in proportions of 10-100%.

Materials and Methods

Peripheral blood samples (15 ml) were collected in sterile heparinised vacuutainers (Becton Dickinson) from four healthy volunteers (3 males and 1 female) aged between 30 and 47 years old (mean 40.3 y). To simulate partial-body exposures, calculated volumes of irradiated blood (exposed *in vitro to* 2.0 Gy of gamma rays with the dose rate was 3.16 Gy/min) were mixed with appropriate amounts of non-irradiated blood from the same volunteers to obtain the following proportions of irradiated blood: 10, 25, 50, 75 and 100%.

Two milliliters of the mixed blood samples were cultured for 48 h, in a humidified atmosphere containing 5% CO_2, at 37°C. The culture medium consisted of 8.0mL of RPMI-1640 supplemented with 10% heat inactivated FBS, 1% streptomycin/penicillin (Gibco BRL). Besides this, 300㎕ of phytohemagglutinin (PHA-m form, Gibco BRL) was added to stimulate cell division. To block the mitotic process of the cells at the metaphase stage, Colcemid (Gibco BRL) was added for the last 4 h of culture at a final concentration of 0.1 ㎎/㎖. The contents of the tube were then centrifuged for ten minutes at 300 xg and re-suspend in 8 ml of 0.075 M KCl (pre-warmed to 37°C) for twenty minutes. At this

stage, 2 ml of fresh Carnoys Fixative (3:1 = methanol : acetic acid glacial) was added into the tube, and fixation step was repeated three times. The yield of metaphase cells was stored in -20°C freezer at least one night until the preparation of slide was made.

Two slides were prepared for each sample, encoded, and then stained with 10% Giemsa (Merck) and mounted. The number of aberrations was observed under a microscope (Nikon Eclipse Japan) connected to Olympus CCD Camera System for taking picture. In the control samples 1000-1200 metaphase cells were analysed per donor. Tricentrics and tetracentrics were considered as two and three dicentric equivalents, respectively.

Results and Discussion

The frequencies of chromosomal aberrations found in four volunteers after simulation of partial body exposures. A high difference in frequency of chromosome aberration was found in 10% samples of each samples, even some of them were almost two times higher than others. In 25% portion, a much lower frequency was seen in subject number 2 than others, this maybe related to a specific characteristic of this samples or some mistakes in preparation of chromosome. In 50% portion of irradiated blood, the numbers of chromosome aberration were quite similar for all samples analyzed. Very high frequency in chromosome aberration both for 50% and 100% portions was observed in female subject, possibly due to typical characteristic of gender.

There were no significant differences in the distribution of chromosome aberration frequencies among the four volunteer's samples in the respective dose points.

Conclusion

Dose estimates are crucial for risk assessment as well as for clinical planning of treatment of highly exposed victims. The results presented in this experiment concluded that the scoring of chromosome aberration is reliable methodology for investigating exposure to ionizing irradiation, such as partial-body ones.

This study constitutes a preliminary step in our process of defining the possibilities of cytological technique for biological dosimetry expertise. Because of the limitations of a number of cases, no clear conclusion could be reached.

Reference

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