BL-27B/2015G065

Cell-killing effect by the targeted cytoplasmic irradiation for normal human fibroblasts with monochromatic X-ray microbeams

Masao Suzuki[1,*] and Noriko Usami[2]
[1]NIRS, QST, 4-9-1 Anagawa, Chiba, 263-8555, Japan
[2] Photon Factory, 1-1 Oho, Tsukuba, 305-0801, Japan

1 Introduction

Targeted irradiations to either cell nucleus or cytoplasm using microbeams enable us to understand biological effects, such as bystander effects, genomic instability and radioadaptive response, more in detail. The study of such biological effects for low linear energy transfer (LET) radiation can surely provide the important implications for evaluating risk such a low-dose (rate) exposure as the accident of Fukushima Daiichi Nuclear Power Plants. However, most studies for such biological effects induced in cells irradiated with microbeams have been carried out using high-LET radiations and so far only limited data is available to understand biological effects induced by low-LET electromagnetic radiations, such as X or gamma rays.

In this study we have been studying low-LET-radiation induced bystander cellular effects irradiated with targeted cell nucleus or cytoplasm using X-ray microbeams. This year, we established how to irradiate just cytoplasm in normal human fibroblasts and examined cell-killing effect of cytoplasmic irradiation, extending to the last year's experiment.

2 Experiment

Targeted cytoplasmic irradiations of monochromatic X-ray microbeams (5.35keV) were carried out using the cell-irradiation system of X-ray microbeams at BL27B [1]. Briefly, we made the microbeam covering the areas of 30 square micrometers in which the center of the microbeams the gold-made mask that was 22 micrometer in diameter and 20 micrometer in height on a thin SiN film was set in order to shield the nucleus (Fig.1).

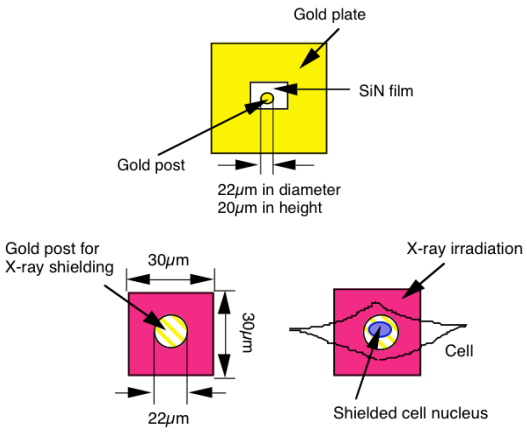

Fig.1: The method for the cytoplasmic irradiation using X-ray microbeams.

Early passaged normal human skin fibroblasts obtained from the Riken BioResource Center were used in this research project. Approximately 1,000 exponentially growing cells were inoculated into the center of each microbeam dish, which was stretching a 2.5μm-thick Mylar film over the bottom of the hole for X-ray window, one day before irradiations. Each cell nucleus stained by Hoechst 33342 was captured by the computerized cell irradiation system (Fig.2). The cytoplasm of all cells captured by the computerized irradiation system was irradiated with 10R and cell-killing effect was measured with a colony-forming assay.

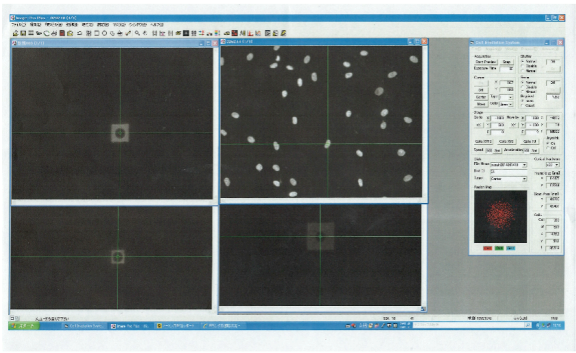

Fig.2: The computerized targeted cytoplasmic irradiation system of X-ray microbeams. The upper photo in the left side shows 40-square-micrometer microbeam and the lower photo in the left side shows 30-square-micrometer microbeam with the gold post.

3 Results and Discussion

So far we have just preliminary results of cell-killing effect with the cytoplasmic irradiation. The plating effeciencies of normal human fibroblasts were to be 34% for non-irradiated control and 35% for 10R cytoplasmic irradiation. The results suggest that no cell-killing effect is induced by the cytoplasmic irradiation. Now we have been examing cell-killing effect by the cytoplasmic irradiation in detail, such as dose-dependent effect.

References

[1] Y. Kobayashi *et al*., *J. Radiat. Res*. **50**, Suppl., A29 (2009).

* suzuki.masao@qst.go.jp