Medical Application

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Synergistic Effect of TRAIL with X-ray Sensitizer on Irradiation-induced Cancer Cell Death

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Introduction

To develop a cancer-specific radiotherapy, we have proposed a novel method that introduces Nb as an X-ray sensitizer into cancer cells and induces cell death with active oxygen species produced by Auger process after X-ray irradiation. We previously reported the introduction of Nb with either Nb-containing cationic liposomes or electroporation into cancer cells, however, no significant increase of cell death after X-ray irradiation was observed [1,2]. We could also detect active oxygen species only at the highest concentration of 1×10^{-2} M Nb atoms after X-ray irradiation. Here, therefore, we report the synergistic effect of TRAIL, one of the apoptosis-inducing ligands of TNF family members [3], with Nb atoms on cancer cell death after X-ray irradiation.

Experiments and Results

Introduction of Nb atoms by electroporation

HeLa cells were suspended in PBS containing 1×10^{-4} M Nb atoms. The cell suspension was charged with 960 μF and 300 V electric pulse using GenePulser (BIO-RAD). Viability of the cells after electroporation was more than 95%, and incorporated Nb atoms were about 3×10^{9} atoms/cell.

X-ray irradiation of the cancer cells containing Nb atoms

Our previous study revealed that the aqueous solution of Nb compounds showed sharp K-edge near 19 keV in the X-ray absorption spectrum. After introducing Nb atoms in HeLa cells by electroporation, the cells were irradiated with 2 Gy of monochromatic X-ray beam from BL-27B at the energy just above or below K-edge of Nb atoms. After irradiation, the culture medium was changed for a fresh medium and a suboptimum dose of hrTRAIL (20 ng/mL) and anti-His_6 antibodies (2 μg/mL) were added simultaneously. The cells were incubated for 18 h, and the number of apoptotic cells was counted by morphological observation. As shown in Figure 1, addition of hrTRAIL increased cell death independent of X-ray irradiation. Upon irradiation coexistence of Nb atoms with hrTRAIL markedly increased cell death, and this tendency was larger at the high energy X-ray irradiation than the low energy one. However, Nb atoms without hrTRAIL did not increase cell death after X-ray irradiation. This synergistic increase of cell death might be caused by low concentration of active oxygen species arisen from Nb atoms by X-ray irradiation, which did not cause cell death by itself, in cooperation with the hrTRAIL-induced apoptotic condition. HrTRAIL might make cancer cells to be highly sensitive to active oxygen species.

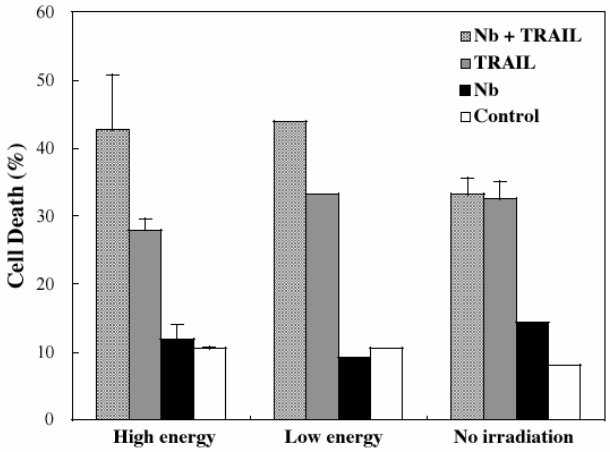

Fig. 1.Synergistic increase of cell death by TRAIL with Nb atoms on X-ray irradiation.

Conclusion

As previously reported [1,2], introduction of Nb atoms at a concentration of 3×10^{9} atoms/cell, which was the highest concentration without any cytotoxicity, produce insufficient concentration of active oxygen species resulted from X-ray irradiation to induce significant cell death in cancer cells. However, suboptimum dose of hrTRAIL showed synergistic increase of cell death specifically for irradiation. These results show that Nb atom is not so eminent to produce active oxygen species enough to induce cell death by X-ray irradiation. Heavier atoms, such as Ba, which have higher efficiency both to absorb X-ray and to produce active oxygen species, may be promising as an X-ray sensitizer for a useful cancer-specific therapy.

References

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