Low-dose high-LET heavy ion-induced bystander signaling (II)

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Radiation-induced bystander response (RIBR) is a cellular response induced in nonirradiated cells that receive bystander signals from directly irradiated cells within an irradiated cell population.[1)] RIBR induced by low doses of high-LET radiations is an important issue concerning the health of astronauts and in heavy-ion radiation cancer therapy. Here, we investigated the underlying molecular mechanisms and biological implications of RIBR induced by such low doses of high-LET radiations.

The clonogenic cell survival of normal human fibroblast WI-38 cells irradiated with Ar ions (310 keV/μm) is shown in Fig. 1. At a higher dose region (0.5 Gy and above), the surviving fractions of cells harvested 16–24 h after irradiation was similar to those of cells harvested immediately (0 h) after irradiation [Fig. 1A]. On the other hand, a strong cell-killing effect at doses below 0.08 Gy was observed in the cells harvested 16–24 h after irradiation [Fig. 1B]. Such an effect was not observed in the cells harvested immediately after irradiation. Previously, we reported that cells irradiated with high-LET Fe ions (1000 keV/μm) showed similar results.[2)] These results suggest that an adequate incubation period is necessary for bystander signal induction and transfer.

Previously, we reported that gap-junction intercellular communication (GJIC), cyclooxygenase-2 (COX-2) protein, and nitric oxide (NO) were involved in high-LET Fe-ion-induced bystander signal transfer.[2)] Figure 2 shows the progress of results reflecting new data. Lindane and NS-398 (an inhibitor of GJIC and COX-2, respectively) were dissolved in DMSO (a scavenger of reactive oxygen species). c-PTIO is a scavenger of NO. DMSO (0.1%), lindane (Lin, 50 μM), c-PTIO (20 μM), or NS-398 (50 μM) was added to the medium 2 h before irradiation[3)] with 0.1 Gy of Fe ions (1000 keV/μm) [Fig. 2A] or 0.05 Gy of Ar ions (310 keV/μm) [Fig. 2B]. The obtained results for the cells irradiated with Fe and Ar ions were almost similar. DMSO did not significantly suppress the bystander cell killing. In contrast, lindane, NS-398, and c-PTIO significantly ($P < 0.05$) suppressed cell death to similar levels. Cells pretreated with both c-PTIO and lindane did not exhibit a significantly higher surviving fraction than those pretreated with lindane or c-PITO alone. These results suggest that bystander signaling through GJIC and the cell culture medium induces the bystander cell killing effect in a coordinated manner.

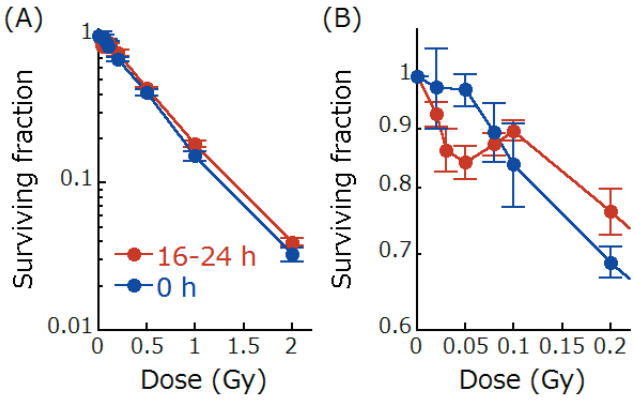

Fig. 1. Survival curves of WI-38 cells. Confluent monolayers of WI-38 cells were irradiated with 95 MeV/u Ar ions and the cells were harvested immediately (0 h) or 16–24 h after irradiation. The surviving fraction was determined by using a colony forming assay. Panel A shows all data obtained in this study. Panel B shows the surviving fractions at doses of 0.2 Gy and below. The error bars represent the standard errors of the mean (SEM) (n = 3–6).

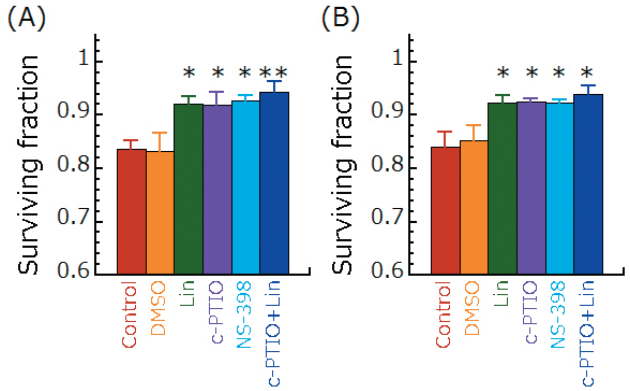

Fig. 2. Effect of inhibitors or scavengers. Panels A and B show the surviving fractions in the cells irradiated with 0.1 Gy of Fe ions and 0.05 Gy of Ar ions, respectively. $*P < 0.05$ and $**P < 0.01$, for comparison with control and drug-treated cultures.

References

1) M. Tomita and M. Maeda: J. Radiat. Res. **56**, 205 (2015).
2) M. Tomita et al.: RIKEN Accel. Prog. Rep. **48**, 302 (2015).
3) M. Tomita et al.: Radiat. Res. **179**, 200 (2013).

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