Phosphorylation and accumulation of low-dose high-LET heavy ion-induced bystander signaling molecules

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Radiation-induced bystander response (RIBR) is a cellular response induced in non-irradiated cells that received bystander signals from directly irradiated cells.[1)] RIBR induced by low doses of high-linear energy transfer (LET) radiation is an important issue for the health of astronauts and in hadrontherapy. Here, we investigated the underlying molecular mechanisms and biological implications of high-LET RIBR.

We found that normal human fibroblast WI-38, cultured confluent, irradiated with high-LET (1000 keV/μm) iron (Fe) ions showed the bystander cell killing effect at low doses ($\leq$ 0.2 Gy).[2)] In addition, we reported that gap-junction intercellular communication (GJIC), cyclooxygenase-2 (COX-2), and nitric oxide (NO) were involved in its signal transfer.[3)]

Figure 1 shows the phosphorylation and accumulation of bystander signaling related molecules in WI-38 cells irradiated with 0.1 Gy of Fe ions. Cells were pretreated with or without c-PTIO (a scavenger of NO) or lindane (an inhibitor of GJIC). Left panels were previously reported.[4)] The phosphorylation of Akt at Ser473 and NF-κB p65 at Ser536 and accumulation of COX-2 were observed in the cell 3 h after irradiation. They were efficiently inhibited by c-PTIO. Phosphorylated histone H2AX at Ser139 (γ-H2AX) is widely used as a surrogate marker of DNA double-strand breaks (DSBs). γ-H2AX was observed 3 and 6 h after irradiation and the one at 6 h was inhibited by c-PTIO. These results suggest that phosphorylation of NF-κB, Akt, and histone H2AX and accumulation of COX-2 were mainly mediated by NO. Meanwhile, phosphorylation of Akt at 3 h after irradiation was also inhibited by linedane but that of NF-κB and γ-H2AX was not (right panels). Surprisingly, COX-2 was overexpressed in the cells pretreated with lindane irrelevantly to irradiation. It is well known that COX-2 overexpressed cancer cells are resistant to the induction of apoptosis.[5)] The ability of lindane to inhibit GJIC is apparent,[1)] whereas COX-2 overexpression in addition to inhibition of Akt may also contribute to the inhibition of bystander cell killing, even if DSBs are induced in the bystander cells.

Continued studies must elucidate the role of GJIC. The obtained results suggested that NF-κB, Akt, and COX-2 were involved in the low-dose of high-LET heavy-ion-induced bystander signaling.

References

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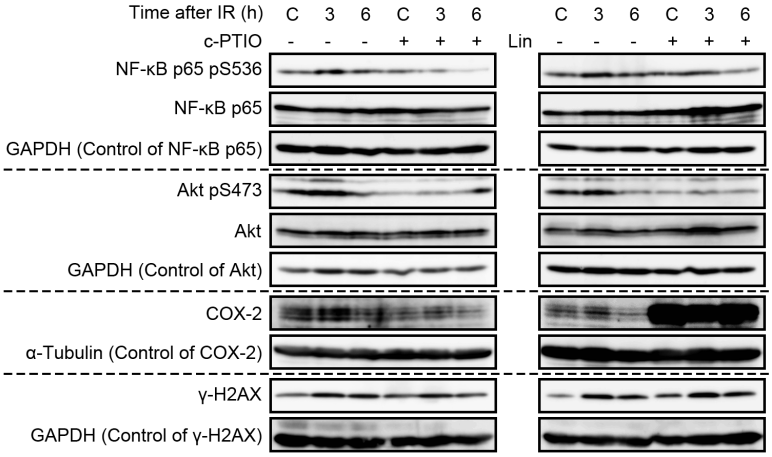

Fig. 1. Phosphorylation and accumulation of bystander signaling related molecules. WI-38 cells were pretreated with or without c-PTIO (20 μM) or lindage (Lin, 50 μM) 2 h before irradiation with 0.1 Gy of 90 MeV/nucleon Fe ions (1000 keV/μm). Cells were harvested 3 and 6 h after irradiation followed by immunoblotting.

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