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Effects of *N*-acetyl-L-cysteine (NAC) on Radiation-induced Cytotoxicity in Fish Hepatoma Cells

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Introduction

N-acetyl-L-cysteine (NAC) is one of well-known sulfhydryl-containing antioxidants, whose role in radioprotection has been explored in several studies [1-3]. The antioxidant property of NAC can be attributed to its ability to provide cysteine and other precursors of glutathione synthesis, as well as its ability to directly scaveng free radicals [4]. With regard to the radioprotective effects of NAC, the majority of studies have been performed *in vitro* [5-6]. NAC were used to protect the CHO (Chinese hamster ovary) cells from radiation-induced apoptosis by controlling the enzyme that triggers programmed cell death [7].

This study was designed to evaluate the radioprotective effects of NAC in different doses on the cell viability in fish cell line induced by ionizing radiation.

Materials and Methods

Cells and culture conditions

PLHC-1 cells were derived from a hepatocellular carcinoma in an adult female *(Poeciliopsis Lucida)*, a topminnow from the Sonoran desert (ATCC®#CRL-2406). PLHC-1 cells were cultured in EMEM suplemented with 5% fetal bovine serum (FBS) and incubated at 30℃ in a humidified atmosphere with 5% CO_2.

Irradiation and NAC treatments of cells

PLHC-1 cells were seeded in 96-well plates at a density of 2 x 10^5 cells/㎖ and incubated for 24 hr. Then cells were treated γ-rays from a ^{60}Co γ-ray source (Korea Atomic Energy Research Institute, Korea) with 50~300 Gy in the presence or absence of 0.05~1.25 mM of NAC which were added 1 hr before.

Cell viability assay

To assess cell viability, 10㎕ of MTT solution was added to each well after removal of 100㎕ supernatant and incubated for another 4 hr at 30℃. The generated formazan crystal was dissolved and the absorbance was detected at 570nm using ELISA reader (Multiskan®EX, Forma Scientific, Inc.).

Results and Discussion

The cell viability of NAC pretreated groups was higher than that of 50 and 100 Gy radiation-

treated groups without NAC. The results showed that NAC prevented cells from radiation-induced death, but it caused cytotoxicity in 300 Gy radiation-treated groups as its concentration increases. While NAC has been used as radioprotective agent in lower concentrations it is cytotoxic at high concentration. The radioprotective effect of NAC was assessed after exposure to $HgCl_2$ combined with 4-nonylphenol [8]. The result showed that cell death was prevented in the group pretreated with NAC . The potential utility of NAC in lower concentrations as a protector against radiation is worth considering here.

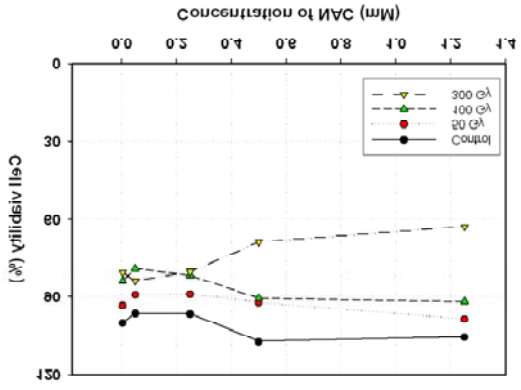

Fig. 1. Protective effects of NAC in radiation-induced cytotoxicity.

Conclusion

Protective effects of NAC were assessed using PLHC-1 cells, irradiated with 50~300 Gy. The data showed that NAC in lower concentrations prevented cell death after irradiation. Theses results suggest that NAC may be able to attenuate radiation-induced cytotoxicity. Also, this investigation give a clue for fundamental theories associated with positive efficacy of radiation applications including radioprotection.

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