Role of poly(ADP-ribosyl)ation in the regulation of DNA damage-induced cell death and inflammatory reactions of the skin

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SUMMARY

MNNG is an alkylating agent, which induces enhanced poly(ADP-ribosyl)ation in different cell types. In the first part of our work we have investigated the mechanism of the PARP-activating and citotoxic effects of MNNG. After MNNG treatment, DNA breakage, increased PARP activity and cytotoxicity could be observed. These effects have been abolished by pretreatment of the cells with thiol antioxidants (glutathione or *N*-acetylcysteine). Inhibition of PARP decreased necrosis but apoptotic parameters increased simultaneously. In aqueous solution of MNNG we found nitrite/nitrate production. Moreover, in MNNG-treated thymocytes intense protein tyrosine nitration could be observed. As MNNG-induced cytotoxicity in thymocytes showed many similarities to peroxynitrite-induced cell death, therefore we have investigated whether peroxynitrite was responsible for the cytotoxicity induced by MNNG. We found that neither NO nor peroxynitrite contributed to MNNG-induced cytotoxicity.

Although PARP-1 is activated mainly by DNA damage, alternative phosphorylation-dependent activation has also been described. It is known, that protein kinase C (PKC) is able to phosphorylate PARP-1 *in vitro*. In our work, we showed PKC-mediated phosphorylation also takes place in a cellular setting leading to inhibition of PARP-1 activity and to reduced cytotoxicity. Cytoprotective effect of PKC-activating phorbol ester was reversed by PKC inhibitors. Our results demonstrate that MNNG-induced DNA breakage leads to PARP activation and PARP-dependent cytotoxicity. PKC inhibits PARP activation by phosphorylation, and thus exerts a cytoprotective effect.

Contact hypersensitivity (CHS) is a form of delayed type of hypersensitivity. Production of proinflammatory cytokines, cellular infiltration by lymphocytes and granulocytes are important events of this process. Infiltration is accompanied by strong oxidative stress. Inhibition of PARP activity or knocking out the PARP-1 gene has been shown to suppress inflammatory reactions. We have investigated the role of PARP-1 in the mouse model of oxazolone-induced CHS. Inhibition of PARP decreased edema, the number of infiltrating cells, induction of proinflammatory cytokines and activity of matrix metalloproteinases. The PARP inhibitor had similar but milder effect in irritant dermatitis indicating that PARP modulates both the antigen-specific immune response and the general inflammatory pathways.

Kulcsszavak: DNS károsodás, poli-ADP-riboziláció, protein kináz C, kontakt hiperszenzitivitás

Keywords: DNA damage, poly(ADP-ribosyl)ation, protein kinase C, contact hypersensitivity