

Pyrrolidine dithiocarbamate augments Hg²⁺ in inducing macrophage cell death through oxidative stress-induced apoptosis and necrosis signaling pathways

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Abstract:

Exposure to mercury causes several types of injuries in mammals, including immune system dysfunction. In addition, environmental factors may enhance the cytotoxic effects of mercury. Thus, it is important to understand and explore the possible toxic mechanisms of mercury combined with environmental factors. Here, we demonstrated that pyrrolidine dithiocarbamate(PDTC), a commonly used antioxidant and metal chelator, augmented HgCl₂-induced cytotoxic effects by facilitating mercury entry into the cultured murine macrophage (RAW 264.7 cells). The Hg²⁺/PDTC complex significantly and rapidly increased ROS formation and decreased intracellular GSH levels in cells. By means of a flow cytometric technique, the number of sub-G1 hypodiploids and Annexin V-FITC binding cells were found to be increased after Hg²⁺/PDTC complex exposure. Several features of mitochondrial-dependent apoptosis were also induced, including mitochondrial dysfunction, activations of poly (ADP-ribose) polymerase (PARP) and caspase 3/7,

and DNA fragmentation. Moreover, both apoptotic and necrotic cells were detected by acridine orange/ethidium bromide dual staining. Meanwhile, depletion of intracellular ATP levels and increased LDH release were observed, suggesting the induction of necrotic cell death processes. All of these Hg²⁺/PDTC complex-induced cytotoxic-related signals could be reversed by pre-treated with an antioxidant, *N*-acetylcysteine. In conclusion, the results obtained suggest that Hg²⁺/PDTC complex-induced oxidative stress caused macrophage cell death via a mixed type of apoptosis and necrosis. These findings imply for first that PDTC may enhance the uptake of Hg²⁺ and dramatically enhance the toxicological effects of Hg²⁺ instead of detoxification.

Keywords: HgCl₂; PDTC; apoptosis; necrosis; oxidative stress; caspase 3/7