Modulation of unfolded protein response by methylmercury (MeHg)

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[Background]

MeHg known as a causative factor of Minamata disease induces neuronal cell death and damage the central nervous system, but the mechanism is poorly understood. Previously, we reported that MeHg induces S-mercuration at cysteine 383 or 386 in protein disulfide isomerase (PDI), and this modification induces the loss of enzymatic activity. Because PDI is a key enzyme for the maturation of nascent protein harboring a disulfide bond, the disruption in PDI function by MeHg results in endoplasmic reticulum (ER) stress via the accumulation of misfolded proteins. However, the effects of MeHg on unfolded protein response (UPR) sensors remain unclear. In the present study, we examined whether UPR is regulated by MeHg.

[Methods]

To investigate the activations of UPR sensors and downstream signaling by MeHg stimulation, we used western blot analysis and RT-PCR. To measure apoptosis cells induced by MeHg, we assessed the chromosomal condensation using the fluorescent dye Hoechst 33342.

[Results]

We found that MeHg activates protein kinase RNA-like endoplasmic reticulum kinase (PERK) and activating transcriptional factor 6 (ATF6) branches. Although phosphorylated inositol-requiring enzyme 1 α (IRE1 α) was detected, MeHg did not induce the cytosolic splicing of immature x-box binding protein 1 (XBP1) mRNA (a selective substrate of IRE1 α). Then, the IRE1 α -null MEFs were transfected with vectors encoding wild-type hemagglutinin (HA)-tagged IRE1 α or cysteine mutants. MeHg-induced inhibition of IRE1 α endonuclease activity was significantly ameliorated in the MEFs that expressed IRE1 α (C931S). These results suggested that C931 in IRE1 α could be a predominant target of MeHg. A previous study has shown that the IRE1 α -XBP1 branch functions as an anti-apoptotic pathway. In contrast, the PERK/ATF6 branches are involved in the induction of cell death. Therefore, these signals may be implicated in the MeHg-induced loss of cell viability. Indeed, treatment with GSK2606414, a specific PERK inhibitor, significantly attenuated MeHg-induced cell death.

[Conclusions]

We demonstrated that MeHg disrupts anti-apoptotic signaling based on the IRE1 α -XBP1 branch and simultaneously promotes pro-apoptotic signaling via the PERK/ATF6 branches. These findings may be utilized in the development of novel therapeutic approaches for Minamata disease.