

The role of HIF-1 α -BNIP3 pathway in acrylonitrile-induced hippocampal neuronal cell toxicity

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Introduction :Acrylonitrile (AN) is an important chemical raw material, and is widely used in the production of fibers, plastics and resins. Exposure to AN occurs in occupational settings. While exposure to AN can cause neurotoxicity in bioorganisms, the exact mechanism remains unclear. Hypoxia-inducible factor (HIF) is an important transcription factor produced under hypoxia. During hypoxia, cytoplasm HIF-1 α binds to stably expressed HIF-1 β and then is translocated into the nucleus to bind with the hypoxia-inducible element (HRE). Multiple downstream target genes are induced.

Methods: CCK assay, LDH release rate, flow cytometry, and Western blot were used to detect the cell viability, toxicity, apoptosis and expression of related proteins in hippocampal neuronal HT22 cells after exposure to AN. Then, cells were pretreated with HIF-1 α activator cobalt chloride (CoCl₂) and BNIP3 overexpression, followed by detection of relevant indicators

Results: We found that the viability of HT22 cells treated with various concentrations of AN (0, 1, 2.5, 5mM) was reduced, but LDH release rate, apoptosis, and ROS were significantly enhanced. The results of Western blotting showed that the expression of HIF-1 α and its downstream proteins, including glucose transporter 1, erythropoietin, Bnip3, and NIX were decreased. In addition, by HIF-1 α CoCl₂ pretreatment, cell viability and expression of HIF-1 α and downstream proteins were increased. CoCl₂ increased the expression of BNIP3, TOM20, BCL2, and decreased the expression of pro-apoptosis protein BAX. Besides, HIF-1 α inhibitor dimethoxyestradiol (2MEOE2) showed opposite results. Furthermore, we found that the cell viability was significantly increased after pretreatment with BNIP3 overexpression, ATP content increased, and BNIP3 and LC3 showed co-localization in confocal microscopy. While the expression of TOM20 decreased, the expression of LC3B and Beclin1 increased significantly, indicating that BNIP3 mitophagy may be involved in the process.

Conclusion: The HIF-1 α -BNIP3 pathway may participate in the neurotoxic mechanism of AN, which providing effective treatment strategy for clinical AN poisoning.

Key words: acrylonitrile; neurotoxicity; HT22; HIF-1 α ; BNIP3; mitophagy