INDUCTION OF APOPTOSIS IN ARSENIC TRIOXIDE-TREATED LUNG CARCINOMA (A549) CELLS

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Abstract: Arsenic and its compounds alter numerous cellular processes such as growth inhibition, apoptosis and cell signaling. Arsenic trioxide has been found to induce apoptosis in leukemia cell lines and clinical remissions in patients with acute promyelocytic leukemia. In this study, we investigated the apoptotic and proliferative effects of arsenic trioxide in lung carcinoma cells. We also studied the effect of arsenic trioxide on the signal transduction pathway in the lung carcinoma cells. To achieve this goal, the lung cancer (A549) cells were cultured following standard protocols, and exposed to various doses (0, 2, 4, 6, 8, and 10 µg/ml) of arsenic trioxide for 48 h. The proliferative response (DNA synthesis) to arsenic trioxide was determined by [³H] thymidine incorporation assay. Arsenic trioxide-induced apoptosis was determined by DNA laddering pattern and caspase-3 activity. DNA laddering was ascertained by the apoptotic DNA laddering assay. Caspase-3 activation was assessed by the caspase-3 fluorescein isothiocyanate (FITC) assay. The p38 MAP kinase activity was examined by immunoblot analysis using phospho p38 MAPK mab primary antibody in the presence of ATP and transcription factor (ATF-2) as a substrate. The [³H] thymidine incorporation assay revealed a dose-related cytotoxic response at higher levels of exposure in A549 cells. The LC₅₀ was found to be 7.8 µg/ml. The results from the DNA laddering assay revealed that arsenic trioxide induced apoptosis in the lung carcinoma cells. We demonstrated that arsenic trioxide stimulated caspase-3 and p38 map kinase activation in A549 cells. In conclusion, the activation of caspase-3 and p38 map kinase and the laddering pattern are cellular markers to validate that arsenic trioxide induces apoptosis in lung carcinoma cells.

Keywords: Arsenic trioxide, lung carcinoma (A549) cells, apoptotic DNA laddering assay, caspase-3 activation, p38 MAP kinase

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