IN VITRO INDICES OF OXIDATIVE STRESS IN LEAD-EXPOSED HEPG₂ CELLS

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Abstract: Lead is a ubiquitous metal that has been used by humans for more than 3 millennia. Its toxic effects on humans are well documented in history. Most research on lead has focused on its effects on organ systems such as the nervous system, the red blood cells, and the kidneys which are considered to be the primary targets of lead toxicity. However, its molecular mechanisms of toxicity are still largely unknown. In this research, we used HepG₂ cells as a model to study the cytotoxicity and oxidative stress associated with exposure to lead nitrate. We hypothesized that oxidative stress plays a key role in lead nitrate induced cytotoxicity. To test this hypothesis, we performed both MTT [3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide] assay and trypan blue exclusion test for cell viability and the thiobarbituric acid test for lipid peroxidation. Data obtained from the MTT assay indicated that lead nitrate significantly reduced the viability of HepG₂ cells, showing a LD₅₀ value of about 28 μg/mL upon 48 hours of exposure, indicating a dose-dependent response. Similar trend was obtained with the trypan blue exclusion test using the hemacytometer to count the cell manually. Data generated from the thiobarbituric acid test showed a significant (p ≤ 0.05) increase in MDA levels in lead nitrate-treated HepG₂ cells compared to control cells. Lead nitrate treatment significantly increased cellular content of reactive oxygen species (ROS), as evidenced by the increase in lipid peroxidation by-products. Taken together, these results indicate that lead nitrate is highly cytotoxic to HepG₂ cells. This cytotoxicity is found to be mediated by oxidative stress, a biomarker of cellular injury.

Keywords: Lead nitrate, lipid peroxidation, MDA, cytotoxicity, and HepG₂ cells

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