

Expression Profiling of Butadiene Diepoxide induce Gene in Androgen Positive and Androgen Negative Prostate Cell lines

*Wesley G. Gray*¹, PI and Shulin Li², Mentor

Chemistry and Environmental Toxicology Southern University, Baton Rouge¹

School of Veterinary Medicine, Louisiana State University, Baton Rouge²

Abstract: Animal and human population studies support the reproductive carcinogenicity of Butadiene and its metabolite BDO₂. However, the toxicity and carcinogenicity of BDO₂ on reproductive tissue such as the prostate or prostate cell culture has not been fully investigated. In the current study we determined whether BDO₂-induced apoptosis involved modulation of Bcl2 (anti apoptotic protein) or Bax (pro apoptotic) expression levels at both the protein and mRNA levels. We subjected LNCaP cells to increasing concentrations of BDO₂ (0.1nM to 100nM) for periods of 6, 12 and 24h. The effect of BDO₂ was evaluated by measuring cellular cytotoxicity using MTT assay, morphological signs of apoptosis using Fluorescent microscopy, prostate cell integrity using PSA secretion by ELISA and gene expression of apoptotic proteins using Western blot analysis and QRT-PCR. The results of MTT assay indicate that BDO₂ induced toxicity with GI₅₀ greater than 100nM. Following 10nM exposure, PSA secretion, was increased by 9% at 6hr, decreased by 32% at 12h and increased by 3.6% at 24h. Fluorescent microscopic examination revealed signs of BDO₂-induced apoptosis at concentrations as low as 10nM at 6hr. Western blot analysis of Bcl2 and Bax expression revealed a 0.5-fold dose-dependent decrease as compared to control. On the other hand, mRNA expression of these proteins decreased 1-1.5-fold at BDO₂ concentrations greater than 10nM. We also observed a corresponding increase in caspase levels which was both time and dose dependent. These results of our study suggest that, the cell death induced by BDO₂ in LNCaP cells may be associated with alteration of three key cellular proteins Bcl2, Bax and Caspases in mediating cytotoxicity and may regulate cellular and molecular processes causing cell death by apoptosis.