

ABSTRACT

Background & Aims: Oxidative stress and apoptosis are proposed mechanisms of cellular injury in studies of xenobiotic hepatotoxicity. The aim of this work is to find early signal markers of drug-induced injury of the liver by focusing on select antioxidant/oxidant and apoptotic genes. As well, to address the relationship between conventional liver dysfunction markers and the measured mRNA and protein expressions in the D-galactosamine/lipopolysaccharide and tert-butylhydroperoxide hepatotoxicity models. Furthermore, potential hepatoprotective capabilities of antioxidant polyphenols quercetin and curcumin were evaluated in relation to its modulation of the oxidative stress and apoptotic parameters in the given xenobiotic hepatotoxicity models.

Methods: Biochemical markers testing the hepatic function included aminotransferases (ALT, AST) and bilirubin. Measurements of TBARS and conjugated dienes were used to assess lipoperoxidation. Plasma levels of catalase and reduced glutathione were used as indicators of the oxidative status of the cell. Real time PCR was used to analyse the mRNA expressions of the inducible nitric oxide synthase (NOS-2), heme oxygenase-1 (HO-1), superoxide dismutase (SOD-1), glutathione peroxidase (Gpx-1), caspase 3 (Casp3), BH3 interacting domain death agonist (Bid) and Bcl-2 -associated X protein (Bax), and tumor necrosis factor α (TNF- α)mRNAs. Additionally, the protein expressions of HO-1 and NOS-2 were assessed with the use of Western blot method. Morphometric evaluation of hepatocytes at the light microscopical level was done on semithin epon sections stained by toluidine blue using Leica IM 500 program for digital recording and measurements. Statistical analysis was performed using ANOVA and post hoc Bonferroni, Tukey- Kramer comparison test or unpaired T-test with Welch correction.

Results: Overall, the results of this study have revealed the early activation of oxidative stress and apoptosis in the given hepatotoxic models as seen by the relevant changes in the tested parameters. Hepatoprotective effects of curcumin and quercetin were demonstrated, where the induction of the antioxidant enzyme HO-1 and its products played the most important cytoprotective role. In case with curcumin, this effect was paralleled with the concomitant reduction of NOS-2 and TNF- α expressions.

Conclusion: Understanding the mutual regulatory mechanisms of the tested parameters in hepatocyte injury should provide important clues to the diagnosis and treatment of liver damage. The research data from the present study paves a way for those interested in further research of these dietary polyphenols, curcumin and quercetin as it gives an overview of the potential cytoprotective mechanisms and effective doses in the given models.

Key words: liver, xenobiotic hepatotoxicity, D-galactosamine, lipopolysaccharide, tertbutylhydroperoxide, hepatoprotection, curcumin, quercetin, heme oxygenase 1, nitric oxide synthase 2, catalase, superoxide dismutase 1, glutathione peroxidase, tumor necrosis factor-alpha, Bid, Bax, caspase 3.