Mol Cell Biochem (2013) 384:59–69
DOI 10.1007/s11010-013-1781-9

Rotenone-induced oxidative stress and apoptosis in human liver HepG2 cells

M. A. Siddiqui · J. Ahmad · N. N. Farshori · Q. Saquib · S. Jahan · M. P. Kashyap · M. Ahamed · J. Musarrat · A. A. Al-Khedhairy

Received: 9 June 2013 / Accepted: 9 August 2013 / Published online: 21 August 2013

Abstract Rotenone, a commonly used pesticide, is well documented to induce selective degeneration in dopaminergic neurons and motor dysfunction. Such rotenone-induced neurodegenration has been primarily suggested through mitochondria-mediated apoptosis and reactive oxygen species (ROS) generation. But the status of rotenone induced changes in liver, the major metabolic site is poorly investigated. Thus, the present investigation was aimed to study the oxidative stress-induced cytotoxicity and apoptotic cell death in human liver cells-HepG2 receiving experimental exposure of rotenone (12.5–250 μM) for 24 h. Rotenone depicted a dose-dependent cytotoxic response in HepG2 cells. These cytotoxic responses were in concurrence with the markers associated with oxidative stress such as an increase in ROS generation and lipid peroxidation as well as a decrease in the glutathione, catalase, and superoxide dismutase levels. The decrease in mitochondrial membrane potential also confirms the impaired mitochondrial activity. The events of cytotoxicity and oxidative stress were found to be associated with up-regulation in the expressions (mRNA and protein) of pro-apoptotic markers viz., p53, Bax, and caspase-3, and down-regulation of anti-apoptotic marker Bcl-2. The data obtain in this study indicate that rotenone-induced cytotoxicity in HepG2 cells via ROS-induced oxidative stress and mitochondria-mediated apoptosis involving p53, Bax/Bcl-2, and caspase-3.

Keywords HepG2 cells · Rotenone · Cytotoxicity · Oxidative stress · Apoptosis

M. A. Siddiqui (✉) · J. Ahmad · Q. Saquib · J. Musarrat · A. A. Al-Khedhairy
Department of Zoology, College of Science, King Saud University, P.O. Box 2455, Riyadh 11451, Saudi Arabia
e-mail: maqsoodahmads@gmail.com; masiddiqui@ksu.edu.sa

N. N. Farshori
Department of Pharmacognosy, College of Pharmacy, King Saud University, Riyadh, Saudi Arabia

S. Jahan · M. P. Kashyap
In Vitro Toxicology Laboratory, CSIR-Indian Institute of Toxicology Research, Lucknow, India

M. Ahamed
King Abdullah Institute for Nanotechnology, King Saud University, Riyadh, Saudi Arabia

Introduction

Rotenone is a plant-derived pesticide, commonly used to protect crops from pests across the world since a long time [1]. It is a neurotoxicant that specifically inhibits the flow of electrons through the mitochondrial respiratory chain by binding with mitochondrial complex-I [2–4], and is known to cross blood brain barrier and gets accumulated in sub cellular organelles [5]. Rotenone is also well known to inhibit the biochemical processes at the cellular level, and can block electron transfer from complex to ubiquinone, resulting blocking of the oxidative phosphorylation, and an increase in reactive oxygen species (ROS) generation [1, 6]. A number of studies have evaluated the effects of rotenone under both in vitro [7] and in vivo conditions [1]. Studies have shown that rotenone is capable to induce apoptosis in various cells derived from human B cell lymphomas [8], promyelocytic leukemias [9], and neuroblastomas [10, 11]. Our group has also reported that rotenone induces oxidative stress-mediated cytotoxicity in rat pheochromocytoma cells-PC12 cell line [12], and in human breast adenocarcinoma-MCF-7 cell line [13]. Rotenone-induced mitochondrial complex-I inhibition results in generation of free radicals and thereby