

Available online at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevier.com/locate/etap

Royal jelly attenuates azathioprine induced toxicity in rats

Walaa M.S. Ahmed^{a,*}, A.A. Khalaf^b,
Walaa A. Moselhy^c, Ghada M. Safwat^d

^a Department of Clinical Pathology, Faculty of Veterinary Medicine, Beni-Suef University, Egypt

^b Department of Toxicology & Forensic Medicine, Faculty of Veterinary Medicine, Cairo University, Egypt

^c Department of Toxicology & Forensic Medicine, Faculty of Veterinary Medicine, Beni-Suef University, Egypt

^d Department of Biochemistry, Faculty of Veterinary Medicine, Beni-Suef University, Egypt

ARTICLE INFO

Article history:

Received 30 May 2013

Received in revised form

14 December 2013

Accepted 20 December 2013

Available online 31 December 2013

Keywords:

Azathioprine

Hepatotoxicity

Royal jelly

Oxidative stress

ABSTRACT

In the present study, we investigated the potential protective effects of royal jelly against azathioprine-induced toxicity in rat. Intraperitoneal administration of azathioprine (50 mg/kg B.W.) induced a significant decrease in RBCs count, Hb concentration, PCV%, WBCs count, differential count and platelet count, hepatic antioxidant enzymes (reduced glutathione and glutathione S-transferase) and increase of serum transaminases (alanine aminotransferase and aspartate aminotransferase enzymes) activities, alkaline phosphatase and malondialdehyde formation. Azathioprine induced hepatotoxicity was reflected by marked pathological changes in the liver. Oral administration of royal jelly (200 mg/kg B.W.) was efficient in counteracting azathioprine toxicity whereas it altered the anemic condition, leucopenia and thrombocytopenia induced by azathioprine. Furthermore, royal jelly exerted significant protection against liver damage induced by azathioprine through reduction of the elevated activities of serum hepatic enzymes. Moreover, royal jelly blocked azathioprine-induced lipid peroxidation through decreasing the malondialdehyde formation. In conclusion, royal jelly possesses a capability to attenuate azathioprine-induced toxicity.

© 2013 Elsevier B.V. All rights reserved.

1. Introduction

Azathioprine (AZA) has been registered in the Netherlands since 1968 as immunosuppressive drug that used in the treatment of hematological malignancies, solid organ transplantation and inflammatory bowel disease (Maltzman and Koretzky, 2003). It is also used for the management of severe, active and erosive rheumatoid arthritis which is not responding to conventional medications (Raza et al., 2003). It is also indicated in cardiac, hepatic and pancreatic transplantations (Heneghan and McFrlane, 2002). However, its use has been

concomitant with a high incidence of serious adverse drug reaction such as myelosuppression, pancreatitis and gastrointestinal disturbances (Rulyak et al., 2003; Sinico et al., 2003) with high incidence of hepatotoxicity (Petit et al., 2008). The hepatotoxicity of azathioprine has been well documented in rats and involves an elevation of reactive oxygen species (ROS) and depletion of reduced glutathione (GSH) which leading to mitochondrial injury and cell death due to necrosis (Lee and Farrell, 2001; Raza et al., 2003). The first step in the biotransformation of azathioprine involves conjugation with reduced glutathione to form 6-mercapotpurin catalyzed by glutathione S-transferase presumably in the liver (Hobara and Watanabe,

* Corresponding author. Tel.: +20 822317586; fax: +20 822327982.

E-mail address: Walaa.msa@yahoo.com (W.M.S. Ahmed).

1382-6689/\$ – see front matter © 2013 Elsevier B.V. All rights reserved.

<http://dx.doi.org/10.1016/j.etap.2013.12.010>

1981). 6-MP is further metabolized to the pharmacologically active 6-thioguanine nucleotides which is responsible for the cytotoxicity (Armstrong and Oellerich, 2001).

Some natural products as royal jelly (RJ) were reported to have a protective role against oxidative damages due to its antioxidant potency and free radical scavenging capacity (Cemek et al., 2010; Silici et al., 2010). Royal jelly is a secretion produced by the hypopharyngeal and mandibular glands of worker honey bees (*Apis mellifera*). It contains many important compounds with biological activity such as free amino acids, proteins, sugars, fatty acids, minerals and vitamins (Nakajina et al., 2009). Royal jelly has been demonstrated to possess antimicrobial (Chan et al., 2009), anti-allergic (Oka et al., 2001), anti-inflammatory (Kohno et al., 2004), immunomodulatory (Simsek et al., 2009) and antioxidant properties (Nakajina et al., 2009).

The present study was conducted to investigate the protective effect of royal jelly administration on rats intoxicated with azathioprine induced hematological, biochemical and histopathological alterations.

2. Material and methods

2.1. Chemicals

Azathioprine (Imuran®) and Royal jelly were obtained from GlaxoSmithKline Company, Egypt. Alanine aminotransferase enzyme (ALT), aspartate aminotransferase enzyme (AST), alkaline phosphatase enzyme (ALP), reduced glutathione (GSH), glutathione s-transferase (G-S transferase) and malondialdehyde (MDA) were estimated using commercial kits (Bio-diagnostic for Research Kits, Egypt). All the other chemicals were of the highest analytical grade and purchased from Sigma-Aldrich Company.

2.2. Animals and treatments

Thirty adult male Wistar Albino rats, weighting 150–200 g were obtained from the Egyptian Organization for Biological Products and Vaccine. They were housed 5 rats per cage with free access to commercial diet and tap water. All animal studies were conducted in accordance with the criteria of the investigations and Ethics Committee of the Community Laws governing the use of experimental animals. After one week of acclimatization, rats were randomly divided into three equal groups of 10 rats each. The group I was kept as a control while group II was injected intraperitoneally with 50 mg of AZA as a single dose. Group III received RJ (200 mg/kg B.W.) by oral gavage for 7 consecutive days pre and 15 consecutive days post intraperitoneal injection of 50 mg/kg AZA.

2.3. Collection and processing of samples

The samples were collected at 24 h according to “Raza et al., 2003; Tabrizi et al., 2009; El-Ashmawy et al., 2010” and 15 days after the last administration of azathioprine. Blood samples were collected from orbital venous plexus under light anesthesia (5 animals from each group). Each sample was divided into two portions; the first was collected in clean

dry Eppendorf tubes containing EDTA (1–2 mg/ml blood) as anticoagulant to be used for hemogram studies. The second part was collected into non heparinized tubes which were centrifuged at 3000 rpm for 10 min for separation of serum. The collected sera were stored at -20°C for biochemical estimation. The rats then were euthanized by cervical dislocation and their liver tissue was excised and part of it was fixed in 10% neutral formalin to be used for histopathological studies. The other part (one gram) was washed using cold saline solution, homogenized in 5 ml of phosphate buffer saline (pH 7.4) and then cold centrifuged at 3000 rpm for 15 min. The obtained supernatant was collected and stored at -20°C for estimation of GSH, G-S transferase and MDA.

2.4. Complete blood count

Total number of erythrocytes (RBCs), total number of leukocytes (WBCs), differential leukocyte count, platelet count, packed cell volume (PCV)%, and hemoglobin (Hb) concentration were estimated by adopting standard procedures. Blood smears were prepared as soon as possible after blood collection on a glass slide and quickly dried and stained with Giemsa and May-Grunwald stain for the differential blood count. Erythrocyte indices like mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were calculated according to standard formulas.

$$\text{MCV (fl)} = \frac{\text{PCV(\%)} \times 10}{\text{RBC count in millions/mm}^3}$$

$$\text{MCH (pg)} = \frac{\text{Hb (g/dl)} \times 10}{\text{RBC count in millions/mm}^3}$$

$$\text{MCHC (g/dl)} = \frac{\text{Hb (g/dl)}}{\text{PCV (\%)}} \times 100$$

2.5. Biochemical parameters

The activities of ALT and AST enzymes were estimated with colorimetric method (Reitman and Franel, 1975). ALP activity was measured according to Belfield and Goldberg (1971).

2.6. Liver antioxidant enzymes and malondialdehyde

The liver tissue homogenate was used for spectrophotometric estimation of GSH and G-S transferase enzymes according to Beutler et al. (1963) and Habing et al. (1974) respectively. The extent of lipid peroxidation was evaluated through monitoring the level of MDA as described by Ohkawa et al. (1979).

2.7. Histopathological examination

Histopathological examination of liver tissue of control and treated groups were performed according to Banchroft et al. (1996).

Download English Version:

<https://daneshyari.com/en/article/2584015>

Download Persian Version:

<https://daneshyari.com/article/2584015>

[Daneshyari.com](https://daneshyari.com)