Available online at www.sciencedirect.com

ScienceDirect

journal homepage: www.jfda-online.com

Original Article

Effect of goat milk on hepatotoxicity induced by antitubercular drugs in rats

Sonam Miglani [a], Rakesh Raman Patyar [b], Sazal Patyar [a,*], Mohammad Rafi Reshi [a]

[a] Department of Pharmacology, School of Pharmaceutical Sciences, Lovely Professional University, Phagwara, India
[b] Postgraduate Institute of Medical Education and Research, Chandigarh, India

ARTICLE INFO

Article history:
Received 23 June 2015
Received in revised form
22 March 2016
Accepted 25 March 2016
Available online 2 June 2016

Keywords:
antitubercular drug-induced hepatotoxicity
goat milk
hepatotoxicity
silymarin

ABSTRACT

Aim of the present study was to assess the hepatoprotective activity of goat milk on antitubercular drug-induced hepatotoxicity in rats. Hepatotoxicity was induced in rats using a combination of isoniazid, rifampicin, and pyrazinamide given orally as a suspension for 30 days. Treatment groups received goat milk along with antitubercular drugs. Liver damage was assessed using biochemical and histological parameters. Administration of goat milk (20 mL/kg) along with antitubercular drugs (Group III) reversed the levels of serum alanine aminotransferase (82 ± 25.1 vs. 128.8 ± 8.9 units/L) and aspartate aminotransferase (174.7 ± 31.5 vs. 296.4 ± 56.4 units/L, $p < 0.01$) compared with antitubercular drug treatment Group II. There was a significant decrease in serum alanine aminotransferase (41.8 ± 4.1 vs. 128.8 ± 8.9 units/L, $p < 0.01$) and aspartate aminotransferase (128.8 ± 8.54 vs. 296.4 ± 56.4 units/L, $p < 0.001$) levels in Group IV (goat milk 40 mL/kg) compared with antitubercular drug treatment Group II. Goat milk (20 mL/kg and 40 mL/kg) was effective in reversing the rise in malondialdehyde level compared with the antitubercular drug suspension groups (58.5 ± 2 vs. 89.88 ± 2.42 μmol/mL of tissue homogenate, $p < 0.001$ and 69.7 ± 0.78 vs. 89.88 ± 2.42 μmol/mL of tissue homogenate, $p < 0.001$, respectively). Similarly, both doses of milk significantly prevented a fall in superoxide dismutase level (6.23 ± 0.29 vs. 3.1 ± 0.288 units/mL, $p < 0.001$ and 7.8 ± 0.392 vs. 3.1 ± 0.288 units/mL, $p < 0.001$) compared with the group receiving antitubercular drugs alone. Histological examination indicated that goat milk reduced inflammation and necrotic changes in hepatocytes in the treatment groups. The results indicated that goat milk prevented the antitubercular drug-induced hepatotoxicity and is an effective hepatoprotective agent.

*Corresponding author. Department of Pharmacology, School of Pharmaceutical Sciences, Lovely Professional University, Jalandhar- Delhi G.T. Road (NH-1) Phagwara, Punjab, 144402, India. Tel.: + 91 9464859178 (Mobile).
E-mail address: sazalpatyar@yahoo.co.in (S. Patyar).

http://dx.doi.org/10.1016/j.jfda.2016.03.012

1. Introduction

Antitubercular drug-induced hepatotoxicity is one of the most challenging clinical problems. It is the main cause of interruption during a tuberculosis treatment course and may lead to hospitalization or life threatening events [1,2]. Antitubercular drug-induced hepatotoxicity has a wide spectrum of presentations, ranging from an asymptomatic mild rise in liver biochemical tests to acute hepatitis and acute liver failure. It is reported to be mediated through oxidative stress, which leads to lipid peroxidation and an alteration in antioxidant levels in the body [3]. Lipid peroxidation results in cell damage due to oxidative degradation of lipids present in cell membranes. In response to deleterious effects of free radical-induced lipid peroxidation, cells activate antioxidant defense mechanisms in which superoxide dismutase and reduced glutathione act synergistically to detoxify the effects of lipid peroxidation [3]. Therefore, it is suggested that the agents which reduce the lipid peroxide content in tissue and increase the intracellular antioxidant defenses may have protective effects on the liver in people taking antitubercular treatment. Currently very few reliable liver-protective drugs are available in the allopathic armamentarium. Their effects are unsatisfactory and they add to the pill burden. Management of drug-induced hepatotoxicity is still a challenge to modern medicine. Therefore, there is a strict need to screen herbal products and nutraceuticals which can be taken by the patients as food during the treatment of tuberculosis.

Goat milk is a food of high nutritional value as it is rich in various physiologically functional components, including proteins, vitamins (such as vitamins E and C), flavonoids, and carotenoids with antioxidant properties [4–7]. Goat milk is considered to possess high antioxidant activity that resists oxidative stability and highly protects consumers from exposure to oxidative stress [8]. It has been reported that goat milk consumption potentiates liver divalent metal transporter 1 expression thereby enhancing Fe metabolism and storage indicating its potential in anemia [9]. Furthermore, few studies have also demonstrated its anti-inflammatory and antioxidant properties which indicate that goat milk may possess hepatoprotective activity [10–13]. Goat milk is easily available, easy to digest, and can be taken as food during drug treatment of tuberculosis. The hepatoprotective activity of goat milk in antitubercular drug-induced hepatotoxicity has not yet been investigated. The rationale of the present study was to explore the effect of goat milk on antitubercular drug-induced hepatotoxicity in rats.

2. Materials and methods

Healthy adult albino rats of either sex of Wistar strain weighing 200–300 g were used after approval of the Institutional Ethics Committee. They were housed in standard laboratory conditions at 25 ± 2°C with a 12 hour light/dark cycle. Animals were given free access to a rat chow diet and water *ad libitum*. Before conducting experiments, animals were acclimatized to laboratory conditions for 7 days.

2.1. Induction of hepatotoxicity

Experimental antitubercular drug-induced hepatotoxicity was produced by administration of isoniazid, rifampicin, and pyrazinamide (H+R+Z) suspension daily orally for 30 days. The doses of antitubercular drugs (H: 27 mg/kg, R: 54 mg/kg, Z: 135 mg/kg/d; Kwality Pharmaceuticals Pvt. Ltd., Amritsar, Punjab, India) were extrapolated from daily human doses using a conversion table [14]. Gum acacia was used as a suspending agent. In the vehicle control group, 2% gum acacia (10 mL/kg) was administered to the rats. For induction of hepatotoxicity, a combination of calculated doses of H, R, and Z suspended in 2% gum acacia were administered to rats orally.

The animals were divided into five groups ($n = 6$). The groups were treated as follows:

Group I: vehicle control, i.e., 2% gum acacia orally for 30 days.
Group II: (H+R+Z) suspension orally for 30 days.
Group III: (H+R+Z) suspension + goat milk 20 mL/kg orally for 30 days.
Group IV: (H+R+Z) suspension + goat milk 40 mL/kg orally for 30 days.
Group V: (H+R+Z) + silymarin (standard) 50 mg/kg orally for 30 days.

Blood samples of animals from all the groups were taken on 30th day by cardiac puncture under ether anesthesia. After sacrificing the animals, livers were removed for histopathological examination and biochemical parameters were investigated.

2.2. Assessment of liver damage

2.2.1. Gross morphological assessment

Livers were excised from the rats and were rinsed with normal saline. They were weighed after blotting with filter paper. The liver indices were calculated as a percentage of the body weight [15]. A gross morphological assessment was then performed for hepatic lesions based on the qualitative procedure developed by Mitchell et al [16]. They were graded as follows: 0, no lesions; 1+, minimal damage; 2+, mild to moderate damage; and 3+, severe damage.

Each liver was excised into two pieces. The right lobe was immersed in isotonic 10% buffered formalin fixative for histological assessment while the left lobe was rinsed using cold physiological saline and then homogenized with cool phosphate buffer saline for malondialdehyde and superoxide dismutase (SOD) assays.

2.2.2. Histopathological examination

All the groups were subjected to histological examination. Microscopic examination was done by a qualified pathologist using hematoxylin and eosin staining in a blinded fashion.

2.2.3. Biochemical estimations

Serum alanine aminotransferase (ALT) and serum aspartate aminotransferase (AST) were estimated using the Reitman and Frankel method [17]. Tissue malondialdehyde and SOD