Original Article

Protective Role of Taurine on Tamoxifen-induced liver damage in Rats: A Morphological Study

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ABSTRACT

Objective: To study the protective role of taurine in tamoxifen- induced hepatic steatosis in albino rats

Study Design: Prospective experimental study

Place and Duration of Study: Department of Anatomy, Khyber Medical College, Peshawar, from July 2011 to November 2011.

Materials and Methods: Four groups of female albino rats, each containing 8 animals, were treated for 21 days as follows:

Group A served control, Group B treated with tamoxifen, Group C treated with tamoxifen plus taurine, Group D treated with taurine alone.

The animals were weighed at the start and end of treatment and then sacrificed under ether anaesthesia. The livers were fixed in 10% formalin and embedded in paraffin. H&E stained 4 µ thick sections were examined microscopically.

Results: There was significant decrease in the body weights of groups B and C as compared groups A and D. Significant increase in the body weights of group C rats in comparison with group B was noted. Histologically the livers of group B animals showed generalized microvesicular and macrovesicular steatosis, with no evidence of hepatocyte necrosis. In group C the fatty change was much less pronounced as compared to group B.

Conclusion: The study data suggest that taurine supplementation can reverse / attenuate the hepatic steatosis caused by tamoxifen treatment for 21 days, in albino rats.

Key Words: Hepatic steatosis, taurine, tamoxifen.

INTRODUCTION

Tamoxifen citrate is a non steroidal anti-estrogen drug used for the treatment and prevention of breast cancer[1]. It has been shown to be hepatotoxic and hepatocarcinogenic in rats[2,3,4]. Cases of liver injury induced by tomoxifen in humans (e.g. toxic hepatitis, steatosis, sub-massive hepatic necrosis and cirrhosis) have been described[6]. Tamoxifen produces oxidative stress in rat liver by overproduction of oxygen radicals during its metabolism. Lipid peroxidation is initiated through removal of hydrogen from unsaturated fatty acids; this leads to the formation of carbon-centered lipid radicals [4]. Addition of molecular oxygen to the lipid radicals form lipid peroxy radicals[5]. It appears that tamoxifen causes hepatic steatosis by derangement of mitochondrial function; this leads to impaired β-oxidation of fatty acids and production of reactive oxygen species and depletion of ATP. The protective effects of natural antioxidants against drug toxicities, particularly where free radical injury is the cause has been the focus of attention lately[6].

Taurine (2-amine ethane sulfuric acid) is a sulphur containing free amino acid, normally present in tissues including liver, brain, kidney and myocardium. The physiological and biological functions of taurine have been reported; it acts as antioxidant, membrane stabilizer, osmoregulator and neuromodulator[7]. Taurine is a non essential amino acid; its synthesis occurs chiefly in the liver from amino acids methionine and cystein[8]. Taurine, by its antioxidant action, has been shown to attenuate the oxidative stress and injury in the urinary bladder and kidney induced by nicotinamide[10], it ameliorates hypoxia induced lactic acidosis in brain, liver and heart[11], and reduces the severity of cyclophosphamide- induced hemorrhagic cystitis in rats[9].

Kerai et al[9] (1999) confirmed the protective role of taurine in ethanol induced hepatic steatosis. They noted that the effects of taurine on reversing hepatic steatosis may be due to the enhanced secretion of hepatic triglycerides, and that increased bile flow as a result of taurine treatment may contribute to the removal of lipid peroxides[9]. Taurine protects the liver against injury induced by agents such as carbon tetrachloride[15], acetaminophen[13], and thioacetamide[13]. Its restorative role in experimentally induced non- alcoholic steatohepatitis has been observed[14].

The purpose of this study was to find out the histological evidence of the effects of tamoxifen on rat liver and to find whether taurine, a sulfur containing amino acid, can offer any protection against the liver injury caused by tamoxifen, in rats.

MATERIALS AND METHODS

This study was conducted in the department of Anatomy, Khyber Medical College, Peshawar.

Tamoxifen citrate (Nolvadex®, ICI) and taurine (GNC, USA) were purchased from the local market.

Thirty two healthy adult female albino rats 90-120 days of age and weighing 200-300 gram, fed on standard chow and water ad libitum, were divided into four group of eight animals each and were treated for 21 days as follows:

Group A served as control.

Group B animals received tamoxifen 45 mg/kg body weight / day dissolved in drinking water and administered orally by gavage[16].

Group C animals received tamoxifen in a dose of 45 mg/kg body weight / day and taurine 1% solution as their sole source of drinking water.

Group D animals received 1% taurine solution as their sole source of drinking water[17].

Body weights of rats were recorded at the start of study and at the time of sacrifice.The animals were sacrificed under ether anesthesia. Livers were removed, weighed and examined grossly. Then they were fixed in 10% formalin and embedded in paraffin. 4 µ thick sections, stained with hematoxylin and eosin, were studied microscopically for cell morphology and lobular architecture. Histological diagnosis was made and results were tabulated.

RESULTS

The body weights of control (group A) and taurine-treated (group D) animals increased significantly (P value < 0.01) without any significant difference across the groups. There was a significant decrease (P value <0.001) between the initial and final body weights in tamoxifen-treated (group B), and tamoxifen plus taurine treated (group C) in comparison with group A (table 1). In comparison between groups B and C there was significant increase in the final body weights (P value <0.01) of group C in comparison with group B rats (Table 2).

Table No.1: *Mean Body Weights (BW) in grams at the beginning and end of three weeks treatment.

Group	Initial BW	Final BW	P Value
A	278.60±3.00	285.20±2.08	<0.01
B	273.60±6.49	233.20±5.21	<0.001
C	271.20±7.47	258.20±8.19	<0.001

*Mean±SEM

Table No.2: Comparison of *Mean Final Body Weights (grams) between group B and C.

Mean Final BW group B	Mean Final BW group C	P Value
233.20±5.21	258.20±8.19	<0.01

*Mean±SEM

Normal histological features were observed in the H&E stained sections of livers of group A and D animals (Figure-1). In the livers of group B animals there was distortion of lobular architecture and marked dilatation of central vein (Figure-2). Swelling of hepatocytes and obliterated sinusoids were seen. Fatty change was more prominent in comparison with an earlier study of one week duration[20]. Ballooned hepatocytes were seen in the zone III but no evidence of hepatocyte necrosis was found. Increase in the number of mononuclear cells in the region of portal triad was observed.

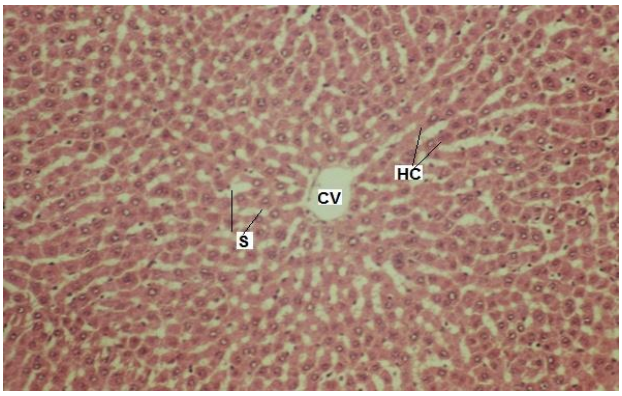

Figure No.1: Photomicrograph of liver lobule of control albino rat showing hepatic cords (HC) radiating from central vein (CV) and normal liver sinusoids (S): (H&E x200).

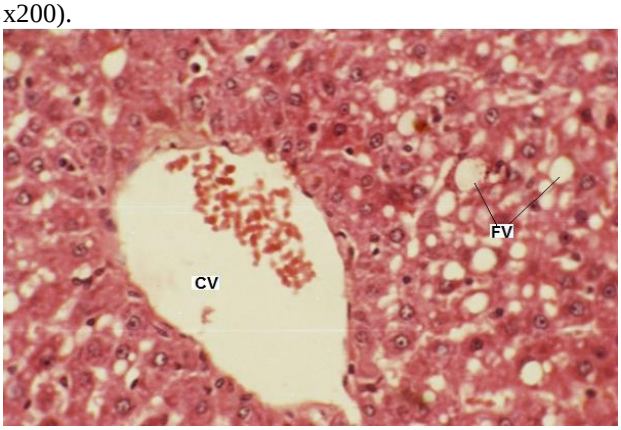

Figure No. 2: Photomicrograph of rat liver treated with tamoxifen for 21 days, showing dilated central vein (CV) and vacuoles of fatty change (FV): (H&Ex400).

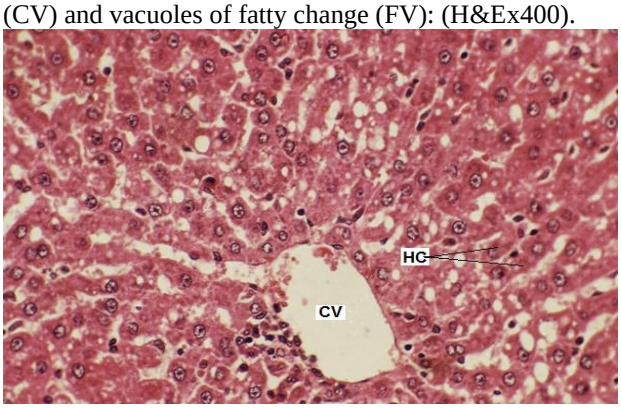

Figure No.3: Photomicrograph of rat liver treated with tamoxifen plus taurine for 21 days still showing dilatation of central vein (CV), restored arrangement of hepatic cords (HC) and reduced fatty change as compared to Figure-2. (H&E, x400).

In group C the lobular architecture showed little distortion but the arrangement of cells in cords was obvious. The central vein showed some dilatation, but the walls were not distorted. The fatty change was not as marked as in group B.

DISCUSSION

Liver injury caused by drugs is a major clinicopathological entity. There are about 900 drugs which are potentially hepatotoxic; the severity of injury ranging from mild to fatal[18].

The triphenylethylene antiestrogen drug, tamoxifen, is used worldwide for the treatment and prevention of breast cancer. Cases of tamoxifen-induced hepatotoxicity have been described, including toxic hepatitis, massive hepatic steatosis or multifocal hepatic fatty infiltration, and sub-massive hepatic necrosis in humans[5].

The decrease in the body weights of animals treated with tamoxifen (group B and C) coincides with the study of Lelliott et al[21]. The authors observed that tamoxifen was known to reduce food intake as well as body weight of tamoxifen-treated rats. Lopez et al[22] also observed that tamoxifen causes tremendous decrease in appetite via its direct action on the hypothalamus.

In the present study, the animals in group C showed significant increase in body weight in comparison with group B animals. This was because of the reduction of tamoxifen toxicity due to taurine treatment. This finding is in agreement with the findings of Naqvi and Ali (2008)[23], who described a weight gain in the rats treated with taurine plus tamoxifen as compared to rats treated with tamoxifen alone in a one week study. Hwang et al[24] studied the effects of taurine administration in rats treated with oxidized fish oil. They suggested that taurine improves the glutathione level in liver which leads to the improvement in body weight gain.

Microscopic examination of group-B animals showed swelling and ballooning of hepatocytes and marked fatty change. These findings correlate with the findings of Lelliott et al[21], who noted that 83% of tamoxifen-treated rats had microvesicular fatty change in their livers.

The finding of macrovesicular fatty degeneration and ballooning of hepatocytes were in agreement with the work of Angulo[25], in which the author has described the liver biopsy features of non alcoholic fatty liver disease. These include steatosis, mixed inflammatory cell infiltration, hepatocyte ballooning and necrosis, Mallory's hyaline and fibrosis. Pratt et el[26] and Hoof et al[27] have reported one case each with same words of macrovesicular fat infiltration, lobular inflammation and portal inflammation with fibrosis, in humans; these observations match with the findings in the present study.

The morphological examination of H&E stained sections of liver in group-C demonstrated that hepatic lobular architecture was comparable to control except mild dilatation of central vein. The fatty change showed a focal distribution and vacuoles were very much reduced in size. These findings can be attributed to the antioxidant, membranoprotective and detoxifying properties of taurine. These findings match with the findings of Chen et al[17], who studied the effects of taurine treatment in experimentally induced steatohepatitis in rats fed on high fat diet. The authors observed significant improvement in both histological and biochemical parameters. Kerai et al[10] suggested that taurine-induced reversal of fatty change is due to increased triglycerides secretion. They also suggested that an increased bile flow enhances the removal of peroxides.

CONCLUSION

The attenuation of fatty change by taurine administration is a finding of great importance. Tamoxifen causes non alcoholic fatty liver disease which can progress into steatohepatitis and cirrhosis in patients of breast cancer. The addition of taurine as a supplemental therapy to these patients can save them from fatty liver disease. Further studies are needed to confirm the findings on this topic.

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