

Original article

Evaluation of hepatoprotective and antidiarrhoeal activity of guduchi, *Tinospora cordifolia* (Willd.) Miers ex Hook. f. & Thoms. in experimental rats

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Abstract

The present study investigated the antidiarrhoeal and hepatoprotective potential of 50 % ethanolic extract of stems of *Tinospora cordifolia* (Willd.) Miers ex Hook. f. & Thoms. in experimental rats. Diarrhoea along with hepatotoxicity was induced by feeding red kidney beans, *i.e.*, *Phaseolus vulgaris*, mixed with rat feed in the ratio of 30:70 (study diet) along with administering CCl₄, mixed with olive oil (1:1; v/v) at 4 ml/kg body weight orally twice a week for trial period of 10 days. Group 1 was healthy control; Group 2 was positive control; Group 3 received silymarin (5 mg/kg b.wt orally) and atropine sulfate (0.1 mg/kg, intra muscular OD); Groups 4 and 5 received 50 % ethanolic extract of *T. cordifolia* at 100 and 200 mg/kg body weight orally, respectively. Results revealed that Group 3 rats restored fecal consistencies from 3rd day onwards while in Groups 4 and 5, fecal consistency restored from 7th and 8th day onwards, respectively. Significant increase in liver enzyme activities due to CCl₄ toxicity was restored well in Group 3, followed by the group receiving *T. cordifolia* at 200 mg/kg. No significant changes were noticed in blood urea nitrogen, creatinine and total bilirubin level within all groups. Oxidative stress indices like lipid peroxidation (3.03 ± 0.12 nmol MDA/ mg protein) and glutathione (6.46 ± 0.38 μmol/mg protein) were significantly improved in Group 3, followed by group receiving *T. cordifolia* at 200 mg, whereas superoxide dismutase (8.13 ± 0.1 U/mg protein) and catalase (7.94 ± 0.3 U/mg protein) were improved better in Group 3, followed by Group 5 receiving *T. cordifolia* at 100 mg. Histopathological study of liver revealed the changes like hepatic degeneration, vacuolation, and dissolution of cytoplasm in negative control group. Silymarin treated group revealed normalcy in hepatic cord with mild vacuolar changes. *T. cordifolia* at 200 mg/kg treatment group revealed only mild fatty changes in liver. This study revealed the dose-dependent hepatoprotectant and antidiarrhoeal effect of *T. cordifolia* in experimental rats.

Keywords: *Tinospora cordifolia* (Willd.) Miers ex Hook. f. & Thoms., diarrhoea, CCl₄ toxicity, oxidative stress

1. Introduction

Guduchi, *Tinospora cordifolia* (Willd.) Miers ex Hook. f. & Thoms., is a large deciduous climbing shrub, belonging to the family Menispermaceae, found throughout India, Srilanka, Nepal, Bangladesh and China. In Hindi, the plant is commonly known as giloe. In ayurvedic system, it is called as Rasayana drug which used for increase body resistance, long life and to alleviate stress (Patwardhan and Gautam, 2005). Giloe is also called as "Amrita" which signifies its use for revitalization and its importance in ayurveda. The plant is also included in various important pharmacopoeias (Anonymus, 1999). The phytochemical constituents of different classes like alkaloids, diterpenoid lactones,

glycosides, phenolics, aliphatic compounds and polysaccharides are isolated from *T. cordifolia* (Singh *et al.*, 2003). The stems of *T. cordifolia* are found positive for presence of alkaloids, phenols, saponins and tannins (Gurav *et al.*, 2014). The anti-inflammatory, antipyretic, antioxidant, antidiabetic, anti-allergic and antiarthritic activities of *T. cordifolia* have been reported in earlier studies (Jana *et al.*, 1999; Bishayi *et al.*, 2002; Stanley and Menon, 2001; Choudhary *et al.*, 2013). This drug is prescribed for various diseases like fever, diabetes, dysentery, cough, asthma, jaundice, diarrhoea, skin diseases and bites of poisonous insects (Devprakash *et al.*, 2011). It has also been indicated in heart disorders, leprosy and helminthiasis (Sharma, 1969; Aiyer and Kolamall, 1963; Kirtikar and Basu, 1933). The ayurvedic drug, guduchi (*T. cordifolia*) is mentioned in various medicine systems like Charak, Sushrut and Ashtang Hridaya and other treatises like Bhavva Prakash and Dhanvantari Nighantu with different names like Amara, Amritvali, Chinnarruha, Chinnodebha and Vatsadani, *etc.* (Sharma *et al.*, 2001; Charkha, 1961; Sushruta, 1992). Several herbs or plant materials have been reported to have antioxidant, antidiarrhoeal and hepatoprotective activity. The objective of the present study was

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to evaluate the antidiarrhoeal and hepatoprotectant potential of *T. cordifolia* stems in experimental animal. The study of two different physiological properties of the same plant will be helpful for therapeutic management of diarrhoeic hepatopathy.

2. Materials and Methods

2.1 Plant material

Stems of Guduchi, *i.e.*, *T. cordifolia* were collected in and around Bareilly of Uttar Pradesh, India area and air dried and grinded to powder form and were subjected to extraction, using 50% ethanol as solvent by columnar Soxhlet method at temperature of 40–41 °C with standard protocol. The extracts were dried at 41 °C temperature.

2.2 Experimental design

Male albino Wistar rats (100–150 g) of 3–4 weeks age, were obtained from Laboratory Animal Resource Section (LARS) of Indian Veterinary Research Institute, Izatnagar (IVRI) and were kept in experimental animal shed of Division of Medicine with standard feeding and managemental conditions. Rats were divided randomly into five groups (n=6). The study was conducted after due approval of Institute Animal Ethical Committee (IAEC) (No. F. 1 - 53 / 2004 / JD (R) dated 09-04-2010 of Joint Director of Research, IVRI).

2.2.1 Induction of hepatotoxicity along with diarrhoea in rats

Hepatotoxicity along with diarrhoea was induced in all groups except healthy control (Group 1) rats by feeding red kidney beans, *i.e.*, *Phaseolus vulgaris*, mixed with rat feed in the ratio of 30:70 (Study diet) along with administering CCl₄ mixed with olive oil (1:1; v/v) at 4 ml/kg body weight orally, twice a week for trial period of 10 days (Shoda *et al.*, 1995 with modification). Group 2 served as positive control which did not receive any treatment. Group 3 received silymarin (5 mg/kg body weight orally) and atropine sulphate (0.1 mg/kg, IM per day) as standard hepatoprotectant and antidiarrhoeal agent. Groups 4 and 5 received 50% ethanolic extract of *T. cordifolia* at 100 and 200 mg/kg body weight orally, respectively. The rats were observed for onset of diarrhoea initially and mortality pattern throughout the study period. Clinical signs or change in behaviour were also noted in all groups of rats and body weight was recorded on day 0, 5th and 10th day of trial. Following the overnight fasting rats were sacrificed by the end of experiment under light chloroform anaesthesia. Blood samples from anaesthetized rats were collected by cardiac puncture, using sterile syringes in heparinised and non-heparinised tubes.

2.3 Parameters of study

2.3.1 Hematology

Hemoglobin, PCV, RBC, WBC, differential leukocyte count was stimulated (Chauhan, 2003).

2.3.2 Serum marker enzymes

Following parameters like serum alanine amino transferase (ALT) and aspartate amino transferase (AST), alkaline phosphates (ALP), serum bilirubin, serum total protein (TP), serum albumin, blood urea nitrogen serum creatinine, serum sodium, potassium and chloride and serum glucose were analysed by using standard methods.

2.3.3 Oxidative stress indices

To assess oxidative stress, the level of LPO (Ohkawa *et al.*, 1979), SOD (Cohen, 1970), catalase (CAT) (Menami and Yoshikawa, 1979) and non-enzymatic antioxidant GSH (Sedlak and Lindsay, 1968) were determined in the liver tissues.

2.3.4 Histopathology

Liver tissue samples were collected at the time of sacrifices and immediately put in 10% formal buffered saline overnight and subsequently processed and stained with routine haematoxylin and eosin for histopathological examination (Culling, 1963).

2.4 Statistical analysis

Data were subjected to statistical analysis, using ANOVA and paired t-test and $p \leq 0.05$ was considered statistically significant. Values are expressed as mean \pm Standard error (SE).

3. Results and Discussion

The study diet was fed initially for few days to induce diarrhoea in experimental rats and after development of diarrhoea in rats, they were taken into account for trial period. As all the groups showed signs of diarrhoea on 3rd day of feeding study diet, we counted it as day 0 of our trial and continued for 10 days with study diet and CCl₄. Presence of lectin (Phytohemagglutinin) in red kidney beans is mostly responsible for induction of diarrhoea which was evidenced by appreciable change in stool consistency in all groups of rats. Diarrhoea due to the lectin in Long-Evans rats was also evidenced in earlier study (Shoda *et al.*, 1995). All groups of rats showed no behavioural or physical changes except alteration in body weights. On day 0 of trial, all rats were normal, alert and showed semisolid faecal consistency (Figure 1). No observable changes in behaviour were noticed in all groups of rats throughout the experiment. The faecal consistency became normal in Group 3 rats from 3rd day onwards. Rats treated with *T. cordifolia* at 200 mg dose, showed normal faecal consistency on day 7th day onwards while the group receiving *T. cordifolia* at 100 mg/kg showed normal faecal consistency from 8th day onwards. Negative control group showed semisolid faecal consistency throughout the trial period. Tannins and alkaloid content of *T. cordifolia* may be responsible for the antidiarrhoeal property (Mukherjee *et al.*, 1998; Gricilda and Molly, 2001). Tannins are responsible for protein denaturation producing protein tannate, which reduces secretion from intestinal mucosa (Tripathi, 1994). *T. cordifolia* also contains tannins which may produce antisecretory activity. The extract significantly protects the rats against diarrhoea, evoked by red kidney beans in a dose dependent manner. The effect of the extract at 200 mg/kg body weight was equal to atropine, which is at present one of the most efficacious and widely employed antidiarrhoeal drug. Alteration in body weights of different rat groups is shown in Table 1. Significant ($p \leq 0.05$) reduction in body weights of negative control group rats were observed on day 10 (65.0 g \pm 10.2) as compared to body weights on day 0 (96.67 g \pm 4.5) while all other treatment groups showed the only insignificant reduction in body weights. Ray and Mehendale (1990) and Nagano *et al.* (2007) reported that animals having a liver injury can lose their body weight and activity. Significant loss in body weight along with reduced feed consumption was reported in rats of CCl₄ toxicity group (Kavitha *et al.*, 2011). The loss in body weights of Group 2

rats may be due to effect of persistent diarrhoea throughout experiment which may lead to fecal energy loss, hence loss in body weight. Since the *T. cordifolia* extract treatment groups did not induce a significant decrease in body weight which may be attributed to inhibition of intestinal transit and also hepatoprotection against CCl₄ toxicity (Yu Li-Li *et al.*, 2000). Healthy control rats showed significant ($p \leq 0.05$) increase in liver weight among all. The haematological profiles in different groups of rats are shown in Table 2. No significant changes were noticed in Hb, PCV and TEC of healthy and other groups of rats, however, nonsignificant increase in PCV was noticed in all the groups as compared to the healthy control which may be attributed to level of dehydration in these groups secondary to diarrhoea. Significant ($p \leq 0.05$) increase in total leukocyte ($16.1 \pm 0.87 \times 10^3 \text{ cmm}^3$) and neutrophil (32 ± 3.88) count in negative control group was observed; however, neutrophil count reduced significantly ($p \leq 0.05$) in treatment groups. Increased TLC in rats may be attributed to general reaction of the immune system to bacterial infection and inflammatory processes in GIT. It was reported that lectin-induced diarrhoea was associated with intraluminal bacterial overgrowth (Shoda *et al.*, 1995). It was also evidenced that lectin-induced diarrhoea in rat can leads to intraluminal bacterial overgrowth (Banwell *et al.*, 1983). Above reports are consistent with our findings.



Figure 1: Semisolid fecal consistency in diarrhoeic rats.

Table 1: Alteration in body weights of different rat groups (mean \pm SE)

Day (s)	Group 1	Group 2	Group 3	Group 4	Group 5
Day 0	130.0 \pm 5.7 ^a	96.67 \pm 4.5 ^b	91.6 \pm 7.2 ^a	105.5 \pm 10.4 ^a	105.9 \pm 7.6 ^a
Day 5	140.0 \pm 7.6 ^a	78.33 \pm 5.8 ^{ab}	85.0 \pm 8.6 ^a	99.8 \pm 14.4 ^a	98.33 \pm 12.1 ^a
Day 10	145.0 \pm 7.6 ^a	65.00 \pm 10.2 ^a	71.6 \pm 12.0 ^a	95.0 \pm 14.8 ^a	88.33 \pm 7.2 ^a

Values bearing different superscripts in the same column vary significantly ($p \leq 0.05$)

Table 2: Haematological profile of different group of rats

Groups /Parameters	Hb(gm/dl)	PCV (%)	TEC (X 10 ⁶ /cmm3)	TLC (X 10 ³ cmm3)	N %	L %	M %	E %	B %
Group 1	13.43 \pm 0.61 ^a	43.04 \pm 1.54 ^a	5.62 \pm 0.34 ^a	8.4 \pm 0.81 ^a	19.0 \pm 3.05 ^a	78.3 \pm 3.18 ^b	1.67 \pm 0.33 ^a	0.67 \pm .33 ^a	0.23 \pm 0.06 ^a
Group 2	12.06 \pm 0.66 ^a	57.22 \pm 2.50 ^a	4.70 \pm 0.28 ^a	16.1 \pm 0.87 ^b	32.0 \pm 3.88 ^b	64.67 \pm 1.20 ^a	2.00 \pm 0.57 ^a	0.33 \pm .33 ^a	0.33 \pm 0.07 ^a
Group 3	13.30 \pm 1.35 ^a	54.20 \pm 4.91 ^a	5.13 \pm 0.49 ^a	12.50 \pm 1.33 ^{ab}	18.0 \pm 2.08 ^a	77.33 \pm 2.60 ^{ab}	3.00 \pm 0.57 ^a	1.00 \pm .00 ^a	0.67 \pm 0.13 ^a
Group 4	11.49 \pm 0.81 ^a	56.10 \pm 4.83 ^a	5.70 \pm 0.26 ^a	13.80 \pm 1.81 ^{ab}	19.0 \pm 1.52 ^a	78.33 \pm 2.18 ^b	1.00 \pm 0.37 ^a	1.00 \pm .00 ^a	0.10 \pm 0.01 ^a
Group 5	13.00 \pm 0.58 ^a	50.40 \pm 2.73 ^a	5.34 \pm 0.31 ^a	12.60 \pm 1.30 ^{ab}	21.0 \pm 2.30 ^a	79.67 \pm 4.09 ^b	1.30 \pm 0.33 ^a	1.00 \pm 577 ^a	0.00 \pm 0.00 ^a

Values bearing different superscripts in the same column vary significantly ($p \leq 0.05$)

Serum biochemical changes in different groups of rats are shown in Table 3. Serum ALT ($318.06 \pm 20.13 \text{ IU/l}$), ALP ($157.53 \pm 7.30 \text{ IU/l}$) and AST ($300.7 \pm 5.93 \text{ IU/l}$) value of increased significantly ($p \leq 0.05$) in Group 2 in comparison to healthy and all other groups. All treatment groups, *viz.*, Group 3 ($70.46 \pm 4.34 \text{ IU/l}$), 4 ($122.3 \pm 8.81 \text{ IU/l}$) and 5 ($83.85 \pm 4.45 \text{ IU/l}$) showed significant ($p \leq 0.05$) reduction in serum ALT value in relation to value of negative control group. Results showed that ALT was best restored in Group 3 followed by Group V. AST level was reduced significantly in silymarin and atropine treatment group ($81.46 \pm 8.84 \text{ IU/l}$) and treatment group receiving *T. cordifolia* at 200 mg/kg, whereas ALP was restored better in rats receiving *T. cordifolia* at 100 mg/kg, followed by standard treatment group (silymarin+atropine). The increased activity of liver marker enzymes such as ALT, AST and ALP in the serum of CCl₄ induced rats, indicates damage to hepatic

cells (Wolf, 1999). Damage to the cell integrity of the liver by CCl₄ is reflected by an increase in the activity of ALT and AST which is released into circulation after cellular damage. ALP is an ectoenzyme of plasma membrane of liver cells. In CCl₄ mediated toxicity increased permeability of the hepatocyte membrane and cellular leakage causes high levels of ALP in serum (Paduraru *et al.*, 1996). The significant ($p \leq 0.05$) reduction in the serum ALT and AST and ALP levels in all treatment groups as compared to the negative control group revealed the hepatoprotective effect by preventing the possible hepatocellular injury. *T. cordifolia* treatment group showed dose-dependent effect of extract in the restoration of increased enzyme levels of ALT and AST in serum. Treatment with *T. cordifolia* extract (100 mg/kg b.wt for 15 days) in CCl₄ intoxicated rats was found to protect the liver as indicated by significant reduction in serum levels of ALT, AST, ALP and bilirubin. Pingale (2010) reported

significant increase in level of blood biochemical parameters in CCl₄ group as compared to those of normal control group. All these biochemical changes returned towards the normalcy in *T. cordifolia* treatment group which indicates the hepatoprotective effect of *T. cordifolia*. Significant reduction in ALT, AST and ALP levels were observed in the groups treated with silymarin and extracts of *T. cordifolia* at 200 mg/kg b.wt orally (Kavitha *et al.*, 2011). Above findings are concurrent with our reports. The level of total protein and glucose (130.40 ± 7.4 mg/dl) were increased significantly ($p \leq 0.05$) in negative control group as compared to healthy rats. Increase in total protein level in negative control rats in our case may be due to the release of proteins from liver in blood due to short term CCl₄ toxicity. No significant changes were noticed in BUN, creatinine and total bilirubin level within all groups.

Oxidative stress indices levels of different treatment groups are shown in Table 4. Significant ($p \leq 0.05$) improvement in LPO (3.03 ± 0.12 nmol MDA/ mg protein) level was noticed in rats, received silymarin and atropine as standard therapeutic agent, followed by group receiving *T. cordifolia* at 200 mg (3.30 ± 0.20 nmol MDA/ mg protein) as compared to LPO level in negative control rats (7.36 ± 0.29 nmol MDA/ mg protein). The level of lipid peroxide is a measure of membrane damage and alterations in structure and function of cellular membranes. Doi *et al.* (1991) who found that rats were given 2 ml CCl₄ with 2 ml olive oil per kg body weight orally produced hepatotoxicity and revealed increased LPO level in the liver of exposed rats. Silymarin is a standardized mixture of antioxidant flavonolignans (silybin and silibinin) extracted from

medicinal plant *Silybum marianum*. It is also a free radical scavenger and membrane stabilizer that prevents lipid peroxidation (Soto *et al.*, 1998). GSH values were found significantly ($p \leq 0.05$) improved in standard, the treatment group (6.46 ± 0.38 µmol/mg protein), followed by group receiving *T. cordifolia* at 200 mg (6.23 ± 0.3 µmol/mg protein) as compared to level of GSH in negative control rats (4.3 ± 0.15 µmol/mg protein). Significant ($p \leq 0.05$) improvement in SOD (8.13 ± 0.1 U/mg protein) and catalase (7.94 ± 0.3 U/mg protein) levels were noticed in the rats of the standard treatment group followed by group receiving *T. cordifolia* at 100 mg (7.26 ± 0.4 U/mg protein; 7.69 ± 0.2 U/mg protein) and *T. cordifolia* at 200 mg (6.76 ± 0.2 U/mg protein; 6.56 ± 0.1 U/mg protein). Critical analysis of oxidative stress indices revealed best antioxidant activity in standard treatment group, followed by group receiving *T. cordifolia* at 200 mg and *T. cordifolia* at 100 mg/kg body weight orally. Pre-treatment of rats with 250 mg/kg b.wt.o of *T. cordifolia* extracts improved SOD, catalase, peroxidase and decreased lipid peroxidation level in comparison to CCl₄ treated group. Pretreatment with 500 mg/kg of *T. cordifolia* extract further improved the antioxidant status which revealed a dose-dependent effect of extract in improvement of antioxidant status of the liver (Singh *et al.*, 2010). Above report is in concomitant with present study. The CCl₄ toxicity effect was reduced significantly by restoration of superoxide dismutase, catalase, peroxidase and decreased lipid peroxidation as compared to the negative control group. Increased levels of oxidative stress enzymes reveals antioxidant potential of *T. cordifolia*.

Table 3: Serum biochemical profile of different group of rats

Group	ALT (IU/L)	AST (IU/L)	ALP (IU/L)	Total Protein (gm/dl)	Albumin (gm/dl)	Glucose (mg/dl)	BUN (mg/dl)	Creatinine (mg/dl)	Bilirubin (mg/dl)
Group 1	67.32 ± 4.3 ^a	73.45 ± 3.96 ^a	50.89 ± 2.86 ^a	6.2 ± 0.26 ^a	4.12 ± 0.16 ^{ab}	73.84 ± 6.02 ^a	10.53 ± 1.18 ^a	1.13 ± 0.12 ^a	0.33 ± 0.13 ^a
Group 2	318.06 ± 20.13 ^c	300.7 ± 5.93 ^c	157.53 ± 7.30 ^b	9.58 ± 0.56 ^b	6.56 ± 0.32 ^b	130.40 ± 7.41 ^b	14.2 ± 1.25 ^a	1.40 ± 0.50 ^a	0.39 ± 0.29 ^a
Group 3	70.46 ± 4.34 ^a	81.46 ± 8.84 ^a	54.09 ± 4.37 ^a	6.48 ± 0.51 ^a	4.3 ± 0.46 ^{ab}	79.08 ± 5.93 ^a	11.5 ± 1.8 ^a	1.10 ± 0.15 ^a	0.34 ± 0.28 ^a
Group 4	122.30 ± 8.81 ^b	170.60 ± 7.73 ^b	43.08 ± 3.86 ^a	6.66 ± 0.39 ^a	4.4 ± 0.60 ^{ab}	101.6 ± 13.8 ^{ab}	11.46 ± 0.99 ^a	1.20 ± 0.23 ^a	0.37 ± 0.26 ^a
Group5	83.85 ± 4.450 ^{ab}	84.49 ± 4.42 ^a	54.50 ± 2.76 ^a	6.13 ± 0.75 ^a	3.8 ± 0.913 ^a	80.1 ± 8.53 ^a	14.93 ± 2.05 ^a	1.40 ± 0.47 ^a	0.29 ± 0.58 ^a

Values bearing different superscripts in the same column vary significantly ($p \leq 0.05$)

Table 4: Oxidative stress indices in rats given different treatment

Group	LPO (nmol MDA/ mg protein)	GSH (µmol/mg protein)	SOD (U/mg protein)	CAT (U/mg protein)
Group 1	2.13 ± 0.20 ^a	6.9 ± 0.15 ^c	8.20 ± 0.23 ^c	8.20 ± 0.23 ^c
Group 2	7.36 ± 0.29 ^c	4.3 ± 0.15 ^a	4.90 ± 0.17 ^a	4.96 ± 0.12 ^a
Group 3	3.03 ± 0.12 ^a	6.46 ± 0.38 ^{bc}	8.13 ± 0.11 ^c	7.94 ± 0.33 ^c
Group 4	4.30 ± 0.41 ^b	5.33 ± 0.31 ^{ab}	7.26 ± 0.44 ^{bc}	7.69 ± 0.27 ^{bc}
Group 5	3.30 ± 0.20 ^{ab}	6.23 ± 0.31 ^{bc}	6.76 ± 0.29 ^b	6.56 ± 0.17 ^b

Values bearing different superscripts in the same column vary significantly ($p \leq 0.05$)

Histopathological study revealed normal architecture and failed to reveal any specific pathological changes liver tissue of healthy rats. The liver tissues revealed of grade 0 by histopathological score system (HPS) (Figure 2). The negative control group showed hepatic degeneration, vacuolation and dissolution of cytoplasm. It was characterized by cytoplasmic vacuolations of variable sizes putting the nucleus to adjacent areas. The changes were mostly confined to perilobular regions. The liver tissues revealed grade 3 by

histopathological score system (Figure 3). Hepatic degenerative changes with postnecrotic lesions in rats treated with CCl₄ observed in 2nd week of trial (Doi *et al.*, 1991). The group receiving silymarin and atropine revealed normalcy in hepatic cord with mild vacuolar changes and graded 0-1 by HPS system (Figure 4). Treatment group receiving *T. cordifolia* at 100 mg/kg orally revealed, hepatic degeneration, mild to moderate vacuolar changes in centrilobular region and vascular congestion and graded 1-2 by HPS (Figure 5).

The Group 5, receiving *T. cordifolia* at 200 mg/kg orally revealed mild fatty changes and vascular congestion with mild infiltration of inflammatory cells and graded 0-1 by HPS system (Figure 6). The rats treated with silymarin and *T. cordifolia* extracts along with CCl_4 toxicant showed sign of protection against the toxicant to considerable extent as evident from formation of normal hepatic cords. Our previous study revealed the presence of flavonoids in the extracts of *T. cordifolia*. Flavonoids usually show better hepatoprotective activity (Scevola *et al.*, 1984). So in present study, the hepatoprotective effect of *T. cordifolia* may be due to its flavonoid content. Critical analysis of effect of *T. cordifolia* as antidiarrhoeal and hepatoprotectant in experimental animals revealed that group receiving *T. cordifolia* at 200 mg/kg showed better potential.

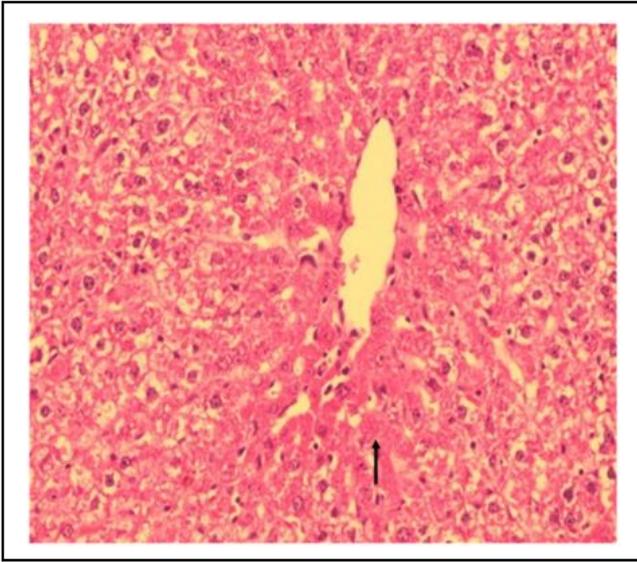


Figure 2: Liver: HP Group 1: Normal architecture of hepatic lobules (H & E, 20 x).

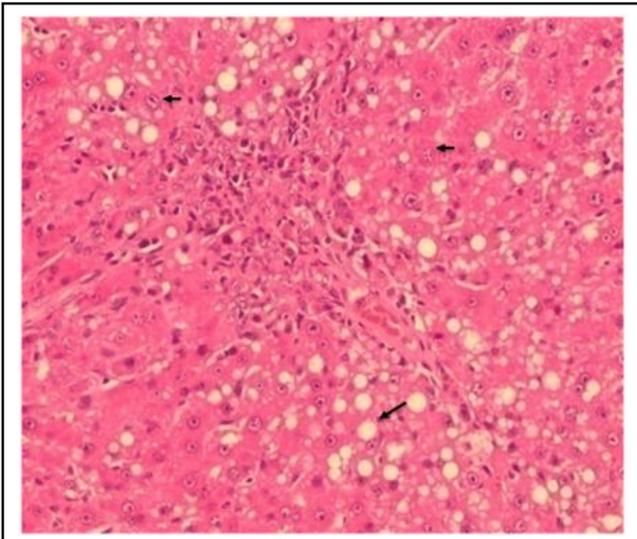


Figure 3: Liver: HP Group 2 : Vacuolar fatty (big arrow) changes in hepatocytes with mild nuclear changes (small arrow) (H & E, 20 x).

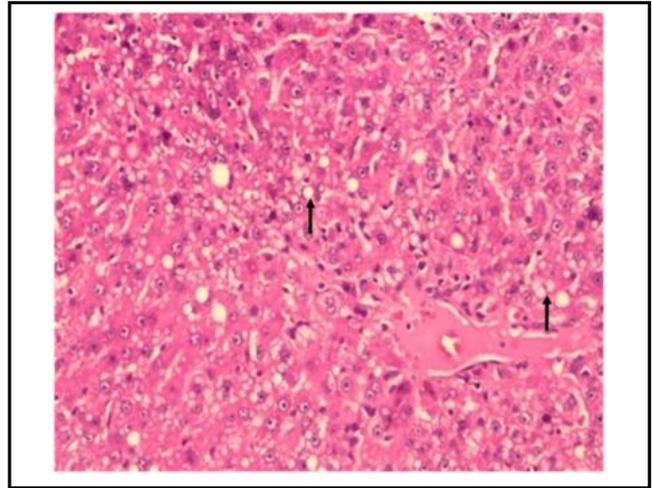


Figure 4: Liver: HP Group 3 : Mild vacuolar changes (arrow) in hepatocytes (H & E, 20 x).

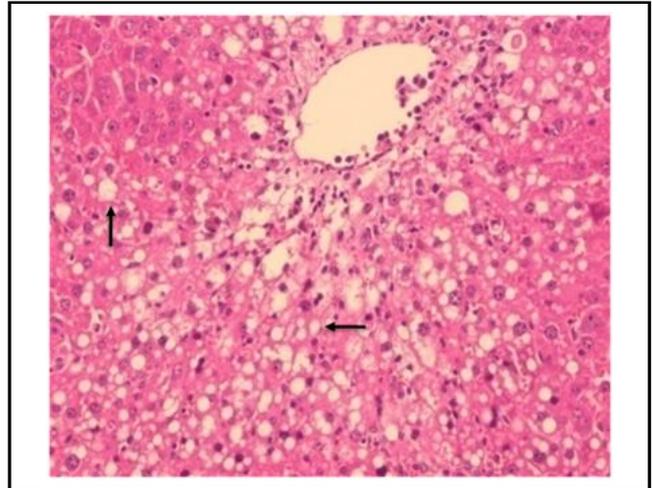


Figure 5: Liver: HP Group 4 : Mild to moderate fatty changes (arrow) in hepatocyte (H & E, 20 x)..

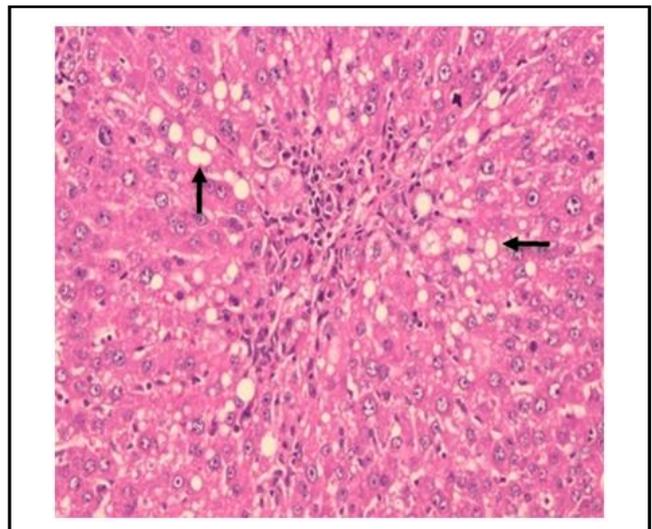


Figure 6: Liver: HP Group 5 : Mild fatty changes (arrow) in hepatocytes (H & E, 20 x).

4. Conclusion

It can be concluded from the study that 50% ethanolic extract of stems of *T. cordifolia* shows anti-diarrhoeal as well as hepatoprotective potential in dose dependent manner. The experimental group receiving *T. cordifolia* at 200 mg/kg body weight showed better anti-diarrhoeal and hepatoprotective potential.

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Conflict of interest

We declare that we have no conflict of interest.

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