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ORIGINAL RESEARCH ARTICLE- EXPERIMENTAL **OPEN ACCESS**

ANTIOXIDANT AND AMELIORATIVE EFFECT OF WHOLE PART OF *FUMARIA PARVIFLORA* AQUEOUS EXTRACT AGAINST CCL_4 INDUCED HEPATOTOXICITY IN ALBINO RATS

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ABSTRACT:

Background: Today, hepatic disorders are one of the most prevalent diseases in modern society. Medicinal plants with antioxidant properties are used to treat hepatic diseases. **Objective:** The present study aimed to investigate antioxidant and protective efficacy of an aqueous extract of whole part of *Fumaria parviflora* (FP) against carbon tetrachloride (CCl_4) induced acute liver injury. **Materials and Methods:** Free radical scavenging activity of FP plant extract was evaluated by DPPH and H_2O_2 assay and *in vivo* hepatoprotective activity of FP plant aqueous extract at three doses (300 mg/Kg, 600 mg/Kg, and 900 mg/Kg, orally) was assessed against CCl_4 (1.5 mL/kg intraperitoneally once only) induced hepatotoxicity in rat animals. The effect of plant extract on Hexobarbitone induced sleep-time and bromosulfophthale in retention time was also determined against CCl_4 intoxication in mice. **Results:** The administration of CCl_4 in experimental animals significantly increased the level of the hepatospecific markers such as serum transaminases (AST and ALT), albumin, hepatic lipid peroxidation, and decreased glutathione levels, which were brought back to normal by the *Fumaria parviflora* treatment. FP extract has also shown modulatory effect on the sleep period produced by hexobarbitone indicated increased the liver's excretory capacity with the BSP retention time. The biochemical findings also supported with histological examinations which were evidenced by restoration of the structural changes and improvement in the cellular morphology by the therapy of *Fumaria parviflora* after CCl_4 induced hepatotoxicity. **Conclusion:** The present study suggests that FP extract has significant antioxidant and hepatoprotective effect against CCl_4-induced acute liver injury and this effect might be due to its free radical scavenging action.

Keywords: *Fumaria parviflora*, Carbon-tetrachloride, Hepatotoxicity, Hepatoprotective, Antioxidant.

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INTRODUCTION

The liver is an important metabolic and drug detoxification organ that helps in the metabolism and transformation of biological molecules and xenobiotics. It plays a crucial role in removal of harmful substances from the body. Liver diseases are group of collective illness caused by exposure of infectious agents (bacteria and viruses), pollutants, and overdose of drugs, synthetic chemicals and pesticides. The prevalence of liver problem is become global public health issues[1]. Despite significant scientific progress in the field of hepatology in recent years, liver diseases such as jaundice, cirrhosis and hepatitis are account for high death rate[2].Due to the side effects of modern medicine, medical professional/researchers are looking towards traditional system medicine for the treatment of the liver diseases. Now days, there is a growing interest in studying various plant products/extract derived from medicinal plants as potential sources of hepatoprotective agents.

Medicinal plants, plant extracts and herbal formulations have been used for the treatment of human diseases since Vedic periods. Herbal medicines are used to treat a variety of liver ailments. Few potential hepatoprotective herbal medicines such as phyllanthin, Silmarin, andrographolide, curcumin, picroside, glycyrrhizin and Liv 52 (mixture of herbs) are clinically tested and used for the treatment of liver diseases[3-4] . Herbalists all over the world used various medicinal plants to prevent and treat liver diseases. *Fumariaparviflora* (F. parviflora) is a folk medicinal plant which are widely used as an hepatoprotective, diuretic, and appetizer in traditional system of medicine[5-6]. It has been reported that it is rich in polyphenolic and alkaloids (protopine, parfumine and fumariline etc.) which are possibly its active components.[7]However, there is a dire need to validate hepatoprotective claim of medicinal plant such as *Fumaria parviflora* by scientific study.

Therefore, the present study focuses to evaluate free radical scavenging and hepatoprotectiveeffect of *F. parviflora*against carbon tetrachloride (CCl_4)induced liver damagein Albino rats of the Sprague-Dawley strain.

MATERIALS AND METHODS

Plant material

The plant sample of *Fumaria parviflora* (FP) was provided in Autumn season (January-March, 2009) by the Central Council for Research in Unani Medicine (CCRUM), New Delhi. The plant material was identified and authenticated by Prof. Ashok K. Jain,

Taxonomist, Department of Botany, Jiwaji University, Gwalior (M.P.).

Preparation of aqueous extract of *Fumaria parviflora* (AFP)

The whole part of FP was washed and cut into small pieces and dried at room temperature for 2 weeks. The dried parts were powdered using the mechanical grinder. The distilled water was mixed (250 g/4L) with the plant and subjected for 18 h with concomitant shaking. After vigorous shaking, the aqueous mixture was filtered via the Whatman filter paper. The extract was further concentrated using rotary evaporator at 45°C and obtain yellowish aqueous extract which was stored in sterile tube at 4°C for further studies.

Chemicals

Chemicals and reagents used were silymarin (Code: S0292, store -20°C) from Sigma Aldrich, Germany and reduced glutathione (GSH), 1,2-dithio-bis nitro benzoic acid (DTNB), thiobarbituric acid (TBA), sodium hydroxide, trichloroacetic acid (TCA) and perchloric acid (PCA) were obtained from SRL, Mumbai-India. Carbon tetrachloride-CCl_4 (99%) was purchased (Code No. 1.02222.2500) from the E-Merck Germany. The assay kits- urea (Code No. 120214) and albumin (Code No. 120223) were purchased from Erba Lachema s.r.o. Czechia & Slovakia. All reagents used were of analytical grade.

Animals

Albino rats of the Sprague-Dawley strain (160±10 gram body weight) were chosen at random from the departmental animal facility, Zoology Department, Jiwaji University, Gwalior, (M.P.) India and maintained in polypropylene cages under consistent husbandry conditions of light (14 h) and dark (10 h), temperature (25±2°C) and relative humidity (60-70%). The animals were fed a commercially available conventional animal diet (Pranav Agro Industries Ltd., Pune, India) and drinking water *ad-libitum*. The experimental protocol was approved by the Institutional Animal Ethical Committee (Regd.no.CPCSEA/501/01/A) in accordance with the rules established by the Indian Committee for the Control and Supervision of Experiments with Animals

Free radical scavenging activity

Free radical scavenging activity of FP extract was evaluated using the dye i.e. 1,1-diphenyl-2-picryl-hydrazil ($DPPH^{\bullet}$)[8].In brief, a 0.1 mM DPPH dye solution was produced in ethanol, and 1 ml of this solution was added to the plant extract(AFP) at the concentration 10-50 µl/ml and 3 ml of water was added to make final volume 5 mL.Vitamin C was used as a positive reference at the same concentration. The mixture was briskly shaken before being allowed to stand at room temperature for 20

minutes. The intensity of the colour product was measured at 517 nm using UV-Vis spectrophotometer. The following equation was used to calculate the percentage of DPPH free radical scavenging.

$DPPH^{\cdot}$ Scavenging effect (%) = [(Ao − A1/Ao) x 100]

Where Ao represents the absorbance of the control and A_1 represents the absorbance of the test sample.

Hydrogen peroxide scavenging activity

The hydrogen peroxide scavenging activity was measured using the method described in Ruch et al.,1989 [9]. A plant extract (AFP) with different concentrations (10-50 µl/ml) was added to 3.4 ml of 0.1 M phosphate buffer (pH 7.4), followed by 0.6 ml of 43 mM hydrogen peroxide. The absorbance of the reaction mixture at 230 nm was measured after 10 minutes. For each concentration, a control mixture without sample and a blank mixture without H_2O_2 were used and the % H_2O_2 inhibition was calculated as follows:

% Inhibition $[H_2O_2] = [(A_0\text{-}A_1)/A_0] \times 100$

Where A_0 was the absorbance of the control, and A_1 was the absorbance of the sample. Ascorbic acid was taken as standard.

Preparation of doses and treatments

The hepatotoxicant CCl_4 was prepared with olive oil (1:1) as a vehicle and injected once only as a single dose of 1.5 ml/kg to the animal via intraperitoneally (i.p.) [10]. Based upon previous studies [11], the doses 300, 600, and 900 mg/kg of plant extract were selected and these doses were prepared in distilled water and given a single dose post orally to the animals.

Hepatoprotective Activity of AFP in CCl_4 induced hepatotoxicity

A total of 42 adult female rats were separated into seven groups of six individuals each. Group 1 treated with Olive oil (once only) and served as normal Control. Groups 2-7 received CCl_4 once only (1.5 ml/kg i.p.). Group 2 was negative control group. Groups 3-6 received single dose of *F. parviflora* at three doses 300, 600 and 900 mg/kg, *p.o.* (once only) respectively after 48 hours of CCl_4 intoxication. Group 7 received silymarin (50 mg/kg, *p.o. once only*). and served as a positive control[12]. .

After 48 hours of the last treatment, all of the animals were euthanized. Blood was drawn by puncturing the retro-orbital venous sinus, and the blood biochemistry and tissue biochemical parameters were performed:

Blood biochemical assay

Blood sample was allowed to clot and centrifuged at 3000 rmp for 15 minutes for serum separation. The separated serum was stored at −20ºC until use. The following biochemical parameters such as aspartate

aminotransferase (AST) and alanine aminotransferase (ALT)[13]. The assay kits- urea (Code No. 120214, stored at 2-8˚C, GLDH Urease method) and albumin (Code No. 120223 stored at 2-8˚C, BCG dye method) were performed as per the manufacture's protocol.

Tissue biochemical assay

Immediately after necropsy, tissues were excised, washed with normal saline, blotted and stored frozen. For enzymatic assays, the tissues were homogenized with a Remi Motor homogenizer (RQ-122) using glass tube and Teflon pestle, in different media according to the protocol of parameters. Tissue homogenates (5% w/v) were prepared in hypotonic solution (0.008% $NaHCO_3$). The required volume of 50 mg/ml was made with chilled hypotonic solution and various suitable aliquots for different estimation were taken. Liver homogenates (10% w/v) was prepared in 0.15 M KCl for LPO, whereas liver homogenates (5% w/v) were prepared in 1% sucrose solution for the estimation of GSH. Adenosine triphosphates (ATPase) and glucose 6 phosphate (G6Pase) homogenates were prepared in hypotonic solution. Lipid peroxidation (LPO) was assessed by determining the thiobarbituric acid reactive compounds (TBARS)[14]. Reduced glutathione (GSH) level was assessed by the method[15]. In addition, the enzyme activities of adenosine triphosphatase (ATPase)[16]and glucose-6-phosphatase (G-6-Pase) were also assessed [17].

Histological observations

For histological analysis, rat liver tissues were quickly dissected, fixed Bouin's fixative (acetic acid, 5% · formaldehyde, 9% · picric acid, 0.9%), and then after 12 hours, fixed sample tissues were dehydrated using various concentrations of ethanol (70, 80, 90, 95, and 100%). The samples were cleared in two changes of xylene after dehydration and then imbedded and blacked out after being soaked twice in hot paraffin wax . Thin sections of 4–5 microns were made from paraffin-embedded block and stained with hematoxylin and eosin for histological examination. Microscopic slides were prepared and observed under a microscope [18].

Hexobarbitone induced sleep time

The effects of hexobarbitoneon sleep were evaluated using the methods described in [19]. Four groups of six swiss albino mice each were separated: Group 1 was the normal control and received vehicles only. Groups 2–4 treated with CCl_4 (1.5 ml/kg, *i.p.*) in which group 2 was the negative control. Groups 3 and 4 received single dose of *F.parviflora* (900 mg/kg) and silymarin (50 mg/kg) respectively after 48 hours. of CCl_4 exposure.

Hexobarbitone (60 mg/kg, *i.p.*) was given to group animals after 48 h. of last administration. The hexobarbitone induced sleep time of *F. parviflora* was determined using the following formula, and the time in minutes from the onset of reflex loss till recovery was used as the duration of sleep.

$$\% \text{ protection} = 1 - \frac{Td - Tn}{Tc - Tn} \times 100$$

where T is the sleep time; Tc is CCl_4 , Td is *F. parviflora* and silymarin and Tn is normal control group respectively.

Bromosulphalein (BSP) retention time

The BSP retention time was performed as per the Kutob SD, Plaa, 1962[20] to determine the effect of FP on CCl4 induced BSP retention time in mice. The mice were divided into four groups (n=6). Group 1 treated as normal control and received only vehicle (olive oil). Groups 2-4 received CCl_4 (1.5 ml/kg, i.p.), while Group 2 served as the negative control. After 48 hours of toxicant administration, F. *parviflora* (900 mg/kg) and silymarin (50 mg/kg) were given orally to the groups 3 and 4, respectively. BSP (100 mg/kg, *i.v.*) was injected into all four groups 48 hours after the last treatment. After 30 minutes, blood was taken in heparinized tubes, processed for plasma separation, and BSP concentration was determined. The following formula was used to calculate excreting capacity based on dye retention.

$$\% \text{ protection} = 1 - \frac{Rd - Rn}{Rc - Rn} \times 100$$

Where R is the retention of BSP in plasma; c, d and n are CCl_4, drug (*F. parviflora* and silymarin) and normal groups respectively.

Statistical analysis

The results are reported as the Mean ±SEM. of six animals in each group. Data were statistically analysed using one way analysis of variance (ANOVA). $P \leq 0.05$ was considered to be significant at the 5% level[21].

Results

Free radical scavenging activity of plant extract

Free radical scavenging activity of *F.parviflora* was performed using the DPPH assay. FP extract at the concentration (10-50 µg/ml) showed DPPH inhibition in dose dependent manner. Upto 90% DPPH inhibition was found at higher concentration (50 µg/ml) when compared with positive control vitamin C. Thus, it was clearly demonstrated that FP extract possesses significant antioxidant properties against DPPH free radicals Figure1.

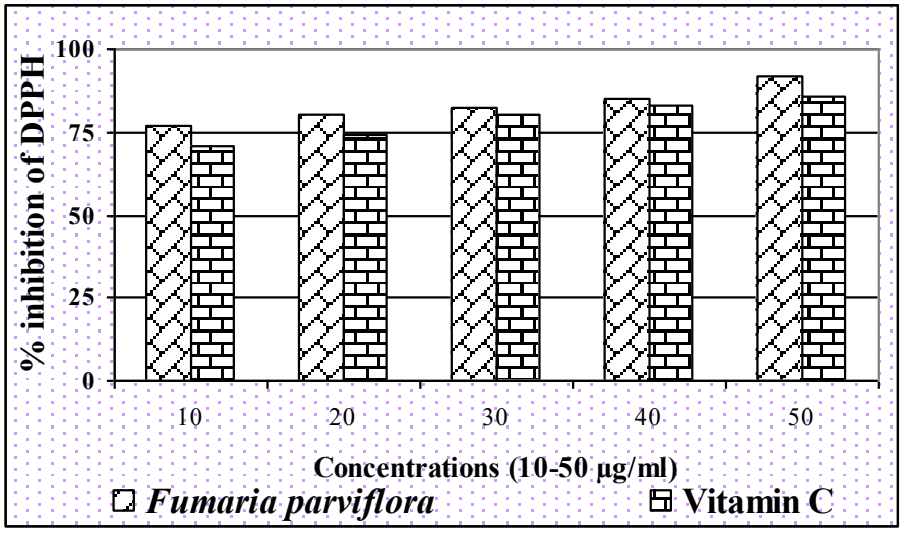

Figure 1. Free radical Scavenging activitiy of *F. parviflora* Aquoes extract

Values are mean ± S.E.M., n=3 in each group

Hydrogen peroxide (H_2O_2) scavenging activity

H_2O_2 scavenging activity of FP at different concentrations (10 to 50 µg/ml) showed significant H_2O_2 scavenging activity which was upto 80% H_2O_2 inhibition at 50 µg/ml concentration (Figure 2).

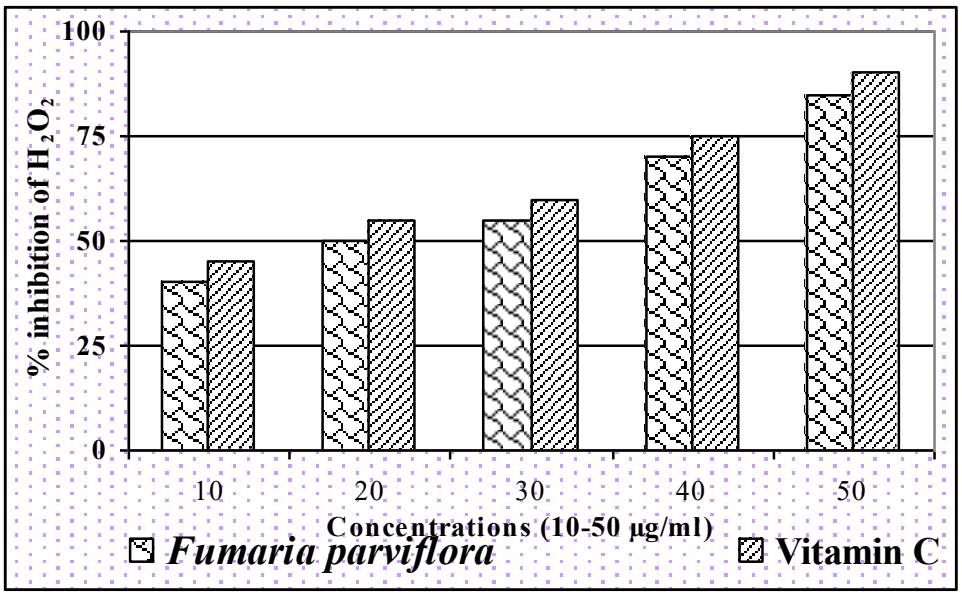

Figure 2. H_2O_2 inhibition activity was of *F. parviflora* Aquoes extract

Values are mean ± S.E.M., n=3 in each group

Blood biochemical assay

The effect of FP extract on CCl4 induced liver damage is shown in Table 1. CCl_4 exposure significantly increased the levels of hepatospecific enzymatic activities of ALT and AST in serum ($P \leq 0.05$) when compared with the control group. The level of albumin and urea level was also enhanced in the CCl_4 treated group. Therapy with FP extract at three doses (300, 600 and 900 mg/kg) showed significant recovery in these parameters in CCl4 treated group. The maximum recovery was observed at the lower dose 300 mg/kg compared to the higher doses 600 and 900 mg/kg ($P \leq 0.05$). No adverse effect were observed on these parameters in FP *per se* group.

Table 1 Effect of *F. parviflora* aqueous extract against CCl_4 induced hepatotoxicity in blood biochemistry.

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Treatments	AST (IU/L)	ALT (IU/L)	Albumin (g/dl)	Urea (mg/dl)
Control	69.5 ± 3.84	44.1 ± 2.43	3.80 ± 0.21	20.1 ± 1.11
F. parviflora per se	70.0 ± 3.86	44.0 ± 2.43	3.75 ± 0.20	20.0 ± 1.10
CCl_4 (Negative control group)	198 ± 10.9#	425 ± 23.4#	5.72 ± 0.31	48.3 ± 2.67#
CCl_4 + FP (300 mg/kg)	104 ± 5.74* (73.1%)	61.5 ± 3.39* (95.4%)	4.90 ± 0.27 (42.7%)	29.1 ± 1.60* (68.0%)
CCl_4 + FP (600 mg/kg)	106 ± 5.85* (71.5%)	70.0 ± 3.86* (93.2%)	5.10 ± 0.28 (32.2%)	30.5 ± 1.68* (63.1%)
CCl_4 + FP (900 mg/kg)	107 ± 5.91* (70.8%)	75.0 ± 4.14* (91.8%)	5.30 ± 0.29 (21.8%)	30.9 ± 1.70* (61.7%)
CCl_4 + S (50 mg/kg)	88.0 ± 4.86* (85.6%)	55.0 ± 3.04* (97.1%)	3.90 ± 0.21 (94.7%)	22.0 ± 1.21* (93.2%)
F value (at 5% level)	29.6@	231@	2.42ns	19.1@

Abréviations: CCl_4= Carbon tetrachloride; FP= *F. parviflora*;S= Silymarin; AST= Aspartate aminotransaminase, ALT=Alanine aminotransaminase; %= Percent protection. ANOVA @ = Significant; ns= non significant; Values are mean ± S.E., N = 6. # $P \leq 0.05$ *vs* Control, * $P \leq 0.05$ *vs* CCl_4

Tissue biochemical assay

The effect of FP extract on tissue biochemistry is presented in Table 2. CCl_4treated group animals showed significant altered the levels of lipid peroxidation, reduced glutathione (GDH) andG-6-Pase and ATP when compared with the control group ($P \leq 0.05$). Enhanced level of lipid peroxidation by CCl4 exposure was significantly declined by the administration of FP extract due to its free radical scavenging activity($P \leq 0.05$).All three doses of FP extract improved the GSH level and the levels of enzyme activities of G-6-Pase and ATP as well when compared with negative

group. However, The lower dose 300mg/kg was found to be significant protective followed by 600 and 900 mg/kg doses(P≤0.05).

Table 2 Effect of *F. parviflora* against CCl_4 induced hepatotoxicity in tissue biochemistry.

Table 2 Effect of *F. parviflora* against CCl_4 induced hepatotoxicity in tissue biochemistry.

Treatments	LPO (n moles TBARS/mg protein)	GSH (µ mole/g)	ATPase (mg Pi/100 ml/min)	G-6-Pase (µmole Pi/min/g liver)
Control	0.27 ± 0.01	7.91 ± 0.43	1998± 110	5.56 ± 0.30
F. parviflora per se	0.28 ± 0.01	7.91 ± 0.43	1997 ± 110	5.57 ± 0.30
CCl_4 (Negative control group)	1.51 ± 0.08#	4.11 ± 0.22#	1088 ± 60.1#	3.31 ± 0.18#
CCl_4 + FP (300 mg/kg)	0.56 ± 0.03* (76.6%)	7.30 ± 0.40* (83.9%)	1781 ± 98.4* (76.1%)	5.28 ± 0.29* (87.5%)
CCl_4 + FP (600 mg/kg)	0.61 ± 0.03* (72.5%)	7.20 ± 0.39* (81.3%)	1755 ± 97.0 * (73.2%)	5.25 ± 0.29* (86.2%)
CCl_4 + FP (900 mg/kg)	0.69 ± 0.03* (66.1%)	7.10 ± 0.39* (78.6%)	1748 ± 96.6* (72.5%)	5.19 ± 0.28* (83.5%)
CCl_4 + S (50 mg/kg)	0.31 ± 0.01* (96.7%)	7.40 ± 0.40* (86.5%)	1910 ± 105* (90.3%)	5.50 ± 0.30* (97.3%)
F value (at 5% level)	60.7@	9.93@	7.30@	7.14@

Abréviations: CCl_4= Carbon tetrachloride; FP= *F. parviflora*; S= Silymarin; LPO= Lipid peroxidation; GSH= Reduced glutathione; ATPase= Adenosine triphosphatase; G-6-Pase= Glucose-6-Phosphatase; %= Percent protection.
ANOVA @ = Significant; Values are mean ± S.E., N = 6. # P ≤ 0.05 vs Control, * P ≤ 0.05 *vs* CCl_4.

Hexobarbitone induced sleep time

CCl_4 administration significantly prolonged the barbiturate induced sleep time (P≤0.05) when compared to normal control group. Significant prolongation in sleeping time in CCl_4 treated group was shortened significantly after administration of *F. parviflora*when compared to the normal control group (Figure 3).

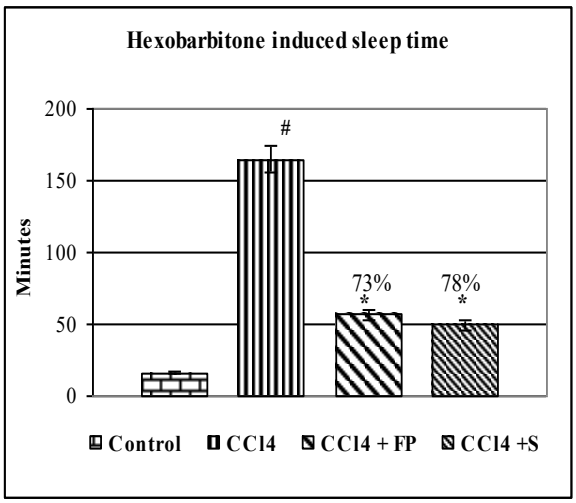

Figure 3 Effect of *F. parviflora* Aquoes extract on Hexobarbitone induced sleep time

Bromosulphalein retention time

BSP retention time after 30 min. of bromoslaphein in injection in normal animals was significantly increased (P≤0.05) after carbon tetrachloride administration. BSP retention time was significantly reduced by the treatment of *F. parviflora* significantly (P≤0.05) indicating improved excretory capacity of liver. Similar results were observed in silymarin treated group (Figure 4).

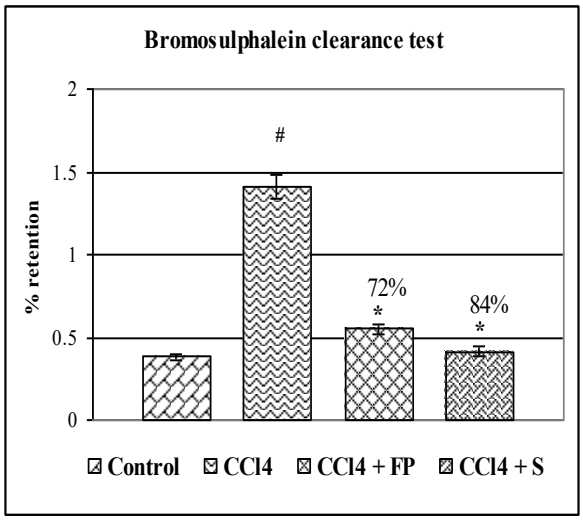

Figure 4 Effect of *F. parviflora* on Bromosulphalein clearance test

ANOVA	Hexobarbitone induced sleep time	BSP
F values	142@	221@

@ = Significant at 5% level

Histological observation

Histological examination of liver showed the clearly hepatic lobules with well-formed hepatocytes and identifiable portal triads. Hepatic cells were grouped in a cord-like pattern, with sinusoids separating them, and the central vein was clearly visible (Figure5a). Fig. 5b showed severe fatty alterations, focal necrosis, ballooning, and degeneration in their hepatic plates, as well as a lack of celluloid borders in CCl_4treated group. Light micrograph of Figure5c exhibited normal architecture of hepatocyte at 300 mg/kg dose of *F. parviflora* indicated significant reduction of structural change against CCl_4 intoxication. The slight degree of necrosis and degeneration was also observed with the treatment at 600 and 900 mg/kg when compared with negative control group indicated mild protective effect of FP (Figure 5d–e). Silymarin-CCl_4treated group showed normal cellular morphology with well-formed, symmetrically placed, and spaced apart by sinusoids in a cord pattern when compared with control group (Figure5f). However, a lower dose (300mg/kg) of the FP therapy was found to be more effective as similar as positive control

Silymarin against CCl_4-induced histological damage.

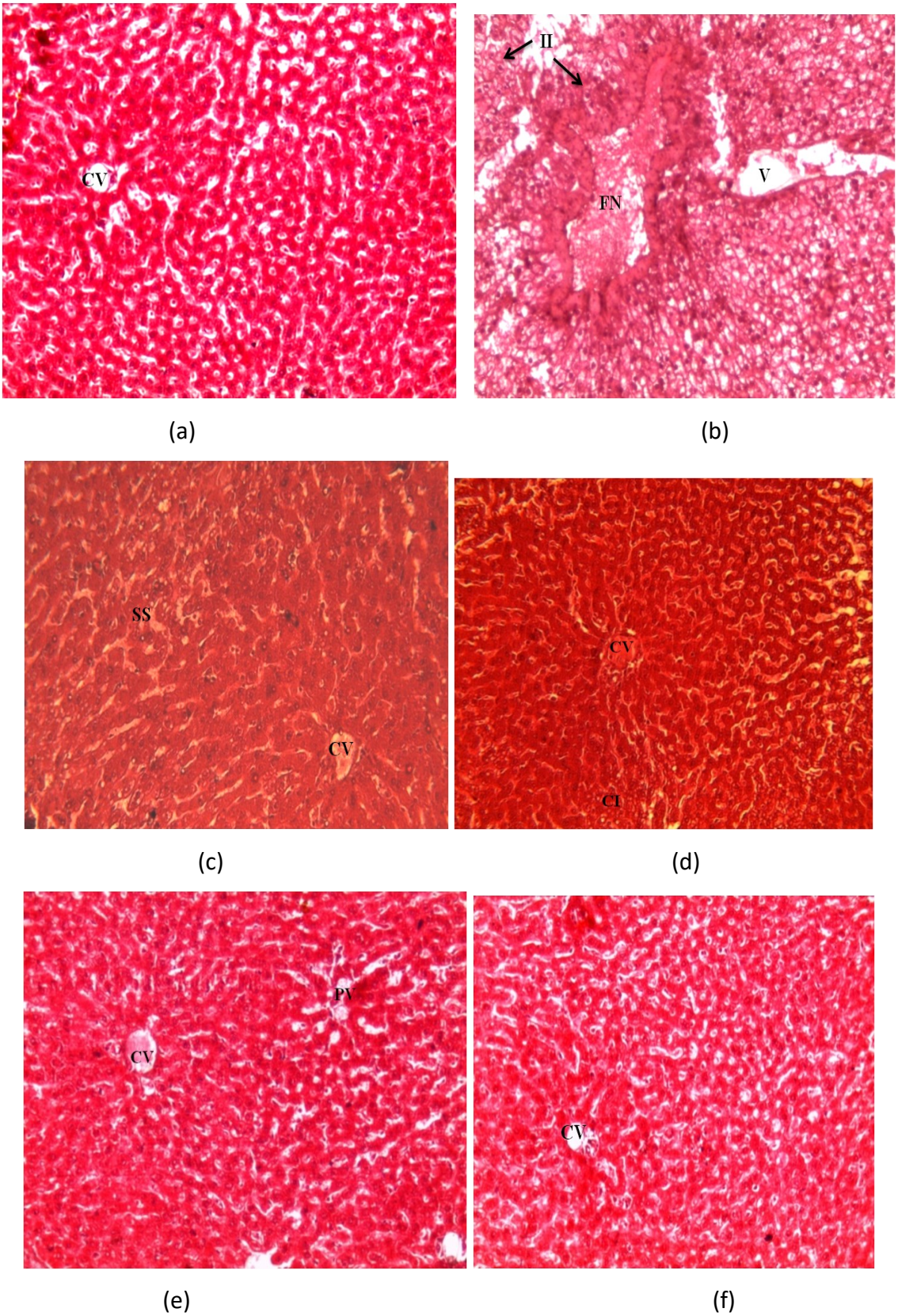

Figure 5 Effect of FP aqueous extract on CCl_4 induced liver toxicity (Histological observation-100X)

Explanation of figure 5 (100X).

a. Showing normal lobular architecture of hepatocytes with clear central vein.

b. Animal treated with CCl_4 showed focal necrosis, vacuolization, inflammatory inflammation and hepatocytes degeneration.

c. *F. parviflora* (300mg/kg) and CCl_4 treated group showed no necrosis, distinct hepatic cells, sinusoidal spaces and necrosis recovery around the central vein showing regenerative activity.

d. *F. parviflora* (600mg/kg) and CCl_4 treated group showed mild cytoplasmic vacuolisation and granulation in hepatocytes.

e. Treatment of *F. parviflora* (900 mg/kg) showed mild improvement in chord arrangement but perinuclear vacuolisation was visible.

f. Treatment of silymarin with CCl_4 exhibited almost normal cellular architecture with proper central vein.

NB: CV: Central vein, V: vacuolation, II: Inflammatory infiltration, FN: Focal necrosis, SS: sinusoidal spaces, PV: Perinuclear vacuolization, CI: Cytoplasmic infiltration

DISCUSSION

CCl_4 is frequently used to screen the hepatoprotective properties of plant extracts in animal model. In current investigation, antioxidant and hepatoprotective activity of *F. parviflora* was evaluated against CCl_4-induced hepatotoxicity. It is reported that the liver cytochrome P450 enzyme converts CCl_4 into trichloromethyl free radical (CCl_3•) and trichloromethyl peroxy (CCl_3O_2•) radicals which induces lipid peroxidation and causes free radical generated liver toxicity[22]. Free radical scavenging activity of *F. parviflora* extract showed dose dependent DPPH inhibition due to the presence of alkaloid and phenolic compounds in it [23-24]. This indicates that plant extract has an antioxidant activity to scavenge CCl_4 reactive metabolites (CCl_3•).

Liver function tests-serum transaminases (AST and ALT) are the main markers of liver damage [25-26]. In the present study, CCl_4 exposure to experimental animal significantly increased the level of serum marker enzymes, AST and ALT into blood circulation due to membrane leakage caused by CCl_4. CCl_4 exposure also altered the level of albumin[27]. Treatment with *F. parviflora* restored the level ofhepatic enzymes i.e. AST, ALT and albumin protein towards normal due to inhibition of membrane leakage. Research studies suggested that there is an inverse relationship between the levels of GSH and lipid peroxidation [28-29]. In our findings, the level of lipid peroxidation was increased and GSH level was diminished after single dose of CCl_4 exposure and these levels were reversed toward normal after administration of FP

indicating its anti-lipidperoxidative activity. The lower dose at 300 mg/kg was found to be significant protective in restoration of these above biochemical parameters followed by higher doses (600 mg/kg and 900 mg/kg) of FP against CCl_4 induced liver damage. The same protection was noticed in silymarin treated group. *Cytisus scorparius* extract is also substantiated our findings[30]. CCl_4 administration also decreased the level of enzymatic activities of ATPase and Glucose-6-Pase which indicated loss of energy (ATP) in the liver. Glucose-6-Pase is an important enzyme involved in regulation of the blood glucose level while ATPase is a mitochondrial enzyme associated with the energy production and transport of intracellular iron. The *F. parviflora*at lower dose (300 mg/kg) reversed the level of the enzyme activity of ATPase and G-6-Pase enzyme declined by CCl^4 which might be due the presence of H_2O_2 scavenging alkaloids in FP extract. Our findings were also supported with the *Rhoicissus tridentate* and *Emblica officinalis*[31-32]. Such protective findings on biochemical parameters were also supported with the histological observations. Light micrograph of CCl_4 treated liver section showed the cellular degeneration, loss of cytoplasm and cord disarrangement which were reversed towards normal control groups after treatment of FP therapy at all doses but lower dose of FP showed significant improvement in cellular morphology compared to the negative control group. In addition, BSP retention time is an important parameter for assessment of excretory ability of the liver cells[33]. Reduction in BSP retention by FP therapy suggested an improvement in the capacity of injured liver to execute its normal function against increased BSP retention time by CCl_4 exposure. The restoration of BSP retention time by FP therapy might be due to its antioxidant nature. Hexobarbitone induced sleep time is regarded as a reliable indicator of hepatic MDME activity [34]. The considerable increased the level of hexobarbitone induced sleep time after CCl_4 treatment in the current study suggests a decreased activity of microsomal drug metabolising enzyme such as CYP4502E1. This finding is supported by Bhadauria *et al*., 2007 [35]. This prolonged hexobarbitone sleep time was decreased by *F. parviflora*, indicating its protective effect on the MDME activity. This protective effect of FP might due its free radical scavenging phenolic contents which may be involved in hepatoprotection against CCl_4-induced toxicity. Other researchers have also reported the same finding in *Eclipta alba* extract [36].

CONCLUSION

The result of the present study suggests that *F. parviflora* aqueous extract has antioxidant and hepatoprotective potential against CCl_4 induced hepatic toxicity which might be due its free radical scavenging activity. Thus, in conclusion, this study indicates that FP extract may be considered as an alternative hepatoprotective plant in the treatment of liver diseases.

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CONFLICTS OF INTEREST

All authors declared no conflict of interest.

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