

Full Length Research Paper

Hepatoprotective and free radical scavenging activities of methanol extract fractions of *Capparis decidua* Edgew (Forssk.) (Capparidaceae)

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The main objective of the study is to evaluate the hepatoprotective and antioxidant activities of different extracts and fractions of a Sudanese traditionally used plant, *Capparis decidua* Edgew (Forssk.) (Capparidaceae). Carbon tetrachloride (CCl₄) induced hepatotoxicity in rats, and DPPH radical scavenging activity (RSA) were the methods used to evaluate the hepatoprotective and antioxidant activities of this plant respectively. The rats were injected i.p. with dichloromethane and methanol 80% extracts at 250 and 500 mg/Kg body weight by the *C. decidua* stem. It was found that the methanol extract showed significant hepatoprotective activity when compared with control group (CCl₄ group); also its liver function parameters were as effective as the positive control (Silymarin group). Fractionation of the methanol extract of *C. decidua* stem was applied by using dichloromethane (DCM), ethyl acetate, n-butanol and aqueous solvents. The fractions were tested on CCl₄- induced hepatotoxicity rats at dose 500 mg/kg body weight where ethyl acetate, DCM and n-butanol fractions showed significant hepatoprotective activity when compared with control group (CCl₄ group); their liver function parameters were within the normal ranges as well as the standard drug group (silymarin group). DPPH radical scavenging activities (RSA) of DCM, ethyl acetate and n-butanol fractions were 86, 92 and 53% respectively. These findings prove the high linkage between hepatoprotective and antioxidant activities. The aqueous fraction showed very weak hepatoprotective property, and antioxidant activity (14% RSA).

Key words: *Capparis decidua* Edgew (Forssk.), methanol extract fractions, hepatoprotective activity, CCl₄, DPPH.

INTRODUCTION

Medicinal plants have been extensively used by mankind in health care system from the earliest history. They are considered as a great warehouse of bioactive compounds which are stored and ready in use with

various pharmacological effects. Therefore, they play an important role and are considered as the main cornerstone in drug discovery and development (Frag et al., 2015; Yadav and Agarwala, 2011; Fabricant and

Fransworth, 2001).

Liver diseases are widely spread over the world. Statistically, there are about 2 million deaths per year, mainly due to complications such as cirrhosis, viral hepatitis and hepatocellular carcinomas; hence liver diseases are considered as one of most important globally burden disease (Asrani, 2019).

Capparis decidua Edgwe (Forssk.) (Capparidaceae) is a perennial woody medicinal plant which is widely distributed in tropical and sub-tropical regions (Dhakad et al., 2016). It is locally known as *Al-toundub*. The plant is traditionally used in treating many ailments, such as rheumatism, asthma, cough, lumbago, toothache, pyorrhea, dysentery, liver infections, diarrhea, febrifuge, cardiac troubles, constipations, ulcers, piles, renal disorders and skin diseases (Nazar et al., 2020; Haq et al., 2011). *C. decidua* has many pharmacological uses. Some of these uses are anti-bacterial, anti-fungal, anti-viral, anthelmintic, anti-termite, insecticidal, anesthetic, anti-hemolytic, antioxidant, anti-diabetic, anti-rheumatic, anti-gout, anti-arthritis, anti-platelet aggregation, hypolipidimic, anti-aging, anti-atherosclerotic, anti-inflammatory, analgesic and nociceptive activities (Nazar et al., 2020; Dev et al., 2015; Mohammed et al., 2014, 2012; Rathee et al., 2012, 2010; Upadhyay et al., 2010; Chahlia, 2009; Sharma and Kumar, 2008; Purohit and Vyas, 2006).

Traditionally, *C. decidua* has been used in treating jaundice (Singh et al., 2011; Aghel et al., 2007). The hepatoprotective activity of its stem part had been proved; its methanolic and aqueous extracts were tested orally at 200 and 400 mg/Kg on CCl₄- induced hepatotoxicity rats for 10 days, and they showed significant hepatoprotective activity through declining the serum levels of aspartate amino transferase (AST), alanine amino transferase (ALT), alkaline phosphatase (ALP) and total bilirubin (TB). The hepatoprotective activity may be due to the presence of flavonoids, tannins, coumarins, sterols, saponins and alkaloids in the stem part of *C. decidua* (Ali et al., 2009). Therefore, the objective of this study is to follow the bio-guided fractionation assay of the most active extract(s) in order to isolate and structurally determine the most active compound(s) responsible for hepatoprotective activity in the future.

MATERIALS AND METHODS

Plant material

The plant material was collected from El-Samrab and Shambat

(Khartoum North) on June 2017 and September 2019 respectively, and was identified by the taxonomist, Dr. Yahia Sulaiman in the herbarium of Medicinal and Aromatic Plant Research Institute, Khartoum, Sudan. The voucher specimens were deposited in Medicinal and Aromatic Plant Research Institute Herbarium (Sudan).

Procurement and preparation of rats

The protocol for this study was approved by the Ethical Committee of the Faculty of Pharmacy, Department of Pharmacology, International University of Africa, Sudan. The document permission no. (IUA,IACE/EXP.Ph.018) was issued on April 2018 and renewed again on December 2019. Rats (both sexes) were obtained from the Animal House of Faculty of Pharmacy, International University of Africa (75- 150 g). All rats were housed under standard laboratory conditions inside suitable cages at room temperature (8-20°C), humidity (about 50%). The rats were exposed to 12 h light/dark cycle and were fed on standard diet and water as required by the Animal House.

Preparation of plant extracts

After collection, authentication, drying and powdering of *C. decidua* stems (1,580 g), extraction process was carried out by maceration; first by dichloromethane (DCM) and next by methanol 80%. The extraction process was repeated for both solvents three times by changing with fresh solvent each time. The extracts were then dried under reduced pressure; yield percentages were calculated and then were kept in tight closed containers for biological assay.

Fractionation of methanol extract

About 100 g of methanol extract was taken and dissolved in distilled water (up to 100 ml). The solution was transferred into separating funnel (500 ml) and DCM was added (about 100 ml). The mixture was shaken gently and allowed to stand till two immiscible layers were separated. The DCM layer was removed and the process was repeated by adding another fresh DCM, several times till the DCM layer became colorless. Next ethyl acetate was added to the aqueous layer and same process was done with DCM applied, after the ethyl acetate layer became colorless. n-butanol was added to the aqueous layer and same process was done with DCM applied. Finally four fractions were obtained from the methanol extract (DCM, ethyl acetate, n-butanol and aqueous fractions). Each fraction was dried under reduced pressure; yield percentages were calculated and then stored in tight closed containers for biological assay.

Hepatoprotective activity model (for DCM and methanol extracts)

The hepatoprotective activity was applied according to the method used by Ali et al. (2009) with mild modifications. Wistar albino rats (75- 150 g/ both sexes) after adaptation period were randomly divided into six groups, four rats per group. Two doses (250 and

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500 mg/ kg) from *C. decidua* stem extracts were tested against CCl₄ induced hepatotoxicity in rats. Silymarin was used as positive control. the groups were: Group A: received only liquid paraffin vehicle (0.2 mg/ kg/ day i.p. for 10 days); Group B: received CCl₄- induced hepatotoxicity (0.2 mg/kg /day i.p. in liquid paraffin (1:9) for 10 days); Group C: served as hepatoprotective drug control, the rats received CCl₄ (0.2 mg/ Kg/ day i.p. in liquid paraffin (1:9) for 10 days) and at the same time they received orally silymarin at dose 100 mg/ Kg/ day; Group D: the rats received CCl₄ (0.2 mg/ kg/ day i.p. in liquid paraffin (1:9) for 10 days) and at the same time they received i.p. *Capparis decidua* methanol extract at 250 mg/ Kg; Group E: the rats received CCl₄ (0.2 mg/ kg/ day i.p. in liquid paraffin (1:9) for 10 days) and at the same time received i.p. *Capparis decidua* methanol extract at 500 mg/ kg and Group F: the rats received CCl₄ (0.2 mg/ Kg/ day i.p. in liquid paraffin (1:9) for 10 days) and at the same time received i.p. *Capparis decidua* dichloromethane extract at 500 mg/ kg.

Dichloromethane extract was dissolved in liquid paraffin, while methanol extract was dissolved in distilled water.

Hepatoprotective activity model of the methanol extract fractions

The hepatoprotective activity was applied to the methanol extract fractions (DCM, ethyl acetate, n-butanol and aqueous fractions) according to the method used by Ali et al. (2009), with mild modifications. Wistar albino rats (90- 150 g/ both sexes) after adaptation period were randomly divided into six groups: four rats per group.

Naming of groups was selected as G1, G2, G3, G4, and G5 in order to differentiate them with the previous hepatoprotective model which was applied with DCM and methanol extracts. Group G1: received CCl₄ to induce hepatotoxicity (0.2 mg/kg /day i.p. in liquid paraffin (1:9) for 10 days); Group G2: the rats received CCl₄ (0.2 mg/ kg/ day i.p. in liquid paraffin (1:9) for 10 days and at the same time received i.p. *Capparis decidua* DCM fraction of methanol extract at 500 mg/ kg; Group G3: the rats received CCl₄ (0.2 mg/ kg/ day i.p. in liquid paraffin (1:9) for 10 days and at the same time received i.p. *Capparis decidua* ethyl acetate fraction of methanol extract at 500 mg/ kg; Group G4: the rats received CCl₄ (0.2 mg/ kg/ day i.p. in liquid paraffin (1:9) for 10 days and at the same time received i.p. *Capparis decidua* n-butanol fraction of methanol extract at 500 mg/ kg; Group G5: the rats received CCl₄ (0.2 mg/ kg/ day i.p. in liquid paraffin (1:9) for 10 days and at the same time they had received i.p. *Capparis decidua* aqueous fraction of methanol extract at 500 mg/ kg.

Collection of blood samples

Blood samples were collected by puncturing rat eyes after day 10. The rats were anesthetized by using diethyl ether after placing the rats inside a desiccator, and then heparinized capillary tubes were used to collect the blood samples. The collected blood samples were centrifuged (5,000 rpm/ 10 min.) and serums were separated. They were stored well in tight containers at -20°C and used for the assessment of liver function tests. The biochemical parameters of liver function tests are alanine amino transeferase (ALT), aspartate amino transeferase (AST), alkaline phosphatase (ALP) and total bilirubin (TB). They were analyzed by using commercial kits (Plasmatic Laboratory Products Ltd., UK) according to their manufacturer's instructions.

Antioxidant activity by using DPPH radical scavenging assay

The DPPH radical scavenging assay was determined according to

Shimada et al. (1992)'s method. About 5 mg from DCM fraction of methanol extract, ethyl acetate fraction of methanol extract, n-butanol fraction of methanol extract and aqueous fraction of methanol extract of *C. decidua* were weighted and dissolved in 1 ml of DMSO. Then, in 96- wells plate the prepared plant samples (to final concentration of 0.5 mg/ ml) were allowed to react with 1, 1-diphenyl- 2- picryl hydrazyl (DPPH) for half an hour at 37°C. The concentration of DPPH was kept as 300 µM (3.72 mg of DPPH was dissolved in 18 ml of ethanol). The positive control propyl gallate (PG) was prepared by dissolving 0.2 mg in 100 µl of distilled water, while the negative control was prepared by adding 10 µl of DMSO to 90 µl of DPPH. After incubation of the plate, a decrease in absorbance was measured at 517 nm by using multiple reader spectrophotometers. The percentage radical scavenging activity by plant samples was determined in comparison to DMSO treated control group which was calculated from the following equation:-

$$\% \text{ inhibition of DPPH radical} = \left(\frac{[A_{br} - A_{ar}]}{A_{br}} \right) \times 100.$$

Where, A_{br} is the absorbance before reaction of the negative control and A_{ar} is the absorbance after reaction for the plant samples.

All tests and analysis were run in triplicate.

Determination of IC₅₀ antioxidant activity

The IC₅₀ (the concentration of the test material which possess 50% inhibition of free radicals) of the most active methanol extract fractions were determined by monitoring the effect of different concentrations of the fractions ranging from 500- 62.25 µg/ ml. The IC₅₀ was calculated using EZ- fit Enzyme Kinetic Program (Perrella Scientific Inc, USA).

Statistical analysis

All obtained hepatoprotective activity results were analyzed using one way ANOVA (Tuckey method of comparison), through using SPSS statistical package application version 20. The p- values less than or equal to 0.05, 0.01 and 0.001 were considered as statistically significant.

RESULTS

Yield percentage

Generally, yield percentage is considered as an important parameter in studying natural products. It is used for determining the desired amount of the extracts or fractions for biological activities and phytochemical screening. Table 1 shows the yield percentages and weights in grams for DCM and methanolic extracts of *C. decidua* stem, as well as its methanol extract fractions.

Hepatoprotective activity results of DCM and methanol extracts

Table 2 and Figures 1 to 8 represent the biological

Table 1. The yield percentage of dichloromethane and methanol (80%) extracts of *C. decidua* stem (1,580 g) and the yield percentage of dichloromethane, ethyl acetate, n-butanol and aqueous fractions obtained from the methanol extract of *C. decidua* stem (100 g).

S/N	<i>C. decidua</i> stem	Weight in grams	Yield percentage (%)
1	Dichloromethane extract	13.746	0.87
2	Methanol extract	164.32	10.40
3	Dichloromethane fraction	7.79	7.79
4	Ethyl acetate fraction	3.27	3.27
5	n-Butanol fraction	22.6	22.60
6	Aqueous Fraction	60.99	60.99

Table 2. Liver function tests results of all groups (mean \pm SD)(n=4).

Group	ALT (IU/ L)	AST (IU/ L)	ALP (IU/ L)	TB (mg/ dl)
	Normal range (52- 224)	Normal range (140- 225)	Normal range (230- 360)	Normal range (0- 0.64)
Group A	57 \pm 9 ^b	187.33 \pm 12.1 ^b	168.33 \pm 71.29 ^b	0.49 \pm 0.03 ^a
Group B	831.67 \pm 296.21 ^d	838.67 \pm 45.79 ^d	733.75 \pm 23.56 ^d	1.215 \pm 0.21 ^d
Group C	56 \pm 4 ^b	131 \pm 3 ^b	228 \pm 8 ^b	0.06 \pm 0.05 ^b
Group D	171 \pm 1.41 ^a	198 \pm 45.25 ^b	277.5 \pm 67.18 ^b	0.275 \pm 0.09 ^a
Group E	104.33 \pm 16.29 ^b	186 \pm 33.06 ^b	253.33 \pm 72.34 ^b	0.17 \pm 0.09 ^b
Group F	453.67 \pm 125.35 ^c	681 \pm 210.46 ^d	976.67 \pm 72.86 ^{a,d}	0.54 \pm 0.39 ^a

Groups: A (control group); B (CCl₄ group); C (standard drug group- silymarin); D (250 mg/kg methanol extract); E (500 mg/kg methanol extract); F (500 mg/ kg dichloromethane extract). ^aMeans that the difference was found to be significant (P- value < 0.01) when compared with Group B. ^bMeans that the difference was found to be significant (P- value < 0.001) when compared with Group B. ^c Means that the difference was found to be significant (P- value < 0.05) when compared with Group C. ^dMeans that the difference was found to be significant (P- value < 0.001) when compared with Group C.

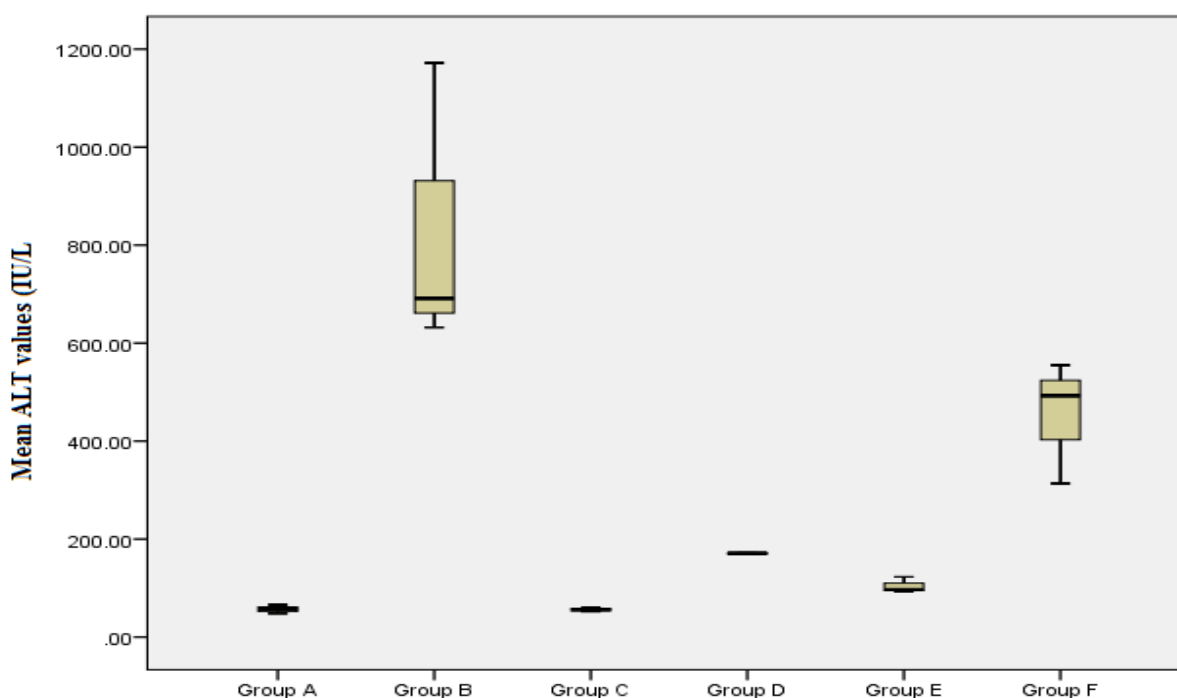


Figure 1. Box plot graph of ALT values (IU/L) of Groups A, B, C, D, E and F.

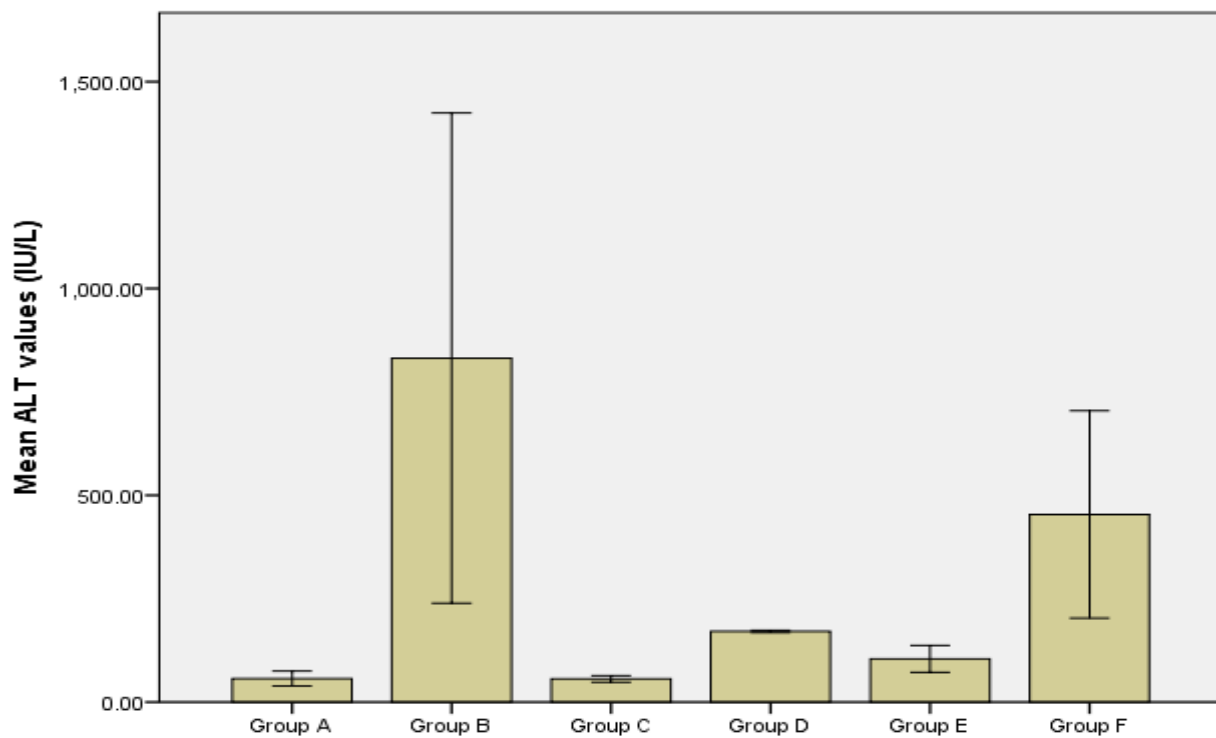


Figure 2. The diagram shows the ALT levels (IU/L) of all groups A, B, C, D, E and F.

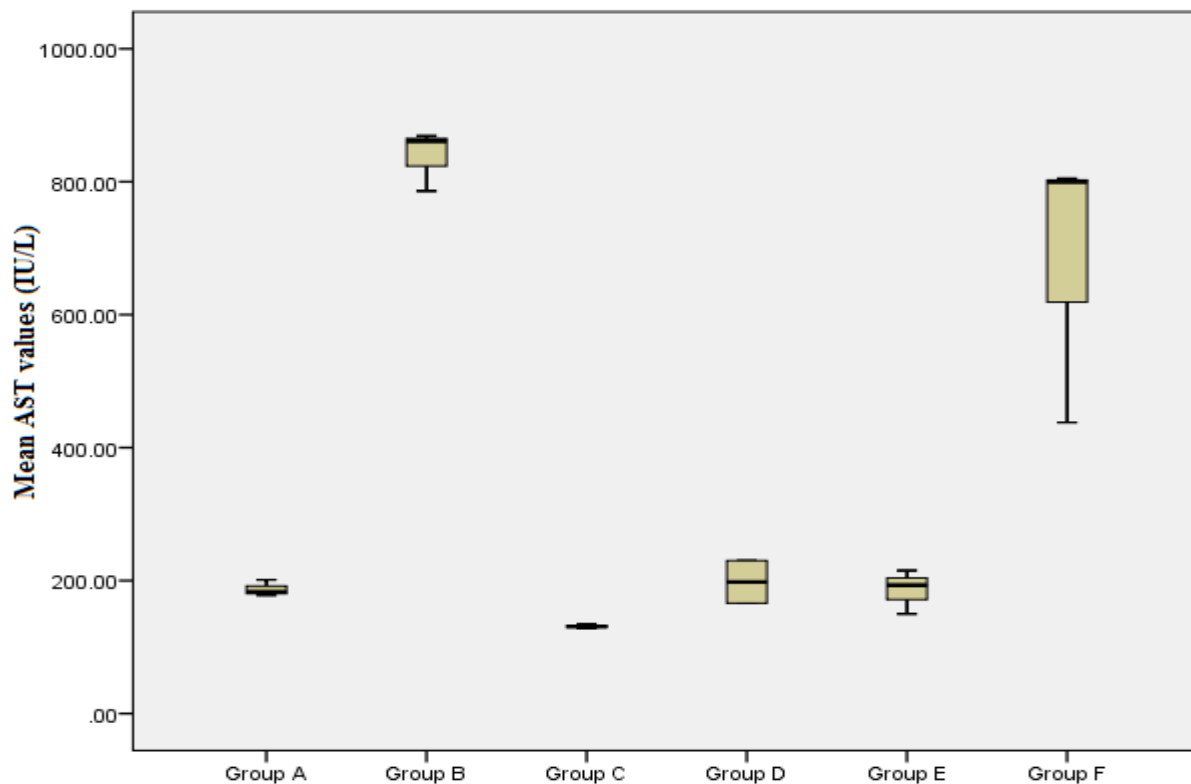


Figure 3. Box plot graph of AST values (IU/L) of Groups A, B, C, D, E and F.

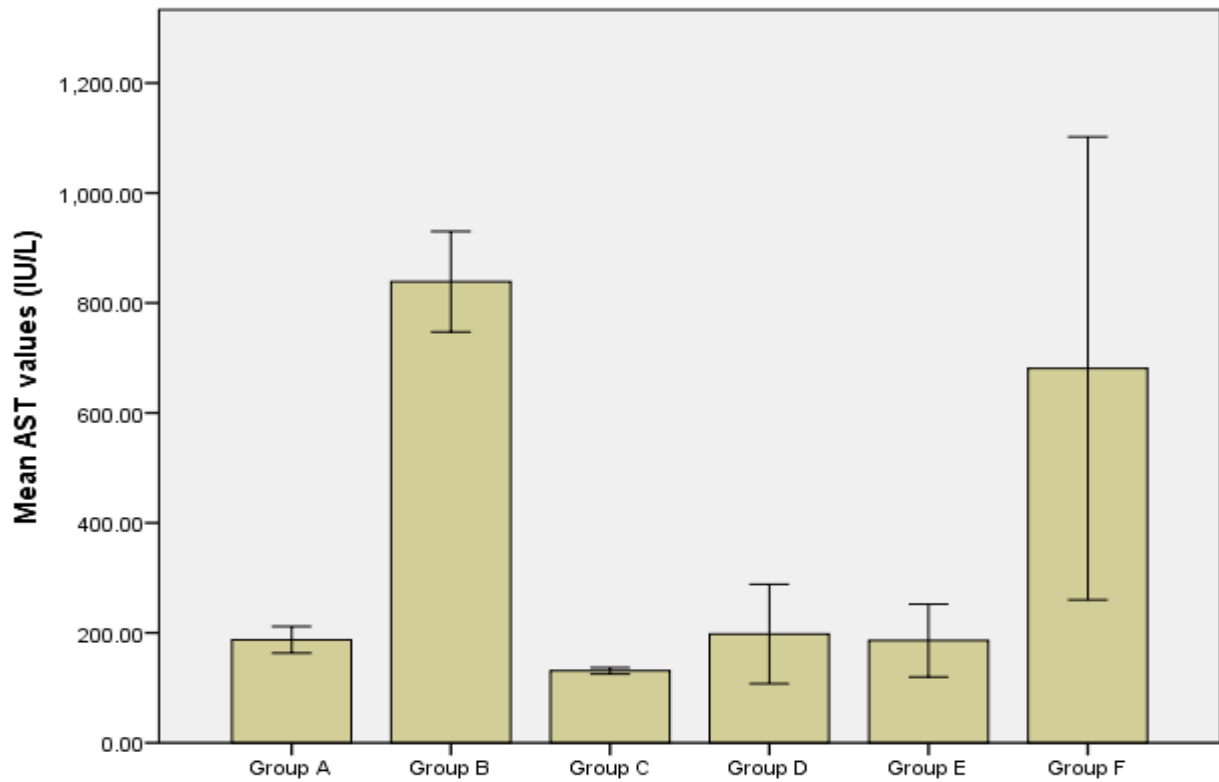


Figure 4. The diagram shows the AST levels (IU/L) of all groups A, B, C, D, E and F.

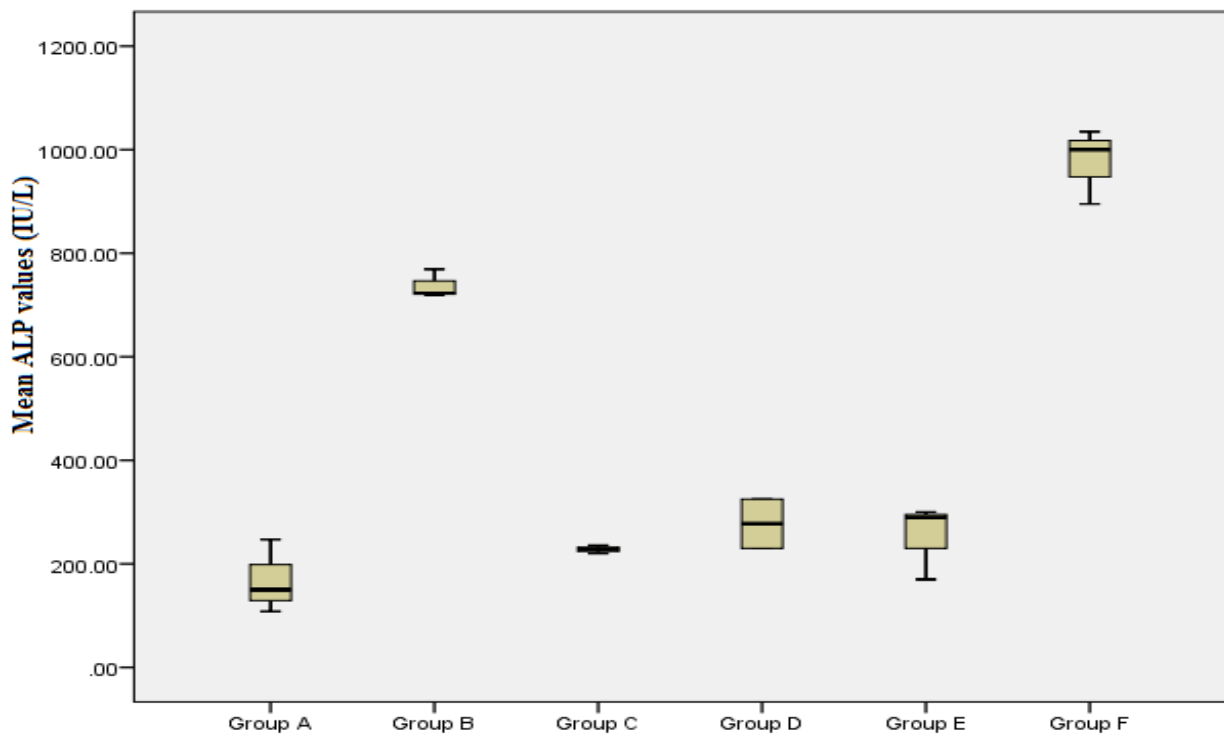


Figure 5. Box plot graph of ALP values (IU/L) of Groups A, B, C, D, E and F.

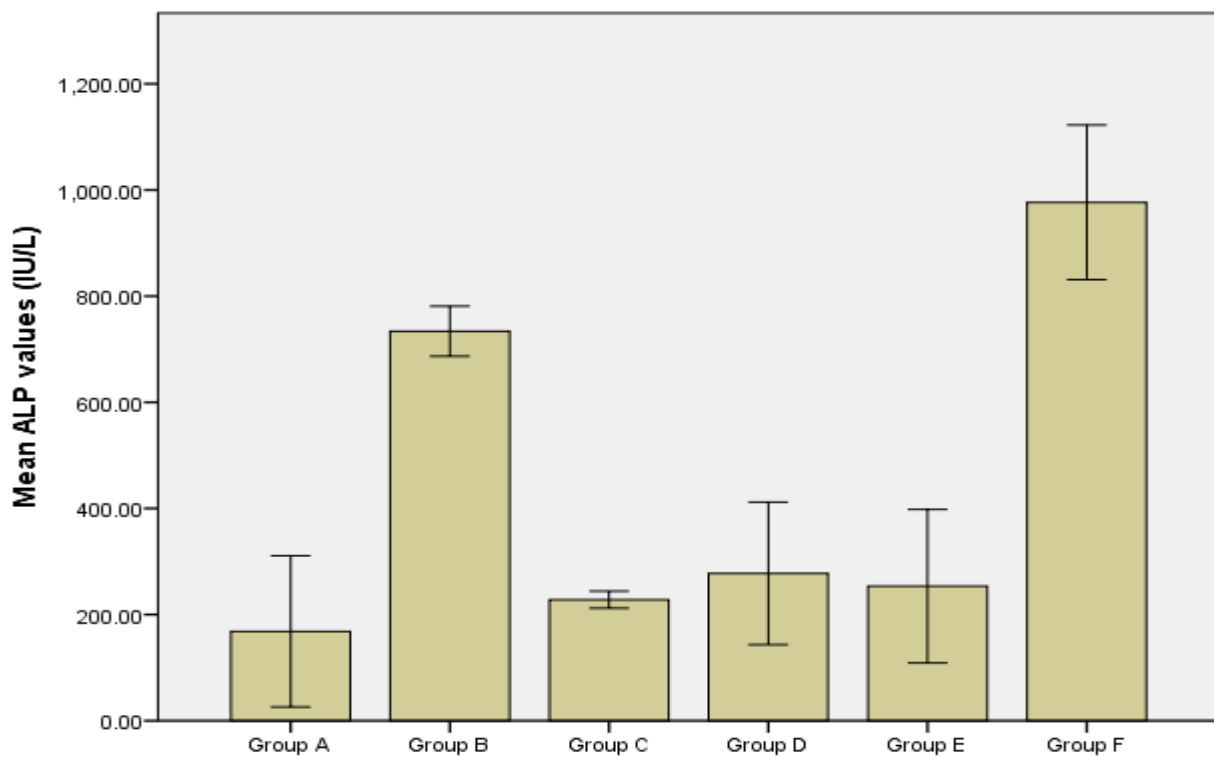


Figure 6. The diagram shows the ALP levels (IU/L) of all groups A, B, C, D, E and F.

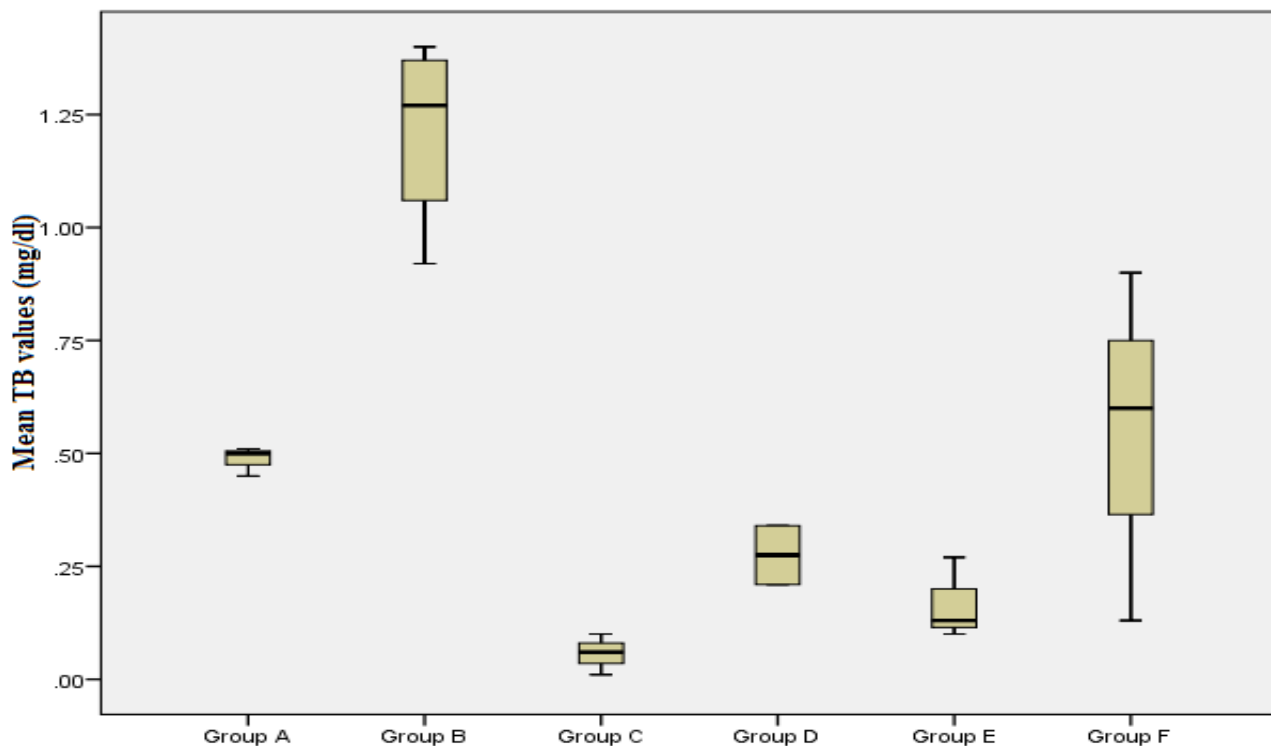


Figure 7. Box plot graph of TB values (mg/dl) of groups A, B, C, D, E and F.

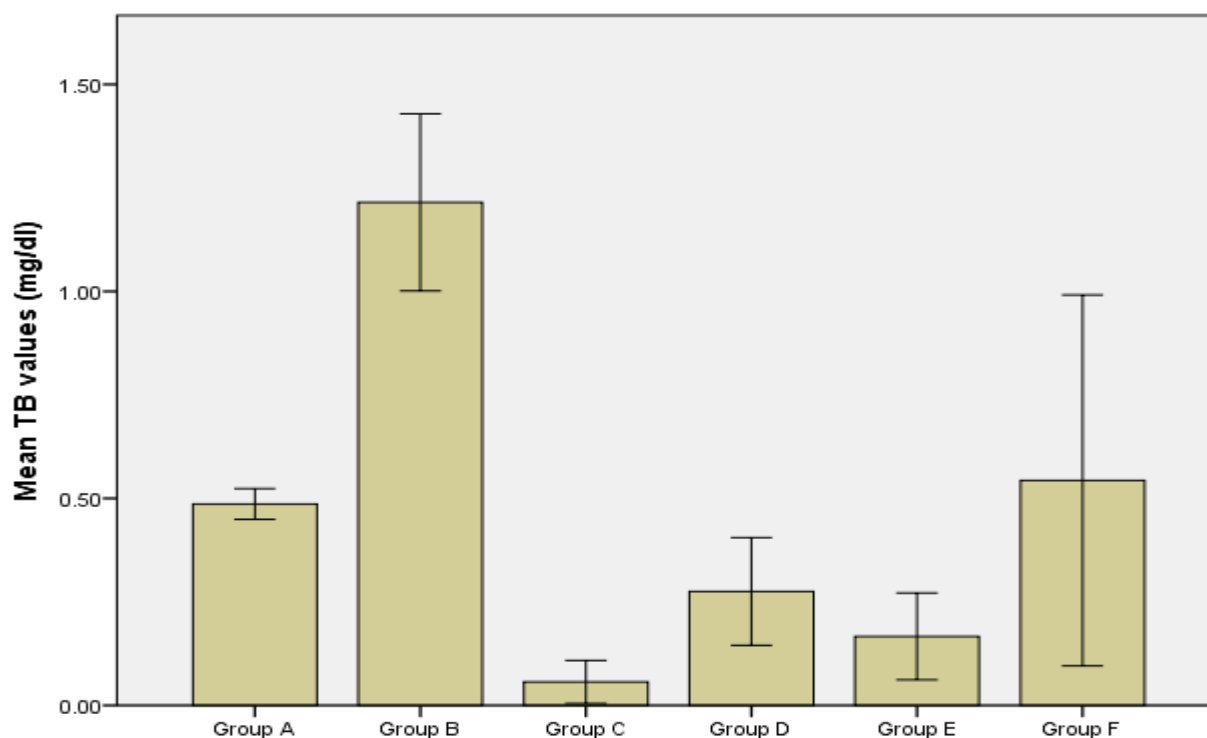


Figure 8. The diagram shows the TB levels (mg/dl) of all groups **A, B, C, D, E** and **F**.

Table 3. Liver function tests results of all groups (mean \pm SD)(n=4).

Group	ALT	AST	ALP	TB
	Normal range (52- 224)	Normal range (140- 225)	Normal range (230- 360)	Normal range (0- 0.64)
Group C	56 \pm 4 ^b	131 \pm 3 ^b	228 \pm 8	0.06 \pm 0.05 ^b
Group G1	831.67 \pm 296.21 ^c	605 \pm 196.77 ^c	201.33 \pm 52.84	1.23 \pm 0.25 ^c
Group G2	52.75 \pm 19.6 ^b	155.25 \pm 60.96 ^b	258.67 \pm 14.36	0.02 \pm 0.01 ^b
Group G3	133.33 \pm 57.74 ^b	107 \pm 16.09 ^b	60.5 \pm 9.19	0.025 \pm 0.01 ^b
Group G4	143.25 \pm 54.52 ^b	202.5 \pm 23.19 ^a	657.25 \pm 85.16 ^{b,c}	0.11 \pm 0.02 ^b
Group G5	1120.67 \pm 107.11 ^c	1066 \pm 176.72 ^{b,c}	160 \pm 74.72	0.11 \pm 0.03 ^b
	IU/ L	IU/ L	IU/ L	mg/ dl

^aMeans that the difference was found to be significant (P- value < 0.01) when compared with Group **G1**. ^bMeans that the difference was found to be significant (P- value < 0.001) when compared with Group **G1**. ^cMeans that the difference was found to be significant (P- value < 0.001) when compared with Group **C**. C: Standard drug group- silymarin, G1: CCl₄ group, G2: 500 mg/Kg dichloromethane fraction group, G3: 500 mg/Kg ethyl acetate fraction group, G4: 500 mg/kg n-butanol fraction group, and G5: 500 mg/kg aqueous fraction group.

parameters results which were obtained from the blood serums isolated from each group.

Hepatoprotective activity results of fractions obtained from methanol extract

Table 3 and Figures 9 to 16 represent the biological

parameters results which were obtained from the blood serums isolated from each group.

Results of antioxidant activity of methanol fractions (by DPPH radical scavenging assay)

Table 4 shows the antioxidant activity results of methanol

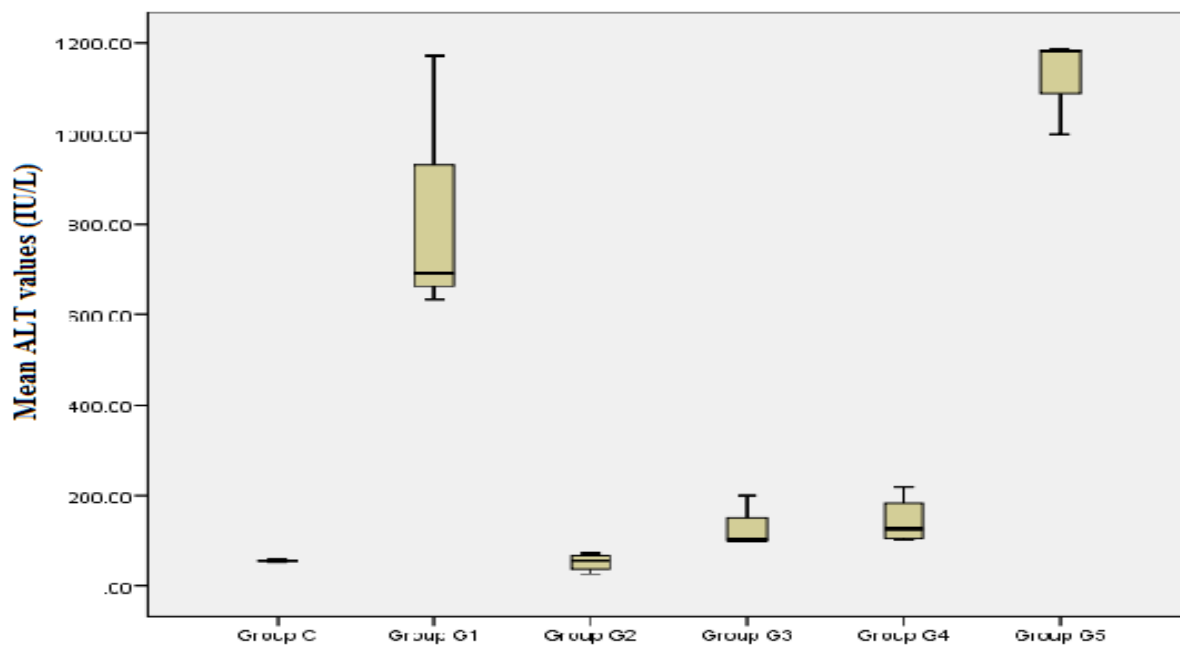


Figure 9. Box plot graph of ALT values (IU/L) of Groups C, G1, G2, G3, G4 and G5.

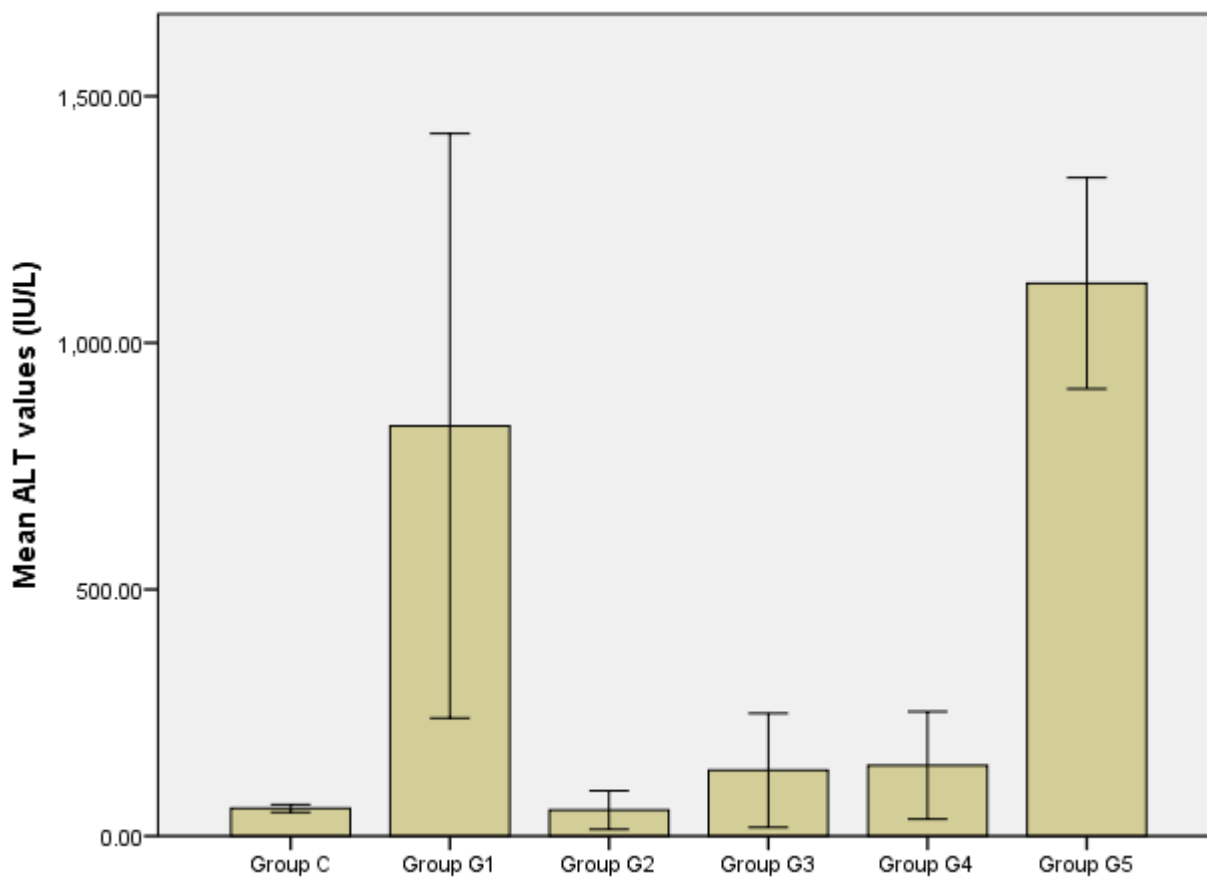


Figure 10. The diagram shows the ALT levels (IU/L) of groups C, G1, G2, G3, G4 and G5.

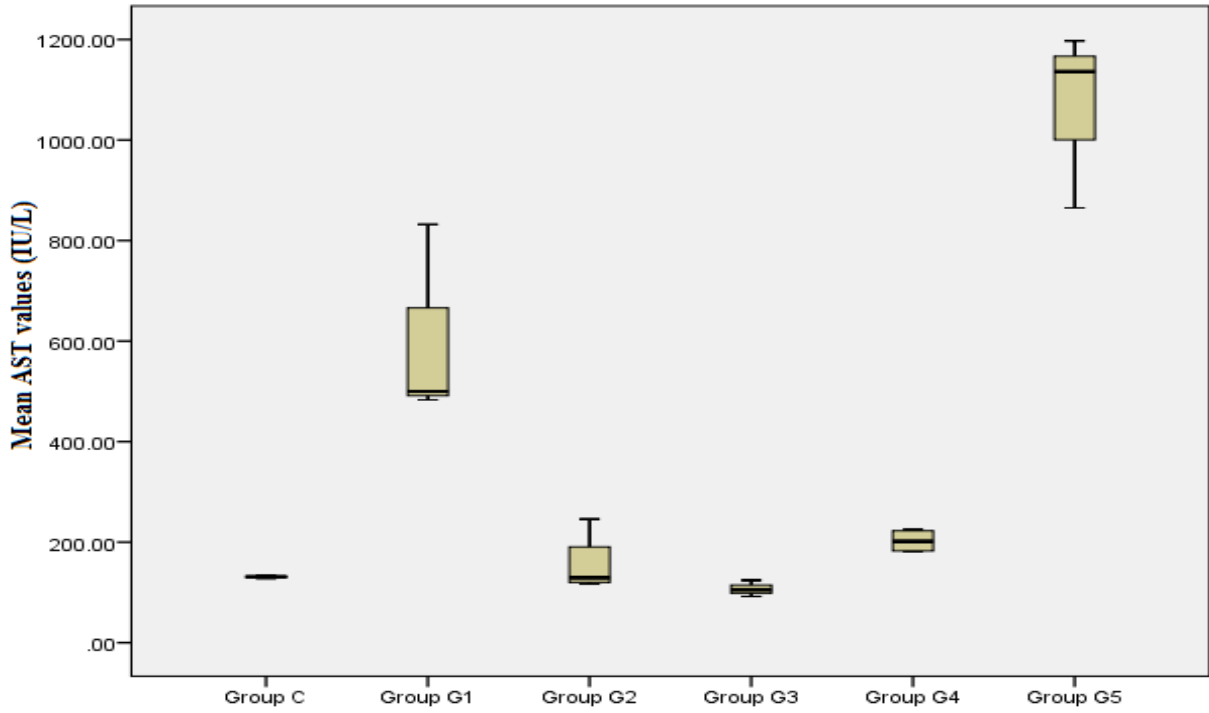


Figure 11. Box plot graph of AST values (IU/L) of Groups C, G1, G2, G3, G4 and G5.

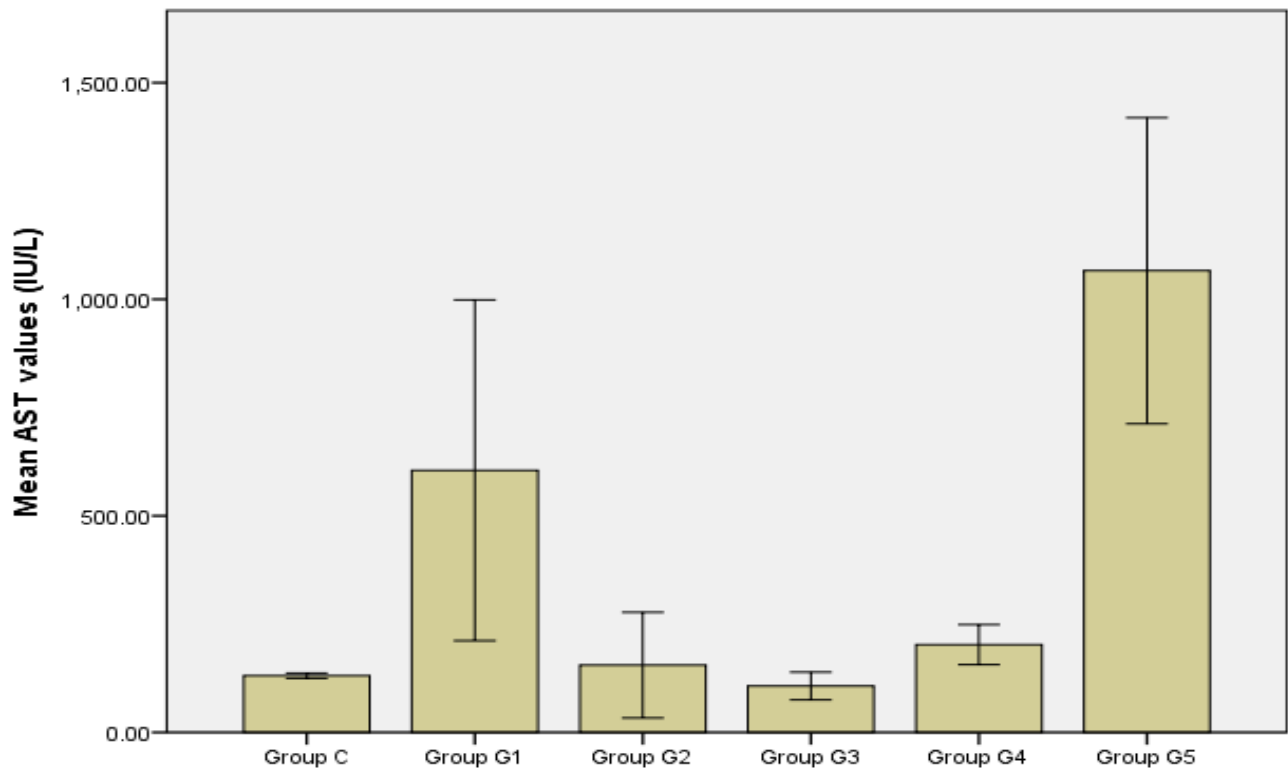


Figure 12. The diagram shows the AST levels (IU/L) of groups C, G1, G2, G3, G4 and G5.

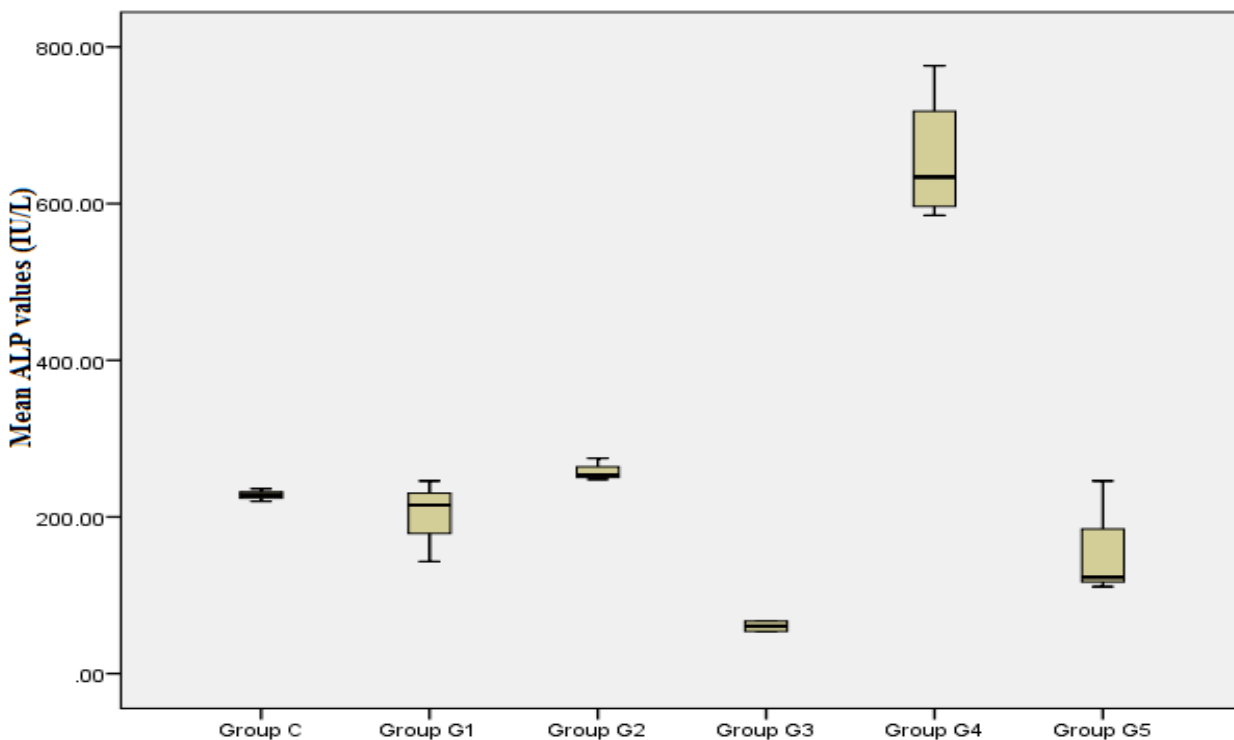


Figure 13. Box plot graph of ALP values (IU/L) of Groups C, G1, G2, G3, G4 and G5.

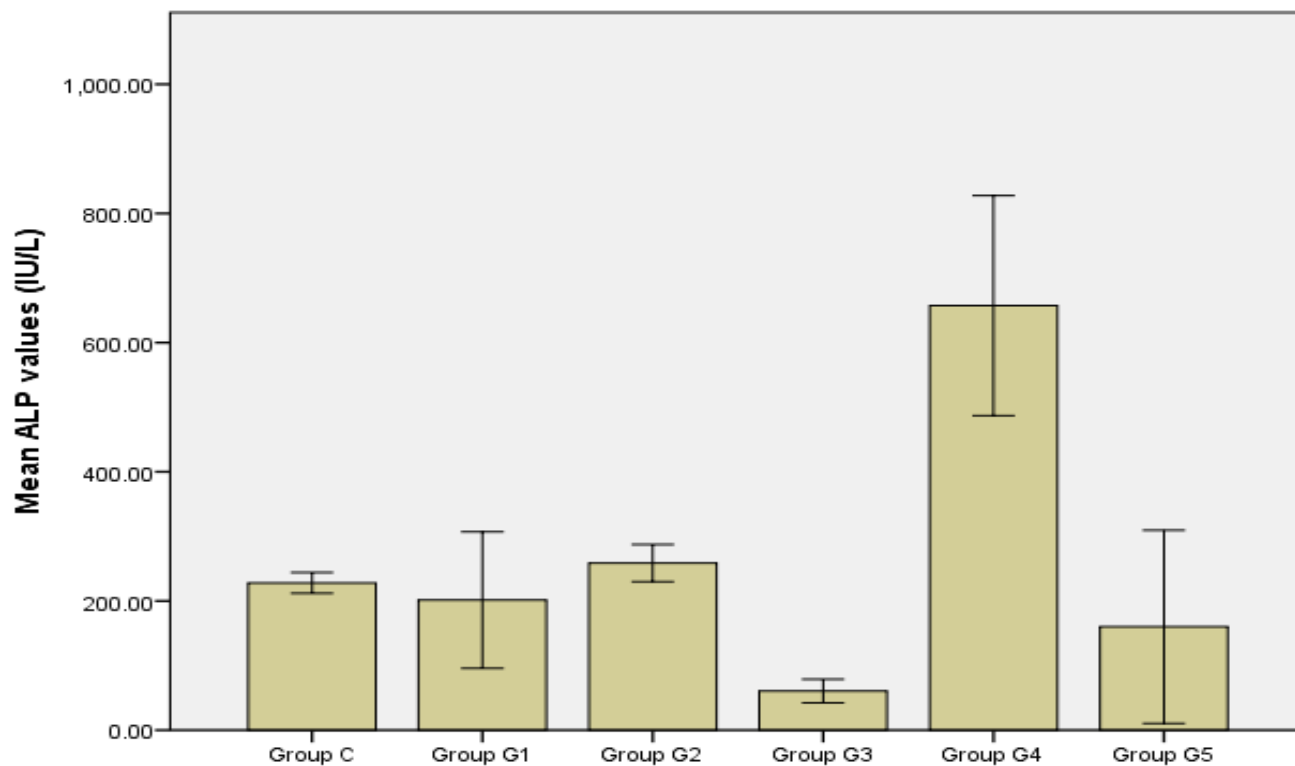


Figure 14. The diagram shows the ALP levels (IU/L) of groups C, G1, G2, G3, G4 and G5.

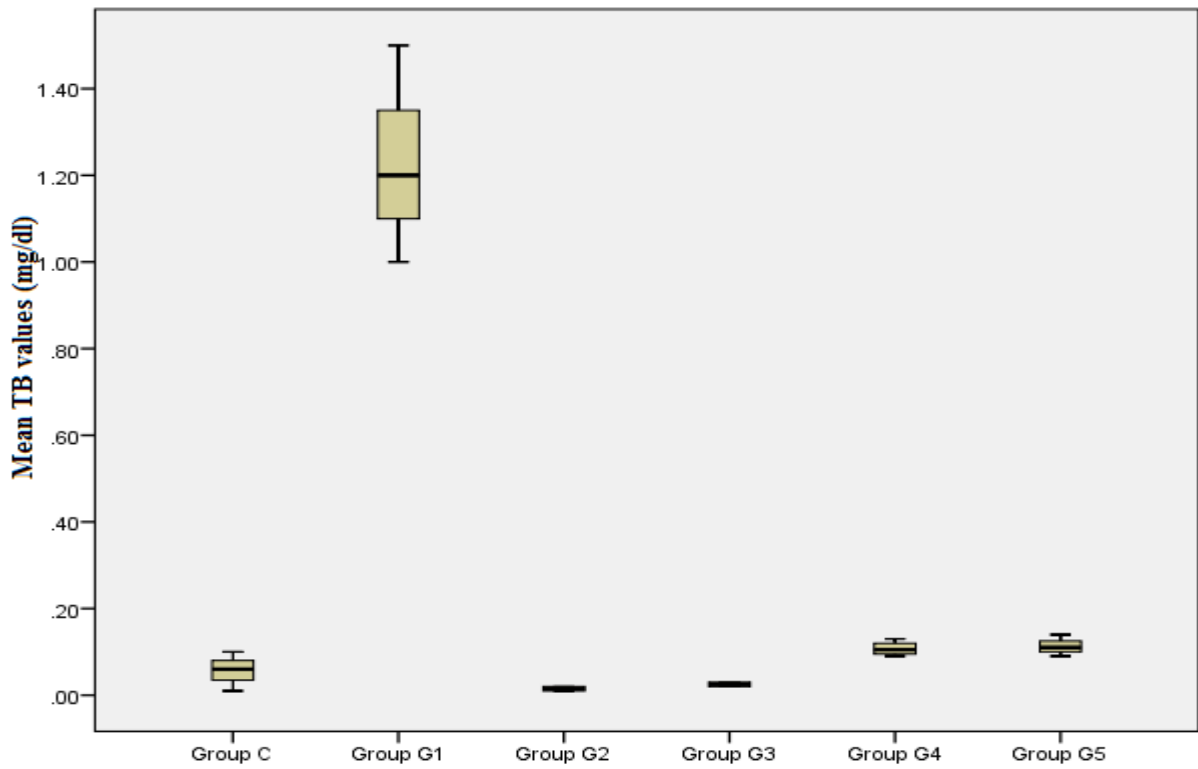


Figure 15. Box plot graph of TB values (mg/dl) of Groups C, G1, G2, G3, G4 and G5.

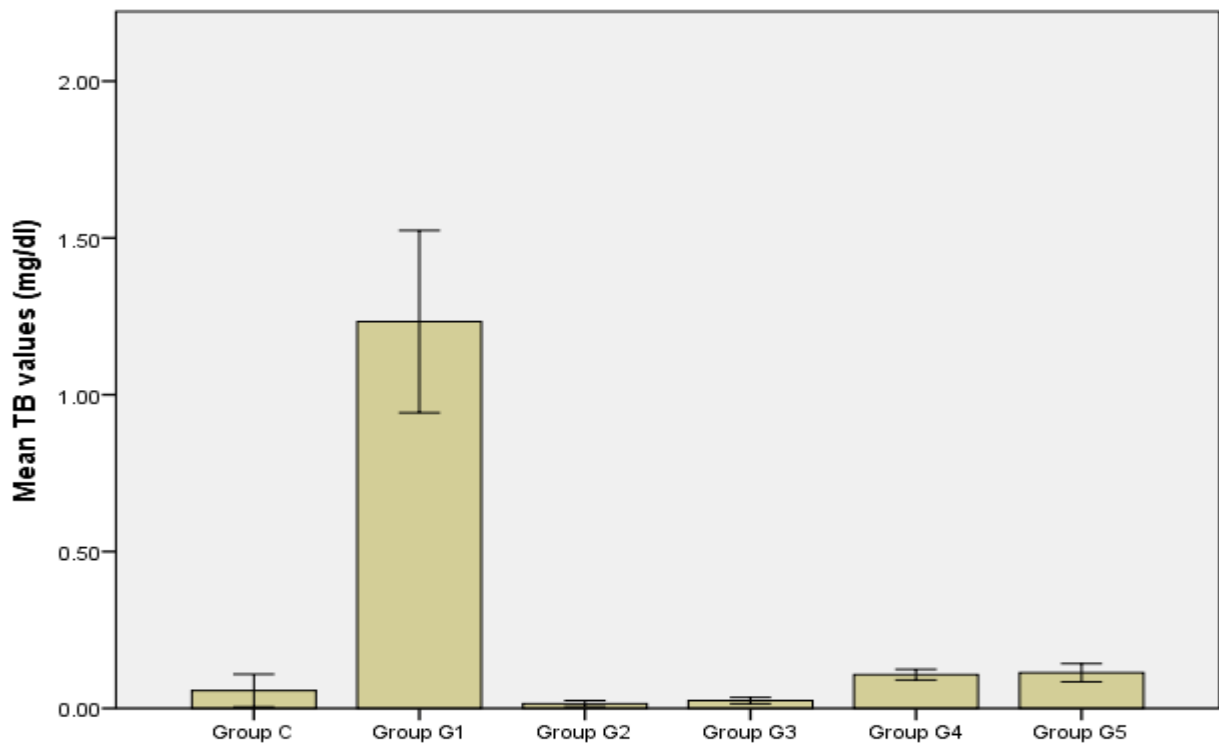


Figure 16. The diagram shows the TB levels (mg/dl) of groups C, G1, G2, G3, G4 and G5.

Table 4. The antioxidant activity of the methanol fractions of *C. decidua*, and IC₅₀ of the most active antioxidant fractions.

S/N	Fraction sample	% RSA (mean ± SD) (DPPH)	IC ₅₀ (mean ± SD) mg/ ml (DPPH)
1	Aqueous fraction	14 ± 0.07	-
2	n-butanol fraction	53 ± 0.09	-
3	Ethyl acetate fraction	92 ± 0.01	0.042 ± 0.3
4	DCM fraction	86 ± 0.02	0.048 ± 0.1
Stan.	Propyl gallate	92 ± 0.01	0.077 µg/ml ± 0.01

extract fractions of *C. decidua*, and the IC₅₀ of the most antioxidant active fractions (the concentration of the fraction which possesses 50% inhibition of free radicals).

DISCUSSION

The yield percentage is a very important parameter in studying natural products; it is used for determining the desired amount of the extract or its fraction for biological activities. Hence, it is used to predict the quantity of any isolated active constituent present in any plant material. In this study, for the general screening two solvents were chosen; non polar (dichloromethane) and polar (methanol 80%) to cover a wide range of phytochemical compounds which are present in *C. decidua* stem part. The weight yields of the methanol and dichloromethane extracts are listed in Table 1. It is obvious that the yield of methanol 80% extract was higher (164.32 g obtained from the total plant material 1,580 g), indicating that the polar phytochemical compounds are abundant. And this observation matches with previous studies carried out on *C. decidua* stem, which showed that many polar phytochemical compounds are present (Ali et al., 2009).

To test the hepatoprotective properties of *C. decidua* stem, the two extracts were tested on CCl₄- induced hepatotoxicity rats' (i.p.) at 250 and 500 mg/kg body weight. The results are shown in Table 2 and Figures 1 to 8. ALT, AST, ALP and TB are the liver function parameters which are used to confirm the hepatoprotective activity, as any increase in their levels above the normal ranges indicates hepatocellular damage due to hepatotoxicity caused by CCl₄. It is obviously seen that the methanol extract at 250 and 500 mg/kg body weight has significant hepatoprotective activity, and the hepatoprotective activity is dose dependent. On the other hand, the dichloromethane (DCM) extract showed very weak hepatoprotective properties at 500 mg/kg body weight, as its liver parameters were above the normal ranges. Our findings match with the work of Ali et al. (2009), in which they used orally methanol and aqueous extracts at doses of 200 and 400 mg/kg body weight. The hepatoprotective activity of *C. decidua* stem was mainly due to the

presence of polar phytochemical compounds in methanol 80% extract as done in this study, or in methanol and aqueous extracts as done in Ali et al. (2009)'s study; so we can safely say that the occurrence of antioxidant polar compounds is probably responsible for the hepatoprotective activity through stabilizing the hepatocellular membranes and hence normalizing the enzymes serum levels of ALT, AST and ALP, as well as the serum level of TB. Many researches demonstrated the linkage between the hepatoprotective property with the antioxidant activity (Ali et al., 2009; Pattanayak and Priyashree, 2008; Satyanarayana et al., 2008; Gupta et al., 2004).

The above mentioned findings encouraged us to start fractionation of methanol extract, by using four solvents starting with nonpolar solvent and ending with polar solvent, that is, dichloromethane, ethyl acetate, n-butanol and aqueous. So the polar phytochemical compounds present in methanol extract were distributed in those selected solvents. The yield of their weights is shown in Table 1. The fractions were tested again in CCl₄- induced hepatotoxicity rats (i.p.) at 500 mg/Kg body weight; the serum liver function parameters were collected and analyzed. The results are given in Table 3 and Figures 9-16. It is obviously seen that DCM, ethyl acetate and n-butanol fractions showed significant hepatoprotective activity when compared with group G1 (CCl₄ control group); their parameters were also within the normal ranges as well as the silymarin group (Group C), except the ALP level of n-butanol group (Group G4); also the hepatoprotective activity of ethyl acetate and DCM fractions (Groups G3 and G2) were higher than the hepatoprotective property of n-butanol fraction (Group G4). This arrangement of the hepatoprotective activity of the 3 fractions also matched with their arrangement of the percentage DPPH radical scavenging activity (antioxidant activity), in which the DCM, ethyl acetate and n-butanol fractions showed 86, 92 and 53% DPPH radical scavenging activity respectively (Table 4). On the other hand, the aqueous fraction does not show any hepatoprotective activity (Table 3 and Figures 9 to 16). Also it showed only about 14% DPPH radical scavenging activity which is considered as very weak antioxidant activity (Table 4). These findings are similar and prove

that the hepatoprotective and antioxidant activities are, in general, linked together.

Conclusion

The methanol extract of *Capparis decidua* stem at 250 and 500 mg/kg body weight has significant hepatoprotective activity, and the hepatoprotective activity was dose dependent. The dichloromethane (DCM) extract of *C. decidua* stem showed very weak hepatoprotective activity at 500 mg/Kg body weight. By linking the above findings with the work of Ali et al. (2009), our results match together in which the hepatoprotective activity was mainly related to the presence of polar active compounds.

Dichloromethane, ethyl acetate and n-butanol fractions obtained from methanol extract of *C. decidua* stem showed significant hepatoprotective activity. Also the hepatoprotective activity of ethyl acetate and DCM fractions was higher than the hepatoprotective property of n-butanol fraction. This arrangement of the hepatoprotective activity of the 3 fractions also matched with their arrangement of the percentage DPPH radical scavenging activity (antioxidant activity), in which the ethyl acetate, DCM and n-butanol fractions showed 92, 86 and 53% DPPH radical scavenging activity, respectively. On the other hand, the aqueous fraction does not show any hepatoprotective activity. Also, it showed only about 14% DPPH radical scavenging activity. These findings are similar and prove that the hepatoprotective and antioxidant activities are, in general, linked together.

Chromatographic methods and techniques must be considered in the future to determine the active constituents present in the active fractions (ethyl acetate, DCM and n-butanol fractions) and to determine the chemical structure of these constituents.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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