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THERAPEUTIC RESPONSE OF ELAEAGNUS PARVIFOLIA'S HYDROETHANOLIC EXTRACT TO RESTORE THE ANTIOXIDANT POTENTIAL, HEPATOPROTECTIVE AND RENOPROTECTIVE EFFECTS.

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Abstract
The goal of this study was to see how albino mice responded to a hydroethanolic extract of *Elaeagnus parvifolia* fruits. Phytochemical analysis, FTIR spectroscopy and HPLC, findings confirmed the existence of functional groups and a wide spectrum of phenolic compounds, respectively. Furthermore, extract did not cause significant hemolysis (5.39±0.176%) when compared to PBS (0.103±0.15%) as a negative control; however, it did have significant clot lysis (21.20 ± 2.25%) potential, as well as antioxidant DPPH scavenging (37.023±0.204%), H_2O_2 scavenging (11.27±0.18%) and reducing power (81.47±0.24%). *In vivo* experiments in albino mice demonstrated that for 28 days giving extract dosages orally, after CCl4 intoxication significantly ($P<0.05$) recovered liver enzymes, renal profiles, lipid profile, serum electrolytes and stress markers, additionally the dosage group (200 mg/Kg b.w.) substantially ($P<0.05$) improved the structural architecture of liver tissue. Elaeagnus parvifolia has a substantial therapeutic response to treat medical difficulties, notably liver disorders, according to the findings.

Keywords: *Elaeagnus parvifolia*, hepatoprotective, renoprotective, antioxidant.

Introduction
Elaeagnus parvifolia belongs to Family Elaeagnaceae and Genus Elaeagnus. Vernacular name is Kankoli and it is a Woody wild shrub, Flowering period May-August. *E. parvifolia* is a significant part of Nepal's flora, as well as Afghanistan, the Himalaya (from Kashmir to Bhutan), Assam, and China. Several species of Elaeagnus, notably *E. parvifolia*, have been isolated of steroids, flavonoids, and triterpenoids, and are regarded folk medicinal plants. Cough and bronchitis have been reported to be cured by its fruits (Singh *et al*.,2014). It is also used as cardioprotective and diuretic. People of hilly areas in Pakistan uses it as a traditional roll and stick forming practice (Khan *et al*., 2010). It is used to treat respiratory and penetrating diseases of patients in Pakistan (Amjad *et al*., 2015). According to textual research, there is no evidence of clinical trials on *E. parvifolia*, however clinical records for different Elaeagnus species are recorded (Liao *et al*., 2012).

Liver plays major role in the body as the body's primary organ to regulate the body's metabolism including biotransformation and the removal of metabolized substances. It also contributes to the achievement of biochemical pathways such as energy production, the provision of nutrients, development, reproduction, and disease defense. liver regulates the body's proteins, carbohydrates and fat metabolism, bile secretion and vitamin concentration. Therefore, healthy liver for good health is essential (Ahsan *et al.,* 2009). Liver damage associated with change in metabolism. Alcohol intake and the spread of xenobiotic drug interactions lead to liver problems (Kumar *et al*., 2013). Hepatitis, jaundice, cirrhosis, and liver cancer are major liver disorders. Toxicity and chronic liver damage are common side effects of several medications including chloroquine and isoniazide. Cirrhosis was reported in 2001, to be the 10th most common cause of mortality in males and in women the 12th most common cause of death, in United States. (Jacob *et al*., 2014). Long-term exposure to toxins from the environment has been linked to hepatitis, cirrhosis, and liver disease in people all over the world. Hepatotoxicity is caused by thousands of synthetic chemicals, medicines, bacteria, fungi, plants, and animal toxin (Raghu *et al*., 2008). Furthermore, some contaminants and hazardous compounds in the environment harm the liver, resulting in some forms of liver disease. Other factors involved in increased liver damage are oxidative stress and pathophysiological activity of free radicals that cause permanent liver damage (Feijoo, 2010). Due to lack of adequate water waste management system, disinfection, limited water availability is convenient; In Pakistan, all types of hepatitis that cause liver illness are common.

Materials and Method

Fruits of *Elaeagnus parvifolia* were collected locally in Rawalakot District, Poonch-Pakistan, and at the University of Poonch Rawalakot's Department of Botany, it was taxonomically recognised and validated. Fruits were sonicated twice in aqueous ethanolic (30:70) at room temperature, for 48 hours. Whatman filter paper number 1 was then used to filter the solution, and then evaporated using a rotary evaporator at decreased pressure.

Phytochemical Analysis

Qualitative analysis

Standard methods were used to investigate phytochemicals in the hydroethanolic *Elaeagnus parvifolia* extract (Jain *et al*., 2014; Sahira Banu *et al*., 2015).

Quantitative analysis

Total flavonoids contents (TFC) and total phenolic (TPC) contents.

Folin-Ciocalteu technique was used to evaluate the total phenolic contents according to Naz *et al*., (2016) reported in plants extract. The standard (gallic acid) was used to calculate the results (Sharif *et al*., 2018). The total phenolic content of plant extract was measured in mg GAE/mL milligrams (mg) gallic acid equivalent (GAE) per milliliter (mL), of plant hydroethanolic extract. The technique described by Rehman *et al*., (2013) has been used to evaluate the total flavonoid content of plant extract. TFC was represented as µg catechin equivalents (CE) per milliliter (mL) (µg CE/mL) of plant extract.

HPLC analysis.

The phenolic components were determined by HPLC, with slight changes, as published by Yue *et al*., (2013). A 20 C18 column (internal diameter 250 4.6 mm) with a 5 mm film thickness, and a 30°C oven was used for liquid chromatography.

Two solutions were used as the mobile phase: solvent A, which was made up of 70% acetonitrile and 30% ethanol, and double distilled water containing solvent B, along with 0.5 percent glacial acetic acid. At 275 nm, UV spectra were collected. Different phytocompounds were identified by comparing spiking samples and retention durations with standards, at a 275 nm wavelength.

FTIR spectroscopy (Fourier-transform infrared)

Elaeagnus parvifolia extract was crushed, and then pressed into pellets with potassium bromide (KBr) powder. By using the frequency range of 400-4,000/cm an FTIR spectrometer after producing the pellets, was used to seek for functional groups that were typical of a wide variety of essential phytoconstituents.

Antioxidant Profiling

DPPH assay (DPPH radical scavenging activity)

The 2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging test was used to assessed the antioxidant capacity, as published by Shahid *et al* (2014). The following formula was used, to compute the % inhibition of DPPH radical samples.

DPPH Inhibition (%) = $\frac{(A_0 - A_1)}{(A_0)} * 100$

Where, A1 = Sample Absorbance and A_0 = Blank Absorbance

Reducing power assay

According to Yadav *et al*., 2014, by converting Fe3+ (CN)6 to Fe2+ (CN)6 the reducing capability of plant extract was determined. The reductive potential of the material was represented by the absorbance, at 700 nm wavelength.

Hydrogen per-oxide scavenging assay

According to procedure Ruch *et al*,. (1989), the H_2O_2 scavenging capacity was determined. Ascorbic acid was utilised as a standard, and H_2O_2 scavenging activities as a percentage of age were computed as

Inhibition (%) = [(Abs Control-Abs Sample)/Abs Control] X 100

Where; A Control is the control reaction absorbance and A Sample is the sample absorbance.

Cytotoxic Potential Determination by Hemolytic Assay

Shahzadi *et al*. (2019) and Rubab *et al*. (2017) employed the approach to investigate the compound's hemolytic activity. Phosphate buffer saline (PBS) was used as a negative control and Triton X-100 (0.1 percent v/v) was used, as a positive control. Using Quant, at 576 nm the absorbance was measured, and the percent RBCs lysis for each sample was computed.

$$\textbf{Percent Hemolysis} = \frac{Ae - Ap}{Ad - Ap} \quad X\ 100$$

Determination of Thrombolytic Activity by Clot Lysis Assay

By using the clot lysis method, the hydroethanolic extract and its in-vitro thrombolytic potential was assessed, as described by Prasad *et al*., 2006. Positive and negative controls were streptokinase and PBS, respectively. The activity of clot lysis in % of age was calculated, as follows:

Clot lysis (%) = (before lysis clot weight _ after lysis clot weight) / clot weight before lysis × 100

In-Vivo Experiment

Animal groups

Male albino mice, weighing between 28 and 32 grammes and aged 10 to 12 weeks, were utilised for in-vivo research as an animal model. Alle animals (n=30) were separated into six groups, each with six individuals (n=6). Group I (n = 6): normal mice treated with carboxymethyl cellulose (1 ml/kg 1 % w/v p/o); Group II (n = 6): intoxicated mice (CCl4 3ml/kg, I/P); Group III (n = 6): standard drugs (CCl4 3ml/kg, I/P single dose and silymarin 100 mg/kg, P/O daily); Group IV (n = 6): Plant extract (CCl4 3ml/kg, I/P single dose and plant extract 200 mg/kg, P/O daily). Based on the results of previous trials, a 28-day treatment plan was devised (Pande *et al*, 2011). This study was conducted

after institutional ethical review committee approval, and held at Government College University (GCU), Faisalabad, Pakistan.

Cyto-toxic Studies

In mice, the hydroetaholic extract toxicity can be determined by performing cytotoxic experiments, performed at doses up to 2000 mg/kg b.w. No hazardous clinical symptoms has been observed in the first 12 hours even after the medication was delivered, at a dosage of 2000 mg/kg.

Blood samples collection

The mice from each group were scarified on the final day of the experiment to obtain blood samples for the evaluation of antioxidant and biochemical parameters and to assess the therapeutic response of plant extract. The serum from non-anticoagulated blood was separated after centrifugation and stored at -20°C until analysis.

Biochemical parameters and stress markers evaluation

The serum was tested for total oxidant status (TOS) and total antioxidant status (TAS). The hepatoprotective activity of hydroethanolic plant extracts assessed by liver enzymes, such as transaminases (ALT, AST, ALP and total bilirubin) were determined as described by (Bergmeyer *et al*.,1986); Thomas, (1998) albumin, total protein, and globulin by (Koller *et al*.,1984), Cholesterol oxidase method was used to measure the cholesterol level quantitatively in the serum samples using the protocol described by Siedel *et al*., (1983). The concentration of triglycerides (TGs) in serum samples was used following the protocol of Shephard and Whiting (1990), HDL and LDL were measured by Conti, (2002); Friedewald *et al*., (1972) and renal function including urea, creatinine and uric acid was measured by method describe by Thomas, (1998) to investigate the impact of medicinal plants on Complete blood cell count (CBC) parameters blood was observed under microscope (Model CX23LEDRFS1, OLYMPUS, Tokyo, Japan) using 10x eyepiece & 40x objective. At least 200 cells were counted under light microscope and DLC calculated (%) as described by Cheesbrough, (2006).

Histological examination of Liver tissue.

Liver fragments were also separated by dissection and washed with water; Buffer formalin (10%) was added for 24 hours until histopathological examination. Liver dehydration was treated with several dilutions of ethyl alcohol (70%, 80% and 100%) and cleared with xylene. 4–5 μm thick portion of liver was prepared using a microtome by dipping the tissue into paraffin-containing bees wax and placing it in a preheated oven at 56oC for 24 hours. After tissue preparation, hematoxylin-eosin (H&E) staining was done after mounting the sections on slides for historical microscopic analysis, to detect changes in hepatocytes (Saleem *et al*., 2020).

Statistical Analysis

In a paired investigation these findings were reported as Mean + SEM, and evaluated using a one-way ANOVA with Tukey's test, to detect the difference between groups.

Results

Phytochemical Constituents

In the hydroethanolic extract of *Elaeagnus parvifolia*, different phytochemical constituents such as glycosides, alkaloids, phenols, flavonoids, steroids, and triterpenoids were investigated (Table 1).

Table 1: Qualitative Phytochemical analysis of Ethanolic Extract of *Elaeagnus parvifolia*

	Reagents/ Test	**Indication**	***Elaeagnus parvifolia***
Alkaloids	Dragendorff's Mayer's Wagner's Hager's	Reddish brown ppt's Creamy precipitate Reddish brown ppt's Yellow ppt's	++ +++ ++ +++
Flavonoids	Shinoda's' Alkaline regent $AlCl_3$	Red or pink color Yellow color Yellow ppt's	++ +++ +++
Coumarins	NaOH	Yellow fluorescence under UV	+++
Phenols	Liebermann's Ferric Chloride Lead acetate	red, green, or blue color. Blue-black color ppt's	++ +++ ++
Phytosterols	Salkowski's Libermann Burchard's	Golden yellow color Green color	- +++
Diterpenes	Copper acetate	emerald green color	++
Starch	Iodine	blue-black speck	++

(+) Indicate week, (++) indicates strong and (+++) indicates very strong presence of phytochemicals and (-) indicate the absence of phytochemicals.
In this study, the total flavonoid (34 ± 0.085 µg CE/ml) and phenolic contents (60.526 ± 0.115 mg GAE/g) were determined using quantitative analysis, and the mean ± SEM (standard error of mean) is shown in Table 2.

Antioxidant Potential

The measurement of free radical scavenging potentials is an important parameter in measuring medicinal plants' antioxidant activity. Table 2 show the findings of the DPPH (1,1-diphenyl-2-picrylhydrazyl) test, power reducing assay, and to assess the antioxidant capacity of hydroethanolic plant extract the hydrogen peroxide scavenging assays used. Plant extracts showed a substantial ($p<0.05$) increase in radical scavenging and lowering activities.

Table 2. *Elaeagnus parvifolia*'s ethanolic extract total phenol, total flavonoid, and antioxidant activities (Mean ± SEM).

Phytochemical Class	***Elaeagnus parvifolia***
TPC (mg GAE/mL)	34±0.085
TFC (mg CE/mL)	60.526±0.115
DPPH Scavenging activity (%)	37.023±0.204
H_2O_2 Scavenging activity (%)	11.279±0.186
Reducing power assay	81.4754±0.248

HPLC and FTIR Spectroscopy

The hydroethanolic extract of *Elaeagnus parvifolia* was analyzed qualitatively using HPLC-UV in this study. Chlorogenic acid (Rt = 2.838), p-cumaric Acid (Rt = 3.213), Gallic acid (Rt = 3.524), Caffeic acid (Rt = 7.158), Vinilic Acid (Rt = 7.495), Kaemphferol (Rt = 11.696), Ferulic Acid (Rt = 12.633), Salicylic Acid (Rt = 15.213), Quercetin (Rt = 17.089), Benzoic Acid (Rt = 18.771) and Rutin (Rt = 23.409) were discovered to be present in *Elaeagnus parvifolia*, which may be responsible for the plant's strong antioxidant potential (Figure 2).
Spectroscopy in the infrared 3,500–4,000, 2974, 2927, 2888, 2359, 1716, 1699, 1653, 1541, 1507, 1455, 1418, 1381, 1321, 1272 1086, 1043, 879 and 803/cm. There were intramolecular and intermolecular hydrogen bonding because the stretching vibration of O-H was linked to the absorption peaks in the range 3,500 to 4,000/cm.
The cause of the absorption peak at 2,927/cm was attributed to the asymmetrically stretched vibration of C-H. Stretching vibrations in the C-H might be the cause of the peak at 2,359/cm. The cause of the absorption peak at 1,716/cm was attributed to the symmetrically stretched vibration of C=O. The absorption maxima at 1,541/cm could be explained by the stretching vibration of C=N

and C=C. The peaks at 1400 to 1500/cm (1455 and 1418/cm) might be explained by strong to medium vibration of the C=C and N=O functional groups. The absorption maxima at 1,381 and 1,321/cm were induced by the stretching vibration of C-O, respectively. The peaks at 1,086 and 1,043/cm were attributed to the asymmetrically stretched vibration of C-O-C. While the peak below 650/cm may be a vibrational S-S stretching, the absorbance peak at 879/cm may represent aliphatic C-I stretching.

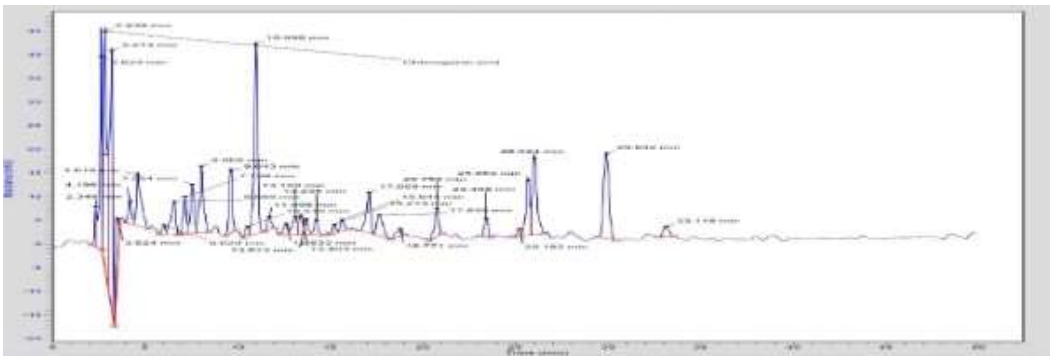

Figure 1: Chromatogram of *E. pervifolia* hydroethanolic extract from HPLC analysis showing the presence of several phenolic compounds.

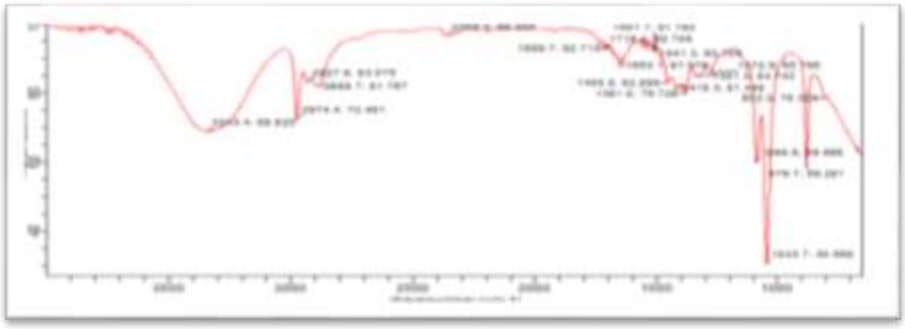

Figure 2. The hydroethanolic extract of *E. pervifolia* exhibits a wide variety of functional groups in its infrared spectra.

Hemolytic and Thrombolytic Activities

The findings of the hemolytic experiment showed that the (percent) hemolysis of plant extract is notably insignificant when compared to the positive control (Triton-X) displayed in Figure. Human clotted blood was used to determine clot lysis activity in order to assess the impact of hydroethanolic extract on blood and semen thinning. Only 5.34+0.46 percent of clot lysis was seen in clots treated with phosphate buffer saline (PBS), and hydroethanolic extract of plant (413.2%) shown substantial (p0.05) thrombolytic activity when streptokinase (SK; 100 mL) was incubated with clot for 90 minutes at 37°C (Figure 3).

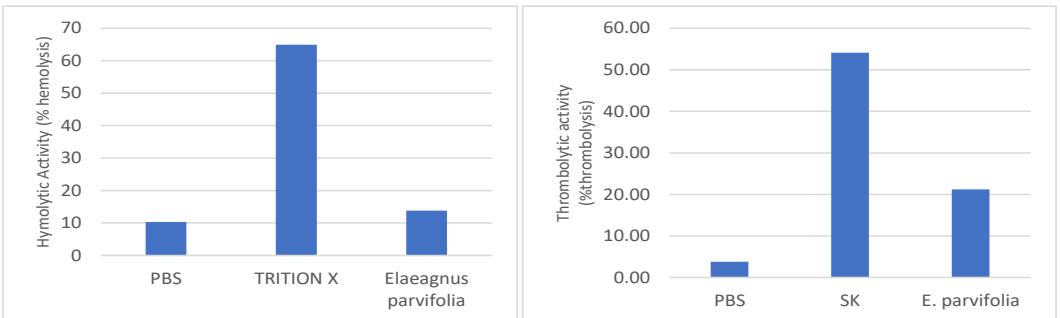

Figure 3. Hemolytic and Thrombolytic Activities of hydroethanolic extract of *E. pervifolia*.

In Vivo Hydroethanolic Extract's Therapeutic Response

Hepatoprotective and renoprotective activities of *Elaeagnus parvifolia* extract.

In drug development, the hepatoprotective and renoprotective characteristics were also investigated, because most herbal remedies offer a wide range of health effects. The statistical analysis demonstrated that CCl4 poisoning resulted in a significant ($P<0.05$) rise in liver enzymes and in total bilirubin, as well as a significant ($P<0.05$) reduction in serum proteins. On the other hand, substantial ($P<0.05$) improvements in liver enzymes and proteins were seen after administration of the plant's hydroethanolic extract (Figure 4A, 4B). Results also revealed that intoxication significantly ($p<0.05$) decreased the serum proteins as compared to healthy control group. On the other hand treatment of mice with *Elaeagnus parvifolia* hydroethanolic extracts and with standard drugs significantly ($p<0.05$) restore the total protein, globulin, albumin, and albumin/globulin ratio. (Figure 4C). Treatment of mice with medicinal plant hydroethanolic extracts significantly ($p<0.05$) lowered cholesterol, TGs, and LDL-Chol concentrations while significantly ($p<0.05$) increasing HDL-Chol as compared to control groups (Figure 4D). When compared to the control group, CCl4 intoxication considerably reduces kidney function as determined by blood urea, creatinine, and uric acid levels, whereas administration of the hydroethanolic extract of *Elaeagnus parvifolia* significantly ($P<0.05$) recovers these renal parameters (Figure 4E).

Biochemical parameters

Intoxication of animals with CCl4 modulated the mineral contents while treating the mice with *Elaeagnus parvifolia* hydroethanolic extract significantly ($p<0.05$) restore to normal as compared to control groups. It was reported that significant increase in potassium ($p<0.05$) due to CCl_4 intoxication, while decreased the calcium, sodium, and chloride which returned to normal significantly ($p<0.05$) on treatment with *Elaeagnus parvifolia* hydroethanolic extract (Figure 3F).

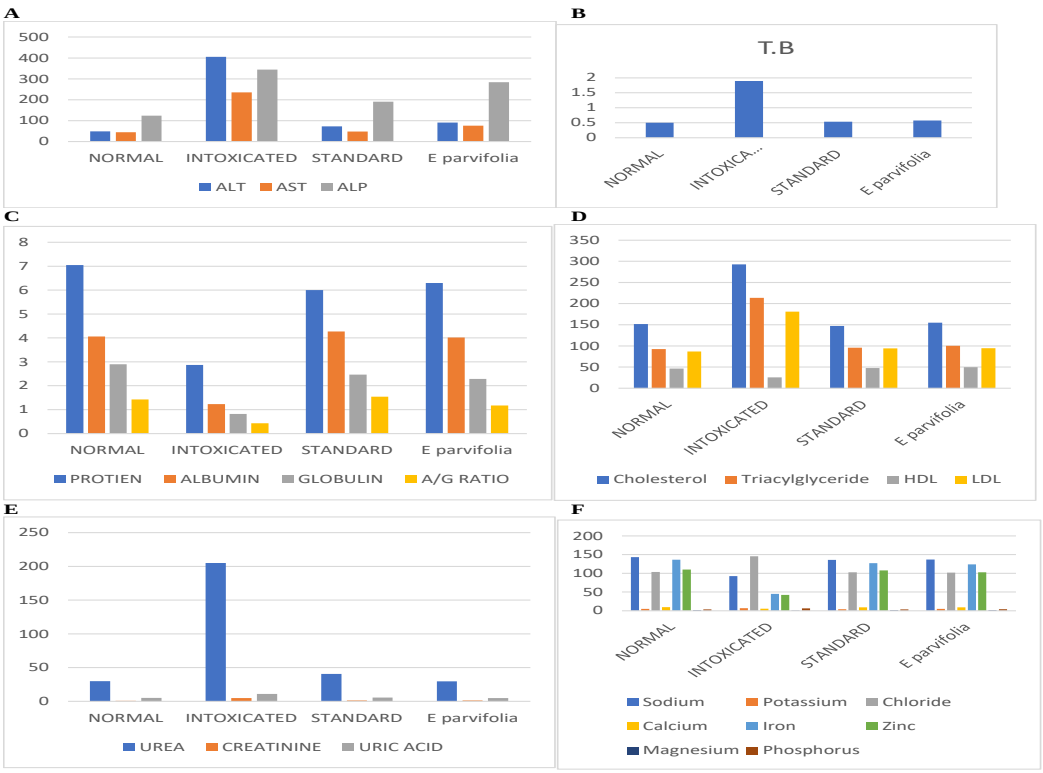

Figure 4. (A): Liver enzymes activities , (B): Serum Total Bilirubin Concentration (C): Serum protein concentration, including albumin, total proteins, and globulin (D): lipid profile, including triglycerides, HDL, and LDL cholesterol (E): The blood urea, creatinine, and uric acid concentrations (F): The serum electrolytes in the blood samples taken from the various experimental groups. Values are in Mean ± SEM

Stress Markers

In the blood of mice, total oxidant status (TOS), total antioxidant status (TAS), and stress markers were analyzed. Induction of CCl4 significantly ($P<0.05$) decreased TAS and raised TOS in mice, whereas daily dose of plant extracts significantly ($P<0.05$) reversed TAS and TOS (Table 3). In comparison to other studied animals, CCl4 intoxicated mice had a significantly higher MDA level (P 0<05). After mice were given E. *pervifloria* extract, their MDA levels returned to near normal (Table 3). Furthermore, after CCl4 toxicity, the activity of SOD (Superoxide dismutase) and CAT (Catalase) as enzymatic antioxidants was shown to be decreased, but in plants extract and positive drug treated mice, both SOD and CAT significantly ($P<0.05$) enhanced (Table 3).

Table 3. Effect of E. parvifolia Extract and Control Drugs on Stress Markers in Albino mice.

Parameters	G-1	G-2	G-3	G-4
TAS	0.612 ± 0.021	0.401 ± 0.032	0.728 ± 0.041	0.734 ± 0.018
TOS	13.865 ± 1.534	25.438 ± 1.323	14.742 ± 1.640	13.435 ± 1.409
MDA	6.899±0.27	15.232±0.07	7.146±0.25	8.351±0.223
SOD	55.248±1.60	25.773±1.90	56.818±2.11	53.191±2.08
CAT	21.666±0.87	13.432±0.05	20.281±0.09	19.800±0.25

G-1; normal control given only diet, G-2; toxic group intoxicated with 3mL 70% CCl4, G-3; Positive control group given 3 mL 70% CCl4 and standard drug, G-4; Test group intoxicated with 3 mL 70 % CCl4 and administered with ethanolic extract *E. parvifolia* 200 mg/ Kg bw, orally daily respectively TAS (Total antioxidant status), TOS (Total oxidant status), MDA (Malondialdehyde), SOD (Superoxide dismutase), CAT (Catalase). Values are mean + SEM (standard error) of means of the study groups. The $p < 0.05$ considered statistically.

Histological examination of liver tissue.

As compared to the normal control group, CCl4 intoxication of mice significantly damaged the liver cells (Figure 5). After CCl4 intoxication, the histoarchitecture of sections from both positive controls and mice given hydroethanolic extract of *E. pervifolia* improved noticeably (Figure 5).

Absolute Control

In this group, the tested animals were normal control with normal histopathology. Figure 5A showed Hepatic parenchyma is normal. Hepatocyte nuclei were also common as holes containing prominent nucleoli and chromatin material.

Positive Control

As a positive control, animals were treated with toxins to stimulate liver damage. Examination of the history of this group showed a high degree of vacuum damage to the cytoplasm of hepatocytes. In some places, there was an excessive amount of single necrosis cells, as evidenced by hepatocyte nuclei. Perivascular tuberculosis, fibrosis, and biliary hypertrophy were also present as shown in (Figure 5B).

Standard Drug

Group 3 experimental animals were given liver-damaging toxins before being given a standard drug (silymarin, 200 mg/kg/body weight) available locally. In liver tissue cells, significant swelling at small sinusoidal openings was demonstrated. Bone vacuoles were found throughout the cytoplasm. A single necrotic cell was also present; No potency effect was shown in (Figure 5C).

E. parvifolia

This set of experimental animals (group 4) received liver-damaging chemicals first, and then they received plant extracts (Elaeagnus parvifolia 200 mg/kg/body weight). The parenchyma of the liver was normal. Hepatocytes from in the soft tissues of the liver and in some places, it was a mild disease. Unicellular necrosis was also present. Pocket space is normal, there was a small degree of hunger pangs in places, and it felt like a casino. The anatomical effect was observed by histopathology (Figure 5D).

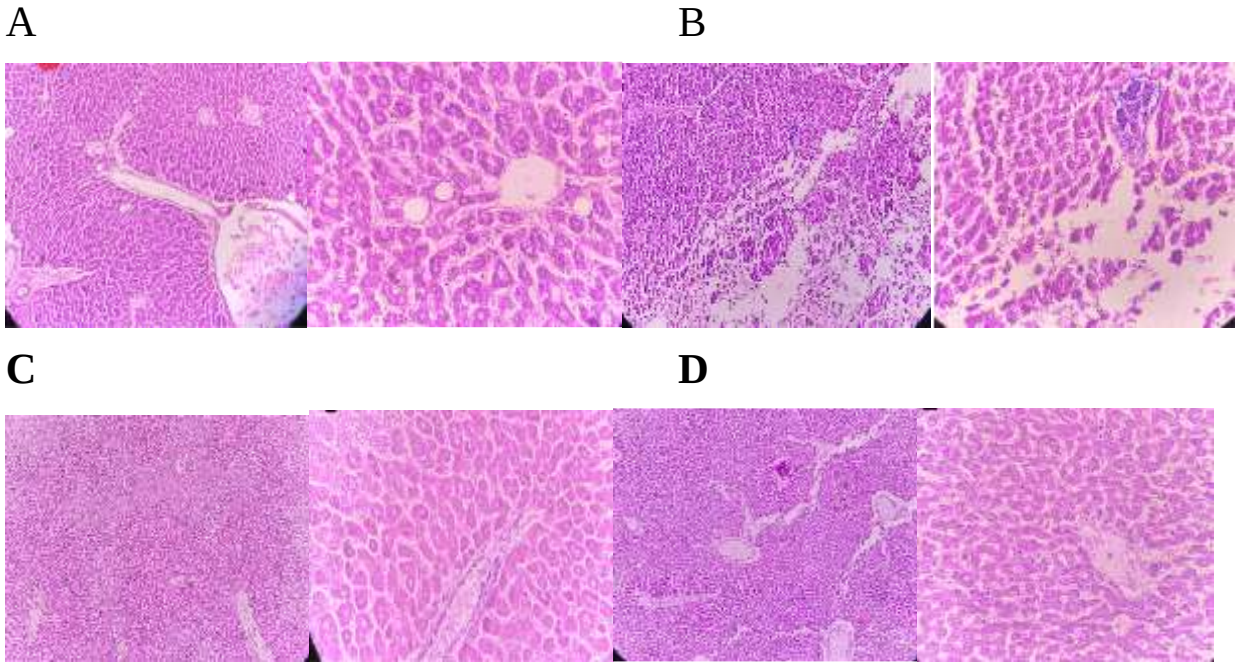

Figure 5: Photomicrograph of liver showing (A): normal hepatic parenchyma in the absolute control group, (B): pyknotic changes and fibrosis in the positive control, (C):mild cell inflammation and vacuolar collapse in the standard drug and (D): mild vacuolar degeneration in the cytoplasm of hepatocytes and individual cell necrosis in the extract from E pervifolia.

Discussion

Constituents of plants and antioxidant potential

Due to the existence of phytochemical components such alkaloids, flavonoids, steroids, glycosides, tannins, and saponins, a wide variety of medicinal plants are used with a diversity of therapeutic potentials. (Edeoga *et al*., 2005) and the antioxidant capacity of herbal medicines has been connected to the presence of phenolic compounds (Zheng *et al*., 2011). Ethanol has also been found to be an excellent solvent for extracting antioxidant phenolic chemicals. The quenching of active oxygen species has been attributed to the presence of various flavonoids in the extract (Leake *et al*., 2001).

Therapeutic Response of Hydroethanolic Extract in Vivo

Elaeagnus parvifolia extract's renoprotective and hepatoprotective properties.

The liver, as the body's major organ, performs vital functions such as synthesis, detoxification, storage, excretion, and secretion; failure of any of these metabolic processes can lead to liver disorders, resulting in high serum liver enzyme levels in the blood circulation (Nweze *et al*., 2015). *Elaeagnus parvifolia* extract may have hepatoprotective properties, based on the improvement in liver enzymes in plants-treated mice.

One of the most important role of liver is to synthesis the serum proteins and these proteins play different vital functions in the body including maintaining circulatory osmotic pressure,

transpotation of metabolites, regulation of certain activities in cells, binding with some waste products and hormones as well as in humoral immune system (Chatterjea and Shinde, 2011).
Results revealed that intoxication significantly ($p<0.05$) decreased the serum proteins as compared to healthy control group. On the other hand, treatment of mice with different concentrations of *E. pervifolia* hydroethanolic extracts and with standard drugs (silymarin) significantly ($p<0.05$) restore the total protein, albumin, globulin and albumin/globulin ratio.
It was reported that on administration of hydroethanolic extracts significantly ($p<0.05$) prevent the renal damage due to CCl_4. It was also reported that CCl_4 intoxication increased significantly ($p<0.05$) urea, creatinine, uric acid in mice which returned to normal on treatment with *E. pervifolia* hydroethanolic extracts. This restoration of renal parameters in treated mice represents the renoprotective potential of medicinal plants.

Total oxidant (TOS), antioxidant status (TAS) and stress markers in mice' blood.
As part of the body's natural defensive mechanism, antioxidant mechanisms, both enzymatic and non-enzymatic, neutralize oxidant chemicals. The body can make enough antioxidants or absorb them as food supplements to detoxify free radical species produced, as well as oxygen free radicals. Total antioxidant status in the body regularly demonstrated the dynamic equilibrium between the body's antioxidant defense and pro-oxidants (Ghiselli *et al*., 2000; Valko *et al*., 2007). *Elaeagnus parvifolia* has shown considerable antioxidant activity in vitro, most likely because of the presence of flavonoids and phenolic compounds as active secondary metabolites (Riaz *et al*., 2016). These substances also provide the antioxidant defense in animals following CCl4 poisoning.

Conclusion
When a therapeutic dose of hydroethanolic extract was given to male albino mice, it was found that in addition to considerable hepatoprotective and renoprotective properties, *Elaeagnus parvifolia* had a wide variety of phytochemical components with significant antioxidant capacity *in vitro* and *in vivo.* The identification of novel components from this medicinal plant is required for further research to solve the hepatoprotective problems.

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