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Research Article

Hepatoprotective activity of *tagetes erecta* Linn. in ethanol induced hepatotoxicity in rats

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Abstract: The plant *Tagetes erecta* Linn., locally known as *Genda Phul* (Marigold) belongs to the family Asteraceae (Compositaeis). Current study focus on hepatoprotective activity of the roots in ethanol extract by ethanol induced hepatotoxicity model. Physical parameters, liver functioning, antioxidant levels and histopathological study of the liver were studied to find out hepatoprotective action of *Tagetes erecta*. Treatment with *Tagetes erecta* root extracts has protected liver from induced hepatotoxicity. This was demonstrated by reducing the elevated levels of biochemical markers and additional histopathological observations have shown that there is an improvement in the structural design of liver due to induced hepatotoxicity.
Keywords: *Tagetes erecta, Genda Phul,* hepatoprotective activity.

INTRODUCTION

The plant *Tagetes erecta* Linn., locally known as *Genda Phul* (Marigold) belongs to the family Asteraceae. It is a stout, branching herb, native of Maxico and other warmer parts of America and naturalized elsewhere in the tropics and subtropics including Bangladesh and India. It is very popular as a garden plant and yields a strongly aromatic essential oil (tagetes oil), which is mainly used for the compounding of high-gradeperfumes [1].Studies of its different parts have resulted in the isolation of various chemical constituents such as thiophenes, flavonoids, carotenoids and triterpenoids . The plant Tagetes erecta has been shown to contain quercetagetin, a glucoside of quercetagetin, phenolics, syringic acid, methyl-3,5-dihydroxy-4- methoxy benzoate, quercetin, thienyl and ethyl gallate [1].

Lutein is an oxycarotenoid, or xanthophyll, containing 2 cyclic end groups (one beta and one alpha-ionone ring) and the basic C-40 isoprenoid structure common to all carotenoids. It is one of the major constituents and the main pigment of *Tagetes erecta* [2].

Different parts of this plants including flower are used in folk medicine to cure various diseases. The leaves are reported to be effective against piles, kidney troubles, muscular pain, ulcers, wounds and earache. The pounded leaves are used as an external application to boils and carbuncles. It is reported to have antioxidant, antimycotic, analgesic activity and 18 active compounds are identified by GC-MS, many of them are terpenoids. The flower is useful in fevers; epileptic fits, astringent, carminative, stomachic, scabies, liver complaints, and also employed in diseases of the eyes. They are said to purify blood and flower juice is given as a remedy for bleeding piles and also used in rheumatism, cold and bronchitis.leaves are used as an external application to boils and carbuncles [3].

MATERIALS AND METHODS

Extraction of *Tagetes erecta* Linn .

The leaves plant of *Tagetes erecta* Linn. was dried under shade and then powdered with a mechanical grinder to obtain a coarse powder (500 gm) the fine powder of whole plant was packed in high quality filter paper, which was then subjected to successive extraction in a soxhlet apparatus using 50% ethanol for about 72 hour, solvent was recovered. Extractive yield of *Tagetes erecta* Linn. was 35 %. . After vacuo evaporation the crude extract was dissolved in distilled water freshly as required.

Animals

Albino rats (*Wistar* strain) weighing 150 - 200 gm of either sex were used for the present study. The animals were housed in polypropylene cages at control temperature (26 ± 2° C) relative humidity (60 ± 5%) and light. Rats were fed with standard laboratory diet and drinking water was given through drinking bottle throughout the experiment. The animals were maintained as per CPCSEA regulation and cleared by

IAEC at Bhupal Nobles' College of Pharmacy, Udaipur (Rajasthan), India.

Drug Formulation

The extract of plant fully dissolves in distilled water. The solution of the whole plant extract (300 mg/ml) was freshly prepared in distilled water.

Experimental Induction of Hepatotoxicity:

All the animals, except normal control group, will be received 40% ethanol (3.76 g/kg, p.o) twice daily for a period of 25 days [4].

Experimental Design

Albino rats of either sex weighing between 200 -250g were selected and divided into six groups of six animals in each. The animals were fasted 24 hours prior to experiment. Group I was maintained as normal control, which was given with distilled water only. Group II received ethanol (3.76 gm/kg, twice daily, p.o) and animals in Group III were treated with Silymarin (200 mg/kg, p.o) which served as standard. Animals in Groups IV, V and VI were treated with three different doses extract of *Tagetes erecta* Linn. (200mg/kg, 400mg/kg and 400mg/kg p.o.,) respectively.Group II, III, IV, V were intoxicated with ethanol for 25 days.

At the end of experimental period, all the animals were sacrificed by cervical decapitation. Blood samples were collected, allowed to clot. Serum was separated by centrifuging at 2500 rpm for 15 min and analyzed for various biochemical parameters.

Assessment of liver function

Biochemical parameters i.e., SGOT [5], SGPT [5], SALP [6], serum bilirubin [7] total cholesterol [8] and total protein [9] were analyzed according to the reported methods. The liver was removed, morphological changes were observed. A 10% of liver homogenate was used for antioxidant studies suchas lipid peroxidation (LPO) [10], glutathione (GSH) [11] and Catalase [12], superoxide dismutase (SOD) [13]. A portion of liver was fixed in 10% formalin for histopathological studies.

Serum hepatospecific markers:

Activities of serum glutamate oxaloacetate transaminase (SGOT) and serum glutamate pyruvate transaminase (SGPT) were estimated by the method of Reitman and Frankel[5]. 0.05 ml of serum with 0.25 ml of substrate (aspartate and α-ketoglutarate for SGOT; alanine and α - keto glutarate for SGPT, in phosphate buffer pH 7.4) was incubated for an hour in case of SGOT and 30 min. for SGPT. 0.25 ml of DNPH solution was added to arrest the reaction and kept for 20 min in room temperature. After incubation 1 ml of 0.4 N NaOH was added and absorbance was read at 505 nm in *uv-vis* spectrophotometer. Activities were expressed as IU/dl.

Based on the method of King and Armstrong alkaline phosphatase activity was assayed using disodium phenyl phosphate as substrate[6]. The colour developed was read at 510 nm in *uv-vis* spectrophotometer after 10 min. Activities of ALP was expressed as IU/dl. Serum total bilirubin level was estimated based on the method of Malloy and Evelyn Diazotised sulphonilic acid (0.25 ml) reacts with bilirubin in diluted serum (0.1 ml serum + 0.9 ml distilled water) and forms purple colored azobilirubin, which was measured at 540 nm in *uv-vis* spectrophotometer. Activities of total bilirubin were expressed as mg/dl. Total cholesterol was determined by method of Richmond[8]. Serum total protein level was estimated based on the method of Gornall *et al*[9] . Biuret reagent (1.0 ml) reacts with serum (10 µL) and the colour developed was read at 578 nm in *uv-vis* spectrophotometer. Activities of total protein were expressed as mg/dl.

Determination of Thiobarbituric Acid Reactive Substances (TBARS)

Lipid peroxidations in liver tissues were estimated colorimetrically by measuring thiobarbituric acid reactive substances (TBARS) by the method of Ohkawa *et al* [10]. To 0.2ml of sample, 0.2ml of 8.1% Sodium dodecyl sulfate, 1.5 ml of 20% acetic acid and 1.5 ml of 0.8% TBA were added. The volume of the mixture was made up to 4 ml with distilled water and then heated at 950 °C in a water bath for 60 min. After incubation the tubes were cooled to room temperature and the final volume was made upto 5 ml in each tube. Then 5 ml of n-butanol: Pyridine mixture was added and the contents were vortexed thoroughly for 2 min. After centrifugation at 3000 rpm for 10 min the upper organic layer was taken and its OD was read at 532 nm against an appropriate blank without the sample.

Determination of reduced glutathione (GSH)

Reduced glutathione (GSH) was determined by the method of Ellman[11]. To 0.1 ml of different tissue homogenate 2.4 ml of 0.02 M EDTA solution was added and kept on ice bath for 10 min. Then 2 ml of distilled water and 0.5 ml of 50 % TCA were added. This mixture was kept on ice for 10-15 min and then centrifuged at 3000 rpm for 15 min. 1 ml of supernatant was taken and 2ml of Tris-Hcl buffer was added. Then 0.05 ml of DTNB solution (Ellman's reagent) was added and vortexed thoroughly. OD was read (within 2-3 min after the addition of DTNB) at 412 nm against a reagent blank. Absorbance values were compared with a standard curve generated from known GSH.

Assay of super oxide dismutase (SOD)

Superoxide dismutase (SOD) activity was determined by the method of Sapakal *et al*.[13]. Prepared 10 % w/v tissue homogenate in 0.15 M Tris HCl .Centrifuged at 15000 rpm for 15 min at 4 °C. Supernatant (0.1 ml) was taken consider it as sample and 1.2 ml sodium pyrophosphate buffer (pH 8.3, 0.052

M) + 0.1 ml phenazine methosulphate (186 μM) + 0.3 ml of 300 μM Nitroblutetrazolium + 0.2 ml NADH (750 μM) were added. Incubated at 30°C for 90 s .0.1 ml glacial acetic acid was added. Stirred with 4.0 ml n-butanol.Allowed to stand for 10 min Centrifuged and separated butanol layer. OD at 560 nm was taken (taken butanol as blank) and concentration of SOD was expressed as units/g of liver tissue. Absorbance values were compared with a standard curve generated from known SOD.

Assay of Catalase (CAT):

Catalase was assayed according to the method of Aebi[12]. The estimation was done spectrophotometrically following the decrease in absorbance at 240 nm. The liver tissue was homogenized in M/150 phosphate buffer (pH 7.0) at 1-40 C and centrifuged at 5000 rpm. The reaction mixture contained 0.01 M phosphate buffer (pH 7.0), 2 mM H_2O_2 and the enzyme extract. The specific activity of catalase was expressed in terms of units/gram of liver tissue. Absorbance values were compared with a standard curve generated from known CAT.

Histology

The tissues of liver were removed from animals, washed with normal saline to remove blood, fixed in 10% formalin and embedded in paraffin wax. Sections of 5 μm thickness were made using rotary microtome and stained with haematoxylin-eosin and histological observations were made under light microscope [14,15].

Statistical analyses

The experimental results were expressed as the Mean ± S.D for six animals in each group. Statistical analyses were performed using the unpaired t test. A p value of 0.05 or less was considered to indicate a significant difference between groups.

RESULTS

Effect of hydroalcholic extract of *tagetes erecta* Linn. on biochemical parameters

Effect on serum marker enzymes, total bilirubin and total cholesterol

Administration of ethanol to the animals resulted in a marked elevation of serum transaminases (SGOT and SGPT), serum alkaline phosphatase (ALP) and total bilirubin (TB), total cholesterol (TC) when compared with those of normal control animals. The rats treated with hydroalcholic extract of *tagetes erecta* Linn. (HATE) and with silymarin showed a significant decrease (P<0.05) in all the elevated serum marker levels, SGOT, SGPT, ALP , TB, TC and significant increase (P<0.05) in total protein (Table 1,2,3,4) which showed the restoration of the level of liver function biochemistry to the near normal values.

Effect on Serum total protein

Ethanol treatment considerably reduced serum total protein levels (P<0.0001). Treatment with silymarin and HATE showed a significant increase in total protein levels (P<0.05) as compared with toxicant control group.

Table: 1: Effect of hydroalcholic extract of *Tagetes erecta* Linn. on serum, SGPT, SGOT, ALT and Total bilirubin on ethanol induced hepatotoxicity in rats:

GROUPS	Treatment	SGPT (IU/dl)	SGOT (IU/dl)	ALP (IU/dl)	TOTAL
I	Vehicle	41.2±3.789	56.2±3.856	105.8±4.985	0.58±0.028
II	Ethanol control	113.4±6.186***	124.2±6.896***	230.2±9.286***	1.9±0.026***
III	Silymerin-(200	62.5±5.189^{+++}	72.2±10.285^{+++}	140.8±7.902^{+++}	0.75±0.018^{+++}
IV	HATE-200 mg/kg	107.8±7.295^{ns}	112.2±7.286^{+}	214.6.6±8.925^{+}	1.87±0.026^{ns}
V	HATE-400 mg/kg	100.2±6.685^{+}	108.5±8.281^{+}	210.2±6.486^{++}	1.84±0.028^{+}
VI	HATE-600 mg/kg	97.9±8.982^{+}	104.6±9.386^{+}	205.6±7.985^{++}	1.80±0.090^{+}

All values are represents mean ± SD; n = 6 animals.

P values: ***‹ 0.0001 when compared with control untreated rats;ns - not signigicant; +++ ‹ 0.0001; ++ ‹ 0.001; +<0.05 when compared with carbon tetrachloride treated rats.

Effect on *In vivo* antioxidant activity (GSH, LPO ,CAT and SOD)

Toxic dose of ethanol significantly reduced the activities of enzymes (SOD and catalase) and non-enzymatic (GSH) antioxidant system and enhanced lipid peroxidation (LPO) level of liver tissue, as were found in group II animals. HATE treatment significantly raised both of the enzymatic and non-enzymatic antioxidant systems as was found in case of silymarin treated group, while the elevated LPO level were found to be reduced back to towards the normal level in HATE as well as silymarin treated rats (Table 2).

As shown in table, the concentration of MDA, an end product of lipid peroxidation, in the rats treated with ethanol was increased to many folds when compared with the vehicle control rats. Consistent with the serum SGOT and SGPT activities, treatment with HATE resulted in a significant dose-dependent decrease in the concentration of hepatic MDA when compared with the ethanol group Conversely, the concentration of hepatic GSH in the ethanol group was decreased by more than 50% when compared with the vehicle control group. However, treatment with HATE also resulted in a statistically significant increase in hepatic GSH

concentration in comparison with that of the ethanol group.

As presented in table, the activity of catalase in the ethanol group were decreased by more than 50% when compared with the vehicle control group. However, treatment with HATE resulted in a statistically significant, dose dependent increase in catalase activity (P<0.05) when compared with the ethanol control group. As shown in table 2, the hepatic SOD activity in the ethanol were also reduced by many folds when compared with the vehicle control group. However, the SOD activity was significantly increased by (P<0.05) HATE treatment in a dose-dependent manner when compared with the ethanol control group.

Table 2: Effect of hydroalcholic extract of *Tagetes erecta* Linn. on SOD, GSH, LPO and Catalase in ethanol induced hepatotoxicity in rats:

GROUPS	Treatment	SOD(unit/mg tissue)	GSH(mmol/mg tissue)	LPO (nmol MDA/mg	CATALASE (unit/mg tissue)
I	Vehicle	12.85±0.695	5.02±0.289	1.65±0.098	15.01±0.698
II	Ethanol control	4.28±0.628***	1.95±0.366***	5.65±0.082***	3.9±0.256***
III	Silymerin(200mg/kg)	10.98±0.256^{+++}	4.6±0.389^{+++}	1.9±0.081^{+++}	13.04±0.286^{+++}
IV	HATA-200 mg/kg	4.35 ±0.586^{ns}	2.05±0.276^{ns}	5.2±0.352^{+}	3.99±0.282^{ns}
V	HATE-400 mg/kg	5.10 ±0.550^{+}	2.38±0.264^{+}	4.95±0.286^{++}	4.35±0.125^{+}
VI	HATE-600 mg/kg	5.35±0.568^{+}	2.45±0.187^{+}	4.81±0.108^{+++}	4.54±0.234^{++}

All values are represents mean ± SD; n = 6 animals.
P values: ***‹ 0.0001 when compared with control untreated rats; ns - not signigicant; +++ ‹ 0.0001; ++ ‹ 0.001; +<0.05 when compared with carbon tetrachloride treated rats.

Table : 3 Effect of hydroalcholic extract of *Tagetes erecta* Linn. on serum Total Protein and Total Cholesterol on ethanol induced hepatotoxicity in rats:

GROUPS	Treatment	Total Protein (µg/mg)	Total Cholesterol (mg/dl)
I	Vehicle	95.8±3.285	95.2±5.686
II	Ethanol control	54.2±6.219***	186.2±9.280***
III	Silymerin (200 mg/kg)	91.6±5.185^{+++}	107.6±11.287^{+++}
IV	HATE-200 mg/kg	50.3±4.854^{ns}	165.8±6.284^{+}
V	HATE-400 mg/kg	69.8±3.196^{+}	145.2±5.981^{+}
VI	HATE-600 mg/kg	71.2±4.67^{++}	125.4±8.978^{++}

All values are represents mean ± SD; n = 6 animals.
P values: ***‹ 0.0001 when compared with control untreated rats; ns - not signigicant; +++ ‹ 0.0001; ++ ‹ 0.001; +<0.05 when compared with carbon tetrachloride treated rats.

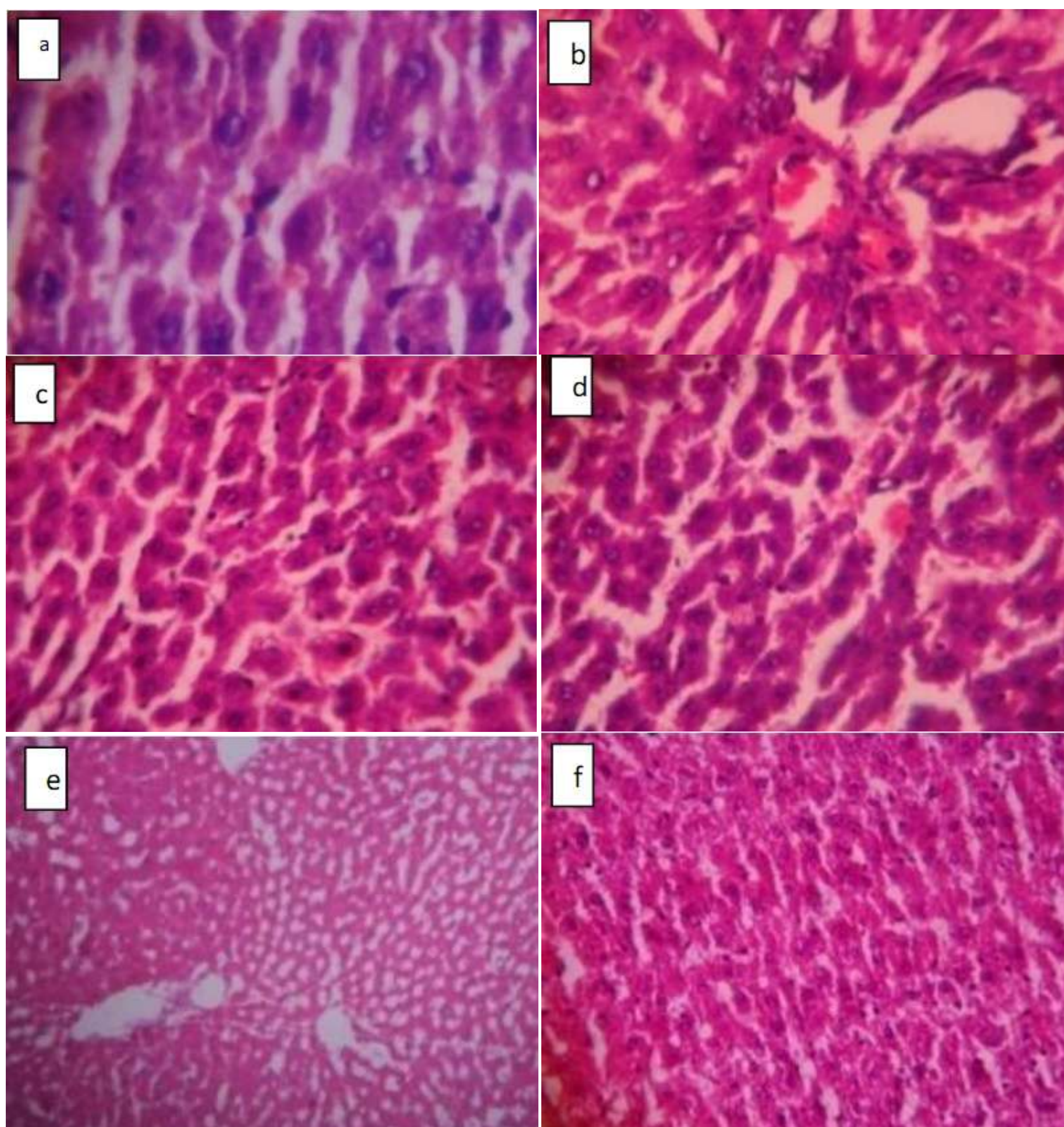

Fig-1: Histopathalogical monograph of extract and standard. a: Control; b: Ethanol (3.76 g/kg) alone; c: Ethanol+ *Tagetes erecta* (3.76 g/kg +200 mg/kg); d: Ethanol + *Tagetes erecta (* 3.76 g/kg+400 mg/kg); e: Ethanol+ *Tagetes erecta* (3.76 g/kg+600 mg/kg); f: Ethanol+Silymarin (3.76 g/kg+ 200 mg/kg).

DISCUSSION

More than ever before, there is an upsurge in alcohol abuse and as a result, alcohol-related disorders are becoming increasingly important causes of morbidity and mortality globally [16]. Ethanol is a natural product that has been available for human consumption for thousands of years. It has well characterized psychophysical and mood-altering effects. It is also a common cause for the generation of reactive oxygen species (ROS), which can damage cellular lipids, proteins, and DNA leading to oxidative stress and induce liver injury [17, 18 ,19, 20].To counteract this oxidative stress, cells have several antioxidant enzymes, including superoxide dismutase (SOD), glutathione peroxidase (GPX), and catalase [21]. Oxidative stress occurs when the production of ROS exceeds the level the body's natural antioxidant defense mechanisms can cope with; causing damage to macromolecules such as DNA, proteins and lipids. This is often characterized by high level of malondialdehyde (MDA), and reduction of reduced GSH molecules [16].

In the body, especially in the liver and the gastrointestinal tract (GIT), ethanol is broken down by a

number of metabolizing enzyme systems by both the oxidative and non-oxidative pathways to generate a number of potentially harmful by products that cause deleterious effects on the body tissues and organs [22,23].About 80–90% of ingested alcohol is metabolized in the liver, where alcohol is oxidized to acetaldehyde. The process is catalyzed by three different enzymes: alcohol dehydrogenase (ADH) in cytoplasm, microsomal ethanol metabolizing system (MEOS) in smooth endoplasmic reticulum and acetaldehyde dehydrogenase (ALDH) . All these biochemical pathways produce acetaldehyde as their toxic by-product. Since acetaldehyde is much more toxic than alcohol, it is associated with a lager number of the metabolic abnormalities in liver disease induced by alcohol [19,24].

The cytochrome P450 enzymes (CYP2E1) and catalase also contribute to alcohol metabolism and alcohol-related cytotoxicity under specific circumstances. Alcohol metabolic rates show a considerable degree of interindividual and ethnic variability, in part due to allelic variants of the genes encoding ADH and ALDH producing functionally different isozymes [25].

Assessment of liver function can be performed by determining the activity of serum enzymes SGPT, SGOT and ALP, originally present in high concentrations in the cytoplasm. When there is hepatic injury, these enzymes leak into the blood stream in conformity with the extent of liver damage [26,27,28,29,30]. Whereas the extracts treated animals had significantly reduced SGPT, SGOT and ALP levels indicating their hepatoprotective effect against alcohol - induced liver cell damage. The elevated activities of these marker enzymes in sera of the ethnol treated rats in the present study were due to the extensive liver damage caused by the toxin. Treatment with the test drug as well as the reference drug silymarin significantly reduced the ethnol induced elevation in the activities of these enzymes.

The leakage of cell membrane participated in the accumulation of these enzymes into the plasma. Elevated activities of SGPT, SGOT, and ALP enzymes indicate liver damage [31,32]. This is because of higher concentration of alcohol dehydrogenase in liver, which catalyzes alcohol to its corresponding aldehyde [33,34] .

The studies on lipid peroxidation, antioxidant enzymes like reduced glutathione, superoxide dismutase and catalase have been found to be of importance parameters in the assessment of liver damage [35,36]. Antioxidant enzymes are components of the cellular defense system against ROS and reactive nitrogen species (RNS). SOD and catalase are defense against oxidative damage by supplying NADPH, which is needed for the regeneration of GSH. Moreover oxidative stress is known to decrease GST activity and also lower GSH content [37]. GSH acts as an antioxidant both intracellularly and extracellularly in conjunction with various enzymatic processes. GSH is a major non-protein thiol in living organisms and plays a central role in coordinating the body's antioxidant defense process [37].

Markedly elevated levels of MDA in liver intoxicated by alcohol suggests that enhanced lipid peroxidation leading to tissue damage. This phenomenon postulates failure of antioxidant defense mechanism to prevent formation of excessive free radicals. Treatment with extacts and its fractions significantly reduced the levels of MDA. It was well accepted that a deficiency of GSH within living cells can lead to tissue injury. The liver injury produced by Ethanol (Alcohol) is known to be related with low tissue levels of GSH. Decrease level of superoxide dismutase (SOD) is a important hepatic oxidative stress parameter during the assessment of liver damage. It scavenges the superoxide anion to form hydrogen peroxide and thus preventing the toxic effect caused by this radical. Treatment by extract causes a significant increase in hepatic SOD and GSH activity.

Ethanol aggravates lipid peroxidation, as evident from the present study. Oral administration of extracts decreased the lipid peroxidation of cellular memberane, suggesting a protective effect. The increase in MDA levels in the livers of ethanol treated rats suggests that main target of oxidative stress is the poly unsaturated fatty acids in cell membranes causing lipid peroxidation and excessive formation of MDA which may lead to damage of the cell structure [34,38,39]. Enhanced peroxidation leading to tissue damage and failure of the antioxidant defense mechanisms to prevent the formation of excessive free radicals [20,35,36]. Treatment with extracts significantly reversed these changes.

GSH has been postulated to be an important pathogenic factor in alcoholic liver injury. The fall inhepatic GSH sensitizes the liver to oxidative injury and sets up a vicious cycle. Chronic ingestion of ethanol resulted in a significant depletion of glutathione (GSH) content in liver [40]. Raised levels of GSH have been reported to elicit a protective response against the toxic manifestations of chemicals, particularly those involving oxidative stress. Explanations of the possible mechanisms underlying the hepatoprotective properties of drugs include the prevention of GSH depletion [41] and destruction of free radicals [42,38].

Decrease level of superoxide dismutase (SOD) is a important hepatic oxidative stress parameter during the assessment of liver damage. It scavenges the superoxide anion to form hydrogen peroxide and thus preventing the toxic effect caused by this radical [35,36]. The presence of superoxide dismutase (SOD)

in various compartments of our body enables SOD to dismutate superoxide radicals immediately [38,39]. In the present work, superoxide dismutase activities increased on ethanol exposure, and reduced significantly due to different treatment including abstination from alcohol. Catalase(CAT) is a haemoprotein and it protects cells from the accumulation of H_2O_2 by dismutating it to form H_2O and O_2 or by using it as an oxidant in which it works as a peroxidase in alcohol treated rats significantly decreased levels of catalase but In the present study application of extracts significantly increased the levels of CAT [35,36]. Catalase activity was decreased in higher concentration of ethanol exposure might be due to loss of NADPH, or generation of superoxide, or increased activity of lipid peroxidation or combination of all [38,39].Different treatment increased its activity in the present study.

Ethanol induces hypercholesteremia and hypertriglyceridemia, which may be due to the activation of enzyme HMG CoA reductase, the rate-limiting step in cholesterol biosynthesis [29]. Treatment with extracts reduced the elevated cholesterol levels, suggesting that the extracts prevented ethanol-induced hyperlipidemia probably due to their hepatoprotective activity. Ethanol is known to have a profound effect on the metabolism of lipids and lipoproteins. Accumulation of lipids in the hepatocytes is the most striking initial manifestation of alcohol-induced liver injury. The concentration of serum and tissue lipids such as free fatty acid (FFA), triacylglycerols (TG), total cholesterol (TC), and phospholipids (PL), is increased, in response to ethanol administration. Serum levels of lipids are principally a balance of production and deprivation by the liver. The observed increase in cholesterol level increases the membrane fluidity, disturbs its permeability, and alters the viscosity and also the internal chemical composition of membranes. [The role of high levels of cholesterol and TG in the early steps of atherosclerosis is well established. Increased levels of FFA may be due to lipid breakdown, which can increase the synthesis of other major lipids and activate NADPH- or NADH-dependent microsomal peroxidation [43].The ethanol acts as a surfactant and suppresses the action of enzyme LCAT to block the uptake of lipoprotein from circulation by extra hepatic tissue, resulting in an increase in blood lipid levels. The hyper-lipidemic condition revealed in the serum of ethanol administered animals was restored to normal after the supplementation of the plant extract. The decreased serum cholesterol in the plant extract administrated rat might be due to increased activity of enzyme LCAT involved in esterification of cholesterol in the plasma [44].

The decrease in the levels of Total protein (TP) observed in the alcohol treated rats suggested that the destruction in the number of hepatic cells which may result in decrease in hepatic capacity to synthesize protein [35,45,46]. Treatment with extracts marked elevated the total protein level which was compared with standard drug silymarin. The reduction in protein concentrations is attributed to the initial damage produced and localized in the endoplasmic reticulum which results in the loss of P450 leading to its functional failure with a decrease in protein synthesis [36] .

Furthermore, in present study in Ethanol intoxicated (Positive Control) group increase in the levels of serum Total bilirubin reflected the level hepatic damage and increase of transaminases level were the clear indications of cellular leakage and loss of functional integrity of cell membrane [47,48].

CONCLUSION

In conclusion, our results of this study reported that hydoalcoholic extract of *tagetes erecta* was effective treatment for the control of hepatotoxicity induced by ethanol. The degree of protection was measured by using biochemical parameters like serum transaminases , alkaline phosphatase, total protein, total cholestrol , bilirubin and antioxidant characters. The ethanolic extracts showed the significant hepatoprotective activity comparable with standard drug silymarin. From this investigation, phenolic compounds of plant leaves may be responsible for the hepatoprotective activity. Our results demonstrated that the plant derived drugs is the best alternative drug for synthetic or chemical drug.

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