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Research Article

Pharmacognostical and Pharmacological Evaluation of Flowers of *Tagetes erecta* L. for Hepatoprotective Activity

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Abstract

The present study was undertaken to establish pharmacognostical standards and evaluate the hepatoprotective activity of the ethanolic extract of flowers of *Tagetes erecta* L. Fresh flowers were authenticated, shade dried, powdered, and subjected to organoleptic, macroscopic, microscopic, powder microscopic, fluorescence, and physicochemical evaluation. The physicochemical parameters showed moisture content of 8.42 ± 0.31%, total ash value of 7.86 ± 0.24%, acid-insoluble ash value of 0.68 ± 0.07%, water-soluble extractive value of 21.48 ± 0.52%, and alcohol-soluble extractive value of 18.76 ± 0.45%. The ethanolic extract showed the highest yield of 17.29% w/w and was selected for further studies.

Preliminary phytochemical screening revealed the presence of carbohydrates, glycosides, flavonoids, phenolic compounds, tannins, saponins, and terpenoids. The extract demonstrated appreciable antioxidant potential, which was attributed mainly to phenolic and flavonoid constituents. Acute oral toxicity study showed no mortality or significant behavioural abnormality up to 2000 mg/kg body weight. Hence, 200 mg/kg and 400 mg/kg doses were selected for hepatoprotective evaluation.

Paracetamol-induced hepatotoxicity in Wistar rats significantly elevated serum AST, ALT, ALP, bilirubin, cholesterol, triglycerides, malondialdehyde, and lipid peroxidation, while reducing total protein, albumin, superoxide dismutase, catalase, reduced glutathione, and glutathione peroxidase levels. Treatment with ethanolic extract of *Tagetes erecta* flowers significantly restored these altered biochemical and antioxidant parameters. The high dose of extract, 400 mg/kg, showed marked hepatoprotective activity comparable to silymarin. Histopathological examination also showed reduced hepatocellular degeneration, necrosis, inflammation, and fatty changes in extract-treated animals.

The study concludes that the ethanolic extract of *Tagetes erecta* flowers possesses significant hepatoprotective activity, which may be attributed to its antioxidant, free-radical scavenging, and membrane-stabilizing properties.

Keywords: *Tagetes erecta* L.; marigold; hepatoprotective activity; paracetamol-induced hepatotoxicity; flavonoids; phenolic compounds; antioxidant activity; silymarin.

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1. Introduction

The liver is a vital organ responsible for metabolism, detoxification, storage of nutrients, synthesis of plasma proteins, and biotransformation of endogenous and exogenous substances [1-2]. Exposure to drugs, alcohol, environmental toxicants, and reactive metabolites may lead to liver injury, oxidative stress, inflammation, fibrosis, and hepatic dysfunction [3-4]. Paracetamol overdose is widely used in experimental studies because its toxic metabolite causes glutathione depletion, oxidative stress, lipid peroxidation, and hepatocellular necrosis [5].

Herbal drugs are receiving increasing attention as potential sources of safer hepatoprotective agents. Plant-derived flavonoids, phenolic compounds, tannins, carotenoids, and terpenoids may reduce hepatic oxidative stress by scavenging reactive oxygen species, preserving endogenous antioxidant enzymes, and stabilizing hepatocyte membranes [6-8].

Tagetes erecta L., commonly known as African marigold or genda, belongs to the family Asteraceae. Its flowers are rich in carotenoids, especially lutein, along with flavonoids and phenolic compounds [9]. Published work has reported antioxidant potential of flower extracts and supports their possible use as functional or nutraceutical ingredients [10].

Previous experimental work has also indicated that *Tagetes erecta* extract may protect against chemically induced hepatic injury through antioxidant and free-radical scavenging mechanisms [11-12]. Therefore, the present study was designed to standardize flowers of *Tagetes erecta* L. pharmacognostically and evaluate the hepatoprotective activity of its ethanolic extract against paracetamol-induced liver injury in Wistar rats.

2. Materials and Methods

2.1 Collection and Authentication of Plant Material

Fresh, healthy, fully bloomed flowers of *Tagetes erecta* L. were collected from the local area during the flowering season. The flowers were cleaned to remove dust, soil particles, and foreign matter. The plant material was authenticated by a qualified taxonomist, and a herbarium specimen was prepared and deposited for future reference.

Particular	Details
Botanical name	*Tagetes erecta* L.
Family	Asteraceae
Common name	African marigold / Genda
Plant part used	Flowers

2.2 Preparation of Powdered Drug and Extraction

The authenticated flowers were shade dried, coarsely powdered, and stored in airtight containers. The powdered drug was successively extracted using petroleum ether, chloroform, ethyl acetate, ethanol, and water. The extracts were concentrated and dried. Percentage yield was calculated using the following formula:

2.3 Pharmacognostical Evaluation

The flower drug was evaluated for organoleptic characters, macroscopic characters, transverse section features, powder microscopy, fluorescence behaviour, moisture content, ash values, extractive values, pH, and foreign matter.

2.4 Preliminary Phytochemical Screening

The ethanolic extract was screened qualitatively for alkaloids, carbohydrates, glycosides, flavonoids, phenolic compounds, tannins, saponins, steroids, terpenoids, proteins, amino acids, fixed oils, and fats using standard phytochemical tests.

2.5 Acute Oral Toxicity Study

The acute oral toxicity study was conducted according to OECD Guideline No. 423 after approval from the Institutional Animal Ethics Committee. The guideline uses an acute toxic class method for oral toxicity assessment.

The ethanolic extract was administered orally at doses of 300 mg/kg and 2000 mg/kg. Animals were observed for 14 days for mortality, behavioural changes, food intake, water intake, locomotor activity, tremors, convulsions, skin and fur condition, and body-weight variation.

2.6 Evaluation of Hepatoprotective Activity

Healthy Wistar rats were divided into five groups containing six animals each.

Group	Treatment
Group I	Normal control: vehicle only
Group II	Hepatotoxic control: paracetamol-treated
Group III	Standard group: silymarin 100 mg/kg + paracetamol
Group IV	*T. erecta* extract 200 mg/kg + paracetamol
Group V	*T. erecta* extract 400 mg/kg + paracetamol

The extract and standard drug were administered orally for the specified treatment period. Hepatotoxicity was induced using a hepatotoxic dose of paracetamol according to the approved experimental protocol. Serum was analysed for AST, ALT, ALP, total bilirubin, direct bilirubin, total protein, albumin, total cholesterol, and triglycerides.

Liver homogenate was analysed for superoxide dismutase, catalase, reduced glutathione, glutathione peroxidase, malondialdehyde, and lipid peroxidation. Liver tissue was preserved in 10% neutral buffered formalin for histopathological examination.

2.7 Statistical Analysis

The data were expressed as mean ± SEM. Statistical analysis was carried out using one-way analysis of variance followed by an appropriate multiple comparison test. A value of $p < 0.05$ was considered statistically significant.

3. Results and Discussion

3.1 Pharmacognostical Evaluation

The flowers of *Tagetes erecta* L. were bright yellow to orange-yellow, aromatic, slightly bitter, mildly pungent, globular to slightly flattened, and soft in texture. The inflorescence was a compact capitulum, with numerous ray and disc florets arranged on a common receptacle.

Microscopic evaluation showed epidermal cells, parenchymatous cells, vascular bundles, xylem vessels, pollen grains, stomata in green bract fragments, and covering trichomes. Powder microscopy showed yellowish-orange powder with pollen grains, trichomes, epidermal fragments, xylem vessels, and pigment-containing parenchymatous cells.

3.2 Physicochemical Parameters

Table 1: Physicochemical Parameters

S. No.	Parameter	Result
1	Loss on drying / Moisture content	8.42 ± 0.31 % w/w
2	Total ash value	7.86 ± 0.24 % w/w
3	Acid-insoluble ash value	0.68 ± 0.07 % w/w
4	Water-soluble ash value	2.14 ± 0.12 % w/w
5	Sulphated ash value	8.95 ± 0.28 % w/w
6	Alcohol-soluble extractive value	18.76± 0.45 % w/w
7	Water-soluble extractive value	21.48± 0.52 % w/w
8	pH of 1% aqueous solution	5.86 ± 0.09
9	Foreign matter	0.32 ± 0.05 % w/w

The low acid-insoluble ash value indicated minimal contamination with sand and siliceous material. The higher water-soluble extractive value suggested the presence of polar phytoconstituents, including phenolic compounds, flavonoids, tannins, and glycosides.

3.3 Percentage Yield of Extracts

Table 2: Percentage Yield of Extracts

Extract	Percentage yield (% w/w)
Petroleum ether extract	3.72
Chloroform extract	4.87
Ethyl acetate extract	7.96
Ethanolic extract	17.29
Aqueous extract	14.43

The ethanolic extract produced the maximum yield and was selected for further evaluation because ethanol effectively extracts polar and moderately polar bioactive constituents.

3.4 Preliminary Phytochemical Screening

Table 3: Phytochemical Screening

Phytoconstituent	Result
Alkaloids	Absent
Carbohydrates	Present
Glycosides	Present
Flavonoids	Moderately present
Phenolic compounds	Moderately present
Tannins	Moderately present
Saponins	Present
Steroids	Absent
Terpenoids	Present
Proteins and amino acids	Absent
Fixed oils and fats	Absent

The phytochemical profile showed predominance of flavonoids, phenolic compounds, and tannins. These constituents are known to possess antioxidant properties and may contribute to the hepatoprotective action of the flower extract.

3.5 Acute Oral Toxicity Study

No mortality or significant toxicity signs were observed at doses of 300 mg/kg and 2000 mg/kg during the 14-day observation period. Animals showed normal alertness, locomotor activity, food intake, water intake, respiration, fur condition, and body-weight gain.

Table 4: General Observation

Dose of Extract	No. of Animals	Mortality	General Observation
300 mg/kg	3	Nil	No signs of toxicity
2000 mg/kg	3	Nil	No mortality or abnormal behavioural signs

The oral LD_{50} was considered to be greater than 2000 mg/kg under the conditions of the study. Therefore, doses of 200 mg/kg and 400 mg/kg were selected for hepatoprotective evaluation.

3.6 Effect on Serum Biochemical Parameters

Paracetamol administration elevated serum AST, ALT, ALP, and bilirubin levels, confirming hepatic damage. Treatment with *T. Erecta* extract significantly restored these parameters towards normal levels. The high dose showed greater protection than the low dose.

Table 5: Effect on Serum Biochemical Parameters

Group	AST (U/L)	ALT (U/L)	ALP (U/L)	Total Bilirubin (mg/dL)
Normal control	82.46 ± 4.15	46.38 ± 3.24	118.62 ± 5.46	0.62 ± 0.05
Hepatotoxic control	214.58 ± 8.72	156.42 ± 7.85	286.36 ± 10.48	2.48 ± 0.14
Silymarin	98.74 ± 5.26***	58.64 ± 4.12***	136.48 ± 6.25***	0.78 ± 0.06***
T. erecta extract 200 mg/kg	148.26 ± 6.84***	101.38 ± 5.96***	198.54 ± 8.37***	1.42 ± 0.09***
T. erecta extract 400 mg/kg	112.68 ± 5.47***	72.16 ± 4.68***	152.36 ± 7.18***	0.96 ± 0.07***

Values are expressed as mean ± SEM, $n = 6$.
**$p < 0.001$ compared with hepatotoxic control group.

3.7 Effect on Protein and Lipid Parameters

The extract improved serum protein and albumin levels, indicating recovery of hepatic synthetic function. It also reduced elevated cholesterol and triglyceride levels, suggesting improvement in hepatic lipid metabolism.

Table 6: Effect on Protein and Lipid Parameters

Group	Total Protein (g/dL)	Albumin (g/dL)	Cholesterol (mg/dL)	Triglycerides (mg/dL)
Normal control	7.42 ± 0.18	4.18 ± 0.12	82.46 ± 3.84	78.24 ± 4.15
Hepatotoxic control	4.68 ± 0.21	2.36 ± 0.14	142.58 ± 5.76	136.42 ± 6.28
Silymarin	7.12 ± 0.16***	4.02 ± 0.10***	88.36 ± 4.12***	84.68 ± 3.96***
T. erecta extract 200 mg/kg	5.86 ± 0.19***	3.18 ± 0.11***	112.46 ± 4.98***	108.52 ± 5.12***
T. erecta extract 400 mg/kg	6.74 ± 0.17***	3.76 ± 0.13***	94.28 ± 4.36***	92.16 ± 4.28***

3.8 Effect on Antioxidant Enzymes

The hepatotoxic control group showed marked depletion of antioxidant defence enzymes. Extract treatment significantly restored SOD, catalase, reduced glutathione, and glutathione peroxidase levels. This suggests that the extract protects liver tissue through antioxidant-mediated mechanisms.

Table 7: Effect on Antioxidant Enzymes

Group	SOD (U/mg protein)	Catalase	Reduced Glutathione	Glutathione Peroxidase
Normal control	12.84 ± 0.56	48.62 ± 2.18	9.48 ± 0.42	8.36 ± 0.35
Hepatotoxic control	5.26 ± 0.34	20.48 ± 1.42	3.82 ± 0.27	3.46 ± 0.24
Silymarin	11.96 ± 0.48***	45.18 ± 1.96***	8.96 ± 0.38***	7.94 ± 0.31***
T. erecta extract 200 mg/kg	8.54 ± 0.42***	33.26 ± 1.68***	6.48 ± 0.31***	5.86 ± 0.28***
T. erecta extract 400 mg/kg	10.64 ± 0.46***	41.72 ± 1.84***	8.12 ± 0.36***	7.18 ± 0.30***

3.9 Effect on Lipid Peroxidation

The extract significantly reduced elevated malondialdehyde and lipid peroxidation values in paracetamol-treated animals. This indicates protection of hepatocyte membranes from oxidative injury.

Table 8: Effect on Lipid Peroxidation

Group	Malondialdehyde (nmol/mg protein)	Lipid Peroxidation (nmol MDA/min/mg protein)
Normal control	2.18 ± 0.16	2.42 ± 0.18
Hepatotoxic control	6.84 ± 0.32	7.26 ± 0.36
Silymarin	2.56 ± 0.18***	2.78 ± 0.21***
T. erecta extract 200 mg/kg	4.26 ± 0.24***	4.58 ± 0.28***
T. erecta extract 400 mg/kg	3.08 ± 0.20***	3.34 ± 0.22***

3.10 Histopathological Findings

The normal control group showed normal hepatic architecture with regularly arranged hepatocytes, normal central vein, and clear sinusoids. The hepatotoxic control group showed severe hepatocellular degeneration, necrosis, inflammatory infiltration, fatty changes, and sinusoidal congestion.

The silymarin-treated group showed near-normal hepatic architecture. The extract-treated groups showed dose-dependent recovery. The high-dose extract group showed mild hepatocellular degeneration, minimal inflammatory infiltration, and substantial restoration of normal liver architecture.

Table 9: Histopathological Observation

Group	Histopathological Observation
Normal control	Normal hepatic architecture
Hepatotoxic control	Severe degeneration, necrosis, fatty changes, and inflammation
Silymarin	Near-normal architecture with minimal degeneration
T. erecta extract 200 mg/kg	Moderate protection with mild degeneration
T. erecta extract 400 mg/kg	Marked protection with minimal degeneration and inflammation

4. Discussion

The pharmacognostical findings established preliminary standards for the flowers of *Tagetes erecta* L. The organoleptic, macroscopic, microscopic, fluorescence and physicochemical parameters can be used for the identification and quality control of the crude drug.

The ethanolic extract demonstrated the highest extractive yield and contained flavonoids, phenolic compounds, tannins, glycosides, saponins, and terpenoids. These classes of compounds may contribute to the observed antioxidant activity. Flower extracts of *Tagetes erecta* have been reported to contain phenolics, flavonoids, and carotenoids, including lutein, with relevant antioxidant properties.

Paracetamol overdose caused hepatic oxidative stress, depletion of glutathione, elevation of liver enzyme markers, and tissue damage. In the present study, the ethanolic extract significantly reduced AST, ALT, ALP, bilirubin, cholesterol, triglycerides, malondialdehyde, and lipid peroxidation levels. It also restored total protein, albumin, SOD, catalase, reduced glutathione, and glutathione peroxidase levels.

Silymarin showed the maximum protective effect, as expected from its established antioxidant and hepatoprotective role in paracetamol-induced injury. However, the high dose of *Tagetes erecta* extract showed substantial restoration of biochemical and histological parameters, indicating promising hepatoprotective activity.

The possible mechanism of hepatoprotection may involve scavenging of reactive oxygen species, prevention of glutathione depletion, inhibition of lipid peroxidation, stabilization of hepatocyte membranes, preservation of endogenous antioxidant enzymes, and attenuation of inflammation. The present findings are in agreement with earlier reports of hepatoprotective action of *Tagetes erecta* extract in chemically induced liver injury models.

5. Conclusion

The present study established preliminary pharmacognostical standards for flowers of *Tagetes erecta* L. The ethanolic extract showed the highest percentage yield and contained flavonoids, phenolic compounds, tannins, glycosides, saponins, and terpenoids.

The extract was safe up to 2000 mg/kg body weight in the acute oral toxicity study. In paracetamol-induced hepatotoxicity, the ethanolic flower extract significantly restored serum liver markers, antioxidant enzymes, protein levels, lipid profile, and histopathological features towards normal values.

The high dose of extract, 400 mg/kg body weight, demonstrated marked hepatoprotective activity and showed effects closer to the standard drug silymarin. The hepatoprotective effect may be attributed to flavonoids, phenolic compounds, carotenoids, tannins, and other antioxidant phytoconstituents present in *Tagetes erecta* flowers.

Further work involving isolation of active constituents, marker-based standardization, chronic toxicity studies, molecular mechanism studies, formulation development, and clinical evaluation is recommended.

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