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Phytochemical and pharmacological study of the flowers of *Capparis spinosa* L. for anti-inflammatory activity

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Abstract

Inflammation is involved in numerous acute and chronic pathological conditions, while prolonged use of conventional anti-inflammatory drugs is associated with important adverse effects. The present investigation evaluated the pharmacognostical, phytochemical and anti-inflammatory properties of a hydroalcoholic extract of *Capparis spinosa* L. flowers (HECSF). Authenticated flowers were subjected to organoleptic, macroscopic, microscopic and physicochemical evaluation. The shade-dried powder was defatted and extracted with 70% ethanol by maceration. Preliminary phytochemical screening and acute oral toxicity testing were performed, followed by carrageenan-induced paw oedema and cotton-pellet-induced granuloma studies in Wistar rats. Extraction of 500 g of flower powder produced 68.40 g of dried extract, corresponding to a yield of 13.68% w/w. The extract showed prominent reactions for phenolics and flavonoids and moderate reactions for tannins, glycosides and terpenoids. No mortality or evident toxicity was observed at 2000 mg/kg during the 14-day acute-toxicity assessment. HECSF produced dose-dependent inhibition of carrageenan-induced paw oedema. At the fifth hour, inhibition was 32.14% and 48.21% at 200 and 400 mg/kg, respectively, compared with 66.07% for diclofenac sodium. In the cotton-pellet model, HECSF produced 20.13% and 34.28% inhibition of granuloma formation, whereas diclofenac produced 45.60% inhibition. The findings indicate that HECSF possesses promising dose-dependent activity against acute and chronic inflammation. Phenolics, flavonoids, tannins and terpenoids may contribute collectively to the observed effects. However, isolation of active constituents, chemical standardization and pathway-specific investigations are required before a definite mechanism or therapeutic application can be established.

Keywords: *Capparis spinosa*; flowers; phytochemical screening; carrageenan; paw oedema; granuloma; anti-inflammatory activity.

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1. Introduction

Inflammation is a protective biological response to infection, tissue injury and chemical or physical irritation. It involves coordinated vascular, cellular and molecular events intended to eliminate the initiating stimulus and facilitate tissue repair. Although an appropriately regulated inflammatory response is beneficial, excessive or persistent inflammation contributes to arthritis, inflammatory bowel disease, cardiovascular disorders, metabolic diseases and several neurodegenerative conditions [1].

The inflammatory response is mediated by histamine, serotonin, bradykinin, prostaglandins, leukotrienes, nitric oxide, reactive oxygen species and cytokines such as tumour necrosis factor-α, interleukin-1β and interleukin-6. Non-steroidal anti-inflammatory drugs reduce inflammation principally by inhibiting cyclooxygenase-mediated prostaglandin synthesis. However, prolonged use may cause gastrointestinal, renal and cardiovascular adverse effects, supporting the search for safer plant-derived anti-inflammatory agents [2].

Capparis spinosa L., commonly known as the caper bush, is a perennial shrub belonging to the family Capparaceae. Its different parts have been used traditionally for rheumatic, digestive and inflammatory complaints. Caper tissues contain chemically diverse secondary metabolites, including flavonoids, phenolic acids, alkaloids, glucosinolates, sterols and terpenoids [3].

Flower buds of *C. spinosa* contain flavonols, hydroxycinnamic acids and other polyphenolic constituents. Their composition and biological activity vary with developmental stage, genotype and environmental conditions. Rutin has been reported in different parts of the plant, with flowering buds showing considerable but geographically variable concentrations [4].

Experimental investigations also support the anti-inflammatory potential of *C. spinosa*. A flower-bud extract reduced carrageenan-induced paw oedema and inhibited TNF-α, IL-1β, leukotriene B_4 and superoxide-anion production. Other preparations modulated IL-17 and IL-4 expression in human peripheral blood mononuclear cells, suggesting immunomodulatory activity [5].

Despite these findings, comparatively limited information is available on the combined pharmacognostical standardization and in vivo anti-inflammatory evaluation of fully developed *C. spinosa* flowers. Therefore, the present study evaluated the identity, physicochemical characteristics, phytochemical composition, acute oral safety and anti-inflammatory activity of a hydroalcoholic flower extract [6].

2. Materials and Methods

2.1 Plant Material and Authentication

Fresh, mature and disease-free flowers of *Capparis spinosa* L. were collected from local market, Bhopal, M.P. during January, 2026. The material was authenticated by Dr. Sana Naz, Safia College, Bhopal, M.P. A voucher specimen bearing number 134/IAEC/2026 was deposited in the institutional herbarium.

The flowers were cleaned, shade-dried at room temperature, coarsely powdered and stored in an airtight container.

2.2 Chemicals and Reagents

Ethanol, petroleum ether, hydrochloric acid, sulphuric acid, ferric chloride, Dragendorff's reagent, Mayer's reagent, aluminium chloride and other analytical-grade reagents were used. Carrageenan was used to induce paw oedema, while diclofenac sodium served as the standard anti-inflammatory drug.

2.3 Pharmacognostical Evaluation

Fresh flowers were evaluated for colour, odour, taste, texture, size, floral symmetry, sepals, petals, stamens, ovary and gynophore.

Thin transverse sections of petals and sepals were prepared, stained and examined under a compound microscope. Powder microscopy was performed to identify epidermal fragments, pollen grains, anther-wall fragments, fibres, parenchymatous cells and vascular elements.

2.4 Physicochemical Evaluation

Foreign organic matter, loss on drying, total ash, acid-insoluble ash, water-soluble ash, alcohol-soluble extractive value, water-soluble extractive value, foaming index and swelling index were determined using standard pharmacognostical procedures. Each determination was performed in triplicate.

2.5 Preparation of Hydroalcoholic Extract

Approximately 500 g of powdered flowers was defatted with petroleum ether. The dried marc was extracted with 70% ethanol by maceration for 72 hours with intermittent shaking.

The extract was filtered and concentrated under reduced pressure at a temperature not exceeding approximately 40–45°C. The concentrated extract was dried to a constant weight and designated HECSF.

Percentage yield=Weight of plant powder/Weight of dried ext ract×100

2.6 Preliminary Phytochemical Screening

HECSF was screened qualitatively for:

- Alkaloids
- Flavonoids
- Phenolic compounds
- Tannins
- Saponins
- Glycosides
- Steroids
- Terpenoids
- Carbohydrates
- Proteins and amino acids

Reaction intensity was recorded as absent (–), weak (+), moderate (++) or strong (+++).

2.7 Experimental Animals

Healthy adult Wistar albino rats weighing approximately 150–200 g were maintained under controlled laboratory conditions with a 12-hour light–dark cycle, standard pellet diet and water ad libitum.

The protocol was approved by the Institutional Animal Ethics Committee under approval number 134/IAEC/2026, and the study was conducted in a CPCSEA-registered animal facility bearing registration number.

2.8 Acute Oral Toxicity

Acute oral toxicity was evaluated according to OECD Guideline 423. Female Wistar rats received a single oral dose of HECSF at 2000 mg/kg and were observed for 14

days for mortality, behavioural changes, salivation, tremors, convulsions, respiratory abnormalities, food and water consumption and body-weight changes. Gross necropsy was performed at study termination. Based on the toxicity findings, 200 and 400 mg/kg were selected as the low and high pharmacological doses.

2.9 Carrageenan-Induced Paw Oedema

The experiment was based on the method of Winter et al. (1962). Animals were divided into five groups of six rats [7]:

Table 1: Experimental Groups

Group	Treatment
I	Normal control
II	Vehicle + carrageenan
III	Diclofenac sodium 10 mg/kg + carrageenan
IV	HECSF 200 mg/kg + carrageenan
V	HECSF 400 mg/kg + carrageenan

Treatments were administered orally before subplantar carrageenan administration. Paw volume was measured before carrageenan and at 1, 2, 3, 4 and 5 hours using a plethysmometer.

$\Delta V = Vt - V0$ Inhibition of paw oedema (%)$= \Delta Vc \Delta Vc - \Delta VT \times 100$

Where ΔVc and ΔVT represent mean paw-volume increases in the carrageenan-control and treated groups, respectively.

2.10 Cotton-Pellet-Induced Granuloma

Sterile cotton pellets weighing 10 mg were implanted subcutaneously under an IAEC-approved anaesthetic and aseptic procedure. The animals received vehicle, diclofenac sodium or HECSF once daily for seven days.

At the end of the treatment period, the pellet–granuloma complexes were removed and weighed. They were then dried to a constant weight.

Net dry granuloma weight$= Wfd - Wip$

Granuloma inhibition (%)$= WcWc - Wt \times 100$

Where Wc and Wt represent mean net dry granuloma weights in the vehicle-control and treated groups.

2.11 Statistical Analysis

Results were expressed as mean ± SEM. Paw-volume data should be analysed using repeated-measures ANOVA or a mixed-effects model, followed by an appropriate multiplicity-adjusted comparison. Granuloma data may be analysed using one-way ANOVA followed by a suitable post-hoc test. A value of $p<0.05$ was considered statistically significant.

3. Results and Discussion

3.1 Pharmacognostical Characteristics

The fresh flowers were white to pinkish-white, with numerous pinkish-purple staminal filaments. They possessed a mild characteristic odour, slightly bitter taste and soft, delicate texture.

The flowers were solitary, bisexual and actinomorphic, with four free sepals, four free petals, numerous elongated stamens and a superior ovary elevated on a conspicuous gynophore.

Microscopy showed single-layered petal epidermises, papillose projections, loosely arranged parenchymatous mesophyll and small vascular strands. Powder microscopy revealed papillose petal-epidermal fragments, sepal epidermis, numerous pollen grains, fibrous anther-wall fragments, parenchymatous cells and spiral and reticulate vessels.

These features may assist in the identification of genuine flower material and the detection of adulteration or substitution.

3.2 Physicochemical Parameters

Table 2: Physicochemical Parameters

Parameter	Result
Foreign organic matter	0.42±0.03%
Loss on drying	8.46±0.17%
Total ash	7.12±0.14%
Acid-insoluble ash	0.86±0.04%
Water-soluble ash	3.14±0.09%
Alcohol-soluble extractive	18.62±0.31%
Water-soluble extractive	23.48±0.37%
Foaming index	<100
Swelling index	2.80±0.10 mL/g

Values are mean ± SD, n=3.

Low foreign-matter and acid-insoluble-ash values indicated satisfactory cleaning and limited contamination with soil or siliceous materials. The higher water-soluble extractive value suggested that polar constituents constituted a substantial fraction of the flowers.

Direct comparison with other reports should be made cautiously because the phytochemical composition of caper material is affected by plant part, developmental stage, geographical source and extraction solvent. Flower-bud polyphenol profiles have been shown to change during development [8].

3.3 Extractive Yield

Extraction of 500 g of flower powder produced 68.40 g of dried HECSF.

Percentage yield$= 50068.40 \times 100 = 13.68\%$ w/w

The yield indicated appreciable recovery of hydroalcoholic-soluble constituents.

3.4 Phytochemical Screening

Table 3: Phytochemical Screening

Phytochemical class	Result
Alkaloids	+
Flavonoids	+++
Phenolic compounds	+++
Tannins	++
Saponins	+
Glycosides	++
Anthraquinone glycosides	–
Steroids	+
Terpenoids	++
Carbohydrates	++
Proteins	–
Amino acids	+

Strong reactions for phenolic compounds and flavonoids were consistent with previous studies demonstrating the polyphenolic richness of caper buds and flowers. Flavonoids such as rutin and related compounds have been detected in different *C. spinosa* tissues, although their abundance varies among plant parts and locations.

The qualitative tests did not identify individual compounds. Therefore, the presence of rutin, quercetin or another specific marker must be confirmed using validated chromatographic analysis.

3.5 Acute Oral Toxicity

No mortality or evident toxicity was observed after oral administration of HECSF at 2000 mg/kg. The animals showed normal activity, feeding, drinking and body-weight gain. Salivation, tremors, convulsions, respiratory abnormalities, paralysis and coma were absent.

No treatment-related gross lesions were identified at necropsy. HECSF was therefore considered tolerated at the tested limit dose under the study conditions. This finding supports only acute oral tolerance and does not establish chronic or clinical safety.

3.6 Carrageenan-Induced Paw Oedema

Carrageenan produced a marked increase in paw volume, reaching a maximum at the third hour.

Table 4: Effect of HECSF on Paw volume

Treatment	3-hour inhibition	4-hour inhibition	5-hour inhibition
Diclofenac sodium	53.95%	61.76%	66.07%
HECSF 200 mg/kg	23.68%	27.94%	32.14%
HECSF 400 mg/kg	35.53%	42.65%	48.21%

HECSF reduced paw oedema in a dose-dependent manner. At the fifth hour, the 400 mg/kg dose produced 48.21% inhibition compared with 32.14% at 200 mg/kg. Diclofenac sodium remained the most effective treatment.

The carrageenan paw-oedema model is a standard method for evaluating acute anti-inflammatory activity. The greater activity observed during the later phase is compatible with an influence on prostaglandin-associated processes, but prostaglandins and cyclooxygenase activity were not measured.

The present findings are directionally consistent with a study in which *C. spinosa* flower-bud extract at 200 and 400 mg/kg reduced carrageenan-induced oedema and inhibited several inflammatory mediators.

3.7 Cotton-Pellet-Induced Granuloma

Table 4: Effect of HECSF on Granuloma

Treatment	Wet weight (mg)	Net dry granuloma weight (mg)	Inhibition
Vehicle control	96.40±2.76	31.80±1.18	—
Diclofenac sodium	62.80±2.11	17.30±0.92	45.60%
HECSF 200 mg/kg	82.10±2.35	25.40±1.06	20.13%
HECSF 400 mg/kg	72.30±2.18	20.90±0.97	34.28%

Values are mean ± SEM, n=6.

HECSF reduced wet and net dry granuloma weights in a dose-dependent manner. Reduction in wet weight suggested decreased inflammatory exudate accumulation, whereas reduced dry weight indicated inhibition of proliferative tissue formation.

The high-dose extract produced 34.28% inhibition, approaching but not equalling the 45.60% inhibition produced by diclofenac sodium. Activity in both the paw-oedema and granuloma models suggested that the extract influenced acute exudative as well as chronic proliferative inflammation.

3.8 Probable Contribution of Phytoconstituents

The observed activity may reflect the combined action of phenolics, flavonoids, tannins, glycosides and terpenoids. These constituent classes can potentially influence oxidative and inflammatory signalling. Previous work has shown that *C. spinosa* preparations can modulate cytokine expression, including suppression of IL-17 and enhancement of IL-4 expression.

Kernouf et al. (2018) reported inhibition of TNF-α, IL-1β, leukotriene B_4 and superoxide-anion generation by a flower-bud extract. These findings support mechanistic plausibility but cannot be directly assigned to HECSF because the extracts, plant material and experimental endpoints differed.

No direct measurements of histamine, serotonin, bradykinin, prostaglandins, COX, LOX, NF-κB or cytokines were performed in the present study. Consequently, the results demonstrate pharmacological activity at the crude-extract level but do not establish a molecular mechanism.

4. Conclusion

The hydroalcoholic extract of *Capparis spinosa* L. flowers demonstrated dose-dependent anti-inflammatory activity in carrageenan-induced paw oedema and cotton-pellet-induced granuloma models.

HECSF at 400 mg/kg produced 48.21% inhibition of paw oedema at the fifth hour and 34.28% inhibition of granuloma formation. The extract was less active than diclofenac sodium but produced measurable effects against both acute and chronic inflammation.

Preliminary phytochemical screening indicated the presence of phenolics, flavonoids, tannins, glycosides, terpenoids and other constituents that may contribute collectively to the observed activity. Nevertheless, a particular constituent or inflammatory pathway cannot be considered responsible without direct chemical and mechanistic evidence.

Further studies should include:

- Quantitative phenolic and flavonoid estimation
- Validated HPTLC, HPLC or LC–MS standardization
- Isolation and structural characterization of active compounds
- Antioxidant and in vitro anti-inflammatory assays
- COX-1, COX-2 and 5-LOX inhibition
- Prostaglandin and cytokine estimation
- NF-κB and iNOS expression studies
- Histopathological confirmation
- Subacute and chronic toxicity assessment
- Pharmacokinetic and bioavailability investigations

Overall, *C. spinosa* flowers represent a promising source of plant-derived anti-inflammatory constituents, but verified experimental replication and mechanistic confirmation are required before therapeutic conclusions can be drawn.

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