Pharmacological Evaluation of Momordica Charantia for Anti-Inflammatory and Wound Healing Potential

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ABSTRACT

The present study aimed to evaluate the anti-inflammatory effects and wound healing potential of Momordica charantia fruit extracts. Plant material was subjected to maceration and Soxhlet extraction, and the resulting extracts were analyzed for physicochemical parameters including water- and alcohol-soluble extractive values, as well as acid and water-soluble ash content. Quantitative phytochemical analysis confirmed the presence of active constituents, with the methanolic extract showing a total phenolic content of 72.22 mg GAE/g and a total flavonoid content of 72.23 mg GAE/g. Pharmacological evaluation was performed using two animal models: carrageenan-induced paw edema in rats for anti-inflammatory activity, and the incision wound model for wound healing potential. The ethyl acetate extract at 20 mg/kg body weight demonstrated maximum inhibition of inflammation (76.92%), which was comparable to the standard drug indomethacin (10 mg, 82.69%). In wound healing studies, the methanolic extract exhibited superior tensile strength (512.62 ± 1.23 g) compared to EMC (489.60 ± 0.58 g), EAMC (338.63 ± 1.66 g), and the standard formulation. Overall, the findings indicate that Momordica charantia possesses significant anti-inflammatory activity and enhanced wound healing properties, with the methanolic extract showing particularly promising results for potential therapeutic and commercial applications.

Keywords: Momordica Charantia, anti-inflammatory, wound, healing, ash value

INTRODUCTION

Inflammation is the body's defense response to harmful stimuli such as allergens or tissue injury. However, when the inflammatory response becomes uncontrolled, it can lead to a wide range of disorders, including allergies, cardiovascular dysfunctions, metabolic syndrome, cancer, and autoimmune diseases [1]. These conditions impose a significant economic burden on individuals and, consequently, on society. Various medicines are available to control and suppress inflammatory crises, including steroids, non-steroidal anti-inflammatory drugs, and natural anti-inflammatory agents [2]. The integration of natural factors into medication therapy aims to enhance pharmacological effectiveness while minimizing unwanted side effects. Phyto-chemical studies have been conducted on approximately 5,000 plant species, with nearly 1,100 species extensively used in Ayurvedic, Unani, and Allopathic medicine. In fact, active plant extract screening programs consistently contribute to the discovery of new drugs, highlighting the ongoing importance of natural sources in modern pharmacology [3].

Figure 1: Fruit of *M. Charantia*

Momordica charantia L. (bitter melon) is widely cultivated and belongs to the gourd family. The ripe fruits, leaves, and seeds of this species are harvested for consumption as a vegetable, while the spices and leaves are often mixed with water to prepare a drink [4]. This plant has been reported to be useful in the treatment of numerous diseases due to its diverse biological activities, including antidiabetic, antioxidant, antibacterial, antimicrobial, antiviral, antifungal, deworming, insecticidal, antitoxic, anti-inflammatory, antithrombotic, anti-allergic, estrogenic, lipid-lowering, anticancer, antiproliferative, and antiasthenic effects [5]. Furthermore, studies have shown that prepared scaffolds from M. charantia exhibit a broad spectrum of antibacterial activity against both gram-positive and gram-negative bacteria [6]. The aim of the present research was to investigate the wound-healing and anti-inflammatory potential of Momordica charantia.

MATERIAL AND METHOD

Plant Collection

Momordica charantia, belonging to the family Cucurbitaceae, was collected in June 2021 from Bhopal, Madhya Pradesh. The plant and fruit were authenticated by Dr. Hussain ul, Bhopal. The specimen, registered under sample number 4825/BBS/BHO/35, has been preserved for future reference and identification. The time of collection plays a crucial role in determining the presence and concentration of active constituents in the plant material. For extraction of these constituents, standard methods such as maceration, percolation, and Soxhlet extraction were employed [7].

Phytochemical Analysis

Determination of Water and Alcohol soluble extractive value

Five grams of powdered drug were macerated with 100 mL of ethanol in a round-bottom flask for 24 hours. The mixture was occasionally shaken for 6 hours and then allowed to stand undisturbed for 18 hours. After filtration, the filtrate was evaporated to dryness in a tarred flat-bottom shallow dish [8]. The residue was dried at 105 °C and weighed. The percentage of ethanol-soluble extractive value was then calculated with reference to the air-dried drug [9].

Determination of total ash

Total ash determination was carried out by weighing 2–3 g of the air-dried crude drug into a tared platinum or silica dish. The sample was then incinerated at a temperature not exceeding 450 °C until free from carbon. After incineration, the dish was cooled and weighed to obtain the total ash value [10].

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Determination of acid insoluble ash

Acid-insoluble ash determination was performed by treating the ash obtained from the previous process with 25 mL of 2 M hydrochloric acid. The mixture was boiled for 5 minutes, and the residue obtained after extraction represented the acid-insoluble portion. The percentage of acid-insoluble ash was calculated with reference to 100 g of the air-dried drug [11].

Determination of water-soluble ash

Water-soluble ash determination was carried out by boiling the ash with 25 mL of water for 5 minutes. The insoluble matter was collected on ashless filter paper, washed with hot water, and then ignited for 15 minutes at a temperature not exceeding 450 °C [12]. The weight of the insoluble matter was subtracted from the total weight of the ash, and the difference represented the water-soluble ash. The percentage of water-soluble ash was calculated with reference to the air-dried drug [13].

Successive solvent extraction

For the extraction of phytoconstituents, solvents such as ethyl acetate, ethanol, and methanol were employed. The powdered drug was subjected to successive Soxhlet extraction using a single-solvent system. Initially, the powdered drug was packed into the apparatus and defatted with petroleum ether. Successive extraction was then carried out sequentially with ethyl acetate, ethanol, and methanol [14]. The resulting residues were dried first at room temperature and subsequently under reduced pressure. Each extract was stored in an airtight container, and the residue was weighed prior to packing. The percentage yield was calculated on a weight-by-weight (w/w) basis with reference to the powdered drug [15].

Qualitative Test Analysis

The dried extracts were subjected to chemical tests for the detection of phyto-constituents such as alkaloids, flavonoids, tannins, sterols, phenolic compounds, terpenoids, and carbohydrates. These preliminary phyto-chemical screenings provided valuable information regarding the presence of bioactive compounds in the plant material, which are responsible for its diverse pharmacological activities [16].

Determination of Total Phenolic Content

The total phenolic content of all extracts was determined using the Folin–Ciocalteu method. A standard calibration curve of gallic acid was constructed by preparing dilutions of 5, 10, 15, 20, 25, and 30 µg/mL in methanol from a stock solution of gallic acid. The procedure followed was as described by Shukla et al. (2018) [17].

Determination of total flavonoid content

The flavonoid content of the extracts was determined using quercetin as the standard, and the results were expressed as quercetin equivalents. The procedure adopted followed the method described by Shukla et al. (2018) [17].

Pharmacological Evaluation

Anti-inflammatory Activity

The fruit extracts Momordica Charantia was evaluated for the anti-inflammatory potential.

Screening for anti-inflammatory (Reticence of Carrageenan induced, paw edema in rats)

The animals were divided into five groups: Panel 1 (Control), Panel 2 (Ethanolic extract of Momordica charantia; EMC), Panel 3 (Ethyl acetate extract of Momordica charantia; EAMC), Panel 4 (Methanol extract of Momordica charantia; MMC), and Panel 5 (Indomethacin). The control group received normal saline (0.5 mL) one hour prior to carrageenan infusion. In this experiment, edema was induced by subcutaneous administration

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of 0.1 mL of 1% carrageenan, and the progression of paw edema was measured using a plethysmometer. Dose–response relationship data were recorded following intraperitoneal or oral administration, with oral dosages showing the most pronounced anti-inflammatory effect. The standard drug, Indomethacin, administered orally at a concentration of 2.5 mg/kg one hour before carrageenan injection, served as the reference treatment for edema management [18].

Wound healing potential/ activity

Incision wound model

In the incision wound model, albino rats were depilated by removing hair from the dorsal thoracic region prior to wounding. The animals were anesthetized with diethyl ether, and paravertebral incisions measuring six centimeters in length were made through the full thickness of the skin on either side of the vertebral column. The wounds were closed with interrupted sutures placed one centimeter apart. Thirty-six albino rats were divided into six panels, each consisting of six animals. For the incision wound studies, Panel I served as the control group, Panel II received the standard drug combination, Panel III was treated topically with ethanolic extract of Momordica charantia (EMC), Panel IV received ethyl acetate extract of Momordica charantia (EAMC), and Panel V received methanol extract of Momordica charantia (MMC). All treatments were applied once daily for ten days, beginning on the day of wounding. Sutures were removed on the eighth post-wounding day, and the tensile strength of the wounds was measured on the tenth day using the continuous water flow technique [19].

Tensile strength in Incision wound model

The tensile strength of the incision wounds was evaluated on the 10th day. The rats were anesthetized and placed on a stack of paper towels positioned on the middle of a board, with the height adjusted so that the wound was aligned with the tips of the arms. Clamps were carefully applied to the skin on opposite sides of the wound, approximately 0.3 cm away from the incision [20]. A fishing line was attached to the clamps, passed over a pulley, and connected to a polyethylene bottle. The board was adjusted so that the bottle received water at a rapid and constant rate from a large reservoir until the wound began to open. The amount of water collected in the polyethylene bottle was weighed, and this weight was considered as the tensile strength of the wound [21].

RESULTS AND DISCUSSION

Physicochemical properties of drugs

The water-soluble extractive value of Momordica charantia was found to be $25.06 \pm 0.13\%$ (w/w), indicating the presence of sugars, acids, and inorganic compounds. In contrast, the alcohol-soluble extractive value was determined to be $12.52 \pm 0.16\%$ (w/w), representing the presence of polar constituents such as phenols, alkaloids, steroids, glycosides, and flavonoids. These values highlight the diverse phytoconstituents present in the plant and provide insight into its chemical nature.

Extraction

Soxhlet apparatus was used for extraction of powder drug of Momordica Charantia. Percentage yield was shown in Table 1.

Table 1: Qualitative estimation

Herbal Drug	Solvent system	Color	Yield (in gm)
Momordica Charantia Fruit	Ethanol: ethyl acetate	Greenish Yellow	3.12
	Ethanolic extract (EMC)	Brown	4.65
	Methanolic extract (MMC)	Dark Brown	8.99
	Ethyl acetate extract (EAMC)	Greenish brown	6.20

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Phytochemical Screening

Total phenolic compounds

The total phenolic content of the extracts of Momordica charantia was quantified using the Folin–Ciocalteu method. Gallic acid was used as the reference standard, and a calibration curve was constructed by preparing dilutions of 5, 10, 15, 20, 25, and 30 µg/mL in methanol. The absorbance values were analyzed by UV spectrophotometry, and the phenolic content was expressed in terms of gallic acid equivalents. The standard curve equation was $y = 0.07x – 0.0322$ ($R^2 = 0.999$), as shown in Figure 2, with the results reported in mg of gallic acid equivalent per gram of extract (Table 2).

Table 2: Standard Curve of Gallic Acid

Concentration (µg/ml)	Absorbance (Mean)
5	0.316±0.020
10	0.652±0.015
15	0.992±0.010
20	1.345±0.020
25	1.720±0.012
30	2.100±0.014

The methanolic extract of fruits of *Momordica charantia* has shown highest amount of total phenolic content (72.220 mgGAE/g) as compared to ethanolic and ethyl acetate extract that contains the 66.306 and 58.125 mgGAE/g phenolic content (Table 3).

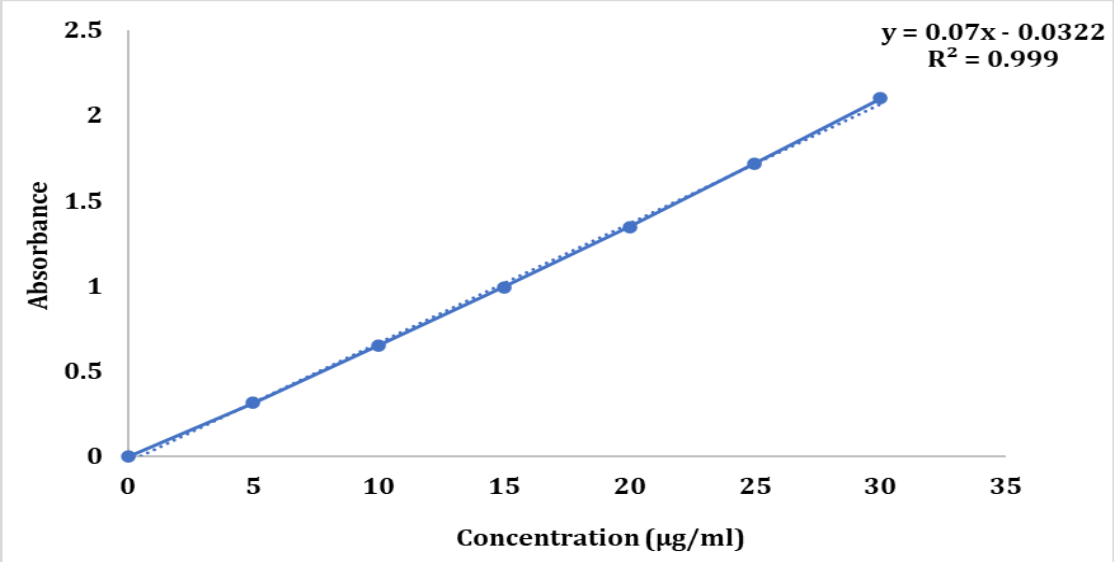

Figure 2: Standard curve of Gallic acid

Table 3: Total Phenolic Content in Different Extracts

Extracts	Conc. (mgGAE/g)
Ethanolic extract of Momordica Charantia (EMC)	66.306
Methanol extract of Momordica Charantia (MMC)	72.220
Ethyl acetate extract of Momordica Charantia (EAMC)	58.125

Determination of Total Flavonoid Content

The flavonoid content of the Momordica charantia plant extract was assessed and quantified using a UV spectrophotometric method. Aluminum chloride ($AlCl_3$) was employed as the reagent for flavonoid determination. A standard calibration curve was prepared using quercetin, which served as the reference

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compound. The regression equation obtained from the standard curve was expressed as: y=0.0176x−0.0051 (R2=0.999). Flavonoid content in the extract was quantified and reported as milligrams of quercetin equivalents (QE) per gram of extract (see Tables 4 and 5). The calibration curve of quercetin is illustrated in Figure 3.

Table 4: Standard Curve of Quercetin

Concentration (µg/ml)	Absorbance (Mean)
5	0.079±0.020
10	0.172±0.030
15	0.252±0.034
20	0.348±0.029
25	0.438±0.024

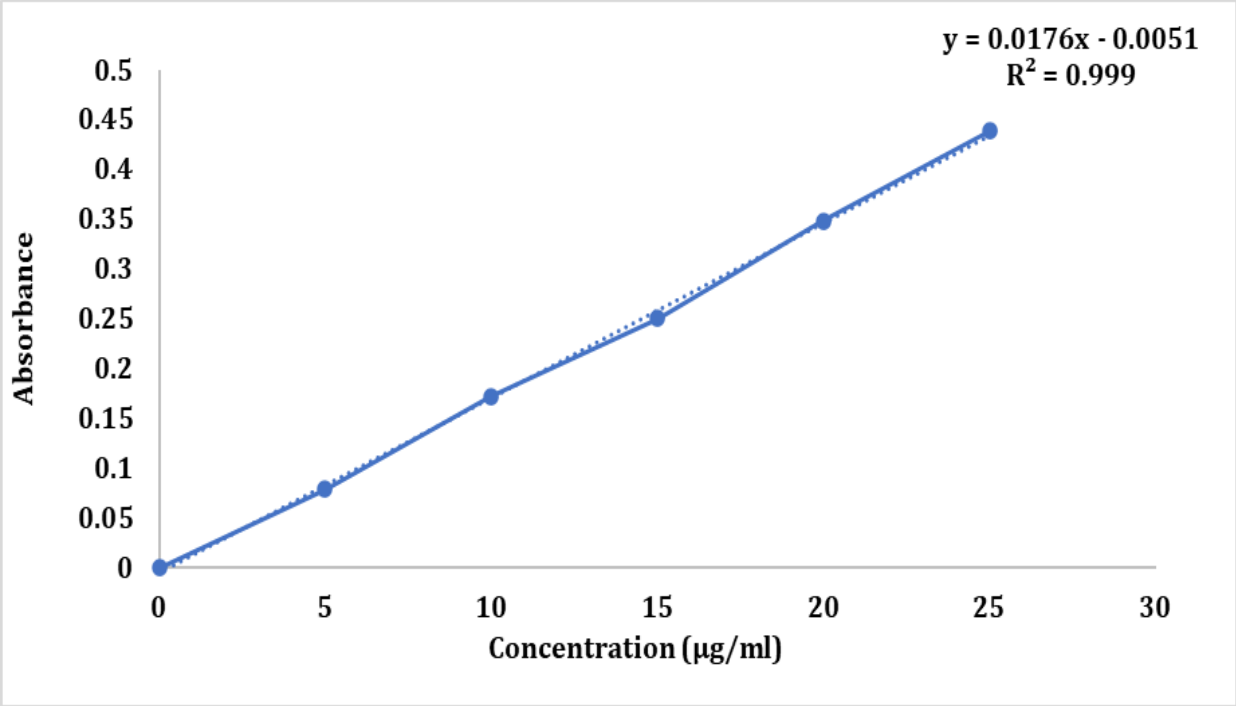

Figure 3: Calibration curve of Quercetin

Table 5: Total Flavonoid content (mg/g)

Extracts	Qty. (mgQE/g)
Ethanolic extract of Momordica Charantia (EMC)	65.32
Methanol extract of Momordica Charantia (MMC)	72.23
Ethyl acetate extract of Momordica Charantia (EAMC)	69.82

The methanolic extract of fruits of *Momordica charantia* has shown highest amount of total phenolic content (72.23 mgGAE/g) as compared to ethanolic and ethyl acetate extract that contains the 65.32 and 69.82 mgGAE/g phenolic content (Table 5).

Pharmacological Activity

The methanolic, ethanolic and ethyl acetate extracts were used for the anti-inflammatory potential.

Screening for anti-inflammatory Activity

The different extract of Momordica Charantia were administered i.p. against the rat infected by carrageenan for paw edema. The observed Reticence are tabulated in Table 6A & B.

Table 6: The data of screening of anti-inflammatory characteristics

(A) At 10 mg/kg b.w.

Group	Isolated fraction	Dose mg/kg i.p.	Mean Edema Volume	Percent Reticence
First	Normal	Normal saline	0.52±0.02	----------
Second	Ethanolic extract (EMC)	10	0.21±0.04	59.61
Third	Methanolic extract (MMC)	10	0.20±0.03	61.53
Fourth	Ethyl acetate extract (EAMC)	10	0.12±0.03	76.92
Fifth	Indomethacin	2.5 mg	0.09±0.02	82.69

At 20 mg/kg b.w.

Group	Isolated fraction	Dose mg/kg i.p.	Mean Edema Volume	Percent Reticence
First	Normal	Normal saline	0.52±0.02	--------
Second	Ethanolic extract (EMC)	20	0.20±0.04***	61.53
Third	Methanolic extract (MMC)	20	0.15±0.03***	71.15
Fourth	Ethyl acetate extract (EAMC)	20	0.11±0.03	78.84
Fifth	Indomethacin	2.5 mg	0.09±0.02	82.69

Incision wound model

The wound healing potential of Momordica charantia extracts was evaluated using the incision model, with results summarized in Table 7. Data were expressed as the mean breaking strength (tensile strength) of the incision wound area. The study demonstrated that MMC exhibited significantly higher tear resistance in the wound site from day 1 to day 10 (512.62 ± 1.23 g). In comparison, EMC and EAMC also showed notable resistance to fracture across the same period, with values of 489.60 ± 0.58 g and 338.63 ± 1.66 g, respectively. Overall, MMC produced a greater enhancement in tensile strength than the standard formulation, indicating superior wound healing efficacy.

Table 7: Effect of different extract of Momordica Charantia on Incision Wound Model	
Groups (n)	**Tensile strength (g)**
Control-1	218.55 ± 0.35
Standard	510.65 ± 4.60
Ethanolic extract (EMC)	489.60 ± 0.58
Methanolic extract (MMC)	512.62 ± 1.23
Ethyl acetate extract (EAMC)	338.63 ± 1.66

Note: n =6 animals in each group, values are expressed as Mean±SEM

CONCLUSION

In the present study, extracts of Momordica charantia were evaluated in an albino rat model to assess their wound healing and anti-inflammatory potential. The findings revealed that the ethyl acetate extract exhibited the strongest anti-inflammatory activity and significantly improved wound healing outcomes. In addition, the methanol extract demonstrated enhanced wound healing efficacy, suggesting its potential for development into a commercially available formulation. Its therapeutic effects may be further optimized through continued application and refinement.

Compliance with ethical standards: Animal (albino rats) used for the study. IAEC Approval No. VEDIC/CCSEA/IAEC/39, Dated 11/04/2025).

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