Biotechnology Frontiers
An International Journal
OPEN ACCESS
ISSN 3117-5791
Original Research Article
Check for updates

Wound Healing Activity of *Monodoramyristica* Seed Extracts in Wistar rats -II

Ibironke A. Ajayi*[1], Victor I. Olaoye[1], Emmanuel N. Ifedi[1] and Olaoluwa O. Ajayi[2]

[1]*Industrial unit, Chemistry Department, Faculty of Science, University of Ibadan, Ibadan, Nigeria*
[2]*Microbiology Department, Faculty of Science, University of Ibadan, Ibadan, Nigeria*

Corresponding author: **Ibironke A. Ajayi** | E-mail: **frajayi@yahoo.com**

Citation: Ibironke A. Ajayi, Victor I. Olaoye, Emmanuel N. Ifedi and Olaoluwa O. Ajayi (2025). Wound Healing Activity of *Monodoramyristica* Seed Extracts in Wistar rats -II. *Biotechnology Frontiers: An International Journal.* **DOI: https://doi.org/10.51470/BF.2025.5.2.22**

05 August 2025: Received | 06 September 2025: Revised | 8 October 2025: Accepted | 02 November 2025: Available Online

Abstract

Introduction: *This studyexamined the wound-healing potential of 100 % methanolic and ethanolic extracts obtained from Monodoramyristica seeds.*

Materials and Methods: *M. myristica seed extract (MMSE) was obtained by a cold extraction method with methanol and ethanol each as solvents, and afterwards subjected to phytochemical examination, antimicrobial assay and minimum inhibitory concentration determination. Wound-healing capability of the extracts was assessed using twenty-four rats divided into four groups of six animals each. Parameters evaluated included wound surface area (mm^2), percentage wound closure, duration of epithelialisation, and histological analysis of both treated and control wounds. Group 1 and 2 rats received 100 % methanolic and ethanolic extract, respectively, whileGroup 3 animals (control 1) served as the untreated control. Group 4 animals (control 2) were treated with a standard wound-healing ointment. All the experimental animals were provided unrestricted access to food and water throughout the study period.*

Results: *Phytochemical analysis revealed that MMSEcontains tannins, alkaloids, terpenoids, steroids, quinones, anthraquinones and reducing sugars.Both extracts demonstrated antibacterial activity against Gram-positive and Gram-negative bacteria at varying concentrations. The observed antibacterial and antifungal effects of the ethanolic extract suggest the presence of biologically active constituents within the seeds of Monodoramyristica. The fastest wound healing was observed in group 1, with an epithelialisation time of 18.00±1.09, followed by group 2 (epithelialisation time of 19.83±1.47). Group 4 rats had faster wound healing (epithelialisation time of 20.33±1.86) than those of group 3 (epithelialisation time of 24.83±0.41). Histopathological findings showed complete epidermal regeneration with keratin formation in the healed wounds of Groups 1, 2 and 4 rats, whereas,Group 3 exhibited numerous endocrine glands with observed deep pink eosinophilic materials.*

Conclusion: *The findingssupport the traditional medicinal use of M. myristica seeds in wound management. 100 % w/w methanol and ethanol seed extracts of M. myristica healed wounds faster than conventional wound healing ointment, and might therefore find application as a wound healing ointment.Overall, the study provides pharmacological evidence validating the traditional use of M. myristica plants for wound-healing purposes.*

Keywords: *M.myristica,seed extract, antimicrobial activity, wound healing, epithelialisation time, histopathology.*

1.0 Introduction

Man has used diverse portions of plants in the prevention and treatment of many illnesses since the dawn of time [1].The concept of generating medications from plants used in indigenous medical systems is far older. Although there are some direct links between a local and biomedical use, the relationship is much more complex in other cases [2]. According to the Wound Healing Society, wounds are defined as physical damages that disrupt the normal structure of the skin. Chronic wounds, in particular, present a serious challenge for both patients and healthcare providers, as they affect a large population and significantly reduce the quality of life [3].Recent estimates show that approximately six million individuals worldwide suffer from chronic wounds. These wounds occur at a rate of about 4.5 per 1000 people, while acute wounds have been reported to occur at nearly twice that frequency, approximately 10.5 per 1000 individuals[4].Medicinal plants play an important role in both traditional and contemporary medicine.Plant-derived compounds with suspected antimicrobial properties require evaluation using appropriate microbial models in order to validate their activity and establish relevant parameters. Consequently, a vast number of researchers from many regions of the world have investigated the effects of plant extracts on bacteria [5].Despite advances in modern medicine, traditional medicines are still used by over 80 % of the global population to treat various skin ailments [6].In order to support the creation of a proper environment for natural healing, herbal remedies in wound management include cleaning, debridement, and maintaining a moist

atmosphere [6].

*Monodora*is a plant genus belonging to the Annonaceae family and comprises approximatelythirty-five species distributed across tropical Africa. *M.myristica* (Dunal) is one of the most commonly utilised species and is widely used as a spice[7].Itis a perennial plant predominantly found in evergreen forests of West Africa and is commonly known as African nutmeg, calabash nutmeg, and ehuru while some common names in Nigeria include:ariwo, ehiri, and airama [7].The seedspossess a nutmeg-like aroma, and widely incorporated into West African cuisine. Countries such as Liberia, Nigeria, Cameroon, Angola, Uganda, and western Kenya are among the regions where the *M. myristica* tree grows naturally in evergreen woods. This tropical shrub belongs to the flowering plant family and produces a berry of about 20 cm in diameter. It is smooth, green, and spherical when young, which becomes woody upon maturation. The fruit is attached to a long stalk that may reach up to 60 cm in length and contains numerous oblong pale brown seeds, each averaging about 1.5 cm in length, embedded in a white, aromatic pulp. Studies show that nearly all parts of the *M. myristica* tree are commercially significant [8].The wood is durable and commonly used for carpentry, household fittings, and joinery, while the seeds are used in the production of jewellery and other crafts. The seeds, which are encased in a white, sweet-smelling pulp of the sub-spherical fruit, are the most economically valued portions. On average, a single fruit contains between 119 and 122 seeds [7].Prior to storage or consumption, the seeds undergo processing methods such as fermentation, washing, drying, and cracking [8].Numerous studies have documented the therapeutic properties of *M. myristica*. In the parts of Central Africa, the stem bark is traditionally used to manage haemorrhoids, gastrointestinal discomfort, fever, and eye conditions, while the seeds are used to treat headaches and hypertension, also as a condiment and postpartum tonic in Eastern Nigeria [9]. Anti-sickling properties have been discovered in *M. myristica*, and the powdered kernel is sometimes added to soup to relieve constipation and manage passive postpartum uterine bleeding. Additionally, the fruit is also utilized for its diuretic qualities [9].Previous studies have reported the use of 5 %and 10 %methanolic seed extracts of *M. myristica*and *M.tenufolia* in wound-healing of albino rats [10].Building on some of this evidence, this present study investigated the wound activity of 100 % methanolic and ethanolic extracts of *M. myristica* seeds.

2.0 MATERIALS AND METHOD

2.1 Plant material

The seeds of *Monodoramyristica* were obtained from Bodijamarket in Ibadan, Nigeria. The plant material was identified and authenticated at the Herbarium Unit, University of Ibadan, Nigeria.

2.2 Preparation of seed extract

The seed coats were carefully removed, after which the seeds were air-dried and milled into a coarse powder using an electronic blender.

A total of 300g each of the dried powdered sample wasdivided equally into two separateaspirator containers. One litre of methanol was added to the first container, and also one litre of ethanol to the second container. Both mixtures were allowed to standat room temperature for four days with constant agitation to enhance extraction. The resulting mixtures were filtered, and the filtrates were concentrated using a rotatory evaporator at 53°C for methanol and 65°C for ethanol. The concentrated extracts were then designated as *M.myristica* methanolic seed extract (MMMSE)and *M.myristica*ethanolic seed extract (MMESE), respectively.

2.3 Phytochemical screening

Qualitative phytochemical analyses wereconducted to identify the presence of secondary metabolites in both methanolic and ethanolic seed extracts. The compounds screened included alkaloids, steroids, carbohydrates, flavonoids, saponins, glycosides, terpenoids, quinones, and anthraquinones,using standard procedures as described by Balakrishnan *et al.* [11].

2.4 Physical Properties

Some of the properties of the purchased *M. myristica* seed were determined. The weight of some randomly selected seeds was determined with and without the hull. The percentage of hull or husk was then calculated.*M. myristica* seed length with hull and without hull was also measured in millimeters and the average seed length was determined.

Formula: $\frac{Wh-Wd/}{Wh} \times 100$

Where W_h = Weight of seed with hull/husk

W_d = Weight of seed without hull/husk

2.5 Test Microorganisms

The microorganisms used in this study were *Staphylococcus aureus, Escherichia coli*, *Bacillus subtilis*, and *Salmonella typhi.* Pure bacterial and fungal strains were obtained from the Department of Pharmacy, University of Ibadan, Nigeria. The bacterial isolates were cultured on nutrient agar (Oxoid, Hampshire, UK) and incubated at 37°C for 24 hours.

2.5.1 Susceptibility tests

The antibacterial activity of MMMSE and MMESEagainst the test organisms was assessed using the disk diffusion method. Gentamycin served as the positive control. The test microorganisms were grown overnight (24 h at 37 °C) innutrient agar, after which the inoculum was prepared by diluting cultures in Muller-Hinton Broth to achieve approximately 10^6 CFU/ml. Sterile paper disks (6 mm diameter) were impregnated with 10 µl of the extracts at concentrations of 1.00 mg/ml, 0.875mg/ml, 0.75mg/ml, 0.5mg/ml and 0.25mg/ml. The disks were placed on inoculated agar plates (90 mm) and incubated at 37°C for 24 hours. Zones of inhibition were measured and compared with those produced by Gentamycin (10 µg/ml). All experiments were conducted in duplicate.

2.5.2 Minimum Inhibitory Concentrations (MIC) of seed extract

The Minimum Inhibitory Concentration (MIC) is defined as the extract concentration capable of preventing visible microbial growth on agar media or turbidity in broth cultures. In this study, the susceptibility of bacterial and fungal strains to the extracts was evaluated using the microdilution technique to establish MIC values.This procedure was applied extensively to extracts that demonstrated inhibition zones of at least 8mm. The extractswere dissolved in 10 % of ethanol 99 % v/v. The assay was conducted using a 96-well microplate following established protocols. The wells of Column 2–11was filled with 100 µl of Muller Hinton Broth (MHB) and served as solvent control for sterility. The first tubes contained only the pure extracts and were used as sterility controls. Gentamycin, prepared in MHB at concentrations ranging from 1 mg/mL, was used as the standard drug for positive control. The agar plates incubated at 37 °C for 24 h, and MIC was recorded as the lowest concentration at which no visible colony growth was observed. All experiments were carried out in triplicate [12, 13, 14].

2.6 FTIR Analysis

Fourier Transform Infrared (FTIR)spectroscopy was employed as the preferred infrared analytical technique. When the infrared radiation passes through a sample, certain wavelengths are absorbed while others are transmitted. The transmitted radiation generates a signal detected as an infrared spectrum, which represents a molecular "fingerprint" unique to the sample. The effectiveness of FTIR spectroscopy lies in its ability to differentiate chemical structures based on their distinct spectral patterns.

2.7 Experimental Animals

Healthy Wistar rats of both sexes, with body weights ranging between 150 and 250 g, were obtained from Laniba Farm, located near Leyket Gas Station in Ibadan. They were given a conventional unlimited food and water and kept in polypropylene cages under conventional laboratory conditions (25-30 ^{0}C, 35-60 % relative humidity). All experimental procedures adhered to the ethical guidelines approved by the University of Ibadan Ethics Committee on Research in Animals, as well as internationally acceptedstandards for the use and care of laboratory animals.

2.8 Evaluation of Wound Healing Activity

The wound-healing potential of *M.myristica* seed extracts was evaluated using an excision wound model. The animals were randomly divided into four groups, each comprising six rats.Ointments were applied topically once daily to the rats in groups 1-3. Group 1 animals received MMMSE ointment, while Group 2 was treated with ointment of MMESE. Group 3 served as the untreated control, whereasGroup 4 functioned as the positive control and was treated with a standard wound-healing ointment containing5%povidone-iodine USP.

Prior to and during wound induction, the rats were anesthetized with ketamine hydrochloride (100 mg/kg, intraperitoneally) following standard procedures.All the animals were monitored closely for signs of illness, and any diseased animals were excluded from the study [15].

2.8.1 Excision wound model

Excision wounds were created on the animals using the method described byAjayi and Omolere[16].An electrical clipper was employed to shave the dorsal fur of the dorsolateral flank area. After preparing the wound region with 70% alcohol, the skin from the planned shaved area was excised to its full thickness with forceps, a surgical blade, and a scissor to obtain a wound area of roughly 300 mm^2. Excision incisions were made 2.0 cm from the spinal column on both sides of the dorsal thoracic region.The wound was left open, and all of the animals were treated with the prepared ointments. The wound healing was tracked by tracing the wound on the 3rd, 11th, 15th, 19th, and 23rd days after wounding. Percentage wound closure and epithelialisation time, which is anindicator of new epithelial tissue formation over the wound surface, were calculated based on wound closure measurements recorded at regular intervals [17].

2.8.2 Wound healing activity study

The wound-healing effectiveness of the formulated extracts was assessed using parameters such aswound surface area (mm^2), rate of wound contraction, epithelialisation time in the excision wound model, and histological examination of theregenerated tissues.

2.8.3 Rate of wound contraction

Wound contraction was evaluated every four days and expressed as the percentage reduction in size. Wound areas were measuredgraphically to monitor the percentage of wound closure, which reflects the formation of new epithelial tissue. Transparent paper and a marker were used to trace wound margins and measurements were carried out. The percentage wound healing was calculated using the formula prescribed by Ajayi *et al.*[10].

$$\text{Percentage wound contraction} = \frac{Ao - At}{At} x\ 100\ \%$$

Where:
Ao = Initial area of wound on day "0" of the experiment.
At = Area of wound at day "t" of experiment.

The epithelialisation time was determined by the number of days required for complete scar formation without any remaining raw wound surface.

2.9 Histological Study

On the 25th day of the experiment, skin tissue samples were collected from all the five experimental groups of animals for histological evaluation of collagen deposition and tissue regeneration. Skin samples from the treated animals were fixed in 10% buffered formalin, sectioned in 6 m thickness slices, and stained with hematoxylin and eosin. The prepared slides were examined under a light microscope to identifyhistological alterations.

2.10 Statistical analysis

Each experimental group consisted ofsix animals (n=6). Data analysis was performed using one-way analysis of variance (ANOVA) with IBM SPSS statistical software version 20.0. The Duncan T test was applied to determine significant differences among group means, with statistical significance set atP<0.05. The results were expressed as mean values±standard deviations obtained from six duplicated tests.

3.0 Results and Discussion

3.1 Some determined physical properties of *M. myristica*seeds

The mean weight and length of hulled and dehulled seeds are presented in Tables 1 and 2 respectively. Ajayi and Ifedi [18] reports kernel percentage of 89.90 % for *P. longifolia* seeds.

3.2 Phytochemical Analysis

Table 3shows the phytochemical screening results of MMMSE and MMESE. The analysis confirmed the presence of tannins, alkaloids, terpenoids, steroids, quinones, anthraquinones, and reducing sugars, along with other bioactive constituents.Tannins can bind to proteins, form complexes that prevent blood flow to wound area thereby reducing loss of blood, and thus cause early healing [19]. Saponins are known to a play a role in stimulating the activity of fibroblasts, increasing collagen production, and helping to repair damaged tissue structure [20].Lestari*et al.*[21] reports that *Azadirachta indica* leaf extract contains secondary metabolites with potential as therapeutic agents for wounds, in particular, diabetic wounds.

3.3 Antimicrobial activities assay

The differences observed in the antimicrobial activity of *M.myristica* seed extractscan be attributed to variations in theirchemical composition (Table 4). [22] reported that *B. subtilis*exhibited the highest sensitivity to ethanol extracts at concentrations of 50-400 mg/ml, whereas*E. coli* and *Proteus mirabilis*showed greater resistance. In contrast, findings from the present study identified *S. aureus* and *B. subtilis* as the most resistant organisms. Previous studies have shown that the minimum inhibitory concentration for *S. aureus* and *S. typhi* was 40 mg/ml. The antibacterial effect of both MMMSE and MMESE the against *S. aureus* in this study aligns with results reported by[23], who recorded inhibition zones of 9 mm. Overall, the presence of both antibacterial and antifungal activities in MMESEconfirms that the plant contains biologically active compounds with potential for multiple therapeutic applications.

3.3 FTIR Analysis of methanol and ethanol *M. myristica* extract

The FTIRspectra ofmethanol and ethanol extracts of *M. myristica* seeds demonstrated notable similarities (Figures 1a and 1b). Both extracts have C-H bonds, C-O bonds, and they also have the O-H bonds; the O-H absorption is a broad one in both molecules. There are also C-H and C-O absorptions. The ethanol molecule has more peaks than the methanol molecule because of the presence of C-C bonds in it. The peaks below 1500 cm^{-1}also indicate the fingerprint region, as shown in Fig. 1 and 2.O-H shows a broad absorption at about 3300 cm^{-1}, C-O has one at near 1200 cm^{-1}, C-H bonds absorbed at about 3000 $cm^{-1,}$ while the C-C bond is unique to only ethanol.

3.4 Weekly weight gain of rats

Table 5 presents the weekly weight changes observed in both the test and control animalsduring the study period. It was noticed that there was a decline in weight after the wound was inflicted(after week 1). This may be due to loss of appetite for about a week due to pain from the wound incision. The weights increased steadily after some days as wound healing progresseduntil week 4.

3.5 Wound contraction and epithelialisation time

The progressive reduction in wound size among the various experimental over the 23-daytreatment period using*M. myristica* seeds is presented in Table 6 and 7, Figure 2. Results from this study revealed that topical application of *M. myristica* seed extracts significantly enhanced wound healing in rats. This effect may be linked to the extracts' ability to neutralize free radicals, suppress inflammatory pathway mediators, and inhibit bacteria growth at the wound site [24].Group 1, treated withMMMSE, exhibited the fastest wound healing response, achieving nearly complete healing (almost 100 % wound contraction) within 18 days, compared to Group 2, which received MMESE. By the 19thday post-wounding, wound of Group 1 rats was markedly faster than those of the rest of the groups. Group 3 animalsshowed the slowest wound healing rate, with an epithelialisationperiod of 24.83±0.14 days. Animals treated with povidone iodine (Group 4) demonstrated epithelialisation times of 20.33±1.86 days, while Group 2 ones recorded 19.83±1.47 days. Complete epithelialisation across all treated groups was observed between the $18^{th}$and 20th post-wounding days. The untreated group (Group 3) showed the poorest healing outcome, characterized by prolonged epithelialisation time, and delayed wound closure.MMMSE ointment healed the wound of the rats faster (epithelialisation time of 18.00±1.09) than MMESE ointment (epithelialisation time of 19.83±1.47).

3.6 Histopathology Analysis

Histological examination of the skin tissuesobtained from the wound sitesof the rats treated for 21 days with MMMSE and MMESEointments revealed the presence of well-developed granulation tissue extending across most layers of the dermis in the excision wound model. Histological features observed in the healed wounds included reformation of the epidermal skin layer, presence of myofibroblasts and fibroblasts, collagen deposition, angiogenesis and infiltration of inflammatory cells. These regenerative characteristics wereevident in both the treated and control groups across all experimental sets(Table 8, Figure 3) indicating active tissue repair and remodelling.

Table 1: Weight of hulled and dehulled Monodoramyristica seeds(g)

Parameters	W_1	W_2	W_3	W_4	W_5	AW*
Weight with hull W_h	1.23	1.13	0.98	1.18	1.00	1.10
Weight without hull W_d	0.95	0.87	0.75	0.93	0.80	0.86
Weight of shell	0.28	0.26	0.23	0.25	0.20	0.24
% of shell in seed	22.76	23.00	23.47	21.19	20.00	21.82

*AW= Average weight

Table 2: Length of hulled and dehulled Monodoramyristica seeds(mm)

Parameters	L_1	L_2	L_3	L_4	L_5	AL*
Length with hull	18.00	18.00	19.00	21.00	18.00	18.80
Length without hull	11.00	11.00	11.00	11.00	12.0	11.20
Difference in length	7.00	7.00	8.00	10.00	6.0	7.60
% of Difference in length	38.89	38.89	42.10	47.62	33.33	40.43

*AV = Average length

Table 3: Phytoconstituents analysis of M. myristicaseed extracts

Phytochemicals	Methanol	Ethanol
Tannins	+	+
Alkaloids	+	+
Saponin	-	-
Terpenoids	+	+
Steroids	+	+
Glycosides	-	-
Quinones	+	+
Anthocyanin	-	-
Flavonoid	-	-
Anthraquinones	+	+
Reducing sugars	+	+

Table 4: Antimicrobial activity of methanol and ethanol M. myristica seed extracts

1.000	0.875	0.750	0.500	0.250	1.000	0.875	0.750	0.500	0.250
1.35±0.07	0.95±0.07	0.70±0.00	0.85±0.07	1.40±0.14	0.85±0.07	0.80±0.14	0.75±0.07	0.65±0.07	0.70±0.00
1.90±0.14	1.50±0.00	1.40±0.00	1.35±0.07	0.75±0.07	–	–	–	–	–
0.07±0.07	1.00±0.00	0.60±0.28	0.75±0.07	0.70±0.00	0.60±0.00	1.45±0.07	1.35±0.07	1.05±0.07	1.00±0.00
1.65±0.21	0.70±0.00	0.70±0.14	0.40±0.14	0.80±0.14	1.65±0.21	0.95±0.07	1.30±0.00	1.15±0.07	0.85±0.07

Table 5: Weekly weight gain of rats(g)

Groups	Week 0	Week 1	Week 2	Week 3	Week 4
1	222.00±39.31	234.17±36.88	203.5±25,26	215.50±21.39	233.33±16.80
2	244.67±36.68	249.67±35.27	221.33±28.77	233.67±28.61	249.67±18.57
3	231.67±32.82	249.33±32.85	228.50±34.19	246.67±27.35	251.00±27.32
4	219.83±9.86	232.00±5.02	219.50±10.62	239.67±12.97	254.33±8.43

*Controls were used in conjunction with other experiments

Table 6: Wound healing area and period of epithelialisation

Groups	Wound healing area (mm^2)						Epithelialisation time
	Day 0	Day 3	Day 7	Day 11	Day 15	Day 19	
1	300.00±0.00	162.73±53.16	88.18±32.80	17.27±6.38	8.18±2.99	0.91±2.22	18.00±1.09
2	300.00±0.00	154.42±24.29	82.65±33.33	29.98±14.11	13.63±9.59	3.63±2.81	19.83±1.47
3	300.00±0.00	274.39±10.11	207.14±11.60	136.25±7.71	67.22±5.63	14.99±4.13	24.83±0.41
4	300.00±0.00	255.28±13.50	167.13±8.89	82.66±8.02	30.00±9.02	6.13±1.37	20.33±1.86

*Controls were used in conjunction with other experiments

Table 7: Wound healing contraction (%)

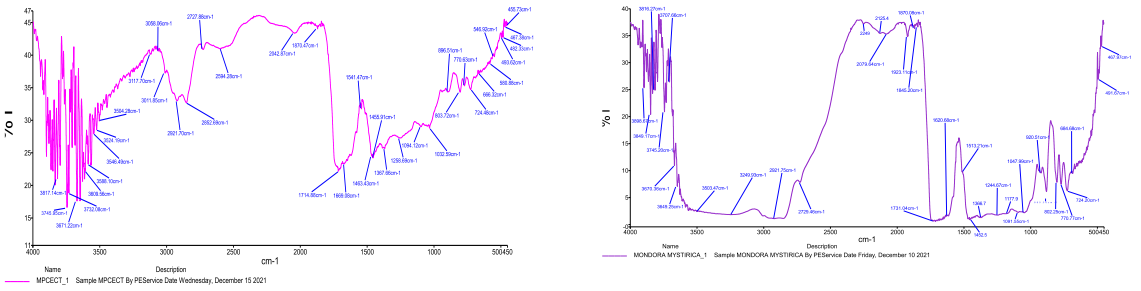
Groups	Day 3	Day 7	Day 11	Day 15	Day 19
1	45.75±17.72	70.61±10.93	94.24±2.13	97.27±0.99	99.69±0.74
2	48.53±8.09	72.45±11.11	90.01±4.70	95.46±3.19	98.78±0.94
3	8.54±3.37	30.96±3.87	54.58±2.35	77.59±1.88	95.00±1.38
4	14.91±4.50	44.29±2.97	72.45±2.67	89.99±3.01	98.64±1.11

*Controls were used in conjunction with other experiments

Table 8: Histopathology of the skin wound area

Groups	Skin layer	Myofibroblasts and Fibroblasts	Collagen deposition	Angiogenesis	Cell infiltration
1	Complete thin layer of epidermis with keratin	Moderate quantity of fibroblasts and myofibroblasts	Moderate densecollagen deposit. Presence of moderate loose collagen insubcutis	Not apparent	Few lymphocytes and macrophages
2	Complete thin layer of epidermis with keratin	Moderate quantity of fibroblasts and myofibroblasts	Moderate dense collagen deposit. Presence of moderate loose collagen in subcutis	Not apparent	Few lymphocytes andmacrophages
3	Numerous sebaceous glands with deep pink eosinophilicmaterials	Abundant myofibroblasts and fibroblasts present	Abundant loose collagen deposition	Numerous sebaceous glands present in the subcutis	Moderate cells include predominantly lymphocytes and a few macrophages and neutrophils
4	Complete epidermal layer with keratin	Numerous fibroblasts and myofibroblasts	Abundant dense collagen in the dermis	Moderate angiogenesis	Moderate predominantly macrophages and lymphocytes

*Controls were used in conjunction with other experiments



Figures 1a and 1b. Spectra of methanol and ethanol extracts of M. myristica seeds

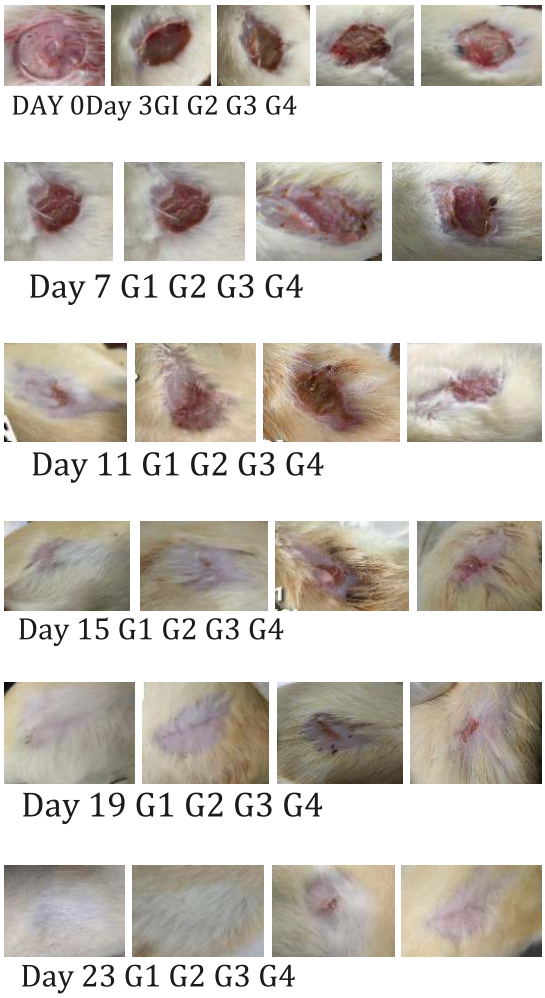


Figure 2: Photograph showing various stages of wound healing of test and control rats

**Controls were used in conjunction with other experiments; G = Group.*

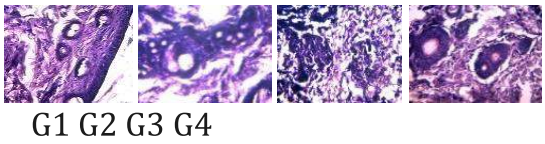


Figure 3: Photomicrograph of rats from test and control groups.

G1: Photomicrograph of group 1 animals treated with MMMSEshowing a locally extensive area of dense collageneous tissue(H&E ×100).
G2: Photomicrograph of group 2 animals treated with MMESE revealing section of dermis with organized collageneous tissue arranged in layer wavy pattern (H&E ×100).
G3: Photomicrograph of group 3 animals that were left untreated with any ointment. There is a severe dispersal/degeneration of the collagen fibres. There are foci of cellular aggregation (H&E ×100).
G4: Photomicrograph of group 4 animals treated with povidone iodine-based ointment. Section shows complete skin with predominantly loose connective tissue in the sub cutis (H&E ×100).
*Controls were used in conjunction with other experiments; G = Group.

4.1 Conclusion

The findings of this study demonstrate that*M. myristica* seed extracts significantly enhancedwound healing in rats.This effect is likely associated with the activity of bioactive compounds present in the extracts. The wound healing impact of *M. myristica* species was also demonstrated by the histological results, as well as the wound contraction rate and duration of epithelialisation. These results suggest that*M. Myristica* seeds possess strong ability for development as a topical wound-healing formulation and many have valuable applications within the pharmaceutical industry.

4.2 Conflict of interest

This study does not have any conflicts of interest.

4.3 Research funding

Research funding was not available for this study.

4.4 Acknowledgement

The Departments of Chemistry, Microbiology and Veterinary Pathology of Faculties of Science and Veterinary, respectively,are acknowledged by the authors for making their facilities available for this research.

REFERENCES

1. Chah, K., Eze, C., Emuelosi, C. & Esimone, C. (2006). Antibacterial and wound healing properties of methanolic extracts of some Nigerian medicinal plants. *Journal of Ethnopharmacology*, 104, 164–167.
2. Heinrich, M. & Gibbons, S. (2001). Ethnopharmacology in drug discovery: an analysis of its role and potential contribution. *Journal of Pharmacy and Pharmacology*, 53, 425–432.
3. Strodtbeck, F. (2001). Physiology of wound healing. *Newborn Infant Nursing Review*, 1, 43–51.
4. Kumar, B., Vijayakumar, M. & Govindarajan, R. (2007). Ethnopharmacological approaches to wound healing exploring medicinal plants of India. *Journal of Ethnopharmacology*, 114, 103–113.
5. Raja, R. & Ramanathan, S. (2009). Studies on wound healing property of coastal medicinal plants. *Journal of Bioscience and Technology*, 1(1), 39–44.
6. Purna, S. & Babu, M. (2000). Collagen based dressings, a review on burns. *Journal of the International Society for Burn Injuries*, 26, 54–62.
7. Burabai, W., Akor, A. & Puyate, Y. (2007). Fracture resistance of African nutmeg (*Monodora myristica*) under compressive loading. *Electronic Journal of Environmental, Agricultural and Food Chemistry*, 6(10), 2434–2442.
8. Nwoso, S., Kasumu, T. & Onyinloye, B. (2015). African nutmeg (*Monodora myristica*) lowers cholesterol and modulates lipid peroxidation in experimentally induced hypercholesterolemic male Wistar rats. *International Journal of Biomedical Science*, 11(2), 86–92.
9. Uwakwe, A. & Nwaoguikpe, R. (2008). In-vitro antisickling effects of *Xylopia aethiopica* and *Monodora myristica*. *Journal of Medicinal Plants Research*, 2, 119–124.

10. Ajayi, I. A., Raji, A. & Umeh, A. (2015). Investigation into the wound healing activity of *Monodora myristica* and *Monodora tenuifolia* seed extracts in albino rats. *American Chemical Science Journal*, 9(4), 1–16.
11. Balakrishnan, N. & Sharma, A. (2013). Preliminary phytochemical and pharmacological activities of *Luffa cylindrica* fruit. *Asian Journal of Pharmaceutical & Clinical Research*, 6, 113–116.
12. Sarker, S., Nahar, L. & Kumarasamy, Y. (2007). Microtitre plate-based antibacterial assay incorporating resazurin as an indicator of cell growth and its application in the in vitro antibacterial screening of phytochemicals. *Journal of Ethnopharmacology*, 42, 321–324.
13. Zander, J., Besier, S., Faetke, S., Saum, S., Muller, V. & Wichelhaus, T. (2010). Antimicrobial activities of trimethoprim/sulfamethoxazole, 5-iodo-2′-deoxyuridine and rifampicin against *Staphylococcus aureus*. *International Journal of Antimicrobial Agents*, 36, 562–565.
14. Marzouk, B., Marzouk, Z., Décor, R., Mhadhebi, L., Fenina, N. & Aouni, M. (2010). Antibacterial and antifungal activities of several populations of Tunisian *Citrullus colocynthis* Schrad., immature fruits and seeds. *Journal of Medical Mycology*, 20, 179–184.
15. Nayak, B., Sandiford, S. & Maxwell, A. (2009). Evaluation of the wound-healing activity of ethanolic extract of *Morinda citrifolia* L. leaf. *Evidence-Based Complementary and Alternative Medicine*, 6, 351–356.
16. Ajayi, I. A. & Omolere, O. B. (2022). Wound healing activity of *Mangifera indica* seed extract in Wistar rats. *Research Journal of Chemical Sciences*, 12(3), 1–10.
17. Sharath, R., Harish, B., Krishna, V., Sathyanarayana, B. & Kumara, H. (2010). Wound healing and protease inhibition activity of bacoside-A isolated from *Bacopa monnieri*. *Phytotherapy Research*, 24, 1217–1222.
18. Ajayi, I. A. & Ifedi, E. N. (2016). Proximate analysis and toxicological studies of *Polyalthia longifolia* seed flour in dietary formulation of albino rats. *American Chemical Sciences Journal*, 18(3), 1–12.
19. Kalalinia, F., Amiri, N., Mehrvarzian, N., Fazly Bazzaz, B. S., Iranshahi, M. & Shahroodi, A. (2020). Topical green tea formulation with anti-hemorrhagic and antibacterial effects. *Iranian Journal of Basic Medical Sciences*, 23(8), 1085–1090.
20. Panche, A. N., Diwan, A. D. & Chandra, S. R. (2016). Flavonoids: an overview. *Journal of Nutritional Science*, 5, e47.
21. Lestari, N. W., Wijianto, D. W. & Wahyuni, A. S. (2025). Potential for diabetic wound healing with hydrogel patch of neem leaf extract (*Azadirachta indica*). *International Journal of Applied Pharmaceutics*, 17(2), 250–258.
22. Darabpour, E., Motamedi, H. & Nejad, M. (2010). Antimicrobial properties of *Teucrium polium* against some clinical pathogens. *Asian Pacific Journal of Tropical Medicine*, 3, 124–127.
23. Sarac, N. & Ugur, A. (2007). Antimicrobial activities and usage in folkloric medicine of some Lamiaceae species growing in Mugla, Turkey. *European Journal of Bioscience*, 4, 28–37.
24. Viddotti, G., Zimmermann, A., Sarragiotto, H. & Nakamura, C. (2006). Antimicrobial and phytochemical studies on *Pedilanthus tithymaloides*. *Journal of Experimental Biology*, 77(1), 43–46.