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FORMULATION AND EVALUATION OF OINTMENT CONTAINING HYDROALCOHOLIC EXTRACT DERIVED FROM THE BARK OF MORINGA OLEIFERA FOR WOUND HEALING ACTIVITY IN RAT MODEL

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ABSTRACT

Background: This study aimed to assess the effectiveness of a hydroalcoholic extract derived from the bark of *Moringa oleifera* in facilitating the healing process of second-degree burns wounds. Moreover, a comprehensive assessment was carried out on standardized *M. oleifera* bark to ascertain its physiochemical characteristics, botanical compound layout, and antioxidant activity, all of which play a crucial role in its capacity to facilitate the healing process of burns. ***Methods:*** For 14 days, the efficacy of ointments containing a hydroalcoholic extract of *M. oleifera* bark at concentrations of 5% and 10% was evaluated for treating second-degree burns in rats. Additionally, histological analysis was conducted on skin tissue samples. ***Results:*** The *M. oleifera* bark extract exhibited TPC (52.56 mg/gm of dried extract) and TFC (84.33 mg/gm of dried extract) value along with antioxidant activity (IC_{50} value of 0.98 µg/ml) for radical scavenging, in the presence of several phytochemicals. The most favorable outcomes were achieved using a 10% ointment composition, demonstrating a wound closure and tissue repair rate of 83.04 ± 0.89%, along with a noteworthy decrease in tissue oxidative stress indicators. Histological investigations have verified the wound-healing properties of *M. oleifera* bark extract. ***Conclusion:*** Due to its significant antioxidant properties and its capacity to create a moist environment for wounds, *M. oleifera* has the potential to serve as a natural treatment for burns. Additional clinical trials are recommended to validate the efficacy of *M. oleifera* bark extract as a therapeutic agent for wound healing.

INTRODUCTION

The healing of wounds occurs naturally as the body repairs damaged tissues. The process occurs in four overlapping stages: hemostasis (blood clotting), proliferation, inflammation, and tissue remodeling. Complex processes are executed by various types of cells, including keratinocytes, fibroblasts, cells that are

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inflammatory, and endothelial cells [1]. Throughout history, people have employed different techniques to address wounds, with modern wound-healing practices emerging in the 20th century. In 2014, the annual expenses for wound care amounted to approximately USD 2.8 billion. It is expected that these costs will increase to USD 3.5 billion by the year 2021. Wound healing is influenced by various factors such as oxygen levels, medical conditions, hormonal changes related to age and gender, anxiety levels, diabetes, obesity, medication usage, alcohol addiction, smoking, and nutritional status. An investigation of the fatality rate of individuals suffering from ulcers caused by diabetes over five years unveiled a comparable rate to that of cancer, underscoring the substantial influence of wounds on the healthcare sector. New therapeutic approaches and technologies are being developed to lessen the strain on healthcare and the economy. Studies are being conducted to assess the effectiveness of traditional wound healing methods like herbs and alternative approaches like leech therapy [2]. An ideal dressing for wounds should be both safe and affordable. Herbal medicine is known for its non-toxic nature, thanks to its extensive history of efficacy and cost-effectiveness. *M. oleifera* is one of the medicinal plants known for its traditional use in promoting wound healing. *M. oleifera* belongs to the Moringaceae family. Ancient Romans, Greeks, Egyptians, and many tropical and subtropical nations in Malaysia, India, Pakistan, Bangladesh, and Afghanistan still used this plant for various purposes [3]. *M. oleifera* has spread across the tropics and is predominantly found in its wild form in northern India. The white flowers emit a delightful fragrance. The fruits have an elongated oval shape, starting off green and maturing to a brown color. In India, leaf paste from *M. oleifera* has been traditionally used for wound healing [4]. In Malaysia, the plant's root is traditionally used for women's health during confinement periods, while the seed oil is applied to the joints to alleviate rheumatism. Other conventional applications include using it as a compress on the abdomen to eliminate intestinal parasites, applying it topically to the breasts to inhibit lactation, ingesting it directly to treat gonorrhea, and combining the leaves with lime to address dropsy. *M .oleifera* is rich in a diverse range of phytochemicals, including vitamins and minerals, carotenoids, isothiocyanates, tannins, saponins, flavonoids, alkaloids, glucosinolates, oxalates, and phytates. These bioactive compounds have positive effects on your health. Research has shown that *M. oleifera* is beneficial in treating different medical conditions because of its antiseptic, antibacterial, antispasmodic, antiulcer, anticancer, anti-hyperthyroidism, hypertensive, and hepato-protective qualities [5]. Earlier reports show that extracts of some of the parts of *M. oleifera* have shown significant wound & burn healing activity, but the effect of its bark on this indication has not yet been studied. Therefore, the bark of *M. oleifera* was chosen for this work. The objective of this study is to evaluate the ability of *M. oleifera* bark to promote wound healing in burn injuries, taking into account its traditional applications and pharmacological features, such as its antiseptic and antibacterial effects. Figure 1 presents the assessment of burn wounds using a hydroalcoholic ointment formulation.

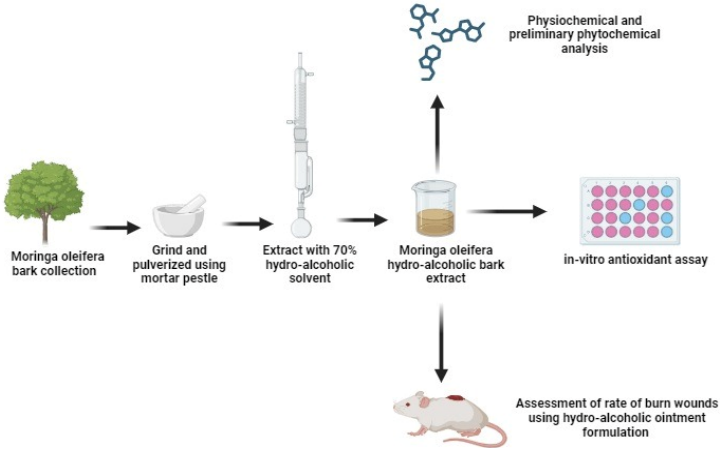

Figure 1: Graphical representation of *Moringa oleifera* for wound healing activity in rat model

MATERIALS AND METHODS

Materials

Ethanol (Chhattisgarh Distilleries Ltd), Methanol (Merck Life Sciences), 2,2-diphenyl-1 picrylhydrazyl (Himalayan Laboratory Ltd), Ascorbic acid (Loba Chime Ltd), Dil. Hydrochloric acid (Loba Chime Ltd), Ethyl acetate (Loba Chime Ltd), Picric acid (Loba Chime Ltd), and Petroleum ether (Loba Chime Ltd). All solvents used in the analysis were purchased from Chhattisgarh Distilleries Ltd, Merck Life Sciences, Himalayan Laboratory Ltd, and Loba Chime Pvt. Ltd.

Collection and Processing of plant material

M. oleifera bark was collected in September 2023 from the Raipur region of Chhattisgarh state. The investigation and experiment were place at the Columbia Institute of Pharmacy, Raipur, Chhattisgarh. *M. oleifera* barks were dried in the air at room temperature until they reached a consistent weight. Barks are dried in the shade and ground into a coarse powder for extraction.

Preparation of *Moringa oleifera* bark extracts

The plant extract was prepared using a composition of 70% alcohol and 30% water because, in this composition, the dissolving power of polar compounds like phenols and flavonoids increases, leaving behind nonpolar compounds that have little biological activity. The dried bark of *M. oleifera* was ground to a coarse powder and extracted using a hydroalcoholic solvent in the Soxhlet apparatus, weighing 100 grams. The extraction process took a total of 72 hours.

Physico-chemical analysis

The extractive values, total ash, water-soluble ash, and acid-insoluble ash were determined on the dried powdered leaves using established protocols [3].

Preliminary phytochemical screening

The preliminary phytochemical screening tests conducted on the bark of *M. Oleifera* tested for saponins, tannins, alkaloids, glycosides, phenols, flavonoids, and steroids [6].

Total Phenolic Content (TPC)

The total phenolic content was quantified using the Folin-Ciocalteu method, as elucidated by Chouhan et al. The UV-spectrophotometer was employed to quantify the absorbance of both standard gallic acid and the samples at a wavelength of 765 nm. The TPC in the samples was measured and expressed as milligrams of gallic acid equivalents (GAE) per gram of dried extract [7].

Total Flavonoid Content (TFC)

The quantitative assessment of the overall flavonoid composition within the unrefined extract was conducted utilizing the aluminum chloride colorimetric technique. The absorbance measurement was conducted at a wavelength of 510 nm, and the outcomes were subsequently conveyed in milligrams per gram of dried extract, namely quercetin equivalents (QUE) [8].

Thin Layer Chromatography (TLC)

Thin layer chromatography (TLC) was employed to identify the constituents present in the extract. For TLC, glass plates were prepared using Silica Gel-GF 254, which acts as an adsorbent. The slurry was made by adding 15gm of silica Gel-G to 30ml of distilled water (1:2). This mixture was triturated in a mortar with a pestle and then poured onto the glass plates. After that, the plates were allowed to air dry for at least one hour, and the layer was fixed by drying it at 110°C for one and a half hours. A micropipette was used to load approximately 10µml of the extract over the prepared plates and air dried. The plates were developed in the solvent system Toluene: ethyl acetate: formic acid (14:10:1). The prepared plates were placed vertically into a saturated TLC chamber. The plates were allowed to develop till the mobile phase moved to about 80% from the line of spotting. Then, the plates were removed and dried. The eluted spots were visualized under a UV chamber at a wavelength of 254 nm [7].

Antioxidant activity

Various extracts' free radical scavenging activity was assessed using a DPPH test. The reduction in DPPH solution absorption after introducing an antioxidant was quantified at 517nm. Ascorbic acid at a concentration of 10 mg/mL in DMSO was utilized as a standard [7].

Preparation of ointment

The ointment was developed using a fusion method with two different concentrations (5% w/w and 10% w/w) by blending the dried hydroalcoholic extract of *M. oleifera* bark into white soft paraffin, hard paraffin, wool fat, and cetostearyl alcohol base. The extract formulations were prepared by melting hard paraffin, cetostearyl alcohol, wool fat, and white soft paraffin at around 75°C in a water bath. The dried hydroalcoholic extract concentrations (5% and 10% w/w) were dissolved in a small amount of distilled water and then added to the mortar. The mixture was stirred continuously until it thickened. The same procedure was followed to prepare blank formulation (without any extract). Next, these formulations were placed in a sterile container designated for applying them to the wounds of the treated rats. The extraction ointment was evaluated by analyzing for organoleptic characteristics (appearance, odor, color, and homogeneity), pH, and spreadability.

Evaluation parameters of Ointment:

All the prepared ointments were characterized by parameters such as appearance, odor, color, homogeneity, pH, and spreadability.

Organoleptic characteristics: All blank formulations (i.e., formulations without active ingredients) and drug-loaded formulations were tested for physical appearance, color, texture, phase separation, and homogeneity. These characteristics were

evaluated by visual observation. Homogeneity and texture were tested by pressing a small quantity of the formulated ointment between the thumb and index finger. The texture and homogeneity of the formulations were evaluated based on their consistency and the existence of coarse particles. Evaluation was also done on the immediate skin feel, which included greasiness, stiffness, and grittiness [9].

pH: 2.5 g of each formulation was combined with 50 ml of water in a beaker. The ointment-filled beaker was heated to between 60 and 70°C in a water bath. The pH of the ointments was determined using a pH meter. Repeating this method three times, the average result was obtained [9].

Spreadability: Spreadability of the formulation was determined by an apparatus suggested previously with some modifications. The device comprises a hardwood block with a fixed glass slide on the block and a pulley at one end. An excess of ointment (3g) was placed on the ground plate. The ointment was sandwiched between this plate and another glass plate with the fixed ground plate dimension and provided with the hook. A one-kilogram weight was placed over the tops of the two plates for five minutes to force out air and create a consistent layer of ointment between them. The extra ointment was removed by scraping off the edges. A 240 g pull was subsequently applied to the top plate. The time needed for the top plate to travel 10 cm was measured with a spring fastened to the hook [10]. A shorter interval indicates better spreadability. Spreadability was calculated using the following formula:

$$S = M \times L/T$$

Where S = Spreadability, M = Weight in the pan (tied to the upper slide), L = Length moved by the glass slide, and T = Time (in seconds) taken to separate the slide from each other.

Experimental Animals

Wistar albino rats weighing 180–200 g were procured from the Animal House facility of the Columbia Institute of Pharmacy, Raipur, Chhattisgarh. Twenty-four rats were controlled and fed a laboratory pellet diet and purified water *ad libitum*. The experimental study was conducted as per the CCSEA guidelines. The experimental protocol was duly approved by the IAEC (Protocol No. CIP/IAEC/2023/210). Animals were assigned into four groups (n=6); group I was taken as control (applied blank formulation); group II as Test group I: applied topically, ointment prepared with hydroalcoholic extract of *M. oleifera* of concentration 5% w/w; group III as test group II: applied topically, ointment prepared with hydroalcoholic extract of *M. oleifera* of concentration 10% w/w; group IV as standard group: the standard group received silver nitrate 0.2% w/w ointment topically.

Induction of Burn wound healing

All of the procedures were performed under anesthesia using 1-3% halothane administered through inhalation. The rat's back (dorsal surface) was shaved using a depilatory cream. The shaved skin and heating device were sanitized with 70% isopropyl alcohol. The skin was allowed to dry and adjust to the ambient temperature at 26°C. The severe second-degree burn injury occurred 24 hours post-shaving. An in-depth second-degree burn wound was created on the rat by applying a heated spatula head to the skin for 10 seconds after being in contact with the flame for up to 5 minutes. The size of the burn wound was about 1.5cm.

Treatment protocol

Different strengths of the prepared ointment containing hydroalcoholic extract of *M. oleifera* and blank and silver nitrate 0.2% w/w ointment were administered topically to the experimental groups of animals daily for 14 days [11].

Assessment of Burn Wound

The progress of wound healing was assessed by calculating the percentage reduction in the initial wound area every 2 days, starting from the third day of treatment, using a digital camera to capture images. The images were analyzed using Image J software to assess the wound area [12]. We determined the wound healing percentage by using the following equation:

$$\%\ \text{wound contraction} = \frac{\text{Initial wound} - \text{Wound size at specific day}}{\text{Initial wound size}} \times 100$$

Histopathological Evaluation

The tissues from the animal's wound were extracted within 14 days. The samples were placed in 10% formalin for one day, then embedded in paraffin, cut into 5µm thick sections, and stained with hematoxylin and eosin for evaluation under light microscopy. They were graded based on epidermal regeneration, granulation tissue formation, fibroblast cell migration, and angiogenesis. A comparison was conducted between the treated and control groups [13].

Statistical analysis

Statistical analysis and graphs were generated using GraphPad Prism 9.0. The results were presented as means ± standard error of the mean (SEM). We conducted multiple comparisons of differences between groups using a one-way ANOVA followed by Dunnett's test. A P-value below <0.05 was deemed statistically significant.

Ethical statement

All animal experiments were approved by the Institutional Animal Ethics Committee (IAEC) at Columbia Institute of Pharmacy, Raipur, Chhattisgarh. Approval no. is CIP/IAEC/2023/210.

RESULTS

Physiochemical and Phytochemical analysis

The physiochemical values provide insight into the quality and standardization of *Moringa oleifera*. Additionally, phytochemical examination reveals the presence of several types of metabolites in the plant material [6]. Table 1 displays the physicochemical, and Table 2 shows a phytochemical estimation of hydroalcoholic extracts derived from the bark of *M. oleifera*.

Assessment quantitative analysis of Phenolic and flavonoid content

In its desiccated state, the hydroalcoholic extract derived from the bark of *M. oleifera* was discovered to possess a concentration of 52.56 mg/g of phenolic compounds and 84.33 mg/g of flavonoids. The results are presented in Table 2.

Table 1: Consolidated table of physiochemical analysis of *Moringa oleifera* bark

Physiochemical analysis			
Ash Value (% w/w)		**Extractive value (% w/w)**	
Total ash	4.5	Ethanol extractive value	6.8
Acid insoluble ash	3.15	Water extractive value	4.8
Water soluble ash	1.65	Methanol extractive value	5.2

Thin layer chromatography (TLC) of *M. oleifera* bark extract

As shown in Figure 2, thin layer chromatography (TLC) analysis of hydroalcoholic extracts from moringa barks was done to evaluate their quality. Under ultraviolet (UV) light with a wavelength of 254 nm, the results show that the solvent system contains phenolic and flavonoid-like antioxidant compounds.

Table 2: Consolidated table of phytochemical analysis of *Moringa oleifera* bark

Phytochemical analysis		
Alkaloids	Dragendorff's Test	Detected
	Wagner's Test	Detected
	Mayer's Test	Detected
	Hager's Test	Detected
Carbohydrates	Fehling's Test	Detected
	Molisch Test	Detected
	Benedict's Test	Detected
Proteins	Biuret Test	Detected
	Xanthoprotein Test	Detected
Tannin	$FeCl_3$ Test	Detected
Flavonoids	Shinoda Test	Detected
Glycosides	Legal's Test	Detected
	Baljet Test	Detected
	Borntrager's Test	Detected
	Keller Kiliani Test	Detected
Steroids	Liebermann Burchard Test	Detected
	Salkowski Test	Detected
	Ninhydrin Test	Detected
Total Phenolic Content (TPC)	52.56 ± 0.76 mg/gm of dried extract of gallic acid equivalent (GAE)	
Total Flavonoid Content (TFC)	84.33 ± 0.44 mg/gm of dried extract of quercetin (QE)	

Solvent system: Toluene: ethyl acetate: formic acid (14:10:1)

Figure 2: TLC of *Moringa oleifera* bark extract

In vitro antioxidant activity

The results showed the highest discoloration activity at 250µg/mL. They indicated significant free radical scavenging

activity regarding the IC50 values of hydroalcoholic *M. oleifera* bark (0.98) and ascorbic acid (0.98). The graph of antioxidant activity is presented in Figure 3.

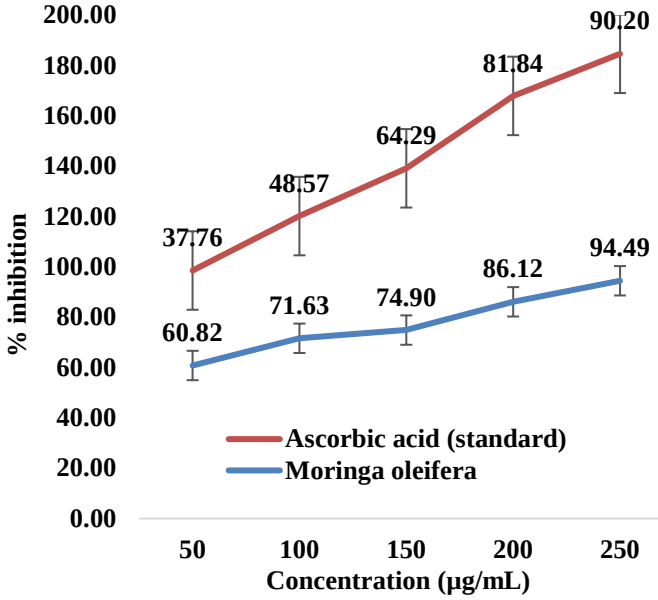

Figure 3: DPPH radical scavenging ascorbic acid standard and hydroalcoholic extracts of *Moringa oleifera* bark

Assessment and examination of the ointment made from the hydroalcoholic extract of *Moringa oleifera*

Several evaluation assessments were conducted to examine the physical characteristics of the *M. oleifera* extract ointment. The ointment formulations were evaluated for physical characteristics, including appearance, color, texture, phase separation, homogeneity, and immediate skin feel. The results indicated that the ointments had a pleasing appearance and a smooth texture and were homogenous with no signs of phase separation. The formulations containing 5% w/w and 10% w/w of plant extract displayed a dark brown color, as indicated in Table 3. All formulations had a pH ranging from 6.5 to 6.9. All formulations have a pH within the normal range of the skin, as shown in Table 3. After determining the spreadability of all formulations, it was noted that formulation 10% w/w exhibited superior spreadability compared to the other formulations, as indicated in Table 3.

In vivo assessment of burn wound healing activity

The images of burn wounds from various treatment groups captured on days 3, 5, 7, 11, and 14 are displayed in Figure 4. Rapid healing of burn wounds was noted in the groups that received treatment compared to those that did not. Measuring the rate of wound contraction is essential for assessing the advancement of wound healing. Research indicates that the burn wound showed 35% and 54% healing within seven days when treated with 5% and 10% hydroalcoholic ointment, respectively. During the same period, the wound treated with silver nitrate showed a healing rate of 63%, while the control group only had a 19% healing rate. Observations show that the wound in the test groups was nearly 75–80% healed by day 11, compared to 87% and 55% healing in the standard and control groups, respectively. In the test groups that used a 10% ointment formulation of *M. oleifera* bark extract, complete healing was observed by the 14th day, while the control groups did not show healing by the end of the protocol. The test groups exhibited noteworthy activity in comparison to both the control group ($p<0.001$) and standard treatment up to the 14th day ($p<0.001$). Experimental groups using 10% hydroalcoholic ointment from *M. oleifera* bark exhibited similar efficacy throughout the study. The empirical investigation revealed that rats were subjected to a hydroalcoholic ointment derived from the bark of *M. oleifera* exhibited noteworthy wound healing efficacy compared to both the standard and control cohorts within burn models. Table 4 displays the rates of wound contraction, and the percentage of wound contraction on different days after burn wound induction has been presented in Figure 5.

Table 3: Evaluation parameters of ointment formulation

Physical evaluation	Formulation containing extract		
	Blank	5% w/w	10% w/w
Organoleptic characteristics			
Physical appearance	Good	Good	Good
Color	Whitish	Dark brown	Dark brown
Texture	Smooth	Smooth	Smooth
Phase separation	No	No	No
Homogeneity	Homogenous	Homogenous	Homogenous
pH	6.80	6.55	6.60
Spreadability	21.05	22.24	22.34

Histopathological studies

The histopathological study showed differences in the healing stages between the control and treated groups. The process of wound healing is complex and consists of three distinct yet interconnected stages: the inflammatory stage, the repair or proliferative stage, and the remodeling stage [13]. Burns can significantly impede the body's healing ability by affecting

angiogenesis, collagen re-organization, and granulation tissue formation. Furthermore, it results in damage from free radicals, which hinders the damaged tissue's healing process. The control group did not show any regrowth of the outer layer of skin (epidermis) in the center of the wound, and white blood cells with a multi-lobed nucleus (polymorphonuclears, or PMNs) were visible at the wound site. The group that received silver nitrate showed the growth of epithelium at the edges of the lesion and the irregular build-up of collagen bundles [14]. There was no new epithelium formation at the wound center when exposed to a 5% extract of *Moringa oleifera* bark. Moreover, an unusual build-up of collagen bundles was observed. In the study, some rats that received *M. oleifera* bark extract showed a deficiency in PMNs and a distinct organization of collagen fibers. The stratum keratinosome did not regenerate, but different layers of skin tissue developed, and cells in the basal layer of epithelium grew almost to the typical level [20]. The study demonstrated that animals treated with a 10% extract of *M. oleifera* bark exhibited enhanced wound healing activity, as indicated by the development of new epidermis and dermis compared to the control groups. Examining tissue samples from the wound area on the 14th day using 100x magnification revealed the images in Figure 6.

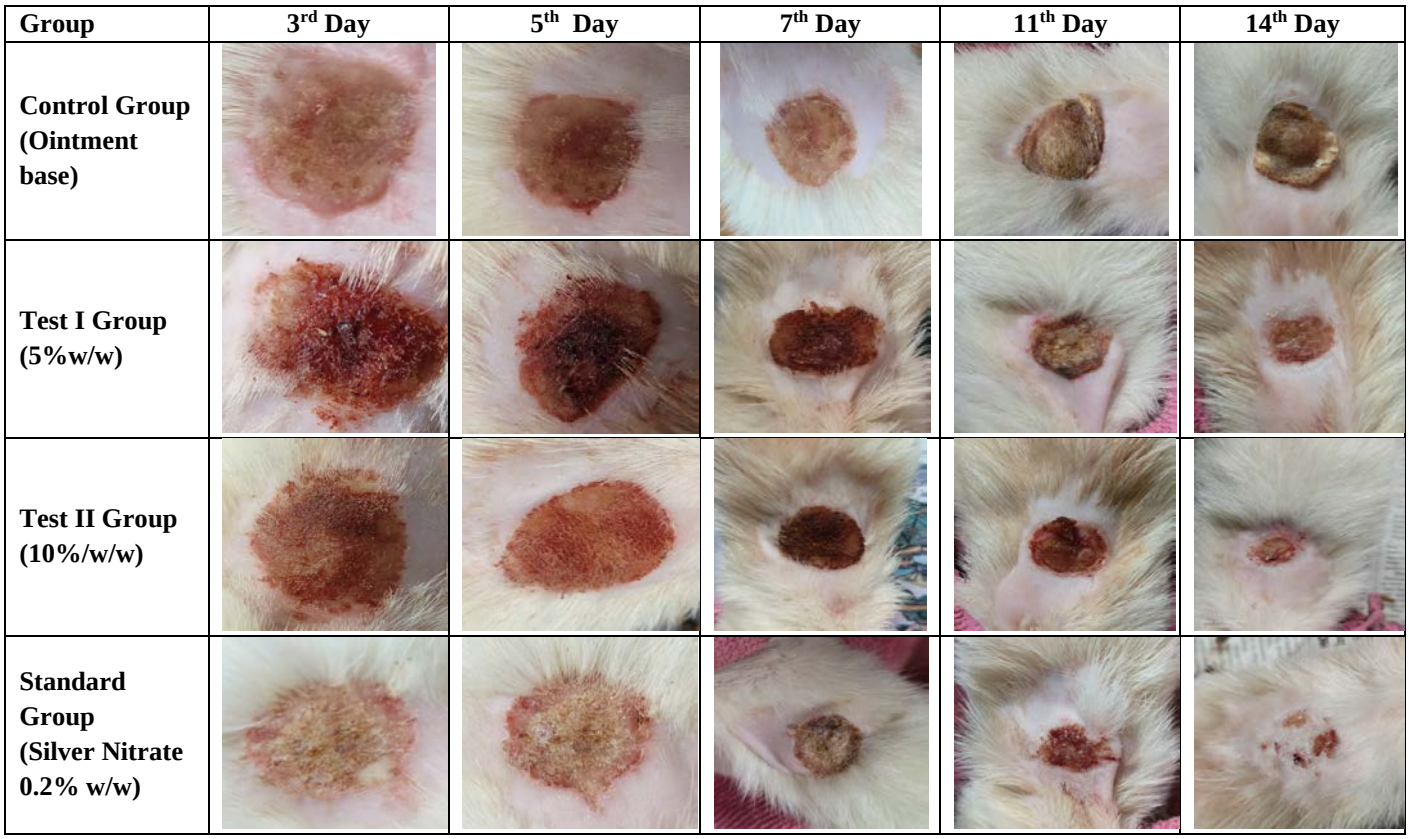

Figure 4: Photographic representation of Progressive improvement of burn wounds in rats treated with *Moringa oleifera* bark extract (5%w/w and 10 % w/w) and standard drug (Silver nitrate 0.2%)

Table 4: Percentage contraction of wound healing of Hydroalcoholic extract of *Moringa oleifera* bark

Animal experimental group	Percentage of wound contraction				
	3rd Day	5th Day	7th Day	11th Day	14th Day
Control Group (Ointment base)	1.66 ± 0.89	9.43 ± 0.89	19.40 ± 0.89	30.36 ± 0.89	55.61 ± 0.89
Test 1 (5% w/w)	3.33 ± 0.89	24.5 ± 0.89	35.73 ± 0.89	55.73 ± 0.89	73.79 ± 0.89
Test 2 (10% w/w)	4.66 ± 0.89	30.08 ± 0.89	54.83 ± 0.89	64.84 ± 0.89	83.04 ± 0.89
Standard (Silver nitrate 0.2 % w/w)	6.66 ± 0.89	35.73 ± 0.89	63.8 ± 0.89	71.37 ± 0.89	87.66 ± 0.89

Values are expressed as mean ± standard deviation (No of Animals, N=6) for each group. Significant differences ($P < 0.05$)

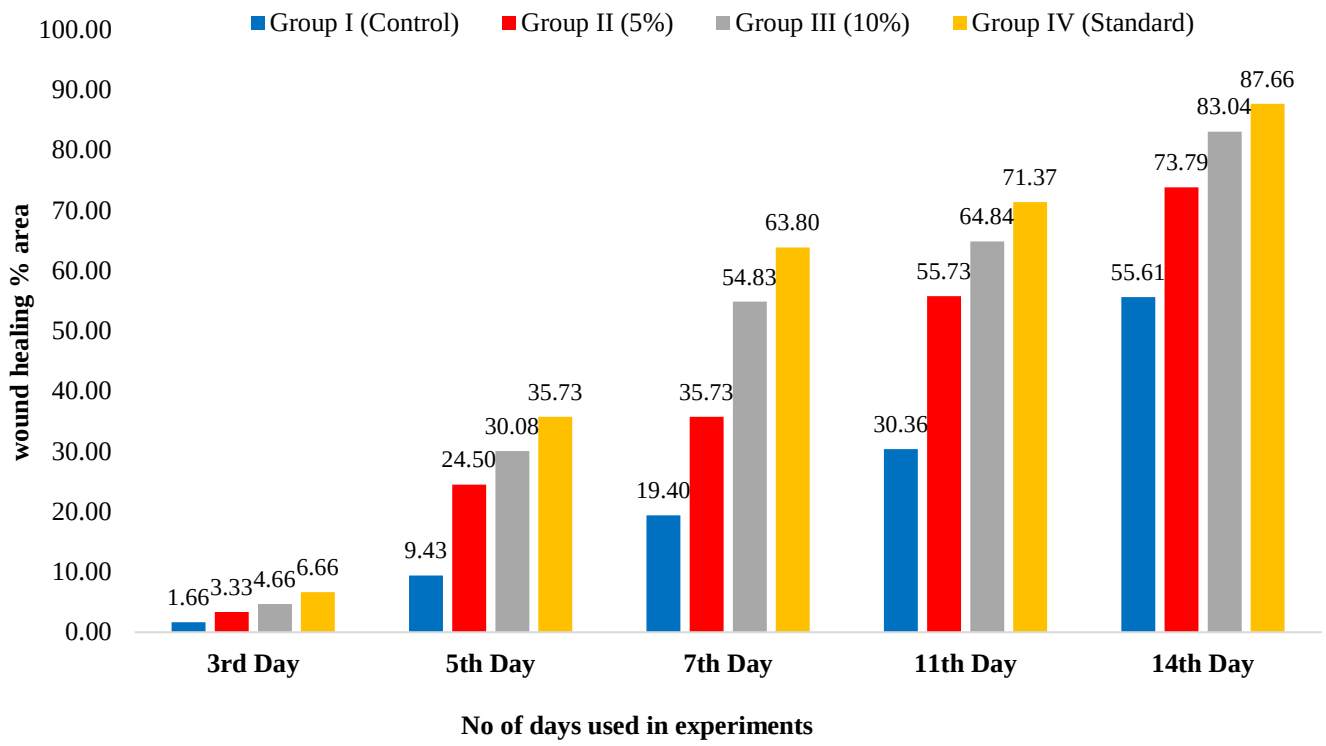

Figure 5: Graphic representation of the %burn wound contraction on different days after burn wound induction

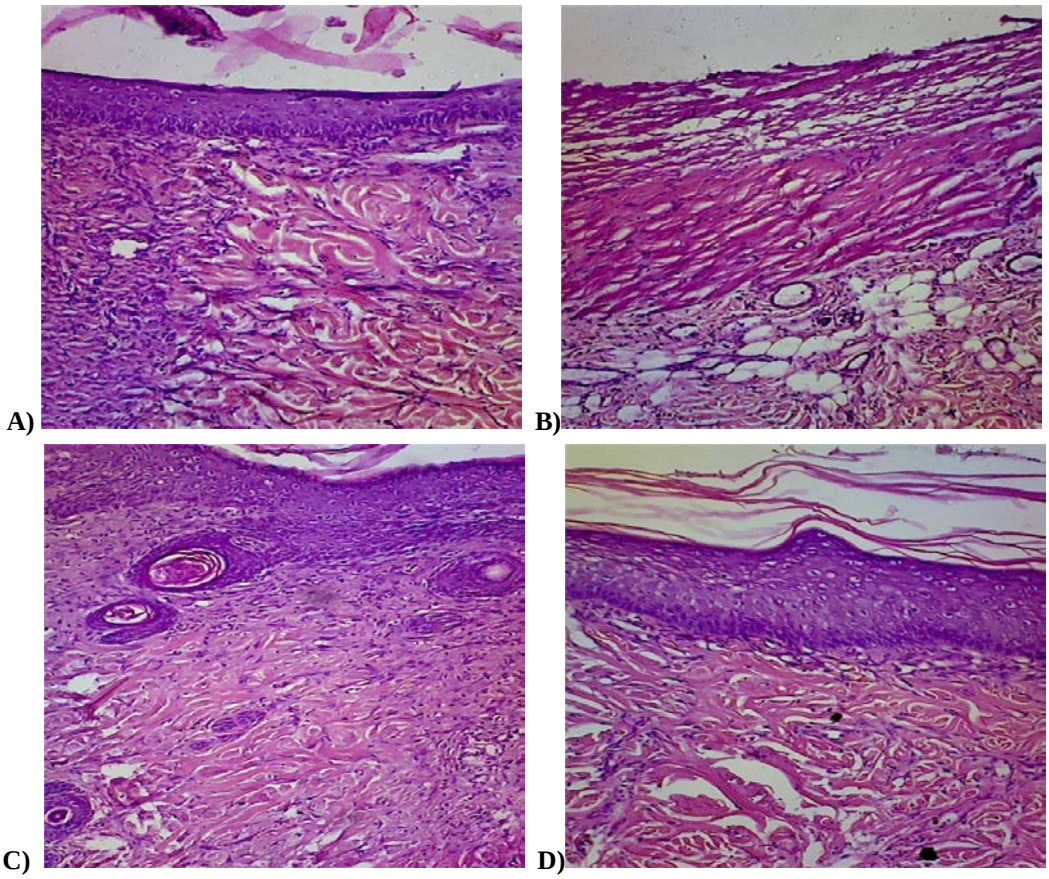

Figure 6: Microscopic images of skin tissue samples of day 14 with a magnification of ×100. (a) Group -I (Control) (b) Test group- I (5%) (c) Test group- II (10%) (d) Group-IV (Standard)

DISCUSSION

This study aimed to investigate the potential wound-healing properties of a hydroalcoholic hydrogel derived from *M. oleifera* bark, building upon previous research findings. Research on *M. oleifera* has identified flavonoids, tannins, saponins, polysaccharides, and polyphenolic compounds in its phytochemical composition [15]. The hydroalcoholic ointment from *M. oleifera* bark exhibits wound-healing potential due to the plant's antioxidant, antibacterial, and wound-healing properties [4]. The hydroalcoholic ointment derived from the bark of *M. oleifera* exhibits promising wound-healing capabilities attributed to the plant's antioxidant, antimicrobial, and wound-healing qualities. Natural antioxidants boost the body's antioxidant system by removing free radicals [16]. There is a notable preference for acquiring antioxidants from natural sources rather than synthetic sources [15]. The *M. oleifera* plant reduces disease risk and severity through its phenolic compounds, which possess antioxidant properties. The hydroalcoholic extract contains a higher level of flavonoids, contributing to the plant's antioxidant properties [17]. Flavonoids and tannins are the main phytochemicals present in plant extracts. Furthermore, hydroalcoholic extracts of moringa with antioxidant properties were formulated. The IC_{50} value of the extract surpassed the IC_{50} value documented in the current study. The hydroalcoholic extract of *M. oleifera* bark shows promise for treating wound infections. In a wound burn model, animals treated with a hydroalcoholic ointment containing *M. oleifera* bark showed notable enhancements in wound healing compared to the standard and control groups. Upon histological examination, the treated instances showed increased cell infiltration in the wound region through staining. The ointment derived from *M. oleifera* bark likely exhibited a heightened chemotactic effect, attracting more inflammatory cells to the wound location [18][19]. The hydroalcoholic ointment from the bark of *M. oleifera* may promote cell division through its mitogenic activity, which is essential for wound healing. The histological observations supported the experimental wound healing research, particularly regarding wound pace and contraction strength. It promotes wound healing by creating an optimal environment on the surface, which encourages the formation of granulation tissue and speeds up the healing process. Infections significantly impact the health and mortality of wound individuals [20]. This ointment aids in infection prevention by lowering the risk of sepsis development and minimizing the duration of the inflammation phase. The lack of this feature in synthetic wound treatment drugs is notable, especially for individuals prone to scar formation, given its cosmetic consequences. Furthermore, the hydroalcoholic ointment from the bark of *M. oleifera* did not cause any adverse effects. Hence, it is reasonable to propose using it for wound healing.

CONCLUSION

Based on the findings, the hydroalcoholic extracts of *M. oleifera* bark show promise as natural remedies for wound healing when formulated as an ointment. Exploring the separation and identification of critical components and how this plant extract equipment impacts different types of microbes could enhance our knowledge of treating infections during wound healing.

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FINANCIAL ASSISTANCE

NIL

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTION

All authors contributed equally. Himanshu Sahu collected, distributed, and organized the data sets and prepared the first draft of the manuscript. Shashikant Chandrakar, Poonam Sahu, and Akhilesh Sahu developed the concept. Trilochan Satapathy and Pushpa Prasad Gupta revised the final manuscript. All the authors approved the final version of the manuscript.

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