



# Therapeutic Efficacy of Topical Sesame and Garlic Ointment Extract on Second-Degree Burn Wounds in Rabbits: A Comparative Histopathological Study

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## Abstract

**Background:** The public health implications of burn injuries are substantial, and conventional burn treatments can result in prolonged healing time and negative side effects. Recently, there has been increased interest in using natural products and their active ingredients for use as alternative therapies for wound care. The purpose of this investigation was to determine the use of a new topical ointment composed of sesame oil and garlic extracts for treating second-degree burn injuries. **Objective:** The effects of sesame/garlic ointment extract on wound healing will be tested by measuring the area of the wound, the time required for the epithelial layer to heal, and the effects of the ointment on the appearance of histopathological changes in the tissue of the rabbits.

**Methods:** A total of fifty New Zealand rabbits were randomly separated into two groups (n=25) as follows: Group A (the control group; received standard petroleum jelly) and Group B (the treatment group; received sesame and garlic ointment). Second-degree burn injuries were inflicted to the dorsum (back) of each rabbit. The sesame and garlic ointment was created using cold-pressed sesame oil containing sesamol and an alcoholic garlic extract (15% concentration containing allicin) blended with a base of petroleum jelly. The ointment was applied to the wounds in both groups twice daily for 14 days, and measurements of the area of each wound were recorded on days 0, 7, and 14. Tissue was also collected from the rabbits on days 7 and 14 for histopathological evaluations.

**Results:** In the current study, the treatment of second-degree burns with sesame and garlic ointment showed that treatment with ointment was statistically better at healing second-degree burns compared to the control group. On day 7, the treated group exhibited good levels of angiogenesis (the production of new blood vessels), abundant amounts of thickly deposited collagen fibers, and an infiltration of macrophages into the wound site. By day 14, there were signs of nearly complete epithelialization of the wound in the treated group, and there were continued signs of both good levels of angiogenesis and an increase in the number of fibroblasts. On the other hand, the delayed healing response, combined with the incomplete epithelialization, disorganized arrangement of collagen fibers, and large amounts of inflammatory infiltrates on the 14th day, was observed in the control group.

**Conclusion:** The application of the sesame and garlic ointment extract produced marked wound healing effects. In summary, the sesame and garlic ointment extract promotes faster epithelialization, improved angiogenesis, and enhanced tissue remodeling. Additionally, these findings suggest that this product may provide an effective natural therapy for the management of burn wounds, and that further clinical studies should be conducted to confirm these observations.

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**Keywords:** Burns, Wound healing, Sesame oil, Garlic extract

## Introduction

Burn wounds are one of the most critical types of trauma, with millions of people affected worldwide each year and significant burdens placed on families and healthcare systems (Peck, 2011). According to the World Health Organization (2018), an estimated 180,000 people die from burn injuries each year, most of which occur in low- and middle-income countries. Because of the complexity of the pathophysiology associated with burn injuries—extensive tissue destruction, acute inflammatory response, potential for infection, and difficulty healing—therapy must address multiple areas involved in healing the wound or the body (Jeschke et al., 2020). Traditional management of the burn wound includes debridement, topical antimicrobial agents (e.g., silver sulfadiazine), and other synthetic or dry dressings (Greenhalgh, 2019).

Despite some success with these approaches, they are associated with limitations. Topical synthetic antimicrobials have the potential to elicit allergic reactions, prevent epithelialization, and contribute to antimicrobial resistance (Atiyeh et al., 2007). Additionally, current large-scale treatments do not sufficiently address all phases of wound healing, including inflammation, angiogenesis, collagen synthesis, and tissue remodeling (Rowan et al., 2015). High costs for systemic treatments limit access to advanced wound care in areas with limited resources. The challenges associated with current therapeutic approaches have led to increased interest in finding new therapies, particularly natural products, that have been shown to be safe with various biological effects (Pereira & Bartolo, 2016).

For centuries, many cultures have used natural herbs and plants to assist in the healing process, and medical research has begun to validate some of the traditional uses of these plants through biochemical identification of bioactive compounds and the explanation of how these compounds work (Biswas & Mukherjee, 2003; Kumar et al., 2007). Natural plants have multiple advantages over synthetic products, including multiple targets, good safety profiles, low likelihood of resistance, and cost-effectiveness (Yuan et al., 2016). Garlic and sesame have emerged as two of the most promising medicinal plants within the

large universe of medicinal plants for possible use in dermatology because of their well-established antimicrobial, anti-inflammatory, and antioxidant activities (Kapoor & Huang, 2006; Ankri & Mirelman, 1999).

Sesame (*Sesamum indicum* L.) is considered one of the world's oldest oilseed crops, cultivated for over 5,000 years, and has been used both for food and therapeutically. The oil from sesame is high in polyunsaturated fatty acids—specifically linoleic and oleic acids—that are important for maintaining the skin's barrier and reducing transepidermal water loss (Kamal-Eldin & Appelqvist, 1996). Additionally, sesame contains a unique group of lignans—sesamin, sesamol, and sesamol—that possess strong antioxidant activity (Namiki, 2007). Sesamol, in particular, has a very strong ability to scavenge free radicals and protect against oxidative stress-induced cellular damage (Joshi et al., 2005). In terms of wound healing, sesame oil has demonstrated effective antimicrobial activity against a number of pathogens associated with wounds, possesses anti-inflammatory activity by modulating cytokine production, and is able to encourage fibroblast proliferation and collagen synthesis (Hsu & Liu, 2004; Kumar et al., 2011).

Garlic (*Allium sativum*) has been used for medicinal purposes as far back as Egyptian, Greek, Roman, and Chinese medical traditions. Over 200 bioactive compounds have been identified in garlic by modern science, but the main active compounds that produce the therapeutic benefits of garlic come from organosulfur compounds, particularly allicin (Borlinghaus et al., 2014). Allicin is produced as a result of the enzymatic conversion of alliin—an amino acid found in garlic that converts into allicin after garlic is crushed or handled; during this process, enzymatic activity creates various sulfur-containing compounds that provide many different types of biological activities (Block, 1985). Garlic possesses a very effective wide range of antimicrobial activities toward Gram-positive bacteria, Gram-negative bacteria, fungi, viruses, and parasites (Bayan et al., 2014). Garlic extracts also work against many antibiotic-resistant organisms (such as methicillin-resistant *Staphylococcus aureus*), which are often involved in the development of burn wound infections (Cutler & Wilson, 2004).

Garlic's anti-inflammatory effects are well studied, with allicin and other associated compounds inhibiting pro-inflammatory mediators including nuclear factor kappa B (NF- $\kappa$ B), cyclooxygenase-2 (COX-2), and a variety of inflammatory cytokines (Arreola et al., 2015). The inflammatory response plays an important role in the repair of burn wounds since excessive or prolonged inflammatory response can hinder healing, potentially resulting in hypertrophic scarring (Martin & Leibovich, 2005). Garlic demonstrates antioxidant effects by protecting cells from oxidative damage through enhancing the activity of endogenous antioxidants and directly scavenging reactive oxygen species (ROS). Recent studies support the idea that garlic may improve angiogenesis and stimulate fibroblast growth, both necessary steps during wound healing (Xie et al., 2017; Shedoeva et al., 2019).

The rationale for combining sesame oil and garlic extracts into a topical preparation is based on the synergy between their mechanisms of action. Sesame oil serves as a lipophilic delivery system to increase the rate of penetration through skin, maintain moisture balance, and provide an antioxidant component via sesamol, while garlic extract provides strong antimicrobial and anti-inflammatory activities via allicin (Abdullahi et al., 2014). Addressing the multiple aspects of burn wound pathophysiology—namely

prevention of infection, control of inflammation, reduction of oxidative stress, and stimulation of tissue growth—may be possible with a multi-targeting combination of sesame and garlic. There is a limited amount of research that has been conducted on the combined effects of these two natural products in a standardized ointment formulation for burn treatment (Dorsett-Martin, 2004).

Animal models provide an essential means of investigating how burn wounds heal and how therapeutic modalities may be applied to the healing process. One animal model that has been used extensively in dermatological research is the rabbit (Sullivan et al., 2001). Advantages associated with the rabbit model include the appropriate size of the animal to allow for standardization of burn wound models, similarity between rabbit and human skin (i.e., skin color, texture, thickness), cost-effectiveness compared to larger animals, and established protocols for both the induction and evaluation of burns. The rabbit model has been used extensively to validate the effectiveness of new creams or other topical agents and provides a predictable pattern of healing, allowing for systematic investigation of treatment effects (Enoch & Leaper, 2008).

Histopathological evaluation has historically been the gold standard for assessing wound healing progress. Histopathology provides detailed information on cellular events at the wound site, tissue architecture, inflammation, angiogenesis, collagen deposition, and epithelialization (Gurtner et al., 2008). While macroscopic measurements may be limited to surface evaluation, histopathology allows complete evaluation of deep tissue changes and determination of specific cellular mechanisms that produce therapeutic effects (Reinke & Sorg, 2012). Important histologic parameters in the evaluation of burn wounds include epithelial regeneration, granulation tissue formation, inflammatory cell infiltration, neovascularization, organization of collagen fibers, and fibroblast activity. These histological parameters reflect various cellular and molecular interactions that comprise wound healing endpoints (Singer & Clark, 1999).

There is limited reliable scientific research supporting the therapeutic use of topically applied sesame and garlic in combination for burn wound healing due to limitations in previous studies, including failure to optimize formulations, ensure stability of active components, and assess depth of tissue penetration. Additionally, a comprehensive evaluation of the sesame and garlic combination compared to standard care has not been conducted in validated animal models, which is needed before clinical development of potentially successful natural products for burn management can proceed. Thus, the present study sought to explicitly examine the therapeutic potential of a standardized sesame and garlic ointment on the healing of burn wounds in rabbits by performing systematic histopathological assessment to clarify the mechanisms associated with any observed positive effects. The results may provide valuable information regarding the scientific basis for developing novel, safe, and effective natural product-based treatments for burn care.

## **Materials and Methods**

### **Study Design and Experimental Animals**

Fifty healthy adult New Zealand White rabbits (*Oryctolagus cuniculus*) aged 6 to 8 months and weighing between 2.5 kg and 3.0kg were received from a certified rabbit breeder facility. The animals

were placed in an environment for one week to become adjusted to their new surroundings before beginning the experiment. Following acclimatization, the rabbits were randomly divided into two groups based on a computer-generated randomization scheme. Group A was the control group and consisted of 25 rabbits ( $n = 25$ ). The rabbits in Group A were treated with a topical application of an inert petroleum jelly base that contained no active ingredients. Group B was comprised of 25 rabbits ( $n = 25$ ) and was treated with an ointment formulation of sesame and garlic.

The sample size for each of the two groups was calculated using standard equations for comparing the results between groups, assuming a 30% difference between the healing rates of the two groups, and based on an alpha level of 0.05 and a power of 80%.

### **Plant Materials and Extract Preparation**

Fresh sesame seeds (*Sesamum indicum*) and garlic bulbs (*Allium sativum*) were obtained from reputable sources and identified by a trained botanist. A voucher sample was created to provide a permanent record of the identification and will be used for future reference in the institutional herbarium.

The preparation of sesame oil was done through cold-press extraction. The sesame seeds were cleaned and allowed to air-dry. Cold-pressing of the cleaned, dried sesame seeds was accomplished using hydraulic presses. The extraction temperature never exceeded 40°C to preserve the bioactive substances, including the thermolabile compounds, such as sesamol. After extraction, the sesame oil was filtered through Whatman filter paper and additionally through Soxhlet extraction using petroleum ether to remove residual impurities. The sesame oil was packaged in dark amber glass bottles and stored at 4°C until time of testing. The amount of sesamol present in sesame oil was confirmed through high-performance liquid chromatography (HPLC). The extraction of garlic was done using a standardized alcohol extraction process. Fresh garlic cloves were peeled, washed thoroughly with distilled water, and sliced into sections approximately 1/4 inch thick (roughly 8 mm). The sliced garlic was dried in a hot air oven at 40 to 45 degrees Celsius (from four to eight weeks). The garlic was then reduced to a fine powder using a grinder and passed through a 40-mesh sieve for particle uniformity. The Soxhlet extraction was performed using 15 g of the garlic powder and 100 ml of absolute ethanol in the Soxhlet apparatus, which was heated at 78 degrees Celsius (boiling point of ethanol) for 6 to 8 hours (4-5 extraction cycles). After Soxhlet extraction, the extraction solvent was evaporated using a rotary evaporator (40 degrees Celsius) under reduced pressure, yielding a 15% concentrated garlic extract. The allicin in the extract was verified and the extracts were standardized to ensure consistency among batches; therefore, the extractors are hermetically sealed in dark, sterile containers and stored at 4 degrees Celsius.

### **Ointment Formulation:**

The ointment base was formulated using pharmaceutical-grade ingredients to provide stability and optimal delivery of the active ingredients. The base formulation consisted of 70 grams of white soft paraffin (petroleum jelly), 10 grams of anhydrous lanolin, and 5 grams of liquid paraffin. The composition of the base ingredients was accurately weighed and combined in a water bath at 40-45 degrees Celsius, providing a homogeneous molten mixture while avoiding degradation of the heat-labile components. The ointment formulation was created with the use of 15 g of standardized garlic extract and subsequently blended using sesame oil during the process of mixing. This was done at a lower speed while being agitated continuously (by use of a glass stirring rod or magnetic stirrer) to allow the

garlic extract and sesame oil to evenly distribute throughout the melted ointment base without entrapping any air thus significantly facilitating uniformity and smoothness of the ointment mixture. Following the formation of a single-massed, smooth ointment base, it was then allowed to cool down to room temperature while still being intermittently stirred. The finished ointment should have been stored in an opaque, sterilized container at 4 °C. The results of all stability tests on the finished product showed no loss in the concentration of the active ingredients in the ointment during the course of the study. The base ointment alone (without active garlic extract and sesame oil) was defined as the control and used as a comparative measure against the active ointments containing the garlic extract.

### **Burn Wound Induction**

Wound burns were performed on each rabbit following national and institutional standards for animal experimentation using ketamine hydrochloride (35 mg/kg) and xylazine (5 mg/kg) delivered by intramuscular injection. The area on the back of the rabbits (dorsal surface) where the burns would be performed was shaved using an electric clipper, cleaned using both 70 percent ethanol and a 5 percent solution of povidone-iodine. A standard second-degree burn was induced by holding a hot metal rod at 100 °C for a period of 10 seconds to each burned site. This was done to ensure a consistent level of burns in that all burns would be of a partial thickness nature affecting both the epidermis and upper layer of the dermis. The burn area measurements were taken immediately after the induction of the burn using electronic calipers, recorded, and photographed.

### **Treatment Protocol**

Treatment of the burn wounds with the respective active ointments was initiated 24 hours after the procedure. Prior to each use of the ointment, all burn wound sites had been cleaned gently using sterile saline solution and allowed to dry. A sterile spatula was used for topical application of approximately half a gram of the assigned ointment (treatment or control) to fully cover the surface of the burn wound. To prevent contamination and mechanical trauma, the burn wound was then covered using a sterile gauze dressing. Both treatments were administered in the morning and evening, with a 12-hour interval between treatments, for a total of 14 consecutive days. Daily monitoring of the study animals included assessing for any signs of infection, distress or adverse reactions resulting from the treatments. The treatments selected for the study were based on preliminary studies, as well as the recommendations found in the literature, for producing optimal results in wound healing.

### **Assessment of Treatment Efficacy**

Several parameters were used to evaluate treatment efficacy for wound healing: measurement of the wound area at day 0 (baseline), day 7, and day 14 using digital planimetry software; the number of days to complete epithelialization (indicated by the days needed to close the wound with an epithelial covering); and histopathologic assessment of the specimens collected at days 7 and 14. The percentage of wound contraction was calculated using the following formula:  $[(\text{Initial Area} - \text{Current Area}) \div \text{Initial Area}] \times 100$ .

### **Histopathological Assessment**

For histopathological evaluation, tissue specimens were collected from the burn wounds on days 7 and

14. Five of the experimental animals from each treatment group were humanely euthanized at each time point through overdose of anesthetic agents. The specimens were composed of full-thickness skin biopsies, including the entire burn wound area as well as a 0.5 cm border of normal surrounding tissue, collected using sterile techniques. The specimens were fixed in 10% neutral buffered formalin to allow for preservation and to minimize tissue decomposition due to the processes occurring after death. Early on in the fixing process, it became apparent that depending on the method used, not all structures would be preserved to the same extent, therefore, when performing preservation through formalin fixing, the tissues were subject to different procedures of dehydrating through an increasing series of alcohol (i.e., 70%, 80%, 90%, 95% alcohol) followed by xylene and subsequently paraffin embedding. The tissues were sectioned into 5 micron thick pieces using a rotary microtome and mounted onto glass slides. Following this, each slide was again subjected to a deparaffinization and rehydration procedure using reverse graded alcohols, and subsequently stained with standard protocols for tissue using the stain of hematoxylin and eosin. Hematoxylin provides the blue-purple staining of nuclei, while eosin stains the cytoplasm and extracellular matrix a pink-red, providing excellent clarity for evaluating structural and cellular characteristics.

Stained sections were examined at 100×, 200× and 400× magnification by two pathologists independently blinded to group assignment. The pathology assessment focused on many areas including: epithelialization status (complete, partial, absent), epithelial layer thickness and organization, stratum basale hyperplasia, inflammatory cell infiltration (type, density and distribution), granulation tissue formation and maturity, an evaluation of collagen fiber characteristics (thickness, density and orientation), neovascularization (blood vessel number and morphology), fibroblast proliferation and activity and scar tissue formation. Digital photomicrographs were taken to allow the pathologists to document their findings and perform detailed analyses. A semi-quantitative scoring system was used to standardize assessments across all samples.

### **Statistical Analysis**

Data analyses utilized appropriate statistical software. Continuous data were presented as means and standard deviations. The Shapiro-Wilk test was used to determine normality of continuous variable distributions. If the variable was normally distributed, independent t-tests were used to make comparisons of means between groups. Repeated measures analysis of variance (ANOVA) was conducted to analyze the change in wound area over time. For categorical variables, comparisons were made using the chi-square or Fisher's exact tests, based on the type of data. Statistical significance was defined as a p-value < 0.05. All statistical tests were conducted at a two-tailed level of analysis.

### **Ethics Approval and Animal Care**

This study was conducted in accordance with the guidelines established by the institution's Institutional Animal Ethics Committee. All procedures and protocols were performed according to international guidelines for laboratory animal care and use. Animals were kept at the institution in a controlled environment in a 12-hour light/dark cycle with a temperature of 22±2°C, and 55±10% relative humidity. The animals were fed standard laboratory chow and water ad libitum for the duration of the study.

## Results

All fifty rabbits are in good health and show no signs of experiencing serious adverse reactions to experimental procedures conducted over the 14 days of testing, and no rabbits died during the testing period. The only side effects that occurred during this period were mild erythema around the edge of the wounds (within the treatment and control groups) of three rabbits in the control group, all of which disappeared on their own without treatment. Neither the treatment group nor the control group exhibited signs of systemic toxicity or allergic reaction as a result of the procedures performed during the study.

### Measurement of Area Wounds

The wound areas at the beginning of the study were similar for the treatment and control groups, confirming that randomization of rabbits had been accomplished successfully. Both groups experienced a decrease in the dimensions of their wounds at a steady rate (over time) through the 14-day study period, but the treatment group exhibited a significantly greater amount of contraction, on average, than did the control group. The following table shows the various wound areas at the different time intervals (post-surgically) through 14 days after surgery.

**Table 1. Wound Area Measurements (cm<sup>2</sup>) Over Time**

Time Point	Group A (Control)	Group B (Treatment)
Day 0	3.14 ± 0.18	3.12 ± 0.16
Day 7	2.08 ± 0.24	1.26 ± 0.19*
Day 14	0.94 ± 0.21	0.28 ± 0.12*

Data presented as mean ± SD. \* $p < 0.05$  compared to control group. Statistical analysis was performed using SPSS version 26.0 (IBM, USA).

### Wound Contraction Percentage

The treatment group exhibited significantly higher wound contraction percentages at both evaluation time points compared to controls. Results are summarized in Table 2.

**Table 2. Wound Contraction Percentage**

Time Point	Group A (%)	Group B (%)
Day 7	33.8 ± 5.2	59.6 ± 4.8*
Day 14	70.1 ± 6.4	91.0 ± 3.2*

Data presented as mean ± SD. \* $p < 0.05$  compared to control group. Statistical analysis was performed using SPSS version 26.0 (IBM, USA).

### Time to Complete Epithelialization

The mean time to complete epithelialization was significantly shorter in the treatment group (11.2 ± 1.4 days) compared to the control group (16.8 ± 2.1 days,  $p < 0.001$ ), representing approximately a 33% reduction in healing time.

### Histopathological Findings

Histopathological examination revealed distinct differences between treatment and control groups at both time points evaluated. Semi-quantitative scoring of key histological parameters is presented in Table 3.

**Table 3. Histopathological Scoring at Day 7 and Day 14**

Parameter	Group A Day 7	Group B Day 7	Significance
Epithelialization	Partial/Mild	Advanced	p<0.01
Collagen Density	Moderate	High/Dense	p<0.01
Angiogenesis	Minimal	Prominent	p<0.001
Inflammation	Severe	Moderate	p<0.05

*Scoring based on semi-quantitative scale: Minimal (1), Moderate (2), Severe (3).*

At day 7, the control group specimens exhibited partial hyperplastic changes (mild) at the level of the basal layer and extensive scar-tissue development. Large quantities of granulation tissue were present and numerous inflammatory cells were observed within the granulation tissue, which indicates there is still an active acute inflammatory process. In comparison, the specimens from the treatment group exhibited evidence of advanced wound-healing characteristics, such as the formation of small calibre blood vessels, collection of red blood cells, thick and coarse collagen fibres, and abundant macrophage accumulation in the dermal layer below the epidermis. Thus, the treatment samples demonstrated a shift into the proliferative phase of healing (Figs. 1 and 2).

By day 14 of healing, the control group tissues had developed a thin crust covering the wound site due to the hyperplastic changes that occurred in the basal layer and the resulting completed and thin epidermal covering. In addition, the collagen fibres present within the healed area exhibited an irregular, coarse, and thin appearance in addition to there being a few inflammatory cells present. There was minimal blood vessel formation observed in the area of the wound site such that now the area surrounding the wound site would be able to properly heal. On the other hand, the treatment group exhibited extensive macrophage accumulation, extensive blood vessel development with highly organized vascular structures, increased numbers of acidophilic collagen fibres indicative of mature extracellular matrix, and significant fibroblast accumulation. All of these findings support the accelerating effects of the treatment on the rate of wound healing and the overall quality of tissue in the area of healing (Figs. 3 and 4).

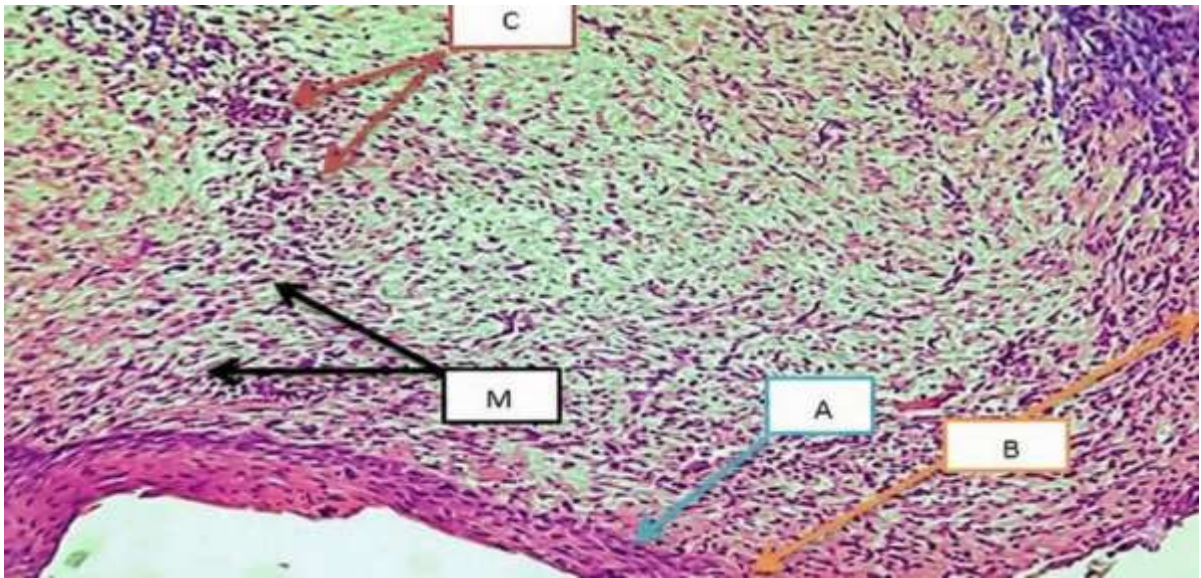


Figure 1. Histopathological section in at 7 day .in group A showed (A) Partial and mild hyperplasia of stratum basale(B) with wide scar tissue(C) . profuse granulation tissue(M) and high infiltration of inflammatory cells. H&E100X

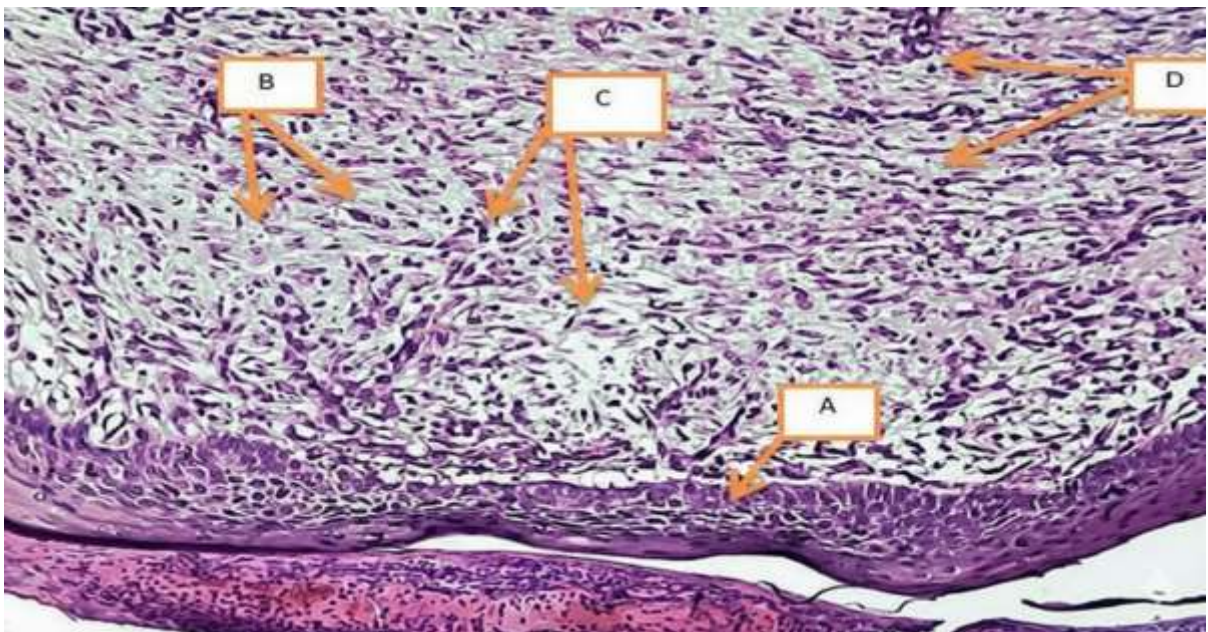


Figure:2 .: Histopathological appearance in skin at 14day .in group A showed (A) thin crust above the wound area. Complete and thin epidermal layer due to hyperplasia of stratum basale (B). Irregular, coarse and thin collagen fibers (C) with scattered inflammatory cells (D) and few formation of new blood vessels (angiogenesis )in the healed area. 100X H&E.

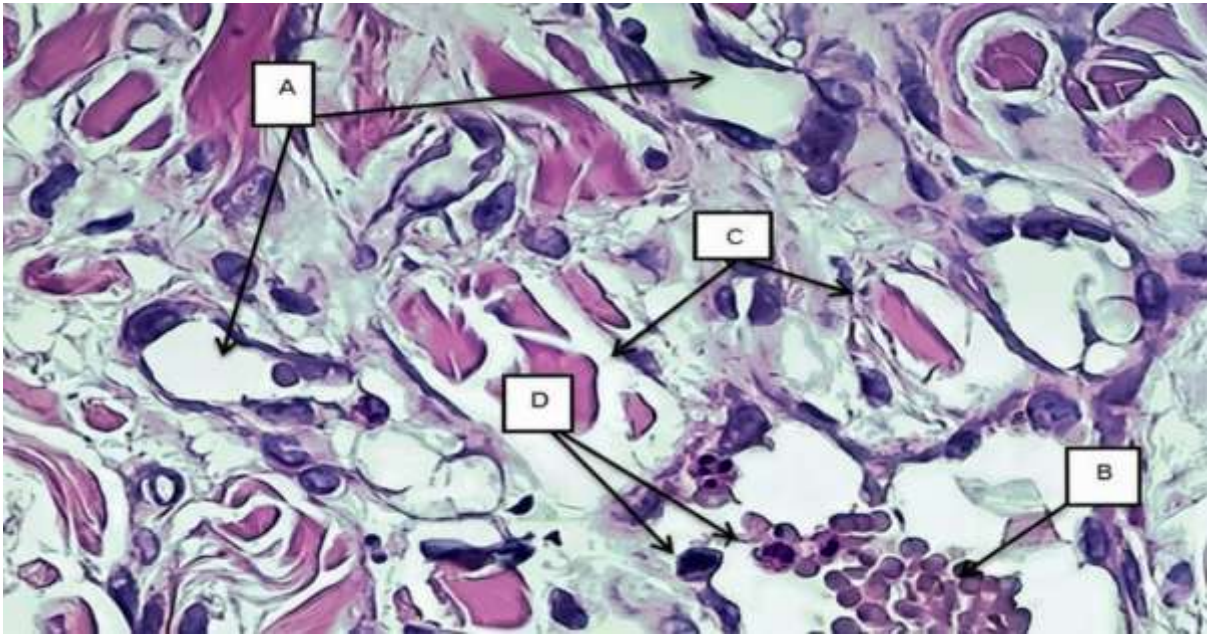


Figure 3. Histopathological appearance of burns in skin at 7 day.in group B showed (A) newly formed small blood vessels(B) and accumulation of RBC in it(C) , thick and coarse collagen fibers(D)and infiltration of macrophages in the dermis. Higher magnification , H&E.400X .

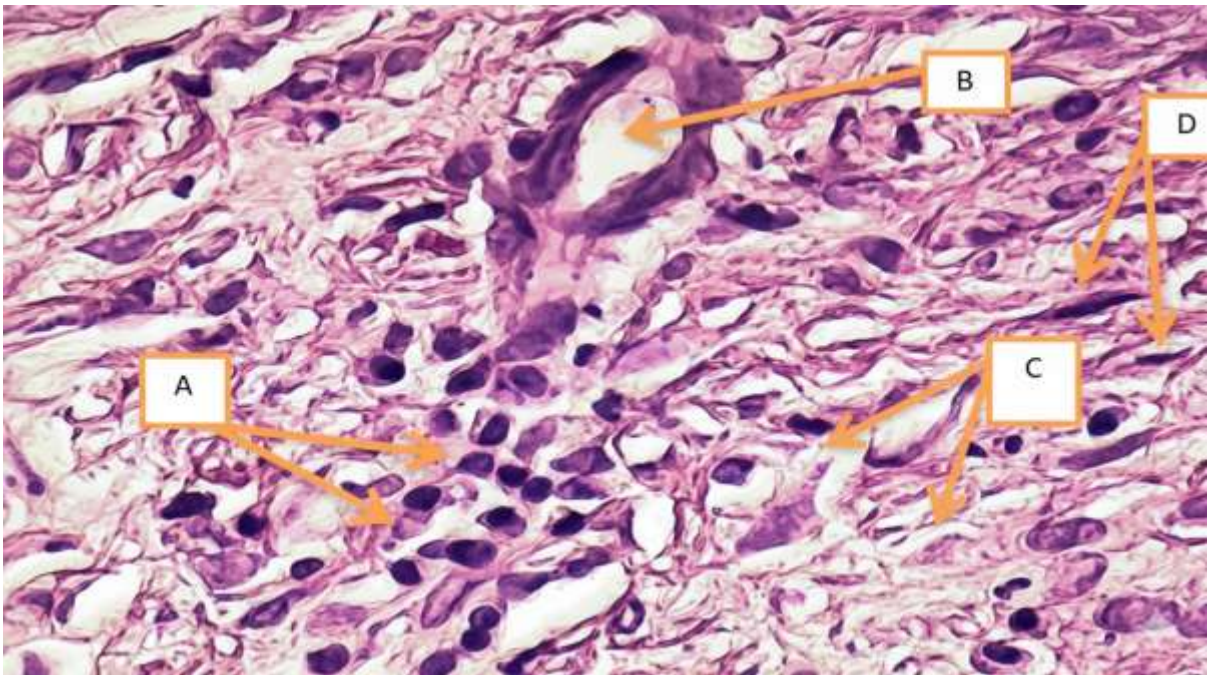


Figure 4. Histopathological appearance burns in skin at 14day ingroup B showed (A) infiltration of macrophages and (B) formation of blood vessel with (C) acidophilic collagen fibers(D) and proliferation of fibroblasts(D). Higher magnification 400X H&E.

## Discussion

The present study demonstrates the therapeutic potential of a novel topical formulation containing sesame oil and garlic extract for burn wound treatment. Results show that this formulation improves all stages of wound healing compared to traditional petroleum-based products, including increased wound closure rate (approximately 91% in the treatment group versus 70% in controls by day 14), accelerated epithelialization, enhanced angiogenesis, and improved overall tissue quality.

The improved wound closure rate observed in the treatment group represents a clinically important difference that could lead to shorter healing times, reduced infection risk, and better patient outcomes. Accelerated wound healing likely results from synergistic effects of bioactive compounds in sesame oil and garlic extract. Sesamol, the primary antioxidant in sesame oil, has been shown to reduce oxidative stress at wound sites, an important step for achieving cellular proliferation and protecting newly formed tissues (Broughton et al., 2006). In addition to reducing oxidative stress, sesame oil's antioxidant activity neutralizes reactive oxygen species generated by burn injury, which can otherwise cause cellular damage and impair healing by interfering with growth factor signaling (Barrientos et al., 2008).

Allicin, the primary organosulfur compound in garlic extract, contributes to wound healing through multiple mechanisms. Beyond its broad-spectrum antimicrobial activity, which plays an important role in preventing wound infection—one of the most significant complications in burn management—allicin may also stimulate angiogenesis and improve healed scar strength (Halstead et al., 2015). Previous studies have demonstrated allicin's ability to inhibit growth of Gram-positive and Gram-negative bacteria commonly associated with burns, including *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Escherichia coli* (Cardoso et al., 2004). Allicin's ability to destroy antibiotic-resistant bacterial strains addresses the growing problem of antimicrobial resistance in wound care (Johnson & Wilgus, 2014).

Histopathological findings provide insight into tissue healing mechanisms. The treatment group on day 7 demonstrated higher vascularization compared to controls, as evidenced by increased numbers of newly formed blood vessels containing red blood cells within the wound bed. Angiogenesis plays a crucial role in wound healing by supplying oxygen and nutrients to healing tissues requiring high cellular activity and by assisting in cellular debris clearance (Li et al., 2007). The pro-angiogenic effects of allicin have been demonstrated in various experimental models through increased vascular endothelial growth factor and other angiogenic cytokines (Chattopadhyay & Raines, 2014). Rapid appearance of functional vascular networks also supports other healing phases, including collagen synthesis and tissue remodeling.

Collagen quantity and structural arrangement are significant determinants of wound strength and scar quality. By day 7, treatment group collagen was heavily vascularized with acidophilic appearance; by day 14, treatment group collagen achieved well-organized architecture with mature extracellular matrix development. In contrast, control group collagen at day 14 remained disorganized with poorly formed structural arrangement. Enhanced collagen production in the treatment group may result from increased fibroblast activity, as evidenced by significant fibroblast proliferation on histological assessment (Tracy et al., 2016). Sesame oil contains essential fatty acids that serve as eicosanoid precursors modulating fibroblast activity, while garlic stimulates collagen production through transforming growth factor-beta (TGF- $\beta$ ) signaling modulation (Wynn & Vannella, 2016).

Inflammatory profiles differed markedly between groups. While both groups exhibited inflammatory

cell infiltration, the treatment group demonstrated a more balanced profile toward resolution, with predominant macrophage infiltration rather than persistent acute inflammatory cell types. Macrophages serve two important functions during wound healing: initial removal of debris and pathogens, and transition to a reparative phenotype promoting angiogenesis and fibroplasia through growth factor and cytokine release facilitating tissue remodeling (Lee et al., 2018). The anti-inflammatory effects of sesame and garlic may prevent excessive or prolonged inflammation that would negatively affect healing and lead to pathological scarring. Both sesame and garlic modulate pro-inflammatory mediators including tumor necrosis factor-alpha, interleukin-1 beta, and cyclooxygenase-2 to maintain inflammatory states necessary for proper wound healing (Rousselle et al., 2019).

The treatment group exhibited markedly enhanced epithelialization rate, characterized by migration and proliferation of new epithelial cells to cover the open wound surface. Time to complete epithelial coverage was significantly shorter in the treatment group, with epithelial layer thickness exceeding that of controls and appearing more organized. Enhanced epithelialization may result from continued stimulation of keratinocyte proliferation and migration through mechanisms regulated by epidermal growth factor and keratinocyte growth factor pathways (Mogoşanu & Grumezescu, 2014). Research suggests sesame oil promotes an optimal wound healing environment, permitting cell migration while preventing excessive drying, particularly during epithelialization. Sesame oil's antimicrobial activity also prevents bacterial colonization that would otherwise delay epithelialization.

Sesame oil's hydrophobic and lipophilic characteristics serve multiple purposes in formulation. First, it acts as a natural carrier for bioactive compounds (sesamol and allicin). Second, sesame oil forms an occlusive barrier minimizing transepidermal water loss, maintaining a moist wound environment, and preventing exposure to environmental contaminants—particularly important for burn wound treatment (Dwivedi et al., 2017). Combined with lanolin and liquid paraffin, the base formulation provides additional benefits, creating an optimal emollient system combining skin penetration enhancement with barrier protection.

Literature review confirms both agreement with and contributions made by this study. While several studies have investigated either sesame or garlic separately as wound healing agents, most report beneficial findings, with at least one study finding both agents positively impact wound healing in validated animal models. However, limited information exists regarding combined formulations and burn histopathology where combination would provide synergistic effects. Therefore, this study advances current understanding of sesame and garlic as wound care agents and introduces potential for superior delivery via combined formulation compared to single applications of each agent used independently (Dorsett-Martin, 2004).

## **Conclusions**

The current study has demonstrated that the topical application of sesame and garlic extract significantly improves the healing of burn wounds using an animal (rabbit) model. Specifically, this treatment accelerates wound closure, increases the rate of epithelialization, enhances angiogenesis, increases collagen deposition and organization, and reduces the degree of inflammation compared to petroleum-based controls. Histo-pathological evaluations provided evidence of the therapeutic effects being due to the synergistic effects of the sesamol and allicin, which acted on multiple aspects of the wound healing process as follows: 1) oxidative stress, 2) inflammation, 3) risk of infection, and 4) tissue regeneration. The results support the potential of this formulation's use in clinical settings as a safe, effective, natural

alternative for managing burn wounds. In order to validate these promising results and facilitate basic research into the possible applications of these products, further clinical studies will be necessary.

## References

- Abdullahi, A., Amini-Nik, S., & Jeschke, M. G. (2014). Animal models in burn research. *Cellular and Molecular Life Sciences*, 71(17), 3241-3255.
- Ankri, S., & Mirelman, D. (1999). Antimicrobial properties of allicin from garlic. *Microbes and Infection*, 1(2), 125-129.
- Arreola, R., Quintero-Fabián, S., López-Roa, R. I., Flores-Gutiérrez, E. O., Reyes-Grajeda, J. P., Carrera-Quintanar, L., & Ortuño-Sahagún, D. (2015). Immunomodulation and anti-inflammatory effects of garlic compounds. *Journal of Immunology Research*, 2015, 401630.
- Atiyeh, B. S., Costagliola, M., Hayek, S. N., & Dibo, S. A. (2007). Effect of silver on burn wound infection control and healing: Review of the literature. *Burns*, 33(2), 139-148.
- Barrientos, S., Stojadinovic, O., Golinko, M. S., Brem, H., & Tomic-Canic, M. (2008). Growth factors and cytokines in wound healing. *Wound Repair and Regeneration*, 16(5), 585-601.
- Bayan, L., Koulivand, P. H., & Gorji, A. (2014). Garlic: A review of potential therapeutic effects. *Avicenna Journal of Phytomedicine*, 4(1), 1-14.
- Biswas, T. K., & Mukherjee, B. (2003). Plant medicines of Indian origin for wound healing activity: A review. *International Journal of Lower Extremity Wounds*, 2(1), 25-39.
- Block, E. (1985). The chemistry of garlic and onions. *Scientific American*, 252(3), 114-119.
- Borlinghaus, J., Albrecht, F., Gruhlke, M. C., Nwachukwu, I. D., & Slusarenko, A. J. (2014). Allicin: Chemistry and biological properties. *Molecules*, 19(8), 12591-12618.
- Broughton, G., Janis, J. E., & Attinger, C. E. (2006). Wound healing: An overview. *Plastic and Reconstructive Surgery*, 117(7 Suppl), 1e-S-32e-S.
- Cardoso, C. R., Souza, M. A., Ferro, E. A., Favoreto, S., Jr., & Pena, J. D. (2004). Influence of topical administration of n-3 and n-6 essential and n-9 nonessential fatty acids on the healing of cutaneous wounds. *Wound Repair and Regeneration*, 12(2), 235-243.
- Chattopadhyay, S., & Raines, R. T. (2014). Collagen-based biomaterials for wound healing. *Biopolymers*, 101(8), 821-833.
- Cutler, R. R., & Wilson, P. (2004). Antibacterial activity of a new, stable, aqueous extract of allicin against methicillin-resistant *Staphylococcus aureus*. *British Journal of Biomedical Science*, 61(2), 71-74.
- Dorsett-Martin, W. A. (2004). Rat models of skin wound healing: A review. *Wound Repair and Regeneration*, 12(6), 591-599.
- Dwivedi, D., Dwivedi, M., Malviya, S., & Singh, V. (2017). Evaluation of wound healing, anti-microbial and antioxidant potential of *Pongamia pinnata* in wistar rats. *Journal of Traditional and Complementary Medicine*, 7(1), 79-85.
- Enoch, S., & Leaper, D. J. (2008). Basic science of wound healing. *Surgery*, 26(2), 31-37.
- Greenhalgh, D. G. (2019). Management of burns. *New England Journal of Medicine*, 380(24), 2349-2359.
- Gurtner, G. C., Werner, S., Barrandon, Y., & Longaker, M. T. (2008). Wound repair and regeneration. *Nature*, 453(7193), 314-321.
- Halstead, F. D., Rauf, M., Bamford, A., Wearn, C. M., Bishop, J. R. B., Burt, R., & Oppenheim, B. A.

- (2015). Antimicrobial dressings: Comparison of the ability of a panel of dressings to prevent biofilm formation by key burn wound pathogens. *Burns*, 41(8), 1683-1694.
- Hsu, D. Z., & Liu, M. Y. (2004). Sesame oil protects against lipopolysaccharide-stimulated oxidative stress in rats. *Critical Care Medicine*, 32(1), 227-231.
- Jeschke, M. G., van Baar, M. E., Choudhry, M. A., Chung, K. K., Gibran, N. S., & Logsetty, S. (2020). Burn injury. *Nature Reviews Disease Primers*, 6(1), 11.
- Johnson, K. E., & Wilgus, T. A. (2014). Vascular endothelial growth factor and angiogenesis in the regulation of cutaneous wound repair. *Advances in Wound Care*, 3(10), 647-661.
- Joshi, R., Kumar, M. S., Satyamoorthy, K., Unnikrisnan, M. K., & Murugesan, T. (2005). Free radical reactions and antioxidant activities of sesamol: Pulse radiolytic and biochemical studies. *Journal of Agricultural and Food Chemistry*, 53(7), 2696-2703.
- Kamal-Eldin, A., & Appelqvist, L. A. (1996). The chemistry and antioxidant properties of tocopherols and tocotrienols. *Lipids*, 31(7), 671-701.
- Kapoor, R., & Huang, Y. S. (2006). Gamma linolenic acid: An antiinflammatory omega-6 fatty acid. *Current Pharmaceutical Biotechnology*, 7(6), 531-534.
- Kumar, B., Vijayakumar, M., Govindarajan, R., & Pushpangadan, P. (2007). Ethnopharmacological approaches to wound healing—Exploring medicinal plants of India. *Journal of Ethnopharmacology*, 114(2), 103-113.
- Kumar, C. M., Sathisha, U. V., Dharmesh, S., Rao, A. G., & Singh, S. A. (2011). Interaction of sesamol with tyrosinase and its effect on melanin synthesis. *Biochimie*, 93(3), 562-569.
- Lee, H. J., Lim, Y., Park, S. Y., Park, Y. S., Kwon, G. T., & Yoon Park, J. H. (2018). Alleviation of UVB-induced photoaging by *Allium hookeri* extract via PPAR- $\gamma$ /NF- $\kappa$ B signaling. *Journal of Photochemistry and Photobiology B: Biology*, 178, 176-184.
- Li, J., Chen, J., & Kirsner, R. (2007). Pathophysiology of acute wound healing. *Clinics in Dermatology*, 25(1), 9-18.
- Martin, P., & Leibovich, S. J. (2005). Inflammatory cells during wound repair: The good, the bad and the ugly. *Trends in Cell Biology*, 15(11), 599-607.
- Mogoşanu, G. D., & Grumezescu, A. M. (2014). Natural and synthetic polymers for wounds and burns dressing. *International Journal of Pharmaceutics*, 463(2), 127-136.
- Namiki, M. (2007). Nutraceutical functions of sesame: A review. *Critical Reviews in Food Science and Nutrition*, 47(7), 651-673.
- Peck, M. D. (2011). Epidemiology of burns throughout the world. Part I: Distribution and risk factors. *Burns*, 37(7), 1087-1100.
- Pereira, R. F., & Bartolo, P. J. (2016). Traditional therapies for skin wound healing. *Advances in Wound Care*, 5(5), 208-229
- Reinke, J. M., & Sorg, H. (2012). Wound repair and regeneration. *European Surgical Research*, 49(1), 35-43.
- Rousselle, P., Braye, F., & Dayan, G. (2019). Re-epithelialization of adult skin wounds: Cellular mechanisms and therapeutic strategies. *Advanced Drug Delivery Reviews*, 146, 344-365.
- Rowan, M. P., Cancio, L. C., Elster, E. A., Burmeister, D. M., Rose, L. F., Natesan, S., & Chung, K. K. (2015). Burn wound healing and treatment: Review and advancements. *Critical Care*, 19, 243.
- Shedoeva, A., Leavesley, D., Upton, Z., & Fan, C. (2019). Wound healing and the use of medicinal

- plants. *\*Evidence-Based Complementary and Alternative Medicine, 2019\**, 2684108.
- Singer, A. J., & Clark, R. A. (1999). Cutaneous wound healing. *New England Journal of Medicine*, 341(10), 738-746.
- Sullivan, T. P., Eaglstein, W. H., Davis, S. C., & Mertz, P. (2001). The pig as a model for human wound healing. *Wound Repair and Regeneration*, 9(2), 66-76.
- Tracy, L. E., Minasian, R. A., & Caterson, E. J. (2016). Extracellular matrix and dermal fibroblast function in the healing wound. *Advances in Wound Care*, 5(3), 119-136.
- World Health Organization. (2018). Burns. World Health Organization.
- Wynn, T. A., & Vannella, K. M. (2016). Macrophages in tissue repair, regeneration, and fibrosis. *Immunity*, 44(3), 450-462.
- Xie, X., Ma, L., Xi, K., Zhang, W., & Fan, D. (2017). In vitro angiogenic effect of platelet-rich plasma in combination with dental implant titanium alloys. *Platelets*, 28(2), 134-143.
- Yuan, H., Ma, Q., Ye, L., & Piao, G. (2016). The traditional medicine and modern medicine from natural products. *Molecules*, 21(5), 559.