



EVALUATION OF COMPARATIVE WOUND HEALING EFFICACY OF *ALOE VERA* AND *ACACIA* HONEY ALONE AND IN COMBINATION ON BURN WOUNDS IN RABBIT MODEL

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Abstract:

Burn injuries are a complex and painful condition that pose many challenges. To reduce tissue scarring and avoid infection, wounds must be managed effectively. While honey is prized for its analgesic, hemostatic, anti-inflammatory, antimicrobial, and wound-healing qualities, aloe vera is known for its wide range of pharmaceutical and therapeutic qualities. The purpose of this study was to assess the effects of honey and aloe vera, both separately and together, on the healing of burn wounds. From local sources, forty healthy adult rabbits of different sexes were acquired and appropriately acclimated. The animals were split into four groups after being given burn wounds to the thoracolumbar region. Group B was treated with honey, and Group A was treated with aloe vera. Group D was given silver sulfadiazine as the control, while Group C was given a mixture of honey and Aloe vera. Tensile strength, healing time, contraction rate, and histopathological analysis of the regenerated tissue were among the metrics used to assess the effectiveness of the healing process. The statistical analysis employed ANOVA, and the mean differences were assessed using the least significant difference (LSD) test. The findings show that aloe vera and acacia honey together significantly increased wound contraction rate and tensile strength, which in turn shortened the healing period. Therefore, it is confidently advised to combine aloe vera and acacia honey for efficient wound healing in both humans and animals.

Keywords: H₂SO₄, parchment paper, *Acacia* honey, *Aloe vera*, silver sulfadiazine.

Introduction

A wound's location, cause, depth, tissue involvement, and clinical presentation are just a few of the variables that can be used to categorise it. To classify burn wounds, general wounds, diabetic foot

wounds, ulcerations, and other kinds of wounds, specific classification tools are used. For example, superficial burns include a variety of injuries whose severity depends on the depth and body part affected; these burns usually involve damage to the epidermis[1]. Partial thickness burns involve damage to deeper structures, whereas full thickness burns affect all layers of skin and underlying tissues[2]. A common way to describe the severity of the damage is as a percentage of the body that was burned. Depending on the extent of the wound, different protocols for managing it should be followed in order to prevent infections. Topical ointments can be used to treat minor burns, while immediate medical attention is required for severe burns. Inadequate wound closure raises the risk of infection and delays the healing process, which can lead to chronic wounds[3]. Although their efficacy varies, antimicrobial ointments like bacitracin, silver nitrate, mupirocin, and silver sulfadiazine are frequently used to reduce the risk of infection and promote wound healing[3]. Because the process of healing wounds is complex and multifaceted, there is always a need for new therapeutic agents. The healing process involves a number of processes, including remodeling, cell division, and inflammation. For a very long time, burn injuries have been treated with aloe vera, a plant[4]. Its gel's antibacterial properties can help remove bacteria that aggravate wounds and promote wound healing[5]. Vitamins A, C, E, B1, B2, folic acid, niacin, choline, and trace amounts of vitamin B12 can all be found in abundance in aloe vera. These nutrients may help promote healing. Similarly, honey has a lot of healthy ingredients[6]. It has a sticky texture, a pale yellow colour, and a unique smell. Fructose and glucose make up the majority of its composition, along with minerals, amino acids, vitamin C, and phenolic compounds. The analgesic, hemostatic, anti-inflammatory, antimicrobial, and wound-healing qualities of honey are well known [7, 8]. Its use as a wound care tool has been demonstrated to accelerate the healing process and lower inflammation, making it an effective natural treatment for a variety of wounds[9].

MATERIALS AND METHODS

Honey: We purchased certified-quality honey that is sold commercially that is made by bees from *Acacia modusta* flowers from the Faisalabad, Pakistan, local market.

Aloe vera: Fresh aloe vera sourced from the agronomy department of the University of Agriculture, Faisalabad.

Experimental animals

Forty clinically healthy adult rabbits, comprising both male and female, were acquired from a local supplier. These rabbits, bred locally and weighing between two to three kilograms each, were randomly allocated into four groups labeled as A, B, C, and D. A standardized pre- and post-operative care protocol was implemented consistently across all rabbits throughout the duration of the experiment to maintain uniformity in their feeding and management routines.

Wound creation

To achieve this objective, the thoracolumbar region of the rabbits underwent shaving to facilitate the creation of burn wounds. Prior to the procedure, the rabbits received premedication in the form of atropine sulfate (administered at a dose of 0.035 mg/kg via intramuscular route). Subsequently, total parenteral anesthesia was induced using ketamine (35 mg/kg via intramuscular injection). The rabbits were then securely positioned in a recumbent posture. Before proceeding, the designated area was meticulously cleaned using pyodine followed by alcohol swabbing.

Treatment protocol

Aloe vera was administered twice a day to Group A. Group B underwent twice-daily treatments with acacia honey, Group C received a combination of aloe vera and acacia honey twice daily, while Group D received twice-daily treatments of silver sulfadiazine.

Results

Wound contraction rate

On the third day following injury, there was no statistically significant difference observed in the rate of wound contraction between groups "B" and "C." However, it was evident that animals in groups "A" and "D" displayed relatively larger wound areas during this specific period of the study. From day 6 to day 15 post-injury, there was no statistically significant difference found between groups "A," "B," and "C," although a significant difference was noted between the treatment and control groups. A similar trend was observed on day 18 after injury, with no statistically significant difference observed between groups "B" and "C." By the 21st day post-injury, a contrasting observation was made for the other groups, as their wounds reached a comparable stage by the 24th day following injury, the wounds of the animals treated with honey and honey and Aloe vera combination completely contracted.

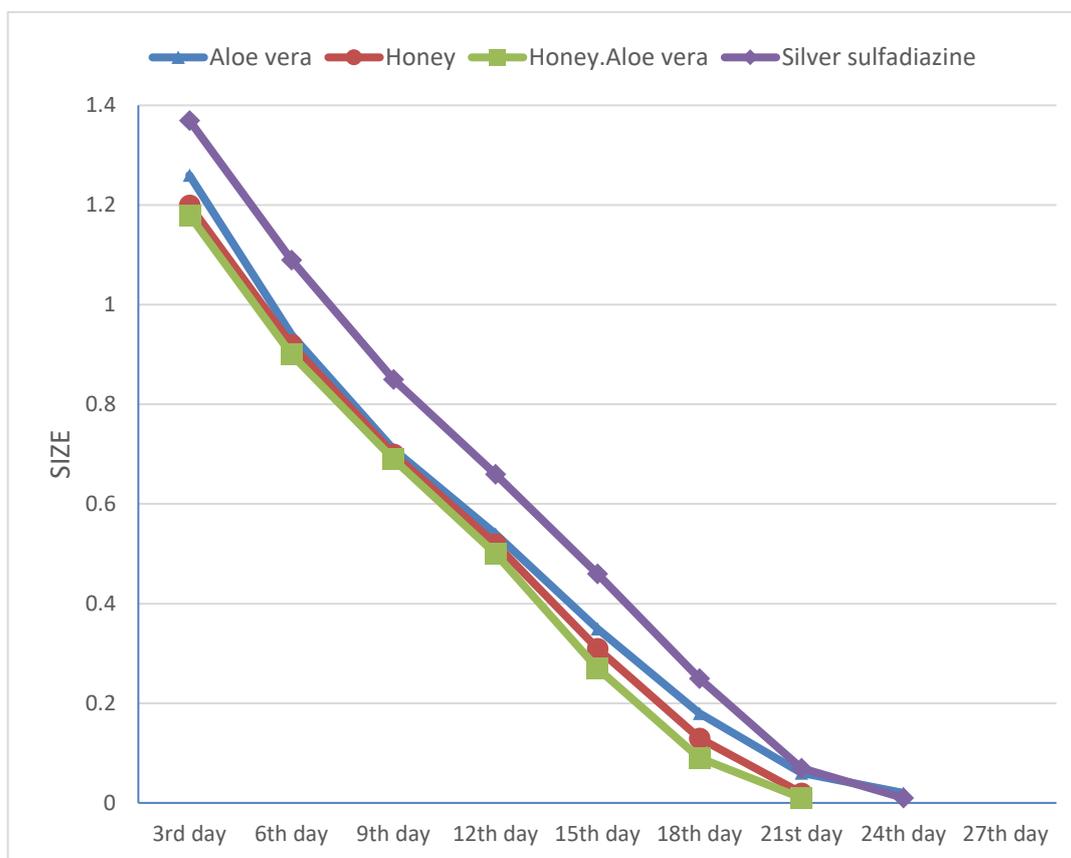


Fig. 1: Evaluation of the contraction rate (measured in cm=%; Mean±SD) at 3-day intervals across four groups: Aloe vera, honey, honey+Aloe vera, and silver sulfadiazine, on wounds in rabbits (n=40)

Healing time: The period from the onset of a wound to the complete shedding of scar tissue is referred to as the healing time. Remarkably, animals in group C, receiving a combined treatment of honey and aloe vera, demonstrated the swiftest wound healing rate compared to the other groups. Conversely, the healing time for group C was notably shorter than that of the remaining groups. Among the four groups, group B, treated with honey, exhibited the second-fastest healing process, while group D, treated with silver sulfadiazine, displayed the slowest healing rate (Fig 1). Specifically, group A treated with aloe vera, group B treated with honey, group C treated with honey and aloe vera, and the control group treated with silver sulfadiazine had mean healing times of 21.90 ± 0.781 , 21.00 ± 0.632 , 19.80 ± 0.663 , and 22.80 ± 0.663 days, respectively.

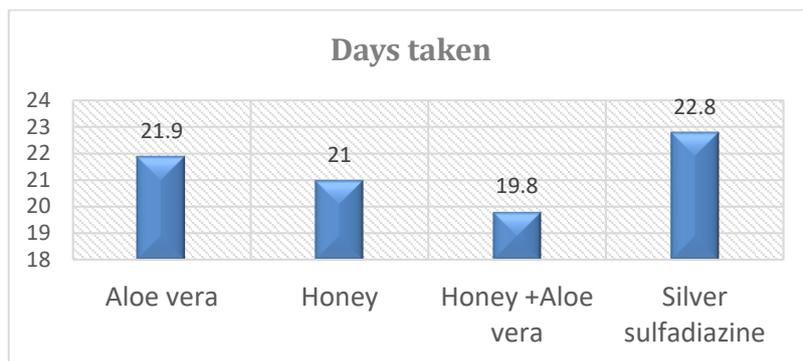


Fig. 2: At the conclusion of the study, an assessment was made on the healing time (in days) across four groups: Aloe vera, honey, honey and Aloe vera combination, and the control group treated with silver sulfadiazine, on wounds in rabbits (n=40)

Tensile strength: Significant variations in tensile strength were observed across all four groups. Specifically, group D, receiving treatment with silver sulfadiazine, exhibited the lowest tensile strength, whereas group C, treated with honey and aloe vera combination, demonstrated the highest tensile strength among all treated groups. The mean values along with the standard error for groups A, B, C, and D were 32.60 ± 0.510 , 38.40 ± 0.510 , 43.00 ± 0.707 , and 25.60 ± 0.748 N, respectively.

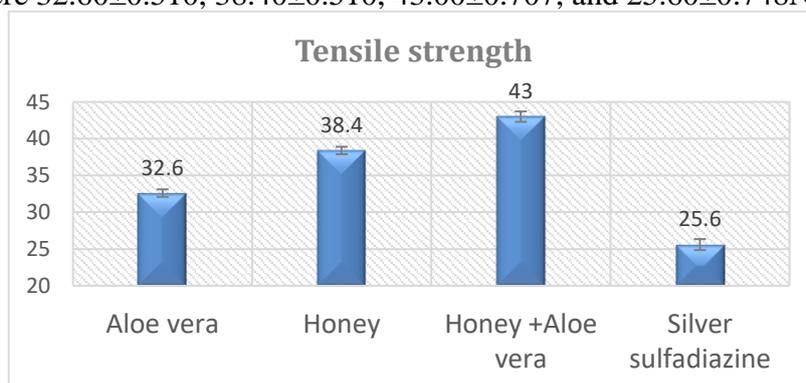


Fig. 3: Evaluation of the impact of topical application of Aloe vera, honey, honey combined with Aloe vera, and silver sulfadiazine on the tensile strength of wounds (n=40) in rabbits at the conclusion of the study

Histopathological evaluation: Upon histopathological assessment, in contrast to groups A (administered aloe vera), B (administered acacia honey), and D (administered silver sulfadiazine), the study's results indicated that animals in group C (treated with honey combined with aloe vera) displayed enhanced thickness of skin layers and well-developed epidermis. Furthermore, when compared to group B (treated with acacia honey) and group C (treated with silver sulfadiazine), animals in group A (treated with aloe vera) exhibited a higher percentage of collagen fiber based on histopathological analysis.

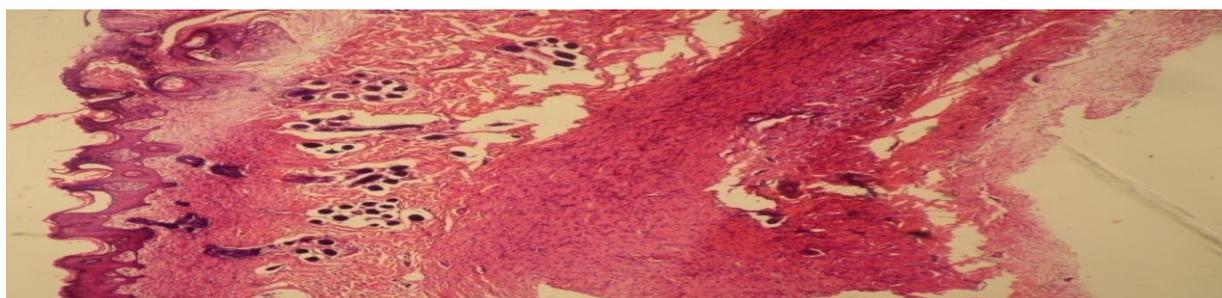


Fig.4: Photomicrograph of a slide revealing a thin epidermis with ample collagen fiber, as well as the presence of blood vessels, in the healed wound tissue treated with Aloe vera

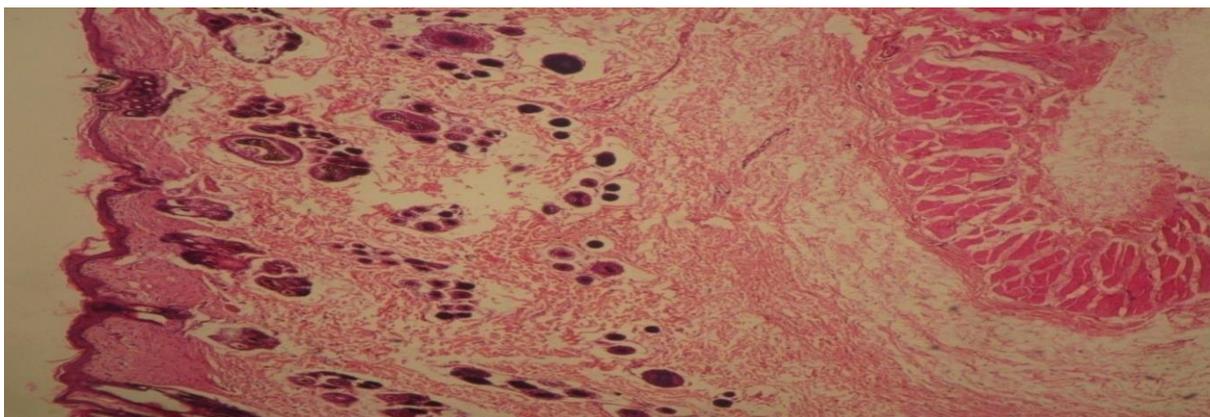


Fig.5: *Photomicrograph of a slide depicting the formation of a thin epidermis and the substantial presence of collagen fiber in the dermis of healed wound tissue treated with honey.*

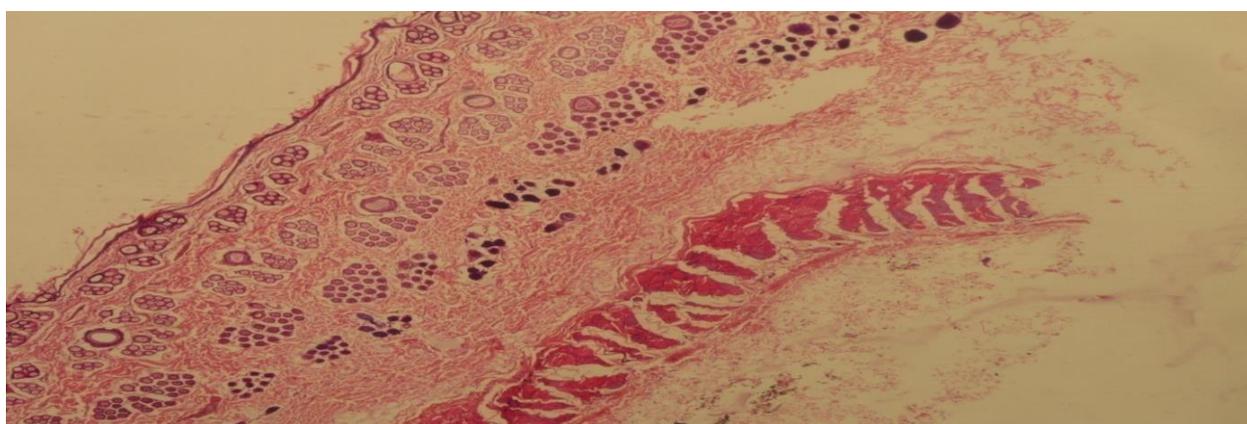


Fig.6: *Photomicrograph of a slide illustrating a thin epidermis with the highest concentration of collagen fibers in the dermis, along with the presence of blood vessels in the tissue of the healed wound treated with a combination of acacia honey and aloe vera*

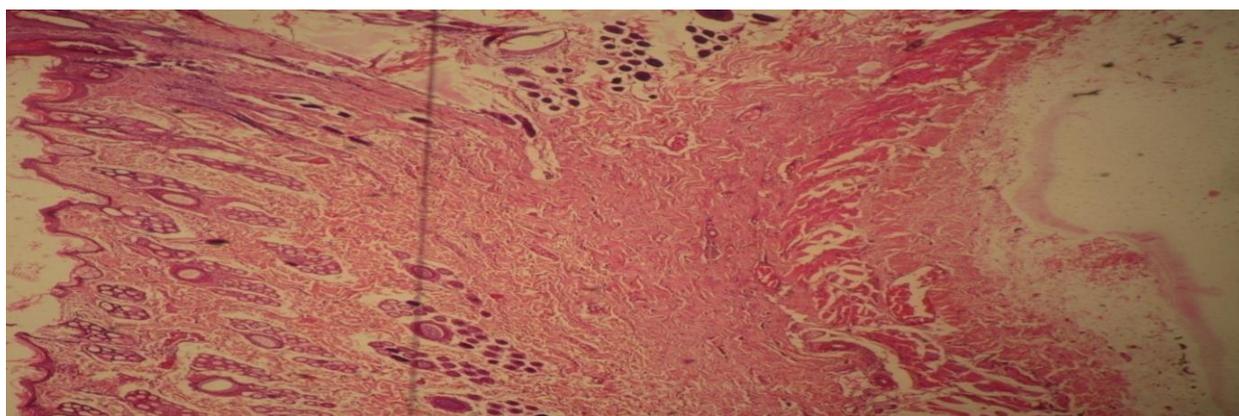


Fig.7: *Photomicrograph of a slide displaying a thin and less developed epidermis with an adequate presence of collagen in the dermis in healed wound tissue treated with Quench (Silver sulfadiazine)*

Discussion: This study aimed to assess the efficacy of aloe vera and honey, both individually and in combination, in facilitating wound healing using a rabbit model with burn wounds. The standard cream, silver sulfadiazine, was compared with these treatments. Tensile strength, healing time, wound contraction rate, and histopathology of second intention burn wounds were among the evaluation parameters. The temperature and respiration rate did not significantly differ between the treatment groups, according to the results ($p>0.05$). Unlike the other treatment groups, wounds

treated solely with honey or in combination with aloe vera demonstrated accelerated healing. In contrast, wounds treated with aloe vera and silver sulfadiazine exhibited delayed healing. Across the treatment groups - Aloe vera (A), honey (B), Aloe vera and honey combination (C), and silver sulfadiazine (control group D) - the mean healing times were 21.90 ± 0.781 , 21.00 ± 0.632 , 19.80 ± 0.663 , and 22.80 ± 0.663 days, respectively. The synergistic combination of minerals, vitamins, antioxidants, and anti-inflammatory properties present in honey and aloe vera may contribute to the expedited healing process[10, 11]. The tensile strength of the healed tissue was also assessed, with group C, receiving the combination treatment of honey and aloe vera, demonstrating higher tensile strength compared to groups A, B, and D, treated with aloe vera, honey, and silver sulfadiazine, respectively. The mean tensile strength values for groups A, B, C, and D were 32.60 ± 0.510 , 38.40 ± 0.510 , 43.00 ± 0.707 , and 25.60 ± 0.748 N, respectively. The enhanced tensile strength observed in group C may be attributed to the presence of enzymes, minerals, and antioxidants in honey, facilitating fibroblast and myofibroblast migration to the wound site and promoting granulation tissue formation and re-epithelization[12, 13]. Histopathological analysis revealed that group C (honey and aloe vera combination) exhibited superior epidermis development with thicker skin layers compared to groups A, B, and D [14]. Furthermore, compared to groups treated with Aloe vera, honey and Aloe vera combination, and silver sulfadiazine (groups A, C, and D, respectively), group B (honey alone) displayed a higher percentage of collagen fibers. This implies that, in comparison to single treatments or conventional therapy alone, the combination of honey and Aloe vera promotes stronger tissue regeneration[15, 16].

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