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Formulation, Optimization, Physicochemical Characterization, and Pharmacological Evaluation of Aloe vera-Enriched WoundRx Cream Against Excision, Incision, and Burn-Induced Diabetic Wounds in Streptozotocin-Induced Diabetic Wistar Albino Rats

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ABSTRACT

The study aimed to formulate and evaluate a natural topical preparation, named WoundRx Cream, designed specifically to manage excision, incision, and burn wounds in streptozotocin-induced diabetic Wistar albino rats. WoundRx Cream was prepared using natural honey, olive oil, beeswax, and varying concentrations of *Aloe vera* extract, employing the fusion method. Six formulations (WRF1–WRF6) were characterized based on physical appearance, pH, spreadability, extrudability, and homogeneity. In-vivo wound healing was assessed using excision, incision, and burn wound models in diabetic rats, comparing untreated diabetic controls, normal rats treated with WoundRx Cream, and diabetic rats treated with WoundRx Cream. The pH of all formulations remained skin-compatible (7.36–7.56), while spreadability and extrudability were optimal, increasing with higher *Aloe vera* concentrations. In-vivo results demonstrated significantly accelerated healing, marked by reduced wound areas and enhanced tissue regeneration, particularly notable in diabetic rats treated with WoundRx Cream compared to diabetic controls ($p < 0.05$). This enhanced healing effect is attributed to the combined antimicrobial, anti-inflammatory, and regenerative properties of the ingredients, particularly honey and *Aloe vera*. These findings highlight the promising therapeutic potential of WoundRx Cream as an effective natural intervention for diabetic wound and burn management, supporting its clinical development and future application.

Keywords: *WoundRx Cream, Aloe vera, Diabetic wound healing, Excision wound model, Incision wound model, Burn wound model, Streptozotocin-induced diabetes, Natural topical formulation.*

1. INTRODUCTION

Diabetes mellitus is a chronic metabolic disorder characterized by elevated blood glucose levels, resulting from impaired insulin secretion, insulin action, or both. Globally, diabetes prevalence continues to rise significantly, with complications such as impaired wound healing becoming major health challenges. Patients with diabetes often experience delayed or

compromised wound healing, increasing susceptibility to chronic wounds, infections, amputations, and reduced quality of life. Impaired wound healing in diabetes primarily occurs due to persistent inflammation, reduced angiogenesis, poor collagen synthesis, and compromised immune responses at the wound site. Thus, diabetic wounds require specialized care and effective therapeutic interventions to promote healing and reduce associated morbidity (Dilworth et al., 2021; Nagesh et al., 2020).

Conventional treatment options, including topical antiseptics, synthetic antibiotics, and wound dressings, often present limitations such as antibiotic resistance, allergic reactions, toxicity, and limited efficacy. Therefore, there is growing interest in developing natural, effective topical formulations capable of enhancing the wound healing process, specifically tailored to diabetic wound conditions. Herbal and natural remedies have gained considerable attention due to their bioactive properties, minimal side effects, and wide availability. Ingredients such as honey, beeswax, olive oil, and plant extracts, especially from *Aloe vera*, have shown promising wound healing potential, attributed to their inherent antimicrobial, anti-inflammatory, antioxidant, and tissue-regenerative properties (Elkordy et al., 2021; Ogidi et al., 2021).

Honey, widely recognized for its antimicrobial and anti-inflammatory properties, accelerates wound healing by promoting tissue regeneration and preventing microbial colonization. Beeswax provides a protective barrier on the wound surface, maintaining moisture, reducing dehydration, and facilitating skin regeneration. Olive oil, rich in polyphenolic compounds and essential fatty acids, offers antioxidant protection and significantly enhances wound contraction and epithelialization. Among medicinal plants, *Aloe vera* (*Aloe barbadensis*) has been extensively studied for its exceptional wound healing abilities. It contains numerous bioactive compounds such as polysaccharides, anthraquinones, flavonoids, and vitamins, demonstrating anti-inflammatory, antimicrobial, analgesic, and collagen-stimulating effects, making it particularly valuable in managing diabetic wounds (Loiacono et al., 2025; Tekiki et al., 2022).

Considering the therapeutic potential of these natural ingredients, the present study focused on formulating a novel topical preparation named "WoundRx Cream." The formulation comprised natural honey, olive oil, beeswax, and varying concentrations of freshly prepared *Aloe vera* extract. The rationale behind combining these ingredients was to leverage their complementary and synergistic wound healing benefits, particularly targeting the compromised wound repair mechanisms observed in diabetic patients. The formulation was evaluated using excision, incision, and burn wound models in streptozotocin (STZ)-induced diabetic Wistar albino rats, well-established animal models mimicking diabetic wound conditions (Ghasemi & Jeddi, 2023).

The objectives of this research included assessing the physical and chemical characteristics of the prepared formulations, evaluating their efficacy in promoting wound healing and tissue regeneration in diabetic conditions, and comparing their healing potential against untreated diabetic controls. By addressing the limitations of current diabetic wound care treatments and utilizing the beneficial properties of natural components, this study aimed to provide valuable insights into developing effective, safe, and affordable topical therapies. Ultimately, this research contributes significantly to advancing natural-based therapeutics for diabetic wound management, thus enhancing patient outcomes and improving the overall quality of life for diabetic patients suffering from wounds and burns.

2. MATERIAL AND METHODS

Materials

Streptozotocin (STZ) was procured from Sigma Aldrich, USA, and normal saline solution (0.9% NaCl) was prepared in the laboratory using analytical-grade chemicals. Ethanol (70%) utilized for sterilization was also laboratory-prepared from analytical-grade ethanol. Natural honey was locally sourced and certified organic, ensuring quality and purity. Extra virgin, cold-pressed olive oil and pure refined beeswax were commercially obtained and selected based on their proven medicinal and pharmaceutical utility. Fresh leaves of *Aloe vera* were collected from the botanical garden and authenticated by a qualified botanist. Healthy male Wistar albino rats (weighing 230-280 g) used for the study were obtained from the institutional animal house facility. Instruments used included a digital pH meter, Vernier caliper (Mitutoyo, Japan), surgical blades and scissors, anesthesia mask with ether anesthetic, brass rod for burn induction, tensiometer for wound evaluation, and glass slides for spreadability assessments. Statistical analyses were performed using GraphPad Prism software.

Preparation of WoundRx Mixture

To optimize the wound healing potential of WoundRx Cream, five different formulations (WRF1 to WRF5) were developed by varying the proportion of *Aloe vera* extract, while maintaining constant volumes of natural honey, olive oil, and beeswax. Fresh *Aloe vera* leaves were collected, washed thoroughly, peeled, and homogenized to obtain the clear mucilaginous gel, which was used as the extract. The formulations were prepared as follows (Table 1):

Table 1. Formulation of WoundRx Mixture

Formulation Code	Honey (% v/v)	Olive Oil (% v/v)	Beeswax (% v/v)	Aloe vera Extract (% v/v)
WRF1	1	1	1	0.5
WRF2	1	1	1	1.0
WRF3	1	1	1	1.5
WRF4	1	1	1	2.0
WRF5	1	1	1	2.5

Each formulation was prepared by gently heating the ingredients over a water bath maintained at 60–65°C, with continuous stirring to ensure complete blending. Once a smooth and homogenous mixture was formed, the blend was allowed to cool to room temperature and then incorporated into the cream base using the fusion method. The final WoundRx Cream formulations were transferred into sterile containers for further evaluation.

Formulation of WoundRx Cream

A natural wax-based cream base was utilized for the final formulation. The fusion method was employed, wherein the wax base was first melted using a water bath at a controlled temperature. Once the base became a clear molten phase, the prepared WoundRx mixture was gradually added with continuous stirring to ensure consistent dispersion of all ingredients. The cream was then allowed to cool slowly at room temperature and was transferred into sterile, airtight containers for storage and subsequent evaluation (Al-Waili, 2003).

Evaluation of WoundRx Cream

***Extrudability* Test**

The extrudability of the WoundRx Cream formulations (WRF1–WRF5) was evaluated to assess the ease with which the product can be expelled from its container under applied pressure—a critical factor influencing user acceptability and dosing accuracy. The method was performed as described earlier with slight modifications. (Aiyalu et al., 2016; Al-Waili, 2003). Approximately 20 grams of the cream formulation was filled into a collapsible aluminum tube and sealed tightly. A standard weight of 1 kg was then placed on the tube for 10 seconds, and the amount of cream extruded from the nozzle was collected and weighed. The extrudability was expressed as the amount (in grams) of formulation extruded per unit force applied. A higher extrudability value indicated better flow characteristics and ensured easy application during clinical or home use.

Measurement of the pH

The pH of each WoundRx Cream formulation was determined to ensure that the formulation remained within the dermally acceptable pH range (typically between 4.5 and 7.0) to minimize skin irritation and enhance compatibility. The procedure was conducted following the method described by Queiroz et al. (2009) (Queiroz et al., 2009). For each formulation, 1 gram of cream was dispersed in 10 mL of distilled water to create a uniform dispersion. A digital pH meter was first calibrated using standard buffer solutions of pH 4.0 and 7.0. Once calibrated, the electrode was immersed into the cream dispersion and the pH was recorded at room temperature. The test was performed in triplicate for each formulation to ensure accuracy and reproducibility (Al-Waili, 2003).

Appearance, homogeneity and Spreadibility

Appearance

The physical appearance of the cream formulations was assessed visually to ensure patient acceptability and product consistency. Each formulation (F1–F5) was inspected for colour, odour, clarity, and overall visual appeal. All batches were observed to be smooth, creamy, and semi-solid in consistency, with colors ranging from pale yellow to light green, depending on the concentration of *Aloe vera* extract. No discoloration, lump formation, or phase separation was observed, confirming acceptable cosmetic properties (Nayak et al., 2005).

Homogeneity

The formulations were evaluated for homogeneity through visual inspection immediately after preparation and during storage. A small quantity of cream from each formulation was spread onto a clean glass slide and observed under light for any visible particles, aggregates, or phase separation. All five formulations demonstrated uniform distribution of components, with no gritty particles, air bubbles, or separation, indicating excellent homogeneity and stability.

Spreadability

Spreadability was evaluated to determine the ease of application and uniform distribution of the cream on the skin surface. The test was carried out using the slip and drag method. A fixed quantity (1 gram) of cream was placed between two clean glass slides. A 500 g weight was applied to the upper slide for 5 minutes, allowing the cream to spread uniformly. The diameter (in mm) of the spread circle was then measured (Jain, 2007).

***In Vivo* Studies**

Animals

Healthy adult male Wistar albino rats, weighing between 230–280 g, were selected for the study. The animals were housed under standard laboratory conditions, with a 12-hour light/dark cycle, controlled temperature and humidity, and had free access to standard pellet diet and water. Prior approval for the experimental protocol was obtained from the Institutional Animal Ethics Committee (IAEC), and all procedures were performed in accordance with CPCSEA guidelines.

Grouping of Animals

The animals were divided into groups consisting of six rats per group (n = 6) for each wound model—excision and incision. Each model included the following groups:

- Group I: Diabetic control (untreated)
- Group II: Diabetic treated with standard wound healing cream
- Group III: Diabetic treated with WoundRx Cream

This grouping enabled comparative analysis of wound healing responses across different treatment conditions.

Induction of Diabetes

Streptozotocin (STZ), procured from Sigma-Aldrich, was used to induce diabetes in the experimental animals. It was freshly prepared in normal saline and administered via a single intraperitoneal injection at a dose of 45 mg/kg body weight. After 72 hours of STZ administration, fasting blood glucose levels were measured using a glucometer. Rats showing blood glucose levels exceeding 120 mmol/L were considered diabetic and selected for further experimentation (Akbarzadeh et al., 2007).

***In Vivo* Wound Healing Studies**

To evaluate the wound healing efficacy of WoundRx Cream, three widely accepted models—excision, incision, and burn wound models—were used in streptozotocin-induced diabetic Wistar albino rats. Animals were divided into three groups: Group I (diabetic control), Group II (standard wound healing treatment), and Group III (WoundRx Cream-treated group), with six animals in each group (n = 6) per model. All procedures were carried out under aseptic conditions and appropriate anaesthesia.

a. Excision Wound Model

In this model, a full-thickness skin excision was created to simulate a deep open wound. Diabetic rats were anesthetized using the open-mask method with light ether. The dorsal thoracic area was shaved, sterilized using 70% ethanol, and a circular full-thickness wound approximately 500 mm² in area was created using a sterile surgical blade. Hemostasis was achieved using sterile cotton. The wound area was monitored regularly, and measurements were recorded on days 1, 3, 7, 14, 21, and 31 using a Vernier caliper and a transparent plastic ruler. The wound contraction was expressed as a percentage reduction in the original wound area using the following formula (Patil et al., 2012):

Wound Contraction (%) = (Initial Wound Area − Wound Area on Specific Day / Initial Wound Area) × 100

This model allowed the comparative evaluation of the wound closure rate in treated and untreated diabetic animals.

b. Incision Wound Model

For assessing tissue regeneration and tensile strength, the incision wound model was employed. After induction of light ether anesthesia, a 6 cm linear incision was made along the dorsal paravertebral region of each rat using a sterile surgical blade. The skin and underlying tissues were incised with care to ensure uniformity across all subjects. The incised edges were approximated and sutured with interrupted stitches at 1 cm intervals using sterile surgical thread. Topical treatments were applied daily as per the group allocation. Sutures were carefully removed on the eighth post-operative day. On the tenth day, wound tensile strength was measured using a tensiometer, which reflects the maximum force required to disrupt the wound. Higher tensile strength is indicative of better collagen cross-linking and skin regeneration (Patil et al., 2012).

c. Burn Wound Model

The burn wound model was used to assess the healing of thermally-induced injuries. After anesthetizing the diabetic rats with ether, the dorsal skin was shaved and sterilized, and a metal rod (2 cm diameter) preheated to 100°C was applied to the

skin for 20 seconds to induce a standardized second-degree burn wound. Care was taken to ensure equal pressure and exposure time across all animals. The resulting wounds were treated with the respective formulations once daily. Wound healing progress was monitored by measuring the wound area on days 1, 3, 7, 14, 21, and 31 using a Vernier caliper. The data obtained were used to calculate wound contraction percentage, similar to the excision model. The healing response was compared among the groups to evaluate the therapeutic potential of WoundRx Cream on thermal injuries.

Statistical Analysis

All quantitative results from the wound models were expressed as mean ± standard error of the mean (SEM) to account for variability within the groups. The data were subjected to statistical evaluation using one-way Analysis of Variance (ANOVA) to determine the overall significance among the different treatment groups. Following ANOVA, Tukey's multiple comparison test was applied to assess intergroup differences. The entire statistical analysis was performed using GraphPad Prism software (version 8). A p-value of < 0.05 was considered statistically significant, indicating a meaningful difference between the control and treated groups in terms of wound contraction, tensile strength, and healing progression.

3. RESULTS

Formulation of WoundRx Cream

The formulation of WoundRx Cream was carried out using natural ingredients known for their wound healing and skin-regenerative properties. The base composition included natural honey, olive oil, and beeswax, combined in equal proportions to provide moisturizing, antimicrobial, and emollient effects. To enhance the therapeutic efficacy, varying concentrations of *Aloe vera* extract were incorporated, resulting in five formulations labeled WRF1 to WRF5. These formulations were prepared using the fusion method, which ensured a uniform and stable cream matrix. The resulting creams were smooth, homogenous, and non-greasy in texture, with acceptable physical appearance and consistency. The increasing concentration of *Aloe vera* imparted a gradual color shift from pale yellow to greenish-yellow, without affecting the stability of the formulation. All batches were successfully prepared and stored for further evaluation, confirming the suitability of this natural composition for wound and burn healing applications.

Fabrication and Characterisation of WoundRx Cream

pH Determination and Homogeneity

The pH evaluation of topical formulations is critical to ensuring patient compatibility and preventing irritation upon application. The prepared WoundRx Cream formulations (WRF1–WRF6) displayed pH values within the acceptable skin-friendly range, varying slightly from 7.36 ± 0.85 (WRF1) to 7.56 ± 0.89 (WRF2). The close proximity of these values to the physiological skin pH (approximately 5.5–7.5) indicated that all formulations were suitable for topical application without causing irritation or discomfort. Additionally, visual inspection confirmed that each formulation exhibited excellent homogeneity, characterized by a smooth, uniform creamy texture, with no visible lumps, granules, or phase separation observed. Such homogeneity ensures consistent therapeutic performance, uniformity in dosage, and enhanced patient compliance.

Spreadability and Extrudability

Spreadability directly influences patient acceptance and ease of application of topical preparations. A higher spreadability value indicates ease of spreading, enhancing the comfort and convenience during topical application. Among all formulations, spreadability improved progressively with increased concentrations of *Aloe vera* extract, ranging from 38.32 ± 1.22 g·cm/sec (WRF1) to a significantly higher 82.74 ± 1.18 g·cm/sec (WRF6). The presence of higher levels of *Aloe vera* extract likely enhanced the gel-like nature, improving consistency and flow. Similarly, the extrudability test demonstrated excellent performance across all formulations, highlighting easy dispensability and patient-friendly attributes. Optimal extrudability ensures that the formulation can be efficiently delivered from the container without excessive pressure, which contributes to patient adherence and satisfaction. Overall, the WRF formulations demonstrated favorable physical and mechanical properties, confirming their suitability as effective and user-friendly topical products for wound and burn management.

Table 2. Characterizations for the WoundRx Cream formulations

Code	pH	Physical Appearance	Spreadability (g cm/sec)	Extrudability
WRF1	7.36± 0.85	Smooth, creamy texture, pale yellow	38.32± 1.22	Excellent
WRF2	7.56 ± 0.89	Smooth, creamy texture, pale	50.48 ± 1.24	Excellent

		yellow		
WRF3	7.48 ± 0.85	Smooth, creamy texture, light greenish-yellow	62.68 ± 1.09	Excellent
WRF4	7.50 ± 0.89	Smooth, creamy texture, light greenish-yellow	69.45 ± 1.26	Excellent
WRF5	7.52± 0.95	Smooth, creamy texture, light greenish-yellow	76.83± 1.19	Excellent
WRF6	7.48 ± 0.77	Smooth, creamy texture, light greenish-yellow	82.74 ± 1.18	Excellent

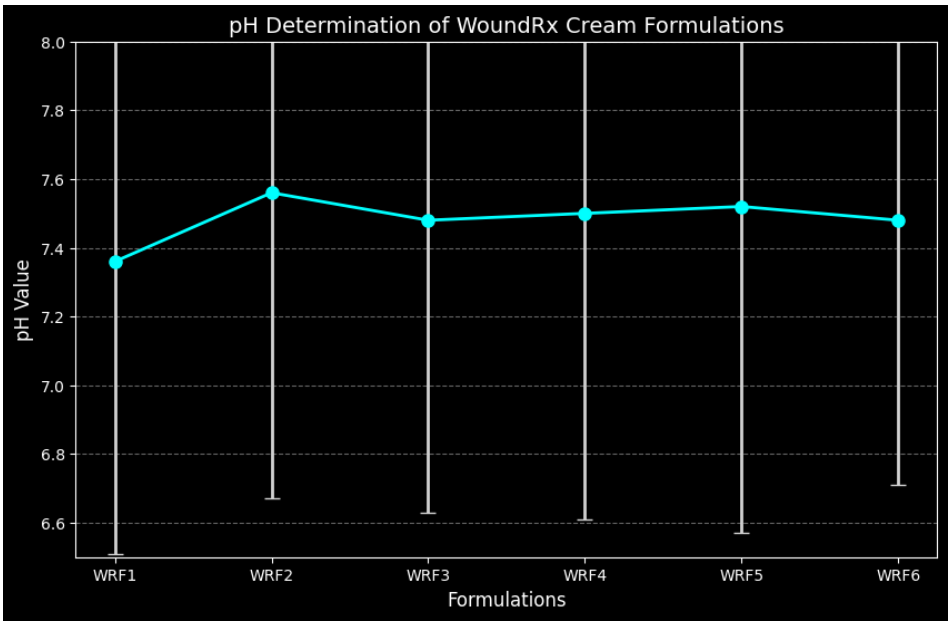

Figure 1. PH Determination of WoundRx Cream Formulations

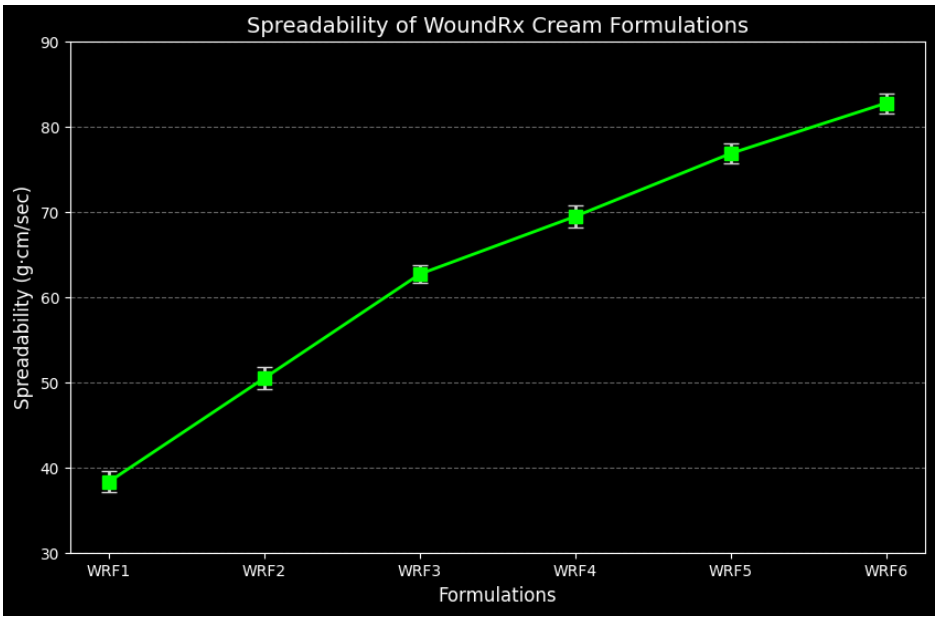

Figure 2. Spreadability of WoundRx Cream Formulations

***In Vivo* Wound Healing Studies**

Excision Wound Model

The results of the excision wound model (Table 3) demonstrated the significant wound healing activity of WoundRx Cream, particularly in diabetic conditions. Initially, on the first day, wound sizes across the three groups were comparable, suggesting uniform wound induction. However, noticeable differences emerged from day 7 onwards. In the diabetic control group, minimal reduction in wound area was observed even after 21 days (23.43 ± 3.70 mm²), indicating impaired wound healing due to diabetes-induced delayed tissue regeneration. Conversely, the group treated with WoundRx Cream under normal conditions (Normal + WoundRx Cream) exhibited remarkable wound contraction by day 14 (7.62 ± 4.13 mm², $p<0.05$) and near-complete healing by day 21 (4.28 ± 2.25 mm², $p<0.05$), confirming the strong regenerative capability of the formulation.

In diabetic rats treated with WoundRx Cream (Diabetic + WoundRx Cream), the wound area significantly reduced to 11.62 ± 4.37 mm² ($p<0.05$) by day 21, highlighting enhanced healing compared to untreated diabetic controls. Although the healing rate in diabetic treated animals was slower compared to normal treated animals, the observed improvement strongly supports the beneficial role of WoundRx Cream ingredients—such as Aloe vera, honey, and olive oil in managing diabetic wounds through their antimicrobial, anti-inflammatory, and collagen-promoting effects. Overall, the findings validate the therapeutic efficacy of WoundRx Cream for accelerating excision wound healing, particularly in diabetic conditions.

Table 3: Effect of WoundRx Cream on Diabetic Rat Excision Wound Healing

Group	1st Day	3rd Day	7th Day	11th Day	14th Day	21st Day
Control Diabetic	30.17 ± 2.44	30.67 ± 1.24	31.17 ± 2.77	30.70 ± 0.93	27.62 ± 6.03	23.43 ± 3.70
Normal + WoundRx Cream	33.95 ± 5.06	32.28 ± 3.91	23.28 ± 5.16	14.62 ± 3.73	7.62 ± 4.13*	4.28 ± 2.25*
Diabetic + WoundRx Cream	32.28 ± 4.85	29.95 ± 5.63	26.62 ± 5.69	17.95 ± 3.34	15.62 ± 5.78	11.62 ± 4.37*

Values are Mean ± SEM (n = 6)

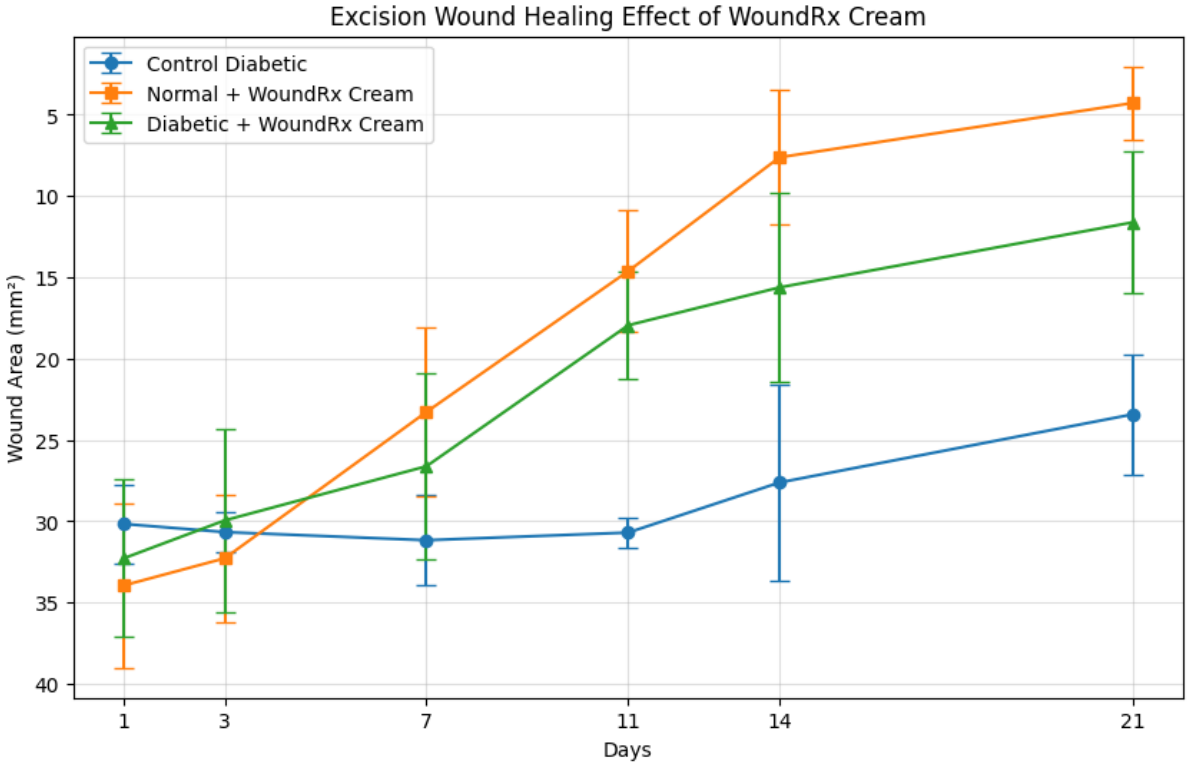

Figure 3. Excision Wound Healing: Effect of WoundRx Cream

Incision Wound Model

The results presented in Table 4 illustrate the wound-healing effectiveness of WoundRx Cream in the incision wound model, especially highlighting its beneficial impact under diabetic conditions. Initially, all groups had comparable wound sizes,

ensuring consistency across the experimental setup. However, significant differences in healing rates emerged notably by day 7 and were highly pronounced by days 14 and 21. The diabetic control group showed limited wound healing, with wound sizes remaining relatively large (24.74 ± 1.35 mm²) even at day 21, indicative of impaired wound healing due to diabetic conditions. In contrast, the application of WoundRx Cream to both normal and diabetic groups significantly accelerated wound healing, with wound areas significantly reduced by day 14 (9.62 ± 1.86 mm² for normal, and 8.95 ± 1.28 mm² for diabetic; $p < 0.05$), and nearly complete closure by day 21 (2.95 ± 0.70 mm² for normal, and 2.63 ± 0.20 mm² for diabetic; $p < 0.05$).

Notably, the diabetic rats treated with WoundRx Cream exhibited wound healing rates comparable to normal rats treated with the cream, demonstrating the formulation's strong potential in mitigating diabetes-associated wound healing delays. The enhanced healing response could be attributed to active phytoconstituents in WoundRx Cream, promoting collagen synthesis, reducing inflammation, and enhancing overall tissue regeneration. These findings confirm the therapeutic value of WoundRx Cream in significantly improving incision wound healing under diabetic conditions.

Table 4: Effect of WoundRx Cream on Diabetic Rat Incision Wound Healing

Group	1st Day	3rd Day	7th Day	14th Day	21st Day
Control Diabetic	27.14 ± 0.35	27.85 ± 1.90	28.46 ± 1.67	27.72 ± 3.53	24.74 ± 1.35
Normal + WoundRx Cream	22.95 ± 3.64	22.62 ± 3.73	18.28 ± 2.21	9.62 ± 1.86*	2.95 ± 0.70*
Diabetic + WoundRx Cream	26.28 ± 3.91	25.62 ± 3.73	22.22 ± 3.01	8.95 ± 1.28*	2.63 ± 0.20*

Values are Mean ± SEM (n = 6

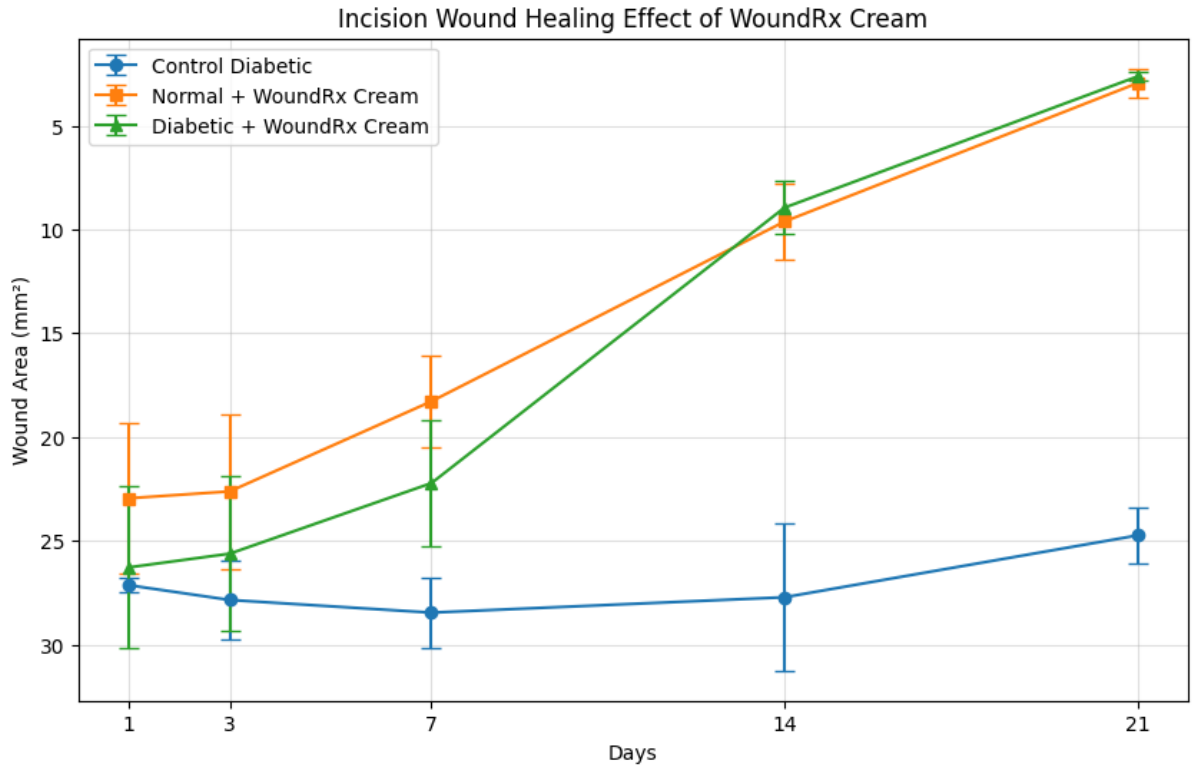

Figure 4. Diabetic Rat Incision Wound Healing: Effect of WoundRx Cream

Burn Wound Model

The burn wound healing study (Table 5) illustrated that WoundRx Cream markedly improved wound recovery in diabetic rats when compared to untreated diabetic animals. Initially, on the first day, wound sizes were similar across all groups, ensuring uniformity in burn injury induction. However, notable differences in healing became evident as early as day 14 and were even more pronounced by days 21 and 31. The diabetic control animals showed minimal wound closure throughout the

experiment, with the wound area remaining relatively large (24.75 ± 4.15 mm²) at day 31, reflecting poor regenerative capacity due to impaired healing pathways associated with diabetes. In contrast, normal rats treated with WoundRx Cream demonstrated significant wound area reduction, with the wound area decreasing substantially to 7.12 ± 3.66 mm² by day 31 ($p<0.05$), highlighting effective healing promoted by the formulation. Diabetic rats treated with WoundRx Cream showed a progressive reduction in wound size, significantly improved (14.12 ± 0.83 mm²) by day 31 compared to untreated diabetic controls ($p<0.05$). Although the healing rate in diabetic treated rats was slower than normal treated rats, the results confirmed the efficacy of WoundRx Cream in enhancing wound contraction even under diabetic conditions. The observed improvement is likely attributable to the anti-inflammatory, antimicrobial, and regenerative bioactive components present in the formulation, particularly *Aloe vera* gel and honey. Overall, the data suggest that WoundRx Cream significantly promotes burn wound healing and supports its therapeutic potential in diabetic burn wound management.

Table 5: Effect of WoundRx Cream on Diabetic Rat Burn Wound Healing

Group	1st Day	3rd Day	7th Day	14th Day	21st Day	31st Day
Control Diabetic	28.25 ± 1.23	28.74 ± 0.16	29.33 ± 1.23	29.77 ± 2.25	28.07 ± 2.33	24.75 ± 4.15
Normal + WoundRx Cream	32.12 ± 2.25	28.12 ± 2.25	27.12 ± 0.83	24.62 ± 7.20	13.12 ± 3.66	7.12 ± 3.66*
Diabetic + WoundRx Cream	25.62 ± 1.54	27.62 ± 0.13	26.12 ± 0.83	24.12 ± 3.63	20.62 ± 1.54	14.12 ± 0.83*

Values are Mean ± SEM (n = 6)

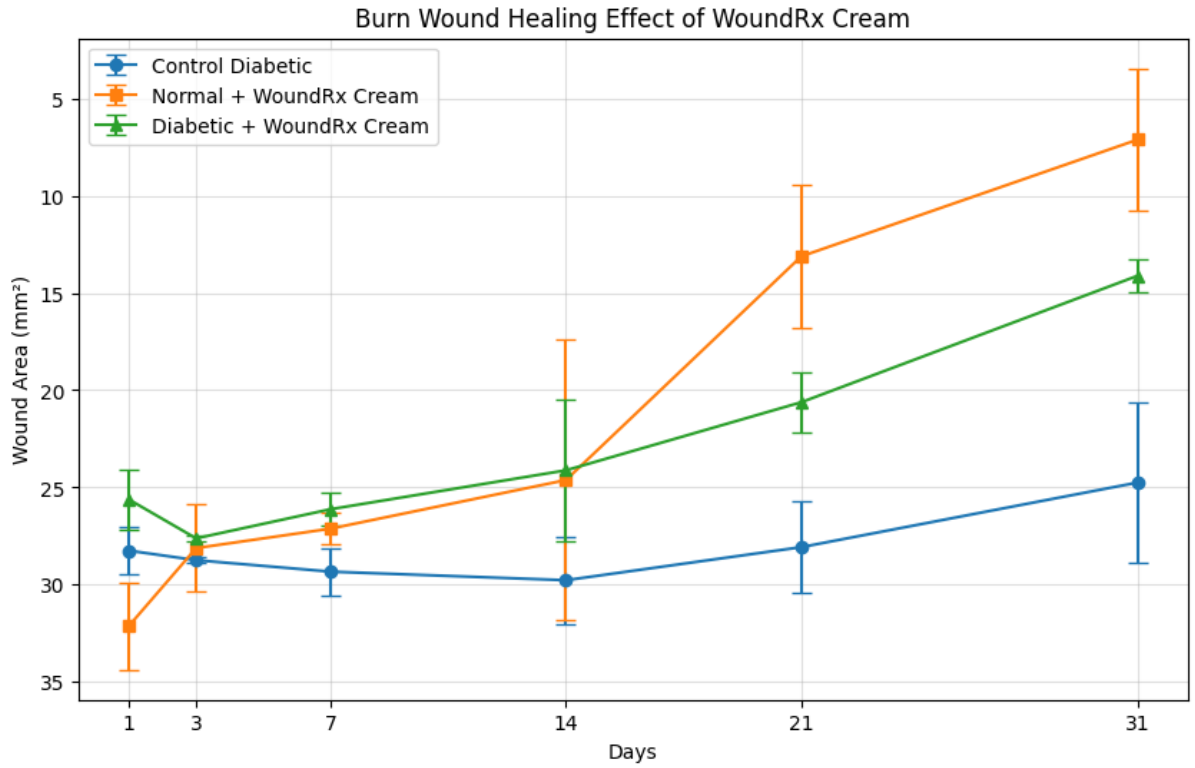

Figure 5. Diabetic Rat Burn Wound Healing: Effect of WoundRx Cream

4. CONCLUSION

The present study successfully formulated and evaluated WoundRx Cream, demonstrating significant wound and burn healing properties in diabetic Wistar rats. The cream, composed of honey, olive oil, beeswax, and *Aloe vera* extract, exhibited optimal physical characteristics, including desirable pH, high spreadability, excellent extrudability, and consistent homogeneity. In-vivo studies across excision, incision, and burn wound models revealed substantial healing efficacy,

significantly reducing wound area and improving healing outcomes in diabetic conditions compared to untreated diabetic controls. The diabetic rats treated with WoundRx Cream showed near-complete wound closure, reaffirming the formulation's potent regenerative activity. These outcomes likely stem from the synergistic therapeutic benefits of its natural ingredients, particularly the anti-inflammatory and antimicrobial actions of honey and *Aloe vera*, which facilitate collagen synthesis and tissue regeneration. The study underscores the therapeutic value of WoundRx Cream as an effective, natural alternative in managing diabetic wounds and burn injuries, presenting a promising approach to overcoming the impaired wound-healing challenges commonly associated with diabetes. Future research should focus on clinical trials and molecular mechanism exploration to further validate and enhance the therapeutic effectiveness and clinical applicability of WoundRx Cream.

REFERENCES

[1] Aiyalu, R., Govindarjan, A., & Ramasamy, A. (2016). Formulation and evaluation of topical herbal gel for the treatment of arthritis in animal model. *Brazilian Journal of Pharmaceutical Sciences*, *52*, 493-507.

[2] Akbarzadeh, A., Norouzian, D., Mehrabi, M. R., Jamshidi, S. H., Farhangi, A., Verdi, A. A., Mofidian, S. M. A., & Rad, B. L. (2007). Induction of diabetes by streptozotocin in rats. *Indian Journal of Clinical Biochemistry*, *22*, 60-64.

[3] Al-Waili, N. S. (2003). Topical application of natural honey, beeswax and olive oil mixture for atopic dermatitis or psoriasis: partially controlled, single-blinded study. *Complementary therapies in medicine*, *11*(4), 226-234.

[4] Dilworth, L., Facey, A., & Omoruyi, F. (2021). Diabetes mellitus and its metabolic complications: the role of adipose tissues. *International journal of molecular sciences*, *22*(14), 7644.

[5] Elkordy, A. A., Haj-Ahmad, R. R., Awaad, A. S., & Zaki, R. M. (2021). An overview on natural product drug formulations from conventional medicines to nanomedicines: Past, present and future. *Journal of Drug Delivery Science and Technology*, *63*, 102459.

[6] Ghasemi, A., & Jeddi, S. (2023). Streptozotocin as a tool for induction of rat models of diabetes: A practical guide. *EXCLI journal*, *22*, 274.

[7] Jain, B. D. (2007). Formulation Development And Evaluation Of Fluconazole Gel In Various Polymer Basesformulation Development And Evaluation Of Fluconazole Gel In Various Polymer BaseS. *Asian Journal of Pharmaceutics (AJP)*, *1*(1).

[8] Loiacono, M., Padovano, L., Malacarne, M. C., Conti, S., & Caruso, E. (2025). Formulation and evaluation of alternative to beeswax for vegan lipsticks. *Int J Cosmet Sci*.

[9] Nagesh, M. R., Vijayakumar, N., & Bhavani, K. (2020). A review on diabetes mellitus-an annihilatory metabolic disorder. *Journal of Pharmaceutical Sciences and Research*, *12*(2), 232-235.

[10] Nayak, S. H., Nakhat, P. D., & Yeole, P. G. (2005). Development and evaluation of cosmeceutical hair styling gels of ketoconazole. *Indian Journal of Pharmaceutical Sciences*, *67*(2), 231.

[11] Ogidi, C. O., Ojo, A. E., Ajayi-Moses, O. B., Aladejana, O. M., Thonda, O. A., & Akinyele, B. J. (2021). Synergistic antifungal evaluation of over-the-counter antifungal creams with turmeric essential oil or Aloe vera gel against pathogenic fungi. *BMC Complementary Medicine and Therapies*, *21*, 1-12.

[12] Patil, M. V. K., Kandhare, A. D., & Bhise, S. D. (2012). Pharmacological evaluation of ethanolic extract of Daucus carota Linn root formulated cream on wound healing using excision and incision wound model. *Asian Pacific Journal of Tropical Biomedicine*, *2*(2), S646-S655.

[13] Queiroz, M. B. R., Marcelino, N. B., Ribeiro, M. V., Espindola, L. S., Cunha, F. R., & Silva, M. V. d. (2009). Development of gel with Matricaria recutita L. extract for topic application and evaluation of physical-chemical stability and toxicity. *Lat. Am. J. Pharm*, *28*(4), 574-579.

[14] Tekiki, A., Charfi, I., Helal, R., & Bornaz, S. (2022). Formulation and optimization of new spreads based on olive oil and honey: A response surface methodology box-benken design. *Food and Nutrition Sciences*, *13*(4), 358-372.