



Research Article

Assessment of the Therapeutic Potential of Snail Mucin on Excision Wound Healing in Wistar Albino Rats

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ARTICLE INFO	ABSTRACT
Date of submission: 23-06-2025 Date of Revision: 25-07-2025 Date of acceptance: 30-07-2025 Key Words: Wound healing, snail mucin, epithelialization period, excision wound, Epithelialization	Wound healing is a multifaceted process involving haemostasis, inflammation, proliferation, and remodelling, all orchestrated by cellular and molecular mediators. Impairment due to systemic or local factors often results in chronic non-healing wounds, posing a major clinical and socioeconomic burden. The search for safe, affordable, and effective alternatives has directed attention toward natural agents, including snail mucin, a secretion rich in glycoproteins, allantoin, hyaluronic acid, and antimicrobial peptides with proven regenerative and protective properties. This study evaluated the wound healing activity of snail mucin ointment in an excision wound model using albino rats. Animals were divided into four groups: control (ointment base), standard (5% povidone-iodine), 5% snail mucin, and 10% snail mucin. Parameters assessed included wound contraction, epithelialization period, and safety. Results demonstrated that 10% snail mucin significantly accelerated wound contraction, achieving ~95% closure by day 16 and complete healing by day 20, outperforming the standard treatment. Epithelialization time was also reduced without signs of dermal toxicity or systemic adverse effects. The findings indicate that snail mucin exerts potent wound healing effects, likely mediated through enhanced fibroblast activity, collagen synthesis, angiogenesis, and anti-inflammatory actions. These results highlight snail mucin as a promising natural therapeutic for wound management, warranting further mechanistic and clinical investigations.

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INTRODUCTION

Wound healing is a complex and dynamic biological process that restores the integrity of injured tissue following physical, chemical, or microbial insult. It involves a highly coordinated cascade of overlapping phases, including homeostasis, inflammation, proliferation, and remodeling. Each stage requires the participation of cellular and molecular mediators such as platelets, neutrophils, macrophages, fibroblasts, keratinocytes, cytokines, growth factors, and extracellular matrix proteins.[1,2] Although healing is an innate physiological process, it can often be delayed or impaired by systemic conditions such as diabetes mellitus, infections, nutritional deficiencies, or local factors including repeated trauma and inadequate blood supply. Chronic non-healing wounds remain a major clinical and socioeconomic burden worldwide, contributing to increased morbidity, prolonged hospitalization, and high treatment costs. Current wound management strategies range from conventional dressings to advanced therapeutic interventions such as growth factor applications, bio-engineered skin substitutes, and negative pressure wound therapy. While effective, many of these modalities are expensive, inaccessible in resource-limited settings, or associated with complications such as infection and

hypersensitivity. Consequently, the search for novel, affordable, and safe wound healing agents remains an important area of biomedical research [4]. Natural products and animal-derived substances have gained increasing attention due to their historical use in traditional medicine and their rich repertoire of bioactive molecules [5,6].

Among these natural substances, snail mucin (also called snail slime or secretion) has emerged as a unique biopolymer with promising dermatological and regenerative properties. It is secreted by land snails such as *Achatina fulica* and *Helix aspersa* to protect their skin and facilitate locomotion [7]. Chemically, snail mucin is a complex mixture of glycoproteins, glycosaminoglycans, allantoin, hyaluronic acid, collagen, elastin, antimicrobial peptides, and trace elements. These constituents confer multiple biological activities including moisturizing, antioxidant, antimicrobial, anti-inflammatory, and tissue-regenerative effects. Historically, snail secretions have been used in folk medicine for skin ailments, burns, and ulcers [8]. In recent years, snail mucin has gained commercial popularity in the cosmetic industry, particularly in formulations marketed for anti-aging, scar reduction, and skin repair. Experimental studies suggest that snail mucin stimulates fibroblast proliferation,

enhances extracellular matrix deposition, promotes angiogenesis, and accelerates epithelialization—hallmarks of effective wound healing. For example, allantoin is known to stimulate keratinocyte migration, while glycosaminoglycans improve hydration and elasticity of skin tissue. Moreover, its antimicrobial peptides may help prevent wound infections, further supporting its use as a wound healing agent. Despite these promising findings, the scientific validation of snail mucin in standardized animal wound models is still limited. Only a few studies have systematically evaluated its efficacy compared to standard wound healing agents such as povidone-iodine or silver sulfadiazine. Most available reports are fragmented, focus on cosmetic outcomes, or lack mechanistic insights. Therefore, rigorous preclinical investigations are required to establish the pharmacological basis of snail mucin in wound healing, particularly in well-established experimental models such as the excision wound model in albino rats. Therefore, the present study was designed to evaluate the wound healing activity of snail mucin ointment using the excision wound model in albino rats [7-9]

MATERIALS AND METHODS

Experimental Animals

Healthy adult Wistar albino rats of either sex, weighing 150–180 g, were procured

from the institutional animal house of Royal College of Pharmacy and Health Sciences (RCPHS), Berhampur, Odisha, India. The animals were housed in polypropylene cages (three per cage) with sterilized rice husk bedding. Environmental conditions were maintained at 22 ± 2 °C temperature, $55 \pm 5\%$ relative humidity, and a 12:12 h light–dark cycle. Rats were provided with standard laboratory diet made in-house as recommended by National Institute of Nutrition (NIN), Hyderabad, and pure drinking water ad libitum. They were acclimatized to the laboratory environment for 7 days prior to the study. The study protocol was reviewed and approved by the Institutional Animal Ethics Committee of RCPHS, Berhampur, Odisha, India (registration number 1018/C/06/CPCSEA). All animal handling and experimental procedures were conducted in accordance with the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Government of India guidelines.

Collection and Extraction of Snail Mucin

Fresh mucin was obtained from adult giant African land snails (*Achatina fulica*). The extraction was performed using a gentle stimulation method to avoid harm to the animals. Briefly, live snails were washed with sterile distilled water and allowed to

secrete mucin by gentle mechanical stimulation of the foot region. The secreted mucin was collected in sterile glass containers and immediately filtered through muslin cloth to remove debris. The crude mucin was then centrifuged at 5000 rpm for 15 minutes, and the supernatant was lyophilized to obtain dry mucin powder. The dried mucin was stored in airtight sterile containers under refrigeration until further use.

Preparation of Snail Mucin Ointment

Snail mucin ointment was prepared using the simple ointment base (British Pharmacopoeia, 1968) consisting of hard paraffin (5%), cetostearyl alcohol (5%), white soft paraffin (85%), and wool fat (5%). The ointment base was melted and homogenized by the fusion method. Snail mucin powder was incorporated into the base to prepare 10% w/w snail mucin ointment [10].

Acute dermal toxicity test

The skin irritation test with 10% w/w snail mucin ointment was conducted on rats. Animals showing normal skin texture were housed in a cage. Three rats were employed for this test and their skin was shaved on the dorsal side, each about 400 mm² areas 24 h before application of the sample. A 10% w/w snail mucin ointment was applied uniformly over the shaved

area for 4 h. the test materials were removed and observed for an adverse skin reaction/ irritation within 24 h. Then, they were further observed daily upto 14 days for any signs of dermal toxicity [11].

Excision Wound Model

The excision wound model was employed as described by Morton and Malone (1972) with slight modifications. The rats were anesthetized with ketamine (50mg/kg, i.p.). The dorsal fur region was shaved and disinfected with 70% ethanol and the anticipated area of the wound to be created was outlined on the back of the animal with marker. The excision wound was made by cutting a circular area of ≈ 300 mm² area and 2 mm depth of skin from the depilated area along the marking using toothed forceps, a surgical blade and pointed scissors. Hemostasis was achieved by blotting the wound with sterile gauze. The wound was left undressed to open environment. No systemic antibiotics or analgesics were administered to avoid interference with the wound healing process [12].

Experimental Design and Grouping

The animals were randomly divided into four groups (n = 6 per group) as per the table 1. Topical applications were performed once daily until complete healing,

Table 1: Different treatment groups

Group	Treatment	Formulation applied once daily
Group I	Control	Ointment base only
Group II	Standard	5% Povidone-iodine ointment
Group III	Test-1	5% Snail mucin ointment
Group IV	Test-2	10% Snail mucin ointment

Parameters Evaluated [13]**a. Wound Contraction (%)**

The progressive reduction in wound area was measured on days 0, 4, 8, 12, 16, and 20 using transparent tracing paper and graph paper method. Wound contraction was calculated as percentage reduction in wound area. Wound contraction (%) was calculated using the relation:

$$\text{Wound contraction (\%)} = \frac{[(WA_0 - WAt) / WA_0] \times 100}{1}$$

Where:

WA₀ = the wound area on day zero,

WAt = the wound area on day t.

b. Epithelialization Period

Defined as the number of days required for complete closure of the wound surface

with new epithelium and for the scab/eschar to fall off naturally without leaving a raw wound.

c. Safety/Tolerability

Animals were observed daily for signs of local irritation (erythema, edema) and systemic toxicity (loss of body weight, abnormal behavior).

Statistical Analysis

All data were expressed as mean $\pm$ standard error of mean (SEM). Statistical analysis was performed using one-way analysis of variance (ANOVA) followed by Tukey's multiple comparison test. A value of $p < 0.05$ was considered statistically significant.

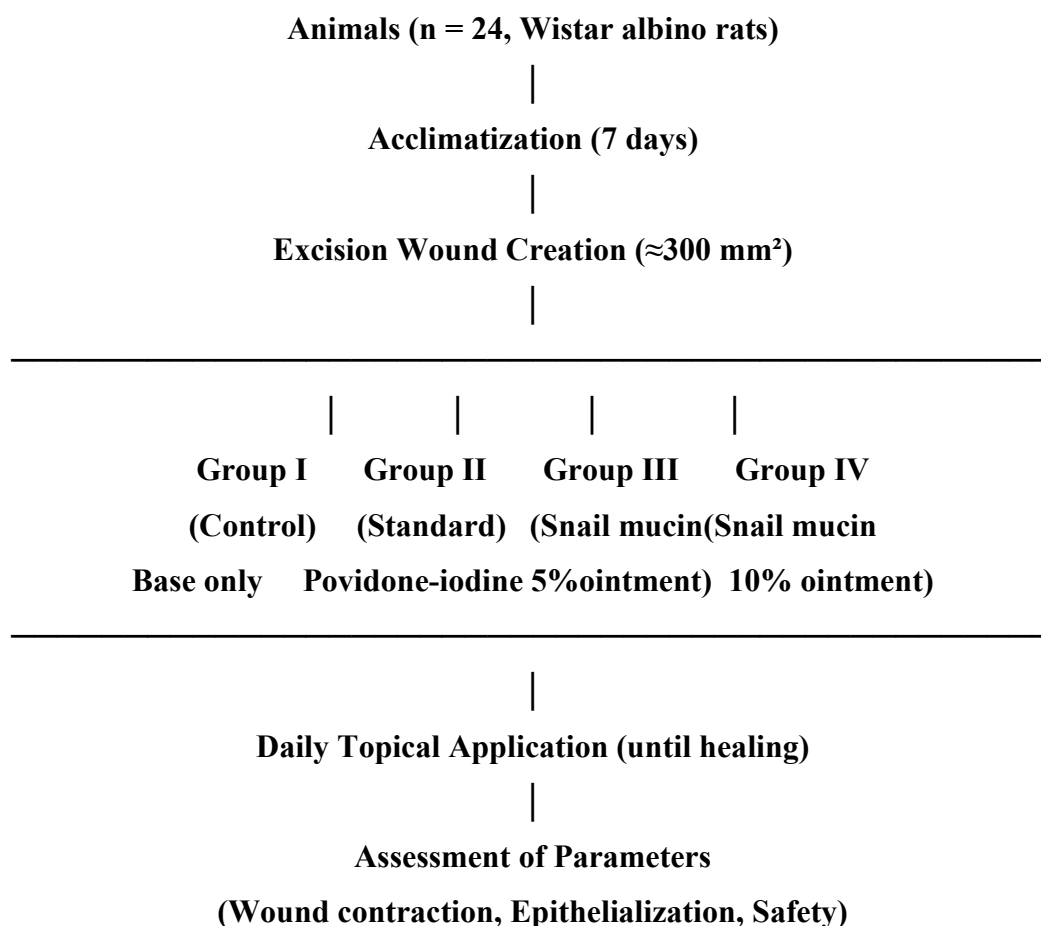


Figure 1. Experimental Design Schematic (Placeholder)

RESULTS

Acute dermal toxicity test

The purpose of this study was to assess the skin irritation potential of 10% w/w snail mucin ointment after a single topical application. During the entire period of experimentation, no animal showed any sign of dermal toxicity.

Excision Wound Model

a. Effect on Wound Contraction

The rate of wound contraction was progressively monitored on days 0, 4, 8, 12, 16, and 20. In the control group, wound contraction was relatively slow, reaching ~72% closure by day 20. The standard group (5% povidone-

iodine) showed enhanced healing with ~93% contraction by day 20. Rats treated with snail mucin 5% exhibited significantly higher wound contraction than control at all time points ($p < 0.05$) and with ~85% contraction by day 20. The snail mucin 10% group demonstrated the fastest contraction, with ~95% wound closure by day 16 and complete healing by day 20 (table 2).

b. Effect on Epithelialization Period

The epithelialization period, defined as the time taken for the eschar to fall off without leaving a raw wound, was markedly reduced in snail mucin-treated groups compared to control (table 3).

Table 2: Percentage wound contraction at different time intervals (mean $\pm$ SEM, n = 6)

Day	Control (Base)	Standard (Povidone-iodine 5%)	Snail mucin 5%	Snail mucin 10%
0	0	0	0	0
4	12.78 $\pm$ 0.92	20.56 $\pm$ 1.22*	18.38 $\pm$ 1.06*	22.98 $\pm$ 1.02*
8	28.56 $\pm$ 1.96	46.44 $\pm$ 2.63*	40.06 $\pm$ 2.14*	51.19 $\pm$ 2.52*
12	48.88 $\pm$ 2.0	71.76 $\pm$ 2.34*	65.38 $\pm$ 2.56*	79.62 $\pm$ 2.78*
16	63.98 $\pm$ 2.9	86.24 $\pm$ 3.22*	78.82 $\pm$ 3.45*	94.66 $\pm$ 3.16*
20	72.46 $\pm$ 3.06	92.78 $\pm$ 2.86*	85.56 $\pm$ 2.72*	100.0 $\pm$ 0.12*

*Significant vs. control, $p < 0.05$ **Table 3. Epithelialization period (days) in different groups (mean $\pm$ SEM, n = 6)**

Group	Epithelialization Period (days)
Control (Base)	25.22 $\pm$ 2.74
Standard (Povidone)	19.78 $\pm$ 1.28*
Snail mucin 5%	21.46 $\pm$ 1.54*
Snail mucin 10%	18.52 $\pm$ 0.96*

*Significant vs. control, $p < 0.05$

c.Safety/Tolerability study by General Observations

All animals tolerated the excision procedure and topical treatments well. No signs of systemic toxicity, weight loss, or local irritation (erythema, edema) were observed in any group throughout the study. Wound areas remained free from overt infection, indicating good tolerability of snail mucin formulations.

DISCUSSION

Wound healing is a complex and dynamic process involving a sequence of cellular and biochemical events that restore the structural and functional integrity of

injured tissue. The present study evaluated the wound healing efficacy of snail mucin in an excision wound model using albino rats. Our findings demonstrate that topical application of snail mucin, particularly at a 10% concentration, significantly accelerated wound contraction, shortened the epithelialization period, and enhanced histopathological features of tissue repair compared to untreated controls and the standard reference drug, povidone-iodine ointment. These observations suggest that snail mucin possesses potent wound healing activity, likely mediated by its bioactive components such as

glycoproteins, allantoin, glycolic acid, and hyaluronic acid, which are known to influence cell proliferation, angiogenesis, collagen deposition, and extracellular matrix remodeling.

ANOVA followed by Tukey's post hoc test showed statistically significant differences ($p < 0.05$) in wound contraction and epithelialization between snail mucin-treated groups and control. The 10% snail mucin group performed significantly better than the standard group. The wound healing activity of snail mucin may be attributed to several interrelated mechanisms, supported by its chemical composition:

Stimulation of fibroblast proliferation: Snail mucin contains glycoproteins and peptides that promote fibroblast migration and proliferation. Fibroblasts are central to collagen synthesis and extracellular matrix deposition, both critical for tissue remodeling.

Collagen synthesis and remodeling: Collagen is essential for tensile strength in newly formed tissue. Studies have demonstrated that mucin upregulates collagen I and III synthesis, accelerating maturation of the wound matrix.

Anti-inflammatory effects: The presence of antioxidants, including superoxide dismutase and glutathione-like peptides in mucin, may help reduce oxidative stress and inflammatory cell infiltration, thereby

expediting the proliferative phase of healing.

Angiogenesis stimulation: Hyaluronic acid and glycolic acid in snail mucin promote endothelial cell migration and capillary formation, ensuring an adequate blood supply for regenerating tissue.

Moisturization and barrier restoration: The hydrating properties of mucin maintain an optimal moist wound environment, known to facilitate keratinocyte migration and re-epithelialization.

These multifaceted actions position snail mucin as a promising therapeutic agent for dermal regeneration.

Implications for clinical use

The findings from this study support the potential application of snail mucin as a topical wound healing agent. Its natural origin, biocompatibility, and multiple regenerative properties make it suitable for Chronic wound management (e.g., diabetic ulcers, pressure sores), Acute wound care (burns, excisions, traumatic injuries), post-surgical wound healing to minimize scarring. Additionally, snail mucin's presence in cosmetic formulations underscores its safety profile and consumer acceptance. Translating these preclinical findings into clinical settings could offer a natural and effective alternative or adjunct to conventional wound therapies.

Future perspectives

Further research should focus on conducting controlled in vivo studies to confirm these findings, exploring the molecular pathways of mucin action, particularly its effect on VEGF, TGF- β , and collagen gene expression, developing mucin-based nanocarriers for sustained drug delivery and performing clinical trials to assess safety and efficacy in human subjects. Such studies will bridge the gap between preclinical evidence and clinical application, paving the way for snail mucin-based wound healing products.

CONCLUSION

This study suggests that snail mucin possesses significant wound healing activity, as evidenced by enhanced wound contraction, shortened epithelialization period, and favorable histopathological features in an excision wound model. The effects were dose-dependent, with the 10% mucin formulation outperforming the standard povidone-iodine treatment. The underlying mechanisms may involve stimulation of fibroblast proliferation, collagen synthesis, angiogenesis, and anti-inflammatory activity, highlighting mucin's multifaceted role in tissue regeneration. Despite the need for experimental validation, these findings underscore the potential of snail mucin as a natural, effective, and safe therapeutic agent for wound management.

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