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Effect of *Gmelina arborea* Roxb. leaves on wound healing in rats

A. Shirwaikar[1]*, S. Ghosh[1], Padma G.M.Rao[2],

1. Department of Pharmacognosy, College of Pharmaceutical Sciences,
2. Department of Pharmacology, Kasturba Medical College,
Manipal - 576 119, Dakshina Kannada Karnataka, India.

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Abstract

Objective: To study the effect of the alcoholic extract of the dried leaves powder of *Gmelina arborea* on wound healing. Materials and method: The alcoholic extract was studied at dose level of 200 mg/kg body weight using incision, excision and dead space wound models in rats. Results: Significant increase in wound contraction rate, skin breaking strength, granuloma breaking strength, hydroxyproline content and dry granuloma weight and decrease in epithelization period was observed. The prohealing actions seem to be due to increased collagen deposition as well as better alignment and maturation. Conclusion: From the results it may be concluded that the plant *Gmelina arborea* is endowed with significant wound healing activity, thereby justifying its use in the traditional medicine.

Keywords: *Gmelina arborea*, incision wound model, excision wound model, dead space wound model.

1. Introduction

Gmelina arborea Roxb. (Verbenaceae) belongs to a genus of trees and shrubs distributed chiefly in South East Asia, tropical Australia and tropical Costa Rica. [1,2] It is a moderate sized unarmed deciduous tree, reaching upto 60ft high. [3] The root of this plant has been used in traditional Indian systems of medicines as a demulscent, stomachic, bitter tonic, refrigerant, laxative, and galactagogue. The tender leaves are used as demulscent, in head ache, fevers, gonorrhea, cough etc. The whole plant is used in snake bite and scorpion sting throughout India [3]. It has also been reported to have anti-inflammatory activity [4], hypoglycaemic [1] and anti-viral activities against Ranikhet disease virus [1]. Since inflammation precedes the process of healing, drugs affecting inflammation are known to slow down the healing. This study was undertaken to assess the effect of this indigenous herb on different parameters related to wound healing in rats.

* Corresponding Author
E-mail: annie.shirwaikar@cops.manipal.edu

2. Materials and methods

2.1 Plant material

Gmelina arborea leaves were collected in September 1999 and identified by Dr. Gopalakrishna Bhat, Department of Botany, Sri Poorna Prajna College, Udupi. A voucher specimen PP502 has been preserved in the College of Pharmaceutical Sciences, Manipal.

2.2 Preparation of drug extract

Shade dried, powdered leaves (300 g) were soxhlet extracted with 90% ethanol. Solvent elimination under reduced pressure afforded a solid greasy residue. (Yield : 32.8%) Preliminary phytochemical screening [5,6] of this extract gave positive results for flavonoid glycosides, steroids, fatty acids, fixed oils and waxes and a small quanitity of alkaloids. For wound healing studies, the extract was prepared in the form of an emulsion using gum acacia.

2.3 Animals

Healthy albino rats of either sex, bred locally, weighing between 150 – 250 g were used. They were housed individually under standard environmental conditions, fed with pellet rodent diet and water *ad libitum.*

2.4 Wound healing studies

Animals were divided into two groups (control and test) of eight animals each. They were orally dosed with emulsion of the ethanolic extract in 1% acacia (200 mg/kg body weight) daily for 10 consecutive days in the incision and dead space wound model, and for 21 days in the excision wound model. The control group was treated with 1% acacia emulsion.

2.5 Wound models

2.5.1 Incision wounds

Two 6 cm long paravertebral incisions were made through full thickness of the skin on either side of the vertebral column of the rat. [7] Wounds were closed with interrupted sutures, 1 cm apart. The sutures were removed on the 7th day. Wound breaking strength was measured on the 10th day by the method of Lee. [8]

2.5.2 Excision wounds

A circular piece of full thickness (approximately 500 mm^2) was cut off from a predetermined area on the back of the rat. [9] The wounds were traced on 1 mm^2 graph paper on the day of wounding and subsequently on alternate days until healing was complete. Changes in wound area were calculated. Reduction in the wound area was expressed as percentage of the original wound size. The number of days required for falling of the eschar without any residual raw wound, gave the period of epithelization.

2.5.3 Dead space wounds

Dead space wounds were made by implantation of polypropylene tube (0.5 cm X 2.5 cm) beneath the dorsal paravertebral skin. On the 10th day, the granuloma tissue formed on the dead space wound was dissected and tensile strength was determined. The excised tissue was cut into 2 approximately equal halves. [8] One half of the granuloma tissue was dried in an oven at 60°C and the dry weight was noted. Acid hydrolysate of the dry tissue was used for the determination of hydroxyproline. [10] The other part of the tissue was kept in 10% formalin solution for histopathological studies to evaluate the effect of the extract on collagen formation.

2.6 Statistical analysis

Results, expressed as mean ± S.E. were evaluated for statistical significance by unpaired Student's *t* - test. Values of $p < 0.05$ were considered statistically significant.

3. Results

Acute toxicity studies of the ethanolic extract of the leaves of *G. arborea* did not exhibit any

sign of toxicity upto a maximum dose of 2 g/kg body weight.

Tables 1a and 1b depict the effect of the ethanolic extract of *G.arborea* on various wound healing parameters using different wound models. Pharmacological studies indicated a significant increase in the tensile strength of drug treated group in incision wound model when observed on the 10th post wounding day (Table 1a). Tensile strength of the granuloma tissue, weight of this tissue and hydroxyproline content also were significantly increased in drug treated *vs.* control group in dead space wound.

From the histopathological study it was observed that collagen formation was more in drug treated as compared to control.

Studies using the excision wound model showed significant decrease in the epithelization period. Epithelization was found to be enhanced significantly ($p<0.001$) by the ethanolic extract of *Gmelina arborea* as evidenced by the shorter period required for eschar dropping (i.e. 15.63 ± 0.263 days) as compared to the control (i.e. 21.75 ± 0.25). The extract also facilitated the wound contraction. (Table 1b).

Table 1a.
Effects of the ethanolic extract of *Gmelina arborea* on wound healing in incision and dead space wound models (Mean ± S.E.)

Wound model	**Incision**	**Dead Space**		
Parameters studied	**Breaking strength (g)**	**Granuloma weight (g/100g b.w)**	**Breaking strength (g)**	**Hydroxyproline (µg/g)**
Control	303.92 ± 7.88	0.017 ± 0.0035	255.00 ± 8.64	721.4 ± 108.59
Ethanolic extract of *G. arborea*	536.25 ± 7.15*	0.140 ± 0.0214**	514.72 ± 19.71*	1235.9 ± 74.99**

Data (Mean ± S.E.) n = 8. *p<0.001; **p<0.005.

Table 1b.
Effects of ethanolic extract of *Gmelina arborea* on excision wound model

		Control	**Ethanolic extract *G. arborea***
Epithelization period (days)		21.75 ± 0.25	15.63 ± 0.26*
% of wound	2	3.5 ± 3.8	35.18 ± 4.82**
contraction by day	4	4.0 ± 3.7	49.14 ± 5.19*
	6	11.0 ± 4.6	55.38 ± 3.05*
	8	24.2 ± 6.4	80.63 ± 1.5*
	10	43.1 ± 3.4	90.34 ± 1.28*
	12	59.6 ± 2.5	93.49 ± 1.13*
	14	68.5 ± 6.78	97.49 ± 0.53**
	16	75.61 ± 5.22	
	18	79.2 ± 8.13	
	20	83.52 ± 5.81	
	22	99.79 ± 1.26	

Data (Mean ± S. E.) n = 8. *p<0.001; **p<0.005 *vs.* Control.

4. Discussion

Normal healing involves an initial inflammatory phase followed by fibroblast proliferation, formation of collagen fibers and shrinking and drying of the scar. These phases are concurrent but independent of each other. *G.arborea* leaves are reported to have anti-inflammatory activity, yet the findings of the present study showed that the indigenous drug had a definite prohealing action.

This is demonstrated by significant increase in (1) wound contraction rate and enhanced epithelization. (2) skin breaking strength. (3) hydroxyproline content, reflecting increased collagen levels as further supported from histopathological reports. (4) granuloma breaking strength, indicating better maturation of collagen by increased cross linking. (5) dry granuloma weight, indicating higher protein content.

Thus it may be concluded that the plant *Gmelina arborea* is endowed with significant wound healing activity, thereby justifying its use in the traditional system of medicine.

References

1. Satyavati GV, Raina MK, Sharma M. (1976) *Medicinal Plants of India,* Vol. 1, Indian Council of Medical Research: New Delhi; 441-444

2. Chopra RN, Nayar SL, Chopra IC. (1986) *Glossary of Indian Medicinal Plants,* Publication and Information Directorate, CSIR: New Delhi; 126

3. Dassanayake MD, Fosberg FR. *Revised Handbook to The Flora of Ceylon,* Vol. 4, 388-394

4. Agrawal VK, Gambhir SS, Wahi AK. (1994) *Indian J. Nat. Prod.* 10(1) : 14-15.

5. Harborne JB. (1984) *Phytochemical Methods*, 2nd edn Chapman and Hall: London.

6. Kokate CK. (1991) *Practical Pharmacognosy,* 3rd edn. Vallabh Prakashan: India; 107-121

7. Ehrlich HP, Hunt TK. (1969) *Ann. Surg.* 170 : 203.

8. Lee KH. (1968) *J. Pharmacol. Sci.* 57 : 1042.

9. Morton JJP, Malone HH. (1972) *Arch. Int. Pharmacodyn. Ther.* 196 : 117.

10. Neuman RE, Logan MA. (1950) *J. Biol. Chem*. 186 : 83.