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EVALUATION OF WOUND HEALING ACTIVITY OF ACALYPHA FRUTICOSA **ROOT EXTRACT IN RATS**

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ABSTRACT:

A wound is a breakage in tissue continuity that can be produced by physical, chemical and thermal damage. It is a generally called as physical injury that cause opening and breaking of skin. Healing of chronic cutaneous wound is a big problem and it involves the restoration of continuity after wounding. The Primary objective of this present study is to evaluate the wound healing activity of Ethanolic extract of roots of Acalypha fruticosa. In this perspective, Ethanolic extract of roots of Acalypha fruticosa was taken and excision wound model was used for the screening of wound healing activity. Mupirocin was taken as standard drug. The percentage of wound contraction and period of epithelization was determined. The wound healing activity of Ethanolic extract of Acalypha fruticosa was showed significant wound healing activity with doses 100 and 200 mg/kg. An acceleration of wound healing in Acalypha fruticosa treated rats as compared to the control. Further detailed phytochemical investigations are required to identify the phytoconstituents responsible for the wound healing activity.

Key words: Wound healing, Acalypha fruticosa, Excision, Mupirocin.

1. INTRODUCTION:

Medicinal plants constitute a source of raw materials for both traditional systems of medicine (e.g. Ayurvedic, Chinese, Unani, Homeopathy, and Siddha) and modern medicine. Now days, plant materials are employed throughout the industrialized and developing world as home remedies, overthe-counter drugs, and ingredients for the pharmaceutical industry. As such, they represent a substantial proportion of the global drug market. Most rural populations, especially in the developing world, depend on medicinal herbs as their main source of primary health care. Although most medicinal herbs are not in their natural state fit for administration but preparations suitable for administration are made according to

pharmacopeial directions. The therapeutic potential of a herbal drugs depends on its form: whether parts of a plant or simple extracts or isolated active constituents. Now a day's many synthetic and herbal medicines are available in the market which is used to treat various ailments [1].

A wound is a breakage in tissue continuity that can be produced by physical, chemical and thermal damage. It is a generally called as physical injury that cause opening and breaking of skin. Healing of chronic cutaneous wound is a big problem and it involves the restoration of continuity after wounding. Wounds are treated with various medicinal herbs or their extracts. Plants provide various remedies to mankind and herbal plants are nature's gift used to treat wound with much lesser side effects. Some of those plants are Acalypha langiana, Tinospora cardifolia, Tragiain volucrata, Napoleona imperialis, Proso piscineria, Lawsonia alba, Ginkgo biloba, Aloe vera and Catharanthus roseus [1].

Acalypha is the fourth largest genus of the Euphorbiaceae family. It consists of 450 species, in the form of evergreen shrubs, trees and annuals, mainly in the tropical regions of Africa, America and Asia [2]. Acalypha fruticosa is an erect shrub, up to 1 m tall, with strong aromatic odour. It has various traditional uses, a cure for stomachache, dyspepsia, venom antidote, rheumatism and dermatitis [3]. The plant has also been subjected to several biological investigations, concerning its antioxidant, anti-inflammatory, and cytotoxic properties [4]. A. fruticosa is also used as sand fly repellent, and used in ethno veterinary practices, as it demonstrates similar medicinal properties in both humans and animals [2].

2. MATERIALS AND METHODS

Plant collection: The Roots of Acalypha fruticosa were collected from Tirupati, Andhra Pradesh, India. The roots were identified and authenticated by Prof. K. Madhava Chetty, Department of Botany, and Sri Venkateswara University. The plant specimen was deposited at Botany Department of Venkateswara University With Voucher number 1252.

Animals: Healthy Wistar albino male rats weighing 150-200 g were used for the study which was procured from mahaveera enterprises, Hyderabad. They were housed individually in polypropylene cages, maintained under standard conditions (12h light and 12h dark cycle, 25±30°C, 35-60% relative humidity), the animals were fed Committee (IAEC), Vaagdevi College of Pharmacy, Warangal, Telangana.

Acute oral toxicity studies: Male swiss albino rats were used and the procedure was followed by using OECD 423[5].

Evaluation of wound healing activity:

Excision Wound Model: Animals in each group were anaesthetized a full thickness of the excision wound of circular area (approx. 500 mm²) and 2mm depth was made on the shave back under mild ether. The animals were divided into four groups. Group I treated with control (simple ointment base), group II treated with test 1 (100 mg /kg), group III treated with test 2 (200 mg/kg), group IV treated with standard drug mupirocin ointment. The wounding day was considered as day 0. Rats were left undressed to open environment. To determine the rate of wound contraction, excision wounds were traced on a transparent paper having a millimeter scale and the change in wound size was calculated as the percentage of wound area that had healed. The period of epithelialilzation of the wound was expressed as the number of days taken for complete epithelialization (so that no raw was left behind) [6]. The wounds are monitored and the area of wound was measured on 4, 6, 8, 10, 12, 16 post-wounding days and the mean % wound closure is reported [6].

The Period of Epithelization: The period of epithelization was calculated as the number of days required for falling of the dead tissues remants without any residual raw wound in excision wound model [6].

Statistical Analysis: All results were expressed as mean ± S.D. and the results were compared statistically by One-Way ANOVA Test using Graph pad Prism software (student version 5). The P value < 0.05 was considered statistically significant.

Table 1: Calculation of % wound contraction					
Post	Wound Area in mm² (% wound contraction)				
wound day	Control	Standard	T1 (100 mg/kg)	T2 (200 mg/kg)	
0	500.76±20.86	517.48±24.64	497.31±20.44	500.76±20.86	
2	455.60±14.24	406.81±24.32***	428.58±15.57***	398.06±29.47***	
	(9)	(21.3)	(13.8)	(20.4)	
4	395.08±28.23	216.35±19.04***	236.26±16.23***	227.18±14.38 ***	
	(18.5)	(58.2)	(52.4)	(54.6)	
6	256.41±21.95	133.05±14.42***	152.45±16.20***	125.55±17.28***	
	(42.8)	(74.2)	(59.3)	(64.9)	
8	249.88±11.95	77.85±14.81***	96.2±87.8***	76.65±16.65***	
	(50.1)	(84.9)	(65.56)	(74.7)	
10	229.30±10.06	43.4±6.71***	60.28±7.18***	35.4±9.34***	
	(54.2)	(91.6)	(70.8)	(82.9)	
12	207.58±13.37	14.33±3.01***	26.20±6.49***	11.81±4.34***	
	(58.5)	(97.2)	(76.56)	(87.6)	
14	152.35±12.68	5.01±2.83***	14.33±3.51***	4.65±2.64***	
	(69.5)	(99)	(89.1)	(91.23)	
16	131.1±7.65	2.75±1.59***	5.81±2.31***	1.78±1.4***	
	(74.8)	(99.4)	(92.23)	(94.26)	

Values were Mean±S.D. (n=6) statistical significance was determined by ANOVA followed by Dunnet's test *** P<0.0001 when compared to control group.

3. RESULTS

Acute toxicity study

In the acute toxicity study, EEAF was found to be safe up a dose of 2000 mg/kg. So, two doses i.e., 100 and 200 mg/kg were selected for the evaluation of wound healing activity.

Table 2: Period of Epithelization in excision				
wound model treated by EEAF ointment				
Group	Period of Epithelization (Days)			
Control	25.3±0.81			
Standard	18.5±1.51***			
T1	22.3±0.51***			
T2	20.7±1.22***			
Values were Mean±S.D. (n=6) statistical significan				

P<0.0001 when compared to control group.

Evaluation of wound healing activity:

was determined by ANOVA followed by Dunnet's test **

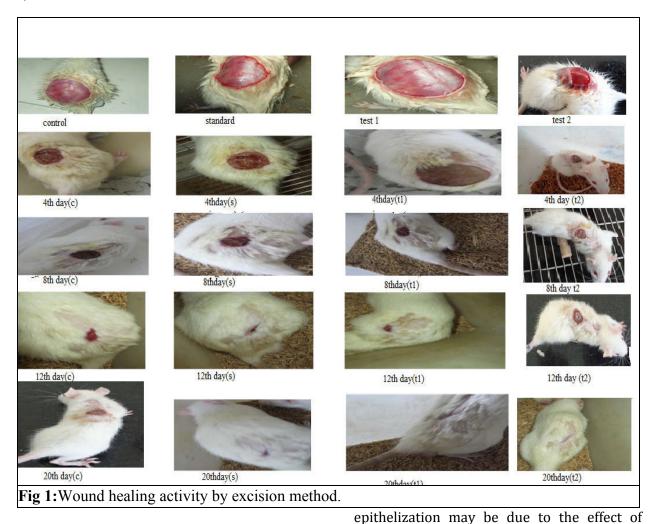
The percentage wound contraction was calculated as 92.23% (100 mg/kg body) and 94.26% (200 mg/kg) as compared to the control group 74.8%.

Effect on period of epithelization:

The period of epithelization was found to be 22.3 days (100 mg/kg) and 20.7days (200 mg/kg) compared to control 25.3.

3. DISCUSSION

The present investigation describes some unique features of the root extract from the plant Acalypha fruticosa with respect to its potential wound healing capacity in rats. Plant products are potential wound healing agents, and largely preferred because of their wide spread availability, non-toxicity, absence of unwanted side effects, and effectiveness as crude preparations. Early dermal and epidermal regeneration in the treated group conformed that the Acalypha fruticosa extract had a positive effect toward cellular proliferation, granulation tissue formation, and epithelization. The treated rats showed marked epitheliazation, a moderate amount of extra cellular matrix synthesis and new blood vessel formation.



Latex Calotropis (200 gigantean mg/kg/day) was evaluated for its wound healing activity in albino rats using excision and incision wound models. Latex treated animal's exhibit 83.42 % reduction in wound area when compared to controls which was 76.22 %. The extract treated wounds are found to epithelize faster as compared to controls. Significant (p<0.001) increase in granulomatous breaking strength (485±34.64) was observed. The Framycetin sulphate cream (FSC) 1 % w/w was used as standard [7].

The results of the present study showed that Acalypha fruticosa possesses a definite prohealing action. In excision wound healing model the ethanolic extract of the roots of the plant Acalypha fruticosa showed significant increase in percentage closure of excision wounds by enhanced epithelization. The

Acalypha fruticosa on enhanced collagen synthesis. Topical application of Acalypha fruticosa extract ointment increased the percentage of wound contraction and completed wound healing by 20th day. Progression of wound healing by 16th day i.e T1 92.23 and T2 94.26 compared with control 74.8. It also reduced the period epithelization from control 25.3 days to T1 22.3days to T2 20.7 days. In excision wound healing model the ethanolic extract of the roots of the plant Acalypha fruticosa showed significant increase in percentage closure of excision wounds by enhanced epithelization. The epithelization may be due to the effect of Acalypha fruticosa on enhanced collagen synthesis. The aqueous extract of C. papaya fruit (100 mg kg-1 day-1 for 10 days) was evaluated for its wound healing activity in streptozotocin-induced diabetic rats using

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excision and dead space wound models. Extract-treated animals exhibited 77% reduction in the wound area when compared to controls which was 59%. The extract treated wounds were found to epithelize faster as compared to controls. The wet and dry granulation tissue weight and hydroxyproline content increased significantly when compared to controls. The extract exhibited antimicrobial activity against the five organisms tested. Carica papaya promotes significant wound healing in diabetic rats and further evaluation of this activity in humans is suggested [8].

Percentage yield of the extract was 62% and its colour was Green with semi solid consistency. Preliminary phytochemical investigations of EEAF showed the presence of triterpinoids, steroids, flavanoids, phenols and saponins.

3. CONCLUSION

In conclusion, Ointment from the root of Acalypha fruticosa exhibited significant prohealing activity in the infected wound when topically applied on rats by affecting various stages of healing process.

4. ACKNOWLEDGEMENT

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5. CONFLICT OF INTEREST

The author(s) confirm that this article content has no conflict of interest.

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