Available online on www.ijppr.com
International Journal of Pharmacognosy and Phytochemical Research 2013; 5(1); 57-59

ISSN: 0975-4873

Research Article

Wound Healing Properties of Methanolic Extract of Seeds of *Mucuna Pruriens*

*[1]Gunde M.C., [1]Meshram S.S., [1]Dangre P.V., [2]Amnerkar N.D.

[1]Kamla Nehru College of Pharmacy,Butibori,Nagpur (M.S.)-India
[2]Sharad Pawar College of Pharmacy, Wanadongri,Nagpur (M.S.)-India.

ABSTRACT
Plants and their extracts have immense potential for the management and treatment of wounds. Wound healing is complicated process that involve four phases viz.(i)Coagulation begins immediately after injury (ii) inflammation which begins with in hours (iii)proliferation which begins with in days (iv)Remodeling up to period of weeks, months, year. *Mucuna pruriens* seed are being used traditionally as anti-inflammatory, antimicrobial agents in various disorders. This study was aimed to investigate the wound healing efficiency of methanolic extract of seeds formulated as 1% and 2% hydogels of carbapol 974 NF. Carbapol is a pH sensitive polymer and widely used as a base for various topical gel preparation. Wound were induced in swiss albino mice divided into five groups as follows: Group-I (negative control) received no treatment. Group-II was treated with carbapol empty gel. Group-III and IV was treated with carbapol gel containing 1and 2% of methanolic seeds extract, Group-V (positive conrol) received the standard drug (Framycetin cream). The efficacy of treatment was evaluated based on the wound contaction, hydroxyproline content and tensile strength of skin.

Keywords: Wound healing, hydrogel, hydroxyproline content and wound contraction.

INTRODUCTION

Wound is defined as a loss or breaking of cellular and anatomical or functional continuity of living tissue [(1)]. Healing of wound is biological process that is initiated by trauma and often terminated by scar formation [(2)]. The process of wound healing occurs in different phases such as coagulation, epithelization, granulation, collagenation and tissue remodeling [(3, 4)]. In India, there has been interest in knowing the potential of natural products obtained plants and animal for development of drug for various diseases. The wound healing properties of plants was taught in popular form of Indian medicine know as Ayurveda [(5)].

Mucuna pruriens Baker (Fabaceae) is a herbaceous twining annual found almost all over India from Himalayan foothills to the plains of Punjab to south India among hedges and bushes. It has been cultivated in certain region for its levodopa content [(6)]. Seed of *M.pruriens* have been shown to possess antispasmodic, anti-inflammatory, antipyretic [(7)], antivenin property [(8)], aphrodisiac activity [(9)], anabolic and fertility property [(10)]. It has also shown to possess CNS depressant activity. Seeds are used in general weakness as a tonic.

MATERIAL AND METHODS

Plant Material: The fresh seeds of *M.pruriens* were collected from the rural area of Nagpur district (M.S.) India and authenticated by macroscopic and microscopic evaluation. The seeds were dried in shade and pulverized to get coarse powder.

Animal and grouping: Swiss albino mice weighing 20-25gm were used. They are allowed to acclimatize to laboratory conditions for not less than 10 days. Wounds were induced in mice by dividing them into following groups: Group-I (negative control) received no treatment. Group-II was treated with carbapol empty gel. Group-III and IV was treated with carbapol gel containing 1and 2% of methanolic seeds extract, Group-V (positive conrol) received the standard drug (Framycetin cream).

Preparation of extract: The weighed quantity of coarsely powder of seeds of *M.pruriens* was passed through sieve no.40 and subjected to hot solvent extraction in a soxhlet apparatus using methanol, at a temperature range of 40-80^{o}C, before and after every extraction the marc was completely dried and weighed. The filtrate was evaporated to dryness at 40 ^{o}C under reduced pressure in a rotary vacuum evaporator. A brownish black waxy residue was obtained. The percentage yield of methanolic extract was 18.7%w/w. The extract was further used for the evaluation of wound healing activity.

Experimental Methods: Wound healing activity was studied using two models viz. excision wound model and incision wound model.

Excision wound model: It was made on the dorsal thoracic region 1 cm away from vertebral column and 5 cm away from ear using round seal of 1.5 cm diameter on the anaesthetized mice. The skin of wound area was excised to the full thickness to obtain a wound area of about 300 sq.mm. A contraction, which contributes for wound closure in first 2 weeks, was studied by tracing the raw wound. Wound area was measured by retracing the

*Author for correspondence: E-mail: mgunde@gmail.com

Table 1: Effect of topical application of ointments containing methanolic extracts of *M.pruriens* seeds on wound contraction of excision wound.

Area of wound closure (sq.mm.±S.E.M.)							
Groups	0 day	3rd Day	6th Day	9th Day	12th Day	15th Day	18th Day
I (Control)	287.83±1.722	272.5±2.289	192.83±0.626	146.33±0.733	109.33±0.58	47.66±1.489	6.75±0.366
II (Empty Carbapol)	290.66±1.303	255.66±1.505	183.16±1.722	135.83±1.471	98.33±1.211	43.5±1.0488	0.833±0.6831
III(1% M.E.in Carbapol)	291.33±1.211	212.83±1.870	151.33±1.632	106.66±1.048	63.66±0.376	17.5±0.3578	0.416±0.58
IV(2% M.E.in Carbapol)	288.33±1.722	173.16±0.310	113±0.31	56.83±0.45	27.16±1.94	2.15±0.567	0.041±0.102
V (Standard Drug)	290.83±1.169	174.66±1.870	117.5±0.703	63.83±0.542	33.5±0.460	5.66±0.429	0.083±0.204

Graph 1: Effect of topical application of ointments containing methanolic extracts of *M.pruriens* seeds on wound contraction of excision wound

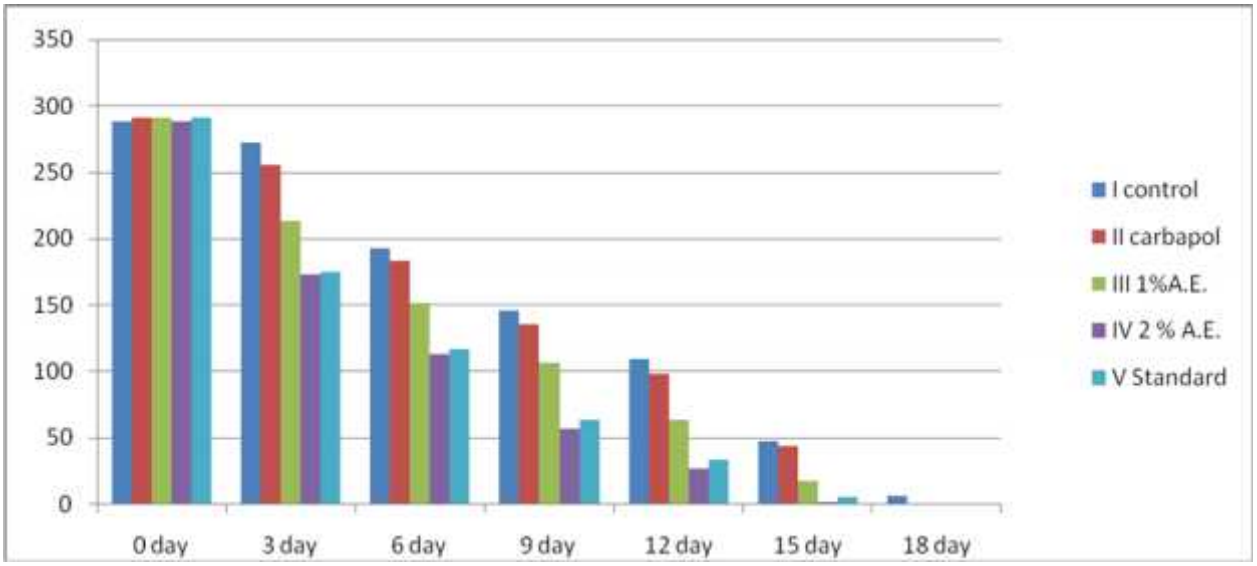

Table 2: Effect of topical application of ointments containing methanolic extracts of *M.pruriens* seeds on hydroxyproline content.

Group	Hydroxyproline content
I (Control)	42.11±5.60
II (Empty Carbapol)	62.23±4.50
III (1% M.E.in Carbapol)	63.35±4.65
IV(2% M.E.in Carbapol)	74.43±4.56
V (Standard)	65.38±6.12

wound on a millimeter scale graph paper. The degree of wound healing was calculated using formula:

1-(wound area on corresponding day/wound area on zero day) x 100. The number of days for complete epithelization was noted [11].

Estimation of Hydroxyproline: The hydroxyproline content was determined by extracting hydroxyproline from scab using conc.HCL followed by reaction between amino group of hydroxyproline with p-aminobenzaldehyde to develop red colour. The red colour thus measured on spectrophotometer at 558nm. [12]

Incision wound model: One Para-vertebral straight incision of 6 cm length each was made through the entire thickness of the skin, on either side of the vertebral column with the help of a sharp scalpel. After complete homeostasis the wound were closed by means of interrupted sutures placed at equidistance points about 1 cm apart and wound healing was noted. When wounds were cured thoroughly, the sutures were removed and the tensile strength of the skin that is the weight in grams required to break open the wound skin was measured by tensiometer[13].

RESULTS AND DISCUSSIONS

The basic principle of optimal wound healing is to minimize tissue damage and provide an adequate tissue

Table 3: Effect of topical application of ointments containing methanolic extract of *M.pruriens* seeds on tensile strength of the skin having incision wound.

Groups	Tensile strength in gram (mean±SEM)
I (Control)	328.83 ± 3.64
II (Empty Carbapol)	513.00 ± 2.63
III (1% M.E.in Carbapol)	739.67 ± 3.29
IV(2% M.E.in Carbapol)	861.83 ± 3.19
V (Standard Drug)	643.00 ± 4.91

perfusion and oxygenation, proper nutrition and moist wound healing environment to restore the anatomical continuity and function of the affected part. The results of excision wound model indicate that in the first 4 days there is no significant increase in the wound contraction in all the groups as compared to the control group. The results of the 8th day indicate that there is significant increase in the percentage wound contraction in the group treated with standard drug, 1% and 2% methanolic extract revealing that the extract has ability to induce cellular proliferation. (Table-1 and Graph-1)

Hydroxyproline is one of the biomarker which increase level indicate the wound healing process. The increase hydroxyproline content in scab of the animal treated with 1and 2% methanolic extract support the wound healing process. Out of these two extracts 2%methanolic extracts is found to be the most effective. (Table-2)

The increase in tensile strength of wounded skin in inscision wound model indicates the promotion of collagen fibers. Highest tensile strength of the wounde skin observed in the animal treated with 2% methanolic extracts. The increase tensile strength reveals that the disrupted surfaces are firmly knit by the collegen (Table 3)

REFERENCES

1. Ayello EA, What does the wound say? Why determining etiology is essential for appropriate wound care, Advances in skin wound care (2005) 18, 98-109.
2. Rubin E, Fabrex JL, Repair, regeneration and fibrosis. In Pathology, 2nd edition J.B.Lippincott, Philaldephia, (1996),7-15.
3. Fulzele SV, Satturvar PM, Joshi SB, Dorle AK, Wound healing activity of *Hingvadya ghrita* in rats. Indian Drugs (2002), 39, 606-609.
4. Toshikazu K, Yuko I., Molecular pathology of wound healing, Forensic Science International (2010), 203,93-98.
5. Biswas TK,Mukherjee B.Plant medicine of indian origin for wound healing activity: a review, International Journal of Lower Extremity Wounds (2003),2,25-29.
6. Rangari VD, Pharmacognosy &Phytochemistry, 1st edition, Vol-II, Career Publication,Nashik (M.S.)(2005), 243.
7. Stephen OM, Ademola A, Solomon U, Oluwatoyan AO, Evaluation of the anti-diabetic properties of *Mucuna pruriens* seed extract, Asian Pacific Journal of TropicalMedicine, (2011),632-636.
8. Nget HT, ShinYF, Si MS, Enrico M, Roberto G, John CA, The protective effect of *Mucuna pruriens* seeds against snake venom poisoning, Journal of Ethnopharmacology, (2009), 123, 356-358.
9. Mohammad KA, Abbas AM, Shukla KK, Jaiswal SP and Sohail A, Effect of *Mucuna pruriens* on semen profile and biochemical parameters in seminal plasma of infertile men, Fertility and Sterility Vol-90 No-3.
10. Dhawan BN, Dubey MP, Mehrotra BN, Rastogi RP, Tandon JS, Screening of Indian plants for biological activity, Indian Journal of Experimental Biology,18,594-606.
11. Nayak BS, Anderson M, Pereire P, Evaluation of wound healing potential of *Catharanthus roseus* leaf extract in rats, Fitoterapia (2007), 78, 540-544.
12. Shivhare Y, Singour PK, Patil UK, Pawar PS, wound healing potential of methanolic extract of *Trichosanthes dioica* Roxb. (fruits) in rats, Journa of Ethanopharmacology (2007),614-619.
13. Diwan PV, Reddy BS, Reddy RKK, Naidu VGM, Madhusudan K, Agwane SB, Ramkrishna S., Evaluation of antimicrobial, antioxidant and wound healing potentials of *Holoptelea integrifolia*, Journal of Ethanopharmacology(2008),115,249-256.