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EVALUATION OF WOUND HEALING ACTIVITY OF *ROSA INDICA LINN.* IN EXPERIMENTAL RATS

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ABSTRACT

Cicatrisation, is an intricate process in which the skin or another organ-tissue repairs itself after injury. In the inflammatory phase, bacteria and debris are phagocytosed and removed, and factors are released that cause the migration and division of cells involved . The importance of this new model becomes more apparent through its utility in the fields of regenerative medicine and tissue engineering. When tissue is first wounded, blood comes in contact with collagen, triggering blood platelets to begin secreting inflammatory factors. Platelets also express glycoprotein on their cell membranes that allow them to stick to one another and to aggregate, forming a mass. Fibrin and fibronectin cross-link together and form a plug that traps proteins and particles and prevents further blood loss. This fibrin-fibronectin plug is also the main structural support for the wound until collagen is deposited. Migratory cells use this plug as a matrix to crawl across, and platelets adhere to it and secrete factors. The clot is eventually lysed and replaced with granulation tissue and then later with collagen. The plants Rosa indica have wide ethno medicinal use. The literatures revealed that there is lack of scientific reports on its leaves. So it is important to provide scientific means in a systematic manner.

KEYWORDS: Cicatrisation, *Rosa indica* , Fibronectin, Collagen

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INTRODUCTION

Immediately after a blood vessel is breached, ruptured cell membranes release inflammatory factors like thromboxanes and prostaglandins that cause the vessel to spasm to prevent blood loss and to collect inflammatory cells and factors in the area. Increased porosity of blood vessels also facilitates the entry of inflammatory cells like leukocytes into the wound site from the bloodstream. Macrophages are essential for wound healing.[1] They replace PMNs as the predominant cells in the wound by two days after injury.[2] Attracted to the wound site by growth factors released by platelets and other cells, monocytes from the bloodstream enter the area through blood vessel walls.[3] Numbers of monocytes in the wound peak one to one and a half days after the injury occurs.[4] Once they are in the wound site, monocytes mature into macrophages. The spleen contains half the body's monocytes in reserve ready to be deployed to injured tissue. The wound may be defined as a loss or breaking of cellular and anatomic or functional continuity of living tissues. Healing of wound is a biological process that is initiated by trauma and often terminated by scar formation. The process of wound healing occurs in different phases such as coagulation, epithelization, granulation, collegenation and tissue remodelling. A method for evaluating and comparing texture properties of hydrogels was established with the Texture analyser and back-extrusion equipment. Gels can be analysed in minutes and the method was reproducible with standard deviations varying with less than 2%. This makes it applicable in comparing variations between different batches of gels as well as the stability. Stability was tested with the preferred concentrations of gels as well as gels. Accelerated stability tests proved that gels are not stable for 30 days at 40 °C. dispersion added to the gels made them less viscous and less stable. The addition of 1% (w/w) glycerine prepared with high molecular weight made the gels more stable.

The Purpose of wound infection management is to prevent or minimise the risk of infection. .

Sterilisation of surgical instruments, sutures etc according to guidelines this study holds great promise for future research in diabetes and obesity

MATERIALS AND METHODS

Chemicals

Framycetin sulphate cream (FSC)_ (1%w/w), diethyl ether, ethanol, sterilized cotton were used . The plant specimen identified should be disease free with all parts intact without any injuries or deformities. The plant can be uprooted, root is to be cleaned and washed. Plant twigs having leaves and flowers must be collected in case of flowering plan After collection of the plant, they should be pressed immediately in the field condition. Wilting the plant material should be avoided.

Procurement and identification of plant material

The Plant material were collected from Local Area of Etawah. The disease free flowers were collected in the morning. The flowers were collected in the month of January by hand picking method. and kept at standard storage conditions for further investigations.

Animals

Healthy wistar albino rats of either sex and of approximately the same age,weighing about 150-250 g were used for the study. They were fed with standard diet and water *ad libitum*. They were housed in polypropylene cages maintained under standard conditions (12/12 hr light/dark cycle; 25ºC ± 30ºC, 35-60%RH).Models were used to evaluate the wound-healing activity of leaves of *Rosa indica.* The study was approved by the Institutional Animal Ethical Committee of Sir Madanlal Institute of Pharmacy Etawah (U.P.), and also fulfils the guidelines of CPCSEA.

Preparation of Rosa indica extract

(i) Selection - The plant specimen identified should be disease free with all parts intact without any injuries or deformities. The plant can be uprooted, root is to be cleaned and washed. Plant twigs having leaves and flowers must be collected in case of flowering plants.

(ii) Pressing - After collection of the plant, they should be pressed immediately in the field condition. Wilting the plant material should be avoided.

(iii) Technique of Pressing - Collected plant specimen can be kept in newspaper sheets. News paper sheets are arranged alternately be blotting paper sheet. These paper sheets are pressed. A wooden press or an aluminum press can be used. Spreading of plant material inside the sheets and weight on press are to be done carefully. Standard size of the press measures 32 X 48cm.

(iv) Drying - Blotting paper sheets are changed 2 to 3 times for proper soaking of moisture from the plant materials. Paper changing in the press is done carefully for fifteen to twenty days by observing the condition of material.

(v) Mounting - Good quality herbarium sheets are used for pasting or fixing materials. Heavy white sheet card boards are good for the mounting. Properly dried materials are fixed on the sheet by glue or gummed cell phone tape.

(vi) Labelling and Identification - Labelling and identification are done after fixing. A label is attached to the right hand corner of the herbarium sheet. The identification information carries locality, botanical name of the plant and vernacular name if any, family, soil, habit, uses, distribution and other entries as it deemed proper. The name of the collector is mentioned last.

(vii) Storage- Wooden or steel cabinets are used for storing. The size of the shelves should be bigger than herbaria sheet.

(viii) Protection - Proper sanitation of storage condition is to be maintained. Mould, fungi, insects also create problem for herbaria sheet. Thoroughly dried and well ventilated warm conditions can save from fungal infection. Otherwise fungicides can be sprayed. Naphthalene balls and moth balls are kept in shelves to provide protection against insect invasion.

Induction of wound:

Excision wounds were used for the study of rate of contraction of wound and epithelization. Animals were anaesthetized with slight vapour inhalation of di-ethyl ether and the right side of each rat was shaved.

Excision wounds sized 300 mm and 2 mm depth were made by cutting out layer of skin from the shaven area.

Experimental design

Rats selected from colonies were randomized in to three groups, comprising of 6 rats each. For the assessment of cutaneous wound healing activity, dose level was chosen in such a way that, dose was approximately one tenth of the maximum dose during acute toxicity studies (200 mg/kg/day).

The treatment schedule was as follows:

Group-I: Received no treatment and served as control

Group-II: Received application of standard drug ointment i.e. Framycetin sulphate cream (FSC)_(1 %w/w)

Group III : Received application of extract of Rosa indica (200 mg/kg/day)

Statistical analysis:

The means of wound area measurement and wound breaking strength between groups at different time intervals were compared using one-way ANOVA, followed by Tukey's tests .

RESULTS AND DISCUSSION

During study of wound healing in normal rats following results were obtained: Acute toxicity studies showed that drug was found to be safe up to maximum dose of 2g/Kg body weight of the animal. In studies using excision wound model, the latex treated group III showed significantly greater wound healing as compared to control animals (Table 1). The standard drug treated animals in normal animals were showed significantly greater wound closure as compared to control and latex treated animals (Table 2).

Table 1. Effect of Leaves of *Rosa indica* on Excision Wound [Wound Area (mm2)]

Day	Group 1	Group 2	Group 3
0	295.83 ± 10.362	229.87 ± 7.032	186.67 ± 9.545
4	255.00 ± 12.547**	175.83 ± 5.866**	153.33 ± 9.972**
8	213.33 ± 9.093**	165.83 ± 5.974**	120.83 ± 8.634**
16	68.33 ± 2.482**	25.50 ± 2.141**	30.83 ± 2.396**

n=6; values are in mean ± SEM, **Significant p<0.001

Table 2. Effect of leaves of Rosa indica on excision wound (% wound closure)

day	Group 1	Group 2	Group 3
0	0	0	0
4	10.88 %	22.52 %	17.86 %
8	25.33 %	39.68 %	33.27 %
16	76.52 %	77.86 %	73.48 %

Table 3 : Effect of leaves of Rosa indica on wound healing in incision wound

Group	**Incision wound breaking strength (g)**
Group 1	299.17 ± 30.90
Group 2	425 ± 80.14*
Group 3	488.17 ± 33.64***

CONCLUSION

Wound infections can complicate illness, cause anxiety, increase patient discomfort and lead to death. It is estimated that surgical wound infections result in an increased length of hospital stay by about 7-10 days. Hence the prevention and management of wound infections has a major impact on both patient health and health economics.

However, more effectiveness can be seen by altering the doses. therefore, further studies are required to establish the efficacy of the Rosa indica extract as a woundhealing drug.

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