

## Wound Healing Activities of *Plantago major* Leaf Extract in Rats

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**Abstract:** Four groups of adult male *Sprague dawley* rats each consist of six animals. All animals were experimentally wounded in the dorsal neck area. Wounds of Group I rats each was treated with blank Vaseline-jelly (control), whereas wounds of Group II and Group III animals were treated with 5 and 10% Vaseline (*Plantago major* aqueous leaf extract in Vaseline), respectively. Wounds of Group IV rats each was treated with commercial solcoseryl-jelly as reference. The effects of vehicles on the rate of wound healing were assessed. Wounds treated with 5 and 10% Vaseline and solcoseryl-jelly significantly healed earlier and much faster as compared to wounds of control. These results strongly document the beneficial effects of plant extract for the acceleration of wound healing process.

**Key words:** Wounds healing, *Planrago major*, solcoseryl-jelly, rats

### INTRODUCTION

The leaves of *P. major* L. are used in wound healing in traditional medicine. Either whole or crushed leaves are used directly on burns, wasp stings and wounds of all kinds to stop bleeding, keep the wound clean and to enhance the healing process. *P. major* was described as a very efficient wound healing remedy. For superficial wounds to heal, it was sufficient to apply the juice from the plant<sup>[1]</sup>.

*P. major* leaves have been associated with various biological properties ranging from anti-inflammatory<sup>[2]</sup>, antibacterial<sup>[3]</sup>, anticandidal activity<sup>[4]</sup> antioxidant activity<sup>[5,6]</sup> and antitumor<sup>[7]</sup>, immunomodulating and antinociceptive effect<sup>[8]</sup>, antiulcerogenic activities<sup>[9,10]</sup> to wound healing<sup>[1,11,12]</sup>.

The aim of the present study was carried out to assess the effects of aqueous extracts of *P. major* on the rate of wound-healing process in rats.

### MATERIALS AND METHODS

**Vaseline (100% pure petroleum jelly):** Vaseline is a brand of petroleum jelly originally produced by Chesebrough-Pond's, USA, and currently by Unilever. Vaseline is obtained from the local pharmacy.

**Solcoseryl-jelly:** Solcoseryl Jelly manufactured by Solco Basle Ltd. (CH-4127) Birsfelden, Switzerland is obtained from the local pharmacy.

***P. major:*** The plant were collected from Machang, Kelantan, Malaysia and identified by comparison with specimens available at the Herbarium of the Forest Research Institute, Kepong, Malaysia. Voucher specimens of the plant material are deposited at Department of Pharmacy, University of Malaya, Malaysia.

**Preparation of aqueous extracts:** *P. major* fresh leaves were cut, labelled, washed with distilled water and dried in oven 50°C for 5-7 days until fully dried. The Leaves were ground to a fine texture form using a grinder. Weighing 40 g of fine texture and mixing it with 800 mL of sterile distilled water in a conical flask using a ratio of 1:20. It was then heated and stirred on a hotplate for 3 h. After being left to cool, the residue was removed by filtration using a mesh and filter funnel. EYELA rotatory vacuum evaporator then extracted the filtered material.

The extracts were then submitted to lyophilization by a freeze-dryer, to produce powdered forms of the extracts. The freeze-dried products were mixed with Vaseline in a concentration of 5% and 10% Vaseline (wt/wt), respectively.

**Experimental animals:** Sprague Dawley adult male rats were obtained from the animal house, Faculty of Medicine, University of Malaya. The rats were divided randomly into 5 groups of 6 rats each. Each rat that weighted between 180-200 gm was housed separately (one rat per cage). The animals were left for 48 h to acclimatize to the animal room conditions and were maintained on standard pellet diet and tap water.

Table 1: Time required for wounds healing by *P. major* in rats

Animal groups	No of animals	Type of dressings	Healing time (days) (Mean±M.S.E)
Group I	6	Blank Vaseline	17.67± 0.72
Group II	6	5% Vaseline	14.50±0.43*
Group III	6	10% Vaseline	11.67±0.42**
Group IV	6	Solcoseryl-jelly	12.17±0.31

\*\*p<0.05 significant from control (Group I) \*\*p<0.05 significant from control (Group I) and Group II

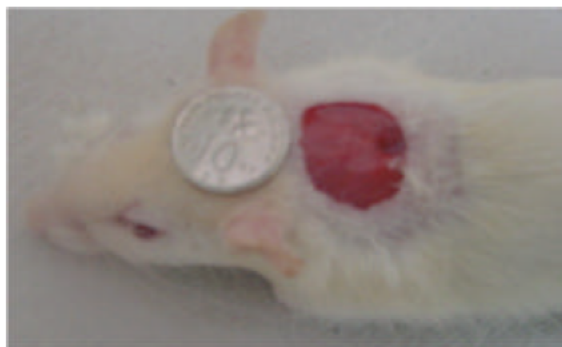


Fig. 1: 2 cm diameter excision skin wound on day 0 before application of vehicle

**Experimentally induced wounds:** An area of uniform wound 2 cm in diameter was excised from the nape of the neck of all rats with the aid of round seal as described by Morton and Melone<sup>[3]</sup>(Fig. 1), in previously shaved, disinfected with 70% alcohol and injected with 1 mL of Lignocaine HCl (2%, 100 mg/5 mL), to the depth of the muscle, avoiding incision of the muscle layer itself and tension of skin was kept constant during the procedure.

**Topical application of vehicles:** Group I animals were treated with a thin layer of blank Vaseline (100% pure petroleum jelly) as positive control twice daily. Group II and Group III rats were treated with a thin layer of 5 and 10% Vaseline wt/wt (50 mg and 100 mg of aqueous leaf extract in one gram Vaseline) twice daily, respectively. A thin layer of commercial solcoseryl-jelly was topically applied twice daily to wounds of Group IV rats as reference.

**Statistical analysis of data:** Results were expressed as mean+S.M.E. The statistical difference between the groups in the term of the mean of wound healing was calculated by using Student's t-test

## RESULTS AND DISCUSSION

The present study showed that aqueous extract of *P. major* had efficient wound healing activity. The topical administration of the preparation with *P. major* extract accelerates the progression of wound healing (Table 1). Wounds treated with 5% and 10% Vaseline and

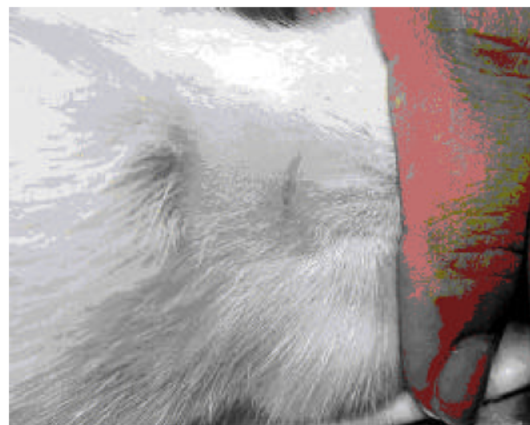


Fig. 2: Complete wound healing in 10% Vaseline treatment on day 11



Fig. 3: Complete wound healing in blank Vaseline treatment on day 17

Solcoseryl-Jelly showed considerable signs of dermal healing and statistically ( $p<0.05$ ) healed earlier than the blank Vaseline (Table 1; Fig. 2 and 3).

The majority of the world's population relies on traditional medicine for the treatment of wounds. In developing countries, remedies prepared from herbal plants have been widely used for the treatment of wounds. Extract from the leaves of *P. major* have been shown to be beneficial for treatment of wounds. The traditional use of *P. major* in wound healing is quite old. The leaves were prescribed for treatment of dog bites<sup>[3]</sup> and for wound healing<sup>[4]</sup>. The fact that the extract produced antibacterial activities suggests that there may be a scientific basis for their utility in traditional medicine for the treatment of skin infections<sup>[4]</sup>. *P. major* extract had good effect on bacterial infections. The extract had weak antibacterial activity *in vitro* but they had an effect on infected wounds *in vivo*. While antibiotics on infected

wounds had no effect, topical treatment with *P. major* extract eradicated the infections and healed the wounds<sup>[9]</sup>. Polysaccharide fraction of a soluble pectin polysaccharide from *P. major* protects against pneumococcal infection in mice when administered systemically prechallenge<sup>[3]</sup>.

The extract from *P. major* contains a mixture of powerful antioxidant compounds that may be one of potential mechanism contributing to enhance wound healing<sup>[13]</sup>. Protection of cells against destruction by inflammatory mediators may be one of the ways in which the extracts from the plant, *P. major*, contribute to wound healing. Several flavonoids have been isolated from *P. major*. Skari *et al.*,<sup>[6]</sup> isolated plantaginidin and homoplantinidin. Many flavonoids are antioxidant<sup>[5]</sup>. Examples of such compounds in *P. major* are baicalein, hispidulin and plantaginidin<sup>[13]</sup>. A number of flavonoids are also known to have free radical scavenging<sup>[15]</sup>. Baicalein, hispidulin, scutallarein and plantaginidin are free radical scavengers and inhibit lipid peroxidation<sup>[6]</sup>. Plantagluconin were isolated from the leaves and have been used to treat ulcers<sup>[14]</sup>. Plantamajoside has some known biological activities. It has an inhibitory effect on arachidonic acid-induced mouse ear edema (anti-inflammatory activity)<sup>[2]</sup>. The fresh leaves of old plants were reported to contain  $\beta$ -carotene (provitamin A) and ascorbic acid<sup>[16]</sup>. In conclusion, aqueous extract of *P. major* significantly accelerate wound healing compare to control and appeared to have several important properties that make it useful ideal as a dressing agent for wounds.

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