

WOUND HEALING ACTIVITY OF THE ETHER-CHLOROFORM BENZENE-95% EXTRACT OF MOMORDICA CHARANTIA FRUITS S IN ALBINO RATS

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The aim of present study was to assess the wound healing activity of benzene extracts of Momordica charantia fruit. Methanol extract of Momordica Charantia fruit was examined for wound healing potential in the form of 1% w/v solution in the excision wound created on the dorsal side of experimental animals, the 1% w/v extract should considerable difference in wound models and the result were compatable to that of the standard drug betadine (5% w/w) in terms of wound contracting ability and wound closure time. Antibacterial activity of methanol extract of the plant was also carried out as a supporting evidence for its wound healing potential. The mean percentage wound closure was calculated on the 4th, 8th, 12th, 16th and 15th wounding days. The extract treated animals showed tastes epithelisation of wound (17.86 ± 0.19) then the control. The period of epithelisation 21.23 ± 0.37 in case of standard drug 5% betadine ointment.

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1. Introduction

Momordica charantia (kerala) is important vegetable in India. The role of dirty compound preventing cancer important area of research. Several study suggested that momordica play vital roll in prevention of cancer. In numerous studies, at least three different groups of constituents found in all parts of momordica have clinically demonstrated hypoglycemic properties or other actions of potential benefit against diabetes mellitus[1]. These blood sugar lowering chemicals include a mix of steroidal saponins (known as charantins), insulin-like peptides, and alkaloids. Charantin is composed of a mixture of beta-sitosterol-beta-D-glucoside and 5,25 stigmadien-3-beta-ol glycoside. This hypoglycemic effect is more pronounced in the fruit of bitter melon where these chemicals are found in greater abundance. To date, close to 100 in vivo studies have demonstrated the blood sugar lowering effect of this bitter fruit. This fruit has also shown the ability to enhance cells' uptake of glucose, to promote insulin release, and to potentiate the effect of insulin [2]. Momordica charantia (MC), a member of the Cucurbitaceae family, is known as bitter melon, bitter gourd, balsam pear, karela, and pare. It grows in tropical areas of the Amazon, East Africa, Asia, India, South America, and the Caribbean and is used traditionally as both food and medicine. The plant is a climbing perennial with elongated fruit that resembles a warty gourd or cucumber. The unripe fruit is white or green in color and has a bitter taste that becomes more pronounced as the fruit ripens. The seeds, fruit, leaves, and root of the plant have been used in traditional medicine for microbial infections, sluggish digestion and intestinal gas, menstrual stimulation, wound healing, inflammation, fever reduction, hypertension, and as a laxative and emetic.³ Clinical conditions for which M. charantia extracts (primarily from the fruit) are currently being used include diabetes, dyslipidemia, microbial infections, and potentially as a cytotoxic agent for certain types of cancer.⁴⁻⁷

2. Experimental

2.1 Preparation of extract

The leaves and bark of *Madhuca longifolia* were collected from Indore, Madhya Pradesh. The leaves were dried under shade, powdered and passed through 40 meshes and stored in closed vessel for further use. The dried powdered material macerated for five days with alcohol with frequent shaking separately. On the five day it was filtered and the benzene extract was concentrated in vacuum under pressure using rotary flash evaporator. The material was air dried under shade, pulverized by a mechanical grinder and passed through a 40 mesh and then stored in airtight containers. The powdered leaves (250 g) were extracted with methanol (50%) for 24 h using a Soxhlet extractor. This ethanolic extract was concentrated to dryness under reduced pressure and controlled temperature (45-50°C) to yield solid masses.

2.2 Phytochemical analysis of the extract

The extract was screened for the presence of various constituents employing standard screening test.⁸ Conventional protocols for detecting the presence of glycosides, saponins, flavonoids, tannins etc. was used. Several phyto constituents like flavonoids and glycosides are known to promote wound healing process due to their antioxidant and antimicrobial activities.⁹

2.3 Toxicity Studies

Toxicity studies of alcoholic extract were carried out in oral doses of 150 to 250 mg/ kg-body weight using albino rats. After test extract administration, animals were observed 72 hr. period. The number of deaths was expressed as a percentile and the LD50 was determined by probate a test using the death percentage versus the log dose.¹⁰

3. Evaluation of wound healing activity

The rats of either sex weighing 250-300 g were selected. Animals were depilated at dorsal thoracic region before wounding. They were housed individually with free access to food and water, the basal food intake and body weight to the nearest gram was noted. Under light chloroform anaesthesia, wounding was performed aseptically. Excision, incision and dead space wound models in rats were selected for assessing the wound healing activity. The extract was suspended in Tween 70 (5% w/v) and administered orally at a dose of 250 mg/ kg body weight for a period of fifteen days to the animals in the incision and dead space models. The animals in excision model received daily doses in the same manner for a period twenty days or till the excised wound healing which ever was earlier and the animals in the first group, which served as a control received the vehicle only.

3.1 Incision wound

The rats were wounded under light Ether-Chloroform anaesthesia method. Two vertebral incisions of 6 cm were made through the entire thickness of the skin, on either side of the vertebral column with help of straight rounded bodied needle. Sutures were removed on 8th post wounding day and the tensile strength was determined on 15th post wounding day by continuous constant water flow technique.

4. Wound healing activity

Excision wound models was used to evaluate the wound – healing activity of *Momordica charantia* fruit.

5. Excision wound

In the excision wound model¹¹⁻¹² rats were anaesthetized lignocaine prior to creation of the wounds. A full thickness of the excision wound of 314 mm² was created by using toothed forceps, a surgical blade and pointed scissors. The entire wound left open. The groups were treated in the same manner as mentioned in the animal experimentation. The ointment was topically applied once in a day. The progressive changes in wound area were monitored planimetrically by tracing the wound margin on graph paper on wounding day, followed by 4th, 8th, 12th, 16th and 18th day.¹³⁻¹⁶

The animals were divided into 3 different groups as follows:-

Total no. Of animals for each group: - 4 mice (Swiss albino)

50% benzene Momordica extract: - 1% w/v (Topical application)

Betadine Ointment (5%)

Group	Treatment	No. of animals
Control	Untreated	4
Standard	Betadine	4
Test	50% benzene Momordica ext.	4

Study Parameters:-

- ❖ Wound closure
- ❖ Epithelialisation Time

6. Result and discussion

Significant wound healing activity was observed in animals treated with momordica extract compared with those who received the standard and control treatment. In excision wound modal, Momordica extract treated animals showed a significant reduction in wound area and period of epithelisation.

The mean percentage wound closure was calculated on the 4th, 8th, 12th, 14th and 17th, wounding days. The extract treated animals showed faster epithelisation of wound (13.26±0.01) then the control. The period of epithelisation was 16.15±0.21 in case of standard drug 5% Betadine ointment.

7. Conclusion

In our study, 1%w/v of the 50% benzene Momordica fruit extract significantly increased the rate of wound closure and rate of epithelisation. The wound was healed in very efficient manner that is very close to the standard Betadine. The drug has equally shown better and similar wound healing property as compared with Betadine. The constituents present in the Momordica fruit extract may be responsible for promoting the wound healing activity. New method developed

References

- [1] <http://www.rain-tree.com/bitmelon.htm> Retrieved Jan. 16, 2007
- [2] A. P. Jayasooriya, et al., Journal of Ethnopharmacology, 2000,
- [3] Tropical Plant Database, Raintree Nutrition. <http://rain-tree.com/bitmelon.htm>. [Accessed July 3, 2007]

- [4] Bitter melon – Wikipedia, the free encyclopedia. http://wikipedia.org/wiki/Bitter_melon. [Accessed July 2, 2007]
- [5] Y. Oishi, T. Sakamoto, H. Udagawa, et al. *Biosci Biotechnol Biochem* **71**, 735 (2007).
- [6] P. Chaturvedi, S. George, M. Milinganyo, Y. B. Tripathi, *Phytother Res* **18**, 954 (2004).
- [7] I. Ahmed, M. S. Lakhani, M. Gillett, et al. *Diabetes Res Clin Pract* **51**, 155 (2001).
- [8] G. E. Trease, W.C. Evans, *Text book of Pharmacognosy*, Edn 12, ELBS Publication, Bailliere Tindall 1985, 334-345.
- [9] Gulcin, Mshvildadze and Elias, *Planta Medica*, Vol. 70(6), 2004, 561.
- [10] Turner RA., *Screening methods of Pharmacology*, Academic press Newyork.1965, 22-41.
- [11] Harinantenaina L, Tanaka M, Takaoka S, et al. *Momordica charantia* constituents and antidiabetic screening of the isolated major compounds. *Chem Pharm Bull (Tokyo)* 2006;54:1017-1021.
- [12] Hunt, T.K., “Basic principles of wound healing”, *J Trauma*, 1990, 30, 1990, 122-128.
- [13] Nayak, B.S., Anderson, M. and Pintopereira, L.M. “Evaluation of wound-healing potential of *Catharanthus roseus* leaf extract in rats” *Fitoterapia*, 78, 2007, 540- 544.
- [14] Kokate CK, Purohit AP, Gokhale SB, *A text book of pharmacognosy*, Nirali Prakashan, New Delhi 25 th Ed, 2003,379.
- [15] Pulok K. Mukherjee, “Quality control of Herbal drugs”, *Horizons pharmaceutical publication* 380-422.

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