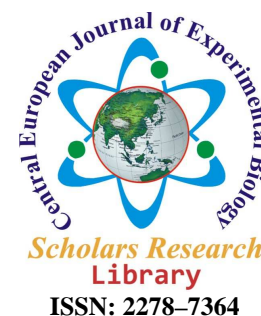




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Central European Journal of Experimental  
Biology, 2012, 1 (1):7-10  
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## Wound healing activity of methanolic extract of leaves of *Gossypium herbaceum*

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### ABSTRACT

The present study was carried out to evaluate the effect of *Gossypium herbaceum* on experimentally induced wounds in rats and compare the effects observed with an antiseptic agent, povidine iodine ointment. The models selected were excision wound, incision wound and dead space wound. In incision and excision models, a significant decrease in period of epithelization and wound contraction was observed in all the treatment groups when compared to control. In the incision wound model, a significant increase in the breaking strength was observed. Granulation tissue formation significantly increased in all treated animals compare to control. It was concluded that extract administered orally (200mg/kg) possesses wound healing activity.

**Keywords:** *Gossypium herbaceum*, Incision wound, Excision wound and Wound contraction.

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### INTRODUCTION

The World Health Organization estimated that 80% of people worldwide rely on herbal medicines for some aspect of their primary healthcare [1]. The aim of herbal treatment is usually to produce persisting improvements in wellbeing. Practitioners often talk in terms of trying to treat the “underlying cause” of disease and may prescribe herbs aimed at correcting patterns of dysfunction rather than targeting the presenting symptoms [2]. Wounds may be defined as loss or breaking of cellular and anatomic or functional continuity of living tissue. In general wounds are classified as acute wound and chronic wound [3]. Wound healing can be defined as a complex dynamic process that results in the results in the restoration of anatomic continuity and function. It is a finely orchestrated and overlapping sequence of events involving vascular response phase/hemostasis, inflammation, proliferation, maturation and remodeling [4, 5]. Many Ayurvedic herbal plants have a very important role in the process of wound healing. Plants are more potent healers because they promote the repair mechanisms in the natural way. So the present study designed to evaluate the wound healing activity of *Gossypium herbaceum*.

## MATERIALS AND METHODS

### Preparation of extracts

The collected leaves were shade dried completely. The dried leaf was then coarsely powdered and was sieved (sieve # 60) to get uniform powdered. The extract was prepared by continuous hot extraction using methanol as a solvent. Extracts obtained was concentrated, dried kept in a desiccators for further use.

### Preliminary phytochemical screening

The methanolic extract of leaves of *Gossypium herbaceum* was screened for the presence of various phytoconstituents like steroids, alkaloids, flavonoids, saponin, mucilage, tannin and phenolic compounds [6].

### Experimental study design

Wistar albino rats weighed about 150-200g were divided into three groups of six rats each. Animals were housed at a temperature of  $23 \pm 2^\circ\text{C}$  and relative humidity of 30–70%. A 12:12 light: day cycle was followed. All animals were allowed free access to water and fed with standard commercial rat chaw pellets.

Group I : Negative control, administer saline 2 ml/kg orally.

Group II: Standard drug povidone iodine ointment applied topically.

Group III: Receive MEGH 200 mg/kg Orally.

### Excision wound [7]

The animals were anesthetized by using ketamine [100 mg/kg, im] and xylazine [16 mg/kg, im]. An impression was making on the dorsal thoracic region 1 cm away from vertebral column and 5 cm away from ear on the anaesthetized rat. The particular skin area was shaved one day prior to the experiment. The skin of impressed area was excised to the full thickness to obtain a wound area of about 500 mm<sup>2</sup>. Haemostasis was achieved by blotting the wound with cotton swab soaked in normal saline. Animals were treated daily with drugs as mentioned above under experimental design from 0<sup>th</sup> day to 15<sup>th</sup> post-wounding day. Wound area is measured on 15<sup>th</sup> days post-wounding for determination of wound contraction and percentage wound contraction was calculated. Falling of scar leaving no raw wound behind was taken as end point of complete epithelization and the days required for this was taken as period of epithelization.

### Incision wound [8, 9]

Para vertebral straight incision of 6 cm length was making 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 was closed by means of interrupted sutures placed at equidistance points about 1 cm apart. Animals were treated daily with drugs as mentioned above under experimental design from 0<sup>th</sup> day to 10<sup>th</sup> post-wounding day. The wound breaking strength was determined on 10th day by tensiometer.

### Dead space wound model [10]

This type of wound was created by implanting subcutaneously 10mg sterilized cotton in the lumber region of dorsal side in anesthetized rats. Animals were treated daily with drugs as mentioned above under experimental design from 0<sup>th</sup> day to 10<sup>th</sup> post-wounding day. On the 10<sup>th</sup> post wounding day, granulation tissue harvested on the implanted cotton was carefully dissected out and dried at 60 °C for 24 h to get a constant weight and weighed.

## RESULTS

### Excision and incision wound model

The results of excision and incision wound model are given in table 1. In this parameter the mean percentage of wound area were calculated 15th post wounding days. In an excision wound model, methanolic extract at a dose 200mg/kg BW p.o. of *Gossypium herbaceum* showed significant wound healing activity (wound contraction on 15th day (120±9.2) compared to control (300± 13.4) and almost equal to standard (118±10.4). It also showed complete epithelization 9.56±0.32 days when compared to control (14.56±0.45). In incision study, the extract showed significant (386±2.7) breaking strength when compared to control (278±3.2).

**Table 1: Effect on period of epithelization, wound contraction and tensile strength in excision and incision wound model**

Group	Excision wound		Incision wound
	Epithelization Period	Wound contraction in mm <sup>2</sup>	Breaking strength (g)
I	14.56±0.45	300± 13.4	278±3.2
II	7.21±0.24*	118±10.4*	425±3.4*
III	9.56±0.32*	120±9.2*	386±2.7*

Significant difference at  $P<0.05$  when compared to control. Values are Mean  $\pm$  SEM from 6 animals in each group.

**Effect of extract of excision wound on 15<sup>th</sup> day**

Figure 1. Control

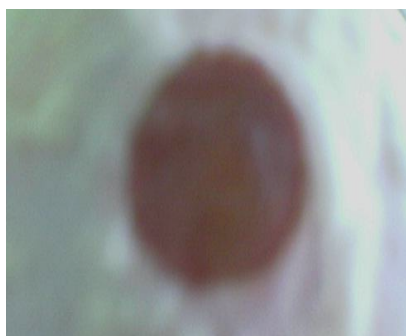


Figure 2. Gossypium extract



Figure 3. povidone iodine



**Effect of extract of incision wound on 10<sup>th</sup> day**

Figure 4. Control



Figure 5. Gossypium extract



Figure 6. povidone iodine



**Dead space wound model**

The results of dead space wound model are given in table 2. The extract showed highly significant increase in wet granuloma (420.23±3.7,  $P<0.05$ ) and dry weight of granulation tissue (99.6±5.6,  $P<0.05$ ) as compared to control (185.65±5.6, 28.7±4.5).

**Table 2: Effect on wet granuloma and dry granuloma tissue weight in dead space wound model.**

Group	Wet granuloma weight (mg)	Dry granuloma weight (mg)
I	185.65±5.6	28.7±4.5
II	386.46±4.7*	85.4±2.3*
III	420.23±3.7*	99.6±5.6*

Significant difference at  $P<0.05$  when compared to control. Values are Mean  $\pm$  SEM from 6 animals in each group.

### DISCUSSION

Wound healing involves various phases. Initially involves acute inflammatory phase followed by the synthesis of collagen and other extra cellular macromolecules, which are later removed to form a scar [11]. Drugs, which influence one phase, may not necessarily influence another. Hence different models have been used in our study to assess the effect of various phases. The treated group of wound showed complete healing of wounds with almost normal architecture of the collagen, reticulin. Increase in tensile strength of treated group wound may be due to increase in collagen concentration. Significant increase in skin breaking strength which was a reflection of increased collagen levels by increased cross linking of collagen fibers. In addition, increase in dry granulation tissue weight indicated the presence of higher protein content [12]. Wound contraction and decrease in period of epithelization in the animals treated with methanolic extract of *Gossypium* may be attributed to their broad spectrum antibacterial activity. The present study shows confirm that methanolic extract of *Gossypium herbaceum* involved in the all the phases of wound healing and the promising wound healing activity may be attributed to presence of different phytoconstituents like flavonoids, tannins etc.

### CONCLUSION

From the study carried out showed that the methanolic extract of *Gossypium herbaceum* possesses a definite wound healing activity, there by justifying its use in the indigenous system of medicine.

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