

EVALUATION OF WOUND HEALING POTENTIAL OF ETHANOLIC LEAVE EXTRACTS OF *Gmelina arborea* ROXB IN WISTAR RATS

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ABSTRACT

Gmelina arborea Roxb. commonly known as Gambhari belong to family (Verbenaceae) is a medicinal tree and its leaves, stem bark, fruits, roots, flowers all are used for medicinal purpose in India since ancient times. Leaf paste is applied to relieve headache and juice is used as wash for ulcers. The present study was an attempt to investigate the wound healing potential of *Gmelina arborea* leaf extract in two different types of wound models in albino rats viz., incision and excision. The various leaf extracts of *G.arborea* such as ethanol, ethyl acetate, methanol and petroleum ether were obtained by successive solvent extraction. The standard drug (povidone iodine ointment) applied topically and all the extracts (250 mg/Kg) of *G.arborea* leaves were given orally. The ethanolic extract showed significant increase in wound contraction and formation of scar in excision wound model. The extract showed significant increase in the breaking strength of resutured incision wound as compared to control group ($p < 0.05$). The result of the present study indicate that the ethanolic extract of *G.arborea* leaf shows more significant wound healing property than the other three extracts in excision and incision wound model.

KEYWORDS: *Gmelina arborea*, povidone iodine ointment, incision and excision, wound models.

INTRODUCTION

In the last few decades there has been an exponential growth in the field of herbal medicine. It is getting popularized in developing and developed countries owing to its natural origin and lesser side effects.^[1] About 80% of people in developing countries depend on traditional systems of medicine for primary health care.^[2] The aim of wound care is to promote wound healing in the shortest time possible, with minimal pain, discomfort, and scarring to the patient and must occur in a physiologic environment conducive to repair and regeneration.^[3] *Gmelina arborea* Roxb. (Verbenaceae) belongs to a genus of trees and shrubs distributed chiefly in South East Asia, tropical Australia and tropical Costa Rica.^[4,5] *Gmelina arborea* Roxb. locally named as Gambhari (Oriya), Gambhar (Hindi), Gambhar (Bengali), Sriparni (Sanskrit) and Gummadi (Telgu).^[6] Flowering takes place during february to april when the tree is more or less leafless whereas fruiting starts from May onwards up to June. Flowers occur in narrow branching clusters at the end of branches. The yellow flower, tinged with brown, is trumpet shaped, 3-4 cm long. The trumpets flare open into a gaping mouth with 5 distinct lobes.^[7] The root of this plant has been used in traditional Indian systems of medicines as a demulcent, stomachic, bitter tonic, refrigerant, laxative, and galactagogue. The tender leaves are used as demulcent,

in headache, fevers, gonorrhea, cough etc. The whole plant is used in snake bite and scorpion sting throughout India.^[8] Ayurvedics prescribe them for alopecia, anemia, consumption, leprosy, thirst, and vaginal discharges; the flowers for blood disorders and leprosy; the root, deemed anthelmintic, laxative and stomachic, for abdominal pains, burning sensations, fever, hallucinations, piles and urinary discharges.^[9,10] According to scientific studies, the root decoction is used as a folk remedy for abdominal tumors. The roots are useful in hallucination, piles, abdominal pains, fevers, 'tridosha' and urinary discharge.^[11,12] Traditional people are using to get relieve from Post-delivery weakness, they are using half glass of boiled root extract. The extract is prepared by boiling roots with one glass of water till it gets reduced to half a glass. The plant has also been reported to have anti-inflammatory activity hypoglycaemic and anti-viral activities against Ranikhet disease virus.^[13]

MATERIALS AND METHODS

Collection of Plant Material

The leaves of *Gmelina arborea* were collected from the campus of Jeypore college of pharmacy, Jeypore, Koraput district.(India) in the month of August 2017. The plant was identified, confirmed and authenticated by the Biju Patnaik Medicinal Plants Garden and Research Centre, Jeypore, Koraput, Orissa,(Letter No. MJ/SS/P-

407/17, dated (6.8.2017). After authentication leaves were collected in bulk and washed under running tap water to remove adhering dirt. Then the leaves were shade dried and the dried leave materials were made into coarse powder by grinding in mechanical grinder then stored in a closed air tight container for further use.

Preparation of Extracts

The coarse powder was taken in Soxhlet apparatus and extracted successively with ethanol, ethyl acetate, methanol and pet ether as solvent. A total amount of 550g coarse powder was extracted with 1000 ml of each solvent. For each solvent, 10 cycles were run to obtain thick slurry. Each slurry was then concentrated under reduced pressure to obtain the crude extract. All crude extracts were kept in closed air tight containers under cool and dark place for further study.^[14,15]

Animals

Healthy adult wister strain of albino rats weighing approximately 180 to 200g were used for study. They were housed in standard conditions of temperature (25 ± 2 °C), 12 hours light per day cycle, relative humidity of 45-55 % in animal house of Jeypore College of Pharmacy. They were fed with standard pellets of food and water. Animals were kept and all operation on animals was done in aseptic condition. All the studies conducted were approved by the Institutional Animal Ethical Committee (1906/PO/Re/S/16/CPCSEA), Jeypore college of pharmacy, Jeypore, Odisha according to prescribed guide-lines of the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Government of India.

Wound healing Activity

Excision wound model

For the excision wound healing study, animals were divided into 6 groups of six rats in each group. Group-I was served as control and received only saline water 2ml/Kg orally, Group-II were given standard drug (povidone iodine ointment) applied topically The ethanol, ethyl acetate, methanol and pet ether extracts (250 mg/Kg) of *Gmelina arborea* leaves were given orally to Group-III, Group-IV, Group-V and Group-VI respectively. An impression was made on the dorsal thoracic central region 5mm away from the ears, by using a round seal of 2.5 cm diameter as described by Morton and Malone [16]. The skin of the impressed area was excised to the full thickness to obtained area of about 500 mm² under light ether anaesthesia in aseptic condition. The animals were housed individually. The methanol extracts in simple ointment base (5% w/w) were applied on the wound once a day for 18 days starting from the day of wounding. The percentage wound closure was observed on 4th, 8th, 12th, 16th, 18th post wounding day. Epithelization time (in days) and size of the scar area was noted.

Incision wound model

Incision wound model was performed according to Ehrlich and Hunt.^[17] In the incision wound healing model, the animals were divided into 6 groups of six rats in each group, and kept in separate cage. Group-I was served as control, received only saline water 2ml/Kg orally, Group-II were given standard drug (povidone iodine ointment) applied topically, the ethanol, ethyl acetate, methanol and petroleum ether extracts (250 mg/Kg) of *Gmelina arborea* leaves were given orally to Group-III, Group-IV, Group-V and Group-VI respectively for 10 days Under light ether anesthesia, the animals were secured to operation table in its natural position. Two paravertebral straight incisions of 6 cm each were made through the entire thickness of the skin, on either side of the vertebral column with help of sharp blade. Removal of the sutures was done on 8th post wounding day. Tensile strength was determined on both wounds by continuous constant water flow technique of Lee.^[18]

Statistical analysis

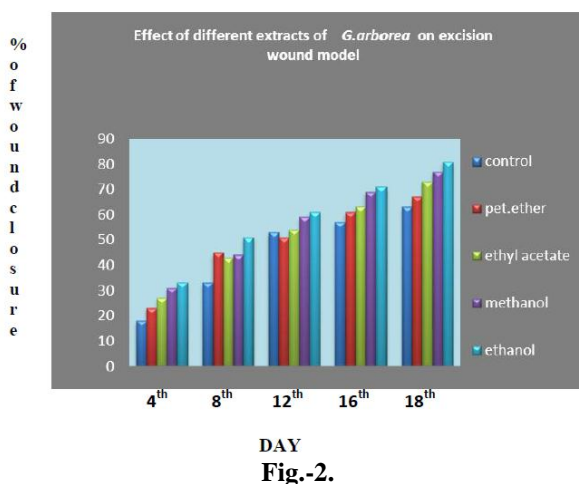
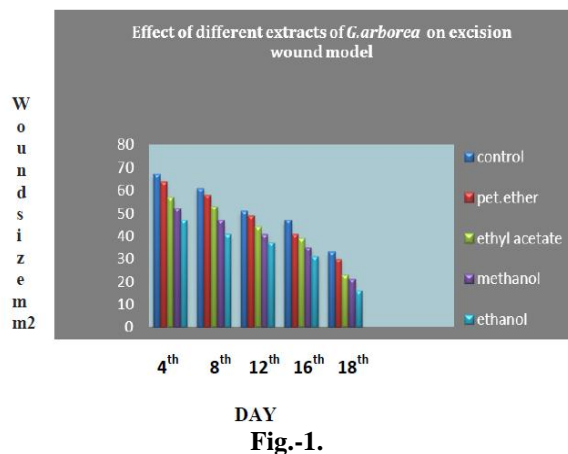
The results are reported as Mean \pm SE. Statistical analysis was done using ANOVA (Tukey-Multiple Copmarision Test). When probability (p) was less than 0.05 was considered as significant.^[19]

RESULT AND DISCUSSION

In the study using excision wound model, animals treated with ethanol extract of *Gmelina arborea* leave showed significant decrease in epithelization period as evidenced by shorter period for fall of eschar as compared to control group ($p < 0.05$) (Fig-1). The extract also facilitated the increase in rate of wound contraction than control group. The petroleum ether extract treated animal (Group-II) showed wound contraction by 63.26%. The ethyl acetate extract treated animals (Group-III) showed wound contraction by 71.31%. The methanol extract treated animal (Group-IV) showed wound contraction by 78.21%, The ethanol extract treated animal (Group-V) showed wound contraction by 83.33% as compared with the control (Group-I) by 63.27%. In all the extract. (Fig-2) The result of present study reveals that ethanolic leave extracts of *G. arborea* possess a prominent prohealing activity in incision wound model. This was demonstrated by significant increase in the skin tensile strength in methanol extract treated groups ($p < 0.05$) on 10th post wounding day are presented in (Table-1).

Table 1: Effect of extracts of *G. arborea* leave on the breaking strength in incision wound.

Sl. no.	Group	Breaking strength
1	Control	239.12 \pm 22.06
2	petroleum ether	327.14 \pm 11.37
3	ethyl acetate	336.22 \pm 11.47
4	methanol	347.12 \pm 141.21
5	ethanol	426.47 \pm 16.22*



CONCLUSION

In the present study, the wound healing activity of *G. arborea* leaf was carried out and the results of the present study suggest that local application and systemic administration of ethanol extract of the leaf has shown more significant wound healing activity in excision and incision wound models and the popular use of plant to open wound in folk medicine. The wound healing property of *G. arborea* leaf has been attributed to its antimicrobial effects. However, further investigation employing isolation of constituents and screening models are needed for further confirmation of wound healing potential of *G. arborea* leaf. Thus The traditional use has been pharmacologically validated.

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