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PHYTOCHEMICAL SCREENING AND EVALUATION OF WOUND HEALING POTENTIAL OF ETHANOLIC FLOWERS EXTRACT OF *BAUHINIA ACUMINATA* L. IN WISTAR RATS

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ABSTRACT: The present study was carried out to evaluate the wound healing potential of ethanolic flowers extract of *Bauhinia acuminata* in two different types of wound models in Albino rat viz., incision and excision, and compare the effects with standard drug povidine iodine ointment. The ethanolic extract at a dose of (250 mg/kg) possesses a significant increase in wound contraction and formation of scar in an excision wound model. The extract showed a significant increase in the breaking strength of resutured incision wound as compared to the control group ($p < 0.05$). The result of the present study indicates that ethanolic flowers extract of *B. acuminata* possesses significant wound healing in excision and incision wound model. The presence of phytoconstituents like flavonoids, saponins, phenols either individually or combined may exhibit the synergistic effect towards the healing of wounds.

INTRODUCTION: Herbal medicine has become an integral part of standard health care. The World Health Organization estimated that 80% of people worldwide rely on herbal medicines for some aspect of their primary healthcare¹. 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^{2,3}. Many Ayurvedic herbal plants have a very important role in the treatment of the wound. Plants are more potent healers because they promote the repair mechanisms naturally. So the present study undergoes to evaluate the wound healing activity of flowers of *B. acuminata*.

The tribal areas of Baipariguda, Koraput (District) of Eastern Orissa, due to its unique varieties geographical and climatic factors have had a wide variety of medicinal plant. *Bauhinia acuminata* (Family: Fabaceae) is a species of a flowering shrub native to tropical Southeastern Asia. Common names white *Bauhinia*. The genus *Bauhinia*, one of the largest genera in sub-family Caesalpiniaceae, represent more than 300 species⁴. *Bauhinia* has been extensively planted as a garden, park, and roadside ornamental tree in many warm temperate and subtropical region⁵. The bark, flower, and root of the *B. acuminata* are used for various skin diseases, worms, tumours, and diabetes⁶.

In Malaysia and Indonesia, the plant is used in the treatment of common cold and cough⁷. While in India, the leaves and bark of this plant are used for treating asthma⁸. Moreover, the leaf of *B. acuminata* is used to treat bladder stone, venereal diseases, leprosy, asthma, and digestive diseases⁹. A different part of this plant such as bark, leaves, stem, flowers, and roots have been used in traditional medicine¹⁰.

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Leaves were used externally and internally in skin disease scabies, and the leaves/root were an ingredient of many popular herbal liver tonic and medicines for liver disorders. People use it also for the treatment of insect bites, snake bite, scorpion sting, constipation, edema, fever, inflammation, rheumatism¹¹. Its roots, leaves, flowers and seeds were used as a laxative and purgative¹². The chemical constituents found in *B. acuminata* were vitamin C (leaves), beta-sitosterol, lupeol, kameferol. Several chemical compounds including palmitic acid, three phthalic acid esters, phthalic acid, gallic acid, ursolic acid, were identified from the leaves of *Bauhinia acuminata*¹³.

MATERIALS AND METHODS:

Collection of Plant Material: The flowers of *Bauhinia acuminata* were collected from the tribal belts of the local area of Baipariguda, Koraput district (India) in December 2018. The plant was identified, confirmed and authenticated by the Biju Patnaik Medicinal Plants Garden and Research Centre, Dr. M. S. Swami Nathan Research Foundation, Jeypore, Koraput (District), Orissa (Letter no. MJ/SS/P-302/18, dated (16.12.2018)). After authentication flowers were collected in bulk and shade dried. The dried materials were made into coarse powder by grinding in a mechanical grinder and stored in a closed airtight container for further use.

Preparation of Extracts: The coarse powder of flowers was taken in Soxhlet apparatus and extracted successively with ethanol as solvent. A total amount of 550 g coarse powder was extracted with 1000 ml of solvent, and 10 cycles were run to obtain thick slurry, and the slurry was then concentrated under reduced pressure to obtain crude extract and kept in closed air tight containers under cool and dark place for further study¹⁴.

Preliminary Phytochemical Screening: The crude ethanolic extracts of the flower of *Bauhinia acuminata* were subjected to preliminary phytochemical analysis to detect the presence of various groups of phytoconstituents by carrying out the chemical analysis¹⁵.

Animal: Healthy adult Wistar strain of albino rats weighing approximately 180 to 250 gm were used.

They were housed in standard conditions of temperature (25 ± 2 °C), 12 h light per day cycle relative humidity of 45-55 % in the 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.

Experimental Protocol: Animals were selected, weighed (25-30 g) and divided into three groups (n=6), namely control, standard drug, and one test groups belonging to *B. acuminata* flower extract. The studies conducted were approved by the Institutional Ethical Committee (1906/PO/Re/S/16/CPCSEA), Jeypore College of Pharmacy, Jeypore, Odisha according to prescribed guidelines of the Committee for Control and Supervision of Experiments on Animals (CPCSEA), Government of India.

Wound Healing Activity:

Wound Model:

Excision Wound: For the excision wound study, animals were divided into three groups of six rats in each group. Group-I served as control and received only saline 2 ml/kg orally, Group-II served as Standard drug (povidone iodine ointment) applied topically. The ethanol extracts (250 mg/kg) were given orally to Group- III of *B. acuminata* flower extract. An impression was made on the dorsal thoracic central region 5 mm away from the ears, by using a round seal of 2.5 cm diameter as described by Morton and Malone.

The skin of the impressed area was excised to the full thickness to the obtained area of about 500 mm² under light ether anesthesia in the aseptic condition the animals were housed individually. The animals were treated daily with drugs as mentioned above under experimental design from 0th day to 18 days starting from the day of wounding. The percentage of wound closure was observed on 4th, 8th, 12th, 16th, 18th post wounding day. Epithelization time (in days) and the size of the scar area was noted¹⁶.

Incision Wound: In the Incision wound model, the animals were divided into three groups of six rats in each group, and kept in a separate cage. Group-I served as the control, received only saline 2ml/kg orally, Group-II served as a standard drug

(povidone iodine ointment) applied topically, the ethanol extracts (250 mg/kg) were given orally to Group- III of *B. acuminata* flower extract. Under light ether anesthesia, the animals were secured to the 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 the help of a sharp blade.

After complete homeostasis, the wound was closed using interrupted sutures placed at equidistance points about 1 cm apart. Animals were treated daily with drugs as mentioned above under experimental design from 0th day to 10th post-wounding day. The wound breaking strength was determined on the 10th day by continuous constant water flow technique of Lee¹⁷.

Statistical Analysis: The results are reported as Mean \pm SE. Statistical analysis was done using ANOVA (Tukey-Multiple Comparison Test). When probability (p) was less than 0.05 was considered as significant¹⁸.

RESULTS AND DISCUSSION: The preliminary phytochemical screening showed that the solvent extracts of the flower of *Bauhinia acuminata* contain carbohydrate, flavonoids, phenolic compound, fat and oils and saponins whereas alkaloid, glycoside and tannins are absent. In the study using excision wound model, animals treated with ethanol extract *B. acuminata* flowers showed a significant decrease in an epithelization period as evidenced by shorter period for fall of eschar as compared to control group (p<0.05) **Fig. 1 & 2**.

TABLE 1: PHYTOCHEMICAL SCREENING OF FLOWERS EXTRACTS OF *B. ACUMINATA*

Extracts		Phytochemicals						
Ethanol	Alkaloids	Flavonoids	Glycosides	Saponins	Tannins	Phenolic compounds	Carbohydrate	Fat & Oils
	--	++	---	++	---	++	++	++

+++ , Strong; ++, moderately; +, poor presence, -- Absent

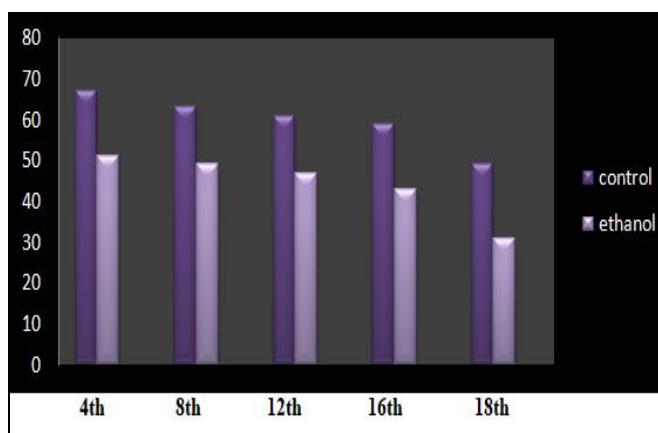


FIG. 1: EFFECT OF FLOWERS EXTRACT OF *B. ACUMINATA* ON EXCISION WOUND MODEL

The extract also facilitated the increase in the rate of wound contraction than the control group. The ethanol extract showed wound contraction by 81.33% as compared with the control (Group-I) by 67.24%.

TABLE 2: EFFECT OF FLOWER EXTRACTS OF *B. ACUMINATA* ON THE BREAKING STRENGTH IN INCISION WOUND

S. no.	Group	Breaking strength
1	Control	243.11 \pm 27.02
2	Ethanol	335.14 \pm 14.31*

Values are mean \pm SE (n=6)* p<0.05 vs. Control.

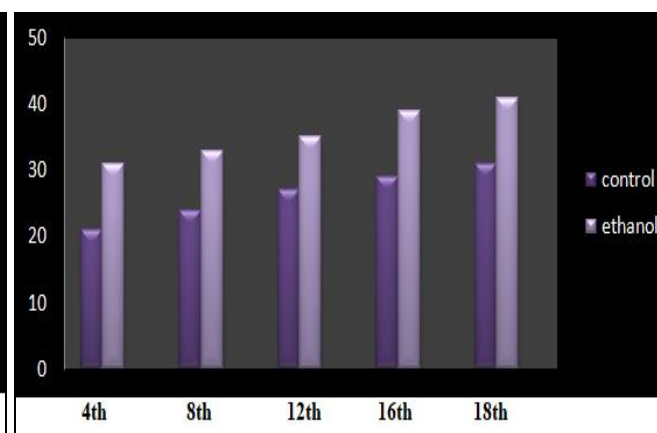


FIG. 2: EFFECT OF FLOWERS EXTRACT OF *B. ACUMINATA* ON INCISION WOUND MODEL

The result of the present study reveals that ethanolic flowers extracts of *B. acuminata* possess a prominent pro-healing activity in an incision wound model. This was demonstrated by a significant increase in the skin tensile strength in methanol extract treated groups (p<0.05) on 10th post-wounding day are presented in **Table 2**.

CONCLUSION: The results of the present study suggest that local application and systemic administration of ethanol extract of the flowers has shown significant wound healing activity in excision and incision wound models and support

the popular use of the plant to open wound in folk medicine.

The wound healing property of *Bauhinia acuminata* has been attributed to its antimicrobial effects. The presence of phytoconstituents like flavonoids, saponins, phenols either individually or combined may exhibit the synergistic effect towards the healing of wounds.

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CONFLICT OF INTEREST: Nil

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