

FORMULATION AND EVALUATION OF OINTMENT CONTAINING NATURAL WOUND HEALING ACTIVITY OF *TRIDAX PROCUMBENS*

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ABSTRACT: The main objective of the work was to formulate and evaluate ointment of *Tridax procumbens* to analyze its wound healing activity. Various phytochemicals were extracted by solvent extraction using Soxhlet apparatus and these phytochemicals were qualitatively analyzed. Herbal ointment containing hydroalcoholic leaf extract of *Tridax procumbens* was formulated and tested for wound healing activities by topical application. Ointment applied on the excision wound in mice caused significantly higher rate of wound healing and showed the highest rate of wound closure as compared to the blank ointment and should be explored in harnessing the potential of plant in treatment of topical diseases.

Keywords- *Tridax procumbens*, herbal ointment, wound healing, Excision wound.

INTRODUCTION

The Compositae (Asteraceae) is an advanced and botanically highly specialized family of herbaceous plants, herbs, shrubs, or less commonly trees and are arguably the largest family of flowering plants, comprising about 1400 species out of which 674 species are found in India. Some of them are tropical trees and shrubs while few are members of herbs. *Tridax procumbens* is a species of flowering plant in the daisy family (Compositae) a common weed in west Africa, sub-region and other tropical zones of world and known as coat button in English, Jayanti Veda in Sanskrit, Ghamra in Hindi and Dagadi Pala in Marathi. It is best known as widespread weed and pest plant can be found in fields, meadows, croplands, disturbed areas, lawns and roadsides. It is a semi prostrate annual creeper herb. Stem is ascending 30-50 cm height, branched, sparsely hairy, rooting at nodes. Leaves are simple, opposite, stipulate, lanceolate to ovate. 3-7 cm long irregularly toothed margin, base wedge shaped, short petiole, hairy on both surfaces. ^[1] The plant is commonly used in Indian traditional medicine as hair tonic, anticoagulant, antifungal and insect repellent, in bronchial catarrh, diarrhoea, dysentery and wound healing. Previously isolated constituents, alkyl esters, sterols, pentacyclitriterpens, fatty acids and polysaccharides.

MATERIAL AND METHOD

Collection of plant material

The fresh leaves of *Tridax procumbens* were collected in the month of January from local area in Bhandara district, India. The plant was authenticated by Prof. Dr. Mahesh Kawle, Department of botany, D.B. Science College, Gondia, and herbarium was deposited for further study. Collected leaves were washed and shade dried and used for the further investigation.

Preparation of extract

The dried plant samples were grinded using mechanical grinder to coarse powder and passed through 40 micron sieve. The powdered drug was extracted by soxhlation and dried under vacuum.

Preparation of ointment

The constituents of base were weighed and melted in a beaker at 70° C using heating burner. The *Tridax procumbens* extract was formulated into the base to attain in the final concentration of 60 mg, 80 mg and 100 mg /g. The formulated ointments were stored at 4°c, till further use.

Wound Healing activity:

- Swiss male albino mice weighing 25-30 g or either sex were used in the study. Animals were procured from Laboratory Animal House of Bajiraoji Karanjekar College of Pharmacy, Sakoli, Dist. Bhandara, Maharashtra
- All animal experiment strictly complied with approval of the Institutional Animal Ethical committee. The animals were kept in polyacrylic cages and maintained under standard housing conditions of temperature (24-27° c) and humidity (60-65/ %) with 12:12 light: Dark cycles. They were acclimatized for 7 days. The food was provided in the form of dry pellets and water ^[9]

Study design for wound healing activity

- Excision wound models were used to assess the wand healing profile of leaf. Extract of *Tridax procumbents* which was formulated in ointment base. The drug treatments were as follows. Six mice were taken for each group.
- Group A: - Received ointment base and served as normal control
- Group B: - Received reference standard Beta dine Ointment (100 mg/gm)
- Group C: - Received *Tridax procumbens* ointment (80 mg/gm)
- Group D: - Received *Tridax procumbens* ointment (100 mg/gm)

Mechanism of wound Healing Activity

The extract of *Tridax procumbens* linn leaves has ability to set the normal and immune compromised wound healing in mice The wound healing process by application of this extract material involves complex interaction between epidermal and dermal cells, the extra cellular matrix controlled angiogenesis and plasma designed protein all coordinated by an array of cytokines and growth factor the plant not only increase lysyl oxidase but also protein and nucleic acid content in the granulation tissue, probably due to increase of glycosamino-glycan content.

Preliminary evaluation of formulation

1. Colours and odour – Colour and odour of prepared ointment was examined by visual examination.
2. pH- the pH of ointment was determined by digital pH meter 1g of ointment was dissolved in 50 ml of distilled water and ph was measured.
3. Spreadability – Spreadability was determined by the apparatus which consist of a wooden block, which was provided by a pulley at one end.

Microbial growth

Nutrient agar media was used in microbial growth study in this method the blank and sample petriplates were used and ointment sample were aseptically transered on to the sample plates in a cross pattern.

RESULTS

Physicochemical Parameters

Parameter	Result
Description	Green
Total ash	12 %
Water soluble ash value	4.05 %
Acid insoluble ash value	2.35 %
Water extractive	2.8 %
Alcohol Extractive	2.4 %
Loss on drying	4.66 %

Table 1. Physicochemical Parameter

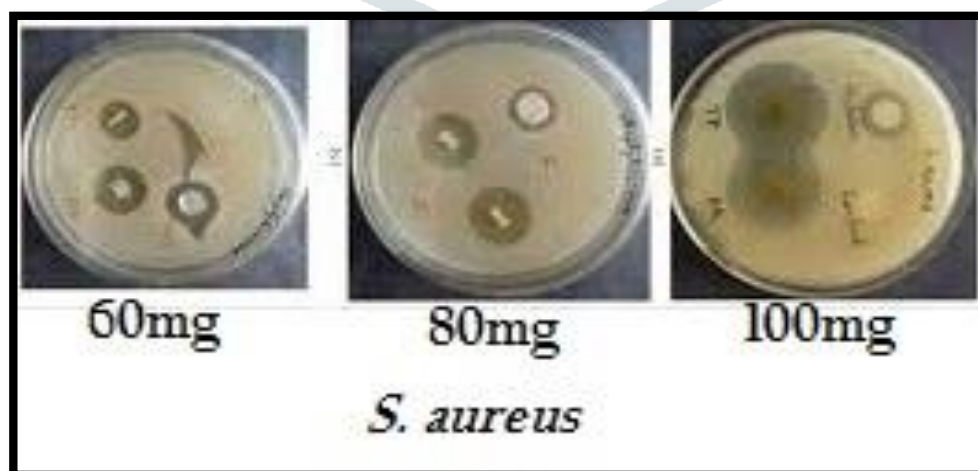
Ointment Preparation:

Sr. No.	Ingredients	F1 (80mg/g) in %	F2 (100mg/g) in %
1.	Sodium Lauryl sulphate	0.85%	-
2.	Propylene glycol	10.32%	-
3.	Stearyl alcohol	21.42%	-
4.	White petrolatum	21.42%	-
5.	Honey	14.28%	-
6.	<i>Tridax procumbens</i> extract	8%	10%
7.	Propyl paraben	0.05%	-

Table 2. Ointment preparation

Antibacterial activity

The antibacterial activity of all the ointment containing extract was tested by well- diffusion using pour plate method against *staphylococcus aureus* (gram+ve) and *Escherichia coli* (gram-ve) .

Fig. 1- The antibacterial activity of ointment containing extract against *Escherichia coli*Fig. 2- The antibacterial activity of ointment containing extract against *staphylococcus aureus*

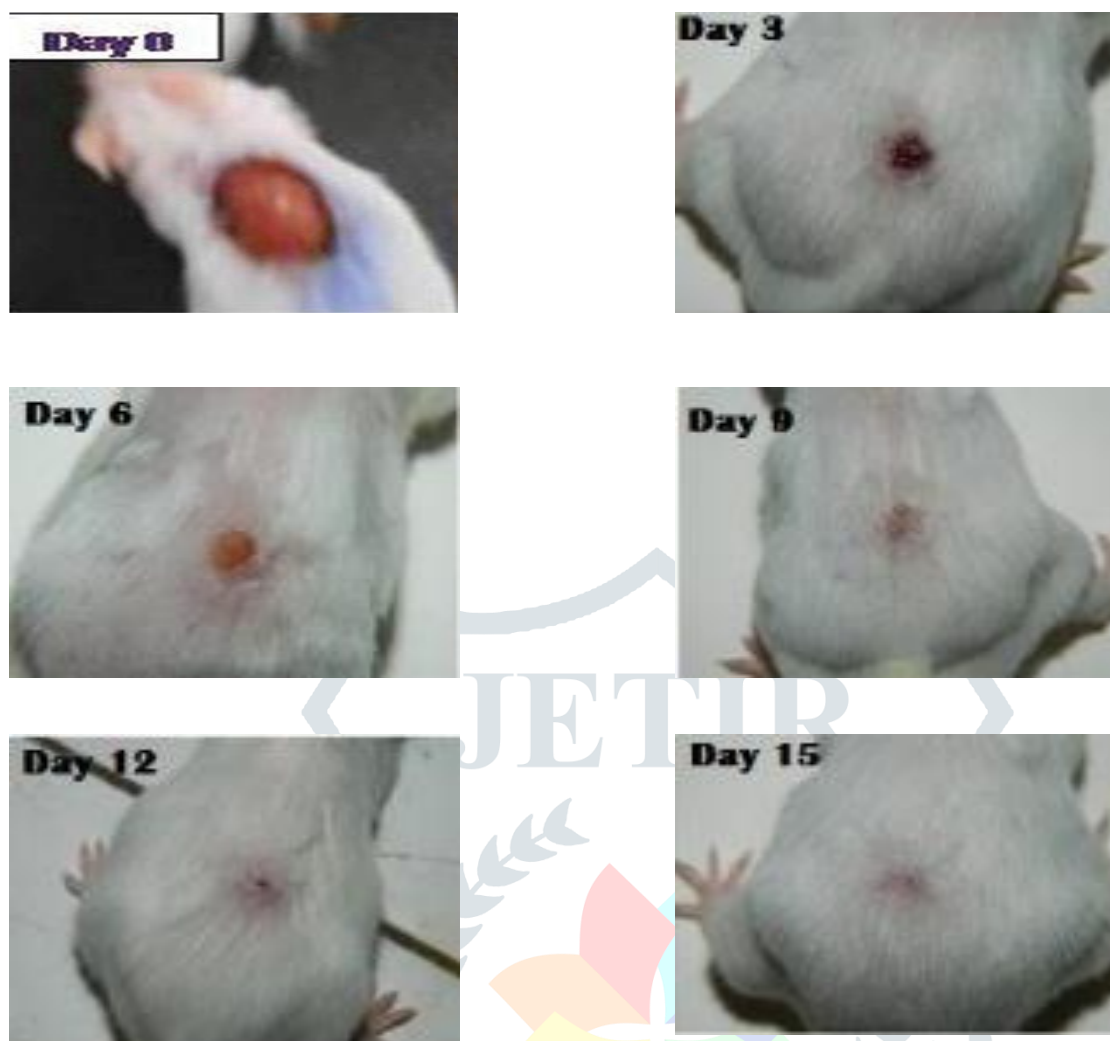


Fig 3- Wound contraction with ointment (100mg)

Preliminary evaluation

pH of the formulation:

The pH of ointment was determined by digital pH meter 1 gm of ointment was dissolved in 50 ml distilled water & the pH was measured.

Sr. No.	Batch	pH
1	F1 (80 mg/gm)	6.4
2	F2 (100 mg/gm)	6.8

Spreadability of formulation

Sr. No.	Batch	Spreadability
1.	F 1 (80 mg/gm)	12.14
2.	F 2 (100 mg/gm)	13.15

CONCLUSION

In Indian system of medicine of herbal product are made by using crude plant or portion of plant parts and their extracts. The leaves extract of *Tridax procumbens* plant belongs to family Asteraceae was taken for this present study and formulated for the topical ointment and its properties. The Ointment prepared using *Tridax procumbens* leaf extracts was found to be good ointment characteristics with respect to homogeneity, spreadability, pH, viscosity, microbial growth, antimicrobial activity & wound healing activity. The ointment prepared using *Tridax procumbens* leaves ethanolic extract exhibited strong antimicrobial activity & especially with 100 mg/g of the extract concentration in the ointment. The result of different chemical and physical tests of ointment showed that the formulation could be used topically in order to protect skin against damage caused by microorganism. The ointment prepared using *Tridaxprocumbens*ethanolic extract shows significant wound healing activity and it significantly help in the wound contraction. Especially with 100 mg/g concentration. But stdbetadine shows more wound contraction at same concentration compare with test. The ointment was stable even after two months. Also *Tridax procumbens* is used for their hepatoprotective, immunomodulatory effect, antidiabetic and anticancer activity. Thus it can be concluded that there is a growing demand for herbal formulation in the world market and they are invaluable gift of nature. Herbal medication considered safer than allopathic medicine as allopathic medicine are associated with the side effects. Similarly this ointment can be tested for skin related bacterial infection & also can be used for commercial production of wound care ointment.

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