

## STUDY ON WOUND HEALING ACTIVITY BY USING PLANT *LANTANA CAMARA* AND *TRIDAX PROCUMBENS*

Ganesh Vivek Kalyankar\*, Vishnukanth Sambhaji Kadam, Karan Vitthal Kalaskar,  
Renuka Yogaji Kale, Bhuvan Ashok Kamble, Meena Pandurang Pisal

D.K. Patil Institute of Pharmacy, Sayal Road, Loha.

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### \*Corresponding Author

Ganesh Vivek Kalyankar

D.K. Patil Institute of Pharmacy, Sayal  
Road, Loha.



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### ABSTRACT

Wound healing is a complex biological process involving inflammation, tissue formation, and remodelling. Traditional medicine has long utilized various medicinal plants for enhancing wound healing. In this study, the wound healing potential of two ethnomedicinal plants, *Lantana camara* and *Tridax procumbens*, was investigated through in vivo models. Extracts from the leaves of both plants were prepared using solvents such as ethanol and aqueous solutions. The phytochemical analysis revealed the presence of flavonoids, tannins, saponins, and terpenoids compounds known to promote tissue regeneration and possess antimicrobial properties. Wistar rats were subjected to excision and incision wound models, and the rate of wound contraction, epithelialization period, and tensile strength were evaluated over a 14-day period. The results showed a significant

improvement in wound contraction and healing rate in groups treated with the combined extracts, compared to the control and standard treatment groups. The synergistic effect of *L. camara* and *T. procumbens* is attributed to their anti-inflammatory, antioxidant, and antimicrobial properties. This study supports the traditional use of these plants and suggests their potential as natural, cost-effective alternatives in wound management.

**KEYWORDS:** Wound healing, *Lantana camara*, *Tridax procumbens*, herbal therapy, anti-inflammatory activity, collagen synthesis.

## INTRODUCTION

### Definition of Wound

A wound is defined as any tissue injury that disrupts anatomical integrity and leads to functional loss. The ability of an organism to repair or regenerate tissues is a definite advantage for survival.<sup>[1]</sup>



**Fig. no. 1: Skin wound.**

Any violation of live tissue integrity may be regarded as a wound. Skin is the largest organ of the human body and one of its key functions is to protect water-rich internal organs from the dry external environment<sup>[2]</sup> Maintaining skin integrity and possessing a robust wound healing capacity are key prerequisites for healthy survival. Furthermore, wound healing can also present a significant challenge and burden on health care systems. Medicare cost estimates for acute and chronic wound treatments ranged from \$28.1 billion to \$96.8 billion during 2014.<sup>[3]</sup> The highest wound-related expenses were attributed to surgical wounds followed by diabetic foot ulcers.<sup>[4]</sup>

### Phases of Wound Healing

Skin epithelial cells are labile elements that are continuously eliminated in the stratum corneum through the keratinocyte desquamation process and are replaced, in the basal layer, by differentiated elements derived from stem cell proliferation and differentiation.

Cell renewal varies according to different factors, such as trauma, hormonal influences, skin conditions and individual wellbeing. However, the cutaneous regenerative process, in reference to a wound lesion, is inversely proportional to the evolution of the considered species.<sup>[5]</sup>

## PLANT PROFILE

### *LANTANA CAMARA*

*Lantana camara* belongs to Verbenaceae family. *Lantana camara* is commonly known as lantana. The genus lantana consists of 150 pantropical species used as traditional medicines all around the world. Some taxa of lantana plant are also found in Africa and tropical Asia. Now, it grows approximately in 50 different countries of the world.<sup>[6]</sup>



Fig no. 2: *Lantana camara*.

### Taxonomical classification<sup>[7]</sup>

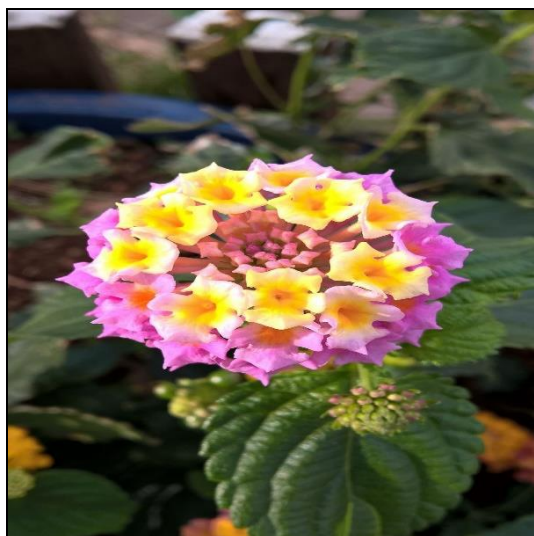
<b>Botanical Name</b>	<i>Lantana Camara</i> Linn
<b>Common Names</b>	Lantana Weed, Raimuniya, Ghaneri, Tantani
<b>Family</b>	Verbenaceae
<b>Plant Form</b>	Shrub
<b>Therapeutic Uses</b>	Plant pacifies vitiated condition of vata and kapha.

### Phytochemistry of *Lantana camara*

The constituents of essential oil of *Lantana camara* are Sabiene (19.6- 21.5%), 1, 8- Cineole (12.6- 14.8%),  $\beta$ -caryophyllene (12.7-13.4%),  $\alpha$ -humulene (5.8-6.3%), two rare sesqui terpenoids humulene epoxide-III and 8-hydroxy bicyclogermacrene<sup>[7]</sup>, 1, 8-cineol (15.8%), sabinene (14.7%) and caryophyllene (8.9%).<sup>[8]</sup>



### Various parts of *lantana camara*



**Fig.no.3 Flower of *L. camara*.**



**Fig.no.4 Stem of *L. camara*.**



**Fig.no.5 Leaves of *L. camara*.**



**Fig.no.6 Fruits of *L. camara*.**

### Medicinal properties of *L. camara*

- 1) Antibacterial activity:** Ethanolic extracts of *L. camara* leaves and roots were reported for antibacterial activity. The in vitro antibacterial activity was performed by microdilution method. The extracts exhibited antimicrobial activity against *Staphylococcus aureus*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Vibrio cholerae*, *Escherichia coli* and two multi resistant strains *E. coli* and *S. aureus*.<sup>[9,10]</sup>
- 2) Antifungal activity:** Antifungal activity of ethanol and hot water extract of *L. camara* was screened against wood destroying white and brown rot fungi. Both extracts exhibited efficient antifungal activity against white and brown rot fungi, however ethanol extract was highly potential at very low concentration (0.01%).<sup>[11,12]</sup>

- 3) **Anti-inflammatory activity:** Aqueous extract of *L. camara* was reported for anti-inflammatory activity in albino rats. Extract treatment (500mg/kg body weight) significantly decreased paw volume in carrageenan induced paw oedema test in rats.<sup>[13]</sup>
- 4) **Wound healing activity:** Wound healing property of ethanol extract of leaf of *L. camara* was reported in adult male Wister rats. Topical application of the extract over the wound significantly increased the wound healing activity. Topical application of the extract on the wound (100 mg/kg/day) significantly enhanced the rate of wound contraction (98%), synthesis of collagen and decreased wound healing time.<sup>[14]</sup>
- 5) **Antimotility activity:** Methanol extract of *L. camara* leaves was reported to possess antimotility activity in mice. Intestinal motility was assayed by charcoal meal test in mice. At a dose of 1 g/kg body weight, the extract completely inhibited the transit of charcoal in normal mice.<sup>[15]</sup>
- 6) **Anti-filarial activity:** Anti-filarial activity of crude extract of *L. camara* stem was reported. The extract and its chloroform fraction resulted in the death of adult *Brugia malayi* and sterilised most of the surviving female worms in the rodent model *Mastomys coucha*.<sup>[16]</sup>

### ***TRIDAX PROCUMBENS***

It is a native to the tropical Americas, but it has been introduced to tropical subtropical, and mild temperate regions worldwide, it is often rooting at node solitary, long stalked, yellow composite, heterogamous, bisexual flower with white flowing heads and very hairy, with coarsely toothed, petiolate, ovate or lanceolate leaf whole arial part is useful medicinally, leaves possesses wound healing, insecticidal, antisecretory and hypotension action, while seeds are used to control bleeding.<sup>[17]</sup>

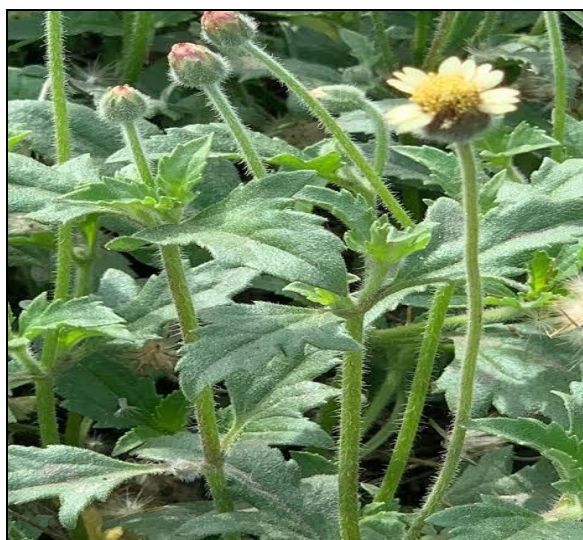


**Fig no. 7: *Tridax Procumbens*.**

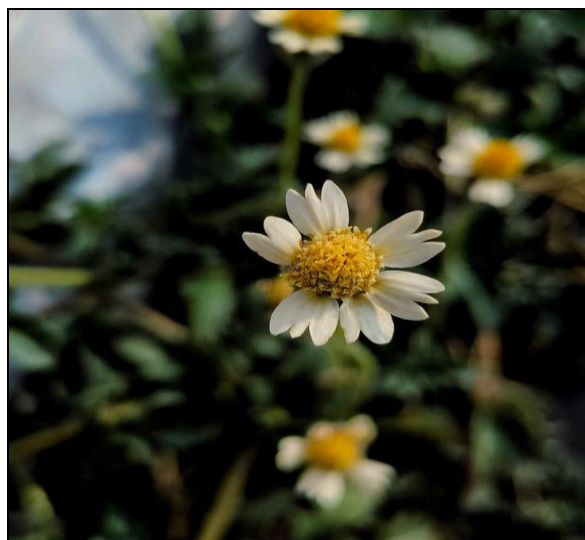


Scientific classification<sup>[18]</sup>

<b>Kingdom</b>	Plantae
<b>Phylum/Division</b>	Magnoliophyte
<b>Class</b>	Eudicots
<b>Order</b>	Asterales
<b>Family</b>	Asterales
<b>Genus</b>	Tridax
<b>Species</b>	<i>Tridax Procumbens</i>
<b>Common Name</b>	+Chrysanthemum Procumbens, Balbisia Canescens, Balbisia divaricate. Balbisia Pediculate, <i>T. Procumbence's</i>



Fig, no.8 Flower.



Fig, no.9 Stem/



Fig, no.10 Leaves.



Fig, no.11 Fruit.

Phytochemistry of *T. procumbens*

*T. procumbens* has high moisture content of 88.30 % in stem and 90.05 % in leaf. It is rich in protein with 37.44 % dry weight (4.38 % wet weight) in the steam and 34.57 % dry weight

(3.44% wet weight) in leaf. The total lipid and carbohydrate content in the stem is 0.85% dry weight (0.1% wet weight) and 41.03 % dry weight (4.80% wet weight) respectively and that in leaf is 6.03% dry weight (0.6% wet weight) and 51.26% dry weight (5.10% wet weight) respectively. The crude Fiber contents is 16.41% dry weight (1.92% wet weight) in stem and 6.13% dry weight (0.61 % wet weight) in leaf The metabolizable vigor per 100 g of *T. Procumbens* is about 321.54 Kcal in dry weight (37.62 Kcal in wet weight) for stem and 397.59 Kcal in dry weight (39.57 Kcal in weight) for leaf.<sup>[19]</sup>

### Medicinal properties of *T. procumbens*

- 1) **Antioxidant Activity:** The concentration (mg/ml) of the methanol extract fractions that signals the generation of DPPH radicals by 50% is known as the IC<sub>50</sub>, and it is used to express the antioxidant activity of the fractions.<sup>[20]</sup>
- 2) **Anti-bacterial Activity:** The entire plant parts of *Tridax procumbens* have been shown to exhibit anti-microbial efficacy against a variety of bacterial species in a previous study. To extract juice from a whole plant, squeeze it between your palms and apply it twice a day for four to five days to wounds and cuts. With the aid of the disc diffusion experiment, the extract of the entire plant demonstrated anti-microbial properties exclusively against *Pseudomonas aeruginosa*.<sup>[21]</sup>
- 3) **Wound Healing Activity:** The plant decoction's ability to cure wounds is attributed to a complex interplay between plasma-derived proteins, extracellular matrix, controlled angiogenesis, and epidermal and dermal cells, all of which are regulated by growth factors and cytokines.<sup>[22]</sup> Although not as much as whole plant decoctions, waterleaf decoctions were also successfully raising lysyl oxidase.
- 4) **Anti-fungal Activity:** To ascertain the antifungal efficacy of the plant decoctions, the disc diffusion method was applied to two fungus strains, *Aspergillus flavus* and *Aspergillus Niger*, in a research study. The decoction of flavonoids had the maximum level of activity against *Aspergillus Niger*, but the decoction of alkaloids demonstrated no action against either of the test fungus.<sup>[23]</sup>
- 5) **Anti-malarial Activity:** The ethanol and water infusions have anti-plasmodial qualities that combat *Plasmodium falciparum*, a parasite resistant to chloroquine. Further animal toxicity studies on the plant are necessary however the decoctions exhibit modest toxicities to human red blood cells.<sup>[24]</sup>
- 6) **Anti-cancer Activity:** Using the MTT assay, the cytotoxicity of the plant-derived compounds was assessed against a human lung cancer cell line. The substance displayed a

90% decrease in cell viability. The substance is lupeol, according to the results of the NMR, MS, and IR spectra. The evaluation of Lupeol's anti-cancer capability against human lung cancer cell line has been conducted by many methods, including clonogenic survival determination, cell cycle control, cell-based assay for COX-2 activity inhibition, and DNA fragmentation.<sup>[25]</sup>

- 7) **Blood Coagulation and Haemostatic Activity:** Because of the strong blood coagulation activity observed in leaf decoctions, water may be employed as a powerful haemostatic agent.<sup>[26]</sup> Since the ethanol extract shortens the clotting time in the blood samples from all the studies, the haemostatic property of the plant's leaves of the various solvent extracts was ascertained in vitro using Lee-White's method.<sup>[27]</sup>
- 8) **Repellence Activity:** In one study, essential oils were isolated from leaves using the steam distillation method, and their ability to resist the malaria parasite *Anopheles Stephens* locally in mosquito cages was examined<sup>[28,29]</sup>. Every essential oil was examined in three different concentrations. The plant's essential oils demonstrated a discernible repellent effect.<sup>[30]</sup>

## MATERIALS AND METHOD OF PREPARATION

### Formulation for wound healing activity by using *L. camara*

The composition of *L. camara linn* (Verbenaceae) was collected. The foreign, earthy matter and residual materials were removed carefully from the leaves and then cleaned and dried in the shade. It was powdered and used for extraction. Powdered leaves were placed inside a thimble made from thick filter paper, which was loaded into the main chamber of the Soxhlet extractor. The Soxhlet extractor was placed onto a flask containing the extraction solvent until it gets exhausted. The extract is filtered and concentrated under reduced pressure. It was stored at 4-8°C until use.<sup>[31]</sup>

### Formulation for wound healing activity by using *T. procumbens*

For experimental study the materials required Albino rats, etc. Animal Ethics committee, the was taken for preclinical study, selection animals (Albino rats), grouping of animals the rats were housed in individual cages and kept in a well-ventilated room under hygienic conditions. This was conducted as per the method developed by Morten and Mallon. The wound healing property of the trial drugs, 5 dosage forms of *T. procumbens* was analysed by excision wound model.<sup>[32]</sup>



**PHYTOCHEMICAL TEST***Lantana camara*<sup>[33,34,35]</sup>

Sr.no.	Phytochemical Constituents	Test Name	Observation / Positive Result
1)	Alkaloids	Mayer's Test	- An orange red ppt indicates the presence of alkaloids.
2)	Flavonoids	Shinoda Test	- If a yellow sol <sup>n</sup> turn colourless then it indicates presence of flavonoids.
3)	Tannins	Ferric Chloride Test	- A dark blue or greenish black colour product shows presence of tannin.
4)	Saponins	Foam Test	- Foaming (small creamy bubbles) was the indication of saponin presence.
5)	Glycosides	Keller–Kilani Test	- A few drops of Fehling's sol <sup>n</sup> A & B were added and red pp. Indicates presence of glycosides.
6)	Terpenoids	i) Salkowski Test ii) Liebermann–Burchard Test	- A reddish-brown colour shows presence of terpenoids. - Forms layer with brown colour at junction upper layer with green colour and lower red colour layers shows terpenoids.
7)	Phenols	Ferric Chloride Test	- Deep blue or black indicates the presence of phenolic compound.

*Tridax Procumbens*<sup>[36,37,38,39,40]</sup>

Sr.no.	Phytochemical Constituents	Test Name	Observation / Positive Result
1)	Flavonoids	i) NaOH test ii) Shinoda test	- Formation of intense yellow colour that didn't become colourless on the addition of a few drops of dil. HCL indicated absence of flavonoids. - Formation of magenta colour indicated the presence of flavonoids.
2)	Tannins	i) Gelatine test ii) Lead acetate test	- Formation of white ppt indicated the presence of tannins. - Formation of yellow or red ppt indicated the presence of tannins.
3)	Saponins	i) Foam test ii) Haemolysis test	- Foam indicated the presence of saponins. - Haemolytic zone appeared indicates presence of saponins.
4)	Alkaloids	i) Iodine test ii) Wagner's test	- Blue colour which disappeared on boiling and reappeared on cooling indicated presence of alkaloids. - Formation of reddish brown ppt indicated the presence of alkaloids.
5)	Steroids	i) Salkowski test	- Colour change from violet to blue or green in some sample indicates presence of steroids.

## EVALUATION TEST

### 1) Plant material & extract preparation

Collect fresh *Lantana camara* leaves, shade-dry, powder and note batch details. Prepare a hydroalcoholic or methanolic extract by maceration or Soxhlet extraction (typical ratio 1:5–1:10 w/v), concentrate under reduced pressure and record yield (% w/w). For topical studies prepare ointment bases containing 5% and 10% w/w extract (common concentrations used in published topical formulations). Document storage and stability. Methods and concentrations follow those reported in preclinical *L. camara* wound-healing reports.<sup>[41]</sup>

### 2) In-vitro screening (optional but recommended before animals)

**a. Cytotoxicity** — test extract on dermal fibroblasts (e.g., L929) or HaCaT keratinocytes with MTT or resazurin to determine non-cytotoxic concentrations for further assays (report CC<sub>50</sub>).

**b. Scratch (wound) assay** — create linear scratch in confluent monolayers, treat with sub-cytotoxic concentrations (e.g., 1–100 µg/mL), image at 0, 12, 24, 48 h and quantify % wound closure with ImageJ. In vitro migration/proliferation benefit was suggested in plant wound-healing screening workflows in the literature.<sup>[42]</sup>

### 3) Animals, grouping & general considerations

Use healthy adult Wistar rats (150–250 g) or Swiss albino mice as appropriate. Randomize animals into groups ( $n \geq 6$  per group): (1) Normal control (no treatment), (2) Vehicle control (ointment base), (3) Standard (e.g., povidone-iodine, silver sulfadiazine or nitrofurazone), (4) *L. camara* ointment 5% and (5) *L. camara* ointment 10%. Follow prior studies' group sizes and controls when possible. Ensure IAEC/IACUC approval and humane handling.<sup>[43]</sup>

### 4) Excision wound model (primary efficacy test)

**Procedure:** Under light anaesthesia, shave dorsal surface, mark and excise a full-thickness circular wound (commonly ~500 mm<sup>2</sup> or 2 cm diameter — adapt to animal size). Apply test topical formulation once or twice daily until complete epithelialization. Trace wound margins on transparent sheets on days 0, 3, 7, 10, 14 and measure area. Calculate wound contraction: % Wound contraction =  $[(\text{Initial area} - \text{Area on day } n) / \text{Initial area}] \times 100$ . Record epithelialization period (days until scab falls with no raw area). This exact excision protocol and measurement approach were used in *L. camara* studies that reported significant contraction and shortened healing time.<sup>[41]</sup>

**5) Incision wound model (tensile strength / breaking strength)**

**Procedure:** Make a 3–4 cm paravertebral linear incision under anaesthesia and suture. Apply topical treatments daily; remove sutures on day 7. On day 10 or 14, measure tensile strength (breaking strength) of healed skin using a tensiometer or continuous water flow method. Increased breaking strength indicates improved collagen maturation — a parameter reported improved by *L. camara* treatment in prior preclinical work.<sup>[41]</sup>

**6) Dead-space or granulation tissue model (collagen & biochemical markers)**

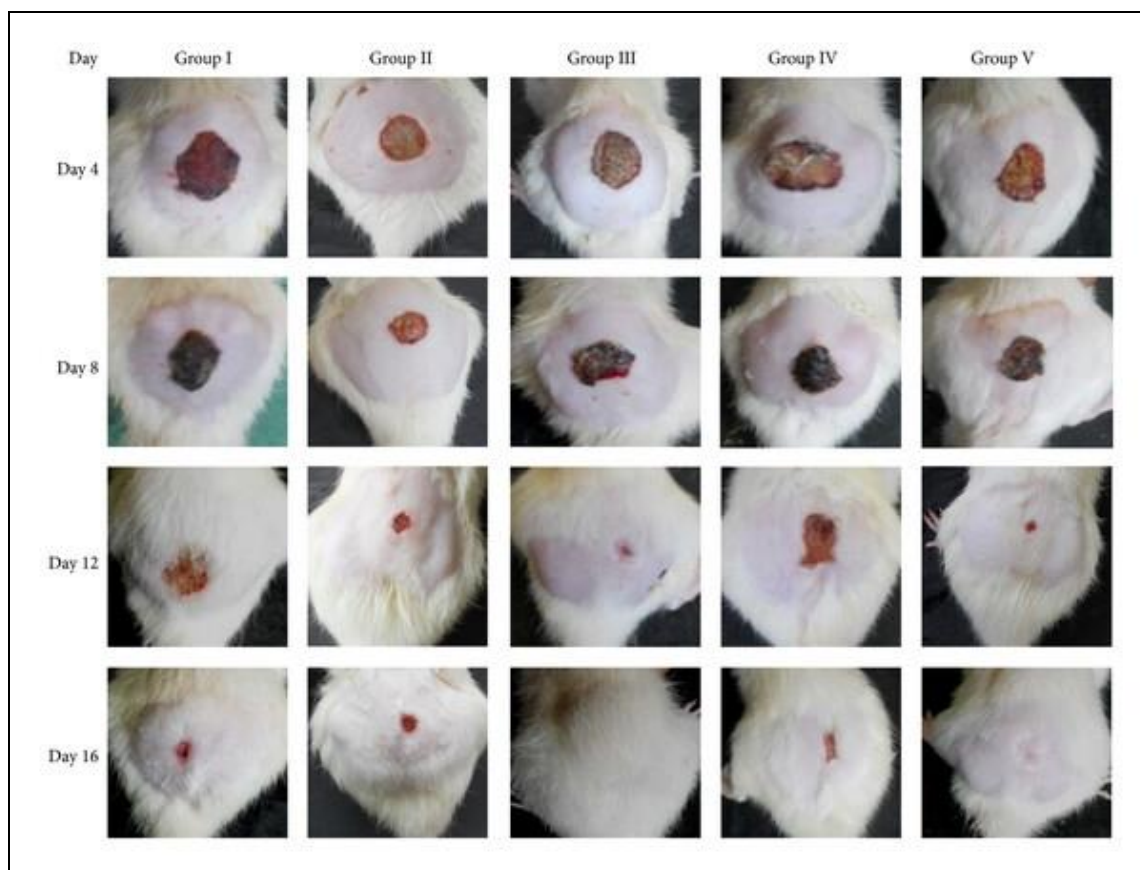
Implant sterile cylindrical tubes or cotton pellets subcutaneously to induce granulation tissue, treat per protocol, then on day 7 or 14 remove tissue, dry and weigh. Assay hydroxyproline content (colorimetric) as a collagen marker (expressed mg hydroxyproline/g tissue) and measure total protein and antioxidant enzyme activities (SOD, CAT) and lipid peroxidation (MDA) if resources permit. Increased hydroxyproline with *L. camara* extract was reported in the literature and correlates with improved collagen deposition.<sup>[41]</sup>

**7) Phytochemical correlation & mechanism insights**

Measure total phenolic and flavonoid contents (Folin–Ciocalteu,  $\text{AlCl}_3$ ) and antioxidant activity (DPPH/ABTS/FRAP) of the extract — these correlate with wound-healing outcomes in many plant studies and were cited as likely contributors to *L. camara*'s activity (antioxidant, anti-inflammatory, antimicrobial). Identification of triterpenoids (e.g., ursolic acid derivatives) in *L. camara* has been linked to wound healing mechanistically via collagen modulation and anti-inflammatory effects.<sup>[42]</sup>

**8) Antimicrobial and Antioxidant Evaluation**

Agar well-diffusion and broth dilution assays against *Staphylococcus aureus*, *E. coli*, *Pseudomonas aeruginosa*, and *Candida albicans* reveal notable inhibition zones and low MIC values for ethanolic extracts. Antioxidant potential is quantified using DPPH and ABTS radical scavenging assays, showing concentration-dependent inhibition. These activities contribute to the protection of wound tissue from oxidative stress.<sup>[42]</sup>



**Fig. no. 12: Wound healing images.**<sup>[43]</sup>

## CONCLUSION

The study confirms that *Lantana camara* and *Tridax procumbens* possess significant wound healing potential due to the presence of various bioactive phytochemicals such as flavonoids, alkaloids, tannins, and terpenoids. These compounds contribute to anti-inflammatory, antimicrobial, and antioxidant effects, which collectively enhance collagen formation, tissue regeneration, and wound contraction. Both plants, individually and in combination, can be effectively used in the formulation of herbal ointments or gels for wound management. Their natural origin, safety, and efficacy make them valuable alternatives to synthetic drugs for promoting faster and safer wound healing.

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