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# Sanitation and Hygiene Condition of Urban Slums: A Study on Slums of Lucknow City, (U.P.) India

***Dipti Maurya, Pradip Kumar and Susmita Srivastava[1]**

Department of Geography,
[1]Department of Zoology
S.K.P.G. College, BASTI-227001(U.P.) INDIA
*Corresponding Author
E-mail: diptimaurya007@gmail.com

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## ABSTRACT

The number of slums has significantly increased in Lucknow over the last three decades along with the expansion of cities and towns. Rapid urbanization, caused largely by a heavy influx of migrants from rural areas, has exerted severe pressure on urban housing and public services in Lucknow City. At the rate at which urban slums are increasing the infrastructure and basic services in Lucknow are not able to cope. Due to this circumstance as well as the impoverished economic circumstances of the poor migrants, a considerable number of slums have been formed where severe service deficiencies have compounded and proliferated, creating dangerous environmental conditions. In the present Study Data have been collected from primary sources, secondary sources and from focused group discussion. The study analyses the practice of sanitation and hygiene regarding water. This paper has highlighted the survey of water resource availability and quality at the source point of consumption; problems faced in getting safe drinking water; and knowledge of hygienic latrines; awareness about health. In addition to the above, the survey maintained a specific focus on, women and the young generation. The study finds that there have been some improvements in terms of knowledge of hygiene, due to the spread of awareness during corona.

Figure : 00 References : 09 Tables : 02
KEY WORDS : Hygiene; Sanitation; Slum; Water resource

## Introduction

United Nations defined slums as communities characterized by insecure residential status, poor quality of housing, overcrowding, and inadequate access to safe water, sanitation, and other infrastructure[9]. With the global trend of rapid urbanization, the proliferation of informal settlements, commonly known as slums, has become a prominent feature of many cities worldwide. In the context of India, urbanization has led to the emergence and expansion of slum communities, posing significant challenges in terms of basic amenities and public health. Among these challenges, ensuring adequate sanitation and hygiene conditions within urban slums stand as a critical priority for both local governments and public health authorities. Slums are characterised by informal and unplanned settlements with poor infrastructure and inadequate sanitation unfit for human habitation[2]. Understanding the challenges faced by slum dwellers will help in formulating targeted involvement that can effectively enhance the health and well-being of slum residents while fostering sustainable urban development. In 1981, the slum population in India constituted 17.5% of the urban population which increased to 35% in 2018. In 2001, there was 23.5 per cent of households in urban areas which were living in slums. In 2011, it came down to 17.4 per cent. But there are still 13.74 million slum households and 68 million people living in the slum areas[1].In the context of Uttar Pradesh, though the state is considered one of the less urbanized states of India, it has the second-largest urban population in the country. About 22% of the population lives in urban areas in Uttar Pradesh, which constitutes more than 44 million[6]. Sanitation and health have invariably been linked together, the former being an essential condition to achieve the latter[4].Sanitation is also one of the major environmental health issues to be addressed. The Joint Monitoring Project (JMP) report of 2013 estimates that 50% of the population in India still defecates in the open. 1.9 billion people gained access to improved sanitation facilities over a period of two decades (1990 to 2011) with an average rate of 240,000 individuals gaining access every day. 8% of the Indian population is still devoid of clean water, and only 25% of the population has access to piped water on premises[9].The lack of recognized space for the urban poor in the cities' master plans has led to a significant rise in the number of slum households in the country over the decade[1].

**TABLE-1 : Profile of families in the selected slums of Lucknow**

| Household Characteristics | Number | Percentage (%) |
|---|---|---|
| **Family Type** | | |
| Nuclear | 316 | 79.2 |
| Joint | 84 | 20.8 |
| **Number of family members** | | |
| Up to 4 | 280 | 70.1 |
| 5 - 7 | 40 | 9.9 |
| >7 | 80 | 20.0 |
| **Religion** | | |
| Hindu | 294 | 73.6 |
| Muslim | 105 | 26.4 |
| **Social Class** | | |
| OBC | 101 | 25.2 |
| SC/ST | 212 | 52.9 |
| General | 87 | 21.9 |
| Education | | |
| Literate | 292 | 73.1 |
| Illiterate | 108 | 26.9 |
| **Socioeconomic Status** | | |
| Upper lower | 76 | 18.9 |
| Lower middle | 94 | 23.5 |
| Lower | 230 | 57.6 |

References – Personal Based Survey 2023

Access to better sanitation conditions in the form of the availability of private toilets is largely governed by the financial status of the family. An improvement in health and privacy will, therefore take place with enhanced accessibility to private toilet facilities[5]. Kaccha slums were mainly responsible for the overall burden of excreta disposal, solid waste disposal, and access to their water supply for drinking and other household purposes[3]. Improved sanitation, hygiene and water can help generate considerable national and household health savings both in terms of cost and time which in turn can be used productively. The over congested locality and house setup might be the reason for inadequate ventilation (47.3%) and lighting (25.0%).Dampness was found in about 72.4% of the surveyed households, which directly revealed the unplanned and improper construction and maintenance of the houses[7].

## Study Area

The city of Lucknow is situated on the banks of river Gomti which passes through the middle of the city. The city lies at 26°30' N and 27°10 'N Latitude and 80°30' E and 81°13' E Longitude. Lucknow is the capital city of Uttar Pradesh state and one of the most prominent cities in India in terms of commerce, education, history, architecture, culture, Urdu literature *etc*.

## Objectives

- To analyse the sanitation and hygiene conditions in the slums of Lucknow.
- To examine the awareness of sanitation and hygiene conditions in residents of the slums of Lucknow.

## Methodology

### 4.1 Sources of data

In the present work, both primary and secondary data have been used. The source of primary data is a questionnaire schedule distributed among sampled respondents and through field observations. The secondary information was gathered from different sources like books, records, journals, National and International reports, reviews, websites of Government and Non-Governmental Organizations (NGOs), and reports of the Committee on Slum Statistics/Census of Lucknow city.

### 4.2 Sampling and collection of data

Lucknow City is divided into 6 Zones out of which 4 Zones have been selected for data collection. Data were collected from 100 households from each of the 4 selected slums. The four selected slums were Mawaiyya, Madiayon, Amausi and Chinhat Bazar.

**TABLE-2 - Sanitation and Hygiene Condition**

| Variables | Parameters | Indicators | Mawaiyya | Madiaon | Amausi | Chinhat |
|---|---|---|---|---|---|---|
| X1 | Main source of Drinking water | Pipe water (municipality) | 31.00 | 41.00 | 36.00 | 49.00 |
| | | Public hand pump | 69.00 | 59.00 | 64.00 | 51.00 |
| X2 | Quality of water consumed | Ppm 200 -400 | 18.00 | 26.00 | 29.00 | 51.00 |
| | | PPM more than 400 | 82.00 | 74.00 | 71.00 | 49.00 |
| X3 | Distance Travell to fetch clean water | 1km – 2km | 83 | 86 | 78 | 91 |
| | | More than 2km | 17 | 14 | 22 | 9 |
| X4 | Duration for which water is available | 2hours a day | 27 | 34 | 33 | 19 |
| | | 4 hours a day | 61 | 43 | 57 | 36 |
| | | 6 hours a day | 12 | 23 | 10 | 45 |
| X5 | Drinking water handling & storage practices | Safe | 25 | 20 | 22 | 30 |
| | | Not safe | 75 | 80 | 78 | 70 |
| X6 | Handwashing practice before eating food | Present | 78.00 | 76.00 | 72.00 | 82.00 |
| | | Absent | 22.00 | 24.00 | 28.00 | 18.00 |
| X7 | Hand wash Practice before cooking food | Present | 52.00 | 64.00 | 65.00 | 53.00 |
| | | Absent | 48.00 | 36.00 | 35.00 | 47.00 |

| Variables | Parameters | Indicators | Mawaiyya | Madiaon | Amausi | Chinhat |
|---|---|---|---|---|---|---|
| X8 | Drainage for sullage | Present | 38.00 | 11.00 | 9.00 | 95.00 |
| | | Absent | 62.00 | 89.00 | 91.00 | 5.00 |
| X9 | Excreta disposal | Open field | 78.00 | 67.00 | 72.00 | 66.00 |
| | | Sanitary latrine | 22.00 | 33.00 | 28.00 | 34.00 |
| X10 | Rodent sites | Present | 58.00 | 77.00 | 66.00 | 53.00 |
| | | Absent | 42.00 | 23.00 | 34.00. | 47.00 |
| X11 | Mosquito breeding sites | Present | 78.00 | 75.00 | 74.00 | 72.00 |
| | | Absent | 22.00 | 25.00 | 26.00 | 28.00 |
| X12 | Habit of cleanliness | Present | 78.00 | 72.00 | 65.00 | 77.00 |
| | | Absent | 22.00 | 28.00 | 35.00 | 23.00 |

References – Personal Based Survey 2023

**4.3 Method and techniques of analysis**

Based on the objectives and problems of the study, the data were processed and analysed through Microsoft Excel[7]. The collected data were further analysed and interpreted with the help of statistical tools like percentages and averages.

## Results and Discussion

The profile of families in the selected slums of Lucknow like family type, number of family members, religion, social class, education and socioeconomic status are given (Table – 1). It shows that 79.2% of families in slums are nuclear type and only 20.8% of families are joint families. The number of family members in slums shows that 70.1 % of households have up to 4 members in the family, around 9.9% of families in slums have family members between 5 members to 7 members, and 20 % of families in slums have family members more than 7 members in the family. Religion-wise profile of respondents 73.6 % are Hindu and 26.4 % are Muslim. In the social caste group, 21.9% belong to the General class, 25.2% of respondents belong to other backward classes (OBC), and 52.9% are in Schedule Caste (SC) and Schedule tribes(ST). The Table shows the education profile of slum respondents shows that 73.1% are Literate and 26.9% are Illiterate. According to the Socioeconomic status of respondents, about 18.9% of respondents belong to the Upper lower class, 23.5% of respondents belong to the Lower middle class and 57.6% of respondents belong to the lower class.

The present study (Table-2) analysed the sanitation and hygiene condition of four selected slums of Lucknow on twelve indicators which include the main source of drinking water, quality of water consumed, distance travelled to fetch clean water, duration for which water is available, drinking water handling and storage practices, handwashing practice before cooking food, Drainage for sullage, Excreta disposal, Rodent sites, Mosquito breeding sites and habit of cleanliness.

**X1-Main source of Drinking water-**This indicator shows the source of water in selected slums. In Mawaiyya, 31 % of households have a piped water supply whereas 70% of households have public hand pumps in the community for water. In Madiaon, 41

households have a piped water supply, and 59 households have a public hand pump. In the Amausi slum, 65% of households have a public hand pump water supply. In Chinhat Bazar slum around 51% of households have public hand pumps installed in the community.

**X2- Quality of water consumed**- In Mawaiyya slum in 82% of households the quality of water is poor, and not good for drinking. In Madiaon slum 74% of households the quality of water is poor, not good for drinking. In Amausi slums in only 30 % of houses, the quality of water is fair enough. In Chinhat bazaar slum, the quality of water is fair in 51% of houses.

**X3-Distance travel to fetch clean water**– It is found that most of the slums don't have a direct supply of clean water and they have to travel long distances to fetch clean water. In Mawaiyya slum 83% of people have to travel for one to 2 km to fetch clean water. In Madiyaon 86% of people have to travel one to 2 km distance. In Amausi 78% of people's households have to travel one to 2 kilometres to fetch clean water. In Chinhat Bazar only 9% of households travel more than 2 kilometres to fetch clean water which shows that the Chinhat Bazar slum has better water conditions.

**X4-Duration for which water is available-** There have been instances in slums where water is not available for the whole day. In Mawaiyya 27 households have access to water for only 2 hours a day. In Madiaon, in 34 households water is available for 2 hours a day. In Amausi Slum 33 households have access to water for 2 hours a day. In Chinhat Bazar slum 45 households have water available for 6 hours aday.

**X5-Drinking water handling and storage practices-** Due to less awareness among people in slums, they do not handle drinking water safely and neither store it properly. This variable indicates the handling and storage practices in slums. In Mawaiyya, only one-fourth of households found safe handling and storage of drinking. In Madiyaon, almost 80% of households do not safely handle and store drinking water. In Amausi, only 22% of households safely handle and store drinking water. In Chinhat bazaar slum, 30% of households know about the safe handling and storage of drinking water.

**X6-Hand washing practice before eating food** Mawaiyya 78% of households have hand washing habits whereas 22% of people did not put much care on washing their hands before eating food. In Madiyaon, 76% of households do hand wash before eating food. In the Amausi slum, 72% of people have a hand-washing habit before eating food. In Chinhat bazar slum 82% of people have a hand-washing habit before eating food.

**X7-Hand washing practice before cooking food**- It is another indicator which indicates the habit of sanitation and hygiene in the slums. In Mawaiyya slum around 50% of people, take hand washing habits casually and don't wash their hands before cooking food. In Madiyaon slum 64% of people do wash their hands before cooking food. In the Amausi slum, 35% of people take it casually and do not wash their hands before cooking food.In Chinhat bazar slums 53% of people prefer to wash their hands before cooking food.

**X8-Drainage for sullage**- In Mawaiyya, over 38 households have drainage facilities for sullage while in 62 households drainage for sullage facilities is not present, in these households open pit is present for sullage. In Madiaon slum only in 11 households drainage for sullage is present whereas in 89 households drainage for sullage is not present. In Amausi slum mere 9 households have drainage for sullage whereas in 91 households no drainage for sullage. In Chinhat Bazaar slums have better living conditions, in this slum around 55 households have drainage for sullage whereas 45 households don't have drainage for sullage facilities.

**X9- Excreta Disposal**–in Mawaiyya slums around 80% of households dispose of excreta in open fields. In the Madiaon slum, 67% of househol dispose off excreta in open fields. In the Amausi slum, 72% of households dispose of excreta in open fields. In Chinhat Bazar slum 34 % of households use sanitary latrines and in this slum, it is observed that major role is played by Local NGOs in spreading awareness.

**X10 – Rodents** – In Mawaiyya slums around 58 households rodent sites are present. In Madiaon slums it is seen that around 77 household rodents are present, only around 23 household rodent sites are not present because of awareness of dengue disease. In the Amausi slum around 66 households rodent sites are present and around 34 households no rodents are present. In the Chinhat Bazaar slum around 53 households, no rodent sites are present because of awareness provided to them by local NGOs during corona time about cleanliness.

**X11- Mosquito breeding sites** – It is found that due to waterlogging in slums there are ample chances of mosquito breeding sites. In this research, it is found that, in Mawaiyya slum mosquito breeding sites are present around 78 households. In Madiaon slum around 75 households mosquito breeding sites are present. In Amausi slum around 74 houses mosquito breeding sites are present. In Chinhat Bazaar slum around 72 houses mosquito breeding sites are present.

**X12 – Habit of cleanliness** – This variable is added to analyse the behaviour of people in slums towards the habit of cleanliness around them. In the Mawaiya slum in 78 houses habit of cleanliness is present whereas in

22 houses habit of cleanliness is not present. In Madiaon slum in 72 houses habit of cleanliness is present. In the Amausi slum in 65 houses habit of cleanliness is present whereas in 35 houses habit of cleanliness is not present. In Chinhat bazar slum, in 77 houses habit of cleanliness is present only 23 houses do not have a habit of cleanliness.

## Conclusion

The present study showed that sanitary and hygiene conditions are very poor in slums. The behaviour of people in slums towards sanitation and hygiene is negligible they completely rely on the support of others. Government support is needed to bring change in the sanitation and hygiene conditions of slums in Lucknow. It has been observed that role of NGOs played a major role in spreading awareness of sanitation and hygiene.In Chinhat Bazar slum the sanitation and hygiene conditions are somehow better because the people of slum are proactive towards sanitation and hygiene. In Mawaiyya slum hygiene conditions are very poor because of less awareness of cleanliness.In Madiaon and Amausi it is observed that clean drinking water and water availability duration is poor, people of slum are struggling to meet their daily basic needs.

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# A phytochemicals based formulation induced faster wound healing of excision wounds in *wistar* rats

***Lalita Kushwah[1], Rupali Dutt[2], Manisha Singh[3], Sadhana Shrivastava[4] and D.K. Sharma[1]**

[1]Department of Zoology,
S.M.S. Govt. Model Science College,
GWALIOR-474009 (MP) INDIA
[2]School of Studies in Microbiology,
Jiwaji University,
GWALIOR-474001 (MP), INDIA
[3]Department of Surgery,
Gajra Raja Medical College,
GWALIOR-474009 (MP) INDIA
[4]School of Studies in Zoology,
Jiwaji University,
GWALIOR-474001 (MP), INDIA
*Corresponding Author
Email. lalitakuashwah@gmail.com

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## ABSTRACT

The present study involved development and scientific validation of wound healing potential of three phytochemical-based formulations at different concentration in experimental rat model. Formulation-I consisted of ethanolic extracts of *T. erecta*, *T. procumbens*, *A. vera* and *C. longa*. Formulation-II consisted of ethanolic extracts of *A. indica, F. benghalensis*, *A. vera* and *C. longa*. Formulation-III contained ingredients of both Formulations-I and II. The extracts were analyzed quantitatively, revealing that *T. erecta* had the highest content of total phenols (41.61±0.10 mg GAE/g) and flavonoids (52.50±0.53 mg QE/g) and tannins were found in *T. procumbens* (59.61±0.15 mg TC/g). Wound healing functions of formulations were assessed by topical application on excision wounds made in *Wistar* rats. The healing process was monitored by measuring the degree of wound contraction on alternate days. The polyherbal Formulation-III (5% concentration) was found to achieve complete wound contraction (Significant pd"0.0001) on 18$^{th}$ day in comparison to other Formulations. The study demonstrated that the Formulation-III was found to have the most potential in achieving rapid wound healing rate.

Figures : 03 References : 33 Tables : 05
KEY WORDS : Excision wound, Phytochemicals, Polyherbal formulation, Wound healing.

## Introduction

Wounds are physical skin injuries that result in an opening or break of the skin that disrupts structural and functional integrities of the skin tissues. These alterations may or may not be associated with the loss or damage of underlying connective tissues (bone, cartilage, fat, blood and lymphatic tissue). Wound healing is the process involving four overlapping phases, *viz.,* hemostasis, inflammation, proliferation and remodeling[4,6,14]. Speedy and rapid wound healing is necessary for the survival and protection against the edema and skin ulcers which may prove lethal to the organism on no treatment[18].

The *Tagetes erecta*, *Tridax procumbens*, *Azadirachta indica, Ficus benghalensis*, *Curcuma longa* and *Aloe vera* exhibit a wide spectrum of biological activities and are used as traditional medicines for household remedies against various human ailments in Ayurveda[20]. *T. erecta* also known as Marigold (Family-Asteraceae)[32] and has anti-microbial, anti-inflammatory, hepatoprotective, anti-parasitic, anti-septic, anti-oxidant, analgesic and wound healing properties[2,9]. Studies have

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**TABLE-1: Herbal components of F-I, F-II and F-III**

| F-I | F- II | F-III |
|---|---|---|
| *T. erecta* | - | *T. erecta* |
| *T. procumbens* | - | *T. procumbens* |
| - | *A. indica* | *A. indica* |
| - | *F. benghalensis* | *F. benghalensis* |
| *A. vera* | *A. vera* | *A. vera* |
| *C. longa* | *C. longa* | *C. longa* |

shown that it can increase platelet and white blood cell count and reduce bleeding and clotting times[16]. Topical application of *T. erecta* flower paste has resulted in faster wound healing in animals and extracts have also exhibited oral anti-ulcer activity[7]. The *T. procumbens* (Ghamara) belonging to the Asteraceae family has been extensively used in the Ayurvedic system of medicine for treatment of cuts, wounds and burns[33] due to the presence of pharmacological activities like anti-inflammatory, anti-bacterial and anti-oxidant potentials[8]. The herb contains bioactive components belonging to alkaloids, phenols, flavonoids, carotenoids, B sitosterol, fumaric acid, luteolin, quercetin, tannin, *etc.*[30]. Neem, scientifically known as *A. indica* (Family- Meliaceae), contains bioactive compounds in its every part like seeds, leaves, roots, bark and trunk. *A. indica* possesses anti-inflammatory, anti-fungal, and anti-bacterial properties that aid in wound healing. It also contains amino acids, vitamins, and main active ingredients such as nimbidin, nimbin and nimbidol that play important role in wound healing processes particularly in the proliferation phase, formation of collagen and angiogenesis[14,29]. The *F. benghalensis* (Family-Moraceae), also known as the Indian Banyan tree, has been used in traditional medicine to treat conditions such as dysentery, diabetes and nervous disorders. Its parts have antimicrobial, antioxidant, anti-inflammatory, anti-ulcer, and wound healing properties[15]. A study reported that ethanolic and aqueous extracts of *F. benghalensis* leaves demonstrated a decrease in epithelization and an increase in the rate of wound contraction in an excision wound model[10]. The *A. vera* plant from the Liliaceae family has been traditionally used to treat burns, allergic reactions, arthritis, indigestion, ulcers, diabetes, skin diseases, and digestive system inflammation[1]. *A. vera* extract has anti-inflammatory properties, promote collagen synthesis, skin regeneration and blood supply essential for wound healing[12]. It contains bioactive compounds such as flavonoids, alkaloids, tannins, terpenoids, polyphenols, amino acids and vitamins and reduces wound-related bacterial infections[11]. The *C. longa* (family- Zingiberaceae) is well known for its anti-microbial functions[22]. Curcumin has a wide range of biological effects including anti-inflammatory, anti-oxidant, anti-tumor, anti-bacterial and anti-viral activities[21]. Curcuminoids, the active ingredients of *C. longa* are known to be beneficial in treatment of skin diseases and enhance the wound healing and skin regeneration[28].

Several other herbs/plants have been reported to play a crucial role in wound healing process[26]. Polyherbal formulations not only accelerate wound healing process with minimum or no side effects *i.e.,* they are safe, non-toxic and can be administered over longer periods[25]. Hence the polyherbal formulations would have longer acceptability in treatment of wounds with high efficacy. More than 70% of wound healing pharma products are of plant origin[24]. The present study demonstrated superior wound healing potentials of different polyherbal formulations on excision wounds made on Wistar rats.

## Materials and Methods

### Collection and identification of plant materials

Mature *A. indica*-leaves, *T. procumbens*-whole plant*, F. benghalensis*-bark, and *A. vera*-leaves were collected from 'Charak Udhyan' (Medicinal plants garden) of Jiwaji University, Gwalior. The used

**TABLE-2 : Quantitative analysis of phytochemical extracts**

| Extracts | Total Phenols (mg GAE /g) | Total Flavonoids (mg QE / g) | Total Tannins (mg TC / g) |
|---|---|---|---|
| ***T. erecta*** | 41.61±0.10 | 52.50±0.53 | 9.920±0.24 |
| ***T. procumbens*** | 17.14±0.23 | 34.00±0.52 | 59.61±0.15 |
| ***A. indica*** | 15.15±0.53 | 9.420±0.60 | 14.17±0.60 |
| ***F. benghalensis*** | 12.23±0.90 | 8.970±1.12 | 44.01±0.41 |
| ***A. vera*** | 34.28±0.36 | 40.27±1.00 | 19.51±0.32 |

Results are expressed in Mean ± SEM (n=3)

*T. erecta*-flowers were collected from the temple of the Jiwaji University campus. The plant specimens were authenticated by the Institute of Ethnobiology, Jiwaji University. The dried plants/plant parts were mounted as herbarium specimens in the Institute and have been assigned code names *viz.*, *T. erecta* (IOE-438), *A. indica* (IOE-439), *T. procumbens* (IOE-440), *F. benghalensis*, (IOE-435) and *A. vera* (IOE-437). Curcumin purchased from Pukhraj herbals, Mandsaur India and it was used directly.

## Preparation of extracts

The selected parts of plants/herbs (*T. erecta*-flowers, *T. procumbens*-whole plant, *A. indica*-leaves and *F. benghalensis*-bark) were washed. Materials were dried at room temperature in shade for 7 to 10 days then grounded to powder separately using a mechanical grinder. Crude powder was Soxhlet extracted separately using 95% ethanol. The extract was dried up at *45ºC in a hot air oven for 2-3 days. F*resh *A. vera* leaves gel was ground to homogenous gel and left for 24 hours on a magnetic stirrer adding 95% ethanol for uniform mixing. The solution was centrifuged at 10,000 rpm for 30 min., supernatant was dried under the hot air oven at *45ºC* for 2-3 days, and the powder was collected and stored at 4°C in an airtight bottle.

## Characterization

## Quantitative analyses

The total phenolic constituents of various extracts were determined by *Folin-Ciocalteu* method[27] using Gallic acid as standard and results are expressed as mg of gallic acid equivalents per g (mg GAE/g) of extract. The flavonoid constituents were determined by colorimetric assay[27] using Quercetin as standard and the results are expressed as mg QE/g of extract. The tannins were determined by the *Folin-Ciocalteu* method[3]. Tannic acid (100 to 1000µg/ml) was used as a standard. The tannin content was expressed as mg of TA/g of extract.

## Preparation of polyherbal ointment/formulation

The base was prepared by melting cetyl alcohol and soft wax paraffin, followed by the addition of liquid paraffin. Each polyherbal formulation (I, II, and III) was created by mixing specific amounts of ingredients/ extracts (refer to Table 1) and then homogenized to form a smooth ointment suitable for topical application on wounds. Four different concentrations (2%, 5%, 10%, and 25% w/w) of each formulation (F-I, F-II and F-III) were made and stored at room temperature in plastic containers.

## Experimental rodents

*Wistar* rats weighting about 200-250g obtained from Animal facility of the Defence Research Development Establishment (DRDE), Gwalior were acclimatized at 25°C±2°C and with a humidity of 50% - 60% for two weeks' time before making excision wounds. The animals were fed on standard pellet diet and provided water *ad libitum*.

## Creation of excision wounds on *Wistar* rats

Rats were anesthetized with diethyl ether, hair of the dorsal thoracic region was shaved off with an electrical shaver and disinfected with 70% alcohol and a skin area of 2 X 2 $cm^2$ (~400 $mm^2$ diameter circular

area) on the dorsal thoracic region was excised with surgical scissor[33]. The day of wound creation was considered a zero-day. The formulation was topically applied evenly on the wound area, once a day till complete healing of the wound achieved.

**Experiment- I: A total of six groups (6 rats in each group) were made as detailed below-**

Group 1- Normal Control (Base)

Group 2- Reference (Betadine)

Group 3- Formulation-I (2%)

Group 4- Formulation-I (5%)

Group 5- Formulation-I (10%)

Group 6- Formulation-I (25%)

**Experiment- II: A total of six groups (6 Rats in each group) were made as detailed below-**

Group 1- Normal Control (Base)

Group 2- Reference (Betadine)

Group 3- Formulation-II (2%)

Group 4- Formulation-II (5%)

Group 5- Formulation-II (10%)

Group 6- Formulation-II (25%)

**Experiment- III: A total of six groups (6 Rats in each group) were made as given detailed below-**

Group 1- Normal Control (Base)

Group 2- Reference (Betadine)

Group 3- Formulation-III (2%)

Group 4- Formulation-III (5%)

Group 5- Formulation-III (10%)

Group 6- Formulation-III (25%)

## Assessment of wound area contraction

Wound area was marked by tracing the raw wound area on transparent paper with a permanent marker and the area was measured on graph paper. The wound area was measured at 4 day intervals till complete healing was achieved following topical application of a given formulation. Photographic image of wound area from a fixed distance of 15 cm was taken and documented. Percentage of wound contraction was calculated by using the formula given below[33].

$$\text{Wound contraction \%} = \frac{[\text{Initial wound area} - \text{Wound area following treatment}]}{\text{Initial wound area}} \times 100$$

The study protocol was approved by the Institutional Animal Ethics Committee of Jiwaji University (Number- IAEC/JU/21).

## Statistical Analysis

Statistical analyses were performed using GraphPad Prism 5.0 software (GraphPad Prism software Inc., La Jolla, Ca). The results were analyzed using one-way analysis of variance (ANOVA). P values *pd"0.05, **p<0.01, ****p<0.001, ****p<0.0001. All values are presented as mean ± the standard error of the mean (SEM).

# Results

## Quantitative analyses

Quantitative estimation of total phenols, flavonoids and tannin were done on ethanolic extracts of all phytochemical ingredients and results are shown in the Table 2. The highest content of total phenols (41.61±0.10 mg GAE/g) and flavonoids (52.50±0.53 mg QE/g) were found in *T. erecta*. Maximum amount of tannins were recorded from *T. procumbens* (59.61±0.15 mg TC/g), followed by *F. benghalensis* (44.01±0.41 mg TC/g), *A. vera* (19.51±0.32 mg TC/g), *A. indica* (14.17±0.60 mg TC/g) and *T. erecta* (9.920±0.24 mg TC/g).

## Wound healing potentials of different phytochemical formulations

Wound contractions recorded on topical application with different concentrations *viz.,* 2%, 5%, 10% and 25% of F-I as measured on day 20$^{th}$ were 85.35%, 100%, 94.04% and 88.50% respectively. F-I 5% on topical application showed the maximum wound contraction (100%) which is significant (pd"0.001) as compared to the reference (Table-3 and Fig. 1).

Wound contractions recorded on topical application with different concentrations of F-II *viz.*, 2%, 5%, 10% and 25% measured on day 22$^{nd}$ were 88.75%, 100%, 98.02% and 82.12% respectively. F-II 5% on topical application showed the maximum wound contraction (100%) which is significant (pd"0.01) as compared to reference (Table-4 and Fig. 2).

Wound contractions recorded on topical application with different concentrations of F-III *viz.*, 2%, 5%, 10% and 25% on day 18$^{th}$ were 91.03%, 100%, 97.37%, and 87.50% respectively. F-III at 5% on topical application showed the maximum wound contraction (100%) which is highly significant (pd"0.0001). 5% of F-III showed superior wound contraction from 4$^{th}$ day onwards and angiogenesis was significantly higher than

**TABLE-3: Wound healing potential of F-I on excision wounds in *Wistar* rat model**

| *Experimental Groups* | Wound area ($mm^2$) | | | | | |
|---|---|---|---|---|---|---|
| | $0^{th}$ day | $4^{th}$ day | $8^{th}$ day | $12^{th}$ day | $16^{th}$ day | $20^{th}$ day |
| **Group 1 (Control)** | 400.0±2.2 (0.0%) | 386.7±6.6 (3.33%) | 336.5±22.0 (12.98%) | 256.2±6.6 (35.95%) | 190.0±5.7 (52.50%) | 121.5±6.6 (69.63%) |
| **Group 2 (Reference)** | 398.0±4.2 (0.0%) | 373.3±6.6 (6.21%) | 336.7±12.0 (15.40%) | 246.7±20.2 (38.02%) | 127.1±13.2 (68.09%) | 62.3±5.6*a (84.35%) |
| **Group 3 F- I (2%)** | 400.0±5.6 (0.0%) | 370.0±11.5 (7.50%) | 333.2±14.5 (16.70%) | 273.5±8.5 (31.63%) | 130.3±102 (67.43%) | 59.2±8.2*a (85.20%) |
| **Group 4 F- I (5%)** | 400.0±4.5 (0.0%) | 321.2±8.1 (19.70%) | 246.7±4.6 (38.33%) | 113.2±10.4 (71.70%) | 23.5±15.3 (94.13%) | 0.0±0.0***a*b (100%) |
| **Group 5 F- I (10%)** | 396.0±8.9 (0.0%) | 350.0±5.1 (11.62%) | 296.7±9.3 (25.08%) | 198.4±10.5 (49.90%) | 89.2±11.5 (77.47%) | 23.6±8.3**a (94.04%) |
| **Group 6 F- I (25%)** | 398.0±4.6 (0.0%) | 370.2±5.3 (6.98%) | 315.0±4.2 (20.68%) | 234.0±10.5 (41.2%) | 121.3±4.8 (69.52%) | 45.5±7.2**a (88.50%) |

Data are expressed as mean ± SEM. Wound area is expressed in $mm^{2.}$ Figures in parentheses indicate percentage of wound area and is taken as measure of wound healing * $p<0.05$, ** $p<0.01$, *** $p<0.001$. Data were analyzed by One way ANOVA, **a** = compared with control , **b** = compared with the reference.

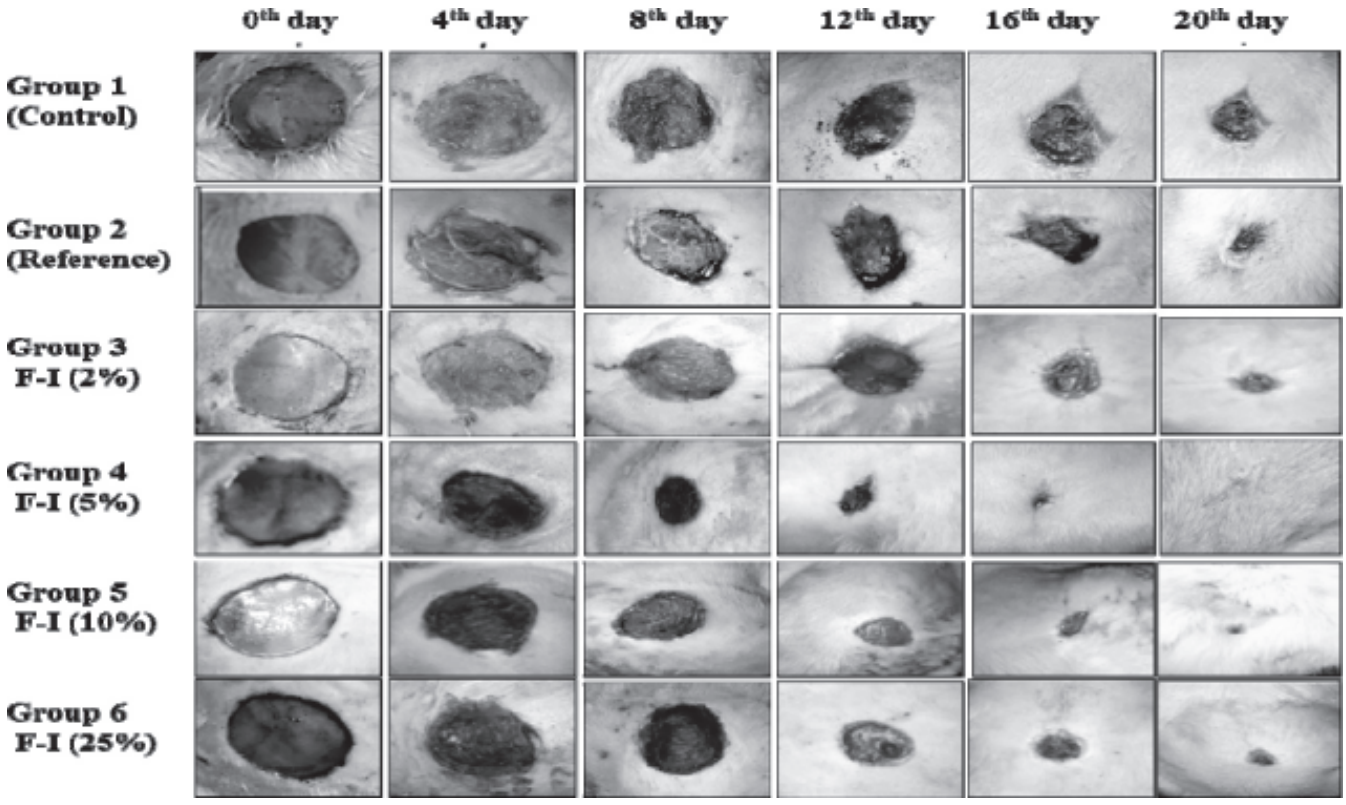


**Fig. 1: Images of wound areas in different groups of experiment I at different time intervals**

**TABLE-4: Wound healing potential of F-II on excision wounds in *Wistar* rats**

| *Experimental Groups* | Wound area ($mm^2$) | | | | | | |
|---|---|---|---|---|---|---|---|
| | 0th day | 4th day | 8th day | 12th day | 16th day | 20th day | 22nd day |
| **Group 1 (Control)** | 400.0±2.2 (0.0%) | 386.7±6.6 (3.33%) | 336.5±22.0 (12.98%) | 256.2±6.6 (35.95%) | 190.0±5.7 (52.50%) | 121.5±6.6 (69.63%) | 76.5±4.2 (80.55%) |
| **Group 2 (Reference)** | 398.0±4.2 (0.0%) | 373.3±6.6 (6.21%) | 336.7±12.0 (15.40%) | 246.7±20.2 (38.02%) | 127.1±13.2 (68.09%) | 62.3±5.6 (84.35%) | 10.8±4.8 (97.11%) |
| **Group 3 F- II (2%)** | 401.0±2.2 (0.0%) | 360.0±1.2 (10.22%) | 315.3±10.3 (21.37%) | 247.2±12.4 (38.35%) | 163.3±6.4 (59.28%) | 89.3±9.7 (77.73%) | 40.5±10.2*a (88.75%) |
| **Group 4 F- II (5%)** | 400.8±6.3 (0.0%) | 343.3±8.4 (14.35%) | 276.4±9.3 (31.04%) | 196.0±10.5 (51.10%) | 62.7±11.8 (84.36%) | 13.1±4.3 (96.73%) | 0.0±0.0**ab (100%) |
| **Group 5 F- II (10%)** | 399.0±5.3 (0.0%) | 366.7±5.8 (8.10%) | 318.0±4.5 (20.30%) | 266.3±11.5 (33.26%) | 196.3±11.0 (50.73%) | 66.5±14.2 (83.33%) | 7.9±5.6*a (98.02%) |
| **Group 6 F- II (25%)** | 400.0±8.5 (0.0%) | 386.7±6.5 (3.33%) | 336.5±4.8 (15.88%) | 293.2±6.5 (26.7%) | 240.0±4.3 (40.0%) | 153.3±6.2 (61.68%) | 73.0±7.1*a (82.12%) |

Data are expressed as mean ± SEM. Wound area is expressed in $mm^2$. Figures in parentheses indicate % of wound area and is taken as measure of wound healing * $p<0.05$, ** $p<0.01$. Data were analyzed by One way ANOVA, **a** = compared with control, **b** = compared with the reference.

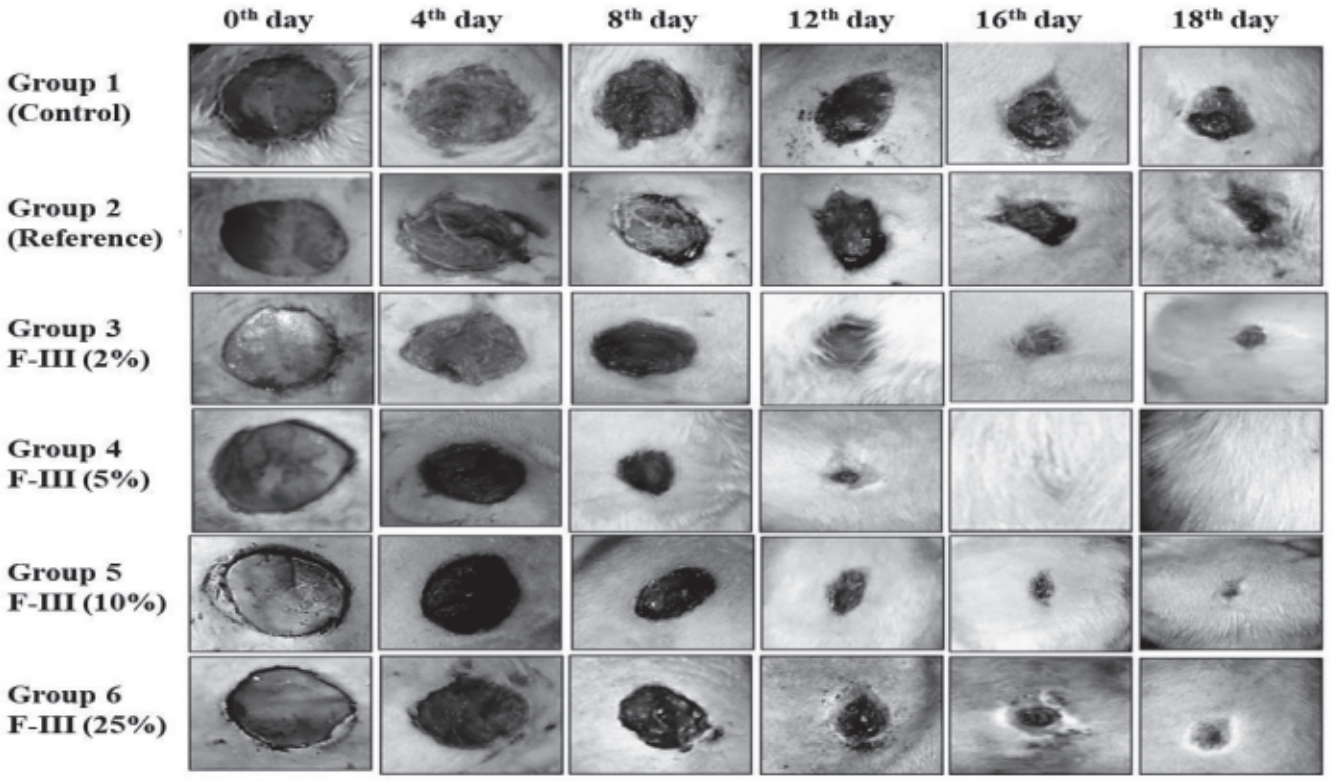


**Fig. 2: Images of wound areas in different groups of experiment II at different time intervals**

**TABLE-5: Wound healing potential of Formulation III on excision wound in *Wistar* rats**

| *Experimental Groups* | Wound area ($mm^2$) | | | | | |
|---|---|---|---|---|---|---|
| | **$0^{th}$ day** | **$4^{th}$ day** | **$8^{th}$ day** | **$12^{th}$ day** | **$16^{th}$ day** | **$18^{th}$ day** |
| **group 1 (Control)** | 400.0±2.2 (0.0%) | 386.7±6.6 (3.33%) | 336.5±22.0 (12.98%) | 256.2±6.6 (35.95%) | 190.0±5.7 (52.50%) | 157.5±6.6 (60.63%) |
| **Group 2 (reference)** | 398.0±4.2 (0.0%) | 373.3±6.6 (6.21%) | 336.7±12.0 (15.40%) | 246.7±20.2 (38.02%) | 127.1±13.2 (68.09%) | 82.3±6.6*a (79.32%) |
| **Group 3 F-III (2%)** | 395.6±3.5 (0.0%) | 355.3±6.6 (10.26%) | 280.4±11.5 (29.17%) | 190.0±10.0 (52.01%) | 73.3±13.2 (81.49%) | 35.5±6.6**ab (91.03%) |
| **Group 4 F- III (5%)** | 402.0±6.0 (0.0%) | 310.0±11.5 (22.90%) | 206.7±8.8 (48.59%) | 70.0±11.5 (82.59%) | 2.5±11.8 (99.38%) | 0.0±0.0****ab (100%) |
| **Group 5 F- III (10%)** | 398.9±8.5 (0.0%) | 340.0±5.7 (14.77%) | 280.0±8.8 (30.81%) | 151.3±12.0 (62.07%) | 37.8±15.2 (90.52%) | 10.5±6.6***a**b (97.37%) |
| **Group 6 F- III (25%)** | 396.0±7.2 (0.0%) | 346.7±6.6 (12.47%) | 273.0±5.7 (31.08%) | 198.0±5.7 (52.0%) | 98.8±4 (75.18%) | 49.5±6.6**a*b (87.50%) |

Data are expressed as mean ± SEM. Wound area is expressed in $mm^2$. Figures in parentheses indicate % of wound area and is taken as measure of wound healing *p<0.05, **p<0.01, *** p<0.001, ****p<0.0001. Data analyzed by One way ANOVA, **a** = compared with control (Base), **b** = compared with the reference.

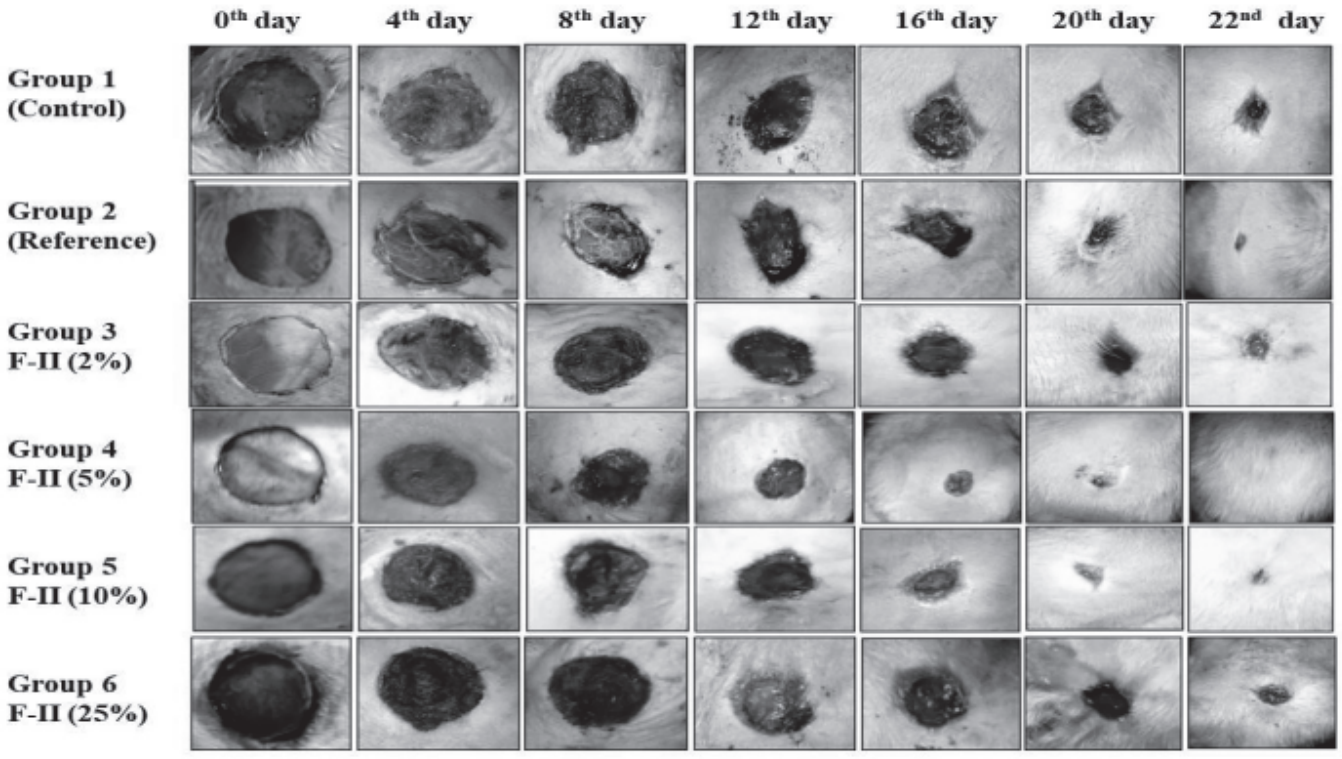


**Fig. 3: Images of wound areas in different groups of experiment III at different time intervals**

in control (Table 5 and Fig. 3). The 5% F-III showed significantly faster completely wound contraction at 18th day with absence of scar compared to F-I (20th day) and F-II (22nd day) and reference group.

## Discussion

Good wound care is essential for effective management. Wound healing involves replacing damaged skin tissue through biological processes like clot formation and tissue generation. Antiseptics may have a toxic effect on tissues and are not suitable for open wounds. It's advised to use them with caution as their toxicity might outweigh any benefits[13]. Some reports suggest that they may exhibit cytotoxicity and are advised against their application on open wounds. Wound healing agents primarily function in the Inflammatory and Proliferative phases[17]. Common antiseptics may have a toxic effect on tissues and caution is advised when using them. It's recommended to combine traditional and modern wound healing agents for optimal care.

Various studies have reported an improved wound-healing process following topical application of herbal products[31]. The results of the present study demonstrated wound healing abilities of ethanolic extracts of a specified set of plants/herbs as Formulations in excision wounds, as evident by the generation of granulated tissue and remarkable increase in the rate of wound contraction compared to the reference formulation *viz.,* Betadine. Phytochemicals-based formulations appear to influence one or more stages resulting in faster wound closure when compared to the control and reference groups.

Medicinal plants have different phytochemicals as secondary metabolites. Phytochemicals such as flavonoids, phenols and tannins present in phytochemical formulations are shown to possess anti-microbial, anti-inflammatory, anti-oxidant activities and are thus responsible for wound healing activity[5], because Tannins have strong astringent property and promote capillary vasoconstriction, which decrease vascular permeability and cause a local anti-inflammatory effect[23]. Flavonoids are well known for their antioxidant potential and antibacterial properties which promote the wound healing process by wound contraction, increased rate of epithelialization and raise the level of hydroxyproline supporting homeostasis[19].

The results of this study seem to confirm the use of F-III consisting of *T. erecta, T. procumbens, A. indica, F. benghalensis, C. longa* and *A. vera* for faster and effective treatment of excision wounds followed by F-I and F-II. The extracts of these plants can be developed into phytomedicines for wider application in the management of wounds.

## Conclusion

After conducting experiments on excision wounds, it can be concluded that the topical application of all phytochemical formulations -I, II, and III separately shows significant wound healing activity. This was evidenced by faster generation of fibrocollagenous tissue, neovascularization, epithelialization, and anti-bacterial functions. F-III (5%) proved to be superior in achieving faster wound healing than the other formulations (F-I and F-II) studied. Since *Tagetes erecta, Tridax procumbens, Azadirachta indica, Ficus benghalensis, Aloe vera* and *Curcuma longa* are widely available and abundant, they could provide a fairly economical wound healing agent for wider application in wound care management. Further studies are required to identify the active compounds participating in processes of wound healing.

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