Published online in http://ijam.co.in ISSN No: 0976-5921

*International Journal of Ayurvedic Medicine, Vol 13 (2), 424-428*

# Evaluation of wound healing activity of *Dhataki Pushpa* ointment (*Woodfordia fruticosa* Kurz.) on *Sadyavrana* (excised wound) - An experimental study

**Research Article**

**Vidhate R Sneha[1*], Meena D S[2], Rajkumar C[3], Nilima W[4], Shamli H[5]**

1. Assistant Professor, Department of Dravyaguna, 3. Assistant Professor, Department of Samhita & Siddhanta, 4. Associate Professor, Department of Agadatantra, 5. Assistant Professor, Department of Panchakarma, Mahatma Gandhi Ayurved College, Hospital & Research Centre, Datta Meghe Institute of Medical Sciences (DMIMS), Wardha, Maharashtra. India. Associate Professor, Department of Dravyaguna, AIIA new Delhi, India.

## Abstract

Background: *Woodfordia fruiticosa* Kurz. of Lythraceae is a shrub whose blooms are called *Dhataki Kusuma*. The fresh flower of W. fruticosa controls bleeding in emergency cuts, whereas the dried flower powder heals wounds faster. Indian tribes utilise it to cure wounds. No scientific evidence supported using W. fruticosa to treat wounds, hence the present study assessed the plant's wound healing ability. Aim: To find out the efficacy of *Dhataki Pushpa* ointment on *Sadyavrana* (excised wound). Material & Methodology: BSI (Botanical Survey of India) verified *Dhataki* flowers. The ointment was prepared and then standardised. This research randomly chose 12 Wistar rats (six for *Dhataki Pushpa* ointment, and six for betadine). Albino rats (Wistar) of both sexes weighing 150-200 g were used in this research to examine the impact of *Dhataki Pushpa* ointment on the healing of Sadyavrana (incised wound). Observations: Progressively wound contraction was assessed on 0 days, 4th, 8th, 12th, and 16th days in both groups using graph paper (mm2) and represented as a percentage. Result: the statistical analysis (unpaired t-test) revealed that there is a significant difference in the wound healing activity of both the ointments on *Sadyavrana* ($p<0.0001$). Discussion: *Dhataki* extract is antibacterial and antifungal. Tannins and alcohol concentration may speed wound contraction and healing. Reduced wound contraction indicates *Dhataki* ointment's superiority over betadine. Conclusion: *Dhataki* flower ointment has a significant effect on *Sadyavrana* when compared with betadine ointment.

**Key Words:** *Dhataki Pushpa, Sadyavrana, Woodfordia fruticosa* Kurz., Wound healing activity.

## Introduction

*Woodfordia fruticosa* Kurz. belongs to the Lythraceae family and maybe a ligneous plant found ordinarily, the flowers of this plant were employed in totally different medicative preparations in India by the name of *Dhataki Kusuma* (1). The market drug that consists of dried fruits, flowers, buds and broken items of inflorescence were sold (2, 3). Dried flowers are authorised with stimulant and astringent properties (4). Therapeutically these are employed in *Vrana* (wound), *Raktastrava* (haemorrhages, menorrhagia) and *Shukradosha* (seminal abnormalities) (5). Excluding these flowers containing self-generated alcohol are used extensively in the preparation of *Asava* and *Arishta*. In Bhrihatrayi, the *Vranaropana* property of *Dhataki Pushpa* has been emphasized in the various conditions of *Vrana* (6,7). A similar study was done like "wound healing potential of flowers extracts of *Woodfordia fruticosa*" (8) and an experimental study to gauge the efficacy of *Dhatakyadi yoga lepa* in the healing of burns (9).

In Ayurveda, Sushrutacharya stated that *Vrana* Should be considered as *Vyadhi* as a result of it causing *Vedana* (pain) to the *Body* and *Mind*. The knowledge of the wound and its healing was abbreviated and classified on the premise of various stages by Vagbhata who advocated the preparation and application of drawn ghee based preparation, mostly an ointment for native use (10). There are several drugs mentioned in Ayurveda, some are extra-pharmacopeial medicines like *Jayanti* veda (*Tridax procumbens*) ordinarily used for *Vranaropana* (wound healing) activity. The body's normal process of renewing dermal and epidermal tissue is known as wound healing or repair. The drug *tridax* exhibits antimicrobial activity against gram-positive and gram-negative microorganisms. Could also be this property helps to prevent infection and leads to wounding healing methods (11). Traditionally *Woodfordia fruticosa* Kurz is used in wound treatment by tribes of Chhattisgarh, India. No scientific data was supporting the use of Woodfordia fruticosa for treating wounds. Therefore it is time to evaluate the *Vranaropana* karma of *Dhataki Pushpa* on the premise of its action or properties as the earlier textual claims of

*** Corresponding Author:**

**Vidhate R Sneha**

Assistant professor, Department of Dravyaguna, Mahatma Gandhi Ayurved College, Hospital & Research Centre, Salod (H), Wardha, 442001. Maharashtra. India.

Email Id: snehavidhate1@gmail.com

Ayurveda. Hence the present study was carried out in Wistar rats to prove its efficaciousness on *Sadyavrana* (excised wound) in the form of topical application as an ointment.

**Aim:**

- To evaluate the wound healing activity of *Dhataki Pushpa* ointment on *Sadyavrana* in wistar rats.

**Objectives:**

- To compare the wound healing activity of *Dhataki Pushpa* ointment and betadine ointment on *Sadyavrana* in wistar rats.
- To assess the wound healing activity of *Dhataki Pushpa* ointment on *Sadyavrana* in wistar rats.
- To assess the wound healing activity of betadine ointment on *Sadyavrana* in wistar rats.

## Materials and methods

### Materials

**Plant Material:**

Initially *Dhataki* flowers were collected from Seva Village, Wardha (Maharashtra). For authentication purposes, a Herbarium was prepared and sent to the Botanical Survey of India (BSI), Pune (Maharashtra) and received authentication letter Specimen NO. SRV No./BSI/WRC/IDEN.CER/2016/692 on dated 5/02/2016. After that, the plant material was collected in March 2016 and allowed to shade in dry. These flowers were used for the preparation of ointment and the same was used for the present experimental study.

**Animals**

Twelve healthy Wistar rats of 150 to 200 gm weight of either sex were obtained from the Animal House of JNMC Sawangi (M) Wardha, Maharashtra under the ambit of Datta Meghe Institute of Medical Sciences (Deemed to be University) and randomly allocated in two groups. All the animals were kept in the animal house maintained at 27 °C ± 2 °C under 12 hrs dark/light cycle. They were fed with standard rat feed and water ad labitum was provided.

**Ethical Clearance**

The procedure was reviewed and approved before the initiation of the experimental study by the Institutional Animal Ethics Committee (Letter No: 571/ a/CPCSEA) on 25/4/2015.

### Methods

**Ointment preparation for topical application**

Water extract of W*oodfordia fruticosa* flowers was used for the preparation of the ointment for topical application. 0.5% (W/W) of *Dhataki Pushpa* extract ointment was formulated using Glycerine 50 gm and Sodium benzoate 2 gm. mixed and stirred well very slowly for 15 minutes (fig 1). After the completion of this procedure, the prepared ointment was sent for analysis at "Unijules Pharmaceuticals", Kamthee, Nagpur for standardization. All the necessary quality control parameters were tested in the prepared ointment such as Ph, microbial content, viscosity, spreadability, heavy metal analysis and HPTLC. After that specification No. F1112- S. (Annexure-1) of the Standard product of *Dhataki Pushpa* ointment was given.

**Wound healing evaluation**

**Excision Wound Model**

Two groups of six animals (Wister rats) weighing 150-200 g either of sex were used in this study. The animals were acclimatized for six days under aseptic laboratory conditions. They were housed in polypropylene cages and maintained at 27 °C ± 2 °C under 12 hrs dark/light cycle and fed with standard rat feed, water ad libitum was provided. Under aseptic conditions, the rats were anaesthetized by using ketamine injection 30 mg/kg/BW (Body Weight) with insulin syringe and 1.5 to 2cm excision was created on the dorsal region of the rat under sterile conditions and haemostasis was achieved by blotting the wound with a cotton swab soaked in normal saline. 12 Wistar rats were randomly divided into two groups (group 1 & group 2). Group 1 was treated with *Dhataki Pushpa* ointment whereas group 2 was treated with betadine ointment in a quantity sufficient (qs) with a sterile spatula for two times a day as the standard method. Wound area was measured on 0 day, 4$^{th}$, 8$^{th}$, 12$^{th}$, and 16$^{th}$ days with the help of tress paper. For post wound for determination of wound contraction calculated through the formula. The completely healed percentage of wound contraction of both groups was compared. (Table-1 & 2).

**Measurement of wound area**

The progressive changes in wound area were measured graphically by tracing the wound margin on a graph paper on 0 day, 4$^{th}$, 8$^{th}$, 12$^{th}$ and 16$^{th}$ days. The changes in the healing of wound i.e. measurement of wound contraction on graph paper were measured as a unit (mm$^2$). Wound contraction was expressed as a percentage reduction of original wound size (12).

**Formula**

$$\% \text{ of Contraction} = \text{Healed Area} \times 100 / \text{Total Wound area}$$

{Healed area = Original wound area – Present wound area}

**Statistical Analysis**

All the data were expressed as mean ± SEM. The statistical significance between the two groups was compared by applying an unpaired t-test. Which showed significant differences. (Table No.3). It was observed that P-value is less than 0.05 hence it can be said that there is a significant difference in wound healing activity between the two groups.

## Results

The standardization of *Dhataki Pushpa* ointment has been done with the help of unijules pharmaceutical

company in which pH, Viscosity, HPTLC, Heavy metal analysis were done and then the efficacy of wound healing in animals. The studies on the excision wound healing model reveal that all the two groups showed the reduction of wound area from 0 day to 16 day. However, on the 16th post wounding day, group 1 (*Dhataki Pushpa* ointment ) showed 100% contraction of wound whereas group 2 (standard betadine ointment) showed 90% contraction of the wound which resulted in significant wound healing activity was observed in *Dhataki* ointment in comparison to standard betadine ointment. (Table-3)

## Discussion

*Woodfordia fruticosa* is also known as *Dhataki* and fire flame bush. *Dhataki* plant was utilized vividly in many ayurvedic formulations by traditional practitioners. Although, all parts of the *Dhataki* plant possess valuable therapeutic properties, flowers are considered a potent part and it has high demand in the market. Ayurveda compendia advocate that *Dhataki Pushpa* is *Kashaya* (Astringent ) in taste, possess *Laghu* (Light), *Ruksha* (dry) properties, *Katu* (Pungent) in *Vipaka* (Metabolism), *Sheeta* (Cold) in *Veerya* (potency), *kapha–Pitta hara*, *Madakari* (narcotic) in *karma* (action), due to these properties, it pacifies *kapha* and *pitta dosha* while aggravates *Vata dosha*; and the flower of *Dhataki* are highly beneficial in the management of *Atisara*, *Vrana*, *Arsha* etc.

Sushruta, the father of ancient Indian surgery, has allocated a separate chapter on *Vrana*, which emphasizes the importance of wound management. In Ayurveda treatises, grouped herbs are called either *Varga* or *Gana*. This *gana or varga* are named based either on Pharmacological action or based on the name of the main plant in Charaka Samhita and Sushruta Samhita respectively. Having healing potential of *Dhataki* is mentioned in *Priyangavadi*, *Ambasthadi* and *Sandhaniya gana* for *vranropana* (13,14). Woodfordia fruticosa in preparation in a substantial increase of the inhibition of both human complement activity and chemiluminescence generated by zymosan-stimulated human polymorphonuclear leukocytes. It was established that the increased biological activity was not due to microbial inference but due to immune active constituent release from the Woodfordia flowers. Significantly raised alcohol content upon addition of *Dhataki* flowers.

*Dhataki* has a wide range of uses which was prescribed in many diseases with or without combination in any preparation. So, there is a definite need to produce advanced treatment modalities in Ayurveda. As the application of ointment in wound healing rather than a paste application, the ointment of *Dhataki Pushpa* was prepared for wound healing purposes in this study. The quality control standards of *Dhataki* ointment was done in which some required parameter was tested such as pH (7.2), Viscosity (169 cps), HPTLC, stability, Spreadability, Penetratrability was complied, the microbial limit was 124 cfu/g, heavy metals complied. Escherichia species, salmonella species were absent in the sample. These all parameter was done to determine the quality standardization of *Dhataki* ointment. An experimental study on the animals was done in which 12 Wistar rats were selected and randomly allocated in each group. In the 1st group, Six animals were treated with *Dhataki* ointment and the 2nd group with Betadine ointment. During the study, on 1st, 4th, 8th,12th and 16th days, the period contraction of the wound was determined in which the contraction of the wound on 1st day of *Dhataki* ointment was 2.77 % whereas in Betadine it was 10.93% which is more but on the 8th day wound contraction was 30.55 % in group 1 and 24.27 % of group 2. On the 12th day, it was 54.86 % of group 1 whereas 71.09% of group 2. On the 16th day contraction of the wound was measured for the group treated by *Dhataki* ointment showed 100% wound contraction whereas 2nd group treated with betadine ointment showed 90 % wound contraction (all the progressive changes with satistical analysis were depicted in Table-1- 3 & figure-1).

The aim of quality control standards of ointment was a healthy skin that has slightly acidic ph and Viscosity was measure a fluid resistance to flow, it describes the internal friction of a moving fluid (15). In spreadability, the principle is related to the contact angle of the drop of a liquid or a semisolid preparation on a standardized substrate and is a measure of lubricating, which is directly related to the coefficient of friction.

A microbial study has also been done to check the absence of any microbes, which is harmful in the finished end product (16). In this way, the quality control parameter is obligatory to generate the efficacy of the finished product. Through its *kashaya rasa, Ruksha*, *Laghu guna Dhataki* flowers absorb moisture and *kleda* from the wound. Therefore, the union of tissue act as styptic action, it heals wound by removing secretions from them. It removes *doshas* residing in the skin and nourishes it (17). Some similar studies showed the effect of *Dhataki* flowers on *vrana*, antimicrobial activity and antiviral activity (18).

### Scope and future study

The study was limited to 6 rats in each group only. The present work is based on an animal experimental study and there can be Variation in pharmacodynamics and pharmacokinetics of drug reactions in animals and on the human being. So there is a definite need to evaluate a clinical study of the wound healing activity of *Dhataki Pushpa*. in further aspects.

## Conclusion

The duration taken for complete wound contraction in *Dhataki* ointment was much earlier when compared with standard betadine ointment. This was an innovative approach of *Dhataki Pushpa* ointment preparation for topical application which indicates to conduct the future studies on various wound conditions. Hence, it is concluded that the result of the *Dhataki* ointment is having potent wound healing activity than betadine ointment in Wistar rats.

**Conflicts of interest**

No conflicts of interest

**Financial Support**

The present work is funded by Datta Meghe Institute of Medical Sciences (Deemed to be University), Wardha, Maharashtra, India.

**Acknowledgements**

This work would not have been possible without the financial support of DMIMS & MGACH & RC. The authors are indebted to Dr. Sandip Joge, Dr. Sarang Despande, Dr. Akshay Pargaokar and our gardener Mr. Shamrao Timade who have been supported positively for this study.

## References

1. Anonyms, Council for Scientific and Industrial Research. A Dictionary of Indian Raw Materials and Industrial Products, the Wealth of India (Raw Materials). Vol. X New Delhi: Publications and Information Directorate; 1998; p 586.
2. Uday M, Kishor D, Raghuvanshi A. A Pharmacognostic and pharmacological overview on *Woodfordia fruiticosa* Kurz. Scholars Academic Journal of Pharmacy, 2014;3(5): p 418-22.
3. Warrier PK. Indian Medicinal Plants, Volume 5, Hyderabad: Orient Longman Private Ltd, 1994; p.412.
4. Finose A, Devaki K. Phytochemical and Chromatographic studies in the flowers of *Woodfordia fruiticosa* (L) Kurz.; Asian J Plant Sci Res 2011;1(3):p 81-5.
5. Khare CP. Indian Medicinal plants- An illustrated Dictionary. New Delhi; Springer (India) Pvt Ltd.; 2007; p 720.
6. Bramhshankar mishra, kashinath shastri ,Charak samhita, Choukhamba Bharti Academy varanasi, part 2, edition 2007, chikitsasthan, chapter 25/89, New Delhi, pp 711.
7. Kaviraj Ambika Datta Shastri, Sushrut samhita, Choukhamba Sanskrit samsthan Varanasi, vol 1,sutrasthan , chapter 38/47,45,46, pp 187.
8. Mukharjee PK. Quality control of Herbal drugs, Business Horizons pharmaceutical publishers, New Delhi, 1st edition 2002; p 131-219.
9. Ramesh killedar, s.v.emmi, original research article an experimental study to evaluate efficacy of dhatakyadi yoga lepa in burn wound healing, Journal of Ayurveda and Holistic Medicine | April, 2014 | Volume 2 | Issue 4
10. Bhisagacharya harisashtri paradakara Vaidya, Ashtang Hridaya, commentaries of Arundatta and Hemadri, Choukhamba Orientalia Varanasi, Sutrasthan 15/38, pp 238.
11. Kethamakka SRp, Meena SD, Jayanti veda (Tridax procumbens) - Unnoticed Medicinal plant by Ayurveda,pp-6-22
12. Mortan,J.J.Pand M.H.Malone, 1972, Arch.Int.Pharmacodynamics,19: 117-126.
13. Tripathi Ravi Datta, Charak Samhita of Agnivesh reducted by Dridbala,Vol 2, 1st ed, Chikitsaaa sthan 25/29 -30, reprint 2001, Chaukhambha bharati Academy, Varanasi, pp 614.
14. Ambikadatta Shastri ed, Sushruta samhita of Sushruta, Vol I, Reprint 2005, Sutrasthan 38/45,46, Chaukhamba Sanskut Sansthan, Varanasi, pp128.
15. Available from https://www.quora.com/what is relationship - viscosity & gel time. Accessed on 25.03.2022.
16. Kartar singh Dhiman, et.al., Quality control evaluation of keshanjana & keshmasi eye ointment, ayu – 2014 Jan- mar 35(1),58-62.
17. Dhyani S.C, Rasapanchaka, Ayurvedic principles of drugs action,Choukhmba krishnadas academy, edition 2, year 2003, pp 43.
18. Neeraj Varma, G. Ambresh et.al, Wound healing potential of flowers extract of *Woodfordia fruticosa* Kurz, Indian journal of biochemistry vol 50 aug 2013, pp 296- 304, revised 11 june 2013, pp 296.

**Table-1: Day-wise wound contraction in *Dhataki Pushpa* ointment group**

| Sr. No | Initial Day- Graphical area (GA) | 4th Day GA | 4th Day % | 8th Day GA | 8th Day % | 12th Day GA | 12th Day % | 16th Day GA | 16th Day % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 144 | 140 | 2.77 | 100 | 30.55 | 65 | 54.86 | 5 | 96.52 |
| 2 | 183 | 154 | 15.84 | 91 | 50.27 | 27 | 85.24 | - | 100 |
| 3 | 109 | 104 | 4.58 | 25 | 77 | 10 | 90 | - | 100 |
| 4 | 189 | 170 | 10.05 | 166 | 12.16 | 85 | 55 | | 100 |
| 5 | 110 | 108 | 1.81 | 68 | 38.18 | 38 | 65.45 | 3 | 97 |
| 6 | 163 | 162 | 0.61 | 35 | 78.52 | 27 | 83.43 | - | 100 |

**Table-2: Day-wise wound contraction in Betadine ointment group**

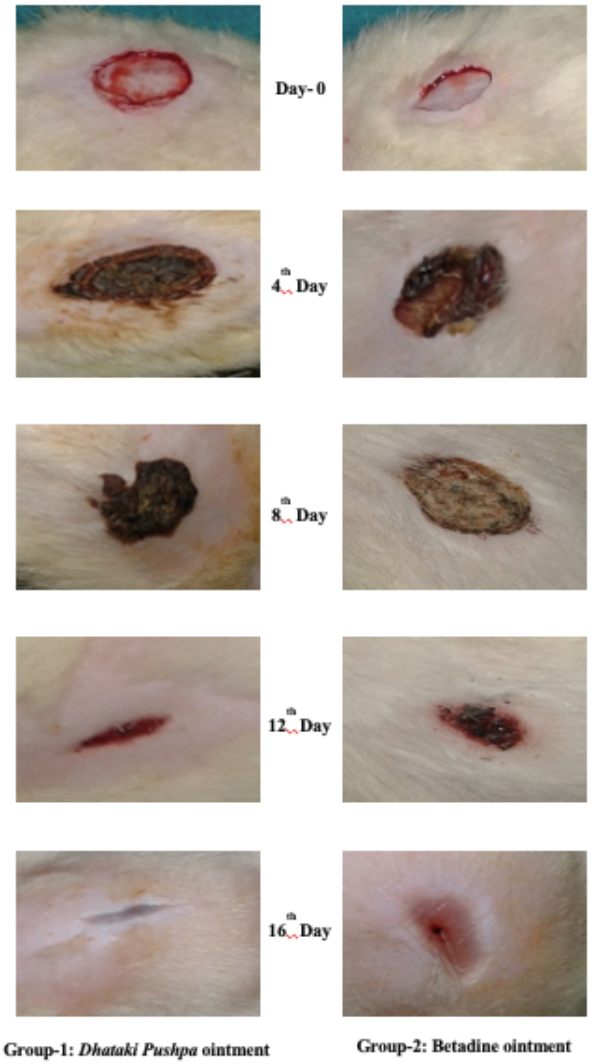
| Sr. No | Initial Day-Graphical area (GA) | 4th Day GA | 4th Day % | 8th Day GA | 8th Day % | 12th Day GA | 12th Day % | 16th Day GA | 16th Day % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 173 | 154 | 10.98 | 131 | 24.27 | 50 | 71.09 | - | 95 |
| 2 | 184 | 178 | 3.26 | 50 | 72.82 | 13 | 92.39 | 6 | 96 |
| 3 | 153 | 133 | 13.07 | 67 | 56.20 | 32 | 79.08 | 2 | 98 |
| 4 | 155 | 139 | 10.32 | 26 | 83.22 | 20 | 87.09 | - | 100 |
| 5 | 141 | 140 | 0.70 | 32 | 77.30 | 25 | 82.26 | 2 | 98 |
| 6 | 100 | 98 | 2 | 15 | 85 | 7 | 13 | 4 | 96 |

**Table- 3: Showing the statistical analysis of wound contraction in both the groups**

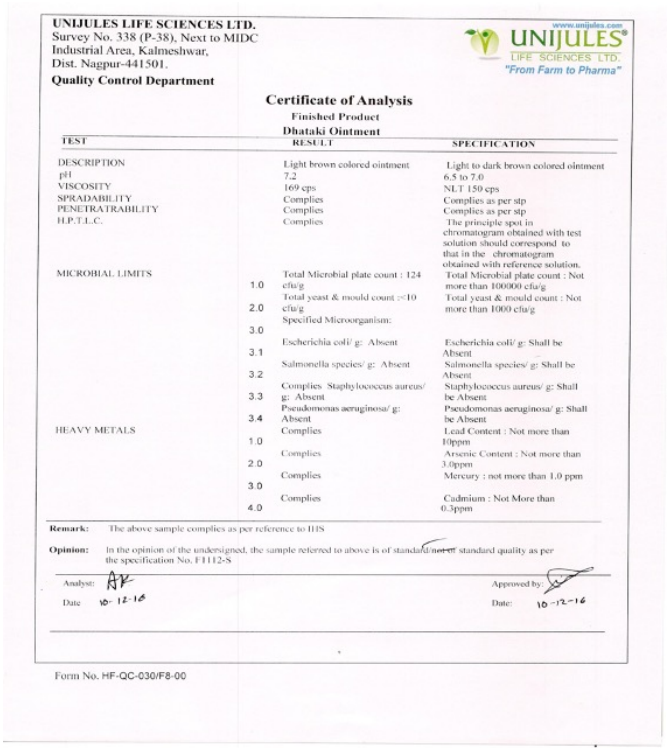
| Days | Mean of GA ( *Dhataki* ointment) | Mean of GA (Betadine | SD | SEM | t- value | p-value* |
|---|---|---|---|---|---|---|
| **Initial Day** | **144** | **173** | **20** | **14.5** | 5.311 | 0.0001* |
| **4th Day** | **139** | **140** | **0.70** | **0.5** | | |
| **8th Day** | **80** | **54** | **19.09** | **13.0** | | |
| **12th Day** | **42** | **25** | **12.05** | **8.5** | | |
| **16th Day** | **2** | **5** | **2.12** | **1.12** | | |

***Unpaired t-test**

**Figure-1: A pictorial presentation depicting the comparative efficacy of *Dhatakai Pushpa* ointment and betadine ointment on *Sadyavrana* (excised wound) in both the groups**




Group-1: *Dhataki Pushpa* ointment Group-2: Betadine ointment



UNIJULES LIFE SCIENCES LTD.
Survey No. 338 (P-38), Next to MIDC
Industrial Area, Kalmeshwar,
Dist. Nagpur-441501.
Quality Control Department

UNIJULES LIFE SCIENCES LTD. "From Farm to Pharma"

**Certificate of Analysis**

Finished Product
Dhataki Ointment

| TEST | | RESULT | SPECIFICATION |
|---|---|---|---|
| DESCRIPTION | | Light brown colored ointment | Light to dark brown colored ointment |
| pH | | 7.2 | 6.5 to 7.0 |
| VISCOSITY | | 169 cps | NLT 150 cps |
| SPREADABILITY | | Complies | Complies as per stp |
| PENETRATRABILITY | | Complies | Complies as per stp |
| H.P.T.L.C. | | Complies | The principle spot in chromatogram obtained with test solution should correspond to that in the chromatogram obtained with reference solution. |
| MICROBIAL LIMITS | | | |
| | 1.0 | Total Microbial plate count : 124 cfu/g | Total Microbial plate count : Not more than 100000 cfu/g |
| | 2.0 | Total yeast & mould count :<10 cfu/g | Total yeast & mould count : Not more than 1000 cfu/g |
| | 3.0 | Specified Microorganism: | |
| | 3.1 | Escherichia coli/ g: Absent | Escherichia coli/ g: Shall be Absent |
| | 3.2 | Salmonella species/ g: Absent | Salmonella species/ g: Shall be Absent |
| | 3.3 | Complies Staphylococcus aureus/ g: Absent | Staphylococcus aureus/ g: Shall be Absent |
| | 3.4 | Pseudomonas aeruginosa/ g: Absent | Pseudomonas aeruginosa/ g: Shall be Absent |
| HEAVY METALS | | Complies | |
| | 1.0 | Complies | Lead Content : Not more than 10ppm |
| | 2.0 | Complies | Arsenic Content : Not more than 3.0ppm |
| | 3.0 | Complies | Mercury : not more than 1.0 ppm |
| | 4.0 | Complies | Cadmium : Not More than 0.3ppm |

Remark: The above sample complies as per reference to IHS

Opinion: In the opinion of the undersigned, the sample referred to above is of standard/~~not of~~ standard quality as per the specification No. F1112-S

Analyst: AP Date: 10-12-16

Approved by: Date: 10-12-16

Form No. HF-QC-030/F8-00

*****