

Creation and Valuation of Ayurvedic Herbal Soothing Balm for Chemical Induced Wound

Mihir Y. Parmar ^{1*}, Kautuk Shah ², Zalak D. Dave ³, Palak Landge ³

¹Head & Professor, Department of Pharmacology.

²Head & Professor, Department of Pharmacognosy.

³Assistant Professor, Department of QA, Krishna School of Pharmacy & Research, Dr. Kiran and Pallavi Patel Global University (KPGU), Vadodara, Gujarat, India.

***Corresponding Author:** Mihir Y. Parmar, Head & Professor, Department of Pharmacology.

Received date: August 31, 2024; **Accepted date:** September 18, 2024; **Published date:** September 30, 2024

Citation: Mihir Y. Parmar, Kautuk Shah, Zalak D. Dave, Palak Landge, (2024), Creation and Valuation of Ayurvedic Herbal Soothing Balm for Chemical Induced Wound, *J. Clinical Case Reports and Studies*, 5(8); DOI:10.31579/2690-8808/219

Copyright: ©, 2024, Mihir Y. Parmar. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Abstract

Wounds are physical injuries that results in a breach or observance of the skin. Proper heal of wounds is very essential for the restoration of disrupted anatomical stability and disturbed functional status of the skin. Wound healing is a multifaceted but generally orderly process. Chronological impressions of dedicated cell types first clear the provocative injury and then progressively build the scaffolding to fill in any resulting defect. Cream is defined as semisolid emulsions which may be oil in-water (o/w) or water- in- oil (w/o) type and these semisolid emulsions are meant for external applications. Presently whole world including the developed country recognized the importance of traditional medicine and encouraging the research on herbal or ethnomedicine as they safe and less toxic. In this study we have formulated an herbal cream (o/w) type satisfying almost all the pharmaceutical parameters which showed better tissue restoration and healing capacity. The wound healing activity of herbal cream was experimentally evaluated by excision wound model. The experimental data of wound size area and histopathological study expressed that, healing in the cream treated group of animal was significant as compared to control group of animal.

Key Words: excision; herbal soothing balm; o/w emulsion; wound

Introduction

Wound may be defined as a loss or breaking of cellular and anatomic or functional continuity of living tissue. It is produced by physical, chemical, thermal, microbial, or immunological damage to the tissue. Wound healing or wound repair is the body's natural process of regenerating dermal and epidermal tissue.

Healing requires the collaborative efforts of various tissues and cell lineages. It involves aggregation of platelets, clotting of blood, fibrin formation, and an inflammatory response to injury, alteration in the ground substances, angiogenesis and re-epithelialisation. Healing process is not complete until the disrupted surfaces are firmly knit by collagen [1-3a, 4].

Cream is defined as semisolid emulsions which are oil in-water (o/w) or water- in- oil (w/o) type and these semisolid emulsions are intended for external applications. Creams are often composed of two phases. Oil-in-water (o/w) emulsions are most useful as water-washable bases, whereas water-in-oil (w/o) emulsions are emollient and cleansing agents. An

emulsifying agent is used to disperse the aqueous phase in the oily phase or vice versa [5-8].

World Health Organization (WHO) as well our country has been promoting traditional medicine because they are less expensive, easily available and comprehensive, especially in developing countries [3, 5].

It is also true that eight percent of the world's population relies on medicinal plants for their primary health care. Whole world including the developed country recognized the importance of traditional medicine and has treatment strategies, guidelines and standard for ethnomedicine [6, 9, 9a].

Although various types of cream is considered for wound healing but these are still appears to be limited in rate of tissue regeneration. Hence after a depth review regarding pathogenesis as well as different traditional and alternative therapy for wound healing, we have taken up the project to develop and formulate an herbal cream which will be effective and has

better rate of tissue regeneration [2, 3]. The herbal cream that is planned to be formulated for wound healing will be oil/water (o/w) emulsion type which will be less oily, less greasy and less sticky in nature so that patient compliance is more and will be beneficial for all kind of people in our society.

After thorough review of Traditional System of Ayurvedic and Homeopathic system of medicine we have selected following herbs to formulate the cream for wound healing action.

- *Panax ginseng* (promotes blood circulation, skin warming) [10],
- *Aloe vera* (regenerate tissue) [11, 12],
- *Calendula officinalis* (heal ulceration) [13],
- *Clerodendrum indicum* (antimicrobial action) [14, 15],
- *Arnica Montana*, (reduce pain due to injury) [16],
- Rose hip oil (rich source of vitamin C) [17, 18].

In Our research our attempt to formulate most complete Herb-O-Seal cream that contains herbs which will satisfy almost all the mechanism to heal a wound effectively.

Materials and Methods

Collection of Plant Materials

The dried crude drugs of *Calendula officinalis*, *Arnica Montana* and extracted powder of *Aloe vera* were collected from G Hakim supplier, Vadodara. The fresh leaves of *Clerodendrum indicum* was collected from Silvasa, Gujarat. The dried root of *Panax ginseng* was collected from G Hakim supplier, Vadodara. Rose hip oil was collected from Vasu Pharma, Vadodara.

Chemicals

The Chemicals used during the experiments were of analytical grade. Lanolin (Lions & Co), White Petroleum (Chemfine), Tween 60, Stearic acid (Hi-media Lab), Mineral oil, Triethanolamine, Propylene Glycol (Merck Lab), Betadine; Povidone-iodine IP 5% w/w, (Cipla medicare) etc were used.

Instruments

Freeze Dryer, Homogenizer, Centrifuge (REMI), Viscometer (Brookfield DV-E viscometer), Digital Balance (Delux Instrument), Digital pH meter (Sigma) etc were used.

Experimental Procedure

Extraction of plant materials

The extractions of crude drugs were carried out by Simple Maceration method using water (aqueous extract) as menstruum with occasional stirring. Liquid mixture is then pressed and filtered to get a clear liquid extract. The clear liquid is then subjected to freeze drying in order to get a solid mass [19, 20].

Formulation of herbal cream (O/W emulsion)

Ingredient of oil phase (A) was melted in a beaker by using water bath on constant stirring. Components of aqueous phase (B) were mixed together and warmed to about same temperature of oil phase (up to 70°C). The preservative methyl paraben and concentrated aqueous extract of the plants were added into aqueous phase and heated. Then oil phase was added to water phase little by little on constant stirring and perfume was added to it when the temperature was 35°C - 40°C. Six different formulations (F1-F6) were prepared by using varying concentration of aqueous extract, stearic acid and liquid paraffin [8, 21, 22].

Pharmaceutical evaluation of cream

The formulations or creams were evaluated for different pharmaceutical parameters: such as Type of emulsion (Dye method and Dilution method), Homogeneity, Appearance, After feel, Type of smear, Removal, Creaming or coalescence, Globule size analysis, pH (Digital pH meter, systronics), Extrudability, Viscosity, (Brookfield Viscometer), stability testing and Spreadability. The best formulation was selected on basis of their pharmaceutical parameters and evaluated for wound healing activity [7, 23-25].

In Vivo Evaluation of Herbal Cream

Skin irritation Test

The cream was evaluated for primary skin irritation test on experimental animals (shaved back of the rats) to evaluate the safety of cream [3,5, 6].

Evaluation of wound healing activity

Wistar albino rats of either sex, weighing between 200-250 gm, were kept under standard environmental conditions of temperature, humidity (25±0.5°C) and 12 hr light/dark cycle. Animals were fed with standard pellet diet and water *ad libitum*. The research was conducted in accordance to the protocol approved by Institute Animal Ethics Committee (IAEC), Krishna School of Pharmacy & Research, KPGU (Registration no-1372/c/10/CCSEA).

The rats were separated into three groups i.e. Group 1-Control (Base Cream treated /no extract), Group 2-Standard (Povidone-Iodine treated/Betadine) and Group 3- Test (Formulated Herbal Cream). On the day of experiments rats were anesthetized by administering Ketamine (50 mg/kg/ip). A full thickness of the excision wound with circular area of 176 mm² (width 1.5 cm and depth 0.2 cm) was made on the shaved back (dorsal thoracic region) of the rats. The wounding day was considered as day 0. The wounds were treated with topical application of the cream once daily till complete epithelisation. The wounds were monitored and the area of wound size was measured on 3, 6, 9, 12, 15 & 18th of post-wounding day. The wound size area and mean % wound closure/contraction were noted. The results were reported in Mean±SEM and the data was analyzed using ANOVA [2, 3, 26-29, 29a].

The Percentage (%) of Wound Closure was calculated using the following formula:

% of wound closure=

$$\frac{\text{Wound Area on Day '0' - Wound Area on Day 'n'}}{\text{Wound Area on Day '0'}} \times 100$$

Where n =number of days.

Histopathological studies of wounded skin

On day 18th the experiment was terminated and tissue of wound area was removed from the surviving animals for histopathological examination. Sample tissues were fixed in 10% formalin and were embedded in paraffin wax. Serial sections (5µm thickness) of paraffin embedded tissues were cut. The tissues stained by haematoxylin and eosin, (H & E) and after that they were examined by electronic microscope [29-31].

Results And Discussion

Pharmaceutical evaluation of herbal cream

F6 Herb-O-Seal Cream was found to be best and satisfactory compared to all other formulations. It had light Yellow appearance, gave a cool and smooth feel on application which was maintained after tested the stability study.

Stability was determined by exposing the formulation to various temperatures such as 4°C, 27°C & 37°C for specified period. The pH of the formulation was found to be 6.60 which is good for skin (pH=6.8). The creams also showed good Spreadability (16.17 g.cm/sec) when measured using slides. Lesser the time taken for separation of two slides resultant better Spreadability. Spreadability was calculated by using the formula. (S= M.L/T). Where S= Spreadability, M= Weight tied to upper slide, L= Length of glass slides and T= Time taken to separate the slides completely from each other. After application of the cream the type of smear formed on the skin was found to be non-greasy and easily removed on washing with tap water. The viscosity of the creams was found to be 17,650 cps, with 25 rpm, which indicates that the prepared cream was easily spreadable with small amount of shear [Table 1].

| Ingredients | Formulation (% w/w gm) |
|---|------------------------|
| Aq. Extract of <i>Panax ginseng</i> | 5.0 |
| Aq. Extract of <i>Calendula officinalis</i> | 5.0 |
| Aq. Extract of <i>Arnica Montana</i> | 2.0 |
| Aq. Extract of <i>Clerodendrum indicum</i> | 1.0 |
| <i>Aloe vera</i> | 3.0 |
| Rose hip oil | 4.0 |
| White petrolatum | 0.8 |
| Liquid paraffin | 8.3 |
| Lanolin | 0.8 |
| Stearic acid | 16.7 |
| Propylene glycol | 3.5 |
| Triethanolamine | 1.0 |
| Tween 60 | 5.0 |
| Methyl paraben | 0.1 |
| Water | q.s |

Table 1: Final composition of Herbal Cream (Formulation F6)

Skin irritation test

This test was conducted to evaluate the irritation caused by the prepared cream on the intact skin of animals. The results showed that the formulation (F6) was devoid of any primary skin irritation or sensation or erythema, or edema even after 48 hrs of application on the rat skin. None of the animal showed any skin reaction.

Wound healing activity

The results of wound healing activity by excision wound model are presented in **Table 2 and Figure 1A, 1B, 1C**. The values of wound area are presented in mm² at 0, 3, 6, 9, 12, 15 and 18th days. The results indicate that standard cream and test herbal cream both significantly ($P < 0.01$) reduces the wound area as compared to the control group.



Figure 1A: Wound at day 0:

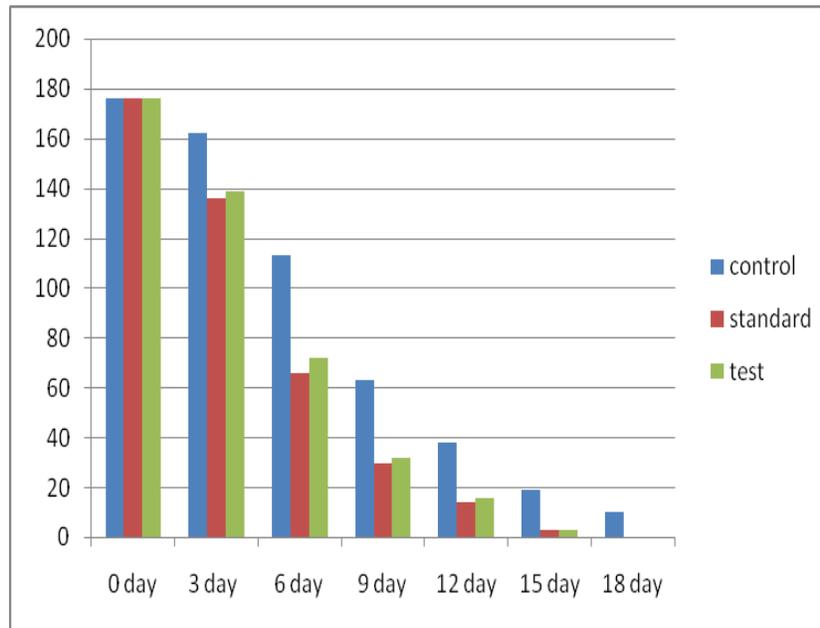


Figure 1B: Wound size area (mm²) at different days interval

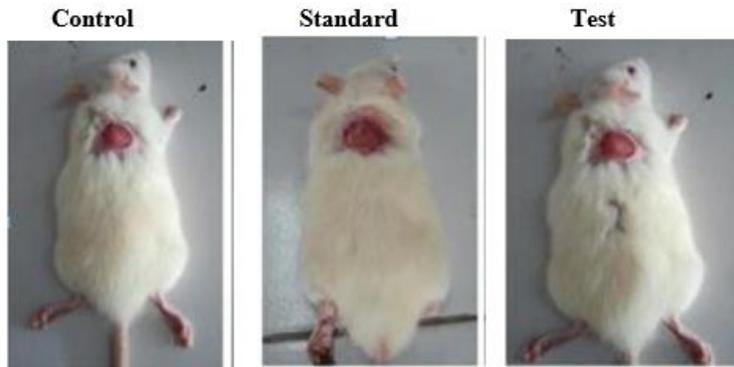


Figure 1C: Wound at day 0;

| Groups | Wound size area in mm ² (Mean ± SEM) | | | | | | |
|----------|---|---------------|---------------|--------------|--------------|--------------|---------------|
| | Day 0 | Day 3 | Day 6 | Day 9 | Day 12 | Day 15 | Day 18 |
| Control | 176.60 ± 0.93 | 162.45 ± 0.83 | 113.00 ± 0.85 | 63.58 ± 0.65 | 38.46 ± 0.93 | 19.62 ± 0.95 | 10.36 ± 0.33 |
| Standard | 172.44 ± 0.83 | 136.66 ± 0.85 | 66.44 ± 0.89 | 30.13 ± 0.63 | 13.83 ± 0.91 | 3.10 ± 0.33 | 0.26 ± 0.05** |
| Test | 174.24 ± 0.85 | 136.78 ± 1.21 | 72.34 ± 0.89 | 30.17 ± 0.63 | 16.62 ± 1.06 | 3.20 ± 0.35 | 0.28 ± 0.05** |

The treated, standard groups are compared with the control group. *** $P < 0.001$. ** $P < 0.01$. * $P < 0.05$.

Table 2: Effect of herbal cream on Wound size at different day's interval

Table 3 and Figure 2 represents percentage (%) wound healing (wound contraction) at 9 and 18th days for control, standard and the test groups. It is observed that wound contracting rate of animals treated with herbal cream

and standard cream significantly higher ($P < 0.01$) on days 9 and 18th as compared to the control group.

| Groups | % Wound contraction of excision wound (Mean ± SEM) | | |
|----------|--|--------------|-----------------|
| | Day 0 | Day 9 | Day 18 |
| Control | 0 | 39.96 ± 3.83 | 75.33 ± 3.85 |
| Standard | 0 | 58.33 ± 1.66 | 95.53 ± 3.85 ** |
| Test | 0 | 57.66 ± 2.33 | 95.53 ± 3.85 ** |

The treated, standard groups are compared with the control group. *** $P < 0.001$. ** $P < 0.01$. * $P < 0.05$.

Table 3: Effect of herbal cream on % wound contraction of excision wound at different days interval

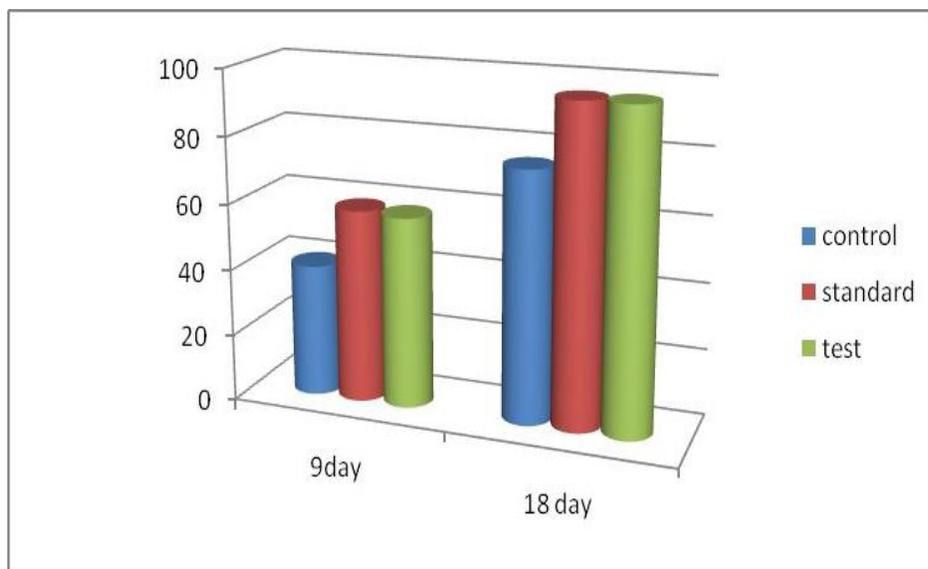


Figure 2: % Wound contraction of excision wound at different days interval

Histopathological study

The characteristics observed during histopathological examination were the proliferation of fibroblasts, granulation tissue, collagen fibre and tissue remodelling etc. Compared to control groups the above mentioned

parameters were more conclusive and decisive in case of both standard and test group. This histopathological observation also provided additional evidence for the experimental wound healing activity. The details of histopathological data are given in Figure 3, 4 & 5.

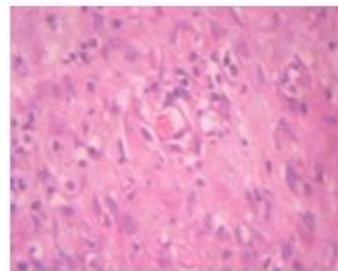
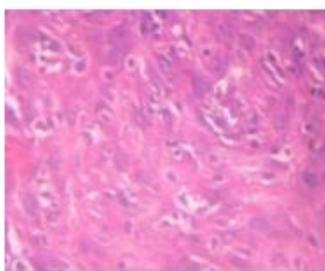
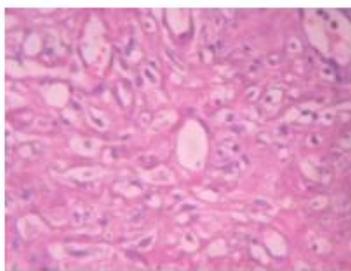


Figure. 3/4/5: Histological image of Control, Standard and Test at HEX20

Conclusion

The prepared cream was pleasant, coolant, easily spreadable and washable thereby there is a chance of increased the patient compliance. Formulated cream significantly promotes wound healing than control or non-medicated group. The activity may be mainly due to free radical scavenging activity, antioxidant activity and anti-inflammatory effect of the polyphenols and flavonoids present in the different extract. Some of herbs reported to act by supplying Vitamin C and by promoting tissue regeneration. However, further depth and structured study, would be beneficial to assess its usefulness and mechanisms more exactly. This study can be helpful for upcoming researchers to select these herbs for the formulation and evaluation of other cosmetic applications which can be claimed for their efficacy with scientific data.

Authors Contribution

MP*, KS, ZD, PL

Disclosures

No conflicts of interest, financial or otherwise, are declared by the authors.

Author Contributions

MP, PL had done review of literature and experimental work. MP drafted manuscript; MP, KS, ZD, evaluated and analyze the data. MP & PL edited and revised manuscript; MP approved final version of manuscript.

Acknowledgement

The authors are thankful to Management of Krishna School of Pharmacy & Research, Dr. Kiran and Pallavi Patel Global University (KPGU), Vadodara, Gujarat, India for providing all the supports to carry out the research work.

Reference

1. Kokane DD, More RY, Kale MB, Nehete MN, Mehendale PC, et. al., (2009), "Evaluation of wound healing activity of root of *Mimosa pudica*", *J Ethnopharmacol.*; 124: 311–315.
2. Reddy AKG, Saranya SC, Kumar ACK. (2012), "Wound healing potential of Indian medicinal plants". *Int J Pharm Rev Res.*; 2(2):75-87.
3. Kumar B, Kumar VM, Govindarajan R, Pushpangadan P. (2007), "Ethnopharmacological approaches to wound healing— Exploring medicinal plants of India". *J Ethnopharmacol.*; 114: 103–113.

- 3a. Mihir Y. Parmar, Mounika B. (2019), Nephroprotective and Antioxidant Potential of Ethanolic Extract of Flowers of Cassia Siamea against gentamicin induced nephrotoxicity. *JOJ Urology and Nephrology.*, 1-6.
4. Rajsekhar S. (2011), "Unseen aspects of wound healing: an overview". *Int J Pharm Biol. Sci.* 2011; 2(4):275-287.
5. Singh M, Sharma S, Khokra LS, Kumar SR. "Preparation and evaluation of herbal cosmetic cream", *Pharmacologyonline.*; 5(2):1258-1264.
6. Das K, Dang R, Machale MU, Ugandar R, Lalitha B. (2012), "Evaluation for safety assessment of formulated vanishing cream containing aqueous Stevia extract for topical application. *Ind J Novel Drug Deliver.* 4(1):43-51.
7. Khalid AS, Saringat HJ, Khan GM. et. al., (2005), "Haruan (*Channa striatus*) incorporated palm-oil creams: Formulation and stability studies". *Pak J of Pharm Sci.*; 18(1):1-5.
8. Mahalingam RC, Xiaoling L, Bhaskara RJ. (2006), "Semisolid Dosages: Ointments, Creams and Gels", *Pharmaceutical Manufacturing Handbook.*; 2(3): 267-274.
9. Upadhyay NK, Kumar R, Mandotra SK, Meena RN, Siddiqui MS, et. al., (2009), "Safety and healing efficacy of Sea buckthorn (*Hippophae rhamnoides* L.) Seed oil on burn wounds in rats". *Food Chem Toxicol.* 3(47):1146-1153.
- 9a. Mihir Y, Parmar, Shravya B. Anti-atherosclerotic potential of Echinops Echinatus against high fat diet induced atherosclerosis in wistar rats. *Bio Engg Bio Med Sci Int J.* 1(1) 2019: 1-9
10. Attele AS, Wu JS, Chun SY. (1999), "Ginseng pharmacology multiple constituents and multiple actions", *Biochemical Pharmacology.*; 58(3):1685-1693.
11. Saeed MA, Ahmad I, Yaqub U, Shazia A, Waheed A, et. al. (2004), "Aloe Vera: A Plant of Vital Significance". *Science Vision.* 9(2): 1-13.
12. Nandal U, Bhardwaj RL. (2012), "Aloe vera for human nutrition, health and cosmetic use -A review". *Int Res J Plant Sci.* 3(3): 38-46.
13. Akhtar N, Zaman S, Khan BA, Haji M, Khan M, et al. (2011), "Evaluation of various functional skin parameters using a topical cream of *Calendula officinalis* extract". *Afr J Pharm Pharmacol.* 5(2): 199-206.
14. Raiman SZ, Biswas P, Monir MM, Biswas SK, Chowdhury A, (2012), "Phytochemical investigation and *in-vitro* antinociceptive activity of *Clerodendrum indicum* leaves". *Pak J Biol Sci.*; 15(3): 152-155.
15. Chhetri HP, Yogol NS, Sherchan J, Anupa KC, Mansoor S, et. al., (2010), "Formulation and evaluation of antimicrobial herbal ointment". *J Sci. Eng Technol.*; 6(1):102-107.
16. Macedo SB, Carvalho JCT, Ferreira LR, Santos-Pinto R. (2005), Effect of *Arnica montana* 6 cH on edema, mouth opening and pain in patients submitted to extraction of impacted third molars. *Ärztzeitschrift für Naturheilverfahren.*; 46(60): 381-387.
17. Ozcan M. (2002), Nutrient composition of rose (*Rosa canina* L.) seed and oils. *J Med Food.* 5 (3):137 –140.
18. Chrubasik C, Duke RK, Chrubasik S. (2006), The evidence for clinical efficacy of rose Hip and seed: a systematic review. *Phytother Res.*; 20:1-3.
19. Handa SS, Khanuja SPS, Longo G, Rakesh DD. (2008), Extraction Technologies for Medicinal and Aromatic Plants. *International centre for science and high technology, Trieste.*; 21-25
20. Tiwari P, Kumar B, Kaur M, Kaur G, Kaur H. (2011), Phytochemical screening and Extraction: A Review. *Int Pharm Scientia.* 1(1): 98-106.
21. Mahapatra AP, Kumar MD, Panda P. (2012), Formulation and evaluation of cream from *Croton sparsiflorus* Morong and their wound healing activity. *Int J res Ayurveda Pharm.*; 3(6): 803-807.
22. Kohli DPS, Shah DS. (1998), Drug formulation Manual. 2nd ed. New Delhi. *Eastern Publication.* 611-650.
23. Lachmann L, Libermann HA, Kanic JL. (1987), The theory and practice of industrial Pharmacy. 18th ed. Bombay: Lea and Febroger. Philadelphia. Varghese Publishing House. 534-563.
24. Kulkarni GT, Gowthamarajan B, Suresh B. (2004), Stability testing of Pharmaceutical products-An overview. *Ind J Pharm Edu.* 38(4): 194-198.
25. Sabale V, Kunjwani H, Sabale. (2011), Formulation and *in vitro* evaluation of the topical antiaging preparation of the fruit of *Benincasa hispida*. *J Ayurveda Integr med.* 2(3): 124-128.
26. Akkol K. E, Koca U, Pesin I, Yilmazer D. (2009), "Exploring the wound healing activity of *Arnebia densiflora* (Nordm.) Ledeb by *in vivo* models". *J Ethnophacol.* 124: 137-141.
27. Ilango K, Chitra V. (2010), "Wound healing and anti-oxident activities of the fruit pulp of *Limonia acidissima* Linn (Rutaceae) in rats". *Trop J Pharm Res.*; 9(3): 223-230.
28. Ramane SB, Syed VB, Biyani KR. (2013), Evaluation of Wound Healing Activity of Polyherbal Gel – A Novel Herbal Formulation. *Int J res Pharm Biomed Sci.*; 4 (3): 788-794.
29. Kiran K, Asad M. (2008), Wound healing activity of *Sesamum indicum* L seed and oil in rats. *Ind J Exp Biol.*; 46: 777-82.
- 29a. Mihir Y. Parmar, Sindhuja S. Nephroprotective Potential of Ethanolic Extract of Barks of *Tricholepis Glaberrima* against gentamicin induced nephrotoxicity. *J Pharmacology & Clinical Research.*
30. Murthy S, Gautam K, Goel S, Purohit V, Sharma H, (2013), Evaluation of In Vivo Wound Healing Activity of *Bacopa monniera* on Different Wound Model in Rats. *BioMed Res Int.*: 1.
31. Singh SDJ, Krishna V, Mankani KL, Manjunatha BK, Vidya SM, (2005), Wound healing activity of the leaf extracts and deoxyelephantopin isolated from *Elephantopus scaber* Linn. *Ind J pharmacol.*; 37(4): 238-242.



This work is licensed under Creative Commons Attribution 4.0 License

To Submit Your Article Click Here: [Submit Manuscript](#)

DOI:10.31579/2690-8808/219

Ready to submit your research? Choose Auctores and benefit from:

- fast, convenient online submission
- rigorous peer review by experienced research in your field
- rapid publication on acceptance
- authors retain copyrights
- unique DOI for all articles
- immediate, unrestricted online access

At Auctores, research is always in progress.

Learn more <https://auctoresonline.org/journals/journal-of-clinical-case-reports-and-studies>