IJARSCT
ISSN (Online) 2581-9429
International Journal of Advanced Research in Science, Communication and Technology (IJARSCT)
Volume 2, Issue 2, June 2022
Impact Factor: 6.252

Formulation and Evaluation of Ocimum Sanctum based Emulgel for Wound Healing Potential

Tikone Dattatray B[1] and Prof. Gaikwad Shital D[2]
Samarth Institute of Pharmacy Belhe Pune, Maharashtra, India[1]
Department of Pharmaceutical Chemistry, Samarth Institute of Pharmacy Belhe, Pune, Maharashtra, India[2]

Abstract: *The purpose of this study was to evaluate the anti-inflammatory effect of leaves of Occimum sanctumin the formulation of hydrogels using different types of animal models. Materials and Methods: O. sanctum leaf ethanol extract was fractionated with methanol to give a methanol fraction. The methanol fraction (1% vw / w) was used in hydrogel formulations in various combinations of Carbopol 940 and sodium CMC. The hydrogels produced were characterized for optimal physical properties, pH, ductility, uniformity, viscosity, release profile, and irritation. Optimized hydrogels were evaluated for anti-inflammatory activity in animals using xylene-induced, croton oil-induced ear edema, and cotton pellet-induced granuloma models. The effect was monitored by , which measures the suppression rate of ears , nitric oxide (NO) and myeloperoxidase (MPO) levels in mice after acute inflammation induced by croton oil. Results and discussion: Physical evaluation confirmed that the resulting hydrogel was brownish in color and had a uniform and smooth appearance when applied. The combination of F3-hydrogel formulations was found to be appropriate for all other evaluation parameters such as pH, viscosity, spreadability and consistency. Therefore, it was observed that the optimal composition of F3 preparation is 1.5 g of Carbopol 940 and 1% sodium CMC. The suppression rate of edema in xylene-induced ear edema in mice was found to be comparable to that of the standard treatment group (65.59%). In this study, the results show that the prepared hydrogel-OSMFH has an inhibitory effect on acute inflammation. The results show the ability of croton oil to induce the influx of neutrophils into the ear tissue of mice.*

Keywords: Ocimum sanctum, Anti-inflammatory, Hydrogel, Voltaren Emulgel, Carbopol.

I. INTRODUCTION

Inflammation is one of the diseases affecting 25% of the world's population. inflammatory diseases that cause the immune system to attack cells and tissues in the body can cause abnormal inflammation, which can lead to chronic pain, redness, swelling, stiffness, and damage to normal tissues. The inflammatory process acts rapidly to destroy and eliminate foreign and damaged cells, isolating infected or damaged tissue from the rest of the body. [1] Ocimum sanctum is an aromatic perennial plant of the mint family, commonly known as the sacred basil, turkey, or tulsi. Various parts of the Tulsi plant, such as leaves, flowers, stems, roots, seeds, traditionally have sputum, painkillers, anticancer agents, antiasthma agents, antiemetics, sweating agents, antidiabetic agents, antifertilizers, It is used as a hepatoprotective agent, antihypertensive agent, hypolipidemia agent, and especially as an antistress agent. Tulsi is also used to treat fever, bronchitis, arthritis, convulsions and more. Tulsi leaf decoction is given to patients suffering from stomach and liver disease. Volatile leaf oil [2] includes eugenol (1-hydroxy-2-methoxy-4-allylbenzene), eugenol (also called eugenol acid), ursolic acid, and carbachlor (5-isopropyl-2-methylphenol). , Linalol (3, 7-dimethylocta-1,6-diene-3-ol), lymatrol, cariophyllene (4,11,11-trimethyl-8-methylene-bicyclo [7.2.0] undeca-4-ene) and Methylcarbicol (also known as estragor: 1-allyl-4-methoxybenzene), volatile seed oil contains fatty acids and citosterol. In addition, the mucus contains some sugar, and the green leaves contain anthocyanins. Sugar is composed of xylose and polysaccharides. [3] Based on previous studies and literature, this study aims to evaluate the anti-inflammatory effect of O. sanctum leaf on hydrogel formulations using various animal models.

II. PLANT PROFILE

Tulsi is an important symbol of the Hindu religious tradition. Although the word Tulsi" gives the implication of the incomparable one, its other name, Vishnupriya means the one that pleases Lord Vishnu. Found in most of the Indian

IJARSCT
ISSN (Online) 2581-9429
International Journal of Advanced Research in Science, Communication and Technology (IJARSCT)
Volume 2, Issue 2, June 2022
Impact Factor: 6.252

homes and worshipped, its legend has permeated Indian ethos down the ages. Known in English as Holy Basil and botanically called Ocimum sanctum, Tulsi belongs to plant family Lamiaceae

2.1 Morphology

Fig 1: Ocimum Sanctum Plant

It is an erect, herbaceous, more-branched, softy hairy, biennial or triennial plant, 30-75 cm high. The leaves are alliptic-oblong, acute or obtuse, entire or serrate, pubescent on both sides, minutely gland dotted; the flowers are puplish or crimson, in racemes, close whorled; the nut-lets are sub- globose or broadly ellipsoid, slightly compressed, nearly smooth, pale-brown or reddish with small, black markings.

- **Class:** Magnoliopsida
- **Order:** Lamiales
- **Family:** Lamiaceae
- **Genus:** Ocimum
- **Species:** O.tenuiflorum
- **Botanical Name:** Ocimum Tenuiflorum.

Three varieties of Tulsi are:

- Rama or Light Tulsi (Ocimum Sanctum)
- Shyama or Dark Tulsi (Ocimum Sanctum)
- Vana Tulsi (Ocimum Gratissimum)

Uses:

- **Heart Disease:** Basil has a beneficial effect on heart disease and the resulting weakness. Lowers blood cholesterol levels (Jyoti et al., 2004). Pediatric Diseases Common pediatric problems such as cough, runny nose, fever, diarrhea and vomiting respond favorably to basil leaf juice. When chickenpox pustules delay the onset, basil leaves with saffron accelerate them (Devi et al, 1999).
- **Stress and Headache:** Basil leaves are considered "adaptogens" or anti-stress agents. Recent studies have shown tha leaves provide important protection against stress. Even healthy people can chew 12 basil leaves

IJARSCT
ISSN (Online) 2581-9429
International Journal of Advanced Research in Science, Communication and Technology (IJARSCT)
Volume 2, Issue 2, June 2022
Impact Factor: 6.252

twice daily to prevent stress. It helps cleanse the blood and prevent some common factors. Basil is a good medicine for headaches. In this disease, you can administer a decoction of leaves. Pound leaves mixed with sandalwood paste can also be applied to the forehead to relieve heat and headaches and provide overall cooling.

- **Eye Disorders:** Basil juice is an effective remedy for sore eyes and night-blindness, which is generally caused by deficiency of vitamin A. Two drops of black basil juice are put into the eyes daily at bedtime.
- **Mouth Infections:** The leaves are quit effective for the ulcer and infections in the mouth. A few leaves chewed will cure these conditions.
- **Insect Bites:** The herb is a prophylactic or preventive and curative for insect stings or bites. A teaspoonful of the juice of the leaves is taken and is repeated after a few hours. Fresh juice must also be applied to the affected parts. A paste of fresh roots is also effective in case of bites of insects and leeches (Sharma et.al. 1998).

III. MATERIALS & METHOD

3.1 Plant Materials

Fresh leaves of Ocimum basilicum were collected from differentlocation of Dera Ismail Khan, Pakistan and were authenticated byProfessor Mushtaq, Department of Botany, QAU Islamabad. A voucher specimen (No. pp1281) has been deposited. The leaves wereshade dried for five days and the dried leaves were then grindedin a grinder and stored in an air resistant container.

3.2 Extraction Method

Grinded OB leaf powder (850 g) was soaked in 6 L of 70% methanol. The mixture was stirred at for 24 hours at intervals of 6-8 hours. The mixture was then held at room temperature of ° C. for 6 days. The mixture was then coarsely filtered using a muslin cloth followed by filter paper. The solvent was evaporated on a rotary evaporator at 45 ° C. The extract was then stored in an airtight bottle. The yield was about 17-19% (Sekar et al., 2009; Amzad et al., 2010).

Fig 2 : Extraction of Ocimum Sanctum.

IJARSCT
ISSN (Online) 2581-9429
International Journal of Advanced Research in Science, Communication and Technology (IJARSCT)
Volume 2, Issue 2, June 2022
Impact Factor: 6.252

3.3 Formulation of Emulgel

As shown in Table 1, we prepared a formulation containing various amounts of ingredients. Emulgel, the gel portion of , was prepared by dissolving Carbopol-934 in cold water and constantly stirring at medium speed of until a uniform mixture was formed. The pH was then adjusted to 6-6.5 using triethanolamine (TEA). Tween 80 was dissolved in distilled water to create the aqueous phase of the emulsion, and was dissolved to create the oil phase of the emulsion. Span 80 was dissolved in liquid paraffin. To preserve the emulsion,methyl parabene was dissolved in propylene glycol and the extractwas dissolved in ethanol then both solutions were mixed with theaqueous phase. Both the aqueous and the oil phase were heated ina water bath at 70 C separately. Then the oil phase was addeddrop wise to the aqueous phase with continuous stirring usinghomogenizer (WiseStir HS-120A, Daihan Scientific,Korea) atspeed of 3000 rpm for 10 min then cold to room temperature. Atthe end the gel and emulsion portions were mixed in 1:1 ratio withmoderately stirring to prepare emulgel.

3.4 Evaluation of Ocium Santum Emulgel

A. Characterization of Emulgel

Determination of Lambdamax (kmax) by UV / Vis spectrophotometer: Lambdamax of plant extract was determined according to a previously reported study using a UV / Vis spectrophotometer (CECIL, CE 2021 Germany). (Bueno et al., 2012; Joseph et al., 2018). For this purpose, 20 mg of OB extract was removed and dissolved in 20 ml of methanol (stock solution). Next, 1 ml of the prepared stock solution was taken and dissolved in a measuring flask. Volume was made up to the mark with methanol. Similarly, the stock solution was serially diluted to three concentrations. Aliquots of various preparation solutions were taken and scanned in the UV range 190- 400 nm to determine the peak absorbance wavelength of the main component of the OB extract (linalool). I used methanol as a blank. 6.2. Fourier Transform Infrared Spectrophotometer Analysis (FTIR) FTIR analysis is Burkietal. Performed according to previous studies published by. It was reported to check thecompatibility of the drug / extract of Osimum basilicum with that of other excipients of the pharmaceutical product(Burki et al., 2020).First, an infrared spectral analysis of the Osimum basilicum extract was performed using a Fourier transform infrared spectrophotometer (Tencor Series 7, Shimadzu, Germany). Next, the spectral analysis of the pharmaceutical product was performed. The spectrum was recorded in the range offrom 4000 cm1 to 400 cm1. 6.3. Stability test First, eight different formulations (FB1-FB8) were prepared and thewas placed in an incubator at 25 ° C for 3 days. Of the eight formulations, the formulation FB8 was relatively stable. The product FB8 was divided into 4 samples and stored in 4different incubators at 8, 25, 40 and 40 ° C + 75% RH (relative humidity), respectively. Theses were sensually observed for color, uniformity, phase separation and liquefaction for one month with various Emulgel formulations.

Table 1: Composition of various emulgel formulations.

Sr. No	Ingredient (%w/w)	FB1
1)	OB extract	5
2)	Carbopol 934	0.50
3)	Liquid paraffifin	2.50
4)	Tween 80	0.50
5)	Span 80	0.75
6)	Propylene glycol	3.50
7)	Methyl parabene	0.01
8)	Distilled water QS	50.0
9)	Triethanolamine few drops	pH=Adjusted to 6-6.5

B. Spreadability Study

Spreadability was determined by apparatus suggested by Mutimer et al. It consists of a block of wood at one end of which, a pulley was connected. On the basis of ‘drag’ and ‘sleep’ method, spreadability was determined. A ground glass

IJARSCT
ISSN (Online) 2581-9429
International Journal of Advanced Research in Science, Communication and Technology (IJARSCT)
Volume 2, Issue 2, June 2022
Impact Factor: 6.252

slide was fifixed on this block. Test emulgel (2 g) was placed on this slide. The emulgel was then sandwiched between these slides and another glass slide having the same dimension of fixed ground side and provided with hook. Weight (40 g) was then placed on the top of this slide. The time required (in seconds) by the top slide to cover a distance of 6 cm was noted. Then spreadability was calculated using the following formula

$$S = M.L/T$$

where S = spreadability, M = Weight tied to upper slide, L = Length of glass slides T = Time taken to separate the slides completely from

Fig 3 : Spredability Study

C. Viscosity / Rheology

The initial viscosities of the eight newly prepared formulations (FB1-FB8) were measured using a Brook-Fifield viscometer with a # 04 spindle. The spindle was lowered vertically to the center of the emulsified product placed in the beaker, and the spindle was rotated at a speed of 2.5 rpm for 5 minutes / minute, being careful not to touch the bottom of the beaker. Viscosity values were recorded. (Basha et al., 2011). Next, the stable formulation FB8 was divided into four samples (ie, FB8A, FB8B, FB8C, and FB8D). The four samples also underwent a viscosity test and were checked regularly for a month.

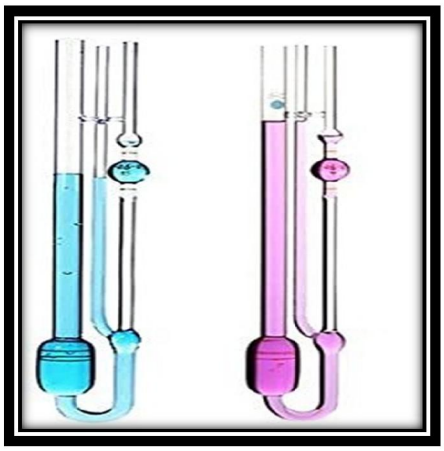

Fig 4: Ostwald viscometer

IJARSCT
ISSN (Online) 2581-9429
International Journal of Advanced Research in Science, Communication and Technology (IJARSCT)
Volume 2, Issue 2, June 2022
Impact Factor: 6.252

D. Patch / Sensitivity Test

volunteers (n = 3) were selected for the patch test. The formulation (1 g) was applied to the volunteer's forearm in the form of a bandage disc and then covered with a surgical bandage. After 24 hours, the patch was removed and the area was washed with saline. Volunteers were asked about irritation and the area of use was examined for edema and erythema (redness of the skin) (Rasul and Akhtar, 2011).

3.5 Aplication of Ocimum sanctum emulgel

Ocimum sanctum (family Lamiaceae), commonly known as Holy Basel or Tulsi, is an annual herb that has been used in the Indian traditional system of medicine for hundreds of years. The leaves of this plant have been traditionally used in coughs, colds, asthma, and bronchitis (Ghosh, 1995)

Fig 5 : Ocimum sanctum

3.6 Result

- **Stability Test:** All four samples of the stable formulation FB8 (ie, FB8A, FB8B, FB8C, and FB8D) were stored for 1 month under different storage conditions for color, phase separation, uniformity, consistency. , And liquefaction. According to cosmetic formulation requirements.
- **Spreadability Study:** Spreadability indicates that the emulgel is easily spreadable and comes out of containers by small amount of shear (Helal et al., 2012). Average spreadability values of different formulations have been given in Table 3. Larger the value of spreadability coeffificient better is its spreadability on the skin.

Sr.no	Evalution Test	Result
1	Spredability study	Do not spread

Table 2: Spreadability study

- **Patch Test:** For confifirming the safety of topical preparations, the important point is that they must not cause any contact dermatitis when

3.7 Viscosity Study

Viscosity is an important parameter to be evaluated because consistency of dosage form and drug content release mainly depend upon viscosity (Ghada et al., 2014). Viscosities of all formu lations were performed by using brook fifield viscometer. Values have been given in Table 4. The most viscous formulation was FB8 (12500 cp). This is due to high level of gelling agent, low level of emulsifying agent and low level of liquid paraffifin.

Sr. No	Evaluation Test	Result
1	Viscosity study	The average of water is :47.66sec The average of ocium santum emulgel:12 min

Table 3: Viscosity study

IJARSCT
ISSN (Online) 2581-9429
International Journal of Advanced Research in Science, Communication and Technology (IJARSCT)
Volume 2, Issue 2, June 2022
Impact Factor: 6.252

IV. CONCLUSION

The findings of this study reveal that OS emulgel contains a variety of phytochemicals such as tannins, sesquiterpenes that are responsible for its in vivo activities including wound healing. It can also be concluded that this study presents a better strategy for the formulation of emulgel with signifificant wound healing potential. Further studies are required to determine formulation of emulgel containing isolated constituents that are responsible for wound healing activity and to make standardized the extract, using standard and sophisticated phytochemical analysis techniques that would enable us to reach a conclusive stage

REFERENCES

[1]. Chen L, Deng H, Cui H, Fang J, Zuo Z, Deng J, et al. Inflammatory responses and inflammation- associated diseases in organs. Oncotarget 2017;9:7204-18.
[2]. Kelm MA, Nair MG, Strasburg GM, DeWitt DL. Antioxidant and cyclooxygenase inhibitory phenolic compounds from Ocimum sanctum Linn. Phytomedicine 2000;7:7-13.
[3]. Shishodia S, Majumdar S, Banerjee S, Aggarwal BB. Ursolic acid inhibits nuclear factor-kappaB activation of IkappaBalpha kinase and p65 phosphorylation: Correlation with down-regulation of cyclooxygenase 2, matrix metalloproteinase 9, and cyclin D1. Cancer Res 2003;63:4375-83.
[4]. Paech K, Tracey MV. Modern Methods of plant Analysis. Vol. 4. Berlin: Springer; 1955. p.367- 74.
[5]. Sim SK. Medicinal Plant Glycosides. 2nd ed. Toronto, Canada: University of Toronto Press; p. 25-7.
[6]. Kokate CK, Purohit AP, Gohkale SB. Pharmacognosy. In: Terpenoids. 21st ed. Pune: Nirali Prakashan; 2002.
[7]. Chirayath RB, Jayakumar R, Biswas R, Vijayachandran LS. Development of Mangifera indica leaf extract incorporated carbopol hydrogel and its antibacterial efficacy against Staphylococcus aureus. Colloids Surf B Biointerfaces 2019;178:377-84.
[8]. Phad AR, Dilip TN, Ganapathy RR. Emulgel: A comprehensive review for topical delivery of hydrophobic drugs. Asian J Pharm 2018;12:S382.
[9]. Gupta P, Yadav DK, Siripurapu KB, Palit G, Maurya R. Constituents of Ocimum sanctum with antistress activity. J Nat Prod 207;70:1410-6.
[10]. Dutta D, Devi SS, Krishnamurthi K, Kumar K, Vyas P, Muthal PL, et al. Modulatory effect of distillate Of Ocimum sanctum leaf extract (Tulsi) on human lymphocytes against genotoxicants. Biomed Environ Sci 2007;20:226-34.
[11]. Draize JH, Woodard G, Calvery HO. Methods for the study of irritation and toxicity of substances applied topically to the skin and mucous membranes. J Pharmacol Exp Ther 1944;82:377-90.
[12]. Yousif MF, Haider M, Sleem AA. Formulation and evaluation of two anti-inflammatory herbal gels. J Biol Act Prod Nat 2011;1:200-9.
[13]. Maxia A, Frau MA, Falconieri D, Karchuli MS, Kasture S. Essential oil of myrtuscommunis inhibits inflammation in rats by reducing serum IL-6 and TNF- α. Nat Prod Commun 2011;6:1545-8
[14]. Redza-Dutordoir M, Averill-Bates DA. Activation of apoptosis signalling pathways by reactive oxygen species. Biochim Biophys Acta 2016;1863:2977-92.
[15]. Bryan NS, Grisham MB. Methods to detect nitric oxide and its metabolites in biological samples. Free Radic Biol Med 2007;43:645-57.
[16]. Krawisz JE, Sharon P, Stenson WF. Qualitative assay for acute intestinal inflammation based on myeloperoxidase activity. Gastroenterology 1984;87:1344-50.
[17]. Bancroft JD, Gamble M. Theory and Practice of Histological Techniques. 6th ed. London, United Kingdom: Churchill Livingstone, Elsevier; 2008. p. 93-133.
[18]. Winter CA, Risley E, Nuss G. Carrageenan-induced edema in hind aw of the rat as an assay for antiinflammatory drugs. Proc Soc Exp Biol Med 1962;111:544.
[19]. Crunkhon P, Meacock S, Mediators of the inflammation induced in the rat paw by carrageenan. Br J Pharmacol 1971;42:392-402.

IJARSCT
ISSN (Online) 2581-9429
International Journal of Advanced Research in Science, Communication and Technology (IJARSCT)
Volume 2, Issue 2, June 2022
Impact Factor: 6.252

[20]. Deuschle VC, Deuschle RA, Bortoluzzi MR, Athayde ML. Physical chemistry evaluation of stability, spreadability, in vitro antioxidant, and photo-protective capacities of topical formulations containing Calendula officinalis L. leaf extract. Braz J Pharm Sci 2015;51:63-75.

[21]. Atta AH, Alkofahi A. Anti-nociceptive and antiinflammatory effects of some Jordanian medicinal plant extracts. J Ethnopharmacol 1998;60:117-24.

[22]. Vogel HG, Vogel WH. Drug Discovery and Evaluation, Pharmacological Assay. Berlin: Springer; 1997. p. 370, 382, 402-3.

[23]. Medeiros R, Figueiredo CP, Passos GF, Calixto JB. Reduced skin inflammatory response in mice lacking inducible nitric oxide synthase. Biochem Pharmacol 2009;78:390-5.

[24]. Xavier-Santos JB, Félix-Silva J, Passos JG, Gomes A, Fernandes JM, Garcia VB. Development

[25]. of an effective and safe topical anti-inflammatory gel containing Jatropha gossypiifolia leaf extract: Results from a pre-clinical trial in mice. J Ethnopharmacol 2018;227:268-78.

[26]. Badgujar VB, Jain PS, Patil RR, Haswani NG, Chaudhari SG. Anti-inflammatory activity of Helicteres isora L. stem bark extracts in rats. Asian J Pharm Clin Res 2009;2:63