

FORMULATION AND EVALUATION OF HERBAL NANOGEL FOR THE TREATMENT OF MOUTH ULCER

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Abstract

In the worlds of fashion and beauty, herbal cosmetics are currently popular. These products are becoming more popular because most women choose natural cosmetics over synthetic ones because they give their bodies the nutrients they need, improve their health, and make them feel good because they don't contain synthetic chemicals and have fewer side effects than synthetic cosmetics. As dispersions of hydrogel nanoparticles based on crosslinked polymeric networks, nanogels in the biomedical field are innovative and promising materials that have been dubbed next-generation drug delivery systems because of their uniformity, tunable size, simplicity of preparation, low toxicity, stability in the presence of serum, and stimuli responsiveness. The use of nanogels in chemotherapy, diagnostics, organ targeting, and the transport of bioactive chemicals all has enormous potential.

Introduction

Ulcer

Mouth ulcers are quite prevalent and typically result from trauma, such as from ill-fitting dentures, broken teeth, or fillings. However, in order to rule out cancer or other dangerous disorders such persistent infections, patients with ulcers that have been present for longer than 3 weeks should be referred for a biopsy or further investigations. After the origin of the ulcer has been removed to maintain excellent oral hygiene, trauma-related ulcers typically heal within a week[1].

Based on their clinical condition, mouth ulcers are categorised into acute and chronic ulcers:

1)Acute ulcer:

Traumatic ulcers:

Acute ulcers include traumatic ulcers. Usually, physical, thermal, or chemical trauma to the oral mucosa results in tissue destruction and subsequent ulceration in this form of ulcer. Physical harm brought on by routine tasks like tooth brushing or flossing, denture or tooth sharp edges, oral piercings, or even self-infection by the patient while under local anaesthesia during a dental operation. Thermal burns frequently result from hot foods or drinks like pizza, coffee, or tea as well as from heated dental instruments used during dental procedures. Patients who use aspirin to treat pain frequently report developing ulcers that are brought on by chemical damage. Traumatic ulcers often disappear within 7 to 10 days.

Primary Herpetic Gingivitis

The most prevalent infection caused by the herpes simplex virus (HSV) in the mouth is primary herpetic gingivostomatitis. HSV-1, which arises above the waist but below the waist, is the infection that causes more than 90% of cases. Anorexia, anaemia, and irritability are among the symptoms. HSV ulcers that come back frequently resemble traumatic ulcers that appear on the palate.

Primary varicellazoster virus infection

Primary varicella zoster virus infection also known as chicken pox, typically affects children between the ages of one and twenty. These ulcers naturally disappear between 10 to 14 days. Skin and mouth

ulcerations can also be a symptom of autoimmune diseases like pemphigus and pemphigoid, however these lesions are persistent.

2) Chronic Ulcer

Decubitus ulcers

Chronic oral mucosal injury can result in traumatic ulcers that last for a long time and are characterised by fibrosis around the ulcerations. The floor of the lingual sulcus, lips, tongue, and buccal mucosa are where they are most frequently found. Traumatic ulcers usually disappear within 7 to 10 days, but some don't, lingering for weeks or months as a result of repeated trauma, irritation, or secondary infection.

Cancer of the squamous cell

More than 90% of mouth malignancies are caused by squamous cell carcinoma, the most prevalent cancer. It might appear as a mixed, exophytic, red and white, ulcerative, or red and white lesion. It tends to affect men over 40 and is most frequently associated with a history of cigarette or alcohol use. The initial symptom is a non-healing ulcer that can last for days or weeks. Typically, the lesion develops asymptotically without the patient being aware of it.

Causes of Mouth ulcer

A mouth ulcer's aetiology and pathology are unknown, but some factors, such as a lack of iron and certain vitamins, particularly B12 and C, poor dental hygiene, infections, stress, indigestion, mechanical injury, skin disease, etc., are thought to be significant.[2]

1)Genetic factors

About 30%–40% of patients with aphthous ulcers have a family history [3], suggesting that a genetic component may play a role in the condition. In some cases, it's clear that the patient has a family history of recurring aphthous ulcers. Young age of onset and symptoms that are more severe than usual are common connections.

2)Physical or psychological stress

Aphthous ulcer incidences have a close correlation to difficult living circumstances [4]. In the development of recurrent aphthous stomatitis, psychological stress may act as a trigger or a moderator. The link between stress and recurrent aphthous stomatitis has not been conclusively established by research.

3)Nutritional deficiencies

Nutritional deficiencies including those affecting iron, folic acid, vitamin B12, B1, and B2 and B6, have been linked to a subset of aphthous ulcer patients. Based on diet and dietary supplementation, different regions' contributions of nutritional deficiencies to aphthous ulcers are anticipated to differ [5].

4) Trauma

Stress and localised trauma are the most common causes of aphthous ulcers. Accidental self-biting, dental work, sharp-edged foods (like potato chips), anaesthetic injections, and tooth brush bristles can all cause damage to the oral mucosa. In addition to this, emotional and environmental stress can also cause aphthous ulcers [6].

5) Food allergies

A variety of foods might result in allergies. Patients with recurrent aphthous stomatitis exhibit anti-cow milk and anti-wheat protein antibodies (celiac illness). Because of this, a lot of typically allergenic foods (including strawberries, tomatoes, and nuts) haven't been directly linked to recurrent aphthous stomatitis [7].

6) Immune disorders

Patients with immune conditions such as cyclic neutropenia, inflammatory bowel disease, Behçet's illness, and HIV disease are more likely to develop aphthous ulcers and to have more severe cases [8].

Jasminum officinale

Herbal medicine has been used to treat sickness pharmacologically for a very long time. Because herbs have such a diverse spectrum of pharmacological and therapeutic properties, they eventually became the source of several significant drugs[9–19]. Alkaloids, coumarins, flavonoids, tannins, terpenoids, glycosides, emodine, leucoanthocyanins, steroids, anthocyanins, phlobatinins, essential oil, and saponins were all found in *Jasminum officinale* according to the results of the phytochemical examination. The herb had antiulcer, antimicrobial, insecticidal, antioxidant, antifertility, and dermatological benefits, according to pharmacological research.



Figure 1: *Jasminum officinale*

Cynodondactylon

Bermuda grass, also known as *Cynodondactylon*, is a perennial grass that may be found all over the globe but is native to mild temperate and tropical climates. In particular, proteins, carbohydrates, minerals, flavonoids, carotenoids, alkaloids, glycosides, and triterpenoids were abundant in the plant. The entire plant of *C. dactylon* maintains a number of biological functions, including wound healing, antiviral, antibacterial, antimicrobial, and antiulcer characteristics. Additionally, it has a long history of usage in traditional remedies to treat a wide range of conditions, including tumours, warts, dropsy, dysentery, haemorrhage, hypertension, hysteria, measles, and snakebite. Other conditions it has been used to cure include cough, headache, diarrhoea, cramps, epilepsy, dropsy, and dysentery[20].



Figure 2: *Cynodondactylon*

Nanogel

Incorporating either copolymerized or non-ionic monomers, nanogels are strongly cross-coupled nano-sized gel formulations [21,22]. Between 20 and 200 nanometers are the dimensions of nanogel[23]. They

are highly soluble, exhibit low viscosity, exhibit good thermodynamic stability, and can withstand sterilisation[24]. In terms of drug loading capacity, stability, and duration spent in contact with the skin surface, nanogels have surpassed traditional and macro-sized delivery systems, making them better suited for transdermal drug administration[25].

Advantages of nanogels:-

1. Nanogels are advantageous as a medication delivery technology due to their great biocompatibility [26].
2. They are non-toxic and won't have any negative or side effects because they are biodegradable by nature and won't build up in bodily organs [26, 30].
3. Because nanogels are inactive in blood and plasma, they do not trigger non-immunologic reactions [26].
4. Nanosized particles prevent phagocytic cells from clearing them from the kidneys quickly, enabling both active and passive focused medication delivery [27].
5. This technique has the enormous advantage of controlling the rate, timing, and target of drug release in the body [26].
6. They are more capable of loading drugs [25].
7. Nanogels can be delivered in many different ways, including orally, nasally, pulmonaryly, transdermally, and topically [26, 28].
8. Drug delivery systems using nanogels can be created for both hydrophilic and hydrophobic drugs [26, 29, 31].

Disadvantages of nanogel

1. Surfactant toxicity can occur sometimes.
2. It necessitates pricy methods.

Materials and suppliers

Table no 1: Materials and suppliers

| Sr. No. | Materials | Suppliers |
|---------|------------------------------------|--------------------------------|
| 1 | <i>Jasminum Officinale</i> extract | AmsarPvt.Ltd, Indore |
| 2 | <i>CynodonDactylon</i> extract | AmsarPvt.Ltd, Indore |
| 3 | Carbapol 934 | Research lab Fine Chem. Mumbai |
| 4 | Propylene glycol | Research lab Fine Chem. Mumbai |
| 5 | Triethanolamine | Research lab Fine Chem. Mumbai |
| 6 | Methyl paraben | Research lab Fine Chem. Mumbai |
| 7 | Propyl paraben | Research lab Fine Chem. Mumbai |
| 8 | Poloxamer 407 | Research lab Fine Chem. Mumbai |
| 9 | Eudragit S100 | Research lab Fine Chem. Mumbai |
| 10 | Glycerine | Research lab Fine Chem. Mumbai |

Formulation of poly-herbal mouth ulcer nanogel

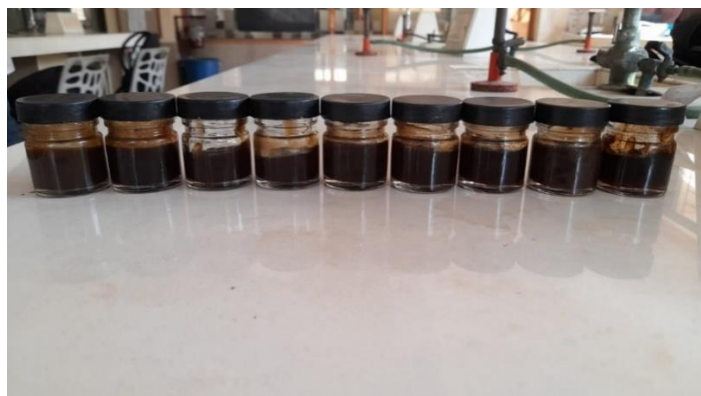


Figure 3: All nine batches of formulation

Table no 2: All ingredients

| Ingredients | F1 | F2 | F3 | F4 | F5 | F6 | F7 | F8 | F9 |
|-----------------------------------------|---------|---------|---------|---------|----------------|---------|---------|---------|---------|
| <i>Jasminum officinale</i> extract (gm) | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| <i>Cynodactylon</i> extract (gm) | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| Carbapol 934 (gm) | 0.11 | 0.4 | 0.2 | 0.68 | 0.4 | 0.2 | 0.4 | 0.6 | 0.6 |
| Eudragit S100(gm) | 0.035 | 0.013 | 0.02 | 0.035 | 0.035 | 0.05 | 0.05 | 0.05 | 0.02 |
| Poloxamer 407(gm) | 0.035 | 0.013 | 0.02 | 0.035 | 0.035 | 0.05 | 0.05 | 0.05 | 0.02 |
| Propylene glycol (ml) | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 |
| Methyl paraben (gm) | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Propyl paraben (gm) | 0.05 | 0.05 | 0.05 | 0.05 | 0.05 | 0.05 | 0.05 | 0.05 | 0.05 |
| Triethanolamine | 2 Drops | 2 Drops | 2 Drops | 2 Drops | 2 Drops | 2 Drops | 2 Drops | 2 Drops | 2 Drops |
| Ethanol (ml) | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| Distilled water | Q.S | Q.S | Q.S | Q.S | Q.S | Q.S | Q.S | Q.S | Q.S |

Evaluation of polyherbal mouth herbal nanogel

- pH measurement

Using an EQUI-TRONICS MODEL-614 digital pH metre, the pH of gel was determined. 100 ml of clean water and 1 g of gel were combined, then the mixture was set aside for 2 hours. Each formulation's pH was assessed three times, with the average results being computed.[33] (View Table No. 5).

- **Spreadability**

A pre-marked circle on a glass plate with a diameter of 1 cm was filled with 0.5 g of gel to test its spreadability. The plate was then put over another plate. A 500 g weight was allowed to lie on the top glass plate for 5 minutes.[34]

In Table No7. The formula was used to calculate it.

o $S = M \times L / T$

o M = Weight putted on upper side of the slide;

o L = Glass Slides' Length;

o T = Time to Separate Slides

- **Viscosity**

At 25 degrees Celsius, the viscosity of several gel formulations including Bombax ceiba thorn extract and psidium guajava leaf extract was determined. The Brookfield viscometer (Model LMDV 60) was used to measure the gel's viscosity. A 50 ml glass beaker was filled with accurately weighed 50 g of gel. Spindle number 6 was chosen, and it is submerged in the gel. The viscometer was run at 10 rpm until the reading stabilised, at which point the reading was recorded in pas. [35](View Table 7)

- **Moisture absorption studies**

One gramme of gel is inserted in the desiccator for this test. Alongside the gel in the same desiccator is a beaker filled with distilled water. After 24 hours, weigh the gel again. Gel formulation would get heavier if it took in any moisture.

- **Drug content**

One gramme of gel was dissolved in twenty millilitres of pH 7.4 phosphate buffer solution, and the mixture was then filtered through paper. Then, an absorbance measurement was made using a Shimadzu UV 1700 (Japan) UV spectrometer at 255 nm. (View Table 9 and Table 10)

- $$\text{Drug content} = \frac{\text{Theoretical concentration} - \text{practical concentration}}{\text{Theoretical concentration}} \times 100$$

- **Tests using centrifugation**

All nine batches of gel were put into centrifuged equipment (a Remi centrifuge) for centrifugation testing, and the separation of two phases was seen after an hour of operation at 1000 rpm. (View the 11th Table.)

- **The freeze-thaw test**

Herbal gels were subjected to a freeze-thaw test in which they were first allowed to thaw at ambient temperature for 24 hours after being frozen for 24 hours at -10 °C. Changes were noted by ocular observation after this cycle had been repeated five times.

- **Gel toughness**

The strength of the gel was assessed by measuring the number of seconds required for the weight to pierce the gel. Each of the optimal batches had a 5 gramme sample collected from it. A weight of 3.5gm was applied to the gel's surface. the time it takes for the weight to stably penetrate 0.5 cm of gel.(View Table 12).

- **Extrudability**

In standard capped collapsible aluminium tubes, the gel compositions were packaged and sealed. To assess extrudability, the thumb pressure was applied. Excellent +++, Good ++, and Satisfactory + were the grades given.[36](View Table 8)

- **Study of Stability**

Stability testing were performed on both closed and open containers. Gel was introduced here and kept at room temperature for three months.[37] (Refer to table 14)

- **Effective trapping**

Microcentrifuge (Remi) was used to centrifuge a small quantity of the nanodispersion for one hour at 10,000 rpm. A UV spectrophotometer (Jasco V530) was used to evaluate the absorbance of the adequately diluted supernatant solution at 274 nm in comparison to a blank or control nanodispersion after the supernatant was removed. The following equation was used to calculate the efficiency of entrapment.

$$\text{Entrapment Efficiency (\%)} = \frac{W_{\text{initial drug}} - W_{\text{free drug}}}{W_{\text{initial drug}}} \times 100 \dots \dots (2)$$

- **In-vitro Drug Release Studies**

The drug release investigations were conducted using Franz diffusion cells, which had an effective diffusion area of 3.14 cm² and a cell volume of 16.5 mL. Cellophane membrane surface was equally coated with gel (1 g). A cellophane membrane was clamped in the diffusion cell between the donor and receptor chambers. Freshly made phosphate buffer solution (pH 6.8) was placed within the receptor chamber. A magnetic stirrer was used to stir the receptor chamber. The samples were collected at the proper intervals. Following the proper dilutions, samples were examined for drug content using a UV visible spectrophotometer at max (nm). For each suitable time interval, the total amount of drug released was calculated as a function of time and replaced with new buffer.

- **Zeta potential**

The Malvern Zetasizer is used to calculate the nanogel preparation's zeta potential. The formulation is placed in a transparent, disposable zeta cell, and the outcome is obtained. Methanol is used to clean the cuvettes before to the experiment, and the sample is then put inside.

- **Antifungal activity**

Using the Cup-plate method, the antifungal activity of all created batches of formulation and without drug containing gel formulation (blank formulation) was compared to marketed antifungal formulations. Bacteria cultures used included *Aspergillus aureus* and *Candida albicans*. The antifungal test was conducted using agar well diffusion. The prepared nourishment was brought in and placed in sterile petri dishes to dry and chill. A micron wire loop was used to spread each bacterial culture. A sterile cork borer with a diameter of 6 mm was used to drill holes 4 mm deep. Following that, insert 0.5 gramme of gel from each batch through the holes. Plates were then incubated at 27°C for 48 hours. The zone of inhibition (in mm) formed for each chemical with each fungal strength was then measured.

- **Cell line study**

Experimental procedure:

1. Cells were incubated at a concentration of 1×10^4 cells/ml in culture medium for 24 h at 37°C and 5% CO₂.
2. Cells were seeded at a concentration (70 μl) 10^4 cells/well in 100 μl culture medium and 100 μl synthesized compounds (10, 40, 100 μL/ml) into micro plates respectively (tissue culture grade, and 96 wells).
3. Control wells were incubated with DMSO (0.2% in PBS) and cell line. All samples were incubated in triplicate. Controls were maintained to determine the control cell survival and the percentage of live cells after culture.
4. Cell cultures were incubated for 24 h at 37°C and 5% CO₂ in CO₂ incubator (Thermo scientific BB150)
5. After incubation, the medium was completely removed and added 20 μl of MTT reagent (5 mg/min PBS).
6. After addition of MTT, cells incubated for 4 hrs at 37°C in CO₂ incubator.

7. Observed the wells for formazan crystal formation under microscope. The yellowish MTT was reduced to dark coloured formazan by viable cells only.
8. After removing the medium completely. Added 200µl of DMSO (kept for 10 min) and incubate at 37°C (wrapped with aluminium foil).
9. Triplicate samples were analyzed by measuring the absorbance of each sample by a Elisa microplate reader (Benesphera E21) at a wavelength of 570nm.

RESULT AND DISCUSSION

UV of *Jasminum officinale*

Table 3: Absorbance range of *Jasminum officinale* extract at different concentration

| Concentration (ug/ml) | Absorbance of extract |
|------------------------|-----------------------|
| 2 | 0.014 |
| 4 | 0.023 |
| 6 | 0.031 |
| 8 | 0.042 |
| 10 | 0.042 |
| R2 | 0.997 |
| Slope | 0.0047 |
| Intercept | 0.0046 |

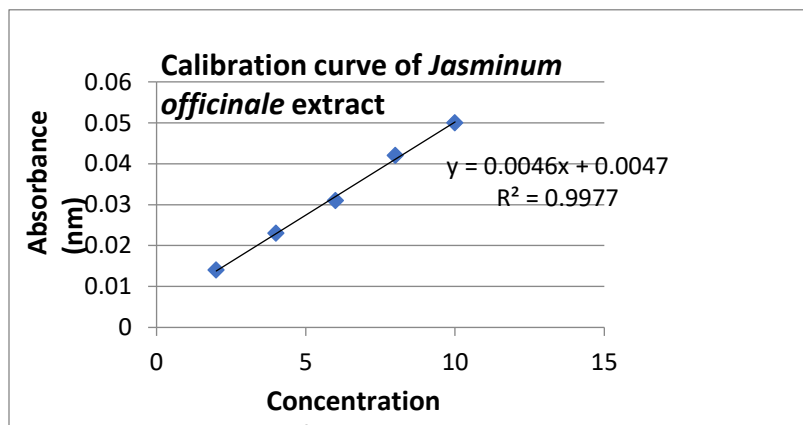


Figure 4: Calibration curve of *Jasminum officinale* extract

UV of *Cynodon Dactylon*

Table 4: Absorbance range of *Cynodon dactylon* extract at different concentration

| Concentration (ug/ml) | Absorbance of extract |
|------------------------|-----------------------|
| 2 | 0.89 |
| 4 | 0.145 |
| 6 | 0.218 |
| 8 | 0.288 |
| 10 | 0.350 |
| R2 | 0.998 |
| Slope | 0.0185 |
| Intercept | 0.0333 |

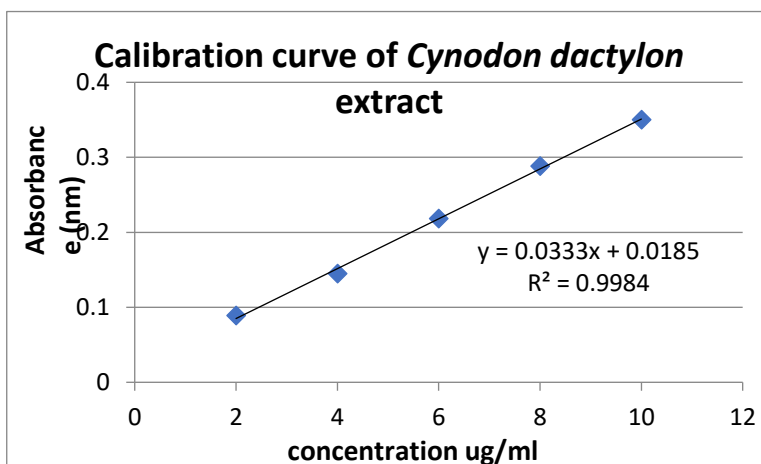


Figure 5: Calibration curve of *Cynodon dactylon* extract

pH of nanogel

Table no 5: pH determination

| Formulations | pH |
|--------------|-------------|
| F1 | 6.82 |
| F2 | 6.74 |
| F3 | 6.60 |
| F4 | 6.70 |
| F5 | 6.50 |
| F6 | 6.59 |
| F7 | 6.43 |
| F8 | 6.65 |
| F9 | 6.52 |

Spreadability

Table no 6: Spreadability

| Formulation | Spreadability (gm.Cm/sec) |
|-------------|---------------------------|
| F1 | 6.8 |
| F2 | 7.1 |
| F3 | 7.3 |
| F4 | 6.7 |
| F5 | 7.9 |
| F6 | 7.4 |
| F7 | 6.9 |
| F8 | 7.5 |
| F9 | 7.2 |

Viscosity

Table no 7: Viscosity

| Formulation | Viscosity(cps) |
|-------------|----------------|
| F1 | 3309 |
| F2 | 2987 |
| F3 | 3086 |
| F4 | 2842 |
| F5 | 3982 |
| F6 | 3521 |
| F7 | 2722 |
| F8 | 3264 |
| F9 | 3720 |

Moisture absorption studies:

Considering that there were no weight changes after the gel was placed next to a beaker holding water in the desiccator, batches F1, F2, F3, F4, F5, F6, F7, and F8 of the formulation passed the test. These formulas remain stable over time. Although batch F9 of the formulation indicates a minor weight gain, this could be because this formulation contains more propylene glycol than other formulations. Stability is impacted by this element.

Extrudability

Table no 8: Extrudability

| Formulation | Extrudability |
|-------------|---------------|
| F1 | ++ |
| F2 | +++ |
| F3 | ++ |
| F4 | + |
| F5 | +++ |
| F6 | ++ |
| F7 | ++ |
| F8 | ++ |
| F9 | + |

Drug content of *Jasminum Officinale*

Table 9: % Drug content of *Jasminum officinale*

| Formulation | Drug content |
|-------------|--------------|
| F1 | 90.42% |
| F2 | 80.85% |
| F3 | 82.97% |
| F4 | 93.61% |

| | |
|-----------|---------------|
| F5 | 96.80% |
| F6 | 92.55% |
| F7 | 95% |
| F8 | 84.04% |
| F9 | 87.23% |

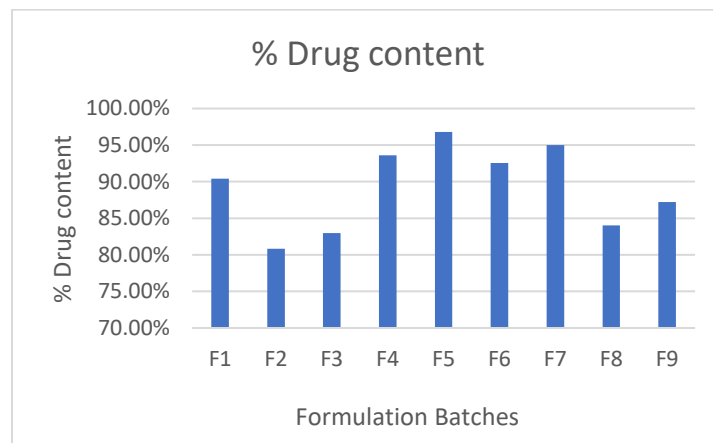


Figure 6: % Drug content of Jasminum officinale extract

Drug content of *CynodonDactylon*

Table no 10: Drug content of *CynodonDactylon*

| Formulation | Drug content |
|--------------------|---------------------|
| F1 | 91.89% |
| F2 | 81.08% |
| F3 | 83.78% |
| F4 | 94.59% |
| F5 | 97.29% |
| F6 | 93.24% |
| F7 | 96.21% |
| F8 | 86.48% |
| F9 | 89.18% |

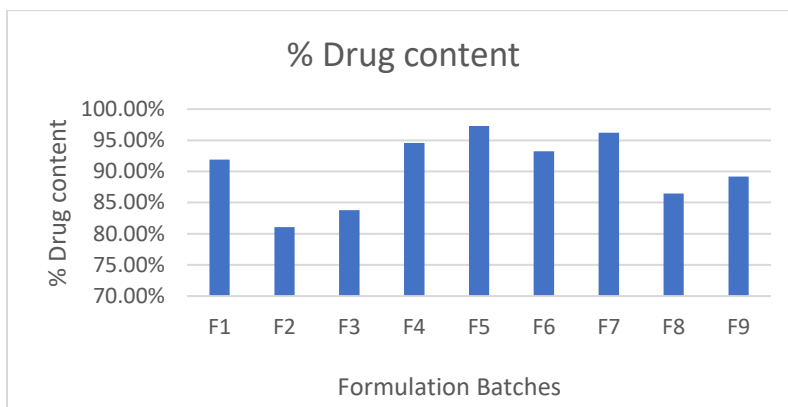


Figure 7: % Drug content of *Cynodon dactylon* extract

Centrifugation test

Table no 11: Centrifugation test

| Formulation | Centrifugation test |
|-------------|---------------------|
| F1 | No phase separation |
| F2 | No phase separation |
| F3 | No phase separation |
| F4 | No phase separation |
| F5 | No phase separation |
| F6 | No phase separation |
| F7 | No phase separation |
| F8 | No phase separation |
| F9 | phase separation |

Freeze thaw testing:

All of the gels were kept at ambient temperature and in freezers. The appearance, colour, texture, and stability of all gels remain the same, and phase separation cannot be seen. Thus, the freeze-thaw test is successful for all gels.

Gel strength

Table no 12: Gel strength

| Formulation | Gel strength (Seconds) |
|-------------|-------------------------|
| F1 | 18 |
| F2 | 25 |
| F3 | 32 |
| F4 | 20 |
| F5 | 36 |
| F6 | 35 |
| F7 | 28 |
| F8 | 31 |
| F9 | 22 |

SEM

The 38.93nm size of the nanogel formulation F5 was detected using SEM, which was carried out using an instrument called IMINA under the magnification of 100x and energy range of 10,000 electron volts. It was noted that the SEM image demonstrates that there is no breakdown of the nanogel.

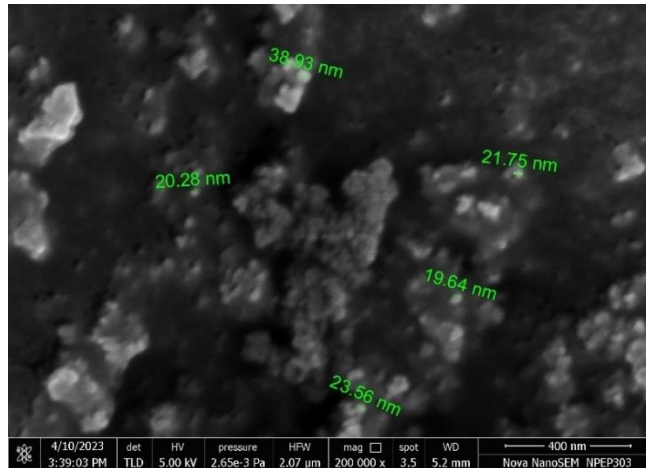


Figure 8-Scanning electron microscopy of nanogel formulation

Particle size

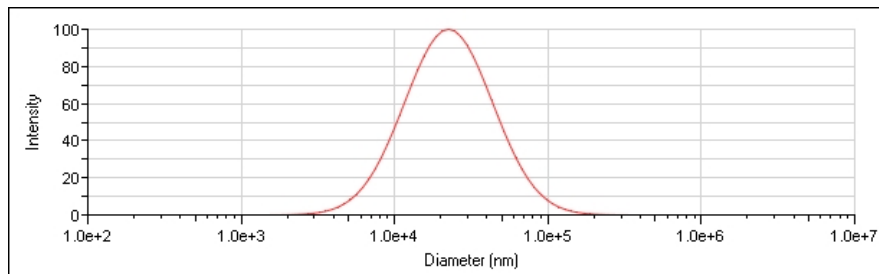


Figure 9: Particle size of optimized formulation

Zeta potential

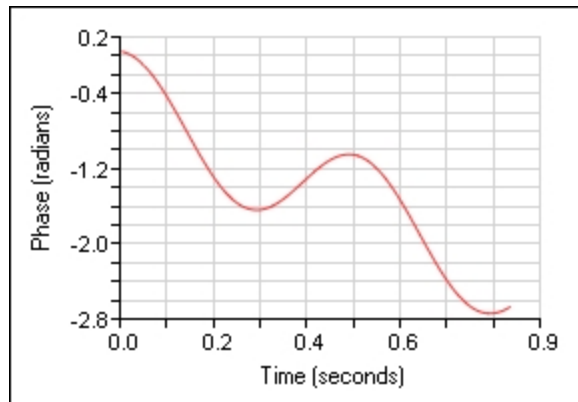


Figure10 : Zeta potential graph of optimized batch

Entrapment Efficiency

Table no 13: Entrapment Efficiency

| Formulation | Entrapment Efficiency |
|-------------|-----------------------|
| F1 | 79.80% |
| F2 | 85.47% |
| F3 | 81.62% |
| F4 | 84.92% |
| F5 | 88.35% |
| F6 | 78.88% |
| F7 | 75.29% |
| F8 | 80.73% |
| F9 | 82.59% |

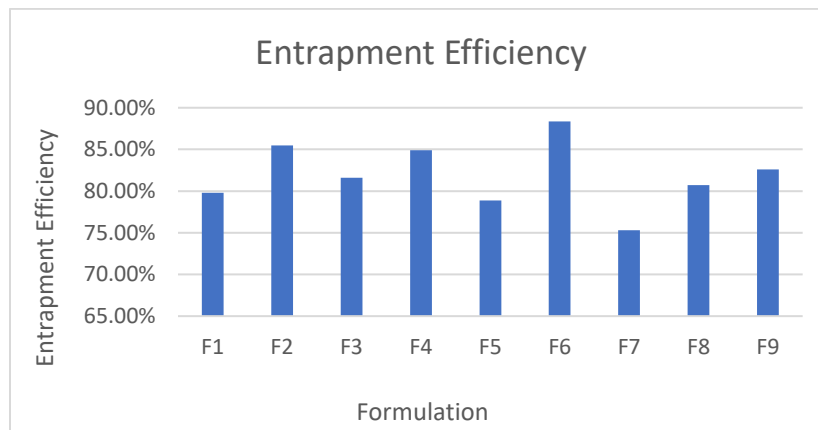


Figure 11: Entrapment Efficiency

% Drug release of *Jasminum officinale*

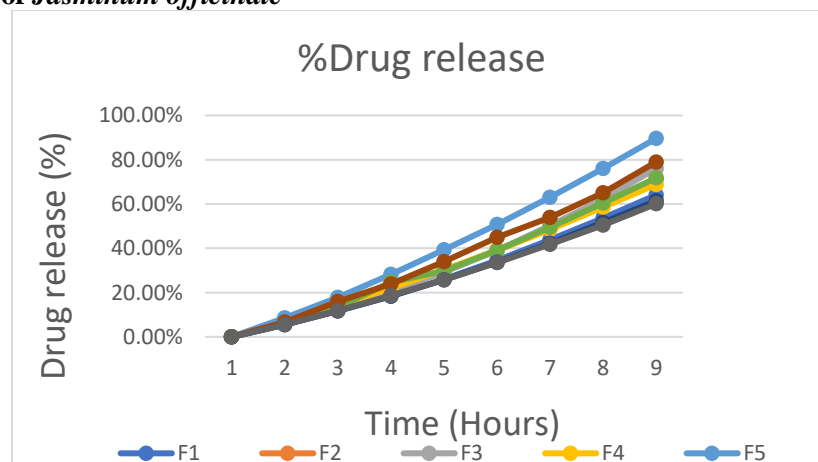


Figure 12: % Drug release of *Jasminum officinale* extract

% Drug release of *Cynodondactylon* extract

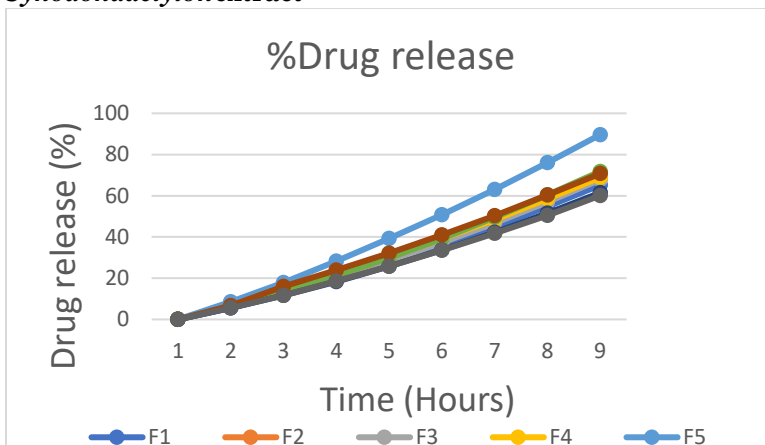


Figure 13 : % Drug release of *Cynodondactylon* extract

Stability study

Table no 14: Stability study

| Temperature and humidity | Parameter | Observation (in months) | (in months) |
|--------------------------|---------------------------|----------------------------|----------------------------|
| | | Stability data for 1 month | Stability data for 2 month |
| 30 ± 2°C / 65 ± 5% RH | pH | 6.54 | 6.56 |
| | Colour | Reddish Brown | Reddish Brown |
| | Texture | Smooth | Smooth |
| | Viscosity (Pa.s) | 3.090 | 3.099 |
| | Spreadability (gm.cm/sec) | 5.86 | 5.72 |
| 40 ± 2°C / 75 ± 5% RH | pH | 6.89 | 6.95 |
| | Colour | Reddish Brown | Reddish Brown |
| | Texture | Smooth | Smooth |
| | Viscosity (Pa.s) | 3.509 | 3.690 |
| | Spreadability (gm.cm/sec) | 4.76 | 4.30 |

Optimized HPTLC condition

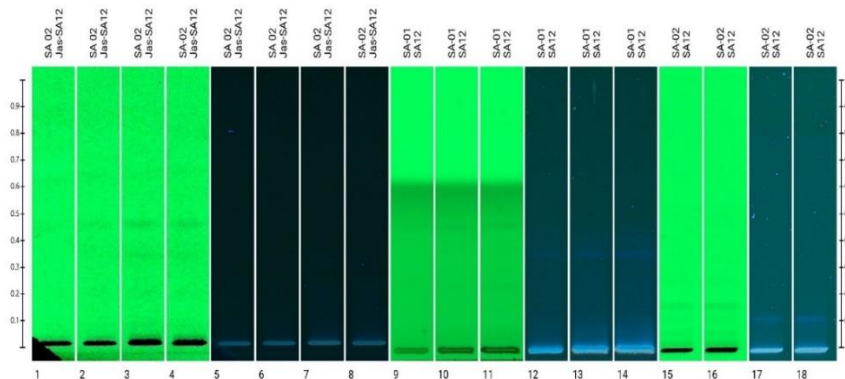


Figure14:HPTLC fingerprint of *Jasminum officinale* extract at R 366nm

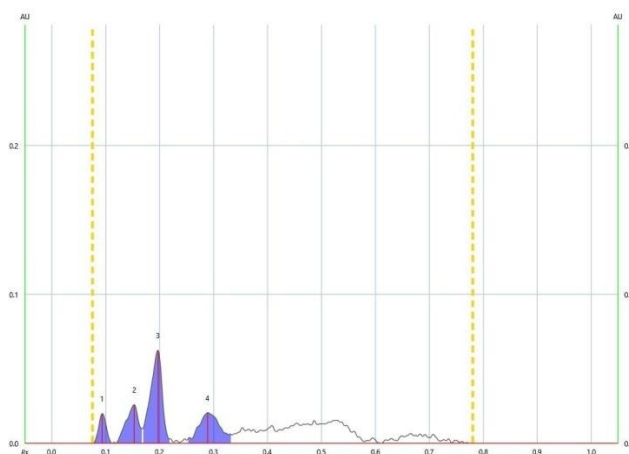


Figure15: Densitogram of *Jasminum officinale* extract

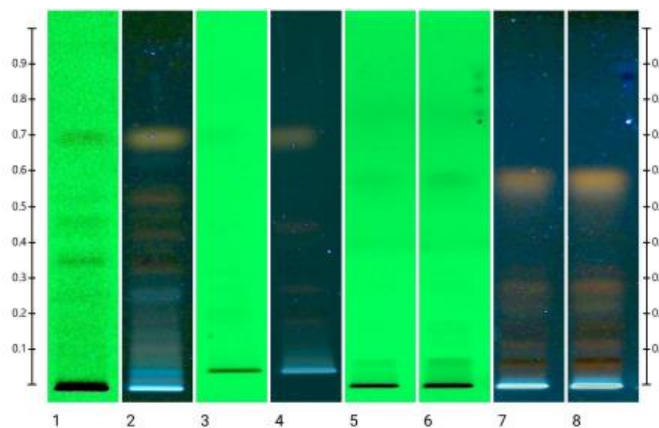


Figure 16:HPTLC fingerprint of *Cynodon dactylon* extract at R 366nm

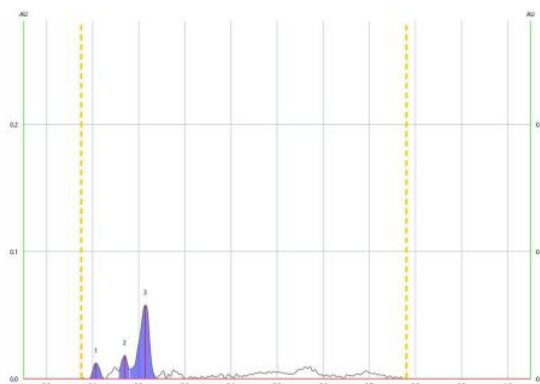


Figure 17: Densitogram of *Cynodon dactylon* extract

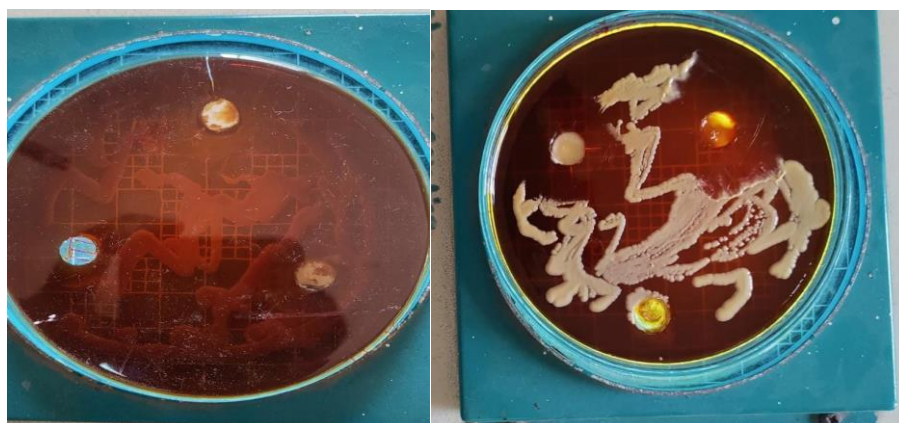


Figure 28: Antifungal activity

Cell line study result:

Table 15: Effects of compound against L-929 (adherent type of mouse fibroblast cell line) by MTT assay

| Sr. no | Sample | Concentration (µg/ml) | OD | Mean | % inhibition | IC 50 (µg/ml) |
|--------|-------------|-----------------------|-------|-------|--------------|---------------|
| 1 | Control | | 0.899 | 0.875 | | |
| | | | 0.891 | | | |
| | | | 0.837 | | | |
| 2 | Std. 5 FU | 10 | 0.212 | 0.203 | 76.08 | 32.07 |
| | | | 0.201 | | | |
| | | | 0.196 | | | |
| | | 40 | 0.105 | 0.115 | 86.85 | |
| | | | 0.117 | | | |
| | | | 0.125 | | | |
| | | 100 | 0.078 | 0.090 | 89.71 | |
| | | | 0.093 | | | |
| | | | 0.100 | | | |
| 3 | Sample – F3 | 10 | 0.389 | 0.374 | 57.25 | 37.50 |
| | | | 0.375 | | | |
| | | | 0.358 | | | |
| | | 40 | 0.332 | 0.327 | 62.62 | |
| | | | 0.330 | | | |
| | | | 0.321 | | | |
| | | 100 | 0.301 | 0.295 | 66.28 | |
| | | | 0.296 | | | |
| | | | 0.290 | | | |

According to Table, at the different Concentration (10µg/ml,100µg/ml,100µg/ml) of Sample–F3 compounds carried out for anticancer activity against L-929 (adherent type of mouse fibroblast cell line). The positive control 5 Flurouracil was used as standard drug. The Sample–F3 showed good activity as compared to standard compound.



Figure 29: Control



Figure 30: Standard 5FU



Figure 31: Sample

Conclusion

The research of *Cynodondactylon* and *Jasminum officinale* extracts for the successful treatment of mouth ulcers may boost drug penetration from the affected area, which may demonstrate both antifungal and antibacterial action. Propylene glycol may improve gel stability because it is present. *Jasminum officinale* extract with antiulcer properties. Additionally, it possesses antioxidant properties that aid in shielding the mouth's surface from oxidative damage. The phenolic acids, flavonoids, terpenoids, glycosides, and saponins in *Cynodondactylon* extract have antibacterial and antiulcer properties. Thus, a polyherbal combination of *Cynodondactylon* and *Jasminum officinale* extracts was added to the gel used to treat mouth ulcers.

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