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RESEARCH ARTICLE

Extraction, Phytochemical Study, Formulation & Evaluation of Antiulcer Activity of Jasminum Grandiflorum L.

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ABSTRACT

The present research has been undertaken with the aim to formulate & evaluate antiulcer mouth gel containing jasminum grandiflorum L. extract. The gel formulation was designed by using 70% methanol extract of Jasminum Grandiflorum L. & evaluated for various parameters. The gel was formulated using accurately weighed the amount of extract along with other additives, poured into the fixed amount of glycerol dispersion with frequent heating & constant stirring. The mouth ulcer gel formulations prepared were subjected to preliminary evaluation such as ph, drug content uniformity, in vitro diffusion study. The parameters were found to be satisfactory. The result of the study reveals significant acceptability of gel formulation of Jasminum grandiflorum L. since evaluation parameters lie in range.

KEYWORDS

Jasminum Grandiflorum L., Antiulcer mouth gel, 70% methanol.

INTRODUCTION

1. Mouth ulcer

A mouth ulcer is an ulcer that occurs on the mucous membrane of the oral cavity. Mouth sores are very common, occurring in association with many diseases and by many different mechanisms, but usually, there is no serious underlying cause. The two most common causes of oral ulceration are local trauma (e.g. rubbing from a sharp edge on a broken filling) and aphthous stomatitis ("canker sores"), a condition characterized by the recurrent formation of oral ulcers for unknown reasons.

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Mouth ulcers often cause pain and discomfort and may alter the person's choice of food while healing occurs (e.g. avoiding acidic or spicy foods and beverages).

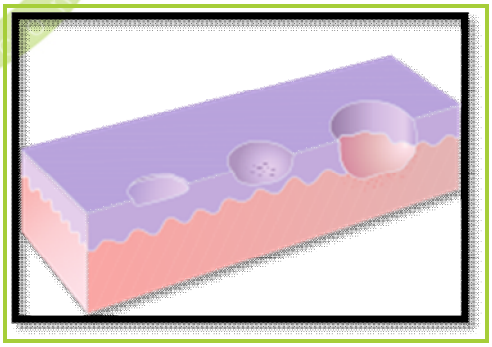

Fig no:-1 Diagrammatic representation of mucosal erosion (left), excoriation (center), and ulceration (right)

They may form individually, or multiple ulcers may appear at the same time (a "crop" of ulcers). Once formed, the ulcer may be maintained by inflammation and/or secondary

infection. Rarely, a mouth ulcer that does not heal may be a sign of oral cancer.

An ulcer is a break in the skin or mucous membrane with loss of surface tissue and the disintegration and necrosis of epithelial tissue. A mucosal ulcer is an ulcer which specifically occurs on a mucous membrane. An ulcer is a tissue defect which has penetrated the epithelial-connective tissue border, with its base at a deep level in the submucosa, or even within muscle or periosteum. An ulcer is a more in-depth breach of the epithelium than an erosion or an excoriation and involves damage to both epithelium and lamina propria.

Erosion is a superficial violation of the epithelium, with little damage to the underlying lamina propria. A mucosal erosion is an erosion which specifically occurs on a mucous membrane. Only the superficial epithelial cells of the epidermis or the mucosa are lost, and the lesion can reach the depth of the basement membrane. Erosions heal without scar formation.

Excoriation is a term sometimes used to describe a breach of the epithelium which is deeper than an erosion but shallower than an ulcer. This type of lesion is tangential to the rete pegs and shows punctiform (small pinhead spots) bleeding, caused by exposed capillary loops. Jasminum Gradiflorium L. (family: - Oleaceae) exhibit a wide ecological range & found extensively all over India. These leaves are used in the treatment of fixing loose teeth, ulcerative stomatitis, leprosy skin diseases, dysmenorrhoea, ulcers, wounds. The leaves of this species have the distinction of being used in the Indian folk medicine for treating ulcers. The objective of the present study was to formulate antiulcer mouth gel by using 70% methanolic extract of Jasminum grandiflorum L.[1]

2. Gels

A gel is a two-component, crosslinked three-dimensional network consisting of structural materials interspersed by an adequate but proportionally large amount of liquid to form an infinite rigid network structure which immobilizes the liquid continuous phase within. A gel is an intermediate state of matter possessing the property of a solid and a liquid, termed as viscoelasticity.

The U.S.P. defines gels as a semisolid system consisting of dispersion made up of either small inorganic particle or large organic molecule enclosing and interpenetrated by a liquid. The inorganic particles form a three-dimensional “house of cards” structure.

I. Properties of Gels

Gels should possess the following properties

1. Ideally, the gelling agent for pharmaceutical or cosmetic use should be inert, safe, and should not react with other formulation components.

2. The gelling agent included in the preparation should produce a reasonable solid-like nature during storage that can be easily broken when subjected to shear forces generated by shaking the bottle, squeezing the tube, or during topical application.

3. It should possess suitable anti-microbial to prevent from microbial attack.

4. The topical gel should not be tacky.

5. The ophthalmic gel should be sterile.

II. Characteristics of Gels

A) Swelling

When a gelling agent is kept in contact with a liquid that solvates it, then an appreciable amount of liquid is taken up by the agent and the volume increases. This process is referred to as swelling. This phenomenon occurs as the solvent penetrates the matrix.Gel-gel interactions are replaced by gel solvent interactions. The degree of swelling depends on the number of linkages between individual molecules of gelling agent and on the strength of these linkages.

B) Syneresis

Many gels often contract spontaneously on standing and exude some fluid medium.This

effect is known as syneresis. The degree to which syneresis occurs, increases as the concentration of gelling agent decreases. The occurrence of syneresis indicates that the original gel was thermodynamically unstable. The mechanism of contraction has been related to the relaxation of elastic stress developed during the setting of the gels. As these stresses are relieved, the interstitial space available for the solvent is reduced, forcing the liquid out.

C) Ageing

Colloidal systems usually exhibit slow spontaneous aggregation. This process is referred to as aging. In gels, aging results in the gradual formation of a denser network of the gelling agent. Theimer suggests that this process is similar to the original gelling process and continues after the initial elation since the fluid medium is lost from the newly formed gel.

D) Structure

The rigidity of a gel arises from the presence of a network created by the interlinking of particles of the gelling agents. The nature of the particle and the type of force that is responsible for the linkages determine the structure of the network and the properties of the gel.

E) Rheology

Solutions of the gelling agents and dispersion of flocculated solid are pseudoplastic i.e. exhibiting Non-Newtonian flow behavior, characterized by a decrease in viscosity with an increase in shear rate. The tenuous structure of inorganic particles dispersed in water is disrupted by applied shear stress due to breaking down of inter-particulate Association, exhibiting a greater tendency to flow. Similarly, for macromolecules the applied shear stress aligns the molecules in the direction of stress, straightening them out and lessening the resistance to flow.

III. Uses

In the pharmaceutical and cosmetic industry, the gel may be enumerated to have the following applications.

- As delivery systems for orally administered drugs.
- To deliver topical drug applied directly to the skin, mucous membrane or the eye.
- As long-acting forms of drug injected intramuscularly.
- As binders in tablet granulation, protective colloids in suspensions, thickeners in oral liquid and suppository bases.
- In cosmetics like shampoos, fragrance products, dentifrices, skin and hair care preparations.

IV. Classification of Gels

Gels can be classified based on colloidal phases, nature of the solvent used, physical nature and rheological properties.

1. Based on colloidal phases

They are classified into

- Inorganic (two-phase system)
- Organic (single phase system)

Two phase system

If the partial size of the dispersed phase is relatively large and form the three-dimensional structure throughout gel, such a system consists of floccules of small particles rather than larger molecules and gel structure, in this system is not always stable. They must be thixotropic-forming semisolids on standing and become liquid on agitation.

Single-phase system

These consist of the major organic molecules existing on the twisted strands dissolved in a continuous phase. This more important organic molecule either natural or synthetic polymers are referred as gel formers, they tend to entangle with each other the in random motion or bound together by Vander Waals forces.

2. Based on nature of solvent

Hydrogels (water based)

Here they contain water as their continuous liquid phase

E.g. bentonite magma, Gelatin, cellulose derivatives, carpooler, and poloxamer gel.

Organic Gels (with a non-aqueous solvent)

These contain a non-aqueous solvent on their continuous phase. E.g. plastic base (low molecular wet. polyethylene dissolved in mineral oil & short Cooled)

Xerogels

Solid gels with low solvent concentration are known as xerogels. These are produced by evaporation of the solvent or freeze drying, leaving the gel framework behind incontact with the fresh fluid, they swell and can be reconstituted. E.g. Tragacanth ribbons, acacia tear β-cyclodextrin, dry cellulose, and polystyrene.

3. Based on rheological properties

Usually, gels exhibit non-Newtonian flow properties.

They are classified into,

a) Plastic gels

b) Pseudoplastic gels

c) Thixotropic gels.

Preparation of gels

Gels are typically in the industrial scale prepared under room temperature. However few of polymers need special treatment before processing. Gels can be prepared by following methods.

1. Thermal changes

2. Flocculation

3. Chemical reaction

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Formulation Considerations for Pharmaceutical Gels

The choice of vehicle/solvent

Purified water is used as a solvent. To enhance the solubility of the therapeutic agent in the dosage form and/or to improve drug permeation across the skin, co-solvents may be used, E.g., alcohol, glycerol, PG, PEG 400, etc.

Inclusion of buffers

Buffers may be involved in aqueous and hydroalcoholic-based gels to control the pH of the formulation. The solubility of buffer salts is reduced in hydroalcoholic-based vehicles.

E.g., Phosphate, Citrate, etc.

Preservatives

Certain preservatives cooperate with the hydrophilic polymers used to prepare gels, thereby reducing the concentration of free (anti¬microbially active) preservative in the preparation. Therefore, to compensate for this, the initial concentration of these preservatives should be improved. E.g., Parabens, phenolics, etc.

Antioxidants

It may be involved in the formulation to improve the chemical stability of therapeutic agents that are prone to oxidative degradation. Its choice is based on the nature of the vehicle used in the preparation of gel. Water-soluble antioxidants are used as the majority of gels are aqueous-based. E.g., Sodium metabisulphite, sodium formaldehyde sulfoxylate, etc.

Flavors/Sweetening agents

- **Manufacture of Gels**

The water soluble excipients are firstly dissolved in the vehicle, in a mixing vessel by using mechanical stirrer.

To prevent aggregation, add hydrophilic polymer to the stirred mixture slowly. Stirring is continued until the dissolution of the polymer has oc¬curred. The excessive stirring results in entrapment of air. The mixing rate must not be extreme, or a mixing vessel may be used to which a vacuum may be pulled, to prevent the entrapment of air.[2]

3. PHARMACOGNOSTIC STUDY

Fig: 2 JasminumGradiflorum L.

Kingdom:-plantae plants

Subkingdom:-tracheobionts-vascular plants

Division:-mgnoliophyta flowering plants

Class;-magnoliopsida-dicotyledons

Order:-scrophulariales

Family:-oleaceae-olive family

Genus:-jasminum

Species:-grandiflorum3

Chemical constituents

The chameli leaves contain resins, salicylic acid, jasmininean alkaloid & an astringent principle. An indoleoxygenase has been isolated from the leaves. Secoiridoid glycosides viz.2"epifraxamoside& demethyl-2"epifraxamoside,jasminanhydride also contains ascorbic acid anthranilic acid & its glycosides.

Uses

The leaves of jasminum grandiflorum are used in folk medicine for treating ulcerative stomatitis, toothache, skin disease, ulcers, wounds, corns,& also as a gargle.

It is also useful in odontalgia, leprosy, ottorhoea, otalgia, strangury.

MATERIALS & METHODS

1. Plant material

Jasminum grandiflorum leaves were collected from Gadhinglaj in the month of February 2017. The plant was identified & authenticated by Mrs. Chivalkar

2. Chemicals

The chemical used for the extraction was 70% methanol.

The chemicals used for the formulations were glycerol, dextrose, amaranth solution, propylene glycol, dill oil, peppermint oil, methylparaben, tartrazine agar etc.

The chemicals used for phytochemical screening were Mayer's reagent, Dragendroff reagent, etc.

3. Instruments

Incubator

Franz diffusion cell

Mechanical shaker

UV- spectrophotometer

4. Preparation of extract

Fresh leaves were collected, shade-dried & powdered mechanically. About 40 gm of leaf powder was extracted with 400ml of 70% methanol by maceration at room temperature for 4 hours using a mechanical shaker. The extract was dried at 40 0C under vacuum, and the yield of the extract was 24 %.4

5. Phytochemical screening

5.1 Test For Alkaloids

Evaporate the alcoholic extract separately. To the residue, add dilute HCL. Shake well and filter. With filtrate, perform the following test:

4.1 (a)Dragendorff's test

To 2-3 ml filtrate, Add few drops Dragendorff's reagent.Orange brown ppt is formed.

4.1(b) Mayer's test

I-3 ml filtrate with few drops Mayer's reagent gives ppt.5

5.2 Solubility test

a. Ethanol: - completely dissolve

Table no:-1

INGREDIENTS	S1	S2	S3
DRUG	0.1 gm	0.1gm	0.1 gm
GLYCEROL	3.1 ml	2.6ml	1.5ml
SUCROSE	0.5gm	0.2gm	0.4gm
COLOURING AGENT	q.s	q.s	-
FLAVOURING AGENT	0.1ml	q.s	-
PROPYLENE GLYCOL	1ml	2ml	2ml
PRESERVATIVE	0.1gm	0.2gm	0.1gm
DILL OIL	0.1ml	-	-
AGAR AGAR	-	-	1gm
TOTAL	5ml	5ml	5ml

b. Water: - sparingly soluble

c. Glycerol: - soluble by frequent heating.

5. EXPERIMENTAL METHOD

5.1 Plant Extract

Fresh leaves were collected, shade-dried & powdered mechanically. About 40 gm of leaf powder was extracted with 400ml of 70% methanol by Maceration at room temperature for 4 hours using a mechanical shaker. The extract was dried at 40 0C under vacuum, and the yield of the extract was 24 %.[4]

Fig: 3 Formulation S1 &S2

5.2.2.Formulation Procedure

- All ingredients were weighed accurately according to the required quantity.
- Then the 0.1gm of drug i.e. Jasminum Grandiflorum extract was taken.
- The Glycerol of about 3.1ml was taken in the beaker & the drug was added to it.
- Constant stirring with heating was given to the above mixture & the drug was completely dissolved in the Glycerol.
- Then 0.5gm of Dextrose was added as a sweetening agent in the formulation.
- The coloring agent Amaranth was added as quantity sufficient
- Then the Peppermint oil was added as a flavoring agent of about 0.1gm.
- Then the Propylene glycol of about 1 ml was added to the formulation.
- Then Methylparaben was added as a preservative about 0.1gm
- And then the Dill oil was added in the formulation.

EVALUATION TESTS

1. Homogeneity

After the gels have been set in the container, all developed gels were tested for homogeneity by visual inspection. They were tested for their appearance and presence of any aggregates.

2. Grittiness

All the formulations were evaluated microscopically for the presence of any appreciable particulate matter which was seen under a light microscope. Hence naturally the gel preparation fulfills the requirement of freedom of particular issue and grittiness as desired for any topical preparation

3. Skin irritation

Skin irritation test was performed for selected gels on human volunteers to find out any irritation problems which could make it unsuitable for topical use.Skin irritation test was conducted, for each gel on three volunteers.Approx,1gm gel was topically applied to the hand near the wrist over a two sq.inch area and observed for any lesions or irritation or redness.

4. Stability Test

Preparation of nutrient media

Weigh the required quantity of all ingredients as shown as below. Mix all ingredients to form a solid mass by frequent heating.

Then add the formulation into the nutrient media in a zig-zag manner. After that kept the media into an incubator at 370 C for 24 hours. They were tested for their stability.

5. pH TEST

The pH of gel formulations was determined by using pH paper. The pH of the formulation was determined by comparing with the standard pH scale.

6. In vitro Diffusion studies

The diffusion studies of the prepared gels can be carried out in Franz diffusion cell for studying the dissolution release of gels through a cellophane membrane. Gel sample (0.5g) was taken in cellophane membrane, and the diffusion studies were carried out at 37 ± 1° using 250 ml of phosphate buffer (pH 7.4) as the dissolution medium.

Five milliliters of each sample was withdrawn periodically at 1, 2, 3, 4, 5, h and each sample was replaced with equal volume of fresh dissolution medium. Then the samples were analyzed for the drug content by using phosphate buffer as blank.[4]

RESULTS & DISCUSSION

The appearance of gel

Table: 2 Physical evaluation of formulations

Sample	Colour	Odour	Texture
S1	Reddish brown	Aromatic	Homogenous
S2	Slightly yellowish	Aromatic	Homogenous
S3	Reddish brown	Aromatic	Homogenous

2. Homogeneity

The formulations were tested by visual inspection,& it was found that there is an absence of aggregates. And formulations having smooth, homogenous texture

3. Grittiness

The formulations were evaluated under light microscope,& it was found that there is absence of particulate matter & free from grittiness

4. Skin Irritancy Test

No allergic symptoms like inflammation, redness, irritation witnessed on human volunteers up to 2hrs.

5. pH Test

The PH of the gel formulations was in the range of 6 to7, which lies in the standard pH range of the mouth(skin) and would not fabricate any skin irritation. There was no considerable change in pH values as a function of time for all formulations, which is shown in table no.

Table: 3 pH range

Formulation Code	pH
S1	7
S2	6
S3	7

6. In vitro diffusion/ release Study

The release of an active pharmaceutical ingredient from the gel was varied according to polymer concentration. The drug release from its emulsified gel formulation can be categorized in the ascending order.

Table: 4 Drug release

Time in hr.	S1	S2	S3
1	0.6937	0.4182	0.5363
2	1.4470	0.9658	0.9851
3	1.6665	1.5634	2.3727
4	2.4834	2.2540	1.7108
5	2.6379	1.8380	1.9329

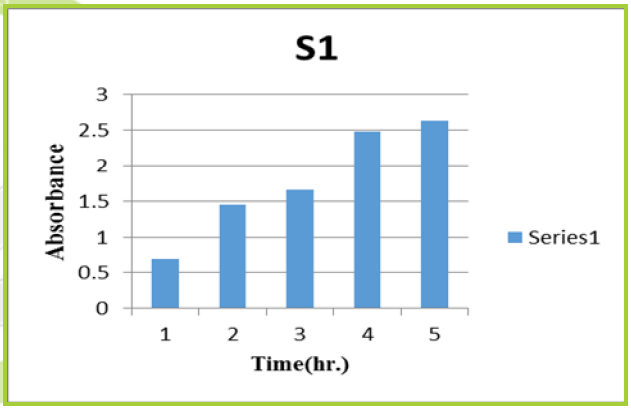

Fig no:-4

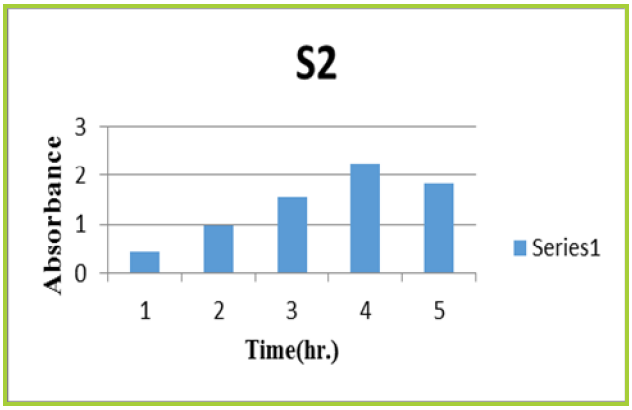

Fig: 5

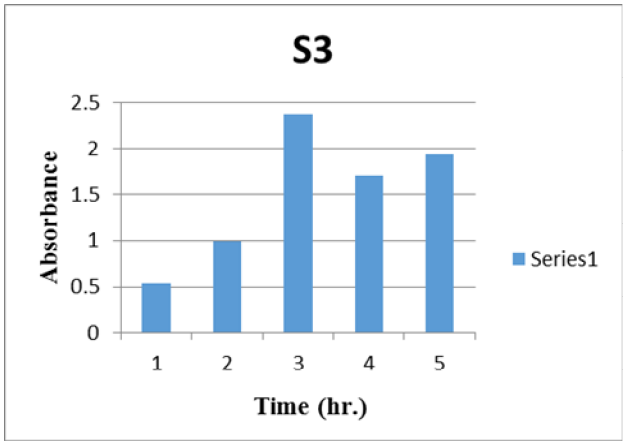

Fig: 6

Comparative study of the formulation (S1, S2, and S3)

Table: 5 comparative study

Time in hr.	S1	S2	S3
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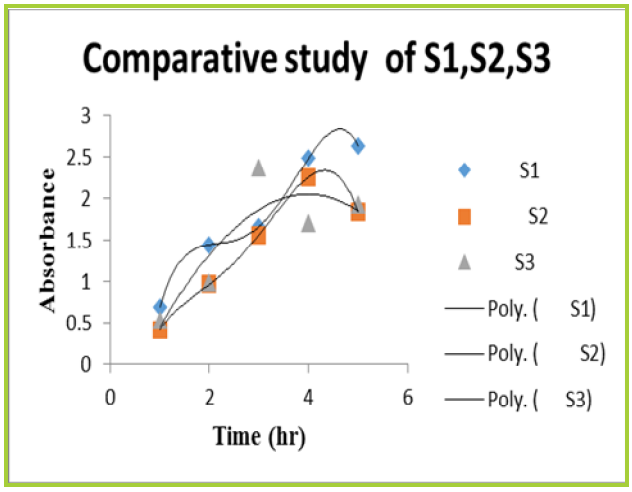

Fig: 7

CONCLUSION

The results of the present study indicated that the crude aqueous extract of Jasminum grandiflorum leaves possesses anti-ulcer activity. The phytochemical analysis shows the presence of alkaloids, flavonoids, glycosides, phytosterols, and carbohydrates. Further studies using in vitro models and to isolate active constituents from extract a required to carry out and established. The mouth ulcer gel formulation was evaluated for various pharmaceutical parameters and results were satisfied. In conclusion this extraction studies it found that the leaves *Jasminum grandiflorum* has potent anti-ulcer activity and the mouth ulcer gel formulation showed a moderate release of drug in an optimum period. By comparing all formulations, formulation S1 gives better result in diffusion study.

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