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Development and Characterization of Gel for Its Anti-Microbial Activity Containing Different Herbal Extracts

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ABSTRACT: Gels are transparent to opaque semisolid containing gelling agent that merges or entangles to form a three-dimensional colloidal network structure. It is responsible for a gel resistance to deformation and its visco-elastic properties. Gels have better potential as a vehicle to administer drug topically in comparison to ointments because they are non-sticky, require low energy during formulation, have aesthetics value and are stable. Herbal Anti-microbial gel containing Capsicum annuum and Cinnamomum verum were successfully formulated and characterized. Firstly the herbal extract was obtained by Soxhlet extraction and aqueous extraction. Herbal anti-microbial gel were formulated using carbopol-934 as gelling agent and NaOH is used as preservative. Using herbal extracts three formulations were formulated based on different formula and concentration. All these formulation (F1,F2,F3,) were evaluated for visual appearance, pH, viscosity, spreadability, centrifuge test, anti-microbial activity was determined for prepared anti-microbial gel. All the formulations showed antimicrobial activity. Among these formulations F3 showed a good antimicrobial activity

KEYWORDS: Gel, Soxhlet extraction and aqueous extraction, Anti-Microbial Activity.

I. INTRODUCTION

[1]. Gels are semi-rigid structures with a three-dimensional network of particles or macromolecules that constrain the movement of the dispersed material.The term "gel" is derived from "gelatine," and both "gel" and "jelly" can be traced back to the Latin Gelu for "frost," and gel meaning "freeze," or "congeal." This origin represents the key idea of a liquid transforming into a solid-like material that does not flow but is elastic and retains some liquid properties.In the late 1800s, chemists used the term "gel" to categorize semisolid substances based on their phenomenological features rather than their chemical makeup. At the time, analytical tools for determining chemical structures were insufficient.The USP defines gels (also known as jellies) as semisolid systems that comprise either suspensions of minute inorganic particles or large organic molecules interpenetrated by a liquid. A gel with a network of tiny particles is considered a two-phase system. The gel mass is frequently referred to as magma in a two-phase system when the particle size of the dispersed phase is particularly big. Single-phase gels are made up of organic macromolecules that are uniformly distributed throughout a liquid, with no obvious boundaries between the dispersed macromolecules and the liquid itself.

[2]. Gel's stiffness is due to the presence of a network formed by the crosslinking molecules of gelling agent particles.The links are determined by the cross-linking molecule's features and the type of force, which determines the network's structure and gel properties.Each hydrophilic colloid particle may include a spherical or isometric group of molecules, as well as a single macromolecule.Linear macromolecules are made up of entwined and entangled molecules, having points of contact that may be tiny or structured in a crystalline series.The attraction between gelling agent particles can range from strong primary valencies (such as in silicic acid gels) to weaker hydrogen bonds and vanderwaals forces.A minor increase in temperature

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can cause gel to liquefy, indicating that these forces are weaker.

[3]. Bacterial resistance is a major global health concern that threatens society's health.The advent of resistant infections has led in a decrease in the effectiveness, if not outright ineffectiveness, of existing antibacterial medications, necessitating the creation of new ones.This study highlights two key points: the structural and antibacterial mechanism similarities to older antibiotics have prompted concerns regarding rapid resistance evolution during clinical usage. Secondly, antibacterial medicines are currently in early stages of investigation as potential alternatives.To retain the efficacy of existing treatments in treating common and life-threatening diseases, it's crucial to reduce the growth of resistant bacteria.Understandingbacterial resistance pathways allows for the development of effective treatments.This may involve antibiotic efflux, inactivation, biofilm development, and target change.This may involve antibiotic efflux, inactivation, biofilm development, and target change.

[4].Bacteria can utilize many mechanisms to hide their resistance to antibiotics.Some bacteria are naturally resistant to antimicrobial agents of more than one class.As a result, all strains of that bacterial species are resistant to all antibacterial treatments.The primary concern is the requisite resistance.This happens when sensitive bacteria grow resistant to an antibacterial agent, leading to their proliferation and spread due to selective pressure.Antimicrobial resistance can easily spread to a wide range of bacteria. To begin, an organism may acquire genes that encode enzymes like β-lactamases, which degrade antibacterial medicines before they become effective. Bacteria may develop efflux pumps that remove antibiotic drugs from the cell before they reach their intended target.Bacterial cell walls can be changed to remove antimicrobial drugs' binding sites, or porin genes can be down-regulated, preventing antimicrobial compounds from accessing intracellular targets.Bacteria that are ordinarily vulnerable to antimicrobial agents can become resistant by mutation, selection, or receiving resistance genes from another bacterium.Genetic mechanisms such as transduction, conjugation, and transduction can be used in the final test.

[5].Extracts from commonly used plants in South India, including Allium sativum (garlic), Myridtica fragrans (nutmug), Gingiber officinale (ginger), Allium cepa (onion), and Piper nigrum (pepper), have antibacterial properties. The current study sought to create formulations including several plant extracts with anti-microbial properties.

II. MATERIALS AND METHOD

The herbal compounds utilized; Cayenne pepper (Capsicumannuum), Cinnamon leaves (Cinnamomum verum). List of the chemicals used Carbopol-934(LOBA Chemie laboratory reagents and fine chemicals), EDTA (NR CHEM Mumbai), NaOH (Genuine chemical Co. Mumbai), Propylene glycol (Genuine chemical Co. Mumbai), Triethanolamine (SDFCL. Mumbai), Flavouring agent (orange syrup) (Sisco Research laboratories Pvt. Ltd. Maharastra).

[6].Capsaicin Extraction. Utilizing a soxhlet extraction apparatus, capsaicin was extracted. 20 g of chillies was bought from a local store, they were then deseeded and grinded. Only the flesh was used for the experiment. After setting up the equipment, the extraction was done 20 times at 100^0C. 95% ethanol served as the solvent.

[7].Cinnamaldehyde Extraction. Three times, the leaves were washed in double-distilled water. In a shady spot, the cleaned leaves were allowed to air dry at room temperature. After drying, the leaves were crushed, and the powder was stored for later use. 10g of crushed Cinnamomum verum leaves were soaked in 100ml of double-distilled water for 24 hours, and then they were cooked for 15 minutes at 600C. Muslin cloth was used to filter the filtrate. After the extract had been passed through Whatmann filter paper grade-1. For 15 minutes, the CTAE was centrifuged at 5000 rpm. The leaf extract was stored at 40C in a colored amber glass jar for subsequent use.

The gel preparation is done in three formulations F1, F2, F3. The F1 and F2 formulation contains capsaicin and cinnamaldehyde respectively. F3 contains 50% of capsaicin and 50% of cinnamaldehyde.

[8]. Preparation of Gel. A homogeneous dispersion was achieved by constantly stirring the carbopol in water at 350 rpm after it had been finely distributed. Next, the carbopol mixture was supplemented with ethanol-dissolved capsaicin or cinnamondehyde extract and propylene glycol EDTA. There were five grams total in the extract. To make the gel, NaOH was added to the dispersion.

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Ingredients	F_1	F_2	F_3
Carbopol-934(gm)	1	1	1
Capsaicin(gm)(extract)	5	-	2.5
Cinnamonum verum(gm)(extract)	-	5	2.5
EDTA(gm)	0.02	0.02	0.02
NaOH(gm)	0.11	0.11	0.11
Propylene glycol(ml)	0.5	0.5	0.5
Tri ethanolamine(optional)	q.s	q.s	q.s
Distilled water	q.s	q.s	q.s
Flavouring agent(optional)	q.s	q.s	q.s

Table No:01. Composition of gel formulation

III. EVALUVATION OF HERBAL GEL

1. [9].PhysicalApperance: The physical appearance was simply assessed by color and odor. The uniformity and texture of the produced gel were also assessed
2. [10]. Measurement of Viscosity: It was measured using a Brookfield digital viscometer. To test viscosity, a 5gm prepared gel sample was placed in the sample holder of the B. Viscometer (spindle no.62) and allowed to settle for 5 minutes before rotating at 50rpm.
3. [11]. Determination of pH: The pH of herbal gel was measured using pH paper. 1g of herbal gel was dissolved in 25ml of distilled water. The pH paper was dipped in the solution for a few minutes until a consistent hue was achieved and noted.
4. [12]. Spreadability: Spreadability was measured using a modified setup that included a glass plate block with a pulley at one end. Two conventional glass slides were taken, and a 20gm weight was carefully tied to the upper slide using a hook. Gel formulation (about 1 gram) was applied on the slide. The other slide was placed on top of the gel, sandwiching it between the two slidesin an area that measured 7.5cm down the slide. To ensure a homogeneous gel film between slides, add 20gm of weight on top for 5 minutes to remove air. Excess gel was scraped off the edges.

The top slide was then exposed to a full weight of 20gms using a string linked to the hook, and the time it took to travel a distance of 7.5cm was recorded. The experiment was repeated three times, and the average computation time was shorter. The spreadability improved as the time necessary to separate the two slides decreased. The formula for calculating spreadability was as follows:

$$S=M\times L /T$$

Where, S=Spreadability, M=Weight tied to the upper slide, L=Length moved by the glass slide (7.5cm) and T=Time (in sec.) taken to separate the slide completely each other.

5. [13].Centrifuge Test: Formulations were separately centrifuge in a test tube of 10cm long and 1cm width for 5,10,20 and 30 minutes with 2000rpm and then studied for sedimentation and gel stability pH determination.
6. [14].Extrudability test: Fill a clean, lacquered aluminum collapsible tube with approximately 5gm of gel mixture. Crimp the end of the tube and apply a clamp to prevent roll back. The cap was removed, and gel was extruded through the tip. The extruded gel was collected and weighed and the percentage of gel extruded was calculated and grades were allotted. (>90% Extrudability: Excellent) (>80% Extrudability: Good) (>70% Extrudability: Fair)
7. [15][16].Antimicrobial Activity: The formulation's antibacterial activity was tested using the Kirby-Bauer disk diffusion method. Standard strains of Escherichia coli (ATCC25922) and Staphylococcus aureus (ATCC29213) were employed, with Amikacin serving as a control or comparator. The organisms were inoculated in peptone water, and the turbidity was measured against McFarland standards. This was inoculated as a grass culture onto Muller Hinton agar. The formulation extract (250µl) was impregnated onto a sterile filter paper disc, which was then placed on a commercially available amikacin disc. Both plates were incubated at 37 degrees Celsius for 24 hours. After 24 hours, the plates

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were checked for zone of inhibition, which was measured in mm.

IV. RESULTS AND DISCUSSION

In the present study a new herbal anti-microbial gel developed by using herbal drugs such as capsaicin, cinnamaldehyde, carbopol, propylene glycol, EDTA, NaOH, trietholamine, were used as excipients for preparing the herbal anti microbial gel. The herbal anti-microbial gel is prepared and then prepared anti-microbial gel is evaluated for different parameters.

Sl. No	Evaluation parameter	Observation		
		F1	F2	F3
1	Colour	Creamish white		
2	Odour	Orange odour		
3	Texture	Smooth		

Table No 02: Organoleptic Evaluation of gel formulation

Sl.No	Parameters	F1	F2	F3
1	pH	6.8	6.7	6.9
2	Viscosity(cps)	11850	11990	11910
3	Spreadability (cm/sec)	31.44	32.39	76.92

Table No 03: Evaluation Results of gel formulation

*All the parameters have been done in triplicate

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Sl.No	Specimen	F1 (mm)	F2 (mm)	F3 (mm)	STANDARD (mm)
1	*E. Coli*	12	14	18	18
2	*S. Aureus*	10	11	15	16

Table No 04: Anti-microbial Activity

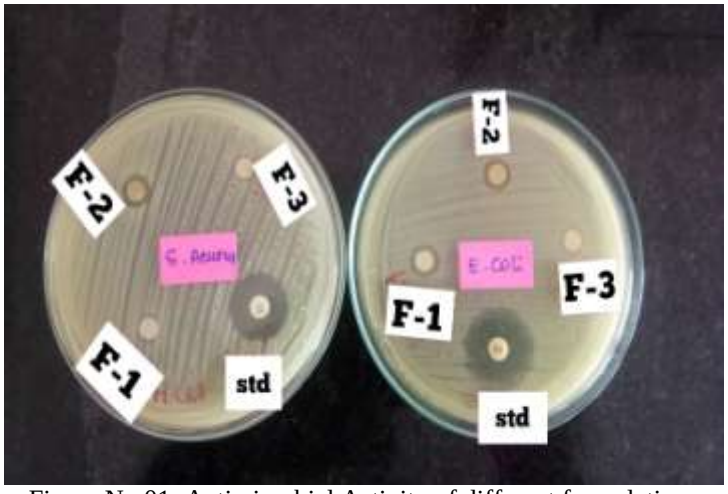

Figure No 01: Antimicrobial Activity of different formulations

- In this present study, our aim is developing a typical herbal anti-microbial gel formulation by using different herbal drugs. The present study makes use of potential herbs like: Cayenne pepper, Cinnamomum verum and excipients like carbopol-934, orange flavor, tri-ethanolamine formulation hence avoiding some kind of synthetic particulates will definitely reduce the side effects. Herbal active ingredients show lesser side effects on skin comparing to the synthetic components.
- The present study makes use of multiple herbal extract.Each individual has its own therapeutic activity of microbes. Capsaicin and cinnamaldehyde has potential effect in defeating against microbes as well as capsaicin shows anti-microbial and antioxidant activity. The orange flavor is presence in formulation which gives good smell upon application.
- In the present study not only the herbs but along with those some of synthetic excipients are also used to develop a typical Anti-microbial gel formulation since for preventing microorganism growth, for these purposes we need synthetic excipients like NaOH, EDTA, etc The some ingredients were collected from the local market and plant source were collected and they were washed thoroughly with double distilled water and were dried under room temperature in shade and These herbal ingredients were further crushed and powdered further used for the herbal extraction with soxhlet method and aqueous extraction method.
- Three different formulations were developed using different concentrations in which each formulations have a fine consistency, good appearance and a smooth texture. These developed formulations were evaluated for various parameters such as pH, viscosity, Spreadability, homogeneity etc. All the formulation shows good and acceptable results for all the evaluation parameters and good

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spreadability, as well as homogeneity along with it as optimum viscosity.

- pH of the prepared formulations F1, F2 were found to be 6.8 and 6.7 respectively. F3 were found to be 6.9 in which this pH is compatible to formulate typical anti-microbial gel formulation. So the extent of pH will not going to affect the condition of a skin.
- Viscosity of prepared formulations F1 and F2 were found to be 11850cps and 11990cps respectively, F3 was found at 11910cps range at 50rpm. This shows a good consistency of gel. This kind of consistency is compatible for easy to apply on skin. This will enhance the contact time of formulation with skin. The carbopol used in the formulation will for made finite consistency and viscosity of formulation.
- Spreadability of the formulation F1 and F2 were found to be 31.44 and 32.39cm/sec respectively. F3 was found to be 76.92cm/sec. Spreadability of formulation was defined by how extents can a formulation spread upon gentle pressure. The present formulation has a moderate spreadability which upon application of simple pressure it spreads to good extent.
- Homogeneity of the all formulation was found to be good. This test was carried out to evaluate the even particle size present in the formulation. By this test found that there are no clumps of particulates in formulation since it has the good and smooth consistency. All the herbal extracts were grinded thoroughly to get fine powders, the homogeneity of anti-microbial gel was found to be good.
- Formulation in case of pH, viscosity, spreadability, anti-microbial gel activity since the results are nearer to standard product the conclusion can be given that the herbal ingredients can safely incorporated into anti-microbial gel formulation as comparing to synthetic products these are good enough. By comparing all the Formulations F3 shows good spreadability, viscosity, homogeneity and optimum pH with effective anti-microbial activity.
- The therapeutic effects produced by herbal ingredients are optimum for example anti-microbial effect by capsaicin and cinnamaldehyde extract. By conducting and comparing all the evaluation parameters with marketed product shows it's better to use of herbal formulations in the treatment of microbial infections.
- From this study we can conclude that the incorporation of herbal ingredients in anti-microbial gel formulation is better way of finding solution for the microbes. In order to overcome these defects with minimal side effects the herbal anti-microbial gel approach is best option.

CONCLUSION

Herbal anti-microbial gel formulations can be prepared by using different plant sources with varying concentrations.The pH of the herbal gel formulations were in the range of 6 to 7 which lies in the normal pH range of the skin. All the herbal gel formulations showed good viscosity and were in the standard range and they were capable to remain in the site of application for prolonged time. All the gel formulations showed good spreadability. Among these formulations F3 shows excellent spreadability. The values of spreadability indicate that the gel was easily spreadable by small amount of shear. The gel formulation of F1 F2 and F3 was performed for anti-microbial potential. Among these formulations F3 has shown the best result for anti-microbial activity.

SUMMARY

The present work of Development and evaluation of herbal anti-microbial work was carried out using various herbal extracts in which each individual herbal ingredients are responsible for individual therapeutic effect on microbes.The present anti-microbial formulation was developed along with some efficient adjuvants suitable for anti-microbial formulation.The aim of present work was to formulate and evaluate gel containing Capsaicin and Cinnamaldehyde extract and study its anti-microbial activity.Anti-microbial gels were prepared using carbopol-934 as gelling agent Triethanolamine, NaOH, and distilled water as other additives.All the herbal anti-microbial gel formulations showed good viscosity and were in the standard range.The values of spreadability indicate that the gel was easily spreadable by small amount of shear.The gel formulations of F1, F2, F3 were performed for anti-microbial activity. Among these formulations F3 has showed the best results for anti-microbial activity when compared to the standard.

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