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Design and Evaluation of Allium Sativum Gel

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Abstract:- Garlic and its preparations have been widely recognized as agents for prevention and treatment of cardiovascular and other metabolic diseases such as, atherosclerosis, hyperlipidemia, thrombosis, hypertension and diabetes. It is also effective against bacterial as well as fungal infections. Garlic oil obtained by cold maceration. Garlic has different active constituents among which ajoene shows antibacterial and antifungal activity. Ajoene is highly unstable it remains stable only in oil macerate form. It is a great challenge for scientists all over the world to make a proper use of garlic and enjoy its maximum beneficial effect as it is the cheapest way to prevent bacterial as well as fungal infection so, an attempt to form topical gel from garlic oil macerate for its antibacterial as well as antifungal activity and evaluation.

Keywords:- *ajoene, cold maceration, evaluation, garlic oil macerate.*

I. INTRODUCTION

Garlic is used for different purposes such as a palliative for the heat of the sun in field labor, preventing heart disease (including atherosclerosis, high cholesterol, and high blood pressure), cancer and cardiovascular benefits of garlic. It is also alleged to help regulate blood sugar blood sugar levels and also shows antimicrobial activity. In modern naturopathy, garlic is used as a treatment for intestinal worms and other intestinal parasites, both orally and as an anal suppository. Garlic cloves are used as a remedy for infectious (especially chest problems), digestive disorders, and fungal infections such as thrush, candidiasis, aspergillosis and cryptococcosis. (1),(2),(3) The garlic is mention as remedy for skin diseases in Ayurveda & it is chosen for antibacterial activity because every antibiotic there is development of resistance by bacteria but in case of garlic there is no resistance developed against garlic. When garlic chopped or crushed, allinase enzyme, present in garlic, is activated and acts on alliin (present in intact garlic) to produce allicin. The enzyme allinase responsible for converting alliin (s-allyl cysteine sulphoxide) to alliicin is inactivated by heat.(4) Allicin is a volatile and short lived (hours or days) compound, which will break down in to compounds, such as diallyl disulphide. In a matter of hours it will further degrade in to an oily withes brew of bisulphides, trisulphides such as allyl methyl trisulphide and vinyldithiins and polysulphides and many others. Allicin is powerful natural antibiotic (about one-fiftieth as powerful as penicillin and one-tenth as powerful as tetracycline) that kill many kind of bacteria (including bacillus, escherischia (e coli), mycobacterium, pseudomonas, staphylococcus an streptococcus). It also has anti-fungal and anti-viral properties.(4),(5) There are different garlic formulations in the market such as oil macerates of fresh garlic formulated commonly in soft gelatin capsules, the dry powder products

of fresh garlic formulated either as sugar or film coated tablets in European market and dry powder products of aged garlic formulated either as sugar or film coated tablets in American market. In India not much work done on garlic oil macerate so it is used for its combined effects against all bacteria. The macerated garlic oil contains the vinyldithinins and ajoenes. The antifungal activity of ajoenes was reported against the Candida albicans and Aspergillus niger.(6),(7) Ajoene remain stable for long period only in form of garlic oil macerate. First garlic oil macerate obtained by Garlic is crushed first and then remain for half an hour exposed to air, so that there is formation of the allicin from alliin by enzyme allicilnase(8),(9) and then allow to macerate for seven to eight days. Maceration is done with two different oils ie sesame and sunflower oil. The two different strains of garlic are used for maceration first single clove and multiple clove garlic. After maceration separate the macerated oil from crushed garlic by filtration through suction pump. After filtration the crushed garlic taken on piece of cloth and all oil content is removed by applying pressure. The concentrated part is also removed along with oil so, filter this oil though suction pump to obtained clear oil macerate. The topical gel using different gelling agents are prepared along with garlic oil macerate.

II. EXPERIMENTAL PROCEDURE

A. Chemicals and reagents

The chemicals and regents along with grade are as listed in table 1.

B. Instruments

Instruments used are as follows electronic balance from Shimdzu, Autoclave from Medica Instrument Pvt. Ltd, Magnetic stirrer from Bio lab, pH meter of Lab India and suction pump from Besto Pvt. Ltd.

C. Maceration procedure

Maceration was done with two different oils i.e. sesame oil and sunflower oil. The two different strains of garlic are used for maceration first single clove and multiple clove garlic. Maceration is the process of extracting drug constituent in to particular solvent. In case of garlic, 500 g was weighed and then crushed with the help of mortar pestle or grinder. The crushed garlic was exposed to environment for 10 to 15 minutes so that there is conversion of alliin to allicin by enzyme allinase. Then 25 ml of oil was poured in crushed garlic and mixed with help of stirrer. Then the container was wrapped with aluminum foil and kept for seven days to macerate. After seven days, the oil was separated from crushed garlic by filtration using suction pump. The crushed garlic was placed on the piece of cloth and oil traces were removed by applying pressure. The concentrated part is also removed along with oil so as to obtain a clear oil macerate. The same procedure was repeated for both types of oils and both varieties of garlic.

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The choice of oils was based on the fact that sesame oil is the most common form of oil used in Ayurveda (including garlic products) as it is considered to have the best medicinal properties. Soyabean oil was selected on the basis of availability, cost and also because physicochemical stability.

D. Different batches for Gels

The method of preparation for each batch as follows

a) Method Batch 1

First weigh the 2g of CMC then mix it with help of magnetic stirrer in 100ml of water. After this break the clumps that are formed during mixing with the help of glass rod. Then add triethanolamine to adjust pH to neutralization point. Check the pH with help of pH paper. Add garlic oil macerate (2.0ml), preservative methyl paraben 0.2, propyl paraben 0.02g and eucalyptus oil to mask the odor of GOM. Kept it for overnight stability.

b) Batch 2

First weigh 1 gm of carbopol 940 then mix it with help of magnetic stirrer in 50ml of water. After this break the clumps that are formed during mixing with the help of glass rod. Then add triethanolamine to adjust pH to neutralization point. Check the pH with help of pH paper. Add tween 80 as a surfactant then GOM and preservatives i.e methyl paraben and propyl paraben. Mask the odor by adding sufficient quantity of camphor to formulation.

III. RESULTS

A. Different trials for Gels

Sr No	Ingredients	Batch 1	Batch 2	
1	GOM	1.0	1.0	
2	СМС	0.5	-	
3	TEA	0.25	1.2	
4	Tween 80	-	1.0	
5	Carbopol 940	-	1.0	
6	Methyl Paraben	0.1	0.2	
7	Propyl Paraben	0.01	0.02	
8	Water	50	50	
GOM : Garlic oil macetrate, CMC: CarboxyMethyl Cellulose,TEA: Triethanolamine				

Table 1: Formulation of batch

- **Batch 2:** Observation: In trial first the gel formed with 1.0% CMC was found very low in viscosity and consistency, so we moved to next trial by lowering the water quantity.
- **Batch 2:** Observation: gel formed with carbopol 940 was found to be of the best viscosity and consistency among all

the gelling agents. The gel formed was also found to be stable.

• **Result:** Among all the trials, gel formed with 2% carbopol 940 was found to be stable and with good consistency, so further evaluation of this gel was performed using different evaluation parameters such as pH, Viscosity and antifungal assay.

B. Evaluations of gels

Batch	рН	Homogeneity	Skin irritation test	Spreadability	
Batch 1	5.4	Uniformly homogenous	Nil	1.2 min	
Batch 2	7.2	Uniformly homogenous	Nil	26 Sec	

Table 2

• pH

The pH was determined using digital pH meter of Lab India.

• Homogeneity

Homogeneity was calculated by visual inspection.

• Skin irritation test

Skin irritation test was performed on five healthy volunteers by applying gel on back side of the hand and covering it with a suitable material and retaining it for 24 hours. After 24 hours, the reports about skin irritation were collected from the concerned volunteers.

• Spreadability

Spreadability was performed using two glass slides, one of which is fixed and the other is movable.

• Antifungal assay

Antifungal assay was performed only for ointment trial 3 and gel trial 6 because from above results, it was concluded that only these products comply with all tests. The culture used for antifungal assay was candida albicans 3471. The medium used for culture growth was potato dextrose agar from Himedia Laboratories. Agar diffusion method was used for antifungal assay.

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- Cultures used: Fungi: Candida Albicans 3471
- Media used:
 For Fungi: Potato Dextroso
- For Fungi: Potato Dextrose Agar (Hi-Media) Inoculum size:
- Fungi: 1 X106 Cells Per Ml
- Concentration of Compound: 100 Mg/Disc (Prepared In DMSO)
- Method used: Agar diffusion assay (well method, well size 6 mm)
- Dilution of drug: Stock prepared 1mg per ml prepared in DMSO [100 μg/disc]
- G. Results:

Results are as shown in table number 3.

Cmpound	Candida Albicans 3471			
DMSO (Control)				
BATCH 1	2.34			
BATCH 2	11.12			
Nystatin	22.1			
Table 3				

The zone of inhibition of Batch 2 gel was found to be 10.12 However, the gel was shown to exhibit anti-fungal activity, comparable to a standard anti-fungal drug- Nystatin because of presence of allicinase enzyme extracted in oil macerate.

Conclusion: There were no studies with macerated oils of *Allium Sativum*. Gel containing the oil macerate extract of Allium sativum showed good antifungal activity. The activity observed may be due to presence of Ajoene constituent in it.

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