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Preparation and Evaluation of Herbal Antiseptic Ointment

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Abstract: *Even in areas where modern Synthetic medicines are available, the interest towards herbal medicines and their utilization have been increasing rapidly in recent years. Plant derived substances and herbal drugs have now a day's attract the peoples and they have great interest towards their versatile application, as medicinal plants are the richest source of bioactive compounds used in traditional and modern medicine [4]. The present work is to formulate and evaluate the ointment of Neem (Azadirachta indica), Turmeric (Curcuma longa) extract and clove oil (Eugenia Caryophyllata). An ointment is a viscous semisolid preparation and it used topically on a variety of body surface. The objective was to formulate and evaluate the antiseptic herbal ointment from local medicinal plants. The ethanolic extracts were prepared and collected by using maceration method. The ointment base was prepared and formulation of ointment was done by incorporating the extract & clove oil in the base by using levigation method. After completion of formulation it was evaluated for its physicochemical parameters like colour, odour, pH, extrudability, consistency, diffusion study, solubility, washability etc.*

Key Words: *- Herbal ointment, Maceration, Levigation, Extrudability, Spreadability.*

1. INTRODUCTION:

Nature has provided source of medicinal compounds from bygone days. The basic herbs have the answer with no side effects and effective remedies and the golden fact is use of herbal treatment is not dependent of any age group. When two or more herbs are used in the formulation they are known as polyherbal formulations. Traditional medicine is an important development of chemotherapeutic agents. The first step towards this goal is the screening of medicinal plants used in popular medicine. Thus antimicrobial research is geared towards the discovery and development of novel antibacterial and antifungal agents. Plant drugs are less toxic and free from side effects than the synthetic ones. Along with other dosage forms, herbal drugs are formulated in the form of ointment. An ointment is a viscous semisolid preparation and it is used topically on a variety of body surfaces. These include the skin and also the mucus membranes of the eye, nose, vagina and anus. An ointment may or may not be medicated. Medicated ointments contain a medicament dissolved, suspended or emulsified in the base. Ointments are used topically for several purposes Numerous studies have been conducted with the extracts of Neem leaves (Azadirachta indica; Family-Meliaceae) and extract of turmeric rhizomes (Curcuma longa; Family-Zingiberaceae) with the combination of many other herbal drugs.

2. LITERATURE REVIEW:

Abhijeet Pandey ,et.al [7] (2010) In the present study, herbal ointment containing Aloe vera, Neem and Turmeric was formulated and evaluated to study antibacterial and antifungal activity. The evaluation is done using cup plate method for zone of inhibition and two fold dilution method for MIC (Minimum Inhibitory Concentration).The study showed that Aloe ointment is exhibiting broad-spectrum antifungal activity against A. varies and antibacterial activity against E.coli. The overall experiment showed that Aloe ointment and Turmeric ointment showed more antifungal activity than Neem ointment.

P.J. Rajasree, et.al [8] (2012) The main objective of the present study is to formulate and evaluate a poly herbal ointment with antiseptic activity. Ointments were formulated using methanolic extracts (by continuous hot percolation-soxhletation) of Azadirachta indica, Chromolaena odorata, Mimosa pudica, Samadera indica and were evaluated for its physicochemical property, antibacterial and antioxidant activity. Ointments were prepared using different concentrations of the extracts such as 2%, 4%, 6% w/w by fusion method using emulsifying ointment as base. Formulations were then tested for its physicochemical properties like loss of drying, pH, spreadability, extrudability and diffusion study and gave satisfactory results.

Shubhangi E. Sawant and Monali D.Tajaneb[4] (2014) The present work is to formulate and evaluate the ointment of Neem (Azadirachta indica) and Turmeric (Curcuma longa) extract. The ethanolic extracts were prepared by using maceration method. The ointment base was prepared and formulation of ointment was done by incorporating the extract in the base by levigation method. After completion of formulation it was evaluated for its physicochemical parameters like colour, odour, pH, spreadability, extrudability, consistency, diffusion study, solubility, washability. Also the formulation was evaluated for its stability at various temperature conditions which shows no change in the irritancy, spreadability and diffusion study.

3. OBJECTIVES:

- To reduce side effects of synthetic formulation.
- To reduce the cost of antiseptic products.
- To avoid irritation of synthetic chemicals to the skin.
- To formulate natural ointment formulation.
- To evaluate herbal antiseptic ointment.

4. MATERIALS AND METHODS:

4.1 Preparation of Neem extract [4]:

Leaves of the Neem plant were collected and washed thoroughly with distilled water and shade dried for 10 days. Dried leaves were ground into powder form. From that 100 gm powder was imbibed with 350ml of 90% Ethanol for 3hrs and transferred to percolator with addition of 150ml of 90% ethanol for maceration process for 7 days with occasional stirring. Finally ethanolic extract of Neem was collected and then concentrated to get blackish green residue. The extract was stored in the airtight container at cool and dark place.

4.2. Preparation of Turmeric extract [4]:

Dried rhizomes of turmeric were ground and the powder obtained was used for extraction. 100gm powder was imbibed with 350ml of 90% ethanol for 3hrs. And then transferred to percolator with addition of 150ml of 90% ethanol for maceration process for 7 days with occasional stirring. Finally ethanolic extract of turmeric was collected and concentrated to get crimson red colour residue. The extract was stored at cool and dark place in air tight container.

4.3. Isolation of clove oil :

Take 50 to 60 g of clove drug with 500 to 600 ml of water in distillation flask. Add a few pieces of porcelain to it (to avoid bumping drugs distillation). Set up the apparatus and fill the tubes (receiver and return tube) with water by introducing it at side tube by means of a pipette. Close the side tube. For heating the flask-heating mantle is used. Lift the flask at intervals and then shake the contents, until the liquid is boiling steadily. Finally adjust the temperature so that the distillate in the graduated tube remains cold. Continue heating till no more oil collects. (This requires two hours or more). Turn off heating mantle and allow the liquid in the condenser to drain for five or ten minutes then read the volume of oil [2].

Figure 1: Extract of Neem, Turmeric & Clove

Formulation of ointment:

I. Formulation of ointment base:

Table 1. List of Ingredients required for ointment base

Sr. No.	Name of ingredients	Quantity to be taken
1.	Wool fat	1 gm
2.	Cetostearyl alcohol	1gm
3.	Hard paraffin	1gm
4.	Yellow soft paraffin	16.64gm

Table 2. List of Ingredients required for ointment preparation

Sr. No.	Name of ingredients	Quantity to be taken
1.	Prepared Neem extract	0.12gm
2.	Prepared turmeric extract	0.12gm
3.	Clove oil	0.12 ml
4.	Ointment base	19.64 gm

Initially ointment base was prepared by weighing accurately grated hard paraffin which was placed in evaporating china dish on water bath. After melting of hard paraffin remaining Yellow soft paraffin, Wool fat and Cetostearyl alcohol were added and stirred gently to aid melting and mixing homogeneously followed by cooling of ointment base. Herbal ointment was prepared by mixing accurately weighed Neem, Turmeric and clove oil extract to the ointment base by levigation method to prepare a smooth paste with 2 or 3 times its weight of ointment base, gradually incorporating more bases until to form homogeneous ointment, finally transferred in a suitable container [4].

Figure 2.-Formulation of ointment

Evaluation of antimicrobial activity:

Preparation of medium and nutrient broth: Weighed about 1.3gm of nutrient broth and dissolved in 100 ml of water. Then broth was suspended in each test tube. The agar plates was prepared which contain 3gm of nutrient agar was suspended in 100 ml of water. The both the medium and broth were kept for sterilization. After sterilization, the nutrient broth was allowed to cool and then organism were inoculated and incubated for 24 hours. The prepared nutrient agar were poured in the Petri dish before cooling and allowed to solidify for 20-30 min under the laminar air flow.

4.4. Methodology: The bacterial culture was spread on the culture medium and a well bored in the middle of the agar. Then samples were poured inside the wells and plates were incubated at 37^0C overnight for observation. The presence of inhibition was noted. The susceptibility of the test organism to tested plants extracts was determined by observing the zone of inhibition around the each well.

5. RESULT & DISCUSSION:

Present study was done to prepare and evaluate the herbal antiseptic ointment. For this the herbal drugs extracts were prepared by using simple maceration process to obtain a good yield of extract and then evaporated to get concentrated. There was no any harm to the chemical constituents and their activity.The levigation method was used to prepare ointment so that uniform mixing of the herbal extract with the ointment base was occurred which was stable during the storage. The physicochemical properties were studied which shows satisfactory results for extrudability, spreadability, solubility, washability, loss on drying and others. Also the formulation was placed for a stability study at different temperature conditions like 2^0C, 25^0C and 37^0C within four weeks. There were no changes observed in ointment herbal formulation.

Antimicrobial activity: The ethanolic extracts of Neem and Turmeric, isolated clove oil and antiseptic ointment were evaluated for their antimicrobial activity on Bacillus subtilis, streptococcus, pseudomonas, Klebsiella pneumoniae & fungi were used as test organism. Each test organism responds in varied manner to ethanolic extracts, antiseptic ointment and clove oil under study. The response of each test organism was observed after proper incubation period. The response of each organism is given below-

Antimicrobial activity of Neem, Turmeric, clove oil and herbal ointment on Gram Positive bacteria's- Bacillus Subsitis and Streptococcus:

Antimicrobial activity on Bacillus Subsitis **Antimicrobial activity on Streptococcus**

Antimicrobial activity of Neem ,Turmeric, Clove oil and herbal ointment on Gram Negative bacteria Pseudomonas and Antifungal activity on Klebsiella:

Antimicrobial activity on Pseudomonas **Antifungal activity on Klebsiella**

Zone of inhibition:

Table 3. Zone of inhibition

Microbes / Sample	Bacillus	Streptococcus	Pseudomonas	Klebsiella
Neem	3 cm	2.8 cm	2.3 cm	1.5 cm
Turmeric	2.6 cm	2.2 cm	2 cm	1.9 cm
Clove oil	3.5 cm	3 cm	2.5 cm	2 cm
Antiseptic ointment	4.2 cm	3.5 cm	3.3 cm	2.8 cm

Physicochemical Evaluation of Formulated Ointment

Table 4. - Physicochemical Evaluation of Formulated Ointment

Sr.No.	Physicochemical parameters	Observation
1.	Colour	Green
2.	Odour	Characteristic
3.	Consistency	Smooth
4.	PH	5.6
5.	Spreadability (second)	8
6.	Extrudability	0.4 gm
7.	Diffusion study (after 60 min)	0.7 cm
8.	Loss of drying	30%
9.	Solubility	Soluble in boiling Water, miscible With alcohol, ether, chloroform
10.	Washability	Good
11.	Non irritancy	Non irritant
12.	Stability study (2^0C,25^0C,37^0C)	Stable

5. CONCLUSION:

From the ancient time Neem, Turmeric and Clove oil is used for their various medical properties like antibacterial, antifungal, anti inflammatory, etc. The purpose of study was to prepare antiseptic herbal ointment using locally available plant. On the basis of antimicrobial efficacy, three different local plants were taken and their ethanolic extracts were incorporated in the ointment base. The final product radially spread on skin surface, showed no irritant effect, diffused well and was stable at different temperature. Thus this ointment could become media to use these medicinal properties effectively and easily as a simple dosage form.

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