

Hydrotopic Cream Preparation from Licorice Extract

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How to cite this paper: Ms. Chetana D. Patil | Ms. Rutika U. More | Mrs. Pooja S. Bhandare "Hydrotopic Cream Preparation from Licorice Extract" Published in International Journal of Trend in Scientific Research and Development (ijtsrd), ISSN: 2456-6470, Volume-3 | Issue-5, August 2019, pp.707-710, <https://doi.org/10.31142/ijtsrd26435>



ABSTRACT

The current work aimed to formulate a stable w/o cream containing Glycyrrhiza glabra extract and studying its effects on skin pigment. Glycyrrhiza glabra Extract obtained by concentrating the alcoholic extract of Glycyrrhiza glabra roots was entrapped in the inner aqueous phase of W/O emulsion. Base containing no active material and a formulation containing ethanolic extract of Glycyrrhiza glabra (1%) were prepared. Samples of base and formulation were stored at different accelerated conditions (8°C, 25°C, 40°C, 40°C+75%RH) for four weeks to predict the stability of creams. Base and formulation were stable at all accelerated conditions regarding color, liquefaction and phase separation. Both creams were good for sensory evaluation.

KEYWORDS: Glycyrrhiza glabra extract, W/O cream, Evaluation test and Antimicrobial activity

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INTRODUCTION

The word **Glycyrrhiza** has been derived from the Greek origin that means *sweet root*; and **glabra** means *smooth* and usually refers to the smooth, pod-like fruit of this particular species. Nevertheless, the fruits of the *glandulifera* variety have a distinct gland like swellings An extract of Glycyrrhiza glabra is rich with natural antioxidants. The best natural antioxidants in the extract of Glycyrrhiza glabra are glycyrrhizin (glycyrrhizic acid) and flavonoids[5]. Glycyrrhiza glabra extract is obtained from the roots of Glycyrrhiza glabra by solvent extraction[6] method and then concentrating the extract[7] by rotary evaporator.

Glycyrrhiza glabra extract is preserved by refrigeration and/or freezing The role of Glycyrrhiza glabra extract on the skin is mainly attributed to its antioxidant activity particularly to its potent antioxidants triterpene saponins and flavonoids[5].Skin whitening[8], skin depigmenting[9],skin lightening[10-11], antiageing, anti-erythemic[12], emollient[13],anti-acne[14-15] and photoprotection effects are mainly attributed to Glycyrrhiza glabra extract. Glycyrrhiza glabra extract is incorporated in the internal aqueous phase of W/O emulsion. The aim of this study was to formulate a stable W/O cream and to measure the effects of this W/O cream of Glycyrrhiza glabra extract on different physiologic functions of skin particularly skin melanin as well as Erythema, skin moisture, skin sebum, pH of human skin and Transepidermal water loss (TEWL).



Figure No:-1- Whole Plant of Licorice

MATERIALS AND METHODS

Paraffin oil and Coconut oil, Beeswax, Glycerin, Lemon oil, Sodium lauryl sulfate. Extract of glycyrrhiza glabra (ethanolic) was prepared in the laboratory

Preparation of Extract of Glycyrrhiza glabra:-

Glycyrrhiza glabra roots were used as plant material. The Shade dried roots of Glycyrrhiza glabra(Family: Fabaceae)

were taken, cleaned and ground in an electric grinder to have a coarse powder. The powder was then passed through sieve number 60 and stored in a well-closed container at 25°C. One kg powdered material of Glycyrrhiza glabra roots was introduced into a glass flask and analytical grade ethanol was added, soaked for three days and kept in the laboratory. The flask was shaken for 10 minutes after each day for three days. Finally the soaked material of plant was filtered through several layers of muslin cloth one by one coarse filtration. The filtrate so obtained was evaporated under reduced pressure at 45 °C in a Rotary vacuum evaporator. The process of evaporation was continued until the little amount of alcohol remained. Then the same procedure of soaking, filtering and evaporating was repeated with the remaining material in the flask for two times more. Then, the final concentrate of three successive extractions was combined and again concentrated on Rotary vacuum evaporator under reduced pressure at 45°C. The syrupy extract so obtained was collected in glass containers and stored in the freezer at 0°C.

Preparation of base and formulation:-

W/O creams were prepared by the addition of the aqueous phase to the oily phase with continuous agitation[16]. The base was prepared by heating paraffin oil, bees wax, coconut oil and surfactant(sodium lauryl sulfate) up to 75°C±1°C. At the same time, the aqueous phase consisting of glycerin and water was heated to the same temperature and then the aqueous phase was added to the oil phase drop by drop. Stirring was continued by the mechanical mixer for about 10 minutes until the complete aqueous phase was added. After the complete edition of the aqueous phase, the speed of the mixer was reduced for homogenization. Then triethanolamine was added and homogenization was carried out for a period of 5 minutes until the formed cream cooled to room temperature[16]. The formulation was also prepared by the same method; the only difference was the addition of Glycyrrhiza glabra extract (active drug) that was added in the aqueous phase consisting of glycerin and water. Each formulation consists of preserved water (propyl paraben, methyl paraben) .the formulations were neutralized by triethanolamine to Ph=5.5.

Stability Tests:

Physical analysis, types of emulsion, pH determination, electrical conductivity and centrifugation tests of creams were analyzed to assure the formulation of desired properties. Stability tests were performed at different conditions for emulsions to note the effect of these conditions on the storage of creams. These tests were performed on samples kept at 80C ± 0.10C (in refrigerator), 250C ± 0.10C (in oven), 400C ± 0.10C (in oven) and 400C ± 0.10C(in oven) with 75% relative humidity (RH).The physical characteristic of creams, i.e. color, creaming and liquefaction were noted at various intervals for 28 days.

Patch Test:

On the first day of skin testing, patch tests were performed on the forearms of each volunteer. 5cm X 4cm regions were marked on both the forearms. 1.0 g of base and formulation each were applied to the 5cm X 4cm marked regions separately on each forearm. The regions were covered with the surgical dressing after application. After 24 hours, dressings were removed and the measurements of erythema and melanin were repeated on both forearms.

EVALUATION TEST OF FORMULATION

A. Centrifugation test:

for Creams In this study centrifugation test was performed for both the base and formulation kept at different storage conditions up to a period of 28 days at different time intervals. No phase separation on centrifugation was seen in any of the samples kept at different storage conditions i.e. 80C, 250C, 400C and 400C+ 75% relative humidity up to 28th day of observation. This indicated both creams were stable at all the storage conditions for 28 days. It is evident that proper homogenization speed during emulsion formulation prevented the base and formulation breakage during stress condition

B. Electrical Conductivity Test:

In this study conductivity test was performed for both base and formulation kept at different storage conditions up to a period of 28 days at different time intervals. No electrical conductivity was seen in any of the samples of base and formulation kept at different storage conditions i.e. 80C, 250C, 400C and 400C+ 75% relative humidity up to 28th day of observation. This indicated both the creams were stable at all the storage conditions for 28 days.

C. Stability:

In this work, both base and formulation were divided into four samples separately and these samples were kept at different storage conditions i.e. at 80C in the refrigerator, at 250C, 400C and at 400C + 75% relative humidity (RH) instability chambers. These samples at different storage conditions were observed for a period of 28 days at different intervals. Samples were observed with respect to change in color, liquefaction and phase separation

ANTIMICROBIAL ACTIVITY

PROCEDURE -

100 mg of powder sample was dissolved in 0.1 ml of respective solvent Then 0.1 ml of bacterial inoculums was introduced on prepared cultured media. Then it was spread with the help of glass spreader. The bore was created on culture media with the help of cork borer. The extract was introduced on that bore. The plate was kept on the deep refrigerator for 1 hr. for penetration and then kept in incubator for incubation for 48 hrs at 37°C. The zone of inhibition was observed and measured.

RESULT AND DISCUSSION:

1. Colour:

The freshly prepared base was creamy white while formulation was pale yellow in color (due to the presence of Glycyrrhiza glabra extract). There was no change in color of any sample of base and formulation at different storage condition i.e. 80C, 250C, 40 0C and at 400C+ 75% relative humidity up to the observation period of 28 days. Polyphenols which are a present extract of Glycyrrhiza glabra have microbicide activities against huge no of bacteria.

2. Liquefaction:

No liquefaction was observed in any of the samples of base and formulation kept at 80C and 250C during the whole observation period of 28 days but slight liquefaction was observed in samples kept at 400C and 400C + 75% RH from 21st day of observation but there was no increase in liquefaction till the end of the study period.

3. Phase Separation:

No phase separation was observed in any of samples of base and formulation kept at 80C, 250C, 400C and 400C+ 75% relative humidity up to observation period of 28 days. This indicated both base and formulation were stable at all the storage conditions for 28 days.

4. Ph:

The pH is a significant parameter so far as the effectiveness of the cream is concerned and it can be used as an indicator of emulsion stability. For the formation of stable emulsions pH value of aqueous phase is the key factor. pH of skin ranges between 5 and 6, and 5.5 is considered to be the average pH of the skin. Therefore, the formulations intended for application to skin should have pH closer to this range. In this study, the pH of freshly prepared base and formulation was 5.51 and 5.34 respectively, which is very close to the skin pH. The pH values of the samples of base kept at different storage conditions i.e. 80C, 250C, 400C and 400C + 75% RH was found to be decreased gradually from the first day to the last day & on 28th day pH of base was 5.33, 5.31, 5.33 and 5.24 respectively, whereas samples of formulation were kept at different storage conditions i.e. 80C, 250C, 400C and 400C + 75% RH was 4.40, 4.80, 4.50 and 4.99 respectively.

5. Patch test:

In this study patch tests were performed on forearms of volunteers for 24 hours for both the formulation and base, to check the safety of the formulation and the base on human skin. It was found that erythema level was decreased after the application of base and formulation by the end of 24 hours and no skin irritation was produced by the two creams after 24 hours. It is concluded that the formulation and the base produced no skin irritation after performing patch test of 24 hours. It may be attributed to the presence of a good emollient glycerin in the base and formulation, and/or Glycyrrhiza glabra natural antioxidant in an extract of glycyrrhiza glabra, in the formulation, which has the ability to reduce skin erythema.

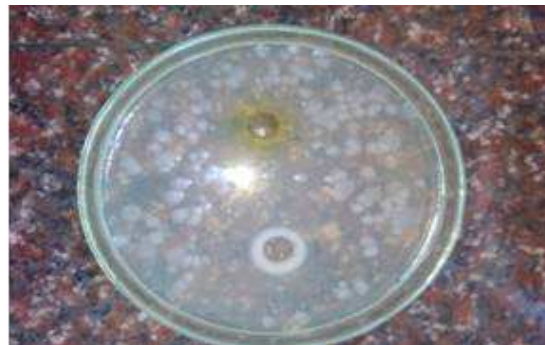
EVALUATION OF CREAM:

Sr. no	Parameter	Observation
1	Colour	Yellowish brown
2	Odour	Sweet
3	Taste	Sweet

ANTIMICROBIAL ACTIVITY



E.coli



Staphylococcus aureus



Bacillus

DISCUSSION:

The current work aimed to formulate stable water in containing glycyrrhiza glabra extract and studying its effect on skin pigment. No phase separation was seen in any of sample of base and formulation kept at 8C,25C and 30C but there was a little bit of separation in samples kept at 40C+75%RH from the 21 days of observation. This indicated both creams were stable at 8C,25C and 30C. In the present, both base and formulation did not show any change in color of samples at different storage condition up to the observation period of 28 days. G. was introduced extract containing polyphenols that have antimicrobial activities and thus prevent a color change of the formulation. In addition, no liquefaction was observed in any of the samples of the base and formulation kept at 8C,25C and 30C. However slight liquefaction was observed in samples kept at 40C and 40C+RH after 3 weeks of observation, but there was no increase in liquefaction till the end of the study period. Thus, the above findings (color, liquefaction and phase separation) indicated that both base and formulation were stable at 8C,25C and 30C storage conditions for 28days.

pH is a significant parameter of the effectiveness of cream stability. Skin pH range between 5-6 and 5.5. is considered to the average pH of the skin. In the present study, the pH of the freshly prepared base and the formulation was 5.51 and 5.34, this is very close to the skin pH. In the present study indicated that erythema induces by application of base and formulation was decrease after 24 hours of their application.

CONCLUSION:

Thus it is concluded that both base and formulation produce no skin irritation after performing a patch test of 24 hours. It may be attributed to the presence of a emollient glycerin in the base and formulation, and G. glabra in the formulation, which has the ability to reduce to skin erythema.

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