

FORMULATION AND EVALUATION CLITORIA TERNATEA LINN. ALCOHOLIC EXTRACT ANTIPIGMENT CREAM

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ABSTRACT:

It is concluded that Clitoria ternatea is a plant with a variety of ethnic medicinal uses. The qualitative analysis of Clitoria ternatea shows the presence of bioactive compounds such as Alkaloids, Tannins, Glycosides, Resins, Steroids, Saponins, Flavonoids and Phenols. The quantitative estimation of total Saponins, Flavonoids and Phenols in roots and of Flavonoids in shoots, flowers and seeds is also reported which is very important for the pharmaceutical industry. This is valuable information for preparation of drugs in pharmaceutical industry and stress the need for more intensive research in this medicinal plant since the compounds play a great role in healthcare. The pH of the cream base was found to be in range of 6.2-6.9 which is good for skin pH. The viscosity of was cream was in the range of 27021-27053 cps which indicates spreadibility of cream. Acid value 5.9, saponification value 25.7. Irritancy test was conducted in this project work. Dye test this dye confirms that formulation is o/w type emulsion cream. Homogeneity: formulation of base produce uniform distribution in cream. This was confirmed by visual appearance and by touch. Appearance When formulation kept for long time, it found that no change in colour of cream base After feel Emolliency, slipperiness and amount of residue left after the application of fixed amount of cream base was found Type of smear After application of cream base, the type of smear formed on the skin were non greasy Removal The cream applied on skin was easily removed by washing with TAP WATER and result found to be satisfactory. The skin irritation study exhibited that no such sign of irritation, itching, redness and inflammation was found over lip over extended period of time.

Keywords: inflammation, Emolliency, spreadibility, saponification value, Acid value, Alkaloids, Tannins, Glycosides, Resins, Steroids, Saponins, Flavonoids and Phenols

MATERIALS AND METHODS

Clitoria ternatea Linn. plants were collected from rural organic nursery garden, place : Bargaon, Sundargarh District, Odisha. The plant parts namely leaves, roots, shoots, flowers and seeds were shade dried and powdered in a mechanical grinder for preparation of extract. Preparation of plant extracts The powdered plant parts were Soxhlet extracted with methanol. The extract, on removal of solvent in vacuum, gave a dark greenish brown semisolid residue. The powdered material or the extracts of the plant parts mentioned above were used for the study. Qualitative analysis It comprised of tests for the presence of Alkaloids, Tannins, Glycosides, Resins, Steroids, Saponins, Flavonoids and Phenols. Test for Alkaloids About 0.5 gm of methanol extract was taken in a test tube and was diluted and homogenized with 10 ml distilled water, dissolved in 20 ml dilute HCl solution and clarified by filtration. The filtrate was tested with Drangendroff's and Mayer's reagent. The treated solution was observed for precipitation of white or creamy colour.

Test for Tannins: Five grams of the ground powder was extracted with 10 ml ammonical chloroform and 5 ml chloroform. The mixture was filtered and the filtrate was shaken with 10 drops of 0.5 M sulphuric acid. Creamish white precipitate was observed for the presence of tannins.

Test for Glycosides: About 0.5 gm of methanol extract was taken in a test tube and 1 ml glacial acetic acid containing traces of ferric chloride was added to it. To this solution, 1 ml concentrated sulphuric acid was added and observed for the formation of reddish brown colour at the junction of the two layers and the upper layer turned bluish green in the presence of glycosides.

Test for Resins: For the tests concerning the presence of Resins, 0.5 gm of methanol extract was taken in a test tube and 5 ml of distilled water was added to it and observed for turbidity which indicates the presence of Resins.

Test for Steroids: About 0.5 gm of methanol extract was taken in a test tube and 2 ml of acetic anhydride was added to it and 2 ml of sulphuric acid was added by the sides of the test tube and observed for the colour change to violet or blue green.

Test for Saponins: About 0.5 gm of methanol extract was taken in a test tube and 5 ml distilled water was added to it. The solution was shaken vigorously and observed for persistent froth. The frothing was mixed with 3 drops of olive oil and shaken vigorously after which it was observed for the formation of an emulsion

Test for Flavonoids: About 0.5 gm of extract was introduced into 10 ml of ethyl acetate in a test tube and heated in boiling water for 1 min. The mixture was then filtered. About 4 ml of the filtrate was shaken with 1 ml 1% aluminium chloride solution and incubated for 10 min. Formation of yellow colour in the presence of 1 ml dilute ammonia solution indicated the presence of flavonoids.

Test for Phenols: About 0.5 gm of extract was taken in a test tube, mixed with 100ml distilled water and heated gently. To this, 2 ml of ferric chloride solution was added and observed for the formation of green or blue colour. Quantitative analysis of the root extract was carried out for total Flavonoids, Saponins and Phenols and the shoot, flower and seed extract for total flavonoids. The root extract was prepared as explained above. Determination of total

Flavonoids: The Aluminium chloride colorimetric method (Chang et al. 2002) with some modifications was used to determine total Flavonoids content. The liquid extract was prepared (with mixing 0.5 gm of root/shoot/flower/seed extract in 100 ml of water) and 1.0 ml of this was mixed with 1.0 ml of methanol, 0.5 ml of aluminum chloride (1.2 %) and 0.5 ml of potassium acetate (0.1176 %). The mixture was allowed to stand for 30 min at room temperature. Later, the absorbance was measured at 415 nm in a spectrophotometer. Quercetin was used as standard. Flavonoid content is expressed in terms of quercetin equivalent (mg/g of extracted compound).

Determination of Saponins: The method of Obadoni and Ochuko (2001) was used for determination of Saponins. The root extract (20 gm) was put into a conical flask and 100 ml of 20 % aqueous ethanol was added. It was heated over a hot water bath for 4 h with continuous stirring at about 55° C. The mixture was filtered and the residue re-extracted with another 200 ml 20 % ethanol. The combined extracts were reduced to 40 ml over water bath at about 90° C. The concentrate was transferred into a 250 ml separator funnel and 20 ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated and 60 ml of n-butanol was added. The n-butanol extract was washed twice with 10 ml of 5 % aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporation, the samples were dried in the oven to a constant weight. The content of Saponins was estimated as mg/gm of extracted compound.

Determination of Phenols The method Gupta et al. (2010) was followed presently. To 5 gm of the root extract in a 250 ml beaker, 200 ml of 10 % acetic acid in ethanol was added, covered and allowed to stand for 4 h. This was filtered and the extract was concentrated on a water bath to one quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitate was collected and washed with dilute ammonium hydroxide and then filtered. The residue comprising of the phenols was dried, weighed and expressed as mg/gm of extracted compound..

Phytochemical screening of medicinal plants is very important in identifying new sources of therapeutical and industrial importance (Salhan et al. 2011).

Antiaging cream and its effect Many anti-aging creams, function in four ways to help the slow skin aging process. It is a very potent antioxidant and it helps maintain the health of the mitochondria, which is the powerhouse of the cell. When this cell is compromised, it cannot perform youthful repair functions. Also, it helps turn off an inflammatory messenger known as nuclear factor kappa B that can do much damage to the skin. Alpha-lipoic acid activates a collagen-regulating factor known as AP-1 that turns on enzymes that digest damaged collagen. Aged skin occurs when the slowdown in production of youthful new cells fail to replace the accumulation of damaged aged cells. Vitamin A stimulates skin cell renewal by increasing the rate of mitotic cell division. Anti-aging creams, make sure it has four important ingredients, such as alphas-lipoic acid, glycolic acid, retinoic acid and Vitamin A. Whether these products work or not, it wouldn't hurt to try.

Drug Formulation The emulsifier (stearic acid) and other oil soluble components (Cetyl alcohol, almond oil) were dissolved in the oil phase (Part A) and heated to 75°C. The preservatives and other water soluble components (Methyl paraban, Propylene glycol, extracted materials). were dissolved in the aqueous phase (Part B) and heated to 75° After heating, the aqueous phase was added in portions to the oil phase with continuous stirring until cooling of emulsifier took place.

Evaluation of Cream[10]

pH of the Cream The pH meter was calibrated using standard buffersolution. About 0.5g of the cream was weighed and dissolved in 50.0 ml of distilled water and its pH was measured.

Viscosity Viscosity of the formulation was determined by Brookfield Viscometer at 100 rpm, using spindle no 7. Dye test The scarlet red dye is mixed with the cream. Place a drop of the cream on a microscopic slide covers it with a cover slip and examines it under a microscope. If the disperse globules appear red the ground colourless. The cream is o/w type. The reverse condition occurs in w/o type cream i.e. the disperse globules appear colourless in the red ground. Homogeneity The formulations were tested for the homogeneity by visual appearance and by touch. Appearance The appearance of the cream was judged by its color, pearlscence and roughness and graded. After feel Emolliency, slipperiness and amount of residue left afterthe application of fixed amount of cream was checked. Type of smear After application of cream, the type of film or smear formed on the skin were checked. Removal The ease of removal of the cream applied was examined by washing the applied part with tap water. Acid value Take 10 gm of substance dissolved in accurately weighed, in 50 ml mixture of equal volume of alcohol and solvent ether, the flask was connected to reflux condenser and slowly heated, until sample was dissolved completely, to this 1 ml of phenolphthalein added and titrated with 0.1N NaOH, until faintly pink color appears after shaking for 30 seconds.

Acid value = $n \times 5.61 / w$ n - number of ml of NaOH required, w - weigh of substance.
Saponification value

Introduce about 2 gm of substance refluxed with 25 ml of 0.5 N alcoholic KOH for 30 minutes, to this 1 ml of phenolphthalein added and titrated immediately, with 0.5 N HCL. Saponification value = $(b-a) \times 28.05 / w$ a - volume in ml of titrant, b - volume in ml of titrant, w -weigh of substance in gm.

Skin irritation test: The skin irritation test was carried out by using Human as animal model. The prepared Antiaging facial cream was applied over skin. In interval of 10 min, any reactions like itching, inflammation, redness etc. were not observed.

RESULT :

The pH of the cream base was found to be in range of 6.2-6.9 which is good for skin pH. The viscosity of cream was in the range of 27021-27053 cps which indicates spreadibility of cream. Acid value 5.9, saponification value 25.7. Irritancy test was conducted in this project work. Dye test This dye confirms that formulation is o/w type emulsion cream. Homogeneity: formulation of base produce uniform distribution in cream. This was confirmed by visual appearance and by touch. Appearance When formulation kept for long time, it found that no change in colour of cream base After feel Emolliency, slipperiness and amount of residue left after the application of fixed amount of cream base was found Type of smear After application of cream base, the type of smear formed on the skin were non greasy Removal The cream applied on skin was easily removed by washing with TAP WATER and result found to be satisfactory. The skin irritation study exhibited that no such sign of irritation, itching, redness and inflammation was found over lip over extended period of time.

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