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FORMULATION AND EVALUATION OF *CASSIA AURICULATA LINN* HERBAL CREAM

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ABSTRACT

The study effort was effort to make a multipurpose herbal cream by using *Cassia auriculata linn*. The task of this work is, to protect the herbal active biomolecules and certify the activity. The herbal cream was prepared by using *Cassia auriculata* flower extract, it was prepared by using two different solvent of ethanol and distilled water. The resultant extract was analyzed by various chemical tests. The results confirms that both the extract have active biomolecules. *Cassia auriculata linn* flower extract incorporated into inert cream base then the biological activity and cream evaluation were piloted. The result shows water extract of *Cassia auriculata* herbal cream was better results than etanol extract using herbal cream in all standard parameters.

KEYWORDS: Biomolecule, biological activity, *Cassia auriculata linn*, cream base, ethanol extract and water extract.

INTRODUCTION

The first protecting sheet of human body is skin, it is the largest organ of human body. Its roles are well demarcated, it provides protection against water loss, microbial infection, and environmental elements. When skin is under attack by microbial agents such as viruses, bacteria and often reacts by local inflammation. UV irradiation also alters immune function and induces skin cell death, further complicating the homeostasis of the skin.[1] Mostly herbal

cosmetic products are also referred to as natural cosmetic products. The herbs does not have any side effects on the human body, it also provide valuable nutrients and minerals to the body. Cosmeceuticals are cosmetic-pharmaceutical mixtures to improve magnificence and health through ingredients.

Cream

Pharmaceutical creams are semisolid dosage form containing one or more drug substances dissolved or dispersed in a suitable base.[2] Then advantages of cream is provide prolong contact in their site of application, Avoidance of first pass metabolism, convenient and easy to apply.

MATERIALS AND METHOD

Flower parts of *cassia auriculata* was collected from Anaikuttam village Virudhunagar district. Liquid paraffin were purchased from Sipali (I) Pharma, Bees wax from Reachem Laboratory Chemicals, and all other chemicals were of analytical grade.

Plant authentication

Plant of *cassia auriculata* was evaluated for its physical state, odor and colour. Plant was authenticated by Dr. L.Lingakumar, Head and associate professor in botany, Ayyanadar Janakiammal College, Sivakasi, Tamilnadu.

METHODS OF EXTRACTION

Maceration with ethanol

In the whole plant flower part was separated and dried under shade for 15 days. The dried product was powered using mechanical grinder. The powder was sieved through sieve no: 22 to get uniform particle size. About 1kg of coarse powder was macerated in ethanol (1.5L) for 5 days with occasional shaking, after completion of the maceration, the ethanolic extract was filtered and concentration at 55^0C on water bath till it acquires ¾ concentration.[3,4]

Maceration with water

About 1kg of coarse powder was macerated with water (1.5L) for 5 days with occasional shaking, after completion of the maceration, the extract was filtered and concentration at 55^0C on water bath till it acquires ¾ concentration.[5]

Preliminary phytochemical test

The Flower extract was subjected to chemical test for proof of identity of its active constituent's presents. Both extracts evaluated to find the following compounds alkaloids, carbohydrates, steroids, glycosides, saponins, tannins, phenolic compounds, proteins and free amino acids, and test for flavonoid.[6-8]

Test for Alkaloids

To 2 mg of the extract, 5 ml of distilled water and 2 M hydrochloric acid were added until an acid reaction occurred. To this 1 ml of Dragendroff's reagent was added. Formation of orange or orange red precipitate indicated the presence of alkaloids.

Test for carbohydrates

Boil 2 ml of Benedict's reagent with a crude extract, a reddish brown color indicated the presence of the carbohydrates.

2 ml of iodine solution mixed with crude plant extract. Purple or dark blue colors prove the presence of the carbohydrate.

Test for glycosides

The extract were boiled with 1 ml of dilute Sulphuric acid in a test tube separately for 5 min, filtered while hot, pipette out the supernatant or filtrate, cooled and shaken with an equal volumes of dichloromethane. The lower levels of dichloromethan separated and shaken with half its volume with dilute ammonia. A rose pink to red color appeared in the ammonical layer, indicating the presence of glycosides.

Test for Steroids

2 mg of dry extract was dissolved in acetic anhydride, heated to boiling, cooled and then 1 ml of concentrated sulphuric acid was added along the sides of the test tube. Formation of green colour indicated the presence of steroids.

Test for phenols and tannins

2 ml of 2% solution of ferric chloride mixed with crude extract. Black or blue-green color indicated the presence of tannins and phenols.

Test for Proteins and Amino acids

3 ml of extract were mixed with 5 ml Million's reagent separately. White precipitate was formed which on heating turned to brick red. It may indicated the presence of amino acids.

Test for Saponins

5 ml of extract, a drop of sodium bicarbonate solution was added. The test tube was shaken vigorously and left for 3 minutes. Formation of honeycomb like froth indicates the presence of saponins.

Tests for flavonoids

2 ml of 2% sodium hydroxide solution was mixed with plant crude extract, intensive yellow color was formed, which turned into colorless when added 2 drops of diluted acid to solution, this result indicated the presence of flavonoids.

Test for terpenoids

2 ml of chloroform was mixed with the plant extract and evaporated on the water path then boiled with 2 ml of sulphuric acid concentrated. A grey color produced indicated the entity of terpenoids.

Preparation of cream

The *cassia auriculata* extract powder was incorporated in to inert cream base. The formula was shown in table No.1.

Table No.1: Herbal Cream formula.

Ingredients	Working formula for F1	Working formula for F2
Cassia auriculata (flower extract)	0.5gm (Water extract)	0.5gm (Ethanol extract)
Liquid paraffin	6.86gm	6.86gm
Bees wax	1.12gm	1.12gm
Paraffin wax	6.86gm	6.86gm
Borax	0.056gm	0.056gm
Cetyl alcohol	0.14gm	0.14gm
Perfume	0.05ml	0.05ml
Distilled water	4.84gm	4.84gm

Evaluation of cream

Physical properties

The cream was observed for colour, odour and appearance.

Determination of pH

5 ± 0.01gm of the cream was weighed accurately in a 100 ml beaker. 45 ml water added and dispersed the cream in it. The pH of the suspension was determined 27^{0}C using a pH meter.[9]

Spreadability studies

Two glass slides of standard dimension were selected. The formulation whose spread ability had to be determined was placed over one of the slides. The other slide was placed on top of the formulation was sandwiched between the two slide across the length of 5cm along the slide. 100gm weight was placed upon the upper slide. So that the formulation between the two slides was pressed uniformity to form a thin layer. The weight was removed and the excess of formulation adhering to the slides was fixed on which the formulation was placed. The second moveable slide was placed over it with one end tied to a sting to which load could be applied by the help of a simple pulley and a pan. A 30gm weight was put on the pan and the time taken for the upper slide to travel the distance of 5cm and the separate away from the lower slide under the direction of the weight was noted. The spread ability was then calculated from the following formula.[10,11]

$$\text{Spreadability} = m \times l/t$$

m= weight tied to the upper slide, l= length of glass slide, t= time taken in seconds.

ANTI-MICROBIAL ACTIVITY

Material used for anti-microbial studies

Alcohol and Water extract were taken for antibacterial studies. Both alcohol and Water extract were dissolved in Dimethyl sulfoxide (DMS).

Micro organism

To clinical strains were used for the antibacterial studies such as *Streptococci*, *E.coli*. They are sub cultured on nutrient agar and stored at 4°C. Active culture for experiments were prepared by transferring loopful of cells from stock of nutrient agar which are incubated without agitation for 24 hours at 37°C.

Standards

The standard drug tetracycline were dissolved in Dimethyl sulfoxide (DMS).

Agar well diffusion method

The antibacterial medium Muller Hinton Agar was used in this study. Pathogenic bacteria were grown in nutrient broth for 24 hours and swapped on the Petridis plates containing Muller Hinton Agar. In Muller Hinton Agar plate, about 6 mm diameter well were made by gel puncture, specified quantity of herbal cream were applied into the well and the plates were incubated at 37°C for 24 hrs. The antibacterial activity was assayed by inhibition zone formed around the well.[12]

RESULTS AND DISCUSSION

The phytochemical characteristics of the two different extracts of *C. auriculata* flowers investigated. Alkaloids, carbohydrates, glycosides, steroids, phenolic compounds, proteins and amino acids, flavonoids, tannins and terpenoids were present in both extracts, in additionally saponins was present in water extract. The results were shown in table no.2.

Table no-2 Preliminary phytochemical tests.

Sl.No	Constituents	Water Extract	Ethanol Extract
1	Alkaloids	+	+
2	Carbohydrates	+	+
3	Glycosides	+	+
4	Steroids	+	+
5	Phenolic compounds	+	+
6	Proteins & amino acids	+	+
7	Saponins	+	-
8	Flavonoids	+	+
9	Tannins	+	+
10	Terpenoids	+	+

The antibacterial activity revealed that both the extracts containing formulation (F1 and F2) of *C. auriculata* flowers possessed antibacterial activity against Streptoccoci *and E.coli.* Among the solvent tested, the water extract containing cream showed highest inhibitory activity when compared to ethanol extract. Highest zone of inhibition 17 mm was observed against streptococci and 13 mm zone of inhibition in E.coli with water extract containing cream. The ethanol extract containing cream showed 8 mm diameter of zone inhibition in streptococci and 8 mm zone of inhibition in E.coli. The results were shown in table no.3.

Table no.3: Antibacterial activity of herbal creams.

Name of organism	Zone of inhibition (mm)			
	Standard	Control	F1	F2
Streptococci	16	3	17	8
E.coli	17	4	13	8

The *C. auriculata* herbal cream physical parameters of appearance, pH, homogeneity and spreadbility was observed, the both the formulation results are satisfied. The results were shown in table no.4.

Table no.4: Physical observation of *C. auriculata* herbal cream.

Physical parameters	F1	F2
Appearance	Pale Yellow colour	Pale Yellow colour
pH	6.1	6.1
Homogeneity A .By visual B. By touch	Homogeneous and smooth	Homogeneous and smooth
A. Spreadability	Easily spreadable	Easily spreadable
B. Wetness	Moisture surface	Moisture surface
Type of smear	Non greasy	Non greasy

CONCLUSION

The *Cassia auriculata flower extract* were successfully formulated as cream by using inert cream base. Microbiological studies confirms the activity of both *Cassia auriculata* multipurpose cream active against gram positive and gram negative bacteria. Though the formulation having activity the best formulation was selected on the basis of phytoconstituents presents, bacterial activity, cost of chemicals and minimal chemical composition. In all those factors derived that the formulation (F1) using water extract containing cream was best and economical.

FUTURE SCOPE

In future this research effort may continue for dose optimization, more microbial study and stability test, which will give a platform for FDA approval and preclinical research study.

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