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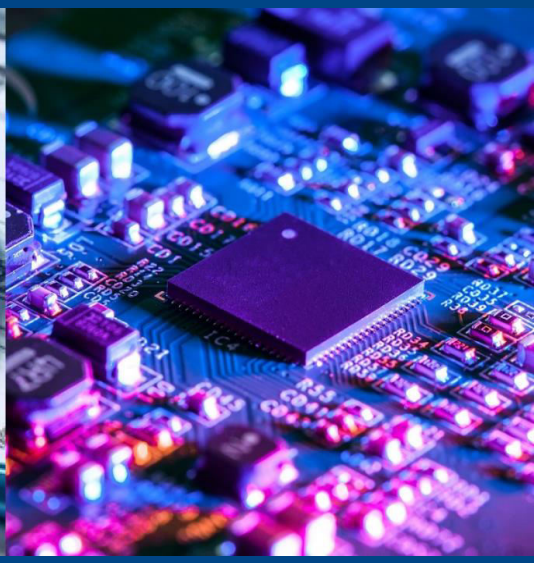

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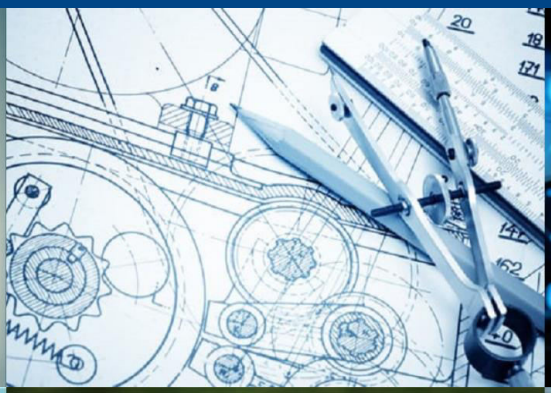

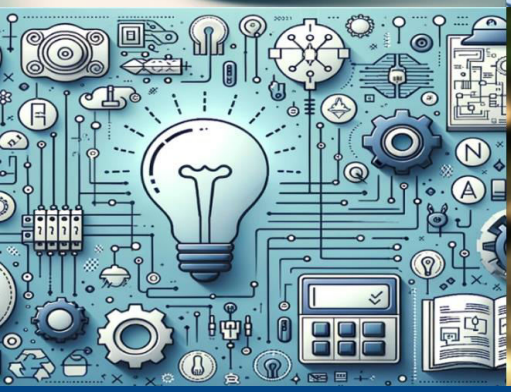

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Topic:- Formulation and Evaluation of a Polyherbal Churna for the Management of Vitiligo

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ABSTRACT: The present study focuses on the formulation and evaluation of a polyherbal churna for the management of vitiligo, a chronic depigmentary skin disorder characterized by the loss of melanocytes. The formulation was developed using four medicinal plants—*Psoralea corylifolia* (Bakuchi), *Azadirachta indica* (Neem), *Curcuma longa* (Turmeric), and *Rubia cordifolia* (Manjistha)—known for their traditional use in skin disorders and their antioxidant, anti-inflammatory, and melanocyte-stimulating properties. The prepared churna was evaluated for organoleptic characteristics, physicochemical parameters such as pH, moisture content, and ash value, and preliminary phytochemical screening. The results indicated acceptable organoleptic properties, stable physicochemical parameters, and the presence of key phytoconstituents including alkaloids, flavonoids, tannins, phenols, and saponins. These findings suggest that the polyherbal formulation possesses potential therapeutic value in promoting repigmentation and improving skin health, supporting its possible use as a natural and cost-effective alternative in the management of vitiligo.

I. INTRODUCTION

Vitiligo is a chronic, acquired depigmentary skin disorder characterized by the appearance of well-defined white patches on the skin due to the destruction or dysfunction of melanocytes, the cells responsible for melanin production. Melanin is the pigment that determines the color of the skin, hair, and eyes. The loss of melanocytes leads to depigmented areas that can appear on different parts of the body such as the face, hands, arms, and feet. According to epidemiological studies, vitiligo affects approximately 0.5–2% of the global population and occurs in individuals of all ages, genders, and ethnic backgrounds.

The exact cause of vitiligo is still not fully understood; however, several mechanisms have been proposed including autoimmune reactions, oxidative stress, genetic predisposition, and environmental triggers. Among these, autoimmune destruction of melanocytes is considered one of the major factors. In addition, increased oxidative stress in the skin may lead to melanocyte damage and impaired melanin synthesis. These factors together contribute to the development and progression of vitiligo.

Conventional treatments for vitiligo include topical corticosteroids, calcineurin inhibitors, phototherapy, and surgical grafting techniques. Although these treatments may help repigmentation in some patients, they are often associated with limitations such as side effects, high cost, long treatment duration, and variable effectiveness. Because of these challenges, there is increasing interest in exploring herbal and natural remedies that may offer safer and more sustainable therapeutic options.

Traditional systems of medicine such as Ayurveda have long used various medicinal plants for the management of skin disorders including vitiligo. Several herbal drugs possess properties such as antioxidant activity, immunomodulatory effects, and stimulation of melanocyte function, which may be beneficial in restoring skin pigmentation. In the present study, a polyherbal formulation has been developed using four medicinal plants: Psoralea corylifolia (Bakuchi), Azadirachta indica (Neem), Curcuma longa (Turmeric), and Rubia cordifolia (Manjistha).

Bakuchi (Psoralea corylifolia) is one of the most important herbs used traditionally for vitiligo treatment. It contains psoralen and related compounds that stimulate melanocyte activity and promote repigmentation of the skin.

Neem (Azadirachta indica) possesses strong antimicrobial, anti-inflammatory, and immunomodulatory properties that help in maintaining skin health.

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Turmeric (Curcuma longa) is well known for its antioxidant and anti-inflammatory effects due to the presence of curcumin, which may help reduce oxidative stress involved in vitiligo pathogenesis.

Manjistha (Rubia cordifolia) is widely used in Ayurveda as a blood purifier and skin rejuvenating herb and is believed to support healthy skin pigmentation.

The combination of these medicinal herbs in a polyherbal formulation may provide synergistic therapeutic effects for the management of vitiligo. Therefore, the present research aims to develop and evaluate a standardized polyherbal churna formulation containing Bakuchi, Neem, Turmeric, and Manjistha for its potential role in supporting repigmentation and improving skin health. Materials and Methods.

MATERIALS AND METHODS

The raw herbal drugs used in the formulation were **Bakuchi (Psoralea corylifolia) seeds, Neem (Azadirachta indica) leaves, Turmeric (Curcuma longa) rhizome, and Manjistha (Rubia cordifolia)** roots. All the crude drugs were collected from a local herbal drug store and authenticated based on their morphological characteristics. The chemicals and reagents used for analytical testing were of analytical grade.

Plant Material

Fresh plant materials that is Leaves, stem and Seed of Psorolia corylifolia L. and Caesulia axillaris Roxb, were collected from different regions Jalgaon city side area. Collected plant materials were taxonomically identified and authenticated by botanical expert. The plant materials were shade dried until all the water molecules evaporated and plants became well dried for grinding. Dry plant material separately grind to a fine powder and stored for further experiment with proper labels.

Preparation of Extract

Dried powder of Leaves (L), Stem (St) and Seed (S) powder for each experimental plant was exhaustively extracted successively in soxhlet apparatus using Hexane, Chloroform and Methanol respectively. The solvents were removed by distillation and the last traces of solvent being removed under reduced pressure. The extracts were weighed and their percentage value was recorded and thereafter, was stored in refrigerator for further experimental work .

Phytochemical analysis

The extractions was tested for the presence of bioactive compounds by using following standard methods

Test for alkaloids: Crude extract was mixed with 2ml of 1% HCl and heated gently. Mayer's And Wagner's reagents were then added to the mixture. Turbidity of the resulting precipitate was taken as evidence for the presence of alkaloids

Test for flavonoid: When dilute sodium hydroxide was added to 0.2 ml of extract creates intense yellow colour, which on addition of HCI turns colourless suggests presence of flavonoids

Test for tannins: Crude extract was mixed with 2ml of 2% solution of FeCl3. A blue-green or black coloration Indicated the presence of tannins .

Test for phenolic: Formation of intense green, purple, blue or black colour with addition of 1% ferric chloride Solution to the extract.

Test for saponin: 200mg plant material was taken in 10 ml chloroform and then filtered. In 2ml filtrate, 2ml acetic anhydride and small amount of H_2SO_4 was added, appearance of blue green ring indicates presence of saponin

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MATERIAL AND METHODS

Collection of sample

Fresh leave of Neem Plant were collected on 15th of July 2018, from Agbani farm in Nkanu West Local Government Area of Enugu State, Nigeria. It was identified by Prof. Helen Nwamba, a Botanist of Enugu State University of Science and Technology.

Sample preparation

It was air dried at room temperature three weeks after which, it was taken to electric grinding machine where the engine was properly washed and dried to avoid contamination. The leave was ground to required texture.

Extraction

For aqueous extraction, the weighing balance was zeroed and the filter papers were placed on the weighting balance, a quantity 0.5 g of the sample were weighed and poured into a reagent bottle. 10 ml of distilled water was added to soak and facilitate extraction. After some minutes, proper filtration was carried out using filter paper after which the filtrate was tested with several reagents to determine the presence and quantity of tannins, alkaloids, saponins glycosides, terpenoids, flavonoids, steroids and phenols.

For ethanol extraction, the weighing balance was zeroed and the filter paper were place on weighting balance. The sample was weighed 0.5 g into 250 ml beaker; 100 ml of 10 % acetic acid in ethanol was added to the sample and covered. In ethanol extraction it was done in closed system for proper extraction.

Test for alkaloids

The method of was adopted in testing for alkaloids in neem plant. A quantity of 0.5 g of sample was dissolved in Hydrochloric acid and filtered using filter paper to the 2 ml of filtrated was treated with dragendroff's reagent (solution Of potassium Bismuth iodide) formation of red precipitate confirmed indicating the presence of alkaloid the test is called Dragendroff's test. To 2 ml of filtrates was treated with Hager's reagent, formation of yellow colour confirmed the Presence of alkaloid.

Test for tannin

This is based on utilization of standard conventional protocols as illustrated by [5]. A quantity, 0.5 g of sample was weighed out and stirred with 10 ml of distilled water and then filtered. To the 2 ml of filtrate measure out in the test tube, few drop of 1% ferric chloride solution were added formation of blue green precipitate was confirmed indicating the presence of tannins.

Test for saponins

The method of [5] was adopted in testing for saponins in neem plant. A quantity of 0.5 g of sample was boiled with 50 ml of distilled water and filtered. To 5 ml of each filtrate, 3 ml of distilled water was added and shaken vigorously for about 5 minutes, formation of frothing was confirmed showing the presence of saponins.and B was added until it turn to alkaline indicating the presence of glycoside.

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Test for flavonoids

The method of [6] was adopted in testing for flavonoids in neem plant. A quantity, 0.5 g of the sample dissolve in distilled Water and filtered to 5 ml of filtration, 3 ml of lead ethanoate solution was added. Appearance of pale yellow-brown (buff-coloured) confirmed the presence of flavonoid.

Test for phenol

The method of was adopted in testing for phenol in neem plant. A quantity of 0.5 g of the sample was boiled with 15 Ml of distilled water and filtered. To 2 ml of the filtrate, few drops of 10% ferric chloride solution were then added.

Formation of violet colour was confirmed indicating the presence of phenolic hydroxyl group.

Materials and Methods

Materials

Collection of Plant Materials-

The Manjishtha used for the study was collected from Rasashastra Pharmacy, National Institute of Ayurveda, Jaipur**.**

Test for Alkaloids

The following Colour tests are used to detect the presence of an Alkaloid in the sample.

Dragon Droff's reagent: It is soln. of Potassium Iodide and Bismuth sub nitrate. They form Orange colour ppt. with the reagent

Test for Carbohydrate

Molich's Test: 2 ml of the Aqueous Extract of Drug is taken in test tube and 2 ml of the Molisch's reagent is added and shaken carefully, then about 1 ml. of conc. H_2SO_4 is poured from side of the test tube and allowed to stand for one 1 minutes. A Red Brown ring at the junction of the two layers indicates the presence of Carbohydrate.

Test for Saponin

About 1 ml of Aqueous Extract is diluted by distilled water up to 10 ml and shaken in a graduated cylinder for 15 minutes. Formation of 1cm layer of froth indicates

presence of Saponin .

Test for Phenols

2 ml of Drug extract is taken in a test tube and added 2 ml of $FeCl_3$ solution. Blue – Violet/ Red or Deep Green colour of the solution. is suggestive to presence of Phenols.

Test for Flavonoids

To dry powder or extract, add 5 ml. 95% ethanol, few drops conc. HCL and 0.5 g magnesium turnings. Pink color observed

Materials and Methods

Source and Preparation of Plant Materials

Fresh rhizomes of ginger and turmeric were collected from local farms located along Laje Road, Ondo-City, Ondo State, Nigeria. The plant materials were taxonomically identified and authenticated in the Environmental Biology laboratory at the Department of Science Laboratory Technology, Rufus Giwa Polytechnic, Owo, Nigeria. The rhizomes were shade dried until all the water molecules evaporated and the rhizomes became dried for grinding. The foreskin of the rhizomes were removed and it was later ground using electrical laboratory blender into a very fine powder and kept in an airtight container with proper labelling prior to analysis.

Source of Reagents

The chemicals and reagents used for these analyses were analytic grade.

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Preparation of Plant Extracts

The method described by Amir et al., 2005 and modified by Arawande et al., 2013 was used. Ten gram of the powdery sample was weighed into each of the five cleaned and dried 250 mL reagent bottles and 100 mL of each solvent (ethanol, chloroform, ethyl acetate, acetone and water) was separately added to each of the bottle and left for 72 h during which it was intermittently shaken on a shaking orbit machine. The mixture was filtered through a 0.45 µm nylon membrane filter. The extracts wereevaporated to dryness under reduced pressure at 40ºC by a rotary evaporator. The obtained extract was weighed and the extractive value of each solvent was calculated thus:

Qualitative Phytochemical Screening of Ginger and Turmeric Extracts

Phytochemical screening was carried out ethanol, chloroform, ethyl acetate, acetone and water Extracts of ginger and turmeric extracts using standard procedures as described by Sofowora, 2008; Trease and Evans 1989; Odebiyi and Sofowora 1978 and Harborne 1973.

Test for tannin

0.5 g of plant extract was mixed with 2mL of water and heated on water bath. The mixture was Filtered and 1mL of 10% FeCl3 solution was added to the filtrate. A blue-black solution indicates the Presence of tannin. **Test for flavonoid**

5 mL of distilled water and about 0.2 g of plant extract were mixed thoroughly. And 1 mL of 1% AlCl3 solution was added and shaken. A light yellow precipitate indicates the presence of flavonoids.

Test for phenol

About 0.5 g of plant extract was added to 1 mL of 10% FeCl3 solution. A deep bluish green Colouration was an indication for the presence of phenol.

Test for saponin

About 0.2 g of plant extract was shaken with 4 mL of distilled water and then heated to boil on A water bath. Appearance of creamy miss of small bubbles (Frothing) shows the presence of saponin.

II. COMPOSITION OF POLYHERBAL CHURNA

The polyherbal formulation was prepared using the following ingredients:

S.NO.	Herbal Drug	Botanical Name	Quantity
1.	Bakuchi	Psoralea corylifolia	20g
2.	Neem	Azadirachta indica	10g
3.	Manjista	Rubia cordifolia	10g
4.	Turmeric	Curcuma longa	10g

Total weight of the formulation: 50 g

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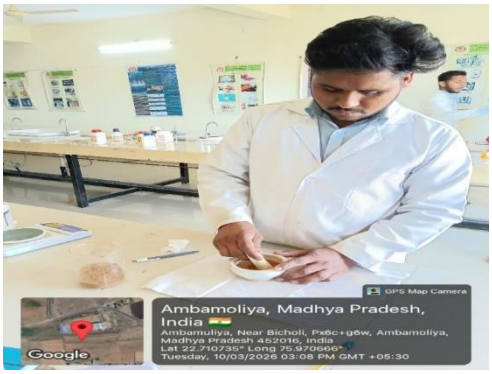
III. PREPARATION OF POLYHERBAL CHURNA

The collected herbal drugs were first cleaned to remove dust, foreign particles, and impurities. The crude drugs were then dried under shade to maintain their active constituents. After complete drying, each herb was separately powdered using a mechanical grinder. The powdered materials were passed through a sieve (No. 80) to obtain a fine powder. All the powders were accurately weighed according to the required quantity and mixed thoroughly using the geometric dilution method to ensure uniform distribution. The prepared polyherbal churna was stored in an airtight container to prevent moisture contamination.



IV. ORGANOLEPTIC EVALUATION

The prepared formulation was evaluated for its organoleptic properties, including colour, odour, taste, and texture, by visual and sensory inspection.

S.NO.	Herbs	Colour	Odour	Taste	Texture
1.	Bakuchi	Brown to dark brown.	strong aromatic odour.	Bitter and slightly pungent taste.	fine, smooth, and slightly oily
2.	Neem	Green to dark green	Strong herbal odour	Very bitter taste	Fine and slightly rough
3.	Manjistha	Reddish-brown to dark brown	Slight characteristic earthy odour	Slightly bitter and astringent taste.	Fine and slightly rough
4.	Turmeric	Bright yellow to orange-yellow	Aromatic and slightly spicy odour.	Slightly bitter, warm, and pungent taste.	Fine, smooth, and slightly dry

Physicochemical Evaluation of Individual Herbal Powders

Physicochemical evaluation of the selected herbal powders such as **Bakuchi (Psoralea corylifolia), Neem (Azadirachta indica), Turmeric (Curcuma longa), and Manjistha (Rubia cordifolia)** was carried out to determine their purity, quality, and physicochemical properties before formulation.

1. pH Determination

About 1 g of each herbal powder was dissolved in 100 ml of distilled water to prepare a 1% solution. The mixture was stirred properly and the pH was measured using a pH paper

1.Bakuchi :- 7
2.Neem :- 5
3.Manjistha :- 8
4.Turmeric :- 6

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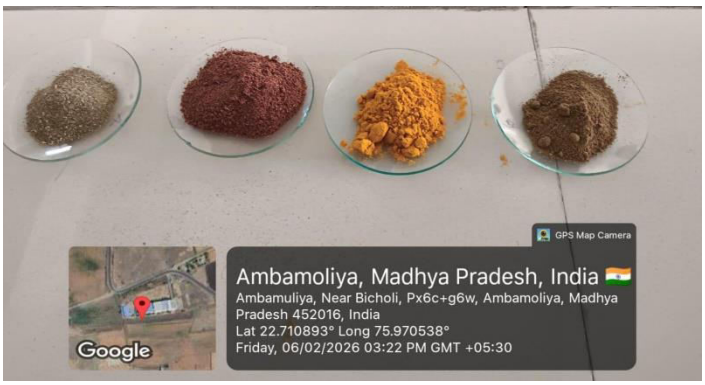
2. Loss on Drying (Moisture Content)

Approximately 3 g of each powdered drug was accurately weighed and dried in an oven at 105°C until a constant weight was obtained. The difference in weight indicates the moisture content present in the sample.

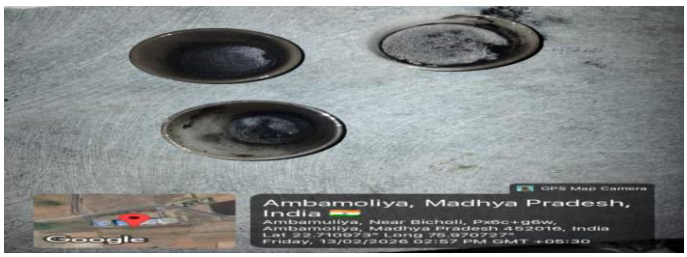
Weight	Bakuchi	Neem	Manjistha	Turmeric
Before drying	3g	3g	3g	3g
After drying	2.83	2.72	2.71	2.74

3. Total Ash Value



About 5 g of each powder was placed in a silica crucible and incinerated at a high temperature until a carbon-free ash was obtained. This determines the total inorganic content present in the drug.

Weight	Bakuchi	Neem	Manjistha	Turmeric
Before burning	5g	5g	5g	5g
After burning	1.35g	1.27g	1.33g	0.95g



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Phytochemical Screening

Preliminary phytochemical screening of the polyherbal formulation containing **Bakuchi (Psoralea corylifolia), Neem (Azadirachta indica), Turmeric (Curcuma longa), and Manjistha (Rubia cordifolia)** was carried out to identify the presence of various phytoconstituents. The powdered formulation was extracted using suitable solvents such as water and alcohol, and the extracts were subjected to standard qualitative tests.

Test	Bakuchi	Neem	Manjistha	Turmeric
Alkaloid	+	+	+	Nil
Carbohydrates	Nil	Nil	+	Nil
Flavonoids	+	+	+	+
Phenol	+	+	+	+
Tannins	+	+	Nil	+
Saponins	+	+	+	+

V. RESULT AND DISCUSSION

The present study focused on the preparation and evaluation of a polyherbal churna formulation for the management of vitiligo using Bakuchi (Psoralea corylifolia), Neem (Azadirachta indica), Manjistha (Rubia cordifolia), and Turmeric (Curcuma longa).

Organoleptic Evaluation

The prepared polyherbal churna was evaluated for organoleptic properties such as colour, odour, taste and texture. The formulation showed a brownish-yellow colour, a characteristic herbal odour, slightly bitter taste, and fine powder texture. These properties indicate good blending of all herbal ingredients.

Physicochemical Evaluation

The formulation was subjected to physicochemical parameters such as pH, moisture content and ash value. The pH of the churna was found to be near neutral, which indicates suitability for herbal consumption and stability. The moisture content was within acceptable limits, which helps prevent microbial growth and improves shelf life. The total ash value indicated the presence of inorganic constituents derived from herbal ingredients.

Phytochemical Screening

Preliminary phytochemical screening of the polyherbal formulation revealed the presence of important phytoconstituents such as alkaloids, flavonoids, tannins, phenols and carbohydrates. These bioactive compounds are known for their antioxidant, anti-inflammatory and skin protective properties.

Discussion

Vitiligo is a skin disorder characterized by depigmentation caused by the destruction or dysfunction of melanocytes. The herbs selected for the formulation are traditionally used for skin disorders.

Bakuchi contains psoralen, which is known to stimulate melanin production and is widely used in vitiligo treatment. Neem possesses antimicrobial and anti-inflammatory properties that help maintain skin health. Manjistha acts as a blood purifier and supports skin detoxification, while Turmeric provides antioxidant and anti-inflammatory effects that protect skin cells.

The combined effect of these herbs in the polyherbal formulation may help in stimulating melanocyte activity, improving skin pigmentation, and reducing oxidative stress, which are important factors in the management of vitiligo.

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Overall, the evaluation results confirmed that the prepared polyherbal churna possesses acceptable physicochemical characteristics and beneficial phytochemical constituents, supporting its potential use as a herbal formulation for vitiligo management.

The present study focused on the preparation and evaluation of a polyherbal churna formulation containing Bakuchi (Psoralea corylifolia), Neem (Azadirachta indica), Manjistha (Rubia cordifolia), and Turmeric (Curcuma longa) for the management of vitiligo.

The formulation was successfully prepared using selected herbal ingredients in appropriate proportions. The organoleptic evaluation showed acceptable characteristics such as brownish-yellow colour, characteristic herbal odour, and fine powder texture. Physicochemical parameters like pH, moisture content, and ash value were found within acceptable limits, indicating the quality and stability of the formulation.

Preliminary phytochemical screening confirmed the presence of important bioactive constituents such as alkaloids, flavonoids, tannins, phenolic compounds, and glycosides, which are known for their antioxidant, anti-inflammatory, and skin protective properties.

Based on the results obtained, the prepared polyherbal churna shows potential therapeutic value in the management of vitiligo due to the combined action of its herbal ingredients that may help stimulate melanocyte activity and support skin pigmentation.

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