



EVALUATION OF ANTIBACTERIAL AND ANTIOXIDANT ACTIVITIES OF FORMULATED *MURRAYA KOENIGII* CREAM

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ABSTRACT

The aim of present study was to formulate and evaluate the topical drug delivery system of *Murraya Koenigii* in form of cream. *Murraya Koenigii* cream was prepared by simple method. Formulated cream was evaluated for physical parameters, viscosity, pH, antibacterial and antioxidant activity. Physical parameters were showed satisfactory results. From antibacterial activity it was found that prepared Batch C of *Murraya Koenigii* leaves showed 19mm zone of inhibition against tested pathogens. *Murraya Koenigii* have also significant source of antioxidant, which might be helpful in preventing or slowing the progress of various oxidative stress including diseases. Formulated Batch C of *Murraya Koenigii* leaves powder showed 12.1% hydrogen peroxide scavenging activity. From above results it was concluded that the formulated herbal cream Batch C have significant antibacterial and antioxidant properties and hence will be better, safe and effective than allopathic medications.

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INTRODUCTION

Many plants are containing natural anti-bacterial and anti-oxidant compound which are used as natural medicine.⁽¹⁾ The plant is known to be active against wide spectrum of bacteria due to presence of flavonoids, Saponin and phenolic compound. Several reports are available on the antimicrobial activity of plant extracts on human pathogenic bacteria. *Staphylococcus aureus* and *Escherichia coli* are the main pathogens that cause skin infection.⁽²⁾

Oxygen is a highly reactive atom that is capable of becoming part of potentially damaging molecules commonly called “free radicals.” Free radicals are capable of attacking the healthy cells of the body, causing them to lose their structure and function. Antioxidants are capable of stabilizing, or deactivating, free radicals before they attack cells.⁽³⁾ Hence the naturally derived antioxidants, which may inhibit reactive oxygen species and may display protective effects. Plant phenolics, in particular phenolic acids, tannins and flavonoids are known to be potent antioxidants. In this context, one such plant is *Murraya Koenigii*.^(8,4)

India is the country which has some of the richest, oldest and diverse cultural tradition associated with the use of medicinal plants since centuries and still persists in living tradition. Herbal Medicines are used in almost 75% over the world with special reference to the developing countries where it is still practiced for the primary health care.

Medicinal and aromatic plants are important for the wellbeing of the population, especially of the rural poor who depend on these resources to treat human and livestock ailments and diseases.^(14,15)

Herbal formulations always have attracted considerable attention because of their good activity and comparatively lesser or nil side effects with synthetic drugs. *Murraya Koenigii* has been reported to have anti-oxidative, cytotoxic, antimicrobial, antibacterial, anti-ulcer, positive inotropic and cholesterol reducing activities. In this study creams were formulated based on the antibacterial and antioxidant activities of herbal extracts and its evaluation.

Murraya Koenigii linn. medicinal plant belongs to family – Rutaceae is commonly called as curry leaf tree, limblee tree. It is an evergreen shrub about 2.5-6 meters in height. This species is native to India. It commonly occurs in the Himalayas, Assam, Sikkim, Kerala, Tamilnadu, A.P, Maharashtra. The leaves of *Murraya Koenigii* Linn. are also used in Ayurvedic medicine. They contain several medicinal properties that include it being anti-diabetic, antioxidant, antimicrobial, anti-inflammatory, anti-carcinogenic and with hepato-protective properties.^(4,5)

Murraya Koenigii, commonly known as curry leaf or kari patta in Indian dialects, belonging to Family Rutaceae which represent more than 150 genera and 1600 species. *Murraya Koenigii* is a highly values plant for its characteristic aroma and medicinal value. A number of chemical constituents from every part of the plant have been extracted. The most important chemical constituents responsible for its intense

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characteristic aroma are P-gurjunene, P-caryophyllene, P-elemene and O-phellandrene. The plant is credited with tonic and stomachic properties. Bark and roots are used as stimulant and externally to cure eruptions and bites of poisonous animals. Green leaves are eaten raw for cure of dysentery, diarrhoea and for checking vomiting. Leaves and roots are also used traditionally as bitter, anthelmintic, analgesic, curing piles, inflammation, itching and are useful in leucoderma and blood disorders. A number of active constituents responsible for the medicinal properties have been isolated and characterized. This plant has been reported to have anti-oxidative, cytotoxic, antimicrobial, antibacterial, anti-ulcer, positive inotropic and cholesterol reducing activities. The available literature and wide spread availability of *Murraya Koenigii* in India thus makes it an attractive candidate for further pre-clinical and clinical research.^(16,17,18)

MATERIALS AND METHODS

Materials: The raw materials like drugs, polymers, excipients and chemicals required for the present work were procured from different sources. Following materials were used for the formulation and evaluation of cream.

Table no. 1 List of Materials

Sr.no.	Drug /polymer /chemical	Manufacture
1	Murraya koenigii	Harika foods.hyderabad.
2	Stearic acid	Lobachemie PVT.Ltd.
3	Spermaceti wax	S.D. Lab. chemical. Centre. mumbai.
4	Cetyl alcohol	S.D. Lab. chemical. Centre. mumbai.
5	Glycerin	Lobachemie PVT.Ltd.
6	Benzyl alcohol	Vishal Chem.mumbai.
7	Triethanolamine	Vishal Chem.mumbai.

Preparation of *Murraya Koenigii* cream by simple method

Oil in water (O/W) emulsion-based cream (semisolid formulation) was formulated. The emulsifier (stearic acid) and other oil soluble components (Cetyl alcohol, Spermaceti wax) were dissolved in the oil phase (Part A) and heated to 75°C. The preservatives and other water soluble components (Glycerin, Benzyl alcohol, Triethanolamine, ethanol extract of *Murraya Koenigii*) were dissolved in the aqueous phase (Part B) and heated to 75°C. After heating, the aqueous phase was added in portions to the oil phase with continuous stirring. An equivalent vigorous high shear type of mixing is equally effective. Stirring with the help of electrical stirrer was kept on until a homogenous cream was formed. After, the cream was allowed to cool to room temperature and then perfumes were added below 35°C.^(19,23)

Table no 2 Composition of *Murraya koenigii* cream

Sr.no.	Ingredient	Batch F1	Batch F2	Batch F3
1.	Stearic acid (gm)	10	12	14
2.	Spermaceti (gm)	5	5	5
3.	Cetyl alcohol (gm)	5	5	5
4.	Glycerin (ml)	1	1	1
5.	Benzyl alcohol (ml)	2	2	2
6.	Triethanolamine (ml)	2	2	2
7.	Extract (gm)	1	1	1
8.	Water upto (ml)	100	100	100

Evaluation of *Murraya Koenigii* cream

Visual Inspection :^(19,23)

The prepared cream was observed for color, odour, and appearance.

pH of the cream:⁽¹⁹⁾

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

Viscosity⁽²⁰⁾

Brookefield Viscometer (Model DV-E) with helipath stand was used for rheological studies. The sample (50 g) was placed in a beaker and was allowed to equilibrate for 5 min before measuring the digital reading using a spindle No. 63 at 50 rpm. At this speed, the corresponding reading on the viscometer was noted.

Antibacterial activity^(20,23,24)

Agar well diffusion method

Nutrient agar medium was prepared and sterilized by autoclaving at 121°C for 15 minutes. The three agar plates were prepared and labeled. The nutrient agar after sterilization was poured into the three plates and allowed to get solidify. After solidification the culture of *E.coli* were applied on each three plates. Then the well were prepared using cork borer. After the well preparation the samples were poured in respective wells using micropipette. The plates then allowed incubating at 37°C for 24 hours.

% Hydrogen peroxide radical scavenging activity^(3,4,21)

A solution of Hydrogen peroxide (20mM) was prepared in phosphate buffer pH7.4. Different concentrations of *Murraya Koenigii* (4 to 20ug/ml) in ethanol (1ml) were added to 2ml of the hydrogen peroxide solution. After 10mins, the absorbance was measured at 230nm using spectrophotometer against a blank solution that contained hydrogen peroxide solution without *Murraya Koenigii*.

$$\text{Radical scavenging activity} = \left(1 - \frac{A_s}{A_c}\right)$$

RESULT AND DISCUSSION

Physical Evaluation of Formulated *Murraya Koenigii* cream

The prepared cream was found to be the following colour, odour, texture and appearance shown in table no.4

Table no 3 Physical Evaluation of *Murraya Koenigii* cream

Batch no.	Colour	Odour	Texture	Appearance
Batch A	Pale Yellow	Characteristic	Smooth	Semi-solid
Batch B	Pale Yellow	Characteristic	Smooth	Semi-solid
Batch C	Pale Yellow	Characteristic	Smooth	Semi-solid

pH Measurement of *Murraya Koenigii* cream

The pH *Murraya Koenigii* cream was found to be 7.2, 7.0 and 6.8 in batch with A, B and C respectively shown in table no. 5

Table no 4 pH Measurement of formulated *Murraya Koenigii* cream

Sr.no	Batch no.	pH
1.	Batch A	7.2
2.	Batch B	7.0
3.	Batch C	6.8

Viscosity measurement of formulated *Murraya Koenigii* cream

The viscosity of cream are shown in table no.6 in batch with A,B and C respectively.

Table no.5 Viscosity measurement of *Murraya koenigii* cream

Sr . no.	Batch no.	Viscosity of cream (Cps)
1.	Batch A	2399
2.	Batch B	2399
3.	Batch C	2399

Antibacterial activity of Formulated *Murraya Koenigii* cream

The antibacterial activity of *M. koenigii* were determined in this study and it was found that the extracts at a concentration of 0.5 µg/ml tested with microbial strain E.coil produced significant zones of growth inhibition (Table 7and Figure 4.A,B &C).

Table no. 6 Antibacterial activity of *Murraya Koenigii* cream

Sr.no.	Batch	Zone of inhibition (mm)
1)	Batch A	12
2)	Batch B	15
3)	Batch C	19

Murraya Koenigii has effective antibacterial activity. From the above result it was found the Batch-C is the most optimized batch having good antibacterial activity.

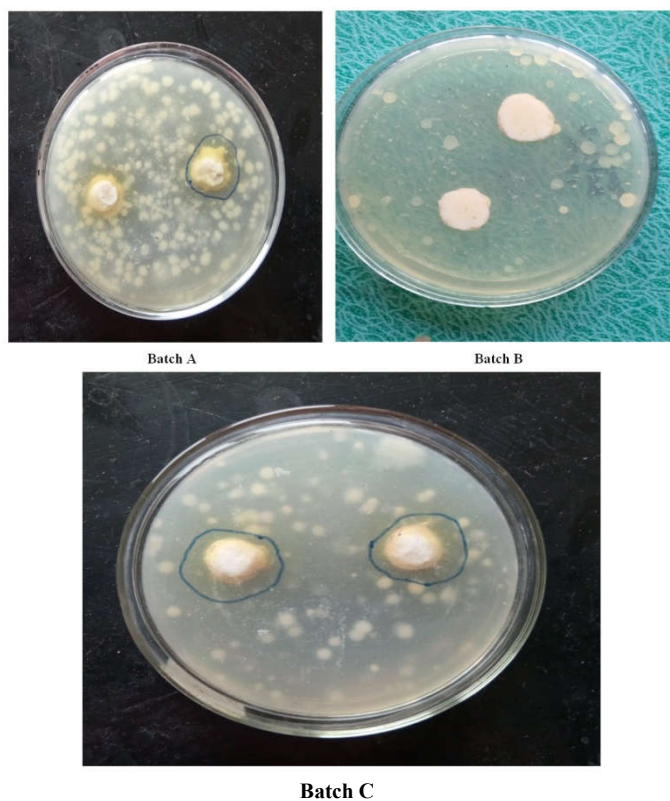


Fig No 1 Images of Antibacterial activity of formulated *Murraya Koenigii* cream

%Hydrogen peroxide radical scavenging activity of Formulated *Murraya Koenigii* cream

The ability of *M.koenigii* to scavenge hydrogen peroxide is shown in Table no.8 Hydrogen peroxide scavenging activities of *M.Koenigii* are found to be 1.3%, 4.6%, 12.1% in batch with A, B, and C respectively.

Table no 7 Hydrogen peroxide radical scavenging activity of *Murraya Koenigii* cream

Sr. no.	Batch	% Hydrogen peroxide radical scavenging activity.
1.	Batch A	1.3 %
2.	Batch B	4.6 %
3.	Batch C	12.1 %

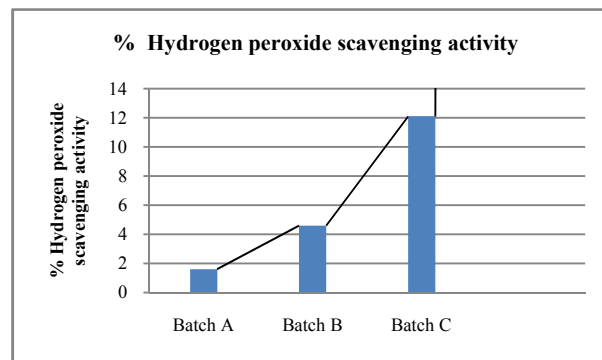


Fig No.2 % Hydrogen peroxide radical scavenging activity

Murraya Koenigii has effective hydrogen peroxide scavenging activity. From the above result it was found the Batch-C is the most optimized batch having good hydrogen peroxide radical scavenging activity.

CONCLUSION

This study is a preliminary evaluation of antimicrobial and antioxidant activity of the *Murraya Koenigii* plants. From the present investigation, it has been found that herbal cream of plant *Murraya Koenigii* leaves powder can be formulated using various ingredients and the evaluation of physical parameters shown satisfactory results. From antibacterial activity it was found that prepared Batch C of *Murraya Koenigii* leaves powder showed 19mm zone of inhibition against tested pathogens. *Murraya Koenigii* have also significant source of antioxidant, which might be helpful in preventing or slowing the progress of various oxidative stress including diseases. Formulated Batch C of *Murraya Koenigii* leaves powder showed 12.1% hydrogen peroxide scavenging activity. Hence, from the overall results, finally it was concluded that the formulated herbal cream Batch C have significant antibacterial and antioxidant properties and hence will be better, safe and effective than allopathic medications.

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