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Formulation and Evaluation of Cream from Fresh Yellow Latex of *Argemone Mexicana* Linn Plant

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***Abstract*: The yellow latex extract of *Argemone Mexicana* is well known by its antibacterial activity and its uses in wound healing. The aim of this study was to evaluate the antibacterial activity of aqueous extract of the yellow latex against *staphylococcus aureus* and *klebsiella* species. Also, to formulate effective, stable antibacterial cream and evaluate its physical and antibacterial properties. Disk diffusion method was used to assess antibacterial activity of aqueous extract. The antibacterial cream was prepared and evaluate for their physical, rheological and antibacterial properties. Finally, the efficacy of the antibacterial cream formulation was compared to one commercial product. The antibacterial activity of the aqueous extract was found to be effective against both *S.aureus* and *klebsiella* especially when the extract concentration increased. Stability studies showed a stable, homogenous appearance and effective during three week storage period at room temperature. It can be concluded that the aqueous extract of yellow latex of *ArgemoneMexicana* exhibited strong antibacterial activity especially with increase of the extract concentration. The prepared cream was found to be natural, stable and safe. The cream could be used topically in order to treat wound healing and burn.**

***Keywords*: *Argemone Mexicana* Linn plant, Yellow latex, berberine, Cream, antibacterial activity**

1. Introduction

Therapy with herbal drugs is an old traditional medicine. Plants have been used over the year for the treatment of numerous health problems including infections and non-infectious skin disorders.

The anti-microbial effect of some plants were attributed the use of conventional medication is often unsatisfactory for many patients with chronic skin disorders because of adverse effects and loss of effectiveness on long term uses. Moreover, the development of drug resistance in human pathogens against commonly used. Therefore, it has become necessary to search for an alternative safe effective medicinal with little side effects.

Argemone Mexicana Linn known as prickly poppy or Ghamoya (family: Papaveraceae) is an exotic weed indigenous in South America but has widespread distribution in many tropical and subtropical countries including Asia and West Africa. This plant is common everywhere by roadsides and field in India as well.

The plant is an erect prickly annual herb of about 1 meter high, leaves usually 5 to 11 cm long and flower become 4 to 5 cm in diameter. The seeds are spherical, shining, black and pitted.

Taxonomical classification:

Kingdom: Plantae – Plants
Subkingdom: Tracheobionta – Vascular plants
Superdivision: Spermatophyta – Seed plants
Division: Magnoliophyta – Flowering plants
Class: Magnoliopsida – Dicotyledons
Subclass: Magnoliidae
Order: Papaverales
Family: Papaveraceae – Poppy family
Genus: *Argemone* L. – prickly poppy
Species: *Argemone mexicana* L. – Mexican Prickly Poppy

Argemone Mexicana is considered as an important medicinal plant in India. Different part of this plant are used in chronic skin disease and also as emetic, expectorant, demulcent and diuretic; the seed and seed oil are employed as a remedy for skin disease ulcers and other intestinal affections. Flowers are found to be expectorant and have been used in the treatment of cough and roots are anthelmintic and also used in skin disease, leprosy and inflammations [1].

Fig. 1. Argemone Mexicana

Objective:

Aim of the present study were

- To formulate topical cream containing fresh yellow latex contains berberine.

- Evaluate the in vitro antibacterial activity of this berberine in the formulation.

2. Material and method

A. *Collection of bacterial isolates*

Bacterial strains were isolated from department of microbiology. All colonies that had grown on MacConkey agar and were incubated at 37°C for 24 hours.

B. *Collection of sample*

The fresh yellow latex of Argemone Mexicana Linn plant was collected from rural area (form kumbhari). The fresh yellow latex was collected from leaf and stem and dried it.

C. *Preparation of crude extract*

After drying of latex, the dried latex containing berberine is dissolved in water and alcohol. The standard Plant material W/solvent volume concentration used was 1/10 and the Mixture is filtered using Whatman filter paper no.1. The filtrate is used for further use.

Table 1
Chemical test for crude extract [2]

Test	Observation	Inference
1. Wagner's test: To small amount of crude drug, add Wagner's reagent(iodine-potassium iodide solution)	Gives brown or reddish brown colour or precipitate	Presence of alkaloid
2. Hager's test: To small amount of crude drug, add Hager's reagent (saturated solution of picric acid)	Gives yellow precipitates	Presence of alkaloid

Wagner's test **Hager's test**

Fig. 2. Wagner's and Hager's test

D. *Antibacterial assay by disc diffusion method*

Antibacterial activity was demonstrated using single diffusion method. A pure colony of each of test organism were sub-cultured into MacConkey agar (49.53 MacConkey agar is dissolved in 1 liter of water) and then autoclaved at 121°C for 15 min.

Table 2
Antibacterial assay by disc diffusion method

Organism	Fresh yellow latex	Distilled water	Zone of inhibition(mm)	Control (Ciprofloxaime)
S.aureus	0.01	0.9	21	0.005mm
Klebsiella	0.01	0.9	20	0.005mm

The test is carried out by using placing each disc in plates and plates are incubated at 37°C for 24hrs to observe formation of clear zone of inhibition [3].

(a) (b)

Fig. 3 (a) MIC of aqueous extract of fresh yellow latex on S.aureus, (b) MIC of aqueous extract of fresh yellow latex on klebsiella

Table 3
Antibacterial assay by disc diffusion method

Organism	Fresh yellow latex	Alcohol	Zone of inhibition (mm)	Control (Ciprofloxaime)
S.aureus	0.01	0.9	17	0.005mm
Klebsiella	0.01	0.9	10	0.005mm

(a) (b)

Fig. 4 (a) MIC of aqueous extract of fresh yellow latex on S.aureus, (b) MIC of aqueous extract of fresh yellow latex on klebsiella

Table 4
Formulation for Cream

S. No.	Ingredient	Quantity	Quantity taken (20g)
1	Stearic acid	24%	4.8
2	Water	64%	12.8
3	glycerin	10.5%	2.10
4	Fresh yellow latex	0.2%	0.02
5	Potassium hydroxide	1%	0.19
6	Perfume	q.s	q.s

E. *Procedure*

Melt stearic acid at water bath and dissolve potassium hydroxide in water add glycerin and add fresh dried yellow latex containing berberine is dissolved in water a heat to 75°C.add slowly melted stearic acid with constant stirring and add perfume at 40°C and transfer into wide mouth container [4].

F. *Principle*

Cream is semisolid preparation intended for external use only. Stearic acid and potassium hydroxide are combines to forms Potassium stearate. It is used as emulsifier. Fresh yellow latex has antibacterial properties due to berberine. The berberine is isoquinoline alkaloid found in yellow latex. Glycerin is used humectant and potassium hydroxide used to control the pH. The perfume is added for fragrance.

Formulation of the cream:

Base cream containing water and oil phase was prepared. The composition and the amount of the formulation ingredients are shown in Table 4. In order to prepare the cream, different amount of ingredient were incorporated together and then the required amount latex extract was added.

Evaluation of physical properties of formulation:

Physical Characteristics

The prepared formula containing latex extract and the commercial preparation were examined.

Determination of pH:

PH of the prepared formula was measured by using pH paper. The pH was found to be 7.

Homogeneity:

Homogeneity of formulation was tested by visual observation. The homogeneity of cream is good.

Spreadability:

The Spreadability determination: excess of sample was applied in between two glass slides and was compressed to uniform thickness by placing 100gm weight for 5 minutes. Weight was added to the pan. The time required to separate the two slides i.e. the time in which the upper slide moved over the lower plate was taken as measure of Spreadability.

$$S = m \times l / t$$

Where,

m = weight tide to upper slide

l = length moved on the glass slide

t = time taken.

Spreadability test also was performed by applying the cream on the skin and noticed that Spreadability is good.

Table 5
Measure of Spreadability

Parameter	Observation
Colour	Pale Yellow
PH	7
Homogeneity	Good
Spredability	14.13g.cm/sec

G. Physical Stability test

One set of 20 g samples of the formulation and the one commercial product, like Krack cream respectively were stored at room Temperature 37°C for 1 month. Then, after one month, their stability was checked regarding antibacterial activity and appearance. After one month there is slight change in pH.

H. In Vitro Antibacterial Activity

The in vitro antibacterial study was performed by measuring and comparing the diameter if zones of inhibition (in nm) for the various products. The zone of inhibition can be defined as the clear region around the well that contains an antimicrobial agent. It is known that the larger the zone of inhibition, the more potent the antimicrobial agent.

In the first step, the yellow latex antibacterial activity was measured individually against S. aureus and klebsiella. It concluded from the result that the antibacterial activity of yellow latex is higher against the tested microorganism. Results are confirmed that yellow latex have a synergistic activity, as shown by their larger zone of inhibition against tested microorganisms.

In the next step, the antibacterial activity of cream with marketed product was studied against S.aureus and klebsiella. The zone of inhibition increased as the concentration of latex increased. This indicated that the antibacterial activity of yellow latex increased against s.aureus and klebsiella as the concentration of the actives was increased. The cream containing 0.1% active ingredient had a similar antibacterial activity to that of the control; the cream containing 0.1% of active ingredient had statistically significantly higher activity against both microorganisms. Based on the result, it can be concluded that active ingredient has to be present of at least 0.1% to achieve a similar or better antibacterial activity as the control against both microorganism.

The final in vitro antimicrobial study was performed to compare the antibacterial activity of selected formulation to those of marketed product against s.aureus and klebsiella. We use krack cream for comparative study.

3. Results and discussion

The aqueous extract of yellow latex of Argemone Mexicana Linn plant was screened for biological activity against different strains of bacteria. The Yellow latex is selected on the basis of data obtained from the literatures, reports of their local traditional uses and for the treatment of various the skin disorders. The antibacterial activity-screening was carried out on organisms that are known to be among the most common causative agents of both primary and secondary infectious skin disorder. On the basis of their antibacterial activity, we go for antibacterial activity against staphylococcus aureus and klebsiella with aqueous extract and alcoholic extracts. As alcoholic extracts has minimum inhibitory activity than the aqueous extract. As aqueous extract have maximum inhibitory activity we selected aqueous extract for formulation of cream. The effect of the aqueous extract of fresh yellow latex with different concentration of the latex extracts ranging from 0.005-0.03mg.

Cream is semisolid dosage forms intended mainly for external use and commonly consist of two immiscible phases an oily phase and an aqueous external phase. Due to emulsified nature of skin surface, drug formulated as cream more effectively interact with skin and more rapidly penetrate through biological membranes.

4. Conclusion

The aqueous extract of yellow latex of Argemone Mexicana Linn exhibited strong antibacterial activity. The antibacterial activity was enhanced with the increase of the extract concentration. The result of different chemical and physical

tests of cream showed that it could be used topically in order to protect skin against damage caused by these this pathogen.

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