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Formulation and evaluation of antibacterial gel containing pongamia pinnata

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ABSTRACT

Background: Most topical preparations are meant to be applied to the skin and therefore basic knowledge of skin and its physiological function and biochemistry is very important for designing topical formulations. Sweat and fatty acids secreted from sebum influence the pH of the skin surface. It is advocated that the acidity of the skin helps in limiting or preventing the growth of pathogens and other organisms. One promising avenue is the use of herbal gels, which harness the power of natural compounds derived from plants to combat microbial infections. The aim of the study is to formulate and evaluate antibacterial gel containing Pongamia pinnata.

Result: Three formulations were developed by using suitable polymer(carbopol 934p). Developed formulations of herbal gel were evaluated for the physico-chemical parameters such as pH, viscosity, absorbance. This led to an outcome of the formulation of stable herbal gel possessing potent antibacterial activity.

Conclusion: Three formulations were developed by using suitable polymer (carbopol 934p). Developed formulations of herbal gel were evaluated for the physico-chemical parameters such as pH, viscosity, absorbance.

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1. Introduction

For the reason that delivery of mankind, there has been a relationship between life, ailment and plant life. There are no facts that humans in prehistoric instances used artificial medicines for his or her ailments however they tried to make use of the things they might without difficulty procure.¹

Microbes that enters the frame thru ingestion, inhalation, direct contact, cutaneous contamination and ascending infection after which stumble upon epithelial cells, macrophages, lymphocytes of limitations formed with the aid of mucous, sub cutaneous junctions or layers of pores

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The skin is a maximum enormous and readily on hand organ of the human body. 28 most topical education are supposed to be implemented to the pores and skin and hence basic expertise of skin and its physiological function and biochemistry could be very crucial for designing topical formulations.² Sweat and fatty acids secreted from sebum affect the pH of the skin floor. It is advised that acidity of the pores and skin facilitates in limiting or stopping the growth of pathogens and other organisms.³

One promising road is using natural gels, which harness the energy of herbal compounds derived from plant life to fight microbial infections.

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and skin.²

1.1. Gel

A gel is a solid or semi-stable machine of at least two parts, which include a condensed mass enclosing and interpenetrated by means of liquid. Combination of solid and liquid consisting of a way product will hold the go with the flow semi-solid in nature regarded a gel.¹

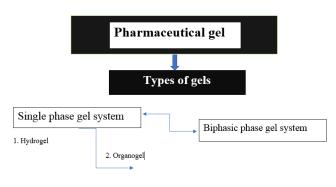


Fig. 1:

1.2. Hydrogels

- 1. Hydrogels are the 3-Dimentional, hydrophilic, polymeric network capable of imbibing the large amount of water or biological fluid.
- 2. Hydrogels also possess a degree of flexibility very similar to natural tissue due to their significant water content.
- 3. When the continuous phase is aqueous media, the gel is called as hydrogel.
- 4. Two types of hydrogels: 1. Natural hydrogel 2. Synthetic hydrogel.

1.3. Organogel⁴

- 1. Organogel is a non-crystalline, non-glassy thermoplastic strong cloth composed of liquid natural segment trapped in a three-D pass-related network.
- 2. The solubility and the particle measurement of the structure isn't important characteristics for the elastic properties and firmness of the organogel.
- 3. Organogel have the potential for use in a number of software such as in prescribed drugs, cosmetics, art conservation and food.
- 4. Examples: natural solvent, mineral oil, vegetable oil.

1.4. Mechanism of action

Gels depends on their specific composition and application. In general, gels work by trapping the liquid component within their three-dimensional network, providing stability and unique physical properties. Some mechanisms involved in gel formation and action include:

- 1. Physical cross-linking: This mechanism involves the entanglement of polymer chains or the formation of weak physical bonds between particles, leading to the creation of a gel network. Physical cross-linking can occur through processes such as cooling, solvent evaporation, or pH change.
- 2. Chemical cross-linking: Chemical cross-linking involves the formation of covalent bonds between polymer chains or particles, leading to a more stable gel structure. Chemical cross-linking can be achieved through the use of cross-linking agents, heat, or radiation.⁵
- 3. Swelling and diffusion: Gels can absorb and retain large amounts of liquid due to their porous structure. The gel network allows for the diffusion of molecules or ions through the gel matrix, which can be utilized for controlled release applications or for providing a barrier or protective effect.

1.5. Advantages of gel⁶

- 1. Gels are used to achieve optimal cutaneous and percutaneous drug delivery.
- 2. Gels are not deactivated by the liver enzymes because the liver bypass.
- 3. Gels are non-invasive and have patient compliance.⁷

1.6. Disadvantages of gel^6

- 1. Gels have possibilities of allergic reaction.
- 2. Gels which are used for the introduction into body cavity or eyes should be sterilized.⁸
- 3. They may cause the skin allergy during the application.

2. Aim and Objective

2.1. Aim

Formulation and evaluation of antibacterial gel containing Pongamia pinnata.

- 2.2. Objective
 - 1. To determine effect of herbal constituent against S. aureus and E. coli.
 - 2. To determine the preparation and formulation of antimicrobial gel.
 - 3. To demonstrate anti-microbial activity of herbal gel.

2.3. Biological source

Biological source of karanj is Pongamia pinnata Linn Pierre belonging to the family (Fabaceae) Leguminosae.

Table 1: Literature survey

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| S. No. | Title | Author | Year | Findings |
|--------|---|---|-------------|--|
| 1. | P. pinnata: Phytochemical constituents, Traditional uses and Pharmacological properties: A review ⁸ | V.V. Chopade, A.N. Tankar1, V.V. Pande1, A.R. Tekade1, N.M. Gowekar1, S.R. Bhandari1, S.N. Khandake1 | 22 Jan 2008 | Concentrated fruits or seeds extract can be found in various herbal preparations are widely available in market today. Pongamia pinnata preparation oil is widely available and employed by practitioner of natural health for treatment of rheumatism. ¹ |
| 2. | A review on Pongami pinnata (L.) Pierre: A great versatile leguminous plant. ⁶ | Sangwan S, Rao DV, Sharma RA. | 2010 | Pongamia pinnata is rightly called as Biodiesel plant, being considered as excellent source of Biodiesel. This plant is a multipurpose tree with immense medicinal and economic value. ⁶ |
| 3. | Bioavailability of karanjin from Pongamia pinnata L. in Sprague dawley rats using validated RPHPLC method. ¹⁴ | Shejawal N, Menon S, Shailajan S. | 2014 | The present work reports oral bioavailability of karanjin in rat plasma and a validated RP-HPLC-UV method for determination of karanjin. ⁹ |
| 4. | Pongamia pinnata (L.): Composition and advantages in agriculture: A Review ¹⁰ | KV-Usharani, Dhananjay Naik and RL Manjunatha. | 2019 | The plant shows the properties for the agriculture like insect pest management, as a biofuel, as a good source of crop macro and micronutrients, as a soil binder etc and medical industry as an anti-microbial, anti-ulcer, anti-diarrhoeal, anti-plasmodial, antiinflammatory, anti-oxidant antiviral, properties. Pongamia oil, leaf and cake was found to be the good nutritional value as a soil fertility management, pesticide, acaricide and nematicide in agriculture. ¹⁰ |
| 5. | Molecules of Interest – Karanjin – A Review | Aina Akmal Mohd Noor1,2, Siti Nurul Najiha Othman1, Pei Teng Lum1, Shankar Mani3, Mohd. Farooq Shaikh4, Mahendran Sekar1, * ¹¹ | 04 May2020 | Karanjin is initially recognized as an antioxidant, several kinds of research have proven that it is trul multifaceted in other industries as well. This precious molecule requires further understanding to fully capture its mechanism in many terms and benefits. Hence, this review has provided a comprehensive view concerning the initial ideas of the medicinal and agricultural purposes of karanjin. ¹² |
| 6. | A Review on Pongamia pinnata (L.): Traditional Uses, Phytochemistry and Pharmacological Properties ¹³ | Akshay G. Fugare 1 *, Rajkumar V. 2 Shete , Vishal S. 3 Adak, Krishna 4 Murthy G. | 19 Jan 2021 | Pongamia pinnata plant is used for anti-inflammatory, cardioprotective, anti- playsmodial, anti- nociceptive, anti-diarrhoeal, anti-ulcer, anti-hyperglycaemic, antihyperammonic and antioxidant, antibacterial, antiviral, anticonvulsant activity. ¹³ |

Table 2: Synonyms

| Marathi | Karanj |
|----------|---------------|
| Hindi | Karanjuaini |
| Tamil | Ponga, pongam |
| Assamese | Korach |



Fig. 2: Image of Pongamia pinnata (a) Seed oil (b) Flower (c) Fruit (d) Stem.

Table 3: Taxonomy 13

| Kingdom | Plantae |
|----------|---------------|
| Division | Magnoliophyta |
| Class | Magnoliopsida |
| Order | Fabales |
| Family | Leguminoseae |
| Genus | Pongamia |
| Species | Pinnata |

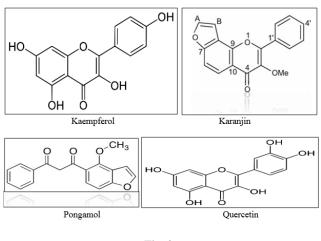
2.4. Phytochemical constituents

There are various chemical constituents isolated from the plant P. pinnata. Flavonoid and its derivatives are the most common constituents for isolation. The derivatives of flavonoids are flavones, flavans and chalcones. Sesquiterpene, diterpene, triterpenes, steroids, amino acids, disaccharides, fatty acids and ester compounds are also detected in this plant. Flavones are the most common constituents extracted from P. pinnata. The flavone class of compounds is distributed in all parts of this plant. Karanjin is considered the first compound to be extracted from this plant.¹⁴ Karanjin is a major chemical constituent of P. pinnata. In the current studies, explored the anti-ulcerative property of karanjin, a furano-flavoid isolated from the seeds of karanj.¹⁵

Karanjin is known as the main active principle in Karanj, which is effective against large number of insects.

2.5. Traditional uses¹⁶

- 1. The leafy foods are utilized in people solutions for stomach growths in India, the seeds for keloid cancers in Sri Lanka and a powder got from the plant for growths in Vietnam 23. In Sanskritic India, seeds were utilized for skin diseases. Today, the oil is utilized as a liniment for stiffness.
- Leaves are dynamic against Micrococcus; their juice is utilized for cold, hacks, the runs, dyspepsia, tooting, gonorrhea and uncleanliness.





- 3. Pulls are utilized for cleaning gums, teeth and ulcers.
- 4. Bark is utilized inside for draining heaps. They are likewise utilized for beriberi.
- 5. Juices from the plant as well as the oil are clean. It is supposed to be a superb solution for tingle, herpes and pityriasis versicolor.¹
- 6. Powdered seeds are esteemed as a febrifuge, tonic and in bronchitis and beating hack.
- 7. Blossoms are utilized for diabetes.
- 8. Juice of the root is utilized for purging foul ulcers and shutting fistulous injuries.
- 9. Youthful shoots have been suggested for ailment.
- 10. Ayurvedic medication depicted the root and bark as alexipharmic, anthelmintic and helpful in stomach amplification, ascites, biliousness, sicknesses of the eye, skin and vagina, tingle, heaps, splenomegaly, growths, ulcers and wounds; the fledglings, considered alexiteric, anthelmintic, aperitif and stomachic, for irritation, heaps and skin sicknesses; the leaves, anthelmintic, stomach related and diuretic, for irritations, heaps and wounds; the blossoms for biliousness and diabetes; the leafy foods for keratitis, heaps, urinary releases and infections of the cerebrum, eye, head and skin, the oil for biliousness, eye illnesses, tingle, leukoderma, ailment, skin sicknesses, worms and wounds.¹⁷
- 11. Unani framework utilizes the debris to reinforce the teeth, the seed, carminative and depurative, for chest objections, ongoing fevers, ear infection, hydrocele and lumbago; the oil is utilized as fuel for cooking and lamps.¹⁸

2.6. Pharmacological activity^{5,19}

- 1. Anti-Plasmodial activity
- 2. Anti-Inflammatory activity
- 3. Anti-diarrhoeal Activity
- 4. Antioxidant and Anti-hyperammonemic Activity

- 5. Anti-ulcer Activity
- 6. Anti-hyperglycaemic and Anti-lipidperoxidative Activity
- 7. Anti-lice Activity
- 8. Neuroprotective Activity
- 9. Anti-viral activity

2.7. Plan of work

- 1. Literature Survey
- 2. Sample collection
- 3. Preparation/Formulation
- 4. Evaluation Tests:
 - (a) Physical examination
 - (b) PH determination
 - (c) Homogeneity
 - (d) Skin Irritation
 - (e) Viscosity

3. Materials and Methods

3.1. Materials

Karanja oil, carbopol934p, triethanolamine, propyl paraben, methyl paraben, propylene glycol, water.

3.2. Methods

Polymer (carbopol934p) and purified water were taken in a beaker and allowed to soak for 24hrs. Take a required quantity of methyl paraben and propyl paraben in sufficient quantity of water which where dissolved by heating on water bath. The solution then cooled and propylene glycol were added. Further, the karanja oil was added to the mixture. The above mixture were added in a soaked carbopol934p and dissolved those mixtures properly until the homogenous gel was formed. Drop wise triethanolamine were added to the formulation for adjust the pH of the gel for the skin(6.8-7).

The gel formulation of karanja oil were prepared by using different concentration of extract (karanja oil).

3.3. Evaluation of herbal gel¹⁸

3.3.1. Physical evaluation

Physical parameters like colour and appearance were checked.

3.3.2. Measurement of pH

pH of a gel was measured by using pH meter.

3.3.3. Spreadability

The steel blocks used to actually take a look at spreadability. Spreadability was estimated by this strategy based on the slip and the medication attributes of the gel put on ground slides and the overabundance gel under the investigation. The gel was then positioned between the slides and 200g weighted for 5minutes was put on the highest point of 2 slides to remove air to give a uniform gel film between the slides where the overabundance gel was rejected off the edges. The time noted by the top slide to cover a distance of 7.5cm should be noted.

S=M.L/T Where, M-weight attempted to upper slide L-Length of the glass slide T-Time taken to isolate the slide

3.3.4. Homogeneity¹⁰

After the gel was set in compartment spread on slide, by visual examination, the created gels were tried for the presence of any bumps, flocculates or totals.

3.3.5. Consistency¹⁰

consistency of not entirely settled by utilizing Brookfield viscometer with Ski lift shaft. The consistency was viewed as 7720cp at 50rpm.

3.3.6. Skin disturbance

The skin disturbance was completed on human workers. For planned gel, two workers were chosen and 1.0g of figured out gel was applied over an area of two square crawls to the rear of the hand the human workers were noticed for aggravation or any skin response.

3.4. Anti- microbial testing of herbal gel

- 1. The antimicrobial exercises of various not entirely set in stone by altered agar well dissemination strategy. Culture of Staphylococcus aureus and *E.coli* at 37 o C in 10 ml Mueller Hinton stock was utilized. The way of life were acclimated to roughly 105 CFU/ml with clean saline arrangement.⁹
- 2. In this technique, supplement agar plates were cultivated with 0.2 ml for 24 hr stock culture of E. coli and *S.aureus*. The agar plates were permitted to harden. A clean 8 mm drill was utilized to cut wells of equidistance in each plate 0.5 ml of detailing hatched at 37 degree Celsius for 24 hr in a hatchery.²⁰
- 3. Agar dispersion procedure was applied to concentrate on the antibacterial impact of the recently referenced separates. Last convergence of 5%, 10%, 15%, 20%, 30%, 40%, half of the sound leaf gel in Mueller agar were acquired for each concentrate independently. Mueller Hinton agar plates were cleaned with a suspension of Staphylococcus aureus arranged as referenced before, utilizing sterile q-tip. Plugs were eliminated from every agar plate creating openings.
- 4. To each opening 1gm of gel weakened in 10L refined water from various centralization of each concentrate was added and permitted to diffuse at room

| Formulation code | Drug | Carbopol | Water | Propyl paraben | Methyl paraben | Propylene glycol | Triethanolamine |
|------------------|------|----------|-------|-------------------|-------------------|---------------------|-----------------|
| F1 | 2g | 1g | 40 | 0.1 | 0.2 | 5ml | 1-2drop |
| F2 | 1.5g | 1g | 40 | 0.1 | 0.2 | 5ml | 1-2drop |
| F3 | 1g | lg | 40 | 0.1 | 0.2 | 5ml | 1-2drop |

Table 4: Formulations of herbal gel

temperature for 20 min, the plates were then brooded vigorously for the time being at 37^{o} C.²¹

5. The antibacterial action was assessed by estimating zone of restraint.

4. Result and Discussion

The prepared formulations were characterized for physical appearance, pH, spreadability, viscosity, in-vitro antimicrobial study and in-vitro skin irritation study.¹

 Table 5: Organoleptic characterization for oil.

| Karanja oil | Characteristics |
|-------------|-----------------|
| Colour | Dark green |
| Odour | Pleasant aroma |
| Taste | Bitter |

pН

7.0

6.8

6.9

| Table 10: Spreadabili | ity |
|-----------------------|---------------------------|
| Batches | Spreadabliity (gm-sm/sec) |
| F1 | 4 cm |
| F2 | 5.7 cm |
| F3 | 5.8 cm |

Table 11:

| Marketed gel | Spreadability |
|------------------|---------------|
| Itraconazole gel | 2.8 |

Table 12: Anti- microbial activity of gel formulation

| Name of formula | Culture media | Zone of inhibition (mm ²) |
|-------------------------------|---------------------|---|
| F1 | S.aureus and E.coli | 5.4 mm and 6 mm |
| F2 | S.aureus and E.coli | 7 mm and 6 mm |
| F3 | S.aureus and E.coli | 7.5 mm and 8.0 mm |
| Standard-drug Azithromycin | S.aureus and E.coli | 10 mm |

Table 7: Homogeneity

Table 6: pH determination.

Formulation

F1

F2

F3

| Batches | Homogeneity characteristic |
|---------|----------------------------|
| F1 | Thick and greasy |
| F2 | Smooth and consistent |
| F3 | Smooth and homogenous |

Table 8: Skin irritation

| Characteristics |
|--------------------|
| No skin irritation |
| No skin irritation |
| No skin irritation |
| |

Table 9: Viscosity

| Batches | Viscosity (cps) |
|---------|-----------------|
| F1 | 6598 cps |
| F2 | 7720 cps |
| F3 | 7560 cps |
| | |



Fig. 4:

5. Conclusion

Three formulations were developed by using suitable polymer (carbopol 934p). Developed formulations of herbal gel were evaluated for the physico-chemical parameters such as pH, viscosity, absorbance. This led to an outcome of the formulation of stable herbal gel possessing potent antibacterial activity. This study indicated that the effectiveness of gel change by using different concentration of oil.

6. Source of Funding

None.

7. Conflict of Interest

None.

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