INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES
[ISSN: 0975-4725; CODEN(USA): IJPS00]
Journal Homepage: https://www.ijpsjournal.com

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Review Paper

Antibacterial and Antifungal Cream of Papaya Seed: A Critical Review of Formulation and Evaluation

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ARTICLE INFO

Published: 29 May. 2025
Keywords:
Carica papaya seed extracts, benzyl isothiocyanate, antibacterial, antifungal, Zone of inhibition
DOI:
10.5281/zenodo.15544220

ABSTRACT

Natural plant-based products have gained significant attention in pharmaceutical research due to their potential therapeutic properties. This study focuses on the formulation and evaluation of an antibacterial and antifungal cream derived from papaya (Carica papaya) seeds. Papaya seeds are known to contain bioactive compounds such as alkaloids, flavonoids, and phenolic compounds, which exhibit antimicrobial properties. The project involves the extraction of active compounds from papaya seeds using appropriate solvents, followed by the formulation of a topical cream. The cream is then evaluated for its physicochemical properties, stability, and antimicrobial efficacy against bacterial strains like Staphylococcus aureus and Escherichia coli, as well as fungal strains like Candida albicans and Aspergillus niger. Standard antimicrobial testing methods, such as agar well diffusion, are employed to determine the cream's effectiveness.

INTRODUCTION

Antibacterial are the substances which can effectively cure the infections caused by the different types of bacteria and Antifungal are the subatance which can effectively cyre the infection caused by different fungi. The frequency of life-threatening diseases caused by the micro-organisms has increased throughout the world and is becoming a main reason of mortality and morbidity in developing countries. The antibacterial properties of several medicinal substances have been analyzed by a number of studies worldwide and many of them substances have been used as therapeutic alternatives because of their very good medicinal properties. Plant based antimicrobials have more therapeutic effect with lesser side effects. Papaya belongs to a family of Caricaceae having four different genera in world. The genus Carica L. is represented by four types of species in India, of which Carica papaya

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Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

L. is the most widely cultivated and the best-known species. It is commonly known as Pawpaw, Tapayas, Papaya Melon tree, Kapaya, Papyas, Papye, papita, papayabaum and papaya. Papaya is basically originated from southern Costa Rica and Mexico, then introduced in to Sri Lanka, Australia, South Africa, Hawaii, Philippines and India all tropical and subtropical regions. It is growing both commercially and in-home garden. Carica papaya tree is an erect, fast-growing treemeasuring 7 - 8m tall, with copious latex and trunk of about 20cm in diameter. Its leaves are soft, lobulated, clustered, long-petiolated and measuring up to 80cm long. Its fruit is a greenish-orange berry about 7.5cm long and bitter in wild types, up to 45cm long with flesh 2.5-5 cm thick, sweet, juicy and of orange color in cultivars. Carica papaya seeds were approved and confirmed in some studies for their effective anthelmintic properties against nematodes found in animals. Chinoy has proved the anti-implantation, abortifacient and anti-fertility properties of extracts from papaya seeds. The seeds of C. papaya are potential anti-fertility drugs. Pawpaw seeds are used to produce an indigenous Nigerian food condiment called 'daddawa', the Hausa word for a fermented food condiment. Phytoconstituents analysis has shown that Carica papaya contains nicotine, carpain, flavonols, tannins, alkaloids and terpinenes as well as enzymes such as papain and chymopapain. Isothiocyanates (ITCs) are natural plant products generated by the enzymic hydrolysis of glucosinolates found in Brassicaceae vegetables. These natural sulfur compounds and their dithiocarbamate conjugates have been previously evaluated for their anti-cancerous properties. Their antimicrobial properties have been previously studied as well, mainly for food preservation and plant pathogen control. Recently, several revelations concerning the mode of action of ITCs in prokaryotes have emerged. This review addresses these new studies and proposes a model to summarize the current knowledge and hypotheses for the antibacterial effect of ITCs and whether they may provide the basis for the design of novel antibiotics.

MATERIAL AND METHOD: -

a. Extraction of Papaya seed.
b. Preparation of cream.

A. Extraction of Papaya seed: -

Plant Materials:

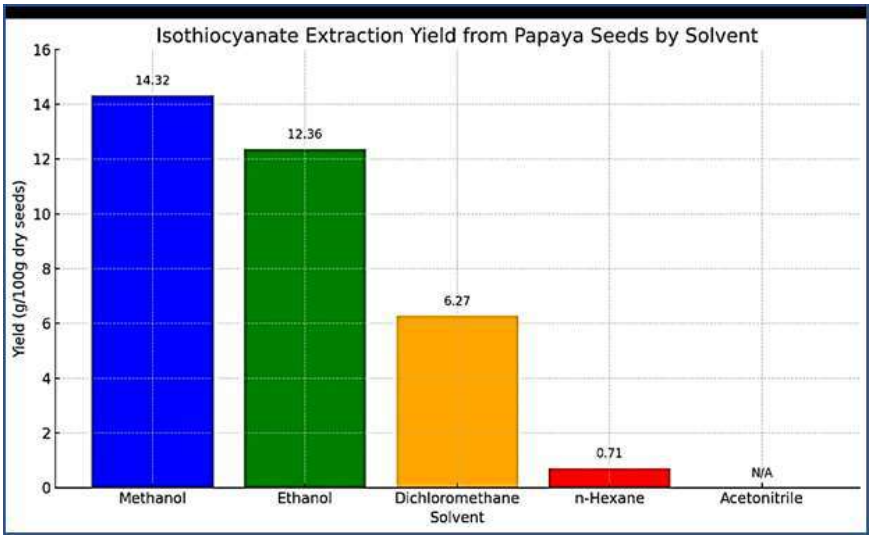
The plant material were used in this work are Carica papaya seeds were collected from in and around of the Alephate, Maharashtra

Preparation of Plant extraction:

Freshly collected seeds of Carica papaya L were thoroughly washed under tap water followed by

sterile water separately. The washed seeds of Carica papaya were dried independently in shade followed by grinding in to a fine powder. 15gm of dry powdered seeds of Carica papya L was extracted with 125ml of ethanol by soxhlet's apparatus for 6 hr (or) till the plant material get colourless. The trace amount of solvent was removed using a rotary vacuum evaporator to give a concentrated extract. In the same method followed for the extraction of Carica papaya L seeds with methanol, chloroform solvents also15. Different concentration (50, 100, 150µg/ml) of carica papaya seed solvent extracts was prepared with Dimethyl Sulfoxide (DMSO).

Fig 2. Liquid extract of Papaya seed



Screening of antimicrobial activity:

Media for test organisms:15.2g of Mueller Hinton Agar was dissolved in 400ml of sterile water and sterilized with autoclave at 121C for 15 mins at 15 lbs pressure. 1.0g of dextrose was dissolved in 10ml of sterile water and sterilized for 15 mins. After cooling both the solutions were mixed and poured into sterile petriplates up to approximately 4 mm and allowed to set at ambient temperature and used.

Media for fungal organism: The media used for antifungal test was Sabouraud's dextrose agar/broth of Hi media Pvt. Bombay, India.

Preparation of Inoculum: Bacterial inoculum was prepared with pure culture of test organism inoculated into 5ml of sterile nutrient broth and incubated at 37°C for 2 to 8 hr till moderate turbidity developed. The inoculum of different microorganisms was standardized by matching with 0.5 McFarland turbidity standards, which corresponds to cell density approximately 108 CFU/ml. The fungal strains were inoculated separately in Sabouraud's dextrose broth for 6h and the suspensions were checked to provide approximately 105 CFU/ml.

Antibacterial and antifungal activity by agar disc diffusion method: Antibacterial and antifungal activity of each carica papaya seed extracts was determined by using a modified Kirby Bauer disc diffusion method16. Broth cultures of test bacterial and fungal organisms were spread on the Mueller Hinton Agar media and sabouraud's medium in petriplates and microbes broth culture under lab condition. The extracts were tested using 5mm sterilized filter paper discs which impregnated with 50, 100, 150µg/ml of ethanol, methanol and chloform extracts of carica papaya seeds and allowed to dry for few minutes at room temp, plates were incubated at 37C about 24 hr.

B. Preparation of cream: -

1. Preparation of cream base: - 10g batch of emulsifying cream base was prepared by melting 3.2 g of emulsifying wax (beeswax) in a water bath maintained at 70 ± 0.5 °C. To the melted wax, 8 ml of liquid paraffin, 0.02g of methyl paraben, and 0.16g of borax was dissolved in 6ml Distilled water in heat bath. Subsequently, melted borax dissolved is added in bees wax in varying concentrations was incorporated into the mixture. The preparation was then removed from the water bath and stirred continuously until it cooled and achieved a uniform consistency.

2. Formulation of Antibacterial and Antifungal cream: - Carica papaya seed ethanol extract creams, 3.2 g portion of the emulsifying cream base was weighed into a demitasse dish and melted in a water bath maintained at 70 ± 0.5 °C. The required quantity of the ethanolic extract was added to the molten cream base and continuously stirred to ensure uniform dispersion. Warm distilled water was gradually added in portions to the mixture while stirring until a homogeneous blend was achieved. The cream was then allowed to cool before being transferred into labeled cream jars, designated as F1, F2, and F3.

Fig 3. of cream

EVALUATION OF CREAM

1.Physical appearance: Visual examination of the cream was conducted to assess its color, odor and texture. After being transferred to containers, all prepared creams were inspected to evaluate their homogeneity.

2. pH measurement: The pH value of the formulated cream was determined by weighing 1 g of the sample and dissolving it in 100 ml of distilled water. The pH measurement for each formulation was conducted individually using a digital pH meter.

3. Spreadability test: To measure the spreadability of the cream, 1 g of the sample was placed as a 1 cm diameter circle on a glass slide. A second glass slide was carefully placed on top of it, and a 250 g weight was applied for 5 minutes. The increase in the cream's diameter was observed, and the time required to separate the two glass slides was recorded. Three measurements were taken for each formulation, and the average values were calculated. The spreadability was determined using the following formula

S =M. L/T

The following was used to calculate the spreadability;

S = Spreadability,
M = Weight on the glass slide
L = Length moved the glass slide
T = Time taken to separate the slides.

4.Washability test: The washability test was carried out by applying the small quantum of cream on the nail of cutlet and also washed under the handling water.

5.Viscosity determination: The sample was placed in the teacup of a Brookfield Viscometer and allowed to rotate at spindle number 12. The measurements were taken independently at rotational speeds of 20 rpm and 30 rpm. For each speed, the readings were recorded, and the process was repeated three times. The average of the three readings at each speed was calculated to ensure consistency and accuracy.

6.Antifungal studies: The antifungal activity was evaluated using the slice prolixity system. Sabouraud's Dextrose Agar (SDA) plates were inoculated with a 72-hour culture of Candida albicans. Sabouraud's Dextrose Broth cultures were prepared in test tubes. Using a sterile cotton swab, the surface of the SDA plates was evenly swabbed to prepare a uniform field culture. Once the agar surface had dried (approximately 5 minutes), wells were aseptically created using a sterile cork borer. The wells were then impregnated with three formulations of the nail cream under study. A standard fluconazole disk was included on the same plate as a reference control. The plates were incubated at 28°C for 24–48 hours. After incubation, the zones of inhibition were measured using a scale to the nearest millimeter for each formulation.

7.Antibacterial Studies: The antibacterial activity was evaluated using the agar well diffusion method. Mueller-Hinton Agar (MHA) plates were inoculated with a 24-hour culture of Pseudomonas aeruginosa. Mueller-Hinton Broth cultures were prepared in test tubes. Using a sterile cotton swab, the surface of the MHA plates was evenly swabbed to prepare a uniform lawn culture. Once the agar surface had dried (approximately 5 minutes), wells were aseptically created using a sterile cork borer. The wells were then impregnated with different concentrations of the papaya seed extract under study. A standard ciprofloxacin antibiotic disk was included on the same plate as a reference control. The plates were incubated at 37°C for 24 hours. After incubation, the zones of inhibition were measured using a scale to the nearest millimeter for each formulation.

8.Test of stability: The stability assessment of the final product was carried out by keeping it under a constant temperature (room temperature) for 1 to

3 months. Further the physical instabilities such as changes in color, odor and constituency of the formulation and any signs of bacterial or fungal growths were checked. The stability of the final product was evaluated by storing it at a constant room temperature for a period of 3 months. During this time, the formulation was regularly monitored for physical instabilities, including changes in color, odor, and consistency. Additionally, the samples were inspected for any signs of bacterial or fungal growth.

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HOW TO CITE: Bhagat Suhani*, Vaid Harshada, Kamble Rachana, Antibacterial and Antifungal Cream of Papaya Seed: A Critical Review of Formulation and Evaluation, Int. J. of Pharm. Sci., 2025, Vol 3, Issue 5, 4714-4720. https://doi.org/10.5281/zenodo.15544220