Formulation and Evaluation of Cassia Alata Containing Anti-Fungal Spray

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***Abstract*—Cassia alata, a medicinal plant widely recognized for its potent antifungal properties, has been traditionally employed in the treatment of various skin infections. The present study focuses on the formulation and evaluation of a topical antifungal spray incorporating Cassia alata extract as the active ingredient. The spray was designed as a film-forming system to enhance drug contact time, permeability, and sustained release at the site of infection. Key formulation parameters such as polymer concentration, solvent system, nozzle type, and spray ability were optimized to achieve uniform distribution and effective drug delivery. The prepared spray was subjected to physicochemical evaluation, including viscosity, pH, drying time, and film-forming ability, along with in vitro antifungal activity against common dermatophytes. Results demonstrated that the Cassia alata-based spray exhibited significant antifungal efficacy, improved bioavailability, and reduced irritation compared to conventional topical preparations. The film-forming nature of the spray provided continuous drug release and accelerated wound healing through moisture control.**

***Index Terms*—Cassia alata; anti-fungal; topical formulation.**

I. INTRODUCTION

Topical Spray:
A topical medication is applied to a specific area of the body, typically the skin or mucous membranes, to treat localized ailments. These medications include creams, foams, gels, lotions, and ointments. Topical products do not cross the blood-brain barrier and are not intended for ingestion by humans or animals. Most commonly, topical drug delivery systems are applied to the skin, where the medicine either acts locally or is absorbed into the bloodstream through the dermis. Film-forming sprays generally consist of active substances, enhancers, and polymers dissolved in organic solvents. These sprays form a thin, non-sticky film that increases contact time and drug permeability, enabling continuous drug release. They also prevent crystallization, making more of the drug available for therapeutic effect compared to conventional topical preparations. Factors such as nozzle type, aperture size, spray pressure, and liquid properties significantly influence the spray ability of film-forming sprays.

Topical sprays offer several advantages over traditional topical preparations:

-Uniform drug distribution and dosing
- Enhanced bioavailability
- Reduced irritation
- Continuous drug release
- Accelerated wound healing through moisture control

II. DISEASES PROFILE

FUNGAL INFECTION
A fungal infection, also called mycosis, is a skin disease caused by a fungus. There are millions of species of fungi. They live in dirt, on plants, on household surfaces, and on your skin. Sometimes, they can lead to skin problems like rashes or bumps. Different types of fungi can cause fungal infections. In some cases, fungi typically found on or inside your body can multiply out of control and cause an infection.
Fungal infections can be contagious. They can spread from one person to another.

Some common types of fungal infection include:

- Athlete's foot
- Jock itch
- Ringworm
- Yeast infection
- Onychomycosis (fungal infection of the nail)

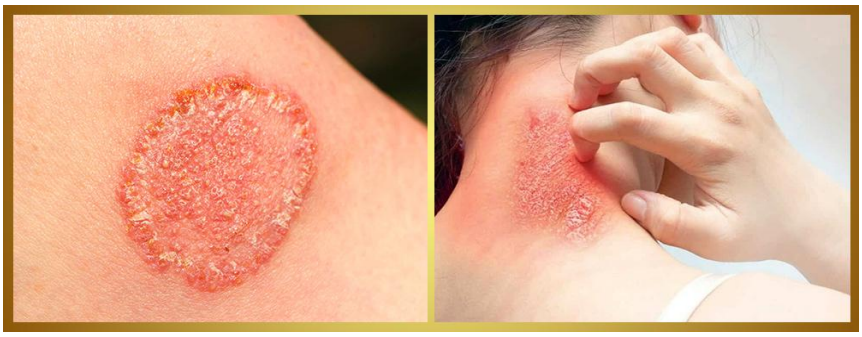

Figure 1: Fungal Infection

III. PLANT PROFILE

Senna alata (L.) Roxb. is a flowering shrub of the Fabaceae family. It has the name "candle bush" owing to the framework of its inflorescence. It is an annual and occasionally biannual herb, with an average height of 1–4 m, burgeoning in sunlit and humid zones. The leaves are oblong, with 5 to 14 leaflet sets, robust petioles (2 to 3mm), caduceus bracts (2=3/1=2cm) and dense flower (20=50/3=4) zygomorphic flowers have bright yellow colour with seven stamens and a pubertal ovary.

Figure 2: Cassia alata plant

IV. MATERIALS AND METHODS

4.1. Selection of plant:

Whole plant of cassia alata was collected from the herbal garden of Thiruvannamalai district of Tamilnadu during October 2021. The plant material was identified and authenticated by Dr. Jayaraman, ph.d. (PARC) the director of plant anatomy research centre, West Tambaram, Chennai. A specimen No.PARC/2021/4559 was preserved for future reference.

4.2. Extraction of Carica Cassia alata leaves:

A Soxhlet extractor is a piece of laboratory apparatus invented in 1879 by Franz von Soxhlet. It was originally designed for the extraction of a lipid from a solid material. However, a Soxhlet extractor is not limited to the extraction of lipids. Typically, a Soxhlet extraction is only required where the desired compound has a limited solubility in a solvent, and the impurity is insoluble in that solvent. If the desired compound has a significant solubility in a solvent then a simple filtration can be used to separate the compound from the insoluble substance.

Normally a solid material containing some of the desired compound is placed inside a thimble made from thick filter paper, which is loaded into the main chamber of the Soxhlet extractor. The Soxhlet extractor is placed onto a flask containing the extraction solvent. The Soxhlet is then equipped with a condenser. The solvent is heated to reflux. The solvent vapor travels up a distillation arm, and floods into the chamber housing the thimble of solid. The condenser ensures that any solvent vapor cools, and drips back down into the chamber housing the solid material. The chamber containing the solid material slowly fills with warm solvent. Some of the desired compound will then dissolve in the warm solvent. When the Soxhlet chamber is almost full, the chamber

is automatically emptied by a siphon side arm, with the solvent running back down to the distillation flask.
This cycle may be allowed to repeat many times, over hours or days. During each cycle, a portion of the non-volatile compound dissolves in the solvent. After many cycles the desired compound is concentrated in the distillation flask. The advantage of this system is that instead
of many portions of warm solvent being passed through the sample, just one batch of solvent is recycled. After extraction the solvent is removed, typically by means of a rotary evaporator, yielding the extracted compound. The non-soluble portion of the extracted solid remains in the thimble, and is usually discarded.

4.3 Preparation of Ethanolic extracts:
Collect fresh whole plant of senna alata were cleaned with water & shade dried until a constant weight was obtained & subsequently powdered & sieved mash no. 40. Powdered material 5kg was defatted with petroleum ether & marc was extracted with 90% of ethanol v/v at 50 degree in Soxhlet apparatus 1L for 72hr. dark brown semi - solid residues. 525g was obtained by evaporating the ethanol extract under reduced pressure.

(SENNA ALATA)
- Pulverized (coarse powder)
- Successive soxhlet extraction with ethanol 90%
- Extract concentrated in vaccum
- Stored in desicator

4.4 Formulation of Spray:

INGREDIENTS	F1	F2	F3
Plant extract	5 ml	5ml	5ml
IPA	2.25ml	2.25ml	2.25ml
IPM	1.5ml	1.5ml	1.5ml
PG	1.5ml	1.5ml	1.5ml
CARBAPOL 940	0.50gm	-	-
HPMC	-	0.75gm	-
CMC	-	-	1.0gm
METHYL PARABEN	0.5gm	0.5gm	0.5gm
MENTHOL	1gm	1gm	1gm
CITRIC ACID	0.8gm	0.8gm	0.8gm
ALCOHOL AND ACETONE	100	100	100

Figure 3: Prepared Anti-fungal spray

V. EVALUATION PARAMETERS

pH: Using the digital pH meter, the pH of the optimized spray solution was calculated. The pH meter was adjusted using phosphate buffer of different pH values (4.0, 7.0 and 9.0) before calculating the pH of the optimized formulation. The pH was determined for the spray solution. Each formulation was measured in triplicate and then the mean values were calculated. VISCOSITY: Viscosity was calculated at 25±1°C using a Brookfield viscometer (digital viscometer model). The rotation of the ULA spindle was kept at 1 rpm.

DRUG CONTENT: The solution equivalent of 10 ml was taken into a Volumetric flask (100 ml) and diluted using methanol (1000 µg/ml) up to the mark. The solution was filtered using a Whatmann filter paper. A final solution of 15 µg/ml was prepared by diluting with methanol. The absorbance of this solution was measured at 297 nm.

DRYING TIME: Evaporation time is the time needed to dry the spray film. It was measured by spraying the formulation on a glass slide and noting down the drying time.

SPRAY ANGLE: First, the distance from nozzle between papers was fixed. After that, one actuation

was sprayed onto paper and the circle size was measured. Spray angle is calculated as:

$$\text{Spray angle } (\theta) = \tan^{-1} (1 / R)$$

Where, 1 and R are the paper's distance from the nozzle and average circle radius, respectively.

5.1 ANTIFUNGAL ACTIVITY:

5.1.1 PREPARATION OF AGAR MEDIUM

- Prepare MHA from the dehydrated medium according to the manufacturer's instructions. Media should be prepared using distilled water or deionized water. Heat with frequent agitation and boil to dissolve the medium completely. Sterilize by autoclaving at 121°C for 15 minutes.
- Check the pH of each preparation after it is sterilized, which should be between 7.2 and 7.4 at room temperature. This is done by macerating a small amount of medium in a little distilled water or by allowing a little amount of medium to gel around a pH meter electrode. Cool the agar medium to 40 to 50°C. Pour the agar into sterile glass or plastic petri dishes on a flat surface to a uniform depth of 4 mm.
- Allow to solidify. Prior to use, dry plates at 30–37°C in an incubator, with lids partly ajar, for not more than 30 minutes or until excess surface moisture has evaporated. Media must be moist but free of water droplets on the surface. Presence of water droplets may result in swarming bacterial growth, which could give inaccurate results. They are also easily contaminated.

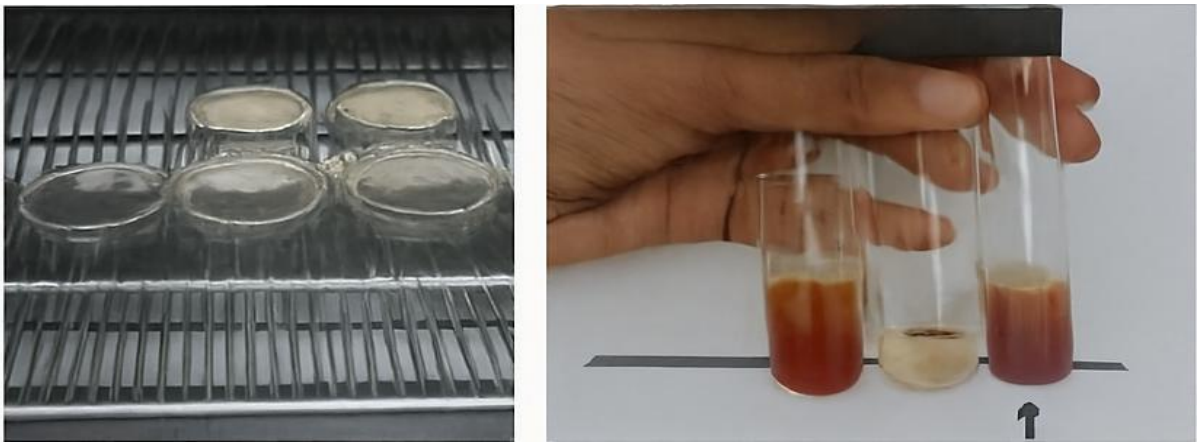

Figure 4: Prepared agar medium

5.1.2 INOCULUM PREPARATION

From a fungal culture (not more than 48 hours old, except for slow-growing organisms), take 4 or 5 colonies with a wire loop. Transfer colonies to 5 ml of trypticase soy broth or 0.9% saline. Incubate the broth at 30°C at an optimum growth temperature until it achieves or exceeds the turbidity of 0.5 McFarland standard (prepared by adding 0.5 ml of 0.048% $BaCl_2$ to 99.5 ml of 0.36% H_2SO_4; commercially available). Compare the turbidity of the test bacterial suspension with that of 0.5 McFarland (vigorously shaken before use) against a white background with a contrasting black line under adequate light. Arrow points to tube with correct turbidity. Reduce turbidity by adding sterile saline or broth.

5.1.3 INOCULATION OF PLATES

1. Dip a sterile cotton swab into the standardized fungal suspension.
2. Remove excess inoculum by lightly pressing the swab against the tube wall at a level above that of the liquid.
3. Inoculate the agar by streaking with the swab containing the inoculum.
4. Rotate the plate by 60° and repeat the rubbing procedure. Repeat two times. This will ensure an even distribution of the inoculum.
5. Allow the surface of the medium to dry for 3–5 minutes but not longer than 15 minutes to allow for absorption of excess moisture.

6.RESULTS AND DISCUSSION

6.1 EVALUATION

S.NO	PARAMETERS	F1	F2	F3
1	PH	7.09±0.05	7.20±0.04	7.19±0.01
2	VISCOSITY	3.48±0.37	3.77±0.25	4.02±0.55
3	DENSITY	0.688±0.01	0.679±0.02	0.689±0.01
4	FLAME EXTENSION	69CM	71CM	70CM
5	FLASH BACK	10CM	12CM	11CM
6	SPRAY ANGLE	220	240	220
7	LEAKAGE TEST	NO LEAKAGE	NO LEAKAGE	NO LEAKAGE
8	DRUG CONTENT	99.87±0.05	101.10±0.01	99.87±0.05

6.2 ANTI-FUNGAL TEST

S.NO	MICROORGANISMS	CONTROL	1	2	3	KETOCONAZOLE
		Zone of inhibition in mm				
1	Aspergillus flavus	-	18	08	12	10
2	Penicillium sps	-	12	10	15	17
3	Candida albicans	-	-	-	-	18

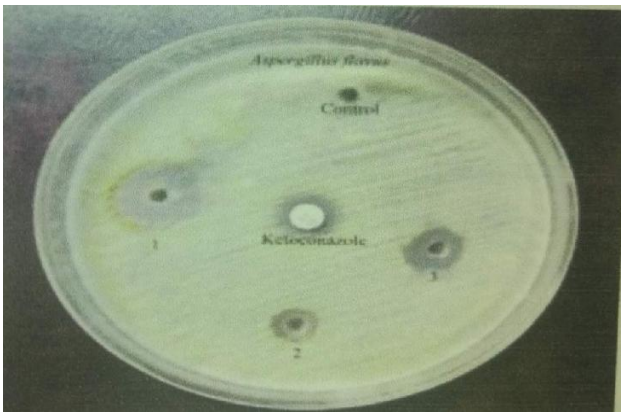

Figure 5: Aspergillus flavus

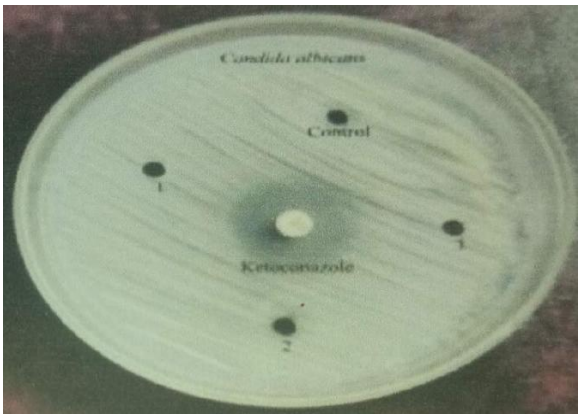

Figure 7: Candida albicans

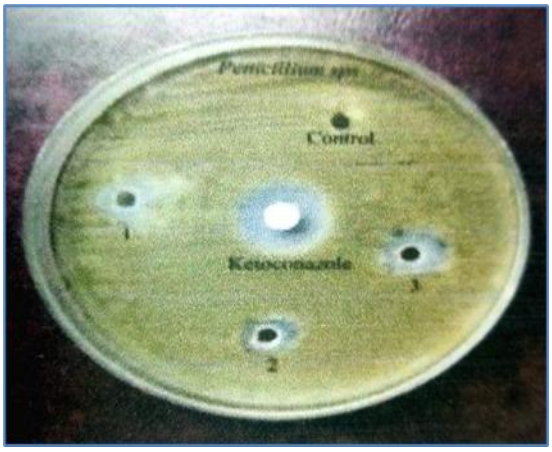

Figure 6: Penicillium sps

VII. CONCLUSION

The evidence suggests that cassia alata has antifungal action in comparison to ketoconazole. Cassia alata was successfully formulated as a solution for spray pattern which can be used in future trials as a topical formula on a number of participants. From the results obtained in the present work, it can be concluded that cassia alata can be an innovative and promising approach for topical administration. The diffusion studies indicated that the permeation of cassia alata formulations through the skin was much higher compared to the diffusion of a simple organic solution of the drug.

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