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Documentation and Comparison of Antifungal Sensitivity of *Cassia alata* Leaf and Flower Extracts against *Candida albicans*

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ABSTRACT

The plant *Cassia alata* is a shrub that has various uses ranging from mild to severe infectious and non-infectious diseases. The study was aimed at comparing the antifungal efficiency of the methanolic leaf and flower extracts of *Cassia alata* against *Candida albicans*. The leaves and flowers of the plant were collected, dried, extracted by continuous hot percolation method using methanol. The extract was used for antifungal sensitivity testing by well diffusion method at different concentrations. At 20 percent, the zone of inhibition for the leaf was 1.90 ± 1.14 and for flower was 1.10 ± 1.58 . At 40% level, the zone of inhibition for leaf was 4.10 ± 0.64 and for flower was 2.80 ± 0.84 . While at 60% level, zone of inhibition for leaf extract and flower was 5.80 ± 0.31 and 3.60 ± 0.54 , respectively and at 80% level, the zone of inhibition for leaf extract and flower was 7.90 ± 1.10 and 4.40 ± 1.10 , respectively. Whereas, the pure extract of leaf depicted 8.80 ± 0.46 and flower depicted 6.20 ± 0.68 zone of inhibition. From the results, it was observed that the leaf extract has better anti-fungal properties than the flower due to the presence of more phyto-pigment anthraquinone in leaves than flower petals.

INTRODUCTION

Ethnomedicine is part and parcel of human life since its beginning. Along the time new medicinal plants were discovered and utilized. Many plants and plant parts were being used as raw materials for extraction of active ingredients of different drugs [1] in addition to the traditional practices like Ayurveda, Siddha and Unani etc. But potentials of plants remained scientifically unexplored to a large extent, especially in Indian sub-continent. India has huge biodiversity with a total of 47,513 plant species [2] out of about 0.4 million hitherto known in the world, representing as much as 11.4% of world flora, with many medicinally valued plants which are explored and yet to be explored. Few of those are very important plants with great medicinal value, one of such plant is *Cassia alata* Linn, and every part of this herb has pharmacological importance. *Cassia alata* is cosmopolitan in existence, it is found in South and Southeast Asia, Africa, Northern Australia and Latin America [3]. It is an ornamental plant with diverse medicinal uses [4]. Phytochemical studies revealed that *Cassia alata* L. leaves contain chemicals such as alkaloids, saponins, anthraquinones, tannins, terpenes, steroids, flavonoids and carbohydrates[5,6]. *Cassia alata* is known for therapeutic properties like anti-fungal, anticancer [7], antibacterial[8] and wound-healing properties[9] etc. Even though there are several uses, there are many questions to be answered. Hence, this herb still has to be explored on scientific lines on full extent.

In the era of pollution the world is facing main threat of skin problems, in such circumstances, there are many chemically prepared drugs available in market. The incidences of increasing drug resistance in human pathogens along with unwanted side effects are most commonly encountered problems during the use of chemical antimicrobial agents. This has prompted the search for safer alternatives with promising effectiveness and safety [10]. Several reports have shown that *Cassia alata* contains antimicrobial substances [11-18] that may be responsible for its reported activity in bacterial and fungal infections. Considering these facts the present investigation is aimed at understanding the comparative effectiveness and antifungal activity of *Cassia alata* leaves and flower in case of *Candida albicans*.

MATERIALS AND METHODS

The leaves and flowers of *Cassia alata* were collected from in and around villages of West and East Godavari districts. The collected leaves and flowers were dried under shade for

seven days and grinded into fine powder by using electric blender and sieved through sieve no. 40 and this fine powder was further used for extraction process.

Process of extraction:

Continuous hot percolation process or Soxhlet extraction has been used to obtain the aliquot. The powdered leaves and flowers were weighed to 20 grams, in a round bottomed flask. Then 150 ml of methanol was added to the flask and subjected to extraction for about 8 hours. The extract was collected for further distilled to separate methanol from the crude extract. The collected extract was further concentrated by heating gently on a heating mantle. Finally, jelly like extract was obtained.

The screening of antimicrobial activity of crude extract from *Cassia alata* was carried out on active cultures of *Candida albicans*. The in-vitro evaluation of antimicrobial activity, diffusion method like agar well diffusion method [19] was used. In this method, 6mm diameter holes were made on agar plate by using sterile borer and the extract was placed in these wells. Thus micro-organisms growing in the agar get inhibited by extract. After incubation for 48 hours at 37⁰C for fungi, the diameter of the zone of inhibition surrounding the wells was measured, 0.1 ml of inoculum was added by spread plate method, inoculum was evenly spread onto the plates. The obtained yield from the Soxhlet extraction is assumed as 100 per cent aliquot from the extracts of leaf and flower. From the aliquot, the extract was diluted with methanol. From these dilutions were prepared by taking 20%, 40%, 60% and 80%. Then the plates were loaded with different concentrations (20%, 40%, 60%, 80%) of crude extract and are left at room temperature for diffusion for 20 minutes. Further, they were moved on to an incubator at 37⁰C \pm 2 for 24 to 48 hours for incubation. The zone of inhibition was obtained around the extract inoculated wells representing the suppression of growth of fungi *Candida albicans* in dextrose agar. After incubation period of 24 hours the petri plates were examined for zone of inhibition and the obtained zone was calculated using vernier calipers and expressed in millimeters.

RESULTS AND DISCUSSION

This research was undertaken to compare the anti-fungal properties of *Cassia alata* of leaf and flower. It was evident that at different percent level, mean and standard error for zone of inhibition of methanol extract against *Candida albicans* for leaf was always greater than flower in well diffusion method. The results revealed that at 20% level for leaf was 1.90 \pm

1.14 and flower was 1.10 ± 1.58 , at 40% level zone of inhibition for leaf was 4.10 ± 0.64 and for flower was 2.80 ± 0.84 . Further, at 60% level zone of inhibition for leaf was 5.80 ± 0.54 and for flower was 3.60 ± 0.54 and at 80% level zone of inhibition for leaf extract was 7.90 ± 1.10 and for the flower was 4.40 ± 1.10 , respectively. While pure extract of leaf depicted 8.80 ± 0.46 and flower depicted 6.20 ± 0.68 zone of inhibition. The results are in comparison with Timothy et.al (2012) [19] who also recorded increased efficiency against *Candida albicans* in ethanolic leaf extract of *Cassia alata*.

CONCLUSION

Anti-fungal effect of *Cassia alata* was evident due to presence of anthraquinone and other phytopigments. This was evident by the zone of inhibitions produced in saubourd dextrose agar against *Candida albicans*. The above results suggest increase in zone of inhibition with increase in concentration, results also proved that the leaf extract has better antifungal properties than the flower as depicted in the fig.1 this may be due to the presence of more phytochemicals in leaves than flower petals.

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Table 1: Comparison of leaf and flower methanolic extract of *Cassia alata* showing inhibition zones for *candida albicans*.

Concentration in percent	Mean \pm S.E zone of inhibition of methanol extract (mm)	
	Leaf	Flower
20	1.90 \pm 1.14	1.10 \pm 1.58
40	4.10 \pm 0.64	2.80 \pm 0.84
60	5.80 \pm 0.54	3.60 \pm 0.54
80	7.90 \pm 1.10	4.40 \pm 1.10
100	8.80 \pm 0.46	6.20 \pm 0.68

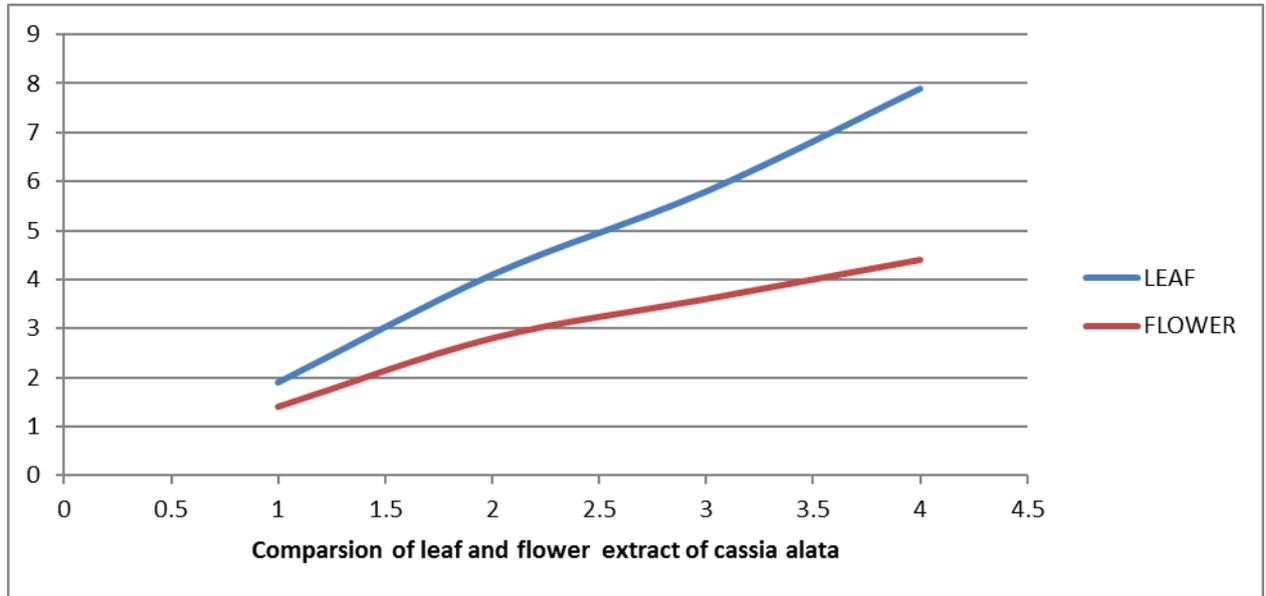


Fig 1: Graphical representation of antifungal efficiency of leaf and flower

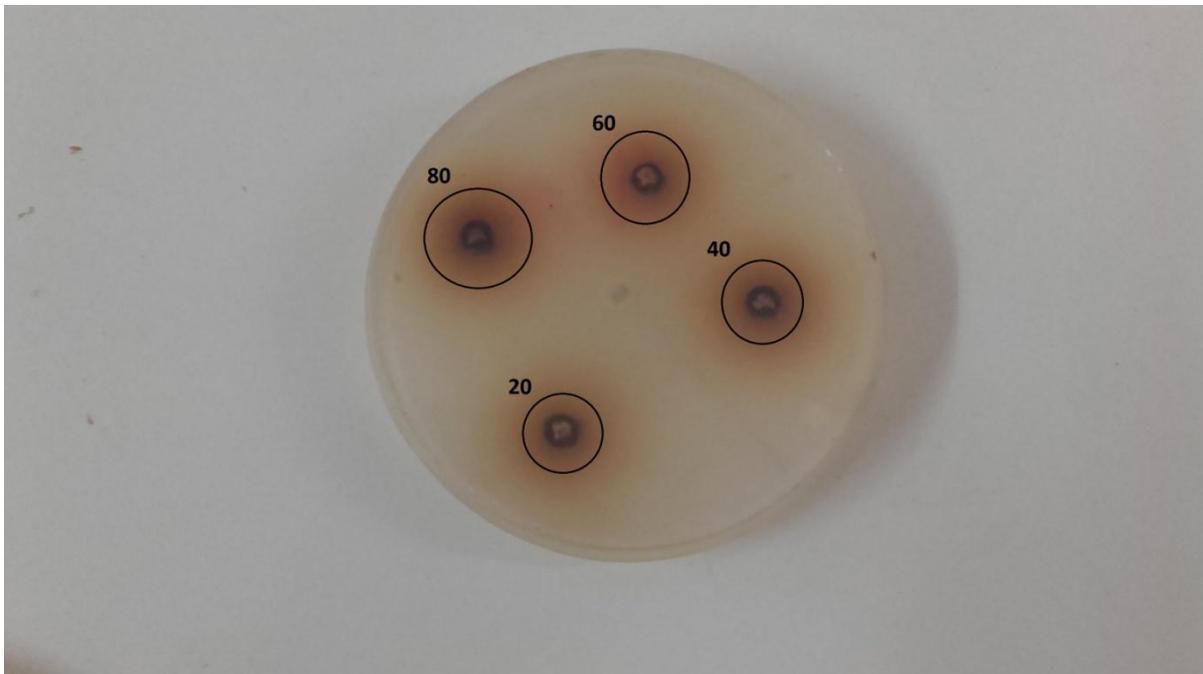


Fig-2: Zone of inhibition with methanol extract of leaves of *C. alata* against *C. albicans*



Fig-3-: Zone of inhibition with methanol extract of flower of *Cassia alata* against *Candida albicans*

