



**Effects of Different Solvent Extracts of *Cassia tora* Leaves
against Gram Positive Bacteria**

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Abstract

Cassia tora antibacterial activity was carried out to explore their effectiveness as drugs with respect to their traditional use. In this study range of solvent system was used for the better extraction of active compound which include the following solvents hexane, chloroform, ethyl acetate, methanol. The extracts from the leaves of *Cassia tora* were tested for antibacterial activity using disc diffusion method. Different concentrations of extract 5 mg and 10 mg were used, standard antibiotic penicillin was used as positive control against different bacteria and the zones of inhibition were measured in millimeters. Among the tested solvents ethyl acetate showed maximum activity against *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Bacillus subtilis* followed by chloroform extract against gram positive bacteria.

Key-Words: *Cassia tora*, Antibacterial activity, Gram positive bacteria, Skin disease

Introduction

Cassia tora is an annual herb found in dry soil in all tropical parts of India. Shrubs grow about 30-120 cm in height, they have erect stem, compound leaves, but does not have a well-developed tap root system. Leaves pinnate with glands, leaflet three pairs and ovate. Flowers are small and yellow in colour. Pods slender, long, four sided, curved, sharp pointed, 12-30 cm long, seeds 20-30 nos, grayish or pale brown, shining, about 2 mm long [1].

Cassia tora is traditionally used in India and China, for skin diseases caused by ring worm and itch. The powder is mixed with butter milk and applied on the itch to ease the irritation [2]. It is used as a mild and gentle laxative and prevents recurring diseases such as malaria, reduces/eliminates fever, kills worms in the body, helps in digestion and used to treat intestinal disorders [3]. The leaves are generally given to children who have intestinal disorders; seeds contain chrysopanic acid and valuable for treatment of skin diseases like ringworm, scabies and eczema. Roots are used as antidote against snake bite [1]. The leaves of *Cassia tora* have several anthraquinone glycosides showing anti-inflammatory and antifungal activity.

The plant is also reported to have a significant hepato-protective effect against the toxicity of carbon tetrachloride in rats [4]. In this study, we report the results of the antibacterial activity of different solvent extract of *Cassia tora* against gram positive and gram negative bacteria.

Material and Methods

Plant material

Leaves of *Cassia tora* were collected from Cuddalore and Poonamalle, Tamil Nadu, South India. It was authenticated by a plant taxonomist Preetam Raj JP from the Department of Plant Biology and Biotechnology, Loyola College, Chennai.

Extraction and isolation

Leaves were collected; shade dried at room temperature and powdered using a blender. The powder was extracted with hexane for period of 48 hrs. The extract was then filtered through a Buchner funnel with Whatman No 1 filter paper. The filtrate was evaporated to dryness under reduced pressure using rotary vacuum evaporator. The residue after hexane treatment was further extracted with chloroform, ethyl acetate, and methanol sequentially in a similar manner.

Microorganisms used

Bacillus subtilis (MTCC 441), *Enterococcus faecalis* (ATCC 29212), *Staphylococcus aureus* (ATCC 25923), *Staphylococcus epidermidis* (MTCC 3615),

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Escherichia coli (ATCC 25922) and *Proteus vulgaris* (MTCC 1771) were used for the experiment.

Antimicrobial assay

Antimicrobial activity was carried out using disc-diffusion method [5]. Petri plates were prepared with 20 ml of sterile Mueller Hinton Agar (MHA) (Hi-media, Mumbai) for bacteria. The test cultures were swabbed on the top of the solidified media and allowed to dry for 10 min. The tests were conducted at two different concentrations of the crude extract (5 and 10 mg per disc). The loaded discs were placed on the surface of the medium and left for 30 min at room temperature for compound diffusion. Negative control was prepared using Dimethyl sulfoxide (DMSO). Penicillin was used as positive control. The plates were incubated for 24 hrs at 37 °C for bacteria to grow and the zone of inhibition was measured in millimeters. The experiment was repeated twice.

Results and Discussion

Majorie [6] state that different solvents have different solubility capacities for different phytoconstituents, hence the differences in the biological activities of the various extracts. In this context the current research was conducted to screen the effect of different solvent extracts of *C.tora* leaves against the pathogenic bacteria. Sharma *et al.*, [7] has already worked on ethanolic and aqueous extract, we decided to work using other solvents, John *et al.*, [8] has worked on hexane, chloroform, ethyl acetate, ethanol extract of *C. tora*, among which hexane and ethyl acetate extract showed antimicrobial activity against *E. coli*. According to the finding of Sonia Singh [9] the aqueous extract showed maximum antibacterial action against gram positive bacteria and less action on gram negative bacteria this was found similar to our findings. In this study we tried to do the antibacterial activity using the solvent acetonitrile, which no other investigators have used earlier. Previous investigators have used various solvents such as hexane, chloroform, ethyl acetate, ethanol [8], which we also tried but could not get the results as reported by them. Our conditions were different from them and we could not find clear zones for *E. coli* in our study, we observed only morphological changes. Ethyl acetate extract of *C. tora* leaf showed maximum zone of 10 mm for *B. subtilis* and *S. epidermidis* and 11 mm for *S. aureus*. In our study, ethyl acetate and chloroform showed maximum activity against *S. aureus* and *S. epidermis* when compared to the standard penicillin. Our study proves that *C. tora* leaf extract can be used against skin diseases caused by *Staphylococcus sp.*, and it can be used against allergies caused by *B. subtilis*. Roopashree

et al [10] reported that *B. subtilis* and *S. aureus* was susceptible to aqueous extracts of *Cassia tora* and methanolic extract was effective against *S. aureus* and *E. coli*. In our screening ethyl acetate extract was found to be effective against gram positive bacteria such as *S. aureus*, *S. epidermidis* and *B. subtilis*. *E. faecalis* showed moderate activity compared to *P. vulgaris* and *E. coli*. *Staphylococcus aureus* the causative organism for various infections and is known to aggravate skin conditions like psoriasis, atopic dermatitis, erythroderma Tomi *et al.*, [11]. In the midst of resistance *Staphylococcus sp* against the existing antibacterial agents especially for treating skin diseases, herbs like *C. tora* with their potential phytochemicals are need to be explored in details for its pharmacological effect.

Conclusion

The antimicrobial activity of *Cassia tora* provides scientific source for its use as a traditional medicine. The plant can therefore be used in the treatment of skin disease, wound infections caused by the screened bacteria. Further research has to be conducted on the activity of the extracts against a wider range of bacteria and fungi and on the toxicology and further purification of the extracts for isolation of the pure active constituents.

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Fig. 1: Plant morphology

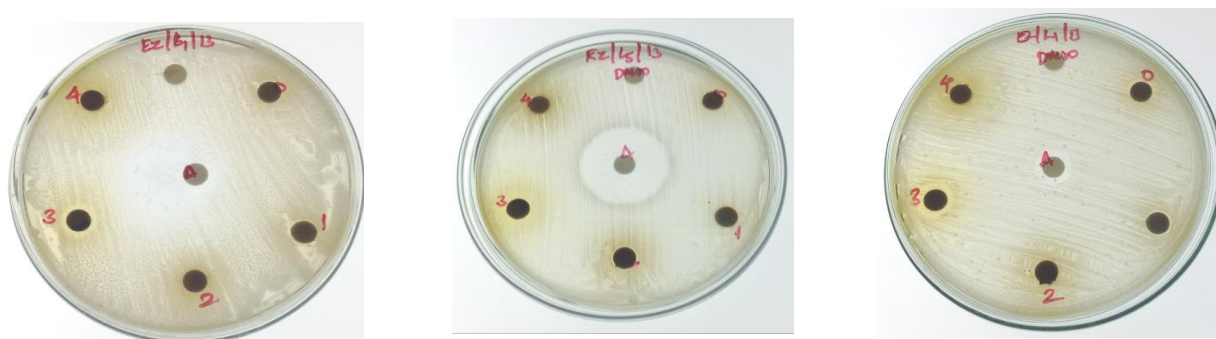


Fig. 2: A- *Bacillus subtilis*, B- *Staphylococcus epidermidis*, C- *Staphylococcus aureus*
0-Acetonitrile, 1- Hexane, 2- Chloroform, 3- Ethyl acetate, 4-Methanol, 5- DMSO, A- Penicillin

Table 1: Antibacterial activity data of *Cassia tora* leaf

Bacterial Culture	Solvent extract of <i>Cassia tora</i> leaf (mg)											
	Acetonitrile		Hexane		Chloroform		Ethyl acetate		Methanol		Control	
	5	10	5	10	5	10	5	10	5	10	Penicillin	DMSO
<i>Bacillu subtilis</i>	-	>10	-	>10	+	>10	>10	10	+	>10	10	-
<i>E. faecalis</i>	-	++	-	++	>10	>10	>10	>10	+	++	17	-
<i>S. aureus</i>	+	>10	+	>10	+	>10	10	11	++	++	>10	-
<i>S. epidermidis</i>	-	-	-	-	-	>10	10	10	+	++	17	-
<i>Escherichia coli</i>	-	+	-	+	-	+	+	>10	+	++	15	-
<i>Proteus vulgaris</i>	-	+	-	+	+	++	++	++	++	++	18	-

*DMSO-Dimethyl sulfoxide, ++ Very Good, + Good

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