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## PHYTOCHEMISTRY AND IN VITRO SCREENING OF ANTIMICROBIAL ACTIVITY OF FLOWERS OF CASSIA TORA L.

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### Abstract:

*Cassia tora is a potent medicinal plant in the Indian system of medicine. The present study mainly focused on the potential antimicrobial value of methanol plant extract of Cassia tora against pathogenic bacteria E.coli, B. subtilis, S.auresus, P. aeruginosa and fungi A. niger and C.albicans by agar well diffusion method. The results revealed that the methanol plant extract were potent in inhibiting these bacteria and fungi and this work highlights that the inhibitory effect is on par with standard antibiotics (streptomycine).*

### KEYWORDS :

Cassia tora, Phytochemistry, microorganisms, agar well diffusion method.

### INTRODUCTION :

India being a botanical garden of the world and is a gold mine of well recorded and traditionally well practised knowledge of herbal medicine. WHO encourages the traditional drugs because of its less side effect and most the European countries expanding towards Ayurvedic medicine. Since ancient times, people have been exploring the nature particularly plants in search of new drugs (Savithamma et al., 2011). Plant materials have been used for the treatment of various diseases throughout the world before the advent of modern clinical drugs. The use of medicinal plants still play an important role to cover the basic health needs in developing countries (Shrikumar and Ravi., 2007). In recent years, secondary plant metabolites previously with unknown pharmacological activities, have been extensively investigated as a source of medicinal agents. Thus, it is anticipated that phytochemical with adequate antimicrobial efficacy will be used for the treatment of bacterial infections.

Cassia tora is an annual small leguminous plant. It leaves and seeds are astringent, laxative. Also they are useful in leprosy, ringworm, constipation and cough. The dried leaves have flavonoid, glucoside. It also controls blood glucose level. Decoctions of parts of Cassia tora are used as an analgesic, anti convulsant, antipyretic, antifungal treatment of glaucoma and hypertension (Perry et al., 1980). The present study intends to study about the phytochemical and antimicrobial activity in the flowers extracts of Cassia tora against some gram negative and gram positive bacterial strains In vitro.

### MATERIALS AND METHOS :

#### Plant material:

Fresh bark of B.racemosa Lamk. were collected from the natural population growing in different areas of Parbhani district of Maharashtra. Plant was identified and authenticated from the herbarium of

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Dept. of Botany Dr. B.A.M . University Aurangabad, flora of Marathawada (Naik et al.,1998) and encyclopaedia of Indian medicinal plants (Khare, 2007).

**Preparation of extracts :**

The fresh pods was dried under shed at room temperature and then powdered with a grinder and stored in air tight container. A soxhlet apparatus was used for extraction of antimicrobial active compounds from the powder. The dried powder of the bark of the plant (20 gm) was successively extracted using methanol, ethanol, aqueous, acetone and petroleum ether solvents respectively. The collected extracts were concentrated by evaporation under room temperature and used for the study.

**Phytochemical screening :**

Different extracts were screened for the presence of alkaloids, glycosides, flavonoids, saponins, tannins, steroids, resins, phytobatanine, oil and fats by using standard protocol (Trease and Evans, 1989; Sofowora, 1982).

**Test microorganisms :**

Bacterial and fungal strain used for testing included authentic pure cultures of human pathogenic bacteria like *Staphylococcus aureus* (SRTCC1073), *Bacillus subtilis* (SRTCC1091) and two are gram negative viz. *Pseudomonas aeruginosa* (SRTCC708), *Escherichia coli* (SRTCC3260). Two species of fungi viz. *Aspergillus niger* (SRTCC1073), *Candida albicans* (SRTCC3971) These were obtained from the school of life sciences, S.R.T.M. university, Nanded (M.S.).

**Preparation of test organisms suspension :**

The test organisms were maintained on slants of medium containing nutrient agar (2.5 gm/ 10ml) and sub cultured once a week. The slants incubated at 37°C for 24 hrs and stored under refrigeration. The inoculum was  $1 \times 10^8$  cells/ml.

**Antimicrobial activity :**

The in vitro antimicrobial activity of different bark extracts of *B. racemosa* Lamk. was determined by agar well diffusion method (Perez et al., 1990). The plant extracts were dissolved in distilled water at concentrated 50 µg/ml streptomycin was used as reference antibiotic. Each plate was inoculated with 20 µl microbial suspension having concentration  $1 \times 10^8$  cells/ml. The 0.1 ml extract was added to each well. The plates containing bacteria were incubated at 37°C for 24 hrs and those containing fungi were incubated at 25°C for 7 days. Positive antimicrobial activity was based on growth inhibition zone and compared with standard drug (Perez et al., 1996). The diameter of zone of inhibition surrounding each of the well was recorded. All the experiments were performed in triplicate.

**STATISTICAL ANALYSIS :**

Results were expressed as mean  $\pm$  S.D. statistical significance was determined using analysis of students t-test.

**RESULT AND DISCUSSION :**

The results of the phytochemical screening of the different extracts of leaves were presented in Table 1. Different standard test were performed and it was revealed that the secondary metabolites such as alkaloid, carbohydrate, flavonoid, triterpenoids and saponin were present in the leaves. Whereas fats and oils, glycosides, saponins, photobatalin and anthraquinonines were found to be absent in all the extracts of the leaves. Among the different extracts methanol extracts of leaves show the presence of alkaloids, carbohydrates, flavonoids and ethanol extracts only show the presence of alkaloids. Aqueous extracts of leaves show the presence of carbohydrates, tannins, flavonoids. In acetone extracts of leaves show the presence of saponins and triterpenes.

Rejiya et al.,(2010) found that Preliminary phytochemical screening of leaf showed the presence of polyphenols which prompted researchers to evaluate its antioxidant and antiproliferative potential.

The results of the antimicrobial activities of the different extracts of flowers are presented in Table 2. The results revealed that all the extracts show antimicrobial activity. Methanol extract was found to be effective against all the test organisms. The aqueous and acetone extracts were effective against E.coli and B. subtilis. Dharmendra Dave et al., (2010), reported that the in vitro antimicrobial activity of Cassia tora shows the maximum activity against Bascillus subtilis (12mm).The inhibition zone especially on ciprofloxacin resistant was (20mm) respectively. The activity was observed on Escherichia coli (10mm) among the gram negative bacteria.

Table 2: In vitro antimicrobial activity of flowers of Cassia tora L.

Microorganisms	Diameter of Zone of inhibition in mm of different extracts of flowers (2mg/ml) Mean± S.D.					Standards reference antibiotic (Streptomycine)
	Methanol	Ethanol	Aqueous	Pet. ether	Acetone	
<b>Bacteria</b>						
<i>Escherichia coli</i>	24±0.1 4	13±0.2 1	20±0.1 0	11±0.1 5	15±0.2 1	14±0.4 1
<i>Staphylococcus aureus</i>	21±0.2 1	12±0.2 4	12±0.1 5	13±0.1 2	10±0.2 0	12±0.2 3
<i>Bacillus subtilis</i>	22±0.1 2	15±0.2 0	15±0.5 4	18±0.1 5	18±0.2 1	16±0.1 3
<i>P.aeruginosa</i>	25±0.2 3	12±0.2 1	12±0.3 5	11±0.1 5	14±0.1 4	18±0.3 6
<b>Fungi</b>						
<i>Aspergillus niger</i>	20±0.2 1	14±0.5 4	18±0.31	13±0. 17	12±0.10	19±0.22
<i>Candida albicans</i>	10±0.1 2	11±0.2 1	15±0.21	10±0. 18	12±0.25	18±0.16

Table.27. Phytochemical analysis of leaves of Cassia tora L.

Sr. No.	Phyto Constituents	Chemical tests	C. tora leaves extract				
			P.E	Met.	Eth	Aq	Ac
1	Alkaloids	1. Mayer's test	-	+	+	-	-
		2. Dragendroff's test	-	+	+	-	-
		3. Wagner's test	-	+	+	-	-
		4. Hagers test	-	+	+	-	-
2	Carbohydrates	1. Molisch's test	-	+	-	+	-
		2. Benedicts test	-	+	-	+	-
		3. Fehling's test	-	+	-	+	-
3.	Glycosides	1. Modified Borntragers	-	-	-	-	-
		2. Legal test	-	-	-	-	-
4	Saponins	1. Foam test	--	-	-	-	-
		2. Froth test	-	-	-	-	+
5	Triterpenes	1. Salkowski test	-	-	-	-	+
		2. Libermann Burchard	-	-	-	-	-
		3. Tschugajew test	-	-	-	-	-
6.	Fats & Oil	1. Stain test	-	-	-	-	-
7	Tannins	1. Alkaline Reagent	-	-	-	-	-
8	Flavanoids	1. Gelatin test	-	+	-	+	-
		2. Lead acetate test	-	+	-	+	-
		3. Shinoda test	-	+	-	+	-
		4. Zn-Hcl reduction	-	+	-	+	-
9	Photobatalin		-	-	-	-	-
10	Anthraquinonines		-	-	-	-	-

( - : Not present , + : Present)

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