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Research Article

EVALUATION OF LEAF AND STEM EXTRACTS FROM *CASSIA GLAUCA* L. FOR ANTIMICROBIAL ACTIVITY

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ABSTRACT

India is endowed with a rich wealth of medicinal plants. Approximately 80% of world population still relies on traditional medicines for the treatment of common illnesses. Rural people were more susceptible for diseases but they couldn't afford synthetic drugs due to their higher import cost. Although large numbers of synthetic antimicrobial agents have been discovered, they have side effects and pathogenic microbes are constantly developing resistance to these agents. *Cassia glauca* leaves and stem are traditionally claimed for various biological activities, and the plant is being utilized without any scientific validation. Hence, in the present study leaves and stems of *C. glauca* have been evaluated for their antibacterial activity against the disease causing organism *S. aureus, E. coli, E. faecalis* and *Klebsiella* and antifungal activity against *A. fumigatus* and *C. albicans* using petroleum ether, ethanol by serial dilution method. All the extracts of *C. glauca* leaf and stem were found to be more potent anti-bacterial against *S. aureus,* and *E. faecalis* (MIC< 0.195 mg/ml) when compared *Klebsiella* and *E. coli.* (MIC= 0.391 – 100 mg/ml). They also exhibited potent anti-fungal activity against the *C. albicans* (MIC< 0.195 mg/ml) and *A. fumigates* (MIC 0.195 –1.562 mg/ml). Thus the result of present work supports the traditional use of *Cassia glauca* and can be further explored for their potential therapeutic use.

Keywords: *Cassia glauca*, Antimicrobial activity, Plant extracts, Serial dilution.

INTRODUCTION

The herbal products today symbolize safety in contrast to the synthetics that areregarded as unsafe to human and environment. Although herbs had been priced for their medicinal, flavoring and aromatic qualities for centuries, the synthetic products of the modern age surpassed their importance, for a while. However, the blind dependence on synthetics is over and people are returning to the naturals with hope of safety and security (Joy *el al*., 2001). Many reports have shown that some *Cassia* species contain anti-bacterial, anti-diabetic, antimalarial, anti-carcinogenic and hepato protective substances (Nanasombat *el al*., 2009; Tona1, 1999; El-Sawi, 2010: Sharma 2000). The genus *Cassia* is closely related to Mimosaceae and Papilionaceae, but can be distinguished by few stamens and five free petals. Caesalpinioideae consist of trees, shrubs and a few woody herbs found in the tropics. Economically, woody Caesalpiniaceae is important for its timber. This diverse genus is native throughout the tropics, with a small number of species reaching into temperate regions. The number of species is usually estimated to be about 260 (Marazzi, 2006) but some authors believe that there are as many as 350 (Randell and Barlow, 1998). About 50 species of *Senna* are known in cultivation (Huxley *el al*., 1992).This genus is distributed all over India, Pakistan, Cylon, Malaysia, China and South Africa (Chopra *el al*., 1956; Jafri *el al*., 1999; Kritikar *el al*., 1990). About 26 species

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of *Cassia* have been reported to contain anthrocene derivatives either in their free form or as glycosides. The anthraquinone molecules are widely distributed in the genus *Cassia* and have remarkable biological properties.

Cassia glauca Lam. is about 10 m h, evergreen shrubs, found throughout India, tropical Asia and Australia. The leaves are long linear, acute, curved in shape. The flower is yellow in color and shorter than the leaves (Kirtikar and Basu, 1944). Phytochemical study of stem of *Cassia glauca*has been indicated the presence of chrysophenol, physcion, stearic acid, β-sitosterol and β-D glucoside (Hemlat and Kalidhar, 1994). In folk medicine, bark and leaves of *C. glauca* are used for the treatment of diabetes and gonorrhea. The leaves are used for blennorrhagia (Kirtikar and Basu, 1944). Bark and leaves are used in diabetes and gonorrhea in folk medicine. This plant is also a good pollution tolerant and reduces chemical pollutants from atmosphere (Warrier *el al.,*….). In the present study leaves and stems of *Cassia glauca* have been evaluated for their antibacterial activity under laboratory conditions.

MATERIALS AND METHODS

Plant material and extracts preparation

The leaves of *Cassia glauca* were collected from in and around Bagalkot Dist located in North Karnataka region, in the month of September–October. A voucher specimen (BSC/Pharmacy/ 2012/1/12) was stored in the department for future reference. Leaves and stem were shade dried at room temperature. The shade dried and coarsely powdered plant material were successively extracted with petroleum ether (60-80° C), Chloroform and ethanol using Soxhlet apparatus. The extracts were dried under reduced pressure at temperature of 30° C to dryness to yield dried extract residue.

Antibacterial and antifungal activity

All the extracts were evaluated for antimicrobial activity against few clinical isolates, by serial dilution method in duplicate (Koneman, 1995). Antimicrobial activity tested against *Staphylococcus aureus* (ATCC 25923), *Enterococcus faecalis* (ATCC29212), *Klebsiella (*ATCC-1705), *Escherichia coli* (ATCC 25922) and antifungal activity against *Aspergilus fumigatus* (ATCC102) and *Candia albicans* (ATCC10231).They are grown on blood agar media, sub cultured and isolated. On the other hand control strains of same organisms were also developed in suitable culture media. The inoculum of both control strains and clinical isolates were standardized by adjusting to McFarld scale (0.5) using Muller-Hinton (10^5CFU/ml). Ciprofloxacin and Fluconazole were used as reference standard. The plant extracts were initially dissolved in minimum quantity of DMSO and then were added to Muller-Hinton broth to reach final concentration of 1mg/ml, 300µl of these extracts were added to first and second tubes further dilutions were made from second tube to ninth tube using 2 fold dilution technique, so that the highest and lowest concentration of each extracts were 300 µl and 0.6 mg/ml respectively. To each of these tubes 100 µl of microbial culture (105CFU/ml) was added and incubated for 24 hrs at 37° C, and were examined from bottom using reflective viewer. The lowest growth was recorded as MIC for each organism.

RESULTS AND DISCUSSION

From antibacterial studies we can find out that all extracts showed anti-bacterial activity against all the bacterial isolates tested. Leaf extracts using Petroleum Ether showed inhibition against *E.coli* (MIC590 mg/ml), followed by *S. aureus* (0.236 mg/ml) and *E. faecalis* (0.287 mg/ml). Chloroform extract showed inhibition against *E. coli* (0.675 mg/ml), followed by *Klebsiella* (0.385 mg/ml), *E. faecalis* (0.259 mg/ml) and against *S.aureus* (0.192 mg/ml). Ethanol extract showed inhibition against *E. coli* (1.030 mg/ml), *Klebsiella* (0.408 mg/ml), *E. faecalis* (0.250 mg/ml), and *S. aureus* (0.239 mg/ml).

Stem extracts using Petroleum Ether showed inhibition (MIC) against *E.coli* (0.903 mg/ml), followed by *Klebsiella* (0.351mg/ml), *E. faecalis* (0.260 mg/ml) and *S. aureus* (0.237 mg/ml).

Chloroform extract of stem showed inhibition against *E. coli* (0.796 mg/ml), *Klebsiella* (0.444 mg/ml), *E. faecalis* (0.216 mg/ml), and *S.aureus* (0.214 mg/ml). Ethanol extract showed inhibition against *Klebsiella* (0.795 mg/ml), *E.coli* (0.526 mg/ml), *S. aureus* (0.186 mg/ml), and *E. faecalis* (0.169 mg/ml) (Table 1).

The leaf and stem extracts of *C. glauca* were shown good inhibition against fungi organisms. Petroleum Ether Leaf extract showed inhibition (MIC) zone against *C. albicans* (0.436 mg/ml) and *A. fumigates* (0.322 mg/ml). Chloroform extract showed inhibition against *C. albicans* (0.441 mg/ml) and *A. fumigatus* (0.320 mg/ml). Ethanol extract showed inhibition against *C. albicans* (0.458 mg/ml) and *A. fumigatus* (0.351 mg/ml). Petroleum Ether Stem extracts showed inhibition (MIC) zone against *C. albicans* (0.430 mg/ml) and *A. fumigatus* (0.329 mg/ml). Chloroform extract showed inhibition against *C. albicans* (0.437 mg/ml) and *A. fumigatus* (0.371 mg/ml). Ethanol extract showed inhibition against *C. albicans* (0.42 8 mg/ml) and *A. fumigatus* (0.305 mg/ml) (Table 2). Antimicrobial and antioxidant activity of seeds of different extracts of *Cassia glauca* as well as their fatty acid composition of petroleum ether extract has been well documented (Deepak Kumar *et al.,* 2013). Several studies have been shown that extracts of *C. glauca* leaf improved blood sugar level and normalization of liver functions (Farswan *et al.,* 2011).

Table 1. Anti-bacterial activity of the plant extracts.

Plant material	Extracts	MIC (mg/ml)			
		Staphylococcus aureus	*Enterococcus faecalis*	*Klebsiella*	*Escherichia coli*
Cassia glauca Leaf extract	Petroleum Ether	0.236	0.287	-	0.590
	Chloroform	0.192	0.259	0.385	0.675
	Ethanol	0.239	0.250	0.408	1.030
Cassia glauca Stem extract	Petroleum Ether	0.237	0.260	0.351	0.903
	Chloroform	0.214	0.216	0.444	0.796
	Ethanol	0.186	0.169	0.795	0.526

Table 2. Anti-fungal activity of the plant extracts.

Plant material	Extracts	MIC (mg/ml)	
		Aspergilus fumigata	*Candida albicans*
Cassia glauca Leaf extract	Petroleum Ether	0.322	0.436
	Chloroform	0.320	0.441
	Ethanol	0.351	0.458
Cassia glauca Stem extract	Petroleum Ether	0.329	0.430
	Chloroform	0.371	0.437
	Ethanol	305	0.428

CONCLUSION

On the basis of our studies we find that plant extracts are active against one or more tested bacterial and fungal diseases. The knowledge of medicinal plants is limited to traditional healers, herbalists and elderly persons who love in rural areas. This study also points out that certain species of medicinal plants are being exploited by the local residents who are unaware of the importance of medicinal plants in the ecosystem as best antifungal agent. The Ethanolic extract of *Cassia glauca* exhibited high antioxidant and antimicrobial properties due to it have high phenolic content which may be responsible for these activities. This finding provides an insight into the usage of the leaves of Cassia species in traditional treatment of wounds or burns associated with bacterial and fungal infections. However, further work is needed in the form of phytochemical screening and pharmacological activity of some more extracts before one could conclude anything definite about the therapeutic potential of these extracts.

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