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Research Article

Phytochemical screening and antibacterial activity of *Bombex ceiba*Srikanth M ^{1, *} and Ganga Rao B ²

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Abstract

Bombex ceiba is a traditional medicinal plant used by tribal people in treatment of different diseases. The present carried out for providing the scientific evidence of its traditional medicinal usage on antibacterial capacity and presence of different phytochemical constituents on *B. ceiba* seeds. Qualitative phytochemical screening, quantitative estimation of total phenolics and alkaloids was carried out using different standard test procedures and antibacterial activity was tested using cup plate method and measured its capacity on zone of inhibition measurement. The all extracts of *B. ceiba* revealed the presence of phenols, alkaloids, carbohydrates, steroids, terpenoids and glycosides and gave negative result to saponins. The hydroalcoholic and ethyl acetate extracts revealed the presence of flavonoids and tannins but the hexane extract gave negative results. The Quantified phenolic contents of *B. ceiba* extracts were ranging from 13.85±1.22 to 34.10±2.62 (mg/gm). The quantitative alkaloid content was ranging from 16.24±2.38 to 31.86±1.88 (mg/gm). All the extracts (hexane, ethyl acetate and hydroalcoholic) of selected medicinal plants at different concentrations (50μg, 100μg, 150μg and 200μg/cup) exhibited antibacterial activity along with standard drug (Rifampicin) against tested bacterial strains. Ethyl acetate and hydroalcoholic extract showed equal antibacterial activity on all bacterial strains.

Keywords: Bombex ceiba, Seeds, Phenolics, Alkaloids and Antibacterial activity.

1. Introduction

The relationship between humans and plants has been close throughout the development of human culture and they are dependent on plants for survival like food, shelter and in the health maintenance. There were many evidences and reports about the use of natural products including plants usage in the treatment of different diseases in ancient societies. The Rig-Veda, which is the oldest book of library of man, supplies the information about some herbs (De Pasquale, 1984; Kokate et al., 2002). The synthesis of many chemicals and their introduction into therapeutics as drugs certainly revolutionized the treatment of diseases. Today we have a large number of synthetic drugs which are effective in different diseased conditions. However, it is a known fact that these drugs are not fully safe to humans and the disease causing microorganisms getting resistance towards current drugs and they produce a large variety of adverse reactions and have been the cause of a number of diseases (Boursi et al., 2015).

Plant derived natural products hold great promise for discovery and development of new

drugs in treatment of different diseases (Mc Chesney et al., 2007). However, a large number of medicinal plants have not been studied in detail for their chemical constituents and pharmacological properties and use in different diseases and prevention of causing microorganisms. In this point of view, we selected the Bombax ceiba plant for the present study. Bombex ceiba is a traditional medicinal plant used by tribal people in different situations for example used as an abortifacient by the Oraon tribe in West Bengal (Mitra and Mukharjee, 2009), to cure gonorrhoea, impotency, spermatorrhea, sterility, nocturnal emission and leucorrhoea (Kosalge and Fursule, 2009), seeds and roots of B. ceiba were used in the treatment of leprosy (Mollik et al., 2009) belongs to the family Malvaceae, widely distributed at tropical and subtropical regions (Smith et al., 2004; Parrotta, 2001). There is less work reported on the seed extracts of B. ceiba. So, B. ceiba seeds were selected to extract with different solvent and to evaluate their phytochemical analysis and Antibacterial capacity.

2. Materials and Methods

2.1 Collection of plant material and preparation of extracts

The plant material seeds were collected at Araku valley, Visakhapatnam district, Andhra Pradesh, India and the plant was authenticated by taxonomist Prof. M. Venkaiah, Department of Botany, Andhra University. The collected seeds was shade dried and milled into powder. The powdered material was used for extraction with different solvents successively (Hexane, Ethyl acetate, and Hydro alcoholic) using maceration process. Then the extracts were used for further study.

2.2 Chemicals and test bacterial species

Muller Hinton agar media was purchased from Sisco Research Laboratories Pvt Ltd., Mumbai. The other chemicals were analytical grade. The microorganisms used for the experiments were procured from MTCC, IMTECH, Chandighar.

Gram +ve organisms:

Bacillus megaterium (B. m), Staphylococcus epidermidis (S. e), and Lactobacillus acedophillus (L. a).

Gram-ve organisms:

Escherichia coli (E. c), Salmonella typhi (S. t) and Klebsiella pneumonia (K. p).

2.3 Qualitative and Quantitative Phytochemical Screening

Qualitative phytochemical screening was carried out using different standard phytochemical tests for different compounds (Kokate 1991; Prashant Tiwari *et al.*, 2011) Quantitative estimation of phenols and alkaloids were carried out using Folin-Ciocalteau reagent (Mallikarjuna Rao *et al.*, 2012) and Bromocresol Green solution (Fazal Sharma *et al.*, 2008).

2.4 Culture Media for anti bacterial activity

The Bacterial species were maintained in the nutrient broth medium on placing shaker in separate culture tubes for each species separately. For Anti bacterial activity Muller-Hinton Agar media was used.

2.5 Standard and test solution preparation

The test compounds (dried extracts) at a

concentration of 50, 100, 150 and $200\mu g/mL$ were dissolved in dimethylsulphoxide and used as stock solution. The reference standard (Rifampicin) as 0.6mg/mL concentration in HPLC grade water and finally added $100\mu L$ in each cup of Petri dish during antibacterial activity.

2.6 Evaluation of antibacterial activity

The antibacterial activity was assayed using cup/cylinder plate method. The method was based on capacity of different drugs by zone of inhibition (Size in mm) on microbial growth on Petri dish (Indian Pharmacopoeia., 1996; Ganga Rao et al., 2012). The tested microorganisms were spread on different plates using spread plate technique, on those plates 4 wells with 4mm diameter were placed using sterile borers. Accurately measured (100µl) solution of each concentration and reference standards were added to the cups with a micropipette and placed at 2-8°C for effective distribution of testing/standard compounds in wells. Later, they were incubated at 37°C for 24 hours, then Petri dishes were observed for presence or absence of definite zone of inhibition. If any zone of inhibition around the well (cup) indicates presence of antibacterial activity. At the same time, the vehicles (DMSO, HPLC grade water) were alson tested for antibacterial activity.

3. Results and Discussion

Qualitative phytochemical screening of B. ceiba extracts revealed the presence of different phytochemical constituents like steroids, terpenoids, flavonoids. alkaloids, glycosides, tannins. carbohydrates, oils and amino acids. The extracts gave negative results for the quinines and saponins. The all extracts of *B. ceiba* revealed the presence of phenols, alkaloids, carbohydrates, steroids, terpenoids and glycosides and gave negative result to saponins. The hydroalcoholic and ethyl acetate extracts revealed the presence of flavonoids and tannins but the hexane extract gave negative results. The hexane and ethyl acetate extracts reveals the presence of minute amount of oils hydroalcoholic extracts gave negative results. All the extracts gave negative result to amino acids but

the hydroalcoholic extracts give minute result for the presence of amino acids. The results were shown in table 1.

Phenolic contents of *B. ceiba* extracts were ranging from 13.85±1.22 to 34.10±2.62 (mg/gm). The hydroalcoholic extract have more phenolic content i.e. 34.10±2.62 (mg/gm) than other extracts. As phenolic contents, alkaloid content was vary from 16.24±2.38 to 31.86±1.88 (mg/gm). The hydroalcoholic extract has more alkaloid content i.e. 31.86±1.88 (mg/gm) than other extracts. The results were shown in table 2.

All the extracts (hexane, ethyl acetate and hydroalcoholic) of selected medicinal plants at different concentrations (50µg, 100µg, 150µg and 200µg/cup) exhibited antibacterial activity along with standard drug (Rifampicin) against tested bacterial strains (Table 3). The antibacterial potency of different medicinal plants extracts depends on type of extraction and nature of components in them and sensitiveness of tested strains. Significantly higher antibacterial activity was observed with hydroalcoholic extract, whereas least activity was observed in case of hexane extract with intermediate values for ethyl acetate extract. Both gram positive and gram negative bacteria were susceptible to selected plants extracts which supports the earlier reports that plant extracts were most active against bacterial strains. Hexane extract of Bombax ceiba seeds showed zone of inhibition on B. megaterium, K. Pneumonia and S.typhi at 50µg/cup. Ethyl acetate and hydroalcoholic extract showed equal antibacterial activity on all bacterial strains but hydroalcoholic extract showed better activity on K. Pneumonia (15mm) and B. megaterium (16mm) strains.

The current study outcome, demonstrates that *Bombax ceiba* seeds posses the significant and considerable antibacterial activity on different bacterial strains as currently using drugs and contain biological active compounds (Phenolics, Alkaloids, Steroids, Glycosides, Flavanoids and Terpenoids) which are effective in resisting the growth of the pathogenic bacteria (David *et al.*, 2011; Rajeswari *et al.*, 2014; Newman *et al.*, 2003)

and further studies are useful in isolation of drugs from these plants for many diseases.

Table 1. Nature of phytoconstituents presents in different extracts of *Bombax ceiba*.

	Bombax ceiba					
Phytochemical constituents	Hexane extract	Ethyl acetate extract	Hydro alcoholic (70%)			
Phytosterols	+	+	++			
Terpenoids	+	+	+			
Glycosides	+	++	++			
Saponins	-	-	-			
Flavonoids	-	+	+			
Tannins	-	+	+			
Carbohydrates	+	+	+			
Alkaloids	+	+	++			
Amino acids	-	-	+			
Oils	+	+	+			
Phenols	+	+	++			

^{+,++=}Present, - = Absent (+=Less Intense; ++= More Intense)

Table 2. Total phenolic and alkaloid contents (mg/gm) of *Bombax ceiba* extracts.

Name of the	Total Phenolic	Total alkaloid		
extract	content (mg/gm)	content (mg/gm)		
Hexane	13.85±1.22	16.24±2.38		
Ethyl acetate	26.28 ± 0.66	22.40±1.36		
Hydro alcoholic	34.10±2.62	31.86±1.88		

Table 3. Antibacterial activities of *Bombax ceiba* seeds extracts.

Name of	dose	Zone of inhibition [#] (in mm)					
the	$(\mu g/$	gram (+)ve			gram (–)ve		
extract	cup)	S.e	B.m	L.a	E.c	S.t	K.p
Hexane extract	50	-	6	-	-	7	6
	100	-	7	7	7	9	8
	150	7	8	9	9	11	10
	200	8	10	11	11	14	12
Ethyl acetate extract	50	7	6	6	-	6	-
	100	9	8	9	7	8	7
	150	11	10	11	9	10	9
	200	13	12	13	11	13	10
Hydro alcoholic extract	50	-	7	6	7	6	7
	100	6	9	8	9	8	10
	150	8	12	11	11	10	12
	200	10	16	13	13	12	15
Rifampic in	50	22	20	19	23	24	21
DMSO	100µ1	-	-	-	-	-	-

⁻⁼No activity; #Values Includes the cup diameter (4mm)

4. Conclusion

The present study provide the scientific evidence to the *Bombax ceiba* plants traditional medicinal usage in the treatment of diseases.

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Conflicts of interest

Author has none to declare.

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