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ORIGINAL RESEARCH ARTICLE

Phytochemical and Antimicrobial Activity of Selected Microorganism of Bark Extract of the Plant *Crataeva religiosa*

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ABSTRACT

Phytochemical and Antibacterial activity of chloroform, ethanol & Hexane extract of *Crataeva religiosa* bark samples was studied with respect to three pathogenic bacterial species *Enterococcus faecalis*, *E. coli & Staphylococcus*. The Ethanolic extract of bark was effective than chloroform and Hexane extract. The bacterium *Enterococcus faecalis* has more inhibition than two other bacterial species studied. Preliminary phytochemical analysis revealed the presence of alkaloids, glycosides, flavonoids, tannin, saponins, steroids & anthroquinone glycosides. Among the phytochemicals, Saponins, glycosides and Anthroquinone glycosides was shown best in all extracts.

Key words: Antibacterial activity, Phytochemical analysis and Crataeva religiosa.

1. INTRODUCTION

Use of herbal medicines in India represents a long of interactions with the environment. Plants used for traditional medicine contain a wide range of substances that can be used to treat chronic as well as infectious diseases ^[1]. The search for herbal remedies to relieve pain and heal diseases drove early man to explore the natural resources surroundings him particularly the plants. Most of the plants are major sources of natural products used as pharmaceuticals, agrochemicals, flavor and fragrance ingredients, food additives and pesticides. Large number of primary metabolites acts as precursors of pharmacologically active metabolites in pharmaceutical compounds for the synthesis of drugs. Millions of the people in the third world still use the herbal drugs ^{[2].} History reveals that many medicines that we use today were isolated from plants sources. Approximately 25-33% of currently available modern medicines in the United States have their origins in plants, Animals or mineral systems. Biotechnologically derived and synthesized medicines have renewed interest to pay attention on herbalism^[3].

Crataeva religiosa hook & frost belonging to family capparidaceae (*cappaceae*) is a tree usually found in the vicinity of temples of central and eastern India^{[4] [5].} It is known as pasugandha in

sanskrit, three legs in capper in English, Varuna in hindi. Crataeva religiosa is globally distributed in India, Myanmar, Sri Lanka, Malaysia, Indonesia and China. In India, it is found in Peninsular India, Western India, Gangetic Plains, and Eastern India, up to Tripura and Manipur^[6]. The plant part used for the medicinal purpose includes Leaves, stem bark and Root bark ^{[5][7]}. Plant is used ethnopharmacologically as diuretic, laxative, lithonotriptic, antirehumatic, antiperiodic, bitter tonic, rubifacient and counterirritant ^{[5] [7]}. The bark is used in the urinary disorders including kidney and bladder stones, antiemetic, and calculous affections and as an antidote in snake bite ^[5]. Scanty literature is available on bactericidal properties of bark of this plant and hence the plant is screened for the bactericidal potential.

2. MATERIALS AND METHODS

2.1. Collection and processing of the plant material

Bark samples of *Crataeva religiosa* were collected from the Nagamalai Hills, Arachalur, Erode district. The sample was authenticated by Dr. S. Saravana Babu, reader in botany, Chikkaiah Naicker College, Erode. The bark samples were cut into pieces, Sun-dried to reduce the moisture level. After the completion of drying, the plant material was pulverized to get coarser powder material, which was stored in air tight plastic container.

2.2. Microorganisms

Staphylococci, Enterococcus and *E. coli* were used for testing antibacterial activity of bark extracts. The test organisms used in this study were obtained from the Biiogenic Institute, Namakkal.

2.3. Qualitative screening of phytochemicals

Different extracts were screened for the presence of alkaloids, glycosides flavonoids, saponins and terpenoid by using standard protocols ^{[8,[9]}.

2.4. Preparation of media

Accurately weighed 28g of nutrient agar (Himedia) was dissolved in the 1000ml of distilled water. The medium was sterilized under 15Lb pressure for 15minutes in an autoclave. 30ml of this sterilized semisolid nutrient agar medium was poured in pre-sterilized 90mm glass petriplates under aseptic conditions in laminar flow. The plates were allowed to cool at room temperature to solidify the medium.

2.5. Determination of antibacterial activity by agar well diffusion method

Agar well diffusion method was employed to determine antibacterial activity. ⁽¹⁰⁾ A loop full of culture was taken from the broth. The culture was swabbed evenly on Muller-Hinton gas. The plates were air dried. Wells were prepared using a gel puncture. Different concentrations of crude extracts of medicinal plants were added on different wells. The plates were incubated aerobically at 37 °C for 24 hrs. Diameters of zones of inhibition were measured (in mm).The data were recorded.

3. RESULTS AND DISCUSSION

Phytochemical screening of ethanol, chloroform and hexane extract of bark revealed the presence of some secondary metabolites like alkaloid, glycosides, flavonoids, saponins, steroid, and tannins. The concentration of saponin, glycosides and anthroquions glycosides is high in all three extracts (Table -1) than other secondary metabolites. Antibacterial activity of bark extracts of C. religiosa against 3 different organisms is shown in (Table 2). From the table 2 it is clear that the Enterococuss faecalis shows maximum zone of inhibition against extracts than Staphylococcus aureus and E.coli.

Bijase *et al* ^[11] reported that three known isoflavonoids from methanolic extract of root bark and eight known isoflayonoids from the stem bark of Bolusanthus specious which exhibits antibacterial activity against. Owalabi et al. [12] reported antibacterial activity of ethanolic extract of kigelia africana stem bark against S.aureus (15mm). According to Mann et al ^[13] noticed that the root bark, stem bark and leaves Terminalia avicennioides extracted in ethanol exhibits bactericidal potential against *E.coli*. Abusaanab^[14] investigated that the crude ethanolic extracts of S.officinals, S.aromaticum, C.verum, R.officinals T.valgaris, S.officinals/ R.officinals and R.officinals / T.valgaris inhibited the growth of multi drug resistant bacterium P.aeruginosa. The bark extract was found to be containing glycosides, alkaloids, tannin steroids and flavonoids which are biologically active ^{[15].} Tannin from Dichrostachys cinerea root bark possesses antibacterial activities against S.aureus, *E.coli and P.aeruginosa*^[16]. Flavonoids have been reported to have both antibacterial and antifungal activities ^[17]. In our study all three extracts revealed the presence of secondar y metabolites in different concentration.

The different rates of inhibition may be probably due to the quantity of the phytochemicals present in the extracts ^[18]. Thus, it is important to know the biochemistry of stem bark in order to isolate and screen the new pharmacological active principals.

S. No	Tests	Extraction				
		Chloroform	Ethanol	Hexane		
1	Tannin	++	++	++++		
2	Saponin	+++	+++	+++		
3	Flavonoid	+	+	+		
4	Steroid	+++	+	+		
5	Glycosides	+++	+++	+++		
6	Alkaloid	+	++	++		
7	Anthroquinone glycosides	+++	+++	+++		

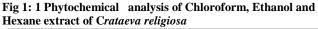
Table 1:Phytochemicalanalysis of Chloroform, Ethanol andHexane extract of Crataeva religiosa

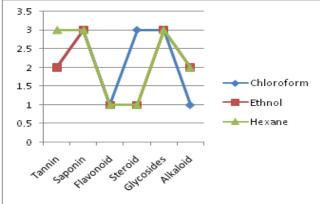
+++: Present in high concentration, ++: Present in moderate concentration, +: Present in low concentration

 Table 2: Antibacterial activity of crataeva religiosa

S. No	Micro-organism	Extracts	Zone of inhibition in mm			
			25 µL	50 µL	75 µL	100 µL
1.	Enterococcus	Chloroform	1	2	4	6
	faecalis	Ethanol	2	3	5	8
		Hexane	2	4	6	7
2.	E.coli	Chloroform	-	1	2	4
		Ethanol	1	2	3	5
		Hexane	-	2	3	4
3.	Staphylococcus	Chloroform	1	2	4	5
	aureus	Ethanol	1	3	4	6
		Hexane	1	2	3	4

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