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RESEARCH ARTICLE

PHYTOCHEMICAL SCREENING AND ANTIBACTERIAL ACTIVITY OF THE SEEDS OF AGLAIA ROXBURGHIANA

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ABSTRACT

Objective Aglaia roxburghiana, family Meliaceae, is a widely used medicinal plant for the treatment of numerous diseases. Although bark, roots, stem, leaves and seeds claim medicinal importance, only a few of the properties have been validated. To provide a scientific base to its popular use, a phytochemical analysis of the aqueous and organic extracts of the seeds were done followed by evaluation of antibacterial activity.

Method The presence or absence of phytochemicals was detected qualitatively by chemical methods. The antibacterial activity of the seed extract was determined by Agar well diffusion method.

Result The aqueous and organic extracts of the seeds revealed the presence of many secondary metabolites. Assessment of antibacterial activity showed positive results.

Conclusion: The results indicate that the ethanolic seed extract of Aglaia roxburghiana could be a safe and potential candidate for the development of new strategies to treat bacterial infections.

INTRODUCTION

Herbs and medicinal plants used in the practice of Ayurveda, ethnic and folklore medicines are rich in phytochemicals effective against a wide range of ailments. Worldwide, it is estimated by the World Health Organization (WHO) that, 65 percent of the population depend on medicinal plants as a primary mode of treatment [1].But of the known medicinal plants known so far, only 6 percent have been screened for biological activity and a reported 15 percent have been evaluated for the presence of useful secondary metabolites [2]. Aglaia roxburghiana, family Meliaceae, is a popular remedy in Ayurveda and among certain ethnic groups for the treatment of numerous diseases. The medicinal use of this plant includes anti-tumour, antiinflammatory, anesthetic, analgesic, antioxidant and antibacterial activity [3]. Leaves are used by the rural folk for curing various ailments like skin diseases, fever, dysentery, cooling, astringent, abdominal pain and hemorrhages [4]. In coastal Karnataka leaves are also used by traditional herbalists for the treatment of Herpes [5].The seeds of Aglaia are also used by certain tribal folks for the treatment of diabetes [6].Many important secondary metabolites have been isolated from the bark, leaves and fruits of the plant.

The present study was designed to assess the traditional claims on the medicinal properties of the seeds, the phytochemicals present in the ethanolic extract of the seeds and the antimicrobial properties of the extracts.

Plant profile - Aglaia elaeagnoidea (A.Juss.) Benth is often seen as an understory tree in the dry as well as moist evergreen forests of the Western Ghats and also in drier parts of India. It is also found in the sacred groves of the plains. Globally it has wide distribution extending from Indo – Malaysia to the Pacific islands [7].Common names include Priyangu - Hindi, Chokkala - Tamil and Punyava - Malayalam. The tree grows to a height of 10 metre. Bark is reddish brown in colour, leaves imparipinnate and alternate, leaflets 5-7, opposite with entire margin. Flowers pale yellow in axilliary branching panicles, fruits berries, buff coloured and globose with 1-2 seeds [7].

MATERIALS AND METHODLOGY

Plant collection and identification

The ripe fruits of Aglaia roxburghiana were collected from the Bonacaud estate region of Western Ghats. The plant was authenticated by eminent botanist, Dr.A.G.Pandurangan, Jawaharlal Lal Nehru Tropical Botanical

Garden and Research Institute, Palode, Thiruvananthapuram, Kerala.

Preparation of the extract and phytochemical screening

The seeds required for study were removed from the fruits, cleaned from all debris and washed thoroughly with running water to free from traces of pulp. The seeds were then dried in shade for a week and ground to a course powder using a mechanical grinder. 200 gram of the seed powder was subjected to continuous soxhlet extraction for 72 hours with 2 Litre each of petroleum ether, ethyl acetate, chloroform, ethanol and distilled water. Each time before extraction with the next solvent, the powdered material was air dried. Extracts were concentrated using a vacuum rotary evapourator and stored at 5° in labelled sterile screw-capped bottles for further use. The extracts were subjected to preliminary phytochemical screening following the method of Trease and Evans [8] and Harborne [9].

Antibacterial activity

All the chemicals and standard antibiotics used for the work were purchased from Hi Media, Mumbai, India and all solvents employed for the work were of analytical grade from Merk, Germany and Sigma Chemicals, USA.

Bacterial cultures and growth conditions:

The test organisms selected were Escherichia coli (MTCC - 1696), Staphylococcus aureus (MTCC - 6908), Salmonella Typhimurium (MTCC- 3224), Sreptococcus pneumoniae (MTCC – 1936) and Vibrio cholerae (MTCC - 3906) which were obtained with their antibiotic resistance profiles from the CSIR- IMTECH Microbial Type Culture Collection and Gene Bank (MTCC), Chandigarh, India. All the test strains were maintained on nutrient agar slants (Hi -Media Laboratories Pvt. Limited, Mumbai, India) at 4°c and sub cultured on to nutrient broth for 24 h prior to testing. These bacteria served as test pathogens for antibacterial activity assay.

Assay method:

Antibacterial activity of the ethanol seed extract was determined by the agar well diffusion method in accordance with National Committee for Clinical Laboratory Standards 1993a (NCCLS)[10].Petri plates containing 20 ml of sterile Muller Hinton Medium were prepared. Each of the plate was inoculated by streaking with a sterile swab moistened with 24 hour culture of the bacterial strains. Wells of 9 mm diameter were bored into the agar medium using a well cutter and filled with 40µl and 80µl of plant extract. Allowed to diffuse at room

temperature for 2 hours. The plates were then incubated in the upright position at 37°c for 24 hours. Wells containing DMSO served as negative control while standard antibiotic discs of streptomycin was used as the positive control. After incubation, the diameters of the growth inhibition zones were measured in mm.

The results indicate that the seed extract could be a safe and potential candidate for the development of new strategies to treat bacterial infections.

RESULTS AND DISUSSION

The present study carried out on Aglaia roxburghiana seed extracts revealed the presence of twelve phytochemicals the results of which are presented in Table 1. Presence and absence of colour was taken as an indication of positive and negative results respectively. In these screening process tannins, phlobatannins, steroids, glycosides, saponins, flavonoids, coumarins, alkaloids, quinones, phenols, sugars and triterpenes gave positive results.

The stem and leaves of Aglaia are employed by certain ethnic groups for painful micturition and urinary discharges[11] .Since these symptoms are associated with Urinary tract infections, antibacterial activity of the ethanol extract of the seeds of Aglaia roxburghiana was analyzed against Escherichia coli[12], Staphylococcus aureus, Salmonella typhimurium, Streptococcus pneumoniae, and Vibrio cholerae. No antibacterial activity was observed against any bacterial organism for a low concentration of the extract (40µg/ml). Antibacterial activity for higher concentration of extract (80µg/ml)was observed against Escherichia coli(15 mm), Staphylococcus aureus(12 mm), Salmonella typhimurium(14 mm), Streptococcus pneumoniae, and Vibrio cholerae(12 mm). Among the four bacterial organisms maximum growth suppression was observed in Escherichia coli (15mm).The results are presented in Table 2, Fig 1.

CONCLUSION

This study has revealed the presence of many important secondary metabolites in the seeds of Aglaia roxburghiana. It has been further confirmed that the plant extract is effective against four of the bacterial strains used for testing, at concentrations of 80 µg/ml. The results lend credence to the use of this plant by tribal folks in treating microbial infections. Secondary metabolites from Aglaia roxburghiana could be exploited for development of new potent antimicrobial agents.

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Table 1: Preliminary phytochemical screening of Aglaia roxburghiana extracts

Sl no	Phytochemicals	Petroleum ether	Ethyl acetate	Chloroform	Ethanol	Distilled water
1	Tannins	-	-	-	+	+
2	Phlobatannins	-	-	-	-	+
3	Steroids	+	+	+	-	-
4	Glycosides	+	+	-	-	-
5	Saponins	-	-	+	+	-
6	Flavanoids	-	-	-	+	+
7	Alkaloids	-	-	+	+	+
8	Coumarin	-	+	+	+	-
9	Quinone	-	-	-	+	+
10	Phenol	-	-	-	+	+
11	Sugar	-	-	-	-	+
12	Steroid	-	-	+	+	-
13	Triterpenes	-	+	+	+	+

+ = presence and - = absence

TABLE – 2: Antibacterial activity of Aglaia roxburghiana by Agar well diffusion method

Test extracts	Salmonella typhimurium MTCC – 3224	Staphylococcus aureus MTCC – 6908	Sreptococcus pneumoniae MTCC- 1936	Escherichia coli MTCC-1696	Vibrio cholerae (MTCC - 3906)
Positive	37	35	35	34	38
Negative	-	-	-	-	-
Test extract 1	-	-	-	-	18
Test extract 2	14	12	-	15	12