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Studies on comparative antibacterial activity of aqueous and ethanolic extracts of *Caesalpinia bonducella* flem. fruit

T.R.Prashith Kekuda*[1], S.Mukunda[1], H.S.Ravikumar Patil[2]

[1]Dept. of Microbiology, S.R.N.M.N College of Applied Sciences, Shivamogga-01, Karnataka, (INDIA)

[2]Dept. of Biotechnology, G.M Institute of Technology, Davangere, Karnataka, (INDIA)

E-mail : prashith_kekuda@rediffmail.com

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ABSTRACT

Caesalpinia bonducella Flem. is traditionally employed to cure various disorders such as urinary disorders, diarrhoea, dysentery, leucorrhoea, piles, wounds, indolent ulcers, skin diseases, leprosy, and toothache. In the present study pod, seed coat and seed kernel of *Caesalpinia bonducella* were subjected to solvent extraction. The aqueous and ethanol extracts were subjected to antibacterial activity by disc diffusion method. Marked antibacterial activity was observed in case of both the extracts against test bacteria. Aqueous extracts were found to be inhibiting test bacteria to more extent than ethanol extracts. The results obtained justify the traditional use of the plant in folklore medicine. Further studies in animal models could reveal the potency of the plant in inhibiting pathogenic bacteria *in vivo*.

KEYWORDS

Caesalpinia bonducella flem;
Solvent extraction;
Antibacterial activity;
Disc diffusion method;
Inhibition zone.

INTRODUCTION

Medicinal plants are important elements of traditional medicine in virtually all cultures. Finding healing powers in plants is an ancient idea. People all over the world have used plants for several remedies. Interest in plants with antimicrobial properties has revived as a result of current problems associated with the use of antibiotics. *Caesalpinia bonducella* (Linn.) Flem is called Bonduc fruit, fever fruit and Physic nut in English and Putikaranja in Sanskrit and belongs to Caesalpiniaceae. It is an armed liane, with glossy branchlets, recurved prickles, yellow flowers and bitter, ovoid to reniform polished seeds in swollen fruit found wild throughout the plains of India and up to an altitude of 1000m in Himalayas. These nuts have been used as anti-periodic for a long time. Pharmacological trials have revealed diuretic and antipyretic activity of the nuts and besides adaptogenic, antimicrobial and muscle contractile activity[1]. The twigs and young leaves of *Caesalpinia bonducella*, a prickly shrub found throughout the hotter regions of India, Myanmar and Sri Lanka, are traditionally used for the treatment of tumors, inflammation, and liver disorders[2]. The present study deals with extraction and antimicrobial activity of pod, seed coat and seed kernel of *Caesalpinia bonducella*.

MATERIALS AND METHODS

Collection and identification of plant material

The plant material was collected from the area near Bidar city, Karnataka, and taxonomically authenticated by the botanist in the Department of Botany, S.R.N.M.N

College of Applied Sciences, Shivamogga and the voucher specimen was retained in the department for future reference.

Extraction of plant material (Solvent extraction)

The plant material was mechanically powdered and the powdered material was subjected to extraction. About 250g of powdered materials were subjected to soxhlet extraction and exhaustively extracted with ethanol for about 48 hours. The extract was filtered and subjected to Vacuum under reduced pressure using rotary flash evaporator, dried in dessicator and stored in airtight containers. For aqueous extract, 10g of powdered material was mixed in distilled water and boiled for about half an hour and filtered through muslin cloth followed by A1 filter paper. The filtrate was condensed to pasty mass. Both ethanol and aqueous extracts were subjected to antibacterial activity.

Screening aqueous extract for antibacterial activity

The bacteria strains were obtained from National Chemical Laboratory, Pune. Gram positive bacteria namely *Bacillus subtilis NCIM 2063, Staphylococcus aureus* NCIM 2079 and Gram negative bacteria namely *Escherichia coli* NCIM 2065, *Enterobacter aerogenes* NCIM 2340 were used. The antibacterial activity was assessed using the simple disc diffusion method[3]. The bacterial strains were inoculated to Muller Hinton broth (Oxoid) medium and incubated. A standard size of inoculum was plated on surface of Muller Hinton agar by spread plate technique using sterile L-shaped glass spreader and plates were allowed for few minutes. Sterile Whatman filter paper discs of 0.5 cm diameter were impregnated with condensed aqueous and ethanol extract (5mg) of pod, seed coat and seed kernel of *Caesalpinia bonducella*, dried and placed on solid medium inoculated with test bacteria. Streptomycin disc (10mcg/disc) was used as reference antibiotic. The plates were left for 30 minutes at room temperature to allow the diffusion of the extract and antibiotic. The plates were allowed to stand at room temperature for two hours and then incubated at 37^0C for 24 hours and zone of inhibition was measured to the nearest millimeter. The test was done in triplicates to arrive concordant results.

TABLE 1: Antibacterial activity of aqueous and ethanol extracts of *Caesalpinia bonducella*

Test bacteria	Inhibition zone in mm						
	Streptomycin (10mcg/disc)	Ethanol extract			Aqueous extract		
		Pod	Seed coat	Seed kernel	Pod	Seed coat	Seed kernel
E.coli	26	17	19	17	19	22	19
E.aerogenes	22	16	16	18	17	19	17
B.subtilis	19	14	15	16	16	16	15
S.aureus	20	16	17	14	16	17	16

RESULTS AND DISCUSSION

TABLE 1 reveals antibacterial activity of aqueous and ethanolic extracts of pods, seed coat and seed kernel of *C.bonducella*. From the results it is evident that both the extracts were found to exert marked antibacterial activity against Gram positive and Gram negative bacteria. Among the extracts tested, aqueous extract was found to be more inhibitory to test bacteria than ethanolic extract. Among different parts tested, it was seed coat that was found to possess more antibacterial activity than other parts tested except in case of *E.aerogenes* and *B.subtilis* by Seed kernel extract. The results of the study supports the folklore use of the plant.

Caesalpinia bonducella Flem. is traditionally employed to cure various disorders such as urinary disorders, diarrhoea, dysentery, leucorrhoea, piles, wounds, indolent ulcers, skin diseases, leprosy, and toothache[4]. The phytochemical constituents flavonoids, triterpenoids, diterpenoids and steroids have been reported in the plant. There are plenty of supporting documents giving information about the pharmacological activities of these phytochemicals. Various parts of the plant are known to possess multiple therapeutic properties like antipyretic, antidiuretic, anthelmintic, antibacterial, anticonvulsant, antiviral, antiasthmatic, antiamoebic, and antiestrogenic activities. Hepatopro tective and antioxidant role of the plant has also been reported[2]. L-y-ethylideneglutamic acid and amino acids were found to be present in the Seeds of *Caesalpinia bonducella*. Fatty acid composition of the seed-oil was reported. a-,b-,y-,d-, e-and j-caesalpins, caesalpin-F and 7-hydroxy-4-methoxyhomoisoflavone (bounducel line) have been isolated form the seed kernels[5]. In a study employing ethanol extracts of 22 Indian medicinal plants, broad spectrum antimicrobial activity of

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C.bonducella was noticed along with few other plants[6]. The methanol extract and four triterpenoids isolated from the seeds of *Caesalpinia bonducella* showed a wide range of inhibiting activity against both gram-positive and gram-negative bacteria[7]. The aqueous and ethanolic extracts of *Caesalpinia bonducella* showed potent hypoglycemic activity in chronic type 2 diabetic model[8]. Methanol extract of *C.bonducella* exhibited significant antitumor and antioxidant activity in Ehrlich ascites carcinoma bearing mice[2]. Ethanolic extract of *Caesalpinia bonducella* seed kernel possesses potent antipyretic and antinociceptive activities in rat model and the findings of the investigation validates its use in the treatment of pain and pyretic disorders[9]. Plants produce a diverse range of bioactive molecules, making them rich source of different types of medicines. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids, and phenolic compounds. Phytomedicines derived from plants have shown great promise in the treatment of various diseases including viral infections. Single and poly herbal preparations have been used throughout history for the treatment of various types of illness. One of the best approaches in search for antimicrobial agents from plant resources is the selection of plants based on ethnomedical leads and testing the selected plants efficacy and safety in light of modern science[2]. The goal is to utilize the impressive array of knowledge assembled by indigenous people about plant and animal products they have used to maintain health[10].

CONCLUSION

Medicinal plants which have been used by human being to treat common infectious diseases are important elements to traditional medicine. Traditional medicine relies on many plants, and many current medicines have been developed from plants. The results of the present *in vitro* study clearly indicate the potential of aqueous extracts of various parts of the plant material to inhibit bacteria. The results provide a scientific basis for the use of this plant as medicine in various tribes and villages. Further studies in animal models could reveal the potential of the plant to inhibit disease producing microorganisms *in vivo*.

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