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ANTIMICROBIAL ACTIVITY OF *CYAMOPSIS TETRAGONOLOBUS* AND *CYPERUS ROTUNDUS* - AN *IN VITRO* STUDY

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ABSTRACT

Cyamopsis tetragonolobus commonly called as Guar is a popular home remedy for gastric disturbances and abdominal discomfort. Cyperus rotundus is used as folklore medicine against spasms, stomach disorders and bowel irritation. The aim of the study was to determine in vitro antimicrobial activity of ethanolic extract of C. rotundus rhizome and C. tetragonolobus fruit using disc diffusion method. Both plants were tested against bacterial species such as Staphylococcus aureus, Pseudomonas aeruginosa, Bacillus subtilis, Klebsiella pneumoniae and fungal species such as Candida albicans, Aspergillus flavus, Fusarium spp, Penicillium spp. It was observed that both the plants showed antibacterial and antifungal activity. The ethanolic fruit extract of C. tetragonolobus was found to be most active against P. aeruginosa and B. subtilis. The ethanolic rhizome extract of C. rotundus was most active against S. aureus and K. pneumoniae. The antifungal activity of C. tetragonolobus was better than C. rotundus.

KEYWORDS : *Antibacterial activity, Antifungal activity, Cyamopsis tetragonolobus, Cyperus rotundus*

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INTRODUCTION

Infectious diseases are the leading cause of death world-wide. Antibiotic resistance has become a global concern in recent years. The screening of natural products has been the source of many therapeutic agents. Due to rapid increase in the rate of infection, antibiotic resistance in microorganisms and side effects of many synthetic antibiotics, medicinal plants are gaining popularity over the drugs. Antimicrobial activities of many plants have been reported by the researchers.

Cyperus rotundus Linn. (Family: Cyperaceae) is found throughout India. It is a pestiferous perennial weed with dark green glabrous culms, arising from underground tubers [1].The tubular part of *C. rotundus* is one of the oldest known medicinal plants used as analgesic, sedative, antispasmodic and to relieve diarrhea. It appears most commonly among Indian, Chinese and Japanese traditional drugs that were used against spasms, stomach disorders and anti-inflammatory diseases [2]. Various pharmacological investigations indicated that *C. rotundus* had remarkable hypotensive, anti-inflammatory and antipyretic effects [3]. Previous phytochemial studies showed that the major chemical components of this herb were essential oil, flavonoids, sesquiterpenes and cardiac glycosides [4]. *C. rotundus* is found to significantly lower blood glucose levels which can be attributed to its antioxidant activity, *in vitro* [5].

Cyamopsis tetragonolobus belonging to Fabaceae, also called as Indian Cluster Bean (Guar) is an annual legume crop mainly grown in India [6]. The seed flour of Guar is the commercial source as a gum used in food, paper, textile and pharmaceutical industries [7].Guar gum is a well-known traditional plant used in folklore medicine. It acts as an appetizer, cooling agent, digestive aid, and laxative and also useful in dyspepsia and anorexia, anti-ulcer, anti-secretory, cytoprotective, hypoglycemic, hypolipidemic and anti-hyperglycemic effects [8]. In addition, guar beans are potentially high sources of phytochemicals [9].

The aim of the study was to determine *in vitro* antimicrobial activity of ethanolic extracts of *C. rotundus* rhizome and *C. tetragonolobus* fruit using disc diffusion method.

MATERIALS AND METHODS:

Collection of Plant Material:

The rhizomes of *Cyperus rotundus* are collected from medicinal vendor and fruits of *Cyamopsis tetragonolobus* are collected from local market, Chennai, Tamil Nadu, India. The selected plant materials were identified and authenticated (PRAC/2010/495 and PRAC/2010/494 respectively) by Prof. P. Jayaraman, Plant Anatomy Research Center, West Tambaram, Chennai, Tamil Nadu, India.

Preparation of powder and extract:

C. rotundus rhizome and *C. tetragonolobus* fruit were shade-dried and pulverized to fine powder in a mechanical grinder. The powder (100g) was extracted with 1 liter of ethanol as solvent. The extracts were concentrated under reduced pressure in a rotary evaporator. The ethanolic extracts were used for antimicrobial study.

Microorganisms:

In vitro antimicrobial studies of the plant extracts were tested against bacterial species such as *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus subtilis*, *Klebsiella pneumoniae* and fungal species such as *Candida albicans*, *Aspergillus flavus*, *Fusarium spp*, *Penicillium spp*. All the stock cultures were obtained from Microbial Type Cell Culture (IMTECH, India).

Assay for antibacterial activity:

Preparation of inoculum:

Stock cultures were maintained at 4°C on nutrient agar (HiMedia) slants. Active cultures for experiments were prepared by transferring a loopful of culture to 10mL of nutrient broth (HiMedia) and incubated at 37°C for 24 hours for bacterial proliferation.

Agar-well diffusion method:

Agar well bioassay was employed for testing antibacterial activity of both the plants [10]. Each extracts were made to a final concentration of 50 mg/mL. 24 hour old cultures of test organisms (0.05mL) were seeded onto Mueller Hinton agar (HiMedia) plate and uniformly spread with a

spreader. Wells (5mm) were made in the agar plate with a sterile cork borer. The plant extract was introduced into the well and the plates were incubated at 37°C for 24 hours. The antibacterial activity of the plant extract was determined by measuring the diameter of the inhibition zone. Controls contained only dimethyl sulfoxide (DMSO). The antibacterial assay for each of the extracts against all microorganisms tested was performed in triplicates. Standard antibiotic streptomycin was maintained as positive control.

Assay for antifungal activity:

Potato dextrose agar (HiMedia) was prepared and 1mL (50mg/mL) of plant extract was added to the medium. After solidification a loopful of culture was placed in the centre of the plate. Controls contained only DMSO. All the plates were incubated at 25°C for 48 hours. The growth of the fungal cultures was measured and compared with the respective control plates. The antifungal assay for each of the extracts against all microorganisms tested was performed in triplicates. Standard antibiotic amphotericin B was maintained as positive control.

RESULTS

The antimicrobial activity of medicinal plants *Cyperus rotundus* and *Cyamopsis tetragonolobus* were tested against four bacteria and four fungi as reported in Figure 1 and Figure 2.

Figure 1: Screening of Antibacterial Activity of Both Medicinal Plants

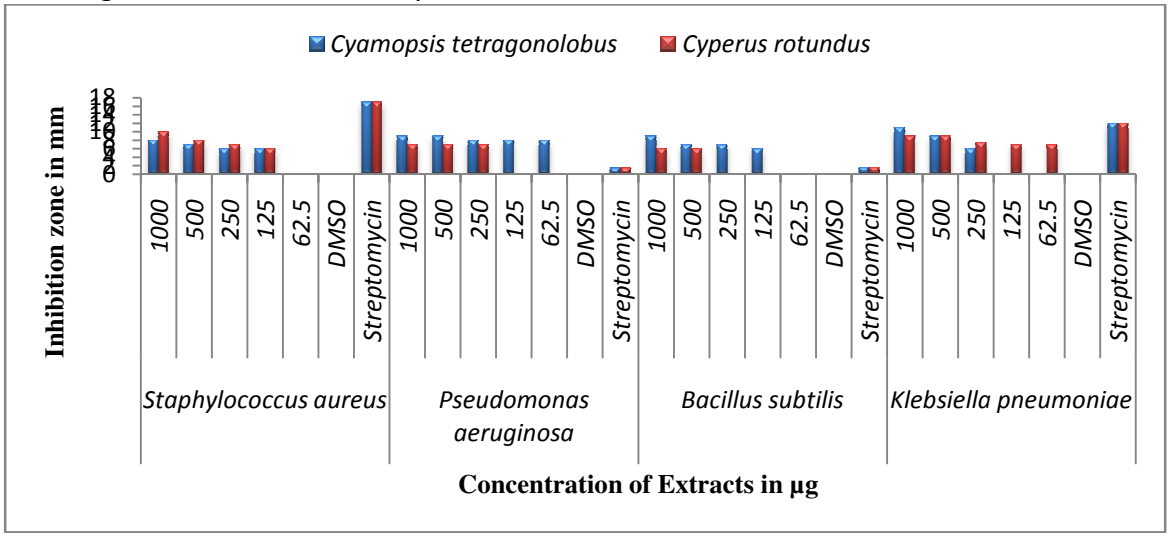
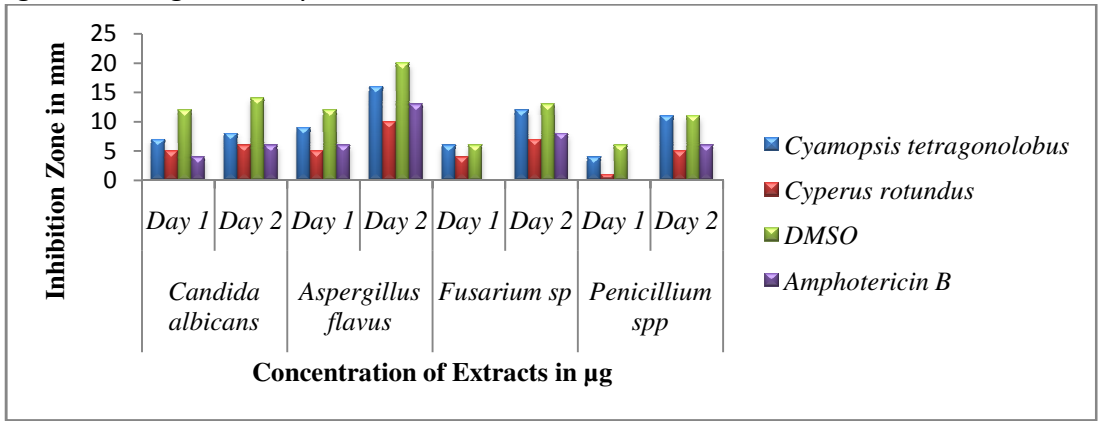


Figure 2: Screening of Antifungal Activity of Both Medicinal Plants



It was observed that both the ethanolic extracts of *C. rotundus* rhizome and *C. tetragonolobus* fruits showed antibacterial and antifungal activity. The ethanolic fruit extract of *C. tetragonolobus* was found to be most active against *P. aeruginosa* and *B. subtilis*. The ethanolic rhizome extract of *C. rotundus* was most active against *S. aureus* and *K. pneumoniae*. The antifungal activity of *C.*

tetragonolobus was more against all the tested fungal species than *C. rotundus*. The susceptibility of the microorganisms to the extracts was compared with each other and with the standard antibiotics streptomycin and amphotericin B.

DISCUSSION

Herbal drugs are proved to be of great commercial and pharmacological importance. Traditional herbal medicines form an important part of the healthcare system. Plants have been used for centuries as remedies for human diseases [11]. The ethanolic extracts of *C. tetragonolobus* fruits showed antibacterial activity. The ethanolic fruit extract of *C. tetragonolobus* was found to be most active against *P. aeruginosa* and *B. subtilis* compared to *S. aureus* and *K. pneumoniae.* The antifungal activity of *C. tetragonolobus* was more against all the tested fungal species than *C. rotundus*. The antimicrobial activities of medicinal plants are attributed due to the presence of flavonoids, tannins and steroidal alkaloids [12]. *C. tetragonolobus* are found to contain flavonoids, tannins, terpenoids and phlobatannins. In our previous study, we have shown the presence of various phytochemicals in *C. rotundus* and *C. tetragonolobus* [13]. Phytochemical studies revealed the presence of saponins, saponin glycosides, sterols, triterpenes, vitamin A, proteins and sugar in *C. tetragonolobus*. The plant also showed gastric anti-ulcer and cytoprotective effects in rats [14]. Guar gum is found to be effective in improving insulin sensitivity in streptozotocin-induced diabetic rats [15]. The hydro-alcoholic extracts of *C. tetragonolobus* possesses hepato-protective activity against CCl4-induced hepatoxicity in rats and this effective in preventive in nature [16].The ethanolic extracts of *C. rotundus* rhizome showed antibacterial activity. The ethanolic rhizome extract of *C. rotundus* was most active against *S. aureus* and *K. pneumoniae* than *P. aeruginosa* and *B. subtilis*. *C. rotundus* rhizomes showed presence of varied essential oil composition [17]. *C. rotundus* contains essential oils, terpenes, flavonoids, b-sitosterol and ascorbic acid [18]. *C. rotundus* significantly lowered the blood glucose levels in alloxan-induced diabetes in rats [19]. *C. rotundus* extracts have found to possess gastroprotective effect against acute gastric mucosal lesions induced by ischemia/reperfusion [20]. *C. rotundus* exhibited significant antioxidant and antimutagenic activities [21].Thus the present study supports the traditional usage of *C. rotundus* and *C. tetragonolobus* suggesting that some of the plant extracts possess compounds with antimicrobial properties. These plants can be used as antimicrobial agents in new drugs for the therapy of infectious diseases caused by pathogens. It is observed from the present study that *C. rotundus* was most active against *S. aureus* and *K. pneumoniae* while *C. tetragonolobus* was found to be most active against *P. aeruginosa* and *B. subtilis*. The antifungal activity of *C. tetragonolobus* was more against all the tested fungal species than *C. rotundus*. However, further work is to be carried out to isolate the active principle and study the nature of the compound.

CONCLUSION

Thus it may be concluded that alcoholic rhizome extracts of *C. rotundus* and fruit extracts of *C. tetragonolobus* possess significant *in vitro* antimicrobial activity and these extracts can be subjected to isolation of therapeutic antimicrobials to carry out further pharmacological evaluation.

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