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Research Article

SCREENING OF CRUDE ETHANOLIC EXTRACT OF THE LEAVES OF *CAPPARIS GRANDIFLORA* WALL.EX HOOK.F & THOMSON FOR ANTHELMINTIC ACTIVITY

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ABSTRACT

Attappady region of Kerala State in India is inhabited by several tribes and they have been using several plants or plant-based preparations for the treatment of various ailments in their traditional system of medicine. During our course of studies on ethno medicine of this region, this plant had a wide reputation among natives of being curative for intestinal-worm infections. Based on this, an attempt has been made to evaluate the anthelmintic potential of the crude ethanolic extract of the leaves of *Capparis grandiflora* using Indian earthworm, *Pheretima posthuma* as test worm. The bioassay involved determination of time of paralysis (P) and time of death (D) of the worms. Piperazine citrate (10mg/ml) was included as reference standard. The crude ethanolic extract exhibited significant anthelmintic activity comparable with the reference drug, Piperazine citrate (10mg/ml). The extract was found to cause significant paralysis and also death of worms especially at higher concentration (100mg/ml). In conclusion, the use of the leaves as an anthelmintic has been confirmed and further studies are suggested to isolate the active principles responsible for the activity.

Keywords: *Capparis grandiflora*, *Pheretima posthuma*, Anthelmintic assay, Phytochemical screening, Tannin.

INTRODUCTION

Helminth infections are among the most widespread infections in humans, distressing a huge population of the world. The majority of infections due to helminths is generally restricted to tropical regions and cause enormous hazard to health and contribute to the prevalence of under nourishment, anemia, eosinophilia and pneumonia[1] .Parasitic diseases cause ruthless morbidity affecting principally population in endemic areas[2]. The parasitic gastroenteritis is caused by mixed infection with several species of stomach and intestinal worms, which results in weakness, loss of appetite, decreased feed efficiency, reduced weight gain and decreased productivity[3].The gastro-intestinal helminthes becomes resistant to currently available anthelmintic drugs therefore there is a foremost problem in treatment of helminthes diseases[4].Indiscriminate use of synthetic anthelmintics can lead to resistance of parasites[5] . Hence there is an increasing demand towards natural anthelmintics. Herbal drugs have been in use since ancient times for the treatment of parasitic disease in human and could be of value in preventing the development of resistance[6,7].

Capparis grandiflora is a climbing shrub with spreading branches found mainly in the adjacent regions of Coimbatore, Nilgiry and Tiruchirappalli[8].Communication with the traditional practitioners have revealed the use of the leaf juice as a stomachic, diuretic , anti rheumatic and anti tumor. *Capparis* species has been reported to have anthelmintic, antimicrobial and anti inflammatory activities[9,10].Modern phytochemical screening has shown the presence of fatty acids, flavonoids and alkaloids in its leaves [11-13] .In this study, an attempt was made to evaluate the anthelmintic activity of the crude ethanolic extract of *C.grandiflora* leaves.

MATERIALS AND METHODS

Plant material: The fresh leaves of *Capparis grandiflora* wall ex. Hook.(Capparidaceae), collected at the flowering stage in the month of March 2010 from the tribal areas of Palakkad district, Kerala State were authenticated by the Botanical survey of India, Combatore, Tamilnadu (BSI).A voucher specimen (no.BSI/SRC/5/23/10-11/Tech-565) is deposited in the departmental herbarium. Leaves were dried in shade for 20 days and then powdered to get a coarse powder. This powder was stored in air tight container and used for further successive extraction.

Preparation of Crude extract: The dried and powdered plant material was extracted with 70% ethanol in a Soxhlet apparatus. Ethanol was removed under vacuum and a semisolid mass was obtained. The extract was subjected to preliminary phytochemical testing for the presence of different chemical classes of compounds.

Preliminary phytochemical screening: Preliminary phytochemical screening was carried out by standard methods[14] .The screening covered mainly alkaloids, glycosides, sterols, terpenes, flavanoids, saponin, tannins, protein and reducing sugar. The presence of phytoconstituents is reported in Table 1.

Anthelmintic assay: The assay was performed on adult Indian earthworm (*Pheretima Posthuma*) due to its anatomical and physiological resemblance with intestinal round worm parasite of human being [15-19].Because of easy availability, earthworms have been widely used for the initial evaluation of anthelmintic compounds in vitro [20-23].All the extracts were dissolved in minimum amount of dimethyl sulphoxide and then volume was adjusted with saline water. 50 ml formulations containing different concentrations of each of the crude extract (25, 50,100mg/ml) in distilled water were prepared and six worms (same type) were placed in it. Time for paralysis was noted when no movement of any sort could be observed except when the worms were shaken vigorously. Time for death of worms were recorded after ascertaining that worms neither moved when shaken vigorously nor when dipped in warm water(50^0C). Piperazine citrate (10mg/ml) was used as a reference standard while distilled water as control.

RESULTS AND DISCUSSION

Preliminary Phytochemical screening of crude extract revealed the presence of carbohydrates, glycosides, alkaloids, tannins and flavanoids .

Table 1: Qualitative analysis of various extracts of *Capparis grandiflora*

Phytoconstiuents	Ethanolic extract
Alkaloids	+
Carbohydrates	-
Flavonoids	-
Glycosides	+
Proteins	-
Saponins	+
Steroids	-
Tannins	+
Terpenoids	-

Table 2: Anthelmintic activity of *Capparis grandiflora*

Normal saline	Conc. (mg/ml)	Paralysis time (min)	Death time (min)
Ethanol extract	25	29± 1.24	33.4± 1.2
Ethanol extract	50	23.63± 0.40	27± 0.56
Ethanol extract	100	11± 0.60	23± 0.64
Piperazine citrate	10	23± 1.15	28.6± 0.8

Values are expressed as Mean ±SEM, n=6.

As shown in Table 2, the ethanolic extract of *Capparis grandiflora* was found to be more potent than the reference control, Piperazine citrate in a dose dependent manner. The potency of the extract was found inversely proportional to the time taken for paralysis / death of worms. Phytochemical analysis has revealed the presence of tannins among the different chemical constituents. Tannins were shown to produce anthelmintic activity[24].

Chemically tannins are polyphenolic compounds[25].Some synthetic phenolic anthelmintics like Niclosamide, Bithionol, Oxyclozamide etc. are shown to interfere with energy generation in helminth parasites by uncoupling oxidative phosphorylation[26].It is possible that tannins contained in the extract of *Capparis grandiflora* produce similar effects. Another possible anthelmintic effect of tannins is that they can bind to free proteins in the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and cause death [27, 28]. The predominant effect of Piperazine citrate in worm is to cause a flaccid paralysis that result in expulsion of the worm by peristalsis. Results reported in the present work constitute rational evidence and a scientific basis to justify and support the folklore claims of the potential anthelmintic activity of the leaves of *Capparis grandiflora*. Further study regarding isolation and characterization of active principles responsible for activity, using *in vivo* models, and establishment of the possible mechanism(s) of action are currently under progress.

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