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Original Article

CUSCUTA SPECIES MEDICINAL APPLICATIONS AND EFFECTS BASED ON THEIR EXTRACTIVE COMPOUNDS

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ABSTRACT

Cuscuta species are plays a very robust and important role in the medicinal applications, this plant stems and seeds have highly important medicinal values. Some research studies say those Indian tribes and other traditional communities are used this plant as purgative, carminatives and external application for skin diseases. And it had antiviral and anti cancerousvactivities. Cuscuta reflexa Roxb, is prevalent in various regions of Bangladesh. And extracts of the stem reportedly demonstrated anti-steroidogenic and antibacterial activities The various pharmacological activities of whole plant or plant parts having particular activity.Two of the widely used plants for treatment of this disease are the stems (vines) of Cuscuta reflexa and leaves of Calotropis procera. It was the objective of the present study to evaluate the hypoglycemic potential of methanol and chloroform extracts of stems of Cuscuta reflexa and methanol extract of leaves of Calotropis procera Further research work is necessary to isolate, characterize the phytochemical constituents with effective pharmacological study.

Keywords: Antimicrobial, antihelmethitic, cuscuta, methanol

Introduction:

Many of the plants used in herbal medicine contain active constituents whose effects can be demonstrated pharmacologically and the action of the whole plant extract can usually be related to that of the isolated constituents. However, for some herbal remedies the situation is complicated by the frequent use of a number of drugs in combination1. And it has long been used for its sedative properties, but the unreliability of its preparations and the lack of association of therapeutic activity with known constituents.The sedative action of the root resided in a group of unstable, the epoxyiridoid esters(valepotriates) and these were marketed as a freeze dried products to avoid decomposition.Most of the heebal compounds are composed and remedies are inherently safer then the potent synthetic drugs, which often produce undesirable side effects. There are extremely toxic plants in the plant kingdom which produce carcinogens, teratogens and other compounds which cause disease sensitization. Thus, comfrey(symphytum officinale), always considered a safe herb has been found to contain small quantities of pyrrolizidine alkaloids which are known to be hepatotoxic and which, when administered to rats, cause liver cancer 2-5. And it exhibit associated with breast cancer, but no such cases have been reported as a result of administration of root extract. A number of cases of toxicity arising from over-consumption of herbal remedies have been reported, and the principal danger appears to be that arising from the uncontrolled supply and administration of these products6,11.

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Herbal medicine used as Anti microbial [18, 19, 20]

The history of prevention of bacterial infection can be traced back to the 19 th century when Joseph Lister (in 1867) introduced antiseptic principal for use in surgery & post-traumatic injury. He used phenol (carbolic acid) for the hands

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wash, as a spray on an incision site, & on bandages applied on wounds. Around 1881 & continuing to 1900 microbiologist Paul Ehrlish disciple of Robert Koch began work with a set of antibacterial dyes & anti- parasitic organic arsenic. His goal to develop compound that retained antimicrobial activity at the expense of toxicity to the human host, he called that agent that he sought “magic bullets”.

Anti microbial methods depends on several factors

Methods of evaluation of anti-microbial activity

I) Diffusion method : a) Agar disk diffusion assay b) Agar well diffusion assay c) Ditch-plate method e) Poison food technique (antifungal) f) Spore germination assay (antifungal)

II) Diluation method : a) Broth micro- diluation

b) Broth macro- diluation

III) Bio-autography : a) Agar diffusion /contact bioautography

b) Immersion /agar overlay bioautography

c) Direct bioautography

Anti helminthic activity [21,22]

The incidence of helminth infections is a global human health concern. Tropical regions of the world, particularly the Sub-Saharan African communities are among the worst hit by the diseases. The majority of infections due to helminths causes enormous hazard to health, contributing to the prevalence of under nourishment, anaemia, eosinophilia and pneumonia. Parasitic diseases such as lymphatic filariasis, onchocersiasis and schistosomiasis cause ruthless morbidity affecting principally population in en-demic areas. The parasitic gastroenteritis is caused by mixed infection with several species of stomach and intestinal worms which result in weakness, loss of appetite, decreased feed efficiency, reduced weight gain and decreased productivity.

Material and methods

Methods for testing acute and sub acute inflammation:

UV-erythema in guinea pigs: Prostaglandin E (PGE) levels in the skin have been shown to be elevated during the 24 h period following exposure of guinea pig skin to ultraviolet radiation from 280-320 nm. The development of increased PGE levels paralleled the development of the delayed phase of erythema. Delay the development of ultraviolet erythema on albino guinea pig skin by systemic pretreatment with clinically equivalent doses of phenylbutazone and other nonsteroidal anti-inflammatory agents. Erythema (redness) is the earliest sign of inflammation, not yet accompanied by plasma exudation and edema. This model depicts the delay in development of UV erythema on albino guinea pig skin by systemic pretreatment with clinically equivalent doses of phenylbutazone and other NSAIDs.

Procedure: Albino guinea pigs of both sexes with an average weight of 350g are used. Four animals are used each for treatment and control group.18 hr prior testing, the animals are shaved on both the flanks and on the back. Then they are chemically depilated by a commercial depilation product or by a suspension of barium sulphide. 20 min later, the depilation paste and the fur are rinsed off in running warm water. On the next day the test compound is dissolved in the vehicle and half of the test

Evaluation: The degree of erythema is evaluated visually by 2 different investigators in a doubleblindedmanner. The followings scores are given:

0 = no erythema

1 = weak erythema

2 = strong erythema

4 = very strong erythema

Animals with a score of 0 or 1 are considered to be protected. The scoring after 2 and after 4 h gives some indication of the duration of the effect. ED50 values can be calculated.

Vascular permeability: During inflammation, vascular permeability increases to allow plasma constituents such as antibodies and complement to access injured or infected tissues. The test is used to evaluate the inhibitory activity of drugs against increased vascular permeability which is induced by phlogistic substances. Mediators of inflammation, such as histamine, prostaglandins and leucotrienes are released following stimulation e.g. of mast cells. This leads to a dilation of arterioles and venules and to an increased vascular permeability. As a consequence, fluid and plasma proteins are extravasated and edemas are formed. The increase of permeability can be recognized by the infiltration of the injected sites of the skin with the vital dye Evan’s blue.

Procedure: Albino Wistar are used each group containing 4 rats. Control group will receive distilled water 1%w/v 1ml/100g by oral route and other group will receive test compound by oral route and standard group will receive diclofenac 10ml/kg by intraperitoneal route. After 1h of these administration rats are injected with 0.25ml of 0.6% v/v solution of acetic acid intraperitoneally. Immediately, 10 ml/kg of 10%w/v Evans blue is injected intravenously via tail vain. After 30 min , the animals are anesthetized with ether anaesthesia and sacrificed. The abdomen is cut open and exposed viscera. The animals are held by a flap of abdominal wall over a Petri dish. The peritoneal fluid (exudates) collected, filtered and made up the volume to 10 ml using normal saline

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solution and centrifuged at 3000 rpm for 15 min. The absorbance (A) of the supernatant is measured at 590 nm using spectrophotometer.

Evaluation: Decreased concentration of dye with respected to absorbance indicates reduction in permeability. The result of test is compared with that of standard. ED50 values can also be calculated.

Oxazolone-induced ear edema in mice: The oxazolone-induced ear edema model in mice is a model of delayed contact hypersensitivity that permits the quantitative evaluation of the topical and systemic anti-inflammatory activity of a compound following topical administration. The oxazolone-repeated challenge increased the level of Th2 cytokines and decreased that of a Th1 cytokine in the lesioned skin. The Th2 cytokines, especially IL-4, play major roles in the development of dermatitis in the present mouse model.

Procedure: Using 12mice in each group, the same skin site of the right ear was sensitized by a single application of 10 μl (each 5 μl for inner and outer of ear) of 0.5% oxazolone in acetone 7 days before the first challenge (day 0), and 10 μl of 0.5% oxazolone in acetone was repeatedly applied to the sensitized right ear 3 times per week. In the nonsensitized animals, acetone alone was applied to the right ear. The mice are challenged 8 days later again under anesthesia by applying 0.01 ml 2% oxazolone solution to the inside of the right ear (control) or 0.01 ml of oxazolone solution, in which the test compound or the standard is solved. Groups of 10 to 15 animals are treated with the irritant alone or with the solution of the test compound. The left ear remains untreated. The maximum of inflammation occurs 24 h later. A this time the animals are sacrificed under anesthesia and a disc of 8 mm diameter is punched from both sides. The discs are immediately weighed on a balance. The weight difference is an indicator of the inflammatory edema.

Evaluation: Average values of the increase of weight are calculated for each treated group and compared statistically with the control group.

Results and discussions:

Before 1947, there was production of quinine from cinchona as a plant based modern drugs in India. After 1965, bulk production of plant-based modern drugs has become an important segment of Indian pharmaceutical industry12-17. Some of the phyto-pharmaceuticals produced in India at present include morphine, codeine, papaverine, thebaine, emetine, quinine, quinidine, digoxin, caffeine, hyoscine, hyoscyamine, xanthotoxin, psoralen, colchicines, rutin, berberine, vinblastine, vincristine, nicotine, strychnine, brucine, ergot alkaloids, senna glycosides, pyrethroids and podophyllotoxin resin. Phytopharmaceuticals for which technology has been developed for undertaking large scale production include L-dopa from Mucuna beans, ajmaline and ajmalicine from Rawolfia serpentiana and Catharanthus roots, respectively, and 18 β-acetyl glycyrrhetic acid from Glycyrrhiza glbra.

Experimental data

Screening of in vivo anti bacterial activity of crude extracts

Table. No-1: Antibacterial activity of Psoralea Corylifolea seed crude extracts

S.No.	Concentration (mg/ml)	Zone of inhibition in mm	
		Methanolic extract	Hexane extract
1	25	22	17
2	50	29	23
3	100	34	31

Zone of inhibition for Standard drug Streptomycin (10μg/ml) - 38mm Zone of inhibition for Standard drug Cefotaxime (10μg/ml) - 42mm From the above results the methanolic extract was found to be having more anti bacterial activity than hexane extract.

Screening of in vitro anti helminthic activity of crude extracts

Table no-2: Anti helmintic potency of methanolic and hexane extract of Psoralea Corylifolea

S.No.	Extract	Concentration (mg/ml)	Pheritima posthuma	
			Paralysis Time (min)	Death Time(min)
1	Control(1% DMSO)	_	_	_
2	Std.Albendazole	20(mg/ml)	25	56
3	Methanolic	25(mg/ml)	73	128
		50(mg/ml)	58	104

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		100(mg/ml)	44	81
4	Hexane	25(mg/ml)	76	135
		50(mg/ml)	63	109
		100(mg/ml)	48	85

From the above results the methanolic extract was found to be having more anti heminthicl activity than hexane extract. The anti helmintic activity of methanol extract could be due to the constituents present.

Screening of in vivo anti bacterial activity of crude extracts

Table. No 3 Antibacterial activity of cuscuta whole plant extracts

The zone of inhibition was examined and measured (The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the well). Streptomycin(10μg/ml) was used as a standard.Howerver Both the plant extracts at dose 100 mg/ml showed almost same effect on these selected bacteria and found to show significant results in dose dependent manner and was comparable to standard streptomycin 10mg/ml.

S.No.	Concentration (mg/ml)	Zone of inhibition in mm	
		S.aureus	E.coli
1	MR 50	12.33±1.52**	15±1.0**
2	EAR100	8.33±0.577**	11.66±1.52**
3	MA 50	10.33±1.52**	9.33±2.08**
4	EAA 100	14±1.0**	12±1.0**
5	Streptomycin 10	16±1.0**	16.33±1.52**

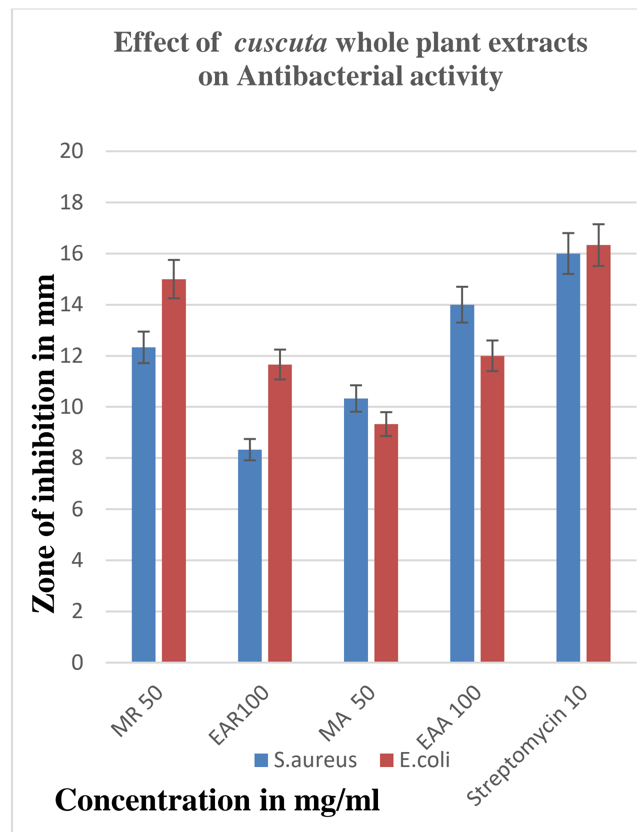

Table 4: Effect of C reflexa whole plant extracts on antifungal Activity

S. No	Fungal pathogen	*Candida albicans*		*Aspergillus niger*	
1	Methanolic extract	Concentration (mg/ml)	Zone of inhibition in mm	Concentration (mg/ml)	Zone of inhibition in mm
		50	15±1.0**	50	13.33±1.52**
		100	22.66±3.21**	100	26±1.0**
2	Ethyl acetate extract	50	15±1.0**	50	12±1.0**
		100	24.66±4.16**	100	27±2.0**
3.	Fluconazole	10	33.33±1.52**	10	31±1.0**

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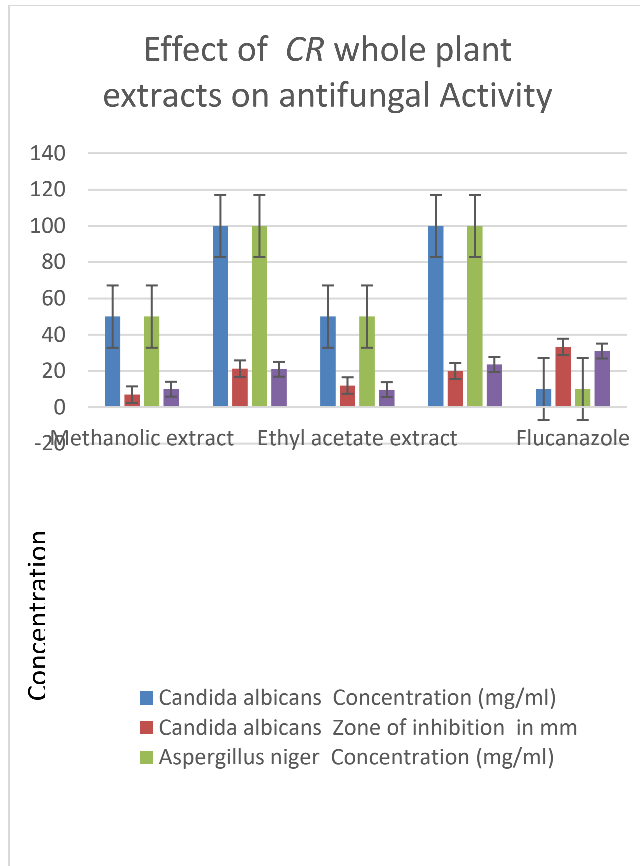


Table 5: Effect of C australis plant extracts on antifungal Activity

S. No	Fungal pathogen	*Candida albicans*		*Aspergillus niger*	
1	Methanolic extract	Concentration (mg/ml)	Zone of inhibition in mm	Concentration (mg/ml)	Zone of inhibition in mm
		50	7±1.0**	25	10±1.0**
		100	21.33±1.52**	50	21±1.0**
2	Ethyl acetate extract	50	12±1.0**	25	9.66±1.52**
		100	20±1.0**	50	23.66±1.52**
3.	Flucanazole	10	33.33±1.52**	10	31±1.0**

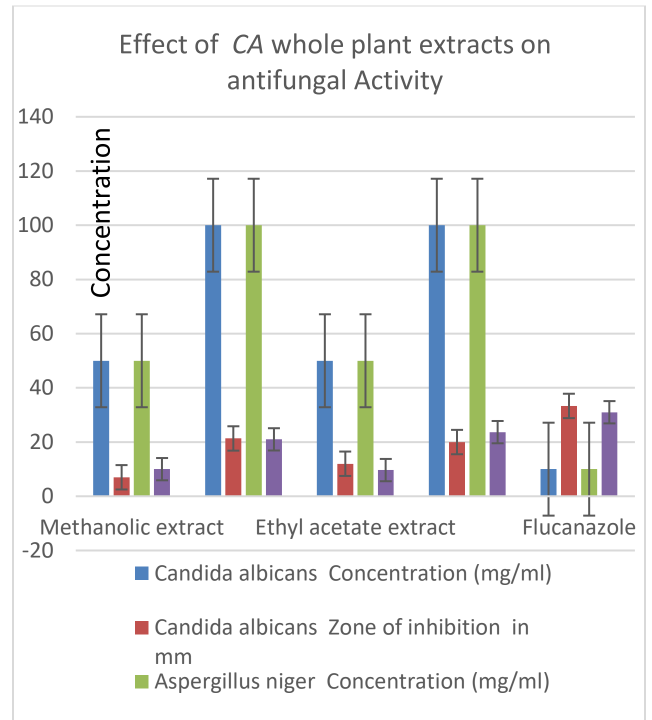
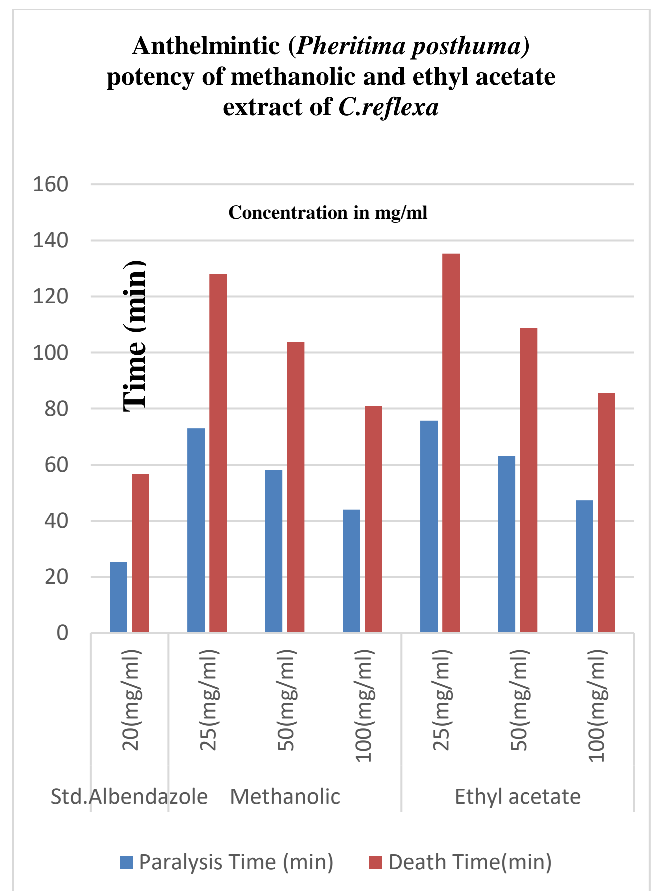


Table 6: Anthelmintic potency of methanolic and ethyl acetate extract of Cuscuta australis



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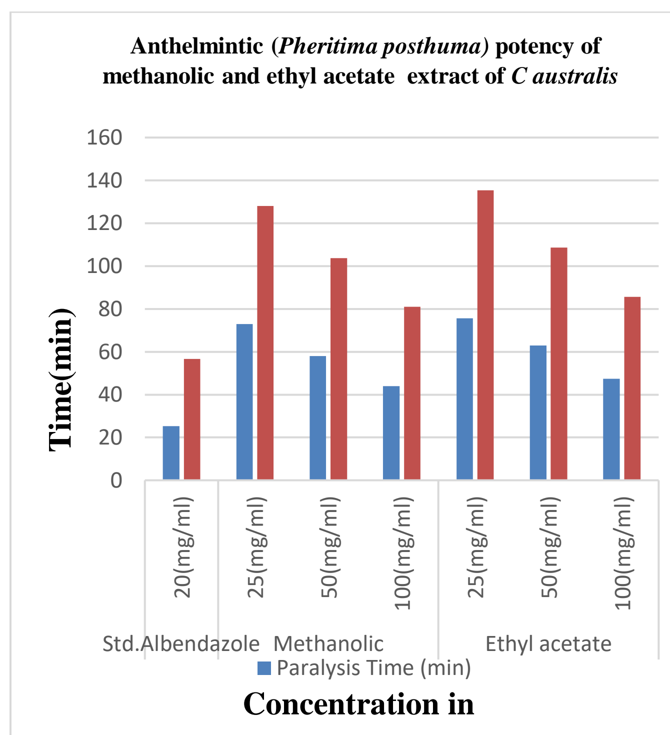
Anthelmintic potency of methanolic and ethyl acetate extract of CA

S.No.	Extract	Concentration (mg/ml)	*Pheritima posthuma*	
			Paralysis Time (min)	Death Time(min)
1	Control(1% CMC)	_	_	_
2	Std.Albendazole	20(mg/ml)	25.33 ± .33***	56.66 ± 0.66***
3	Methanolic	25(mg/ml)	80.33±2.51**	127±1.73**
		50(mg/ml)	76.33±1.52**	111±3.606**
		100(mg/ml)	57.66±2.51**	82±5.29**
4	Ethyl acetate	25(mg/ml)	93±3**	134.66±1.52**
		50(mg/ml)	65.66±2.51**	120.33±0.57**
		100(mg/ml)	51.66±1.52**	94.33±4.04**

Values are given as Mean± S.D (n=3 (n=3); ***p<0.001;p<0.01;*p<0.05 compared with control**



From the above results the methanolic extract was found to be having more anti heminthic activity of the effect of standard drug albendazole at adose of 100 mg/ml concentration.

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