

SHORT COMMUNICATION***In Vitro* Evaluation of *Capparis spinosa* against *Lumbricus terrestris* (Annelida)****Fatin A. A. Mustafa**

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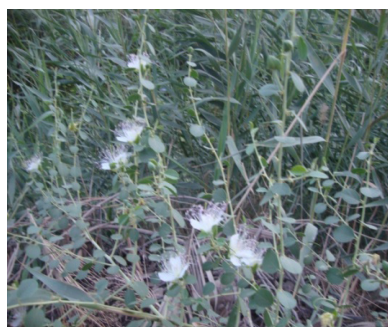
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Parasitic diseases are highly prevalent in third world countries⁽¹⁾. Although many antihelminthics are available, yet development of drug resistance^(2,3) has urged the need for safe alternatives. Medicinal plants have been used safely against parasitic infections in man and animals^(4,5), and have been reported as a rich source of botanical antihelminthics^(6,7). This has encouraged the proposal of screening medicinal plants for their antihelminthic activity.

Capparis spinosa (Figure 1) belonging to the family Capparidaceae is a xerophytic plant growing in a broad range of climatic conditions, varying from dry deserts to cooler altitudes of mountains⁽⁸⁾. This plant also known as the caper bush, is a perennial winter deciduous species that bears rounded, fleshy leaves and large white to pinkish flowers⁽⁹⁾. The caper bush is included in the floristic composition of almost all circum-Mediterranean countries. Although, the flora of the Mediterranean region has considerable endemism, it is uncertain whether the caper bush is indigenous to these regions. However, it could have originated in the tropics and later naturalized to the Mediterranean basin⁽⁸⁾. Many species of *Capparis* are reported from Iraq, from northern to southern plateaux of the country⁽¹⁰⁾. The plant is best known for the edible buds and fruits (caper berry), which are consumed pickled⁽¹¹⁾.

**Figure (1):** *Capparis spinosa*

C. spinosa was reported to be effective in treatment of paralysis and toothache⁽¹²⁾, and was found to exhibit potent anti-hyperglycemia activity as well as prevent chemically induced pappillomagenesis in mouse skin^(13,14). Its ethanolic and aqueous extracts reduced carrageen induced edema in rats⁽¹⁵⁾ and showed anti-hepatotoxic activities^(16,17). Extracts of different parts of *C. spinosa* have been shown to possess biological activity against a large number of pathogens⁽¹²⁾. Antifungal, antibacterial, anti-amoebic, and anti-worm activities have been demonstrated⁽¹⁸⁻²¹⁾. In one report its extract agglutinated *Leishmania* (parasite) and killed it in the vector *Phlebotomus papatasi*⁽²²⁾. In addition, *C. spinosa* is extensively used as an antihelminthic⁽²⁰⁾. The aim of the present study was to investigate the antihelminthic effect of aqueous and alcoholic extracts (soxhlet) of the aerial parts of *C. spinosa* (L.) on the adult earthworm *Lumbricus terrestris*, as a model for future application in treatment of human helminthes.

Fresh aerial parts of the plant *C. spinosa* (Linn Juss) were collected from the garden of Education College, Basrah University, close to Qarmat Ali river in Basrah city during May and June, 2011. The aerial parts of *C. spinosa* were dried in the shade and ground by a mechanical grinder, and the powdered plant was exhaustively extracted in a soxhlet apparatus. In this extraction process, 100 gm of the dried powder was treated separately with 250 ml ethanol or distilled water as solvent. The extracts were cooled at room temperature, filtered and concentrated under reduced pressure in a rotary evaporator, then dried. The dry extracts were subjected to various chemical tests to detect the presence of different phytoconstituents^(23,24).

The qualitative phytochemical investigation of both extracts of *C. spinosa* showed the presence of an array of active chemical constituents including alkaloids, glycosides, tannins, and phenolic carbohydrates, steroids,

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flavonoids, triterpenoids, and saponins. The alkaloids were tested using Dragendroff reagent and Mayer's reagent⁽²³⁾, the glycoside was tested by Benedict's reagent⁽²³⁾, triterpenoids was tested by Hirschorn test⁽²³⁾, and tannins and phenolic compound were tested by lead acetate and ferric chloride, respectively⁽²³⁾.

Because of easy availability, earthworms have been used widely for the evaluation of antihelminthic compounds *in vitro*^(25,26,27,28). Therefore as an experimental model the adult earthworm (Annelida) was collected from water logged areas from the nearby locality of Qarmat Ali River. The antihelminthic assay was carried out according to Ajaiyoba *et al.*⁽²⁹⁾. Twenty ml formulas, each containing one concentration of the crude ethanolic or aqueous extracts (200 mg/ml or 400 mg/ml) were prepared. Six worms were placed in each formula and the time for paralysis was recorded when no movement of any sort was observed, except when worms were shaken vigorously. Time for death of worms was recorded after ascertaining that the worms neither moved when shaken vigorously nor when dipped in warm water at 50°C⁽²⁸⁾.

Albendazole (20 mg/ml) was used as reference standard and distilled water was used as control^(27,29).

Preliminary screening of the alcoholic extract revealed the presence of alkaloids, glycosides, carbohydrates, tannins, phenolics, flavonoids and triterpenoids while the aqueous extract showed the presence of steroids, glycosides, carbohydrates, flavonoids and saponins (Table 1).

Both the alcoholic and aqueous extracts of *C. spinosa* displayed significant antihelminthic properties at high concentrations. Both extracts showed antihelminthic activities in a dose-dependent manner giving short time of paralysis and death with 400 mg/ml concentration. The alcoholic extract induced paralysis of the earthworm *L. terrestris* in 6.16 minutes and death in 9.1 minutes, while the aqueous extract showed paralysis and death in 21.83 and 34.5 minutes. In the mean time, albendazole (20 mg/ml) caused paralysis of the earthworm in 8.6 minutes and death in 32.23 minutes. The mean \pm SEM values calculated for each parameter are shown in table (2).

Table (1): Qualitative phytochemical analysis of *Capparis spinosa* (aerial parts) extract.

Chemical Tests	Ethanolic Extract	Aqueous Extract
Alkaloids	+	+
Steroids	-	+
Glycosides	+	
Carbohydrates	+	+
Flavonoids	-	+
Tannins	+	+
Phenols	+	+
Triterpenoids	+	-
Saponins	-	+

(+): shows the presence of the given chemical constituent.

(-): shows the absence of the given chemical constituent.

Table (2): Antihelminthic activity of ethanolic and aqueous extracts of *Capparis spinosa* aerial parts.

Extracts	Conc. (mg/ml)	Paralysis time (min)	Death time (min)
Aqueous	200	32.33 \pm 1.71	42.9 \pm 61.99
Aqueous	400	21.83 \pm 1.16	37.13 \pm 1.34
Ethanolic	200	10.96 \pm 1.99	23.46 \pm 1.01
Ethanolic	400	6.16 \pm 1.90	9.1 \pm 0.41
D. W.	-	51.2 \pm 0.41	74.83 \pm 2.24
Albendazole (standard)	20	8.6 \pm 0.12	32.23 \pm 0.40

Each value represents mean \pm SEM (N=6).

An antihelminthic drug can act by causing paralysis of the worm or damage to the body cuticle leading to partial digestion. In the host, the drug may promote rejection by immune mechanisms. Antihelminthic drugs can also interfere with the metabolism of the worm⁽³⁰⁾. The mechanism of action of albendazole on the worm is to breakdown microtubules (which are the main components of the cytoskeletal structure of the worm). This is accompanied by selective and irreversible inhibition of glucose uptake. The final result is depletion of parasite glycogen stores, reduced formation of ATP, disruption of metabolic pathways, and ultimate worm death⁽³¹⁾.

The antihelminthic activity of *C. spinosa* extract may be related to the presence of tannin, which is a polyphenolic compound capable of producing the same effect as that of some synthetic phenolic antihelminthics, such as niclosamide, oxiclozanide and bithionol. These compounds are known to interfere with energy generation in helminth parasites by uncoupling parasite specific fumarate reductase mediated oxidative phosphorylation⁽³²⁾. Another possible antihelminthic effect of tannins is that they can bind to free proteins of host GIT⁽³³⁾ or glycoprotein on the parasite cuticle⁽³⁴⁾ and ultimately causing parasite death.

In conclusion, the present study confirms the value of the traditional use of the aerial parts of *C. spinosa* as an antihelminthic agent; as both the ethanolic and water extracts showed good antihelminthic activity against.

Further studies are needed to isolate the possible active phytoconstituents which are responsible for the antihelminthic activity, and to study the mechanism of action using parasitic models.

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