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Antifungal Activity of Medicinal Plants from Jordan Environment

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Abstract: Medicinal plants collected from different locations in Jordan were tested for their antifungal activities against 5 plant pathogenic fungi: *Phytophthora infestans, Fuusarium oxysporum, Rhizoctonia solani*, *Stemphylium solani* and *Mucor* sp. Data of this study showed that the highest growth inhibition of all fungi was observed with *Salvia indica*, which gave (66.3%), of inhibitions for *Stemphylium*, followed by *Mucor* (60.5%), *R. solani* (51.7%), *F. oxysporum* (48%) and *P. infestans* (28.8%).

Key words: Medicinal plants, extracts, plant pathogenic fungi, Jordan

INTRODUCTION

Medicinal plants have been used for centuries to cure human diseases[1,2]. It has been proved that many of these plants exhibit general antifungal and antibacterial activities.

Plant diseases caused by plant pathogenic fungi are among the most important factors that limit vegetable production in Jordan. To reduce yield losses, farmers apply large quantities of fungicides every year. The continuous application of chemicals will lead to destroy the ecosystem and result in outbreaks of new strains of fungi that are difficult to control. To minimize the side effects of chemical application many efforts have been done to utilize the antimicrobial activity of plant extracts. Several studies showed the importance of natural chemical as a possible source of non-phytotoxic, systemic and easily biodegradable alternative[3-6].

Studies on the antifungal activity of medicinal plants against plant pathogens are rare. Therefore, the aim of this study was to investigate the antifungal activity of various medicinal plants (milfoil, chamomile, field southern wood, small flowered bind wood white sage and large flowered sage) collected from different location in Jordan against the following plant pathogenic fungi (*Phytophthora infestans, Fusarium oxysporum, Rhizoctonia solani, Stemphlium solani* and *Mucor* sp.).

MATERIALS AND METHODS

Plant material: Six plants collected from different areas in Jordan (between March and May of 2003) were used in this study (Table 1).

Fungal strains: The plant pathogenic fungi used in this study were collected from various location in Jordan: *phytophthora* sp., *Fusarium oxysporum, Rhizoctonia solani* and *Mucor* sp. (Table 2). All fungal isolates were identified at species or genus level and deposited in fungal collection bank at Department of Biotechnology of Al-Balq'a Applied University, Al-Salt, Jordan.

Fungal isolates were maintained on Potato Dextrose Agar (PDA, Difco Laboratories, Detroit, MI USA) and the culture were stored at room temperature and subculturing once a month. The isolates were allowed to grow for 7-10 days before they were used in the microbial studies.

Preparation of extracts: Plant material was dried in shade at room temperature and then ground by using a blander. A 250 g of powdered plant material were soaked in

Table 1: Medicinal plants collected from Jordanian environment and used from extraction preparation

Scientific name	English name	Plant part used for extraction	Area from which plant collected
Achillea tomentosa	Milfoil	Aerial part	Mafraq
Anthemis nobilis	Chamomile	Aerial part	Upper, Lower Jordan, Valley, Dead Sea, Amman, Petra, Salt
Artemisia arboresceus	Field Sathern wood	Aerial part	Azraq, Um-Jamal
Convolvulus siculus	Small flowered bindwood	Aerial part	Salt, Amman, North Part of Jordan
Savia triloba	White sage	Leaves	Amman, Irbid, Mezar
Salvia indica	Large flowered sage	Leaves and branches	Ajloun, Jarash and Salt

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Table 2: Plant pathogenic fungi collected from various locations in Jordan

Fungus	Host	Plant part	Location
Phytophthora sp.	Potato	Leaves	Jordan valley
Fusarium oxysporum	Cucumber	Root	Jerash
Rhizoctonia solani	Cucumber	Roots and stem	Jerash
Stemphylium solani	Tomato	Stem and leaves	Jerash
Mucor sp.	Gerber	Roots	Amman

1.25-1.5 L of 95% ethanol for 5 days at room temperature. The mixture was stirred daily by shaker for regular infusion. After a five-day period, the extract was filtered by using Whatman filter paper No. 1 (ALBET^R). The filtrate was dried using a rotary evaporator at 60°C. The final dried extract was stored in labeled sterile glass bottles at -20°C until used[7].

Antifungal activity: Extracts from the six test plants were diluted in (Dimmethyl Sulfoxide) DMSO (10 mL as final volume). The 10 mL of DMSO including the plant extract was added to 240 mL of PDA to give a final concentration of 100, 250 and 500 ppm for each extract and then the resulting medium was poured in plates. Control plates received only DMSO in PDA without plant extract. Inoculum plugs from the actively growing margin of petri plate cultures of each fungal isolate was placed face down in the center of each pertri plate using a 10 cm long spring loaded plunger of 5 mm diameter. Each isolate was inoculated on 3 plates for each extract and incubated for 7-10 days at 28°C. Control plates were run along each fungal isolate and crude extract, following the same procedure as above.

Starting two days after inoculation, radial growth was recorded daily for 7 days or until the plates were overgrown. The percentage of fungal growth inhibition = [(growth in control - growth in sample)/(growth in control) X 100] where growth was measured in mililiter m as colony diameter[8]. The values reported for minimum inhibitory concentration were average of three readings.

RESULTS AND DISCUSSION

Extracts from 6 medicinal plants from Jordan were tested against 5 phytopathogenic fungi to determine their antifungal. Antifungal activity, was measured by the MIC (Minimum Inhibition). Different concentrations of each extract were tested: 100, 250 and 500 ppm. All the fungi tested in the growth inhibition assay showed various degrees of sensitivity to the 6 plant extracts obtained (Table 3).

For *S. solani* 66.2% of fungal growth was inhibited with E1 at 100 ppm, followed by E6 (44.4%) at 500 ppm, E_2 (31.4%) at 500 ppm, E_3, E_4 and E_5 had a weak antifungal effect against *S. solani* (Table 3).

All extracts had a weak effect on *Mucor* sp. Except extract (E1) (60.4% inhibition at 250 ppm).

R. solani showed 51.6% of fungal growth inhibition by E_1 at 500 ppm followed by E_5 (29.8%) at 500 ppm, E_6, E_3, E_2 and E_4 had very weak antifungal effect against *R. Solani* (Table 3).

Fusarium oxysporum showed 47.9% of fungal growth inhibition by E_1 at 500 ppm, followed by E_5 (14.1%) at 500 ppm, E_2, E_3, E_6 and E_4 had very weak antifungal effect against *F. oxyporum*.

For *Phytophthora* sp. 28.81% of fungal growth was inhibited with E_1 at 500 ppm, followed by E_4 (20.3%) at 100 ppm. E_5, E_6, E_3 and E_2 had very weak antifungal effect against *phytophthora* sp.

The highest growth inhibition of all fungi was observed with *S. indica* (E_1), which gave (66.2%) of inhibition for *stemphylim solani*, followed by *Mucor*

Table 3: Antifungal activity of plant extracts from six medicinal plants pathogenic fungi isolated in Jordan

Plants	Extract concentration (ppm)	Minimum Inhibition Concentration (MIC) (%)				
		Fusarium oxysporum	*Phytophthora* sp.	*Mucor* sp.	*Rhizoctonia solani*	*Stemphylium solani*
	100	22.10	25.4	28.8	34.3	66.2
Salvia indica	250	31.60	21.1	60.4	43.2	51.4
(E_1)	500	47.90	28.8	51.1	51.6	41.7
	100	05.80	03.8	03.2	05.7	10.7
Convolvulus siculus	250	04.60	-12.7	05.7	06.5	32.8
(E_2)	500	10.10	-13.1	11.4	05.3	31.4
	100	03.30	-02.5	05.8	05.0	24.9
Salvia triloba	250	07.00	04.6	15.8	08.8	15.1
(E_3)	500	02.40	-11.8	10.9	12.4	36.9
	100	-01.80	20.3	03.0	02.0	16.3
Anthemis nobilis	250	00.93	08.8	-03.3	01.1	20.2
(E_4)	500	-00.00	02.9	03.7	06.8	29.7
	100	00.60	21.1	07.0	16.1	05.8
Artemisia arborescens	250	08.90	18.6	17.3	20.5	00.0
(E_5)	500	14.10	16.5	18.3	29.8	31.9
	100	06.40	-13.9	-00.4	08.9	16.3
Achillea tominotosa	250	02.10	04.2	01.9	15.7	04.4
(E_6)	500	06.10	11.0	10.7	18.5	44.4

(60.46%), *Rhizoctonia solani* (51.6%), *Fusarium* (47.9%) and *phytophthora* (28.8%) (Table 3).

All extracts showed high antifungal activity against *stemphylim solani* E_1 (66.28%), E_6 (44.4%), E_2 (32.8%) E_3 (36.9%), E_5 (31.9%) and E_4 (29.7%) (Table 3).

The significance of indigenous products for plant disease control has been investigated in other studies and encouraging results are reported[9-11].

This is the first report of antifungal activity (against plant pathogenic fungi) of extracts from medicinal plants collected from the Jordan environment. The preliminary results presented in this study shed the light on the ability of extracts from medicinal plants to be used against different fungi that cause plant diseases, but further studies are need to investigate the inhibitory activities of extracts from other medicinal plants on various fungi and plant pathogenic bacteria that cause severe yield losses to different crops in Jordan. In addition, efforts should be done to identify the active compounds that cause growth inhibition and try to integrate these compounds in the control programs used to reduce yield losses caused by different plant pathogens.

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