J. Agric. Sci. Mansoura Univ., 28 (10): 7419 - 7436, 2003

ANTIMICROBIAL ACTIVITIES OF CERTAIN EGYPTIAN MEDICINAL PLANT EXTRACTS AND ESSENTIAL OILS

Abdel-Fatah, M.K.*; M.E. Osman; F.H. Ahmed*; Mona I. Mabrouk*.**

* National Organization for Drug Control and Research (NODCAR)

** Faculty of Science, Helwan University.

ABSTRACT

Sixteen Egyptian medicinal plants namely Harmela , Arghel, Chaste tree, Egyptian acacia, Lemon grass, Fennel, Garden chrysanthemum, Oregano, Jojoba, Common rue, Great plantein, Camel's hey, Ben-nut, Cypress, Juniper and Egyptian Balsam were screened for their antimicrobial activity *in vitro* against six Gram - positive bacteria [*Bacillus subtilis* (CAIM1007), *Bacillus pumilus* (CAIM 1303), *Staphylococcus aureus* (CAIM 1352), *Staphylococcus epidermidis* (CAIM 1353), *Streptococcus* sp. (CAIM 1221) and *Micrococcus lutues* (CAIM 1246)] and seven Gram negative - bacteria [*Escherichia coli* (CAIM 1357), *Pseudomonas fluorescens* (CAIM 1221), *Salmonella typhimurium* (CAIM 1350), *Salmonella typhi*, *Salmonella treforest*, *Shigella* sp. (NMRO) and *Bordetella bronchiseptica* (ATCC 4617)] as well as two yeasts [(*Candida albicans* (CAIM 22) and *Saccharomyces cerevisiae* (CAIM 14)] and two fungal strains [*Aspergillus niger* and *Aspergillus flavus*]. The obtained results indicated that the water extract of Oregano (*Origanum vulgare*) and Ben–nut (*Moringa peregrina*) only gave highly antimicrobial activity, while the organic solvent extracts (Ethanol and Petroleum ether) of Oregano, Juniper, Harmela, Arghel and Camel's hey were found to possess moderate activities against different microorganisms. Data also revealed that all oils have high activities in comparison with other extracts and oil of (*Origanum vulgare*) and (*Moringa peregrina*) were the most effective against all the tested microorganisms.

INTRODUCTION

Since prehistoric times, in Egyptian traditional medicine the use of plants in the form of crude extracts, infusions or plasters is a widespread practice to common infection. There is rich local ethnobotanical bibliography describing the species most frequently used by the population to cure gastrointestinal, respiratory, urinary tract and skin infections. (Hewedi, 1977; El-Hadidy *et al.* 1981; El-Ballal *et al.* 1991; El-Shourbagy *et al.* 1993).

Literature are rich in information about plants that possess variable efficacy of antimicrobial activity (Khan *et al.* 1980), based on their use in the folk medicine and sometimes randomly, plants are selected to screening studies to clarify the efficacy of their crude extracts on different types of microbes (Farnsworth *et al.* 1972). The later represents Gram-positive bacteria, Gram-negative bacteria, yeast, fungi, actinomycs and viruses .... etc. However, few number of these plants have been chemically studied for the purpose of isolation and identification of their active agents, which are responsible for the claimed activity.

Alicia *et al.* (1981) reported that antimicrobial agents from higher plants are few (14%), while Paster *et al.* (1990) found that origanum oil completely

inhibited mycelial growth of *A. niger* and *A. flavus*. Also Deans *et al.* (1992) found that oil of origanum and lemon grass were highly active against certain bacterial species. While Baratta *et al.* (1998) showed that origanum oil has the highest braadest activity against sevral bacteria and fungal species. Hammar *et al.* (1999) and Ozcan and Erkmen (2001) found that oregano oil have highly activity against 10 genera of test organisms.

Ramadan *et al.* (1994) found that alcoholic extracts from 20 wild medicinal plants has strong activity against different test organisms.

Hassanein and Eldoksch (1997) found that some of oily petroleum ether extracts have antimicrobial activity.

Jit *et al.* (1980) and Brantner *et al.* (1996) found that ethyl ether and ethanolic extract of medicinal plants have activity against test organisms.

In Egypt, although folk medicine comprises some antimicrobial plant drugs, most of them have not been subjected to fulfill the above purpose. Therefore, the present study was designed to investigate the antimicrobial activity of different extracts and the oils of some medicinal plants.

MATERIAL AND METHODS

Materials :

Sixteen Egyptian medicinal plants were collected from Arab Co. for Pharmaceutical and Medicinal plants, Agriculture Station, Sharkiah Governorate.

Test organisms :

Six Gram - positive bacteria and seven Gram negative - bacteria as well as two yeasts and two fungal species (Table 1) were kindly supplied from the Microbiological Resource Center, Ain shams Univ., Fac. of Agric., Egypt (CAIM, Cairo Mircen), and from Bacteriological Dep. of (NODCAR), ATCC, American Type Culture Collection.

Media :

Nutrient agar medium (Jacobs and Gerstein, 1960) was used for growing the Gram-positive and Gram-negative bacteria. Sabouraud dextrose agar medium (Jacobs and Gerstein, 1960) was used for growing yeasts.

Methods :

1- Preparation of the plant materials :

The plant samples were collected from cultivated fields. There was no treatment with chemical fertilizers or pesticides. The collected plant samples were air dried in shade at room temperature and then subjected to dryness in the oven at 45 °C to constant moisture content, then pulverized. The powders were kept away from moisture in well-closed plastic containers ready for investigation. Plants obtained from herbalists were treated similarly. However, the seeds were powdered just before examination (Khalifa, 1998).

2- Preparation of water extracts:

Fifty-gram samples of every plant material were ground using small-sized mortars. The content of each mortar was transferred to a flask

Table (1) : The antagonistic effect of some plant oils (100%) against some microorganisms.

Test organisms		Diameter of inhibition zone in mm.														
		Harmela	Arghel	Chaste tree	Egyptian acacia	Lemon grass	Fennel	Garden chrysanthemum	Oregano	Jojoba	Common rue	Great plantain	Camel's hay	Ben - nut	Cypress	Juniper	Egyptian Balsam
Gram- positive	B. subtilis	0	0	0	0	8	13	0	33	9	15	0	15	25	9	10	0
	B. pumilus	0	0	0	0	7	12	0	30	9	11	0	10	30	10	10	0
	Staph. aureus	0	7	0	0	7	15	12	30	9	12	0	17	32	0	0	0
	Staph. epidermidis	0	0	0	0	10	14	9	9	13	10	0	15	32	0	0	0
	Streptococcus sp.	0	12	0	0	9	0	11	17	9	11	0	10	30	15	0	0
	M. luteus	0	10	0	0	13	18	7	15	9	0	0	12	37	6	0	0
Gram- negative	*E. coli*	0	9	0	0	11	13	0	18	10	0	0	18	0	0	0	0
	Ps. fluorescens	0	0	0	0	10	13	7	17	9	10	0	12	30	8	8	0
	S. typhimurium	0	10	0	0	11	12	0	30	20	0	0	14	30	7	8	0
	S, typhi	0	0	0	0	8	0	12	35	9	0	0	9	19	0	6	0
	S. treforest	0	7	0	0	10	15	9	20	10	8	0	14	26	0	9	0
	Shigella spp.	0	9	0	0	9	20	0	22	10	0	0	15	27	0	7	0
	Bordetella spp.	0	0	0	0	7	20	0	18	10	7	0	19	37	0	12	0
Yeasts	*C. albicans*	0	11	0	0	10	15	0	11	10	0	0	15	0	10	0	0
	Sacch.cerevisiae	0	0	0	0	12	15	0	10	9	0	0	17	0	0	0	0
Fungi	*Asp. niger*	0	0	0	0	0	0	0	14	11	0	0	0	25	9	0	0
	Asp. flavus	0	0	0	0	0	0	0	0	0	0	0	0	20	13	0	0

containing 200 ml of water, then boiled for 10 minutes and cooled. The mixture was left to settle at room temperature. After settling, the supernatant was aspirated in a sterile conical flask. All watery extracts were sterilized by filtration through zeits filter using positive pressure and glass filter G_5 then, the filtrate was kept in refrigerator at 4±C till use, according to the Egyptian Pharmacopoeia (1984).

3- Preparation of organic solvent extracts:

Fifty grams of the air-dried powdered sample of every plant material, was successively extracted with 200 ml of petroleum ether (40-60±C) and 200 ml of ethanol 95 % in a soxhlet apparatus. The solvent in each was evaporated at 40±C under reduced pressure to constant weight.

4- Preparation of volatile oils:

The dried powdered sample (500 gm) was covered with sufficient water in closed flask and left for 2hr at room temperature, it was then subjected to steam distillation in a modified likens and Nickers on apparatus which allows the simultaneous extraction of the volatile oil fraction, with a characteristic odor (Guenther,1950). Oils stocks and extracts were kept at 4±C throughout the whole investigation.

5- Preparation of fixed oils (*Moringa peregrina*) :

The oils were extracted by boiling seeds with water and collecting the oil from the surface of the water according to the method described by Somali *et al.*(1984).

6- The antimicrobial activities of the medicinal plant extracts:

The antimicrobial activities of medicinal plant extracts were determined using the diffusion method as described by British Pharmacopoeia (1968). Each medium was sterilized by autoclaving at 121±C for 15 minutes, cooled to 45±C and inoculated with 1ml /100 ml of the test organisms and thoroughly mixed by rotatory motion of the flasks and poured into a sterile plates and left for solidification at room temperature. Four holes (diameter 10 mms) in each plate were filled with sterile supernatant to be tested and the plates were left at room temperature for one hour and then incubated at 37°C for 24 hr in case of bacteria; and at 28°C for 48 hr in case of yeast. The diameter of inhibition zones was measured in millimeters. The mean value of inhibition zones was the average of three readings.

7- The antimicrobial potentialities of medicinal plant oils :

The antimicrobial activities of the tested oils were carried out by disc diffusion technique as described in British Pharmacopoeia (1968). Nutrient agar was melted at 45°C and inoculated by the cell suspension 1 ml /100 ml bacteria or yeast.

The flask was shaken vigorously and poured into petri–dishes (15-cm in diameter). Filter paper discs 6 mm diameter whatman No.2 were thoroughly moistened with (50 ig / ml) at different concentrations crude tested oils (0.1%, 1%, 10%, 100%), acetone was used as a solvent for all the tested oils (Ahmed *et al.* 1994).

The treated discs were aseptically transferred and left for evaporation of the solvent and then placed upon the surface of the inoculated plates with tested organisms, and kept in a refrigerator for one hour to permit diffusion of antimicrobial substances and the plates were incubated at the same conditions. The zone of inhibition was measured in the same way.

8- The antimicrobial activities of antibiotics as comparative studies :

The antimicrobial activities were carried out by disc diffusion technique as described in British Pharmacopoeia (1968). Nutrient agar was melted at 45°C and inoculated by the cell suspension (1 ml /100 ml) bacteria or yeast. The flask was shaken well and poured into a petri–dish (15-cm in diameter). Filter paper discs (6 mm) Whatman No.2 were thoroughly moistened by antibiotics (50 μg), the treated discs were aseptically transferred and placed upon the surface of the inoculated plates with tested organisms, and kept in a refrigerator for one hour to permit diffusion of antimicrobial substances. The plates were incubated at the same conditions. The zone of inhibition was measured in the same way.

9- Selected plants :

Origanum vulgare and *Moringa peregrina* were chosen for further studies because of their high antimicrobial activity.

RESULTS

The antimicrobial activity of sixteen medicinal plant oils were tested against 17 test organisms which belonged to six Gram-positive, seven Gram-negative bacterial species, two yeast species and two fungal species are presented in Tables (1-4) which showed that the different concentrations of plant oils (100%, 10%, 1%, 0.1%) are differ greatly in their antimicrobial activities.

Data revealed that oregano, jojoba, lemon grass, fennel, camel's hay and ben-nut oils were the most effective against 15-16 test organisms at different concentrations (1.0-100%). While arghal garden chrysanthemum, common rue, cypress and juniper were the most effective against 8 test organisms at the same concentration. On the other hand, harmela, chaste tree, Egyptian acacia, great plantain and Egyptian balsam haven't any activity against any test organisms.

Concerning the antimicrobial activity of water extracts, 14 medicinal plants were record active against Gram-positive, Gram-negative bacteria, yeasts and fungi (Table 5). The water extract of ben-nut showed that the most effective extract against *M. lutes*, *Bordetella spp.*, *B. subtilis*, *B. pumilus*, *Staph. aureus*, *Staph. epidermidis*, *Streptococcus spp.*, *Shigella spp.*, *A. niger* and *A. flavus*. Certain water extracts of the tested plants exhibited an antimicrobial activity against Gram-negative bacteria like (*E. coli*, *S. typhi* and *Shigella spp.*).

Petroleum ether extracts of the tested medicinal plants were tested as antimicrobial agents Gram-positive, Gram-negative bacteria, yeasts and fungi (Table 6).The obtained results showed that juniper was more effective

Table (2) : The antagonistic effect of some plant oils (10%) against some microorganisms.

Test organisms		Diameter of inhibition zone in mm.													
		Harmela	Arghel	Chaste tree	Egyptian acacia	Lemon grass	Fennel	Garden chrysanthemum	Oregano	Jojoba	Common rue	Great plantain	Camel's hay	Ben - nut	Cypress	Juniper	Egyptian Balsam
Gram- positive	*B. subtilis*	0	0	0	0	0	8	0	14	8	0	0	12	19	6	0	0
	B. pumilus	0	0	0	0	0	10	0	10	9	9	0	9	20	6	6	0
	Staph. aureus	0	7	0	0	7	12	10	10	8	0	0	9	15	0	0	0
	Staph. epidermidis	0	0	0	0	0	10	0	9	10	0	0	12	10	0	0	0
	Streptococcus sp.	0	9	0	0	0	0	0	6	9	0	0	9	10	7	0	0
	M. luteus	0	9	0	0	0	12	0	6	0	0	0	9	20	0	0	0
Gram- negative	*E. coli*	0	8	0	0	0	12	0	12	9	0	0	10	0	0	0	0
	Ps. fluorescens	0	0	0	0	0	13	0	16	0	0	0	11	18	0	8	0
	S. typhimurium	0	7	0	0	8	11	0	19	0	0	0	13	16	0	7	0
	S, typhi	0	0	0	0	7	0	0	16	8	0	0	9	10	0	0	0
	S. treforest	0	0	0	0	0	11	0	9	0	0	0	10	19	0	6	0
	Shigella spp.	0	9	0	0	8	14	0	6	9	0	0	9	10	0	6	0
	Bordetella spp.	0	0	0	0	6	13	0	9	0	0	0	9	20	0	10	0
Yeasts	*C. albicans*	0	7	0	0	0	12	0	10	0	0	0	0	0	9	0	0
	Sacch.cerevisiae	0	0	0	0	0	13	0	7	7	0	0	0	0	0	0	0
Fungi	*Asp. niger*	0	0	0	0	0	0	0	6	0	0	0	0	15	0	0	0
	Asp. flavus	0	0	0	0	0	0	0	0	0	0	0	0	13	0	0	0

Table (3) : The antagonistic effect of some plant oils (1%) on some microorganisms

Test organisms		Diameter of inhibition zone in mm.													
		Harmela	Arghel	Chaste tree	Egyptian acacia	Lemon grass	Fennel	Garden chrysanthemum	Oregano	Jojoba	Common rue	Great plantain	Camel's hay	Ben - nut	Cypress	Juniper	Egyptian Balsam
Gram- positive	*B. subtilis*	0	0	0	0	0	7	0	13	0	0	0	7	10	6	0	0
	B. pumilus	0	0	0	0	0	10	0	9	8	0	0	9	15	6	0	0
	Staph. aureus	0	6	0	0	7	10	0	9	0	0	0	9	10	0	0	0
	Staph. epidermidis	0	0	0	0	0	8	0	7	0	0	0	11	9	0	0	0
	Streptococcus sp.	0	8	0	0	0	0	0	0	7	0	0	8	10	6	0	0
	M. luteus	0	7	0	0	0	12	0	0	0	0	0	7	10	0	0	0
Gram- negative	*E. coli*	0	0	0	0	0	11	0	0	0	0	0	7	0	0	0	0
	Ps. fluorescens	0	0	0	0	0	12	0	11	0	0	0	9	8	0	0	0
	S. typhimurium	0	7	0	0	6	8	0	11	0	0	0	11	9	0	0	0
	S, typhi	0	0	0	0	7	0	0	0	0	0	0	0	6	0	0	0
	S. treforest	0	0	0	0	0	8	0	0	0	0	0	7	10	0	0	0
	Shigella spp.	0	8	0	0	6	14	0	0	8	0	0	9	10	0	0	0
	Bordetella spp.	0	0	0	0	6	11	0	7	0	0	0	0	10	0	6	0
Yeasts	*C. albicans*	0	6	0	0	0	11	0	8	0	0	0	0	0	0	0	0
	Sacch.cerevisiae	0	0	0	0	0	12	0	0	0	0	0	0	0	0	0	0
Fungi	*Asp. niger*	0	0	0	0	0	0	0	0	0	0	0	0	9	0	0	0
	Asp. flavus	0	0	0	0	0	0	0	0	0	0	0	0	13	0	0	0

Table (4) : The antagonistic effect of some plant oils (0.1%) on some microorganisms.

Test organisms		Diameter of inhibition zone in mm.												
		Harmela	Arghel	Chaste tree	Egyptian acacia	Lemon grass	Fennel	Garden chrysanthemum	Oregano	Jojoba	Common rue	Great plantain	Camel's hay	Ben - nut	Cypress	Juniper	Egyptian Balsam
Gram- positive	*B. subtilis*	0	0	0	0	0	7	0	11	0	0	0	6	8	0	0	0
	B. pumilus	0	0	0	0	0	8	0	0	7	0	0	8	9	0	0	0
	Staph. aureus	0	6	0	0	7	7	0	0	0	0	0	8	9	0	0	0
	Staph. epidermidis	0	0	0	0	0	7	0	6	0	0	0	9	6	0	0	0
	Streptococcus sp.	0	0	0	0	0	0	0	0	0	0	0	7	6	0	0	0
	M. luteus	0	0	0	0	0	10	0	0	0	0	0	7	9	0	0	0
Gram- negative	*E. coli*	0	0	0	0	0	10	0	0	0	0	0	7	0	0	0	0
	Ps. fluorescens	0	0	0	0	0	11	0	9	0	0	0	7	6	0	0	0
	S. typhimurium	0	7	0	0	0	8	0	10	0	0	0	7	6	0	0	0
	S. typhi	0	0	0	0	7	0	0	0	0	0	0	0	6	0	0	0
	S. treforest	0	0	0	0	0	7	0	0	0	0	0	7	6	0	0	0
	Shigella spp.	0	7	0	0	6	13	0	0	7	0	0	6	0	0	0	0
	Bordetella spp.	0	0	0	0	6	10	0	0	0	0	0	0	0	0	0	0
Yeasts	*C. albicans*	0	0	0	0	0	10	0	0	0	0	0	0	0	0	0	0
	Sacch.cerevisiae	0	0	0	0	0	10	0	0	0	0	0	0	0	0	0	0
Fungi	*Asp. niger*	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Asp. flavus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Table (5) : The antagonistic effect of water extracts of plants on some microorganisms.

Test organisms		Diameter of inhibition zone in mm.													
		Harmela	Arghel	Chaste tree	Egyptian acacia	Lemon grass	Fennel	Garden chrysanthemum	Oregano	Jojoba	Common rue	Great plantain	Camel's hay	Ben - nut	Cypress	Juniper	Egyptian Balsam
Gram- positive	*B. subtilis*	0	0	0	0	0	27	0	21	15	25	0	0	29	0	19	21
	B. pumilus	0	0	0	0	0	26	0	16	0	15	21	0	31	0	23	0
	Staph. aureus	0	0	16	0	0	26	0	21	0	14	25	0	33	23	0	0
	Staph. epidermidis	0	0	0	0	0	25	0	19	0	0	0	0	30	17	0	0
	Streptococcus sp.	20	0	0	0	0	0	0	24	0	21	23	0	29	0	20	0
	M. luteus	0	0	0	0	0	22	0	0	0	22	0	0	38	0	0	0
Gram- negative	*E. coli*	0	0	0	0	0	26	0	29	0	22	0	0	0	23	0	0
	Ps. fluorescens	0	0	18	0	0	24	0	16	0	15	20	0	32	0	0	0
	S. typhimurium	25	0	16	0	0	25	0	30	0	18	0	0	32	0	20	21
	S, typhi	40	0	0	0	0	0	0	33	0	26	0	0	0	0	0	28
	S. treforest	28	0	0	0	0	27	0	18	0	22	0	0	33	0	0	25
	Shigella spp.	32	0	0	0	0	25	0	22	0	0	24	0	31	0	43	35
	Bordetella spp.	26	0	0	0	0	26	0	25	0	22	0	0	38	0	0	14
Yeasts	*C. albicans*	0	0	0	0	0	40	0	15	0	0	0	0	0	0	0	0
	Sacch.cerevisiae	0	0	0	0	0	30	0	13	0	0	0	0	0	0	0	23
Fungi	*Asp. niger*	0	0	0	0	0	0	0	0	0	0	0	0	20	0	0	0
	Asp. flavus	0	0	0	0	0	0	0	0	0	0	0	0	10	0	0	0

Table (6) : The antagonistic effect of Petroleum ether (40-60°C) extracts of plants on some microorganisms.

Test organisms		Diameter of inhibition zone in mm.													
		Harmela	Arghel	Chaste tree	Egyptian acacia	Lemon grass	Fennel	Garden chrysanthemum	Oregano	Jojoba	Common rue	Great plantain	Camel's hay	Ben - nut	Cypress	Juniper	Egyptian Balsam
Gram- positive	*B. subtilis*	0	0	0	10	8	0	0	7	8	8	9	0	7	10	11	0
	B. pumilus	0	0	0	11	9	0	0	8	8	12	9	8	12	10	8	0
	Staph. aureus	0	0	0	11	0	0	0	0	0	12	10	0	0	11	0	0
	Staph. epidermidis	0	0	0	8	7	0	0	12	8	8	10	20	7	8	9	0
	Streptococcus sp.	0	0	0	11	0	0	0	11	0	8	8	0	0	12	15	11
Gram- negative	*M. luteus*	13	13	0	9	29	0	0	14	0	9	9	0	10	9	30	0
	E. coli	0	0	0	9	0	0	0	0	0	10	11	0	0	0	0	0
	Ps. fluorescens	0	0	0	9	13	0	0	11	8	9	9	0	0	7	10	0
	S. typhimurium	0	7	8	8	11	0	0	8	8	8	10	8	10	9	8	0
	S. typhi	0	0	0	11	0	0	0	0	7	9	10	0	0	12	0	0
	S. treforest	0	0	0	7	17	0	0	14	0	8	0	10	20	9	27	0
	Shigella spp.	0	0	0	9	0	0	0	0	0	12	12	0	0	11	0	0
	Bordetella spp.	7	8	0	9	7	0	0	16	0	8	8	0	7	8	12	7
Yeasts	*C. albicans*	0	0	0	7	0	0	0	0	0	8	8	10	0	10	0	0
	Sacch.cerevisiae	0	0	0	0	0	0	0	0	0	0	0	13	0	12	0	0
Fungi	*Asp. niger*	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Asp. flavus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

against *B. subtilis*, *Streptococcus spp.*, *M. lutes* and *S. treforest*. While common rue was the most effective against *B. pumilus*, *Staph. aureus* and *Shigella spp.* . The Camel's hay was the most effective against *Staph. epidermidis* and *Sacc. cerevisiae*. Also, the results showed that cypress was the most effective in the inhibition of *C. albicans* and *Sacc. cerevisiae*.

Also, the alcoholic extracts of the tested medicinal plants were investigated as antimicrobial activity against 17 test organisms (Table 7). Results revealed that alcoholic extract of harmela was the most effective against *B. subtilis, Staph. epidermidis, Streptococcus spp., M. lutes, E. coli, Ps. fluorescens, S. typhimurium, S. typhi, S. treforest, Shigella spp.* . Also, alcoholic extract of oregano was the most effective against *A. niger*, alcoholic extract of Egyptian balsam was the most effective against *Staph. aureus* and *Sacc. cerevisiae*, alcoholic extract of lemon grass was the most effective against *B. pumilus*, alcoholic extract of juniper was the more effective against *S. typhimurium* and *Sacc. cerevisiae* and alcoholic extract of oregano, juniper, Egyptian acacia, arghel and Harmela showed marked activity against most Gram-positive and Gram-negative bacteria.

The volatile oils of *Origanum* and *Moringa* possessed a potent antimicrobial activity against test organisms as revealed by comparison with standard antibiotics (Table 8).

Data reveled that all oils have high activities in comparison with other extracts and oil of *Origanum vulgare* and *Moringa peregrina* were the most effective. Therefore subjected for further studies.

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DISCUSSION

The discovery of the relationship which exists between microorganisms and diseases which occurred around the beginning of the 19th century, brought forth a more extensive use of the volatile oils and their derivatives for purpose of controlling microbial growth and of the effects of the later on the host or on the environment in which it occur, *i.e.*, as a preservative against decomposition due to fermentation and /or putrefaction.

At first only the crude materials were employed, such as spies, barks, herbs, flowers, etc.. later, after the process of distillation was discovered and perfected, the respective constituent oils were obtained from the crude sources.

Antimicrobial agents from higher plants are few (14%) as compared to the total antibiotics obtained from different forms of life (Alicia *et al.* 1981). Antibiotics from higher plants may be less toxic to the host, cheaper, more available especially during war time and since they are from local sources, no need for foreign exchange to import them as compared to other antibiotics.

So, this study was carried out to investigate the antimicrobial activity of some medicinal plants oils and their extracts, namely (Harmela , Egyptian Balsam, Arghel , Camel's hey, Lemon grass, Egyptian acacia , Ben – nut,

Table (7) : The antagonistic effect of Alcoholic extracts of plants on some microorganisms.

	Test organisms	Diameter of inhibition zone in mm.													
		Harmela	Arghel	Chaste tree	Egyptian acacia	Lemon grass	Fennel	Garden chrysanthemum	Oregano	Jojoba	Common rue	Great plantain	Camel's hay	Ben - nut	Cypress	Juniper	Egyptian Balsam
Gram- positive	B. subtilis	31	11	0	17	8	0	0	0	0	12	8	0	0	9	7	0
	B. pumilus	14	0	0	9	16	0	0	0	0	0	0	14	0	0	0	0
	Staph. aureus	13	10	0	14	0	0	0	8	0	0	0	14	0	0	10	16
	Staph. epidermidis	18	11	9	12	8	0	0	0	0	8	0	7	0	10	11	12
	Streptococcus sp.	16	12	14	14	7	0	0	15	15	0	0	0	0	0	12	0
	M. luteus	22	10	15	17	12	0	0	0	0	10	9	7	0	8	11	0
Gram- negative	*E. coli*	28	11	12	12	8	0	0	0	0	8	0	0	0	0	11	7
	Ps. fluorescens	26	18	9	15	17	0	0	20	0	10	11	20	0	11	18	22
	S. typhimurium	21	0	13	13	0	0	0	0	0	9	0	0	0	8	20	0
	S. typhi	18	0	0	13	0	0	0	0	0	9	10	0	0	8	12	0
	S. treforest	14	0	0	14	7	0	0	0	0	11	8	0	0	8	0	0
	Shigella spp.	12	0	0	12	8	0	0	0	10	9	8	0	0	8	0	0
	Bordetella spp.	12	7	7	14	13	0	0	9	0	8	8	9	0	10	0	9
Yeasts	*C. albicans*	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Sacch.cerevisiae	0	0	0	0	0	0	0	0	0	0	0	0	0	0	18	23
Fungi	*Asp. niger*	0	12	14	0	20	0	0	24	8	0	0	16	0	0	0	13
	Asp. flavus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Table (8) : Effect of some antibiotics on some microorganisms in comparison with *Origanum* and *Moringa* oils.

Test organisms		Diameter of inhibition zone in mm								
		Tetracycline	Amoxicillin	Ampicillin anhydrous	Ampicillin	Chloramphenicol	Gentamicin	Mycostatin	*Origanum* oil	*Moringa* oil
Gram-negative	*Escherichia coli*	25	0	0	0	30	0	0	18	0
	Pseudomonas fluorescens	30	26	25	22	25	0	0	17	30
	Shigella spp.	20	18	21	19	25	0	0	22	27
	Bordetella spp.	32	38	36	37	26	0	0	18	37
	Salmonella typhimurium	35	30	28	25	20	0	0	30	30
	Salmonella treforest	24	22	26	25	24	0	0	20	26
Gram-positive	*Staphylococcus aureus*	26	37	36	35	25	0	0	30	32
	Staphylococcus epidermidis	34	32	31	25	32	0	0	9	32
	Bacillus subtilis	30	20	20	18	22	0	0	33	25
	Bacillus pumilus	28	22	21	20	25	0	0	30	30
	Micrococcus luteus	26	55	60	60	18	0	0	15	37
Yeasts	*Candida albicans*	0	0	0	0	0	25	0	11	0
	Saccharomyces cerevisiae	0	0	0	0	0	28	0	10	0

Fennel, Common rue, Cypress, Jojoba, Oregano, Garden chrysanthemum, Juniper, Great plantain, and Chaste tree).While the test organisms included 6 strains (Gram-positive bacteria), 7 strains (Gram-negative bacteria), 2 yeast strains and 2 fungal strains. This part of the study was carried out by agar diffusion method.

Concerning the effects of the plant oils and extracts on the tested organisms, the results showed that the higher dilution of these plant oils (0.1% of the original) gave the lower activity against the same test organisms.

Data also revealed that *Origanum* oil was the most effective against *B. subtilis*, *B. pumilus*, *C. albicans*, *Sacc. cerevisiae*, *A. niger*. The results are in agreement with Paster *et al.* (1990), who found that *Origanum* oil completely inhibited mycelial growth of *A. niger*, and *A. flavus*, they also reported that growth was inhibited in the case of *Staph. aureus* and *S. typhimurium*.

Also, Deans *et al.* (1992) found that oil of *Origanum* and Lemon grass were highly active against bacterial species including plant and animal pathogens and food spoilage organisms. While Baretta *et al.* (1998) showed that origanum oil has the highest and broadest activity against 25 genera of bacteria and fungal species. Montes *et al.* (1998) found that Origanum vulgare has activity against 8 strains of bacterial species.

Hammar *et al.* (1999) and Ozcan and Erkmen (2001) found that oregano oil have antimicrobial activity against *S. typhimurium*, *B. cereus*, *Staph. aureus*, *E. faeolis*, *E. coli*, *Y. enterocolitica*, *Sacc. cerevisiae*, *C. rugosa*, *Rh. oryzae* and *A. niger* at different concentration.

Therefore, *Moringa peregrina* also gave high activity against different test organisms Jahn (1981 and 1986) and Sattaur (1993) found that domestic water treatment with Moringa became a low cost technology to be utilized in improving water and health and used as efficient elimination of indicator bacteria and also Skandamis *et al.* (2001) found that of addition oregano essential oil at the time of inoculation caused increase in the lag phase, decrease in the maximum growth rate and reduced final population size of *E. coli*.

Results revealed that Lemon grass oil has activity against 14 test organisms. Such results are in line with those of Alam *et al.* (1994) who reported that Lemon grass oil has activity against *B. subtilis* and *E. coli*, also are in agreement with Pattnaik *et al.* (1996) who proved that Lemon grass oil has antimicrobial activity against 22 bacterial strains and 12 fungal.

Concerning the antimicrobial activity of water extracts, 14 medicinal plants were record active against Gram positive, Gram negative bacteria, yeasts and fungi (Table5). The water extract of ben-nut showed that the most effective extract against 13 test organisms. Certain water extracts of the tested plants exhibited an antimicrobial activity against Gram-negative bacteria like (*E. coli*, *S. typhi* and *Shigella spp.*). Theses findings were in accordance with Ramadan *et al.* (1994) found that aqueous extract of 20 wild medicinal plants have strong activity against *Staph. aureus*, *Salmonella typhi*, *E. coli* and *Ps. aeruginosa*. Also are in line with Qureshi *et al.* (1997) who showed that extracts from 18 plant species have antifungal activity. Also, Thiribhuvanamala and Narasimhan (1998) found that aqueous leaf extracts

from 22 plant species inhibited the spore germination, mycelial growth and spore production of seed born pathogens.

The obtained results of petroleum ether extracts of the tested medicinal plants showed that juniper was more effective against *B. subtilis*, *Streptococcus spp.*, *M. luteus* and *S. treforest*. While common rue was the most effective against *B pumilus*, *Staph. aureus* and *Shigella spp.* . The camel's hay was the most effective against *Staph. epidermidis* and *Sacc. cerevisiae*. Also, the results showed that cypress was the most effective in the inhibition of C. albicans and *Sacc. cerevisiae*. The results were in agreement with Hassanien and Eldoksch (1997) who found that some of oily petroleum ether extracts have antimicrobial activity.

Results in (Table7) revealed that alcoholic extract of Harmela was the most effective against 10 test organisms. Also, alcoholic extract of oregano was the most effective against *A. niger*, alcoholic extract of Egyptian balsam was the most effective against *Staph. aureus* and *Sacc. cerevisiae* , Lemon grass was the most effective against *B. pumilus* , while alcoholic extract of oregano, juniper, Egyptian acacia Arghel and Harmela showed marked activity against most Gram-positive and Gram-negative bacteria.

The results are in agreement with Jit et al. (1980) who found that ethyl ether and 50% ethanolic extract of 5 medicinal plants were active against *Staph. aureus*, *E. coli* and *C. albicans* while El Hady *et al.* (1994) indicated that methanol water extract of Solenstemma argel have activity against 8 bacterial strains and 14 fungi.

Ramadan et al., (1994) found that alcoholic extracts from 20 wild medicinal plants from qassim area (Saudi Arabia) have strong antimicrobial activity against *Staph. aureus*, *E. coli*, *Ps. aeroginosa* , but tested fungi and yeast were less sensitive.

Brantner *et al.* (1996) found that ethanolic extracts prepared from various plant parts (leaves, flowers, fruits, bark and roots) have inhibitory activity against *Staph. aureus*, *Strept. Faecalis*, *M. luteus* and *Shigela sonnei*.

Akhtar *et al.* (1997) reported that crude extracts of acacia nilotica have a high level of inhibition on the tested bacterial strains.

Nelson, (1997) indicated that juniper oil is effective against *Staph. aureus* and *Streptococcus spp.* .

Generally speaking, extraction of the tested medicinal plants by different solvents led to various effects on the tested organisms, either bacteria or fungi. These results may be due to the presence of different compounds of the extracts also, the variation of the plant organs as well as the method of extraction led to various antimicrobial effects since the inhibition of microbial growth caused by volatile oils especially of *Origanum* and *Moringa* oils are much higher than the inhibition caused by other extracts, so the oils of these plants were used in further investigation.

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النشاط المضاد لمستخلصات بعض النباتات الطبية المصرية والزيوت الأساسية على بعض الميكروبات

محمد كمال عبد الفتاح* ، محمد السيد عثمان ،فتح الله حسن احمد*و منى ابراهيم مبروك***

*** الهيئة القومية للرقابة والبحوث الدوائية**

**** كلية العلوم- جامعة حلوان**

تم دراسة النشاط التضادى لستة عشر نباتا طبيا مصريا على بعض الميكروبات الممرضة سواء كان مستخلص مائى أو كحولى أو أثيرى أو زيت.

فقد تمت الدراسة على ستة أنواع من البكتريا الموجبة لجرام وسبعة أنواع من البكتريا السالبة لجرام وأثنين من الخمائر وأثنين من الفطريات الممرضة .

أوضحت الدراسة أن النشاط التضادى للخلاصة الكحولية والأثيرية والمائية لنبات الأوريجانو ونبات الحبة الغالية أكثر فاعلية وكذلك زيوت هذه النباتات أيضا أكثر فاعلية من غيرها من النباتات الطبية المختبرة.