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Antimicrobial Activity of Crude Extracts of Some Plant Leaves

[1]Maher Obeidat, [1]Mohamad Shatnawi, [2]Mohammad Al-alawi, [1]Enas Al-Zu'bi, [3]Hanee Al-Dmoor, [4]Maisa Al-Qudah, [3]Jafar El-Qudah and [1]Ismael Otri

[1]Department of Biotechnology, Faculty of Agricultural Technology, Balqa' Applied University, 19117, Al-Salt, Jordan

[2]Department of Plant Production and Protection, Faculty of Agricultural Technology, Balqa' Applied University, 19117, Al-Salt, Jordan

[3]Department of Nutrition and Food Processing, Faculty of Agricultural Technology, Balqa' Applied University, 19117, Al-Salt, Jordan

[4]Department of Allied Medical Sciences, Faculty of Al-Zarqa, Balqa' Applied University, Al-Zarqa, Jordan

Corresponding Author: Maher Obeidat, Department of Biotechnology, Faculty of Agricultural Technology, Balqa' Applied University, 19117, Al-Salt, Jordan Tel: +962777271071 Fax: +96253530469

ABSTRACT

The effect of ethanol, methanol, acetone and water extracts of leaves of 11 plant species, used in the folk medicine, against six antibiotic resistant clinical pathogens was evaluated by the agar-well diffusion method. The obtained results indicate that most of the extracts revealed antimicrobial activity. The water extract of *A. discoridis* leaves exerted significant effect and recorded the lowest MIC and MMC. Ethanol leaf extraction method is the best. It produced broad-spectrum of antimicrobial activity followed by methanol leaf extraction. Interestingly, methanol extraction method was found to be the most effective extraction method of anticandidal agents. Among the pathogenic bacteria tested, *S. pneumonia* was the least sensitive. Nevertheless, the anticandidal MIC and MMC values are higher than antibacterial values suggesting that *C. albicans* is less sensitive to plant leaf extracts. In conclusion, aqueous extracts of *A. discoridis* leaves exhibited the highest potency against all pathogens tested. Thus, this study confirms the efficacy of some plant extracts as natural antimicrobials and suggests the possibility of employing them in drugs for treatment of infectious diseases caused by the test pathogens.

Key words: Antimicrobial activity, inhibition zone, leaf extract, MIC

INTRODUCTION

Plants have been classified as an essential source of medicinal agents for centuries and a huge number of novel drug components have been isolated from natural plant sources. Many of these plants and their extracts were used in traditional medicine. Medicinal plants play a key role in health care with about 80% of the world's populations relying on the use of traditional medicine which is predominantly based on plants (Owolabi *et al*., 2007). According to WHO, medicinal plants would be the best source to obtain a variety of drugs. Plant derived medicines have made large contributions to human health (El-Astal *et al*., 2005). This is due to the significant healing power of the traditional medicinal systems (Adebolu and Oladimeji, 2005). Medicinal plants are distributed worldwide but they are most abundant in tropical countries (Elvin-Lewis, 2001; Naovi *et al*., 1991). Natural products from plant may offer new agents for antimicrobial use. A special feature of higher

plants is their capacity to produce a large number of organic chemicals of high structural diversity the so called secondary metabolites (Naovi *et al.*, 1991). Plants are rich in a wide variety of secondary metabolites with antimicrobial properties, such as tannins, terpenoids, alkaloids and flavonoids (Al-Momani *et al.*, 2007; Bisignano *et al.*, 2000; Bouzada *et al.*, 2009; Chakraborty and Brantner, 1999; Cowan, 1999; Setzer *et al.*, 2000; Sohail *et al.*, 2011).

The abundance of medicinal plants in nature and the traditional knowledge increase the understanding of the medicinal plants properties, safety and efficacy (Nascimento *et al.*, 2000). This concern has been expressed because of the resistance of clinically pathogenic microorganisms to the antibiotics that have been produced in the last decades (Cohen, 1992; Nascimento *et al.*, 2000). In the last decade, studies based on extraction of biologically active compounds from plant species used for medicinal purposes are intensively increased (Nascimento *et al.*, 2000; Rios and Recios, 2005).

Jordan has a unique location which was diverse in climate, geology and topography. Although Jordan is relatively a small country, it is characterized by a great variation in wild plants. Jordan has around 2500 wild plant species, of which 100 species are endemic, belonging to about 700 genera. From these plants, more than 500 species are classified as medicinal plants which are widely distributed all over the country and massively used in traditional medicine (Afifi and Irmaileh, 2000; Al-Eisawi, 1982; Oran, 1994; Oran and Al-Eisawi, 1998). These medicinal plants are used in Jordan for different biological and pharmaceutical activities including antifungal, antibacterial and insecticidal properties (Al-Mughrabi, 2003).

The rising prevalence of antibiotics resistant pathogenic microorganisms in the last decades raises the demand for finding new alternative antimicrobial agents. Therefore, the current study aim was to evaluate the antimicrobial activity of some local natural plants which have potential of treating infectious diseases and with lesser side effects compared to the synthetic drug agents. Leaf extracts of local 11 plant species have been screened for their possible antimicrobial activities on resistant strains of bacteria and yeast isolated from a local hospitalized patient. Relative few studies have been carried out to evaluate the antimicrobial properties of these plants. To our knowledge, the current study appears to be the first that point up the antimicrobial activity of *Arum discoridis* water extracts. In general, these plants are used in traditional medicine in the treatment of skin diseases, gastrointestinal tract diseases and respiratory problems. The plants used in this study and their properties are listed in Table 1.

Table 1: Uses of the selected plant species in local folk medicine

Scientific name	Common medicinal uses
Achillea membranacea	Stomach ailments, anti-inflammatory and anti-diarrhea.
Arum hygrophilum	Treatment of circulatory system problems and internal bacterial infection.
Arum discoridis	Treatment of circulatory system problems and internal bacterial infection.
Ecbalium elaterium	Treatment of edema.
Eminium spiculatum	Used as edible food.
Gundelia tournefortii	Decrease liver total cholesterols.
Lupinus varius	Treatment of kidney problems.
Mandragora autumnalis	Treatment of eye diseases, inflammation and ulcers.
Paronychia argentea	Treatment of tuberculosis.
Ruta chalepensis	Treatment of eye diseases and menstrual problems.
Urtica pilulifera	Anti-asthmatic and hypoglycemic.

MATERIALS AND METHODS

Collection of plant materials: Materials of 11 plant species (Table 1) were harvested in March 2010 from Irbid province, Jordan. They were carefully washed, oven-dried for 1 h at 160°C and put in the shade in an aerated place till complete drying, then were ground into a fine powder.

Preparation of plant extracts: The prepared powder was soaked in each of water, ethanol, methanol and acetone solvents (plant material to solvent ratio was 1:10, w/v) and extracted for 24 h at room temperature with shaking at 150 rpm. Filtrates of the extracts were dried at 40°C. The dried extracts were resuspended in Phosphate Buffered Saline (PBS) to bring to 500 mg mL^{-1} concentration.

Test microorganisms: A total of six clinical antibiotic-resistant microorganisms, including; five bacterial strains and one fungal strain mostly recorded in hospital infection were used in this study (Table 2), they were obtained from the Hospital of Jordan, Amman.

Microbial inoculums: A 24 h microbial cultures grown in the bacteriological Mueller-Hinton Broth (MHB) at 37°C and in the fungal Sabouraud Dextrose Broth (SDB) at 30°C were adjusted at 2×10^6 colony forming units (CFU mL^{-1}) and 2×10^5 spore mL^{-1}, respectively.

Antimicrobial activity screening tests: The antimicrobial activity of the test organisms to the 11 plant extracts was screened by using the agar-well diffusion method (Perez *et al*., 1990). An inoculum suspension was swabbed uniformly to solidified 20 mL Mueller-Hinton Agar (MHA) for bacteria and Sabouraud Dextrose Agar (SDA) for fungi and the inoculum was allowed to dry for 5 min. Holes of 6 mm in diameter were made in the seeded agar using sterile cork borer. Aliquot of 50 μL from each plant crude extract (500 mg mL^{-1}) was added into each well on the seeded medium and allowed to stand on the bench for 1 h for proper diffusion and thereafter incubated at 37°C for 24 h. The resulting inhibition zones were measured in millimeters (mm). The same procedure was followed for the fungus *C. albicans* but incubated at 30°C. Negative controls using 50 μL PBS were also run in the same manner and parallel to the treatments. These studies were performed in triplicate.

Determination of minimum inhibitory concentration and minimal microbial concentration: The Minimum Inhibitory Concentration (MIC) and Minimal Microbial Concentration (MMC) were determined for the active plant extracts that showed the widest

Table 2: Resistance of clinical test microorganisms to some standard antibiotics

	Standard antibiotics[b]							
Test microorganisms[a]	AMP10	CHL30	ERY15	NA30	NV30	P10	VA30	NYS10
Escherichia coli	R	R	R	S	R	R	R	ND
Salmonella typhimurium	R	S	S	R	S	R	S	ND
Pseudomonas aeroginosa	R	R	R	S	S	R	R	ND
Streptococcus pneumonia	R	R	R	S	S	R	R	ND
Staphylococcus aureus	R	S	R	S	S	R	R	ND
Candida albicans	R	R	R	R	R	R	R	S

[a]Bacteria tested in MHA medium, yeast in SDA. [b]AMP10: Ampicillin 10 μg, CHL30: chloramphenicol 30 μg, ERY15: Erythromycin 15 μg, NA30: Nalidixic acid 30 μg, NV30: Novobiocin 30 μg, P10: Penicillin G (10 Units), VA30: Vancomycin 30 μg, NYS10: Nystatin 10 μg. R: Resistant, S: Sensitive, ND: Not determined

spectrum of antimicrobial activity against test microorganisms. To determine MIC and MMC values, the methods of Dulger and Aki (2009) and Nakamura *et al.* (1999) were used and modified as previously described (Obeidat, 2011). The MIC was considered the lowest concentration of the sample that prevented visible growth. The MMC was defined as the lowest concentration yielding negative subcultures or only one colony. All samples were examined in triplicate.

Statistical analysis: The mean values were expressed as the Mean±Standard Deviation (SD) and were analyzed using one-way ANOVA (Tukey's studentized range) using the program SPSS 19.0 for Windows. Differences were considered significant at $p<0.05$.

RESULTS

The antimicrobial activities of extracts of 11 medicinal plant species were assessed. Table 3 displays that water extract of *A. discoridis* revealed the highest significant antimicrobial activity with inhibition zone more than 32 mm. Although *A. discoridis* and *A. hygrophilum* were belonged to the same genus but their leaf extracts exhibited different antimicrobial activity. Ethanolic and methanolic extracts of *A. discoridis* showed antibacterial and antifungal activities. Whereas, ethanolic and methanolic extracts of *A. hygrophilum* produced only antifungal activity (Table 3). Likewise, methanol extract of *E. elaterium* and ethanol extract of *M. autumnalis* showed no antibacterial activity but exhibited antifungal activity to *C. albicans* with mean inhibition zone equal to 16.3±0.6 and 18.7±1.5 mm, respectively. On the other hand, all extracts of *E. elaterium* and *E. spiculatum* showed no antibacterial and antifungal activity, respectively.

Ethanol extracts of *A. membranacea* as well as ethanol and acetone extracts of *R. chalepensis* had an antimicrobial activity against all test microorganisms except *S. pneumonia* (Table 3). The bacterium *S. pneumonia* was the least sensitive microorganism to plant extracts investigated in the current study.

Interestingly, it was noticed that *U. pilulifera* ethanolic extract had only antibacterial effect against all Gram-negative bacteria used in this study (Table 3). Moreover, it was found that acetone extract of *A. membranacea*, methanol extract of *E. spiculatum*, ethanol extract of *G. tournefortii* and ethanol extract of *P. argentea* exhibited only antibacterial activity against Gram-negative *S. typhimurium* and *P. aeroginosa* but not against *E. coli*. Those extracts, except *P. argentea* extract, were also appeared to show no antifungal activity against *C. albicans*.

In general, it was observed that both *E. coli* and *S. aureus* were sensitive to the same plant extracts (Table 3); they are sensitive to ethanol and methanol extracts of *A. membranacea*, acetone and water extracts of *A. discoridis*, methanol extracts of *G. tournefortii* and *P. argentea*, ethanol and methanol as well as water extracts of *L. varius* and to ethanol, methanol and acetone extracts of *R. chalepensis*.

Table 3 illustrated that ethanol and/or methanol extracts of *A. membranacea, A. hygrophilum, A. discoridis, E. elaterium, G. tournefortii, L. varius, M. autumnalis, P. argentea* and *R. chalepensis* exhibited antifungal activity against *C. albicans* with mean values ranging from 12.0±1.7 to 24.3±0.6 mm. However, water extracts of *A. discoridis* exhibited the highest anticandidal activity with inhibition zone value equal to 32.2±1.5 mm.

Acetone and/or water extracts of *A. membranacea, A. hygrophilum, E. elaterium, E. spiculatum, G. tournefortii, M. autumnalis, P. argentea, R. chalepensis* and *U. pilulifera* showed no antimicrobial activity against microorganisms examined in the current study (Table 3). In addition, ethanol extracts of *E. elaterium* and *E. spiculatum* as well as methanol extract of *U. pilulifera* had

Table 3: Antimicrobial activity of four leaf extracts of some medicinal plants against six microorganisms

		Diameter of inhibition zone (mm)[a]					
Plant species	Solvent	*E. coli*	*S. typhimurium*	*P. aeroginosa*	*S. pneumonia*	*S. aureus*	*C. albicans*
Achillea membranacea	Ethanol	17.3±1.5^{de}	20.0±2.0^{def}	25.7±1.5^{i}	0^{a}	19.0±1.7^{ef}	19.3±0.6^{ef}
	Methanol	16.3±0.6^{cde}	0^{a}	0^{a}	0^{a}	12.3±0.6^{b}	17.3±1.5^{cde}
	Acetone	0^{a}	12.0±1.0^{b}	11.7±0.6^{bc}	0^{a}	0^{a}	0^{a}
	Water	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
Arum hygrophilum	Ethanol	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	23.3±1.5^{gh}
	Methanol	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	16.3±0.6^{cd}
	Acetone	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Water	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
Arum discoridis	Ethanol	0^{a}	16.7±2.5^{c}	17.0±2.0^{de}	15.3±3.1^{b}	0^{a}	16.0±2.0^{c}
	Methanol	0^{a}	0^{a}	0^{a}	0^{a}	22.3±2.1^{gh}	17.3±1.5^{cde}
	Acetone	15.7±1.2^{cd}	17.7±1.5^{cd}	22.7±1.5^{h}	23.7±2.5^{c}	18.3±0.6^{ef}	0^{a}
	Water	34.7±1.5^{g}	35.0±1.0^{h}	32.7±1.5^{j}	36.7±0.6^{d}	33.0±0.0^{i}	32.3±1.5^{i}
Ecbalium elaterium	Ethanol	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Methanol	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	16.3±0.6^{cd}
	Acetone	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Water	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
Eminium spiculatum	Ethanol	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Methanol	0^{a}	19.3±0.6^{cde}	22.0±2.0gh	0^{a}	0^{a}	0^{a}
	Acetone	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Water	23.3±1.5^{f}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
Gundelia tournefortii	Ethanol	0^{a}	16.7±2.1^{c}	12.7±1.2^{c}	0^{a}	0^{a}	0^{a}
	Methanol	14.3±1.5^{c}	0^{a}	0^{a}	0^{a}	16.7±0.6d^{e}	12.0±1.7^{b}
	Acetone	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Water	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
Lupinus varius	Ethanol	18.7±1.5^{e}	21.3±1.5^{ef}	20.0±1.7^{fg}	0^{a}	15.0±1.0^{cd}	0^{a}
	Methanol	16.3±1.5^{cde}	0^{a}	15.3±2.^{5d}	0^{a}	22.0±2.0^{g}	21.3±2.1^{fg}
	Acetone	0^{a}	12.0±2.0^{b}	0^{a}	0^{a}	12.7±1.2^{bc}	0^{a}
	Water	17.0±1.0^{de}	0^{a}	0^{a}	0^{a}	20.0±2.0^{fg}	0^{a}
Mandragora autumnalis	Ethanol	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	18.7±1.5^{de}
	Methanol	0^{a}	21.0±1.0^{ef}	0^{a}	0^{a}	0^{a}	22.7±1.5^{gh}
	Acetone	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Water	0^{a}	0^{a}	0^{a}	0^{a}	24.7±2.5^{h}	0^{a}
Paronychia argentea	Ethanol	0^{a}	10.3±0.6^{b}	10.0±1.0^{b}	0^{a}	0^{a}	16.3±1.5^{cd}
	Methanol	16.7±1.2^{cde}	21.3±3.1^{ef}	20.7±1.5^{fgh}	0^{a}	13.7±1.5^{bc}	0^{a}
	Acetone	10.3±0.6^{b}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Water	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
Ruta chalepensis	Ethanol	17.0±2.0^{de}	18.7±1.5^{cde}	19.0±1.0^{ef}	0^{a}	17.0±2.7^{de}	17.0±0.0^{cde}
	Methanol	23.0±2.0^{f}	30.7±1.5^{g}	0^{a}	0^{a}	20.7±1.5^{fg}	24.3±0.6^{h}
	Acetone	16.7±0.6^{cde}	18.0±2.0^{cd}	18.7±1.5^{ef}	0^{a}	18.0±2.0^{ef}	18.3±2.1^{cde}
	Water	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
Urtica pilulifera	Ethanol	11.0±2.0^{b}	22.7±1.5^{f}	12.7±1.5^{c}	0^{a}	0^{a}	0^{a}
	Methanol	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Acetone	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}
	Water	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}	0^{a}

[a]Inhibition zone diameters are expressed as Mean±SD. The Mean±SD within column followed by the same letter are not significantly different (Tukey's studentized range test: $\alpha = 0.05$)

Table 4: Minimum inhibitory concentration (MIC) of the promising solvent extracts of selected medicinal plants

	MIC (mg mL^{-1})						
			L. varius			*R. chalepensis*	
	A. membranacea	*A. discoridis*			*P. argentea*		
Micoorganism	Ethanol	Water	Ethanol	Methanol	Methanol	Ethanol	Acetone
E. coli	32 (64)[a]	4 (8)	16 (16)	16 (64)	32 (32)	16 (16)	16 (16)
S. typhimurium	16 (64)	4 (8)	8 (64)	-	8 (16)	8 (8)	8 (8)
P. aeroginosa	4 (32)	4 (8)	8 (64)	16 (16)	8 (32)	8 (16)	8 (32)
S. pneumonia	-	4 (8)	-	-	-	-	-
S. aureus	16 (32)	4 (8)	16 (32)	4 (8)	64 (64)	16 (64)	16 (32)
C. albicans	64 (128)	4 (16)	-	32 (128)	-	64 (64)	64 (64)

[a]Numbers between parentheses are the minimal microbial concentration (MMC) in mg mL^{-1}

also no antimicrobial activity. Nevertheless, water extracts of *A. discoridis* developed the highest antimicrobial activity against all test microorganisms. Furthermore, acetone extracts of *A. discoridis* and *R. chalepensis* showed noteworthy inhibitory effects to most test microorganisms with mean inhibition zone values ranging from 15.7±1.2 to 23.7±2.5 mm and from 16.7±0.6 to 18.7±1.5 mm, respectively. It was found that only acetone extract of *R. chalepensis* exhibited antifungal activity (18.3±2.1 mm).

Significant antimicrobial effects, expressed as MIC and MMC, of promising crude extracts against test microorganisms are shown in Table 4. Extracts of selected plants were among the most active with the MIC values ranging from 4 to 64 mg mL^{-1}. Among the plant tested, water extracts of *A. discoridis* showed the strongest activity against all test microorganisms with the best MIC (4 mg mL^{-1}). The MMC values of the promising plant extracts ranged from 8 to 128 mg mL^{-1}; the lowest MMC for test bacteria and *C. albicans* was obtained from *A. discoridis* extracts and was 8 and 16 mg mL^{-1}, respectively. Whereas, the highest MMC was 128 mg mL^{-1} for *C. albicans* and obtained from ethanol and methanol extracts of *A. membranacea* and *L. varius*, respectively.

DISCUSSION

Antimicrobial activity of 11 medicinal plants has been evaluated *in vitro* against 5 bacterial and one fungus (*C. albicans*) species. They are frequently incriminated in humans infections. Most of the tested plant extracts showed some level of antimicrobial activity. The obtained results show that, ethanol extract of nine medicinal plant species, namely: *A. membranacea*, *A. hygrophilum*, *A. discoridis*, *G. tournefortii*, *L. varius*, *M. autumnalis*, *P. argentea*, *R. chalepensis* and *U. piluliferah*as revealed a wide antibacterial spectrum against most test bacterial strains. These findings are consistent with those obtained in some previous studies (Ahmad *et al*., 1998; Eloff, 1998; Lin *et al*., 1999). Also, the obtained results indicate that methanol stands as the second effective solvent after ethanol. Similar findings were reported previously (Heisey and Gorham, 1992; Leven *et al*., 1979; Naovi *et al*., 1991). Therefore, ethanol followed by methanol are better solvents for more consistent extraction of antimicrobial substances from medicinal plants compared to water and acetone so1vents. Nevertheless, methanol extraction method was found the most successful to extract antifungal metabolites from the used plants.

Extraction of antimicrobial metabolites from the selected plant species by acetone and water was observed to be the least effective methods (Table 3). However, acetone extraction method was found to be useful for extraction of antimicrobial metabolites from *A. discoridis* and *R. chalepensis*. On

the other hand, water extracts of *A. discoridis* showed the highest significant antibacterial and antifungal activities. However, all solvent extracts of *A. hygrophilum* showed no antimicrobial activity except the antifungal effect of ethanol and methanol extracts. The results of the current investigation clearly indicate that the antibacterial and anticandidal activity vary with the species of the plants. Therefore, the antimicrobial activity of plant extracts depends on the species of plant, the type of solvent and the type of tested microorganism.

In general, all the tested microorganisms were inhibited by several plant extracts of different solvents used in this study. For example, while water extracts of *A. discoridis* inhibited all six microorganisms, acetone extracts inhibited five (all bacteria), ethanol extracts inhibited four, methanol extracts inhibited two. Thus, the efficacy of plant extracts evaluated as antimicrobial agents was dependent on the solvent of extraction. Alzoreky and Nakahara (2003) reported that, both methanol and acetone were proved to be good solvents in extracting inhibitory substances from medicinal plants. In contrast, Eloff (1998) and Cowan (1999) found that methanol was more efficient than acetone in extracting phytochemicals from plant materials. In accordance with these dissimilar findings, the results of the current study revealed that the solvent type is not the only factor that should be taken in consideration during extraction of plant constituents but also the plant species as well as the test microorganism played an important role in the antimicrobial efficacy.

Among the plants screened, ethanol extracts of *A. membranacea*, water extracts of *A. discoridis*, ethanol and methanol extracts of *L. varius*, methanol extracts of *P. argentea* and methanol and acetone extracts of *R. chalepensis* showed promising activity against tested microorganisms. Since that, no significant difference in the inhibitory effects of the ethanol and acetone extracts of *R. chalepensis* was recorded, they can be considered as suitable extraction solvents for the antimicrobial agent of this plant.

The tested plant extracts showed limited or even no effect against the Gram-positive bacterium *S. pneumonia*. However, several previous findings (Branter *et al*., 1996; Nostro *et al*., 2000; Ojala *et al*., 2000) reported that Gram-negative bacteria were not susceptible to plant extracts when compared to Gram-positive bacteria. The resistance of Gram-negative bacteria towards antibacterial substances is related to lipopolysaccharides in their outer membrane (Gao *et al*., 1999; Sawer *et al*., 1997). This is controversial with the obtained results since most of the extracts showed prominent activity against gram-negative bacteria. Therefore, water extracts of *A. discoridis* which exhibited the highest inhibitory effects against gram-negative bacteria can be developed for the use in drugs industry.

The test microorganisms exhibited slight or no susceptibility to extracts of *A. hygrophilum*, *E. elaterium, E. spiculatum*, *G. tournefortii*, *M. autumnalis* and *U. pilulifera*. Therefore, the MIC and MMC were determined on extracts of *A. membranacea*, *A. discoridis*, *L. varius*, *P. argentea* and *R. chalepensis*, expectedly, having promising antimicrobial activity. Interestingly, the antifungal MIC and MMC values of extracts of such plants were higher than those of antibacterial values, suggesting that the fungus *C. albicans* is less sensitive to plant extracts than bacteria. Such a difference in susceptibility between the eukaryotic cells of *C. albicans* and the prokaryotic cells of bacteria might be attributed to their difference in cell type. This finding is in agreement with Oskay and Sari (2007) who reported that *C. albicans* MIC and MMC values were higher than that obtained with bacteria. Lower MIC and MMC values and higher zones of inhibition for *A. discoridis* water extracts implies higher solubility of phytoconstituents in the water compared to the other solvents used. Different solvents have various degrees of solubility for different phytoconstituents (Cowan, 1999).

The obtained results in this study present the first report on the antibacterial and antifungal activities of *A. discoridis* aqueous extract which revealed a broad spectrum activity against the test pathogenic microorganisms. Further phytochemical studies are invited to purify and to characterize the active ingredient(s) of this plant species.

In conclusion, the results obtained confirm the folkloric anticipation of the antimicrobial effectiveness and the therapeutic applications of the examined plants.

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REFERENCES

Adebolu, T.T. and S.A. Oladimeji, 2005. Antimicrobial activity of leaf extracts of *Ocimum gratissimum* on selected diarrhoea causing bacteria in Southwestern Nigeria. Afr. J. Biotechnol., 4: 682-684.

Afifi, F.U. and B.A. Irmaileh, 2000. Herbal medicine in Jordan with special emphasis on less commonly used medicinal herbs. J. Ethnopharmacol., 72: 101-110.

Ahmad, I., Z. Mehmood and F. Mohammad, 1998. Screening of some Indian medicinal plants for their antimicrobial properties. J. Ethnopharmacol., 62: 183-193.

Al-Eisawi, D., 1982. List of Jordan vascular plants. Mitt. Bot. Munchen, 18: 79-182.

Al-Momani, W., E. Abu-Basha, S. Janakat, R.A.J. Nicholas and R.D. Ayling, 2007. *In vitro* antimycoplasmal activity of six Jordanian medicinal plants against three Mycoplasma species Trop. Anim. Health Prod., 39: 515-519.

Al-Mughrabi, K., 2003. Antimicrobial activity of extracts from leaves, stems and flowers of *E. macroclada* against plant pathogenic fungi. Phytopathol. Mediterranea, 42: 245-250.

Alzoreky, N.S. and K. Nakahara, 2003. Antibactrial activity of extracts from some edible plants commonly consumed in Asia. Int. J. Food Microbiol., 80: 223-230.

Bisignano, G., R. Sanogo, A. Marino, R. Aquino and V. D'Angelo *et al.*, 2000. Antimicrobial activity of *Mitracarpus scaber* extract and isolated constituents. Lett. Applied Microbiol., 30: 105-108.

Bouzada, M.L.M., R.L. Fabri, M. Nogueira, T.U.P. Konno, G.G. Duarte and E. Scio, 2009. Antibacterial, cytotoxic and phytochemical screening of some traditional medicinal plants in Brazil. Pharm. Biol., 47: 44-52.

Branter, A., Z. Male, S. Pepelinfak and A. Atolic, 1996. Antimicrobial activity of *Paliurus spina.* Chrisfi mill. (*Christ-thorn*). J. Ethnopharmacy., 52: 119-122.

Chakraborty, A. and A.H. Brantner, 1999. Antibacterial steroid alkaloids from the stem bark of *Holarrhena pubescens*. J. Ethnopharmacol., 68: 339-344.

Cohen, M.L., 1992. Epidemiology of drug resistance: Implications for a Post-antimicrobial Era. Science, 257: 1050-1055.

Cowan, M.M., 1999. Plant products as antimicrobial agents. Clin. Microbiol. Rev., 12: 564-582.

Dulger, G. and C. Aki, 2009. Antimicrobial activity of the leaves of endemic *Stachys pseudopinardii* in Turkey. Trop. J. Pharmaceut. Res., 88: 371-375.

El-Astal, Z.Y., A.E.R.A. Ashour and A.A.M. Kerrit, 2005. Antimicrobial activity of some medicinal plant extracts in Palestine. Pak. J. Med. Sci., 21: 187-193.

Eloff, J.N., 1998. Which extract should be used for the screening and isolation of antimicrobial components from plants. J. Ethnopharmacol., 60: 1-8.

Elvin-Lewis, M., 2001. Should we concern about herbal medicine. J. Ethnopharmacol., 75: 141-164.

Gao, Y., M.J. van Belkum and M.E. Stiles, 1999. The outer membrane of Gram-negative bacteria inhibits antibacterial activity of Brochocin C. Applied Environ. Microbiol., 65: 4329-4333.

Heisey, R.M. and B.K. Gorham, 1992. Antimicrobial effects of plant extracts on *Streptococcus mutans, Candida albicans, Trichophyton rubrum* and other microorganisms. Lett. Applied Microbiol., 14: 136-139.

Leven, M., D.A. van den berghe, F. Merten, A. Vlictinck and E. Lammens, 1979. Screening of higher plants for biological activities. I. Antimicrobial activity. Plant Medica, 36: 311-321.

Lin, J., A.R. Opiku, M. Geneeb, A. Hubings, S.E. Terblanche, A.K. Jager and J.V. Staden, 1999. Preliminary screening some traditional Zulu medicinal plants for anti-inflammatory and antimicrobial activities. J. Ethnopharmacol., 68: 267-274.

Nakamura, C.V., T. Ueda-Nakamura, E. Bando, A.F.N. Melo, D.A.G. Cortez and F.B.P. Dias, 1999. Antibacterial activity of *Ocimum gratissimum* L. essential oil. Mem. Inst. Oswaldo. Cruz., 94: 675-678.

Naovi, S.A.H., M.S.Y. Khan and S.B. Vohora, 1991. Antibacterial, antifungal and anthelmintic investigations on Indian medicinal plants. Fitoterapia, 62: 221-228.

Nascimento, G.G.F., J. Locatelli, P.C. Freitas and G.L. Silva, 2000. Antibacterial activity of plant extracts and phytochemicals on antibiotic resistant bacteria. Braz. J. Microbiol., 31: 247-256.

Nostro, A., M.P. Germano, V. D'Angelo, A. Marino and M.A. Cannatelli, 2000. Extraction methods and bioautography for evaluation of medicinal plant antimicrobial activity. Lett. Applied Microbial., 30: 379-384.

Obeidat, M., 2011. Antimicrobial activity of some medicinal plants against multidrug resistant skin pathogens. J. Med. Plant Res., 5: 3856-3860.

Ojala, T., S. Remes, P. Haansuu, H. Vuorela, R. Hiltunen, K. Haahtela and P. Vuorela, 2000. Antimicrobial activity of some coumarin containing herbal plants growing in Finland. J. Ethnopharmacol., 73: 299-305.

Oran, S., 1994. Genetic resources of medicinal plants in Jordan. Proceedings of the National Seminar on Plant Genetic Resources of Jordan, August 2-4, 1994, Jordan.

Oran, S.A. and D.M. Al-Eisawi, 1998. Check list of medicinal plants in Jordan. Dirasat. Med. Biol. Sci., 25: 84-112.

Oskay, M. and D. Sari, 2007. Antimicrobial screening of some turkish medicinal plants. Pharmaceut. Biol., 45: 176-181.

Owolabi, O.J., E.K.I. Omogbai and O. Obasuyi, 2007. Antifungal and antibacterial activities of the ethanolic and aqueous extract of *Kigella africana* (Bignoniaceae) stem bark. Afr. J. Biotechnol., 6: 1677-1680.

Perez, C., M. Paul and P. Bazerque, 1990. An antibiotic assay by the agar well diffusion method. Acta Biol. Med. Exp., 15: 113-115.

Rios, J.L. and M.C. Recios, 2005. Medicinal plants and antimicrobial activity. J. Ethnopharmacol., 100: 80-84.

Sawer, I.K., M.I. Berry and J.L. Ford, 1997. Effect of medium composition, agitation and presence of EDTA on the antimicrobial activity of cryptolepine. Lett. Applied Microbiol., 25: 207-211.

Setzer, W.N., M.C. Setzer, R.B. Bates and B.R. Jakes, 2000. Biologically active triterpenoid of *Syncarpia glomulifera* bark extract from Paluma, North Queensland, Australia. Planta Med., 66: 176-177.

Sohail, M.N., A. Karim, M. Sarwar and A.M. Alhasin, 2011. Onion (*Allium cepa* L.): An alternate medicine for Pakistani population. Int. J. Pharmacol., 7: 736-744.