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Discovering antimicrobial powers of some herbs used by Bedouin in the Jordanian Petra

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ABSTRACT

Bedouin of Petra-Jordan have been using herbs in their foods and folk medicine since ages. Although phytochemical properties of many of these plants were investigated in some neighboring countries; many of these herbs are still veiled. In this study; antimicrobial activity of three herbs (*Origanum jordanicum* Danin and Kuenne, *Plantago afra* L. and *Equisetum ramosissimum Desf.*); which are endemic to the historical city of Petra were investigated. Three types of extracts (methanol; ethanol or water) were prepared from each plant to investigate their effect against seven bacterial strains (*Bacillus subtilis* ATCC 6633, *Staphylococcus aureus* ATCC 25923, *Staphylococcus epidermidis* ATCC 12228, *Klebsiella pneumoniae* ATCC 31488, *Escherichia coli, Salmonella* sp., and *Erwinia carotovora*) in addition to one yeast strain; *Candida albicans* ATCC 10231 using both disk diffusion and micro dilution (MIC) assays. Promising antimicrobial reactions were obtained from the experimented plant extracts from each plant type. In disk diffusion assay, the most inhibited bacteria species was *Staphylococcus aureus* ATCC 25923 exposed to ethanol extract of *Origanum jordanicum* Danin and Kuenne, and, *Plantago afra* L., with inhibition zones of (30±0.71 and 24.9±0.81 mm; respectively). On the other hand; the ethanol extract of *Equisetum ramosissimum* Desf. had strongly prohibited growth of *Bacillus subtilis* ATCC 6633 with inhibition zone of (25.0±0.84 mm). For MIC method; ethanol extract *of Plantago afra* L. and methanol extract of *Equisetum ramosissimum* Desf., were most effective against *Staphylococcus aureus* ATCC 25923 at MIC value of 0.391 mg/mL. *Origanum jordanicum* Danin and Kuenne methanol extract gave the best result against *Staphylococcus epidermidis* ATCC 12228 at 0.781 mg/mL MIC., while it was effective in both assays against *Candida albicans* ATCC 10231. Our data represent the first report for the antimicrobial activities of three plants had been used for ages by Bedouin of Petra for food and medicine. Furthermore; *Origanum jordanicum* Danin and Kuenne an endemic species to Petra was screened against different microbes for the first time. However, more research must be done to ensure the safety of those herbs and the possibility of being introduced to phytondustry.

Key words: *Antimicrobial activities, Bedouin, Extract type, Folk medicine, Inhibition zone assay, Medicinal plants, Petra, MIC assay.*

Introduction

Petra is one of the most ancient cities in Jordan and has been recently listed as one of the seven wonders of the world (Urban *et al.*, 2013). Petra is rich in medicinal plants that are spreading in its different parts (Oran, 2014). Bedouin of Petra depends on these plants for their folk medicine to treat or mitigate the effect of different pains (Al-Qura'n, 2007). In spite of the touristic importance of Petra, very little research is available on wild herbs of Petra used by the Bedouins living there; especially those used for either food or medication. Bedouin live in the desert where hospitals or medical centers are

few (Abdelhalim *et al.,* 2017), and consequently, Bedouin used herbs in their folk medicine as a part of their traditions for treatments of different diseases and some of them are known as traditional healers (Al-Qura'n, 2007; Abdelhalim *et al*., 2017; Friedman *et al*., 1986).

One the most important endemic medicinal plant from Petra region in Jordan is; *Origanum jordanicum* Danin and Kuenne; which called locally as (Zater Petra) (Taifor and El-Oqlah., 2014; Danin and Künne, 1996). *Origanum jordanicum* Danin and Kuenne grows in some restricted areas of Jordan and is subjected to some growing threats of habitats degradation (Taifor and El-Oqlah, 2014). This herb is not registered in any other countries before. The genus *Origanum* has different medicinal importance including antimicrobial, antioxidant; antihyperglycaemic and other activities (Aligiannis *et al*., 2001; Boskovic *et al*., 2015; Chishti *et al*., 2013). In this study *Origanum jordanicum* Danin and Kuenne was evaluated for its antimicrobial activities as its used widely by Bedouin of Petra for either therapeutic or food purposes (Al-Qura'n, 2007). Furthermore, no literature had evaluated its antimicrobial activities against different microbes before.

Plantago afra L., is another wild medicinal plant; commonly name as (Arrableh). It belongs to the Plantaginaceae family (Taiforand El-Oqlah., 2016). *Plantago* spp are usually used in food and dietary research as their seed husk powder can be used as a dietary supplement and as a natural, safe powder that can be used in obesity regime for weight loss (Turnbull, 1995; Van Craeyveld *et al*., 2009). In Jordan *Plantago afra* L., is considered as a less explored medicinal plant although it has large populations in Petra (Taifor and El-Oqlah, 2016; Oran, 2005). *Plantago afra* is widely used by Arabian healers of Petra to treat many diseases mainly the digestive tract ailments such as diarrhea beside urethritis and hemorrhoids problems (Newall *et al.,* 1996). In most previous research only the seeds or seed husk of *Plantago spp* were studied for their chemical activities (Bemiller and Whister, 1996; Ahmadi *et al*., 2012).

Equisetum ramosissimum Desf., is a wild medicinal plant that had been listed in the endangered plant species list in Jordan (Taifor and El-Oqlah, 2016). It belongs to the Equisetaceae family and can be found in some valleys of southern part of Jordan; it has been recorded in Wadi Rum; Maan; Dana and Petra (Taifor and El-Oqlah, 2014). It is common name in Jordan "branched horsetail" or "Thail Al Hussan" (Taifor and El-Oqlah, 2014). It is widely used in medicine to treat some diseases such as hemorrhage, hepatitis; urethritis, and jaundice (Oh *et al*., 2004; Li *et al*., 2016). Only; few previous studies had demonstrated the inhibitory effects of *Equisetum ramosissimum* Desf. extracts against some microbes (Abu-Mejdad, 2014; Kelmanson *et al*., 2000; Van Staden *et al*., 2017; Eslamiyan *et al*., 2015). There is a need to investigate more anti-infectious properties of *Equisetum ramosissimum* Desf. Against different pathogen types due to its wide usage by Bedouin of Petra.

In general; few research were conducted on these three plants in terms of their anti-microbial effect. So, as those plants are used widely in the folk medicine by Bedouin in Petra-Jordan; this study was conducted to evaluate the effect of different extracts types taken from the whole parts of *Origanum jordanicum* Danin and Kuenne, *Plantago afra* L. and *Equisetum ramosissimum* Desf. plants from Petra against some pathogenic microbes.

Materials and Methods

Plant material and preparation of extract: Aerial parts of wild medicinal plants of *Origanum jordanicum* Danin and Kuenne, *Plantago afra* L. and *Equisetum ramosissimum* Desf.; were collected from Petra; at the southern parts of Jordan (Latitude: 30°19'43''N, Longitude: 35°26'31''E) in spring 2019. Plant materials were identified by the plant Taxonomist Hatem Taifor from the Royal Botanic Garden (RBG); Tall Arrumman- Jordan. Samples were dried at room temperature until all excess water was removed and the dried samples were grounded and weighed. The extraction was carried out on the base of dry weight with different solutions (methanol; ethanol or water) with the ratio of 1:10 (g of sample: 10 mL of solution). The extracts with solution, used were left with continuous stirring for three days to let good mixing and extracting of the most soluble active components. After that, mixtures were filtered using filter paper and evaporated with a rotary evaporator until dryness. Then; the dried extracts were dissolved in determined DMSO (mL) which were calculated according to the following equation; DMSO (mL) = weight of evaporated extract × 10); to achieve a concentration of 100 mg/mL (Tahtamouni, 2018).

Tested microorganisms: The antimicrobial potential of *Origanum jordanicum* Danin and Kuenne,

Plantago afra L. and *Equisetum ramosissimum* Desf. extracts were screened against seven bacterial strains (*Bacillus subtilis* ATCC 6633, *Staphylococcus aureus* ATCC 25923, *Staphylococcus epidermidis* ATCC 12228, *Klebsiella pneumoniae* ATCC 31488, *Escherichia coli*, *Salmonella sp.*, and *Erwinia carotovor*a) and one yeast strain; *Candida albicans* ATCC 10231. The tested microorganisms were obtained from Hamdi Mango center for Scientific Research at the University of Jordan- Amman –Jordan.

Antibacterial screening: Antibacterial activity of *Origanum jordanicum* Danin and Kuenne, *Plantago afra* L. and *Equisetum ramosissimum* Desf. extracts were determined using both disk diffusion assay and microdillution assay. Disc diffusion assay was done according to (Karlsmose, 2010). Muller Hinton agar was prepared then the bacterial suspension was adjusted to 0.5 McFarland with 10 mL of NaCl. After that about 10 mL were taken from the bacterial suspension and inoculated in the discs which were about 6 mm in diameter. After that about of 10 mg of each plant extract were diffused in the discs and then incubated at 37 °C for 24 h. The antibiotic; Tetracycline (Sigma Co., St. Louis, MO) was used as reference or positive control at the concentration of 10 mg/mL. The inhibition zone was measured after 24 h of incubation. Five replicates of five plates were used for each treatment and data were means of each treatment with standard errors. The other method; Microdilution assay; minimum inhibitory concentration (MIC) was determined by microdilution method using serially diluted extracts according to NCCLS (2000). Ten dilutions were prepared from each concentrated extract (100 mg/mL) in Muller Hinton, potato dextrose broth; by equal division for each concentration to get (50; 25; 12.5; 6.25; 3.125; 1.562; 0.781; 0.391; 0.195; and 0.097 mg/mL). Plant extract dilutions were diffused in wells with bacteria inoculate of about 10^5 CFU/mL. The antibiotic Tetracycline at the concentration (10 mg/mL) was used as a control. After that samples were incubated in plate shaker incubator at 350 rpm and 37 °C for 24 h and results were collected visually.

Anti-candida screening: for well diffusion assay; Candida cultures were inoculated to petri plates containing potato dextrose agar, 6.0 mm diameter wells were made in the plates which were loaded with 10 mg from each plant extract. After 24-48 hr incubation at 30 °C, the antifungal activity was assayed by measuring the zone of inhibition. A standard antifungal fluconazole (Pfizer INC., USA); with the concentration of 10 mg/mL was used as positive control against *Candida albicans* ATCC 10231. Data were collected for Disc diffusion method for the inhibition zone diameter in mm after 24 h of incubation. Five replicates were used for each treatment and data were means of each treatment with standard errors. Microdilution assay was assayed for *Candida albicans* as described by Fromtling *et al*., 1993. The minimum concentrations were determined as the lowest MICs value that inhibited visible Candida growth. For minimum inhibitory concentration (MIC); the results were recorded for the well that has the lowest dilution number of each extract type from (1-10) diluents as described above in MIC method for bacteria. The positive control was fluconazole (10 mg/mL) with nutrient broth.

Results and Discussion

Disk diffusion assay: A promising data of microbes growth suppression were obtained from the employed plant extracts (Table 1). The highest two inhibited bacteria species were *Staphylococcus aureus* ATCC 25923 and *Staphylococcus epidermidis* ATCC 12228. For *Origanum jordanicum* Danin and Kuenne plant extract; *Staphylococcus aureus* ATCC 25923; was inhibited with the ethanol and methanol extracts with inhibition zones of (30±0.71 mm, 25.4±0.84; respectively). Amazingly; this was more effective than antibiotic (Tetracycline) results (Table 1). This results were in accordance with (Marques *et al*., 2015) were they found that *Origanum vulgare* L. and *Origanum majorana* L. oils were very effective against *Staphylococcus aureus* isolated from poultry meat. Beside that; *Staphylococcus epidermidis* ATCC 12228; also has inhibition zones of (22.40±0.51; 18.0±0.63) with ethanol or methanol extracts; respectively. The results of *Klebsiella pneumoniae* ATCC 31488 were also very closed to the positive control when *Origanum jordanicum* Danin and Kuenne ethanol extract was used. Furthermore *Escherichia coli* inhibition results with *Origanum jordanicum* Danin and Kuenneethanol extract was a little more than the control (Table 1). In general *Origanum jordanicum* Danin and Kuenne ethanol extract was more effective than other two types of extracts methanol or water extracts in prohibition of bacterial growth (Table 1). *Origanum* spp have shown antimicrobial activities against different microbes.

Chishti *et al.,* (2013) reported that ethanol of *O. majorana* L. have shown highest antimicrobial activity against different bacteria species than water extract. On the other hand Brđanin *et al*., (2015) found that *O. vulgare* extracts showed a moderate activity against gram negative or positive bacteria used.

For *Plantago afra* L. the ethanol extract was the most effective for bacterial growth prevention. *Staphylococcus aureus* ATCC 25923 was highly inhibited with 24.9±0.81 mm of inhibition zone; and its growth was more inhibited than when treated with the used antibiotic (Tetracycline) (Table 1). Furthermore; for the same extract type of the *Plantago afra* L.*; Staphylococcus epidermidis* ATCC 12228 resulted in 20.0±0.32 mm of inhibition zone which was higher than the control (Tetracycline: 14.0±0.84 mm inhibition zone). In contrary; methanol extract gave the lowest results compared to ethanol extract of *Plantago afra L.*. Beside that water extract was the least extract used for the inhibition of the bacteria in *Plantago afra L.* These results were in accordance with previous studies that had been evaluated *Plantago* spp. seeds husk against microbes (Motamedi *et al*., 2010; Erum *et al.,* 2015). Motamedi *et al*., (2010) found that ethanol and methanol extracts from seeds husk of *Plantago ovata* were highly inhibiting *Staphylococcus epidermidis* and *Staphylococcus aureus*. On the other hand; Erum *et al*., (2015) found that *S. epidermidis* and *S. aureus* were inhibited by carboxymethylarabinoxylan extract isolated from *P. ovata* husk.

In *Equisetum ramosissimum* Desf. experiment, ethanol extract gave the highest inhibition zone with *Staphylococcus aureus* ATCC 25923 and *Staphylococcus epidermidis* ATCC 12228 (Table 1). Their inhibition zones were (25.0±0.84 & 22.0±0.32 mm; respectively) and were higher than control (Table 1). Water extract was the least extract that inhibited

Table 1. Inhibition zones diameters (mm) against different bacteria strains obtained from disc diffusion assay for *Origanum jordanicum* Danin and Kuenne*, Plantago afra* L. and *Equisetum ramosissimum* Desf. plant extracts

Bacterial strain	Methanol extract	Ethanol extract	Water extract	Positive control Tetracycline
		Inhibition zone (mm)*		
Plant species / *Origanum jordanicum* Danin and Kuenne				
Bacillus subtilis ATCC 6633	10.46±0.40	15.04±0.32	0.0±0.0	21.0±0.32
Staphylococcus aureus ATCC 25923	25.0±0.84	30.0±0.71	10.0±0.32	15.0±0.32
Staphylococcus epidermidis ATCC 12228	22.40±0.51	18.0±0.63	12.0±0.63	14.0±0.84
Klebsiella pneumoniae ATCC 31488	10.10±0.12	20.0±0.63	10.0±0.32	20.0±0.63
Escherichia coli,	11.06±0.21	12.0±0.63	10.0±0.63	10.06±0.17
Salmonella sp.	10.0±0.32	18.2±0.58	8.0±0.32	22.20±0.66
Erwinia carotovora	7.94±0.41	12.0±0.32	8.0±0.32	15.0±0.32
		Plantago afra L.		
Bacillus subtilis ATCC 6633	5.50±0.59	11.0±0.32	0.0±0.0	21.0±0.32
Staphylococcus aureus ATCC 25923	10.0±0.32	24.9±0.81	6.0±0.63	15.0±0.32
Staphylococcus epidermidis ATCC 12228	8.0±0.63	20.0±0.32	8.0±0.32	14.0±0.84
Klebsiella pneumoniae ATCC 31488	0.0±0.0	8.0±0.32	0.0±0.0	20.0±0.63
Escherichia coli,	4.0±0.32	10.0±0.32	0.0±0.0	10.06±0.17
Salmonella sp.	0.0±0.0	0.0±0.0	0.0±0.0	22.20±0.66
Erwinia carotovora	8.80±0.58	11.8±0.37	6.0±0.32	15.0±0.32
		Equisetum ramosissimum Desf.		
Bacillus subtilis ATCC 6633	8.0±0.32	25.0±0.84	0.0±0.0	21.0±0.32
Staphylococcus aureus ATCC 25923	12.0±0.63	22.0±0.32	0.0±0.0	15.0±0.32
Staphylococcus epidermidis ATCC 12228	10.0±0.63	10.0±0.63	0.0±0.0	14.0±0.84
Klebsiella pneumoniae ATCC 31488	0.0±0.0	15.0±0.63	0.0±0.0	20.0±0.63
Escherichia coli,	8.0±0.44	10.0±0.32	0.0±0.0	10.06±0.17
Salmonella sp.	6.0±0.45	12.0±0.32	0.0±0.0	22.20±0.66
Erwinia carotovora	8.0±0.32	25.0±0.84	0.0±0.0	15.0±0.32

*Data are means of five replicates (n = 5) ± standard error.

different bacteria species for the three plant species used; however *Equisetum ramosissimum* Desf. water extract has no effect on bacterial inhibition. No inhibition was found in *Salmonella sp.* in response to application of the different extracts. In another study ethanol extract of *Equisetum ramosissimum* from east and central Nepal gave a moderate zone of inhibition (8 mm to 11 mm) against all the tested bacteria (Subba and Basnet, 2014).

Minimum inhibitory concentration method (MIC)

Results obtained from the extracts varied in their potency against microbes with plant extract type and bacteria species as shown in (Table 2). *Origanum jordanicum* Danin and Kuenne, methanol extract was effective at MIC values of (0.781 mg/mL) against *Staphylococcus epidermidis* ATCC 12228 and it was much lower than control (Table 2). *Origanum jordanicum* Danin and Kuenne, ethanol extract was more effective for *Klebsiella pneumoniae* ATCC 31488 and *Escherichia coli* at MIC values of (6.25 mg/mL) (Table 2). Water extract of *Origanum jordanicum* Danin and Kuenne, was not effective against the tested bacteria species as there was no inhibition obtained with four bacteria species of (*Bacillus subtilis* ATCC 6633; *Klebsiella pneumoniae* ATCC 31488, *Escherichia coli, Salmonella sp*. and *Erwinia carotovor*a) (Table 2). *Origanum* spp extracts varied for their activities to the bacteria inhibition. *Origanum vulgare* were approved to have bactericidal properties against Bacillus strains (Boughendjioua and Seridi, 2017). Beside that; Brðanin *et al.,* (2015) found that the minimal inhibitory concentration (MIC) for the Gram positive bacteria were between 62.5 and 125 µg/mL when they used *Origanum vulgare* L. extracts.

The results obtained from *Plantago afra* L. ethanol extract against *Staphylococcus aureus* ATCC 25923 was very interesting. Growth inhibition was maximized at low MIC value of 0.391 mg/mL; while it

Table 2. MIC of *Origanum jordanicum* Danin and Kuenne, *Plantago afra* L. and *Equisetum ramosissimum* Desf. extracts against tested microorganisms by micro dilution assay.

Bacterial strain	Methanol extract	Ethanol extract	Water extract	Positive control Tetracycline
	MIC (mg/mL)			
Plant species/*Origanum jordanicum* Danin and Kuenne				
Bacillus subtilis ATCC 6633	12.5	12.50	0.0	1.562
Staphylococcus aureus ATCC 25923	3.125	12.50	3.125	0.781
Staphylococcus epidermidis ATCC 12228	0.781	1.562	6.25	1.562
Klebsiella pneumoniae ATCC 31488	3.125	6.25	0.0	1.562
Escherichia coli	12.50	6.25	3.125	0.391
Salmonella sp.	12.50	3.125	0.0	0.781
Erwinia carotovora	0.0	3.125	0.0	1.562
	Plantago afra L			
Bacillus subtilis ATCC 6633	6.25	3.125	12.5	1.562
Staphylococcus aureus ATCC 25923	0.781	0.391	1.562	0.781
Staphylococcus epidermidis ATCC 12228	1.562	1.562	12.5	1.562
Klebsiella pneumoniae ATCC 31488	12.5	6.25	12.5	1.562
Escherichia coli	12.5	6.25	25	0.391
Salmonella sp.	0.0	0.0	0.0	0.781
Erwinia carotovora	6.25	3.125	12.5	1.562
	Equisetum ramosissimum Desf.			
*Bacillus subtilis*ATCC 6633	0.0	0.0	0.0	1.562
Staphylococcus aureus ATCC 25923	0.391	0.781	1.56	0.781
Staphylococcus epidermidis ATCC 12228	3.125	6.25	12.5	1.562
Klebsiella pneumoniae ATCC 31488	0.0	0.0	0.0	1.562
Escherichia coli	6.25	3.125	12.5	0.391
Salmonella sp.	0.0	0.0	0.0	0.781
Erwinia carotovora	6.25	12.5	0.0	1.562

required to increase the positive control to (0.781 mg/mL) to obtain such response. The ethanol extract of *Plantago afra* L. has a wide range of inhibition for all bacteria used except for *Salmonella sp.* This is also applicable on *Plantago afra* L.methanol and water extract, but with lower inhibition values than ethanol extract. *Plantago ovata* seeds extracts were also evaluated against *Staphylococcus aureus* and the highest MIC was 100 mg/mL (Bokaeian *et al*., 2015). Also, MIC for ethanol or methanol extract of *Plantago Ovata* seeds against *S. aureus* were (20 mg mL^{-1} (Motamedi *et al*., 2010). Karami *et al*., 2017; also found that *P. ovata* seeds husk extract had a high and moderate effect against *Bacillus sphaericus* and *Pseudomonas aeruginosa*. *Equisetum ramosissimum* extracts inhibition was restricted to less bacterial species. *Staphylococcus aureus* ATCC 25923 was inhibited highly with methanol extract of *Equisetum ramosissimum* at 0.391 mg/mL. In another study of *Equisetum ramosissimum* using MBC method; the methanol and ethanol extracts at the minimum concentration of 25 mg/ml showed a lethal effect on standard strain of *E. coli* (Eslamiyan *et al*., 2015).

We can observe from the above results of the three employed plant extracts; that ethanol and methanol extracts have high inhibitory effects with priority of ethanol in inhibition. These results were in agreement with Bacon *et al*., (2017). This may be due to these solvent properties in extraction which may be had ability to catch up all the important antimicrobial constituents as well as bacterial strains used. On the contrary to this, water extract was the least effective extract used against microbes and this was confirmed previously with different studies (Bacon *et al*., 2017). Other studies suggested that hydrophobicity characteristics of the plant extract may be the reason that permit those extracts to react with membranes of the microbes cells (Friedman *et al*., 2004; Tiwari *et al*., 2009). According to disc diffusion assay, our results represent the first report for *Origanum jordanicum* Danin and Kuenne as the best plant for prohibition of the tested microbes. While for the other method MIC; *Plantago afra* L. worked as the best extract against bacteria used.

Anti-candida activity: Results obtained in both applied assays for *Origanum jordanicum* Danin and Kuenne revealed that methanol extract gave the best results. Inhibition zone diameter of (25.22±0.34 mm) was obtained using inhibition zone assay, while this extract was extremely powerful at MIC value of (1.562 mg/mL) while it needed to increase the positive control to (6.25 mg/mL) to get such effect (Table 3). Brðanin *et al.,* (2015) found that extracts of other *Origanum* species; *O. basilicum* extract were active against *Candida albicans* (MIC 125 µg/mL). *Plantago afra* L methanol extract was effective also against *Candida albicans* ATCC 1023 with 19.20±0.25 mm zone of inhibition. While MIC value were 3.125 mg/mL for *Plantago afra* L methanol extract. This results were in opposite with *Plantago* major where *Candida* had no effect on it (Karima *et al*., 2015). Beside that; *Equisetum ramosissimum* Desf. results were only obtained with methanol extract. Furthermore This result was in accordance with Eslamiyan *et al*. (2015) and Abu-Mejdad, (2014) who found that *Equisetum ramosissimum* can be used to treat fungal infections either lonely or in combination with other antimicrobial agents. Water extract was not effective

Table 3. Anti-Candida activity of *Origanum jordanicum* Danin and Kuenne, *Plantago afra* L. and *Equisetum ramosissimum* Desf. using disc diffusion assay (mm) and micro dillution assay (MIC) (mg/ml) against tested fungus *Candida albicans* ATCC 10231.

Yeast strain	Method	Plant species	Methanol extract	Ethanol extract	Water extract	Positive control Fluconazole
Candida albicans ATCC 10231	Disc diffusion assay (mm)	*Origanum jordanicum* Danin & Kuenne	25.22±0.34	22.0±0.63	0.0	20.0 ±0.46
		Plantago afra L	19.20±0.25	10.1±0.32	0.0	20.0 ±0.46
		Equisetum ramosissimum Desf.	6.28±0.13	0.0	0.0	20.0 ±0.46
	Microdillution assay (MIC); (mg/ml).	*Origanum jordanicum* Danin & Kuenne	1.562	6.25	0.0	6.25
		Plantago afra L	3.125	12.5	0.0	6.25
		Equisetum ramosissimum Desf.	6.25	0.0	0.0	6.25

*Data are means of five replicates (n = 5) ± standard error.

on *Candida albicans* ATCC 1023 for three plant species extracts. We can see from this study that *Origanum jordanicum* Danin and Kuenne have the highest anti-Candida effects using methanol extract. The other two plant species also have a moderate effect against *Candida albicans* ATCC 10231.

Conclusion

From this study, we can conclude that; these plants species used by Bedouin of Petra have promising antimicrobial activities against most tested microbial strains, especially for *Origanum jordanicum* Danin and Kuenne which is an endemic species to Petra-Jordan. In this study; the most inhibited microbes species were *Staphylococcus aureus* ATCC 25923 and *Staphylococcus epidermidis* ATCC 12228. Our results indicated that the natural extracts of the experimented plants were either stronger than the antibiotics used or close to them. Further studies should focus on the isolation and identification of compounds that may be contributing to inhibition of these foodborne or human pathogenic microbes.

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