J.Agric.Chem.and Biotechn., Mansoura Univ.Vol. 7(10): 269 - 274 , 2016

Determination of Phytochemicals and Antibacterial Activity of Some Medicinal Plants Against Pathogenic Bacteria .

Aida, H. Afify ; F. I. A. Hauka and Amany K. E. El-Namla
Microbiology Dept., Fac. Agric., Mansoura Univ., El-Mansoura, Egypt

ABSTRACT

The present work was achieved to find the active chemical constituents and to evaluate the antimicrobial activity of some medicinal plant extracts on six pathogenic bacteria include ; *Escherichia coli, Salmonella* sp., *Shigella flexneri, Enterobacter sakazakii*, *Staphylococcus aureus* and *Bacillus cereus* by agar well diffusion method. A qualitative phytochemical analysis was performed for the detection of alkaloids, saponins, terpenes, flavonoids, tannins and glycosides. The results revealed that *Rosmarinus officinalis* (rosemary), *Citrus sinensis* (orange peel), *Eucalyptus globulus* (camphor), *Psidium guajava* (guava), *Matricaria chamomilla* (chamomile), *Allium cepa* (onion), *Allium sativum* (garlic) and *Nigella sativa* (black seed) contain alkaloids, flavonoids, tannins, terpenes, saponins and glycosides. Also the plant extracts of both rosemary, orange peel, camphor, guajava, showed highest significant effect against all tested pathogenic bacteria followed by chamomile, onion and garlic showed lowest significant effect but black seed showed no effect. *Salmonella* sp. exhibited resistance to most plant extracts. This study revealed the presence of bioactive compounds in these plants which have antibacterial properties, can be useful as an alternative antimicrobial agent for treatment infectious diseases and in pharmaceutical industries.
Keywords: Phytochemical analysis– plant extracts – pathogenic bacteria – antibacterial activity.

ITRODUCTION

There has been an increase in the number of poisoning outbreaks caused by food-borne pathogenic bacteria. Diseases are the major causes of death in the developing countries. The extensive use of the antibiotics to treatment these diseases has led to the emergence of multidrug resistance (Westh *et al.*, 2004). Bacterial infectious diseases represent an important cause of morbidity and mortality worldwide. Bacterial resistance to antibiotic is becoming increasingly common (Lesse, 1995). The increased usage of specific antimicrobial correlates with the increased levels of bacterial resistance to those agents (Mordi and Erah, 2006). There has been increasing incidence of multiple resistances in pathogenic bacteria attribute to the indiscriminate use of commercial antimicrobial drugs for treatment diseases (Shareef, 2011), therefore recently research toward to develop alternative drugs from medicinal plants for cure infectious diseases. Drugs from the plants are easily available, less expensive, safe, efficient and rarely have side effects. Plants are rich in a wide variety of secondary metabolites such as alkaloids, tannins, terpenoids and flavonoids which exhibited the antimicrobial properties for treatment of a wide range of diseases(Cowan, 1999). Also plant contain some organic compounds which have physiological action on the human body and these bioactive compounds include tannins, alkaloids, carbohydrates, terpenoids, steroids and flavonoids (Edoga *et al*., 2005). These compounds are synthesized by primary or secondary metabolism of organisms. Secondary metabolities are chemically and taxonomically extremely diverse compounds with obscure function. They are widely used in the human therapy, veterinary, agriculture (Vasu *et al*., 2009). Plant products have been part of phytomedicines since time immemorial. This can be derived from barks, leaves, flowers, roots, fruits, seeds (Criagg and David, 2001). The chemical constituents of plants is desirable because such information will be value for synthesis of complex chemical substances (Mojab *et al*., 2003 ; Parekh and Chanda, 2007 , 2008). Herbs and spices have many phytochemicals which are sources of natural antioxidant such as flavonoids, phenolic, diterpenes, tannins and phenolic acid. These compounds have antioxidant, putrefaction and anticancer properties (Chitravadivu *et al*., 2009). These substances have been used as food, medicine and having medicinal value have been extensively used for treating various disease conditions. Herbs being easily available to human beings, have medicinal properties (Perumal and Krishnakone, 2007). Extraction of bioactive compounds from medicinal plants permits of demonstration of their physiological activities, lead to the synthesis of a more potent drug with reduced toxicity .

The aim of present work to determine the phytochemical analysis and antibacterial activity of some plant extracts against pathogenic bacteria.

MATERIALS AND METHODS

Pathogenic bacteria : Six bacterial species among Gram negative such as *Salmonella* sp., *Shigella flexneri* and *Enterobacter sakazakii* and Gram positive such as *Staphylococcus aureus* and *Bacillus cereus* were used in this work. These microbes were obtained from faculty of Medicine, Mansoura University and Agricultural Research Center (ARC) , Giza , Egypt .

Collection and extraction of plants: Eight plants such as *Rosmarinus officinalis* (leaves), *Citrus sinensis* (peels), *Eucalyptus globulus* (leaves), *Psidium juagava* (leaves), *Matricaria chamomilla* (flowers), *Allium cepa* (bulbs), *Allium sativum* (cloves) and *Nigella sativa* (seeds) were collected and purchased from the market of Mansoura City. Each part cut into pieces, dried under shaded condition at room temperature and then powdered. The powder was macerated with four solvents (water, methanol, ethanol and acetone) for two days. The mixture was filtered using centrifuge . The filtrates were evaporated in a rotary evaporator at $45^{o}C$ to concentrate and preserved in refrigerator at 4°C untile further use (Ababutain 2011)

Qualtitative phytochemical analysis : The extracts were tested for the presence of terpenes, alkaloids, flavonoids, tannins, saponinis and glycosides compounds by using qualtitative analysis according to Harborne (1988) .

Preparation of bacterial cultures : Loopful of stock cultures were transferred into test tubes contain nutrient broth and incubated at 37^{o}C for 24 h. The bacterial culture was enumerated by using the serial dilution method. Final concentration was 10^{6} colony forming units (cfu / ml) according to NCCLS (1999).

Antibacterial assay : All the plant extracts were studied to ascertain their antibacterial activity against pathogenic bacteria by using the well diffusion agar method (Araujo *et al*., 2003). One ml of active bacterial culture containing (10^{6}cfu) was poured to Petri dishes containing nutrient agar. Well was made in nutrient agar plate by sterile cork borer (9 mm). 0.1 of each of each concentration was placed in the well. Also 0.1 ml of each solvent without bacteria was placed in the well as a control, then the plates were incubated at 37^{o}C for 24 h. The antimicrobial activity of the different extracts was determined by measuring the diameter of zone of inhibition (ZI) exhibited by the extracts compared with control.

Statistical analysis : Using SAS (2001) and Dancan (1955) numerical data collected were statistcally analyzed for analysis of variance and least significant difference at propability (P) level of ≤ 0.05 .

RESULTS AND DISCUSSION

Phytochemical analysis : The phytochemical constituents of eight medicinal plants were summarized in Table (1). From the phytochemical screening, terpenes, flavonoids, flavonoids and glycosides were found in all tested plant extracts. Alkaloids were observed in different plant extracts except the aqueous extract of both camphor and guava also the acetone extract of rosemary did not have alkaloids, while alkaloids did not found in all different extracts of onion and black seed. Saponins were absent in all extracts of orange peel, chamomile, onion and garlic but they were present in the methanol and ethanol extracts of rosemary, also saponins were present in the methanol, ethanol and acetone extracts of camphor and guava, all plant extracts of black seed possess saponins.

Antibacterial activity : From the results of antimicrobial screening are shown in Tables (2 – 8).

The antibacterial activity of ethanol extract of *Rosamarinus officinalis* in Table (2) showed highest effect with non significant difference between methanol and acetone extracts against *E. coli* but the aqueous extract showed lowest significant effect. The methanol and acetone extracts had high effect with non significant difference between them against *Salmonella* sp., while the aqueous and ethanol extracts exhibited non effective. The acetone extract was highest inhibition zone with non significant difference between all extracts against *Shigella flexneri* and *Enterobcter sakazakii* but the aqueous extract showed lowest significant effect with *Shigella flexneri,* ethanol extract showed lowest significant effect against *Enterobacter sakazakii*. The methanol extract exhibited the highest significant effect with *Staphylococcus aureus* and *Bacillus cereus*, but the aqueous extract showed lowest significant effect. Also highest effect was at concentration 400 mg/ml, lowest significant effect was in 100 mg/ml .

Table 1. Phytochemical constituents of eight medicinal plants

Plant extracts	Compounds	Terpenes	Alkaloids	Flavonoids	Tannins	Saponnins	Glycosidess
Rosmarinus officinalis	aqueous extract	+	+	+	+	-	+
	methanol extract	+	+	+	+	+	+
	ethanol extract	+	+	+	+	+	+
	acetone extract	+	-	+	+	-	+
Citrus sinensis	aqueous extract	+	+	+	+	-	+
	methanol extract	+	+	+	+	-	+
	ethanol extract	+	+	+	+	-	+
	acetone extract	+	+	+	+	-	+
Eucalyptus globulus	aqueous extract	+	-	+	+	-	+
	methanol extract	+	+	+	+	+	+
	ethanol extract	+	+	+	+	+	+
	acetone extract	+	+	+	+	+	+
Psidium guajava	aqueous extract	+	-	+	+	-	+
	methanol extract	+	+	+	+	+	+
	ethanol extract	+	+	+	+	+	+
	acetone extract	+	+	+	+	+	+
Matricaria chamomilla	aqueous extract	+	+	+	+	-	+
	methanol extract	+	+	+	+	-	+
	ethanol extract	+	+	+	+	-	+
	acetone extract	+	+	+	+	-	+
Allium cepa	aqueous extract	+	-	+	+	-	+
	methanol extract	+	-	+	+	-	+
	ethanol extract	+	-	+	+	-	+
	acetone extract	+	-	+	+	-	+
Allium sativum	aqueous extract	+	+	+	+	-	+
	methanol extract	+	+	+	+	-	+
	ethanol extract	+	+	+	+	-	+
	acetone extract	+	+	+	+	-	+
Nigella sativa	aqueous extract	+	-	+	+	+	+
	methanol extract	+	-	+	+	+	+
	ethanol extract	+	-	+	+	+	+
	acetone extract	+	-	+	+	+	+

\+ = present - = absent

Table 2. Effect of *Rosmarinus officinalis* extracts against pathogenic bacteria (diameter of inhibition zone mm)

Plant extracts	Pathogenic bacteria *E. coli*	*Salmonella* sp.	*Shigella flexneri*	*En. sakazakii*	*St. aureus*	*B. cereus*
Aqueous extract	9.67 ^{b}	0.00 ^{b}	5.67 ^{d}	8.33 ^{c}	9.00 ^{c}	6.33 ^{c}
Methanol extract	17.67^{a}	20.3^{a}	15.67 ^{c}	17.67 ^{b}	18.00 ^{a}	20.22^{a}
Ethanol extract	18.67^{a}	0.00 ^{b}	17.33 ^{b}	7.67 ^{c}	17.33 ^{ab}	16.67 ^{b}
Acetone extract	18.00^{a}	20.0 ^{a}	18.33 ^{a}	19.33 ^{a}	15.33 ^{b}	16.67 ^{b}
LSD	1.104	1.517	0.99	1.315	2.068	1.444
Concentrations mg / ml						
100	14.75^{c}	8.75 ^{b}	11.75 ^{b}	11.75 ^{b}	13.00 ^{b}	13.50 ^{b}
200	15.75^{b}	10.0 ^{b}	12.75 ^{b}	12.75 ^{b}	15.25 ^{a}	14.25 ^{b}
400	17.50^{a}	11.50^{a}	15.25 ^{a}	15.25 ^{a}	16.50 ^{a}	17.17 ^{a}
LSD	0.95	1.313	0.86	1.139	1.791	1.251

Mean in each column in each followed by the same letter are not significantly different according to LSD test (p = 0.05) .

The antibacterial activity in Table (3) indicated that *Citrus sinensis* had highest significant effect in their ethanolic extract against *E. coli*, *Salmonella* sp. , *Enterobacter sakazakii* and *Bacillus cereus*, while the aqueous extract showed lowest significant effect, but the aqueous extract did not effect on *Salmonella* sp. The methanol and ethanol extracts showed highest effect with non significant difference between them against *Shigella flexneri*, while the aqueous extract showed lowest significant effect, The methanol extract showed highest significant effect with *Staphylococcus aureus* but the aqueous extract showed lowest significant effect. Also highest effect was at concentration 400 mg/ml, lowest significant effect was in 100 mg/ml .

Table 3. Effect of *Citrus sinensis* extracts against pathogenic bacteria (diameter of inhibition zone mm)

Plant extracts	Pathogenic bacteria					
	E. coli	***Salmonella* sp.**	***Shigella flexneri***	***En. sakazakii***	***St. aureus***	***B. cereus***
Aqueous extract	13.67^{c}	0.00^{d}	10.33^{b}	13.67^{b}	12.33^{c}	6.67^{c}
Methanol extract	14.44^{c}	18.0^{ab}	18.00^{a}	16.67^{a}	19.78^{a}	15.00^{b}
Ethanol extract	20.00^{a}	21.00^{a}	18.00^{a}	17.33^{a}	17.33^{b}	17.00^{a}
Acetone extract	16.00^{b}	10.0^{c}	17.67^{a}	15.00^{b}	14.33^{c}	16.67^{a}
LSD	1.281	2.435	1.577	1.606	2.056	1.267
Concentrations mg / ml						
100	12.58^{c}	9.25^{b}	13.25^{c}	11.50^{c}	11.00^{c}	10.50^{c}
200	17.00^{b}	12.75^{a}	16.00^{b}	15.75^{b}	15.08^{b}	14.00^{b}
400	18.50^{a}	14.75^{a}	18.75^{a}	19.75^{a}	21.75^{a}	17.00^{a}
LSD	1.109	2.109	1.348	1.391	1.780	1.0977

Mean in each column in each followed by the same letter are not significantly different according to LSD test (p = 0.05) .

Data from results in Table (4) showed that the aqueous extract of *Eucalyptus globulus* showed highest significant effect with *E. coli* while the acetone extract showed lowest significant effect. *Salmonella* sp. was resistance for all extracts. The methanol extract exhibited highest significant activity against *Shigella flexneri*, *Enterobacter sakazakii*, *Staphylococcus aureus* and *Bacillus cereus*, but the ethanol extract showed lowest significant effect against *Shigella flexneri and Staphylococcus aureus* The acetone extract showed lowest significant effect with *Enterobacter sakazakii* while the aqueous showed lowest significant effect with *Bacillus cereus*. Also highest effect was at concentration 400 mg/ml, lowest significant effect was in 100 mg/ml .

Table 4. Effect of *Eucalyptus globulus* extracts against pathogenic bacteria (diameter of inhibition zone mm)

Plant extracts	Pathogenic bacteria					
	E. coli	***Salmonella* sp.**	***Shigella flexneri***	***En. sakazakii***	***St. aureus***	***B. cereus***
Aqueous extract	16.33^{a}	0.00	14.22^{a}	14.00^{b}	12.78^{ab}	9.33^{c}
Methanol extract	13.33^{b}	0.00	15.00^{a}	17.11^{a}	14.67^{a}	13.33^{a}
Ethanol extract	15.0^{ab}	0.00	10.33^{b}	14.33^{b}	11.00^{b}	11.67^{b}
Acetone extract	10.67^{c}	0.00	11.67^{b}	12.00^{c}	13.67^{a}	10.56^{bc}
LSD	1.961	0.00	1.618	1.699	2.237	1.593
Concentrations mg / ml						
100	11.25^{c}	0.00	10.00^{c}	11.00^{c}	9.25^{c}	7.50^{c}
200	13.75^{a}	0.00	12.00^{b}	14.08^{b}	13.08^{b}	10.92^{b}
400	14.75^{a}	0.00	16.42^{a}	18.00^{a}	16.75^{a}	15.25^{a}
LSD	1.698	0.00	1.402	1.4719	1.937	1.379

Mean in each column in each followed by the same letter are not significantly different according to LSD test (p = 0.05) .

The present finding in Table (5) revealed that the methanol and aqueous extracts of *Psidium guajava* exhibited the best antibacterial activity with non significant different between them against *E. coli*, while the ethanol extract showed the lowest significant effect. The ethanol extract showed highest significant effect against *Salmonella* sp. while the acetone extract showed the lowest significant effect, the aqueous extract did not show any effect. The ethanol and aqueous extracts showed highest effect with non significant difference betweem them against *Shigella flexneri.* but the methanol and acetone extracts showed the lowest effect with non significant different between them. The aqueous extract showed highest significant effect against *Enterobacter sakazakii* but the acetone extract showed the lowest significant effect. The methanol extract showed highest significant effect against *Staphylococcus aureus* but the acetone extract showed the lowest effect. The methanol extract showed highest significant effect against *Bacillus cereus* but the aqueous extract showed the lowest significant against *Bacillus cereus*. Also highest effect was at concentration 400 mg/ml, lowest significant effect was in 100 mg/ml.

Table 5. Effect of *Psidium guajava* extracts against pathogenic bacteria (diameter of inhibition zone mm)

Plant extracts	Pathogenic bacteria					
	E. coli	***Salmonella* sp.**	***Shigella flexneri***	***En. sakazakii***	***St. aureus***	***B. cereus***
Aqueous extract	15.67^{a}	0.00^{d}	14.44^{a}	15.67^{a}	14.00^{ab}	10.67^{c}
Methanol extract	16.67^{a}	15.33^{b}	12.00^{b}	14.33^{ab}	15.00^{a}	15.22^{a}
Ethanol extract	11.67^{b}	18.78^{a}	14.89^{a}	13.00^{b}	13.00b^{c}	12.33^{b}
Acetone extract	13.00^{b}	9.67^{c}	12.00^{b}	11.56^{c}	12.33^{c}	12.00^{b}
LSD	1.486	1.137	1.501	1.380	1.309	1.395
Concentrations mg / ml						
100	11.75c	9.75^{b}	10.92^{b}	11.50^{c}	11.50^{c}	10.00^{c}
200	14.00^{b}	10.58^{b}	14.00^{a}	13.67^{b}	13.50^{b}	12.42^{b}
400	17.00^{a}	12.50^{a}	15.08^{a}	15.75^{a}	15.75^{a}	15.25^{a}
LSD	1.287	0.98	1.300	1.197	1.134	1.177

Mean in each column in each followed by the same letter are not significantly different according to LSD test (p = 0.05) .

From the Table (6) it could be seen that the acetone extract showed the highest significant activity against *E. coli* and *Enterobacter sakazakii* but the aqueous extract showed lowest significant effective. The methanol extract showed the highest significant effect against *Salmonella* sp., *Shigella flexneri Staphylococcus aureus* and *Bacillus cereus*, but the aqueous extract showed lowest significant effect. Also highest effect was at concentration 400 mg/ml, lowest significant effect was in 100 mg/ml .

The antibacterial efficacy of the ethanol and methanol extracts in Table (7) were the highest activity with non significant between them against *E. coli* but the acetone extract showed lowest significant effect. The methanol and acetone extracts showed the highest effect with non significant difference between them against *Salmonella* sp, but the ethanol extract showed lowest significant effect. The methanol and aceton extracts showed highest effect with non significant difference between them against *Shigella flexneri* and

Enterobacter sakazakii, while ethanol extract showed lowest significant effect with *Shigella flexneri*, non significant effect with *Enterobacter sakazakii*. The acetone extract showed the highest significant effect against *Staphylococcus* and *Bacillus cereus* but the methanol extract showed lowest significant effect. The aqueous extract showed no effect against all tested bacteria. Also highest effect was at concentration 400 mg/ml, lowest significant effect was in 100 mg/ml .

Table 6. Effect of *Matricaria chamomilla* extracts against pathogenic bacteria (diameter of inhibition zone mm)

Plant extracts	Pathogenic bacteria					
	E. coli	***Salmonella* sp.**	***Shigella flexneri***	***En. sakazakii***	***St. aureus***	***B. cereus***
Aqueous extract	`1.67[c]	4.00[c]	2.33[c]	5.33[c]	1.67[c]	0.33[c]
Methanol extract	11.33[b]	14.11[a]	11.44[a]	14.33[a]	16.33[a]	12.33[a]
Ethanol extract	12.6[ab]	0.00[d]	8.33[b]	11.67[b]	11.00[b]	10.33[b]
Acetone extract	13.33[a]	10.33[b]	10.67[a]	15.00[a]	15.00[a]	10.00[b]
LSD	1.591	1.497	1.547	1.172	1.486	1.236
Concentrations mg / ml						
100	7.25[c]	5.58[c]	6.25[c]	10.00[c]	8.75[c]	6.75[c]
200	9.75[b]	7.00[b]	8.33[b]	11.50[b]	10.75[b]	8.00[b]
400	12.25[a]	8.75[a]	10.00[a]	13.25[a]	13.50[a]	10.00[a]
LSD	1.378	1.296	1.340	1.015	1.287	1.071

Mean in each column in each followed by the same letter are not significantly different according to LSD test (p = 0.05) .

Table 7. Effect of *Allium cepa* extracts against pathogenic bacteria diameter of inhibition zone mm)

Plant extracts	Pathogenic bacteria					
	E. coli	***Salmonella* sp.**	***Shigella flexneri***	***En. sakazakii***	***St. aureus***	***B. cereus***
Aqueous extract	0.00[c]	0.00[b]	0.00[c]	0.00[b]	0.00[c]	0.00[c]
Methanol extract	8.33[a]	6.89[a]	9.67[a]	7.78[a]	7.00[b]	8.00[b]
Ethanol extract	8.67[a]	0.89[b]	5.00[b]	6.67[a]	7.67[ab]	8.67[ab]
Acetone extract	6.00[b]	6.67[a]	8.67[a]	7.44[a]	8.67[a]	9.33[a]
LSD	1.261	1.775	1.495	1.559	1.389	1.192
Concentrations mg / ml						
100	4.50[b]	2.17[b]	4.25[c]	3.50[c]	4.50[c]	5.00[c]
200	5.50[b]	3.25[b]	5.75[b]	5.00[b]	5.75[b]	6.50[b]
400	7.25[a]	5.42[a]	7.50[a]	7.92[a]	7.25[a]	8.00[a]
LSD	1.092	1.537	1.278	1.350	1.203	1.032

Mean in each column in each followed by the same letter are not significantly different according to LSD test (p = 0.05) .

Data obtained in Table (8) showed that the ethanol and methanol extracts showed the highest effect with non significant difference between them against *E. coli*, but the acetone extract showed lowest significant effect. The methanol extract showed high significant effect against *Salmonella* sp., but other extracts showed no effect The methanol extract showed highest effect with non significant effect against *Shigella flexneri*, *Enterobacter sakazakii* and *Bacillus cereus*, but the ethanol extract showed lowest significant effect against *Shigella flexneri*, the acetone extract showed lowest significant effect against *Enterobacter sakazakii* and *Bacillus cereus*. The aceton showed highest significant effect against *Staphylococcus aureus* while methanol extract showed lowest significant effect. The aqueous extract showed no effect on all bacteria. Also highest effect was at concentration 400 mg/ml, lowest significant effect was in 100 mg/ml .

Table 8. Effect of *Allium sativum* extracts against pathogenic bacteria (diameter of inhibition zone mm)

Plant extracts	Pathogenic bacteria					
	E. coli	***Salmonella* sp.**	***Shigella flexneri***	***En. sakazakii***	***St. aureus***	***B. cereus***
Aqueous extract	0.00[c]	0.00[b]	0.00[c]	0.00[c]	0.00c	0.00[c]
Methanol extract	9.33[a]	9.33[a]	9.67[a]	7.67[a]	7.67b	11.44[a]
Ethanol extract	9.44[a]	0.00[b]	2.78[c]	0.00[c]	0.00c	9.78[b]
Acetone extract	7.33[b]	0.00[b]	6.22[b]	3.44[b]	9.33a	9.33[b]
LSD	1.306	1.905	3.277	1.577	1.577	1.133
Concentrations mg / ml						
100	5.00[b]	1.50[b]	3.75[a]	1.83[b]	3.00[b]	6.75[b]
200	6.75[a]	1.75[b]	5.00[a]	2.50[b]	4.50[a]	7.58[b]
400	7.83[a]	3.75[a]	5.25[a]	4.00[a]	5.25[a]	8.58[a]
LSD	1.320	1.650	2.838	1.104	1.365	.098

Mean in each column in each followed by the same letter are not significantly different according to LSD test (p = 0.05) .

The results obtained in this study revealed that the identified phytochemical compounds may be the bioactive constituents responsible for the efficacy of the tested plant. These compounds which found in the plants have antimicrobial activity and are known to exhibit medicinal physiological activities (Sofowra, 1993). Several phenolic compounds like tannins are potent inhibitors of many hydrolytic enzymes such as pectolytic macerating enzymes used by pathogenic bacteria. Also many plants which contain non toxic glycosides can get hydrolyzed to release phenolic are toxic to pathogenic bacteria (Aboaba and Efuwape, 2001).

Their inhibitory effect on the tested bacteria attribute to the crude form which contained the phytochemicals in large quantities, composition of bacterial cell wall and type of solvents which used in the extraction the bioactive compounds from the plants such as phenolic compound. Shan *et a*l. (2005) reported that methanol extract of rosemary leaves and stem contain a phenolic concentration of 5.07 gm gallic acid (dry weight) but the concentration in water extract 185 mg gallic acid (Dorman *et al*., 2003). The polarity of the solvents play role in the extraction of natural products from plants and chemical constituents are partially separated attribute to their polarity and the polarity of the solvent used. Alkaloids are used in medicine as anesthetic agents also the presence of saponin in plant responsible for tonic, stimulating activities and anti-inflammation.

The rosemary and orange peel were greater antibacterial activity against all pathogenic bacteria while *Salmonella* sp. was resistance , this resistant due to the presence of their cell wall lipopolysaccharaide. Followed by camphor and guava. Chamomile showed moderate effect. Garlic and onion exhibited weakest activity. Black seed did not inhibit the growth on all bacteria. The medicinal properties of the plants could be attributed to the presence one or more of plant natural products.

Rosemary have a potent antioxidant and antibacterial activity (Erkan *et al.*, 2009) due to flavonoids and hydroxyl group which are phenolic compounds. These possess biological properties, have antimicrobial properties against microorganisms. The presence of active compounds in orange peels makes it useful in folk medicine to treat many diseases therefore orange peel used for treatment infection disease (McGravey and Croteau, 1995). D- limonene terpene derived from the peels of citrus fruits it is a cyclic monoterpene. Also citrus contain flavonoids which are hydroxylated phenolic substance have antimicrobial and antioxidant properties.

Camphor have antimicrobial activity because it contain bioactives such as comarins, essential oils, flavonoids, triterpenes, triterpenes and ellagitannins are useful for treatment diseases. Guava leaf extracts contain several compounds such as comarins, essential oils, phenolic compound, glycosides, flavonoids, triterpenes and ellagitannins. These compounds showed antimicrobial and antioxidant properties. Tannin which found in guava is phenolic compound can be soluble in various solvent such as water, alcohol and acetone. It used as natural defense mechanism against microorganisms by binds with protein, complex with polysaccharide (Chung *et al*., 1988). Chamomile have antimicrobial activity attributed to the presence of tannins which are water soluble polyphenols (Cinco *et al*., 1983). Chamomile used for treating colic, cloup convulsions, fiver, diarrhea, indigestion insomania, infantile, convulsions, toothache and bleeding. Garlic and onion extracts showed weakest effect against all tested bacteria may be related to the methods extraction of secondary metabolism and its concentration. Many studies reported that garlic has antimicrobial activity against Gram negative and Gram positive bacteria (Ross *et al*., 2001). Azu and Onyeagba (2007) reported that garlic is more effective than onion and may be due to the high molecular weight of the onion extract. Black cumin did not effect on all tested bacteria may be presence of polysaccharides in cell wall of gram negative bacteria prevent reaching the active compounds , essential oils and extracts to the cytoplasmic membrane (Duraipandiyan *et al*., 2006) .

The present study provides that solvent extract of rosemary, orange peel, guava, camphor, chamomile, onion, garlic and black seed contains medicinally bioactive compounds and they can use as traditional medicine for treatment of various diseases.

REFERENCE

Ababutain, I.M (2011). Antimicrobial activity of ethanolic extracts from some medicinal plant. Australian J. Basic and Appl. Sci. 5 (11) : 678 – 683 .

Azu, N.C and Onyeagba, R.A (2007). Antimicrobial properties of extract of *Allium cepa* (onion) *Zingiber officinale* (ginger) on *E. coli*, *Salmonella typhi* and *B. subtilus*. The interned J. tropical medicine.3 . 2.

Chung, K.T ; Wong, T.Y ; Wei, C.I ; Huang, Y.W and Lin, Y (1988). Tannins and human health : a review. Crit. Rev. Food. Sci. Nutr. 38 (6) : 421 – 464.

Cinco, M ; Banfi, E and Tubaro, A (1983). A microbiological survey on the activity of a hydroalcoholic extract of chamomile. Int J Drug Res. 145 – 151 .

Cowan, M.M (1999). Plant products as antimicrobial agents. Clin Microbiol Rev. 12 (4) : 564 – 582 .

Criagg, G.M and David, J.N (2001). Natural product drug discovery in the next millennium. J. Pharm. Biol. 39 : 8 – 17 .

Dorman, H.J.D ; Peltoketo, A ; Hiltunen, R and Tikkanen, M.J (2003). Characterisation of the antioxidant properties to de-odourised aqueous extracts from selected amiaceae herbs. J. Food Chem. 83 : 255 – 262.

Duncan, D.B. (1955). Multiple range multiple F-test. Biometrics,11: 1-42

Duraipandiyan, V ; Ayyanar, M and Igancimuthu, S(2006). Antimicrobial activity of some ethnomedicinal plants used by paliyar Tribe from Tamil Nadu, India. BMC Complement Altern Med. 6 : 35

Edoga, H.O ; Okwu, D.E and Mbaebie, B.O (2005). Phytochemicals constituents of some Nigerian medicinal plants. Afr. J. Biotechnol. 4 (7) :685 – 688.

Erkan, N ; Ayranic, G and Ayranci, E (2008). Antioxidant of rosemary (*Rosmarinus officinalis* L) extract, black seed (*Nigella sativa* L) essential oil, carnosic acid, rosmarinic acid and SESAMOL. J. Food Chem. 110 : 76 – 82.

Harborne, J.B (1988). Phytochemical methods A Giude to Modern techniques of plants analysis 2nd ed. London. Chapman & Hall, 1 – 226 .

Lesse, A.J (1995). Penicillins and other cell wall active agents. Essentials of pharmacology, WBSanders Co, Philadelphia, 362 – 373 .

Mojab, F ; Kamalinejad, M ; Ghaderi, N and Vanidipour, H.R (2003). Phytochemical screening of some species of Iranian plants. Iran. J. Pharm. Res. 3 : 77 – 82 .

Mordi, R.M and Erah, P.O (2006). Susceptibility of common urinary tract isolates to the commonly used antibiotics in a tertiary hospital in southern Nigeria. Afr. J. Biotechnol. 5 (11) : 1067 – 1071 .

NCCLS (1999). Methods for determining bactericidal activity of antimicrobial agents. Approved guideline, M26-A. National Committee for Clin. Laboratory Standards, Wayne, Pa.

Parkh, J and Chanda, S (2007). Antibacterial and phytochemical studies on twelve species of Indian medicinal plants. Afr. J. Biomed Res. 10 : 175 – 181 .

Parkh, J and Chanda, S (2008). Phytochemicals screening of some plants from western region of India. Plant Arch. 8 : 657 – 662 .

Perumal, S.R and Gopala, K.P (2007). Current status of herbal and their future perspectives, Nature Precedings .

Ross, Z.M ; O,Gara, E.A ; Hill, D.J ; Sleightholme, H.V and Maslin, I.J (2001). Antimicrobial properties of garlic oil against human enteric bacteria : Evaluation of methodologies and comparisons with garlic oil sulfides and garlic powder. Appl. Environ. Microbiol. 67 : 475 – 480 .

SAS (2001). SAS / STAT Guide for personal computer. SAS Inst. Cary. N.C .

Shan, B ; Cai, Y.Z ; Sun, M and Corke, H (2005). Antioxidant capacity 26 spice extracts and characterization of their phenolic constituent. J.Agricul and Food Chem. 53 : 7749 – 7759.

Shareef, A.A (2011). Evaluation of antibacterial activity of essential oils of *Cinnamomum* sp. and *Boswellia* sp. J. Basrah Res. 37 (5) : 60 – 71 .

Sofowra, A (1993). Medicinal plants and traditional and medicines in Africa. Spectrum Books Ltd. Ibadan, Nigeria, 191 – 289 .

Vasu, K ; Goud, J.V Suryam, A ; Singara, Chary, M.A (2009). Biomolecular and phytochemical analyses of three aquatic angiosperms. Afr. J. Microbiol. Res. 3 (8) : 418 – 421 .

Westh, H ; Zinn, C.S and Rosdahl, V.T (2004). An international multi cancer study of antimicrobial consumption and resistance in *S. aureus* isolates from 15 hospitals in 14 countries. Microbe Drug Resistance, 10 : 169 – 176 .

تقدير المكونات الكيميائية والنشاط المضاد للبكتيريا لبعض النباتات الطبية ضد البكتيريا الممرضة .

عايدة حافظ عفيفى , فتحى إسماعيل على حوقه و أمانى كمال الدسوقى النملة
قسم الميكروبيولوجى – كلية الزراعة – جامعة المنصورة –المنصورة – مصر

اجريت هذه الدراسة لتقدير المكونات الكيميائية الفعالة والنشاط المضاد للبكتيريا لبعض المستخلصات النباتية على البكتيريا الممرضة وتشمل : *Escherichia coli* , *Salmonella* sp. , *Shigella flexneri*, *Enterobacter sakazakii* *Staphylococcus aureus* and *Bacillus cereus* واجرى التحليل الكيميائى بطريقة التحليل الوصفى للكشف عن القلويدات , الصابونينات , التريبينات , الفلافونيدات , التانينات و الجليكوسيدات . وإختبرت فعالية هذه المستخلصات النباتية بإستخدام طريقة الإنتشار فى الأجار وذلك بقياس قطر منطقة التثبيط . وأظهرت النتائج الأتى : أظهر التحليل الوصفى للمستخلصات النباتية لكل من حصالبان , البرتقال , الكافور , الجوافة , البابونج , البصل , الثوم وحبة البركة أنها تحتوى على التريبينات – القلويدات – الفلافونيدات – التانينات – الصابوبينات – الجليكوسيدات وقد أظهر النشاط المضاد للبكتيريا للمستخلصات النباتية لحصالبان , البرتقال , الكافور والجوافة أعلى تأثير معنوى ضد البكتيريا الممرضة , بينما أظهرت المستخلصات النباتية لكل من البصل والثوم أقل تأثير معنوى ضد البكتيريا الممرضة أما حبة البركة كانت عير مؤثرة على جميع البكتيريا الممرضة . وبالتالى توصلت هذه الدراسة إلى إحتواء هذه النباتات على مواد فعالة هامه لها خواص مضادة للبكتيريا ويمكن أستخدامها كعوامل طبيعية بديلة مضادة للميكروبات الممرضة لمعالجة الأمراض وكذلك فى الصناعات الدوائية.