

Screening of Some Natural and Cultivated plants in Sudia Arabia Fight Infections And inhibit growth of pathogenic Bacteria

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Abstract: Aqueous, ethanol, methanol and petroleum ether, extracts of 8 Medicinal plant species, Grow naturally on the land of the Kingdom Sudia Arabia, traditionally used in for treatment of ailments of infectious and/or inflammatory nature were screened for in vitro antibacterial activities. Antibacterial activity was tested using the agar diffusion method. All the antibacterial activity was against 10 bacterial species Gram-positive and Gram-negative; *Staphylococcus aureus*. ATCC 33591, *Proteus Mirabilis* ATCC 43071, *Escherichia. Coli* ATCC 2592, *Klebsiella pneumoniae*. ATCC 700603, *Klebsiella pneumoniae*. ATCC 13883, *Salmonella typhi*. ATCC 14028, *Enterococcus faeculis*. ATCC 29212, *Pseudomonas aeruginosa*. ATCC 27853 *Staphylo coccus aureus*. ATCC 25923, *Proteus mirabils*. ATCC 12153 with 8 plants species showing some activity against all tested bacteria with different degree. The highest activity was found in the aqueous, ethanol and methanol extracts respectively of *Abutilon pannosum*, *Salavadora persicalin*, *Matricaria chamomilla*, *Citrullus colocynthis*, *Ricinus Communis* *Azadirachta indica* and *Ocimum basilicum L*. In most cases, aqueous, ethanol and methanol extracts show stronger inhibition of bacteria than other extracts. Minimum Inhibitory Concentration of Aqueous extract of plants used (microgram/ml) (MIC) was determined for each plant for all strains of bacteria..

[Amnah A.H. Rayes. **Screening of Some Natural and Cultivated plants in Sudia Arabia Fight Infections And inhibit growth of pathogenic Bacteria**. *Researcher*. 2012;4(7):17-28]. (ISSN: 1553-9865). <http://www.sciencepub.net/researcher>. 4

Keywords: Medicinal plants; Antibacterial activity; agar diffusion method; Sudia Arabia.

Introduction:

Traditional medicine is widely practised in the world and about 400 plant species have been recorded to be used in traditional remedies (**Matu and Staden, 2003**). In the rural areas, reliance on traditional medicine is high and is attributed to both economic and cultural factors (**Aketch, 1992; Haniffa and Kavitha, 2012**). From an economic point of view, the high cost of imported conventional drugs and/or inaccessibility to western health care facilities implies that traditional mode of health care is the only form of health care that is affordable and available to the rural people. The use of plants as a source for the treatment of many diseases due to the prehistoric times to the peoples of all continents and this old tradition led to a search for a cure for infectious diseases long before where people were aware of the presence of microbes (**Rajashree et al., 2011**). Where it has proven successful treatment of many diseases using plants and herbs or extracts for this purpose. In recent years, resistance has been the indiscriminate use of chemical drugs trade resistance of micro-organisms that cause diseases in both humans and plants. (**Davis, 1994 ; Service, 1995**).

Antibacterial activity is the ability of the chemical effective to inhibit or kill bacterial cells are being used different types of antibiotics for the treatment

of diseases has been getting most of these antibiotics from micro-organisms and the recent trend now is to get antibiotics from natural sources. (**Karaman et al., 2003; Abad et. al., 2007**).

Saudi Arabia has a wide variety of plants and medicinal herbs, so this study was aimed to use some medicinal plants located in the region of Mecca (Abutalon, Arak, Chamomile, Mental, Bitter melon, Castor, Neem and Basil) so as to provide many of the active substances from natural sources for using in the pharmaceutical industry. As a trial to survey for the natural wealth of the active substances stored and abundant in plants existing and widespread abundance in the environment of Saudi Arabia, through the extraction active substances from plants using a variety of organic solvents to determine the ability of the different active substances on the inhibition of bacterial growth for many bacterial strains.

Material and methods:

Plants collection used in the study: Plants used In this Study grow naturally in the environment and the other planted. The selection of these plants due to the abundance of growth in an environment of Saudi Arabia and get it and some of which are commonly used in some types of traditional medicine. Plants

that grow naturally a publican, Abutalon, bitter melon has been obtained from various places of Makkah and Azizia include Awali and Wadi Fatima. As for the plant and keeping Mary have lost arak obtained from merchants Clarke next to the Grand

Mosque in Mecca. The plants are cultivated Mental, chamomile, castor, Neem and Basil some purchased from nurseries in and others were purchased from shops (table1).

Table 1 plants species used in the study and part used in extraction

Plant species (common name)	Latin name	Part of plant used
Abutalon Plant	<i>Abutilon pannosum</i>	Leaves
Arak Plant	<i>Salavadora persicalin</i>	Roots
Chamomile Plant	<i>Matricaria chamomilla</i>	Flowers
Mental plant	<i>Mentha logifolia</i>	Leaves
Bitter melon	<i>Citrullus colocynthis</i>	Fruits
Castor Plant	<i>Ricinus Communis</i>	Seeds
Neem plant	<i>Azadirachta indica</i>	Leaves
Basil	<i>Ocimum basilicum L.</i>	Leaves

Preparation of plant extracts:

Method is used for the preparation of extracts of eight medicinal plants under study, drying the plant in room temperature then transfer the dry plant samples to a fine powder with a different degree of smoothness depending on the type of plant used and the extent of fiber it contains. Some plant samples used in the study were broken into small pieces. To get the powder of plant and the preparation of plant extracts. Weight of the dry powder of the samples of plant was taken and put it in clean and sterile glass beaker, **Sule and Agbabiaka (2008)**.

Solvents used to get a medical plant extracts as follows:

Aqueous extract (Water Extract), Ethanol 70 % Extract, Methanol Extract, Petroleum ether extract, Placed on a shaking flasks for 3 hours and then left in the temperature of the laboratory for 24 hours. medical plant extracts filtered using a cotton flannel. Re-filtered using filter paper to get the clear filtrate free of any residue or impurities.

Bacterial strains used for inhibitory activity:

Staphylococcus aureus. ATCC 33591, *Proteus Mirabilis* ATCC 43071, *Escherichia. Coli* ATCC 2592, *Klebsiella pneumoniae*. ATCC 700603, *Klebsiella pneumoniae*. ATCC 13883, *Salmonella typhi*. ATCC 14028, *Enterococcus faeculis*. ATCC 29212, *Pseudomonas aeruginosa*. ATCC 27853 *Staphylo coccus aureus*. ATCC 25923, *Proteus mirabilis*. ATCC 12153 were the test microorganisms which were obtained from King Abd-El Aziz Medical Hospital (National Guard) in Jeddah, Kingdom of Sudia Arabia.

Preparation of Growth Media:

Culture media used by a number of medium, including, liquid and solid for development of

strains of bacteria and maintain for a long time to conduct sensitivity tests ; Nutrient agar media, Blood Agar media, nourishing broth, and Molar Henton agar media.

Study the inhibitory effect of plant extracts to bacterial growth:

The effect of bacterial growth inhibitor of plant extracts test sensitivity disk Diffusion Susceptibility test was studied according to (**Washington, 1981**). activated bacterial strains used in the study, stored on blood agar re-grown on nutrient agar media for single colonies spaced attended the hanging of the bacterial strains tested by choosing the number of 3-5 single bacterial colony and far in between, it was placed in a sterile test tube containing 5 ml of liquid media (Molar Hinton) then incubated at a temperature of 37°C for 24 hours. for test sensitivity and dip needle inoculation with a sterile cotton node (Swab) in the bacterial suspension for several times in the suspense and pressure on the inner wall of the tube above the liquid level to remove the increase of the bacterial suspension of the needle. Inoculate your Molar Hinton agar by moving the loop on the surface of agar in three different directions to make sure the distribution of bacterial suspension. it was left for several minutes to dry bacterial suspension. Developed three discs of filter paper of diameter 6 mm and a fixed size (0.15 ml) of extract of plant to test the impact on the bacterial strains tested on the surface of agar (Molar Hinton) the disks was pressed gently to ensure contact with the full surface of the agar in order not to move disks from the first set, which proved to him from the beginning and put dishes in the temperature of the laboratory for a period ranging between 3-5 hours before you put them in the nursery, repeated the experiment three times. used in these experiments officer (Control is a

papers saturated with solvent used in the preparation of extract of plant you want to know the impact damper size equal to the size of the plant extract user to know the impact of inhibiting bacterial growth and were distributed to the disks on the dishes an average of three tablets each dish incubate at 37 ° C for 24 hours dishes were examined after incubation period and results were recorded measuring the diameter of the zone of bacterial growth (inhibition zone diameter) in millimeters.

Determination of Minimum Inhibitory Concentration (MIC):

Was determined according to **Zain et al. (2012)**, briefly the least concentration of inhibitor of plant extracts and less concentration of bacteria, where he attended the aqueous extract of medicinal plants under study using 100 g per plant and then dried aqueous extracts of oven drying at a temperature of 40° C to get the weights of dry for five medicinal plants from plants used in the study, which recorded the ability of a clear to cause inhibition of the growth of bacterial strains under study. Reduced the aqueous extracts of the medicinal plants used under study upward to determine the lowest concentration inhibiting bacterial growth of strains of bacteria under study (MIC), where the weight of dry plant of 1 g / liter in order to obtain concentrations of the following: (0, 10, 20, 40, 60,80,100, micrograms / ml). 5 ml of each concentration of the aqueous extracts of the eight plants was taken in test tubes and added to 5 ml of nutrient broth (Nutrient broth) with sterile 0.1 ml bacterial suspension concentration (1,2 x10⁷cfu/ml⁻¹ 0.5 Mcfarland standards) Two types of controls (positive and negative) and incubated at 37⁰ C for 24 hours as follows: Positive Control, a tube containing nutrient broth and commentator with the bacterial strain and plant extract and negative Control, It is a tube containing nutrient broth and sterile plant extract. the lowest concentration inhibiting bacterial growth (MIC) of aqueous extracts of medicinal plants used under study was recorded and (which does not show the turbidity of the inability of bacterial strains on growth).

Results and discussion:

Regarding to effect of plant extraction inhibitory of bacterial growth, The effect of active substances of the different parts of eight medicinal plants used under study was studied. the effect of plant extracts and organic solvents of different test sensitivity against bacterial growth in a method of the spread of the disk was also studied and showed the results tables (2&3) that the inhibitory effect of bacterial growth due to the active chemicals derived from

plants, not by the organic solvents used in extraction.

Regarding to effect of *Abutilon pannosum* leaves extraction inhibitory effect of bacterial growth, is clear from the results table (2&3) that the examination of extracts of leaves of Abutalon have a counter effect to the growth of most bacterial strains tested with the strength of inhibitory effects differed between a very strong inhibitory effect to weak and this is consistent with **Mohamed et. al.(2010)** who make extraction of 23 plants for 19 families of plants found in Sudan, including the plant Abutilon which are used traditionally in the treatment of various diseases and survey the chemical that's the tannins and steroids and Alterbanued where they study the impact of bio-against pathogenic bacteria such as: *Escherichia coli* NCTC 8196; *Staphylococcus aureus* NCTC 6447 and *Klebsiella pneumonia* ATCC 35657 and the results showed that the methanol extract has the ability to inhibit the same strains tested according to this study and this confirms the effect of the active ingredient in the methanol extract.

Regarding to effect of *Salavadora persicalin* roots extraction inhibitory of bacterial growth, The results listed in table (2&3) we find that the plant extracts of the roots of a plant arak (distilled water - ethanol (70%) - methanol) affect as a common inhibitor for the growth of most bacterial strains studied. Did not have any other effect of extracts of an inhibitor of the growth of all bacterial strains tested. Where different force of impact between the inhibitory effect of a strong inhibitor to the influence of a weak inhibitor.

This is agree with the results obtained by **Akpata and Akinrimisi (1977)** who examined the extractors of water and alcohol to the roots of arak plant which has disincentive effect for the growth of the following bacterial strains: *Streptococcus pyogenes*; *Staphylococcus aureus*; *Escherichia coli* and *Pseudomonas aeruginosa*.

The results of present study nearly agree with the results conducted by **Saadabi et. al., (2006)** who confirmed in their study the presence of medium for inhibiting growth of bacterial strains Gram positive and negative *Staphylococcus aureus*; *Bacillus subtilis*; *Escherichia coli* and *Pseudomonas aeruginosa*. Also **Al-Shmma et. al. (2006)** demonstrated that minimum inhibitory concentration of aqueous extracts of the roots of a plant arak (tooth brushing) (MIC 20% and that the effect of the extract vital against the following bacterial strains *Staphylococcus aureus*; *Streptococcus mutans*; *Pseudomonas aeruginosa* and *Escherichia coli*.

Table 2: Showing the inhibitory effect of plant extracts with different solvents to investigated bacterial strains

Plant sp.	Solvent	Bacterial strain				
		<i>Staph. aureus.</i> ATCC 33591	<i>Prot. mirabilis</i> ATCC 43071	<i>E. coli</i> ATCC 25922	<i>K. pneumoniae.</i> ATCC 700603	<i>K.pneumoniae.</i> ATCC 13883
<i>Abutilon pannosum</i>	Water	+	+	+++	-	-
	Ethanol	++	+	++++	-	+
	Methanol	-	+	++	-	+
	Petroleum ether	-	-	-	-	-
<i>Salavadora persicalin</i>	Water	++	++	+	++	+
	Ethanol	+++	++	+	+	+
	Methanol	+	+	++	+	+
	Petroleum ether	-	-	-	-	-
<i>Matricaria chamomilla</i>	Water	+	++++	++	+	+
	Ethanol	+	++++	+	+	+
	Methanol	-	+++	+	-	-
	Petroleum ether	-	-	-	-	-
<i>Mentha logifolia</i>	Water	-	+	+	+	-
	Ethanol	-	-	-	-	-
	Methanol	-	+	-	-	-
	Petroleum ether	-	-	-	-	-
<i>Citrullus colocynthis</i>	Water	+	++	++++	+	++
	Ethanol	-	++	++++	+++	+
	Methanol	-	+	+	-	+
	Petroleum ether	-	-	-	-	-
<i>Ricinus Communis</i>	Water	+	+	+	++	+
	Ethanol	+	++	+	++	+
	Methanol	-	++	-	-	-
	Petroleum ether	-	-	-	-	-
<i>Azadirachta indica</i>	Water	-	-	-	-	-
	Ethanol	-	-	-	-	-
	Methanol	-	+	+	+	-
	Petroleum ether	-	+	++	-	-
<i>Ocimum basilicum L.</i>	Water	-	-	-	-	-
	Ethanol	++	+	+	+	-
	Methanol	+	+	+	-	-
	Petroleum ether	-	-	+	-	-

	ether	-	-	-	-	-
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Table 3: Showing the inhibitory effect of plant extracts with different solvents to investigated bacterial strains

Plant sp.	Solvent	Bacterial strain				
		<i>S. typhi</i> . ATCC 14028.	<i>E. faeculis</i> . ATCC 29212	<i>P. aeruginosa</i> . ATCC 27853.	<i>Staph. aureus</i> . ATCC 25923	<i>Prot. mirabilis</i> . ATCC 12153

<i>Abutilon pannosum</i>	Water	-	++++	+++	+	+
	Ethanol	-	+	++	+	+
	Methanol	-	+	++	+	+++
	Petroleum ether	-	-	-	-	-
<i>Salavadora persicalin</i>	Water	++	++	++	+	++
	Ethanol	+	+	+	+	+
	Methanol	+	-	-	+	-
	Petroleum ether	-	-	-	-	-
<i>Matricaria chamomilla</i>	Water					
	Ethanol					
	Methanol					
	Petroleum ether	+	+	+	+	+++
<i>Mentha logifolia</i>	Water	+	+	+	-	++
	Ethanol	-	-	-	-	-
	Methanol					
	Petroleum ether					
<i>Citrullus colocynthis</i>	Water	-	+	+	-	+
	Ethanol	-	-	-	-	-
	Methanol	-	-	-	-	-
	Petroleum ether	-	-	-	-	-
<i>Ricinus Communis</i>	Water	+	++	++	-	+
	Ethanol	+	++	+	+	+
	Methanol	+	+	+	+	+
	Petroleum ether	-	-	-	-	-
<i>Azadirachta indica</i>	Water	-	-	+	+	-
	Ethanol	-	+	++	++	-
	Methanol	-	-	-	-	-
	Petroleum ether	-	-	-	-	-
<i>Ocimum basilicum</i>	Water					
	Ethanol					
	Methanol	+	+	+	+	-
	Petroleum ether	-	++	-	-	-
<i>Ocimum basilicum</i>	Water	-	+	+	+	-
	Ethanol	-	-	-	-	-
	Methanol	-	-	-	-	-
	Petroleum ether	-	-	-	-	-
<i>Ocimum basilicum</i>	Water	+	+	+++	++++	-
	Ethanol	+	-	++	+++	-
	Methanol	-	-	-	+	-
	Petroleum ether	-	-	-	-	-

Diameter of inhibition zone = The mean of three replicates of recorded zones of inhibition

(ATCC) American Type Culture Collection

(-) = no effect (+) = weak effect (5-10mm) (++) = medium effect (10-15mm)

(+++)= strong effect (15-20mm) (++++)= very strong effect (20-30mm)

Al-Bayati and Sulaiman (2008) that the extractors of water and methanol extract of the roots of a plant arak effect inhibiting the growth of seven bacterial strains isolated from the mouth and causing diseases using the spread of the disk on the agar *Staphylococcus aureus*, *Streptococcus mutans*,

Streptococcus faeculis, *Streptococcus pyogenis*, *Lactobacillus acidophilus*, *Pseudomonas aeruginosa*, has been observed that the aqueous extracts of the roots of a plant arak effective disincentive for the growth of all bacterial strains isolated from the mouth and especially strain

Pseudomonas aeruginosa has reached the diameter region inhibition 22.3 mm at the lowest concentration of extract (0.781 mg / ml) on the strain of *Streptococcus faecalis*. This is consistent with the current study. The ability of the aqueous extract to make a moderate inhibiting effect (10-15) for the growth of bacterial strain of Gram negative ATCC 27853 *Pseudomonas aeruginosa*.

Concerning the effect of *Matricaria chamomile* flowers extraction inhibitory of bacterial growth, results of the listed plant extracts, we find that the three plant chamomile flowers (distilled water - ethanol (70%) - methanol) have a joint effect of an inhibitor of the growth of most bacterial investigated. Extracts had no effect on any other all strains tested. The different strength of the inhibitory effect between strong inhibitory effect to the impact of a weak inhibitor, and noted that the bacterial strain of Gram negative *P. mirabilis* ATCC 43071 are most sensitive to plant extracts of chamomile flowers, three (distilled water - ethanol (70%) - methanol).

Dulger and Gonuz (2004) showed that for a number of extracts of medicinal plants used in the treatment of different microbial diseases and had chamomile plant ethanol extract against the antimicrobial activity and their impact on the growth of many micro-organisms and the resistance of the activity of nine bacterial strains positive and negative character gram (*Esharichia coli* ATCC 11230; *Staphylococcus aureus* ATCC 6538p; *Klebsiella pneumonia* UC57; *Pseudomonas aeruginosa* ATCC 27853; *Proteus vulgaris* ATCC 8427; *Bacillus cereus* ATCC 7064; *Mycobacterium smegmatis* CCM 2067; *Listeria monocytogenes* ATCC 15313 and *Micrococcus luteus* CCM 169), The study carried out by **Essawi and Srour (2000)** a clear difference in the activity of the impact of organic extracts and water for the same plant, and this is what was observed with the current study of a clear difference in the effect of extracts and other organic, where they noticed that the effect of extracts organic medicinal plants used in the study may have an impact similar to or more active influence of aqueous extracts of the same plant on the bacterial strains tested.

Regarding to the inhibitory effect of *Mentha logifolia* leaves extraction on bacterial growth, The results of the study in Table (2&3) all the extracts of mental plant leaves from all other resistant bacterial strains under study only one bacterial strain *P. mirabilis* ATCC 43071, was observed with a weak inhibition (7-10 mm) for growth only under the influence of ethanol extract of mental leaves. This extract has an impaired ability to stop the growth of

bacterial strain mentioned and there is no effect of the other extracts (ethanol and Petroleum ether) for the growth of any strain of the bacterial strains studied.

This is consistent with the study carried out by **Abu Shanab et al. (2006)** using ethanol 80% and water extract of mental plant and its impact on the strain of bacteria, and only one is defined as the resistance to methylene, *Staphulococcus aureus*, where as much as the lowest concentration inhibiting bacterial growth (MIC) between 3.125 - 12.50 µg/ml.

Both the **Hajlaoui et al. (2008)** determined that with a different geographical location of the plant as part derived from it (leaves or stems) are different compounds present in plants as well as its opposite AEA was test the effect of extracts of leaves of mental using the method of spread of the disk, concluded that the extracts of mental leaves has the ability to inhibit bacterial activity of all bacterial strains tested, and this was consistent with the results of the current study, the inability of extracts of different mental leaf to inhibit bacterial growth of strains of bacteria tested for this study except the aqueous extract was a weak effect (7-10 mm).

The study also showed that the reward of all **Al-Bayati (2009)** who mentioned that a known compound was isolated from the essential oil of the plant and was identified as (-) menthol. The isolated compound was investigated for its antimicrobial activity against seven selected pathogenic and non-pathogenic microorganisms: *Staphylococcus aureus*, *Streptococcus mutans*, *Streptococcus faecalis*, *Streptococcus pyogenis*, *Lactobacillus acidophilus*, *Pseudomonas aeruginosa* and the yeast *Candida albicans*. Menthol at different concentrations (1:1, 1:5, 1:10, 1:20) was active against all tested bacteria except for *P. aeruginosa*, and the highest inhibitory effect was observed against *S. mutans* (zone of inhibition: 25.3 mm) using the disc diffusion method. Minimal inhibitory concentration MIC values ranged from 15.6–125.0 µg/ml, and the most promising results were observed against *S. aureus* and *S. mutans* (MIC 15.6 µg/ml) while, *S. faecalis*, *S. pyogenis* and *L. acidophilus* ranked next (MIC 31.2 µg/ml). Furthermore, menthol achieved considerable antifungal activity against the yeast *C. albicans* (zone of inhibition range: 7.1–18.5 mm; MIC: 125.0).

Rajashree et al. (2011) mentioned that the activity of aqueous and ethanol extracts and acetone to the mental leaf against the growth of some types of bacterial strains that cause human diseases, whether Gram positive or Gram negative,

(*Micrococcus luteus* ; *Staphylococcus aureus* ; *Salmonella typhi* ; *Klebsiella pneumoniae* ; *Escherichia coli* ; *Pseudomonas aeruginosa*) in a manner that the spread of the disk on the agar was to ethanolic extract effect inhibiting strong on all bacterial strains studied, and this is contrary to the results of the present study the ability of the ethanolic extract not to make a chilling effect, but the aqueous extract is the one who has had a inhibitory effect of bacterial growth of the bacterial strains tested, including *Staphylococcus aureus* ATCC 25923; *Klebsiella pneumoniae* ATCC 700603; *Pseudomonas aeruginosa* ATCC 27853; *Escherichia coli* ATCC 25922.

The present study coincided with the study conducted by **Gulluce et al. (2007)** to verify the effect of ethanol extract of mentha leaf against bacterial growth for a number of micro-organisms has been observed that the ethanol extract of mentha leaf has inhibitory effect forces.

Regarding the effect of *Citrullus colocynthis* fruits extraction inhibitory of bacterial growth, the results showed in Table (2&3) ethanol extract (70%) of the fruits of bitter melon has very strong inhibitory effect (20-30 mm) on the growth of bacterial strain, negative Gram *E. Coli* ATCC 25922), and a strong inhibitory effect (15-20 mm) on the growth of bacterial strain Gram positive, *K. pneumoniae* ATCC 700603). The present results confirmed by the results of studies conducted by **Gurudeeban et al. (2010)** who reported that the aqueous extract of the fruits of bitter melon has high activity against the growth of strains ; *E. coli* & *Staphylococcus aureus* and influence against the low growth of the bacterial strains *Bacillus subtilis*; *Klebsiella pneumoniae*. The impact of ethanol extract of the fruits of bitter melon has a strong anti-bacterial growth of strains of *Bacillus subtilis*, *Streptococcus pyogenes*, *Salmonella typhi* and the effect is very weak against the growth of bacterial strains *Streptococcus faecalis* *Proteus mirabilis* & *Proteus vulgaris*.

The results in Table (2&3) illustrate that the Effect of ethanol extract (70%) of the fruits of bitter melon was average inhibitory effect (10-15 mm) for the growth of bacterial strain, Gram positive *E. faecalis* ATCC 29212 and other Gram negative *P. mirabilis* ATCC 43071. The ethanol extract (70%) of the fruits of bitter melon is a weak inhibitor (10-15 mm) on the bacterial growth of the bacterial strains ; Gram positive *S. aureus* ATCC 25923 and four bacterial strains of Gram negative *P. mirabilis* ATCC 12153, *P. aeruginosa* ATCC 27853, *K. pneumoniae* ATCC 13883 & *S. typhi* ATCC 14028).

Resistance was observed for the bacterial strain *S. aureus* (MRSA) ATCC 33591 Gram positive a for, ethanol extract (70%) of the fruits of bitter melon. Studying the impact of ethanol extract of the fruits of bitter melon, inhibitory effect is weak (5-10 mm) for the growth of two strains of bacteria, Gram positive (*E. faecalis* ATCC 29212 & *S. aureus* ATCC 25923) and six Gram negative *P. mirabilis* ATCC 12153, *P. mirabilis* ATCC 43071, *S. typhi* ATCC 14028; *P. aeruginosa* ATCC 27853; *E. coli* ATCC 25922; *K. pneumoniae* ATCC 13883.

Paul (2008) studied the activity of three extracts of the fruits of bitter melon (ethanol and, petroleum ether) against the growth of seven bacterial strains (*Bacillus subtilis*; *Escherichia coli*; *Klebsiella pneumoniae*; *Proteus vulgaris*; *Pseudomonas aeruginosa*; *Salmonella typhi* and *Staphylococcus aureus*) using the method of spread of the disk and consistent with the results of the present study the ability of the ethanol extract to cause a active significant impact against bacterial strain (*Escherichia coli*; *Klebsiella pneumoniae*; *Pseudomonas aeruginosa*; *Salmonella typhi* and *Staphylococcus aureus*). It also agreed with the study of **Memon et al. (2003)** who reported that the capacity of ethanol extract of the fruits, leaves, stems, and roots of bitter melon on the inhibition of bacterial growth (Gram positive and Gram negative) *Bacillus pumilus*, *Staphylococcus aureus*. Where it was observed with a stronger effect to suppress bacterial growth using the ethanol extract of the fruit and the roots of bitter melon extract all of the bacterial strains

Regarding to effect of *Ricinus communis* seeds extraction inhibitory of bacterial growth, the results shown in Table (2&3) the effectiveness of aqueous extract of the castor plant seeds to inhibit the growth of different bacterial strains, but the impact is medium (10-15 mm) for the growth of bacterial strain and one Gram negative *K. pneumoniae* ATCC 700603 and the effect of aqueous extracts was weak inhibitor (5-10 mm) for the growth of six bacterial strains, two of which are positive for Gram *S. aureus* ATCC 33591 & *S. aureus* ATCC 25923 and four bacterial strains of Gram negative *P. mirabilis* ATCC 43071, *P. aeruginosa* ATCC 27853, *E. coli* ATCC 25922 & *K. pneumoniae* ATCC 13883. Search results also showed resistance to three strains of the bacterial aqueous extracts of seeds of castor plant bacterial strain and one Gram positive *E. faecalis* ATCC 29212 bacterial strains and Gram negative *P. mirabilis* ATCC 12153 & *S. typhi* ATCC 14028. The results also showed outlined in the table (2&3) Effect of ethanol extract (70%) of the seeds of

the castor plant, where the observed average effect (10-15 mm) to inhibit bacterial growth of the bacterial strain and one Gram positive *S. aureus* ATCC 25923 and three bacterial strains of Gram negative (*K. pneumoniae* ATCC 700603; *P. mirabilis* ATCC 43071 & *P. aeruginosa* ATCC 27853).

Ethanol extract of the seeds of castor has little effect (7-10 mm) for the growth of two strains of bacteria positive Gram & *S. aureus* (MRSA) ATCC 33591 ATCC 29212 *E. faecalis* and two strains of Gram negative *E. coli* ATCC 25922 & *K. pneumoniae* ATCC 13883 strains also showed resistance of Gram negative *P. mirabilis* ATCC 12153 & *S. typhi* ATCC 14028 of ethanolic extract (70%) of the seeds of the castor plant.

There is agreement between the study of **Jombo and Enenebeakup (2007)** who conducted the influence of water and ethanol extracts of plant seeds of the castor on different strains of bacteria for susceptibility testing using the agar diffusion method, appeared to the strains of bacterial *Klebsella Pneumoniae*; *E. coli*; *Proteus vulgaris*; *Staphylococcus aureus* sensitivity for each of the ethanol, methanol and water extract of plant the seeds of castor, this is consistent with the results of the present study the ability of the extract of castor oil seeds make an impact in the medium for the growth inhibitory bacterial strain *E. coli* ATCC 25922.

Regarding to effect of *Azadirachta indica* leave extraction for inhibitory effect of bacterial growth The results of effect of *Azadirachta indica* Leaves Extraction for Inhibitory bacterial growth showed in tables (2&3) the effectiveness of aqueous extract of *Azadirachta indica* leaves to inhibit bacterial growth is weak (7-10 mm) for the growth of seven bacterial strains, including strains of Gram positive *E. faecalis* ATCC 29212 & *S. aureus* ATCC 25923 and five bacterial strains of Gram negative (*P. mirabilis* ATCC 43071, *P. aeruginosa* ATCC 27853; *E. coli* ATCC 25922; *K. pneumoniae* ATCC 700603 & *S. typhi* ATCC 14028) The rest of the bacterial strains tested were recorded resistance of aqueous extracts of oleander leaves to the growth of bacterial strain of Gram positive (*S. aureus* (MRSA) ATCC 3359) and two strains of Gram negative *P. mirabilis* ATCC 12153 & *S. typhi* ATCC 14028). From the results of Table (2&3) observed that the extract of *Azadirachta indica* leaves ethanol (70%) a strong inhibitory effect (15-20 mm) for the growth of bacterial strain and one Gram negative *E. coli*

ATCC 25922 and the effect of inhibitor medium (10-15 mm) for the growth of bacterial strain and one Gram positive *E. faecalis* ATCC 29212 and influenced by a weak inhibitor (7-10 mm) for the growth of bacterial strain and one Gram negative *P. mirabilis* ATCC 43071 and with the use ethanol extract (70%) of *Azadirachta indica* leaves were observed resistance to two strains of Gram positive bacteria *S. aureus* (MRSA) ATCC 33591 & *S. aureus* ATCC 25923 and five bacterial strains of Gram negative *P. mirabilis* ATCC 12153; *P. aeruginosa* ATCC 27853; *K. pneumoniae* ATCC 700603; *K. pneumoniae* ATCC 13883 & *S. typhi* ATCC 14028. The effect of ethanol extract of leaves of *Azadirachta indica*.

The results in tables (2&3) explain that the ethanol extract of the leaves of *Azadirachta indica* strong influence (20-30 mm) for the growth of bacterial strain and one Gram negative *E. coli* ATCC 25922, and the effect is weak (7-10 mm) for the growth of two bacterial strains one positive for the dye Jtram *E. faecalis* ATCC 29212 and other Gram negative *P. mirabilis* ATCC 43071 and other strains of bacterial resistance recorded against ethanol extract of *Azadirachta indica* leaves a Gram positive strains of *S. aureus* (MRSA) ATCC 33591 & *S. aureus* ATCC 25923 and five bacterial strains of Gram negative (*P. mirabilis* ATCC 12153, *P. aeruginosa* ATCC 27853, *K. pneumoniae* ATCC 700603, *K. pneumoniae* ATCC 13883 & *S. typhi* ATCC 14028).

Results of the study showed that plant extracts of *Azadirachta indica* leaves (ethanol 70% and methanol) have a joint effect of an inhibitor of bacterial growth of three strains differed depending on the strength of the impact damper for their ability to resist bacterial strains of the different extracts. It is clear from these results that extracts of *Azadirachta indica* leaf water, ethanol (70%) and methanol extract have the ability to make a counter-influence to the growth of most bacterial strains tested with different forces of impact between the inhibitory effect of a strong inhibitor to the influence of a weak inhibitor. The results nearly agree and confirmed by **Siswomihardjo et al.,(2007)** who reported that the ethanolic extract of neem leaves and sticks had strong inhibitor activity to the growth of *streptococcus mutans*. Also **Vank et. al. (2001)** confirmed that mentioned that indigenous Neem *Azadirachta indica* has anti-bacterial growth for *streptococcus mutans* and *lactobacilli*.

Table 4: Showing the Minimum Inhibitory Concentration of Aqueous extract of plants used (micrograms/ml) (MIC)

Bacterial strain	Aqueous extract of plant							
	<i>Abutilon</i>	<i>Salavadora</i>	<i>Matricaria</i>	<i>Mentha</i>	<i>Citrullus</i>	<i>Ricinus</i>	<i>Azadirachta</i>	<i>Ocimum</i>

	<i>pannosum</i>	<i>persicalin</i>	<i>chamomilla</i>	<i>logifolia</i>	<i>colocynthis</i>	<i>Communis</i>	<i>indicas</i>	<i>basilicum</i>
<i>Staph.aureus</i> . ATCC 33591	80	100	60	30	90	70	60	50
<i>Prot.mirabilis</i> ATCC 43071	100	100	40	70	100	80	50	--
<i>E. coli</i> ATCC 25922	80	80	40	90	30	70	50	90
<i>K.pneumoniae</i> . ATCC 700603	80	--	80	80	50	40	40	80
<i>K.pneumoniae</i> . ATCC 13883	80	80	100	--	70	60	50	70
<i>S. typhi</i> . ATCC 14028.	--	--	--	50	80	--	60	--
<i>E. faeculis</i> . ATCC 29212	80	80	60	60	80	--	40	50
<i>P. aeruginosa</i> ATCC 27853.	40	80	60	40	70	--	40	40
<i>Staph. aureus</i> . ATCC 25923	--	70	60	30	90	40	60	30
<i>Prot. mirabilis</i> . ATCC 12153	100	--	40	70	50	90	30	30

Regarding to effect of basil leave extraction for inhibitory of bacterial growth, Basil now grows in many regions throughout the world, but it was first native to India, Asia and Africa The name "basil" is derived from the old Greek word *basilikohn*, which means "royal," reflecting that ancient culture's attitudes towards an herb that they held to be very noble and sacred. The tradition of reverence of basil has continued in other cultures. In India, basil was cherished as an icon of hospitality. Basil is an excellent source of vitamin K and a very good source of iron, calcium and vitamin A. In addition, basil is a good source of dietary fiber, manganese, magnesium, vitamin C and potassium.

From the results listed in table (2&3) we find that the plant extracts of the leaves of basil (distilled water - ethanol (70%) - methanol) affect as a common inhibitor for the growth of most bacterial strains studied. Where different force of impact between the inhibitory effect of a strong inhibitor to the influence of a weak inhibitor. Research studies on basil have shown unique health-protecting effects in two basic areas: basil's flavonoids and volatile oils. basil have shown DNA Protection Plus Anti-Bacterial Properties. The unique array of active constituents called *flavonoids* found in basil provide protection at the cellular level. *Orientin* and *vicenin* are two water-soluble flavonoids that have been of particular interest in basil, and in studies on human white blood cells; these components of basil protect cell structures as well as chromosomes from radiation and oxygen-based damage. In addition,

basil has been shown to provide protection against unwanted bacterial growth. These anti-bacterial properties of basil are not associated with its unique flavonoids, but instead with its volatile oils, which contain *estragole*, *linalool*, *cineole*, *eugenol*, *sabinene*, *myrcene*, and *limonene*. Laboratories studies show the effectiveness of basil in restricting growth of numerous bacteria, including: *Listeria monocytogenes*, *Staphylococcus aureus*, *Escherichia coli* O:157:H7, *Yersinia enterocolitica*, and *Pseudomonas aeruginosa* **Bagamboula et al., (2004)**.

Basil (*Ocimum basilicum* L.) is a popular culinary herb, and its essential oils have been used extensively for many years in food products, perfumery, and dental and oral products. Basil essential oils and their principal constituents were found to exhibit antimicrobial activity against a wide range of Gram-negative and Gram-positive bacteria, yeast, and mold. The study reviews primarily the topic of basil essential oils with regards to their chemical composition, their effect on microorganisms, the test methods for antimicrobial activity determination, and their possible future use in food preservation or as the active antimicrobial (**Orafidiya et al.2001 ; Suppakul et al. 2003**).

Regarding the determination of Minimum Inhibitory Concentration (MIC):The lowest concentration of plant extracts inhibiting the growth of bacterial strains Gram positive and negative and tested under study and after cultivation of a fixed volume (0.1 ml) of suspended bacterial (inoculums

10⁷ cfu / ml 0.5 McFarland standards) in a test tube containing different concentrations of aqueous extracts of medicinal plants under study table (4), the results nearly agree with the results obtained by **Abu Shanab et al. (2006); Al-Shmma et al. (2006), Hajlaoui et al. (2008) and Al-Bayati (2009).**

From present study we can concluded that all the plant used in investigation(*Abutilon pannosum*, *Salavadora persicalin*, *Matricaria chamomilla*, *Citrullus colocynthis*, *Ricinus Communis* *Azadirachta indica* and *Ocimum basilicum*) have inhibitory effect on bacterial growth of bacterial strains used *Staphylococcus aureus*.(MRSA) ATCC 33591, *Proteus Mirabilis* ATCC 43071, *Escherichia. Coli* ATCC 2592, *Klebsiella pneumoniae*. ATCC 700603, *Klebsiella pneumoniae*. ATCC 13883, *Salmonella typhi*. ATCC 14028, *Enterococcus faeculis*. ATCC 29212, *Pseudomonas aeruginosa*. ATCC 27853 *Staphylococcus aureus*. ATCC 25923, *Proteus mirabilis*. ATCC 12153, the effect differ between weak and strong for different strains and can be used as alternative to commercial chemical antibiotics.

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7/10/2012