

Antibacterial effect of watery crude plants extract and antibiotics sensitivity

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Abstract

The objective of the present study was to investigate antimicrobial activity a number of *Origanum* spp, *Slavia* spp and *Syzygium* spp extract raw dried on normal flora and pathogenic bacteria. The study was done in the collage of sciences in missan city . The isolates was *E. coli*, *Staphylococcus aureus* and pathogenic bacteria was *Klebsiella*. Pathogenic bacteria *Klebsiella* were isolated from patients sever from ulcer skin and identified by biochemical test and two types of bacteria as normal flora *E.coli* from stool sample and *Staphylococcus* from nose then plants were extracted and tested by disc diffusion methods also antibiotics was done. The *Staphylococcus* is sensitive to ciprofloxacin (CIP) (27mm) than amikacin (AK) (22mm) in diameter and resistant to anther antibiotics. *Klebsiella* resistant all of antibiotics except Amikacin (23mm) in diameter. The *Origanum* spp, *Saliva* spp have no effect on all normal flora and pathogen bacteria but *Syzygium* spp is more effective.

Key words: Antimicrobial agents, plant extracts, *Origanum* spp, *Slavia* spp , *Syzygium* spp, antimicrobial activity.

INTRODUCTION:

A wide variety of antibiotics are commonly used for the treatment of serious infections caused by bacteria [1].In recent years, multiple drug resistance has developed due to indiscriminate use of existing antimicrobial drugs in treatment of infection disease. Antimicrobial resistance is a threat to mankind because most of the infectious causing bacteria has become multidrug resistance [2]. Because of the concern about the side effects of conventional medicine , the use of natural products as an alternate to conventional treatment in healing and treatment of various disease has been on the rise in the last few decades [3] .

Bacteria have the genetic ability to transmit and acquire resistance to drugs used as therapeutic agent [7]. *Salvia* genus has 900 species throughout the world and 88 species in Turkey, are rather important in Turkish folk medicine. *Salvia officinalis* is in parks and gardens as ornamental plants used to treat chill, sniffles, abdominal pain and stomach disorders. Some aqueous extract of this genus especially *S. officinalis*, *S. viridis*, *S. multicaulis* are known that have antioxidant activity [6]. *Salvia heldreichiana* is an endemic plant is known that essential oil has antimicrobial activity on pathogen *Staphylococcus aureus*, *E. coli*, *Salmonella typhimurium* and *Pseudomonas aeruginosa*. Its aqueous extract hasn't been yet researched. We presented data on antimicrobial activity of herbal extract and essential oil of two endemic *Salvia heldreichiana*. Oregano plays a primary role among temperate culinary herbs in world trade [8]. Oregano is cultivated in European countries and is one of the most popular herbs in Mediterranean cooking. It is treated both as "whole" dried leaves and in ground form [9]. The leave and dried herb of oregano as well as its essential oil of oregano has been used traditionally for respiratory disease, indigestion, dental caries, rheumatoid arthritis and urinary tract disorders [10]. Carvacrol is a major active component of **Oregano** and has potential uses as a food preservative [11]. Antimicrobial resistance is a threat to mankind because most of the infection causing bacteria has become multi drugs resistant [12]. Children the elderly and those with weak immune system is not very vigorous as those of healthy adults [13]. Because of the concern about the side effect of conventional medicine treatment in healing and treatment of various disease has been the rise in the last few decades [14]. **Cloves (*Syzygium aromaticum*)** are the aromatic dried flower buds of a tree in the family Myrtaceae used in Indian Ayurveda medicine, Chinese medicine [15-16]. Cloves are western herbalism. It is also used in dentistry where the essential oil of clove is used as anodyne for dental emergencies [17],[18]. ***Syzygium*** species have been reported to possess antibacterial and anti-inflammatory activity [19].

The aim of study was effects of plant extract on normal flora and pathogenic bacteria and compared with antibiotics sensitivity.

MATERIAL AND METHODS:

Plants crude extracts preparations:

Preparation of extract of *Origanum vulgare*: *Origanum vulgare* leaves were purchased from the local market of Missan city in Iraq and grounded to a powder then kept in dry container. The type of extract were prepared in the present study is watery based .Extracts was prepared by mixing 20gm of *Origanum* powder with 200ml distilled water in flask and left for 24 hours at room temperature and filtered by filter paper (Whatman No.1).The filtrate infusion was dried [4].

Preparation of infusion:

The aqueous infusion was prepared by taking 10gm clove in 100ml distilled water and left for 24 hours at room temperature with occasional shacking and filtered to obtain clear infusion[4].

Preparation of decoction: The aqueous decoction was prepared by boiling 10gm clove in 100ml distilled water in a flask for 20 minutes. The flask was removed from heat and allowed to cool. The content of flask was filtered to obtain clear decoction[4].

Preparation of infusion (*Salvia*):

The dried plant samples were first ground to fine powder. For aqueous extraction 1gm of the fine powder was extracted with 10ml of distal water at 100 °C for 30min in a water bath. The sample were then cooled down to room temperature and sterilization by filtration 0.22Mm Millipore filter. Final concentration of 50mg/ml were stored at -20°C and used for antimicrobial test [4].

Antibacterial testing:

Antibacterial activity test was carried out using the agar disc diffusion method. Muller Hinton Agar was prepared and poured in to the sterile petri plate, allowed to solidify. Organisms was swabbed on the surface of the M.H. agar and put medium plant extract disc after 10min and use filter paper punched 6mm by cork borer, each filter disc was filled with stock solution and dilution solution 10^{-1} , 10^{-2} . The inoculated agar plates were incubated at 37°C for 24 hours. After incubation period the diameter of inhibition zone is the area surrounding the filter disc is positive result and there is no growth of the inoculated microorganism. Ciprofloxacin was used as control to check its inhibitory activity against the test organism.

RESULTS AND DISCUSSION

Antimicrobial plant products have gained a special attention because of increase resistance to antibiotic acquired of some microorganisms [5]. In table (1) show the **Staphylococcus** is sensitive to ciprofloxacin (CIP) (27mm) than amikacin (AK) (22mm) in diameter and resistant to another antibiotics. **Klebsiella** resistant all of antibiotics except Amikacin (23mm) in diameter show in table [3]. **E. coli** was sensitive to Chloramphenicol (19mm), Ciprofloxacin (20mm) and Amikacin (20mm) and resistant to another antibiotics in table [5]. No value of antibacterial activity to **Origanum spp** and **Salvia spp** on normal flora (**E.coli** and **Staphylococcus**) and pathogenic bacteria **Klebsiella spp**. The aqueous infusion of clove exhibited maximum activity against **Staphylococcus aureus** was (12mm) in diameter zone of inhibition and use a ciprofloxacin as control and give (14mm) in diameter in table [2]. **Klebsiella** (10mm) in diameter and ciprofloxacin as control (16mm) in table (4). In case of **E. coli** (13mm) in diameter and use ciprofloxacin as control was (20mm) in table (6).

Table (1) .The value of resistance of **Staphylococcus aureus** to antimicrobial agents.

Type of antibiotics	Concentration (mcg/g)	Type of resistance	Diameter of inhibition zone
Ciprofloxacin CIP	5	S	27 mm
Ampicillin AM	10	R	
Chloramphenicol C	30	R	7 mm
Gentamicin GM	10	R	10 mm
Lincomycin L	10	R	
Amikacin AK	30	S	22 mm
Tetracycline TE	30	R	-----
Penicillin P	10	R	-----
Novobiocin NV	30	R	-----

Table (2) .The value of effect plant watery extract on **Staphylococcus aureus**.

Plant watery extract	I.Z (Stock solution)	I.Z (Dilution 10 ⁻¹)	I.Z (Dilution 10 ⁻²)
Salvia spp	-----	-----	-----
Origanum spp	-----	-----	-----
Ciprofloxacin CIP	S (25 mm)		
Syzygium spp	(12 mm)	-----	-----
Ciprofloxacin CIP	(14 mm)		

CIP: as a control. I.Z: inhibition zone.

Table (3). The value of resistance **Klebsiella** to antimicrobial agents.

Type of antibiotics	Concentration (mcg/g)	Type of resistance	Diameter of inhibition zone
Ciprofloxacin CIP	5	R	-----
Lincomycin L	10	R	-----
Chloramphenicol C	30	R	-----
Amikacin AK	30	S	23 mm
Penicillin P	10	R	-----
Tetracyclin TE	30	R	-----
Novobiocin NV	30	R	-----

Plant watery extract	I.Z (Stock solution)	I.Z (Dilution 10^{-1})	I.Z (Dilution 10^{-2})
Salvia spp	-----	-----	-----
Origanum spp	-----	-----	-----
Ciprofloxacin CIP	S (25 mm)		
Syzygium spp	10 mm	-----	-----
Ciprofloxacin CIP	16 mm		

Table (4) .The value of effect plant watery extract on **Klebsiella**

Table (5) .The value of resistance of **Escherichia coli** to antimicrobial agents

Type of antibiotics	Concentration (mcg/g)	Type of resistance	Diameter of inhibition zone
Chloramphenicol C	30	S	9 mm
Cefotaxime CTX	30	R	----
Penicillin P	10	R	----
Novobiocin NV	30	R	----
Ampicillin AM	10	R	----
Ciprofloxacin CIP	5	S	20 mm
Lincomycin L	10	R	----
Amikacin AK	30	S	20 mm
Tetracycline TE	30	R	----
Gentamicin GM	10	R	----

S = (Sensitive)

Table (6) .The value of effect plant watery extract on **Escherichia coli**.

Plant watery extract	I.Z (Stock solution)	I.Z (Dilution 10^{-1})	I.Z (Dilution 10^{-2})
Salvia spp	-----	-----	-----
Origanum spp	-----	-----	-----
Ciprofloxacin CIP	S (40 mm)		
Syzygium spp	10 mm	-----	-----

Ciprofloxacin CIP	20 mm		
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Present study exhibited the medical importance of three plant extracts (**Origanum spp**, **Salvia spp** and **Syzygium spp**). Through the existence of antimicrobial activity in the crude, diluted extracts of these plant extract. Microorganisms show a variable resistant to the action of oregano extracts. It was observed that **E. coli**, **Staphylococcus aureus** as normal flora and **Klebsiella** as pathogenic bacteria where resistant to crude and diluted extract this study is disagree with [20]. He founded the diameter of the inhibition zone to **Staphylococcus aureus** was (18mm) and no inhibition zone with **E. coli** and **Klebsiella spp**. This study is agree with [21] that show no inhibition zone. In this study no antimicrobial activity salvia to **E. coli**, **Staphylococcus** and **Klebsiella**. This study disagree with [22] that show diameter inhibition zone in **Staphylococcus aureus** is (13mm) and **E. coli** was (19mm) in diameter. The result of antibacterial effect of clove watery extract against **Staphylococcus aureus**, **E. coli** and **Klebsiella spp** and agree with [21]. Watery extract showed maximum zone of inhibition zone (15mm) against **Staphylococcus aureus** while minimum was (10mm) against **Klebsiella spp**.

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