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STUDY OF THE EFFECT OF THE PLANT EXTRACT OF *PROSOPIS FARCTA* ON THE GRAM NEGATIVE AND GRAM POSITIVE BACTERIA, ISOLATED FROM DIFFERENT INFECTIONS

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Abstract

The effect of *Prosopis farcta* extract was studied on positive and negative bacterial species of gram that isolated from different infections. The effectiveness of active isolates from the leaves of *P. farcta* was tested for the growth of four types of positive and negative gram in Agar-well Diffusion test, these isolates were obtained from the Samarra General Hospital in Salah al-Din Governorate. These isolates included Gram positive bacterial strains Staphylococcus. *aurous*, while the Gram negative bacteria, used in this study, are *Escherichia coli*, *Pseudomonous aeruginosa*, *Salmonella typhi* and *Brucella militensis*. These isolates were first diagnosed and tested according to their phenotypic properties. Using a number of biochemical tests, the results showed that all active substances isolated and diagnosed had an inhibitory effect on the growth of these bacterial strains and discrete diameters vary according to the active substances and their concentrations.

Key words : *Prosopis farcta,* gram negative and gram positive, phenotypic properties, discrete diameters.

Introduction

Oxidation of lipids that is the main reason behind quality deterioration in several food systems, could result in off-flavors and formation of deadly compounds, and will lower the standard and organic process worth of foods. What is more, lipid oxidization is additionally related to aging, membrane harm, cardiovascular disease and cancer (Ramarathnam *et al*., 1995).

Antioxidants can be defined as compounds that inhibit or delay the oxidation of other molecules by inhibiting the initiation or propagation of oxidizing chain reactions (Velioglu *et al*., 1998). The most commonly used antioxidants at the present time are butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), propyl gallate (PG), and tert-butylhydroquinone (TBHQ) (Sherwin, 1990). However, they are suspected of being responsible for liver damage and carcinogenesis in laboratory animals (Grice, 1986; Wichi, 1988). Therefore, there is a growing interest on natural and safer antioxidants (Moure *et al*., 2001; Gülçin, 2006; Oktay *et al*., 2003) Many inhibitor compounds, present in plant sources, are known as free radicals or active oxygen scavengers. Recently, interest has significantly increased find present antioxidants to be used in food or healthful materials to switch artificial antioxidants, that square measure being restricted because of their aspect effects like carcinogenicity. Natural antioxidants will defend the physical body from free radicals and retard the progress of the many chronic unwellness additionally as lipoid aerophilic rancidity in foods (Gülçin *et al*., 2004). most of the plants of the thorns belonging to the *P. farcta* species belonging to the leguminosae family have medicinal value, since the water and alcohol extract of the fresh leaves of *P. farcta* has a microbial effect against *Staphylococcus aureus*, *E. coli*. Vinaline and alkaloids have been isolated and contain a toxic substance in the cells of this plant and the teramine. It has been shown to be used as a preservative and medicine to treat scorpion and open wounds and to treat rheumatism. As for the importance of medicinal plants at present, the plant (*Prosopis farcta*) has been selected for its medicinal

and nutritional importance. The fruit of the plant of *P. parcta* contains a type of Gum composed of units of Manose and Galactose (Qutb, 1981). (*Prosopis* spp.) on prosopinine, prosopine and the active ingredients of the plant thorns are alkaloids in general, which vary in different types of thorns, (Gronsnevor, 1995).

Materials and Methods

Collection of Specimens

Our samples were collected, from patient in Samarra general hospital during the period from October month to December month of the year 2016.

Bacterial Isolates: samples were planted on the following media.

1- Blood agar

2 - MacConkey's agar

3-Nutrient agar

4-Mannitol Salt Agar to diagnose bacteria *S.aureus*.

All Medias attended and dissolved in distilled water and then sterilized by autoclave at a temperature 121 C^0 and pressure 15 pound for 15 minutes, the dishes were incubated aerobically and at a temperature 37 C^0 for 24 hour.

Diagnosis of bacterial isolates

The morphological and chemical properties of developing colonies were observed.

A- Microscopy and agricultural characteristics

The bacteria were first identified by observation agricultural characteristics of the developing colonies on the media used form where size, height and colonial color and attended thin swabs and a pigment with a Gram stain to observe cell shapes, arrange and their susceptibility to pigmentation (Collee *et al.*, 1996; Manual of WHO, 2003).

B- Bacteriological test

IMVIC test were conducted includes (Indol test, Methyl red, Vog's proskauer, Simmon citrate (Collee *et al.*, 1996; Manual of WHO, 2003) As well as tests Oxidase, Catalase, Coagulase are used to confirm the isolated bacterial species (Koneman *et al.*, 1997; Harley and Prescott, 2002).

Collection and extraction of *Prosopis farcta*

The aerial parts (leaves and flowers) were collected during the period 2015-2016 from Al-Dulueaya town which is located in region 80 km north-east Baghdad, Iraq. The plant material was identified by Dr. Ibraheem Omar, department of Biology, College of Science, University of Tikrit, Salah Uddin, Iraq. The fresh aerial parts were washed thoroughly with tap water at room temperature to remove dirt prior to the drying process. These washed aerial parts washed again by a distil water and then dried in the shade at room temperature for seven days. Then, they were crushed into final powder one hundred grams of DPP were extracted with 200ml diethyl ether using soxhlet apparatus for three hours to remove fatty contents. The defatted plant materials was dried at 35°C in an air oven, and then extracted twice with 250ml (70%) ethanol at 90°C for 2 hours. The solution was filtered and centrifugation at 3000 rpm for 15 minutes. The solvent was evaporated and the aqueous extract was condensed under reduced pressure. The extract was weight, labeled and protected from light in a refrigerator at 4°C in a glass container until use (Rafah Alsamarrai *et al*., 2006).

Isolation and Identification of Bacterial species that isolated from variety infections

Some bacterial species isolated from different infections were diagnosed based on morphological, cultivation, and biochemical tests. Isolated *E. coli* appeared as a short chromium-negative sticks, they also showed ability to ferment lactose in the Maconkey medium, in circular colonies, Convex with full edge and pink color. While on Eosin Methylene Blue Agar they appeared as a black colonies emerged with metallic green brilliant. It was a producer of catalase and indole enzyme since it had the ability to split tryptophan. Whereas *Staphylococcus aureus* appeared as a Spherical shape and positive for Gram stain. Also appeared Fermented with lactose on the Maconkey medium. They appeared in the form of colonies resembling grape clusters, didn't grown on Eosin Methylene Blue Agar and can produce catalase and indole enzyme due to the ability of this bacteria to split tryptophan. *S. typhi* bacteria were negative for Gram stain, negative for uric acid test and positive for red methyl as shown in table1. While *B. melitensis* this is Gram negative bacteria, aerobic, and has ability to movement although it doesn't has a fragelle or Capsules (Mahon *et al.*, 2015).

Results and Discussion

The similar characters indicate that there are no significant differences between the groups (probability ± 0.05), while the different letters indicate significant differences between the groups (probability ± 0.05). The capital letters refer to the vertical comparison, whereas the small letters refer to the horizontal comparison.

Table 2 shows that the concentration of *Prosopis farcta* was more effective on *Staphylococcus aureus*

Table 1 : Tests used to diagnosis our bacterial species.

العزلات البكتيرية	Gram Stain	Urase Test	Oxidase Test	Indol Test	Red Methyle	MR-VP	Lactose fermentation	Hemolysis
E.coli	-	+	-	+	+	-	+	+
S.aureus	+	+	-	-	+	+	-	+
P.aeroginosa	_	+	+	-	+	+	-	-
B.melitensis	-	+	_	_	+	+	_	+
S.typhi	_	_	_	_	+	_	-	_

Table 2 : Inhibitor diameter is measured in ml for bacterial isolates.

concentration mg/ml	Inhibitor diameter is measured in ml for bacterial isolates				
	E. coli	***P. aeruginosa***	***S. typhi***	***S. aureus***	***B. melitensis***
100	12.00±1.00 Ab	9.33±1.52 Bc	12.66±1.52Ab	15.33±1.52Aa	0.00Ad
75	0.00Bc	0.00Cc	9.00±1.00Bb	15.66±1.52Aa	0.00Ac
50	0.00Bd	10.00±1.00Ab	8.33±1.52Bc	11.00±1.00Ba	0.00Ad

significant differences were observed between them and other bacteria, it was also observed that there was no effect of the *Prosopis farcta* extract on bacteria *B. melitensis* in the same concentration, the remaining concentrations (50 and 75) were similar in their effect to the concentration (100) on the bacterial species under study, concentrations (50 and 100) were more effective on bacteria *S. aureus* as compared to other bacteria as well as not affecting the species *B. melitensis*. When comparing the different concentrations of each type of bacteria we find there are significant differences between concentrations (100) of *Prosopis farcta* extract with other concentrations for *S. aureus, S. typhi* and *E. coli*. While no significant differences were observed between concentration (100) and the other concentrations for *B. melitensis*. Concentrations (50 and 100) were more effective than the concentration of 100 on S. typhi bacteria.

The results showed that the plant extract had an anti-microbial effect on the growth of all bacterial species except bacterial (*Brucella melitensis*) which has no positive result towards the plant extract under study and as shown in Fig. 1, Gram-negative bacteria were found to be most affected by the plant extract. The Inhibition diameters of E. coli was (12, 0, 0 mm) at concentrations (100, 75, 50 mg/ml) respectively as shown in fig. 2 and this result was correspond to (Boukhbaty 2010) which found that the rate of inhibition of this bacteria (13, 10, 8 mm) But they differ from what (Al-Daheri *et al.*, 2010) has been reached, Which found that the rate of inhibition

Fig. 1: Inhibitor diameter of B. melitensis

Fig. 2: Inhibitor diameter of *E. coli*

Fig. 3: Inhibitor diameter of *P. aeruginosa*

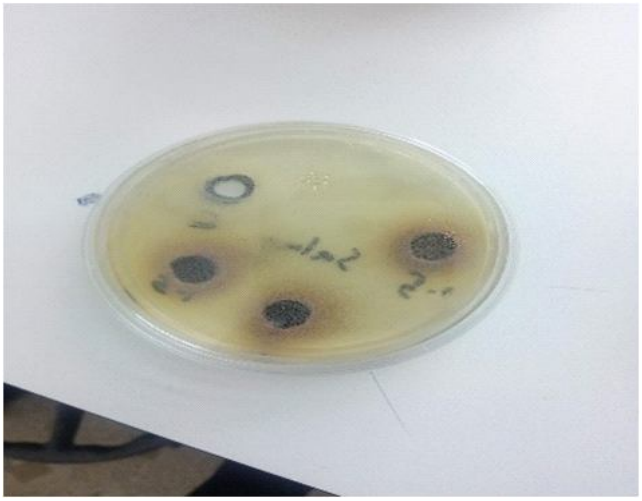

Fig. 4: Inhibitor diameter of *S. typhi*

of these bacteria respectively (0, 8.0), This is due to reasons related to the bacterial strains used and the different environments isolated from the breeds or plant-related causes, such as the different environmental conditions that directly affect the plant content of active compounds (Aggett 1994). While the rate of inhibition for *P. eroginosa* bacteria as shown in fig. 3 (0, 0, 11 mm) at concentrations (50, 75, and 100%) respectively, the ability of *P.farcta* seeds to inhibit bacterial strains because they contain some phenolic compounds such as thymol, which has a good effect against bacteria and fungi, has a direct effect on bacterial cell walls by dissolving the bacterial cell wall fat and forming the hydrogen atom with water molecules and nitrogen amino acids inside the cell, also have antimicrobial efficacy by disrupting the cytoplasmic membrane mechanism of microorganisms, thus inhibition of microorganism growth, as well as containing some mono-cytoplasmic terpenoid compounds, which interfere with membranes, mainly

Fig. 5: Inhibitor diameter of *S. aureus*

affect the suspension of the effective transport mechanism (Da Porto and Decorti, 2009; Bakkali *et al.*, 2008). While *Salmonella typhi* bacteria as shown in fig. 4 has a diameter of inhibition (14, 10, 8 mm) at concentrations of (100, 75, 50%) respectively. The results also showed a positive inhibitory activity of the plant extract against Gram positive bacterial strains, which include *Staphylococcus aureus* bacteria as shown in fig. 5.

Our selection of the studied species was not coincidental but was one of the most famous species in our region and in the temperate regions, they are also known for hybrids, as plant species are known to differ morphologically, biologically, in chemical components, depending on the plant environment in which they are present, climatic conditions and other factors affecting the plant.

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