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Evaluation inhibition activity for radish seed extract on bacteria inhibition which isolated for Urinary Tract Infections in Al-Alam city/Tikrit

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ABSTRACT
This study was conducted to isolation and identify of bacterial isolated from urinary tract infection, in the study was collected 190 a simple of patients with UTI and isolated the Gram negative bacteria for different ages and races, Diagnosed these bacteria by Biochemical tests biochemical by using API 20E system to confirm the tests.
The *Escherichia .coli* isolates had the highest rate of isolation. The isolated were tested to determine their resistance to antibiotics Penicillin, Amoxycillin, Cefriaxxone, Cefotaxime, Gentamicin, Amikacin, Ciprofloxacin, Azithromycin and Doxycycline. The isolates showed high resistances to most antibiotics, the resistance of *E.coli* to the antibiotics was P, CTX in the rate of 100%, 93.3% respectively. The effect of aqueous and alcohol extracts of *Radish* seeds on isolates has been studies. The effect of alcohol extract was more effective on the isolates than the aqueous extract.
The highest concentration was inhibition of the alcohol extract at the concentration of 100% in the *Proteus* bacteria where it was 23 mm and the lowest inhibition diameter in *Klebsiella* bacteria where it was 8mm, the aqueous extract was highest concentration in the *Proteus* where it was 16mm and lowest effects in *Klebsiella* bacteria where the diameter was 7 mm. The higher the concentration of the extract the greater the diameter of the inhibition.

1- Introduction
UTIs is one of most common diseases, followed by diseases of the respiratory system, which leads to the death of many people infected with it [1]. The number of people infected by it reaches millions, including males and females if all ages[2]. Women are more likely to get the diseases than men[3].
The important increased in study of Gram negative bacteria not only because of the fact that has ability to configure the diseases but also because of increasing the ability to resist antibiotics, Which is of health problems scattered in the world[4]. The most prevalent causes of diseases is Gram negative bacteria including *E.coli* which is the cause of the main the diseases , some of them lactose fermented bacteria and some are lactose not fermented bacteria[5].
They found that the largest living organisms that cause this disease are bacteria, including the negative , Enterobacteraceae and Speudomonaceae and the positive bacteria , including the *Staphylococcus* spp and Spongy[6]. Treatment with plants and medicinal herbs is one of the method used throughout the ages in treating various diseases. Many plants have been used for treatment by civilizations, including Indian and Chinese. The *redish* plant has been studied and its effect on the growth of negative bacteria has been used in its seeds. The aim of the research:
Isolation and diagnosis of Gram negative bacteria isolated from UTI-
-Know how effect resistant bacteria are to antibiotics.
-Test the effect of plant extracts on the growth of bacteria

2- Material and Methods
This study were collected 190 samples of Gram negative bacteria from patients infected with urinary tract during the period from of 18\11\2017 to 17\2\2018. Was diagnosed bacteria after their development at the medium blood agar that contain the 5% of the blood of the human in petri dish's by used loop and medium macConkey and EMB sterile autoclave degree 121 degree Celsius and for 15 minutes. After bacterial growth, we isolated it and

conduct the Biochemical test of IMViC and the test of oxidase and catalase to diagnose it as well as the work of chromium dye to determine its shape and methods of collection and thus depends on the official characteristics and biochemical tests of the diagnosis [7]. Also use the system API 20E make sure of the diagnosis[8].

2-1 Antibiotic

These antibodies are used as a treatment for humans and animals and have several mechanisms that work within the body. Some of them work on the wall of the cell, some stop the formation of the protein and some make DNA or RNA, and there are many antibiotics that have treated many diseases caused by bacteria, Some antibiotics have been used to determine the resistance of different bacteria to these antibiotics: Amoxicillin (AM), Penicillin (P), Cefotaxime(CTX), Ceftriaxone (CTR), Ciprofloxacin (CIP), Amikacin (AK), Doxycycline (DO), Azithromycin (AZM), Gentamycin (GM).

2-2 Medium antibiotics

The medium of the muller-Hinton is mentioned on the packaging and sterilize and cool to 45-50 C. Bacterial suspension is prepared and spread on the surface of the medium by swab antibiotic tablets are placed and incubated for 24 hours and 37C. Then reading the inhibition diameter and compare the Results with the standard diameters of the inhibition of antibiotic as stated in[9].

2-3 Plant extract preparation

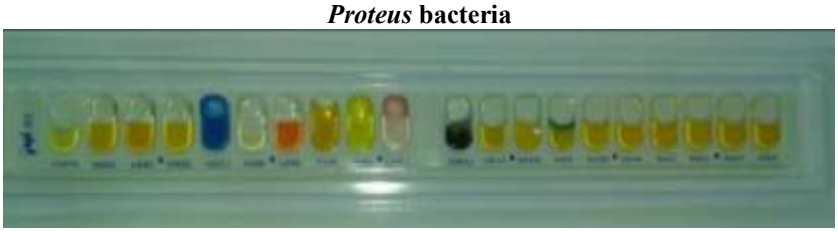
Radish seeds were used to determine their effect on the bacteria, and the water aqueous and alcohol extract of the plant was mixed with known quantities of seeds, aqueous or alcohol and incubated in a humidifier for 24 hours and at 37 degrees Celsius and then filtered with gauze cut and placed in the centrifuge cycle 3000/5000 cycles/minute, The active substances were not through PHLC technique the active substances in it are the Raphanin and Raphaiol[10].

3- Results and Discussion

Collect samples Gram negative bacteria isolated from patients UTIs, The most isolated of bacteria are *E.coli,* These isolates were tests laboratory and biochemical tests and using the API 20E system for confirmation by method [8]. Figure (1) shows the diagnosis of bacteria by the API 20E system.

***E.coli* bacteria**

***Proteus* bacteria**



***Klebsiella* bacteria**

Figure (1) Diagnosis of bacteria by API 20E system

3-1 Antibiotic resistance

Some bacteria show antibiotic resistance because they possess certain defensive means such as biofilm and capsule, etc. In our stud, we took some antibiotics CTX, CTR, P, AM, AK, AZM, GM, CIP, DO. The table(1) shows percentages of bacteria resistance to antibiotics.

Table (1) show result effect antibiotic on the gram negative bacteria

CTR	AK	AM	DO	CIP	GM	P	CTX	AZM	No isolated	Anti / Isolated
no (%)	no (%)	no (%)	no (%)	no (%)	no (%)	no (%)	no (%)	No (%)		
12 80%	8 %53.3	13 86.6%	14 93.3%	15 100%	13 86.6%	14 93.3%	15 %100	10 66.6%	15	***E.coli***
0 0%	2 25%	6 75%	3 37.5%	2 25%	6 75%	7 87.5%	6 75%	4 50%	8	***Klebsiella***
0 0%	1 50%	2 100%	2 100%	2 100%	1 50%	2 100%	0 0%	2 100%	2	***Proteus***

Azithromycin (AZM) Gentamicin (GM) Penicillin (P) Amikacin (AK) Ciprofloxacin (CIP) Doxycycline (DO) Amoxycilin (A) Cefotaxime (CTX) Ceftriaxone (CTR)

Bacterial isolated showed variation significantly in resistances of antibiotics which used in table (1). As it was a higher proportion of resistances to bacteria *E.coli* was antibiotic CTX, P it 100% and 93.3% respectively, where this ratio of resistance is not consistent with [11, 12]. As for bacteria *klebsiella* the highest resistance was in the antibiotic a rate of P 87.5% and CTX a rate of 75%. This result is consistent with [13]. As for bacteria *proteus* was a higher proportion of resistance in antibiotic P and AM is rate of 100% to both, the resistance ratio of AM was consistent with[12].

3-2 Effect of plant extract

The results of the current study showed that the alcoholic extract of radish seeds it has greater effectiveness than the aqueous extract of radish seeds in inhibiting the growth of bacteria. The cause of this deadly effect is that the active compounds in the plant are dissolved in alcohol which spread in the ocean and inhibit living cells[14].

Table (2) Shows that the highest diameter inhibition of the alcohol extract in the *proteus* bacteria at the concentration of 100% and the diameter of 23 mm and less diameter was in bacteria *E.coli* where it reached 14 mm at the concentration of 100%, In the aqueous extract of radish seeds aqueous was the highest inhibition at the concentration of 100% in the *Proteus*, where the diameter of 16 mm and less diameter of inhibition of 10 mm in the bacteria *Klebsiella*, our results are consistent with the findings[15]. Effect of radish seeds inhibition microbiology growth belong to have a raphanin in which have effect anti bacteria , result agree with[10]. They were found effect alcohol extract radish seed anti bacteria. Also agree with [15] indicate role alcohol extract radish seeds anti *E.coli*, *Pseudomonas* and *Klebsiella*. Figure (2) shows the effect of radish extract on bacteria.

Table (2) The inhibition zone % alcohol and aqueous extract against bacteria (mm).

Extract / Bacteria	Aqueous Concentration (%)			Alcohol Concentration (%)		
	25	50	100	25	50	100
E.coli	8	10	12	11	12	14
Klebsiella	7	8	10	8	12	15
Proteus	10	14	16	19	21	23

effect of radish extract aqueous on *E.coli* - effect of radish extract alcohol on *Proteus*

Figure (2) Effect of radish seeds extract on bacteria

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تقييم الفعالية التثبيطية لمستخلص بذور الفجل على نمو البكتريا المعزولة من مرضى المسالك البولية في مدينة العلم/ تكريت

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الملخص

تم جمع 190 عينه من البكتريا السالبة لصبغة كرام والمعزولة من مرضى المسالك البولية بمختلف الاعمار والاجناس، شخصت هذه البكتريا عن طريق الاختبارات الكيميوحيوية وباستخدام نظام API 20E التشخيصي لتأكيد الاختبارات، كانت بكتريا *E.coli* اعلى نسبة عزل والمسببة لهذه الاصابة، والنساء اكثر اصابة بهذا المرض من الرجال .

تم اختبار العزلات لتحديد مقاومتها للمضادات الحيوية ومن المضادات المستخدمة هي Penicillin(P), Cefotaxime(CTX), Amoxycillin (AM), Ceftriaxone (CTR), Gentamicin (GM), Doxymycin (DO), Amikacin(AK), Ciprofloxzcin (CIP), Azithromycin (AZM)، هذه العزلات اظهرت مقاومة عالية تجاه المضادات، فمقاومة *E.coli* للمضادات كانت اعلى في P, CTX وبنسبة 100%، 93.3% على التوالي. اما مقاومة *Klebsiella* فكانت اعلى عند مضاد P وبنسبة 87.5 % ومضاد CTX وبنسبة 75%، اما عزلات *Proteus* فكانت اعلى نسبة مقاومة تجاه مضادي P, AM وبنسبة 100%.

درس تأثير المستخلص المائي والكحولي لبذور الفجل وتأثيره على نمو البكتريا، تأثير المستخلص الكحولي اكثر فعالية على العزلات من المستخلص المائي، اعلى قطر للتثبيط في المستخلص الكحولي عند التركيز 100% في بكتريا *Proteus* حيث بلغ 23 mm واقل قطر تثبيط كان 8mm في *Klebsiella* ، واعلى قطر تثبيط للمستخلص المائي كان 16mm في *Proteus* واقل قطر تثبيط في المستخلص المائي عند تركيز 25% في بكتريا *Klebsiella* حيث بلغ قطر التثبيط 7mm، كلما زاد تركيز المستخلص ازداد قطر التثبيط.