Isolation and Identification and Antibiotics resistance of some Pathogenic bacteria from ants

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Abstract

This study was carried out on 60 ants samples collected from different places included: Gardens House, Food, died Cockroaches and W.C. Two methods were used: Isolation of bacteria from external surface and internal component, that to show the importance of this insect and their role in transferring of pathogenic bacteria.

The results showed isolation of 30 isolates of different bacterial genus at 50%, included *Staphylococcus aureus* at highest ratio 26.23% then *Escherichia coli*, *Bacillus subtilis*, *Klebsiella spp.*, *Proteus spp.*, *Pseudomonas areuginosa* and *Sarcinia* at (23.34, 13.34, 13.34, 10, 6.67, 6.67)% respectively. The study revealed that the ants ply an important role in transferring these microbes. The majority of that isolates were on their external surface 63.33% in comparison to that isolated from internal component.

The antibiotic sensitivity test for isolates against (7) antibiotics were resistance to multi antibiotics specially Vancomycin, whilst most isolates were sensitive to Nitrofurantin.

Introduction

Ants are often a serious problem in hospitals, restaurants and houses, since they may be found in large number difficult to control. The occurrence and abundance of ants are frequently high because of the strict characteristics of the buildings, such as easy access to the outside environments, and proximity the inadequate management practices are also important factors. Improper storage of medication in people and presence of food aggravate the ant problem(1). Ants colonize hospital equipment, invade sense as intensive care units and become a nuisance to staff and patients. Also visitors to patients could play a role in transportation of ants inside hospitals facilities(2).

Many ant species that display synanthropic behavior have spread successfully in urban areas(3). Ants can act as mechanical vectors, transporting

bacteria, including those resistant antimicrobials, and creating pathway of resistance dispersion in the external environments(4). Bacteria that can be transported include Gram negative bacilli, facultative anaerobic bacteria, with high counts indicating contamination, inadequate cleaning and sanitization, ineffective heat treatments, or multiplication processing or storage(5).

In this study we investigated the presence of different bacterial species on ants collected at the different places, with the purpose of evaluating the contaminating potential of the different genera and evaluate their resistance level to antibiotics.

Material and Methods

1. Ants collection:

Sixty samples of ants were collected from different places in Baghdad city included: Gardens, House room, died Cockroaches, Food and W.C. Each samples individual ant placed in screw vials containing 1g of sugar to keep the ants live until arrive to the laboratory. The vials marked with the collection date and location.

2. Ants Classification.

Ants classify according to (6)

3. Isolation and identification of bacteria from ants.

3-1 Isolation from external surface.

Each sample were washed with 1 ml of normal saline (Nacl. 0.85%) were added to each vial. Shacked gently to prepare suspension. Then 0.1ml of the suspension were cultured in nutrient broth (nutrient broth were prepared as manufacturing Oxoid). Shaking the medium for 5 min then incubated at 37 C° for 24hr. For demonstration of colonies, 0.1ml of broth culture was plated onto Nutrient ager (Oxoid company) and incubated for 24-48hr at 37C°. After appearance of colonies, one colony was transferred to the same medium of isolation for purification. The purified strains were identified according to the conventional bacterial colony, Gram stain and biochemical test (7,8).

3-2: Isolation from internal ants component.

As mention previously in (3-1), each ant sample after washing with 70% a alcohol was transferred to vial containing 1 ml of normal saline and crushed using sterilized glass rod to prepare ant suspension. Then the last suspension was treated as in (3-1) for isolation and identification of bacteria.

4-Antibiotic sensitivity test.

The antibiotic sensitivity test for isolated bacteria was carried out by means of the disc diffusion technique as described by(9).

Result and Discussion

Table (1) shows the types of microorganisms isolated from ants. A total of 30 isolates were obtained, from 60 individual ants sample (50%). The isolates

include 8 *S. aureus* 26.23%, 7 of *E. coli* 23.34%, followed by 4 (13.34)% of *Bacillus subtilis* and 4 (13.34%) of *Klebsiella spp.* and 3(10%) of *Proteus spp.* and 2 (6.67%) of *Pseudomenas aeruginosa* and *Sarcinia spp.* The results obtained are in agreement with Moreiro, *et al* (10) they also investigated the presence of many bacterial species recarried by ants including *S.aureus, Klebsiella spp* and *Streptococcus spp.*

Results in table (2) revealed the comparison between the occurrence of bacterial species isolated from the external insect surface and the internal insect components, which shows 19 (63.3%) isolates from the external surface comparing with 11 (36.6%) isolates from the internal components, these results emphasize the importance of considering ants as potential vectors of diseases caused by virulent and pathogenic bacteria like *S.aureus*.(11).

S.aureus is among the microorganisms able to from biofilms and it has been 65% of nosocomial infections are associated with biofilms (12), which aid the bacterial adherence to the ants surface.

Table 1 : The types of bacteria isolated from ants

Isolate species	No. of isolate	Occurrence %
Staphylococcus aureus	8	26.23
Escherichia coli	7	23.43
Bacillus subtilis	4	13.34
Klebsiella spp.	4	13.34
Proteus spp.	3	10
Pseudomonas aeruginosa	2	6.67
Sarcinia spp	2	6.67
Total	30	50% from 60 samples

Table 2: Bacterial species isolated from ants percentage according isolation method

Isolate species	Total No. of isolates	External surface		Internal component	
		No	%	No	%
Staphylococcus aureus	8	4	50	4	50
Escherichia coli	7	4	57.14	3	42.85
Bacillus subtilis	4	2	50	2	50
Klebsiella spp.	4	3	75	1	25
Proteus spp.	3	2	66.6	1	33.3
Pseudomonas aeruginosa	2	2	100	0	0
Sarcinia spp.	2	2	100	0	0
Total	30	19	63.3	11	36.6

As shown in table (3) the most isolates of *S. aureus* were obtained from the garden, while most *E. coli* isolates were isolated from ants who feeding on died

Cockroaches.

Also 40% of all isolates were isolated from the Garden comparing with 33.34% from the house, Among these pathogenic isolates presented the highest level of contamination of ants with these species. Further more An overall analysis indicated that the bacterial isolates from ants show to a large number of antimicrobials, these results are in agreement with (14).

Table 3: Bacterial species isolates from ants according to their collection position

Isolates species	No. of isolates	No. of isolates according collection position			
		Garden	Hose room	Cockroaches	W.C
S. aureus	8	4	3	1	0
E. coli	7	1	2	3	1
B. subtilis	4	2	2	0	0
Klebsiella spp	4	1	2	0	1
Proteus spp.	3	1	0	1	1
Pseudomonas aeruginosa.	2	1	1	0	0
Sarcinia spp.	2	2	0	0	0
Total of isolates	30	12	10	5	3

In the present study, there was a considerably high degree of resistance of different bacterial species isolated from ants to various antimicrobial agents, it can be seen in table (4).

That isolates of *S.aureus* and *E. coli* were highly resistant to Vancomycin (100%) comparing to the other antibiotics, Although many published studies on this subject revealed that most of the isolated strains of *S.aureus* and *E. coli* were resistant to Vancomycin (12) it appears that most isolated bacteria are highly sensitive to Nitrofurantin, when 25 isolates of 30 isolates were sensitive to this antimicrobial agent., Carek and Sack (13) stated that the initial antibiotic regimens for patients with bacterial infection (*S.aureus*) are Nitrofurantin. It can therefore be concluded that ants are important vectors of bacteria, presenting a high level contamination [(50%) of sample] and can be considered important vectors in house environments. Further more carried by ants show higher levels of resistance than the bacteria isolated from the environment ants are disseminators of bacterial resistance.

Table (4) antibiotic susceptibility of different bacterial species isolated from ants

Bacterial species	No. of isolates	Types of antibiotics													
		N		G		A		V		Am		C		Ci	
		S	R	S	R	S	R	S	R	S	R	S	R	S	R
S. aureus	8	8	0	5	3	3	5	-	8	5	3	3	5	5	3
E. coli	7	6	1	3	4	4	3	-	7	3	4	2	5	3	4
Bacillus spp.	4	2	2	3	1	3	1	2	2	1	3	4	-	1	3
Klebsiella spp	4	3	1	3	1	1	3	1	3	3	1	3	1	3	1
Proteus spp.	3	2	1	1	2	-	3	1	2	1	2	1	2	3	-
P. aeruginosa	2	2	-	1	1	-	2	-	2	-	2	1	1	2	-
Sarcinia spp.	2	2	-	1	1	1	1	-	2	1	1	2	-	1	1
Total of isolates	30	25	5	17	13	12	18	4	26	14	16	16	14	18	12

G = Gentamicin N = Nitrofurantin Am = Amikacin Ci = Ciprofloxacin
A = Ampicillin V = Vancomycin C = Cefataxin

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عزل وتشخيص والمقاومة للمضادات الحياتية لبعض البكتريا المرضية من النمل

الخلاصة

أجريت الدراسة على 60 نموذج من حشرات النمل، جمعت من مواقع مختلفة شملت: الحدائق والمنازل والأغذية والصراصر الميتة والمرافق الصحية. استخدم في العزل الجرثومي طريقتين: عزل البكتريا من السطح الخارجي لجسم الحشرة والعزل من المحتويات الداخلية لجسم الحشرة وذلك لبيان الأهمية الصحية لهذه الحشرة ودورها في نقل البكتريا المرضية.

أظهرت النتائج عزل 30 عزلة من الأجناس البكترية المختلفة وبنسبة 50% تضمنت:*Staphylococcus aureus* بأعلى نسبة 26.23% يليها كل من *Escherichia coli* ، *Bacillus subtilis*، *Klebsiella spp.*، *Proteus spp*، *Pseudomonas aeruginosg* و*Sarcinia spp* وبنسبة (6.67,6.67,10,13.34.13.34,23.34) % وعلى التوالي، حيث تبين أن للنمل دورا مهما في نقل هذه الجراثيم حاملا إياها على السطح الخارجي للجسم مع عدم ظهور الإعراض أو التأثيرات المرضية على الحشرة، وكانت معظم العزلات هي تلك المعزولة من السطح الخارجي لجسم الحشرة 63.33% مقارنة بعدد العزلات من الأحشاء الداخلية للحشرة.

اجري اختبار حساسية العزلات للمضادات الحيوية باستخدام (7) مضادات، أذا أظهرت العزلات مقاومتها لمعظم المضادات وخاصة الفانكومايسين، فيما كانت معظم العزلات حساسة لمضاد النايتروفيورانتين.