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The Incidence of Multirsistance *Kocuria spp*. among Different Clinical Samples in Erbil City, Iraq

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Background: Gram positive *Kocuria spp.* are a part of normal of the skin and oropharynx commensals in mammals but also described as opportunistic pathogens in human infections including urinary tract infection, pneumonia, wound infection, Ear infection, bronchial infection and also peritonitis and brain abscess, isolated from different clinical specimens increasing resistance to some antibiotics all over the world by distribution between patient with high morbidity and mortality rates.
Objective: Our study aimed to carry out retrospective study on *Kocuria spp.* isolated from various clinical samples in Erbil city and analyzed its epidemiology, antibiotic resistance pattern and pathogenic potential.
Methods: A total of thirty *Kocuria spp.* isolated distributed according to their source of isolation such as blood, wound, CSF, ear swab, throat swab, ascetic fluid and bronchial wash isolated and

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identified by using microscopical, morphological, biochemical tests and API and Vitek 2 compact system. Antibiotic susceptibility test was performed by manually by using 8 antibiotics (Ciprofloxacin, Gentamicin, Amikacin, Nitrofurantion, Cefotaxime, Ceftriaxone, Cifixime, Amoxicillin-Clavulanate (2:1)). Phenotypic screening of ESBLs was undertaken using Vitek 2 compact system.
Results: Out of 4399 samples, 30 samples were tested positive for *Kocuria spp*., the highest percentage of *Kocuria spp*. In female 20(0.76%) was higher than those in male patients 10(0.57%) Results showed that 30 isolates of Kocuria distributed as (13) isolates were indicated as *Kocuria kristinae,* (5) *Kocuria varians,* and (6) *Kocuria rosea Kocuria spp.*isolates showed high resistance (43.3%) to Cifixime, (30%) to Amoxicillin-Clavulanate 2:1, (26.7%) to Amikacin, (20%) to Nitrofurantion and (13.3%) to both Gentamicin and Cefotaxime. *Kocuria spp.* was surprisingly (100%) sensitive to Ciprofloxacin and Ceftriaxone with all the 30 samples that we had. Relation between bacteria species and gender, all species showed higher number in female than male.

Keywords: Gram positive Kocuria spp.; clinical samples; antibiotic sensitivity.

1. INTRODUCTION

"*Kocuria* belongs to Micrococcaceae family and compromises at least 18 different species of aerobium gram positive cocci" [1]. "These microorganisms are found as normal flora and about 5 species have been described as opportunistic pathogens in human infections. The name kocuria is honor to the Slovenian microbiologist Miroslav kocur, rosea came from "pink" and *kocuria rosea* was known as *Micrococcus rosea* but since 1995 has its actual name" [2]. "Kocuria belongs to the Family Micrococcaceae, Suborder Micrococcineae, order Actinomycetales, class actinobacteria" [3]. "kocuria species are skin and oropharynx commensals in mammals(including man), as well as environmental organisms including the soil and several other ecological niches" [4]. "*K.aegyptia* was isolated from saline, alkaline desert-soil sample from Egypt" [5]. "*K.marina* was found to inhibit marine sedimentation in the East seberian sea" [6]. "*K.carniphila* was observed colonize meat" [7]. "Both *K.koreensis* and *K.atrinae* were isolated in korea from jeotgal, a traditional fermented seafood made from comb pen shell" [8]. "Recently there have been reports of gram-positive cocci which are morphologically similar to both *Staphylococci* and *Micrococci*. These bacteria have been identified as *Kocuria* species with the help of automated identification system and other molecular methods including 16S rRNA(ribosomal ribonucleic acid)evalution . Kocuria belongs to the family Micrococcaceae which also includes *Staphylococcus* species and *Micrococcus* species. Isolation and clinical significance of these bacteria from human specimens warrant great caution as it does not necessarily confirm infection due to their ubiquitous presence, and as a normal flora of skin and mucous membrane in human and animals. Most clinical microbiology laboratories ignore such bacteria as laboratory and specimen contaminants. With increasing reports of infections associated with these bacteria. Its now important for microbiologists to identify and enumerate the virulence and antibiotic susceptibility patterns of such bacteria and assist clinician in improving the patient care and management" [9]. "Kocuria is a gram positive cocci arranged in pairs,short chains,tetrads,cubical packets of eight and irregular clusters" [10]. "The organism was preliminarily as *Kocuria* based on the phenotypic test results ,such as positive reactions for catalase,oxidase,nitrate reduction and and grow in 5% of NaCl and motility test negativity. Subsequent additional tests,such as bacitracin susceptibility, lysozyme sensitivity at 200mg, resistance to furazolidone and lysostaphin, helped to discriminate *Micrococci* from *Staphylococci*" [11]. "Major criteria for the conventional discrimination between *Staphylococci* and *Micrococci* are the sensitivity of Kocuria to bacitracin and lysozyme(while *Staphylococci* are resistant to both) and the resistance of *Kocuria* to netrofurantoin/ furazolidone and lysostaphin(*Staphylococci* are susceptible to the latter, although they may express resistance to the former)" [12]. "*Kocuria* species do not produce hemolysis on blood agar, unlike most clinical isolates of *Staphylococci* .This bacterium is normally misidentified in the clinical microbiology laboratories as coagulase negative *Staphylococci* (CoNS) based on its gram reaction, catalase positive and coagulase negative properties. Other physiological and biochemical properties of *Kocuria* are the formations of non-hemolytic colonies on blood agar, non-capsulated, non-spore forming, non-motile, non-acid fast and positive for Voges-Proskauer test (VP).Kocuria spp. have been

reported to be normal flora of human skin and oral cavity and are usually regarded as laboratory contaminants and ignored when isolated in the clinical specimens undermining its pathogenic potential.. Infections associated with isolation of *Kocuria* include urinary tract infections, cholecystitis, catheter-associated bacteremia, dacryocystitis, canaliculitis, keratitis, native valve endocarditis, peritonitis, descending necrotizing mediastinitis, brain abscess and meningitis" [13]. "The predisposing factors associated with infections related to *Kocuria spp*. include congenital deformities (short bowel syndrome), chronic catheterization (in cases of total parenteral nutrition), *Kocuria* appears to mostly affect compromised hosts suffering from haematological malignancies, solid tumours or metabolic disorders, although only *K. kristinae, K. marina* and *K. rhizophila* have been observed to cause infections in humans, to the best of our knowledge" [3]. "Currently, these bacteria are gaining importance as emerging pathogens in hosts with cancer, immunocompromised status and metabolic disorders. Two species, *Kocuria rosea* and *Kocuria kristinae* have been reported to cause catheter-related bacteremia" [14]. "A recent research report has highlighted the significance of *Kocuria* in causing hospital acquired infections" [3]. "The same study has also noted that although Kocuria spp. are commensals of humans, animals and are present in the environment, they should be considered as potential pathogens in patients who are immunocompromised, undergoing critical care treatment and neonates. A study which included 12 pediatric age patients suffering from underlying debilitating conditions like premature birth and cancer had noted that more than 50% of patients suffered from invasive infections with *Kocuria* spp" [15]. "The disc-diffusion method revealed that *Kocuria spp.* was susceptible to ampicillin, cefotaxime, ciprofloxacin, cloxacillin, gentamicin, erythromy- cin, amikacin, imipenem, linezolid, teicoplanin and vancomycin, but showed intermediate resistance to ceftazidime [16], *Kocuria sp* is sensitive to a variety of drugs (amoxicillin, cephalosporin, aminoglycoside, vancomycin, clindamycin); variable sensitivity to quinolones and sulfa. Amoxicillin-clavulanate has been proposed as the initial antibiotic treatment. Potentially contaminated catheters, if present, must be removed" [12]. Becker et al. [17] have reported that "the *Kocuria rhizophila* isolated in blood from a case of sepsis in pediatric age patient revealed resistance only to norfloxacin". "Resistance to ciprofloxacin and erythromycin was observed in *K. rhizophila* isolated from blood of a 3-year-old catheterized patient" as reported by Moissenet et al. [18]. "*K varians* isolated in peritoneal fluid resistant only to levofloxacin was reported by Meletis et al in a patient undergoing continuous ambulatory peritoneal dialysis" [19].Decreased cell wall permeability and the presence of efflux pumps are implicated in the resistance of Kocuria species [20]. Aim of our study to detect this *Kocouria spp*.and determine the sensitivity againdst antibiotics.

2. MATERIALS AND METHODS

2.1 Sample Collection

A total of (4399) samples were collected from seven different sources (urine, wound swab, branchial wash,ascitic fluid, CSF, sputum and Ear swab)from male and female . After collection all species isolates were subjected to a series of confirming tests.. Clinical samples were collected from Rizgary hospital, Raparin hospital, Central lab and Estern emergency hospital in Erbil city/Iraq during the period (July 2017 to February 2018) from male and female patients with the age of 10-79 years .For isolation of microorganisms, the specimen was directly inoculated on culture media; Blood culture and macConkey agar plates were incubated aerobically at 37°C for (24-48) hours. Pure colonies of isolated microorganisms were identified using morphological, biochemical tests, Species identification and antibiograms for pathogens were performed using Vitek 2 system [21].

2.2 Antimicrobial Susceptibility test by Vitek 2 System

All isolates of were screened for their susceptibility to 13 used antibiotics by using Vitek 2 system **w**ith its ability to provide accurate "fingerprint" recognition of bacterial resistance mechanisms and phenotypes, the AES is a critical component of Vitek 2 technology .The Vitek 2 card contains 64 micro wells. Each well contains identification substrates or antimicrobial. Vitek 2 offers a comprehensive menu for the identification and antibiotic susceptibility testing of organisms. The Vitek 2 test card is sealed, which minimizes aerosols, spills, and personal contamination. Disposable waste is reduced by more than 80% over microtiter methods [21].

2.3 Phenotypic Screening for ESBL

Each isolate was tested using the VITEK 2 system with the antimicrobial susceptibility test

extend AST-EXN8 card. This system was designed to perform both screening and confirmatory tests for phenotypic detection of ESBL on the same plate. The test comprises a panel of six wells containing ceftazidime 0.5 mg⁄L, cefotaxime 0,5 mg⁄L and cefepime 1,0 mg⁄L, the rest of three wells were filled with same three antibiotics in combination with clavulanic acid.

(4,4 -10 mg⁄L, respectively). Growth in each well was quantitatively assessed by means of an optical reader. The proportional reduction in growth in wells containing cephalosporin + clavulanate compared with those containing the cephalosporin alone was considered to be indicative of ESBL production. All phenotypic interpretations of ESBLs were reported as a positive ESBL screening result. Strains were reported as ESBL-negative whenever phenotypic interpretations other than ESBLs were proposed by the An expert system (AES) [21].

3. RESULTS

3.1 Frequency of *Kocuria species* in 4 Different Labrotories

In present study among 4399 samples, 30 samples were diagnosed as *Kocuria spp.* Frequency of *Kocuria spp.* Isolated, a total of (1082) samples were collected from patients attending Raparin hospital only (8) were tested positive.In Rizgary hospital, our results showed that among (1471) samples only 10 were tested positive with *Kocuria spp*. while 1461 were negative as in Table 1.In Central lab, from (1144) samples only (9) were tested positive. Estern emergency lab, a total of (702) samples (3) samples were tested positive meanwhile (699) samples were negative as in Table 2.

3.2 Relation between *Kocuria spp.* with Gender (Female, Male)

Relation between *Kocuria spp*. with gender (female, male). Out of 4399 samples, 30 samples were tested positive for *Kocuria spp*. ,the highest percentage of *Kocuria spp*. In female 20(0.76%) was higher than those in male patients 10(0.57%) as in Table 2 and Fig. 1.

3.3 Relation between Species of Kocuria Depending on Gender

Out of 2350 samples 10 sample *Kocuria spp*. isolates distribution according to species and gender.The highest species found among the 30 positive samples are *Kocuria kristinae* which is 13 samples 7 in female and 6 in male which is almost a near ratio between male and female. *Kocuria rosea* and Kocuria spp. comes after *Kocuria kristinae* with 6 samples for each one but in Kocuria rosea 5 in female and 1 in male while in *Kocuria spp.*4 in female while 2 in male. And last we have *Kocuria varians* with 5 samples 4 in female and 1 in male as in Table 3 and Fig. 2.

3.4 The Number of Antibiotics (Sensitive and Resistant) for *Kocuria spp.*

Regarding antibiotic resistance pattern in *of Kocuria spp.* isolates 30 isolates were screened for their susceptibility to but in general most isolates of *Kocuria spp.*were multi drug resistance to more than three antibiotics as in Table 4.

Table 1. Distribution of *Kocuria spp*. according to location in Erbil city

Samples	Positive samples		Negative samples		Total	
	No.	%	No.	%	No.	%
Rizgary Lab	10	0.68%	1461	99.32%	1471	100%
Raparin Lab	8	0.74%	1074	99.26%	1082	100%
Cenral Lab	9	0.79%	1135	99.21%	1144	100%
Eastern Emergency Lab	3	0.43%	699	99.57%	702	100%
Total	30	0.68%	4369	99.32%	4399	100%

Table 2. Relation between male and female in *Kocuria spp.*

Patients	Culture positive		Culture negative		Total	
	No.	%	No.	%	No.	%
Male	10	0.57%	1750	99.43%	1769	100%
Female	20	0.76%	2619	99.24%	2639	100%
Total	30	1.33%	4369	98.67%	4399	100%

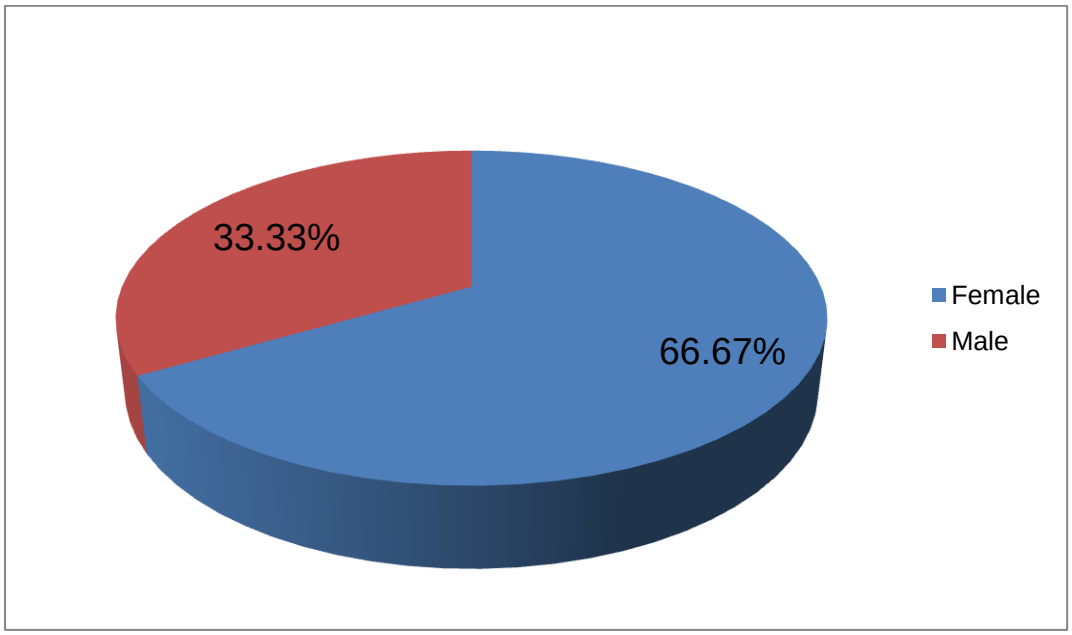

Fig. 1. Relation between *Kocuria spp*. with gender

Table 3. Distribution of *Kocuria spp*. according to species in male and female

Species	Positive samples in female		Negative samples in male		Total	
	No.	%	No.	%	No.	%
Kocuria kristinae	7	53.85%	6	46.15%	13	100%
Kocuria varians	4	80%	1	20%	5	100%
Kocuria rosea	5	83.33%	1	16.67%	6	100%
Kocuria spp.	4	66.66%	2	33.34%	6	100%

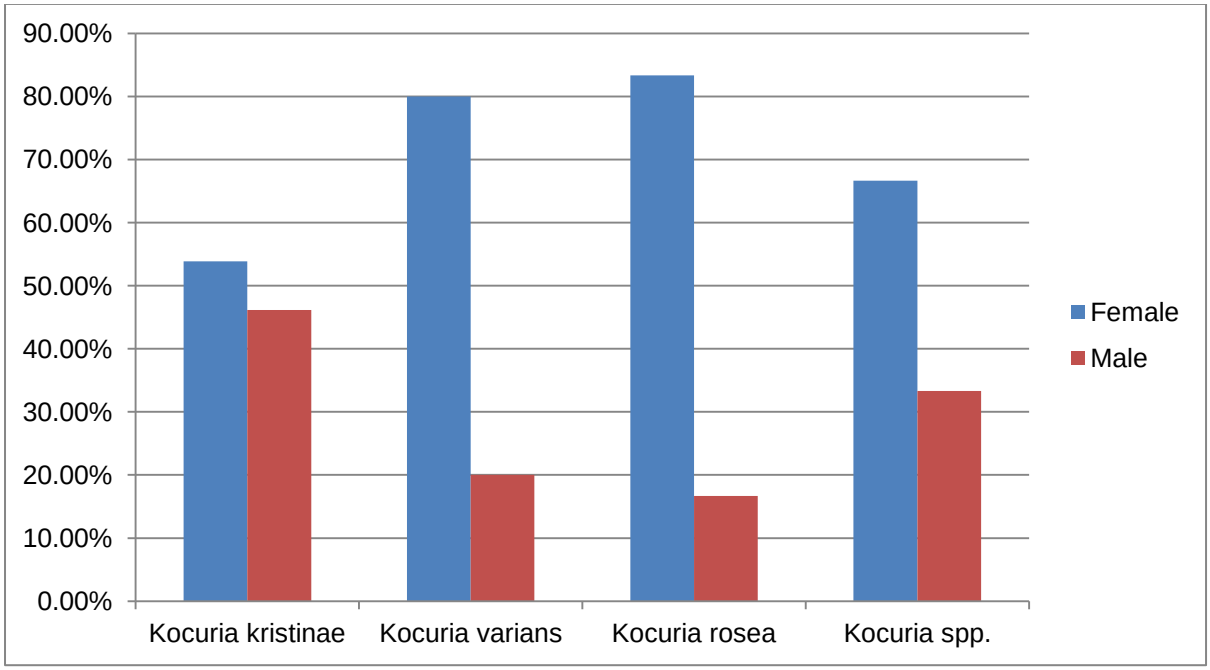

Fig. 2. Relation between *Kocuria spp*. depending on gender

3.5 The Incidence of *Kocuria spp.*i n Different Clinical Specimens

Out of 2350 samples 10 sample isolates distribution according to their source of isolation as in Table 5. The most isolates of *Kocuria spp* is isolated from urine 16 (0.37%) followed by 4(0,09%), 3(0.07%), 2(0.05%) isolates from CSF and sputum and throat swab respectively and only 1(0,02%) isolates from Acetic acid, bronchial, ear swab, blood and wound for each on the other hand the highest of *Kocuria* species found among the 30 positive samples are *Kocuria kristinae* which is 13 samples followed by 6 (0.13%) *Kocuria varians* and other species of *Kocuria* and only 5(0.11%) for *Kocuria rosea.*

3.6 Antibiotic Susceptibility Patterns of *Kocuria spp.*

The bacterial isolates revealed remarkable variation in their resistance and sensitive antibiotics used, Regarding antibiotic resistance pattern in *of Kocuria* isolates . 30 isolates of *of Kocuria spp.* were screened for their susceptibility to 8 used antibiotics (Ciprofloxacin, Gentamicin, Amikacin, Nitrofurantion, Cefotaxime, Ceftriaxone, Cifixime, Amoxicillin-Clavulanate (2:1)).Results indicate that most effective antibiotic were Ciprofloxacin 100%, and Ceftriaxone 100% and 86.7% to Gentamicin and Cefotaxime followed by 80% sensitive to Nitrofurantion while less effective to antibiotics like Amikacin (73.3%) followed by Amoxicillin-Clavulanate (70%) as in Table 6.

4. DISCUSSION AND CONCLUSION

"Kocuria previously classified as one of the six genera of *Micrococcus*, have been reclassified as a separate genus based on its phylogenetic and chemotaxonomic analysis" [10]. "Reports of infection with *Kocuria* species have gained prominence in the late twentieth century and are showing an increased trend, signifying its pathogenic potential. Infections associated with isolation of *Kocuria* include urinary tract infections, cholecystitis, catheter-associated bacteremia, dacryocystitis, canaliculitis, keratitis, and native valve endocarditis, peritonitis,

Table 4. Patterns of sensitivity of *Kocuria spp.*

Isolates		Antibiotics									
		CIP	GEN	AK	NIT	CTX	CRO	CFM	AMC	NO.R	NO.S
1	*Kocuria kristinae*	S	S	S	S	S	S	R	R	2	6
2	*Kocuria kristinae*	S	S	S	R	R	S	R	S	3	5
3	*Kocuria kristinae*	S	S	S	S	S	S	S	R	1	7
4	*Kocuria kristinae*	S	S	S	S	S	S	S	R	1	7
5	*Kocuria kristinae*	S	S	S	S	S	S	S	R	1	7
6	*Kocuria kristinae*	S	S	S	S	S	S	S	S	—	8
7	*Kocuria kristinae*	S	R	S	R	S	S	S	S	2	6
8	*Kocuria kristinae*	S	S	S	S	R	S	R	R	3	5
9	*Kocuria kristinae*	S	S	S	R	R	S	R	S	3	5
10	*Kocuria kristinae*	S	S	S	S	S	S	R	S	1	7
11	*Kocuria kristinae*	S	S	S	S	R	S	S	R	2	6
12	*Kocuria kristinae*	S	R	S	S	S	S	S	S	1	7
13	*Kocuria kristinae*	S	S	S	S	S	S	R	S	1	7
14	*Kocuria kristinae*	S	S	S	S	S	S	R	S	1	7
15	*Kocuria rosea*	S	S	R	S	S	S	S	S	1	7
16	*Kocuria rosea*	S	S	R	S	S	S	R	S	2	6
17	*Kocuria rosea*	S	S	R	S	S	S	R	S	2	6
18	*Kocuria rosea*	S	S	R	S	S	S	R	S	2	6
19	*Kocuria rosea*	S	S	R	S	S	S	S	S	1	7
20	*Kocuria rosea*	S	S	R	S	S	S	R	S	2	6
21	*Kocuria varians*	S	R	S	S	S	S	S	S	1	7
22	*Kocuria varians*	S	S	S	R	S	S	S	R	2	6
23	*Kocuria varians*	S	R	S	S	S	S	R	S	2	6
24	*Kocuria varians*	S	S	R	S	S	S	S	S	1	7
25	*Kocuria spp.*	S	S	S	R	S	S	S	S	1	7
26	*Kocuria spp.*	S	S	S	S	S	S	S	S	—	8
27	*Kocuria spp.*	S	S	S	S	S	S	S	R	1	7
28	*Kocuria spp.*	S	S	S	S	S	S	S	R	1	7
29	*Kocuria spp.*	S	S	R	S	S	S	R	S	2	6
30	*Kocuria spp.*	S	S	S	R	S	S	S	S	1	7

S: Sensitive R: Resistance
CIP:Ciprofloxacin GEN:Gentamicin AK:Amikacin NIT:Nitrofurantion CTX:Cefotaxime CRO:Ceftriaxone CFM:Cifixime AMC:Amoxicillin-Clavulanate (2:1)

Table 5. Frequency of *Kocuria spp.* in different clinical specimens

	No. and % 0f *Kocuria spp.*									
	Urine		**CSF**	**Sputum**	**Ascitic fluid**	**Bronchial wash**	**Throat swab**	**Ear swab**	**Blood**	**Wound**
	Number and %									
Kocuria kristinae	Positive (%)	8 (0.18%)	1(0.02%)	2 (0.05%)	—	1(0.02%)	—	1(0.02%)	—	—
	Negative (%)	1673(38.03%)	1018(23.14%)	784 (17.82%)	98(2.23%)	125(2.84%)	195(4.43%)	186(4.23%)	198(4.50%)	109(2.48%)
Kocuria rosea	Positive (%)	2 (0.05%)	1(0.02%)	—	—	—	1(0.02%)	—	1(0.02%)	—
	Negative (%)	1679(38.17%)	1018(23.14%)	786(17.86%)	98(2.23%)	126(2.86%)	194(4.41%)	187(4.25%)	197(4.48%)	109(2.48%)
Kocuria varians	Positive (%)	3(0.07%)	1(0.02%)	—	1(0.02%)	—	1(0.02%)	—	—	—
	Negative (%)	1678(38.15%)	1018(23.14%)	786(17.86%)	97(2.21%)	126(2.86%)	194(4.41%)	187(4.25%)	198(4.50%)	109(2.48%)
Kocuria Spp	Positive (%)	3(0.07%)	1(0.02%)	1(0.02%)	—	—	—	—	—	1(0.02%)
	Negative (%)	1678(38.15%)	1018(23.14%)	785(17.85%)	98(2.23%)	126(2.86%)	195(4.43%)	187(4.25%)	198(4.50%)	108(2.46%)
Total	Positive (%)	16(0.36%)	4(0.09%)	3(0.07%)	1(0.02%)	1(0.02%)	2(0.05%)	1(0.02%)	1(0.02%)	1(0.02%)
	Negative (%)	1665(37.85%)	1015(23.07%)	783(17.80%)	97(2.21%)	125(2.84%)	193(4.39%)	186(4.23%)	197(4.48%)	108(2.46%)

Table 6. Antimicrobial susceptibility testing for *Kocuria spp.*

Antibiotics	Sensitive No. (%)	Resistance No.(%)
Ciprofloxacin	30 (100%)	—
Gentamicin	26 (86.7%)	4 (13.3%)
Amikacin	22 (73.3%)	8 (26.7%)
Nitrofurantion	24 (80%)	6 (20%)
Cefotaxime	26 (86.7%)	4 (13.3%)
Ceftriaxone	30 (100%)	—
Cifixime	17 (56.7%)	13(43.3%)
Amoxicillin-Clavulanate (2:1)	21 (70%)	9(30%)

descending necrotizing mediastinitis, brain abscess and meningitis" [13] "Identification of *Kocuria spp.* remains elusive because most clinical microbiology laboratories have limited or no access to advanced molecular techniques. Laboratory identification of *Kocuria spp.* can be made conventionally only after high laboratory suspicion. Properties such as morphological variability between these bacteria and other similar gram-positive cocci, as well as biochemical properties including the antimicrobial susceptibility patterns against selective antibiotics could be used to presumptively identify *Kocuria* spp" [9]. "laboratory identification *of Kocuria spp*. by biochemistry methods is difficult due to similarity with other pathogens, especially coagulase negative *staphylococci*, which delays the proper treatment" [12] .

A total of (4399) samples were collected from six different sources (Urine, Sputum, Wound, CSF, Bronchial wash, Ascetic fluid). After collection and all bacterial isolates were subjected a series of confirming tests to ensure that these isolates recovered belong to *Kocuria spp* identified using microscopical, morphological, biochemical tests, and Vitek 2 system. Result showed that only 30(1.33%) isolated were indicated positive as gram positive *Kocuria spp.* All bacterial isolates were grown on selective media which was for cultural and morphological characteristics. The colonies on blood agar were whitish, small, round, raised, convex colonies on initial isolation and might develop non-diffusible yellowish pigmentation after prolonged incubation. *Kocuria* spp. do not produce hemolysis on blood agar, the most distinctive colony morphology characteristics of *Kocuria spp.* colonies on tryptic soy agar (TSA) are large round, diffusible yellowish pigmentation, not grow in macConkey agar .Bacterial cells from gram stain are gram positive cocci showing large sized cocci arranged in pairs, short chains, tetrads clusters and deeply stained very large cocci. Oxidase and catalase reaction positive but for coagulase enzyme test negative results.*Kocuria* is a gram positive cocci arranged in pairs, short chains, tetrads, cubical packets of eight and irregular clusters [10]. The organism was preliminarily as *Kocuria* based on the phenotypic test results, such as positive reactions for catalase, oxidase, nitrate reduction and and grow in 5% of NaCl and motility test negativity. "Cultures are more of a [12] challenge and requires detailed clinical history before labeling the organism as contaminant or commensal". "The newer diagnostic methods such as Vitek 2 and 16S RNA based genotypic assay are more accurate in identifying this organism and thus help preventing its erroneous identification" [22]. "By routine conventional tests, it can easily be mistaken for *Micrococcus* and coagulase negative *Staphylococci* cultures is more of a challenge and requires detailed clinical history before labeling the organism as contaminant or commensal. The newer diagnostic methods such as Vitek 2 and 16S RNA based genotypic assay are more accurate in identifying this organism and thus help preventing its erroneous identification" [23].

In present study among 4399 samples, 30 (1.33%) samples were diagnosed as *Kocuria spp.* (8) were tested positive with *Kocuria spp. from* Raparin hospital, (10) were tested positive with *Kocuria spp*. from Rizgary hospital ,while (9) were tested positive from Central lab, only (3) samples were tested positive from Eastern emergency lab . This variation might have been either due to sample size, the prevalence of bacterial varies widely among different areas and communities within the country, the varies prevalence may be because of various reasons such as differences in economic status and educational background, study population and method used for diagnosis of bacteria.

Out of 4399 samples, 30 (1.33%) isolates of *Kocuria spp*., the percentage of *Kocuria spp*. in female 20(0.76%) was higher than those in male patients 10(0.57%), our result not agree to

results recorded by Horino et al. [23] reported cases 38 of *K.kristinae* infections 12 female and 16 male hemodialysis patient with diabetes mellitus, while Pauli et al. reported that the apparently healthy woman of acute 3940 bacterial meningitis due to *Kocuria rosea* with fulminant course and fatal outcome. The differences of number of *Kocuria spp*. isolates in male and female might due to sample size were more in female than in male (2639,1769) respectively because our target populations only patient attending the hospital randomly in Erbil city,The most isolates of *Kocuria spp* is isolated from urine 16 (0.37%) followed by 4(0,09%), 3(0.07%), 2(0.05%) isolates from CSF and sputum and throat swab respectively and only 1(0,02%) isolates from Acetic acid, bronchial, ear swab, blood and wound for each on the other hand the highest of *Kocuria* species found among the 30 positive samples are *Kocuria kristinae* which is 13 samples followed by 6 (0.13%) *Kocuria varians* and other species of *Kocuria* and only 5(0.11%) for *Kocuria rosea.* Our results concordant with other previous study by Lai *et al [24]* who recorded a total 41of 21 blood isolates from five patients were identified as *Kocuria* species by conventional biochemical methods, twenty isolates of *K. kristinae*, which were initially identified by the Phoenix automated system (confidence value 99%) and VITEK 2 system (probability of identification 99%), describe five patients with positive blood culture for other *Kocuria* species, three patients had catheter-related bacteraemia and one had infective endocarditis caused by *Kocuria kristinae*, and one had a *K. marina* isolate, which was considered to be a contaminant. 41 .Also our results agreement with Altuntas et al. who reported 23 cases of 17infection due to Kocuria species have been reported with *K. kristinae* as the most common pathogen followed by *K. rosea, K. marina, K. rhizophila and K. varians w*hile disagree with other results when the most common infections reported are central venous catheter related blood-stream infections in patients with some underlying disease [25,26]. The need to consider *Kocuria* as an emerging neuropathogen in life threatening CNS infections and an outmost microbiological vigilance is required for its specific identification.

Regarding antibiotic resistance pattern in *of Kocuria* isolates. 30 isolates of *Kocuria spp.* were screened for their susceptibility to 8 used antibiotics .The bacterial isolates revealed remarkable variation in their sensitive to antibiotics used,but in general the most sensitive antibiotics against *Kocuria spp* were Ciprofloxacin and Ceftrixon 30(100%) and the result revealed that most of isolates of *Kocuria spp* resistance against Cefixime13(43.3%)and then 9(30%) against Amoxicillin-Clavulanate similar results were recorded by Chen et al. [15] who found that treatment for isolated *K. kristinae*. 24has been successful using monotherapy with Oxacillin, Vancomycin, Piperacillin/ Tazobactam, and Ciprofloxacin and combination therapy with Teicoplanin and Vancomycin, Ciprofloxacin, and Clindamycin as well as Ceftriaxone and Ofloxacin. Also Becker et al. [17] have reported that the *Kocuria rhizophila* isolated in blood from a case of 14sepsis in paediatric age patient revealed sensitive against most of antibiotics and resistance only to norfloxacin14 and Studies by Lee et al. [3] who reported “multiple cases 3involving both extreme age groups noted that *Kocuria marina* isolated from peritoneal fluid was resistant only to tetracycline. Other observations by the same authors showed that *K. kristinae* isolated from bacteremia cases revealed resistance to Oxacillin, Cefazolin and intermediately sensitive to cefotaxime”. On the other hand in the four different hospitals in Erbil city non of them used antibiotics (Ampicillin and Amoxicillin) because *Kocuria spp* are 100% resistance to both antibiotics. The high rate of resistance to Ampicillin and Amoxicillin may reflect the fact that these are the most commonly subscribed antibiotics in hospitals and also the most easily available in the community without prescription and because they are very cheap terms of cost, and so subject to abuse and misuse of antibiotic. The resistance may be also due to the spontaneous and there are no control on take of the drugs, and about 50% of it given to outpatients without physicians prescription are from outside of hospital, as well as the occurrence of any infection in the organ of patient, they are taking antibiotics without culturing and determination of antibiotic susceptibility for the side effect, the emergence of different types of antibiotics. The presence of antimicrobial agents at low concentration through continued usage lead to the development of drug resistant isolates and multiple antibiotic resistance in bacteria reduced efficiency of antibiotic treatment for human and animal diseases. Clinicians also frequently commence patients on antibiotic therapy before sending samples to the microbiology laboratory such that many of the samples are negative when cultured. Abuse and misuse of antibiotics have been known to contribute to the development of antibiotic resistance.

ETHICAL APPROVAL AND CONSENT

The bacterial strains used in this research were extracted from clinical routine specimens, and patients were given written consent. This study has been accepted by the College of Health Sciences / Hawler Medical University Scientific and Research Ethics Committee.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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