Practical Study of Some Antibiotics and Their Effect on Some Pathogenic Bacteria

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Abstract

Despite the discovery of many new antibiotics, bacteria over time develop the ability to resist these antibiotics, a total of one hundred and seventy-three samples of different bacterial isolates of bacteria (158 isolates, and 15 isolates are not cultured) were taken from different patients in Iraqi Kurdistan hospitals from May 2021 to September 2021. Bacteria were isolated from a vaginal swab, a urine sample and ear swab samples from two patients with otitis media. Bacterial susceptibility to biofilm formation was examined. An antibiotic susceptibility test was examined for all bacterial isolates for thirty-one antibiotics. Where it was found that out of the complete of one hundred and sixty-eight patients, it was found that women are less susceptible than men. The bacteria that were isolated from the patients, the gram-positive bacteria were more isolated than the gram-negative bacteria. Most of the isolated bacteria are resistant to antibiotics and biofilm formation.

Keywords: *Antibiotics, Bacteria, Biofilm. Pathogenic.*

دراسة عملية لبعض المضادات الحياتية وتأثيرها على بعض أنواع البكتيريا الممرضة

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الخلاصة

على الرغم من اكتشاف العديد من المضادات الحياتية الجديدة ، إلا أن البكتيريا بمرور الوقت تطور قدرتها على مقاومة هذه المضادات الحياتية ، تم أخذ مائة وثلاثة وسبعون عينة من عزلات بكتيرية مختلفة (158 زرع موجب ، و 15 عزلة زرع سالب) من مختلف المرضى في مستشفيات كردستان العراق. تم عزل البكتيريا الممرضة من مسحة مهبلية وعينة بول ومسحة أذن من مريضين مصابين بالتهاب الأذن الوسطى. تم فحص القابلية البكتيرية لتكوين الغشاء الحيوي. تم فحص اختبار الحساسية للمضادات الحياتية لجميع العزلات البكتيرية لواحد وثلاثين من المضادات الحياتية . من إجمالي مائة وثمانية وستون مريضاً، تبين أن النساء أقل عرضة للإصابة من الرجال. البكتيريا التي تم عزلها من المرضى كانت البكتيريا الموجبة لصبغة جرام أكثر من البكتيريا السالبة لصبغة جرام. معظم البكتيريا المعزولة مقاومة للمضادات الحياتية ولها القدرة على تكوين الأغشية الحيوية.

الكلمات المفتاحية: *المضادات الحياتية، البكتيريا ، الغشاء الحيوي ، الممرض.*

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Introduction

Mortality and morbidity are caused by multiple resistance to antibacterials in addition to increased costs and treatment failures. Antibiotic resistance has been reported when a drug loses its capacity to successfully prevent bacterial growing [1]. Bacterias, when duplicates so in the existence of the antibiotics, are termed resistant bacterias. Antibiotics are commonly active against them, however once the bacteria quieten down susceptibility, it needs a better than the conventional levels of an equivalent medication to possess a bearing. The emergence of antimicrobial resistance was ascertained shortly when the introduction of recent anti-bacterial compounds [2]. There square measure several details behind the event of antibiotic resistance, starting from microorganism reasons to human aspects like overuse and over-prescription of antimicrobials, agricultural and industrial request of antimicrobials within the animal sector, and human behavioral factors [3]. Our ability to treat common pathogens becomes difficult owing to antibiotic, leading to magnified period of ill health, costs, range of difficulties, and deceases. Antibiotics contest to elimination of microorganisms. Therefore, microorganisms possess immune activity. The method of resistance occurs through sequence level mutations [4]. Antibiotics caused selective pressure and genes also act in the context of selective pressure [5]. Bacteria have a criterion for transferring genetic material directly between each other by transferring plasmids, showing that the natural process is not the only mechanism that develops through that resistance. A wide range of antibiotics are prescribed in hospitals as an answer to infections in healthcare facilities; But it will increase the resistance [6]. Antibiotics usually remove the bulk of the bacteria in an overgrowth. Though, a unique colony of genetically mutated bacteria may occur that may cause resistance [7،8]. The extent of antibiotic-resistant infections began to correlate closely with the degree of antibiotic depletion [9، 10].

Materials and Methods

Antimicrobial susceptibility testing for some bacterial strains include (*Staphylococcus* spp, *Escherichia coli* , *Streptococcus* spp , *Gardnerella vaginals, klebsiella pneumonea, Enterococcus faecalis*) that collected from patients in Iraqi Kurdistan hospitals, isolated from vaginal swab, urine, blood, ear swab, seminal fluid , synovial fluid , throat swab, genital lesion swab, sputum and stool .Were cultured on bacterial culture media represented by Nutrient agar, MacConkey agar, Blood agar, Mannitol agar, Chocolate agar at 37°C for (18-48) hours, and diagnosed using Gram stain and microscopic examination, biochemical tests were also used to isolate and diagnose bacterial isolates [11]. The following sample have been taken (Vaginal swab, urine, ear swap, synovial swap, seminal fluid, blood and stool specimen). Antimicrobial test include antibiotics (Nalidixic acid ,Ofloxacin,

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Ciprofloxacin, Gentamycin, Streptomycin, Refampin, Clindamycin, Trimethoprim, Nitrofurantion, CLarithromycin, Amikacin , Norofloxacin , lincomycin, Tobramycin, Cephalothine, cefem, Amoxiclave , Vancomycin, Meropenem, Cefadroxil, Azithromycine, Nitofumation, Cefriaxone , Cefexim , T_S (septrin), Suprax, Cepadroxil, Tobromycin, Metronidazole, Tetracycline, Imipenem, Amoxicillin , CLavulanic acid and Ampicillin), by using of Mueller-Hinton agar and depending on inhibition zone CLSI, Bioanaylyse (Turkey). The activity of biofilm formation was measured using microtiter plate (M.T.P) [12].

Results and discussions

The study involved the investigation of 173 patients suffering from different infections, vaginal swab from 91 patients, and urine sample 70 patients suffering from UTI and ear swab samples from two patients at otitis media, and including seminal fluid and stool (n=two patients) and sputum (n=one patient), seminal fluid (n=2), synovial fluid (1) and throat swab (1). The study also included identifying the pathogen most causing infection, and among the patients (Figure 1).

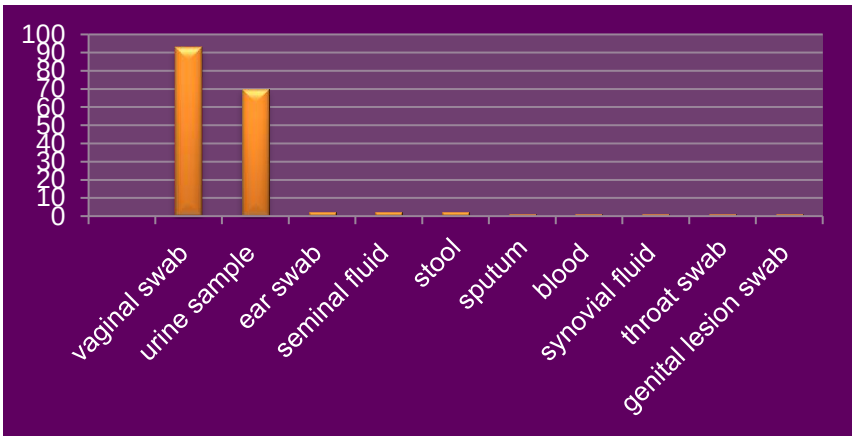

Fig. (1): Showing the number of specimen taken from patients.

The results exhibited that the gram positive bacteria that cause infections are more dominant than the gram negative bacteria that isolated from different specimen, as 103 bacterial (G +ve) isolates were isolated compared to 55 bacterial (G-ve) isolates, as shown in Table (1). The presence and spread of bacteria may be play a role in infection, particularly in suitcases of a pathetic immune system. Furthermore, the study focused on the year in which bacterial infections increase, which is due to its capacity to attack the efficiency of antibacterial substances current in saliva in addition to their capability to abandon to the epithelial tissues [13].

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Table (1): Distribution of bacterial infections in patients with different specimens**.**

The type of sample	Isolates		
	G +V	G --V	Total
vaginal swab	56	29	85
urine	42	24	66
blood	-	-	-
ear swab	2	-	2
seminal fluid	-	1	1
synovial fluid	-	-	-
throat swab	1	-	1
genital lesion swab	1	-	1
sputum	1	-	1
stool	-	2	2
total	103	55	158
%	65.19	34.81	

The table (1) shows the rate of infection between males and females, and it was renowned that the incidence of altered infections was higher in males than females, it is found that further than the overall 173 patients, the number of the diseased women is 76 (43.93%) and 97 men (56.07%), as presented in Figure (2). This may be because of the incidence or spread of certain bad behaviors in one sex over the other, including the prevalence of smoking and alcohol habits among males in excess of it is in females, which growths the rate of diseases [13] specified that this might flow from to the physiological and immunologic variations between the genders, which can change or increase the speed of infection or it should flow from to the immune status and also the secretion and purposeful variations between the sexes [14].

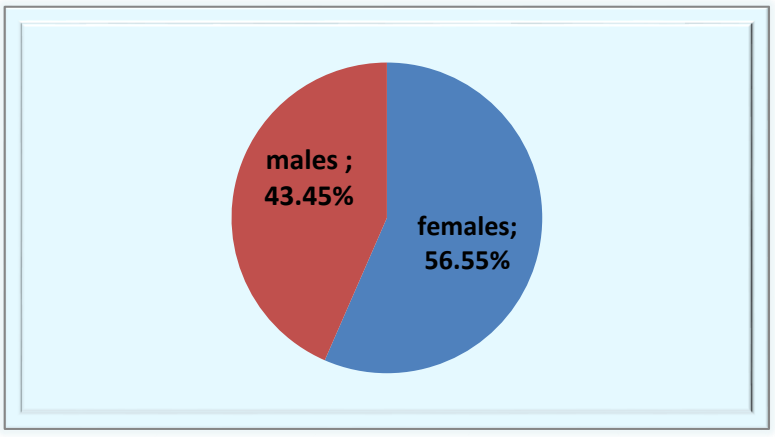

Fig. (2): Distribution of infections according to the sex of the patient.

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A total 173 tests of culturing different strains of bacteria from different specimen of Kurdish population, 158 results was positive and 15 was negative result. isolated vaginal swab (n = 93) which is 8 test negative, 85 tests positive, (42 / % 49.41 of strains are *staphylococcus spp)* , (29 / % 34.11 was *Escherichia coli*), (11 / % 12.94) was *Streptococcus spp)* , (3 / % 3.52 was *Gardnerella vaginals*) as shown in Table (2) , urine (n=71) which is 5 test negative , 66 test positive (34 / %51.51 was *Staphylococcus spp*) , (20 / %30.30 was *Escherichia coli*), (5 / %7.57 was *Streptococcus spp),* (3 / %4.54 was *Klebsiella pneumonia*), (2 / %3.03 was *Enterococcus faecalis*) , (1 / % 1.51 was *Pseudomonas aeruginosa*), (1 / %1.51 was *Lactobacillus* spp, *)* , blood (n=1 was negative test) , ear swab (n=2 was *Staphylococcus spp*) , Seminal fluid (n = 2 1 was negative , 1 was *Escherichia coli)* , synovial fluid (n = 1 was negative result), throat swab (n = 1 was *Streptococcus spp*), genital lesion swab (n = 1 was *Staphylococcus spp*), Sputum (n = 1 was *streptococcus spp*) and stool (n = 2 was *Escherichia coli)* .

Table (2): Types of bacteria isolated from patients according to the type of specimens.

Type of bacteria	Total of number		Vaginal swab		Urine		Ear swab		Other specimens	
	Number of isolate	%	Number of isolate	%	Number of isolate	%	Number of isolate	%	Number of isolate	%
Staphylococcus spp.	79	76.69	42	75	34	80.95	2	100	1	33.3
Streptococcus spp	18	17.48	11	19.64	5	11.91	-	-	2	66.7
Enterococcus fascial	2	1.94	-	-	2	4.76	-	-	-	-
Lactobacillus spp.	1	0.97	-	-	1	2.38	-	-	-	-
Gardnerella vaginals	3	2.92	3	5.36	-	-	-	-	-	-
Total	103	65.19	56	65.88	42	63.64	2	100	3	60
Escherichia coli	51	92.73	29	100	20	83.33	-		2	100
Pseudomonas aeruginosa	1	1.82	-	-	1	4.17	-		-	
Klebsiella pneumonia	3	5.45	-	-	3	12.5	-		-	
Total	55	34.81	29	34.12	24	36.36	-		2	40
Total Summation	158		85		66		2		5	

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The study showed that there is quite one microorganism cause concerned in infections, as quite one microorganism sort was isolated at constant website of infection, and this relies on the sort of infection and therefore the immune standing of the host yet because the impact that some pathogens wear the organs, yet because it was observed that the quantity of infections triggered by Gram-positive microorganisms was higher compared to infections produced by Gram-negative microorganism , as shown in Table (1,3). This might result to their ability to secrete some toxins and enzymes, and this plays a job in resisting bodily process additionally to its presence naturally within the organ. Justify the explanation for this as a result of these gram-positive microorganism area unit naturally found within the bodily cavity region and their presence facilitates the method of their invasion [15].

Table (3): Number & percent of bacteria isolates.

Bacterial isolated	***Number & Percent of bacteria isolates***	
	Number	***%***
Staphylococcus spp	*79*	*50*
Escherichia coli	*51*	*32.28*
Streptococcus spp	*18*	*11.39*
Klebsiella pneumonia	*3*	*1.89*
Gardnerella vaginals	*3*	*1.89*
Enterococcus fascial	*2*	*1.27*
Lactobacillus spp.	*1*	*0.64*
Pseudomonas aeruginosa	*1*	*0.64*
Culture isolated	*158*	
Non-culture isolated	*15*	
Total	*173*	

Results in Table 3 showed samples cultures revealed 158 had bacterial isolates from 173 pateints (while 15 non- culture isolate). The most predominant pathogen was *Staphylococcus spp* (*n* = 79 , 50 %), the second most important microorganism which was isolated *Escherichia coli* (*n* = 51 , 32.28%) followed by both *Klebsiella pneumonia* and *Gardnerella vaginals* (*n* = three , 1.89%), followed by *Enterococcus fascial* (*n* = 2, 1.27 %), *Lactobacillus spp.* (*n* = one, 0.64%) and

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Pseudomonas aeruginosa (*n = one , 0.64%)* isolated from study groups (vaginal swab, urine, blood samples, ear swab, seminal fluid, synovial fluid, throat swab, genital lesion swab and sputum.

High rate resistance of antibiotics of *E. coli* is to cephalexin (15/15), amoxicillin (12/12) metronidazole (6/6), clarithromycin (11/11) clindamycin (32/37), metronidazole (6/6), Rifampin (37/46), naldixic acid (28/35), spiramycin (6/6)), clavulanic acid (6/6), and high rate of susceptibility to levoflaxin (23/25), followed gentamycin (22/45). High rate resistance of *Staphylococcus spp* is to ceftriaxone (33/33), cefatoxime (36/45), nalidixic acid (23/28), Erythromycin (23/29), metronidazole (31/31), trimethoprim (20/26) ofloxine (23/33) , lincomycin (45/59) other in Table-1, and susceptible in high rate to imipenem (31/31) , vancomycin (25/33) , meropenim (27/35) , Amikacin (24/32) .The high resistance rate of *streptococcus spp* is to Naldixic acid (11/13), ceproflaxin (16/19), ceftriaxone (11/12), lincomycin (11/14), norofloxacin (14/18), trimethoprim (12/16) and the high rate susceptibility is to nitrofurantoin (12/18), followed by Rifampin (10/19). *Lactobacillus spp is* resistance to nalidixic acid, levoflaxin, lincomycin, streptomycin in high rate, and susceptible to nitrofurantoin, tetracycline, streptomycin in high rate. *Pseudomonas aeruginosa* susceptible to amikacin, azithromycin, ceproflaxin, gentamycin, imipenem, levoflaxin, meropenim and resistance to amoxicillin , tetracycline , metronidazole , trimethoprim *klebsiella pneumonea* resistance to amoxicillin , tetracycline , Erythromycin, metronidazole in high rate , and susceptible to meropenim , levoflaxin , nitrofurantoin, in high rate .*Gardnerella vaginals* resistance to cephalexin , metronidazole, tetracycline in high rate , and susceptible to ceproflaxin , Azithromycin , levoflaxin, imipenem , vancomycin .*Enterococcus faecalis* has high resistance to amoxicillin , ampicillin , metronidazole, streptomycin , and susceptible to imipenem , levoflaxin, Rifampin , nitrofurantoin in high rate Table.4 .

Table (4): Antimicrobial susceptibility of some bacterial isolates.

Antibiotic	***Staphylococcus spp***			***Escherichia coli***			***Streptococcus spp***			***Lactobacillus spp***		
	S(%)	I(%)	R(%)	S(%)	I(%)	R(%)	S(%)	I(%)	R(%)	S(%)	I(%)	R(%)
Imipenem	31/31	---	---	3/7	4/7	----	4/5	---	1/5	---	---	---
clavulanic acid	8/31	11/31	12/31	-----	---	6/6	---	---	1/1	---	---	---
Levofloxacin	31/56	10/56	15/56	23/25	2 /25	10/25	7/13	2/13	4/13		1/2	1/2
Rifampin	30/70	7/70	33/70	8/46	1/46	37/46	10/19	1/19	8/19	1/2	1/2	
Amoxicillin	7/52	12/52	35/52	----	-----	12/12	---	3/5	2/5	---	---	---
Cefotaxime	3/45	6/45	36/45	-----	-----	6/6	---	1/3	2/3	---	---	---
Naldixic acid	3/28	2/28	23/28	3/35	4/35	28/35	1/13	1/13	11/13			2/2
Ciprofloxacin	24/83	14/83	45/83	15/52	17/52	20/52	2/19	1/19	16/19			2/2
Azithromycin	2/29	11/29	16/29	----	2/6	4/6	3/5	2/5	1/5	---	---	---
Clarithromycin	16/73	3/73	44/73	----	----	11/11	2/5	1/5	3/5	---	---	---
Clindamycin	13/67	10/67	44/67	4/37	1/37	32/37	5/12	2/12	5/12			1/1
Erythromycin	2/29	4/29	23/29	---	---	6/6	2/5	1/5	2/5	---	---	---

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Ofloxine	10/33	----	23/33	7/23	2/23	14/23	1/7	---	6/7	1/2		1/2
Gentamycin	28/70	16/70	26/70	22/45	9/45	14/45	3/17	4/17	10/17	---	---	---
Doxycline	11/20	1/20	8/20	------	2/2	__	2/2	---	---	---	---	---
Lincomycin	11/59	3/59	45/59	2/18	----	16/18	2/14	1/14	11/14	---	---	2/2
Vancomycin	25/33	5/33	3/33	-----	----		5/6	1/6	---	---	---	---
Meropenem	27/35	7/35	1/35	14/15	-----	1/15	4/5	1/5	---	---	---	---
Metronidazole	---	---	31/31	----	----	6/6	---	---	5/5	---	---	---
Nitrofurantoin	41/70	4/70	2570	15/45	15/45	1545	12/18	5/18	1/18	2/2	---	---
Norfloxacin	23/70	7/70	40/70	20/43	----	2343	2/18	2/18	14/18	---	1/2	1/2
Ampicillin	6/31	5/31	20/31	----	----	6/6	2/4	2/4	---	---	---	---
Spiramycin	1/29	10/29	18/29	----	----	6/6	1/5	1/5	3/5	---	---	---
Streptomycin	22/55	6/55	27/55	10/25	1/25	14/25	5 /11	1/11	5/11	---	---	2/2
Tetracycline	5/12	1/12	6/12	2/5	1/5	3/5	---	---	2/2	---	---	---
Trimethoprim	18/50	3/50	39/50	19/45	3/45	23/45	2/16	2/16	12/16	2/2		
Trimethoprim / sulphamethoxazole	4/26	2/26	20/26	----	1/6	5/6	---	---	4/4	---	---	---
Amikacin	24/32	----	8/32	16/33	----	19/33	8/13	3/13	2/13	---	---	1/1
Ceftriaxone	---	----	33/33	---	---	8/8	--	1/12	11/12	---	---	---
Cephalexin	---	----	26/33	---	---	15/15	2/8	1/8	5/8	-	---	2/2

S (sensitive); I (intermediate); R(resistant)

At current, Antibiotic resistance thought-about a worldwide health alternative, Multiple treatment resistance bacterium area unit on growth and more at the time changing into a vital downside of all inhabitants, so area unit being concerned in Increased morbidity among patients. infection with resistant species that work for a long stay in hospitals; Immunocompromised individuals, combined with exposure to multiple antibiotics, are the most common risk factors for infection and multi-antibiotic resistance. Antibiotic resistance appears in varied bacterium from completely divers samples is related to significant Negative results [16]. *staphylococcus spp* is the most prevalent bacteria isolated from vaginal swab (% 49.41) which is released and in urine (%51.51). The increase in resistance has a abundant impact on the cheap of both developed and emerging countries as it is most likely to affect the lab our force through mortality and morbidity.

Capacity of *Staphylococcus* spp and *Escherichia coli* to form biofilm can be indirect by phenotypic characteristic. In table 5, presented the 18 isolates of *Staphylococcus* spp were strong biofilm producers, while 6 isolates were moderately productive and one isolate was weak or non-productive. *Escherichia coli* Fourteen isolates were of strong productivity, 9 of medium production, and two isolates of non- or low-production.

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Table (5): Number and percentage of biofilm form by *Staphylococcus spp and Escherichia coli* detection by M.T.P.

Isolated Bacterial Number (25)	biofilm formation	*Staphylococcus* spp. NO. (%)	*E. coli* NO. (%)	Total NO. (%)
	High	18(72)	14(56)	32(64)
	Moderate	6(24)	9(36)	15(30)
	Weak / none	1 (4)	2(8)	3(6)
	Total	25(100)	25(100)	50(100)

Biofilm development may be a key consider the institution and resolve of coccus infections in humans, animals, and on medical devices, the flexibility to make biofilms on plastic devices is a crucial virulence issue for microorganism.

Conclusions

It was concluded from this study that bacteria isolated from different pathogens from different clinical sources, were Gram-positive bacteria more frequent than Gram-negative bacteria. and that men are more susceptible to infection than women. In addition to that the bacteria that, form the biofilm are more resistant to antibiotics.

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