Antibacterial resistance of burn infections in Al-Hussain Teaching Hospital/Thi-Qar Province

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Abstract:

Burn infection is a main cause of morbidity and mortality. Burn patients are more susceptible to get infections in comparison with other patients. This study has been conducted to determine bacterial pathogens frequently causing burn infections and their antimicrobial resistance patterns.

This study was carried out on thirty patients admitted to the burn unit at Al-Hussain Teaching Hospital/Thi-Qar Province during a five-month period from August 2015 to December 2015. A swab has been taken from each patient. Antibiotic sensitivity was performed by the disc diffusion method. Tested antibiotics used in this study were ampicillin, augmentin, ciprofloxacin, cefepime, ceftazidime, ceftriaxone, gentamicin, imipenem, tobramycin, vancomycin, levofloxacin, piperacillin, ticarcillin-clavulanic acid, netlimicin, amikacin, chloramphenicol, ticarcillin, , tetracycline, Co-Trimethaxazole and rifampicin.

Thirty four bacterial isolates were obtained from thirty swabs. We found that the most predominant bacterial isolate was *Pseudomonas*spp. (49%), followed by *Klebsiella* spp. (21%), *Staphylococcus*spp. (12%), Escherichia coli (*E.coli*) (6%), *Proteus* spp. (6%), *Acinetobacter* spp. (3%) and *Enterococcus* spp. (3%). Polymicrobial infection was obtained in (13.3%) of patients.

All isolated pathogens were multi-drug resistant.*Acinetobacter* spp. and *Enterococcus* spp. were the most resistant pathogens (100%), followed by *E. coli* (90.47%), *Staphylococcus*spp. (79.17%) , *Klebsiella* spp. (68.75%), *Proteus* spp.(68.75%) and *Pseudomonas* spp. (65.88%). Gram-positive bacteria showed complete resistance against ampicillin, while Gram-negative bacteria showed high resistance against augmentin, cefepime, ceftriaxone, tobramycin ,piperacillin, ticarcillin-clavulanic acid, amikacin, chloramphenicol, and tetracycline.

Non-significant difference was found among the tested antibiotics concerning resistance (P value >0.005). However ciprofloxacin, gentamicin and imipenem were significantly effective against most types of pathogens compared to all tested antibiotics (P value <0.005).

In conclusion *Pseudomonas* spp. were the most predominant pathogen. All isolated pathogens were multi-drug resistance which is an emerging problem. We need more periodic

studies to evaluate bacterial resistance from time to time to help in treatment policy.

Keywords: Burn infection, antibacterial resistance.

المقاومة للمضادات البكتيرية لعدوى الحروق في مستشفى الحسين التعليمي/محافظة ذي قار

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الخلاصة:

عدوى الحروق هي سبب رئيسي للمرض والوفيات. مرضى الحروق هم أكثر عرضة للعدوى مقارنة مع المرضى الآخرين. تم إجراء هذه الدراسة من اجل تحديد نمط البكتيريا المسببة غالبا لعدوى الحروق وأيضا لتقييم المقاومة للمضادات البكتيرية.

تم اجراء هذه الدراسة على (٣٠) مريض في ردهة الحروق في مستشفى الحسين التعليمي / محافظة ذي قار لمدة خمسة أشهر ابتداءا من **آب**٢٠١٥ ولغاية كانون الأول ٢٠١٥. المضادات الحيوية التي تم اختبارها في هذه الدراسة كانت الامبيسيلين، الاوكمنتين،السيبروفلوكساسين،السيفيبيم، السيفتازديم، السيفترياكسون، الجنتامايسين، الامبنيم، التوبرامايسين، الفانكومايسين، الليفوفلوكساسين، البيبراسيلين،التيكارسيلين- حامض الكلافولنك، النتيلمايسين، الأميكاسين،الكلورامفينيكول، التيكارسيلين، التتراسايكلين، الكوترايموكسازول والريفامبسين.

تم الحصول على (٣٤) عزلة من أصل (٣٠) مسحه. وجد أنالزائفة كانت المسبب الرئيسي لعدوى الحروق (٤٩٪). فيما وجد ان الكلبسيله مسؤولة عن (٢١٪)،المكورات العنقودية(١٢٪)،الاشريكية القولونية (٦٪)،المتقلبة (٦٪)، الراكدة (٣٪) والمكورات المعدية (٣٪). وقد تم الحصول على عدوى متعدد المكروبات في (١٣.٣٪) من المرضى. كل أنواع البكتريا كانت مقاومة لأدوية متعددة. الراكدة و والمكورات المعدية كانت البكتريا الأكثر مقاومة (١٠٠%)،تليها الاشريكية القولونية(٩٠.٤٧٪)،المكورات العنقودية (٧٩.١٧٪)،الكليبسيله (٦٨.٧٥٪)،المتقلبة (٦٨،٧٥٪) والزائفة (٦٥.٨٨٪). البكتيريا الإيجابية الجرام أظهرت مقاومة تامة ضد الأمبيسلين، بينما البكتيريا السلبية الجرام أظهرت مقاومة عالية ضد الاوكمنتين،السيفيبيم،السيفترياكسون،التوبرامايسين،البيبراسيلين،التيكارسيلين- حامض الكلافولنك،الأميكاسين،الكلورامفينيكولوالتتراسايكلين.لم يكن هناك فرقمهم إحصائياً بين المضادات الحيوية التي تم اختبارها فيما يتعلق بالمقاومة (P>0.005) ومع ذلك كان السيبروفلوكساسين، الجنتامايسينوالامبنيم فعالة بشكل كبير(P>0.005) ضد معظم أنواع البكتريا بالمقارنة مع جميع المضادات الحيوية التي تم اختبارها.

نستنتج من هذه الدراسة أن الزائفة كانت المسبب الرئيسي لعدوى الحروق وكانت كل أنواع البكتريا مقاومة لأدوية متعددة لذلك نحن بحاجة إلىا لمزيد من الدراسات الدورية لتقييم المقاومة البكتيرية من وقت لآخر.

الكلمات المفتاحية: عدوى الحروق، المقاومة للمضادات البكتيرية.

Introduction:

Burn can be defined as a tissue damage caused by electrical or chemical agents(1). Burn infection is a main cause of morbidity and mortality yet to be managed and highlighted(2, 3). Burn patients are more susceptible to get infections in comparison with other patients because burned skin provides fertile media for bacterial growth and part of the trauma response leads to immune suppression, in addition burn patients stay longer period at hospital than other patients (4). Severity of infection is highly variable which ranges from asymptomatic infection to a fatal bacteremia (5). In spite of the major progression of wound care or management at the clinical practice, infection still stand as a challenge of burn management and major source of burden(6). Mortality rate figures are high in burn centers which are linked to the age and burn percent, however 73 % of dead out of burn is due to septic process in the first 5 years after burn injury (7).

Type of bacterial infection is varied from a hospital to another; and variations of type and virulence can occur at the same hospital depending on the stages of wound healing which might change bacterial type markedly(8). Urinary tract infection, pulmonary infections, bacteremia and sepsis are the most common infections in burn patients(2, 9). *Staphylococcus aureus*, Coagulase-negative staphylococci, *Pseudomona saeruginosa*, *E. coli*, *Klebsiella pneumoniae*, *Proteus* spp., *Enterobacter* spp. and *Acinetobacter* spp. are the most common types in burn wound infections(4, 10). The efficacy of commonly used antimicrobial agents in burn units is dynamic due to the ability of micro-organisms to develop resistance quickly(2, 11). The emergence of resistance to antibiotics at burn units left treatment with limited options and consequently, this resistance can be considered as a leading cause of mortality for burn patients(12-14). Numerous types of bacteria have been reported for wounds in burn patients, unfortunately the dominant type of these pathogens are multidrug resistance microbes which are attributed for nosocomial outbreaks at the same time (4, 15).

Materials and methods:

This study was carried out in the burn unit of Al-Hussain Teaching Hospital from August 2015 to December 2015. Thirty wound swabs were taken from patients and transported to the bacteriological laboratory. Medical history about previous use of broad spectrum antibiotics during the last year was taken from patients 'medical records and a questionnaire asked to the patients themselves or their relatives. Swabs were cultured on Blood agar, MacConkey agar and Nutrient agar at 37°C for (24-48) hours. Morphological, microscopical and biochemical characteristics of bacterial isolates have been studied according to the correlated references(16, 17) . For identification of isolates, API 20E kit (BioMeriux) has been used. Antimicrobial susceptibility was performed on Mueller-Hinton agar by the standard disc diffusion method according to the Clinical Laboratory and Standards Institute Guidelines (18). The tested antibiotics were ampicillin, augmentin, ciprofloxacin, cefepime, ceftazidime , ceftriaxone, gentamicin, imipenem, tobramycin, vancomycin, levofloxacin, piperacillin, ticarcillin clavulanic acid , netlimicin , amikacin, chloramphenicol, ticarcillin, tetracycline, Co-Trimethaxazole and rifampicin.

Statistics:

The collected data were analyzed by using SPSS, applied Chi square and one-way ANOVA test.

Results:

This study was carried out on thirty patients, 11 male patients (36.7%) and 19 female patients (63.3%). The age of patients ranged between (1.5 to 55 years). A total number of (30) samples were processed and (34) bacterial isolates were obtained. The most predominant bacterial isolate was *Pseudomonas* spp. (49%), followed by *Klebsiella* spp. (21%), *Staphylococcus* spp. (12%), *E.coli* (6%), *Proteus* spp. (6%),*Acinetobacter* spp. (3%) and *Enterococcus* spp. (3%) as shown in (Figure 1). Monomicrobial infection was obtained from(26) patients (86.7%), while polymicrobial infection was obtained from(4)patients(13.3%).Mixed infections caused by *Pseudomonas* and *Klebsiella* spp. accounted for (75%) of mixed infections, while *Proteus* spp. and *Klebsiella* spp. accounted for (25%) as shown in (Figure 2).

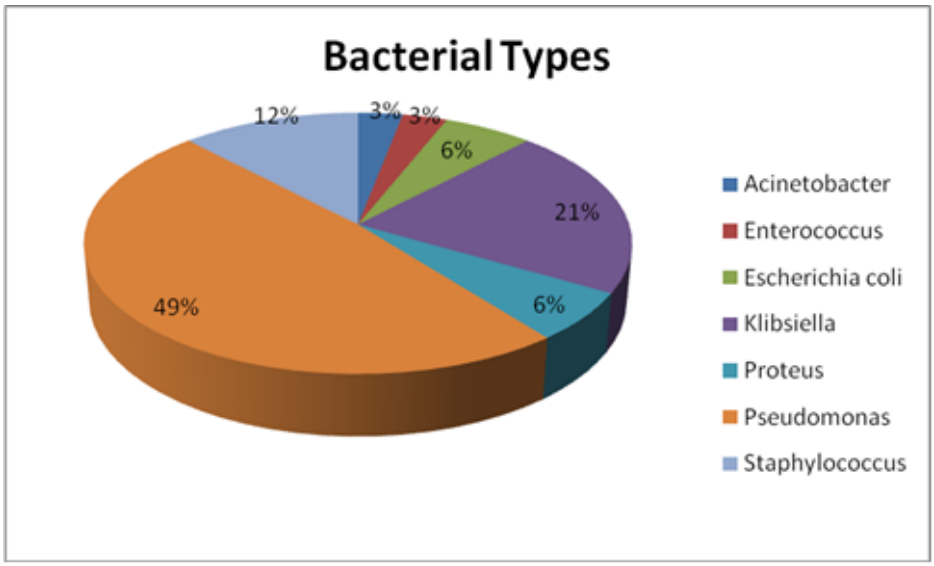

Figure 1. Frequency of the isolated bacterial strains.

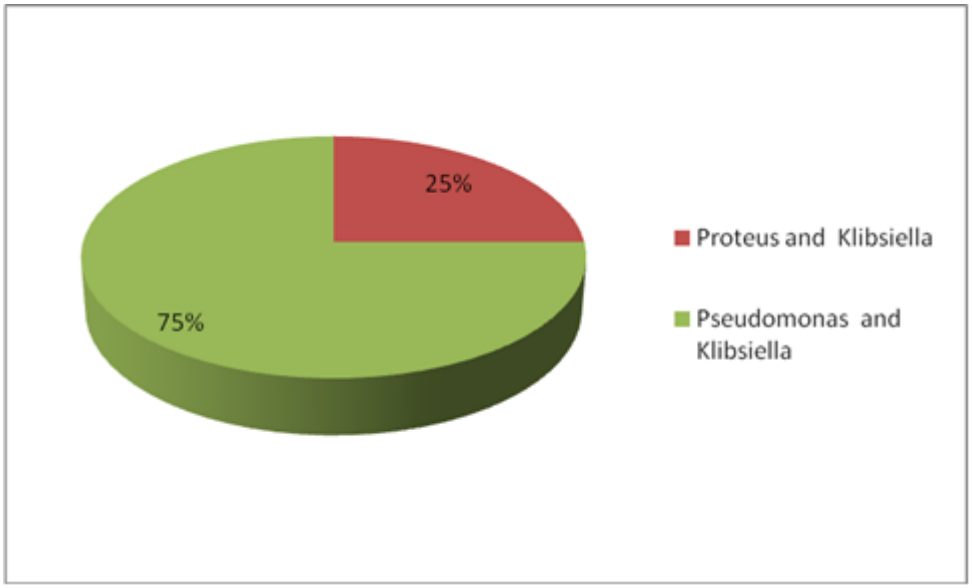

Figure 2. Frequency of mixed bacterial strains in mixed infections.

Gram-positive bacteria like *Staphylococcus* spp.showed complete resistant to ampicillin, ciprofloxacin, imipenem, tetracycline, Co-Trimethaxazole and rifampicin with intermediate resistance against gentamicin, vancomycin, netlimicin and chloramphenicol. *Enterococcus* spp.were completely resistant to ampicillin, vancomycin, chloramphenicol and rifampicin(Table 1).

Gram-negative bacteria like *Pseudomonas* spp. showed complete resistant to tobramycin, levofloxacin and ticarcillin-clavulanic acid with intermediate resistance against ciprofloxacin and imipenem. *Klebsiella* spp. Showed complete resistant to augmentin, gentamicin, amikacin, chloramphenicol and tetracycline with intermediate resistance against ciprofloxacin. *Proteus* spp. were completely resistant to ceftazidime, amikacin, chloramphenicol and tetracycline with intermediate resistance against augmentin, ciprofloxacin, ceftriaxone, gentamicin,

imipenem and Co-Trimethaxazole. *E.coli* showed complete resistant to augmentin, cefepime, ceftriaxone and tobramycin with intermediate resistance to gentamicin, piperacillin, amikacin, chloramphenicol, tetracycline and Co-Trimethaxazole. *Acinetobacter* spp. were completely resistant to ciprofloxacin, cefepime, ceftazidime, ceftriaxone, gentamicin, imipenem, tobramycin, levofloxacin, piperacillin, ticarcillin clavulanic-acid, amikacin, ticarcillin, tetracycline and Co-Trimethaxazole. (Table 2).

Acinetobacter spp. and *Enterococcus* spp. were the most resistant pathogens (100%), followed by *E. coli* (90.47%), *Staphylococcus* spp. (79.17%), *Klebsiella* spp. (68.75%), *Proteus* spp.(68.75%) and *Pseudomonas* spp. (65.88%) (Figure 3).

All tested antibiotics showed non-significant difference regarding resistance by comparing each one with the others (P value >0.005). However, ciprofloxacin, gentamicin and imipenem were significantly effective against most types of pathogens compared to all tested antibiotics (P value <0.005) (Figure 4,5,6)

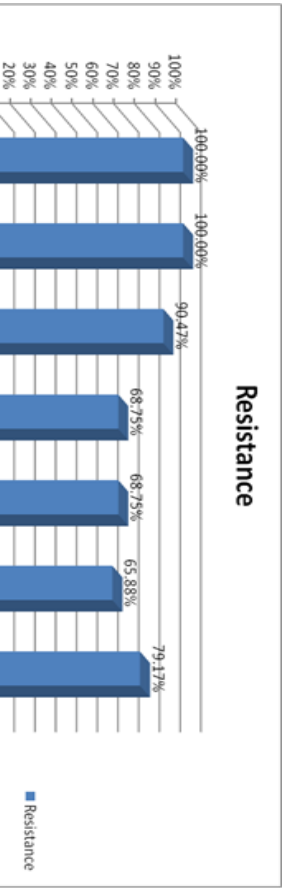

Table 1. Antibacterial resistance among Gram-positive bacteria

Isolates	AMP	CIP	GEN	IPM	TOB	VA	NET	AK	C	TE	COT	RA
Staphylococcus spp.	100%	100%	50%	100%	75%	50%	50%	75%	50%	100%	100%	100%
Enterococcus spp.	100%	ND	ND	ND	ND	100%	ND	ND	100%	ND	ND	100%

AMP:ampicillin; CIP:ciprofloxacin; GEN:gentamicin; IPM:imipenem; TOB:tobramycin; VA:vancomycin; NET:netlimicin; AK:Amikacin; C:chloramphenicol; TE:tetracycline; COT:Co-Trimethaxazole; RA:rifampicin; ND:note done.

Table 2. Antimicrobial resistance among Gram-negative bacteria

Isolates	AMC	CIP	FEP	CAZ	CTR	GEN	IPM	TOB	LEV	PI	TCC	NET	AK	C	TI	TE	COT
Pseudomonas spp.	ND	50%	93.8%	87.5%	ND	81.3%	62.5%	100%	100%	75%	100%	93.8%	81.3%	ND	87.5%	ND	ND
Klibsiella spp.	100 %	57.1%	ND	ND	85.7%	100%	85.7%	85.7%	ND	ND	ND	ND	100 %	100%	ND	100%	85.7%
Proteus spp.	50 %	50 %	ND	100 %	50%	50 %	50.0%	ND	ND	ND	ND	ND	100 %	100%	ND	100%	50 %
E. coli	100 %	ND	100 %	ND	100%	50 %	ND	100%	ND	50%	ND	ND	50%	50 %	ND	50 %	50 %
Acinetobacter spp.	ND	100%	100%	100%	100%	100%	100%	100%	100%	100%	100%	ND	100%	ND	100%	100%	100%

AMP:ampicillin; AMC:augmentin; CIP:ciprofloxacin; FEP:cefepime; CAZ:ceftazidime; CTR:ceftriaxone; GEN:gentamicin; IPM:imipenem; TOB:tobramycin; VA:vancomycin; LEV:lovofloxacin; PI: piperacillin; TCC: ticarcillin-clavulanic acid; NET:netlimicin; AK:Amikacin; C:chloramphenicol; TI: ticarcillin; TE:tetracycline; COT:Co-Trimethaxazole; RA:rifampicin; ND:note done.

Figure 3. Frequency of resistance of gram-positive and gram-negative bacteria isolated from burn wounds.

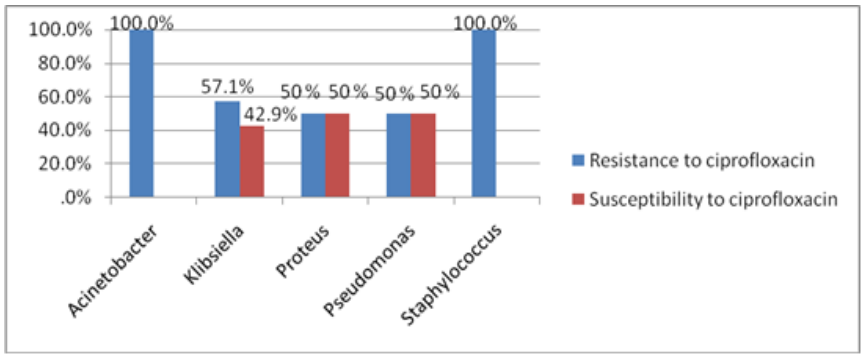

Figure 4. Evaluation of susceptibility and resistance to ciprofloxacin.

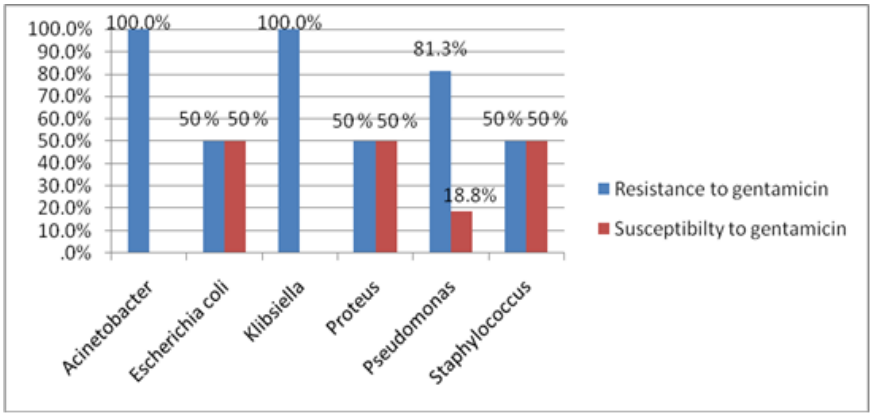

Figure 5. Evaluation of susceptibility and resistance to gentamicin.

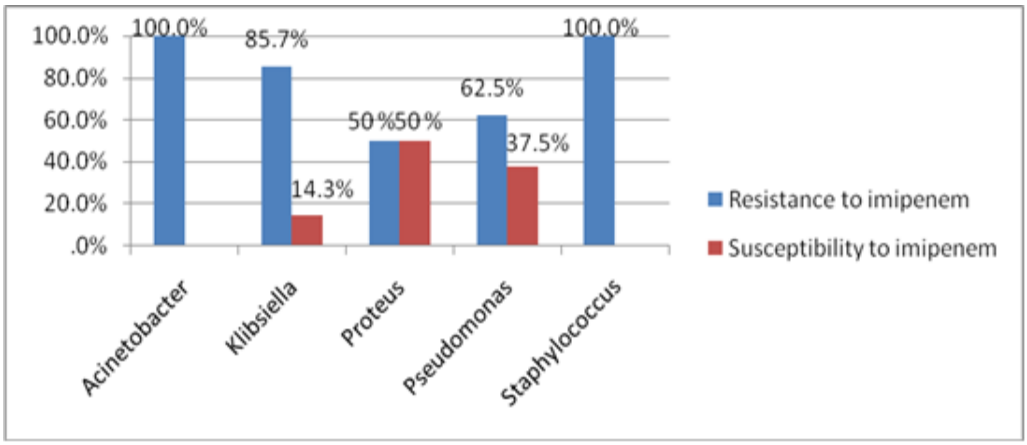

Figure 6. Evaluation of susceptibility and resistance to imipenem.

From patient history, it has been found that (36.36%) of patient used broad spectrum antibiotics last year, while (63.64 %) did not (Figure 7).

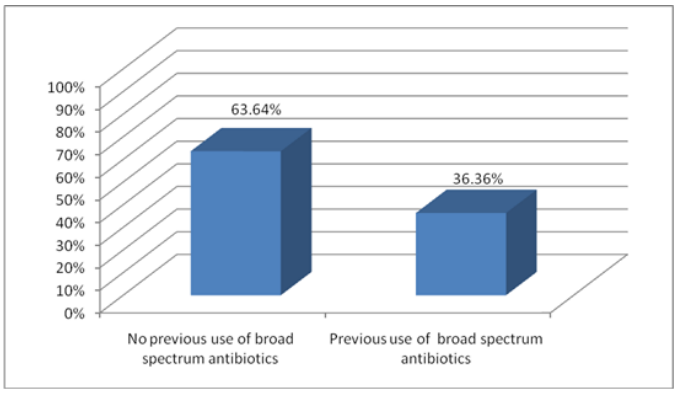

Figure 7. Percentage of patients regarding previous use of broad spectrum antibiotics.

The broad spectrum antibiotics used previously were cefotaxim (58.3%), ampiclox (25%) and ceftriaxone (16.6%) (Figure 8).

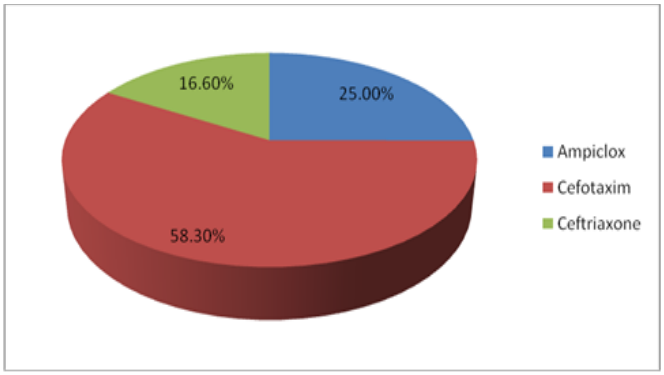

Figure 8. Percentage of broad spectrum antibiotics previously used.

By comparing the resistance to each antibiotic with other tested antibiotics, there was significant difference in resistance to gentamicin in patients who have history for previous use of broad spectrum antibiotics in the last year compared to those who have not (P value <0.005) (Figure 9).

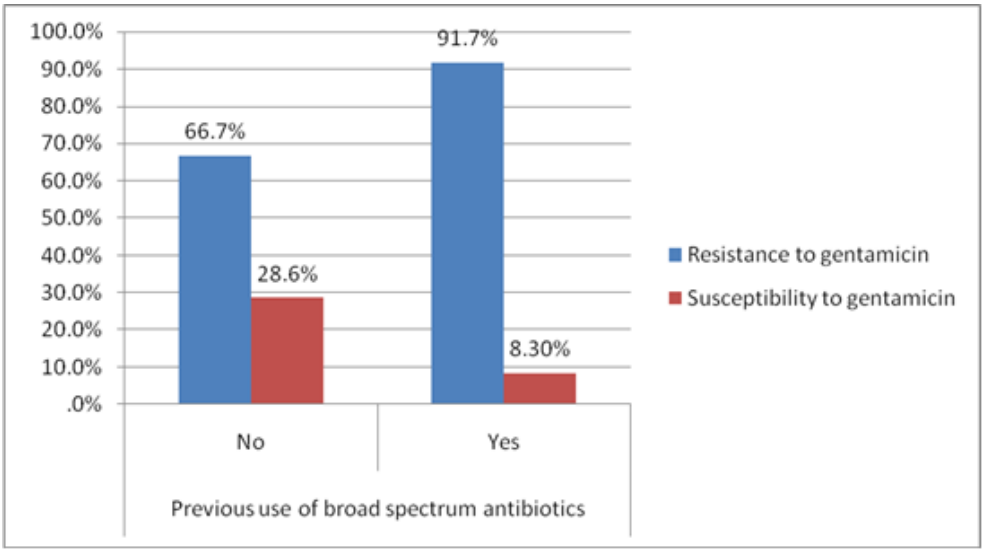

Figure 9. Evaluation of susceptibility and resistance to gentamicin regarding previous use of broad spectrum antibiotics.

Discussion :

Burn patients are more liable to get infections in comparison with other patients because of their damaged skin barrier and suppressed immune system, in addition to extended hospital stay and invasive therapeutic and diagnostic procedures (11).The most common pathogen isolated from burn wounds in our study was Pseudomonas spp. (49%), followed by Klebsiella spp. (21%), Staphylococcus spp. (12%), E.coli (6%), Proteus spp. (6%), Acinetobacter spp. (3%) and Enterococcus spp. (3%). Pseudomonas spp. were the most predominant isolate which is consistent with other studies conducted in Sulaymaniyah Province of Iraq (19), India (20) Bangladesh (21) and Iran (22). However, it contrasts with some studies done in Nigeria, India, Egypt and Yemen where it has been reported that Staphylococcus aureus as the most common organism (23-26(

Acinetobacter spp. were one of the two pathogens that has the least frequency among the isolates, which is incompatible with a study done in Turkey which showed that Acinetobacter spp. were the most predominant bacterial isolate (27). This difference in results coincides with the fact that the spectrum of infective agents varies from time to time and from place to place (28(

Acinetobacter spp. and Enterococcus spp. were the most resistant pathogens (100%), followed by E. coli (90.47%), Staphylococcus spp. (79.17%), Klebsiella spp. (68.75%), Proteus spp.(68.75%) and Pseudomonas spp. (65.88%). Pseudomonas spp. were the most

sensitive pathogens. Our finding concerning Acinetobacter spp. is consistent with a study conducted in Egypt that showed Acinetobacter spp. as the most resistant pathogen (24). However, non- significant difference was found concerning resistance to each antibiotic.

Staphylococcus spp. were completely resistant to ciprofloxacin, imipenem and tetracycline. In comparison with a study done also in the burn unit at Al-Hussain Teaching Hospital/Thi-Qar Province, Staphylococcus spp. showed high resistance to the above mentioned antibiotics (29). Our study showed that Enterococcus spp. were completely resistant to ampicillin and vancomycin which is inconsistent with a study conducted by Bayram et al. (27), where Enterococcus spp. were completely susceptible to the above mentioned antibiotics. Pseudomonas spp. were completely resistant to tobramycin and highly resistant to cefepime, ceftazidime, gentamicin, piperacillin and amikacin with intermediate resistance to ciprofloxacin and imipenem. The same pattern of resistance against ceftazidime and gentamicin has been reported in a study conducted in Iraq/Sulaimaniyah Province (30). In comparison with a study done by Dash et al. (20), Pseudomonas spp. were highly resistant to ceftazidime which is similar to our result, but with intermediate resistant to gentamicin, piperacillin, amikacin and tobramycin and high susceptibility to imipenem which is incompatible with our findings. Our results were also inconsistent with another study, where it has been reported that Pseudomonas spp. have low resistance against ciprofloxacin, ceftazidime, gentamicin and amikacin (27). Our findings showed that Klebsiella spp. were completely resistant to augmentin, gentamicin, amikacin, chloramphenicol and tetracycline, and highly resistant to imipenem and ceftriaxone with intermediate resistance to ciprofloxacin. Melake et al. reported similar findings concerning resistance to amikacin and ciprofloxacin (31). However, in the same mentioned study (31), the low resistance to imipenem and chloramphenicol was inconsistent with our results. Our results were in agreement with a study conducted in Egypt which revealed that Klebsiella spp. were completely resistant to augmentin and gentamicin (24), while complete resistance has been reported against ceftriaxone which is inconsistent with our finding. In our study, Proteus spp. were completely resistant to chloramphenicol and tetracycline with intermediate resistance against augmentin, ciprofloxacin, gentamicin and imipenem. In comparison with a study done by Abbas et al. (24), it has been reported that Proteus spp. were completely resistant to tetracycline which is similar to our finding, but our results were inconsistent with the same mentioned study (24), where resistance was intermediate against chloramphenicol, high to augmentin and gentamicin, and low against ciprofloxacin and imipenem. In our study, E.coli was completely resistant to augmentin with intermediate resistance to gentamicin, amikacin, chloramphenicol and tetracycline. Similar finding of resistance pattern against gentamicin and amikacin has been reported by Magnet et al. (21). However in the same mentioned study (21), E.coli was completely susceptible to chloramphenicol and tetracycline

which is inconsistent with our results. Yasidi et al. noted that E.coli showed high resistance to augmentin (25), while in our study it was completely resistant. Our study revealed that Acinetobacter spp. were completely resistant to all tested antibiotics, ciprofloxacin, cefepime, ceftriaxone, gentamicin, imipenem, tobramycin, amikacin and tetracycline. Our results are in agreement with the results reported by Abbas et al. (24) which revealed that Acinetobacter spp. were also completely resistant to cefepime, ceftriaxone, gentamicin and tetracycline. However in the same mentioned study (24), Abbas et al. reported that Acinetobacter spp. have high resistance to ciprofloxacin but low resistance against imipenem which is inconsistent with our results. In another study, low resistance of Acinetobacter spp. against amikacin has been reported (27), which contrasts with our findings .

In our study, non- significant difference has been found concerning resistance to the tested antibiotics. However ciprofloxacin, gentamicin and imipenem were significantly effective against most types of pathogens compared to all tested antibiotics. Such a high frequency of resistance to antibiotics may be due to inappropriate use of antibiotics and the dispensing policy of antibiotics without a prescription in our community. It has been reported that the high frequency of multidrug resistant pathogens is probably due to empirical use of broad-spectrum antimicrobials before development of infection, extended duration or previous hospitalization and non-adherence to hospital antimicrobial policy (20.(

From patients history, (36.36%) of patients used broad spectrum antibiotics during the last year, while (63.64%) did not. The antibiotics that have been used were cefotaxim, ampiclox and ceftriaxone. Cefotaxim was the most widely used one (58.3%), followed by ampiclox (25%) and ceftriaxone (16.6%). By comparing resistance to antibiotics between patients who have previous history for using broad spectrum antibiotics and those who have not, significant difference was found concerning resistance to gentamicin in patients used broad spectrum antibiotics previously. Yasidi et al. has mentioned that the extensive use of antimicrobial agents for wide range of disease condition in the community because of their affordability and accessibility had encouraged the emergence of resistant strain (25), which is the similar reason in our community

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Conclusion and recommendations:

1- Burn patients were most commonly infected by Pseudomonas spp. followed by staphylococcus spp.

2- All isolated bacterial pathogens were multi-drug resistant, which is an alarming trend that could be a leading cause for mortality in burn patients.

3- Proper isolation of contaminated patients, sterilisation of equipments and awareness of the hygiene would significantly reduce contamination .

4- Periodic studies are necessary to evaluate bacterial resistance from time to time.

5- Once bacterial resistance is identified, this should be reported to health care professionals, clinicians and public health carers.

6- Different types of surveillance and audit should be conducted on regular basis by different specialists i.e. microbiologists, clinicians and pharmacists.

7- Establishment of "infection control team" which is a multidisciplinary team network where they can manage infection together by select of empirical treatment in case of local resistance.

8- Comparison of resistance both quality and quantity with the other local, national or international scales which mandate changes of antibiotic choice and/or policy on local level.

9- People should be educated to use antibiotics when necessary only as the aggressive unnecessary use of antibiotics could result in resistance .

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