Prevalence of Pathogenic Bacterial Isolates Infecting Wounds and their Antibiotic Sensitivity

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Abstract

Wound infection is one of the health problems that are caused by pathogenic bacteria and antibiotic resistance profiles of Gram negative bacteria increased treatment cost especially in diabetic patients. The objective of this work was to isolate, purification and identification of pathogenic bacteria from wound infection then determination of susceptibility testing against antibiotics. 41 isolated from wound infections were collected from microbiology laboratory of Zagazig University Hospitals and some private microbiology laboratory Cairo, Egypt. Bacterial isolates were identified by API 20E Enterobacteriaceae. These pathogenic bacterial isolates belong to these genera (Klebsiella, Enterobacter, Pseudomonas, Providencia, Serratia and Citrobacter). The frequency of the bacterial isolates was 24%, 20%, 14%, 12%, 6% and 6% of the bacterial pathogens isolates in this study respectively. Finally, the antimicrobial susceptibility was performed by disc diffusion method against 12 types of antibiotics covered all mode of action of bacteria.

Keywords: Pathogenic bacterial; Wound infection; Multidrug resistance; Susceptibility patterns; Gram negative bacteria

Introduction

Wounds infection by bacteria and resistance to common antibiotics are the common post-surgical and medical challenges. Wounds bacterial contamination are the common hospital acquired infections causing more than 80% of the mortality [1]. The most common bacterial genera infecting wounds are Enterococci, Escherichia, Pseudomonas, Klebsiella, Enterobacter, Proteus and Acinetobacter [2,3]. Wounds infection have been a recognized as the most critical problem especially in the presence of foreign materials that increases the risk of serious infection even with relatively small bacterial infection [4]. Nosocomial infection is usually higher in burn patients that correlate with other factors like nature of burn injury, age of patient, extent of injury and burn depth. Other microbial factors such as type, number of organisms, enzymes, toxins production, colonization of the burn wound site, systemic dissemination of the colonizing organisms, have a strong effect on severity of bacterial wound infection [5,6]. As well as, widespread using of vast groups of antibiotics together with the length of time causes a significant development of antibiotic resistance to wound infecting bacteria [7], that subsequently increase the complications and costs of treatment [8].

Thus, the objective of this work was to isolate and characterize various bacterial isolates infecting wounds, and determination of their susceptibility to various common antibiotics.

Material and Methods

Sample collections

Swabs from abscesses lesions or pus were collected from microbiology lab. It was cultivated immediately without delay. Each sample was inoculated into nutrient agar medium and McConkey agar media. The plate cultures were incubated for 24 h at 30-37°C [5] and the growth was observed thoroughly.

Identification of isolated pathogenic bacteria

Identification of Gram negative pathogenic bacterial isolates were carried out by API 20E Enterobacteriaceae (BioMérieux Co.). API system has been recognized as a rapid test for bacterial identification. The strips were inoculated by single colony in the suspension media and incubated at 35-37 °C for 18-24 h. The results were recorded based on the special chart, three tests were given a code number, the obtained seven digit numbers were expressed to the corresponding organisms regarding to the API index.

Antimicrobial susceptibility of the pathogenic bacterial isolates

The susceptibility of the recovered bacterial isolates to antibiotics was determined using disc diffusion assay, Kirby-Bauer method [9] as described in of guidelines of the National Committee for Clinical Laboratory Standards [10]. Disks of Amikacin 30 µg, Amoxicillin/Clavulanic 20/10 µg, Cefazidime 30 µg, Ciprofloxacin 5 µg, Colistinsulphate 10 µg, Cefotaxim 30 µg, Cefepime 30 µg, Nitrofurantoin 300 µg, Levofloxacin 5 µg, Rifampicin 5 µg, Sulphamethoxazole/Trimethoprim 25 µg and Tobramycin 10 µg.
Results and Discussion

Isolation, identification and prevalence of pathogenic bacterial isolates

Bacterial wound contamination is a serious problem in the hospital and the treatment of wound infections remain a significant concern for surgeons. The risk of developing wound infection depends on the number of bacteria colonies on the wound. The problem has been magnified due to the unrestrained and rapidly spreading resistance to the available array of antimicrobial agents. Fifty cases from wound swabs were collected from different types of wounds, cultured on nutrient and MacConkey media. Fifty bacterial isolates were isolated from wounds infection out of the total cases. These bacterial isolates were identified based on the API 20E system. From the results, there was 41 cases (82%) recorded as a positive bacterial growth and only 9 samples were recorded as negative bacterial growth. Gram positive bacteria already isolated in nutrient agar media but the majority for Gram negative bacteria so that used the Gram negative bacteria for application. The identified forty-one pathogenic bacterial isolates were belonging to sex genera (Table 1). These genera were Klebsiella pneumoniae (12 isolates; 24%) followed by Pseudomonas fluorescens (10 isolates; 20%), Providencia stuartii (7 isolates; 14%) and Enterobacter cloacae (6 isolates; 12%) and Serratia rubidaea and Citrobacter freundii (3 isolates; 6%). Similarly, high percentage of microbial growth was reported by Mama et al. 2014 [11], 91.6% of cases from wound infection out of the total cases. These bacterial isolates were identified to the genera (Table 1). These genera were Klebsiella pneumoniae (12 isolates; 24%) followed by Pseudomonas fluorescens (10 isolates; 20%), Providencia stuartii (7 isolates; 14%) and Enterobacter cloacae (6 isolates; 12%) and Serratia rubidaea and Citrobacter freundii (3 isolates; 6%). Similarly, high percentage of microbial growth was reported by Mama et al. 2014 [11], 91.6% of culture was positive microbial growth and 12.7% had no bacterial growth. Coincident results for K. pneumoniae (24%), P. fluorescens (20%), P. stuartii (14%) and E. cloacae (12%), S. rubidaea and C. freundii (6%) was reported [12]. Among the recovered isolates, Pseudomonas species were the most common isolates (48.9%) followed by Citrobacter spp (13.3%), Enterobacter spp (11.1%), Proteus vulgaris (6.6%), Klebsiella spp (2.2%) and Serratia rubida (2.2%) infecting wounds [12].

Table 1: The different species of bacteria isolated from wound infection.

| Bacterial Isolates          | Total No. | Percentage % |
|----------------------------|-----------|--------------|
| Klebsiella pneumonia        | 12        | 24%          |
| Pseudomonas fluorescens     | 10        | 20%          |
| Providencia stuartii        | 7         | 14%          |
| Enterobacter cloacae        | 6         | 12%          |
| Serratia rubidaea           | 3         | 6%           |
| Citrobacter freundii        | 3         | 6%           |

Antibiotic susceptibility of the pathogenic bacterial isolates

The antibiotic sensitivity of isolated bacterial strains was carried out by Kirby-Bauer disk diffusion assay against 12 antibiotics (Table 2). Out of the forty-one bacterial isolates, Pseudomonas fluorescens was a completely resistance to all the tested antibiotics. Five pathogenic bacterial isolates belong to the genera Klebsiella, Pseudomonas, Providencia, Enterobacter and Serratia showed a strong resistance to the tested antibiotics by about 91.67%. Seven bacterial isolates belong to Klebsiella pneumoniae (No.9), Pseudomonas fluorescens (No.16, 19, 21, 22) and Citrobacter freundii (No. 39, 40) showed 83.33% resistance to all tested antibiotics. Whereas four pathogenic bacterial isolates of Gram negative bacilli namely; K. pneumoniae (No.3,12), P. fluorescens (No.13) and E. cloacae (No.30) showing 75% resistance to the utilized antibiotic. Four bacterial isolates belonging to K. pneumoniae (No. 6), P. stuartii (No. 28,29) and E. cloacae (No. 34) showed 66.67% resistance to the different tested antibiotics. While three bacterial isolates belong to Gram negative bacilli namely K. pneumoniae (No. 7), P. fluorescens (No. 20) and P. stuartii (No. 24) showed 58.3% resistance to the tested antibiotics. Also, three pathogenic bacterial isolates K. pneumonia (No. 10), E. cloacae (No. 33) and C. freundii (No. 90) showed 50% resistance to the tested antibiotic. Three pathogenic bacterial isolates K. pneumoniae (No.4,5) and P. stuartii (No. 27) showed 41.67% resistance to the different tested antibiotics and six bacterial isolates namely K. pneumoniae (No. 8), P. fluorescens (No. 18), P. stuartii (No. 25,26) and S. rubidaea (No. 38) showed 33.33% resistance. In addition, K. pneumoniae (No. 11), E. cloacae (No. 35) and S. rubidaea (No. 37) showed 25% resistance to the tested antibiotics. While E. cloacae (No.32) and S. rubidaea (36) showed 16.67% antibiotic resistance.

Figure 1: Profile of antibiotics antibiotic susceptibility of Klebsiella pneumoniae isolates (Resistance, Intermediate and Sensitive).

The frequency antibiotic resistance of K. pneumoniae isolates was summarized in Figure 1. K. pneumoniae isolate No. 1 was only sensitive to Colistin Sulphate, with moderate resistant to the other experimented antibiotics, while, and isolate No. 2 of K. pneumoniae was sensitive to seven antibiotics (Amikacin, Amoxicillin\Clavulanic acid, Ciprofloxacin, Colistin Sulphate, Nitrofurantoin, Levofloxacin and Tobramycin); with moderate resistant to Ceftazidim, Isolate No. 3 of K. pneumoniae was sensitive to Colistin Sulphate and moderately resistant to Ciprofloxacin and Levofloxacin. K. pneumoniae (No.4) isolate was sensitive to Amikacin, Ciprofloxacin, Colistin Sulphate, Levofloxacin and Tobramycin with moderate resistance to Amoxicillin \Clavulanic acid and Nitrofurantoin. K. pneumoniae (No.5) isolate was sensitive to six types of antibiotics (Amikacin, Amoxicillin\Clavulanic acid, Ciprofloxacin, Colistin Sulphate, Levofloxacin and Tobramycin) and moderate resistant to Nitrofurantoin. K. pneumoniae (No.6) isolates was sensitive to Amoxicillin\Clavulanic acid, Ciprofloxacin and Levofloxacin) while K. pneumoniae (No.6) moderate resistant against Nitrofurantoin and K. pneumoniae (No.7) moderately resistant against Cefotaxim and Nitrofurantoin. K. pneumoniae (No.8) isolate was
sensitive to seven types of antibiotics (Amikacin, Cefazidim, Ciprofloxacin, Colistin Sulphate, Nitrofurantoin, Levofloxacin and Tobramycin) and moderately resistant against Amoxicillin/Clavulanic acid. While K. pneumoniae (No.9) isolate was sensitive only to Colistin Sulphate and moderately resistant against Cefepime. Also K. pneumoniae (No.10) isolate was sensitive to (Amikacin, Colistin Sulphate, Nitrofurantoin, Levofloxacin and Sulphamethoxazole/Trimethoprim) and moderately resistant against Ciprofloxacin. K. pneumoniae (No.11) isolate was sensitive to eight types of antibiotics (Amikacin, Cefazidim, Ciprofloxacin, Colistin Sulphate, Cefotaxim, Nitrofurantoin, Cefepime and Levofloxacin) while moderately resistant against Tobramycin. K. pneumoniae (No.12) isolate was sensitive to Colistin Sulphate and Nitrofurantoin with moderate resistant to Amikacin.

| Isolate No. | Species of bacteria | Mean inhibition zone (mm) | Reaction to antibiotics | % species Resistance (IR+R) |
|-------------|---------------------|----------------------------|-------------------------|---------------------------|
| 1           | Klebsiella pneumoniae | 6IR 7IR 6IR 6IR 13IR 6IR 6IR 6IR 7IR 6IR 6IR | susceptibility breakpoint AK ≥ 17 AMC ≥ 18 CAZ ≥ 18 CIP ≥ 21 CT ≥ 11 CTX ≥ 23 F ≥ 17 FEP ≥ 18 LEV ≥ 17 RO ≥ 20 SXT ≥ 16 TOB ≥ 15 | 91.6 |
| 2           | Klebsiella pneumoniae | 20S 19S 17IR 31S 13S 8IR 21S 10R 27S 12R 6IR 15S | 33.3 |
| 3           | Klebsiella pneumoniae | 6IR 6IR 6IR 19IR 13S 6IR 14IR 6IR 16IR 7IR 6IR 6IR | 75 |
| 4           | Klebsiella pneumoniae | 21S 17IR 10IR 25S 13S 6IR 16IR 10IR 21S 9IR 6IR 17S | 41.6 |
| 5           | Klebsiella pneumoniae | 23S 18S 11R 25S 12S 6IR 16IR 10IR 21S 9R 6IR 16S | 41.6 |
| 6           | Klebsiella pneumoniae | 6IR 18S 12R 22S 10R 6IR 16IR 11R 20S 7R 6IR 6IR | 66.6 |
| 7           | Klebsiella pneumoniae | 6IR 20S 14IR 22S 10R 16IR 15IR 7R 20S 7R 6R 6R | 58.3 |
| 8           | Klebsiella pneumoniae | 19S 14IR 20S 21S 17S 6IR 18S 12R 20S 9R 6R 20S | 33.3 |
| 9           | Klebsiella pneumoniae | 6IR 9R 6IR 6R 12S 13R 9R 17IR 6R 7R 6R 6R | 83.3 |
| 10          | Klebsiella pneumoniae | 19S 8R 6IR 18IR 12S 7IR 17S 10R 24S 9R 6IR 12R 50 | 50 |
| 11          | Klebsiella pneumoniae | 20S 9R 20S 25S 11S 25S 17S 21S 25S 6R 8R 14IR 25 |
| 12          | Klebsiella pneumoniae | 15IR 13S 14R 6R 12S 6IR 18S 6IR 7R 11R 6R 6R | 75 |
| 13          | Pseudomonas fluorescens | 10R 13IR 41R 15S 6IR 6R 20R 12R 18IR 6R 17S 75 |
| 14          | Pseudomonas fluorescens | 16IR 6R 10R 19IR 9R 6IR 6IR 14IR 6R 6R 6R 6R | 100 |
| 15          | Pseudomonas fluorescens | 11IR 17IR 6IR 6IR 13S 7R 8IR 7R 16IR 6R 6R 7R 6R 9R 6R 91.67 |
| 16          | Pseudomonas fluorescens | 19S 6R 6IR 16IR 14S 6IR 6IR 6IR 8R 11R 6R 10IR 83.33 |
| 17          | Pseudomonas fluorescens | 6IR 6IR 6IR 6IR 13S 6IR 15IR 6R 6R 7R 6R 6R 91.67 |
| 18          | Pseudomonas fluorescens | 18S 21S 17IR 26S 6IR 21IR 21S 18S 18S 29S 24S 12IR 33.33 |
| 19          | Pseudomonas fluorescens | 14R 6R 9R 10R 12S 6IR 6IR 9R 7R 9R 6R 15S 83.33 |
| 20          | Pseudomonas fluorescens | 20S 18S 21S 6R 11S 6IR 18S 9R 8R 10R 6R 11R 58.33 |
| 21          | Pseudomonas fluorescens | 6IR 6IR 6IR 6IR 12S 7R 17IR 6R 6R 7R 6R 6R 83.33 |
| 22          | Pseudomonas fluorescens | 6IR 6IR 12R 28S 6R 6IR 7R 9R 25S 6R 6R 6R 83.33 |
| 23          | Providencia stuartii | 15IR 16IR 6R 6IR 13S 6IR 9R 6IR 15IR 11R 6R 6R 91.67 |
| 24          | Providencia stuartii | 22S 6IR 6IR 30S 8IR 6IR 10R 6IR 34S 14IR 22S 18S 58.33 |
| 25          | Providencia stuartii | 20S 10R 18S 24S 18S 23S 9R 20S 21S 9R 17S 13IR 33.33 |
| 26          | Providencia stuartii | 18S 12R 26S 22S 6R 28S 8IR 25S 24S 11R 21S 19S 33.33 |
| 27          | Providencia stuartii | 22S 14IR 21S 30S 8IR 21IR 11R 25S 26S 8R 20S 16S 41.67 |
28 Providencia stuartii 19/S 7/R 6/R 25/S 13/S 7/R 16/R 13/R 22/S 6/R 8/R 13/R 66.67
29 Providencia stuartii 15/IR 6/R 6/R 15/S 7/R 19/S 10/R 22/S 6/R 6/R 7/R 66.67
30 Enterobacter cloacae 17/S 18/S 6/R 15/R 6/R 14/R 12/R 11/R 15/R 10/R 6/R 8/R 75
31 Enterobacter cloacae 6/R 6/R 6/R 12/S 6/R 6/R 6/R 6/R 6/R 6/R 8/R 91.6
32 Enterobacter cloacae 24/S 16/IR 24/S 17/IR 17/S 26/S 11/R 28/S 21/S 7/R 16/S 17/S 16.6
33 Enterobacter cloacae 6/R 17/IR 32/S 18/R 16/S 6/R 6/R 6/R 22/S 12/R 18/S 8/S 50
34 Enterobacter cloacae 13/R 6/R 16/IR 15/R 14/S 6/R 6/R 12/R 12/R 17/R 6/R 15/S 66.6
35 Enterobacter cloacae 18/S 6/R 20/S 16/R 15/S 16/R 6/R 22/S 16/R 17/R 6/R 18/S 25
36 Serratia rubidaea 23/S 18/S 32/S 22/S 14/S 34/S 29/S 30/S 24/S 14/R 14/R 15/S 16.6
37 Serratia rubidaea 21/S 24/S 25/S 19/IR 13/S 23/S 17/S 30/S 18/S 8/R 6/R 20/R 25
38 Serratia rubidaea 23/S 22/S 29/S 20/IR 15/S 32/S 16/R 30/S 21/S 11/R 6/R 15/S 33.3
39 Citrobacter freundii 6/R 6/R 6/R 18/IR 11/S 7/R 15/IR 7/R 17/S 6/R 6/R 8/S 83.33
40 Citrobacter freundii 17/S 16/R 13/R 6/R 11/S 6/R 16/R 7/R 6/R 6/R 11/R 8/R 11.6
41 Citrobacter freundii 20/S 16/R 20/S 6/R 15/S 11/R 21/S 13/R 6/R 7/R 18/S 19/S 50

Table 2: Antimicrobial susceptibility patterns of the pathogenic Gram negative bacilli against different antibiotics by disc diffusion method.

_Pseudomonas fluorescens_ (No.13) was sensitive to (Colistin Sulphate, Cefepime and Tobramycin) and moderately resistant against Amikacin, Cefazidim, Ciprofloxacin and Rifampicin (Figure 2).

![Figure 2](image2.png)

**Figure 2:** Profile of antibiotic susceptibility of _Pseudomonas fluorescens_ (Resistant, Intermediate Resistant and sensitive).

While _P. fluorescens_ (No.14) was resistant for all antibiotics. _P. fluorescens_ (No.15) was sensitive to Colistin Sulphate only and moderately resistant against two types of antibiotics Amoxicillin/Clavulanic acid and Levofloxacin. _P. fluorescens_ (No.16) was sensitive to Amikacin, Colistin Sulphate and moderately resistant to Ciprofloxacin. Whereas _P. fluorescens_ (No.17) was sensitive to Colistin Sulphate and moderately resistant to Nitrofurantoin. _P. fluorescens_ No. 18 was sensitive to Amikacin, Amoxicillin/Clavulanic acid, Ciprofloxacin, Nitrofurantoin, Cefepime, Levofloxacin, Rifampicin and Sulphamethoxazole/Trimethoprim, while it moderately resistant to Cefazidim and Cefotaxim. Also _P. fluorescens_ No.19 was sensitive to Colistin Sulphate and Tobramycin. _P. fluorescens_ No.20 was sensitive to Amikacin, Amoxicillin/Clavulanic acid, Cefazidim, Colistin Sulphate and Nitrofurantoin, while _P. fluorescens_ No.21 was sensitive to Colistin Sulphate and Nitrofurantoin and finally _P. fluorescens_ No. 22 was sensitive to Ciprofloxacin and Levofloxacin.

_Providencia stuartii_ (No.23) was sensitive to Colistin Sulphate and moderately resistant to Amikacin, Amoxicillin/Clavulanic acid, Levofloxacin and Rifampicin (Figure 3).

![Figure 3](image3.png)

**Figure 3:** Profile of antibiotic susceptibility of _Providencia stuartii_ (Resistant, Intermediate and sensitive).

_P. stuartii_ (No.24) was sensitive to Amikacin, Ciprofloxacin, Levofloxacin, Sulphamethoxazole/Trimethoprim and Tobramycin without appearance resistant to other antibiotics. While _P. stuartii_ (No. 25) was sensitive to eight types of antibiotics (Amikacin, Cefazidim, Ciprofloxacin, Colistin Sulphate, Cefotaxim, Levofloxacin and Sulphamethoxazole/Trimethoprim) and moderately resistant to Tobramycin. _P. stuartii_ (No.26) was sensitive to Amikacin, Cefazidim,
Ciprofloxacin, Cefotaxim, Cefepime, Levofloxacin, Sulphamethoxazole/Trimethoprim and Tobramycin. *P. stuartii* (No.27) was sensitive to Amikacin, Cefazidim, Ciprofloxacin, Cefepime, Levofloxacin, Sulphamethoxazole/Trimethoprim and Tobramycin and moderately resistant to Amoxicillin/Clavulanic acid and Cefotaxim. *P. stuartii* (No.28) was sensitive to Amikacin, Ciprofloxacin, Cefepime, Levofloxacin, Sulphamethoxazole/Trimethoprim and Tobramycin and moderately resistant to Amoxicillin/Clavulanic acid and Cefotaxim. *P. stuartii* (No.29) was sensitive to Ciprofloxacin, Colistin Sulphate, Nitrofurantoin and Levofloxacin and moderately resistant to Amikacin.

*Enterobacter cloacae* (No.30) was sensitive to (Amikacin and Amoxicillin/Clavulanic acid) and moderately resistant against Levofloxacin (Figure 4).

![Figure 4: Relative number of susceptibility profile (Resistant, Intermediate Resistant and sensitive) of *Enterobacter cloacae* against antibiotics.](image)

*E. cloacae* (No.31) was sensitive to only for Colistin Sulphate and without moderately resistant against antibiotics. Also *E. cloacae* (No. 32) were sensitive to (Amikacin, Cefazidim, Colistin Sulphate, Cefotaxim, Cefepime, Levofloxacin, Sulphamethoxazole/Trimethoprim and Tobramycin) and moderately resistant against Amoxicillin/Clavulanic acid and Ciprofloxacin. *E. cloacae* (No.33) were sensitive to (Cefazidim, Colistin Sulphate, Levofloxacin and Sulphamethoxazole/Trimethoprim). While moderately resistant against Amoxicillin/Clavulanic acid and Ciprofloxacin. *E. cloacae* (No. 34) were sensitive to (Colistin Sulphate and Tobramycin) and moderately resistant against Cefazidim and Rifampicin. Finally, *E. cloacae* (No.35) were sensitive to Amikacin, Cefazidim, Colistin Sulphate, Cefepime and Tobramycin and moderately resistant to Ciprofloxacin, Cefotaxim, Levofloxacin and Rifampicin.

*Serratia rubidaea* (No.36) was sensitive to Amikacin, Amoxicillin/Clavulanic acid, Cefazidim, Ciprofloxacin, Colistin Sulphate, Cefotaxim, Nitrofurantoin, Cefepime, Levofloxacin and Tobramycin, while moderately resistant to Sulphamethoxazole/Trimethoprim (Figure 5).

*S. rubidaea* (No.37) was sensitive to Amikacin, Amoxicillin/Clavulanic acid, Cefazidim, Colistin Sulphate, Cefotaxim, Nitrofurantoin, Cefepime, Levofloxacin and Tobramycin and moderately resistant to Ciprofloxacin. *S. rubidaea* (No.38) was sensitive to Amikacin, Amoxicillin/Clavulanic acid, Cefazidim, Colistin Sulphate, Cefotaxim, Cefepime, Levofloxacin and Tobramycin and moderately resistant to Ciprofloxacin and Nitrofurantoin.

*Citrobacter freundii* (No.39) was sensitive to Colistin Sulphate and Levofloxacin and moderately resistant to Ciprofloxacin and Nitrofurantoin (Figure 6). *C. freundii* (No.40) was sensitive to Amikacin and Colistin Sulphate while moderately resistant to Amoxicillin/Clavulanic acid and Nitrofurantoin. *C. freundii* (No.41) was sensitive to Amikacin, Cefazidim, Colistin Sulphate, Nitrofurantoin, Sulphamethoxazole/Trimethoprim and Tobramycin and moderately resistant to Amoxicillin/Clavulanic acid.

![Figure 5: Relative number of susceptibility profile (Resistant, Intermediate Resistant and sensitive) of *Serratia rubidaea* against antibiotics.](image)

![Figure 6: Relative number of susceptibility profile (Resistant, Intermediate Resistant and sensitive) of *Citrobacter freundii* against antibiotics.](image)

Antimicrobial susceptibility pattern of bacterial isolates was tested against selected 12 antibiotics. From Table 2 the results obtained showed that the bacterial isolates varied in their susceptibility to all the antibiotics and showed that maximum sensitivity for Colistin Sulphate (78.1%), both Amikacin and Levofloxacin (53.7%), Ciprofloxacin (46.3%), Tobramycin (39.1%), Cefazidim (31.7%), Nitrofurantoin (29.3%), both Amoxicillin/Clavulanic acid and Cefepime (24.4%), Sulphamethoxazole/Trimethoprim (21.9%), Cefotaxim (17.1%) and Rifampicin (2.4%).
| Isolates                      | Antimicrobial agents (%) |
|-----------------------------|--------------------------|
|                             | AK ≥ 17 | AMC ≥ 18 | CAZ ≥ 18 | CIP ≥ 21 | CTX ≥ 23 | F ≥ 17 | FEP ≥ 18 | LEV ≥ 17 | RD ≥ 20 | SXT ≥ 16 | TOB ≥ 15 |
| Klebsiella pneumonia       |         |          |          |          |          |        |          |          |        |          |          |
| (*n=12*)                   | S       | 6 (50)   | 4 (33.3)| 2 (16.6)| 7 (58.3)| 10 (83.3)| 8 (66.7)| 1 (8.3)  | 4 (33.3)|          |          |
|                            | R       | 5 (41.7)| 6 (50) | 9 (75)  | 3 (25)  | 2 (16.6)| 10 (83.3)| 3 (25)   | 10 (83.3)| 3 (25)   | 12 (100)| 11 (91.7)| 7 (58.3)|
|                            | IR      | 1 (8.3) | 2 (16.6)| 1 (8.3) | 2 (16.6)| 0       | 1 (8.3) | 4 (33.3) | 1 (8.3) | 1 (8.3)  | 0         | 1 (8.3)  |
| Pseudomonas fluorescens    | (*n=10*)| S       | 3 (30) | 2 (20) | 1 (10)  | 2 (20) | 7 (70)  | 3 (30)   | 0        | 2 (20)   | 1 (10)   | 1 (10)   | 2 (20)  |
|                            | R       | 6 (60)  | 7 (70) | 8 (80) | 6 (60)  | 3 (30) | 9 (90)  | 6 (60)   | 8 (80)   | 7 (70)   | 8 (80)   | 9 (90) | 8 (80) |
|                            | IR      | 1 (10)  | 1 (10) | 1 (10) | 2 (20)  | 0       | 1 (10)  | 1 (10)   | 0        | 1 (10)   | 1 (10)   | 0       | 0      |
| Providencia stuartii       | (*n=7*)  | S       | 5 (71.4)| 0      | 3 (42.8)| 6 (85.7)| 4 (57.1)| 2 (28.6)| 1 (14.3) | 3 (42.8) | 6 (85.7) | 4 (57.1) | 6 (85.7)|
|                            | R       | 0       | 5 (71.4)| 4 (57.1)| 1 (14.3)| 3 (42.8)| 4 (57.1)| 5 (71.4)| 4 (57.1) | 0        | 7 (100)  | 3 (42.8) | 2 (28.6)|
| Enterobacter cloacae       | (*n=6*)  | S       | 3 (50) | 1 (16.7)| 3 (50) | 5 (83.3)| 1 (16.7)| 0        | 1 (16.7) | 2 (33.3) | 0        | 2 (33.3) | 3 (50)  |
|                            | R       | 3 (50)  | 3 (50) | 2 (33.3)| 3 (50) | 1 (16.7)| 4 (66.7)| 6 (100) | 4 (66.7) | 2 (33.3) | 4 (66.7)| 4 (66.7)| 3 (50)  |
|                            | IR      | 0       | 2 (33.3)| 1 (16.7)| 0      | 0       | 1 (16.7)| 0        | 1 (16.7) | 2 (33.3) | 2 (33.3) | 0        | 0      | 0      |
| Serratia rubidae           | (*n=3*)  | S       | 3 (100)| 3 (100)| 3 (100)| 1 (33.3)| 3 (100)| 3 (100) | 2 (66.6) | 3 (100) | 3 (100)  | 3 (100) |
|                            | R       | 0       | 0       | 0      | 0      | 0       | 0       | 0        | 0        | 0       | 3 (100)  | 2 (66.6)|
|                            | IR      | 0       | 0       | 0      | 2 (66.6)| 0       | 0       | 1 (33.3)| 0        | 0       | 3 (100)  | 2 (66.6)|
| Citrobacter freundii       | (*n=3*)  | S       | 2 (66.6)| 0      | 1 (33.3)| 3 (100)| 1 (33.3)| 0        | 1 (33.3) | 0        | 1 (33.3) | 1 (33.3)|
|                            | R       | 0       | 1 (33.3)| 2 (66.6)| 2 (66.6)| 3 (100)| 0        | 3 (100) | 2 (66.6) | 3 (100) | 2 (66.6) | 2 (66.6)|
|                            | IR      | 0       | 2 (66.6)| 0      | 1 (33.3)| 0       | 0       | 2 (66.6) | 0        | 0       | 0        | 0      |
| Total (*n=41*)             | S       | 22 (53.7)| 10 (24.4)| 13 (31.7)| 19 (46.3)| 32 (78.1)| 7 (17.1)| 12 (29.3)| 10 (24.4)| 22 (53.7)| 1 (2.4) | 9 (21.9)| 16 (39.1)|
|                            | R       | 14 (34.1)| 22 (53.7)| 27 (65.9)| 15 (36.6)| 9 (22) | 30 (73.2)| 20 (48.8)| 29 (70.7)| 14 (34.1)| 37 (90.2)| 31 (75.6)| 22 (53.7)|
|                            | IR      | 4 (28.6)| 9 (21.9)| 3 (7.3) | 7 (17.1)| 0       | 4 (28.6)| 9 (21.9)| 2 (4.9)  | 5 (12.2) | 3 (7.3)  | 1 (2.4) | 3 (7.3) |

**Table 3:** Antibiotic sensitivity/Intermediate resistance pattern (%) of gram negative bacteria isolated from wound infection.

Whereas bacterial isolates was resistant to Rifampicin (90.2%), Sulphamethoxazole/Trimethoprim (75.6%), Cefotaxim (73.2%), Ceftazime (70.7%), Cefazidim (65.9%), bacterial isolates was resistant to both (Amoxicillin/Clovanlic acid and Tobramycin) (53.7%), Nitrofurantoin (48.8%), Ciprofloxacin (36.6%), both (Cefotaxim and Sulphamethoxazole/Trimethoprim) (34.1%) and bacterial isolates was resistant to Colistin Sulphate (22%). The other results of tested Gram negative bacteria are listed in Table 3.

In our study, *Klebsiella pneumoniae* (24%) followed by *Pseudomonas fluorescens* (20%), *Providencia stuartii* (14%) and *Enterobacter cloacae* (12%). Both *Serratia rubidaea* and *Citrobacter freundii* (6%) with agreement [12]. *Pseudomonas species* was found to be the most common isolate (49.8%) followed by *Citrobacter braakii* (13.3%), *Enterobacter spp.* (11.1%), *Proteus vulgaris* (6.66%), *Klebsiella spp.* (2.22%) and *Serratia rubidaea* (2.22%). As well as, *Klebsiella pneumoniae* was (83.3%) sensitive to Colistin Sulphate, (66.7%) in Levofloxacin, (58.3%) in Ciprofloxacin, (50%) in Amikacin, (41.7%) in Nitrofurantoin, (33.3%) in both Amoxicillin/Clovanlic acid and Tobramycin. Also (16.6%) in Cefazidim, finally (8.3%) in both Cefotaxim and Sulphamethoxazole/Trimethoprim. While the

**Figure 7:** Sensitivity pattern (%) of gram negative bacteria isolated from wound infection.
**Klebsiella pneumoniae** was (100%) resistance to Rifampicin, (91.7%) in Sulphamethoxazole/ Trimethoprim, (83.3%) in both (Cefotaxim and Cefepime), (75%) in Ceftazidim, (58.3%) in Tobramycin, (50%) in Amoxicillin/ Clavulanic acid (41.7%) in Amikacin, (25%) in both of (Ciprofloxacine, Nitrofurantoin and Levofloxacine) and (16.6%) in Colistin Sulphate. Similar results were reported for sensitivity and resistance of Gram negative bacteria against selected antibiotics (Figure 7).

In conclusion, wound infection by pathogenic bacteria and increasing antibiotics resistance are of the most serious health threats facing the patients, especially diabetic foot patients. Thus, the objective of this work was to isolate and identify pathogenic bacteria infecting wounds. Antimicrobial susceptibility of the isolated pathogenic bacteria to different antibiotics covering all mode of action of antibiotics was conducted.

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