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Extended Spectrum β -lactamase (ESBL)-producing *Klebsiella pneumoniae* isolated from Burn Wound Infections

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ABSTRACT

To detect the extended spectrum beta lactamase (ESBL) producing *Klebsiella pneumoniae* isolated from burn infections using phenotypic methods, and then testing their resistance against antibiotic. Forty two bacterial isolates were collected from infected burn wounds at burn center in Al-Fayhaa General Hospital in Basra city/Iraq, then cultured on MacConkey agar to detect *Klebsiella*, and then cultured on CHROM agar orientation to identify *K. pneumoniae*. Nineteen of *K. Pneumonia* isolates were screened for antibiotic resistance, including: Amoxicillin (AX), Doxycycline (DO), Gentamicin (CN), Amikacin (AK), and screening it for ESBL producing. 19 (45.23%) isolates identified as *K. pneumoniae*, 15 (78.9%) of them were ESBL producer and 4 (21%) were ESBL nonproducer. All the nineteen isolates were resistant to Amoxicillin, 11 isolates to Doxycycline, 16 isolates to Gentamicin, and 18 isolates to Amikacin, while 15 isolates were ESBL resistant. Increasing prevalence of ESBL producing *K. pneumoniae* in nosocomial infections, especially in infected burn wounded this adds difficulty in curing post burn infections and may increase the rate of post burn mortality and morbidity. High number of isolates were antibiotics resistant, and this reflect the importance of implementation of susceptibility tests for successful treatment. Using CHROM agar media gives better results and faster in identifying ESBL.

Keywords

Extended-spectrum β -lactamases (ESBL), *Klebsiella pneumoniae*, β -lactamases, burn infections, CHROMagar.

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Introduction

Klebsiella pneumoniae is a Gram-negative, nonmotile, encapsulated, lactose-fermenting, facultative anaerobic, rod-shaped bacterium. It is found in the normal flora of the mouth, skin, and intestines (Ryan *et al.*, 2004), it can cause many infections like destructive changes to human and animal lungs if aspirated, specifically to the alveoli resulting in bloody sputum. Infection is the most common cause of morbidity and mortality in

burned patients (Gomez *et al.*, 2009). The type and quantity of microorganisms that colonize the burn wound may influence the risk of invasive wound infection. The pathogens that infect the wound are primarily gram-positive bacteria such as methicillin-resistant *Staphylococcus aureus* (MRSA) and gram-negative bacteria such as *Acinetobacter baumannii-calcoaceticus* complex, *Pseudomonas aeruginosa*, and

Klebsiella species. These latter pathogens are notable for their increasing resistance to multiple antimicrobial agents (Keen *et al.*, 2010; Albrecht *et al.*, 2006).

Length of hospital stay and performance of invasive procedures are risk factors for acquisition of resistance strains. Current evidence implicates plasmids as the primary source of the resistance genes in *Klebsiella* species (Hudson *et al.*, 2014). With the ability to produce extended-spectrum beta-lactamases (ESBL), *Klebsiella* is resistant to many classes of antibiotics. The most frequent are resistance to aminoglycosides, fluoroquinolones, tetracyclines, chloramphenicol and trimethoprim/ sulfamethoxazole (Nathisuwan *et al.*, 2001).

Increasing the incidence of infections with extended-spectrum β -lactamase (ESBL)-producing organisms has resulted in a therapeutic dilemma, as the choice of antibiotics is limited because of ESBL production (Paterson, 2000). The interactive effects of β -lactams are mediated through the inhibition of the bacterial cell wall and protein synthesis, respectively. Combination therapy with β -lactams and aminoglycosides is well accepted for the treatment of bacteremia caused by ESBL-producing *K. pneumoniae* isolates (Paterson *et al.*, 2004) as well as other systemic infections (Korvick *et al.*, 1992).

Prevent spreading *Klebsiella* infections between patients is a very important application, healthcare personnel must follow specific infection-control precautions (Guideline for Isolation Precautions: Preventing Transmission of Infectious Agents in Healthcare Settings (2007), including strict adherence to hand hygiene and wearing gowns and gloves when they dealing with patients with *Klebsiella*-related illnesses. Healthcare facilities also must follow strict cleaning procedures to prevent the spread of *Klebsiella*.

Methodology

Forty two bacterial isolates were collected from burn center in Al-Fayhaa General Hospital in Basra city Basra/ Iraq during the period from 01 March to 30 May 2015. Nineteen isolates were identified as *Klebsiella* by culturing on MacConkey agar, then identified as *K. pneumoniae* by culturing on CHROMagar Orientation (CHROM agar™, Paris, France) with supplement, then incubated aerobically at 37°C for 18 to 24 h. Appearance of coloured colonies on the chromogenic medium was considered a positive test result.

Susceptibility against Amoxicillin (AX), Doxycycline (DO), Gentamicin (CN), Amikacin (AK) and Extended spectrum β -lactamase antibiotics by using ISBL CHROM agar (Paris, France).

Results and Discussion

A total of 19/42(45.23%) *K. pneumoniae* isolates were obtained from infected burn wounds. *K. pneumoniae* colonies identified by morphological and biochemical characterization. Also CHROM agar Orientation was used for selective and differential colonies of *K. pneumoniae*.

Among 19 isolates of *K. pneumoniae*, 15(78.9%) isolates exhibited extended spectrum β -lactamase activity and showed metallic blue colonies on CHROMagar Orientation. The activities of 4antibioticsagainst isolates as shown in table (1), Figure (1).

In addition to pneumonia, *Klebsiella* can also cause infections in the urinary tract, lower biliary tract, and surgical wound sites. The range of clinical diseases includes pneumonia, thrombophlebitis, urinary tract infection, cholecystitis, diarrhea,

upper respiratory tract infection, wound bacteremia and septicemia.
infection, osteomyelitis, meningitis, and

Table.1 Antibiotics sensitivity of *K. pneumoniae* isolates

No. of isolates	Antibiotics				
	AX(25 µg)	DO(30 µg)	CN(10 µg)	AK(10 µg)	ISBL
1	R	S	S	I	R
2	R	I	R	R	R
3	R	R	R	R	R
4	R	R	R	R	R
5	R	R	R	R	R
6	R	S	S	S	S
7	R	R	R	R	R
8	R	R	R	R	R
9	R	R	R	R	S
10	R	I	R	R	S
11	R	R	R	R	R
12	R	R	R	R	R
13	R	R	R	R	R
14	R	I	R	R	R
15	R	S	S	S	R
16	R	R	R	R	R
17	R	S	R	S	S
18	R	S	R	S	R
19	R	R	R	R	R

R= Resistant, S= Susceptible, I= Intermediate *

Fig.1 Resistant isolates to AX, DO, CN, and AK antibiotics

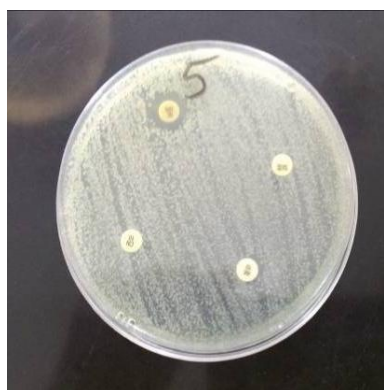


Fig.2 Metallic blue colonies of ESBL-producing *K. pneumoniae* on CHROMagar Orientation



So treatment depends on the organ system involved and the choice of a specific antimicrobial agent depends on local susceptibility patterns because *Klebsiella* organisms are resistant to multiple antibiotics (Paneru, 2015).

Table (1) showed that all the nineteen isolates were resistant to Amoxicillin, 11 isolates to Doxycycline, 16 isolates to Gentamicin, and 18 isolates to Amikacin, while 15 isolates were ISBL resistant. The high number of resistant isolates reflect the importance of implementation of susceptibility tests for successful treatment.

Agents with high intrinsic activity against *K. pneumoniae* should be selected for severely ill patients. Examples of such agents include third-generation cephalosporins (eg, cefotaxime, ceftriaxone), carbapenems (eg, imipenem/cilastatin), aminoglycosides (eg, gentamicin, amikacin), and quinolones. These agents may be used as monotherapy or combination therapy. Some experts recommend using a combination of an aminoglycoside and a third-generation cephalosporin as treatment for non-ESBL-producing isolates. Others disagree and recommend monotherapy.

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