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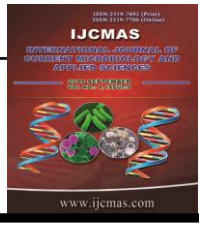

Original Research Article

Isolation and Antibiogram of *Klebsiella* species from Various Clinical Specimens

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Keywords

Klebsiella pneumoniae, ESBL

A B S T R A C T

Klebsiella is an important nosocomial human pathogen that has the potential to cause severe infections. *K. pneumoniae* is gaining renewed interest because of emergence of multidrug resistance due to ESBL production. Total 100 samples showing *Klebsiella* species on culture were studied. *Klebsiella pneumoniae* was the most common species followed by *Klebsiella oxytoca*. Males were most affected. Diabetes, alcoholism & previous surgeries were commonly associated factors. Common infections caused by *Klebsiella* were wound infections, urinary tract infections and respiratory infections. Around a quarter of all *Klebsiella* isolates were ESBL producers exhibiting multidrug resistance to commonly used antibiotics like cephalosporins. All isolates were sensitive to carbapenems.

Introduction

Klebsiella species is a Gram negative opportunistic nosocomial pathogen and is known to cause community acquired infections. *Klebsiella* is known to produce bacterial pneumonia, urinary tract infection, wound infections, blood infections and infections in the intensive care unit (Podschun and Ullmann, 1998; Brisse *et al.*, 2006).

Nosocomial *Klebsiella* infections are mainly caused by *Klebsiella pneumoniae* followed by *Klebsiella oxytoca* (Jadhav *et al.*, 2012). *Klebsiella* spp have several virulence factors such as capsular polysaccharides, lipopolysaccharide and iron-scavenging systems (siderophores).

Major risk factor for colonisation or infection with ESBL producing organisms are long term antibiotic exposure, prolonged intensive care unit stay, nursing home residency, severe illness, residence in an institution with high rates of ceftazidime and other third generation cephalosporins use and instrumentation or catheterization (Sarma *et al.*, 2011). In recent years, outbreaks of infection caused by multidrug-resistant ESBL-KP have been

reported throughout the world (Christian *et al.*, 2010; Filippa *et al.*, 2013).

The study has been conducted to know the prevalence of *Klebsiella* species in terms of samples type, various risk factors and to find the multidrug resistance including ESBL producing rates.

Materials and Methods

Study constituted 100 non-repetitive isolates from various samples in Microbiology lab of M.R. Medical College Gulbarga. All the samples were collected aseptically. Samples were inoculated on Blood agar and MacConkeys agar and incubated at 37°C for 24–48 hours. After Gram's staining, colonies were subjected to preliminary tests like catalase, oxidase & to various biochemical tests like IMViC, TSI, urease, sugar fermentation tests-glucose, lactose, sucrose & mannitol. The biochemical characters used for identification were positive Voges-Proskauer test, positive citrate utilization test, positive urease test, acid and abundant gas production from glucose, lactose, sucrose, maltose and mannitol sugar fermentation tests. Antibiotic sensitivity was done on Mueller-Hinton agar plates by Kirby-Bauer disc diffusion method according to the CLSI guidelines (2011). A log phase broth culture inoculum of the isolate with a turbidity equivalent to McFarland 0.5 standard was used. Lawn cultures on the Mueller-Hinton agar were prepared and allowed to dry. Antibiotic discs were applied to the Mueller Hinton agar surface with the help of sterile forceps. The antibiotics tested were ampicillin (10µg), gentamicin (10µg), amikacin (30µg), amoxicillin/clavulanic acid (20/10µg), piperacillin/tazobactam (100/10 µg), cefepime (30µg), cefotaxime (30µg), ceftriaxone (30µg), ceftazidime (30µg), ciprofloxacin (5µg), ofloxacin (5µg), imipenem (10µg), meropenem (10µg), cotrimoxazole (25µg) nitrofurantoin (300µg). Detection of ESBL was done using CLSI phenotypic confirmation test.

Results and Discussion

During the study period, a total of 100 non-repetitive isolates from various clinical samples were processed. Among the 100 *Klebsiella* spp, 89% were *Klebsiella pneumoniae* and 11% were *Klebsiella oxytoca*. Of the 100 *Klebsiella* isolates, 62 were from males and 38 were from females with a male: female ratio of 1.7: 1.

The highest percentage of *Klebsiella* spp were isolated from pus (50%) followed by urine (21%), sputum (18%), blood (7%), throat swab (3%) and CSF (1%).

Various risk factors associated with *Klebsiella* infection observed were diabetes mellitus, hypertension, alcoholism, smoking, post-operative state and, catheterization.

The isolates were 100% sensitive to imipenem & meropenem, cefepime (82%), piperacillin/tazobactum (77%), amikacin (73%), ceftriaxone (69%), cefotaxime (67%), nitrofurantoin (66%), ceftazidime (63%), amox-clav (62%), gentamicin (62%), ofloxacin (59%), ciprofloxacin (56%), cotrimoxazole (33%) and ampicillin (06%).

Of 100 isolates of *Klebsiella*, 24 isolates were found to produce extended spectrum beta lactamases detected by CLSI phenotypic confirmation test.

Klebsiella pneumoniae (KP) is one of the leading causes of nosocomial infections seen worldwide, causing pneumonia, bloodstream infections, urinary tract infections, surgical site infections and meningitis (Peleg and Hooper, 2010). In the present study 100 isolates of *Klebsiella* were studied. In the current study, isolation rate of *K.*

pneumoniae was more compared to K. *oxytoca*. *Klebsiella pneumoniae* incidence was 89%. This finding is comparable to the report of Asmaa (2012).

Males (62%) were more affected than females, and this could be due to higher prevalence of alcoholism and smoking in males.

Table.1 Percentage of *Klebsiella* species isolated

Species	No of isolates	Percentage
Klebsiella pneumoniae	89	89%
Klebsiella oxytoca	11	11%

Table.2 Percentage of isolation of *Klebsiella* species from different clinical specimens

Sample	No. of isolates	percentage
Pus	50	50%
Urine	21	21%
Sputum	18	18%
Blood	7	7%
Throat swab	3	3%
CSF	1	1%

Table.3 Various Risk factors in patients with *Klebsiella* infection

Risk factors	No of patients
Diabetes	36
Hypertension	16
Post-surgical	27
Catheterisation	08
Alcohol & smoking	21

Table.4 Antibiotic sensitivity profile

Antibiotic	Sensitivity
Ampicillin	06%
Gentamicin	62%
Amikacin	73%
Amox-clav	62%
Piperacillin/Tazobactum	77%
Cefepime	82%
Cefotaxime	67%
Ceftriaxone	69%
Ceftazidime	63%
Ciprofloxacin	56%
Ofloxacin	59%
Imipenem	100%
Meropenem	100%
Cotrimoxazole	33%
Nitrofurantoin	66%

The highest percentage of *Klebsiella* spp were isolated from pus (50%) followed by urine (21%), sputum (18%), blood (7%), throat swab (3%) and CSF (1%). Various studies done on *Klebsiella* found similar results, but most common sample being urine in their study (Amit Kumar Singh, 2015). Most common risk factor associated with *Klebsiella* infections was found to be diabetes followed by alcoholism and previous surgeries. Several studies done suggest these as the most common risk factors in patients (Namratha *et al.,* 2015; Abhilash *et al.,* 2010).

Antimicrobial susceptibility pattern of isolates showed 100% sensitivity to carbapenems (imipenem & meropenem) while resistance to cephalosporins ranged from 50 to 70%. A low susceptibility pattern was observed for Ampicillin and cotrimoxazole. Study done by Menon *et al.* (2006) found 100% susceptibility to imipenem. Harada *et al.* (2013) had also demonstrated the antimicrobial resistance profile of ESBL-KP and showed a higher degree of susceptibility to amikacin (92.5%) and nitrofurantoin (67.43%). 24 isolates of *Klebsiella* were ESBL producers which showed resistance to most of the antibiotics used. Confirmatory phenotypic confirmation test for detection of ESBL is one of the most simple and reliable test for detection of ESBL.

Klebsiella especially K.pneumoniae is gaining renewed interest because of emergence of multidrug resistance among *Klebsiella* associated infections. Males are most commonly affected and risk factors like diabetes, alcoholism increase the risk of developing *Klebsiella* infections. Wound infections, urinary tract infection and respiratory infections are usual in *Klebsiella* infection. *Klebsiella pneumoniae* was the most common species isolated followed by *Klebsiella oxytoca*. *Klebsiella* were still 100% sensitive to carbapenems, but sensitivity to cephalosporins ranged from 50–70%. *Klebsiella* are now being recognized as one of the major threats to effective management of patients in hospital, especially in a developing country like India.

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