Original Article

Antimicrobial resistance pattern of Klebsiella species isolated from various clinical specimens

Begum T[1], Nahar A[2], Quamruzzaman M[3] , Haq AI[4], Mahmood IH[5], Bhuiyan ME[6]

Abstract

Background:

Objective: The present study was carried out to identify Klebsiella species and their antimicrobial resistance pattern from various clinical specimens.

Methods: This cross study was conducted at a tertiary care hospital, Dhaka, Bangladesh from July2020 to June 2021. Written consent was taken from the concerned authority. A total 6739 clinical samples including urine, wound swab/ pus, sputum, blood and endotracheal aspirates were collected and processed for isolation of bacteria by following standard guidelines from adult patients. All the urine, wound swab, blood and endotracheal aspirates were inoculated in both blood agar and MacConkey agar media, for blood samples blood culture was done in BacT/ALERT 3D 60 aerobic bottles irrespective to antibiotics administration. After incubation at 370C aerobically for 24 hours incubated plates were examined. Phenotypic identification of the organisms were done by observing colony morphology, hemolytic properties, pigment production, Gram staining and biochemical tests (oxidase test, catalase test and biochemical reactions after inoculation in TSI, MIU, citrate agar media). The Kirby – Bauer disk diffusion method was used for antimicrobial susceptibility testing of the isolated organism using Mueller-Hinton agar and commercially available antibiotic discs (Oxoid Ltd, UK). The data were analyzed using SPSS software Version-20 (SPSS Inc., Chicago, IL, USA). Categorical variables were expressed as numbers (n) and percentages (%).

Results: A total 6739 samples were included in the present study. Of which, 170 were urine, 162 were wound swabs, 71 were sputum, 52 were endotracheal aspirates and 45 were blood samples. Culture positivity among the samples is shown in table I.

Conclusion: Most of the Klebsiella species are highly resistant to commonly used antibiotics. This higher resistance is the major cause morbidity and mortality related with infections in hospitalized patients. So, a regular surveillance of antibiotic susceptibility pattern is needed to prevent indiscriminate use of antibiotics.

Keywords: Antimicrobial resistance, Klebsiella spp., co-trimoxazole, tigacycline.

1. Dr. Taslima Begum M.Phil, MBBS
   Associate professor
   Department of microbiology
   Holy Family Red Crescent Medical College
2. Dr. Azizun Nahar M. Phil, MBBS
   Associate professor
   Department of microbiology
   Bangladesh Medical College, Dhanmondi, Dhaka
3. Dr. Md. Quamruzzaman MBBS
   Junior Consultant, Dept. of Neuromedicine
   Bangladesh Medical College Hospital
4. Dr. Arman Ibne Haq MBBS, FCPS (Psy)
   Assistant Professor, Dept. of Psychiatry
   Bangladesh Medical College
5. Dr. Iqbal Hasan Mahmood FCGP, MBBS
   Registrar, Dept. of Nephrology
   Bangladesh Medical College
6. Dr. Md. Elias Bhuiyan MBBS, MRCP (London), FCPS (Internal Medicine)
   Assistant Professor, dept. of Medicine
   Bangladesh Medical College & Hospital

Correspondence to:
Dr. Taslima Begum M.Phil, MBBS
Associate professor
Department of microbiology
Holy Family Red Crescent Medical College
pannataslima@gmail.com

Introduction:

Enterobacteriaceae family is most commonly colonizing the gastrointestinal tract causing various human infections. The genus Klebsiella is the second most common organism among Enterobacteriaceae family which is a gram negative, lactose fermenting, non- motile, rod shaped and facultative anaerobic bacilli. Klebsiella species is responsible for 3 – 8% of all nosocomial infections. They are widely recognized as important pathogens in genitourinary infections, bronchopneumonia, wound, soft tissue and blood stream infections (BSI).[1] Depending on the site of infections, Klebsiella species were isolated from different a clinical sample that causes various infections. The most common hospital acquired pathogen is Klebsiella pneumoniae causing abscess, wound infection, urinary tract infection, meningitis, lung infection, sepsis and even death of newborn in intensive care unit.[2] Antimicrobial resistance among clinical isolates of Klebsiella species has become an increasingly serious problem over the past twenty years.[3] Multidrug resistant strain of K. pneumonia emerged due to indiscriminate use of various antibiotics.[4]

Due to a drastic increase in the antibiotic resistance pattern encountered among Klebsiella species, it is imperious to know the institutional prevalence and susceptibility profile.

The present study was carried out to identify Klebsiella species and their antimicrobial resistance pattern from various clinical specimens. This will be beneficial for medical practitioners to select empirical antimicrobial therapy which will play an important role in minimizing the emergence rate of antimicrobial resistance.

Materials & Methods:

This cross study was conducted at a tertiary care hospital, Dhaka, Bangladesh from July2020 to June 2021. Written consent was taken from the concerned authority.

Sample collection

A total 6739 clinical samples including urine, wound swab/ pus, sputum, blood and endotracheal aspirates were collected and processed for isolation of bacteria by following standard guidelines from adult patients.

Culture

All the urine, wound swab, blood and endotracheal aspirates were inoculated in both blood agar and MacConkey agar media, for blood samples blood culture was done in BacT/ALERT 3D 60 aerobic bottles irrespective to antibiotics administration. After incubation at 370C aerobically for 24 hours incubated plates were examined.

Isolation and identification of the organisms from culture

Phenotypic identification of the organisms were done by observing colony morphology, hemolytic properties, pigment production, Gram staining and biochemical tests (oxidase test, catalase test and biochemical reactions after inoculation in TSI, MIU, citrate agar media).[5]

Antimicrobial susceptibility test

The Kirby – Bauer disk diffusion method was used for antimicrobial susceptibility testing of the isolated organism using Mueller-Hinton agar and commercially available antibiotic discs (Oxoid Ltd, UK).[6] Sensitivity discs of amoxicillin/clavulanic acid (20/10 µg), ceftriaxone (30 µg), ceftazidime (30 µg), cefuroxime (30 µg), cefepime (30 µg), cotrimoxazole (1.25/23.75 µg), ciprofloxacin (5 µg), gentamicin (10 µg), amikacin (30 µ g), meropenem (10 µg), piperacillin-tazobactam (100/10 µg) & tigecycline (15 µg) were used. In addition, the disc contents and the zone of inhibition were used as recommended by the Clinical Laboratory Standards Institute (CLSI)[7] & United States Food and Drug Administration (FDA) guideline for tigecycline.[8]

Data analysis

The data were analyzed using SPSS software Version-20 (SPSS Inc., Chicago, IL, USA). Categorical variables were expressed as numbers (n) and percentages (%).

Results:

A total 6739 samples were included in the present study. Of which, 170 were urine, 162 were wound swabs, 71 were sputum, 52 were endotracheal aspirates and 45 were blood samples. Culture positivity among the samples is shown in table I.

Table 1: Culture positive isolates from various clinical samples

Samples	Number of samples	Culture positive
Urine	4030	557 (13.82%)
Blood	2371	85 (3.58%)
Wound swab/ Pus	134	102 (76.11%)
Sputum	95	38 (40%)
Endotracheal aspirate	109	70 (64.22%)
Total	6739	852 (12.64%)

Table 2: Distribution of organisms isolated from culture positive samples (N= 852)

Isolated Organisms	n (%)
E. coli	288 (33.80)
Klebsiella spp.	295 (34.62)
Pseudomonas spp.	87 (10.21)
Acinetobacter spp.	65 (7.63)
Staph. aureus	107 (12.56)
Salmonella Typhi	10 (1.17)
Total	852 (99.99)

N= Total number of bacteria
n= Total number of bacterial species

Distribution of organisms isolated from various clinical samples is shown in Table 2. Among the total 852 culture positive samples, majority of the isolates were Klebsiella spp. 295 (34.62%) followed by E. coli 288 (33.80%), Staph. aureus 107 (12.56%), Pseudomonas spp. 87 (10.21%), Acinetobacter spp. 65 (7.63%) & Salmonella Typhi 10 (1.17%).

Table 3: Isolation rate of Klebsiella species from different clinical samples (N= 295)

Samples	Culture positiven (%)
Urine	193 (65.42%)
Blood	15 (5.08%)
Wound swab/ Pus	24 (8.14%)
Sputum	20 (6.78%)
Endotracheal aspirate	43 (14.58%)
Total	295 (100%)

N= Total number of bacteria
n= Total number of bacterial species

The highest isolation of Klebsiella spp. (37.33%) was observed from urine followed by endotracheal aspirate 43 (14.58%), wound swab/ pus 24 (8.14%), sputum 20 (6.78%), blood 15 (5.08%) (Table 3).

Table 4: Antibiotic resistance pattern of isolated Klebsiella species (N= 295)

Antimicrobial drugs	Resistancen (%)
Amoxyclav	263 (89.15%)
Cefuroxime	266 (90.17)
Ceftriaxone	278 (94.24)
Ceftazidime	266 (90.17 %)
Cefepime	263 (89.15 %)
Co-trimoxazole	92 (31.19%)
Ciprofloxacin	264 (89.49%)
Gentamicin	237 (80.33%)
Amikacin	226 (76.61%)
Meropenem	238 (80.68%)
Piperacillin/Tazobactam	120 (40.68%)
Tigecycline	16 (5.42%)

N= Total number of bacteria
n= Total number of bacterial species

Antibiotic resistance pattern of isolated Klebsiella species was shown in Table 4. Cephalosporin group of antibiotic showed higher resistance, 278 (94.24%) isolates were resistant to ceftriaxone, 266 (90.17%) to ceftazidime, 266 (90.17%) to cefuroxime & 263 (89.15%) to cefepime. Resistance to ciprofloxacin was 264 (89.49%). Gentamycin & amikacin resistance were 237 (80.33%) & 226 (76.61%) respectively. Low resistance were observed in co-trimoxazole 92 (31.19%), piperacillin/tazobactam 120 (40.68%). Very low resistance was seen in tigacycline 16 (5.42%).

Discussion:

A total of 6739 clinical samples were processed for isolation, identification and culture & sensitivity testing where bacterial growth was obtained in 852 (12.64%) samples. This study is closer to a study done in India who reported 22.43% culture positivity (Table 1). [9]

In this study, among the culture positive samples Klebsiella species 295 (34.62%) was the most common organism followed by E. coli 288 (33.80%), Staph aureus 107 (12.56%), pseudomonas species 87 (10.21%), Acinetobacter species 65 (7.63%) & Salmonella species 10 (1.17%). Among the culture positive samples 27% Klebsiella species was identified which is similar to our study (Table 2).[10]

The isolation rate of Klebsiella species was highest from urine 193 (65.42%), followed by endotracheal aspirate 43(14.58%), wound swab/pus 24 (8.14%), sputum 20 (6.78%) & blood 15(5.08%) (Table 3). Highest K. pneumoniae (46.70%) were obtained from sputum and other lower respiratory tract secretions followed by blood (31.30%) and wound swab (24.20%) which is dissimilar with our study.[11] The isolation of Klebsiella species vary from different samples may be due to variation in the sample size. In the present study, 94.24% isolates were resistant to ceftriaxone, 93.22% to ceftazidime, 90.17% to cefuroxime & 89.15% to cefepime (Table 4). Higher rate of resistance to cephalosporin group have also reported by another author.[12] Resistant to ceftriaxone was reported 100%. [13] Resistance to ciprofloxacin was 89.49% in this study (Table 4). A similar study where ciprofloxacin was 88% resistant was also observed.[14] The resistance to flouroquinolones could be due to mutations in the chromosomal genes encoding DNA gyrase of the bacteria or due to efflux of the drug. [15] The higher rate of resistance to ciprofloxacin might be due to widely use of this drug. A high level of resistant is also observed to the aminoglycosides. Gentamycin resistance was 80.33% & amikacin resistance was 76.61% respectively (Table 4), similar with another study.[9] Lower rate of resistance (31.19%) was observed against cotrimoxazole, which is dissimilar with other observations.[13, 16] In the present

study, 42.71% isolates were meropenem resistant. Here, piperacillin/tazobactam resistant was 40.68%. Tigacycline which is a reserve drug was 5.42% resistant (Table 4). Lower resistance may be due to restricted use of this drug. But higher resistance (74%) of tigecycline was observed in a study.[9]

Conclusion:

Most of the Klebsiella species are highly resistant to commonly used antibiotics. This higher resistance is the major cause morbidity and mortality related with infections in hospitalized patients. So, a regular surveillance of antibiotic susceptibility pattern is needed to prevent indiscriminate use of antibiotics.

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