academicJournals

Vol. 8(4), pp. 316-319, 22 January, 2014
DOI: 10.5897/AJMR2013.6409
ISSN 1996-0808 ©2014 Academic Journals
http://www.academicjournals.org/AJMR

African Journal of Microbiology Research

Full Length Research Paper

Multi-drug resistant *Klebsiella pneumoniae* causing urinary tract infections in children in Pakistan

Iqra Jamil[1], Aizza Zafar[1], Muhammad Usman Qamar[2]*, Hasan Ejaz[1], Junaid Akhtar[2] and Abdul Waheed[2]

[1]Department of Microbiology, The Children Hospital Lahore, Pakistan.
[2]Department of Microbiology University of Health Sciences, Lahore, Pakistan.

Accepted 8 January, 2014

Multi-drug resistant (MDR) *Klebsiella pneumoniae* has been associated with different types of infections and the most important aspect is the emergence of MDR strains particularly in hospitalized children. They have the proficiency to cause bacteremia, septicemia and urinary tract infections (UTI). The objective of this study was to determine the susceptibility of MDR *K. pneumoniae* causing UTI in children. One thousand and fifteen (1015) urine samples were collected aseptically. Specimens were cultured on blood agar, MacConkey agar and cysteine lactose electrolyte deficient (CLED) agar. Antimicrobial susceptibility was determined using Kirby-Bauer disc diffusion method as per CLSI 2011 guidelines. Of the 1015 urine specimens, 230 (22.6%) were positive for bacterial growth. Out of these positive cultures predominantly Gram-negative rods (90%) were isolated and major pathogens were *K. pneumoniae* (40%) and *Escherichia coli* (33%). Antimicrobial susceptibility pattern of *K. pneumoniae* showed that more than 70% of these pathogens were resistant to cephalosporins, 69% to ciprofloxacin and amoxicillin-clavulanic acid and 63% to norfloxacin and nalidixic acid while most effective drugs were pipracillin-tazobactam and meropenem. This study concludes that MDR-*K. pneumoniae* is a great threat particularly in children in our locality.

Key words: Multi-drug resistant (MDR), *Klebsiella pneumoniae*, children, antimicrobial resistance (AMR).

INTRODUCTION

Urinary tract infections (UTIs) have become one of the most common disease encounters in clinical practice. There is an estimation that over 150 million UTIs cases occur worldwide annually (Gobernado et al., 2007). This is associated with a high risk of morbidity, mortality, extra financial budget and even, fatal consequences particularly in children (Hasan et al., 2007). UTIs are mainly caused by Gram negative rods (GNR) which account for 80 to 85% and the leading causative organisms are *Escherichia coli* (75 to 95%) and *Klebsiella pneumoniae* (Tanvir et al., 2012).

K. pneumoniae are very often isolated in hospital set up and a significant proportion being multidrug resistant (MDR) that become a formidable challenge nowadays (Rampure et al., 2013). The ability of MDR *K. pneumoniae* to easily spread makes this bacterium an important nosocomial pathogen (Ramirez et al., 2012). These pathogens are also responsible for an important cause of children infections such as bacteremia, septicemia and urinary tract infections (UTIs) (Manisha and Pratibha, 2012). Treatment of UTI patients infected with *K. pneumoniae* has become more difficult due to increasing antimicrobial resistance which severely limits the therapeutic options. These strains have typically been resistant to two or more different classes of antibiotics such as β-lactams, β-lactamase inhibitors, aminoglycosides

*Corresponding author. E-mail: usman9785@gmail.com. Tel: +92-301-3054705.

Table 1. Frequency of urinary isolates (n = 230) from UTI patients.

Organism (n = 230)	Frequency	(%)
Gram-negative rods (n = 209)		
Klebsiella pneumoniae	92	40
Escherichia coli	75	32.6
Pseudomonas aeruginosa	18	7.8
Acinetobacter spp.	15	6.5
Citrobacter spp.	5	2.1
Proteus mirabilis	4	1.7
Gram-positive cocci (n = 21)		
Enterococcus faecalis	13	5.6
Coagulase negative staphylococci (CoNS)	5	2.2
Staphylococcus aureus	3	1.3

and quinolones. This is believed that the best empirical therapy to treat mild to moderate infections due to MDR *K. pneumoniae* would be the ertapenem (Brink, 2008). This is expected that MDR *K. pneumoniae* will become more and more resistant to antimicrobials with the passage of time because of the generation of their new mutant strains (Khameneh and Afshar, 2009). Therefore, we design this study to determine the frequency and antimicrobial resistant pattern in clinical isolates of *K. pneumoniae* in a tertiary care hospital, Lahore.

MATERIALS AND METHODS

Prior to start of the study, permission was taken from ethical review committee, School of Allied Health Sciences, tertiary care hospital Lahore, Pakistan.

Study population

A total of 1015 urine specimens suspected for UTIs were collected from children (age: 0 - 12 years) in a tertiary care hospital Lahore, Pakistan. Mid stream urine samples were collected aseptically.

Identification of isolates

Urine specimens were cultured on blood agar, MacConkey agar and cysteine lactose electrolyte deficient (CLED) agar and incubated at 35°C for 24 h. The isolates were preliminary identified on the basis of morphology and cultural characteristics. Gram-positive isolates were biochemically identified by catalase, slide and tube coagulase and DNAse test whereas, Gram-negative isolates were biochemically identified by cytochrome oxidase and confirmed by API 20E and 20NE (BioMerieux France).

Antimicrobial susceptibility testing

Antimicrobial susceptibility of *K. pneumoniae* isolates was performed by Kirby-Bauer disk diffusion method using Mueller-Hinton agar (Oxoid UK), according to Clinical Laboratory Standards Institute (CLSI) 2011 guidelines. The plates were prepared and incubated at 35°C for 24 h. Implanted antibiotics were amikacin (30 µg), amoxicillin-clavulanic acid (20 µg/10 µg), cefotaxime (30 µg), nalidixic acid (30 µg), cefuroxime (30 µg), piperacillin-tazobactam (100/10 µg), nitrofurantoin (300 µg), norfloxacin (10 µg), cefixime (5 µg), ceftazidime (30 µg), ciprofloxacin (5 µg) and meropenem (10 µg).The interpretation of susceptibility results were done as per CLSI 2011 guidelines (Wikler et al., 2009). Statistical analysis was done using SPSS 16.0.

RESULTS

Of 1015 urine specimens, 230 (22.6%) were positive for bacterial growth. Out of these positive cultures (n = 230), 209 GNR and 21 GPC were isolated. Among GNR major pathogens were *K. pneumoniae* (n = 92) followed by *E. coli* (n = 75), *P. aeruginosa* (n = 18) and *Acinetobacter spp* (n=15) whereas among Gram positive Cocci (GPC), *Enterococcus faecalis* (n = 13) was the main pathogen (Table 1).

Antimicrobial susceptibility pattern showed that more than 70% of *K. pneumoniae* were resistant to cephalosporins, 69% to ciprofloxacin and amoxicillin-clavulanic acid and 66% to nalidixic acid and norfloxacin. Most effective drugs were piperacillin-tazobactam followed by meropenem that showed resistance of 5.4 and 14.1%, respectively (Figure 1).

DISCUSSION

Organisms causing UTIs particularly in children are becoming a public health problem. In present study, 22.6% (n=230) urine specimens were positive for significant bacterial growth which is in accordance with previous studies (Ahmad, 2013; Chaudhary et al., 2013). However few data suggested variable results ranging from 9 to 34% of urine positive cultures (Oh et al., 2013; Jahanzeb et al., 2008). This difference could be due to the difference in samples size, handling and processing of sample techniques.

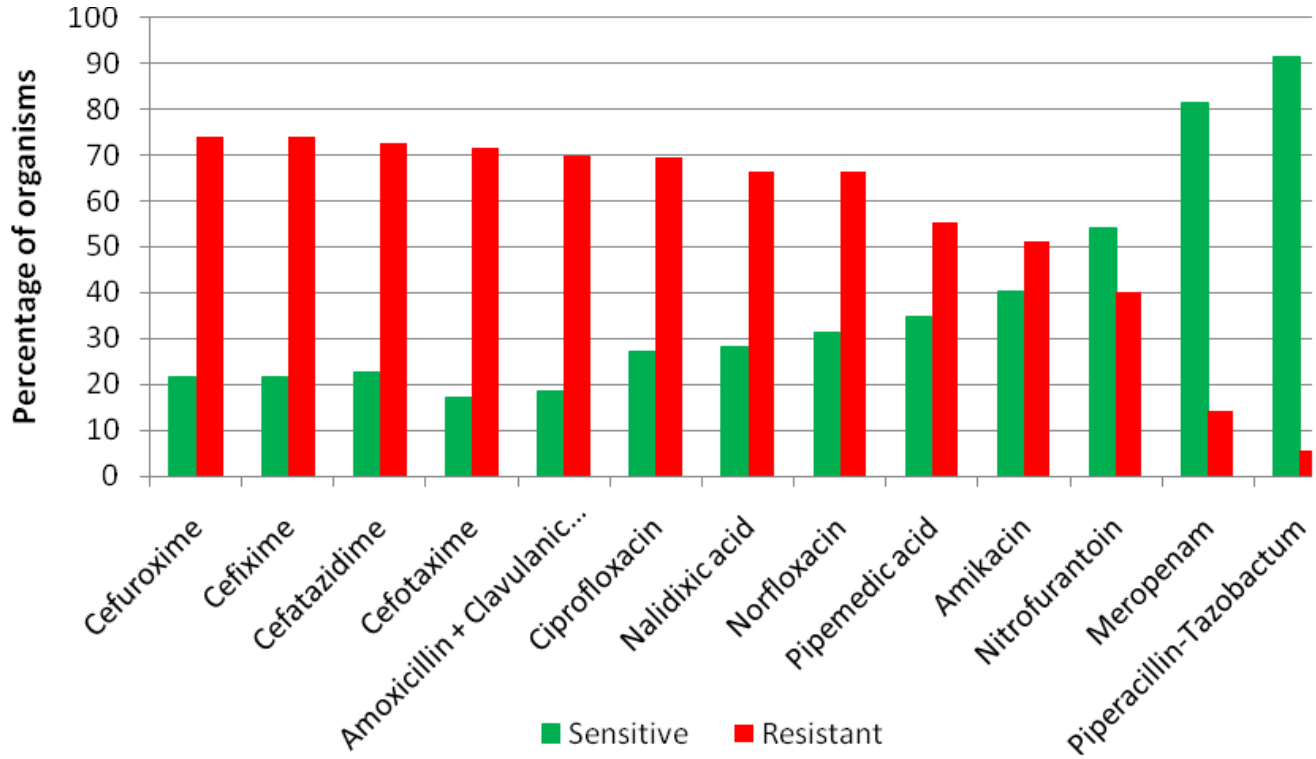

Figure 1. Overall percent susceptibility pattern of *K. pneumoniae* in which high drug resistance was observed against cephalosporins, Amoxicillin+clavulanic acid and ciprofloxacin whereas most effective drug was piperacillin-tazobactam.

Overall, GNR are the commonest cause of UTIs as compare to GPC. In this study, 91% of GNR were isolated which are comparable with previous studies conducted in Iran (90.3%), Turkey (89%) and Australia (96%) (Ayazi et al., 2010; Mehr et al., 2004; Arslan et al., 2002). This has been identified in earlier studies that *Enterobacteriaceae* are the predominant pathogens in UTI patients. These organisms are part of gut normal flora and individuals can easily get infected with poor personal hygiene (Afsharpaiman et al., 2012). Overall, *E. coli* still remains the most frequent isolated pathogens in UTIs but in the present study, the predominant pathogen was *K. pneumoniae* (40%). These results are in agreement with studies conducted in the Children hospital Lahore, Pakistan and Turkey (Ejaz et al., 2006; Biyikli et al., 2004). Contrary to this study, various other data also documented that *E. coli* was the predominant pathogen in UTIs patients (Al-Momani, 2006; Qureshi, 2005). In the current study, *K. pneumoniae* showed high resistance against commonly used antibiotics (cephalosporins, ciprofloxacin, amoxicillin- clavulanic acid and amikacin). These findings are almost in accordance with previous studies conducted in Iran, Pakistan, Mexico and India that reported MDR *K. pneumoniae* (Rampure et al., 2013; Langarizadeh et al., 2011; Ullah et al., 2009). Contrary few studies from Turkey, Korea and Iran showed low to moderate resistance (Yoon et al., 2011; Jalalpoor, 2011; Senel et al., 2010). Spread of MDR- *K. pneumoniae* is associated with inappropriate infections control practices. Various other factors includes contaminated intravenous catheters, environment surfaces and colonized hands of health care staff. Substandard and disinfection practices are also common. Another major factor to acquire resistance in our setup is the irrational use of empirical therapy which is not according to WHO criteria (Hannan et al., 2013).

It is concluded that MDR *K. pneumoniae* play a crucial role in spreading UTIs and these pathogens are also extended from the hospital to community. Now, it is the need of the hour to improve infections control practices, avoid irrational use of antibiotics and empirical regime should be revisited to prevent further resistance.

ACKNOWLEDGEMENTS

We are grateful to the School of Allied Health Sciences, The Children Hospital, Lahore for the financial and logistic support for this research project.

REFERENCES

Afsharpaiman S, Bairaghdar F, Torkaman M, Kavehmanesh Z, Amirsalari S, Moradi M, and Safavimirmahalleh MJ (2012). Bacterial pathogens and resistance patterns in children with community-acquired

urinary tract infection: A cross sectional study. J. Compr. Pediatr. 3:16-20.
Ahmad S (2013). Pattern of urinary tract infection in Kashmir and antimicrobial susceptibility. Bangladesh Med. Res. Counc. Bull. 38:79-83.
Al-Momani T (2006). Microbiological study of urinary tract infection in children at Princess Haya Hospital in south of Jordan. Middle East J. Family Med. 3(5):2.
Arslan S, Caksen H, Rastgeldi L, Uner A, Oner AF, Odabas D (2002). Use of urinary gram stain for detection of urinary tract infection in childhood. Yale J. Biol. Med. 75:73.
Ayazi P, Mahyar A, Jahani HH, Khabiri S (2010). Urinary Tract Infections in Children. Iran. J. Pediatr. Soc. 2(1):9-14.
Biyikli NK, Alpay H, Ozek E, Akman I, Bilgen H (2004). Neonatal urinary tract infections: analysis of the patients and recurrences. Pediatr. Int. 46:21-25.
Brink A (2008). Multidrug-resistant (MDR) Gramnegative fermenters: Our worst nightmare. South. Afr. J. Epidemio. Infect. 22:2-4.
Chaudhary R, Ojha CR, Sijapati K, Singh SK (2013). Bacterial Pathogen Responsible For Urinary Tract Infect. Med. J. Shree Birendra Hosp. 11:13-16.
Ejaz H, Zafar A, Anwar N, Cheema T, Shehzad H (2006). Prevelance of bacteria in urinary tract infections among children. Biomedica 22:139-142.
Gobernado M, Valdes L, Alos JI, Garcia-Rey C, Dal-Re R, Garcia-de-Lomas J (2007). Antimicrobial susceptibility of clinical Escherichia coli isolates from uncomplicated cystitis in women over a 1-year period in Spain. Rev. Esp. Quimioter. 20:68-76.
Hannan A, Qamar MU, Usman M, Ahmad K, Waheed I, Rauf K (2013). Multidrug resistant microorganisms causing neonatal septicemia: In a tertiary care hospital Lahore, Pakistan. Afr. J. Microbiol. Res. 7(19):1896-1902.
Hasan AS, Nair D, Kaur J, Baweja G, Deb M, Aggarwal P (2007). Resistance patterns of urinary isolates in a tertiary indian hospital. J. Ayub Med. Coll. Abbottabad 19:39-41.
Jahanzeb M, Siddiqui T, Idris M (2008). Frequency and clinical presentation of UTI among children of Hazara Division, Pakistan. J. Ayub Med. Coll. Abbottabad 20: 63.
Jalalpoor S (2011). Antibiotic resistant pattern in ESBLs producer klebsiella pneumoniae strains isolated of hospitalized and out patients acquired urinary tract infection. J. Isfahan Med. Sch. 29(142):695-705.
Khameneh ZR, Afshar AT (2009). Antimicrobial susceptibility pattern of urinary tract pathogens. Saudi J. Kidney Dis. Transpl. 20:251.
Langarizadeh N, Ahangarzadeh RM, Aghazadeh M, Hasani A (2011). Prevalence of multi-drug resistant (MDR) *klebsiella pneumoniae* among children and adults with urinary tract infection referred to tabriz teaching hospitals. Quarterly J. Biol. Sci. 4:9-17.
Mehr S, Powell C, Curtis N (2004). Cephalosporin resistant urinary tract infections in young children. J. Paediatr Child Health 40:48-52.
Oh E, Lee H, Lim HS, Park Y (2013). Epidemiology and Resistance Patterns of Bacterial Pathogens in Urinary Tract Infections in the Northern Gyeonggi-do Area during 2007-2011. Lab. Med. Online 3:34-39.
Qureshi AM (2005). Organisms causing urinary tract infection in pediatric patients at Ayub Teaching Hospital Abbottabad. J. Ayub Med. Coll. Abbottabad 17:72-4.
Ramirez M, Xie G, Marshall S, Hujer K, Chain P, Bonomo R, Tolmasky M (2012). Multidrug-resistant (MDR) Klebsiella pneumoniae clinical isolates: a zone of high heterogeneity (HHZ) as a tool for epidemiological studies. Clin. Microbiol. Infect. 18:254-258.
Rampure R, Gangane R, Oli AK (2013). Prevalence of MDR-ESBL producing Klebsiella pneumoniae isolated from clinical Samples. J. Microbiol. Biotech. Res. 3(1):32-39.
Senel S, Karacan C, Erkek N, Gol N (2010). A single-center experience of antimicrobial resistance patterns in pediatric urinary tract infection. Med. Prin. Prac. 19:359-363.
Tanvir R, Hafeez R, Hussain S (2012). Prevelance of Multi Drug Resistant E. coli in patients of UTI Registring at a Diagnostic Laboratory in Lahore, Pakistan. Pak. J. Zool. 44:707-712.
Ullah F, Malik SA, Ahmed J (2009). Antimicrobial susceptibility pattern and ESBL prevalence in Klebsiella pneumoniae from urinary tract infections in the North-West of Pakistan. Afr. J. Microbiol. Res 3:676-680.
Wikler MA, Cockerill FR, Bush K, Dudley MN, Eliopoulos GM (2009). Performance Standards for Antimicrobial Susceptibility Testing; Nineteenth International Supplement:29.
Yoon JE, Kim WK, Lee JS, Shin KS, Ha TS (2011). Antibiotic susceptibility and imaging findings of the causative microorganisms responsible for acute urinary tract infection in children: a five-year single center study. Korean J. Pediatr. 54:79-85.