Original Article

IDI: JCDR/2012/3490:1900

Microbiology Section

Prevalence of Ciprofloxacin Resistance Among Gram-Negative Bacilli in a Tertiary Care Hospital

MATHAVI SURESHKUMAR, SASIKALA GOPINATHAN, KONDIAN RANGACHARI RAJESH, INDRA PRIYADHARSINI

ABSTRACT

Background: Bacterial resistance to antibiotics is a growing therapeutic problem, both in the community and the hospitals, which involves all the antibiotics including fluoroquinolones. The decreased susceptibility to fluoroquinolones arises mainly due to single-step mutations in the *gyrA* and the *parC* genes, which encode the fluoroquinolones targets, the topoisomerase enzymes, thus conferring cross-resistance to the fluoroquinolones. In 1998, some mobile elements with a potential for the horizontal transfer of the quinolone resistance genes were described. The loci which are responsible for this plasmid-mediated quinolone resistance, which have been designated as qnr A, qnr B and qnr S, have been identified in the *Enterobacteriaceae* species.

Aim: This study was undertaken to evaluate the susceptibility pattern of the isolates to various antibiotics and to know the prevalence rate of ciprofloxacin resistance in our hospital.

Materials & Methods: A total of 734 gram-negative bacilli (GNB) which were isolated from various clinical samples over a period of six months, were subjected to antibiotic susceptibility testing. Isolates with resistance or with a decreased susceptibility to ciprofloxacin (≤20 mm) were then screened for their minimum inhibitory concentration (MIC) by using the E-test.

Results: Out of 734 GNB, 235 (32%) isolates were resistant to ciprofloxacin. The MIC of these isolates ranged from 4 to > 32µg/ml.

Conclusion: The resistance rate to ciprofloxacin was 32% in our study. Most of the ciprofloxacin resistant isolates were from urinary tract infections (UTI). The ciprofloxacin resistance was also closely associated with multi-drug resistance, thus limiting the treatment options. Ciprofloxacin resistance can be used as a general surrogate marker of multidrug resistance, thus limiting the already restricted treatment options. The considerably high MIC values for ciprofloxacin in this study reflected the extent of the treatment problems for these resistant isolates and a need for the continuous evaluation of the commonly used antibiotics.

Key Words: Fluoroquinolones, Gram-negative bacilli, Ciprofloxacin, MIC

INTRODUCTION

Ciprofloxacin is a broad-spectrum antibiotic which is active against both gram-positive and gram-negative bacteria, which belongs to the fluoroquinolone class [1]. Bacterial resistance is a growing therapeutic problem, both in the community and the hospitals, involving all the antibiotics, which include fluoroquinolones. A decreased susceptibility to fluoroquinolones arises mainly due to single-step mutations in the *gyrA* and the *parC* genes, which encode the fluoroquinolones targets, the topoisomerase enzymes [2]. In 1998, some mobile elements which were responsible for the horizontal transfer of the quinolone resistance genes were described [3,4]. This study was undertaken to evaluate the susceptibility of GNB to various antibiotics and to know the prevalence rate of ciprofloxacin resistance in our hospital.

MATERIALS AND METHODS

A total of 734 gram-negative bacilli which were isolated from various clinical samples i.e., urine, pus, sputum, blood etc, received in the Microbiology Laboratory over a period of six months (May 2011-October 2011) were subjected to the study. The identification of the isolates was done, based on their colony morphology on MacConkey's agar and blood agar and on the standard biochemical reactions [5]. Antibiotic susceptibility testing was done on Muller Hinton agar (MHA) after standardizing the suspension to 0.5 McFarland's standards (5) for the antibiotics, amoxycillin (25 µg), cotrimoxazole (23.75/1.25 µg), cephatoxime(30µg), ceftazidime(30 µg), nalidixic acid (30 µg), ciprofloxacin (5 µg), amikacin (30 µg), imipenem (10 µg) and nitrofurantoin (300 µg-for urinary isolates). The results were interpreted as were recommended by the CLSI guidelines [6]. Quality controls were carried out once a week with strains of E. coli ATCC 25922 and P. aeruginosa ATCC 27853. Isolates with resistance or with decreased susceptibility to Ciprofloxacin (≤20mm) were subjected to further study.

E-TEST

The resistance to ciprofloxacin was confirmed by breakpoint minimum inhibitory concentration (MIC in µg/ml) by using E-test strips. The isolates with MIC value ≥ 4 µg/ml were defined as resistant isolates, as outlined by CLSI guidelines [6].

RESULTS

Escherichia coli (27.92%) was the predominant isolate which was found among the GNB, followed by Klebsiella species (25.74%) and Pseudomonas species (24.93%), as shown in [Table/Fig-1].

Out of 734 gram-negative bacilli, 235(32%) isolates were resistant to ciprofloxacin. High rates of resistance were observed for amoxycillin, followed by cotrimoxazole, nalidixic acid and cephatoxime, while low levels of resistance were observed for nitrofurantoin, amikacin and imipenem, as shown in [Table/Fig-2]. The resistance rate for ciprofloxacin was 32%. The MIC of ciprofloxacin for these isolates ranged from 4 to >32 µg/ml [Table/Fig-3]. The isolated bacteria showed wide differences in their susceptibility to ciprofloxacin.

A high rate of resistance to ciprofloxacin was observed among Pseudomonas species, Acinetobacter, Proteus and Klebsiella, followed by E. coli.

DISCUSSION

The resistance rate for ciprofloxacin was 32% in our study. Most of the ciprofloxacin resistant isolates were obtained from UTI samples. This may be because fluoroquinolones are preferred as the initial agents for empiric therapy in UTI, because of their excellent activity against the pathogens which are commonly encountered in UTI [7]. This emphasises the importance of the re-assessment of the antibiotics which are used in the empiric treatment of UTIs. Most of the isolates from UTIs were susceptibile to nitrofurantoin, amikacin and imipenem. This finding was in accordance with the finding of a study which was conducted by Zakaria El Astal [8]. These data suggest that nitrofurantoin can still be successfully used in the treatment of UTI.

The ciprofloxacin resistance was also closely associated with multi-drug resistance, thus making the treatment options limited [9]. Ciprofloxacin resistance can be used as a general surrogate marker of multi-drug resistance. Hence, it severely limits the already restricted treatment options. This finding was in accordance with the finding of a study which was conducted by David et al [10]. The high resistance pattern which was seen in our study was probably due to the inappropriate prescribing of antibiotics and the poor infection control strategies. But the antibiotic history could not be properly elicited from the patients in this study.

The drugs which showed maximum activity against most of the isolates were imipenem and amikacin. Though carbapenems remain the final options for treating these infections, there is a possibility that the increasing use of carbapenems may lead to a rapid emergence of carbapenem resistance.

CONCLUSION

The considerably high MIC values for ciprofloxacin in this study reflect the limited treatment options which are available for these resistant isolates and a need for the continuous evaluation of the commonly used antibiotics. Repeated surveillance, the formulation of an antibiotic policy, the prudent prescription of antibiotics and the recycling of antibiotics are the possible routes which can be used to curb the rapid emergence and the spread of these resistant isolates.

Sl. No.	Organism	Total number isolated	Percentage
1.	Escherichia coli	205	27.92%
2.	Klebsiella species	189	25.74%
3.	Pseudomonas aeruginosa	183	24.93%
4.	Pseudomonas species	86	11.71%
5.	Proteus species	45	6.13%
6.	Acinetobacter	26	3.54%

[Table/Fig-1]: Total number of Gram-negative Bacilli isolated from various clinical samples (n=734)

Sl.No.	Antibiotics	Total no of Sensitive isolates (%)	Total no of Resistant isolates (%)
1.	Amoxycillin	198 (27%)	536 (73%)
2.	Cotrimoxazole	249 (34%)	485 (66%)
3.	Nalidixic acid	352 (48%)	382 (52%)
4.	Cephatoxime	433 (59%)	301 (41%)
5.	Ceftazidime	558 (76%)	176 (24%)
6.	Ciprofloxacin	499 (68%)	235 (32%)
7.	Amikacin	602 (82%)	132 (18%)
8.	Imipenem	631 (86%)	103 (14%)
9.	Nitrofurantoin (for urinary isolates=308 GNB)	253 (82%)	55 (18%)

[Table/Fig-2]: Antibiotic Susceptibility pattern of the isolates to various antibiotics (n=734)

Ciprofloxacin MIC values	4µg/ml	8µg/ml	16µg/ml	32µg/ml	>32µg/ml
Total No. of isolates	59 (25%)	33 (14%)	28 (12%)	35 (15%)	80 (34%)

[Table/Fig-3]: MIC values of the resistant isolates to Ciprofloxacin (n=235)

REFERENCES

[1] Drlica K, Zhao XK. DNA gyrase, topoisomerase IV and 4-quinolone. *Microbiol Mol Biol Rev* 1997; 61 (3): 377-92.
[2] Hooper DC. The emerging mechanisms of fluorquinolone resistance. *Emerg Infect Dis* 2001; 7:337-41.
[3] Martinez-Martinez L, Pascual A, Jacoby GA. Quinolone resistance from a transferable plasmid. *Lancet* 1998; 351:797-9.
[4] Nordmann P, Poirel L. The emergence of plasmid-mediated resistance to quinolones in Enterobacteriaceae. *J Anti-microb Chemother* 2005; 56:463-9.
[5] Mackie and McCartney. Textbook of Practical Medical Microbiology. 14th edition; p.153, 416.
[6] Clinical and Laboratory Standards Institute. *Performance Standards for Anti-microbial Susceptibility Testing*. Wayne, Pa :Eleventh Informational Supplement. : 2001; Document M100-S11.
[7] Schaeffer AJ. The expanding role of fluoroquinolones. *Am J Med* 2002; 113(suppl 1A):45S–54S.
[8] Astal ZE. The increasing ciprofloxacin resistance among prevalent urinary tract bacterial isolates in Gaza Strip. *Singapore Med J* 2005; 46(9):457-60.
[9] Kang CI, Kim SH, Kim DM, et al. The risk factors for ciprofloxacin resistance in bloodstream infections which were caused due to extended-spectrum β-lactamase producing Escherichia coli and Klebsiella pneumonia. *Microb Drug Resist* 2004; 10: 71-76.
[10] Paterson DL, et al. Epidemiology of ciprofloxacin resistance and its relationship to extended spectrum β-lactamase production in Klebsiella pneumoniae isolates, which caused bacteremia. *Clin Infect Dis* 2000; 30 (3): 473-78.

AUTHOR(S):
1. Mathavi Sureshkumar
2. Sasikala Gopinathan
3. Kondian Rangachari Rajesh
4. Indra Priyadharsini

PARTICULARS OF CONTRIBUTORS:
1. Assistant Professor,(Microbiology)
2. Assistant Professor (Microbiology)
3. Assistant Professor (Microbiology)
4. Assistant Professor (Microbiology)

Name Of Department(S)/Institution(S) To Which The Work Is Attributed:
Vinayaka Mission's Kirupananda Variyar Medical College, Salem, Tamil Nadu, India.

NAME, ADDRESS, TELEPHONE, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Dr. S. Mathavi M.D(Microbiology), Assistant Professor in Microbiology, Vinayaka Mission's Kirupananda Variyar Medical College, Salem-636308, Tamilnadu, India. Phone: 09600251333
E-mail: drmathavi@dr.com

FINANCIAL OR OTHER COMPETING INTERESTS:
None.

Date Of Submission: **Oct 24, 2011**
Date Of Peer Review: **Dec 22, 2011**
Date Of Acceptance: **Dec 29, 2011**
Date Of Publishing: **Apr 15, 2012**