Original Article

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Microbiology Section

Fosfomycin: An Alternative Therapy for the Treatment of UTI Amidst Escalating Antimicrobial Resistance

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ABSTRACT

Introduction: Urinary tract infections (UTIs) are the most commonly encountered infectious diseases. The current study was undertaken with a dual purpose, to provide an insight into the current scenario of the microorganisms causing UTI, their antimicrobial sensitivity patterns and also try and evaluate the activity of fosfomycin against *E. coli*, both ESBL producers as well as non-producers.

Materials and Methods: The study was conducted prospectively in the Department of Microbiology of a tertiary care hospital from January to June 2014. A total of 358 isolates from the urinary samples of the patients with a diagnosis of urinary tract infection were included in the study. Antibiotic sensitivity testing and extended spectrum beta lactamase (ESBL) production testing was done as per CLSI guidelines.

Results: These represented 297 (82.9%) gram-negative isolates and 61 (17%) gram-positive isolates. The 297 gram-negative isolates represented 265 (89.2%) members of the *Enterobacteriaceae*, 185 (69.8%) of which were *Escherichia coli*, 66 (24.9%) *Klebsiella* spp. and 14 (5.28%) *Proteus* spp. Non-fermentative *Pseudomonas* spp were isolated from 8.9% cases. Amongst the Gram negative isolates tested, 78 (21.8%) formed extended spectrum beta-lactamases. Of the total 358 isolates tested, 338 (94.4%) were found to be susceptible to fosfomycin.

Conclusion: Fosfomycin showed good activity against both ESBL-producing and ESBL-negative *E. coli* isolates. The main finding of our study is that fosfomycin exhibits excellent antimicrobial activity even against the isolates with relatively high levels of antimicrobial resistance and hence can be a useful drug in our armamentarium.

Keywords: Antimicrobial agents, Antimicrobial sensitivity, Extended spectrum beta lactamase (ESBL)

INTRODUCTION

Urinary tract infections (UTIs) are the most commonly encountered infectious diseases by clinicians in developing countries [1,2]. Very often, in clinical practice, we encounter symptomatic uncomplicated UTIs being empirically treated. Normally bacteriological cultures are not very aggressively done and if at all done the culture reports are not available at the time when the treatment is initiated. Besides, contamination of organisms grown is also commonly reported which could be because of various factors like improper sample collection or an increased time lag between receipt of sample in the laboratory and its processing and many more. In such a scenario it is not wise to withhold treatment till the availability of the results. Also, antibiotic susceptibility profile of the commonly isolated bacterial pathogens varies not only from time to time but also from one geographical place to another.

Increasing multidrug-resistant (MDR) pathogens contribute considerably to increasing proportion of urinary tract infections (UTIs) as they limit treatment options [3,4]. The emergence of a plethora of multidrug-resistant (MDR) organisms has prompted re-evaluation of non-traditional antibiotics. The introduction of antimicrobial agents that are not much used in clinical practice, may show a ray of hope. One such drug that has caught attention of clinicians in recent time is Fosfomycin, a phosphonic acid derivative and also known as phosphomycin or phosphonomycin. Fosfomycin was discovered in Spain in 1969 from cultures of *Streptomyces* and has been used in Europe since then, but introduced in other countries recently. It is a broad spectrum antibiotic, active against both Gram-positive and Gram-negative bacteria. Fosfomycin is used against treatment of sepsis, soft-tissue infection, UTI and cystitis and can be used synergistically with many beta-lactams and aminoglycosides too.

Although known for more than four decades clinical data regarding the use of fosfomycin for the treatment of UTIs due to various MDR pathogens is very limited. The current study was therefore undertaken with a dual purpose, to provide an insight into the current scenario of the microorganisms causing UTI, their antimicrobial sensitivity patterns and also try and evaluate the activity of fosfomycin against *E.coli*, both ESBL producers as well as non producers.

MATERIALS AND METHODS

The study was conducted prospectively in the Department of Microbiology, Mahatma Gandhi Medical College during a six month period from January 2014 to June 2014 after receiving ethical clearance from the institution review board. In-patients and out-patients with clinical evidence of cystitis were included in the study. Fresh, mid-stream urine samples were collected aseptically in sterile containers and were submitted to the clinical microbiology laboratory.

The samples received were inoculated onto Blood Agar and Mac Conkey Agar. After an aerobic incubation at 37°C, the plates showing significant growth as per the Kass count (single species count of more than 10^5 organisms per ml of urine) were processed further and the isolates were identified up to the species level by using standard biochemical tests [5-8]. Antibiotic sensitivity testing was done by the Kirby Bauer disc diffusion method as per CLSI guidelines [9,10]. The following antibiotic discs (drug concentrations in µg) were used: Amoxicillin (20), Amoxicillin/clavulanic acid (20/10), Gentamycin (10), Cefuroxime (30), Cefotaxime (30), Ceftazidime (30), Ceftazidime/clavulanic acid (30/10), Cefepime (30), Cotrimoxazole (1.25/23.75), Ciprofloxacin (5), Amikacin (30), Nitrofurantoin (300), Imipenem (10), Cefoxitin (30), fosfomycin (200), Aztreonam (30) and Nalidixic acid (30).

The isolates were also tested for the production of extended spectrum beta lactamases (ESBL) according to the CLSI guidelines [10]. Cefotaxime (30 µg), Ceftazidime (30 µg) and Ceftriaxone (30 µg) discs were used to screen for the ESBL production. The isolates which tested positive by the screening test were subjected to confirmatory test. Ceftazidime (30 µg) and Ceftazidime /clavulanic acid (30 µg /10µg) discs were used for the confirmatory test. The results were interpreted according to the CLSI guidelines.

RESULTS

The study was performed on 358 isolates from the urinary samples of the patients with a diagnosis of urinary tract infection. Of the patients, 78 were outpatients, and 280 were inpatients (185 from wards and 95 from ICU) [Table/Fig-1]. There were 105 male and 253 female patients.

The 358 urinary bacterial isolates represented 297 (82.9%) gram-negative isolates and 61 (17%) gram-positive isolates. The 297 gram-negative isolates represented 265 (89.2%) members of the *Enterobacteriaceae*, 69.8% of which were *Escherichia coli*, followed by *Klebsiella* spp. and *Proteus* spp. Non fermentative *Pseudomonas* spp were isolated from 8.9% cases. The 61 tested Gram positive isolates consisted of 54.09% Coagulase negative Staphylococcal isolates, 32.7% *Enterococci* isolates and 13.11% *Staphylococcus aureus* isolates [Table/Fig-1].

Bacteria (n = 358)	No. of strains	OPD	Ward	ICU
Gram negative				
Escherichia coli	185	49	102	34
Klebsiella	66	16	31	19
Proteus spp.	14	01	07	06
Pseudomonas aeruginosa	32	07	20	05
Gram positive				
CoNS	33	03	07	23
Enterococci	20	02	12	06
Staphylococcus aureus	08	00	06	02
Total	358	78	185	95

[Table/Fig-1]: Stratified distribution of different bacterial species in subjects with uncomplicated urinary tract infection.

Amongst the Gram negative isolates tested, 78 (21.8%) formed extended spectrum beta-lactamases (ESBL), which include 26 (33.3%) from the outpatients and 52 strains (66.6%) from the inpatients. Statistically the rate of ESBL formation amongst the inpatients is found to be significantly higher (p=0.001).

The antibiotic susceptibility profile of all the urinary tract isolates is presented in [Table/Fig-2]. Of the total 358 isolates tested, 338 (94.4%) were found to be susceptible to fosfomycin. These included 277 Gram negative and 61 Gram positive strains.

Specifically, fosfomycin was active against almost all tested *S. aureus*, Coagulase negative staphylococcal isolates and *Enterococcus* spp. Considerable rates of susceptibility to fosfomycin were found for *E. coli*, *Proteus mirabilis*, *Pseudomonas aeruginosa* as well as *Klebsiella pneumoniae.*

Comparison of Antimicrobial Susceptibilities to ESBL-Producing and Non ESBL Isolates

In this study, the rates for extended spectrum beta lactamases (ESBL) production were found to be 52.9%, 48.5%, 46.8%, and 42.8% for *E. coli*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa* and *Proteus mirabilis*, respectively. The ESBL producing isolates showed significantly higher resistance rates to cefepime, ciprofloxacin, co-trimoxazole, Nalidixic acid and gentamycin than the ESBL-negative isolates. Fosfomycin showed good activity against both ESBL-producing and ESBL-negative *E. coli* isolates [Table/Fig-3].

	Fosfomycin sensitivity %	Fosfomycin resistance %
ESBL Producer	95	5
Non ESBL Producer	98.4	1.6

[Table/Fig-3]: Comparison of Fosfomycin sensitivity (%) amongst ESBL producing and non ESBL producing strains of *E. coli*

The main finding of our study is that fosfomycin exhibits excellent antimicrobial activity even against the isolates with relatively high levels of antimicrobial resistance.

DISCUSSION

The spread of extended-spectrum β-lactamases (ESBLs) among isolates of Enterobacteriaceae both from community and health-care settings is quite disturbing. While options like carbapenems, tigecycline, β-lactam/β-lactamase inhibitor combinations exist, they are flawed by various factors like their unfriendly regimens and parenteral use, thereby negating their role in the outdoor patients. There is definitely need for a newer drug that is orally active, has low

	E. coli n=185(%)	*Proteus Mirabilis* n=14(%)	*Klebsiella* n=66(%)	*Pseudo* n=32(%)	CoNS n=33(%)	*Staph.* n=8(%)	*Entero* n=20(%)
Nitrofurantoin	175 (94.5)	9 (64.2)	55 (83.3)	26 (81.2)	33 (100)	4 (50)	16 (80)
Imipenem	173 (93.5)	11 (78.5)	62 (93.9)	21 (65.6)	ND	ND	ND
Fosfomycin	180 (97.2)	13 (92.8)	56 (84.8)	28 (87.5)	33 (100)	8 (100)	20 (100)
Cefoxitin	ND	ND	ND	ND	31 (93.3)	4 (50)	ND
Amikacin	105 (56.7)	9 (64.2)	48 (72.7)	10 (31.2)	23 (69.6)	6 (75)	ND
Gentamycin*	114 (61.6)	8 (57.1)	46 (69.6)	4 (12.5)	14 (42.4)	4 (50)	7* (35)
Cefotaxime	130 (70.2)	7 (50)	40 (60.6)	ND	8 (24.2)	2 (25)	ND
Aztreonam	133 (71.8)	11 (78.5)	50 (75.7)	18 (56.2)	ND	ND	ND
Ciprofloxacin	93 (50.2)	6 (42.8)	31 (46.9)	13 (40.6)	20 (60.6)	8 (100)	ND
Cefuroxime	ND	ND	ND	ND	23 (69.6)	4 (50)	ND
Nalidixic Acid	139 (75.1)	5 (35.7)	22 (33.3)	8 (25)	9 (27.2)	4 (50)	ND
Augmentin	105 (56.7)	8 (57.1)	43 (65.1)	23 (71.8)	19 (57.7)	6 (75)	ND
Co-trimoxazole	85 (45.9)	5 (35.7)	19 (28.7)	ND	21 (63.6)	3 (37.5)	ND
Amoxycillin	44 (23.7)	4 (28.5)	13 (19.6)	ND	2 (6)	1 (12.5)	0 (0)
Cefepime	172 (92.9)	11 (78.5)	57 (86.3)	32 (100)	20 (62)	7 (87.5)	ND
Vancomycin	ND	ND	ND	ND	33 (100)	8 (100)	20 (100)
ESBL	98 (52.9)	6 (42.8)	32 (48.4)	15 (46.8)	-	-	-

[Table/Fig-2]: Antibiotic sensitivity (%) in urinary isolates.
* Genatmycin (120 ug) for *Enterococcus* spp.

levels of existing resistance and also doesn't encourage antimicrobial resistance in future.

Fosfomycin is an old broad-spectrum bactericidal antibiotic agent that acts by inactivating the enzyme phosphoenolpyruvate synthetase, required in assembly of glycan and peptide portion of peptidoglycan, thus disrupting bacterial cell-wall synthesis. It has a broad spectrum of activity against wide range of bacteria such as methicillin-sensitive *Staphylococcus aureus* (MSSA), cephalosporin- and penicillin-resistant *Streptococcus pneumoniae*, methicillin-resistant *S. aureus* (MRSA), and *Enterococcus species*, even the vancomycin-resistant strains [11]. Recent reports show very encouraging in *vitro* activity against MDR Gram negative pathogens also [7,8]. Fosfomycin is mainly used in the treatment of UTIs, particularly those caused by *E. coli* and *Enterococcus faecalis* [11].

Due to its improved pharmacokinetics Fosfomycin is encouraged for use in UTIs; the mean peak urinary concentration of an oral single dose of 3 g fosfomycin tromethamine occurs within 4 hour, while concentrations sufficient to inhibit the majority of the urinary pathogens can be maintained for 1 to 2 days [10]. This easy dosage schedule ensures compliance thereby discouraging any resistance that may occur precisely because of faulty patient habits. Also, fosfomycin is spared from the effect of various mechanisms of multiple resistances to antimicrobial drugs, because of its unique chemical structure and mechanism of action [12]. Oral fosfomycin is well tolerated with a low incidence of adverse events and is very pocket friendly. It has been approved as an oral single-dose treatment for acute uncomplicated cystitis [13].

In a study by de Cueto et al., the invitro susceptibility of 428 extended-spectrum beta-lactamase (ESBL) producing *Escherichia coli* and *Klebsiella pneumoniae* strains was determined to fosfomycin and it showed high activity against all these strains [14]. Falagas et al., and Maraki et al., in their respective studies at Greece have also shown very encouraging susceptibility results. In Study by Maraki et al., fosfomycin was reported to be active in vitro against a considerable percentage of urinary isolates, which exhibited high antimicrobial resistance against the conventionally used antimicrobial agents for the treatment of UTIs [12,15,16]. In their retrospective chart review of 41 hospitalized patients, infected with multi-drug resistant urinary tract infections (UTIs) and treated with fosfomycin tromethamine, Neunar et al., found *Invitro* fosfomycin susceptibility to be 86% [16]. In a study, conducted in 12 medical centres in China, the clinical efficacy rates of fosfomycin tromethamine for acute uncomplicated cystitis, recurrent lower urinary tract infection and complicated lower urinary tract infection were reported to be 94.71%, 77.22% and 62.69%, respectively [17].

In their retrospective matched cohort study, at the University of Michigan Health System, Nagel et al., reported identical clinical success rate in both the fosfomycin and control groups. However, the average days of treatment was found to be lower in the fosfomycin group [18].

Similar to results of given study [Table/Fig-3], Karlowsky et al., too reported 99.4% fosfomycin susceptibility against urinary isolates of *Escherichia coli,* collected from 2010 to 2013 as part of the Canadian national surveillance study CANWARD. β-lactamase-producing isolates and AmpC-producing isolates of *E. coli also showed* 94.9% and 96.6% susceptibility respectively [19]. In a study from Southern part of India, Sahni RD and co-workers reported Fosfomycin susceptibility of 83 and 99% for *E. coli* and *Enterococcus* spp, respectively. Among these, 81% of extended-spectrum β-lactamase producing *E.coli* was susceptible to fosfomycin, while 75.7% of multidrug resistant were found to be susceptible to fosfomycin [20].

Our study has found that fosfomycin is a reliably active antimicrobial drug against Enterobacteriaceae, even those that produce ESBL, particularly *E coli*. The susceptibility of fosfomycin against all isolates tested was 93.2%, with *E.coli* showing the highest susceptibility rate of 97.7%. This finding might be important for the treatment of community-acquired ESBL-associated urinary tract infections, which are mostly caused by *E coli*. Similar results have been presented by other contemporary studies [7,10,15]. Further RCTs are required to evaluate the efficacy of intravenous fosfomycin for the management of infections due to MDR pathogens.

CONCLUSION

Though fosfomycin is used rampantly in Europe for the management of UTI, the drug is yet to be introduced in Indian markets. Ours being a fosfomycin naïve population, has shown high susceptibility towards urinary isolated, including MDR isolates. However, more such studies and clinical trials are needed, before the clinicians and infectious disease specialists can whole heartedly and comfortably use this no-less-than wonder drug. It is active invitro against a large percentage of urinary isolates, and in this era of high drug resistance rates, reported even among community-acquired uropathogens, it may provide a valuable alternative option for the treatment of cystitis. Fosfomycin can therefore become a useful antibiotic agent in our armamentarium for the treatment of UTIs.

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