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ANTIMICROBIAL SUSCEPTIBILITY OF FOSFOMYCIN IN BACTERIAL ISOLATES FROM URINARY TRACT INFECTION : A STUDY FROM TERTIARY HEALTHCARE CENTER IN HIMACHAL PRADESH

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ABSTRACT **BACKGROUND:** Urinary tract infections (UTIs) are common with an annual global incidence of at least 250 million cases; and Escherichia coli is the most common pathogen. Many antibiotics once used to treat UTI are now ineffective due to the development of antimicrobial resistance. Fosfomycin, discovered in late 1960s, has rekindled clinical interests because of reported susceptibilities of current pathogens to the agent. This study was done with the objective to determine in vitro fosfomycin susceptibility of common uropathogens and determining the antimicrobial susceptibility pattern of these organisms. **MATERIAL AND METHOD:** Retrospective study was conducted in the Department of Microbiology for a duration of 2 years from October 2019 to September 2021. Urine samples were received and culture was done on MacConkey agar and AST was performed on significant bacteriuria ≥105 CFU /ml with fosfomycin as per CLSI guidelines. **RESULT:** Total of 9442 urine samples were collected during the duration of 2 years. Out of these 1657(17.5%) showed significant growth. Male to female ratio was 1:1.9 in our study. Majority of the bacteria isolated were uropathogenic E.coli 960(57.9%) followed by Klebsiella spp 185(11.1%). Susceptibility to fosfomycin was seen majorly in all enterobacterales with 96.7% sensitivity to E.coli and 100% sensitivity towards Staphylococcus aureus and Enterococcus spp. **CONCLUSION:** In conclusion, our study indicates that fosfomycin is active in vitro against a considerable percentage of urinary isolates, which simultaneously exhibit high rates of antimicrobial drug resistance to the conventionally used antimicrobial agents.

KEYWORDS :

INTRODUCTION:

Urinary tract infections (UTIs) are common with an annual global incidence of at least 250 million cases; and Escherichia coli is the most common pathogen[1]. Antimicrobial resistance is increasing worldwide, and this has resulted in infections that are more difficult to treat and are associated with higher mortality, morbidity and cost. Many antibiotics once used to treat UTI are now ineffective due to the development of antimicrobial resistance[2]

Enterobacterales are the main causative agent of uncomplicated urinary tract infections in immunocompetent hosts and opportunistic infections in immunocompromised hosts. In recent years, fluoroquinolone-resistant and/or extended spectrum β-lactamase-(ESBL-) producing isolates have been frequently isolated from such patients [3–6].Fosfomycin, discovered in late 1960s, has rekindled clinical interests because of reported susceptibilities of current pathogens to the agent.

This old 'forgotten' drug is now being re-explored for the treatment of cystitis. It has the unique properties of not sharing any structural similarity and lack of cross-resistance with other antimicrobial agents.[7]

Fosfomycin is a phosphonic acid derivative with antibacterial activity against a wide range of gram-negative pathogens and some gram-positive pathogens. It inhibits bacterial cell wall synthesis and is bactericidal against most Enterobacterales isolates, as well as many strains of other members of the family Enterobacteriaceae.[8] It is best absorbed if given before food intake and is excreted in urine.[9] It achieves high concentration in urine of 2000 µg/ml and maintains high levels for over 24 hours with minimal impact on gastrointestinal flora .[10] Hence, single-time oral therapy with fosfomycin has been recommended in uncomplicated UTI.[11]

This study was done with the objective to determine in vitro fosfomycin susceptibility of common uropathogens and determining the antimicrobial susceptibility pattern of these organisms.

MATERIAL AND METHOD:

This retrospective study was conducted in the Department of Microbiology DRPGMC, Kangra at Tanda for a period of 2 years from October, 2019 to September 2021. Urine samples were received in the laboratory during the time period in sterile urine containers. Samples were processed on the same day. A semi quantitative method was adopted for primary isolation of organisms using a calibrated loop of 4 mm diameter which delivers 10µl of urine.[12]

The specimens were inoculated on Mac Conkey agar plates and incubated aerobically at 37°C for 24-48 hours. Culture plates with colony counts of ≥ 105 cfu/ml were considered positive for UTI. Cultures that showed no growth in 24 to 48 hours indicated absence of infection as sterile. From positive cultures, Uropathogenic isolates were identified according to the standard biochemical reactions.[13] A significant bacterium was considered if urine culture yielded $\geq 10^5$ CFU /ml and $<10^5$ CFU/ml is taken as insignificant.[14]

According to the standard operational procedures, in vitro antimicrobial susceptibility testing was done on Mueller-Hinton agar (Hi-Media Lab Ltd, India) using Kirby-Bauer disc diffusion method. A suspension of the test organism was made in sterile normal saline and turbidity adjusted to 0.5 McFarland standards. Uropathogenic isolates were uniformly seeded over the surface of Mueller Hinton agar plates. The plates were allowed to dry for 10 minutes before application of antibiotic discs. The plates were incubated at 37°C for 16-18 hours. After incubation clear zones around the antibiotic discs were measured with a ruler and recorded in millimeters. Fosfomycin (200µg) antibiotic disc was used. Susceptibility and resistance data was interpreted according to Clinical laboratory Standards Institute guidelines.[15]

RESULTS:

A total of 9442 isolates were received in the Department of

Microbiology at DRPGMC Kangra at Tanda, during the period of two years from October 2019 to September 2021. Significant growth was seen in 1657(17.5%) urine samples and the rest 7785(82.5%) were either sterile ,contaminants or insignificant.(figure 1)

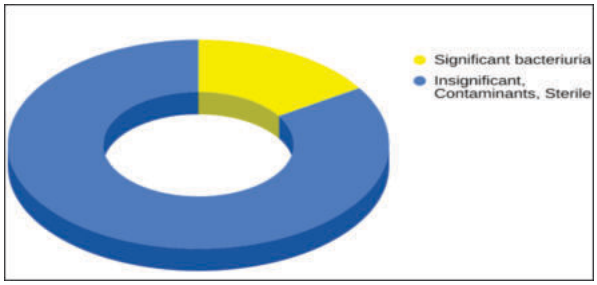

Figure 1: Distribution Of Samples As Significant And Insignificant, Contaminants Or Sterile

Out of 1657 significant bacteriuria samples 570 (34.3%)were males and 1087(65.6%) were females. Male to female ratio was 1:1.9(Figure 2)

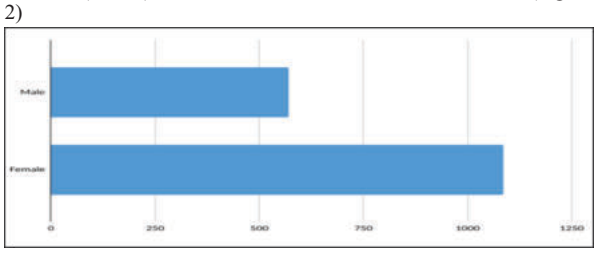

Figure 2: Distribution On The Basis Of Sex

Uropathogenic Escherichia coli was in majority with 960(57.9%) followed by Klebsiella spp 185(11.1%), Acinatobacter spp 107(6.4%) and least were Morganella spp 2(0.1%) followed by Enterbobacter spp 20(1.2%) respectively (Figure 3)

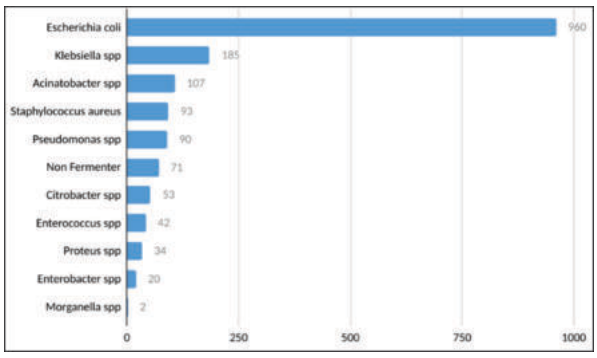

Figure 3: Distribution Of Bacterial Isolates

Susceptibility of fosfomycin was seen in the majority of the bacterial isolates obtained from urinary tract infection. 100% susceptibility was seen in Staphylococcus aureus and Enterococcus. (Table 1)

Table 1: Antimicrobial Susceptibility Of Fosfomycin To Various Bacterial Isolates

Bacterial Isolate	Susceptible(S)	Resistance(R)
Escherichia coli	929(96.7%)	31(3.7%)
Klebsiella spp	154(83.3%)	31(16.7%)
Acinatobacter spp	87(81.3%)	20(18.7%)
Staphylococcus aureus	93(100%)	0%
Pseudomonas spp	71(78.9%)	19(21.1%)
Non fermenter	57(80.2%)	14(19.8%)
Citrobacter spp	44(83%)	09(17%)
Enterococcus spp	42(100%)	0%
Proteus spp	25(73.5%)	09(26.5%)
Enterobacter spp	14(70%)	06(30%)
Morganella spp	02(100%)	0%
Total	1518(91.6%)	139(8.4%)

DISCUSSION:

With the increases in antibiotic resistance among Enterobacterales and other gram negative isolates over the past several decades, surveillance data have become critical for appropriate empiric selection of antibiotic therapy.[16]

In our study only 1657(17.5%) urine samples showed significant bacterial growth. Rana et al in their study showed similarity results with 15.6% of significant bacterial growth of $\geq 10^5$ CFU /ml from urine samples.[16] Females were in majority in our study with male to female ratio of 1:1.9..Females were more susceptible to UTI as compared to males due to the short length of urethra, absence of prostatic secretion, pregnancy and easy contamination of the tract with faecal flora.[17] Similar results were seen in by Rana et al [18] in their study with male to female ratio of 1:1.9.[18]

Escherichia coli was the major bacterial isolate obtained from the urine samples in our study with 960(57.9%) of isolates followed by Klebsiella spp 185(11.1%) from total positive samples.Maraki et al and shreenivas et al also showed similarity with our study with majority of Escherichia coli and Klebsiella spp in their studies respectively.[19]

In the present study we focused on the susceptibility of fosfomycin, an older antibiotic in the era of newer antibiotics towards bacterial isolates obtained from urine samples. Escherichia coli as a major pathogenic bacteria of urinary tract infection show high susceptibility to fosfomycin with 96.7%. Similarly other bacterias from family enterobacterales showed good in vitro results with fosfomycin Klebsiella showed 83.3%, Citrobacter spp with 83%, Enterobacter with70%, Proteus with 73.5% and Morganella with 100% susceptibility. Gram positive bacterial isolates showed excellent results with the fosfomycin with 100% susceptibility to Staphylococcus aureus and Enterococcus spp. Similarly Maraki et al in their study showed susceptibility thoroughly.[19]

Emerging resistance to many regularly used antibiotics, attributable to the irrational use and to the ease of availability of antibiotics over the counter, there is definitely a need for introducing newer drugs that can be used to combat the current situation. Hence, in this scenario, fosfomycin, finds its utility and could be a preferable option for treatment of UTI having demonstrated an excellent in vitro activity against common uropathogens, including MDR isolates. Similar outcomes were seen by other authors too in their studies in various countries[20, 21]

CONLCLUSON

In conclusion, our study indicates that fosfomycin is active in vitro against a considerable percentage of urinary isolates, which simultaneously exhibit high rates of antimicrobial drug resistance to the conventionally used antimicrobial agents for the treatment of UTIs. Consequently, fosfomycin may be considered a useful antibiotic agent in our armamentarium for the treatment of UTIs.

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