Original Research Article

Comment [K1]:

Antimicrobial Susceptibility Profile of Fosfomycin and Nitrofurantoin Despite Dissemination of Fluoroquinolones and Trimethoprim/Sulfamethoxazole Resistant Urinary Tract Isolates

Abstract:

The widespread growth of multidrug-resistant (MDR),extended drug resistant(XDR) uropathogens and the shortage of new antimicrobials are the most significant obstacles challenging the treatment of urinary tract infections. The study is aimed to identify the antimicrobial susceptibility profile against MDR and XDR uropathogens. A total of 2485 urine cultures, 361 uropathogens showed growth. The antimicrobial susceptibility had been determined by the Kirby-Bauer disk diffusion method, following the Clinical and Laboratory Standards Institute's guidelines. Eighty-nine percent of the patients had Multidrug-resistant microorganisms. In comparison to fluoroquinolones and trimethoprim/sulfamethoxazole, fosfomycin and nitrofurantoin demonstrated a significantly higher sensitivity rate against uropathogens, including MDR and XDR uropathogens, in both gender groups with community-acquired and nosocomial UTIs (P<0.001). Fosfomycin revealed the highest sensitivity rate, about 94.8%. Klebsiella pneumonia and E.coli showed the highest resistance rate against fosfomycin in 3.7% and 3.4% of the cases. Nitrofurantoin showed a similar sensitivity rate both in community and hospitalized patients in 86.1%. Fluoroquinolones (61%) and trimethoprim/sulfamethoxazole (86.6%) revealed the highest resistance rate against uropathogens. The prevalence of extended-spectrum beta-lactamases producing pathogens was 10.2%. Fosfomycin and nitrofurantoin revealed a higher sensitivity rate against gram-negative MDR uropathogens in community and nosocomial UTIs compared to fluoroquinolones and trimethoprim/sulfamethoxazole.

Comment [K2]: A totla of 2485 urine samples were processed from - -----patients? 361 uropathogens were grown.

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Comment [K4]: patients or samples. How many patients were included since more than one urine samples can be from one and same patient

Comment [K5]: how many were XDR?

Keywords: Urinary Tract Infection; Fosfomycin; Nitrofurantoin; fluoroquinolones.

Comment [K6]: MDR and XDR pathogens

Introduction:

Urinary tract infections (UTIs) are the most common infections in both community and nosocomial infections (1,2). Globally, over 150 million UTI cases occur each year, costing the global economy over $6 billion US dollars (3). The possibility of colonization and progression to symptomatic UTIs is responsible for host factors, including anatomical or functional abnormalities and genetic predisposition, and microbial factors such as virulence characteristics (4). The widespread growth of multidrug-resistant (MDR) and extensively drug-resistant (XDR) uropathogens and the shortage of new antimicrobials against such pathogens are the most significant obstacles challenging the treatment of bacterial infections (5). Despite the spread of antimicrobial resistance and the lack of new antimicrobials, physicians reverted to older antimicrobials such as fosfomycin and nitrofurantoin, which gained favor due to their activity against gram-positive and gram-negative uropathogens (6). Extended-spectrum beta-Lactamase (ESBL)-producing uropathogens is increasing in community and hospitalized patients(7). Escherichia coli is the most frequently isolated uropathogen in uncomplicated and complicated urinary tract infections. It is resistant to most oral antibiotics, including fluoroquinolones, trimethoprim-sulfamethoxazole, and beta-lactam antibiotics (8). Other common uropathogens include Klebsiella spp, Staphylococcus, Pseudomonas Aeruginosa, Proteus mirabilis, and Candida spp (9).

Antimicrobial resistance is a global problem considerably in developing countries [10,11]. Fluoroquinolone (FQ) is a widely used antimicrobial drug in UTI patients, although various up-to-date articles had identified an alarming increasing resistance toward FQs (12). Fosfomycin is an antibiotic with a broad spectrum of activity against Gram-positive pathogens such as Staphylococcus aureus and Enterococcus and Gram-negative bacteria such as Pseudomonas aeruginosa and Klebsiella pneumonia. Fosfomycin has adequate distribution into tissues, and it is a well-tolerated drug with a low incidence of adverse events. It was discovered for the first time in 1969 in Spain (13). Nitrofurantoin is another helpful drug with potent bactericidal properties against various multidrug-resistant gram-positive and gram-negative uropathogens. It is used to prevent and treat urinary tract infections. As a result of their frequent use, an increased antimicrobial resistance rate was reported recently (14). To the best our konwledge, this is the first study reported from Somalia. The main objective of the study is was to identify the antimicrobial susceptibility profile of Fosfomycin and NF against MDR and XDR uropathogens.

Comment [K7]: Delete the most since ARI are the most common infections

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Comment [K10]: Their ? fosfomycin and NF or QNs

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Materials and Methods:

This retrospective study has reviewed a total of 2485 urine cultures performed in the microbiology unit of Mogadishu Somali Turkish Training and Research Hospital between 2019-2020. This research included 361 uropathogens were isolated from 2485 urine samples that showed bacterial growth. The urine samples were obtained from suspected patients in clean-catch midstream urine specimens and collected, transported, and stored safely in the laboratory unit. Bacterial identification was made by the phenotypic study of the culture, looking for typical characteristics and gram staining, and a series of standard biochemical analyses to recognize the bacteria of interest was also done (15, 16). The antimicrobial susceptibility had been determined by the Kirby-Bauer disk diffusion method following the Clinical and Laboratory Standards Institute's (CLSI) guidelines. Eosin methylene blue agar had used for the identification of uropathogens. Antimicrobial sensitivity and resistance were assessed by Mueller-Hinton agar. The antimicrobials studied against uropathogens were nitrofurantoin (300 mcg), and fosfomycin (200 mcg), ciprofloxacin (5mcg), levofloxacin (5mcg), and Trimethoprim/Sulfamethoxazole (1.25/23.75 mcg). For confirming the findings of AmpC production, E-test strips were used. Uropathogens that were resistant to two or more antibiotic classes were considered multidrug-resistant microorganisms (MDR). The extended-spectrum beta-lactamase production screening test had done according to CLSI recommendations. Analyzed parameters included age, gender, uropathogens obtained from the culture, antibiotic sensitivity, and resistance spectrum. This retrospective study received medical ethical committee approval from the institutional review board of Mogadishu Somalia-Turkey Recep Tayyip Erdogan Training and Research Hospital, Mogadishu, Somalia (Ref. MSTH-4127). Data were collected from medical records, and no potential harm to the patients.

The findings were analyzed in descriptive univariate cross-tabulations using SPSS software for Windows (version 23 SPSS).

Limitations of the study: 1. This a retrospective study using electronic medical records of the patients, 2. The study only focused on the antimicrobial sensitivity and resistance pattern based on culture results, but further prospective studies are needed to evaluate the efficacy and safety of these drugs.

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Comment [K13]: May be given after discussion

Results:

A total of 2485 urine samples were processed for culture and 361 uropathogens were grown. The mean age of the patients was 50 + SD?years. Females constitute 51% of the cases, while males were 49%,. *Escherichia coli* was the most common uropathogen (63.4%), followed by *Klebsiella pneumonia* in 13.3% of the samples. Table 1 shows the distribution of uropathogens identified from urine culture. Eighty-nine percent of the patients had Multidrug-resistant microorganisms. *A*cinetobacter baumannii has the most MDR and XDR patterns in 69.1% of cases, while Escherichia coli and K. pneumonia showed a similar MDR spectrum in 35.2% of the cases.

Fosfomycin exhibited the highest sensitivity rate against MDR and XDR uropathogens in 94.8% in community-acquired and nosocomial UTIs. In our study, patients classified into two groups; inpatients accounted for 44.3% of the cases, and outpatients were 65.7% of the patients. Ninety-six percent of fosfomycin was sensitive against nosocomial uropathogens. A slight evolving resistance against fosfomycin was seen in community-acquired UTI about 5.7% despite their higher sensitivity rate (Figure 1).

Klebsiella pneumonia and E.coli revealed the highest resistance rate against fosfomycin in 3.7% and 3.4% of the cases sequentially. Pseudomonas Aeruginosa and Acinetobacter Baumannii were the most multidrug-resistant and extensively drug resistant uropathogens, but fortunately, they produced a zero resistance rate against fosfomycin (Table 2). One-quarter of extended-spectrum beta-lactamases producing pathogens had shown resistance toward fosfomycin. Nitrofurantoin showed a higher sensitivity rate against extended-spectrum beta-lactamases producing uropathogens. Nitrofurantoin had an 86.1% sensitivity rate against uropathogens, and the drug showed a nearly similar sensitivity rate in patients with the community and hospital-acquired UTIs. As cited above, Klebsiella pneumonia produced the most resistance rate against nitrofurantoin in 27.6% that was three times more than E.coli that developed a resistance rate against nitrofurantoin in 9.4% of the cases. More than two-thirds of resistance against nitrofurantoin is produced by ESBL pathogens.

Fluoroquinolones with relation to fosfomycin and nitrofurantoin showed a lesser sensitivity rate toward uropathogens in both gender groups, uncomplicated and complicated UTIs (Figure 2). Ciprofloxacin had a 67.7% resistance rate in total cases. Acitinobacter Buaminni (100%), E.coli (68%), Klebsiella Pneumonia (60.6%), and Pseudomonas Aeruginosa in 60% resistance pattern

against Ciprofloxacin had gained through the study. Levofloxacin showed a resistance level near that of Ciprofloxacin but slight inferiority regarding the total resistance rate of about 54.2%of the cases. Moreover, levofloxacin was better in community and hospital-acquired UTIs and cases of ESBL producing pathogens. Trimethoprim/sulfamethoxazole showed the highest resistance rate against uropathogens in 86.6% of the cases. Fosfomycin and nitrofurantoin with relation to fluoroquinolones and trimethoprim/sulfamethoxazole showed a higher sensitivity rate against uropathogens, including multidrug-resistant and extensively drug-resistant uropathogens in both gender groups with community-acquired and nosocomial UTIs ($P<0.001$).

Out of 361 urine cultures that showed growth, 37 cases produced extended-spectrum beta-lactamases producing pathogens that make a prevalence of 10.2%. E.coli was the leading ESBL uropathogens in about 70.3%, followed by Klebsiella pneumonia in 27% and Enterobacter cloacae in 2.7%. Patients with hospital-acquired urinary tract infections were more susceptible to develop multidrug-resistant and ESBL producing uropathogens.

Discussion

The main challenges confronting the treatment of urinary tract infections are disseminating multidrug-resistant gram-negative uropathogens and the lack of new antimicrobials against such pathogens (5). The main objective of the study is to identify the antimicrobial susceptibility profile against MDR and XDR uropathogens.

The prevalence of MDR uropathogens in our study was 89% of the cases that is three times higher than previously reported studies (17). Overprescription and improper use of antibiotics are the leading factors that contribute to the widespread antimicrobial resistance. In our study, the urine cultures that showed bacterial growth were 361 (14.5%) cases out of 2485 patients who enrolled in our study that corresponds to other previous studies (18). According to the gender distribution of pathogens, females were more vulnerable to UTI (57.2%) compared to males (42.8%), which matches in the previously reported studies (18).

Fosfomycin had a potential antimicrobial activity against ESBL producing uropathogens in both inpatient and outpatient settings which corresponds to the previous studies (19,20). Fosfomycin showed significant efficacy against uropathogens (ESBL, Enterobacter, Enterococcus, gram-positive and gram-negative bacteria MDR, and XDR) and gained a sensitivity rate of 94.8% of the cases but expressed an evolving resistance in community-acquired UTIs about 5.7% (21).

Klebsiella pneumonia and E.coli revealed the highest resistance rate against fosfomycin in 3.7% and 3.4% of the cases sequentially that oppose a study reported by Annika I. Nilsson and colleagues(22).

Nitrofurantoin showed a satisfactory sensitivity rate against ESBL uropathogens in all age groups. 34.7% resistance rate toward Nitrofurantoin against ESBL expressed in our study that matches research reported by J. Garau from Canada and the USA (23). The authors reported a 9.4% nitrofurantoin resistance rate against E.coli that opposes Kashanian J and associates study from the USA that reported a 2.3% resistance rate (24).

In the present study, E.coli was the leading cause of ESBL producing uropathogens in 70.3% of the total cases, followed by Klebsiella pneumonia in 27%. ESBLs-producing E. coli are the most common cause of increased morbidity in patients with UTI. ESBL-producing organisms are known to show significant resistance implications to antimicrobial drugs such as Fluoroquinolones. Regarding the antibiotic susceptibility pattern, ESBL-producing E. coli showed higher resistance against Ciprofloxacin (68%) and Levofloxacin (62.54%). This higher resistance is in contrast to the other previous studies and could be the overuse of fluoroquinolones in the community and hospital-acquired UTIs before the initial empirical therapy (25,26).

Conclusion:

Fosfomycin and nitrofurantoin revealed a higher sensitivity rate against gram-negative MDR uropathogens in community and nosocomial UTIs compared to fluoroquinolones and trimethoprim/sulfamethoxazole. Pseudomonas Aeruginosa and Acinetobacter Baumannii were the most multidrug-resistant and extensively drug-resistant uropathogens. Fortunately, they exhibited a zero resistance rate toward fosfomycin and nitrofurantoin.

Informed Consent Statement: All patients obtained informed consent.

Data Availability Statement: Data included in the manuscript.

Abbreviations

CDC: Centers for Disease Control and Prevention, CLSI: Clinical and Laboratory Standards Institute's, EMB: eosin methylene blue agar, *ESBL:* Extended-Spectrum Beta-Lactamases, FQ: Fluoroquinolone, MDR: multidrug-resistant, UTI: Urinary tract infection, XDR: extensive drug-resistant.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors

Reference:

1. Manhal F. Urinary tract infection in hemodialysis patients with renal failure. Journal of the Faculty of Medicine. 2012;54(1):38-41.
2. Momtaz H, Karimian A, Madani M, Safarpoor Dehkordi F, Ranjbar R, Sarshar M, et al. Uropathogenic Escherichia coli in Iran: serogroup distributions, virulence factors and antimicrobial resistance properties. *Ann Clin Microbiol Antimicrob*. 2013;**12**:8. doi: 10.1186/1476-0711-12-8. [PubMed: 23627669]. [PubMed Central: PMC3651382].
3. Stamm WE, Norrby SR. Urinary tract infections: disease panorama and challenges. The Journal of infectious diseases. 2001 Mar 1;183(Supplement_1):S1-4.
4. Foxman B. The epidemiology of urinary tract infection. Nature Reviews Urology. 2010 Dec;7(12):653.
5. Falagas ME, Vouloumanou EK, Samonis G, Vardakas KZ. Fosfomycin. Clin Microbiol Rev. 2016 Apr;29(2):321-47. doi: 10.1128/CMR.00068-15. PMID: 26960938; PMCID: PMC4786888.
6. Baquero-Artigao F, Del Rosal Rabes T. Fosfomycin in the pediatric setting: Evidence and potential indications. Rev Esp Quimioter. 2019 May;32 Suppl 1(Suppl 1):55-61. PMID: 31131593; PMCID: PMC6555161.
7. Li B, Sun JY, Liu QZ, Han LZ, Huang XH, Ni YX. High prevalence of CTX-M β-lactamases in faecal Escherichia coli strains from healthy humans in Fuzhou, China. Scandinavian journal of infectious diseases. 2011 Mar 1;43(3):170-4.
8. Ronald A. The etiology of urinary tract infection: traditional and emerging pathogens. The American journal of medicine. 2002 Jul 8;113(1):14-9.
9. Tolkoff-Rubin NE, Rubin RH. Urinary tract infection: significance and management. Bulletin of the New York Academy of medicine. 1986 Mar;62(2):131.
10. Mathai D, Lewis MT, Kugler KC, Pfaller MA, Jones RN, Clarion Health Methodist Hospital I, Staley C, System SH, Center GS, Hospital DG, Hospital A. Antibacterial activity of 41 antimicrobials tested against over 2773 bacterial isolates from hospitalized patients with pneumonia: I—results from the SENTRY Antimicrobial Surveillance

Program (North America, 1998). Diagnostic microbiology and infectious disease. 2001 Feb 1;39(2):105-16.

11. Okeke IN, Fayinka ST, Lamikanra A. Antibiotic resistance in Escherichia coli from Nigerian students, 1986 1998. Emerging Infectious Diseases. 2000 Jul;6(4):393.
12. Mohamed AH, Mohamud MF, Mohamud HA. Epidemiology and Antimicrobial Susceptibility Pattern of Uropathogens in Patients with the Community-and Hospital-Acquired Urinary Tract Infections at a Tertiary Hospital in Somalia. Jundishapur Journal of Microbiology. 2020 Sep 30;13(9).
13. Michalopoulos AS, Livaditis IG, Gougoutas V. The revival of fosfomycin. International journal of infectious diseases. 2011 Nov 1;15(11):e732-9.
14. Cunha BA. Nitrofurantoin: an update. Obstetrical & gynecological survey. 1989 May;44(5):399-406.
15. Baron EJ, Peterson LR, Finegold SM. Bailey and Scott's diagnostic microbiology. St. Louis: Mosby; 1994 Jan.
16. Lu CL, Liu CY, Huang YT, Liao CH, Teng LJ, Turnidge JD, Hsueh PR. Antimicrobial susceptibilities of commonly encountered bacterial isolates to fosfomycin determined by agar dilution and disk diffusion methods. Antimicrobial agents and chemotherapy. 2011 Sep 1;55(9):4295-301.
17. Giancola SE, Mahoney MV, Hogan MD, Raux BR, McCoy C, Hirsch EB. Assessment of Fosfomycin for Complicated or Multidrug-Resistant Urinary Tract Infections: Patient Characteristics and Outcomes. *Chemotherapy*. 2017;**62**(2):100-4. doi: 10.1159/000449422. [PubMed: 27788499].
18. Abejew AA, Denboba AA, Mekonnen AG. Prevalence and antibiotic resistance pattern of urinary tract bacterial infections in Dessie area, North-East Ethiopia. BMC research notes. 2014 Dec 1;7(1):687.
19. Prakash V, Lewis JS, Herrera ML, Wickes BL, Jorgensen JH. Oral and parenteral therapeutic options for outpatient urinary infections caused by Enterobacteriaceae producing CTX-M extended-spectrum β-lactamases. Antimicrobial agents and chemotherapy. 2009 Mar 1;53(3):1278-80.
20. Falagas ME, Kastoris AC, Kapaskelis AM, Karageorgopoulos DE. Fosfomycin for the treatment of multidrug-resistant, including extended-spectrum β-lactamase producing,

Enterobacteriaceae infections: a systematic review. The Lancet infectious diseases. 2010 Jan 1;10(1):43-50.

21. Falagas ME, Vouloumanou EK, Samonis G, Vardakas KZ. Fosfomycin. Clinical Microbiology Reviews. 2016 Apr 1;29(2):321-47.
22. Nilsson AI, Berg OG, Aspevall O, Kahlmeter G, Andersson DI. Biological costs and mechanisms of fosfomycin resistance in Escherichia coli. Antimicrobial agents and chemotherapy. 2003 Sep 1;47(9):2850-8.
23. Garau J. Other antimicrobials of interest in the era of extended-spectrum β-lactamases: fosfomycin, nitrofurantoin and tigecycline. Clinical Microbiology and Infection. 2008 Jan;14:198-202.
24. Kashanian J, Hakimian P, Blute Jr M, Wong J, Khanna H, Wise G, Shabsigh R. Nitrofurantoin: the return of an old friend in the wake of growing resistance. BJU international. 2008 Dec;102(11):1634-7.
25. Yasmeen BN, Islam S, Islam S, Uddin MM, Jahan R. Prevalence of urinary tract infection, its causative agents and antibiotic sensitivity pattern: A study in Northern International Medical College Hospital, Dhaka. Northern International Medical College Journal. 2015 Nov 16;7(1):105-9.
26. Oluremi BB, Idowu AO, Olaniyi JF. Antibiotic susceptibility of common bacterial pathogens in urinary tract infections in a Teaching hospital in South-western Nigeria. African journal of microbiology research. 2011 Oct 16;5(22):3658-63.

Table 1: Uropathogens

Pathogen	**No. of patients**	**Percentage %**
E.coli ESBL	229 26	63.4%
Klebsiella Pneumonia ESBL	48 10	13.3%
Pseudomonas Aeruginosa	14	3.9%
Staph Aureus	13	3.6%
Acinetobacter Baumannii	7	1.9%
Enterobacter cloacae ESBL	5 1	1.4%
Enterococcus faecium	5	1.3%
Streptococcus species	2	0.6%
Citrobacter freundii	2	0.6%
Staphylococcus haemolyticus	1	0.3%
Cedecea lapagei	1	0.3%
Candida	34	9.4%
Total	361	100.0%

Table 2: Antimicrobial resistance pattern against uropathogen including extended-spectrum beta-lactamase producing uropathogens

Medications	**Resistance Rate %**	**Resistant Level against individual pathogens**				
		E.coli	Klebsiella Pneumonia	Pseudomonas Aeruginosa	Acinetobacter Baumannii	ESBL
Ciprofloxacin	67.7%	68%	60.6%	60%	100%	84.2%
Levofloxacin	54.2%	62.5%	25%	50%	100%	72.4%
Fosfomycin	5.2%	3.4%	3.7%	0%	0%	25%
Nitrofurantoin	13.9%	9.4%	27.6%	0%	0%	34.7%
Trimethoprim/Sulfamethoxazole	86.6%	84.2%	84.7%	92.8%	100%	83%

Figure 1: Pattern of antimicrobial resistance in respect to outpatient and inpatient units

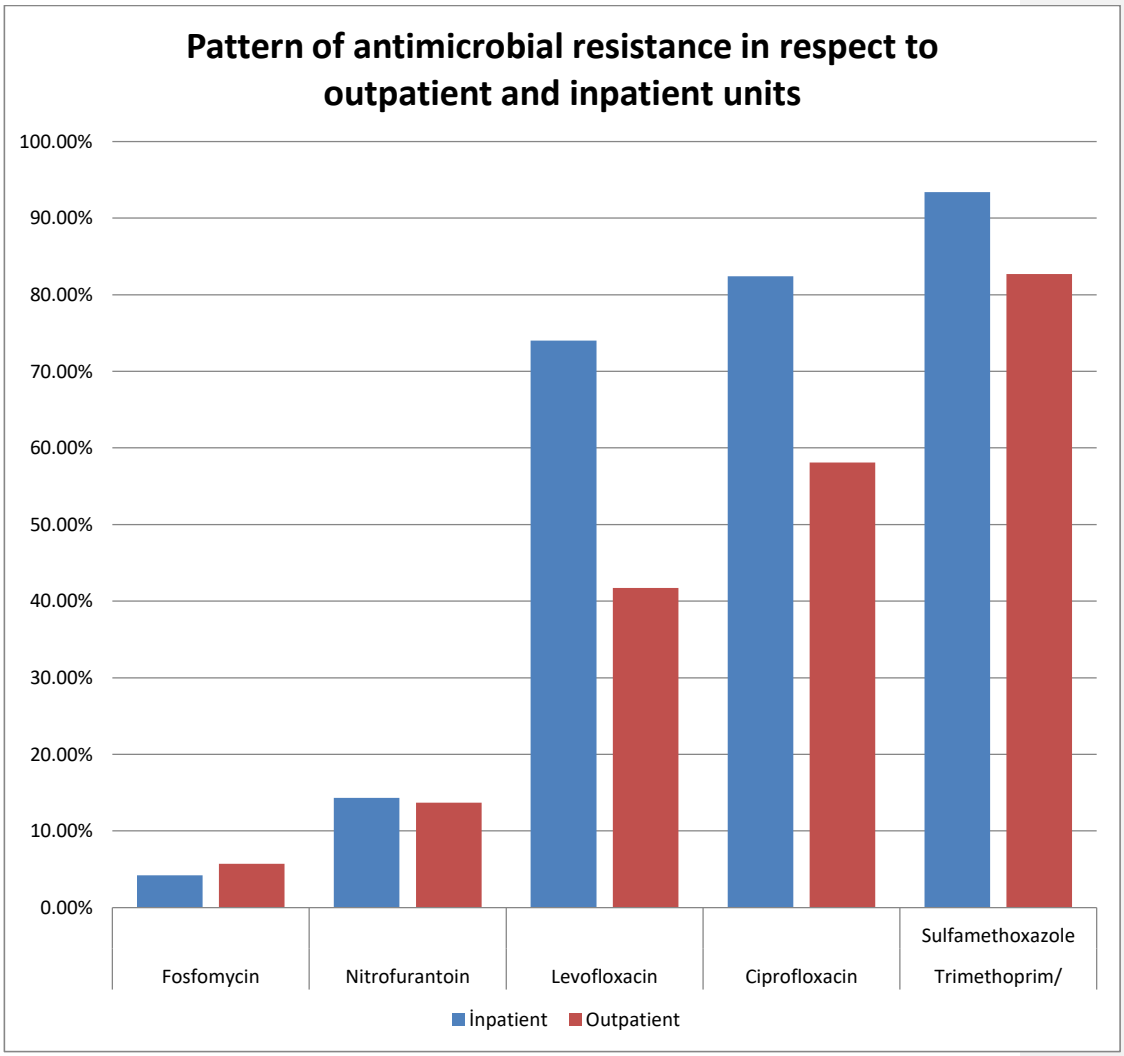

Figure 2: Antimicrobial resistance according to the gender distribution