

Sensitivity of extended spectrum of β -lactamase producing *Escherichia coli* and *Klebsiella species* to Fosfomycin

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Abstract

Objective: To investigate extended spectrum β -lactamase production and Fosfomycin resistance rates of *Escherichia coli* and *Klebsiella species* isolates from patients' urine culture.

Methods: Chromogenic agar was used to identify *Escherichia coli* and *Klebsiella species* strains. All antibiogram processing was carried out on a fully automated VITEK 2 identification and antibiogram system. The results obtained between January 2015 and December 2018 were retrospectively screened.

Results: *Escherichia coli* and *Klebsiella species* were isolated from total 2868 urine cultures. Thus, 844 (34.9%) of 2418 *Escherichia coli* and 305 (67.8%) of 450 *Klebsiella species* produced ESBL. Sensitivity rate of ESBL producing *Escherichia coli* to Fosfomycin was 96.5%, and 98.8% for ESBL negative *Escherichia coli*. Sensitivity rate of ESBL producing *Klebsiella pneumoniae* to Fosfomycin was 70.5%, and 53.1% for ESBL negative *Klebsiella pneumoniae*. None of the three ESBL producing *Klebsiella oxytoca* strains were found to be resistant to Fosfomycin, though five of 15 ESBL negative strains were found to be resistant to Fosfomycin.

Conclusion: Taking into consideration the advantages of low resistant rate, it is concluded that Fosfomycin may be a good alternative for first step empirical treatment in urinary tract infections, especially in *Escherichia coli* positive cases. However, the rate of resistance identified in *Klebsiella* strains informs the onset of resistance problems in Fosfomycin.

Keywords: Fosfomycin, Extended spectrum β -lactamase, *Escherichia coli*, *Klebsiella*, Nosocomial infection.
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Introduction

Urinary tract infections are the most common community and nosocomial (hospital acquired) infections requiring antibiotic therapy.¹ Gram-negative bacilli of Enterobacteriaceae family in particular, *Escherichia coli* and *Klebsiella pneumoniae* are the most commonly reported agents causing this infection.¹ Frequent failure to treatment is often attributed to inappropriate use of antibiotics.² Extended spectrum β -lactamase (ESBL) producing bacteria isolated from UTI are resistant to many antibiotics. For this reason, different antibiotic options are sometimes required for successful treatment.

The increasing frequency of ESBL producing *E. coli* and *K. pneumoniae* strains is a major problem in terms of community health. These strains are usually resistant to many other antibiotics, that reduce the chance of successful treatment thus/ therefore. ESBL producing resistant bacterial infections have higher morbidity and mortality.^{1,2}

Most frequently detected mechanism of antibiotic

resistance detected in *K. pneumoniae* and *E. coli* operates by the enzymatic action of β -lactamase which cleaves the amide bond of β -lactam ring of the antibiotic.² Genes encoded this enzyme are spread by transferring from bacterium to bacterium via plasmid and transposon. ESBL enzymes, the largest β -lactamases in the spectrum, can also disrupt third-generation cephalosporins and aztreonam. Because of the spread of ESBL enzyme genes among species through plasmids, these bacteria produce outbreaks and cause difficulties in treatment. Although, ESBL producing strains are resistant to penicillins, cephalosporins, and aztreonam, they may become susceptible during routine antibiogram process. However, even if they are sensitive in vitro, infections of these strains may not respond to treatment with these antibiotics. For this reason, these antibiotics are not effective in the treatment of patients and cause time loss. In accordance with the recommendation of the Clinical and Laboratory Standards Institute (CLSI), ESBL producing microorganisms must be detected in microbiology laboratories. However, determining of the prevalence of ESBL producing *E. coli* and *Klebsiella* strains will guide the selection of antibiotics in the treatment of infections associated with these strains.³

Nowadays, the increase of resistance to antibiotics such as

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β -lactams and β -lactamase inhibitor combinations, quinolones, trimethoprim-sulfamethoxazole and β -lactam antibiotics commonly that were used in the empirical treatment of community-acquired UTI associated with *E. coli* and *Klebsiella species* was led to the search for alternative drugs in treatment.⁴ Fosfomycin trometamol is one of these alternative drugs. Fosfomycin has a bactericidal activity that effects by disrupting bacterial cell wall synthesis. Fosfomycin acts by inhibiting pyruvyl transferase, which plays a part in bacterial cell wall synthesis. Apart from this effect, Fosfomycin disrupts fimbriae synthesis of bacteria and reduces their motility. This antibiotic inhibits the adherence of bacteria to the epithelium of urinary system and to the inner surface of the urinary catheters. At the same time, Fosfomycin affect the bacteria in biofilm layer. Fosfomycin is one of the rare antibiotics with a very low incidence of resistance. In addition to its pharmacokinetic and pharmacodynamic advantages, it has important advantages such as being in vivo activity, clinical efficacy, highly tolerability and reliability.⁵

The purpose of this study is to determine Fosfomycin resistance rates and ESBL rates of *E. coli* and *Klebsiella* spp. that were isolated from urine cultures of inpatients and outpatients that were treated at various outpatient clinics and clinics of our hospital.

Materials and Methods

The ESBL production and Fosfomycin resistance rates of *E. coli* and *Klebsiella* strains that were isolated from urine cultures of patients admitted to various polyclinics and clinics of Afyonkarahisar Health Sciences University Hospital in Turkey between January 2015 and December 2018 have been examined in this study. Mid stream urine specimens were quantitatively cultured on Chromogenic agar media. The media were left to incubate at 37°C for 18-24 hours. Bacterial cultures which have been a uniform bacterial colony and which have been bacterial colony count of 100.000 CFU/mL were considered significant.

Chromogenic agar was used in identification of *Klebsiella* spp. and *E. coli* strains. In addition to this, fully automated VITEK 2 identification and antibiogram system (bioMérieux, Inc. Hazelwood, MO, USA) was used for identification of *Klebsiella* spp. and *E. coli* strains, and for all antibiogram procedures.⁶ The urine culture results obtained for the last three years for *E. coli* and *Klebsiella* species isolated from urine cultures were evaluated retrospectively. It was confirmed all bacteria by biochemical tests, conventionally.

In order to check the accuracy of the study, external (Oneworld Accuracy Company, Turkey) and internal (*E.*

coli ATCC 25922, *E. faecalis* ATCC 29212, and *S. aureus* ATCC 29213 strains were used) quality control studies are carried out regularly for VITEK 2 system. (Ethical approval for this retrospective research was obtained from the local ethics committee of Afyon Kocatepe University).

Statistical Analysis: The relationship between variables was examined by Chi-square statistic test. SPSS Version 18.0 for Windows Software was used to analyze the data ($P < 0.05$). For the sample size estimation, the formula $n = P \cdot Q \cdot Z_{\alpha}^2 / d^2$ was used. The P and Q ratios [0.5 (50%)] were determined as $Z_{0.01} = 2.58$ for the significance level of 0.01, and effect size $d = 0.025$ (2.5%). The minimum sample calculated as 2663. According to this study, the total sample size consisted of 2868 bacteria and sufficed. (n =sample size, Z_{α} =level of statistical significance/ d =effect size/ P = prediction of occurrence rate/ Q = prediction of non-occurrence rate).

Significance level was determined as $P < 0.05$ for statistical analyses.

Results

In this study, culture and antibiotic sensitivity test results of total 2868 bacteria were examined. Chromogenic agar and VITEK 2 were used in identification and all antibiogram processing of strains. Chromogenic agar cultures of a ESBL positive *E. coli* and *K. pneumoniae* strains were showed in Figure-1. *E. coli*, *K. pneumoniae* and *K. oxytoca* were detected in 2418, 432 and 18 of the total 2868 urine cultures, respectively. All the bacteria were examined according to the distribution of ESBL (Table-1). ESBL was found positive in 844 (34.9%) *E. coli*, in 302 (69.9%) *K. pneumoniae* and in 3 (16.7%) *K. oxytoca* strains. Relationship between bacteria species and ESBL

Table-1: ESBL production rates of *E. coli* and *Klebsiella* spp. isolated from UTI.

Strains	ESBL(+)	ESBL(-)	P value
<i>E. coli</i>	844 (34.9%)	1574 (65.1%)	0.001*
<i>Klebsiella</i> spp.	305 (67.8%)	145 (32.2%)	

* $P < 0.01$.

Table-2: The resistance and sensitivity of bacteria to Fosfomycin.

Strains	Fosfomycin		P value
	Sensitivity	Resistant	
<i>E. coli</i>	2369 (98.0%)	49 (2.0%)	0.001*
<i>K. pneumoniae</i>	282 (65.3%)	150 (34.7%)	
<i>K. oxytoca</i>	13 (72.2%)	5 (27.8%)	
Total	2664 (92.9%)	204 (7.1%)	

* $P < 0.01$.



Figure-1: Chromogenic agar cultures of ESBL positive *E. coli* and *K. pneumonia*.

Table-3: The resistance and sensitivity of ESBL (+) and ESBL (-) bacteria to Fosfomycin.

ESBL(+) Strains	Fosfomycin		P value
	Sensitivity	Resistant	
<i>E. coli</i>	814 (96.5%)	30 (3.5%)	0.001*
<i>K. pneumoniae</i>	213 (70.5%)	89 (29.5%)	
<i>K. oxytoca</i> **	3 (100.0%)	Nil	
Total	1030 (89.6%)	119 (10.4%)	
ESBL(-) Strains	Sensitivity	Resistant	0.001*
<i>E. coli</i>	1555 (98.8%)	19 (1.2%)	
<i>K. pneumoniae</i>	69 (53.1%)	61 (46.9%)	
<i>K. oxytoca</i>	10 (66.7%)	5 (33.3%)	
Total	1634 (95.1%)	85 (4.9%)	

*P<0.01, ***K. oxytoca* was not included in the chi-square statistical test.

production was found statistically significant (P<0.01).

All the bacteria were examined according to the distribution of Fosfomycin sensitivity (Table-2). Thus, 2369 (98.0%) of the 2418 *E. coli* strains were found to be sensitive to Fosfomycin and 49 (2.0%) were resistant. However, 150 (34.7%) of the 432 *K. pneumoniae* and five (27.8%) of 18 *K. oxytoca* strains were found to be resistant to Fosfomycin. Relationship between bacteria species and Fosfomycin resistance was found statistically significant (P<0.01).

Similarly, the whole of bacteria were examined according to the relationship between ESBL production and Fosfomycin resistance (Table-3). The sensitivity rate of ESBL producing *E. coli* strains to Fosfomycin was found to be 96.5%, but the sensitivity rate of ESBL negative *E. coli* to Fosfomycin was found to be 98.8%. The sensitivity rate

of ESBL producing *K. pneumoniae* strains to Fosfomycin was found to be 70.5%, but the sensitivity rate of ESBL negative *K. pneumoniae* to Fosfomycin was found to be 53.1%. The none of the 3 ESBL producing *K. oxytoca* strains were found to be resistant to Fosfomycin, though five of 15 ESBL negative strains were to be resistant to Fosfomycin. Relationship between ESBL production and Fosfomycin resistance was found statistically significant (P<0.01).

Discussion

The last decade an increased number of drug resistant *E. coli* and Klebsiella spp. cases have been reported from Eastern Mediterranean, Asia and African countries. This indeed causing a serious problem in treating the infected population.^{3,7} In accordance with the CLSI recommendations, presence of ESBL should be routinely examined in *E. coli* and Klebsiella strains isolated in laboratories.⁷

According to the results of our study, 844 of 2418 *E. coli* strains and 305 of 450 Klebsiella spp. strains produced ESBL. Thus, ESBL producing *E. coli* and ESBL producing *K. pneumoniae* rates were respectively determined as 34.90% and 67.8%, between 2015-2018. In a meta-analysis conducted in Turkey, it was reported that ESBL positivity for *E. coli* was determined as 8.1% between 1996-2001, 10.6% between 2002-2007 and 28.2% between 2008-2012.⁸ According to our study, ESBL production rate of *E. coli* was found to be higher than these earlier reports indicating an emerging threat of drug resistant. In other study, ESBL production rate for *K. pneumoniae* was determined as 55%.⁹ In a study in 2012, ESBL producing in *K. pneumoniae* strains was

reported as 67%.¹⁰ Results of this study is also consistent with the results of Terzi et al, for *K. pneumoniae*. According to 2007 data of MYSTIC (Meropenem Yearly Susceptibility Test Information Collection) which is a multicenter study in Turkey, 40.5% of *K. pneumoniae* strains were found to be ESBL positive.¹¹ A follow up study in Turkey, spread between 2006 to 2010 revealed that ESBL producing rates of *E. coli* steadily increased from 4.6 to 10.4%, while *Klebsiella* climbed from 25.9 to 37.2% for the same period.¹² A slight variation to this order is reported from South America where ESBL producing strains of *K. pneumoniae* dominated (45%) to *E. coli* strains (8.5%).¹³ In Asia, this rate varies from country to country, ranging from 5% in Korea to 23.3% in Indonesia for *E. coli*. ESBL positivity in *Klebsiella* spp. was found to be 48.8% in Korea and 20-40% in Southeast Asia, China and Japan.¹³ The present study, found that the rates of ESBL-production of *Klebsiella* spp. were comparatively higher than that of other countries (Table-1).

Resistance to other commonly used antibiotics in the empirical treatment of UTI is increasing. Therefore, new alternative drugs are needed for effective treatment. Fosfomycin is one of the best options in terms of sensitivity. Fosfomycin effects most of different bacteria isolated from UTI resulting from *E. coli* and *Klebsiella* strains, especially.^{4,5,14} Fosfomycin provides several advantages in treatment. For example, it has low toxic effect and low side effect. In addition, it is present at a higher concentration in urine and does not make cross resistance to different antibiotics.¹⁵ Furthermore, Fosfomycin is found to have a broad spectrum bactericidal activity.¹⁶ Knowledge of national resistance data is crucial in determining empirical treatment and avoiding unnecessary antibiotic use. According to the results obtained in this study, the rate of resistance to Fosfomycin (2%) was found to be quite low for *E. coli* strains. Fosfomycin can easily be preferred in the treatment of infections associated with *E. coli*. According to this study, it was found that *K. pneumoniae* has the highest resistance (34.7%) to Fosfomycin (Table-2).

One report from Turkey, found Fosfomycin resistance to be 0.4% in ESBL producing *E. coli* strains. ESBL positivity in *E. coli* strains was determined to be 19.5%. In our study, these rates are quite high (Tables-1,3). In the study conducted by Hosbul and colleagues, Fosfomycin resistance was not found in ESBL negative isolates, whereas in our study Fosfomycin resistance was determined as 1.2% in ESBL negative *E. coli*

isolates.¹⁷

Other studies from Turkey, reported low resistance rates of Fosfomycin.³ In this study, the Fosfomycin resistance of *K. pneumoniae* was found to be quite high (34.7%).

Similar studies conducted in Korea, Japan and China, the rate of Fosfomycin resistance in ESBL-producing *E. coli* strains was 7.1%, 1%, and 4.3%, respectively.¹⁸ In a study by Fagalas et al in Greece, a total of 152 MDR Enterobacteriaceae isolates were studied. Of the examined 152 isolates, 85 (55.9%) were extensively drug-resistant (XDR), of which 78 (91.8%) remained susceptible to Fosfomycin. Sensitivity to Fosfomycin of the 34 extended-spectrum-lactamase-producing isolates was determined to be 94.1%. Thus, 105 (90.5%) of 116 *K. pneumoniae* strains were determined to be susceptible to Fosfomycin, and 26 (100%) of the *E. coli* strains were determined to be susceptible to Fosfomycin. In other words, eight (9.6%) of 116 *K. pneumoniae* strains were determined to be resistant to Fosfomycin.¹⁶ According to the results in the Netherlands, sensitivity rates to Fosfomycin were determined to be 95.9% for *E. coli* and 87.6% for *K. pneumoniae*.¹⁹ In a study in Taiwan, sensitivity rates of Fosfomycin were 94% for ESBL producing *E. coli*.²⁰ According to a study in India, 99.6% of 384 ESBL producing *E. coli* strains were found susceptible to Fosfomycin, while 87.7% of 80 ESBL producing *K. pneumoniae* strains were found to be susceptible to Fosfomycin.²¹

A study in Thailand, 359 ESBL producing *K. pneumoniae* and 394 ESBL producing *E. coli* strains were susceptible to Fosfomycin at rates of 88.4% and 97.3%, respectively.²²

According to a study by Cueto et al, in Spain, Fosfomycin has exhibited an excellent activity against ESBL producing 290 *E. coli* and 138 *K. pneumoniae* isolates.²³

When the results of study conducted in the Netherlands, Taiwan, India and Thailand were compared with the results of our study, it was found which the susceptibility rates of Fosfomycin for *E. coli* were similar. However, it was reported the sensitivity rates of Fosfomycin for *K. pneumoniae* were higher than those of our studies. In addition, the results of the study conducted in Spain compared to our study has high sensitivity rates.

In a previous study in Northern Taiwan, sensitivity rate to Fosfomycin was determined to be 56.7% for 66 ESBL producing *K. pneumoniae* strains isolated from patients with UTI.²⁴ Our study results were generally found to be compatible with the results of many studies conducted

in different countries with some exceptions. In Czech Republic, Fajfr et al have demonstrated that there were no significant differences in Fosfomycin susceptibilities among the *E. coli* and *K. pneumoniae* isolates regardless of their β -lactamase-producing status. They have profounded to be indicating that lactamase production is not a mechanism for Fosfomycin resistance.²⁵ However, according to the results of our study, the relationship between ESBL production and Fosfomycin resistance was found statistically significant. Our data exhibited that the Fosfomycin resistance rates of ESBL producing strains were higher than all ESBL non-producing strains except *K. oxytoca*.

Conclusion

Properties of antibiotic resistance of bacteria isolated from UTI may vary regionally. Therefore, the results of this study and other similar studies will help in determining of the regional distribution of the antibiotic resistance profiles of ESBL producing *E. coli* and *Klebsiella* spp., thereby assist in the treatment success and in the selection of antibiotics to be used in treatment. Based on the results of this study, Fosfomycin displayed very high activity against all ESBL producing strains. For this reason, it has been concluded that Fosfomycin, use could be of significant advantage, and is a good option for first-line empirical treatment of UTI caused by community-acquired *E. coli*. The increase in Fosfomycin resistance was observed in direct proportion to the presence of ESBL in general. Therefore, this and similar studies indicated that the problem of resistance in Fosfomycin treatment has started and, this problem may increase with higher resistance rates observed in *Klebsiella* strains. In line of these information, it would be useful to demonstrate the clinical success of Fosfomycin treatment in vivo and in vitro experiments by carrying out studies by geographic region, periodically.

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References

- Kang CI, Kim J, Park DW, Kim BN, Ha US, Lee SJ, et al. Clinical Practice Guidelines for the Antibiotic Treatment of Community-Acquired Urinary Tract Infections. *Infect Chemother* 2018;50:67-100. doi: 10.3947/ic.2018.50.1.67.
- Elsayed TI, Ismail HAF, Elgamel SA, Gad AHA. The occurrence of multidrug resistant *E. coli* which produce ESBL and cause urinary tract infections. *J Appl Microbiol Biochem* 2017;1:1-8. DOI: 10.21767/2576-1412.100008
- Cagan Aktas S, Gençer S, Batirel A, Haciseyitoglu D, Ozer S. Fosfomycin susceptibility of urinary *Escherichia coli* isolates producing extended-spectrum beta-lactamase according to CLSI and EUCAST recommendations. *Mikrobiyol Bul* 2014;48:545-55.
- Peralta G, Lamelo M, Alvarez-García P, Velasco M, Delgado A, Horcajada JP, et al. Impact of empirical treatment in extended-spectrum beta-lactamase-producing *Escherichia coli* and *Klebsiella* spp. bacteremia. A multicentric cohort study. *BMC Infect Dis* 2012;12:e245. doi: 10.1186/1471-2334-12-245.
- Yeganeh-Sefidan F, Ghotaslou R, Akhi MT, Sadeghi MR, Mohammadzadeh-Asl Y, Bannazadeh Baghi H. Fosfomycin, interesting alternative drug for treatment of urinary tract infections created by multiple drug resistant and extended spectrum β -lactamase producing strains. *Iran J Microbiol* 2016;8:125-31.
- Banerjee S, Sengupta M, Sarker TK. Fosfomycin susceptibility among multidrug-resistant, extended-spectrum beta-lactamase-producing, carbapenem-resistant uropathogens. *Indian J Urol* 2017;33:149-54. doi: 10.4103/iju.UU_285_16.
- Obeng-Nkrumah N, Twum-Danso K, Krogfelt KA, Newman MJ. High levels of extended-spectrum beta-lactamases in a major teaching hospital in Ghana: the need for regular monitoring and evaluation of antibiotic resistance. *Am J Trop Med Hyg* 2013;89:960-4. doi: 10.4269/ajtmh.12-0642.
- Aykan SB, Ciftci IH. Antibiotic resistance patterns of *Escherichia coli* strains isolated from urine cultures in Turkey: a meta-analysis. *Mikrobiyol Bul* 2013;47:603-18. doi: 10.5578/mb.6383
- Kahraman EP, Karakeçe E, Erdogan F, Uluyurt H, Köroğlu M, Çiftçi IH. The evaluation of antibiotic resistance status of *klebsiella pneumoniae*. *Ortadogu Tip Dergisi*. 2017;9:12-8.
- Terzi HA, Karakeçe E, Çiftçi IH. The evaluation of antibiotic susceptibilities of *klebsiella* spp. isolates. *ACU Saglik Bil* 2013;4:68-71.
- Eraksoy H, Basustaoglu A, Korten V, Kurt H, Ozturk R, Ulusoy S, et al. Susceptibility of bacterial isolates from Turkey--a report from the Meropenem Yearly Susceptibility Test Information Collection (MYSTIC) Program. *J Chemother* 2007;19:650-7. doi: 10.1179/joc.2007.19.6.650
- Parlak M, Çikman A, Bektas A, Bektas M. Extended spectrum beta-lactamase production and antibiotic resistance in *Escherichia coli* and *Klebsiella pneumoniae* strains: five-year follow up results. *Sakarya M J* 2012;2:11-5. doi: 10.5505/sakaryamj.2012.57441
- Choi T, Yoo KH, Lee SJ. Changing epidemiology of extended spectrum beta-lactamases pathogen of urinary tract. *Urogenit Tract Infect* 2015;10:74-83. doi: 10.14777/uti.2015.10.2.74
- Shakya P, Shrestha D, Maharjan E, Sharma VK, Paudyal R. ESBL Production Among *E. coli* and *Klebsiella* spp. Causing Urinary Tract Infection: A Hospital Based Study. *Open Microbiol J* 2017;11:23-30. doi: 10.2174/1874285801711010023.
- Sardar A, Basireddy SR, Navaz A, Singh M, Kabra V. Comparative Evaluation of Fosfomycin Activity with other Antimicrobial Agents against *E.coli* Isolates from Urinary Tract Infections. *J Clin Diagn Res* 2017;11:26-9. doi: 10.7860/JCDR/2017/23644.9440.
- Falagas ME, Giannopoulou KP, Kokolakis GN, Rafailidis PI. Fosfomycin: use beyond urinary tract and gastrointestinal infections. *Clin Infect Dis* 2008;46:1069-77. doi: 10.1086/527442.
- Hosbul T, Ozyurt M, Baylan O, Bektöre B, Ardiç N, Ceylan S, et al. In vitro activity of fosfomycin trometamol in the treatment of *Escherichia coli* related uncomplicated urinary tract infections. *Mikrobiyol Bul* 2009;43:645-9.
- Lee SY, Park YJ, Yu JK, Jung S, Kim Y, Jeong SH, et al. Prevalence of acquired fosfomycin resistance among extended-spectrum β -lactamase-producing *Escherichia coli* and *Klebsiella pneumoniae*

- clinical isolates in Korea and IS26-composite transposon surrounding *fosA3*. *J Antimicrob Chemother* 2012;67:2843-7. doi: 10.1093/jac/dks319.
19. van den Bijllaardt W, Schijffelen MJ, Bosboom RW, Cohen Stuart J, Diederens B, Kampinga G, et al. Susceptibility of ESBL *Escherichia coli* and *Klebsiella pneumoniae* to fosfomycin in the Netherlands and comparison of several testing methods including Etest, MIC test strip, Vitek2, Phoenix and disc diffusion. *J Antimicrob Chemother* 2018;73:2380-2387. doi: 10.1093/jac/dky214.
 20. Tseng SP, Wang SF, Kuo CY, Huang JW, Hung WC, Ke GM, et al. Characterization of Fosfomycin Resistant Extended-Spectrum β -Lactamase-Producing *Escherichia coli* Isolates from Human and Pig in Taiwan. *PLoS One* 2015;10:e0135864. doi: 10.1371/journal.pone.0135864.
 21. Tulara NK. Nitrofurantoin and Fosfomycin for Extended Spectrum Beta-lactamases Producing *Escherichia coli* and *Klebsiella pneumoniae*. *J Glob Infect Dis* 2018;10:19-21. doi: 10.4103/jgid.jgid_72_17.
 22. Tharavichitkul P, Khantawa B, Bousoung V, Boonchoo M. Activity of Fosfomycin against extended spectrum beta-lactamase producing *K. pneumoniae* and *E. coli* in Maharaj Nakorn Chiang Mai Hospital. *J Infect Dis Antimicrob Agents* 2005;22:121-26.
 23. de Cueto M1, López L, Hernández JR, Morillo C, Pascual A. In vitro activity of fosfomycin against extended-spectrum-beta-lactamase-producing *Escherichia coli* and *Klebsiella pneumoniae*: comparison of susceptibility testing procedures. *Antimicrob Agents Chemother* 2006;50:368-70. doi: 10.1128/AAC.50.1.368-370.2006
 24. Liu HY, Lin HC, Lin YC, Yu SH, Wu WH, Lee YJ. Antimicrobial susceptibilities of urinary extended-spectrum beta-lactamase-producing *Escherichia coli* and *Klebsiella pneumoniae* to fosfomycin and nitrofurantoin in a teaching hospital in Taiwan. *J Microbiol Immunol Infect* 2011;44:364-8. doi: 10.1016/j.jmii.2010.08.012.
 25. Fajfr M, Louda M, Paterová P, Ryšková L, Pacovský J, Košina J, et al. The susceptibility to fosfomycin of Gram-negative bacteria isolates from urinary tract infection in the Czech Republic: data from a unicentric study. *BMC Urol* 2017;17:e33. doi: 10.1186/s12894-017-0222-6.
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