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ORIGINAL ARTICLE

Treatment of recurrent complicated urinary tract infections in children with vesicoureteral reflux

Tsung-Hua Wu [a,b], Fang-Liang Huang [a,c], Lin-Shien Fu [a], Chia-Man Chou [d], Ya-Li Chien [a], Chung-Ming Huang [a], Chin-Fu Lin [e], Po-Yen Chen [a,*]

[a] *Department of Pediatrics, Taichung Veterans General Hospital, Taichung, Taiwan*
[b] *Department of Pediatrics, Show Chwan Memorial Hospital, Changhua, Taiwan*
[c] *Hungkuang University, Taichung, Taiwan*
[d] *Division of Pediatric Surgery, Department of Surgery, Taichung Veterans General Hospital, Taichung, Taiwan*
[e] *Microbiology Section of the Medical Laboratory, Taichung Veterans General Hospital, Taichung, Taiwan*

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Background: Urinary tract infections (UTIs) in children with vesicoureteral reflux (VUR) are often caused by uropathogens with a high rate of drug resistance and are associated with a high rate of recurrence with a single pathogen. In this study, we evaluated the incidence of recurrent UTI and the drug resistance pattern of *Escherichia coli* in children with VUR. We also evaluated whether combination therapy comprising fosomycin plus one other antimicrobial agent is effective for treatment of recurrent UTIs.
Methods: We retrospectively reviewed the medical records of all children with VUR who developed at least one episode of UTI during the period January 1, 2003 to December 31, 2013 at a single medical center. The effectiveness of fosfomycin plus amikicin for *Enterobacteriaceae* or ceftazidime for *Pseudomonas aeruginosa* infections was prospectively studied in six children with recurrent relapsing UTIs.
Results: The study population comprised 129 children (age range, from 1month to 15 years; mean ± standard deviation, 2.37 ± 2.91 years) with VUR who developed at least one UTI during the 10-year study period; 68 (52.7%) had recurrent UTIs. The presence of an underlying urinary tract anomaly was predictive of recurrence ($p = 0.028$). The rates of susceptibility of *E. coli* to

* Corresponding author. Division of Infection, Department of Pediatrics, Taichung Veterans General Hospital, Number 1650 Taiwan Boulevard Section 4, Taichung 40705, Taiwan.
E-mail address: pychen@vghtc.gov.tw (P.-Y. Chen).

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cefazolin ($p < 0.001$) and cefotaxime ($p < 0.001$) were significantly lower in patients with recurrent UTIs. Combination therapy with fosfomycin plus amikacin or ceftazidime was shown to be an effective therapeutic option for recurrent UTIs due to a single uropathogen.
Conclusion: The rates of susceptibility of *E. coli* to commonly used antimicrobials were significantly lower in children who developed more than one episode of UTI. The empiric choice of cefazolin or cefotaxime was usually ineffective. Administration of fosfomycin plus amikacin or ceftazidime was an effective therapeutic and preventive strategy in children with VUR and recurrent relapsing UTI.

Introduction

Urinary tract infections (UTIs) are one of the most common bacterial diseases in children, with a prevalence rate of 7.0% among febrile infants.[1] Recurrent UTI is a major cause of renal scarring, which can cause hypertension, chronic renal failure, and end-stage renal disease.[2,3] Vesicoureteral reflux (VUR) is diagnosed in 18–40% of children who are investigated for a first episode of UTI. These children are at an increased risk of developing recurrent urinary tract and kidney infections, which, over time, can result in renal damage.[2,3] Risk factors of recurrent UTI in children with VUR include high reflux grade, delayed contrast passage on voiding cystourethrograms, bilateral VUR, and age < 1 year.[3–5]

Preventive strategies to reduce the risk of acute infection and renal injury in children with VUR include the administration of prophylactic antibiotics, endoscopic injection of dextranomer hyaluronic acid, antireflux surgery, maintenance of good perianal hygiene, and adequate hydration.[6] However, breakthrough infections due to drug-resistant uropathogens sometimes occur, leading to significant morbidity.

Management of recurrent UTIs in children with VUR is difficult. Our hypothesis is that multiple factors may lead to treatment failure in patients with recurrent UTI, including drug-resistant uropathogens, the mechanism and duration of antibiotics, surgical damage, and biofilm formation. The aims of this study were to evaluate the incidence of recurrent UTI among children with VUR, to evaluate the incidence of drug resistance among *Escherichia coli* isolates from children with recurrent UTI, and to initiate a pilot study to evaluate the therapeutic effectiveness of fosfomycin for preventing recurrent UTI.

Methods

We retrospectively enrolled all children who were treated for a first episode of UTI and who subsequently received a diagnosis of VUR at the Taichung Veterans General Hospital, a 1200-bed tertiary hospital in central Taiwan, during the period from January 1, 2003 to December 31, 2013. Risk factors for recurrence, causative uropathogens, and antimicrobial susceptibility test results were obtained from the medical records.

Children with VUR were separated into a recurrent UTI group or a nonrecurrent UTI group. Clinical parameters for statistical analysis included gender, age, grade of VUR, and additional urinary tract anomalies. The degree of reflux on voiding cystourethrography was determined according to the International Reflux Study classification.[7] Additional urinary tract anomalies included ureteropelvic junction stenosis, renal collecting system anomalies (duplication, renal dysplasia, or ureterocele), bladder exstrophy, and neurogenic bladder. Children with recurrent UTI were also stratified into a reinfection or relapse group. Receipt of surgical intervention including endoscopic injection of dextranomer hyaluronic acid or antireflux open surgery was also evaluated.

A UTI was defined in this study as a single pathogen with adequate colony forming units (CFU)/mL cultured from a urine specimen obtained from voiding urine (> 100,000 CFU/mL), catheterized urine (> 10,000 CFU/mL), or suprapubic puncture (> 1000 CFU/mL).[8] Recurrent UTI was defined as significant bacteriuria in children with fever or dysuria during a 12-month follow-up period. Reinfection was defined as a recurrent UTI with a new bacterial isolate and relapse was defined as a recurrent UTI with the same bacterial isolate as the primary isolate.

E. coli is the most common cause of community-acquired UTI in children. In this study, children with UTI due to *E. coli* were divided into a first episode group or a recurrent episode group. Sensitivity of isolates to ampicillin, cefazolin, gentamicin, amikacin, trimethoprim–sulfamethoxazole, cefotaxime or ceftazidime, cefepime, ciprofloxacin, meropenem, and piperacillin–tazobactam was tested *in vitro* using the disk diffusion method according to the Clinical and Laboratory Standards Institute standards.[9]

In our pilot study to evaluate the therapeutic effectiveness of fosfomycin for preventing recurrent UTI, we prospectively recruited six children with VUR and recurrent UTI. Patients with UTI due to *Enterobacteriaceae* species were administered fosfomycin 100–200 mg/kg/d every 8 hours for 7–10 days plus amikicin 15 mg/kg/d every 12 hours for 5 days. Patients with UTIs caused by *Pseudomonas aeruginosa* were given fosfomycin 100–200 mg/kg/d every 8 hours for 7–10 days plus ceftazidime 100–150 mg/kg/d every 8 hours for 7–10 days.

Statistical analysis

Ordinal variables are expressed as means ± standard deviation (or median, range) and were compared between

groups using the Mann–Whitney *U* test. Categorical variables, expressed as percentages, were analyzed using the two-tailed Yates correction of contingency (when the expected value was > 5 with a 2×2 table) or the Pearson's Chi-square test. Kendall's τ-c (when there was a rectangular table) was used for tests of trend. A p value < 0.05 was considered to represent statistical significance. All analyses were performed using the statistical package SPSS for Windows (version 20; SPSS Inc., Chicago, IL, USA).

Results

During the 10-year study period, a total of 129 children with VUR developed UTIs; 68 (52.7%) had recurrent UTIs. The rate of recurrence was 52.7%. VUR in each patient was diagnosed after the first UTI. There were no significant differences in age at diagnosis of the first UTI, gender, reflux laterality, receipt of operation, or severity of reflux grade between the recurrent and nonrecurrent UTI groups. Underlying urinary tract anomalies were found in 21 (30.9%) patients in the recurrent group and in eight (13.1%) patients in the nonrecurrent group. Children with additional urinary tract anomalies were at greater risk of recurrent UTI than children without said anomalies ($p = 0.028$; Table 1).

A total of 148 *E. coli* isolates were obtained from these patients. The rates of antimicrobial susceptibility to some antimicrobial agents empirically used as first-line treatment, such as cefazolin, gentamicin, and cefotaxime were markedly lower in patients with recurrent infection than in those with nonrecurrent infection (Fig. 1). Rates of sensitivity to cefazolin ($p < 0.001$) and cefotaxime ($p < 0.001$) were significantly lower when used in patients with recurrent UTIs. Isolates remained highly susceptible to amikacin, cefepime, ciprofloxacin, meropenem, and piperacillin–tazobactam in both groups.

Of the 68 children in the recurrent group, 44 (64.7%) developed reinfection, and 36 of them required surgical correction. The majority ($n = 33$, 91.7%) did not develop further episodes of UTI after surgical correction. The other 24 (35.3%) children in the recurrent group had relapsing UTI, and 18 (75.0%) required surgical correction. Of those patients, 13 (72.2%) relapsed after surgery. A total of 45 isolates of uropathogens were collected from the 24 children with relapsing UTI. *E. coli* (53.3%) was the most commonly isolated uropathogen. Non-*E. coli* isolates included *Klebsiella pneumoniae* (15.6%), *P. aeruginosa* (11.1%), *Proteus mirabilis* (6.7%), *Alcaligenes xylosoxidans* (4.4%), *Citrobacter freundii* (2.2%), *Klebsiella oxytoca* (2.2%), *Morganella morganii*, and *Serratia marcescens* (2.2%; Table 2).

In the pilot study, six children with recurrent UTI due to a single uropathogen were prospectively enrolled to receive combination therapy comprising fosfomycin plus amikacin or ceftazidime (Patients A–F). During the follow-up period, one patient (Patient F) did not show evidence of further UTI and four patients (Patient B, C, D, and E) had recurrent episodes of UTI due to different uropathogens. One patient (Patient A), however, had a recurrent episode due to the same pathogen after combination therapy (Table 3).

Discussion

We found that children with reinfection responded well to surgical correction. The majority (91.7%) of children did not have further episodes of UTI after surgery during the 12 months of follow-up. Surgical intervention for persistent VUR or recurrent UTI, including both open and endoscopic methods, can provide curative treatment for some patients.[6] However, the recurrence rate after surgical correction in patients with relapsing UTI due to a single pathogen is as high as 75.0%. Urinary and ureteral catheters as well as surgery-related damage to structures can lead to biofilm formation, which is associated with a recurrent single uropathogen. *E. coli* isolates from UTI have been

Table 1 Characteristics of the children with UTI and VUR

Variable		Recurrent UTI group ($n = 68$)	Nonrecurrent UTI group ($n = 61$)	p
		n (%)	*n* (%)	
Age of first UTI (y)		2.38 ± 3.15	2.35 ± 2.66	0.240[a]
Sex	Male	35 (51.5)	27 (44.3)	0.521[b]
	Female	33 (48.5)	34 (55.7)	
VUR	Bilateral	35 (51.5)	35 (57.4)	0.620[b]
	Unilateral	33 (48.5)	26 (42.6)	
VUR grade	Grade 1	1 (1.5)	2 (3.3)	0.573[c]
	Grade 2	5 (7.4)	6 (9.8)	
	Grade 3	22 (32.4)	26 (42.6)	
	Grade 4	24 (35.3)	16 (26.2)	
	Grade 5	16 (23.5)	11 (18.0)	
Surgical correction	Yes	55 (80.9)	55 (90.2)	0.216[b]
	No	13 (19.1)	6 (9.8)	
Additional urinary tract anomalies	Yes	21 (30.9)	8 (13.1)	0.028[b]
	No	47 (69.1)	53 (86.9)	

[a] Mann–Whitney *U* test.
[b] Yates's correction of contingency.
[c] Pearson Chi-square test.
UTI = urinary tract infection; VUR = vesicoureteral reflux.

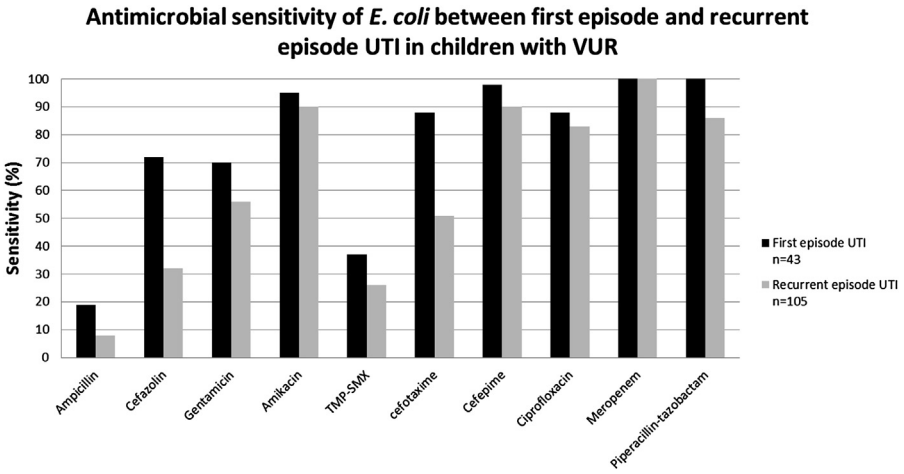

Figure 1. Drug susceptibility of *Escherichia coli* among the two groups. The antimicrobial agents cefazolin (72.1% vs. 32.4%, $p < 0.001$) and cefotaxime (88.4% vs. 51.4%, $p < 0.001$) had significantly decreased susceptible rates of *E. coli*. TMP-SMX = trimethoprim—sulfamethoxazole; UTI = urinary tract infection.

Table 2 Urine culture results in the relapse group ($n = 45$)

Strain of urine culture	*n* (%)
Escherichia coli	24 (53.3)
Klebsiella pneumoniae	7 (15.6)
Pseudomonas aeruginosa	5 (11.1)
Proteus mirabilis	3 (6.7)
Alcaligenes xylosoxidans	2 (4.4)
Citrobacter freundii	1 (2.2)
Klebsiella oxytoca	1 (2.2)
Morganella morganii	1 (2.2)
Serratia marcescens	1 (2.2)

shown to form biofilm *in vitro*.[10] Biofilm represents a structured community of bacterial cells embedded in a self-produced polymeric matrix that adheres to natural or artificial surfaces. The community is, therefore, protected from antimicrobial agents and host immune defenses. A recent review reported that the recurrence of UTI was mainly caused by a relapse of the preceding *E. coli* infection, which supports evidence that *E. coli* can invade and replicate within murine bladders, forming stable biofilm-like intracellular reservoirs for recurrent UTI.[11] It has recently been suggested that biofilm plays an important role in the pathogenesis or recurrence of UTI and persistent colonization with the same strain of uropathogen.

Table 3 Uropathogens involved in and antimicrobial treatment for each recurrent episode in children with vesicoureteral reflux

No.	Age (y)	Sex	Episode of UTI						
			1st	2nd	3rd	4th	5th	6th	7th
A	0.2	M	*P. aeruginosa*[c]	*P. aeruginosa*[*]	*P. aeruginosa*[*]	*P. aeruginosa*[b]	P. aeruginosa[**]	*P. aeruginosa*[**]	*P. aeruginosa*[**]
B	0.2	M	*S. marcescens*[g]	*S. marcescens*[*]	*P. aeruginosa*[**]	*P. aeruginosa*[**]	*P. aeruginosa*[**]	*M. morganii*[f]	*E. coli*-ESBL[e]
C	0.2	M	*E. coli*[a]	*P. aeruginosa*[c]	*P. aeruginosa*[*]	*P. aeruginosa*[**]	*P. aeruginosa*[**]	*P. aeruginosa*[**]	*K. pneumoniae*[d]
D	0.1	M	*P. aeruginosa*[*]	*E. cloacae*[*]	*P. aeruginosa*[*]	*P. aeruginosa*[**]	*P. mirabilis*[e]	*A. xylosoxidans*[**]	*T. beigelii*[**]
E	0.2	F	*E. coli*[a]	*E. coli*-ESBL[*]	*E. coli*[a]	*E. coli*[b]	*P. mirabilis*[e]	—	—
F	0.2	F	*E. coli*[a]	*E. coli*[a]	*E. coli*-ESBL[g]	*E. coli*[*]	*E. coli*[*]	—	—

[a] 1st cephalosporin.
[b] 3rd cephalosporin.
[c] 4th cephalosporin.
[d] Ciprofloxacin.
[e] Amoxicillin—clavulanate.
[f] Ampicillin.
[g] Meropenem.
[*] Fosfomycin for 7—10 days plus amikacin for 5 days.
[**] Fosfomycin plus ceftazidime for 7—10 days.

A. xylosoxidans = *Achromobacter xylosoxidans*; Age = age at first episode of UTI; *E. cloacae* = *Enterobacter cloacae*; *E. coli* = *Escherichia coli*; ESBL = extended-spectrum β-lactamases; F = female; *K. pneumoniae* = *Klebsiella pneumoniae*; *M. morganii* = *Morganella morganii*; M = male; *P. aeruginosa* = *Pseudomonas aeruginosa*; *P. mirabilis* = *Proteus mirabilis*; *S. marcescens* = *Serratia marcescens*; *T. beigelii* = *Trichosporon beigelii*; UTI = urinary tract infection.

Based on our results, therapeutic strategies for recurrent UTI in patients with VUR are becoming less effective. Administration of first-generation cephalosporins, such as cefazolin, as first-line empiric therapy can lead to treatment failure because strains of many common causative pathogens of UTI, particularly *E. coli*, are highly resistant. Broad-spectrum antibiotics such as meropenem and piperacillin—tazobactam are increasingly being used in children with recurrent UTI. One systematic review reported that fosfomycin, a very small hydrophilic compound with activity against several Gram-negative and Gram-positive aerobic bacteria, has a high level of antimicrobial activity against extended-spectrum β-lactamase (ESBL)-producing *Enterobacteriaceae* isolates with advanced resistance to antimicrobial drugs.[12] Moreover, it has been reported that combinations of fosfomycin with other antimicrobial drugs are successful against multidrug-resistant Gram-negative pathogens and biofilm-related bacteria *in vitro*.[13] In our pilot study, we evaluated the effectiveness of combination therapy comprising fosfomycin for 7—10 days plus amikacin for 5 days to treat infections due to *Enterobacteriaceae* species, and fosfomycin for 7—10 days plus ceftazidime for 7—10 days to treat infections due to *P. aeruginosa* in six children with recurrent UTIs (Table 3). One of the six children had a successful response and no further episodes of infection were noted during 12 months of follow-up. Recurrent episodes occurred in four of the six children; however, in all four patients the uropathogens responsible for the recurrent episodes differed from those in previous episodes. Only one of the six children in the pilot study failed to respond to combination therapy and had persistent recurrent infections with the same uropathogen, namely *P. aeruginosa*. Treatment failure in that child might have been due to placement of a ureteral catheter to treat underlying ureteropelvic junction stenosis. Combination therapy comprising fosfomycin plus a second antimicrobial drug was shown to be an effective therapeutic strategy; however, further studies are needed to determine the efficacy of this treatment.

The most common uropathogen in primary and recurrent UTIs is *E. coli* and cefazolin is the drug of choice for treating such infections in children. In our previous study, we found that 86.5% of *E. coli* isolates from children with a first episode of community-acquired UTI were susceptible to cefazolin alone.[14] In another study conducted in Taiwan during 2003—2010, the rates of susceptibility to cefazolin among *E. coli* isolates obtained from children with community-acquired UTI ranged from 62% to 73%.[15] In the current study, the sensitivity of *E. coli* to cefazolin in the first episode group was 72.1%, but decreased to 32.4% in the recurrent episode group. Younis et al[16] demonstrated that urine cultures of isolates from children with recurrent or complicated UTIs are becoming increasingly resistant to commonly used antibiotics such as trimethoprim—sulfamethoxazole or cephalexin.

In our study, the only significant predictor of recurrent UTI was the presence of additional urinary tract anomalies (30.9% vs. 13.1%, $p = 0.028$), such as ureteropelvic junction stenosis, renal collecting system anomalies (duplication, renal dysplasia, ureterocele), or neurogenic bladder. One possible reason for this finding is that some of our patients were referred from local community hospitals with severe infections or for early surgical intervention. Another possible reason is that we included children with VUR and urinary tract anomalies in our study. These children are often exposed to other risk factors for recurrence such as additional surgical procedures, the need for urinary catheter placement, use of broad-spectrum antibiotics, and longer hospital stay, variables that are well-known to increase the rate of recurrence of UTI.

This study has some limitations, including its retrospective design and small sample size, which may have limited our ability to identify differences between groups. In addition, some important variables were not investigated because of the retrospective nature of the study, including the timing of diagnosing VUR, use of chemoprophylaxis, and adequate imaging to evaluate the presence of renal scarring. The definitions of reinfection and relapse utilized in this study were purely clinical in nature and are limited by the lack of clonal analysis. Finally, our pilot study on the effectiveness of fosfomycin therapy was limited to only six patients. Large-scale, prospective studies on fosfomycin alone and in combination with other antimicrobials are needed.

In conclusion, the rates of susceptibility of *E. coli* to many commonly used antibiotic agents decreased in children with VUR who were treated for recurrent UTI, especially in patients with additional urinary tract anomalies. The empiric choice of cefazolin or cefotaxime alone for recurrent UTI was usually ineffective. Surgical correction is suggested for children with recurrent reinfection. Clinical intervention comprising fosfomycin plus a second antimicrobial drug is a promising therapeutic and preventive strategy for managing recurrent relapsing UTI in children with VUR.

Conflicts of interest

All contributing authors declare no conflicts of interest.

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References

1. Shaikh N, Morone NE, Bost JE, Farrell MH. Prevalence of urinary tract infection in childhood: a meta-analysis. *Pediatr Infect Dis* 2008;**27**:302—8.
2. Duzova A, Ozen S. Vesicoureteral reflux in childhood. *Saudi J Kidney Dis Transplant* 2003;**14**:290—5.
3. Shiraishi K, Yoshino K, Watanabe M, Matsuyama H, Tanikaze S. Risk factors for breakthrough infection in children with primary vesicoureteral reflux. *J Urol* 2010;**183**:1527—31.
4. Park S, Han JY, Kim KS. Risk factors for recurrent urinary tract infection in infants with vesicoureteral reflux during prophylactic treatment: effect of delayed contrast passage on voiding cystourethrogram. *Urology* 2011;**78**:170—3.

5. Jang HC, Park YJ, Park JS. Predicting factors of breakthrough infection in children with primary vesicoureteral reflux. *Yonsei Med J* 2012;**53**:748—52.
6. Peters CA, Skoog SJ, Arant Jr BS, Copp HL, Elder JS, Hudson RG, et al. Summary of the AUA Guideline on Management of Primary Vesicoureteral Reflux in Children. *J Urol* 2010;**184**: 1134—44.
7. Elder JS, Peters CA, Arant Jr BS, Ewalt DH, Hawtrey CE, Hurwitz RS, et al. Pediatric Vesicoureteral Reflux Guidelines Panel summary report on the management of primary vesicoureteral reflux in children. *J Urol* 1997;**157**:1846—51.
8. Subcommittee on Urinary Tract Infection, Steering Committee on Quality Improvement and Management, Roberts KB. Urinary tract infection: clinical practice guideline for the diagnosis and management of the initial UTI in febrile infants and children 2 to 24 months. *Pediatrics* 2011;**128**:595—610.
9. Clinical and Laboratory Standards Institute (CLSI). *Performance standards for antimicrobial susceptibility testing; 21st informational supplement (CLSI document M100-S21)*. Wayne, PA: Clinical and Laboratory Standards Institute; 2011.
10. Salo J, Sevander JJ, Tapiainen T, Ikäheimo I, Pokka T, Koskela M, et al. Biofilm formation by *Escherichia coli* isolated from patients with urinary tract infections. *Clin Nephrol* 2009; **71**:501—7.
11. Ejrnæs K. Bacterial characteristics of importance for recurrent urinary tract infections caused by *Escherichia coli*. *Dan Med Bull* 2011;**58**:B4187.
12. Falagas ME, Kastoris AC, Kapaskelis AM, Karageorgopoulos DE. Fosfomycin for the treatment of multidrug-resistant, including extended-spectrum beta-lactamase producing, Enterobacteriaceae infections: a systematic review. *Lancet Infect Dis* 2010;**10**:43—50.
13. Roussos N, Karageorgopoulos DE, Samonis G, Falagas ME. Clinical significance of the pharmacokinetic and pharmacodynamic characteristics of fosfomycin for the treatment of patients with systemic infections. *Int J Antimicrob Agents* 2009; **34**:506—15.
14. Lu KC, Chen PY, Huang FL, Yu HW, Kao CH, Fu LS, et al. Is combination antimicrobial therapy required for urinary tract infection in children? *J Microbiol Immunol Infect* 2003;**36**: 56—60.
15. Chen PC, Chang LY, Lu CY, Shao PL, Tsai IJ, Tsau YK, et al. Drug susceptibility and treatment response of common urinary tract infection pathogens in children. *J Microbiol Immunol Infect* 2014;**47**:478—83.
16. Younis N, Quol K, Al-Momani T, Al-Awaisheh F, Al-Kayed D. Antibiotic resistance in children with recurrent or complicated urinary tract infection. *JNMA J Nepal Med Assoc* 2009;**48**:14—9.