



OUR EXPERIENCE WITH CONTRAST-ENHANCED VOIDING UROSONOGRAPHY

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SUMMARY – Vesicoureteral reflux (VUR) is one of the most common anomalies of the urinary system in children. Contrast-enhanced voiding urosonography (ceVUS) is one of the best methods in VUR diagnosis. This study compared characteristics associated with VUR specific images and categorized patients according to a particular VUR grade. The study included 183 children. VUR was detected in 38.9% of patients, mean age 1.7 ± 1.1 years. Grade II VUR was most common (60.3%), followed by grade III (29.4%). Study results showed that VUR occurred irrespective of age, gender, previous ultrasound findings, causative agent, and severity of urinary tract infection (UTI). VUR was more common in children with recurrent UTI. In the group of children with the first UTI not caused by *Escherichia coli* or with recurrent UTI, boys more commonly suffered from severe VUR (grade IV-V; 66.7%), while girls suffered from moderate VUR (grade II-III; 100%). In this study, the incidence of VUR in prenatally diagnosed hydronephrosis was 28.6%. It is necessary to develop an algorithm for the treatment of children after UTI in Croatia, which should include ceVUS. All children with possible VUR should be referred to a specialized center where it is possible to perform ceVUS.

Key words: *Contrast-enhanced voiding urosonography (ceVUS); Vesicoureteral reflux; Children*

Introduction

Vesicoureteral reflux (VUR) is an anatomic and functional abnormality caused by non-physiologic retrograde flow of urine from the bladder into the ureter and renal canal system. VUR is one of the most common anomalies in children and the most common cause of renal scarring in children with a consequence of renal function damage¹. There is primary and secondary VUR; in primary VUR, the function of the lower urinary tract is normal, whereas secondary VUR is associated with obstruction or impaired function of the lower urinary tract (posterior urethral valve, neurogenic bladder, overactive bladder, urinary disorders after bladder surgery, ureterocele, calculus, etc.)². VUR is associated with renal nephropathy and scarring, which can cause hypertension and renal failure. It is impor-

tant to diagnose morphological or functional abnormality of the urinary tract in children with proven urinary tract infection (UTI) as a predisposition for kidney damage¹. It is recommended to exclude VUR in high-risk patients, i.e. those with hydronephrosis, kidney scarring, atypical UTI, in children with complex conditions (urogenital anomalies, neurogenic bladder, or bowel dysfunction), and those with a positive family history of VUR^{3,4}.

During childhood, about 8% of girls and 2% of boys have a UTI, and it is estimated that there is underlying VUR in 18%-40% of them^{5,6}. It is recommended to exclude VUR in the first febrile UTI in a child from 2 months to 2 years of age in cases of atypical bacteria isolated from urine culture (all except for *Escherichia (E.) coli*) and pathologic findings of kidney and urinary tract ultrasound⁶⁻⁹. Kidney ultrasound has been shown to have neither specificity nor sensitivity for VUR detection¹⁰.

Diagnosis of VUR is based on 3 methods, i.e. voiding cystourethrography (VCU), radionuclide cystogra-

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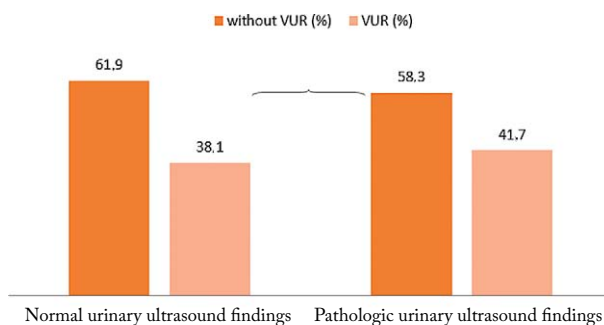


Fig. 1. Comparison of vesicoureteral reflux (VUR) in children with normal urinary ultrasound finding and pathologic urinary ultrasound findings of urinary tract ultrasound.

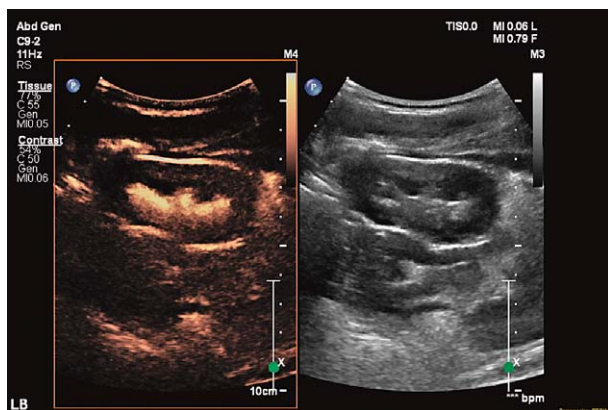


Fig. 2. Grade III-IV vesicoureteral reflux (active and passive) of the left kidney in a 2-year-old girl with recurrent urinary tract infection.

phy, and contrast-enhanced voiding urosonography (ceVUS)^{1,3,11}.

Today, the sensitivity of ceVUS is very high (ranging from 80% to 100%) with high specificity (ranging from 77% to 97%)^{12,13}. It has become a routine and professionally recognized method for the diagnosis of VUR in children^{1,11}. CeVUS is performed without ionizing radiation, sedation, and use of permanent catheter, it is highly sensitive, affordable, and it provides insight into anatomic details^{1,14,15}. The sensitivity of ceVUS is higher than that of micturating cystourethrogram (47% increase in the detection of VUR was recorded when using ceVUS only)¹². It is a real-time imaging method that provides continuous extended imaging with a high possibility of detecting VUR as an intermittent phenomenon¹¹. According to the recommendations of the European Federation of

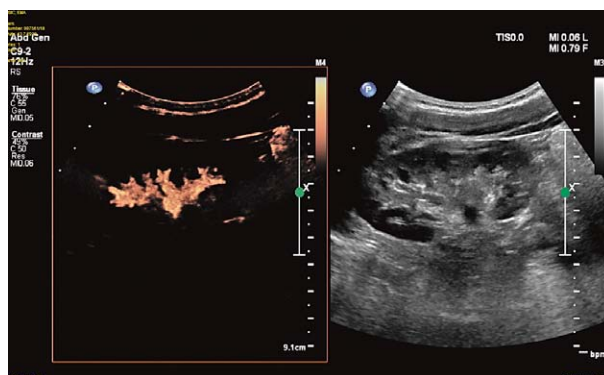


Fig. 3. Grade III-IV vesicoureteral reflux of the left kidney in a 3.5-year-old girl with single umbilical artery syndrome.

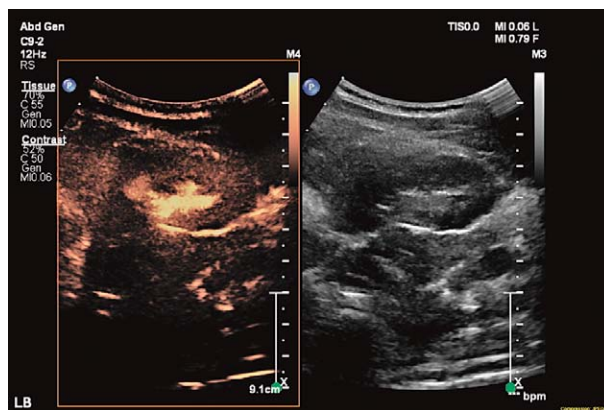


Fig. 4. Grade III-IV vesicoureteral reflux of the left kidney in a 2.5-year-old girl with recurrent urinary tract infection.

Societies for Ultrasound in Medicine and Biology, European Society of Urogenital Radiology, and European Society of Paediatric Radiology, ceVUS is the method of choice in the detection and grading of VUR¹. The advantage is that, in addition to functional evaluation of the existence of VUR, ceVUS also provides anatomic evaluation of the kidney and bladder *via* ultrasound⁵. However, it should be noted that this is a highly subjective procedure, and it depends on the knowledge and experience of the practicing physician¹⁶. There are certain limitations of the procedure as well, such as possible artifacts due to ultrasound contrast, limited possibility in the examination of fistulas in children with anorectal anomalies or anatomic details of the bladder neck, poor cooperation of the child, or inability to clearly show the kidney due to severe scoliosis. Furthermore, this diagnostic procedure should

Table 1. Characteristics of all study children

	n	%
Sex		
Male	71	38.8
Female	112	61.2
Indication for ceVUS		
First UTI	115	62.8
Recurrent UTI	50	27.3
Hydronephrosis	7	3.8
Urogenital anomalies	6	3.3
Neurogenic bladder	2	1.1
Other	3	1.6
ceVUS findings		
Normal	107	61.1
Pathologic	68	38.9
grade I	2	2.9
grade II	41	60.3
grade III	20	29.4
grade IV	4	5.9
grade V	1	1.5
Kidney ultrasound		
Normal	110	60.1
Pathologic	73	39.9

ceVUS = contrast-enhanced voiding urosonography; UTI = urinary tract infection

not be performed during acute infection but after the infection has been treated^{1,3,11}.

Determining VUR grade in ceVUS is similar to determining grades in voiding cystourethrography according to the 1985 international system of radiographic grading of vesicoureteric reflux (based on ureteral dilatation and renal duct system). In ceVUS, grading is based on the presence of microbubbles of the ultrasound contrast agent in the ureter and renal duct system, as well as on the grades of dilatation, as follows: grade I – reflux into the distal part of the undilated ureter; grade II – reflux reaches the proximal part of the drainage system without dilatation; grade III – reflux into the dilated ureter and calyces with minimally blunted fornices; grade IV – reflux to the heavily dilated ureter; and grade V – severe reflux to the gross ureteral dilatation and tortuosity with a loss of papillary impressions^{6,17,18} (Figs. 1-3).

Indications for ceVUS include the first febrile UTI with pathologic findings of urinary tract ultrasound

Table 2. Correlation of VUR with the characteristics in children with UTI

	VUR (%)	p
Sex		
Male	37.93	0.739
Female	41.35	
Age (yrs)		
<1	34.48	0.141
≥1	47.06	
Cause		
<i>Escherichia coli</i>	41.0	0.5
Other	42.0	
UTI		
First	34.78	0.035
Recurrent	53.19	
Urinary tract ultrasound findings		
Normal	38.1	0.739
Pathologic	41.35	
Leukocyte count (x10⁹/L)		
<20	40.28	0.7872
≥20	43.75	
CRP (mg/L)		
≤50	37.5	0.46
>50	45.45	

VUR = vesicoureteral reflux; UTI = urinary tract infection; CRP = C-reactive protein

(e.g., ureterohydronephrosis, scarring, anomalies and malformations of the urinary tract), the first febrile UTI caused by a bacterium other than *E. coli* and first febrile UTI with a positive family history of VUR, recurrent UTI with normal urinary tract ultrasound, prenatal hydronephrosis or other urinary system abnormality, and monitoring of the pre-existing VUR⁷.

The aim of this study was to examine the indications for ceVUS in children, to determine the correlation of individual characteristics with VUR findings, and to present first results after the introduction of this diagnostic procedure at the Division of Nephrology, Department of Pediatrics, Sestre milosrdnice University Hospital Centre, Zagreb, Croatia.

Materials and Methods

Data were collected retrogradely from medical records of 183 children scheduled for ceVUS scan at the

Division of Nephrology, Department of Pediatrics, Sestre milosrdnice University Hospital Centre, from September 2016, when the test was first introduced at the Department, until the end of December 2018.

The test was conducted on a Philips Affiniti 50G ultrasound machine, with a 2.5-5 MHz convex probe. Sulfur hexafluoride (SonoVue 8 $\mu\text{L}/\text{mL}$) was used as the ultrasound contrast agent. The ceVUS method detects microbubbles of the ultrasound contrast agent, which are shown in color and on the surface of the ultrasound B-mode images. The precondition is a sterile urine culture and clean urine within 7 days, as well as the informed consent signed by parents. The diagnostic procedure includes initially filling the bladder with the contrast agent (sulfur hexafluoride (1 mL SonoVue) was injected into the saline solution). During the filling and discharging phase (spontaneous urination), the bladder and kidneys undergo ultrasound examination *via* longitudinal and transverse images using contrast-specific software during which the child lies on the abdomen¹¹.

Gender distribution, age, ceVUS indications, frequency of pathologic findings on urinary tract ultrasound, and frequency of VUR were analyzed. In children with UTI, we observed gender, age, UTI characteristics (relapse, febrility), distribution of pathogens, inflammatory parameters at the time of infection (leukocytes and C-reactive protein (CRP)), and frequency of pathologic findings on urinary tract ultrasound. We analyzed the correlation of all of the above-mentioned characteristics with the incidence of VUR. CRP values were divided into three groups: low CRP (0-50 mg/L), medium-high CRP (51-100 mg/L) and high CRP (>100 mg/L), and leukocyte count into two groups: low leukocyte count (<20x10⁹/L) and high leukocyte count ($\geq 20 \times 10^9/\text{L}$). According to the ceVUS findings, we divided VUR into groups according to severity, as follows: mild (grade I), moderate (grade II-III) and severe (grade IV-V).

Patient characteristics were expressed as percentage and mean. The χ^2 -test was used to determine differences between two categorical variables and Student's t-test was used to determine differences between arithmetic means. Values of $p < 0.05$ were considered statistically significant. Approval for the study was granted by the Ethics Committee of the Sestre milosrdnice University Hospital Centre.

Results

All children

Contrast-enhanced voiding urosonography was indicated in 183 children, mean age 1.7 ± 1.1 years, with a higher prevalence in girls ($n=112$) compared to boys ($n=71$); M:F=61.2%:38.8%. The most common indication for ceVUS in children was the first UTI ($n=115$; 62.8%), followed by recurrent UTI ($n=50$; 27.3%), perinatal hydronephrosis ($n=7$; 3.8%), urogenital tract abnormalities ($n=6$; 3.3%), suspected unstable bladder ($n=2$; 1.1%), and other reasons ($n=3$; 1.6%) (Table 1).

Of the total of 183 children having undergone urinary tract ultrasound, 39.9% (M 28, F 45) had pathologic findings, most often duct system dilatation (90.5%), more often unilateral than bilateral (61% *vs.* 39%), followed by duplex canal system (4%), and others (5.5%).

Of the total of 183 children indicated for ceVUS, 175 of them underwent the procedure, while the remaining eight did not show up for the procedure on the day it was scheduled. Out of 175 children having undergone ceVUS, VUR was detected in 38.9% ($n=68$), equally in both genders (M 32.3% *vs.* F 40.2%; $p=0.47$). Grade II VUR was most commonly diagnosed (60.3%), followed by grade III (29.4%) (Table 1). In children with prenatal hydronephrosis, VUR was diagnosed in 28.6% ($n=2$).

Children with UTI

Of the total number of children with UTI, 90.1% ($n=160$) were referred to ceVUS. The most common indications were the first UTI (62.8%) and febrile UTI (both first and recurrent) (75.8%). Girls were significantly more likely to suffer from recurrent UTI (F 34.6% *vs.* M 18.97%; $p=0.0466$). As expected, the most common cause of UTI was *E. coli* (78.6%), followed by *E. coli* ESBL (6.1%) and *Klebsiella pneumoniae* (6.1%), *Pseudomonas aeruginosa* (1.5%) and others (7.6%). There was similar distribution of pathogens regardless of the first or recurrent UTI.

Of the 183 children, CRP value was recorded in 49% and leukocyte count in 48%. Low CRP value (62%) and low leukocyte count (81%) were most commonly found. Medium CRP was recorded in 13% and high CRP in 37% of patients, while high leukocyte count was recorded in 19% of patients.

Pathologic findings of urinary ultrasound were found in 35.8% of children with UTI, significantly more frequently in boys (M 65.5% *vs.* F 21.15%; $p=0.0001$) and in children with the first UTI (first UTI 42.61% *vs.* recurrent UTI 23.4%; $p=0.0308$).

In children with UTI, VUR was found in 39.4% of them, equally in the following categories observed: (a) both genders (M 37.93% *vs.* F 41.35%; $p=0.739$); (b) children with normal and pathologic ultrasound findings (normal ultrasound 38.1% *vs.* pathologic ultrasound 41.35%; $p=0.739$); (c) children with UTI caused by *E. coli* and children with UTI caused by other pathogens (41% *vs.* 42%; $p=0.5$); (d) children with low and moderate to high CRP values (37.5% *vs.* 45.45%; $p=0.46$); (e) children with low and high leukocyte count (40.28% *vs.* 43.75%; $p=0.787$); and (f) children younger and older than 1 year (before 1st year 34.48% *vs.* after 1st year 47.06%; $p=0.141$). VUR was found significantly more frequently in children with recurrent UTI (recurrent UTI 53.19% *vs.* first UTI 34.78%; $p=0.035$) (Table 2).

In the group of children with the first UTI caused by *E. coli*, the incidence of VUR was not associated with ultrasound finding (normal ultrasound 36% *vs.* pathologic ultrasound 35.5%; $p=1.00$). There was no severe case of VUR in this group; all children suffered from moderate VUR grade regardless of the ultrasound findings. In the group of children with the first UTI not caused by *E. coli* or with recurrent UTI, severe VUR (grade IV-V; 66.7%) was more common in boys and moderate VUR (grade II-III; 100%) in girls.

Discussion

The incidence of VUR after the first febrile UTI is 30%–40%, significantly higher than the incidence of VUR in a healthy population, which is <1%^{5,6,8,20}. This is consistent with our results, where VUR was found in 34.78% of children after the first febrile UTI. The overall incidence of VUR in the observed sample was 39.4%, and among them, 41.7% had pathologic ultrasound findings. The results of this study indicated a similar prevalence of VUR in children with normal ultrasound findings and those with pathologic ultrasound findings during UTI. The above-mentioned leads to a conclusion that ultrasound findings cannot be considered a relevant screening tool for VUR after the first febrile UTI^{1,10,21,22}. Observing the children af-

ter the first UTI caused by *E. coli*, all these patients had moderate VUR (grade II-III), which is not the case with other infectious agents that lead to severe VUR (grade IV-V). These findings are consistent with the current research⁶. The results of this study are in line with the currently applied guidelines issued by the American Academy of Pediatrics and the National Institute for Health and Care Excellence, where no further testing is required in children aged 2–24 months after the first febrile UTI in which *E. coli* has been isolated as a causative agent and in the case of normal ultrasound findings of the kidney and urinary tract^{6,7,24–26}. Further testing for VUR is indicated in the case of recurrent UTI, the first UTI with the causative agent other than *E. coli*, and in pathologic ultrasound findings^{7,25}. Somewhat similar results were obtained in this study; VUR was confirmed in 53.19% ($n=25$) after recurrent UTI, which is slightly lower than that reported in the reference works (70%–75%)¹⁰. However, other characteristics observed, such as the causative agent of the infection and ultrasound findings, were not significant indicators of VUR in our study, contrary to the above-mentioned guidelines.

In this study, we did not find the severity of UTI to influence the incidence of VUR. Thus, CRP and leukocyte count in UTI alone cannot be considered indicators of VUR. Furthermore, the age of children was not in correlation with VUR incidence in this study. We did not find a statistically significant gender difference in the prevalence of VUR, but we did find that girls had recurrent UTI more often than boys, whereas boys had pathologic ultrasound findings of urinary tract more frequently during UTI. However, these insights do not help us predict the presence of VUR. This study indicated that children with the first UTI caused by *E. coli* suffered from lower grade VUR (100% had VUR grade I-III *vs.* 86.1% of VUR grade I-III in children with recurrent UTI and the first UTI caused by bacteria other than *E. coli*), which has also been reported elsewhere^{6,19}.

Four percent of patients were referred to ceVUS due to prenatal hydronephrosis, which is consistent with other studies. The reported incidence of VUR in prenatally diagnosed hydronephrosis is 17%–37%, and in this study, it was 28.6%^{7,24}. The genetic component of multifactorial polygenic or autosomal-dominant VUR inheritance needs to be mentioned as well. In a child with the first febrile UTI whose siblings have

proven VUR, VUR should be considered due to the 30%-60% incidence of VUR in closest relatives^{5,6,8}. It is even recommended to consider VUR in siblings and children of parents with severe VUR^{23,27}.

In studies where VUR was diagnosed using only ceVUS or only VCU, there was a 47% increase in the detection of VUR when using only ceVUS (ceVUS 56% *vs.* VCU 9%). The exception is suspected pathology of the urethra where the method of choice is still VCU¹⁶.

Analyzed data point to a higher likelihood of underlying VUR in recurrent UTI, whereas gender, age, urinary tract ultrasound examination, severity, and other causative agents of UTI have no predictive value for VUR detection. CeVUS is one of the best choices in modern nephrology for detection of VUR, with high sensitivity and negligible side effects. Also, girls suffer from recurrent UTI significantly more often than boys. Every child with indications for VUR should be referred to a center that has experience in ceVUS examination²⁸.

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Sažetak

NAŠA ISKUSTVA S ULTRAZVUČNOM KONTRASTNOM CISTOGRAFIJOM

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Vezikoureteralni refluks (VUR) je jedna od najčešćih anomalija mokraćnog sustava u dječjoj dobi. Kontrastom pojačana mikcijska cistouretografija (ceVUS) postaje priznata metoda za dijagnozu VUR-a u specijaliziranim centrima. U našem radu htjeli smo utvrditi indikacije za izvođenje ceVUS-a, provjeriti postoje li karakteristike povezane s VUR-om te time možda moći predvidjeti koja su djeca u većem riziku za VUR. VUR je dokazan u 38,9% od 183 obrađene djece, srednje dobi 1,7±1,1 godina, a najčešće je dijagnosticiran VUR II. stupnja (60,3%) i III. stupnja (29,4%). Naši rezultati pokazuju da je pojava VUR-a neovisna o spolu, dobi, prethodnom nalazu ultrazvuka, uzročniku i vrijednostima upalnih parametara kod uroinfekcije, ali je zato statistički značajno povezana s recidivom uroinfekcije. Treba napomenuti da se u skupini djece s prvom uroinfekcijom koja nije uzrokovana bakterijom *Escherichia coli* ili kod recidiva većinom kod dječaka otkriva VUR visokog stupnja (IV.-V. stupanj u 66,7% slučajeva), dok djevojčice imaju VUR umjerenog stupnja (II.-III. stupanj u 100% slučajeva). U našem istraživanju je incidencija VUR-a kod prenatalne hidronefroze bila 28,6%. Nameće se potreba izrade algoritma obrade djece u Hrvatskoj nakon preboljele uroinfekcije i uvrštenje ove nove pretrage u algoritam. Kod sumnje na VUR djecu treba uputiti u specijalizirani centar gdje je moguće raditi ceVUS.

Ključne riječi: Mikcijska cistouretografija pojačana kontrastom (ceVUS); Vezikoureteralni refluks; Djeca