Urinary Interleukin-8 in Acute Pyelonephritis of Children
A Before-After Study

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Introduction. The aim of this study was to assess urinary interleukin-8 (IL-8) levels in pyelonephritis and its relation with the clinical course of the infection and of inflammatory changes detected by renal scintigraphy.

Materials and Methods. In this quasi-experimental before-after study, we evaluated 91 children aged 1 to 144 months (mean 34.4 ± 35.2 months) with pyelonephritis. Inflammatory markers including erythrocyte sedimentation rate, C-reactive protein, leukocyte count, and urinary IL-8, together with the results of ultrasonography, voiding cystourethrography, and dimercaptosuccinic acid renal scintigraphy were evaluated in these children. The ratios of urinary IL-8 to creatinine (IL-8/C) before and after the treatment were compared with each other.

Results. Urinary IL-8/C levels were significantly higher after the empirical treatment in comparison with those before the treatment (0.19 ± 0.21 versus 0.51 ± 0.53, $P < .001$). No correlation was found between the urinary IL-8 levels and leukocyturia, urine culture results, other inflammatory markers, or findings of imaging examinations.

Conclusions. We found high urinary IL-8 levels in children with pyelonephritis. We also documented its increasing after the treatment. We conclude that evaluation of urinary IL-8 can be a noninvasive test for diagnosis of upper urinary tract infection and its response to treatment.

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INTRODUCTION

Urinary tract infections (UTIs) are common in children. At least 8% of girls and 2% of boys are estimated to have at least 1 episode of UTI during childhood.[1] They may present with a range of severity from cystitis to febrile UTI or pyelonephritis. Special attention has been given to early diagnosis and treatment of acute infectious episodes in children with UTI, in addition to the reduction of chronic kidney damage and its clinical consequences. The presence of renal scarring has been documented in 5% to 15% of the children assessed after the first febrile UTI.[1] However, manifestation of the disease may be vague with nonspecific symptoms. While the presence or absence of a true UTI is occasionally difficult to determine, distinction between cystitis and pyelonephritis is even more problematic. No clinical findings (such as fever or flank pain) and no laboratory studies (such as erythrocyte sedimentation rate [ESR], C-reactive protein [CRP], and leukocyte count) are accurate in distinguishing pyelonephritis from cystitis.[2] In

the past decade, dimercaptosuccinic acid (DMSA) renal scintigraphy has been considered an objective method for the localization of the UTI site.[3-4] It is a useful but an invasive method, and there are some concerns about the accuracy of DMSA scan in children, and especially in infants.[5] Therefore, physicians prefer to use a noninvasive method for diagnosis of pyelonephritis in children.

Urinary excretion of enzymes, particularly β 2-microglobulin, N-acetyl-β-D-glucosaminidase (NAG), and interleukins, is considered a relatively simple, fast, and noninvasive method for detection of pyelonephritis.[6-7] Interleukin-8 (IL-8) is a neutrophil-activation protein excreted from the renal tubules due to acute inflammatory insults. Sheu and colleagues showed significantly higher initial serum and urine IL-8 levels in children with acute pyelonephritis.[8] Also, Krzemien and coworkers found significantly higher IL-6 and IL-8 levels in children with febrile UTI and elevated inflammatory markers. However, they showed that IL-6 and IL-8 levels do not differentiate acute pyelonephritis from lower UTI in children aged less than 24 months.[9] The aim of this study was to assess the value of urinary IL-8 in acute pyelonephritis and to compare it with other indexes traditionally used for this purpose in children.

MATERIALS AND METHODS

We conducted a quasi-experimental (before-after) study from April 2005 to September 2007 on children who were admitted to Mofid Children's Hospital due to pyelonephritis. Diagnosis of pyelonephritis was based on clinical manifestations (fever, abdominal pain, anorexia, vomiting, frequency, and dysuria) and paraclinical findings (leukocyturia, positive urine culture for microorganisms, increased ESR, positive CRP, leukocytosis, and abnormal DMSA scan). All of our patients had normal kidney function, and they did not have any sign of glomerular or tubular disease or other infectious diseases.

The first fresh morning urine sample was collected from the patients before and 7 days after the treatment and analyzed for creatinine and IL-8 concentrations. Urinary IL-8 was measured by enzyme-linked immunosorbent assay (Sanquin Kit, Amsterdam, The Netherlands) and urinary creatinine, by the Jaffee method (RA-1000). The patients were treated with a uniform empirical treatment protocol (75 mg/kg of intravenous ceftriaxone for 10 days). We evaluated our patients with DMSA renal scintigraphy, voiding cystourethrography, kidney ultrasonography, and biochemical studies. Renal scintigraphy and ultrasonography were performed during the first 3 days of admission and voiding cystourethrography, at the end of treatment. The ethics committees of Pediatric Infectious Research Center (Shahid Beheshti University [MC]) approved this study.

Continuous data were expressed as mean ± standard deviation. Comparisons were done using the Wilcoxon signed rank test and Mann-Whitney test, and correlations were tested using the Pearson correlation coefficient. Values of *P* less than .05 were considered significant. The SPSS software (Statistical Package for the Social Sciences, version 13.0, SPSS Inc, Chicago, Ill, USA) was used for the analyses.

RESULTS

Urine samples were obtained from 132 children diagnosed with pyelonephritis, of whom 91 provided another sample after the treatment, too. The remainder of the children were lost to follow up, discharged before completion of the treatment course, or refused to complete the study. The mean of age of the 91 children who completed the study was 34.4 ± 35.2 months and they were 19 boys (20.9%) and 72 girls (72.1%). The demographic and clinical characteristics of the patients are shown in the Table.

Of the 91 children diagnosed with pyelonephritis, 41 (45.0%) were febrile (mean, 39.6°C; range, 38.3°C to 41.2°C) and 29 (31.9%) had positive urine cultures (*Escherichia coli* in most cases). Initial urinalysis revealed pyuria in all of the children, ranging from 3 to 5 leukocytes per high-power field to "too numerous to count." In 83 patients (91.2%), CRP was reported positive and in 44 (48.2%), proteinuria was detected in urinalysis. In 80 (87.9%) of the children, ultrasonography reported normal kidneys and urinary tract. Of all voiding cystourethrography studies performed, 11 (16.9%) had reportedly abnormal findings, demonstrating vesicoureteral reflux. Finally, abnormality in cortical uptake was found in all of the patients on DMSA scintigraphy, and scar formation, in 10.0%.

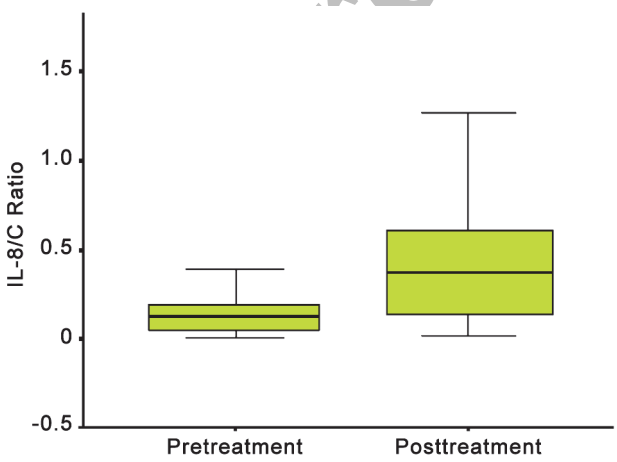
High concentrations of cytokines were demonstrated in the after-treatment urine

Demographic and Clinical Data of Children With Pyelonephritis*

Parameters	Mean ± SD	Range
Age, mo	34.4 ± 35.2	(1 to 144)
Body weight, kg	13.81 ± 8.40	(3.1 to 47.0)
Systolic BP, mm Hg	94.0 ± 11.6	(70 to 115)
Diastolic BP, mm Hg	62.0 ± 9.6	(40 to 80)
Leukocyte count, × 10^3/L	11.71 ± 4.59	(3.0 to 20.6)
Hemoglobin, g/dL	11.39 ± 1.41	(8.4 to 14.2)
ESR, mm/h	45.45 ± 31.35	(2.0 to 122.0)
BUN, mg/dL	10.35 ± 7.99	(2.0 to 28.0)
Serum electrolytes		
Creatinine, mg/dL	0.78 ± 1.47	(0.2 to 1.0)
Calcium, mg/dL	9.53 ± 1.30	(7.0 to 12.0)
Phosphate, mg/dL	4.93 ± 1.47	(3.6 to 6.8)
Sodium, mEq/L	136.29 ± 12.67	(128 to 146.0)
Potassium, mEq/L	4.66 ± 1.80	(3.5 to 5.4)
Urine parameters		
Specific gravity	1009.9 ± 89.8	(1003 to 1035)
Nitrite, μmol/L	1.218 ± 0.415	(1.00 to 2.00)
IL-8/C ratio		
Before treatment	0.185 ± 0.208	(0.01 to 1.00)
After treatment	0.511 ± 0.527	(0.02 to 2.46)

*SD indicates standard deviation; BP, blood pressure; ESR, erythrocyte sedimentation rate; BUN, blood urea nitrogen; IL-8, interleukin-8; and C, creatinine.

samples, with substantially lower concentrations in the before-treatment samples. The differences between the cytokine-creatinine ratios in the initial urine samples and the follow-up samples were significant (0.19 ± 0.21 versus 0.51 ± 0.53; $P < .001$; Figure). No significant correlations were seen between urinary IL-8 and urine leukocyte count, urine culture results, ESR, CRP, blood leukocyte count, kidney ultrasonography results, or voiding cystoureterography studies.



The pretreatment and pottreatment levels of the ratio of urinary interleukin-8 to creatinine (IL-8/C). The increase of this ratio after the treatment period was significant ($P < .001$).

DISCUSSION

The lipid A component of endotoxin and P fimbriae present in *Escherichia coli* and other gram-negative bacteria induce an inflammatory reaction that has been linked to kidney scarring. Previous studies have shown that IL-1β, IL-6, and IL-8 participate in this response, all having been found in elevated quantities in the urine of patients with UTI.[8-10] Interleukin-8 is a cytokine that acts as a chemotactic factor for neutrophils, T-cell subsets, and basophils that activates neutrophils to release lysosomal enzymes, undergo a respiratory burst, and degranulate. Production of IL-8 by mesangial cells has been demonstrated in response to IL-1β and tumor necrosis factor-α, but not to lipopolysaccharide.[10] Sheu and colleagues showed significantly higher initial serum and urine IL-8 levels in children with acute pyelonephritis.[8] Krzemien and coworkers found significantly higher IL-6 and IL-8 levels in children with febrile UTI and elevated inflammatory markers. They also showed that IL-6 and IL-8 levels do not differentiate acute pyelonephritis from lower UTI at least in children younger than 24 months.[9] On the other hand, Tullus and colleagues recommended that there was no correlation between urine IL-8 and DMSA uptake defect in patients with pyelonephritis.[10] Findings of these studies show that there is no consensus on the role of interleukins in upper and lower UTIs. Thus, we still have to further study on the diagnostic and differentiating values of IL-8 in children with UTI.

We did not have any conclusive results about the effect of treatment on urinary IL-8, either. Kassir and colleagues' study revealed that the urinary tract cytokine response to infection is intense, but disappears shortly after the initiation of treatment with antibiotics. This suggests that kidney damage due to inflammation begins early in the course of infection, underscoring the need for rapid diagnosis and intervention.[11] Sharifian and associates evaluated the urinary cytokines including urinary IL-8 in pyelonephritic patients before and after the treatment and concluded that antibiotics combined with dexamethasone significantly decreased urinary IL-8 level in patients with pyelonephritis.[12]

In our study, the level of urinary IL-8 was higher in children with pyelonephritis, but we found an increasing level of this cytokine after appropriate treatment of pyelonephritis. In order to explain the significant difference between the pretreatment and

posttreatment cytokine levels, we sought the other potential reasons for elevation of urinary cytokines. The treatment course and antibiotic therapy might be the cause of elevated urinary cytokine levels in our patients. Prins and associates revealed that serum and urine IL-8 levels increased up to 10% to 40% after 4 hours of treatment with ceftazidime, while they did not observed any increase in patients treated with imipenem.[13] On the other hand, Kumar and coworkers showed that after instillation of bacillus Calmette-Guerin, the mean IL-8 levels elevated in responders to this treatment of bladder cancer but not in nonresponders.[14] Results of this study recommend that the level of urinary interleukin-8 can be increasing during the treatment of different diseases such as infections or malignancies. Therefore, the increase in the level of urinary IL-8 in our patients might be due to previous or current antibiotic therapy and the effect of antibiotics on the renal tubules; however, researches in different situations with larger sample sizes would be able to confirm this hypothesis.

CONCLUSIONS

We conclude that urinary IL-8 is increased in pyelonephritis especially after the treatment. We also found that there is no correlation between the level of urinary IL-8 and the other parameters in children. Thus, urinary IL-8 may have the potential to be a noninvasive test for diagnosis of UTI and response to treatment.

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CONFLICT OF INTEREST

None declared.

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