Urinary N-Acetyl-Beta-D-Glucosaminidase as a Diagnostic Marker of Acute Pyelonephritis in Children

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Introduction. Prompt diagnosis and localization of pyelonephritis are of great importance in children. The urinary excretion of enzymes, and in particular N-acetyl-beta-D-glucosaminidase (NAG), is considered a simple noninvasive marker for detection of renal tubular dysfunction due to pyelonephritis. This study was performed to determine the diagnostic value of urinary NAG in acute pyelonephritis.

Materials and Methods. In a quasi-experimental study conducted on 72 children with confirmed pyelonephritis, we measured urinary NAG, creatinine, and NAG-creatinine ratio before and after the treatment. Diagnostic values of these parameters were evaluated by considering the patients before and after the treatment as disease-positive and disease-negative groups, respectively.

Results. The patients were 18 boys (25.0%) and 54 girls (75.0%) with a mean age of 43.0 ± 39.0 months. The mean levels of urinary NAG were 12.20 ± 6.14 U/L and 5.46 ± 7.98 U/L before and after the treatment, respectively ($P < .001$). The sensitivity and specificity of urinary NAG-creatinine ratio for diagnosis of pyelonephritis were 73.6% and 77.3%, respectively, with a cutoff point of 10.16 U/g (area under the curve = 0.76, 95% confidence interval, 0.67 to 0.76). Significantly higher levels of urinary NAG were found in those who had a negative urine culture at diagnosis (8.8 ± 10.4 U/L) compared to those with a positive urine culture (4.5 ± 8.7 U/L).

Conclusions. We concluded that urinary NAG is elevated in children with pyelonephritis and it can be considered as a further criterion in the diagnosis of upper urinary tract infection.

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INTRODUCTION

Urinary tract infection (UTI) is a common clinical problem in infancy and childhood. Prompt diagnosis of the infection and the localization of its level are of great importance in determining the duration of treatment and the appropriate investigation and prognosis of the patients. In the past decade, dimercaptosuccinic acid (DMSA) scintigraphy has been considered an objective method for the localization of the UTI site.[1,2] It is a useful method, but it is invasive and there are some concerns about the accuracy of DMSA renography in pediatric group, especially in infancy.[3] Therefore, physicians prefer to use a noninvasive method for diagnosis of pyelonephritis in children.

The inflammatory markers, which have primarily been used as indexes for the diagnosis of pyelonephritis are fever, leukocytes, blood neutrophil count, C-reactive protein (CRP), erythrocyte sedimentation rate (ESR), procalcitonin,

interleukins, N-acetyl-beta-glucosaminidase (NAG), and beta 2-microglobulin. Several researchers have tried to correlate these parameters with the UTI site, but the results have been rather conflicting.[4,5] The urinary excretion of enzymes, and in particular NAG, is considered a relatively simple fast noninvasive method in the detection and follow-up of renal tubular function under various conditions. The determination of urinary NAG provides an indicator of damages to the kidney, such as injury or dysfunction due to inflammation and nephrotoxic drugs.[6-8] N-acetyl-beta-glucosaminidase is a lysosomal enzyme present in the proximal convoluted tubule that may be used as a marker of proximal tubular damage.[9] It can be checked by noninvasive techniques and it can be used in a clinical setting to detect functional disorders of the kidney.[10] Increased urinary excretion of NAG has been shown in urinary tract infections. Hence, NAG has the potential to be considered as an additional marker in diagnosis of pyelonephritis and interstitial tubular damage.[11] This study was performed to determine the diagnostic value of urinary NAG in acute pyelonephritis and to compare it with other indexes conventionally used for this purpose in children.

MATERIALS AND METHODS

In a Quasi-experimental before-after study conducted from April 2005 to May 2006, we enrolled children who were admitted to Mofid Children's Hospital due to pyelonephritis. The ethics committees of Shaheed Beheshti University of Medical Sciences and Pediatric Infectious Research Center approved this study and parents of the children provided informed consent. Diagnosis of pyelonephritis was based on clinical manifestations (fever, abdominal pain, anorexia, vomiting, frequency, and dysuria) and paraclinical findings (leukocyturia, positive urine culture for bacterial growth, increased ESR, positive CRP, leukocytosis, and abnormal findings on DMSA renography).

Seventy-two pediatric patients were recruited by a sampling method of census. All of the patients had been previously healthy with no medical or drug history or signs of any kidney diseases. Glomerular filtration rate was calculated according to Schwartz formula[12] and was in normal range in all of them. Fresh random urine samples were obtained on the admission time and at the 48th hour of treatment. Urine samples were tested for NAG (enzyme-linked immunosorbent assay colorimetric, Diazyme Laboratories, La Jolla, California, USA) and creatinine (Jaffe reaction, auto-analyzer, RA 1000) levels. The patients were treated with a same treatment protocol (intravenous ceftriaxone, 75 mg/kg, for ten days). We also evaluated our patients with DMSA renography (at the 3rd day of admission), voiding cystoureterography (at the end of the treatment period), and biochemical studies.

After complete treatment, the patients were considered as the disease-free group. The findings before and after the treatment were compared in the patients using the Wilcoxon signed rank test. The Kruskal-Wallis test, 1-way analysis of variance, and post hoc analysis were used for comparisons and evaluation of correlations between groups. The receiver operating characteristic curve as used to define the diagnostic value and the best cutoff points of each urinary parameter. Continuous variables were expressed as mean ± standard deviation. Statistical tests were two-tailed and the results were considered significant if a *P* value less than .05 would be achieved. The SPSS software (Statistical Package for the Social Sciences, version 11.5, SPSS Inc, Chicago, Ill, USA) was used for these analyses.

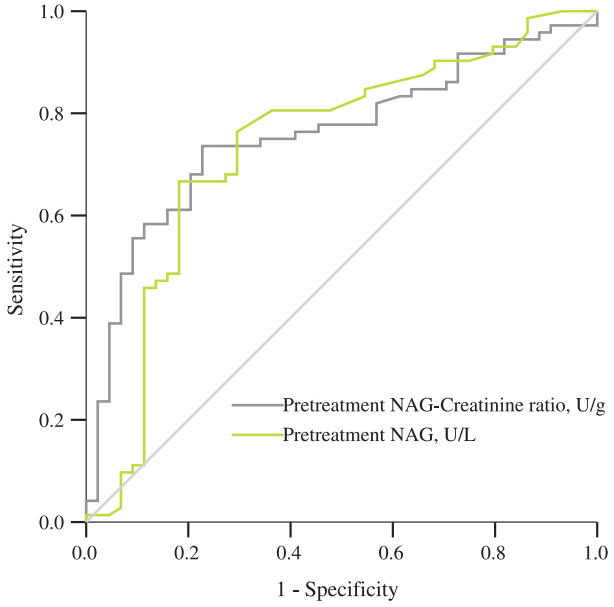
RESULTS

Seventy-two children with pyelonephritis were evaluated. They were 18 boys (25.0%) and 54 girls (75.0%) with a mean age of 43.0 ± 39.0 months. The mean levels of urinary creatinine, NAG and NAG-creatinine ratio are shown in the Table. There was a significant decrease in the posttreatment

Pretreatment and Posttreatment Levels of Urinary Creatinine and NAG in Children With Pyelonephritis*

Urinary Parameter	Pretreatment Value	Posttreatment Value	P
Creatinine, g/L	0.62 ± 0.69	0.77 ± 0.82	.37
NAG, U/L	12.20 ± 6.14	5.46 ± 7.98	< .001
NAG/Creatinine, U/g	46.83 ± 47.96	11.90 ± 24.43	< .001

*NAG indicates N-acetyl-beta-glucosaminidase.



The receiver operating characteristic curves for urinary NAG and urinary NAG-creatinine ratio for diagnosis of pyelonephritis. The area under the curve was 0.735 for urinary NAG and 0.758 for urinary NAG-creatinine ratio. NAG indicates N-acetyl-beta-glucosaminidase.

urinary ratio of NAG to creatinine compared to the pretreatment level ($P < .001$).

We considered the patients before and after the treatment as disease-positive and disease-negative groups, respectively. Based on the receiver operating characteristic curves, the sensitivity and specificity of urinary NAG-creatinine ratio for diagnosis of pyelonephritis were 73.6% and 77.3%, respectively, with a cutoff point of 10.16 U/g (area under the curve = 0.76, 95% confidence interval, 0.67 to 0.76; Figure). Significantly higher levels of urinary NAG were found in those who had a negative urine culture at diagnosis (8.8 ± 10.4 U/L) compared to those with a positive urine culture (4.5 ± 8.7 U/L). No significant correlation was seen between urinary NAG and the patients' sex, age, body weight, and the levels of ESR, CRP, and urinary or blood leukocytes counts. In order to evaluate whether there is a correlation between vesicoureteral reflux or DMSA changes and the level of urinary NAG, we used Kruskal-Wallis test, 1-way analysis of variance, and post hoc analysis; we found no correlation between the urinary NAG level and the above other variants.

DISCUSSION

The prompt diagnosis of pyelonephritis is of great importance for appropriate management of the disease. Variable markers have been used for this purpose, but none of them has been considered adequately sensitive and specific for such a diagnosis and differentiation between upper and lower urinary tract infections.[13] Previous studies have documented that urinary excretion of NAG is significantly greater in patients with upper UTI than in patients with lower UTI in adults and children.[9,14] In a study by Cottone and colleagues, it was noted that by the end of target chemo-antibiotic treatment, all patients presented a reduction in the inflammation, urinary NAG level, and hence, the tubular distress.[15] Belli and associates showed that urinary NAG levels were elevated in children with pyelonephritis in the presence or absence of urinary tract abnormality.[10] In contrast to previous studies that demonstrated urinary NAG as a good indicator of pyelonephritis, Johnson and coworkers reported that this marker is of no value in localizing the site of UTI, and an NAG level within 1 standard deviation of the mean in a child with cystitis indicates a low risk of urologic abnormalities.[16] In addition, Zachariah evaluated urinary NAG level in patients with spinal cord injury and demonstrated that there was a nonselective increase in urinary NAG excretion in the control group and those with lower UTI; however, in patients with upper UTI, there was a selective increase in NAG isoenzyme B excretion.[17] The overlap in enzyme values between the different groups suggests that the test may not be clinically useful in localizing the site of UTI.[17] Based on our knowledge, pyelonephritis involves a high risk of kidney damage, especially in the proximal tubule, due to interstitial nephritis; therefore, NAG excretion should be higher in children with upper UTI than in those with lower UTI or in healthy children. Nonetheless, varying findings of these studies put forward a question on the reliability of NAG in practice.

Because of these conflicting results, we decided to investigate the possible value of urinary NAG in the diagnosis of pyelonephritis. Our data showed that urinary NAG is increased in the acute phase of pyelonephritis and it decreases following appropriate treatment with antibiotics. Our analysis showed the sensitivity and specificity of urinary NAG in diagnosis of pyelonephritis to be 723.6% and 77.3%, respectively. This is in agreement with

the other published data such as those Jantausch and colleagues who reported that the sensitivity and specificity of NAG in predicting UTI in febrile patients, regardless of the site of infection, were 88% and 88%, respectively.[5]

The urine of patients with pyelonephritis often has alkaline pH caused by urease-producing bacterial species. The effect of urine pH on urinary NAG isoenzymes have been reviewed and shown that measurement of NAG isoenzyme B would be more reliable than that of total NAG in alkaline urine, and the increase of NAG isoenzyme B in pyelonephritis might be due to the instability of NAG isoenzyme A in alkaline urine.[18,19]

We also aimed to identify correlation between urinary NAG and the other paraclinical variables and found that the level of urinary NAG was higher in patients with a negative urine culture in comparison with those whose urine cultures were positive. We suppose that may be the chemical effects of urinary bacterial organisms that induce some changes in the level and activity of this marker, but we did not find any confirmation for our idea.

Although, no significant correlation was seen between the level of urinary NAG and ESR, CRP, blood or urinary leukocytes, and voiding cystoureterography or DMSA scan findings in our study, Tomlinson and coworkers showed that elevated NAG levels were seen mostly in the 65 of 93 children with bilateral scarring and severe vesicoureteral reflux.[20] Carr and colleagues demonstrated that urinary NAG levels were elevated with higher grades of reflux, concluding that this relatively simple assay may have clinical usefulness in the assessment of tubular dysfunction associated with reflux.[21] In contrast to these studies, but in agreement with ours, Linne and colleagues reported that a substantial number of infants with pyelonephritis appeared to have normal scans with abnormal urinary NAG; therefore, they supposed that early determination of the urinary NAG might further improve the diagnostic approaches in this age group.[3] It is difficult to make direct comparison between their study and the other studies, and it is unclear why the results of the studies on correlation between the level of this marker and DMSA or voiding cystoureterography changes have been varied.

CONCLUSIONS

We conclude that urinary NAG may be considered as a further criterion in the diagnosis of upper UTI, although studies with greater numbers of patients are needed.

CONFLICT OF INTEREST

None declared.

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