Urine neutrophil gelatinase-associated lipocalin identifies unilateral and bilateral urinary tract obstruction

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Abstract

Background. Urinary tract obstruction (UTO) is a common problem that can lead to permanent loss of kidney function. Unilateral UTO may be difficult to diagnose. Urinary neutrophil gelatinase-associated Lipocalin (uNGAL) may identify unilateral and bilateral UTO.

Methods. Retrospective case–control study of patients undergoing hospital admission at three sites. UTO was determined by review of medical records and cases were matched to control patients. uNGAL was measured by immunoblot.

Results. Twenty-four unilateral UTO and 15 bilateral UTO cases were identified. Admission serum creatinine (sCr) (milligram per decilitre) was significantly higher in bilateral UTO, 2.0 (1.1–5.3), but not unilateral UTO, 1.1 (0.8–1.5), compared to controls, 0.9 (0.8–1.2). uNGAL (nanogram per millilitre) was significantly higher both in patients with bilateral UTO, 140 (40–450), and unilateral UTO, 50 (20–100), compared to controls, 20 (10–45).

Discussion. uNGAL identifies kidney injury in unilateral and bilateral UTO even in the absence of an elevated sCr.

Keywords: acute kidney injury; biomarkers; neutrophil gelatinase-associated lipocalin; obstructive nephropathy; urinary tract obstruction

Introduction

Urinary tract obstruction (UTO) is defined as a structural or functional interference of urine flow in any location along the urinary tract. Approximately 400 000 hospitalizations due to UTO occur annually in the USA alone [1]; if left untreated, UTO results in progressive irreversible loss of kidney function [2]. Although bilateral UTO is classically detected by an elevated serum creatinine (sCr) and decreased urine output, unilateral UTO may be more difficult to diagnose; sCr and urine output may be unremarkable while only imaging may detect an abnormality. Unfortunately, kidney imaging is unlikely to occur without a clinical indication; therefore, unilateral UTO may go undetected for prolonged periods, increasing the likelihood of permanently decreased kidney function.

Novel methods to detect subclinical kidney injury associated unilateral UTO may permit its early identification and treatment. Neutrophil gelatinase-associated lipocalin (NGAL) is a 23–27 kDa protein. Its expression is upregulated by renal tubular epithelia in response to injury [3–6]. Our group previously demonstrated that urine NGAL (uNGAL) distinguished acute kidney injury (AKI) from prerenal azotemia, stable chronic kidney disease and normal kidney function [2]. We also noted that UTO generated the highest uNGAL values among all patients with AKI (T. Nickolas, unpublished data). We hypothesized that uNGAL was induced by pathogenic mechanisms producing UTO and may detect both unilateral and bilateral UTO, even in the presence of an unremarkable sCr.

Materials and methods

Patient enrollment and sample collection

This retrospective case–control study was performed on adults undergoing admission to The Allen Hospital of New York Presbyterian Hospital, Staten Island University Hospital, New York, and the Helios Clinic, Charité—Universitätsmedizin Berlin, Berlin, Germany, between June 2008 and December 2008. Informed consent was obtained. The first urine sample obtained in the emergency department (ED) was collected for uNGAL measurement.

Diagnosis of UTO and matching of controls

UTO was identified by review of chief complaint (flank pain, dysuria, cystalgia and decreased urine output) and by review of admission and discharge diagnoses. Unilateral UTO was defined as obstruction of one ureter with radiographic evidence of hydronephrosis or hydroureter. Bilateral UTO was defined as obstruction occurring at either the level of the bladder or urethra with retention of >500 mL of urine, with or without hydronephrosis on imaging. Controls were selected from subjects with stable kidney function with a wide variety of chief complaints and were age-, sex-, race-, baseline sCr- and urinary tract infection (UTI)-matched to cases. Baseline sCr was determined by review of the prior 12 months of records, or if unavailable, baseline sCr was assumed from the lowest recorded sCr during the hospital course.
Table 1. Patient characteristics, sCr and uNGAL levels, by diagnostic group\textsuperscript{a,b}

<table>
<thead>
<tr>
<th></th>
<th>Controls ((n = 39))</th>
<th>Unilateral obstruction ((n = 24))</th>
<th>Bilateral obstruction ((n = 15))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean age (years)\textsuperscript{a}</td>
<td>57.2 (20.5)</td>
<td>50.0 (20.2)</td>
<td>71.0 (15.1)\textsuperscript{a}</td>
</tr>
<tr>
<td>Female gender (%)</td>
<td>38.5</td>
<td>58.3</td>
<td>6.6</td>
</tr>
<tr>
<td>Non-Hispanic Black (%)</td>
<td>10.3</td>
<td>8.3</td>
<td>13.5</td>
</tr>
<tr>
<td>Non-Hispanic White (%)</td>
<td>61.5</td>
<td>62.5</td>
<td>66.7</td>
</tr>
<tr>
<td>Mean systolic blood pressure (mmHg)\textsuperscript{a}</td>
<td>137 (28)</td>
<td>140 (23)</td>
<td>138 (28)</td>
</tr>
<tr>
<td>Mean diastolic blood pressure (mmHg)\textsuperscript{a}</td>
<td>79 (17)</td>
<td>77 (13)</td>
<td>80 (18)</td>
</tr>
<tr>
<td>Mean hematocrit\textsuperscript{a}</td>
<td>37.9 (4.8)</td>
<td>38.7 (4.5)</td>
<td>34.4 (4.8)\textsuperscript{a,b}</td>
</tr>
<tr>
<td>Mean serum WBC count (1000 cells/mm\textsuperscript{3})\textsuperscript{a}</td>
<td>10.2 (5.3)</td>
<td>10.6 (3.6)</td>
<td>12.8 (5.9)</td>
</tr>
<tr>
<td>Urinary tract infection (%)</td>
<td>30.8</td>
<td>29.2</td>
<td>33</td>
</tr>
<tr>
<td>Mean length of stay (days)\textsuperscript{a}</td>
<td>6.0 (5.7)</td>
<td>3.9 (2.7)</td>
<td>9.7 (7.7)\textsuperscript{a,b,c}</td>
</tr>
<tr>
<td>Median baseline GFR (mL/min/1.73m\textsuperscript{2})\textsuperscript{b}</td>
<td>84 (61–107)</td>
<td>98 (71–114)</td>
<td>75 (39–99)</td>
</tr>
<tr>
<td>Median baseline sCr (mg/dL)\textsuperscript{b}</td>
<td>0.9 (0.7–1.2)</td>
<td>0.8 (0.7–1.1)</td>
<td>1.0 (0.8–1.8)</td>
</tr>
<tr>
<td>Median admission sCr (mg/dL)\textsuperscript{b}</td>
<td>0.9 (0.8–1.2)</td>
<td>1.1 (0.8–1.5)</td>
<td>2.0 (1.1–5.3)\textsuperscript{a,b}</td>
</tr>
<tr>
<td>Median presenting uNGAL (ng/mL)\textsuperscript{b}</td>
<td>20 (10–45)</td>
<td>50 (20–100)</td>
<td>140 (40–450)</td>
</tr>
</tbody>
</table>

\textsuperscript{a}Data presented as mean (SD), median (Interquartile range) or frequency as indicated.

\textsuperscript{b}Data compared with Mann–Whitney U-test.

\textsuperscript{c}Data compared with Mann–Whitney or Kruskal–Wallis test if non-normally distributed. Normally distributed variables were presented as mean (SD).

\textsuperscript{d}\(p < 0.05\) compared to controls.

\textsuperscript{e}\(p < 0.01\) compared to controls.

\textsuperscript{f}\(p < 0.01\) compared to unilateral obstruction.

Laboratory measurements

Urine sample was centrifuged at 12,000 r.p.m (11,752 g) for 10 min. Supernatants were stored at \(-80^\circ\text{C}\). uNGAL (10 μL) was batch assayed by immunobLOTS with non-reducing 4–20% gradient polyacrylamide gels (Bio-Rad Laboratories, Hercules, CA) and monoclonal antibodies (Bioporte Diagnostics, Gentofte, Denmark) or rabbit polyclonal antibodies together with standards (0.2–10 ng) of human recombinant NGAL protein to authenticate monomeric NGAL.

Statistical analysis

Statistical analyses were conducted with SPSS (v16.0; Chicago, IL) and SAS (v9.1; Cary, NC). For categorical parameters, group differences were compared by chi-square tests with a \(\alpha = 0.05\). For continuous parameters, group differences were compared by analysis of variance if normally distributed or Mann–Whitney U-test if non-normally distributed. Normally distributed variables were presented as mean (SD).

Role of the funding source

Abbott Diagnostics provided funding for data collection and analysis. Abbott did not contribute to the study design, execution, analysis or manuscript preparation. The authors had access to all data and took final responsibility for the study’s content and conclusions.

Results

Baseline characteristics

We enrolled 1635 patients admitted from the ED. Thirty-nine patients with UTO (24 unilateral and 15 bilateral) were identified and age-, sex-, race-, baseline glomerular filtration rate (GFR)- and UTI-matched to 39 controls (Table 1). 40% were female and 70% were Caucasian. Mean age was 57 \(\pm\) 20 years. 30% had UTI. Primary UTO etiologies were nephrolithiasis (51%), prostatic hypertrophy (26%), malignancy (5%), neurogenic bladder (8%) and other (10%).

UTO and kidney injury biomarkers

In comparison to controls, admission sCr was significantly higher in patients with bilateral UTO but did not differ between controls and patients with unilateral UTO (Table 1).

In contrast, uNGAL values were significantly higher in both patients with unilateral and bilateral UTO in comparison to controls. Figure 1 shows the box plots for admission sCr and uNGAL; there was less overlap in uNGAL levels between controls and patients with unilateral UTO.

Discussion

These data suggest that uNGAL is elevated in UTO. These are the first data we know of reporting that unilateral UTO in adults may be detected by uNGAL measurement, even when admission sCr is unremarkable.

NGAL is rapidly expressed in the urine of patients with AKI [4, 6–11]. Our data suggest that uNGAL detects AKI in patients with either unilateral UTO or bilateral UTO, even at a sub-clinical stage when GFR has not been affected by UTO-associated tubular injury. This is consistent with a recent study in children with severe hydrenephrosis due to congenital obstructive nephropathy [12]. Our data expand upon those findings by identifying elevated uNGAL levels with unilateral UTO. Furthermore, our data highlights advantages of a direct biomarker of renal tubular injury rather than a surrogate of GFR. Limitations of our study include the small sample size and we therefore were unable to evaluate relationships between uNGAL and severity of UTO. Our enrollment algorithm did not allow us to determine the duration of UTO. Although kidney biopsy is the gold standard to define AKI, it is not used to diagnose UTO; we utilized standard imaging and clinical criteria. Finally, this study is subject to limitations due to its case–control design; a prospective study of consecutive patients is needed.

In summary, these data suggest that uNGAL identifies kidney injury in unilateral and bilateral UTO even in the absence of an elevated admission sCr. uNGAL testing may be particularly useful among populations of patients at risk for unilateral obstruction, such as patients with a history of nephrolithiasis, patients with intermittent obstruction, such as polycystic kidney disease.
as men with prostatic hypertrophy, or among patients receiving medications that predispose the development of nephrolithiasis, such as chemotherapeutic medications. It may also be used to follow patients with asymptomatic stone disease; a positive uNGAL might provide compelling evidence for renal imaging to assess for progression to obstruction. Larger studies are needed to determine the timing of uNGAL elevation if levels decrease after alleviation of UTO and if detection and treatment of UTO by uNGAL measurement decreases UTO-associated morbidity, particularly for patients with unilateral obstruction.

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Conflict of interest statement. Two authors, J.M.B. and T.L.N., hold patent applications relevant to the data presented. Relevant patents are US20100233739, US20070037232 and US 20100047837. T.L.N. has a consultancy agreement with Abbott Diagnostics. Abbott did not contribute
to study design, execution, analysis or manuscript preparation. The authors had access to all data and take final responsibility for the study’s content and conclusions.

**References**


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