Role of urinary NGAL and microalbuminuria in the detection of subclinical acute kidney injury in pediatric intensive care unit and diabetic children

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Abstract

Subclinical Acute Kidney Injury (AKI) describes patients who did not fulfill the classical criteria for AKI diagnosis but showed elevated levels of new biomarkers reflecting tubular injury. One of these biomarkers is Neutrophil Gelatinase-Associated Lipocalin (NGAL). The aim of this study is to investigate the role of urinary NGAL and microalbuminuria as non-invasive biomarkers in the detection of subclinical AKI. Analysis of urinary NGAL and microalbuminuria in 91 subjects [30 pediatric intensive care unit (PICU) patients, 31 diabetic patients and 30 healthy controls] recruited from Cairo University Pediatric Hospital was done. Our study revealed that urinary NGAL was significantly higher in the PICU group followed by the diabetic group and lowest in the controls group (p=0.022). A positive correlation was found between urinary NGAL and microalbuminuria in the PICU group (R-value=0.585, p-value=0.001). In diabetic group, a positive correlation was found between urinary NGAL and fasting blood glucose, 2 hours post prandial and HbA1C (R-value=0.421; p-value=0.021; R-value=0.426; p-value=0.019; R-value=0.438; p-value=0.018 respectively). Urinary NGAL may be a potential biomarker to detect subclinical AKI before actual functional renal damage leading to early intervention and reduction of mortality.

Introduction

The expression Acute Kidney Injury (AKI) has replaced the expression acute renal failure, and new definitions have been suggested to allow earlier detection. AKI is a complex and heterogeneous process associated with significant mortality and morbidity. In critically ill patients admitted to the Intensive Care Units (ICU), the prevalence of AKI reaches up to 60%. It occurs in 10-20% of patients in the Pediatric Intensive Care Unit (PICU). AKI in ICU is associated with an increased hospital stay, initiation of dialysis and increased mortality. Early detection of AKI is significant, as early intervention and regime modification can prevent further kidney injury and improves renal outcome.[1]

The Acute Dialysis Quality Initiative created the RIFLE classification (Risk, Injury, Failure, Loss, and End-stage Kidney) for the definition AKI, which provides three grades of severity based on changes in serum creatinine or Urine Output (UOP). This led to the classification of patients with AKI into one of the following classes: risk (class R), injury (class I) and failure (class F) and two outcome classes (loss and end-stage kidney disease).[2]

However, this classification criteria still relies on imperfect

parameters such as serum creatinine and UOP, which indicate functional damage, and does not incorporate parameters that detect structural damage.[3] Serum creatinine concentrations may not change until 25–50% of the kidney function has already been lost; hence, it may take days after an injury before a significant rise in serum creatinine is detected. At a lower Glomerular Filtration Rate (GFR), serum creatinine will overestimate renal function due to tubular secretion of creatinine. Levels of serum creatinine vary according to muscle mass, hydration status, age, and gender as well as method of measurement.[1]

The limitations of serum creatinine and UOP could not be ignored and research for new biomarkers possibly reflecting tubular structural damage was pursued. The Acute Dialysis Quality Initiative proposed a diagnostic approach to AKI incorporating kidney damage markers as well as kidney function markers (serum creatinine and UOP). Subclinical AKI is the term given to the subgroup of AKI patients who did not fulfill the classically used criteria for AKI diagnosis, however, they showed elevated levels of new biomarkers that reflect tubular injury.[4] Most of these biomarkers are urinary proteins upregulated in renal tubule cells related to cell (or tissue) injury, in contrast to serum creatinine which rises due to renal function changes. Therefore, they have been referred to as "structural" AKI biomarkers.[5] One of the subclinical or structural AKI biomarkers is Neutrophil Gelatinase-Associated Lipocalin (NGAL). NGAL, also named lipid-transport protein 2 (lipocalin-2), is a member of the lipocalin superfamily. It is expressed in abundance in neutrophils and monocytes/macrophages and is a mediator of the innate immune response. NGAL is extruded into the urine with AKI, involved in injury and repair of renal tubule cells.[1] Studies have presented the diagnostic and prognostic usage of both urinary and plasma NGAL in patients with subclinical AKI.[3]

NGAL can be derived from cardiac tissue, neutrophils as well as kidneys, which makes the plasma level of NGAL less specific than the urinary NGAL as a predictive tool of AKI. A meta-analysis was performed to assess the diagnostic value of urine, serum and plasma NGAL for the early diagnosis of AKI and found that urinary NGAL presented a superior performance for the diagnosis of AKI with the highest AUC and other diagnostic accuracy values, compared with serum and plasma NGAL. To detect NGAL level in newborn infants; urinary sample is non-invasive, and it avoids the difficulties and complications of withdrawing a blood sample from newborns, which may lead to a hematoma, bleeding, or infection.[6]

In critically ill patients without pre-existing kidney disease, both plasma NGAL and urinary NGAL measured at admission could predict AKI occurrence in the first 72 hours post-ICU admission. Shoaib et al. reported that urinary NGAL was an accurate marker of AKI in critically ill patients, and thus should be included in the diagnostic workup of AKI in its early stages.[1] Urinary NGAL was also suggested as a reliable biomarker of renal function in diabetic patients.[7]

Furthermore, AKI biomarkers such as NGAL, interleukin-18 (IL-18) and Liver-Type Fatty Acid-Binding Protein (LFABP) have shown promising results for early AKI diagnosis in children undergoing cardiac surgery. Only a few AKI biomarkers studies have been published in the non-cardiac surgery PICU. Validating subclinical AKI biomarkers may help improve AKI management.[5]

From another perspective, albuminuria is recognized as one the most important risk factors for progression of kidney diseases. Microalbuminuria is defined as 30–300 mg/day of albumin excretion in urine. It occurs rapidly after an acute inflammatory incident, persisting in patients with complications. It is common in critically ill patients, where it has shown promise as a predictor of organ failure, vasopressor requirement and mortality.[8] Microalbuminuria was also investigated as a biomarker of renal dysfunction in diabetic patients. The presence of microalbuminuria can be an important marker for cardiovascular and renal risk in diabetes mellitus and is recognized as a risk marker for kidney disease and its complications in humans.[9]

The aim of this study is to investigate the role of urinary NGAL and microalbuminuria levels as non-invasive biomarkers in the detection of subclinical AKI in PICU and diabetic children. It also explores the relation between urinary NGAL and microalbuminuria in these patients.

Materials and Methods

Patients' population

This study is an observational prospective case-control study. Ninety-one subjects were recruited from Cairo University Pediatric Hospital (CUPH). Thirty patients were selected from the PICU and 31 patients from the diabetes outpatient clinic. Thirty healthy control subjects were recruited from different outpatient clinics at CUPH.

Patients aged 1 month to 16 years were included in the study. In the PICU group, we excluded patients with baseline estimated Glomerular Filtration Rate (eGFR) <60 ml/min/1.73m^2; post-renal transplantation or cardiac surgery cases, those with primary renal disease (e.g., nephritis) and less than 2 days stay in PICU. As for the diabetic and controls group, patients with cardiac or acute renal complications were excluded.

The study was performed in line with the principles of the Declaration of Helsinki.[10] Approval was granted by the departmental research ethical committee in the Faculty of Medicine, Cairo University. All legal guardians of participants signed a written informed consent.

Data collection

For both patients' groups, demographic data, anthropometric data, cause of admission to PICU, duration of stay, outcome, comorbidities, family history of renal diseases and laboratory tests results were collected. Pediatric Risk of Mortality (PRISM) score, risk factors (in 1st 3 days; including nephrotoxic drugs, fluid balance, sepsis, etc.) were recorded. Abdominal ultrasound and Risk, Injury, Failure, Loss of kidney function, and End-stage kidney disease (RIFLE) staging were also incorporated. Additionally, follow up and Albumin/Creatinine Ratio (ACR) data were collected for the diabetic patients' group.

Sample collection, measurement of urinary NGAL and microalbuminuria

Urine samples were collected from all study participants (after 48 hours of admission for PICU group) directly into a sterile container (mid-stream first urine of the day), centrifuged and stored at -20°C until analysis time.

The analysis of urinary NGAL level was performed by Magnetic Luminex® Performance Assay Human Lipocalin-2/NGAL Kit (R&D Systems, Inc., USA, Cat. No. LHK1757) using LABScan3D Advanced Multiplex Analyzer (One Lambda, Inc., U.S.A. Catalogue number: LABSCNXS4) based on Luminex® xMAP® technology and use Luminex® xPONENT® software for data acquisition.

Microalbuminuria was analysed using Cobas 6000 analyser (photometric unit of c501 module; Roche Diagnostics International Ltd, Switzerland).

Statistical analysis

Microsoft excel 2016 was used for data entry and SPSS (version 21) was used for data analysis. All collected data were revised for competences and logical consistency. Data exploration as normal or skewed distribution was done by Kolmogorov–Smirnov/Shapiro–Wilk's test. Simple descriptive statistics in the form of arithmetic mean and standard deviation (median and range) was used for the summary of numerical variables, while frequencies and percentages were used for categorical variables. Bivariate relationship was displayed in cross-tabulations, also comparison of proportions was performed by chi-square and Fisher's exact tests where appropriate. Kruskal Wallis test was performed to assess the association between ICU, diabetic and control groups. A Receiver Operating Characteristic (ROC) curve was used to evaluate the sensitivity and specificity of microalbuminuria and urinary NGAL levels in diagnosing AKI. The univariate relation between microalbuminuria, urinary NGAL and other laboratory parameters in ICU, diabetic and control groups was assessed by Spearman's rank correlation analysis. A p-value less than 0.05 revealed a significant difference between variables.

Results

A total of 91 subjects were included in the study. They were divided into 3 groups: 30 PICU patients (18 males and 12 females, age range 0.16-13 years), 31 diabetic patients (18 males and 13 females, age range 4-16 years), and 30 control subjects (19 males and 11 females, age range 0.16-12 years).

For the PICU patients' group, the commonest causes of admission were pneumonia (26.6%), septic shock (13.3%) and heart failure (10%). About 43.3% took nephrotoxic drugs, 26.7% had negative fluid balance, 80% had sepsis infection, 30% needed mechanical ventilation, 40% needed vasopressor and 90% needed a bladder catheter. RIFLE AKI staging, and abdominal ultrasonography were normal in the whole group, and the outcome was satisfactory in 30% of them (improved and discharged). The median PRISM score was 4 (range 2-8), the median duration of stay (DOS) in PICU was 9.5 days (range 5-21), median albumin (baseline) was 3.55 g/dL (range 3.1-4.2), median albumin (after 48 hours) was 3.95 g/dL (range 3.5-4.2), median UOP after 24 hours was 2 mL/kg (range 0.9-4) and median UOP after 48 hours was 2.45 mL/kg (range 1.6-4).

In the diabetic group, median duration of Diabetes Mellitus (DM) was 4.42 years (range 0.5-8.84), median fasting blood glucose was 110 (range 80-266), median two-hours post-prandial blood glucose was 160 (range 110-330), median HbA1C was 7.85 % (range 5.7-13.3) and median Albumin/Creatinine Ratio (ACR) was 13.25 (range 3-43.6). All patients received short and long-acting insulin, 90.3% had complications, and 6.5% had manifestations of CKD.

Table 1 shows statistically significant differences between baseline creatinine, microalbuminuria and urinary NGAL in the three studied groups. A positive correlation was found between urinary NGAL and microalbuminuria in the PICU group (R value=0.585, p-value=0.001) but not in the diabetic group (Table 2).

Correlations between microalbuminuria, urinary NGAL and laboratory parameters in PICU and diabetic groups are demonstrated in Tables 3 and 4. In diabetic group, a positive correlation was found between microalbuminuria and ACR (R-value=0.766; p-value=0.007). A positive correlation was also found in the diabetic group between urinary NGAL and fasting blood glucose, 2 hours post prandial blood glucose and HbA1C (R-value=0.421; p-value=0.021; R-value=0.426; p-value=0.019; R-value=0.438; p-value=0.018 respectively).

ROC analysis revealed that urinary NGAL was a significant discriminator for subclinical AKI in PICU patients after 48 hours ($p<0.05$), where urinary NGAL showed AUC=0.708 with 95% CI 0.572 – 0.844. The best cut-off point of urinary NGAL was 93.05 pg/mL with 70.0% sensitivity and 66.7% specificity (Figure 1A). When ROC analysis was performed in the diabetic group, it revealed that urinary NGAL was not a significant discriminator for AKI ($p>0.05$), with AUC=0.608 and 95% CI=0.462 – 0.754. The best cut-off point of urinary NGAL was 49.70 pg/mL with 60.0% sensitivity and 43.3% specificity (Figure 1B).

We compared the age and gender of the patients above and below our detected NGAL cut-off values. The median age of patients in the PICU group with an NGAL level less than or equal to the cut-off (93.05 pg/mL) was 10 years (range 4-14 years), 7 of them were males and 5 were females. The median age of the higher cut-off group was 9 years (range 4-16 years), including 11 males and 7 females. Age and gender weren't significantly different between the lower and higher cut-off groups with p values 0.450 and 0.879 respectively.

Regarding the diabetic group, there was also no significant difference in age and gender between the patients with NGAL values less than or equal the cut-off value (49.7 pg/mL) and the group with NGAL above the cut-off value (median age 10, range 7-14 versus 9.5, range 4-16 p 0.381; 8 males and 5 females versus 10 males and 8 females, p 0.739).

Table 1. Comparison between the studied groups for creatinine, microalbuminuria and urinary NGAL.

Group	PICU Median (IQR)	Diabetics Median (IQR)	Controls Median (IQR)	p-value
Creatinine (baseline) (mg/dL)	0.85 (0.6-1.1)	0.55 (0.4-0.7)	0.5 (0.4-0.7)	0.001*
Microalbuminuria (g/mL)	24 (11.25-56.95)	13 (3.9-20)	-	0.011*
Urinary NGAL (pg/mL)	195.5 (48.9-589.15)	93.5 (48.9-318.6)	74.95 (48.9-115.5)	0.022*

*Statistically significant difference (P-value≤0.05).

Table 2. Correlation between urinary NGAL and microalbuminuria in the PICU and diabetic groups.

	Urinary NGAL in PICU		Urinary NGAL in Diabetic	
	R-value	p-value	R-value	p-value
Microalbuminuria	0.585	0.001*	0.285	0.127

Discussion

Acute kidney injury is a well-known serious complication affecting hospitalized patients worldwide. AKI diagnostic criteria does not incorporate parameters that directly indicate tubular damage. This has led to the emergence of subclinical AKI where structural damage has occurred but without functional affection yet. Diagnosing patients at this stage helps in early intervention, reduction of morbidity and mortality and helps in early hospital discharge. Understanding the early stress response of the kidney to acute injuries has revealed several potential new biomarkers.[3,11]

NGAL is a promising biomarker in the urine and plasma for several processes such as early prediction of subclinical AKI and for the early detection and prognosis of AKI. NGAL testing is easy, rapid, measurable and non-invasive in the laboratory with standard clinical platform.[12]

In this study, we measured urinary NGAL and microalbuminuria concentrations in 30 PICU patients, 31 diabetic patients and 30 healthy children as a control group. Our study revealed that urinary NGAL was significantly higher in the PICU group followed by diabetic group and the lowest in controls group (p=0.022).

A prospective study by Nickolas et al. recruited patients treated in an emergency department and assessed the short-term prognostic relevance of biomarker based (subclinical) AKI. They found that urinary NGAL was elevated, whereas serum creatinine level was less than 1.4 mg/dl in 15% of patients. These patients were at increased risk of subsequent Renal Replacement Therapy (RRT) and increased hospital mortality.[13] Another similar study by Di Somma et al. reported that 4.3% of patients were considered to have subclinical AKI (plasma NGAL positive/serum creatinine negative) on admission at the emergency unit. These patients had significantly higher rates of clinical events for the combined endpoint of RRT or in-hospital mortality than the patients who were serum creatinine negative/plasma NGAL negative.[14]

Urinary NGAL excretion has been shown to be increased in many pathological conditions including ischemia, inflammation,

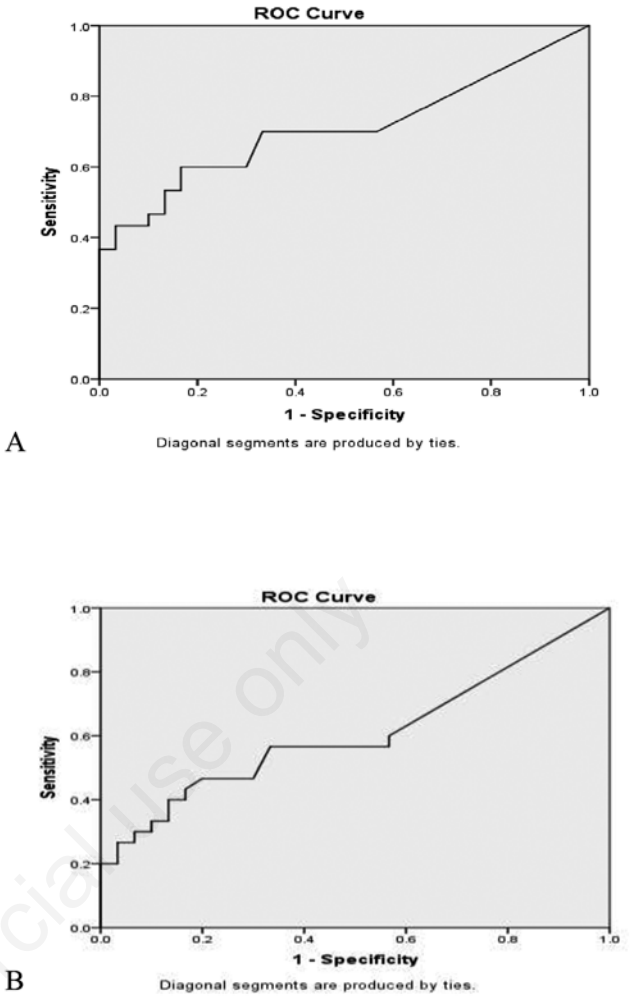

Figure 1. Characteristics of ROC curve to identify the discriminant ability of urinary NGAL to detect subclinical AKI in A) PICU patients and B) in diabetic patients.

Table 3. Correlation between laboratory parameters with microalbuminuria and urinary NGAL in PICU group.

Laboratory parameter	Microalbuminuria		Urinary NGAL	
	R-value	p-value	R-value	p-value
Urea	0.223	0.246	0.238	0.206
Creatinine (baseline)	0.188	0.328	0.157	0.408
Creatinine (after 48 hours)	-0.065	0.739	-0.157	0.406
Albumin (baseline)	-0.083	0.667	-0.221	0.240
Albumin (after 48 hours)	0.066	0.735	-0.104	0.586
UOP (after 24 hours)	-0.159	0.409	-0.302	0.105
UOP (after 48 hours)	0.238	0.213	0.212	0.261

Table 4. Correlation between laboratory parameters with microalbuminuria and urinary NGAL in diabetic group.

Laboratory parameter	Microalbuminuria		Urinary NGAL	
	R-value	p-value	R-value	p-value
Creatinine (baseline)	0.667	0.064	0.147	0.492
Fasting blood glucose	-0.209	0.259	0.421	0.021*
2 hours PP	0.259	0.199	0.426	0.019*
HbA1C	0.284	0.082	0.438	0.018*
Alb/Creatinine ratio	0.766	0.007*	0.284	0.135

cardiac surgery, and renal injury.[15] Nielsen et al. found that urinary NGAL was elevated in Type 1 Diabetic (T1DM) patients with or without albuminuria, demonstrating tubular damage at an early stage.[7] Thus, urinary NGAL level appeared to increase in the very early phase of T1DM before the development of microalbuminuria. Patients with T1DM should be considered to have diabetic kidney injury from the time of diagnosis and preventive interventions should be initiated at an early stage to preclude the progression to end-stage renal disease.[16]

Our groups of patients did not experience AKI according to the RIFLE scoring system in PICU patients and serum creatinine in diabetic patients. Albert et al. study focusing on urinary NGAL data only; showed that 66.8% were classified as having no renal injury (NGAL negative/RIFLE negative), 21.1% as subclinical AKI (NGAL positive/RIFLE negative), 4.5% as isolated functional AKI (NGAL negative/RIFLE positive), and 7.5% as higher risk functional AKI (NGAL positive/RIFLE positive). The subclinical AKI patients group had a 10-fold increase in in-hospital mortality when compared to the group with no renal injury. Biomarker positive group has worse long-term survival than versus biomarker negative group, independent of their RIFLE status.[17]

In agreement with our results, Shoaib et al. showed in their study that the accuracy of urinary NGAL was 90.7% in diagnosing AKI in adult patients admitted to ICU.[1] They had one case where creatinine remained normal or increased by less than 0.3 mg/dL in 48 hours whereas urinary NGAL came out to be positive. They explained this by the fact that serum creatinine increases after more than 50% damage to the kidney has been done. If the renal injury involves less than 50% of the renal parenchyma, then serum creatinine will remain normal, hence this renal injury will be missed by Acute Kidney Injury Network (AKIN) criteria, which utilizes serum creatinine as a tool to diagnose AKI. But in these circumstances, urinary NGAL will be increased as shown in many studies that urinary NGAL as a biomarker of tubular injury and even trivial injuries to the tubular system will result in increased urinary NGAL levels. In another study, there was significant increase in the level of serum NGAL in the critically ill children group compared to control group.[1] Serum creatinine values in critically ill children group showed no statistically significant difference when compared with controls.[18] These findings agree with Bailey et al. who reported that there was a significant difference in serum NGAL between critically ill children and the control group.[19]

Kari et al. evaluated urinary NGAL in critically ill PICU children. None of these children was diagnosed with AKI by RIFLE before entering the PICU. They compared urinary NGAL level at multiple time points with RIFLE classification in diagnosing AKI during their PICU stay. Of these children, 55% developed AKI according to RIFLE criteria. They found that there was a twofold increased risk of incident AKI in those patients with high baseline urinary NGAL at PICU admission.[20] Another study done by Zwiers et al. studied patients with evidence of AKI within the first 48 hours post PICU admission: 69% already had AKI when they entered the ICU and 31% developed AKI later within 48 to 72 hours post-admission. For biomarkers analysis preceding AKI, urinary NGAL was the most sensitive biomarker for prediction of AKI, with sensitivity of 84%, specificity 86%, positive predictive value 70% and its negative predictive value 93%.[21]

The Renal Angina Index is a scoring system that was found to act as a potential biomarker for detecting early AKI. It depends on risk factors (ICU stay, comorbidities and ventilator or vasopressor need) and serum creatinine.[22] The Renal Angina Index was found to predict the risk of developing severe AKI on day 3 only at 12 hours of PICU admission.[5] The combination of Renal Angina Index with biomarkers such as urinary NGAL or L-type Fatty Acid-Binding Protein (L-FABP) was found to improve the predictive performance for AKI.[5,22]

Our ROC analysis revealed that urinary NGAL was a significant discriminator for subclinical AKI in PICU patients after 48 hours ($p<0.05$). Where urinary NGAL showed AUC=0.708 with (95% CI 0.572 – 0.844). The best cut-off value of urinary NGAL was 93.05 ng/mL with 70.0% sensitivity and 66.7% specificity. Similar results were demonstrated by Youssef et al. where ROC curve of urinary NGAL for early detection of AKI showed an AUC of 0.63 with a 95% CI of 0.50–0.77. A a cutoff value of 89.5 ng/mL, urinary NGAL showed sensitivity of 84.6%, specificity of 59.6%, positive predictive value of 36.7%, and negative predictive value of 68.4%.[18] Kari et al. also demonstrated that urinary NGAL levels were highly predictive of AKI (AUC=0.76, 95% CI 0.61–0.92). The cutoff point with the highest correctly classified proportion was 223 ng/mL, which correctly predict 80.0% patients with AKI, with a corresponding sensitivity of 72.7% and a specificity of 89.9%.[20]

In our study, microalbuminuria was statistically significantly higher in PICU group compared to the diabetic group ($p=0.011$). A positive correlation was found between urinary NGAL and microalbuminuria in the PICU group (R value=0.585, p-value=0.001) but not in the diabetic group. Barath et al. concluded that microalbuminuria at 48 hours has predicted AKI in patients. Prolonged ICU stay was directly proportional to high levels of microalbuminuria. Microalbuminuria estimation is a simple and inexpensive test, and it can be used if it helps in the management of sepsis, especially in terms of early prediction of organ failure and requirement of inotropic support.[23]

Papadopoulou-Marketou et al. reported that urinary NGAL has a predictive role for early diabetes nephropathy and probably asymptomatic cardiovascular morbidity, independent of microalbuminuria. New predictive biomarkers can be supplementary to urinary albumin in early diagnosis of T1DM microvascular complications and would help in effective management thus minimizing the rates of severe reno- and cardiovascular as well as all-cause-associated morbidity in young people with T1DM.[24]

In our diabetic group, a positive correlation was found between microalbuminuria and ACR ($r=0.766$; $p=0.007$). Karar et al. observed that microalbuminuria and urine ACR were sensitive and early detectors of early stages of nephropathy, as a positive correlation was seen between microalbuminuria and urine ACR as well as between microalbuminuria and plasma creatinine levels.[25]

Also in our diabetic group, a positive correlation was found between urinary NGAL and fasting blood glucose, 2 hours PP blood glucose and HbA1C ($r=0.421$; $p=0.021$; $r=0.426$; $p=0.019$; $r=0.438$; $p=0.018$), respectively. Kahdem et al. reported that serum NGAL and urinary NGAL have been indicated as marker of AKI and accumulation hyperglycemia and hypertension induce an incipient and specific alteration in the tubular handling of NGAL resulting increased in plasma and urine. So, it is suggested the long duration of diabetes and poor glycemic control lead to increase NGAL in serum and urine.[26]

Studies have suggested that there are two types of subclinical AKI. The first type can be detected in patients who never develop functional AKI with the disappearance of the structural after the exposure to the insult has stopped.[3,27,28] This type is usually seen during the exposure to potentially nephrotoxic substances (aminoglycosides, NSAIDs, contrast media), and the biomarkers should not be considered as only markers of "exposure", reflecting "renal handling of the xenobiotic". This is because the renal handling of many drugs imposes a stress on the tubular cells, which can generate structural damage sufficient to provoke frank nephrotoxic AKI. Some of these biomarkers can thus predict subsequent clinically relevant nephrotoxicity.[29]

The second type of subclinical AKI is more frequent and occurs in the early period, when serum creatinine level is not sufficiently elevated to fulfill the Kidney Disease Improving Global Outcomes (KDIGO) criteria for AKI.[17] The fact that the serum creatinine level is not yet increased does not exclude that the GFR has not declined due to functional renal reserve. The proportion of subclinical AKI patients who develop functional AKI versus those who never develop functional AKI is not established.[3]

The concern about cost-effectiveness of urinary NGAL was addressed and resolved. A cost simulation model was developed by Parikh et al. for clinical algorithms to diagnose AKI based on sCr alone vs. uNGAL plus sCr (uNGAL+sCr). A cost-minimization analysis was performed to determine total expected costs for patients with AKI in two centres.[30] The sensitivity analysis done proved that the superiority of using uNGAL and sCr is contingent on the total hospital costs, length of stay, and costs of additional testing when AKI is suspected. Using uNGAL with sCr as a clinical diagnostic test for AKI may improve patient management and reduce expected costs. Any cost savings would likely result from avoiding delays in diagnosis and treatment and early patient management which may prevent future development of renal complications that may lead to dialysis and the need for transplantation. Although the additional use of uNGAL may not result in cost savings at the beginning, the early detection of AKI with improved patient management and better clinical outcomes lead to economically beneficial results.[30]

NGAL may help to diagnose AKI earlier in the disease state and differentiate different stages of AKI, so proper management to the pediatric patient with AKI will be provided. Following early detection of AKI; several interventions should be applied to help in determining the underlying cause, eliminate further damage to the injured kidney and monitor the response to therapies. Diagnostic evaluation to detect the underlying etiology includes determination of the risk factors for AKI, sepsis, nephrotoxic agents, signs of glomerulonephritis and vasculitis syndrome and the presence of ischemia, bloody diarrhea, volume loss and urine obstruction.

Strategies to prevent renal replacement therapy and support renal function include optimization of hemodynamics via monitoring hemodynamics and urine output when fluid challenge is prescribed and correcting the underlying causes of hypoperfusion to restore GFR. Strategies also include minimizing the exposure to nephrotoxic medications including radiocontrast material and maximizing nutrition. If fluid issues hinder the ability to adequately provide nutrition, renal replacement therapy should be strongly considered. Initiation of goal directed therapy with conservative fluid management after an initial resuscitation is also recommended. Recording cumulative fluid overload at least daily provides a sense of how the patient is handling fluids over the course of the hospitalization. Diuretics and/or ultrafiltration needs to be considered in fluid overloaded patients to optimize the support for the dysfunctional kidney.

Conclusions

The current AKI diagnostic criteria do not incorporate parameters that directly indicate subclinical AKI stage. Potential new biomarkers detecting renal tubular damage such as urinary NGAL helps in early intervention, reduction of morbidity and mortality and helps in early hospital discharge. Urinary NGAL testing is easy, rapid and non-invasive in the laboratory with standard clinical platform.

References

1. Shoaib M, Mahmud SN, Safdar M. Early diagnosis of acute kidney injury by urinary neutrophil gelatinase-associated lipocalin in adult critically ill patients. J Ayub Med Coll Abbottabad 2019;31:12-5.
2. Bellomo R, Ronco C, Kellum JA, et al. Acute renal failure – definition, outcome measures, animal models, fluid therapy and information technology needs: the Second International Consensus Conference of the Acute Dialysis Quality Initiative (ADQI) Group. Crit Care 2004;8:R204–R212.
3. Vanmassenhove J, Van Biesen W, Vanholder R, Lameire N. Subclinical AKI: ready for primetime in clinical practice? J Nephrol 2019;32:9-16.
4. Haase M, Kellum J, Ronco C. Subclinical AKI—an emerging syndrome with important consequences. Nat Rev Nephrol 2012;8:735–9.
5. Roy JP, Devarajan P. Acute kidney injury: Diagnosis and management. Indian J Pediatr 2020;87:600-7.
6. Zhou H, Cui J, Lu Y, et al. Meta-analysis of the diagnostic value of serum, plasma and urine neutrophil gelatinase-associated lipocalin for the detection of acute kidney injury in patients with sepsis. Experiment Therapeutic Med 2021;21:386.
7. Nielsen SE, Schjoedt KJ, Astrup AS, et al. Neutrophil gelatinase-associated lipocalin (ngal) and kidney injury molecule 1 (kim1) in patients with diabetic nephropathy: A cross-sectional study and the effects of lisinopril. Diabet Med 2010;27:1144-50.
8. Basu S, Bhattacharya M, Chatterjee TK, et al. Microalbuminuria: A novel biomarker of sepsis. Indian J Crit Care Med 2010;14:22-8.
9. Chin'ombe N, Msengezi O, Matarira H. Microalbuminuria in patients with chronic kidney disease at Parirenyatwa Hospital in Zimbabwe. Pan Afr Med J 2013;14:39.
10. World Medical Association Declaration of Helsinki. Ethical Principles for Medical Research Involving Human Subjects. JAMA 2013;310:2191-4.
11. Devarajan P. NGAL for the detection of acute kidney injury in the emergency room. Biomark Med 2014;8:217-9.
12. Devarajan P. Biomarkers for the early detection of acute kidney injury. Curr Opin Pediatr 2011;23:194-200.
13. Nickolas TL, Schmidt-Ott KM, Canetta P, et al. Diagnostic and prognostic stratification in the emergency department using urinary biomarkers of nephron damage: a multicenter prospective cohort study. J Am Coll Cardiol 2012;59:246-55.
14. Di Somma S, Magrini L, De Berardinis B, et al. Additive value of blood neutrophil gelatinase-associated lipocalin to clinical judgement in acute kidney injury diagnosis and mortality prediction in patients hospitalized from the emergency department. Crit Care 2013;17:R29.
15. Mishra J, Dent C, Tarabishi R, et al. Neutrophil gelatinase-associated lipocalin (NGAL) as a biomarker for acute renal injury after cardiac surgery. Lancet 2005;365:1231-8.
16. Yuruk Yildirim Z, Nayir A, Yilmaz A, et al. Neutrophil gelatinase-associated lipocalin as an early sign of diabetic kidney injury in children. J Clin Res Pediatr Endocrinol 2015;7:274-9.
17. Albert C, Albert A, Kube J, et al. Urinary biomarkers may provide prognostic information for subclinical acute kidney injury after cardiac surgery. J Thorac Cardiovasc Surg 2018;155:2441-52.e13.
18. Youssef DM, Esh AM, Helmy Hassan E, Ahmed TM. Serum NGAL in critically ill children in ICU from a single center in Egypt. ISRN Nephrol 2013;2013:140905.
19. Bailey D, Phan V, Litalien C, et al. Risk factors of acute renal

failure in critically ill children: A prospective descriptive epidemiological study. Pediatr Crit Care Med 2007;8:29-35.
20. Kari JA , Shalaby MA , Sofyani K, et al. Urinary neutrophil gelatinase-associated lipocalin (NGAL) and serum cystatin C measurements for early diagnosis of acute kidney injury in children admitted to PICU. World J Pediat 2018;14:134-42.
21. Zwiers AJ, de Wildt SN, van Rosmalen J, et al. Urinary neutrophil gelatinase-associated lipocalin identifies critically ill young children with acute kidney injury following intensive care admission: A prospective cohort study. Crit Care 2015;19:181.
22. Matsuura R, Srisawat N, Claure-Del Granado R, et al. Use of the renal angina index in determining acute kidney injury. Kidney Int Rep 2018;3:677-83.
23. Bharath G, Kumar P, Mahendra SV. Microalbuminuria can predict the development of acute kidney injury in intensive care unit admission. Int J Adv Med 2020;7:1372-5.
24. Papadopoulou-Marketou N, Margeli A, Papassotiriou I, et al. NGAL as an early predictive marker of diabetic nephropathy in children and young adults with type 1 diabetes mellitus. J Diabetes Res 2017;2017:7526919.
25. Karar T, Alniwaider RA, Fattah MA, et al. Assessment of microalbuminuria and albumin creatinine ratio in patients with type 2 diabetes mellitus. J Nat Sci Biol Med 2015;6:S89-92.
26. Kahdem AA, Dyab Allawi AA, Nada SZ, Turki KM. Neutrophil gelatinase-associated lipocalin (NGAL) in early detection of nephropathy in type 2 diabetic Iraqi patients. J Fac Med Bagdad 2017;59:74-8.
27. Nehus E, Kaddourah A, Bennett M, et al. Subclinical kidney injury in children receiving nonsteroidal anti-inflammatory drugs after cardiac surgery. J Pediatr 2017;189:175-80.
28. McWilliam SJ, Antoine DJ, Jorgensen AL, et al. Urinary biomarkers of aminoglycoside-induced nephrotoxicity in cystic fibrosis: Kidney injury molecule-1 and neutrophil gelatinase-associated lipocalin. Sci Rep 2018;8:5094.
29. Kane-Gill SL, Smithburger PL, Kashani K, et al. Clinical relevance and predictive value of damage biomarkers of drug-induced kidney injury. Drug Saf 2017;40:1049-74.
30. Parikh A, Rizzo JA, Canetta P, et al. Does NGAL reduce costs? A cost analysis of urine NGAL (uNGAL) & serum creatinine (SCR) for acute kidney injury (AKI) diagnosis. PLoS One 2017;12:e0178091.