
NGAL to predict Acute Kidney Injury – Potential applications and limitations

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Introduction

In a genomewide screening for novel renal biomarkers Neutrophil Gelatinase-associated lipocalin (NGAL) was identified as the gene with highest mRNA and protein levels and with the earliest rise and peak after renal ischemia (1, 2). Renal injury also translated into increased NGAL levels in urine and in plasma. Human NGAL was originally identified as a 25 kDa protein covalently bound to gelatinase from neutrophils (3). From the renal perspective, NGAL (Synonym Lipocalin-2, Siderocalin) is a protease-resistant polypeptide, is released from the distal tubules, secreted with the urine or backleaking to the plasma, freely filtered, reabsorbed in the proximal tubules through endocytotic megalin-receptors or finally secreted with the urine (4).

NGAL is responsible for growth and differentiation of (renal tubular) epithelial cells and exerts bacteriostatic effects in the distal urogenital tract by interference with bacterial siderophore-mediated iron acquisition (4). As siderophore-iron-complex NGAL limits proximal tubular damage and reduces apoptosis (5). NGAL is normally expressed at very low levels in several human tissues, including kidney, lungs, stomach, and colon (6). However, kidney epithelia express and excrete massive quantities of NGAL within 30 minutes in the urine when damaged by ischemia-reperfusion injury, nephrotoxins, sepsis and chronic progressive changes (1). Initially demonstrated in rats, mice, and pigs findings were confirmed in adults, children and neonates (1, 4, 7, 8). NGAL appears to be an early, potentially even 'real-time' biomarker indicating structural damage in contrast to all markers of renal function available in current clinical practice.

In a cross-sectional study, subjects in the intensive care unit with established acute kidney injury (AKI) displayed a greater than 100-fold increase in urine NGAL concentration when compared to normal controls (4). Kidney biopsies

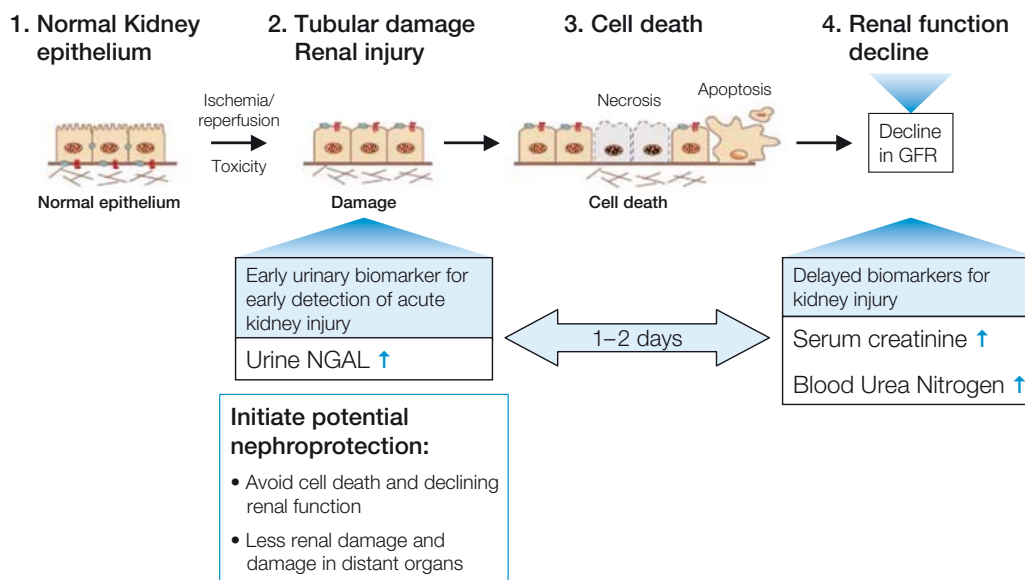
in these patients showed intense accumulation of immuno-reactive NGAL in 50 % of the cortical tubules.

These findings have initiated a number of translational studies and have now been confirmed in several prospective studies of adults and children who developed AKI after defined clinical events such as cardiac surgery, contrast administration, kidney transplantation (9–13) and with unknown timing of kidney injury, such as subjects admitted to emergency or critical care settings (14–17).

AKI represents a frequent and devastating problem in hospitalized patients, with persistently high rates of morbidity and mortality (8). While recent advances have suggested novel therapeutic approaches in animal models, translational efforts in human have yielded disappointing results. The reasons for this include an incomplete understanding of the underlying pathophysiology and the lack of early, sensitive and specific biomarker for AKI (18). In clinical practice, the diagnosis of AKI is mostly based on an increase in serum creatinine. This surrogate parameter of renal function is affected by several non-renal factors, the hydration status and is considered a late indicator of AKI as it typically increases only when the glomerular filtration rate has decreased to >50 % of normal. NGAL has emerged as biomarker that predict development of AKI in heterogeneous groups of patients 1–3 days earlier than serum creatinine (9, 10, 12), Fig. 1 and 2.

In conclusion, NGAL represents a promising early predictive biomarker of AKI. Such novel biomarkers might facilitate earlier diagnosis, timely and individualized patient management decisions including administration of putative therapeutic agents and specific preventative strategies, potentially resulting in fewer complications and improved outcomes (19, 20).

Fig. 1: Early diagnosis of Acute Kidney Injury with urine NGAL



Clinical Utility of Urine NGAL

The Potential Use of NGAL after Cardiac Surgery

Cardiac surgery with cardiopulmonary bypass (CPB) is one of the most common major surgical procedures worldwide often associated with AKI – a frequent and serious complication of cardiac surgery and affects up to 50% of patients depending on definition (12). Even minor degrees of post-operative AKI, as manifest by only a 0.2 to 0.3 mg/dL rise in serum creatinine from baseline, predict a significant increase

in short-term mortality (21). AKI after cardiac surgery is also associated with a number of adverse outcomes, including prolonged intensive care and hospital stay, dialysis dependency, diminished quality of life, and increased long-term mortality. NGAL rises already 1–2 hours after cardiac surgery in case an acute kidney injury develops (Fig. 2).

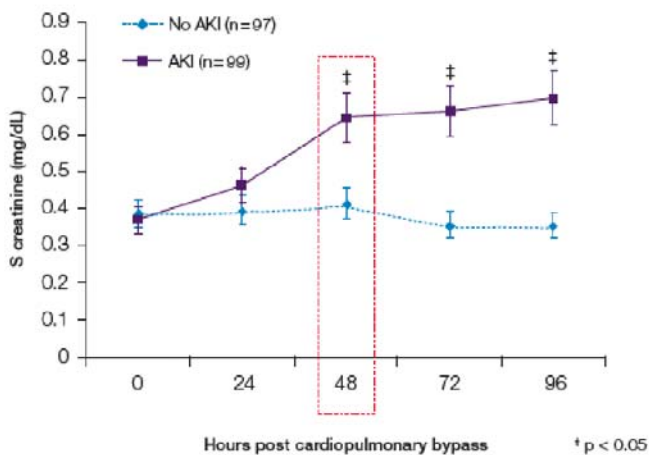
Practical recommendations:

Pending confirmation in larger trials, NGAL measured immediately on admission to the Intensive Care Unit after surgery could identify and stratify patients with acute structural renal damage independent from their preoperative risk and procedure performed. For example, although only investigated in a limited number of patients, NGAL has got a high predictive value for postoperative initiation of renal replacement therapy (RRT) (10, 12). This finding and the potentially earlier initiation of RRT in NGAL-positive patients need confirmation in larger prospective trials.

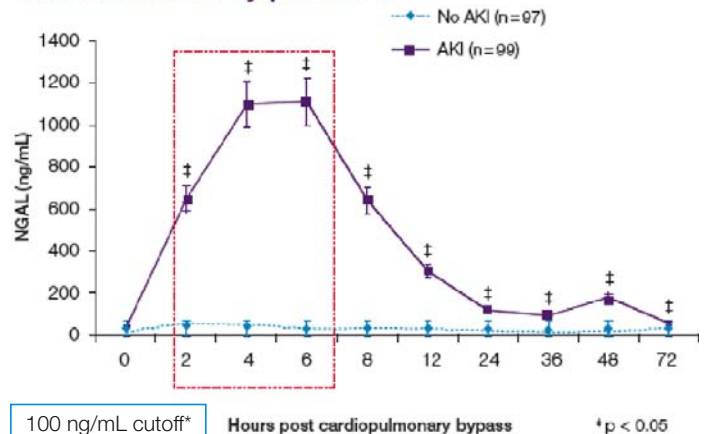
Bennett et al. (10) used the clinical laboratory ARCHITECT platform and found, in children undergoing cardiac surgery, urine NGAL > 150 ng/mL in those subsequently developing AKI.

Fig. 2: NGAL in Diagnosis of AKI after Cardiopulmonary Bypass Surgery

Serum creatinine post-CPB



Urine NGAL measurements obtained by ARCHITECT assay post-CPB



* This represents the value at which AKI can be diagnosed

With the ARCHITECT Urine NGAL* acute kidney injury after cardiopulmonary bypass (CPB) operation could be identified 1–2 days earlier compared to creatinine.

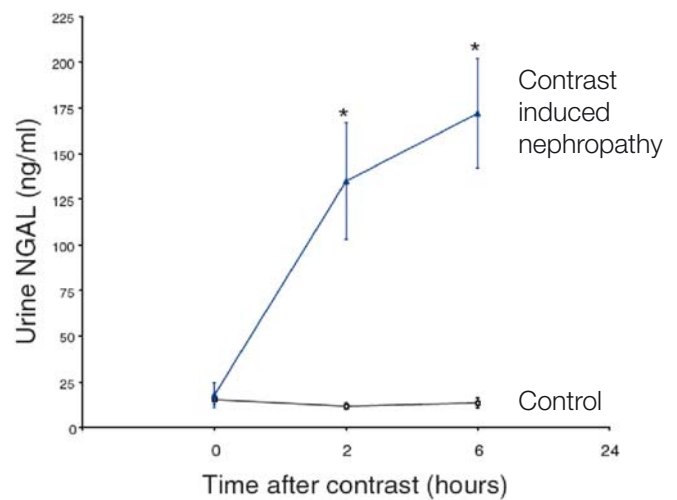
The Potential Use of NGAL after PCI

With many million procedures a year, coronary artery angiography is the most common major cardiac intervention worldwide. Contrast induced nephropathy (CIN) is a frequent complication of percutaneous coronary intervention (PCI) affecting 10–20% of patients and is independently associated with substantially increased risk of hemodialysis and mortality and carries significant costs (22, 23).

Although some preventive measures are available to reduce the incidence of CIN, an early and accurate marker of CIN is lacking. None of the established renal biomarkers is able to diagnose CIN before 24 to 72 hours post-procedure – a time when most of the patients who underwent elective catheterization are already discharged home.

Several investigators have examined the role of NGAL as a predictive biomarker of nephrotoxicity following contrast administration (11, 24). In these prospective studies, NGAL predicted CIN with high sensitivity and specificity within 2–4 hours after contrast administration, Fig. 3.

Fig. 3: NGAL in Diagnosis of AKI after Contrast Nephropathy



Rapid and strong elevation of Urine NGAL

Hirsch et al. *Pediatr Nephrol* 2007

Practical recommendations:

Different cut-off values for urine NGAL have been described (> 10 ng/mL [ref. 25], > 60 ng/mL [ref. 26] and > 100 ng/mL [ref. 11]) measured immediately after coronary artery angiography before the patients are discharged would potentially identify these patients developing AKI. Such patients may require therapy and prolonged hospitalization compared to those without actual renal injury.

The Potential Use of NGAL at the Intensive Care Unit

Several studies have shown that AKI is an independent risk factor for ICU patients (27, 28). To prevent development and progression of AKI in critically ill patients would require an early and accurate diagnosis of renal injury in the ICU, which is often complicated by other risk factors such as sepsis and nephrotoxins (e. g. contrast media, cardiac surgery, aminoglycosides).

Meanwhile several studies have indicated that NGAL might be a useful biomarker of AKI in this setting (15–17). For example, NGAL measured at admission to the ICU allows the diagnosis of AKI up to 48 hours prior to a diagnosis of AKI using the AKI Network criteria (16).

Practical recommendations:

The timing of NGAL measurement during ICU stay deserves further investigation. It is conceivable, that within 6 hours of each pathophysiological 'hit' to the kidney, NGAL should be measured to identify relevant renal damage and adjust treatment accordingly.

The Potential Use of NGAL in the Emergency Department

Nickolas et al. (14) found that a single measurement of urine NGAL in patients admitted to the Emergency Department helps to distinguish AKI from normal function, pre-

renal azotemia, and chronic kidney disease and predicts poor inpatient outcomes.

Practical recommendations:

Patients, with increased serum creatinine concentration at admission and negative NGAL (< 130 ng/mL in this study [ref. 14]) appear not to develop AKI and might not be planned for prolonged hospitalization.

What can be done if AKI is diagnosed earlier?

Implications of urine NGAL are summarized in Figure 4. Ideally, an early increased novel renal biomarker would initiate a paradigm shift in the clinical management of the patient. Clinicians informed of structural renal damage would be aware of the potential for development of clinical AKI. Biomarkers, such as NGAL, may predict AKI. Such patients might benefit from closer monitoring and perhaps adjusted target values of hemodynamics, intravascular status, urine output and renal perfusion. Patients would benefit from the diligent avoidance or discontinuation of nephrotoxins (non-steroidal inflammatory drugs, ACE-inhibitors, gentamicin, cephalosporins, contrast media,

cardiopulmonary bypass). Pre-emptive intervention with extracorporeal blood purification therapy (e. g. immunomodulation using large-pore membranes or new adsorbers) may be considered for subjects with elevated biomarker levels who are developing fluid overload but will not display increased serum creatinine for several days due to hemodilution and time required for reestablishment of a steady state (10). Finally, the availability of promising early biomarkers may enable the timely initiation of interventions, such as natriuretic peptide (29) or sodium bicarbonate (19) that have been beneficial in pilot studies but not yet tested in larger trials.

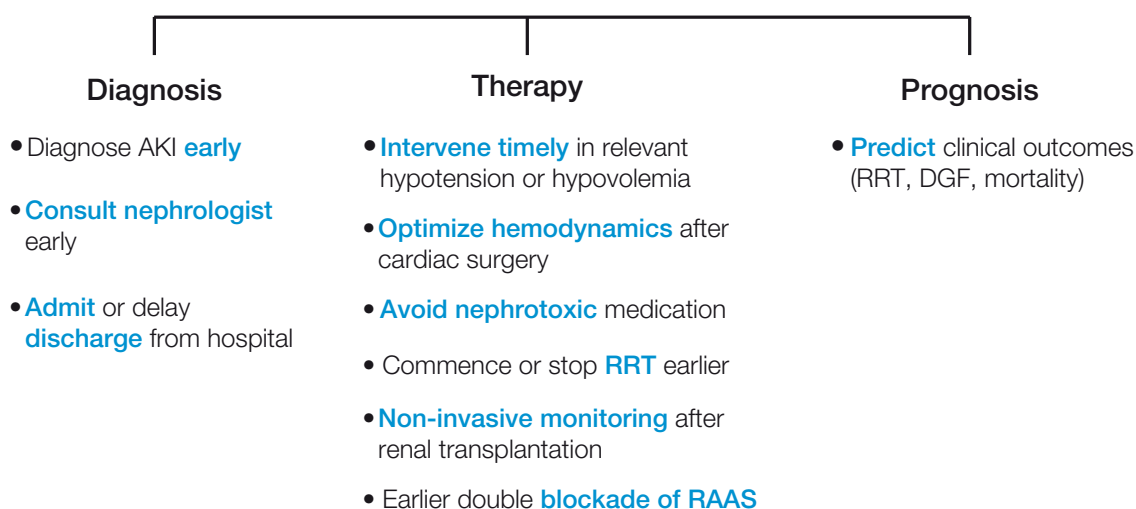
Limitations

Conflicting observations raise concern about the interpretation on NGAL results. Different definitions of AKI after cardiac surgery have been used to assess the predictive value of NGAL in different adult studies creating a definition-related confounder. Recently, a study reported that the predictive value of NGAL increases with grade of AKI (30).

Age, method of measurement ('research'-based vs. routine laboratory platform), urinary tract infections and co-morbidities associated with non-renal release of NGAL appear to be further modifiers of the predictive performance of NGAL. Such limitations of NGAL deserve further investigation in large multicenter studies.

Fig. 4: Implications of urine NGAL

Implications of urine NGAL



AKI: acute kidney injury
RRT: renal replacement therapy
RAAS: renin angiotensin aldosterone system
DGF: delayed graft function

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