Chapter 3

Role of Biomarkers in the Early Detection of Renal Injury

Hema Prabha M P[a], Aanu G S[a], Ramya A[b*]

[a]*B.Pharm Student, Department of Pharmacology, School of Pharmaceutical Sciences, Vels Institutes of Science, Technology & Advanced Studies, Chennai*

[b]*Assistant Professor, Department of Pharmacology, School of Pharmaceutical Sciences, Vels Institutes of Science, Technology & Advanced Studies, Chennai*

** Corresponding Author: ramya.sps@vistas.ac.in*

Abstract

Acute kidney injury (AKI) is a major global health problem associated with significant morbidity and mortality. Conventional diagnostic markers such as serum creatinine and urine output often fail to detect kidney injury at early stages. Therefore, the identification of accurate biomarkers for early detection has become an important focus in nephrology research. Among the emerging biomarkers, urinary neutrophil gelatinase-associated lipocalin (uNGAL) and urinary kidney injury molecule-1 (uKIM-1) have shown considerable promise in identifying renal injury at early stages. These biomarkers are rapidly released following renal tubular damage and can be detected in urine within a few hours after injury. Their high sensitivity and specificity allow earlier diagnosis compared with traditional indicators. This chapter discusses the role of novel biomarkers, including NGAL, KIM-1, IL-18, and L-FABP, in the early detection of

ISBN 978-816855388-0

renal injury and highlights their potential clinical applications in improving diagnosis, monitoring, and management of AKI.

Keywords: Acute kidney injury (AKI); Biomarkers; urinary neutrophil gelatinase-associated lipocalin (uNGAL); urinary kidney injury molecule-1 (uKIM-1).

1. Introduction

Acute Kidney Injury (AKI) is characterised by a decline in renal function that leads to the accumulation of urea and other waste metabolites together with imbalance of fluid and electrolytes. The Kidney Disease Improving Global Outcomes (KDIGO) guidelines characterize acute kidney injury as a rise in serum creatinine levels of ≥0.3 mg/dl (26.5 µmol/litre) within 48 hours, an elevation in serum creatinine levels to ≥1.5 times the baseline within 7 days, or a urine output <0.5 ml/kg/hour for 6 hours. The Prevailing definition and clinical diagnosis of AKI depend on evaluation of serum creatinine and blood urea nitrogen (BUN). However, these parameters rise only after substantial loss of kidney function, often delaying diagnosis and treatment. Serum creatinine may increase only after 48–72 hours of injury, which limits its effectiveness for early detection [1,2]. To overcome these limitations, considerable research has focused on identifying biomarkers capable of detecting renal injury at earlier stages of disease progression [1,10]. Recent advances in molecular biology and proteomics have led to the discovery of several novel biomarkers such as urinary neutrophil gelatinase-associated lipocalin (uNGAL), urinary kidney injury molecule-1 (uKIM-1), cystatin C, interleukin-18 (IL-18), and liver-type fatty acid-binding protein (L-FABP)[5,6,8,12]. These biomarkers are released in response to

renal injury and may appear in blood or urine within hours of damage [1,11].

1.1. Pathophysiology of Acute Kidney Injury

The pathophysiology of AKI involves a significant decline in the glomerular filtration rate (GFR) caused by reduced blood flow to the kidneys, inflammation and tubular injury. The core mechanism includes hemodynamic instability, tubular injury, endothelial dysfunction and intrarenal vasoconstriction [1,10]. The major etiological factors of AKI include ischemia-reperfusion injury, nephrotoxicity and urinary tract obstruction [1,11].

1.2. Characteristics of Ideal Biomarkers

A Biomarker should possess several important characteristics for it to be an ideal one. The characteristics are listed below

- Higher sensitivity and specificity
- Ability to recognize injury at an early stage
- Non-invasive evaluation
- Rapid and reliable assay methods
- Prognostics and predictive value
- Ability to identify the type of renal injury
- Cost-effectiveness for clinical use

Different combinations of biomarkers are often employed to promote the precision of diagnosis because no single biomarker satisfies all the above conditions.

2. Novel Biomarkers in Early Detection of Renal Injury

2.1 Neutrophil Gelatinase-Associated Lipocalin

NGAL is one of the most extensively studied biomarkers for AKI. It is a small protein expressed in neutrophils and epithelial cells. Initially

ISBN 978-816855388-0

discovered in human neutrophils, neutrophil gelatinase-associated lipocalin (NGAL), also known as siderocalin or lipocalin-2 is a 25 kDa polypeptide that is resistant to protease enzymes. The kidney, lungs, and colon are some of the human epithelia where it is expressed in trace amounts. In 2005, Mishra et al. demonstrated that NGAL was a reliable early biomarker of subclinical AKI in pediatric patients following cardiopulmonary bypass; its elevation was maintained throughout the course of AKI and anticipated any increase in serum creatinine by one to three days [1]. The degree of kidney damage is correlated with the level of NGAL expression, which may help identify patients who are at a higher risk of rapid deterioration in renal function [13]. Increased cell proliferation, cytogenesis, renal injury, and the advancement of AKI are all consequences of NGAL expression [1,13]. NGAL has demonstrated a strong association with serum creatinine, cystatin C, and estimated GFR [1]. Additionally, urine NGAL is a measure of normoalbuminuric renal illness in type 2 diabetes mellitus and a good predictor of renal injury prior to apparent changes in eGFR [1, 13].

2.2 Kidney Injury Molecule-1

Kidney injury molecule-1 (KIM-1) is a 38.7 kDa type I transmembrane glycoprotein that contains an extracellular immunoglobulin-like domain [8]. Additionally, KIM-1 functions as a phosphatidylserine receptor and converts epithelial cells into semi-professional phagocytes [15]. It is expressed at low levels in the kidney and other organs, but is markedly elevated in kidney injury, particularly following ischemia-reperfusion injury, in some renal tubulointerstitial disorders, and in polycystic kidney disease[8, 15]. Patients with acute tubular necrosis soon after injury had a soluble form of human KIM-1 in their urine, which was linked to the extent

of the lesion, suggesting that KIM-1 could serve as a biomarker for renal proximal tubule damage and related healing processes[7]. KIM-1 is a sensitive biomarker for chronic proximal tubular injury in patients with acute kidney injury (AKI), which is linked to the incidence, progression, and prognosis of the illness. Chronic KIM-1 expression in renal tubules encourages the production of monocyte chemotactic protein 1, which, in turn, promotes fibrosis and a proinflammatory microenvironment [15]. Its expression increases as the illness progresses and is associated with proteinuria and podocytopenia [8].

2.3 *Interleukin-18 (IL-18)*

The cytokine interleukin-18 (IL-18) belongs to the IL-1 superfamily [6]. It is produced by several cell types, including monocytes, macrophages, proximal tubular cells, and epithelial cells [6]. IL-18 is synthesized as a 23-kDa inactive precursor, which is cleaved by caspase-1 to form the biologically active 18.3-kDa cytokine [6]. The amount of IL-18 released into the urine following its induction in proximal tubular cells and cleavage by caspase-1 can be measured using an enzyme-linked immunosorbent assay (ELISA) [6]. According to Melnikov et al., kidney IL-18 increases in the context of ischemic acute kidney injury (AKI) and causes tubular necrosis by promoting ischemia-reperfusion injury and neutrophil and monocyte infiltration of the renal parenchyma in early animal models [6]. IL-18's predictive accuracy for AKI has been investigated in a number of clinical contexts, such as after cardiac surgery, in critical patient care, following cardiac catheterisation, and following organ donation [1,6]. According to this research, urine IL-18 levels can be easily and inexpensively quantified and rise early in ischaemic kidney injury

ISBN 978-816855388-0

(about 12 hours before clinical AKI). Its ability to predict AKI is enhanced when IL-18 is combined with other biomarkers [1].

3. Liver Fatty Acid-Binding Protein (L-Fabp)

Liver fatty acid–binding protein (L-FABP), also known as FABP1**,** is a 14 kDa soluble protein predominantly found in the cytoplasm of hepatocytes, enterocytes, renal proximal tubular cells, and alveolar epithelial cells [4,12]**.** Under physiological conditions, albumin is primarily reabsorbed in the proximal tubules attached to free fatty acids after being filtered from the glomeruli [14].

Table 1. Major Biomarkers Used in the Early Detection of Renal Injury

Biomarker	Source	Type of Sample	Time of Elevation After Injury	Clinical Significance	Limitations
NGAL	Renal tubular epithelial cells	Blood, Urine	2–4 hours	Early detection of AKI, cardiac surgery AKI	Elevated in infections
KIM-1	Proximal tubular cells	Urine	6–12 hours	Specific marker of tubular injury	Limited routine availability
IL-18	Proximal tubules, immune cells	Urine	4–6 hours	Ischemic and inflammatory AKI	Less specific in systemic inflammation
L-FABP	Renal tubular cells	Urine	1–2 hours	Marker of oxidative stress and tubular ischemia	Limited clinical validation

Following reabsorption, cytosolic albumin enters lysosomes and releases fatty acids to L-FABP [4]. Long-chain fatty acids are bound by L-FABP, which contributes to intracellular signalling, fatty acid

metabolism, and the excretion of lipid peroxidation products, all of which lead to renal protection [4,16]. Urinary L-FABP levels can be measured using a solid-phase enzyme-linked immunosorbent assay (ELISA) based on the sandwich principle with a functional time of 3.5 hours[14]. Elevated urinary L-FABP levels indicate underlying ischemic tubular stress and have been reported to possess moderate prognostic value for predicting the onset of acute kidney injury (AKI) [12]. Combining biomarkers such as KIM-1 and L-FABP improves diagnostic accuracy and early detection of AKI compared with the use of a single biomarker [3].

4. Clinical application of biomarkers for AKI

The identification of AKI biomarkers has evolved, but their application in clinical practice is still relatively new. Acute kidney injury often has multifactorial causes, but the lack of insight into the principal etiology of the acute kidney injury will no doubt hinder studies directed at prevention or treatment of acute kidney injury given the lack of specificity [1, 10]. To date no single biomarker has been shown to be consistently better across a variety of clinical settings. Alternatively, using a panel of biomarkers that are raised under various situations may yield more diagnostic information [3]. The presence of a predictive biomarker or biomarker panel, suggestive of an underlying cause of acute kidney injury, would allow early identification of acute kidney injury and targeting management to the underlying cause, aimed at preventing acute kidney injury. Therefore, continued research and large-scale clinical validation studies are essential to establish reliable biomarker panels for the early diagnosis, prognosis, and effective clinical management of renal injury [1, 10]. The major biomarkers used for the early detection of acute

ISBN 978-816855388-0

kidney injury, along with their sources, sample types, clinical significance, and limitations, are summarized in Table 1.

5. Conclusion

Acute kidney injury (AKI) remains a major clinical concern and is associated with significant morbidity, mortality, and healthcare burden worldwide [1,2]. Conventional diagnostic markers such as serum creatinine and blood urea nitrogen are widely used in clinical practice; however, these markers often increase only after considerable loss of renal function, limiting their usefulness for early detection of kidney injury [1]. Consequently, there has been increasing interest in the development of novel biomarkers that can identify renal injury at earlier stages and allow timely intervention.

Several biomarkers, including neutrophil gelatinase-associated lipocalin (NGAL), kidney injury molecule-1 (KIM-1), interleukin-18 (IL-18), and liver fatty acid-binding protein (L-FABP), have demonstrated significant potential for the early detection and prognosis of AKI [6,8,12,13]. These biomarkers reflect different pathological activities involved in renal injury, such as tubular damage, inflammation, oxidative stress, and ischemia [6,8,12]. Because AKI is a complex and multifactorial condition, the use of a single biomarker may not provide sufficient diagnostic accuracy [3].

Despite the promising potential of these biomarkers, several challenges remain before their routine clinical implementation. Variability in study designs, differences in diagnostic cut-off values, and limited validation across diverse patient populations continue to restrict widespread clinical application [1,10]. Thus, further large-scale clinical studies and standardized protocols are necessary to establish reliable biomarker-based diagnostic strategies.

In conclusion, AKI biomarkers represent an important advancement in the early detection and management of renal injury. With continued research, validation, and integration into clinical practice, biomarker-guided approaches may improve early diagnosis, enable targeted therapeutic interventions, and ultimately reduce the progression of kidney injury, the need for renal replacement therapy, and associated mortality [1,2,3].

References

[1] Mahrukh S Rizvi, Kianoush B Kashani, Biomarkers for Early Detection of Acute Kidney Injury, *The Journal of Applied Laboratory Medicine*, Volume 2, Issue 3, 1 November 2017, Pages 386–399, https://doi.org/10.1373/jalm.2017.023325

[2] Silver, S.A., Long, J., Zheng, Y. and Chertow, G.M. (2017), Cost of Acute Kidney Injury in Hospitalized Patients. Journal of Hospital Medicine, 12: 70-76. https://doi.org/10.12788/jhm.2683

[3] Parikh, C.R., Thiessen-Philbrook, H., Garg, A.X., Kadiyala, D., Shlipak, M.G., Koyner, J.L., Edelstein, C.L., Devarajan, P., Patel, U.D., Zappitelli, M. and Krawczeski, C.D., 2013. Performance of kidney injury molecule-1 and liver fatty acid-binding protein and combined biomarkers of AKI after cardiac surgery. *Clinical Journal of the American Society of Nephrology*, *8*(7), pp.1079-1088.

[4] Yamamoto, T., Noiri, E., Ono, Y., Doi, K., Negishi, K., Kamijo, A., ... & Sugaya, T. (2007). Renal L-type fatty acid–binding protein in acute ischemic injury. *Journal of the American Society of Nephrology*, *18*(11), 2894-2902.

[5] Herget-Rosenthal, S., Marggraf, G., Hüsing, J., Göring, F., Pietruck, F., Janssen, O., Philipp, T. and Kribben, A., 2004. Early detection of acute renal failure by serum cystatin C. *Kidney international*, *66*(3), pp.1115-1122.

[6] Parikh, C.R., Jani, A., Melnikov, V.Y., Faubel, S. and Edelstein, C.L., 2004. Urinary interleukin-18 is a marker of human acute tubular necrosis. *American Journal of Kidney Diseases*, *43*(3), pp.405-414.

[7] Liangos, O., 2006. Urinary kidney injury molecule-1 level is an early and sensitive marker of acute kidney injury following cardiopulmonary bypass. *J Am Soc Nephrol*, *17*, p.403A.

ISBN 978-816855388-0

[8] Ichimura, T., Hung, C.C., Yang, S.A., Stevens, J.L. and Bonventre, J.V., 2004. Kidney injury molecule-1: a tissue and urinary biomarker for nephrotoxicant-induced renal injury. *American Journal of Physiology-Renal Physiology*, *286*(3), pp.F552-F563.

[9] Kjeldsen, L., Johnsen, A.H., Sengeløv, H. and Borregaard, N., 1993. Isolation and primary structure of NGAL, a novel protein associated with human neutrophil gelatinase. *Journal of Biological Chemistry*, *268*(14), pp.10425-10432.

[10] Kramer, A.A., Sutherland, S.M., Hsu, R.K., Kellum, J.A., Bagshaw, S.M., Chawla, L.S., Kane-Gill, S.L., Goldstein, S.L. and Ronco, C., 2016. Utilizing electronic health records to predict acute kidney injury risk and outcomes: Workgroup statements from the 15th ADQI Consensus Conference.

[11] Vaidya, V.S., Ford, G.M., Waikar, S.S., Wang, Y., Clement, M.B., Ramirez, V., Glaab, W.E., Troth, S.P., Sistare, F.D., Prozialeck, W.C. and Edwards, J.R., 2009. A rapid urine test for early detection of kidney injury. *Kidney international*, *76*(1), pp.108-114.

[12] Susantitaphong, P., Siribamrungwong, M., Doi, K., Noiri, E., Terrin, N. and Jaber, B.L., 2013. Performance of urinary liver-type fatty acid–binding protein in acute kidney injury: a meta-analysis. *American Journal of Kidney Diseases*, *61*(3), pp.430-439.

[13] Makris K, Markou N, Evodia E et al. Urinary neutrophil gelatinase-associated lipocalin (NGAL) as an early marker of acute kidney injury in critically ill multiple trauma patients. Clin Chem Lab Med. 2009;47(1):79–82. https://doi.org/10.1515/CCLM.2009.004.

[14] Geng, J., Qiu, Y., Qin, Z. and Su, B., 2021. The value of kidney injury molecule 1 in predicting acute kidney injury in adult patients: a systematic review and Bayesian meta-analysis. *Journal of Translational Medicine*, *19*(1), p.105.

[15] Bonventre, J.V., 2009. Kidney injury molecule-1 (KIM-1): a urinary biomarker and much more. *Nephrology Dialysis Transplantation*, *24*(11), pp.3265-3268.

[16] Kamijo-Ikemori, A. and Kimura, K., 2015. Urinary liver-type fatty acid binding protein and chronic kidney disease. *Indian Journal of Nephrology*, *25*(5), p.263.