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Urinary liver fatty acid-binding protein: another novel biomarker of acute kidney injury

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Liver fatty acid-binding protein (L-FABP) binds selectively to intracellular free unsaturated fatty acids and lipid peroxidation products during hypoxic tissue injury. Urinary L-FABP is a potential biomarker for the detection and assessment of acute kidney injury (AKI). Ferguson *et al.* have demonstrated in a cross-sectional study that urinary L-FABP is an excellent biomarker of AKI and may be useful in predicting dialysis-free survival. This study did not assess utility for early diagnosis of AKI.

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Several candidate biomarkers for the early detection and assessment of acute kidney injury (AKI) are in development, as a result of a concerted effort to improve the timeliness and specificity of interventions in future clinical trials in this patient population. Liver fatty acid-binding protein (L-FABP), which is among these promising new AKI biomarkers, is a member of a family of FABPs. In recent years, a number of human clinical studies have explored the potential utility of urinary L-FABP as a biomarker for the early diagnosis of AKI.[1]

In this issue, Ferguson and colleagues[2] provide data that further establish the potential clinical utility of urinary L-FABP for the diagnostic assessment of AKI in hospitalized patients. This cross-sectional study in 92 patients with AKI and 68 controls (26 healthy volunteers and 42 hospitalized patients, including 29 pre-coronary catheterization patients and 13 intensive care unit patients) establishes that the performance of L-FABP in the detection and assessment of AKI satisfies the most basic requirements for a useful diagnostic test in this syndrome: urinary levels of the marker are significantly higher in those with AKI than in hospitalized control patients without AKI. This is reflected in an excellent area under the receiver operating characteristic curve (ROC-AUC) of 0.93 (95% confidence interval 0.88–0.97, with a sensitivity of 83% and a specificity of 90% at a cutoff value of 47.1 ng/mg creatinine).[2] Sensitivity (95%) and AUC (0.96) were improved when normal volunteers were included among the controls, although specificity decreased to 84% with the lower cutoff value of 26.1 ng/mg creatinine. Regardless, the use of healthy volunteers in the assessment of a diagnostic test for AKI seems more likely to falsely improve test performance than to provide clinically useful information. Although tending to be lower in patients with radiocontrast nephropathy as compared with other causes of AKI (acute tubular necrosis, sepsis, other nephrotoxins), urinary L-FABP levels were not significantly different between these etiologic AKI subgroups.

The authors also directly compared the diagnostic performance of urinary L-FABP with that of other emerging biomarkers of AKI—neutrophil gelatinase-associated lipocalin, kidney injury molecule-1, *N*-acetyl-β-glucosaminidase, and interleukin-18—and found it comparable to the former 3 markers, and superior to the latter. These observations correlate with earlier studies of human L-FABP transgenic mice exposed to ischemia–reperfusion injury. Urinary L-FABP was superior to other biomarkers, such as blood urea nitrogen and *N*-acetyl-β-glucosaminidase, for the detection of significant histological injuries and functional declines by ROC curves.[3]

Finally, Ferguson *et al.*[2] performed an exploratory age-adjusted logistic regression analysis of the prognostic value of L-FABP to predict renal and nonrenal clinical outcomes, and found that urinary levels were significantly predictive of the need for acute renal replacement therapy (RRT) and the composite end point of death/RRT, but not of in-hospital mortality. Thus, despite these encouraging results, the validation of L-FABP for the diagnostic and prognostic assessment of AKI is still in its early stages. In particular, the ability of L-FABP to provide early diagnosis of AKI was not tested in this study;[2] the study was relatively small to establish the value of this marker for the differential diagnosis of AKI etiologies; and the sample size for prognostic evaluation was suitable only for exploratory analysis. Nonetheless, these encouraging results will add impetus to the development of L-FABP as an AKI biomarker.

Although it is exactly 40 years since its discovery by the isolation of two hepatic cytoplasmic proteins, our current understanding of the mechanism by which L-FABP, or FABP1, activates putative antioxidant signals remains surprisingly incomplete.[1] Originally known as Z protein, L-FABP is an intracellular lipid chaperone that binds selectively to free unsaturated fatty acids and lipid peroxidation products during tissue injury from hypoxia. In the kidney, two types of FABPs have been found in renal tubule cells: L-FABP (FABP1) and heart and muscle type (FABP3, or H-FABP). L-FABP is solely localized to the cytoplasmic region of proximal tubular cells. Increased cytosolic L-FABP in proximal tubular epithelial cells may derive not only from

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Table 1 | Clinical studies of L-FABP in AKI

Reference	Year	Subjects/Methods	Key results
Ferguson *et al.*[2]	2010	92 patients with established AKI and 68 control subjects	Elevated urinary L-FABP in established AKI of various etiology. Urinary L-FABP as a predictor of adverse renal/nonrenal outcomes.
Nakamura *et al.*[6]	2009	40 patients with septic shock	Urinary L-FABP levels increased in patients with septic shock and may correlate with severity of septic shock and response to treatment. No change in serum L-FABP levels. Urinary L-FABP levels not correlated with need for renal replacement therapy.
Portilla *et al.*[4]	2008	40 children undergoing cardiac surgery	Urinary L-FABP levels increased 94- and 45-fold at 4 and 12 hours, respectively, following surgery in the 21 patients who developed AKI.
Fukuda *et al.*[5]	2009	27 patients with stable angina or acute coronary syndrome undergoing coronary angiogram and 12 control volunteers	Urinary L-FABP levels were significantly higher in acute coronary syndrome than in stable angina. However, none of these patients developed contrast-induced nephropathy; L-FABP may be a predictor of acute coronary syndrome.
Nakamura *et al.*[8]	2006	66 patients undergoing non-emergency coronary angiography who had a serum creatinine level greater than 1.2 mg/dl (>106 μmol/l) and less than 2.5 mg/dl (<221 μmol/l) and 30 healthy volunteers	Urinary L-FABP levels were detectable before angiography and predicted those who developed contrast medium–induced nephropathy. After 14 days, serum creatinine levels returned to normal and L-FABP levels remained high.
Yamamoto *et al.*[7]	2007	12 patients with kidney transplants from living related donors	Urinary L-FABP levels were measured in the first urine produced after reperfusion of the transplanted organs. Correlations were found between urinary L-FABP level and both peritubular capillary blood flow and ischemic transplant time. Transition of L-FABP from the cytoplasm of proximal tubular cells to the tubular lumen in human L-FABP transgenic mice subjected to renal ischemia–reperfusion.

Abbreviations: AKI, acute kidney injury; L-FABP, liver-type fatty acid–binding protein.

endogenous expression but perhaps also from circulating L-FABP, which might be filtered at the glomeruli and reabsorbed by tubular cells. In this scenario, liver injury may be a potential contributor to the increased urinary levels of L-FABP during AKI. This point was nicely addressed by Portilla *et al.*,[4] who measured serum and urine L-FABP in patients undergoing cardiac surgery. There was a significant increase of urinary L-FABP levels 4 h after the surgery, whereas serum L-FABP levels did not begin to increase until 12 h postoperatively. These observations support the concept that the human urinary L-FABP level is mostly determined by proximal tubule injury, rather than extrarenal production (from liver injury, or systemic inflammation associated with cardiopulmonary bypass or sepsis). In another study, by Fukuda *et al.*,[5] urinary L-FABP showed a higher sensitivity and specificity for the diagnosis of acute coronary syndrome and was not a predictor of AKI in patients undergoing coronary angiogram. The rise in urinary L-FABP levels did not result from contrast-induced nephropathy. The study highlighted the extrarenal source of urinary L-FABP. Nakamura *et al.*[6] demonstrated that urinary L-FABP levels did not correlate with Acute Physiology and Chronic Health Evaluation (APACHE) scores in septic patients. Urinary L-FABP could serve as a biomarker of AKI primarily in septic patients presenting with septic shock and multiorgan failure, in whom the potential for liver involvement could present. This important concept was not addressed by Ferguson *et al.*[2] in the current study. The authors did not include information on any of the following: serum L-FABP levels with or without AKI; APACHE scores; liver function tests; or organ injury in patients with septic shock, multiorgan failure, and probable liver injury.

Once in the proximal tubular cells, L-FABP may act as a surrogate molecule to reduce lipid peroxidative stress during reperfusion by binding fatty acid oxidation products and limiting the toxic effects of oxidative intermediates on cellular membranes. Overexpression of L-FABP in mice subjected to renal ischemia–reperfusion injury offers protection against renal tissue hypoxia.[6] The urinary levels of L-FABP have been reported in several kinds of human clinical studies[4,6–8] (Table 1) and animal studies[3,9] of AKI. Taken together, these results indicate that L-FABP not only contributes to the trafficking of fatty acids but also serves as a diagnostic indicator of acute renal diseases. Although Ferguson and colleagues[2] have provided further evidence that urinary L-FABP can aid in the diagnostic and perhaps the prognostic evaluation of AKI, several steps in the validation process for this urinary biomarker remain to be addressed.

What else needs to be done to evaluate the utility of this promising AKI biomarker? Let's place current knowledge of L-FABP biology within a conceptual framework of AKI that describes a series of stages, antecedents, and outcomes (Figure 1).[10] In this model, the stages of development (Figure 1, left to right) or recovery (right to left) of AKI are highlighted. Antecedents (in yellow) of AKI

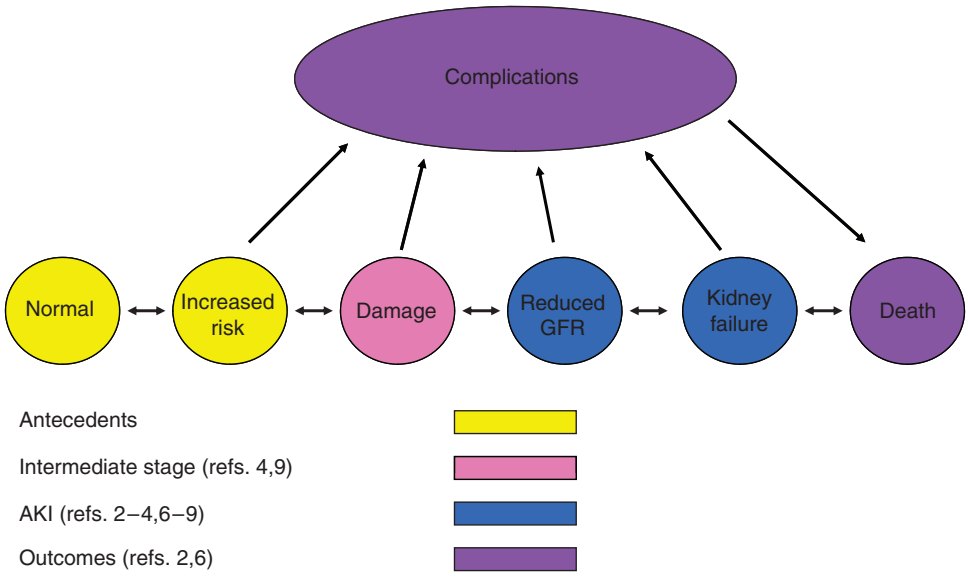

Figure 1 | L-FABP in a conceptual framework of AKI. (Adapted from ref. 10.) AKI, acute kidney injury; GFR, glomerular filtration rate; L-FABP, liver-type fatty acid–binding protein.

include increased risk in certain patient population groups, such as older persons. Outcomes (in purple) include fatal or non-fatal complications in other organ systems. Kidney damage (in pink) is an intermediate stage preceding AKI (signaled by glomerular filtration rate loss and acute elevation of serum creatinine). This stage is emphasized here because of the potential importance of biomarkers in predicting AKI several hours to days before the rise in serum creatinine, which may allow early AKI diagnosis and more rapid implementation of protective measures (volume expansion, vasoactive drug therapy, avoidance of nephrotoxins). Although some clinical data suggest potential utility of L-FABP for the early diagnosis by AKI (Table 1; Figure 1),[4,6,9] the study by Ferguson *et al.*[2] does not address this point. In animal models, urinary L-FABP was observed at 1 h after ischemia–reperfusion injury.[3] In pediatric patients undergoing major cardiac surgery, urinary L-FABP levels were detectable 4 h postoperatively and were predictive of the subsequent development of AKI with an ROC-AUC of 0.81.[4] In addition to a careful assessment of the ability of L-FABP to achieve early diagnosis of AKI, future studies should increase the depth of the assessment of other nonrenal outcomes (length of intensive care unit stay, functional status assessment, economic analysis) and renal outcomes (such as RRT duration, recovery of renal function, and/or progression to chronic kidney disease). In one cohort study of 40 septic patients, 28 survived and 12 died. Among the surviving patients, urinary L-FABP levels were reduced by treatment.[6] However, the non-surviving patients showed higher urinary L-FABP levels with smaller decreases after the treatment as compared with the survivors. These results suggested that urinary L-FABP levels might be able to reflect the severity of sepsis and the response to treatment.[6]

In conclusion, the current data from a modest number of human subjects suggest a potential role of urinary L-FABP in the clinical diagnostic evaluation of AKI. Larger multicenter studies that include earlier serial urine sampling (to evaluate early diagnosis), larger patient cohorts (including important subsets such as septic patients), and comparison with other emerging AKI biomarkers are required to continue the development of this promising biomarker.

DISCLOSURE

PTM is a consultant to Abbott Laboratories, Argutus Medical, and Inverness Inc.

REFERENCES

1. Noiri E, Doi K, Negishi K *et al.* Urinary fatty acid-binding protein 1: an early predictive biomarker of kidney injury. *Am J Physiol Renal Physiol* 2009; **296**: F669–F679.
2. Ferguson MA, Vaidya VS, Waikar SS *et al.* Urinary liver-type fatty acid-binding protein predicts adverse outcomes in acute kidney injury. *Kidney Int* 2010; **77**: 708–714.
3. Negishi K, Noiri E, Doi K *et al.* Monitoring of urinary L-type fatty acid-binding protein predicts histological severity of acute kidney injury. *Am J Pathol* 2009; **174**: 1154–1159.
4. Portilla D, Dent C, Sugaya T *et al.* Liver fatty acid-binding protein as a biomarker of acute kidney injury after cardiac surgery. *Kidney Int* 2008; **73**: 465–472.
5. Fukuda Y, Miura S, Zhang B *et al.* Significance of urinary liver-fatty acid-binding protein in cardiac catheterization in patients with coronary artery disease. *Intern Med* 2009; **48**: 1731–1737.
6. Nakamura T, Sugaya T, Koide H. Urinary liver-type fatty acid-binding protein in septic shock: effect of polymyxin B-immobilized fiber hemoperfusion. *Shock* 2009; **31**: 454–459.
7. Yamamoto T, Noiri E, Ono Y *et al.* Renal L-type fatty acid–binding protein in acute ischemic injury. *J Am Soc Nephrol* 2007; **18**: 2894–2902.
8. Nakamura T, Sugaya T, Node K *et al.* Urinary excretion of liver-type fatty acid-binding protein in contrast medium-induced nephropathy. *Am J Kidney Dis* 2006; **47**: 439–444.
9. Yokoyama T, Kamijo-Ikemori A, Sugaya T *et al.* Urinary excretion of liver type fatty acid binding protein accurately reflects the degree of tubulointerstitial damage. *Am J Pathol* 2009; **174**: 2096–2106.
10. Murray PT, Devarajan P, Levey AS *et al.* A framework and key research questions in AKI diagnosis and staging in different environments. *Clin J Am Soc Nephrol* 2008; **3**: 864–868.