Diagnostic value of neutrophil gelatinase-associated lipocalin for renal injury in asphyxiated preterm infants

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Abstract. The aim of the present study was to analyze the diagnostic value of neutrophil gelatinase-associated lipocalin (NGAL) in renal injury in asphyxiated preterm infants. In total, 48 cases of asphyxiated preterm infants, 45 cases of premature infants and 45 cases of normal newborn infants were included in the study. Using ELISA we evaluated the level of urine NGAL, kidney injury molecule-1 (KIM-1), cystatin C (Cys-C) and serum creatinine (Scr). We also calculated the estimated glomerular filtration rate (eGFR). Our results showed that NGAL, KIM-1 and Cys-C levels in the group of asphyxiant renal injury within 24 and 48 h were markedly higher than the other two groups (P<0.05), while changes in Scr and eGFR within 24 h were not significant (P>0.05). Scr level in renal injury group within 48 h was markedly elevated while the eGFR level was visibly reduced (P<0.05). According to analysis of receiver operating characteristic curve, area under curve for NGAL in the group of asphyxiant renal injury within 24 and 48 h was significantly higher than KIM-1 and Cys-C. Susceptibility and specificity were improved. In conclusion, diagnosis of renal injury in asphyxia neonates using NGAL was more efficient compared to other methods.

Introduction

Renal injury in asphyxiated preterm infants has a high incidence rate (30-55%) and the mortality rate is 60-66% (1). According to the definition of acute kidney injury (AKI) and classification of RIFLE (2) which was presented by AKI network (AKIN) in 2007, the likelihood of diagnosing renal injury in asphyxiated preterm infants was poor (3). Results obtained from at least one study showed that neutrophil gelatinase-associated lipocalin (NGAL), kidney injury molecule-1 (KIM-1) and level of cystatin C (Cys-C) could be used as sensitive biochemical markers for diagnosing renal injury in the early stages (4).

The same study showed that NGAL, KIM-1 and Cys-C in vivo levels were higher and more stable compared to serum creatinine (Scr) and urea nitrogen levels in the early stages of the disease (4).

Through comparative analysis, it was shown that these three indexes (NGAL, KIM-1 and Cys-C) had peak changes at different time-points and were closely related to the degree of renal injury (5,6). The present study explored the significance of using NGAL for diagnosing renal injury in asphyxiated preterm infants and whether NGAL had a better diagnostic value.

Materials and methods

Patients. From October 2014 to October 2015, we enrolled 48 cases of asphyxiated preterm infants which were admitted to our hospital. We excluded cases with: i) Congenital monstrosity; ii) inherited metabolic diseases; iii) hypoxic ischaemia encephalopathy; iv) severe asphyxia with no chance of recovery; and v) serious conditions with expected survival time of less than one month. There were 27 males and 21 females with an average age of 15.6±7.2 h (range, 1-24 h). Average birth weight was 2.1±0.6 kg (range, 1.7-2.3 kg), and the average gestational period was 35.6±2.4 weeks (range, 34-37 weeks). We also enrolled 45 cases of premature infants without renal injury after asphyxia at the same time. Of these cases, there were 25 males and 20 females with an average age of 15.6±7.2 h (range, 1-24 h). Average birth weight was 2.1±0.6 kg (range, 1.7-2.3 kg).

Average gestational period was 35.6±2.4 weeks (range, 34-37 weeks). We also enrolled 45 cases of premature infants without renal injury after asphyxia at the same time. Of these cases, there were 25 males and 20 females with an average age of 15.9±7.5 h (range, 1.5-28 h). The average birth weight was 2.3±0.9 kg (range, 1.6-2.5 kg), and the average gestational period was 35.9±2.6 weeks (range, 35-37 weeks).

Forty-five cases of normal newborn infants were also enrolled and of those there were 26 males and 19 females with an average age of 17.5±6.6 h (range, 2-30 h). The average birth weight for this group was 2.6±0.8 kg (range, 2.3-3.1 kg).

Differences in gender, birth time, birth weight and gestational period for premature infants with renal injury and that of premature infants without renal injury had no statistical
Statistical analysis. We analyzed data using SPSS 19.0 statistical software (SPSS, Inc., Chicago, IL, USA). Measurement data were indicated as mean ± standard deviation. We compared among groups using one-way ANOVA and countable data were presented as samples or percentage. The $\chi^2$ test was used to compare among groups. Analysis of diagnostic susceptibility and specificity of NGAL, KIM-1 and Cys-C were realized using receiver operating characteristic curve (ROC). P<0.05 was considered to indicate a statistically significant difference.

Results

Comparisons of observation index of each group. NGAL, KIM-1 and Cys-C levels in the group of asphyxiant renal injury at 24 and 48 h were markedly higher than the other two groups (P<0.05). Changes in Scr and eGFR within 24 h were not significant (P>0.05). Scr level in the renal injury group in 48 h was significantly elevated while eGFR level decreased considerably (P<0.05) (Table I).

Analysis of NGAL, KIM-1 and Cys-C. Area under the curve (AUC) for NGAL in the group of asphyxiant renal injury at 24 and 48 h was significantly higher than KIM-1 and Cys-C. Susceptibility and specificity was improved (Table II, and Figs. 1 and 2).

Discussion

Compared with glomerular, proximal tubular in premature infants has poorer development, reabsorption and its ability to treat water-electrolyte as well as acid-base balance is seriously undermined. Compared to mature infants, premature infants with lower gestational age and lower birth weight suffer from lower rate of nephron and lower glomerular filtration. In premature infants, GFR that influence renal vasoconstriction and relaxation is usually low. Thus AKI is more likely to happen in premature infant (7-9).

NGAL is involved in the formation and reparation of renal tubular epithelial cells. It has been established that NGAL level is high in proximal tubular epithelial cells and in the case of AKI, renal tubular epithelial cells are damaged and NGAL levels in blood and urine is very high (10).

Devarajan showed that in renal ischemia reperfusion injury model in rat, NGAL could be detected when AKI occurs for 2 h. However, at the same time, Scr level remained at its normal level. It was concluded that compared with Scr,
NGAL was more sensitive and a better marker for early detection of AKI (11).

Results obtained by Yavuz et al on 22 cases of burned children who were admitted to hospital within 12 h revealed that there were no significant differences between the Scr level in the 1-5 days AKI group and that in the non-AKI group. By contrast, blood and urine NGAL levels increased considerably in the AKI group which indicated the sensitivity of NGAL for early detection of AKI (12).

Results obtained from another related study on 60 cases of neonates in critical condition showed that NGAL level had little to do with sepsis (13). In that study, according to diagnostic code of AKIT, the cases were divided into the AKI group and the non-AKI group. The results of that study revealed that, the blood NGAL level markedly increased in the early stages of AKI. It was shown that when the NGAL level in blood surpassed the 117.5 ng/ml mark it was an indication of AKI. Susceptibility was 82.0% and specificity was 88.5% (13).

The results presented by Askenazi et al demonstrated that the baseline assessment of urine NGAL in premature infant and gestational age were negatively correlated, while an elevated level of urine NGAL and the degree of renal injury were positively correlated (14).

KIM-1 is a member of the immunoglobulin superfamily. KIM-1 is a transmembrane glycoprotein which comprised mucoprotein and immunoglobulin. It usually engages in recovering process of renal tubule epithelial cells, renal interstitial fibrosis and immunological reactions. It has the functions of adhering and cleaning up the apoptotic cells (15). Under normal physiological conditions, KIM-1 expression level in normal kidney tissue fluctuates between zero to very low. In a case of AKI caused by ischemia or toxic kidney injury, KIM-1 expression level in kidney tissue was markedly increased (16). When proximal tubular injury occurs, extracellular KIM-1 enters into urine in the form of soluble fragment which is relatively stable and easy to detect (17).

Cys-C is a tyep of cystatin, which is produced by all the karyocytes in the body. It is a non-glycosylated alkaline protein with a low molecular weight that can only be cleaned up by passing through glomerular filtration. It is reabsorbed in proximal convoluted tubule, then decomposed totally and metabolized. It is an endogenous marker that reflects GFR (18).

We showed that NGAL, KIM-1 and Cys-C levels in the group of asphyxiant renal injury within 24 and 48 h were markedly higher than the two other groups. Changes in Scr and eGFR within 24 h were not significant. Scr of renal injury within 48 h was markedly elevated while eGFR reduced obviously. Our results indicated that urine NGAL, KIM-1 and Cys-C could respond to AKI in the early stage. Compared with KIM-1 and Cys-C, AUC of NGAL in the group of asphyxiant renal injury within 24 and 48 h was obviously higher and both susceptibility and specificity was improved. Results also suggested that urine NGAL may have higher diagnostic accuracy compared with KIM-1 and Cys-C.

In conclusion, the critical value of detecting urine NGAL in renal injury in asphyxia preterm infants at 24 h was 48.5 ng/ml,
susceptibility was 86.4% and specificity was 83.7%. The critical value of detecting urine NGAL in renal injury in asphyxia preterm infants within 48 h was 123.7 ng/ml, susceptibility was 87.5% and specificity was 84.3%.

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