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Changes of serum neopterin and its significance as biomarker in prediction the prognosis of patients with acute pancreatitis

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Abstract

Objectives: To investigate the dynamic changes of serum neopterin and its significance as biomarker in prediction the prognosis of patients with acute pancreatitis.

Methods: 54 cases with confirmed diagnosis of acute pancreatitis were included in the present work. Of the included 54 cases, 21 were mild acute pancreatitis and other 33 were server diseases. For the 33 severe cases, nine were finally dead and 24 were survived. The serological neopterin level of the 54 acute pancreatitis was continuously examined at the time point of days 0 (diagnosis), 1 (24 h after diagnosis), 2, 4, 8 and 14 by the enzyme linked immunosorbent assay (ELISA). The severity or death risk of the acute pancreatitis patients was predicted by the serological neopterin.

Results: The serological neopterin was gradually increasing from days 0 to 8, but descending at day 14 in mild and survival groups. For days 8 and 14, the serological levels of neopterin in severe group were higher than those of mild group with statistical difference ($p < 0.05$). The serum neopterin was statistical different in the time point of day 8 and day 14 between death and survival groups ($p < 0.05$). For day 8, the serological neopterin as biomarker for death prediction sensitivity and specificity were 88.89% (95% CI: 51.75–99.72%) and 83.33% (95% CI: 62.62–95.26%) respectively with the AUC of 0.95 (95% CI: 0.88–1.00). For day 14, the death prediction sensitivity and

specificity were 77.78% (33.99–97.19%) and 95.83% (78.88–99.89%) respectively with the AUC of 0.94 (95% CI: 0.87–1.00).

Conclusions: Serological neopterin level was elevated with the development of the pancreatitis. Continuously monitoring the serum neopterin may helpful for prediction death risk of acute pancreatitis. In the later phase of disease beginning on day 8, neopterin levels may be used for risk assessment and possibly change of therapy regiment.

Keywords: acute pancreatitis; death prediction; diagnosis; neopterin; serological marker.

Introduction

Acute pancreatitis (AP) is a common and frequently diagnosed disease clinically. The clinical manifestations are quite different between mild and severe AP cases [1]. The treatment and prognosis of mild and severe cases are also quite different. Severe acute pancreatitis (SAP) had the characteristics of onset urgently, rapid progression, complex condition, multiple complications and high mortality, which require timely and urgent effective treatment [2]. Although receiving proper treatment, the mortality rate of SAP is remain about 20–30% [3–5]. Therefore, it is important to predict and evaluate the severity of AP in early stage.

In recent years, with the continuous progress of diagnosis and treatment of SAP, a variety of clinical scores and laboratory indicators that can predict the prognosis of the disease in early stage, such as Rason score, acute physiology and chronic health evaluation II (APACHE II) score, computed tomography severity index (CTSI) score, and sequential organ failure assessment (SOFA) score. The above scores for AP severity evaluation have their own advantages in clinical application, but also had their own limitations such as low sensitivity or specificity. Therefore, it is important to identify clinical markers, which are accurate, efficient, and operable with high sensitivity and specificity for AP severity or death risk prediction.

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Neopterin is a catabolic product of guanosine triphosphate (GTP) which belongs to the chemical group known as pteridines. Neopterin is one of the important markers reflecting the cellular immune state mediated by lymphocyte macrophage axis *in vivo* [6], and its clinical significance has been paid more and more attention. It has been confirmed that neopterin concentration was obviously elevated in serum or urine in patients with inflammatory disease [7, 8], autoimmune disease. However, the dynamic changes of serum neopterin and its significance as biomarker in prediction the prognosis of patients with AP is unclear yet.

Materials and methods

Patients

This work was approved by the Ethical Committee of the Second People's Hospital of Lishui. The signed written informed consent was obtained from all the included subjects. 54 cases with confirmed diagnosis of AP were included in the present work. Of the included 54 cases, 21 were mild AP and other 33 were server diseases. For the 33 severe cases, nine were finally dead and 24 were survived. The patients inclusion criteria were: (1) The age of the subjects raged from 18 to 70 without limitation of gender; (2) AP diagnosed according to the AP Guideline [9]; and (3) Patients provide the written informed consent for the research. Exclusion criteria were: (1) Patients with a medical history of chronic pancreatitis; (2) Subjects with malignant carcinoma; (3) Pregnant women; and (4) Patients died within 14 days after diagnosis.

Serological neopterin detection

The serological neopterin of the 54 AP was continuously examined at the time point of days 0 (diagnosis), 1 (24 h after diagnosis), 2, 4, 8 and 14 by the enzyme linked immunosorbent assay (ELISA) (Tecan Group Ltd., Männedorf, Switzerland). Two millilitre of peripheral venous blood was taken from cases at each time points and the upper serum was separated and stored in a refrigerator at -20°C for testing after centrifugation.

Statistical analysis

The data were calculated by SPSS17.0 software (SPSS, Inc., Chicago, IL, USA). The plot was drawn by

Graphpad Prism 7.0 software (<http://www.graphpad.com/scientific-software/prism/>). The measurement data was analyzed by student-t test. Diagnostic performance was calculated according to the equation of sensitivity=true positive/(true positive + false negative), specificity=true negative/(true negative + false positive). The receiver operating characteristics (ROC) curve was used to evaluate the feasibility of serum neopterin as biomarker for AP severity or death predication.

Results

Serum neopterin levels of different time points between mild and severe groups

The serum neopterin were tested continuously at the time points of 0 (diagnosis), day 1 (24 h after diagnosis), day 2, day 4, day 8 and day 14. The serological neopterin levels were not statistical different from days 0 to 4 between mild

Figure 1: Scatter plot of serological neopterin level at days 8, 14 in different groups.

(A) Scatter plot of serological neopterin level at day 8; (B) Scatter plot of serological neopterin level at day 14.

Figure 2: Serological neopterin dynamic changes from day 0 to day 14 in different groups. (A) Neopterin dynamic changes between mild and severe acute pancreatitis groups. The neopterin levels were 7.10 ± 2.05 (nmol/L) and 5.37 ± 2.22 (nmol/L) for day 8 and day 14 in mild group which was significant lower than those of sever group 4.76 ± 2.68 (nmol/L), 10.71 ± 6.31 (nmol/L) with statistical difference (all $p < 0.05$); (B) The neopterin levels were 7.81 ± 2.65 (nmol/L) and 6.55 ± 2.59 (nmol/L) for day 8 and day 14 in survival group was significant lower than those of death group 18.45 ± 6.82 (nmol/L), 19.09 ± 7.31 (nmol/L), with statistical difference (all $p < 0.05$). * $p < 0.05$.

and SAP groups. However, for days 8 and 14, the serological levels of neopterin in sever group were significant high than those of mild group with statistical difference ($p < 0.05$) (Figure 1). There was also no statistical difference between the death and survival groups form day 0 to day 4 for serum neopterin ($p > 0.05$). However, the serum neopterin was statistical different in the time point of day 8 and day 14 between death and survival groups ($p < 0.05$) (Figure 1). The serological neopterin were gradually increasing from days 0 to 8, but descending at day 14 in mild and survival groups (Figure 2).

Death and severity prediction for neopterin as serological marker in acute pancreatitis

Due to the difference of serological neopterin between groups at the time point of days 8 and 14, the diagnostic performance of serological neopterin as biomarker for AP severity or death risk was evaluated, Table 1. For day 8, the serological neopterin as biomarker for death prediction sensitivity and specificity were 88.89% (95%CI: 51.75–99.72%) and 83.33% (95%CI: 62.62–95.26%) respectively with an area under the curve (AUC) of 0.95 (95%CI: 0.88–1.00). For day 14, the death prediction sensitivity and specificity were 77.78% (95%CI: 33.99–97.19%) and 95.83% (95%CI: 78.88–99.89%) respectively with the AUC of 0.94 (95%CI: 0.87–1.00) (Figure 3).

Discussion

In the present work, we found that the serological neopterin was gradually increased from days 0 to 8, but descending at day 14 in mild and survival groups. However, at day 14, the serum neopterin level was not obviously decreased for death group. This indicated that the serum neopterin level may maintain longer period of time for the increase trend for seriously groups. At days 8 and 14, the serological levels of neopterin in sever and death group were significant high than those of mild and survival groups with statistical difference ($p < 0.05$). Due to the significant difference in days 8 and 14, the neopterin can be used as biomarker for death prediction. We also found that serological neopterin level was elevated with the development of the pancreatitis in the first 4 days. Continuously monitoring the serum neopterin may be helpful for prediction the disease severity and death risk for AP.

It is important to evaluate the risk factors of early death in SAP [10]. The Japanese severity score (JSS) [11], which covers 18 risk factors, is generally used to assess the severity of AP and predict the progress of AP to SAP. However, the JSS was complex for clinical application. In addition, Ranson score [12, 13], BISAP score [14], APACHE II score [15] and SOFA score

Table 1: Diagnosis performance of serum neopterin as biomarker for acute pancreatitis severity and death prediction.

Diagnosis	Sensitivity (95% CI)	Specificity (95% CI)	AUC (95% CI)	Cut off
Severity				
Day 8	63.64% (45.12–79.63%)	66.67% (43.03–85.41%)	0.68 (0.54–0.82%)	7.69
Day 14	69.70% (51.29–84.41%)	61.90% (38.44–81.89%)	0.72 (0.59–0.86%)	5.56
Death				
Day 8	88.89% (51.75–99.72%)	83.33% (62.62–95.26%)	0.95 (0.88–1.00%)	10.34
Day 14	77.78% (33.99–97.19%)	95.83% (78.88–99.89%)	0.94 (0.87–1.00%)	11.91

Figure 3: ROC curve for serum neopterin on day 8 and 14 as biomarker in prediction acute pancreatitis severity and death. (A) ROC for serum neopterin as biomarker in prediction of severity of acute pancreatitis by using neopterin level of day 8; (B) ROC for serum neopterin as biomarker in prediction of severity of acute pancreatitis by using neopterin level of day 14; (C) ROC for serum neopterin as biomarker in prediction of death of acute pancreatitis by using neopterin level of day 8; (D) ROC for serum neopterin as biomarker in prediction of death of acute pancreatitis by using neopterin level of day 14. ROC, Receiver operating characteristic curve.

system [16] were also applied for severity evaluation in patients with AP, but with their own limitations such as low sensitivity or low specificity. In addition, for clinical practice, it was found that the above scoring systems are complex and cumbersome with limited clinical application.

The mortality rate of SAP is high. According to the AP guideline, the mortality rate of AP patients was less than 10% [17]. However, the mortality rate of SAP patients was as high as 36–50% [18].

In the present work, 33 patients with SAP died in nine cases, the mortality rate was 27.3%, which was consistent with the previous publications. The prognosis of patients with SAP is complicated. The high mortality rate is mainly caused by uncontrollable systemic or local inflammation leading to systemic inflammatory response syndrome (SIRS), which leads to multiple organ failure and death. This situation mostly occurs in the early stage (within 2 weeks), so most of the clinical deaths occur about 2 weeks after diagnosis.

At present, sepsis, septic shock and multiple organ dysfunction syndromes are considered to be the dynamic process of pathophysiological changes and clinical severity changes of severe pancreatitis [19, 20]. The essence is that the body's systemic inflammatory response continues to intensify and worsen, and the balance between the metabolic anti-inflammatory responses is destroyed, which is one of the main causes of the occurrence and death of the severe acute pancreas [21, 22]. Neopterin can promote the synthesis and participates in the pathological process of inflammatory reaction [23]. Ci et al. found

that the serum neopterin level of trauma patients and critical ill patients was higher than that of the healthy controls and serum neopterin level was positively correlated with the severity of the disease [24]. Studies have demonstrated that serum neopterin levels in patients with sepsis were elevated and can be used as a promising biomarker for multiple organ dysfunction syndrome (MODS) diagnosis in patients with sepsis [25–27]. In our work, the findings demonstrated that serum neopterin may reflect the severity of AP at a late stage and may be a potential serological maker for death prediction, which was in accordance with the previous studies. However, patients died within 14 days were not included and analyzed. The serum neopterin may be elevated more early in early death patients. Another limitation was that disease severity prediction sensitivity and specificity were not high, which indicated that measuring of neopterin in the beginning may not change any clinical decision. Therefore, combination of several makers included scores such as Ranson score, APACHE II score, Glasgow score and serum neopterin may improve the death prediction value. Therefore further, studies of comprehensive analysis of inflammatory biomarkers and clinical scores are need.

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Informed consent: Informed consent was obtained from all individuals included in this study.

Ethical approval: The work was approved by the Ethical Committee of the Second People's Hospital of Lishui.

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