Research Article

Open Access

Zemin Xiang[#], Yijun Gu[#], Yuanyuan Huang, Lefeng Zhang, Xiaohong Zhang, Huafeng Xu, Hong Liu, Yebo Zhong^{*}

Elevated serum neopterin concentration increases mortality risk in patients with acute pancreatitis

<https://doi.org/10.1515/pteridines-2019-0005>

received November 18, 2018; accepted January 14, 2019.

Abstract: Background The aim of this study was to investigate serum neopterin levels in patients with acute pancreatitis (AP) and its predictive value for mortality in cases of severe acute pancreatitis (SAP).

Methods Eighty-two patients with confirmed acute pancreatitis (AP) were included and divided into two groups, mild acute pancreatitis (MAP, n=51) and severe acute pancreatitis (SAP, n=31) according to severity of disease. From the SAP group, 8 cases went on to develop multiple organ dysfunction syndrome (MODS) and 6 subsequently died. Thirty healthy subjects from routine medical examination were included as the control group. The neopterin serum concentrations were determined and compared between groups.

Results Serum neopterin concentrations from control, MAP, SAP, SAP_Non-MODS, MODS_survival and MODS_death groups were 6.85 ± 2.42 (nmol/L), 0.91 ± 4.83 (nmol/L), 33.11 ± 11.67 (nmol/L), 30.39 ± 9.97 (nmol/L), 36.40 ± 4.48 (nmol/L) and 41.75 ± 15.64 (nmol/L) respectively, with statistical significant difference ($p < 0.05$). The sensitivity and specificity for mortality risk were 66.67% (95%CI:22.28-95.67%) and 88.00% (95%CI:68.78-97.45%) respectively, with area under the ROC curve (AUC) of 0.71 (95%CI:0.50-0.9), under the cut off value of 40.18.

Conclusion: Serum neopterin levels in patients with acute pancreatitis were significantly elevated and correlated with the severity of disease. Neopterin may also be used a serological biomarker of mortality risk in patients with SAP.

Keywords: Acute pancreatitis; Multiple organ dysfunction syndrome; Death; Diagnosis; ROC curve.

Introduction

Acute pancreatitis refers to the activation of pancreatic enzymes by various causes, followed by local inflammation of the pancreas [1, 2]. Systemic inflammatory response syndrome (SIRS) can occur in severe cases and be accompanied by organ dysfunction [3 - 5]. Early diagnosis of severe acute pancreatitis and correct treatment is important in order to improve overall prognosis [6]. Studies have shown that once pancreatic inflammatory factors were activated, pancreatic cells could be damaged and in turn activate a large number of white blood cells, specifically neutrophils, leading to severe pancreatitis [7].

Neopterin is synthesized by human macrophages upon stimulation with interferon-gamma and is indicative of a pro-inflammatory immune status. Neopterin can serve as a marker of cellular immune system activation. Serum neopterin level usually correlates with the extent and activity of the disease and is also a useful maker for treatment response. Correlation between neopterin level and severe infection has been previously reported [8 - 10]. In our present work, we determined neopterin serum levels in patients with AP and evaluated its performance as a potential biomarker for mortality risk in patients with MAP.

Material and methods

Patients

Eighty-two patients with acute pancreatitis were divided into two groups, mild acute pancreatitis (MAP, n=51) and severe acute pancreatitis (SAP, n=31) according to severity of disease. In the SAP group, 8 cases developed multiple organ dysfunction syndrome (MODS) and 6 subsequently

***Corresponding author: Yebo Zhong**, Department of Emergency, Lishui People's Hospital of Zhejiang Province 323000, E-mail: 1012967280@qq.com

Zemin Xiang, Yuanyuan Huang, Lefeng Zhang, Xiaohong Zhang,

Huafeng Xu, Hong Liu: Department of Emergency, Lishui People's Hospital of Zhejiang Province 323000

Yijun Gu: Department of Emergency, Zhuji People's Hospital of Zhejiang Province 311800

[#] Equal contributor: Zemin Xiang and Yijun Gu contributed equally to this work

died. Thirty healthy controls from routine medical examination were included in the healthy control group. The patient inclusion criteria stipulated an age range between 18 to 80, with a confirmed diagnosis of acute pancreatitis. The MAP group contained 30 male and 21 females, with the mean age of 48.6 ± 21.4 . The SAP group contained 18 male and 13 females with a mean age of 52.6 ± 19.6 . The control group, contained 15 males and 15 females with a mean age of 46.8 ± 20.4 . All patients within the AP group had their diagnosis of acute pancreatitis confirmed with written informed consent.

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

Ethical approval: The research related to human use has been complied with all the relevant national regulations, institutional policies and in accordance the tenets of the Helsinki Declaration, and has been approved by the ethics committee of the Lishui People’s Hospital of Zhejiang Province.

Serum neopterin examination

After diagnosis had been confirmed, 5mL venous blood was collected from the patients within 12 hours. Serum neopterin levels were evaluated by enzyme-linked immunosorbent assay (ELISA) (Immuno-Biological Laboratories Co., Ltd), in strict accordance with manufacturer’s instructions provided with the kit.

Statistical methods

Statistical analyses were carried out using SPSS 18.0 statistical software package (IBM, Armonk, NY, USA). Serum neopterin levels were shown by $\bar{x} \pm s$ and comparisons between groups were made based on the variance analysis of the sample mean. Diagnostic or prediction sensitivity and specificity was calculated by the equation of sensitivity=true positive/(true positive+ false negative), specificity=true negative/(true negative+ false positive). The area under the receiver operating characteristic (ROC) curve was used to evaluate the feasibility of serum neopterin as a biomarker for diagnosis of AP or as a predictor of mortality in patients with MODS

Figure 1: Scatter plot of serum neopterin for each group(Tested by ANOVA, F=63.33, p<0.0001).

Table 1: Serum neopterin concentration comparison between groups.

Group	Number	Neopterin (nmol/L)/mean±sd
SAM	31	33.11 ± 11.67
Non-MODS	23	30.39 ± 9.97
MODS_survival	2	36.40 ± 4.48
MODS_death	6	41.75 ± 15.64
MAP	51	10.91 ± 4.83
Control	30	6.85 ± 2.42

Results

Serum neopterin concentration

The serum neopterin concentration of control, MAP, SAP, SAP_Non-MODS, MODS_survival and MODS_death groups were demonstrated by Figure 1. Significant statistical differences were found between each group (p<0.05), Table 1.

Serum neopterin as biomarker for AP diagnosis and mortality risk

The diagnostic performance of serum neopterin as biomarker for AP diagnosis and mortality risk were defined in Table 2, with ROC curves shown in Figure 2.

Table 2: Diagnosis performance and corresponding cut off value.

Diagnosis	Sensitivity(95%CI)	Specificity(95%CI)	AUC(95%CI)	Cut off
AP vs Control	74.39%(63.56%-83.40%)	76.67% (57.72%-90.07%)	0.86 (0.79-0.93)	9.04
SAP vs MAP	87.10% (70.17%-96.37%)	92.16% (81.12%-97.82%)	0.97 (0.94-1.00)	19.68
MODS vs non-MODS	75.00% (34.91%-96.71%)	60.87% (38.54%-80.29%)	0.72 (0.50-0.94)	33.22
Survival vs death	66.67% (22.28%-95.67)	88.00% (68.78%-97.45%)	0.71 (0.44-0.99)	40.18

Figure 2: Receiver operating characteristic (ROC) curves of serum neopterin as biomarker for AP diagnosis and mortality risk (a)ROC curve for AP diagnosis; b) ROC curve for AP severity differential diagnosis; c) ROC curve for MODS diagnosis; d) ROC curve for mortality risk).

Discussion

Neopterin is a low molecular mass compound derived from the metabolism of guanosine triphosphate in the human body [11]. It is an important marker of cellular immunity mediated by the lymphocyte-macrophage axis [12]. Clinical studies have shown neopterin levels in serum and urine of patients with bacterial or viral infections (HIV [13 - 16], HBV [17 - 19]), immune-related diseases [20, 21], tumors [22] and in those with organ transplants [23] were significantly elevated. Longitudinal examination of serum neopterin would allow the monitoring of cellular immune

activation and inflammatory response changes. Neopterin is an essential cofactor of nitric oxide synthase (NOS), an effective vasodilator and an important influencing factor of vasodilation and edema in the initial stages of inflammatory disease.

The occurrence and development of acute pancreatitis is the result of the continuous aggravation and deterioration of systemic inflammatory response, the breakdown of balance between compensatory anti-inflammatory response and the disturbance of immune function. The continued development of this process will eventually lead to MODS. The lymphocyte-macrophage

axis *in vivo* is important for reflecting the activation status of the system. Neopterin is an important marker of cellular immunity mediated by the lymphocyte-macrophage axis which can reflect the severity of AP. Kaufmann [24] reported elevated serum neopterin in AP patients which correlated with disease severity. In addition, Uomo [25] concluded that neopterin may be considered a reliable prognostic indicator for AP patients.

Our findings revealed serum neopterin levels in patients with MAP were slightly higher than those from healthy controls, due to the increased activation of monocytes and macrophages. In patients with SAP, neopterin levels were significantly higher than the control with MAP groups, indicative of severe systemic inflammatory reaction. In addition, significant differences were found between serum neopterin levels from patients that died and those that survived within the severe group, correlating to severity of disease. Using neopterin as a serological biomarker, the sensitivity and specificity for predicting mortality were 66.67% and 88.00% respectively, with area under the ROC curve (AUC) of 0.71. This indicated severity of disease correlated with a higher incidence of MODS and an increased mortality risk.

Conclusion

Early examination of serum neopterin level in patients with acute pancreatitis may help to predict and identify the occurrence of SAP and MODS as early as possible, thus providing the basis for early intervention. Serum neopterin may also be used as a predictor of mortality in patients with SAP. However, it must be noted that the sample size was small, only six patients were included in this study, therefore, the predictive ability of serum neopterin for mortality is limited and further investigation is recommended.

Conflict of interest: Authors state no conflict of interest

References

- Shah AP, Mourad MM, Bramhall SR. Acute pancreatitis: current perspectives on diagnosis and management. *J Inflamm Res.* 2018;11:77-85.
- Algül H, Schmid RM. [Acute pancreatitis: etiology, diagnosis and therapy]. *Med Monatsschr Pharm.* 2009;32:242-7.
- Bhatia M. Acute pancreatitis as a model of SIRS. *Front Biosci (Landmark Ed).* 2009;14:2042-50.
- Hirota M, Sugita H, Maeda K, Ichibara A, Ogawa M. [Concept of SIRS and severe acute pancreatitis]. *Nihon Rinsho.* 2004;62:2128-36.
- Brivet F, Chary I, Jacobs F. [SIRS during acute pancreatitis: an interesting pathophysiologic concept but with poor clinical relevance]. *Gastroenterol Clin Biol.* 2001;25:1534-41.
- Powell JJ, Parks RW. Diagnosis and early management of acute pancreatitis. *Hosp Med.* 2003;64:150-5.
- Zhang S, Peng XG, Liu CC, Liu H, Lu Y. Low-dose dopamine reduces inflammatory factors of acute pancreatitis in rats. *Hepatobiliary Pancreat Dis Int.* 2007;6:646-9.
- Sapa A, Rak A, Wybieralska M, Machoń J, Krzywonos-Zawadzka A, Zawadzki K, et al. Diagnostic usefulness of sCD163, procalcitonin and neopterin for sepsis risk assessment in critically ill patients. *Adv Clin Exp Med.* 2017;26:101-108.
- Tasdelen FN, Aliyazicioglu Y, Tanyel E, Coban AY, Ulger F, Zivalioglu M, et al. The value of neopterin and procalcitonin in patients with sepsis. *South Med J.* 2010;103:216-9.
- Yao YM, Yu Y, Wang YP, Tian HM, Sheng ZY. Elevated serum neopterin level: its relation to endotoxaemia and sepsis in patients with major burns. *Eur J Clin Invest.* 1996;26:224-30.
- Blau N, Niederwieser A. Guanosine triphosphate cyclohydrolase I assay in human and rat liver using high-performance liquid chromatography of neopterin phosphates and guanine nucleotides. *Anal Biochem.* 1983;128:446-52.
- Lissoni P, Rovelli F, Tisi E, Brivio F, Ardizzone A, Barni S, et al. Relation between macrophage and T helper-2 lymphocyte functions in human neoplasms: neopterin, interleukin-10 and interleukin-6 blood levels in early or advanced solid tumors. *J Biol Regul Homeost Agents.* 1995;9:146-9.
- Skogmar S, Schön T, Balcha TT, Sturegård E, Jansson M, Björkman P. Plasma Levels of Neopterin and C-Reactive Protein (CRP) in Tuberculosis (TB) with and without HIV Coinfection in Relation to CD4 Cell Count. *PLoS One.* 2015;10:e0144292.
- Immanuel C, Victor L, Chelvi KS, Padmapriyadarsini C, Rehman F, Iliayas S, et al. Serum neopterin levels in HIV infected patients with & without tuberculosis. *Indian J Med Res.* 2005;121:220-5.
- Wirleitner B, Schroecksnadel K, Winkler C, Fuchs D. Neopterin in HIV-1 infection. *Mol Immunol.* 2005;42:183-94.
- Stein DS, Lyles RH, Graham NM, Tassoni CJ, Margolick JB, Phair JP, et al. Predicting clinical progression or death in subjects with early-stage human immunodeficiency virus (HIV) infection: a comparative analysis of quantification of HIV RNA, soluble tumor necrosis factor type II receptors, neopterin, and beta2-microglobulin. Multicenter AIDS Cohort Study. *J Infect Dis.* 1997;176:1161-7.
- Demirturk N, Demirdal T, Aktepe OC, Aykin N, Orhan S, Cevik F. Serum neopterin levels in patients with HBV infection at various stages. *Hepatogastroenterology.* 2007;54:903-5.
- Kaleli I, Demir M, Cevahir N, Yilmaz M, Demir S. Serum neopterin levels in patients with replicative and nonreplicative HBV carriers. *BMC Infect Dis.* 2006;6:157.
- Kiliç D, Boyunağa H, Kaygusuz S, Akgül E, Al RM, Kenar L, et al. Neopterin levels in nonreplicative HBV carriers. *Hepatol Res.* 2002;24:18.
- El-Lebedy D, Hussein J, Ashmawy I, Mohammed AM. Serum level of neopterin is not a marker of disease activity in treated rheumatoid arthritis patients. *Clin Rheumatol.* 2017;36:1975-1979.

21. Shady MM, Fathy HA, Ali A, Youness ER, Fathy GA. Association of neopterin as a marker of immune system activation and juvenile rheumatoid arthritis activity. *J Pediatr (Rio J)*. 2015;91:352-7.
22. Melichar B, Spisarová M, Bartoušková M, Krčmová LK, Javorská L, Študentová H. Neopterin as a biomarker of immune response in cancer patients. *Ann Transl Med*. 2017;5:280.
23. Zachariah NY, Nikaein A, Gonwa T, Alivazatos P, Klintmalm G, Husberg B. Neopterin: a biochemical marker in the process of organ transplantation. *Transplant Proc*. 1989;21:2294-6.
24. Kaufmann P, Tilz GP, Demel U, Wachter H, Kreijs GJ, Fuchs D. Neopterin plasma concentrations predict the course of severe acute pancreatitis. *Clin Chem Lab Med*. 1998;36:29-34.
25. Uomo G, Spada OA, Manes G, Feola B, Misso S, Cavallera A, Rabitti PG. Neopterin in acute pancreatitis. *Scand J Gastroenterol*. 1996;31:1032-6.