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Protein Z G79A Polymorphism in Patients With Severe Sepsis

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The aim of the study is to investigate whether the presence of a protein Z polymorphism is a risk factor for the development and outcome of sepsis. Sepsis is a clinical syndrome characterized by the presence of systemic signs and symptoms of inflammation. When sepsis leads to organ failure, the term severe sepsis and septic shock is used. The genetic causes of severe sepsis are not fully explained. Protein Z is a vitamin K–dependent glycoprotein and a member of the coagulation cascade. The study included 53 patients with severe sepsis and 70 control healthy volunteers without a familial history of thrombosis. The G79A polymorphism of intron F of the *protein* Z gene was analyzed by the method of polymerase chain reaction–based DNA analysis. The protein Z intron F G79A polymorphism frequencies of the patients and controls were 43.4% and 40%, respectively. Carrying 79 AA genotype could be a risk factor for severe sepsis and septic shock (OR = 4.5, 95% CI: 0.45-46.1), but it could not find any difference between survivor and nonsurvivor groups. They concluded that the frequency of intron F G79A polymorphism of *protein* Z gene was higher in patients than controls, and carrying 79 AA genotype could be a risk factor for severe sepsis and septic shock.

Keywords: sepsis; severe sepsis; *protein* Z gene; polymorphism

Introduction

Protein Z (PZ) is a vitamin K–dependent glycoprotein and a member of the coagulation cascade. Protein Z has an important role in the inactivation of factor Xa. There are some polymorphisms that have been reported in *PZ* gene. Several researches have examined the association between *PZ* gene polymorphisms and thrombophilia such as venous thromboembolism, ischemic stroke, and recurrent pregnancy loss (RPL).[1-5] The genetic causes of severe sepsis are not fully explained. Several genetic polymorphisms have been identified in patients with severe sepsis and septic shock such as *tumor necrosis factor (TNF)* α and *TNF-*β genes, the interleukin 1 family, plasminogen activator inhibitor type 1, factor V Leiden, the Toll-like receptors.[6-10]

Genetic epidemiologic studies suggest that variations in host genetic factors influence the outcome of sepsis. To our knowledge, there is no published study that investigates the association between the *G79A PZ* gene polymorphism in intron F and the development and outcome of sepsis in pediatric patients. Therefore, we planned a study to determine whether the presence of a PZ polymorphism is a risk factor for the development and outcome of sepsis.

Materials and Methods

We evaluated 53 patients with severe sepsis (27 women, 26 men) who met the criteria for sepsis and severe sepsis as outlined previously[11] and 70 control healthy volunteers (35 women, 35 men) without a familial history of thrombosis. The G79A polymorphism of intron F of the *PZ* gene was analyzed according to the previously reported method.[12] Intron F of the *PZ* gene was amplified by polymerase chain

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Table 1. Distribution of PZ Intron F G79A Polymorphism in Patients and Controls

Intron F G79A	n	GG, n (%)	GA, n (%)	AA, n (%)	A Allele, n (%)	*P*	OR (95% CI)
Controls	70	41 (58.5)	28 (40)	1 (1.4)	30 (21.4)	.28	1
Patients	53	27 (51)	23 (43.4)	3 (5.6)*	29 (27.3)		1.38 (0.76-2.48)

NOTES: CI = confidence interval; OR = odds ratio.
*$P = .61$; OR = 4.5 (0.45-46.1).

Table 2. Distribution of PZ Intron F G79A Polymorphism in Patients (Live and Dead) and Controls

Intron F G79A	n (%)	GG (%)	GA (%)	AA (%)	A Allele	*P*	OR
Controls	70 (100)	41 (58.5)	28 (40)	1 (1.4)	30 (21.4)		1
Live	34 (100)	18 (52.9)	14 (41.2)	2 (5.9)*	18 (26.4)	.57	1.32 (0.67-2.58)
Death	19 (100)	9 (47.4)	9 (47.4)	1 (5.2)**	11 (28.9)	.66	1.49 (0.66-3.35)

NOTES: OR = odds ratio.
*$P = .50$; OR = 4.5 (0.38-53.5).
**$P = .82$; OR = 4.5 (0.25-79.8).

reaction (PCR) using primers as forward 5′-TAACACCATAGACAGAGTCCGATATTCGC-3′ and reverse 5′-ATGAACTCGGCATTAGAACATGGTTGGAA-3′. The G79A polymorphism, in intron F, was analyzed by KspAI (Fermentas, Lithuania) restriction endonuclease enzyme digestion. A written consent was obtained from each individual's parents.

Statistical Analysis

Data were analyzed by using Fisher exact test. A *P* value <.05 was considered significant. The odds ratio (OR) and the 95% confidence interval (CI) were calculated.

Results

The study population consisted of 53 patients and 70 controls. Patients were divided into 2 groups: survivors (34 patients) and nonsurvivors (19 patients). The protein Z intron F G79A polymorphism frequencies of the patients and controls were 43.4% and 40%, respectively (Table 1). The distributions of the protein Z G79A polymorphism were as follows: GG genotype in 27 (51%) patients, GA in 23 (43.4%), and AA genotype in 3 (5.6%) of 53 patients. The prevalence of protein Z G79A polymorphism between the patients and control group was found to be statistically insignificant ($P = .28$).

The frequency of the A allele was higher in patients (27.3%) than in controls (21.4%); however, the difference was not significant ($P = .61$). Carrying 79 AA genotype could be a risk factor for severe sepsis and septic shock (OR = 4.5; 95% CI: 0.45-46.1), but it could not find any difference between survivor and nonsurvivor groups (Table 2); $P = .50$, OR = 4.5 (95% CI: 0.38-53.5) and $P = .82$, OR = 4.5 (95% CI: 0.25-79.8), respectively.

Conclusion

There are some contrast findings about the role of intron F G79A polymorphism of the *PZ* gene on the thrombosis or thrombophilia. Some authors reported the 79A allele as a protective factor for the ischemic stroke.[1] Van Goor et al reported no association between the *G79A PZ* gene polymorphism and the occurrence of stroke recently.[13] Eroglu et al reported that there were no differences in PZ intron F G79A polymorphism between cancer patients with and without thrombosis.[14] It was also studied in patients with RPL and was found the isolate presence of the PZ intron F79A allele was protective against RPL.[5]

There are many studies about genetic basis of severe sepsis and septic shock. Some genetic variants influence the risk of severe sepsis in children, but we could not find any published data about the *G79A PZ* gene polymorphism in progression of severe sepsis in pediatric patients. In this study, we found that the prevalence of PZ G79A polymorphism between the patient and control groups was statistically insignificant ($P = .28$; Table 1). However, the frequency of the A allele was higher in patients than in controls.

Patients with 79AA allele had a 4.5-fold risk of severe sepsis (95% CI: 0.45-46.1), but we could not find any effect on mortality (Table 2).

In conclusion, several genetic polymorphisms have been identified in patients with severe sepsis, and they influence the outcome of sepsis and septic shock. Our findings suggest that the frequency of the A alleles of the G79A polymorphism was higher in patients than control, and carrying 79 AA genotype could be a risk factor for severe sepsis and septic shock. However, it is necessary to compare patients with severe sepsis to those without severe sepsis in the same underlying disease. Further studies are necessary with a larger patient group to confirm these findings.

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