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ORIGINAL

Endothelial protein C receptor polymorphisms and risk of severe sepsis in critically ill patients

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Abstract *Purpose:* Endothelial protein C receptor (EPCR) is expressed mainly in endothelial cells and is involved in regulation of the cytoprotective and anticoagulant pathways of protein C. We assessed whether haplotypes in the EPCR gene modify the risk of severe sepsis and/or septic shock (SS/SS) development in critically ill patients. *Methods:* Three polymorphisms in the EPCR gene were genotyped in 389 Caucasian critically ill patients, hospitalized in the intensive care units of two major hospitals in Athens, Greece. Multivariate logistic regression analysis controlling for age, acute physiology and chronic health evaluation (APACHE) II and sequential organ failure assessment (SOFA) scores, sex, and diagnosis was performed to determine the effect of haplotypes H1 and H3 in the EPCR gene on the development of SS/SS. *Results:* H2 carriers versus all other genotypes combined had a nonsignificant excess of SS/SS ($p = 0.087$). SS/SS occurred in 38.8 % of critically ill patients carrying minor alleles belonging to both H1 and H3 haplotypes, in 58.0 % of H1 carriers, 64.3 % of H3 carriers, and 65.2 % of patients carrying all common alleles (H2). Compared with H2 carriers, the odds ratios (OR) for developing SS/SS were 0.34 [95 % confidence interval (CI) 0.16–0.76, $p = 0.008$] for simultaneous H1 and H3 carriers, 0.65 (95 % CI 0.37–1.13, $p = 0.123$) for H1 carriers, and 0.82 (95 % CI 0.39–1.70, $p = 0.590$) for H3 carriers. *Conclusions:* Our results indicate that simultaneous carriers of minor alleles belonging to both the H1 and H3 haplotypes may be at reduced risk of developing SS/SS in this cohort of critically ill patients.

Keywords EPCR · Severe sepsis · Haplotypes · Critically ill · Septic shock

Introduction

Sepsis, a systemic inflammatory response to infection, is the most common cause of death among hospitalized patients in noncardiac intensive care units (ICUs) with mortality of 30–50 % [1, 2]. Severe sepsis is distinguished from sepsis by the presence of organ dysfunction and the associated reduction in survival [3–5]. Traditionally, risk stratification and outcome prediction of sepsis patients have been based on disease severity scales, but lately emphasis has been placed also on genetic markers. These are hoped to deliver not only mechanistic understanding of pathogenesis, but also useful information regarding identification of particularly susceptible individuals.

The protein C (PC) system is a major participant in sepsis and activation of coagulation, and it is both a consequence of and a contributor to ongoing injury [6]. Protein C activation has been shown to be critical to host defense against septic shock [7]. Activated protein C (APC) modulates coagulation by proteolytically inactivating cofactors Va and VIIIa. In addition, it promotes endothelial cell survival and barrier integrity. Collectively, these properties of APC constitute important countermechanisms against the septic process [8, 9]. Conversion of protein C to APC requires the presence of endothelial protein C receptor (EPCR) [10].

EPCR is a 46-kDa type I transmembrane protein constitutively expressed by endothelial cells of larger vessels, liver sinusoids, monocytes, leukocytes, and several tumor cells [11–16].

Deletion of EPCR or blocking of APC binding to EPCR in animals with experimental sepsis exaggerates the host response to lipopolysaccharide (LPS), resulting in increased mortality [17, 18], suggestive of a critical role of EPCR in host defense against bacterial sepsis. Even though human cases of absent EPCR expression have not been reported, there have been reports of functionally relevant variations in the EPCR gene, among which are a rare 23-bp insertion in exon 3 (position 6367), a polymorphism in intron 2 (C6333T), a polymorphism in the 5′-untranslated region (UTR) (C1651G), a polymorphism in exon 4 (A6936G) that predicts an amino acid change (Ser219Gly) in the transmembrane region of the receptor, and a polymorphism in the 3′-UTR (G7999A) (numbering according to GenBank accession number AF106202) [19–21].

Based on these findings, human EPCR genotypes may be divided into four haplotypes, namely H1, H2, H3, and the very rare H4, three of which (H1, H3, and H4) contain one or more haplotype-specific single-nucleotide polymorphisms (htSNPs). One haplotype (H2) consists of the common alleles of all haplotype-tagging (ht)SNPs [20, 21]. The functional importance of these mutations has been investigated in patients with venous and coronary thrombosis [22–27], but their role in severe sepsis or septic shock is unknown.

To investigate the role of EPCR mutations as predisposing factors to severe sepsis or septic shock development, we undertook a genotyping study of EPCR haplotypes in critically ill patients, controlling for potential confounding factors, such as disease severity, age, sex, and diagnostic category.

Patients and methods

Ethical standards statement

The study was approved by the Research Ethics Committees of both participating hospitals, and all procedures carried out on our patients were in compliance with the World Medical Association Helsinki Declaration. Informed written consent was obtained from all patients' next-of-kin prior to any study procedure.

Study population

A power calculation indicated that a sample size of 389 patients would ensure a power of 80 % to detect an approximately 15 % relative change in risk for developing severe sepsis and/or septic shock (SS/SS) when comparing the H2 haplotype with the remaining haplotypes combined, at a significance level of 5 %. Prior to enrollment, we screened all consecutive admissions to two ICUs in the Athens area, Greece over a 12-month period for eligibility. The catchment areas of the two hospitals are the same, and there were essentially no differences in demographic characteristics and diagnostic categories of patients admitted to the two ICUs. Exclusion criteria were as follows: age <18 years, malignancies, no need for intubation and mechanical ventilation during the ICU stay, contagious diseases (human immunodeficiency virus, hepatitis), and oral intake of corticosteroids at an equivalent dosage of ≥1 mg/kg prednisone/day for a period of more than 1 month. Out of 576 subjects screened, 389 subjects (as it happened all Caucasians; 262 males and 127 females) were enrolled in the study based on the above-mentioned criteria and consent to participate. The patients recruited into the study suffered from medical, surgical, and trauma-related pathologies. Medical conditions included infections and nonsurgical conditions of the respiratory, gastrointestinal, urinary, and hematopoietic systems, while surgical conditions included thoracic surgery (excluding cardiothoracic procedures), neurosurgery, and abdominal surgery; trauma was considered a separate category. Clinical data and blood samples were obtained from all patients enrolled. Following study enrollment, baseline (upon ICU admission)

anthropometric data (height, weight) and detailed organ system-oriented medical history were recorded. A venous blood sample was drawn and processed as described in "Blood collection." The patients were followed daily, and several clinical and laboratory parameters were recorded, including body temperature, arterial blood pressure, arterial blood gases, urine output, complete blood cell count and differential, serum chemistries, presence and source of infection, administration of vasopressors, mechanical ventilation, and renal replacement therapy. Patients were followed until discharge from hospital or death. The diagnoses of severe sepsis and septic shock were based on the criteria set by the American College of Chest Physicians/Society of Critical Care Medicine (ACCP/SCCM) consensus conference [28], while diagnosis of acute lung injury/acute respiratory distress syndrome (ALI/ARDS) relied on the criteria set by the American–European consensus conference (AECC) on ARDS [29]. Patients were assigned to groups based on the presence ($N = 228$) or absence ($N = 161$) of SS/SS at any point during their ICU stay.

Blood collection

Venous blood was collected during the first 24 h post ICU admission in tubes containing 0.129 M (3.8 %) trisodium citrate. High-molecular-weight DNA was isolated from leukocytes using standard methods (Nucleospin Blood L from Macherey–Nagel) and stored at −20 °C.

Genetic analysis

Genotyping was performed by the 5′-nuclease/TaqMan assay [30] using the polymerase chain reaction with fluorescent allele-specific oligonucleotide probes (assay-by-design/assay-on-demand; Life Technologies Corporation, CA, USA) and the Type-it Fast SNP probe PCR kit from Qiagen (QIAGEN GmbH, Hilden, Germany). Fluorescence endpoint reading for allelic discrimination was performed on a PTC-200 (MJ Research Inc., Waltham, MA, USA) using Opticon Monitor 2 software. We chose three htSNPs to cover the three common haplotypes (H1, H2, and H3) in the EPCR gene (Fig. 1). The frequency of the H4 haplotype is only 0.05, thus we assumed that patients not carrying minor alleles belonging to the H1 or H3 haplotype most likely belong to the H2 haplotype. Specifically, C1651G (rs2069940) and A6936G (rs867186) were used to define haplotype H3, while C6333T (rs2069952) corresponded to haplotype H1. The presence of the minor allele determines the respective haplotype, while absence of all three minor alleles determines H2 [21].

In a group of 106 healthy Caucasian volunteer subjects, the frequency of the studied alleles was 0.34 for the 6333C allele (H1 haplotype), 0.05 for the alleles belonging to the H3 haplotype (1651G and 6936G alleles), and 0.61 for the H2 haplotype.

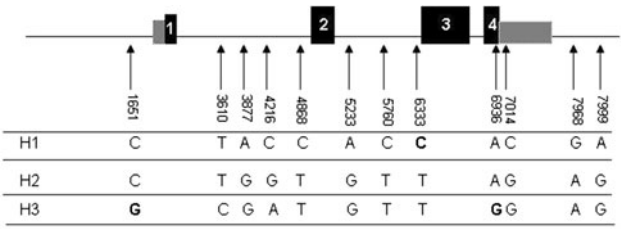

Fig. 1 The three EPCR gene haplotypes. The three polymorphisms studied were C1651G (H3), A6936G (H3), and T6333C (H1) (*bold*). Nucleotides are numbered according to the GenBank sequence (accession number AF106202). Adapted from [20, 21]

Statistical analysis

Descriptive data are presented as mean ± standard deviation (SD), median (25–75 % interquartile range), and percentages of total (%). For continuous variables, the Mann–Whitney test was used for two-group comparisons. Associations between qualitative variables were examined by the chi-square test. Multiple logistic regression analysis was performed with the haplotypes used as key predictor variables (categorical: H2, only H1, only H3, both H1 and H3 present) and development versus no development of "severe sepsis and/or septic shock" as a binary outcome, controlling for APACHE II score (continuous variable), SOFA score (continuous variable), age (continuous variable), diagnosis (categorical: medical, surgery, and trauma) and sex (categorical: female versus male). The categorical variables were introduced as $k - 1$ dummy variables in the model (where k is the number of categories per variable). For continuous variables, the log-linear relationship with the outcome prevalence was checked. The best logistic regression model was selected by the backward elimination method [likelihood ratio (LR) method]. The Hosmer–Lemeshow statistic was calculated to assess the goodness-of-fit of model to data. Two-tailed p values <0.05 were considered as statistically significant.

Results

Characteristics of the study population

Among the 389 critically ill patients studied, 262 were men (67.5 %) and 127 were women (32.5 %). The mean patient age in our sample was 59 years (range 18–94 years). The median (25–75 % interquartile range) APACHE II and SOFA scores at ICU entry were 20

Table 1 Characteristics of the 389 critically ill patients enrolled in the study

Age (years)	59 ± 19
Sex	
Male	262 (67.35 %)
Female	127 (32.65 %)
Diagnosis	
Medical	199 (51.16 %)
Surgical	132 (33.93 %)
Trauma	58 (14.91 %)
SS/SS	228 (58.61 %)
Site of infection	
Lung	100 (43.86 %)
CNS	16 (7.02 %)
Abdomen	33 (14.47 %)
Skin	7 (3.07 %)
Urinary tract	3 (1.32 %)
Blood	69 (30.26 %)
APACHE II score	20 (14–25)
SOFA score	8 (5–10)
Haplotype[a]	
Only H1 (H1H1/H1H2)	169 (43.44 %)
Only H3 (H3H2/H3H3)	56 (14.40 %)
H1 and H3 (H1Hx/H3Hx)	49 (12.60 %)
H2 (no H1 no H3)	115 (29.56 %)

Data expressed as number of patients (*N*) and percentages of total (%), mean ± SD, or median (25–75 % interquartile range)

APACHE II and SOFA scores were calculated upon admission to the intensive care unit

CNS central nervous system, *APACHE* acute physiology and chronic health evaluation, *SOFA* sequential organ failure assessment, *SS/SS* severe sepsis and/or septic shock

[a] "H2" indicates that neither H1 nor H3 haplotype-specific alleles were detected (the frequency of H4, at 0.05, is negligible). "H1 and H3" refers to carriers of minor alleles belonging to both the H1 and H3 haplotypes

Table 2 Allelic and genotypic frequencies of the single-nucleotide polymorphisms in the SS/SS-positive and SS/SS-negative patients

EPCR polymorphism position	SS/SS positive ($N = 228$) (%)	SS/SS negative ($N = 161$) (%)
6333 T/C		
T allele	310 (67.98)	192 (59.62)
C allele	146 (32.02)	130 (40.38)
TT genotype	111 (48.68)	60 (37.27)
TC genotype	88 (38.60)	72 (44.72)
CC genotype	29 (12.72)	29 (18.01)
1651 C/G		
C allele	398 (87.28)	270 (83.85)
G allele	58 (12.72)	52 (16.15)
CC genotype	173 (75.88)	111 (68.95)
CG genotype	52 (22.81)	48 (29.81)
GG genotype	3 (1.31)	2 (1.24)
6936 A/G		
A allele	398 (87.2)	270 (83.85)
G allele	58 (12.77)	52 (16.15)
AA genotype	173 (75.88)	111 (68.95)
AG genotype	52 (22.81)	48 (29.81)
GG genotype	3 (1.31)	2 (1.24)

H1 haplotype: 6333TC or CC; H3 haplotype: 1651CG or GG and/or 6936AG or GG. Both H3 polymorphisms (1651 C/G and 6936 A/G) were detected in the same patients

EPCR endothelial protein C receptor, *SS/SS* severe sepsis and/or septic shock

(14–25) and 8 (5–10), respectively, resulting in ICU mortality of 40.5 %. Approximately half of the study sample consisted of surgical or trauma patients, while the other half had medical diagnoses. SS/SS occurred in 228 patients (59 %) of the total patient cohort. Out of these, 192 patients had severe sepsis or septic shock at baseline, while 36 developed SS/SS during their ICU stay. The most common sites of infection were the lung (43.86 %) and blood (30.26 %). ALI/ARDS was present in 40 % of patients. Table 1 lists basic characteristics of the patient population, including distribution by haplotype.

EPCR polymorphisms and SS/SS

All patients were successfully genotyped, and the genotypes for all three htSNPs could be determined in all 389 patients (Fig. 1). Allelic and genotypic frequencies of the 6333C (H1 haplotype), 1651G and 6936G (H3 haplotype) SNPs in EPCR are presented in Table 2 for this critically ill study population. The frequency of the H1 haplotype was higher in the SS/SS-negative group than in the SS/SS-positive group ($p < 0.05$). The frequency of the H3 haplotype tended to be higher in the SS/SS-negative group. The distributions of the genotypes were in Hardy–Weinberg equilibrium (chi-square test, $p > 0.05$).

Table 3 presents the distribution of patients by presence of SS/SS on the one hand, and the various genotype combinations, as well as key clinical and demographic variables on the other. Although a 2 × 2 contrast of H2 patients versus all other genotypes grouped together generated a p value of 0.087, the distribution of SS/SS-positive and SS/SS-negative patients across genotypes was statistically significantly different (chi-square with three degrees of freedom = 10.79, $p = 0.013$). Thus, 39 % of patients carrying minor alleles belonging to both H1 and H3 haplotypes developed SS/SS, whereas in the remaining genotypes approximately 60 % of patients per genotype developed SS/SS.

To clarify associations between these haplotypes and SS/SS, we performed logistic regression analysis. On univariate analysis, the presence of the H1 haplotype (compared with non-H1 carriers) was associated with reduced risk of developing SS/SS. For the H1 haplotype the odds ratio was 0.63 (95 % CI 0.42–0.95, $p = 0.026$), while for H3 the odds ratio was 0.71 (95 % CI 0.45–1.11, $p = 0.130$). We proceeded to an exploratory analysis to study the effect of the simultaneous presence of minor alleles belonging to the EPCR H1 and H3 haplotypes (compared with the absence of these minor alleles) on the risk of SS/SS.

Table 3 Comparison of the characteristics of the SS/SS-positive group of patients versus the SS/SS-negative group

Parameter	SS/SS positive	SS/SS negative	*p*-Value
Number of patients (*N*)	228	161	
Presence of H2[a] (6333T, 1651C, and 6936A alleles)	75 (65.2 %)	40 (34.8 %)	0.087
Presence of H1 and/or H3 alleles (6333C and/or 1651G and 6936G alleles)	153 (55.8 %)	121 (44.2 %)	
Genotype combination			
Presence of only H1 allele (6333C allele)	98 (58.0 %)	71 (42.0 %)	
Presence of only H3 alleles (1651G and 6936G alleles)	36 (64.30 %)	20 (35.70 %)	0.013
Presence of both H1 and H3 alleles (6333C, 1651G, and 6936G alleles)	19 (38.80 %)	30 (61.20 %)	
Presence of neither H1 nor H3 alleles (H2) (6333T, 1651C, and 6936A alleles)	75 (65.20 %)	40 (34.80 %)	
Diagnosis			
Medical	134 (67.34 %)	65 (32.66 %)	
Surgical	71 (53.79 %)	61 (46.21 %)	<0.001
Trauma	23 (39.65 %)	35 (60.35 %)	
APACHE II score	22 (17–27)	16 (12–22)	<0.001
SOFA score	9 (7–11)	6 (4–9)	<0.001
Age (years)	62 ± 18	56 ± 20	0.001
Sex			
Male	150 (57.30 %)	112 (42.70 %)	0.434
Female	78 (61.40 %)	49 (38.60 %)	

Data expressed as number of patients (*N*) and percentages of total (%), mean ± SD, or median (25–75 % interquartile range). The Mann–Whitney or chi-square test was used, as appropriate. SS/SS positive, group of patients who developed SS/SS during any time point during their stay in the intensive care unit (ICU); SS/SS negative, group of patients who did not develop SS/SS during their ICU stay

SS/SS severe sepsis and/or septic shock

[a] "H2" indicates that neither H1 nor H3 haplotype-specific alleles were detected (the frequency of H4, at 0.05, is negligible)

Table 4 presents the odds ratios (and 95 % confidence intervals) derived from the univariate analysis and the multiple logistic regression analysis for the development of SS/SS per studied genotype, compared with the absence of both H1 and H3 haplotypes. On univariate analysis, the odds ratio for developing SS/SS among patients carrying minor alleles belonging to both H1 and H3 haplotypes compared with patients carrying all common alleles (H2) was 0.34 (95 % CI 0.17–0.67, $p = 0.002$). The multivariate models controlled for potential confounding factors, notably APACHE II and SOFA scores at ICU entry, age, sex, and diagnostic category. The results from the multivariate models indicated that simultaneous carriage of minor alleles belonging to the H1 and H3 haplotypes is statistically significantly associated with a protective effect on the risk of critically ill patients for developing SS/SS (adjusted OR = 0.34, CI 0.16–0.76, $p = 0.008$).

Discussion

To the best of our knowledge this is the first study to investigate the association of the three commonest haplotypes in the EPCR gene with the risk of developing SS/SS in ICU patients. We reasoned that EPCR mutations could be implicated in severe sepsis or septic shock development by modulating the levels of APC and its downstream cytoprotective and anticoagulant properties. Increased levels of APC, as can be expected with H1 haplotypes, could be protective against severe sepsis, as suggested by previous studies on diverse vascular diseases [24–26]. As opposed to H1, H3 has been shown to be a candidate risk factor for deep vein thrombosis, possibly by increasing the levels of soluble EPCR and promoting the procoagulant effects due to APC sequestration [20, 27, 31]. A soluble form of EPCR sequesters APC and may act in a prothrombotic fashion [32]. Medina and coworkers [24] have further shown that these two haplotypes have an additive protective effect in myocardial infarction, probably due to the association of H1 with increased APC plasma levels and H3 due to its association with high sEPCR levels, but the mechanism remains unknown. Another related study has shown that the rare 23-bp insertion was significantly more common among patients with severe sepsis [33], probably due to the fact that the truncated protein product of this mutation is not localized on the cell surface and does not bind APC [19]. Animal data in sepsis have highlighted a role of EPCR in the control of thrombosis and inflammation and in the host response to Gram-negative infection. Inhibition of protein C binding to EPCR exacerbates the baboon response to sublethal *Escherichia coli*, converting it into a

Table 4 Effect on the development of SS/SS of the presence of alleles belonging only to the H1 or only to the H3 haplotype or simultaneously to both the H1 and H3 haplotypes compared with the presence of all common alleles (H2)

Genotype combination	Percentage of patients with SS/SS	Crude OR (95 % CI)	Adjusted OR (95 % CI)
H2 (no H1 no H3) 6333TT/1651CC/6936AA	65.20 %	Referent	Referent
H1 haplotype only 6333CT or 6333CC/ 1651CC/6936AA	58.0 %	0.74 (0.45–1.20) $p = 0.221$	0.65 (0.37–1.13) $p = 0.123$
H3 haplotype only 6333TT/ 1651GC or 1651GG/ 6936GA or 6936GG	64.30 %	0.96 (0.49–1.87) $p = 0.905$	0.82 (0.39–1.70) $p = 0.590$
Both H1 and H3 haplotypes present 6333CT or 6333CC/ 1651GC or 1651GG/ 6936GA or 6936GG	38.80 %	0.34 (0.17–0.67) $p = 0.002$	0.34 (0.16–0.76) $p = 0.008$

Best logistic regression model selected by the backward elimination method. Odds ratios derived either from univariate analysis or from multiple logistic regression model controlling for APACHE II score, SOFA score, age (continuous variables), diagnosis, and sex (categorical variables)
OR odds ratio, *CI* confidence interval, *SS/SS* severe sepsis and/or septic shock

lethal syndrome of disseminated intravascular coagulation (DIC) and exuberant inflammation [17]. In animals challenged with lethal levels of *E. coli*, infusion of APC was able to block microvascular thrombosis and leukocyte activation [34], indicating crosstalk between both pathways via APC and other mediators. In humans, however, the originally demonstrated efficacy of recombinant human activated protein C (drotrecogin alfa activated) for treatment of severe sepsis and septic shock [35] was not recently confirmed [36]; this led to drug withdrawal and modification of the latest Surviving Sepsis Campaign guidelines [5, 37].

To assemble a patient cohort with high likelihood to manifest SS/SS and sufficient power to detect potential haplotype differences, we included 389 Caucasian patients admitted to two ICUs in the Athens area, Greece. The frequency of the H3 haplotype in our critically ill cohort was higher than that in the healthy volunteer group, leading us to genotype two H3 htSNPs to avoid any genotyping errors. Similar findings have been described in thrombosis-related studies where patients experiencing thrombotic events appeared to also exhibit higher H3 allele frequencies than the corresponding healthy controls [20, 27].

Using multiple logistic regression analysis and adjusting for potential confounding factors, we found that the simultaneous presence of alleles belonging to the H1 and the H3 haplotypes was associated with significantly lower risk of developing SS/SS. We can only speculate as to the mechanism. In this regard, the H1 haplotype consists of a combination of T at nt 3787, A at nt 3877, C at nt 4216, C at nt 4868, A at nt 5233, C at nt 5760, C at nt 6333, C at nt 7014, G at nt 7968, and A at nt 7999, while H3 differs from the H2 haplotype at four nucleotide positions (G at nt 1651, C at nt 3610, A at nt 4216, and G at nt 6936). Therefore, any of these nucleotides may be responsible for the low rates of severe sepsis or septic shock in carriers of alleles belonging to both H1 and H3 haplotypes. The molecular mechanism by which the presence of both H1 and H3 haplotypes decreases the risk of developing severe sepsis or septic shock, however, remains to be identified. Several polymorphisms defining the H1 and H3 haplotypes are located within intronic regions. The SNP affecting nt 1651 is located in the 5′-UTR and the nt 6333 in an intron. Polymorphisms in promoter regions, intronic enhancer regions, or 3′-UTR that augment transcription efficiency and protein synthesis have been described in other genes [38]. Alterations in DNA sequence may affect RNA stability, protein function, or other cellular mechanisms. Hence, developing high-throughput approaches for analysis of alternative mechanisms by which SNPs can cause disease remains one of the challenges for genomic research.

Although we were not able to document the primary hypothesis, that is, that carriers of the common alleles (assessed as H2) were at significantly increased risk of SS/SS in comparison with all other genotypes combined, we found a statistically significant deficit of SS/SS among carriers of minor alleles belonging to both H1 and H3 haplotypes, while carriers of alleles belonging to haplotype H1 only or H3 only had no significant associations with SS/SS risk. A limitation of our study is that the power calculated in advance, although sufficient to evaluate an overall association (Table 3), was not adequate to document possible effects of alleles belonging only to H1 or only to H3. Therefore, pending future investigations, we cannot confidently exclude the possibility of a false-positive result concerning the effect of the simultaneous presence of alleles belonging to both the H1 and H3 haplotypes, but this also applies to all but the largest genetic association studies. Moreover, residual statistical concerns would be best addressed through the generation

of functional data. Strengths of our study are the employment of state-of-the-art technology for genotyping and the fact that the diagnosis of severe sepsis and septic shock in both participating hospitals was based on criteria set in international guidelines.

The potential of genetic biomarkers for prognostic purposes is well known for specific, mostly uncommon/rare inherited disorders, but is also emerging for sepsis [39, 40]. In this context, our study provides new evidence that EPCR polymorphisms may be implicated in determining sepsis severity; that is, in our cohort of critically ill patients, those who carried alleles belonging to both H1 and H3 haplotypes in the EPCR gene had reduced risk of developing SS/SS. These findings generate hope that successful use of modern molecular diagnostics could enable rapid identification of particularly susceptible or less susceptible individuals, leading to tailored therapeutic approaches.

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Conflicts of interest The authors declare that they have no competing interests.

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