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Research Article

Effect of CD14 −260C>T polymorphism on the mortality of critically ill patients

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Summary The CD14 receptor seems to be an important part of the innate immune system. A mutant CD14 can produce a reduced signal in response to infection, as a result of which an adequate inflammatory innate response is not induced, leading to a systemic infection. Defects in the innate immunity increase patient susceptibility to systemic infections and can produce a deregulated inflammatory response causing sepsis, organ failure or death in critically ill patients. We evaluated the CD14 −260C>T polymorphism genotyping as a genetic tool for risk evaluation of critically ill patients admitted to an intensive care unit (ICU) in Southern Brazil. We monitored the patients daily during their entire ICU and post-ICU (hospital) stay (measured from the ICU admission day to a maximum of 224 days). A total of 85 patients, aged 19–95 years (mean = 56 years, median = 58 years), were included in this study. Patient mortality was 58.8%. The genotypic (TT = 0.27, TC = 0.41, CC = 0.32) and allelic (T = 0.48, C = 0.52) frequencies did not differ from the values expected by the Hardy–Weinberg model and genotype distribution was random for all clinical characteristics at ICU admission. We found a statistically significant difference favouring the survival of patients with TT genotype ($P = 0.042$), suggesting that this CD14 gene polymorphism could be a candidate for further study in the search for a complementary prognostic tool for patient risk evaluation. Our study describes, for the first time, the effect of the CD14 gene polymorphism in critically ill Brazilian patients. Our data suggest that patients carrying the TT genotype have a better survival outcome.

Key words: critically ill patient, genetic risk factor, −260C>T CD14 polymorphism.

Introduction

Critically ill patients are those who require sophisticated monitoring procedures that involve frequent clinical, physiological, and biochemical data collection. Analyses of the human genome have suggested the use of genetic information as a complementary prognostic tool for many severe illnesses.[1,2] Rapid genetic screening of patients may enable physicians to tailor treatment to meet the specific needs of individual patients. Although several genes play an important role in critical illness, certain genes have a greater and more direct influence. We propose that this could be the case with CD14.

CD14 is a 53–55 kDa glycoprotein with a high-affinity receptor for LPS that is found anchored to the surface (membrane-bound CD14 or mCD14) of monocytes, macrophages, neutrophils and other non-myeloid cells such as endothelial and epithelial cells,[3] by a glycosyl-phosphatidyl-inositol tail, or is found in a soluble form (sCD14).[4] In both forms, the CD14 receptor seems to be an important part of the innate immune system.[5] LPS from Gram-negative bacteria are recognized by CD14 that activates, through the Toll-like receptor (TLR)-4 and nuclear factor (NF)-κB pathways, the pro-inflammatory cytokine gene expression (IL-1, TNF-α, IL-8, IL-12, IL-6) and activates the innate immunity against the infection.[6,7] Therefore, defects in the innate immunity could increase the susceptibility to systemic infections and produce a deregulated inflammatory response, causing sepsis, organ failure and death in critically ill patients.

The CD14 human gene is located in the locus 5q23-31 occupying approximately 1500 bp organized into two exons that code for a protein of 375 residues.[8,9] In the gene promoter sequence, four regions that interact with specific nuclear proteins of monocytes and three other regions where the transcription factor Sp1 binds were identified.[10] A single nucleotide polymorphism (SNP) has been detected in the CD14 promoter. A promoter CD14 SNP presents a transition from cytosine (C) to thymine (T) in the position −260 from the translation starting site of the gene.[10] This mutation is found near the recognition site for the transcription factor Sp1 and interferes in the transcriptional capacity of the gene.[10] It has been observed that TT homozygous individuals present increased levels of mCD14 and sCD14, in comparison to individuals who carry the C allele.[11–15] The TT genotype can result in a differential susceptibility to the development of severe diseases,[11,16,17] although this association has not been verified in all populations studied.[18–21]

The aim of this study was to verify whether the differential inheritance of CD14 variants for the −260C>T can be a genetic marker candidate for the prognosis of sepsis, organ failure and outcome of critically ill patients.

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Materials and methods

Study design and approval

This is a cross-sectional study of random, unrelated, critically ill patients admitted to the general intensive care unit (ICU) of the São Lucas Hospital (HSL) of the Catholic Pontifical University of Rio Grande do Sul (PUCRS), Brazil, between 1 May 2002 and 28 November 2002. Everyday of the study period, one patient recently admitted to the ICU was invited to participate in the study. When the patient was unconscious or not in a condition to give consent for medical reasons, consent was obtained from the patient's family. The patients were included if they fulfilled the following criteria: (i) should have signed the consent term and (ii) should be 18 years and older. The PUCRS Research Ethics Committee approved this study in April 2002 (process # 01/01088 and 838/03-CEP).

Study subjects

A total of 236 patients were admitted to the ICU at HSL/PUCRS during the study period. Eighty-eight (37.3%) patients consented to participate in the study. The patients were enrolled in the following distribution by month: May, 20; June, 12; July, 9; August, 15; September, 9; October, 8; November, 15. Three patients were excluded because of lack of data. A total of 85 critically ill adult patients (45 men and 40 women) were included in this study. We monitored the patients daily during their entire ICU and post-ICU (hospital) stay (measured from the ICU admission day to a maximum of 224 days). All the patients were southern Brazilians and all the personnel involved in patient care were blinded to the selection process and genotyping results.

Blood collection

A 5-mL blood sample was collected in a sterile system with EDTA from each patient at ICU admission and refrigerated at 4°C or frozen at −20°C until DNA extraction. Blood samples for DNA were obtained in the period from May to November 2002.

DNA analysis

Genomic DNA was extracted from leucocytes by a standard method.[22] The biallelic −260C>T polymorphism in the promoter of the CD14 gene was determined for the 85 subjects according to the PCR–RFLP method of Hubacek *et al.*[11] PCR was carried out with a total volume of 50 μL with about 0.5 μg of genomic DNA, 1 U *Taq* DNA Polymerase in *Taq* Buffer (Invitrogen Brasil, São Paulo, SP, Brazil), and final concentration of each dNTP was 0.2 mmol/L and 2 mmol/L $MgCl_2$. The promoter of the CD14 receptor gene was amplified using 0.6 μmol/L of each primer CD14F, 5′-TTG GTG CCA ACA CAT GAG GTT CAG-3′; and CD14R, 5′-TTC TTT CCT ACA CAC GGC ACC C-3′ (Life Technologies do Brasil, INVITROGEN) in a PTC-100 thermocycler (MJ Research, Watertown, MA, USA) as follows: an initial denaturation at 94°C for 10 min followed by 35 cycles at 94°C for 1 min, 65°C for 35 s and 72°C for 1 min. The final extension step was prolonged to 10 minutes. The 560-bp PCR-amplified product (40 μL) was cleaved in appropriated buffer with 10 U of *Hae*III (GibcoBRL-Life Technologies, Rockville, MD, USA) in a total volume of 50 μL at 37°C for 3 hours.

Each individual's genotype was determined by electrophoresis in agarose gel (2%). Genotyping was based on the following information: (i) in the 560-bp PCR-amplified product, all the individuals present a restriction site for *Hae*III in the -417 to -414 position (considering the beginning of the translation) and all the amplified segments are digested at this restriction site; (ii) homozygotic CC individuals present, in both chromosomes, the C nucleotide in the −260 position of the gene promoter, where the restriction site GGCC is located, and after digestion, CC individuals present three fragments—204, 201 and 155 bp; (iii) homozygotic TT individuals present in both chromosomes a nucleotide substitution C→T at the CD14 promoter −260 position, which eliminates one recognition site for *Hae*III showing, as a result of the digestion, only two DNA fragments of 359 and 201 bp; and (iv) heterozygotic TC individuals show four fragments of DNA with 359, 204, 201 and 155 bp (Fig. 1).

The CD14 promoter DNA sequence and both −260C>T CD14 alleles are registered in the EMBL database (GI: 4557416 and GenBank accession numbers X74984 and U00699).

Data collection

For the diagnosis of sepsis and septic shock, we used the American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference criteria.[23] To measure mortality, we used the time in days, instead of the usual categories of until 21 or 28 days mortality, because we consider it more precise and informative. For illness severity evaluation, we used the Acute Physiology and Chronic Health Evaluation II (APACHE-II) score obtained on ICU admission day. For organ dysfunction evaluation, we used the Sequential Organ Failure Assessment (SOFA) score obtained during the first 7 days from the ICU admission. For those patients with multiple ICU admission during the study period, only data from the first entrance was considered.

Statistical analysis

Statistical calculations, including multivariate analysis, were carried out using the statistical package SPSS 11.5 (SPSS, Chicago, IL, USA). Unless otherwise stated, continuous variable results are expressed as mean ± standard deviation (SD) and the categorical variables as frequencies and percentages. Non-normally distributed scalar variables were analysed as non-parametric using Kruskal–Wallis and Mann–Whitney tests. For categorical data, we used Pearson Chi-squared test. To test Hardy–Weinberg equilibrium, the Chi-squared test was used. As in-hospital and ICU mortalities were our main end-points of interest, we used binary logistic regression and hazard statistical procedures to evaluate the influence of individual genotype on the patient outcome. For these analyses, Wald (backward stepwise selection) statistic and the Kaplan–Meier (Log-rank statistic) procedure were applied, respectively. All reported P values are two-tailed, with 0.05 or less taken as significant.

Results

Demographics

Eighty-five patients were studied. Patient were aged 19–95 years (mean = 56 years, median = 58 years). The diagnoses at ICU admission, according to APACHE-II were: sepsis (n = 30), infection (n = 18), cardiac arrest (n = 9), neurological disease (n = 5), metabolic/renal (n = 4), respiratory disease (n = 4), cerebral haemorrhage (n = 2), trauma (n = 2), gastrointestinal tract perforation/obstruction (n = 2) and one patient each for congestive heart failure, digestive haemorrhage, cardiovascular disease, cardiac rhythm disturbance, gastrointestinal disease, coronary artery disease, renal transplantation, surgery for neoplasm of gastrointestinal tract, and hemorrhagic shock after surgery.

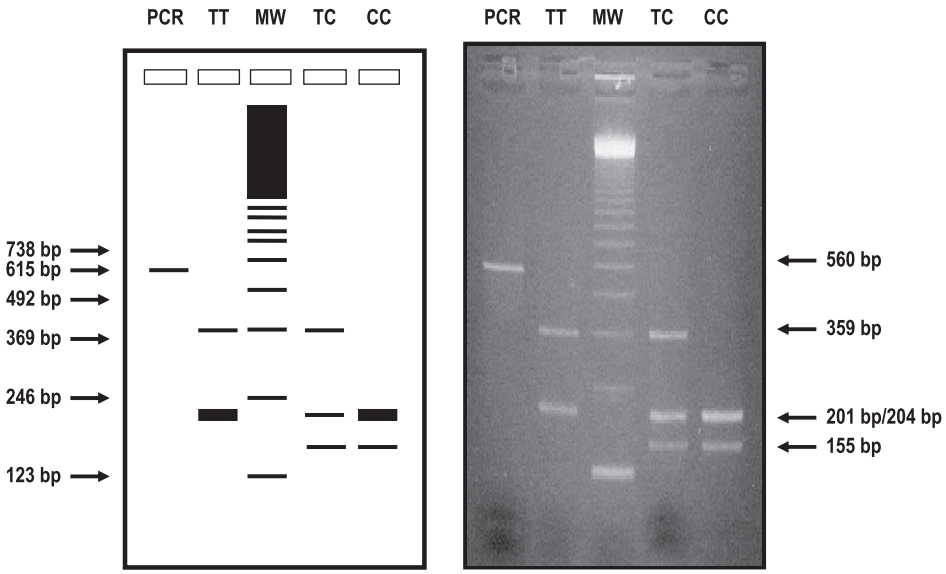

Figure 1 Electrophoretic schematic representation and image from a representative sample in agarose gel. Scale on the right: PCR, amplified product; TT, banding pattern for TT homozygous; TC, banding pattern for heterozygous; CC, banding pattern for CC homozygous. Scale on the left: MW, banding pattern from a 123 bp molecular weight marker (GibcoBRL-Life Technologies, Rockville, MD, USA).

During the ICU internment period, 61% (n = 52) of patients were diagnosed as having sepsis. The results from the bacteriological cultures of patients with sepsis were as follows: 42.3% (22/52) patients with at least one focus of infection caused by the presence of Gram-negative organisms; 13.5% (7/52) with absence of Gram-negative organisms; and 44.2% without an identified germ. Forty-two per cent (n = 36) of patients were diagnosed with septic shock.

Table 1 shows patient data by mortality. Patient mortality (monitored during ICU and post-ICU stay) was associated with older age (P = 0.001), higher APACHE-II (P = 0.004) and SOFA (P < 0.001) scores, a longer stay in the ICU (P = 0.002), sepsis (P = 0.046), and septic shock (P = 0.002).

Genotype distribution

The genotypic (TT = 0.27, TC = 0.41, CC = 0.32) and allelic (T = 0.48, C = 0.52) frequencies from our sample did not differ from the values expected by the Hardy–Weinberg model (P = 0.271). Table 2 shows patient data according to the CD14 −260C>T genotype frequency. We did not evaluate the effect of the CD14 genotype by segregating patients accordingly to bacteria type or disease group because the sample size became much reduced when patients were grouped by such data. There were no statistically significant differences between genotypes when we segregated them by age, sex, APACHE-II score, SOFA score, sepsis, septic shock, length of ICU stay and length of hospital stay. The mortality during the period of ICU plus post-ICU stay presented a statistically significant difference between genotypes, especially between TT and non-TT groups (P = 0.025). Organ dysfunction distribution was not associated with −260C>T CD14 genotypes in any of the six systems at ICU admission (Table 3) or during the first 7 days following ICU admission (data not shown). Promoter CD14 −260C>T allele frequencies presented no statistically significant differences for sepsis and septic shock; however, the frequency of the C allele was weakly associated (not statistically significant) with increased

Table 1 Patients' clinical and demographic data by mortality

Variables	All patients	Survivors	Deceased	P value†
Patients, n (%)	85 (100)	35 (41.2)	50 (58.8)	NA
Age (years), mean (SD)	56 (18.4)	48.4 (18.6)	61.4 (16.5)	0.001 *ST
Female sex, n (%)	40 (47)	16 (45.7)	24 (48)	0.835 χ^2
APACHE-II score, mean (SD)	21.1 (8.2)	18 (7.8)	23.2 (7.9)	0.004 *ST
SOFA score, median (min/max)‡	7 (0/15)	5 (0/13)	7 (0/15)	0.000 *MW
ICU LOS (days), median (min/max)	13 (2/54)	9 (2/43)	21 (2/54)	0.002 *MW
ICU + H LOS (days), median (min/max)	24 (2/224)	21 (5/96)	24.5 (2/224)	0.961 MW
Patients with sepsis, n (%)	52 (61)	17 (32.7)	35 (67.3)	0.046 *χ^2
Patients with septic shock, n (%)	36 (42)	8 (22.2)	28 (77.8)	0.002 *χ^2

*P < 0.05. †Comparison between ICU + H survivor (n = 35) and non-survivor (n = 50) groups. ‡Related to SOFA score at ICU admission. APACHE-II, Acute Physiology and Chronic Health Evaluation II; ICU, intensive care unit; ICU + H, ICU plus hospital stay; LOS, length of stay; MW, Mann–Whitney U-test; NA, not applicable; SD, standard deviation of the mean; SOFA, Sequential Organ Failure Assessment; ST, Student's t-test; χ^2, Pearson Chi-squared test.

Table 2 Patients' clinical and demographic data according to CD14 genotype

Variables	CC	TC	TT	Non-TT	*P* value†	*P* value‡
Frequency, n (%)	27 (31.8)	35 (41.2)	23 (27)	62 (73)	0.271§	NA
Age, (years), mean (SD)	55.7 (19)	58.8 (19)	52.1 (17.5)	57.5 (19)	0.398 AN	0.233 ST
Female sex, *n* (%)¶	13 (48.1)	17 (48.6)	10 (43.5)	30 (48.4)	0.922 χ^2	0.687 χ^2
APACHE-II score, mean (SD)	20.3 (7.1)	21.9 (8.5)	20.8 (9.2)	21.2 (7.9)	0.736 AN	0.850 χ^2
SOFA score, median (min/max)††	6 (2/15)	7 (0/14)	7 (0/13)	6 (0/15)	0.373 KW	0.544 MW
Patients with sepsis, *n* (%)	17 (63)	19 (54)	16 (70)	36 (58)	0.492 χ^2	0.337 χ^2
Patients with septic shock, *n* (%)	9 (33.3)	16 (45.7)	11 (47.8)	25 (40.3)	0.511 KW	0.536 MW
ICU LOS (days), median (min/max)	14 (3/54)	17 (2/38)	12 (2/44)	15 (2/54)	0.698 KW	0.494 MW
ICU + H LOS, (days), median (min/max)	22 (6/72)	25 (2/96)	21 (3/224)	24 (2/96)	0.713 KW	0.847 MW
Mortality, *n* (%)	17 (63)	24 (68.6)	9 (39.1)	41 (66.1)	0.073 χ^2	0.025*χ^2

*$P < 0.05$. †Comparison between genotypes (CC: $n = 27$, TC: $n = 35$, TT: $n = 23$). ‡Comparison between TT ($n = 23$) and non-TT ($n = 62$) genotype groups. §Comparison with the expected Hardy–Weinberg distribution (Pearson Chi-squared test). ¶Related to the genotype frequency. ††Related to SOFA score at ICU admission. AN, anova test; APACHE-II, Acute Physiology and Chronic Health Evaluation II; ICU, intensive care unit; ICU + H, ICU plus hospital stay; KW, Kruskal–Wallis test; LOS, Length of stay; MW, Mann–Whitney *U*-test; NA, not applicable; Non-TT, C allele carrier; SD, standard deviation of the mean; SOFA, Sequential Organ Failure Assessment; ST, Student's *t*-test; χ^2, Pearson Chi-squared test.

Table 3 Organ dysfunction related to Sequential Organ Failure Assessment (SOFA) score at ICU admission, according to CD14 −260C>T genotypes

Organ dysfunction	CC	TC	TT	Non-TT	*P* value†	*P* value‡
Respiratory, *n* (%)	19 (70)	28 (80)	14 (60)	47 (76)	0.280 χ^2	0.174 χ^2
Cardiovascular, *n* (%)	19 (70)	20 (57)	11 (47)	39 (62)	0.262 χ^2	0.210 χ^2
Renal, *n* (%)	15 (44)	14 (40)	11 (47)	26 (42)	0.836 χ^2	0.627 χ^2
Cerebral, *n* (%)	10 (37)	10 (28)	4 (17)	20 (32)	0.306 χ^2	0.176 χ^2
Hepatic, *n* (%)	5 (18)	7 (20)	7 (30)	12 (19)	0.547 χ^2	0.276 χ^2
Haematological, *n* (%)	6 (22)	9 (25)	5 (21)	15 (24)	0.923χ^2	0.813χ^2

†Comparison between genotypes (CC: $n = 27$; TC: $n = 35$; TT: $n = 23$). ‡Comparison between TT ($n = 23$) and non-TT ($n = 62$) genotype groups.Non-TT, C allele carrier; χ^2, Pearson Chi-squared test.

mortality during the period of ICU plus post-ICU stay ($P = 0.078$) (Table 4).

We carried out a hazard function analysis by the Kaplan–Meier procedure using the TT genotype as a discriminating factor. Taking all patients together, we observed that patients carrying the TT genotype had a better outcome ($P = 0.042$) when compared with those carrying the non-TT genotype. Both groups (TT and non-TT) reacted nearly equally up to the third week after ICU admission. However, changes in the survival rate in favour of the TT group were observed after this time (Fig. 2). The same analysis was carried out with patients with sepsis ($n = 52$) and septic shock ($n = 36$) and the survival distribution patterns were very similar although not statistically significant (data not shown).

Table 4 Patients' data according to CD14 −260C>T alleles

Variables	T allele	C allele	*P* value
With sepsis, *n* (%)	51 (49)	53 (51)	0.648 χ^2
Without sepsis, *n* (%)	30 (45)	36 (55)	
With septic shock, *n* (%)	38 (53)	34 (47)	0.250 χ^2
Without septic shock, *n* (%)	43 (44)	55 (56)	
ICU + H deceased, *n* (%)	42 (42)	58 (58)	0.078 χ^2
ICU + H survivors, *n* (%)	39 (56)	31 (44)	

ICU, intensive care unit; ICU + H, ICU plus hospital stay; χ^2, Pearson Chi-squared test.

We carried out binary logistic regression analysis, incorporating both TT and non-TT genotypes and main clinical predictors such as age and illness severity at ICU admission (APACHE-II and SOFA scores) to exclude other risk factors that could influence the outcome. Taking all patients together, only age ($P = 0.015$) and SOFA score ($P = 0.006$) were significantly associated with the outcome, whereas the TT genotype was negatively associated with death outcome, although not significantly ($P = 0.051$; OR = 0.342 [CI 0.11–1.05]). When patients were segregated as those with and without sepsis, we found that the CD14-TT genotype presented in patients with sepsis ($n = 52$) a very significant negative correlation to the death outcome ($P = 0.016$; Table 5).

Discussion

We evaluated, for the first time in a southern Brazilian population, the influence of the −260C>T CD14 SNP in critical ill patients. The genotype and allele frequencies in the sample studied were similar to those in other studies with more than 1000 subjects from different populations with the same European ethnic component.[15,18,24,25] In our subjects, we found that the genotype and allele frequencies were at Hardy–Weinberg equilibrium and that the genotype distribution was random for clinical characteristics at ICU patient admission. We found evidence of a correlation between genotype distribution and patient mortality in the ICU and post-ICU period. The CD14

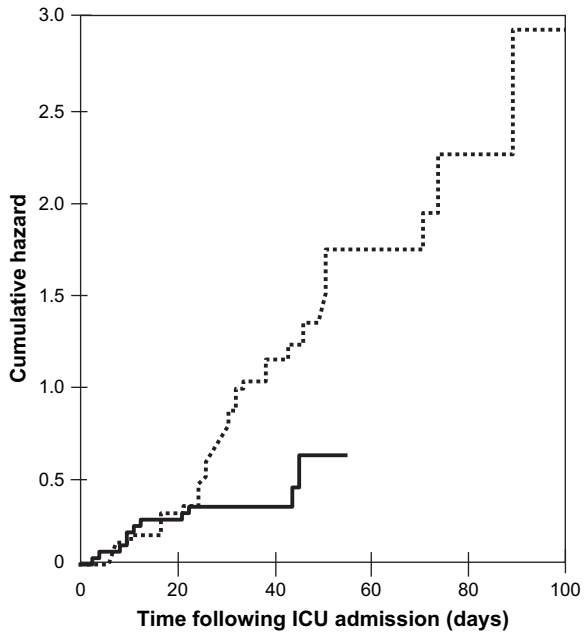

Figure 2 Cumulative hazard for critically ill patients (n = 85). TT (continuous line, n = 23) and non-TT (dotted line, n = 62) −260C>T CD14 genotype groups (Log-rank statistic, P = 0.042). ICU, intensive care unit.

TT patients presented lower mortality when compared with the non-TT patients.

Our result is partially in conflict with a previous study on patients with septic shock, wherein the authors found an association between TT genotype and susceptibility to and risk of death.[16] Nevertheless, it should be emphasized that in our study the patients without sepsis were critically ill, in contrast to the control patients in the study carried out by Gibot *et al.*[16] and that the follow up of our patients was extended up to the total time that the patients stayed in the hospital (up to 224 days). Because sepsis and septic shock are complex and multifactorial diseases, it is possible that an imbalance in the immune response could be responsible for the differences observed over a long period of time.

One can argue that if the hypothesis that abnormal CD14 may lead to increased or more severe infection is correct, the genotype distribution should be skewed. Interestingly, in our sample, the genotype distribution was similar when we compared the patients with sepsis with those without sepsis at ICU admission (P = 0.492; Table 2). However, when we consider the distribution at the time of discharge from the hospital, it was much skewed (alive/dead: TT = 9/7, TC = 3/16, CC = 5/12; P = 0.036). This observation suggests that the genotype lacked sufficient power to affect patient susceptibility to infection. However, the CD14 polymorphism seems to affect the course of the disease and/or the response to the medication.

Here, infectious diseases have been considered as multifactor events that produce organic reactions that alternate between pro-inflammatory and anti-inflammatory responses.[26] Thus, the ability of a patient to overcome a severe infection depends mainly on the body's capacity to adequately modulate the equilibrium between these opposing forces. Glück *et al.*[27] showed that mCD14 expression was reduced in all patients with sepsis but returned to normal levels during the course of the disease in survivors and mCD14 was found to be inversely correlated with the severity of disease, leucocyte elastase and C-reactive protein. These authors observed that among patients with severe disease and APACHE-II scores ≥20, sCD14 levels were significantly higher in patients who survived to day 28 as compared with non-survivors (P = 0.02). They suggested that higher sCD14 levels may be beneficial in sepsis and that persistently reduced mCD14 expression could be a marker for severity of disease in critically ill patients.

An increase in mCD14 and sCD14 concentrations has been closely associated with the TT genotype and T allele in several

Table 5 Patient's risk factors

Sepsis (yes/no)			B	SE	Wald	d.f.	Sig.	Exp(B)	95% CI for Exp(B)	
									Lower	Upper
No	Step 1†	SOFA	0.221	0.128	2.982	1	0.084	1.248	0.971	1.604
		Age	0.034	0.022	2.443	1	0.118	1.035	0.991	1.080
		CD14-TT	−0.153	1.100	0.019	1	0.890	0.859	0.099	7.416
		Constant	−3.260	1.568	4.324	1	0.038	0.038		
	Step 2†	SOFA	0.222	0.128	3.026	1	0.082	1.249	0.972	1.605
		Age	0.035	0.021	2.967	1	0.085	1.036	0.995	1.079
		Constant	−3.356	1.415	5.621	1	0.018	0.035		
Yes	Step 1†	SOFA	0.185	0.112	2.720	1	0.099	1.203	0.966	1.499
		Age	0.046	0.021	4.659	1	0.031	1.047	1.004	1.092
		CD14-TT	−1.780	0.742	5.752	1	0.016	0.169	0.039	0.722
		Constant	−2.511	1.357	3.424	1	0.064	0.081		

B, regression coefficient; CI, confidence interval; d.f., degrees of freedom; Exp(B), odds ratio; SE, standard error; Sig., significance (P value); SOFA, Sequential Organ Failure Assessment; Wald, statistic. †Variable(s) entered at step 1: Age, C D14-TT genotype and SOFA score.

studies that have compared −260C>T CD14 polymorphism in different populations.[12–16] The TT patients, who have an increased CD14 gene activation and produce more effective CD14,[10] potentially generate higher response to LPS and thus could present a stronger innate immunity through immediate activation of the TLR4 and NF-κB and synthesis of IL-1, TNF-α, IL-8, IL-12 and IL-6. However, by producing a reduced signal in response to LPS, the non-TT individuals could not induce an immediate inflammatory response strong enough to clear the bacteria, which spread and become systemic. The systemic infection is what triggers an unregulated inflammatory response that could produce different organ dysfunctions.

Therefore, considering the −260C>T CD14 SNP as a potentially functional polymorphism, we propose that our TT patients had more flexibility in controlling the availability of CD14 during the evolution of the illness. We also suggest, in conformity with Glück *et al.*,[27] that an increase in the CD14 concentration in TT patients could be a protective factor that helps the immune system to improve the balance of its pro-inflammatory and anti-inflammatory actions.

Our results showed that −260C>T CD14 SNP could be considered a useful candidate to better understand the outcome of patients who have been through critical illnesses. Thus, we suggest that −260C>T CD14 polymorphism genotyping could be a good genetic marker for risk evaluation of critically ill patients and that non-TT patients present a higher mortality risk when compared with TT patients. To confirm this hypothesis, CD14 levels should be monitored during the entire period of hospital stay and data compared between TT and non-TT patients, something that was beyond the scope of this study.

Despite the small sample size of our study, we can conclude that there is some evidence to suggest that TT patients could have a positive survival prognosis in cases of critical illness. Identification of genetic polymorphisms that predispose critically ill patients to the most severe manifestations would allow early targeting of high-risk individuals for aggressive or novel treatment, potentially improving their clinical outcome. We also conclude that the CD14 gene is a good candidate for further study in the search for genetic markers that, complementarily to other evaluation tools, such as APACHE and SOFA, may help the physician to better evaluate the critically ill patient's risk.

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