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Elevated Cell-Free Plasma DNA Level as an Independent Predictor of Mortality in Patients with Severe Traumatic Brain Injury

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Abstract

Trauma is the leading cause of death in individuals less than 45 years old worldwide, and up to 50% of trauma fatalities are because of brain injury. Prediction of outcome is one of the major problems associated with severe traumatic brain injury (TBI), and research efforts have focused on the investigation of biomarkers with prognostic value after TBI. Therefore, our aim was to investigate whether cell-free DNA concentrations correlated to short-term primary outcome (survival or death) and Glasgow Coma Scale (GCS) scores after severe TBI. A total of 188 patients with severe TBI were enrolled in this prospective study; outcome variables comprised survival and neurological assessment using the GCS at intensive care unit (ICU) discharge. Control blood samples were obtained from 25 healthy volunteers. Peripheral venous blood was collected at admission to the ICU. Plasma DNA was measured using a real-time quantitative polymerase chain reaction (PCR) assay for the β-globin gene. There was correlation between higher DNA levels and both fatal outcome and lower hospital admission GCS scores. Plasma DNA concentrations at the chosen cutoff point ($\geq$171,381 kilogenomes-equivalents/L) predicted mortality with a specificity of 90% and a sensitivity of 43%. Logistic regression analysis showed that elevated plasma DNA levels were independently associated with death ($p < 0.001$). In conclusion, high cell-free DNA concentration was a predictor of short-term mortality after severe TBI.

Key words: biomarkers; outcome; plasma DNA; real-time PCR; traumatic brain injury

Introduction

Traumatic brain injury (TBI) is the leading cause of injury, death, and disability in young persons worldwide. In the period of 2002 through 2006, approximately 1.7 million U.S. civilians sustained a TBI annually; of these, approximately 1.4 million were treated and discharged from emergency departments (EDs), 275,000 were hospitalized and discharged alive, and 52,000 had a fatal outcome.[1,2] Severe TBI is associated with a 30–70% mortality rate and, generally, the survivors are, to a greater or lesser extent, permanently disabled.[3]

Prediction of outcome is one of the major problems associated with severe TBI.[4] Early assessment of patients' brain damage may be quite difficult during the stay in the intensive care unit (ICU). ICU scores are used to predict hospital outcome in critically ill patients, but have shortcomings.[5,6] Therefore, the wide range of conditions associated and the relatively variable predictive value of clinical assessments in severe TBI complicate the identification of patients at higher risk for development of secondary brain injury and fatal outcome.[7] Clearly, a practical and sensitive biomarker is needed to identify these patients, as early as possible, to initiate intervention and to indicate those persons to be targeted with higher risk therapeutic strategies.[8,9]

In the last few decades, a rapidly growing number of molecules have been tested as potential biomarkers of TBI. Several potential biomarkers have been proposed, among which are: Protein S100B, neuron-specific enolase (NSE), glial fibrillary acidic protein (GFAP), myelin basic protein, creatine-kinase-BB, Hsp 70, sFas, tumor necrosis factor alpha (TNFα), brain-derived neirotrophic factor (BDNF), plasma von Willebrand factor, cleaved tau protein, spectrin breakdown products (SBDPs) , ubiquitin C-terminal hydrolase-L1 (UCH-L1), plasma DNA, and serum interleukin-10.[8–28] Nevertheless, at present, no single molecule presented adequate specificity and sensitivity as a comprehensive clinical diagnostic tool to predict the extent of neural tissue damage, or to aid in monitoring care and forecasting outcome.

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The presence of extracellular nucleic acids in the bloodstream was first described by Mandel and Metals (1948).[29] Only in the last 15 years, however, has the potential use of circulating cell-free DNA in the plasma or serum been investigated for the establishment of diagnosis, prognosis, and monitoring of a variety of conditions. Tumor, fetus, and donor-derived sequences have been detected in the plasma and serum of cancer patients, pregnant women, and transplant recipients, respectively.[30–34] In addition, plasma DNA has also demonstrated potential as a clinical biomarker in several acute pathologic conditions with high mortality risk.[35–39]

Significant increases of circulating DNA in the plasma of trauma patients have been reported as a promising marker for risk stratification of patients with minor, moderate, and severe injury and presented correlation with injury severity and development of posttraumatic complications.[40,41] A previous study of our group showed that severe TBI is associated with elevated DNA plasma levels and suggested that persistent DNA elevations correlated with mortality.[22] Recently, Macher and associates[27] investigated plasma DNA in 65 patients after severe TBI in the first 96 h after ICU admission. After an initial peak, a higher decrease was detected within the first 24 h among survivors compared with non-survivors.[27]

Therefore, our aim was to investigate in a large sample of patients with severe TBI whether plasma DNA concentrations correlated to short-term primary outcome (survival or death) and Glasgow Coma Scale (GCS) scores within the first 24 h after injury.

Methods

Patients and control subjects

Ethical approval for the study protocol was granted by the Research Ethics Committee of the Universidade Luterana do Brasil (CEP ULBRA 2008-239H). From September 2008 to September 2011, in three regional trauma centers, 188 persons with severe TBI (GCS 3–8 at emergency department admission) were enrolled in this prospective study. Only patients ≥ 16 years old and without a history of neurological or psychiatric disease were included in this cohort. On admission to the trauma emergency department, patients were initially evaluated, resuscitated (with crystalloids), and underwent emergency operation when necessary. Only the patients transferred to the trauma ICU within 12 h of the head injury were included in the study.

Clinical outcome variables of severe TBI comprised short-term survival, time for ICU discharge, and neurological assessment using the GCS at the ICU discharge. The circulatory function and GCS scores were monitored at hospital admission, ICU admission, and during ICU stay. All patients were sedated and mechanically ventilated. Corticosteroids were not administered. To establish normal values of plasma DNA, a negative control group was included consisting of 14 healthy male and nine female volunteers without a history of brain damage (median age 30 years; range 19–45 and 44 years; range 34–65 years, respectively).

Blood sampling

Peripheral venous blood was collected into ethylenediaminetetraacetic acid-containing tubes at ICU admission. Blood samples were centifuged at 1000 g for 10 min; then the plasma was removed (with great care taken not to disturb the pellet) and stored at −20°C until batch evaluation. Blood samples from the control group were collected and processed in the same way.

DNA extraction from plasma samples

DNA from plasma samples was extracted according to the protocol developed by Boom and associates.[42] Briefly, 100 μL of plasma samples were lysed in 900 μL of a guanidine thiocyanate (GuSCN) buffer. After lysis, nucleic acids were bound to silica particles and subsequently washed with several solvents (a GuSCN-containing wash buffer, 70% ethanol and acetone) in consecutive steps. After being dried, the nucleic acids were released from the silica particles in 50 μL of elution buffer.

Real-time quantitative polymerase chain reaction (PCR)

Theoretical and practical aspects of real-time quantitative PCR were described by Heid and colleagues.[43] Real-time quantitative PCR analysis was performed using StepOnePlus™ Real-Time PCR System (Life Technologies, Carlsbad, CA). The amplification and product reporting system was based on the 5′ nuclease assay (Taqman assay).[44] Plasma DNA was measured using a real-time quantitative PCR assay for the β-globin gene.[31] Our study used primers and probe sequences from human beta globin gene, based in several previous studies related with circulating cell-free DNA. This target is the most studied for this purpose.[31,32,35,40,41,45] The β-globin Taqman system consisted of the amplification primers beta-globin-354F (5′-GTG CAC CTG ACT CCT GAG GAG A-3′), beta-globin-455R (5′-CCT TGA TAC CAA CCT GCC CAG-3′), and a dual-labeled fluorescent probe beta-globin-402T [5′-(FAM) AAG GTG AAC GTG GAT GAA GTT GGT GG (TAMRA)-3′].[31] The expression of quantitative results as kilogenome-equivalents/L was described previously.[31] One genome-equivalent was defined as the amount of a target sequence contained in a single diploid human cell.

Statistical analysis

Continuous variables were analyzed by the Kolmogorov-Smirnov test to determine the distribution type. Those with a normal distribution were analyzed by the Student *t* test, while those with a non-parametric distribution were analyzed by the Mann-Whitney *U* test or Kruskal-Wallis analysis followed by the Dunn post-test. Correlations were analyzed using the Spearman non-parametric correlation method or linear regression method. The extent to which the DNA concentrations differed between persons surviving or dying in the ICU after severe TBI was assessed using receiver operator characteristics (ROC) plots. The ROC plot is obtained by calculating the sensitivity and specificity for every distinct observed data value, and plotting sensitivity against 1-(specificity). The ROC curve was used to evaluate the optimal cutoff values measured at study entry for prediction of unfavorable outcome. A cutoff point on the curves was chosen to attain the best compromise between sensitivity and specificity for death in the ICU. Logistic regression analysis was performed to eliminate confounding factors, and the dependent variable was the primary outcome (dead/alive). The independent variables tested were age, associated injury, craniotomy, GCS score at hospital admission, and plasma DNA levels. All *p* values presented are two-tailed and the values of $p < 0.05$ were considered statistically significant.

Results

Plasma DNA concentrations were determined in all 188 patients with severe TBI at ICU admission (mean time for blood sampling after hospital admission was 6.0 ± 4.9 h). Characteristics of the severe TBI population stratified for the primary outcome measure (survivors/non-survivors) are depicted in Table 1. The mean age was 34.8 ± 13.9 years and 88.0% of the persons were males. Forty-one percent of the patients presented isolated severe TBI. There were no significant differences concerning age, incidence of pre-hospital care, mechanism of injury, and proportion of associated extracranial injuries between survivors and non-survivors. Craniotomy was performed in 55.3% of the patients, and there were no

Table 1. Characteristics of the Severe Traumatic Brain Injury Study Population Stratified for the Primary Outcome Measure (Survivors/Non-Survivors)

*Characteristic**	*All patients* (n = *188*)	*Survivors* (n = *122*)	*Non-survivors* (n = *66*)	p *value*
Age, years	34.8 (13.9)	33.4 (12.6)	37.2 (15.8)	0.081
Pre-hospital care, n (%)	124 (66)	84 (69)	40 (61)	0.863
Sex, male, n (%)	165 (88)	107 (88)	58 (88)	0.462
GCS at hospital admission	5.7 (2.2)	6.3 (2.6)	5.2 (2.0)	< 0.001
Systolic blood pressure	128 (29)	129 (28)	126 (33)	0.552
Diastolic blood pressure	77 (22)	77 (23)	76 (19)	0.892
Mechanism of injury, n (%)				0.168
Motor vehicle accident	80 (43)	55 (45)	25 (38)	
Assault	41 (22)	24 (20)	17 (26)	
Auto pedestrian	40 (21)	28 (23)	12 (18)	
Fall	27 (14)	15 (12)	12 (18)	
Craniotomy, n (%)	104 (55)	63 (52)	41 (62)	0.236
Associated injuries, n (%)	110 (59)	77 (63)	33 (50)	0.090
Mortality, n (%)	66 (35)	-	66 (100)	
Time between trauma and outcome, days	11.9 (10.8)	15.5 (11.5)	5.0 (3.9)	< 0.001
GCS at discharge from ICU	11.1 (3.4)	11.1 (3.4)	-	

*Data are shown as mean (standard deviation) or, when indicated, number (%) of patients.
GCS, Glasgow coma scale; ICU, intensive care unit.

significant differences in the rate of craniotomy between survivors and non-survivors. Severe TBI was associated with a 35.1% mortality rate, mostly occurring within 72 h after ICU admission. The mean time between the traumatic event and death was 5.0 ± 3.9 days. In contrast, in the survivors group, the mean time between trauma and ICU discharge was 15.5 ± 11.5 days, and mean GCS score at ICU discharge was 11.1 ± 3.4 (Table 1). GCS scores at hospital admission differed significantly between the survivor and non-survivor groups, with the non-survivors presenting lower scores than the survivors (5.2 ± 2.0 and 6.3 ± 2.6, respectively, $p = 0.002$) (Table 1).

Characteristics of the TBI population stratified for the type of severe TBI (isolated TBI or TBI associated with multitrauma) are shown in Table 2. There were no significant differences in age, incidence of pre-hospital care, GCS scores at either hospital or ICU admission, and diastolic blood pressure at hospital admission. There were significant differences in systolic blood pressure, craniotomy rate and time between event and outcome between isolated TBI and TBI associated with multitrauma (Table 2).

The plasma concentration of cell–free DNA was estimated for all persons enrolled in the study. The control group ($n = 25$) presented a mean plasma DNA concentration of 2156 ± 591 kilogenomes-equivalents/L (mean ± standard error of the mean [SEM]). There were no significant differences in cell-free DNA levels among men and women ($p = 0.109$, Mann Whitney U test). In addition, cell–free DNA was estimated in patients with severe TBI at ICU admission (mean time 6.0 ± 4.9 h after hospital admission). Mean plasma DNA concentrations were significantly higher in the severe TBI group (429856 ± 162311 kilogenomes-equivalents/L, mean ± SEM) when compared with the control group ($p < 0.001$, Mann-Whitney U test) (Fig. 1). Noteworthy, mean plasma DNA concentrations were significantly higher in the non-survivor group (986750 ± 260548 kilogenomes-equivalents/L, mean ± SEM) when compared with the survivor group (130699 ± 427496 kilogenomes-equivalents/L, mean ± SEM) ($p < 0.001$, Mann-Whitney U test) (Fig. 1). In fact, there was a significant correlation between higher plasma DNA concentrations and fatal outcome (Spearman's rho = 0.320, $p < 0.001$). There were no significant correlations between plasma cell-free DNA and either age (linear regression, $p = 0.718$) or craniotomy (Spearman rank, $p = 0.724$) (data not shown).

Furthermore, when plasma cell-free DNA levels and GCS scores were analyzed, a significant correlation between higher DNA levels and lower GCS scores at hospital admission was observed (1234043 ± 491479, 436573 ± 257432, 770442 ± 592283, 130611 ± 58329, 130269 ± 84581, 66347 ± 21288, mean plasma cell-free

Table 2. Characteristics of the Traumatic Brain Injury Study Population Stratified for the Type of Trauma (Isolated or Associated with Multitrauma)

*Characteristic**	*Isolated TBI* (n = *78*)	*TBI + multitrauma* (n = *110*)	P *value*
Age, years	37.1 (14.8)	33.2 (12.8)	0.063
Sex, male, n (%)	69 (88)	96 (87)	0.876
GCS at hospital admission	5.9 (2.6)	5.8 (2.2)	0.434
Systolic blood pressure	135 (31)	124 (27)	0.023
Diastolic blood pressure	80 (23)	75 (20)	0.251
Time for blood sampling (h after ICU admission)	5.4 (3.5)	6.3 (5.4)	0.219
Mechanism of injury, n (%)			0.710
Motor vehicle accident	33 (42)	47 (43)	
Assault	20 (26)	21 (19)	
Auto pedestrian	15 (19)	25 (23)	
Fall	10 (13)	17 (15)	
Craniotomy, n (%)	63 (81)	41 (37)	0.004
Mortality, n (%)	33 (42)	33 (29)	0.090
Time between trauma and outcome, days	9.9 (9.7)	13.3 (11.4)	0.040
GCS at discharge from ICU	10.3 (3.4)	11.1 (3.3)	0.215

*Data are shown as mean (standard deviation) or, when indicated, number (%) of patients.
GCS, Glasgow coma scale; ICU, intensive care unit.

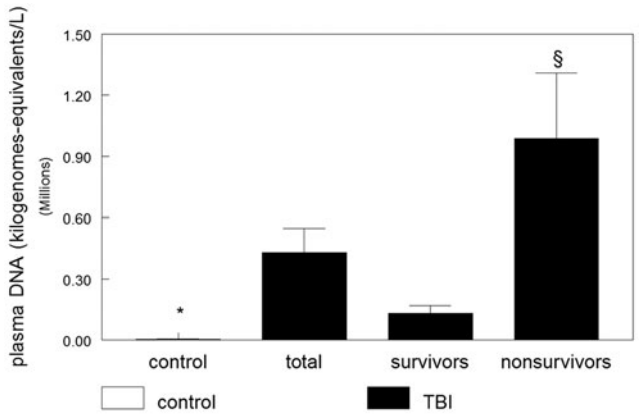

FIG. 1. Plasma DNA concentrations in control and severe traumatic brain injury (TBI) persons stratified by the primary outcome (survival or death). Data are shown as mean ± standard error of the mean stratified by primary outcome. There was a significant correlation between higher plasma DNA concentrations and fatal outcome (Spearman rho = 0.320, $p = 0.001$). *Significantly different from other groups ($p < 0.001$, Kruskal-Wallis, followed by Dunn test). §Significantly different from control and TBI survivor groups ($p < 0.001$, Kruskal-Wallis, followed by Dunn test).

DNA levels for 3, 4, 5, 6, 7, and 8 GCS scores, respectively; Spearman rho = −0.327, $p = 0.001$) (Fig. 2). Indeed, this correlation between higher plasma cell-free DNA levels and lower GCS scores was detected despite the type of severe TBI the patient had (either isolated TBI [Spearman rho = −0.335, $p = 0.003$] or TBI associated with extracerebral lesions [Spearman rho = −0.275, $p = 0.004$]) (Fig. 2B, C).

ROC curve was plotted (Fig. 3) and a cutoff point that would ensure the detection of the highest proportion of persons with fatal outcome with the least compromise of specificity was chosen. Therefore, a cutoff point of 171381 kilogenomes-equivalents/L plasma DNA concentrations within 12 h after hospital admission was chosen. The diagnostic characteristics of this cutoff point was a specificity of plasma DNA concentration for predicting mortality of 90% and a sensitivity of 43%. The area under the curve for cell-free DNA plasma concentration was 0.694 ($p < 0.001$) (Fig. 3). Interestingly, considering the effect of the type of severe TBI (isolated or associated with extracerebral lesions) on the diagnostic characteristics of the chosen cutoff point of plasma cell-free DNA, we observed that isolated severe TBI ensured higher specificity than TBI associated with multitrauma for predicting mortality within the first 12 h after trauma (specificity of 92% and 89% and sensitivity of 33% and 55% for isolated TBI or TBI associated with extracerebral lesions, respectively) (Fig. 4).

Logistic regression analysis was performed to assess the independent influence of cell-free DNA plasma levels on the TBI primary outcome (dead/alive). After adjusting for confounding variables we found that lower GCS at hospital admission ($p = 0.007$), absence of associated injuries ($p = 0.018$), and higher plasma cell-free DNA levels (> 171381 kilogenomes-equivalents/L; $p < 0.001$) were variables independently associated with poor outcome (death).

Discussion

In this study, we evaluated circulating plasma DNA as a predictor of fatal outcome in patients with severe TBI. The study

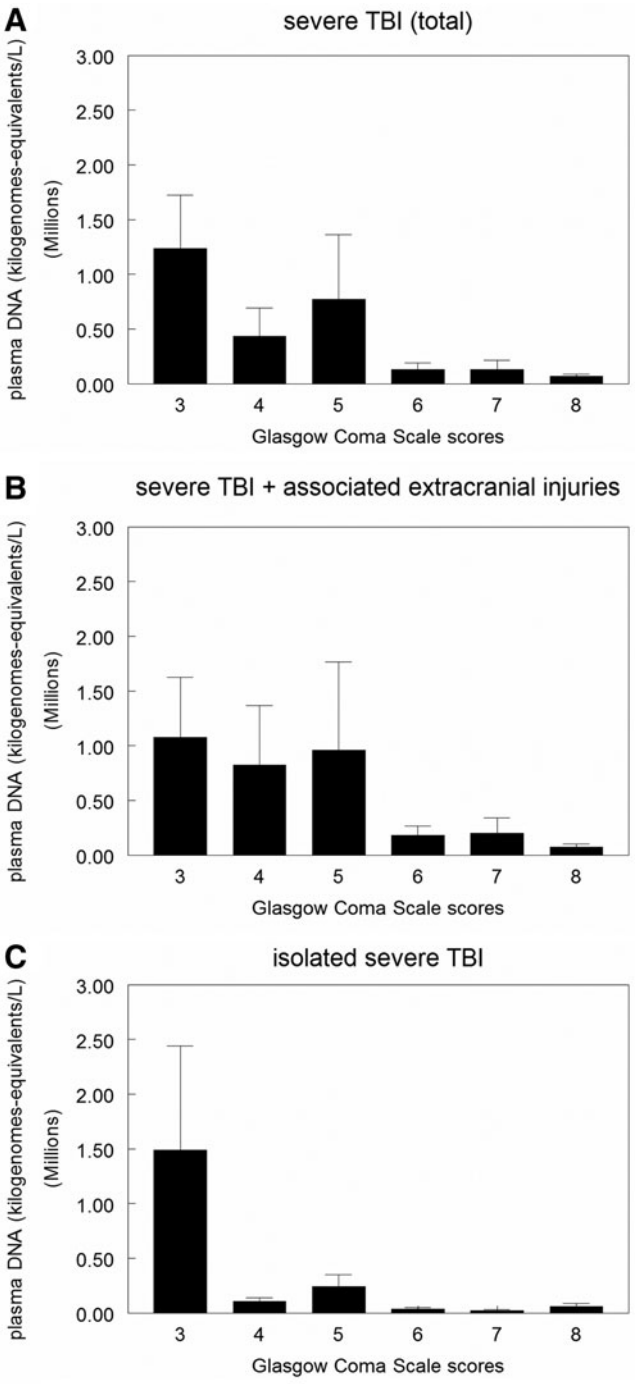

FIG. 2. Plasma DNA concentrations and hospital admission Glasgow Coma Scale scores in severe traumatic brain injury (TBI) persons stratified by the type of TBI lesion. In (**A**), data represent correlation between plasma DNA concentrations and hospital admission GCS scores in all persons with severe TBI ($n = 188$). There was a significant correlation between higher DNA levels and lower GCS scores ($p = 0.001$). In (**B**), data represent correlation between plasma DNA concentrations and hospital admission GCS scores in the group with severe TBI associated with extracranial lesions ($n = 110$). There was a significant correlation between higher DNA levels and lower GCS scores ($p = 0.003$). In (**C**), data represent correlation between plasma DNA concentrations and hospital admission GCS scores in the isolated severe TBI group ($n = 78$). There was a significant correlation between higher DNA levels and lower GCS scores ($p = 0.004$). Data are expressed as mean ± S.E.M.

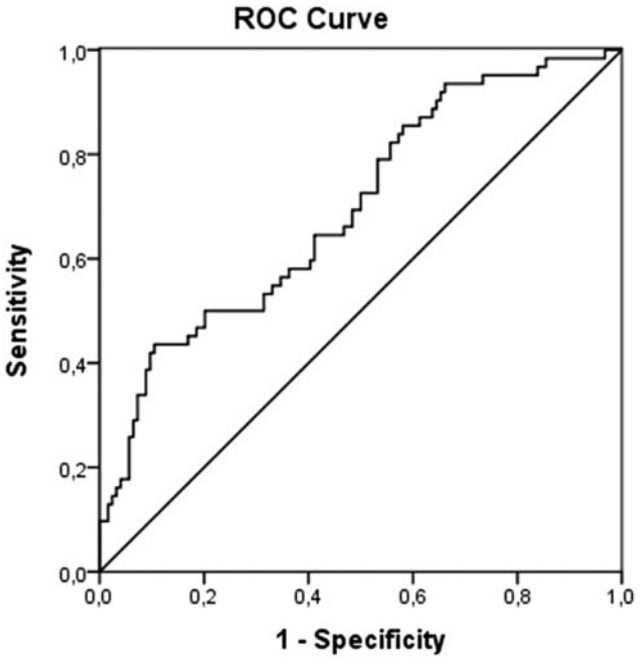

FIG. 3. Receiver operator characteristics (ROC) curves plasma DNA concentrations for predicting fatal outcome after severe TBI. ROC curve analysis showed area under the curve of 0.694 ± 0.040 (standard error of the mean) ($p < 0.001$). A cutoff point of 171381 kilogenomes-equivalents/L plasma DNA concentrations was chosen and presented a specificity and sensitivity for predicting mortality of 90% and 43%, respectively.

showed a positive correlation between high plasma cell-free DNA levels and both lower GCS scores and fatal outcome within the initial 12 h after hospital admission despite the presence of associated extracerebral injuries. To our knowledge, cell-free DNA has not hitherto been investigated as an early predictor of mortality in a large series of exclusively patients with severe TBI. In accordance with the literature, victims of severe TBI enrolled in our study were mostly young men involved in motor vehicle accidents and interpersonal violence. Lower GCS scores at hospital admission were associated with worst prognosis, and the short-term mortality rate was of 35%.[1,]

Previous studies have reported elevated plasma cell-free DNA in trauma; correspondingly, in the present study, we observed that patients with severe TBI presented higher cell-free DNA levels than healthy persons.[22,37,40,41,45] The precise mechanism by which DNA is released into the bloodstream remains uncertain, because both necrosis and apoptosis have been observed in this scenario.[46] Further, decreased efficiency of DNA clearance mechanisms after injury may play a role in the increase of cell-free DNA in trauma.[40] In fact, it is possible that direct damage or hemodynamic compromise of the organ systems responsible for circulating DNA clearance could also lead to increased plasma cell-free DNA.[40] Recently, it has been suggested that circulating plasma DNA may play a role in cell communication. Indeed, evidence supporting the active release of free circulating DNA by living cells has been reported.[47,48]

Previous studies investigating plasma cell-free DNA and trauma showed association of increased plasma cell-free DNA levels with both injury severity and the development of post-traumatic complications.[27,40,41] In a previous study, our group has demonstrated that high concentrations of plasmatic cell-free DNA correlated to

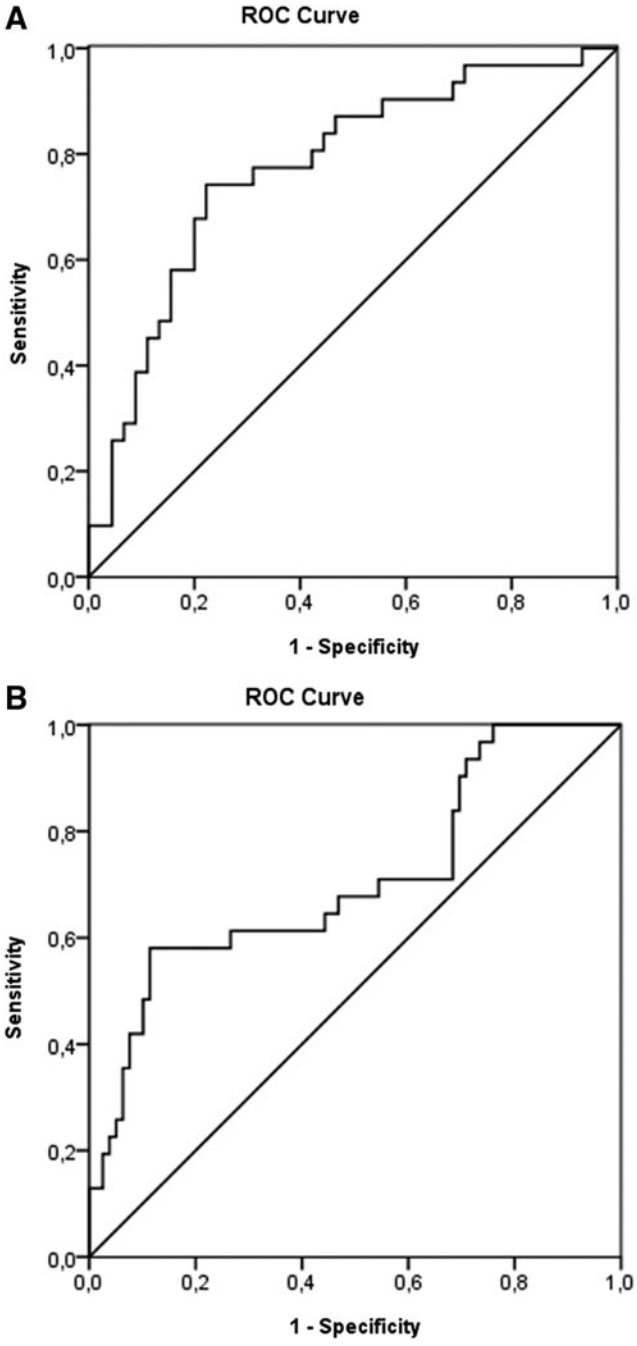

FIG. 4. Receiver operator characteristics (ROC) curves plasma DNA concentrations stratified by type of lesion for predicting fatal outcome after severe TBI. In (**A**), plasma DNA concentrations in the isolated severe TBI group, ROC curve analysis showed area under the curve of 0.777 ± 0.056 (standard error of the mean [SEM]) ($p < 0.001$). In (**B**), plasma DNA concentrations in the severe TBI associated with the extracerebral lesions group, ROC curve analysis showed area under the curve of 0.679 ± 0.060 (S.E.M.) ($p = 0.003$). A cutoff point of 171381 kilogenomes-equivalents/L plasma DNA concentrations was chosen. The specificity of plasma DNA concentration predicting mortality according to the chosen cutoff point was 92% and 89% for isolated severe TBI or severe TBI associated with extracerebral injuries, respectively, and sensitivity was 33% and 55%, respectively.

fatal outcome after severe TBI in males.[22] In the present study, we enrolled a larger sample of patients with severe TBI (either isolated severe TBI or TBI associated with extracerebral lesions) from both sexes and determined plasma cell-free DNA concentrations at ICU admission, within 12 h after trauma. In addition, we analyzed the association between plasma cell-free DNA levels and hospital admission GCS scores.

To our knowledge, this is the largest series investigating plasma cell-free DNA for short-term outcome prediction in patients with severe TBI. We observed a positive correlation between higher plasma cell-free DNA levels and fatal outcome within 12 h after trauma. In contrast with previous studies, in the present study, a single determination early after the trauma (mean 6 h) predicted fatal outcome of the patients with severe TBI admitted to the ICU.[22,27] Temporal profile post-injury may influence assessment of the predictive value of plasma cell-free DNA after severe TBI. Accordingly, Lo and coworkers[49] reported in a study investigating healthy pregnant women that DNA has a rapid clearance from plasma, with a mean estimated half-life of 16.3 min (range 4–30 min). Therefore, because of the short half-life of plasma cell-free DNA, an early determination to establish prognosis in patients with severe TBI may be more suitable and cost-effective in clinical practice.

Since Teasdale and Jennett[50] introduced the Glasgow Coma Scale in 1974, GCS scores have been widely used as a quantative measure of the level of consciousness in patients with TBI.[50] Nevertheless, few studies investigated the GCS as the sole variable in predicting outcome in patients with head injury.[51–53] Interestingly, in the present study, a positive correlation between higher plasma cell-free DNA levels and lower hospital admission GCS scores was demonstrated. This correlation was also demonstrated when patients were stratified for type of severe TBI (isolated or associated with extracerebral lesions). Therefore, we established a correlation between plasma cell-free DNA levels and the severity of injury at hospital admission despite the presence of extracerebral lesions. In fact, major extracranial injury has been reported as a prognostic factor for mortality in patients with TBI, but the strength of the effect is smaller in patients with more severe brain injury as is the most likely to occur in our series of patients who presented a high rate of isolated severe TBI from assault.[54,55]

Besides the brain, the release of cell-free DNA from injured cells may occur peripherally, because DNA is present in the nucleus/mitochondria of the majority of cell types and can be released into the circulation. Thus, extracerebral sources may contribute to the increase plasma DNA levels after multitrauma. Because many patients with severe head injury have extracranial injuries, they were included in the present study to investigate the value of cell-free measurements after severe TBI in the scenario of clinical practice. Associated extracranial injuries was observed in both the survivor (63%) and non-survivor (50%) TBI groups and did not correlate with outcome. Consequently, the impact of associated injuries on mortality seems to be limited. We recognize, however, that for a blood biomarker to be valuable in therapeutic trials in TBI, the ability to contribute to the successful prediction of the level of the neurological outcome of the survivors rather than solely mortality is essential. Indeed, in our study, the remarkably high levels of cell-free DNA observed in non-survivors early after TBI could be reflecting extensive tissue necrosis eliciting greater DNA release into the circulation. We could not establish a direct correlation, however, between cerebral tissue destruction and plasma DNA concentrations. In this sense, it would also have been important to cross-validate the findings for cell-free DNA with other paraclinical measures. A shortcoming of the present study is that image analysis was not defined as a primary outcome measure and therefore was not standardized.

The specificity of plasma cell-free DNA concentrations for prediction of poor outcome (death) at the chosen cutoff point ($\geq$171381 kilogenomes-equivalents/L) was high (specificity of 90%, 92%, and 89%, for all patients with severe TBI, isolated severe TBI, and severe TBI associated with extracerebral lesions, respectively). These rates for prediction of mortality were higher than those of clinically established predictors such as GCS scores and age.

Conclusion

The overall mortality rate in the severe TBI group was 35%, with most deaths occurring in the first 72 h after injury, and high plasma cell-free DNA concentrations within the initial 12 h after hospital admission was associated with lower GCS scores and increased death risk despite the presence of associated extracerebral injuries.

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Author Disclosure Statement

No competing financial interests exist.

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