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## EFFICACY AND SAFETY OF RECOMBINANT HUMAN ACTIVATED PROTEIN C FOR SEVERE SEPSIS

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FOR THE RECOMBINANT HUMAN ACTIVATED PROTEIN C WORLDWIDE EVALUATION IN SEVERE SEPSIS  
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### ABSTRACT

**Background** Drotrecogin alfa (activated), or recombinant human activated protein C, has antithrombotic, antiinflammatory, and profibrinolytic properties. In a previous study, drotrecogin alfa activated produced dose-dependent reductions in the levels of markers of coagulation and inflammation in patients with severe sepsis. In this phase 3 trial, we assessed whether treatment with drotrecogin alfa activated reduced the rate of death from any cause among patients with severe sepsis.

**Methods** We conducted a randomized, double-blind, placebo-controlled, multicenter trial. Patients with systemic inflammation and organ failure due to acute infection were enrolled and assigned to receive an intravenous infusion of either placebo or drotrecogin alfa activated (24  $\mu$ g per kilogram of body weight per hour) for a total duration of 96 hours. The prospectively defined primary end point was death from any cause and was assessed 28 days after the start of the infusion. Patients were monitored for adverse events; changes in vital signs, laboratory variables, and the results of microbiologic cultures; and the development of neutralizing antibodies against activated protein C.

**Results** A total of 1690 randomized patients were treated (840 in the placebo group and 850 in the drotrecogin alfa activated group). The mortality rate was 30.8 percent in the placebo group and 24.7 percent in the drotrecogin alfa activated group. On the basis of the prospectively defined primary analysis, treatment with drotrecogin alfa activated was associated with a reduction in the relative risk of death of 19.4 percent (95 percent confidence interval, 6.6 to 30.5) and an absolute reduction in the risk of death of 6.1 percent ( $P=0.005$ ). The incidence of serious bleeding was higher in the drotrecogin alfa activated group than in the placebo group (3.5 percent vs. 2.0 percent,  $P=0.06$ ).

**Conclusions** Treatment with drotrecogin alfa activated significantly reduces mortality in patients with severe sepsis and may be associated with an increased risk of bleeding. (N Engl J Med 2001;344:699-709.)

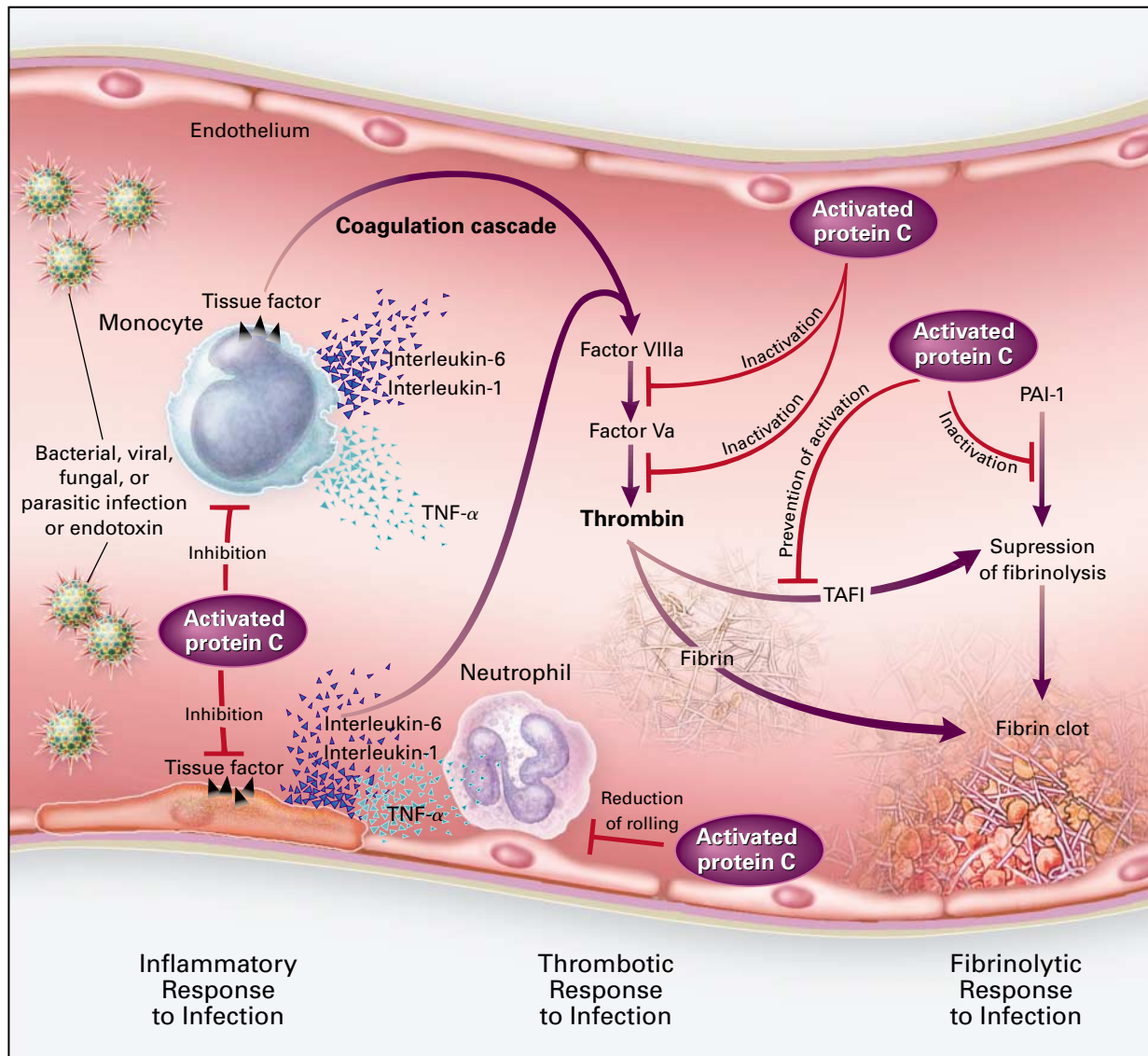
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SEVERE sepsis, defined as sepsis associated with acute organ dysfunction, results from a generalized inflammatory and procoagulant response to an infection.<sup>1</sup> The rate of death from severe sepsis ranges from 30 to 50 percent despite advances in critical care.<sup>2-5</sup> In the United States, approximately 750,000 cases of sepsis occur each year, at least 225,000 of which are fatal.<sup>6</sup>

The inflammatory and procoagulant host responses to infection are closely related.<sup>7</sup> Inflammatory cytokines, including tumor necrosis factor  $\alpha$ , interleukin-1 $\beta$ , and interleukin-6, are capable of activating coagulation and inhibiting fibrinolysis, whereas the procoagulant thrombin is capable of stimulating multiple inflammatory pathways.<sup>7-11</sup> The end result may be diffuse endovascular injury, multiorgan dysfunction, and death. Activated protein C, an endogenous protein that promotes fibrinolysis and inhibits thrombosis and inflammation, is an important modulator of the coagulation and inflammation associated with severe sepsis (Fig. 1).<sup>18</sup> Activated protein C is converted from its inactive precursor, protein C, by thrombin coupled to thrombomodulin.<sup>18</sup> The conversion of protein C

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\*Additional institutions and investigators participating in the study are listed in Appendix 3.



**Figure 1.** Proposed Actions of Activated Protein C in Modulating the Systemic Inflammatory, Procoagulant, and Fibrinolytic Host Responses to Infection.

The inflammatory and procoagulant host responses to infection are intricately linked. Infectious agents and inflammatory cytokines such as tumor necrosis factor  $\alpha$  (TNF- $\alpha$ ) and interleukin-1 activate coagulation by stimulating the release of tissue factor from monocytes and the endothelium. The presentation of tissue factor leads to the formation of thrombin and a fibrin clot. Inflammatory cytokines and thrombin can both impair the endogenous fibrinolytic potential by stimulating the release of plasminogen-activator inhibitor 1 (PAI-1) from platelets and the endothelium. PAI-1 is a potent inhibitor of tissue plasminogen activator, the endogenous pathway for lysing a fibrin clot. In addition, the procoagulant thrombin is capable of stimulating multiple inflammatory pathways and further suppressing the endogenous fibrinolytic system by activating thrombin-activatable fibrinolysis inhibitor (TAFI). The conversion of protein C, by thrombin bound to thrombomodulin, to the serine protease activated protein C is impaired by the inflammatory response. Endothelial injury results in decreased thrombomodulin levels. The end result of the host response to infection may be the development of diffuse endovascular injury, microvascular thrombosis, organ ischemia, multiorgan dysfunction, and death. Activated protein C can intervene at multiple points during the systemic response to infection. It exerts an antithrombotic effect by inactivating factors Va and VIIIa, limiting the generation of thrombin. As a result of decreased thrombin levels, the inflammatory, procoagulant, and antifibrinolytic response induced by thrombin is reduced. In vitro data indicate that activated protein C exerts an antiinflammatory effect by inhibiting the production of inflammatory cytokines (TNF- $\alpha$ , interleukin-1, and interleukin-6) by monocytes and limiting the rolling of monocytes and neutrophils on injured endothelium by binding selectins. Activated protein C indirectly increases the fibrinolytic response by inhibiting PAI-1.<sup>12-17</sup>

to activated protein C may be impaired during sepsis as a result of the down-regulation of thrombomodulin by inflammatory cytokines.<sup>19</sup> Reduced levels of protein C are found in the majority of patients with sepsis and are associated with an increased risk of death.<sup>20-23</sup>

Previous preclinical and clinical studies showed that the administration of activated protein C may improve the outcome of severe sepsis. The administration of activated protein C was protective in a baboon model of lethal *Escherichia coli* sepsis.<sup>24</sup> In a placebo-controlled phase 2 trial in patients with severe sepsis, an infusion of drotrecogin alfa (activated), or recombinant human activated protein C (Eli Lilly, Indianapolis), hereafter referred to as drotrecogin alfa activated, resulted in dose-dependent reductions in the plasma levels of D-dimer and serum levels of interleukin-6, markers of coagulopathy and inflammation, respectively.<sup>25</sup> We therefore evaluated whether the administration of drotrecogin alfa activated would reduce the rate of death from all causes at 28 days in patients with severe sepsis and have an acceptable safety profile.

## METHODS

### Patients

From July 1998 through June 2000, eligible patients were enrolled in this randomized, double-blind, placebo-controlled trial, which was conducted at 164 centers in 11 countries. The institutional review board at each center approved the protocol, and written informed consent was obtained from all participants or their authorized representatives. The clinical coordinating center (Vanderbilt Coordinating Center, Nashville) was available 24 hours a day throughout the study to answer investigators' questions regarding patients' eligibility and safety and the reporting of serious adverse events.

### Selection Criteria

The criteria for severe sepsis were a modification of those defined by Bone et al. (Appendix 1).<sup>26</sup> Patients were eligible for the trial if they had a known or suspected infection on the basis of clinical data at the time of screening and if they met the following criteria within a 24-hour period: three or more signs of systemic inflammation and the sepsis-induced dysfunction of at least one organ or system that lasted no longer than 24 hours. Patients had to begin treatment within 24 hours after they met the inclusion criteria. Exclusion criteria are summarized in Appendix 2.

### Treatment Assignments

Patients were randomly assigned in a 1:1 manner to receive drotrecogin alfa activated or placebo (0.9 percent saline with or without 0.1 percent human serum albumin) at each center. Block randomization stratified according to site was used, and all assignments were made through a central randomization center. Drotrecogin alfa activated, at a dose of 24  $\mu\text{g}$  per kilogram of body weight per hour, or placebo was administered intravenously at a constant rate from foil-wrapped bags for a total duration of 96 hours. The patients, investigators, and the sponsor were unaware of the patients' treatment assignments. Drotrecogin alfa activated was produced from an established mammalian cell line into which the complementary DNA for human protein C had been inserted.<sup>27</sup>

The infusion was interrupted 1 hour before any percutaneous procedure or major surgery and was resumed 1 hour and 12 hours later, respectively, in the absence of bleeding complications. The study protocol did not call for a standardized approach to critical care (e.g., the use of antibiotics, fluids, vasopressors, or ventilatory support).

### Evaluation of Patients

Patients were followed for 28 days after the start of the infusion or until death. Base-line characteristics including demographic information and information on preexisting conditions, organ function, markers of disease severity, infection, and hematologic and other laboratory tests were assessed within 24 hours before the infusion was begun. Blood samples obtained at base line, on days 1 through 7, and on days 14 and 28 were assayed for D-dimer levels (Liatest D-D1 latex agglutination test kit, Diagnostica Stago, Asnieres, France) and for interleukin-6 levels (Quantikine HS enzyme immunoassay kit, R & D Systems, Minneapolis). All measurements were performed by a central laboratory (Covance Central Lab Services, Indianapolis). Blood samples for the measurement of neutralizing antibodies against activated protein C were collected on days 14 and 28 or at the time of discharge from the hospital if it occurred before one or both of these dates. Microbiologic-culture results were assessed each day beginning 48 hours after the initiation of the infusion and continuing through day 28. Patients were defined as having a deficiency of protein C if their plasma protein C activity level was below the lower limit of normal (81 percent) within 24 hours before the initiation of the infusion and defined as having septic shock if they met the criteria for cardiovascular dysfunction at any time within 6 hours before the start of the infusion.

### Statistical Analysis

The primary efficacy end point was death from any cause and was assessed 28 days after the initiation of the infusion. Our prospectively defined primary analysis included all patients who received the infusion for any length of time, with patients analyzed according to the treatment group to which they were assigned at randomization. The trial was designed to enroll 2280 patients; two planned interim analyses by an independent data and safety monitoring board occurred after 760 and 1520 patients had been enrolled. Statistical guidelines to suspend enrollment if drotrecogin alfa activated was found to be significantly more efficacious than placebo were determined a priori and used the O'Brien-Fleming spending function according to the method of Lan and DeMets.<sup>28</sup>

Data were analyzed according to a prospectively defined plan. The primary analysis was based on a Cochran-Mantel-Haenszel test in which the groups were stratified on the basis of three base-line covariates: severity of disease, as reflected by the score on the Acute Physiology and Chronic Health Evaluation II (APACHE II)<sup>29</sup> (3 to 19, 20 to 24, 25 to 29, or 30 to 53, with higher scores indicating more severe disease); age (younger than 60 years or 60 years or older); and plasma protein C activity level (40 percent or less, 41 to 60 percent, 61 to 80 percent, 81 percent or more, or unknown). The corresponding relative risk and 95 percent confidence interval were calculated with use of the logit-adjusted method. The time from the start of the infusion to death was compared in the two groups in a similar manner with use of a stratified log-rank test. Results of both stratified and nonstratified analyses are reported. We evaluated the consistency of the effects of treatment on the risk of death in the subgroups by determining whether the relative risk and 95 percent confidence interval for each subgroup included the observed relative risk for the entire population.

Changes from base-line levels of plasma D-dimer and serum interleukin-6 were analyzed in patients who had subsequent measurements with the use of analysis of variance of ranked data. For patients with missing data, we used the last-observation-carried-forward method of imputation. The proportion of patients who had serious adverse events and new infections was compared in the two groups with the use of Pearson's chi-square tests. All reported P values are two-sided.

## RESULTS

At the time of the second interim analysis of data from 1520 patients, enrollment was suspended because the differences in the mortality rate between the two groups exceeded the a priori guideline for stopping

the trial. Results presented here include data from additional patients who were enrolled before the completion of the second interim analysis.

#### Base-Line Characteristics of the Patients

Of 1728 patients who underwent randomization, 1690 received the study drug or placebo. Thirty-eight patients (17 in the placebo group and 21 in the drotrecogin alfa activated group) never received any study drug. In the drotrecogin alfa activated group, 14 patients met at least one exclusion criterion, 4 patients became moribund before the infusion could be started, and consent was withdrawn before the infusion in the case of 3 patients. In the placebo group, 15 patients did not meet the entry criteria for the study, and 2 patients became moribund before the infusion was begun.

All randomized patients were followed for the en-

tire 28-day study period except for one patient in the drotrecogin alfa activated group who did not receive the study drug. This patient was classified as having died on day 28 in the mortality analysis of all randomized patients.

At base line, the demographic characteristics and severity of disease were similar in the placebo group and the drotrecogin alfa activated group (Table 1). Approximately 75 percent of the patients had at least two dysfunctional organs or systems at the time of enrollment. The lungs and the abdomen were the most common sites of infection, occurring in 53.6 percent and 19.9 percent of the patients, respectively, in the two groups combined (Table 2). The incidence of gram-positive and gram-negative infections was similar within each group and between the two groups. A blinded clinical evaluation committee determined that clinically appropriate antibiotic therapy that was based on the site of infection and available culture and susceptibility data was started within 48 hours of the diagnosis of severe sepsis and continued for at least five days or until death in 776 patients in the drotrecogin alfa activated group (91.3 percent) and in 766 patients in the placebo group (91.2 percent). Base-line levels of indicators of coagulopathy and inflammation were also similar in the two groups (Table 3). Protein C deficiency was present in 87.6 percent of the patients (1379 of 1574) for whom levels were obtained. In addition, plasma D-dimer and serum interleukin-6 levels were elevated in 99.7 and 98.5 percent of the patients, respectively. Among treated patients, 82.4 percent of those in the placebo group and 81.8 percent of those in the drotrecogin alfa activated group received at least 90 percent of the intended infusion and 8.2 percent and 6.4 percent, respectively, died during the 96-hour period of infusion.

#### Efficacy

Twenty-eight days after the start of the infusion, 259 of 840 patients in the placebo group (30.8 percent) and 210 of 850 (24.7 percent) of the patients in the drotrecogin alfa activated group had died. This difference in the rate of death from any cause was significant ( $P=0.005$  in the nonstratified analysis) (Table 4) and was associated with an absolute reduction in the risk of death of 6.1 percent. The prospectively defined primary analysis in which the groups were stratified according to the base-line APACHE II score, age, and protein C activity produced similar results ( $P=0.005$ ), as did the analysis that included the 38 patients who underwent randomization but who never received the infusion ( $P=0.003$ ). The results of the prospectively defined primary analysis represent a reduction in the relative risk of death of 19.4 percent (95 percent confidence interval, 6.6 to 30.5) in association with treatment with drotrecogin alfa activated, as compared with placebo. A Kaplan–Meier analysis of survival yielded similar results ( $P=0.006$ ) (Fig. 2). The absolute dif-

TABLE 1. BASE-LINE CHARACTERISTICS OF THE PATIENTS.\*

CHARACTERISTIC	PLACEBO GROUP (N=840)	DROTRECUGIN ALFA ACTIVATED GROUP (N=850)
Age (yr)	60.6±16.5	60.5±17.2
Age (%)		
<60 yr	43.6	44.1
<65 yr	53.5	51.4
<75 yr	78.5	75.9
Male sex (%)	58.0	56.1
White race (%)	82.0	81.8
Prior or preexisting conditions (%)		
Hypertension	35.0	38.2
Myocardial infarction	14.4	12.1
Congestive cardiomyopathy	9.0	6.4
Diabetes	22.4	20.7
Pancreatitis	3.9	3.4
Liver disease	2.6	2.1
COPD	26.1	22.2
Cancer	18.8	17.1
Recent trauma	5.1	3.3
Recent surgical history (%)		
Elective surgery	6.2	5.8
Emergency surgery	21.2	20.7
No history of surgery	72.6	73.5
APACHE II score	25.0±7.8	24.6±7.6
Other indicators of disease severity (%)		
Mechanical ventilation	77.6	73.3
Shock	71.7	70.4
Use of any vasopressor	75.5	71.8
Use of dobutamine	13.5	13.9
No. of dysfunctional organs or systems (%)		
0	0	0.1
1	24.2	25.3
2	32.5	31.8
3	26.0	25.2
4	13.8	14.0
5	3.6	3.6
Time from first organ dysfunction to start of drug infusion (hr)	17.4±9.1	17.5±12.8

\*Plus-minus values are means ±SD. COPD denotes chronic obstructive pulmonary disease, and APACHE II Acute Physiology and Chronic Health Evaluation II. Because of rounding, not all percentages total 100.

**TABLE 2. SITES AND CAUSES OF INFECTION IN PATIENTS WITH SEVERE SEPSIS.**

VARIABLE	PLACEBO GROUP	DROTRECOGIN
	(N=840)	ALFA ACTIVATED GROUP (N=850)
	percent	
Site of infection*		
Lung	53.6	53.6
Abdomen	19.9	20.0
Urinary tract	10.2	10.0
Other†	16.3	16.4
Positive blood culture	32.5	32.7
Results of Gram's staining of bacterial pathogen		
Purely gram-negative	23.3	21.8
Purely gram-positive	25.1	25.8
Mixed	13.9	15.6
Unconfirmed	5.4	3.3
Culture negative or not obtained	32.3	33.5
Type of organism‡		
Gram-positive		
<i>Staphylococcus aureus</i>	14.4	14.1
Other staphylococcus species	6.2	7.1
<i>Streptococcus pneumoniae</i>	11.3	12.5
Other streptococcus species	9.2	8.6
Enterococcus species	6.5	7.4
Other gram-positive	3.0	3.9
Gram-negative		
<i>Escherichia coli</i>	17.4	15.5
Klebsiella species	6.8	5.5
Pseudomonas species	5.1	6.6
Enterobacter species	4.2	4.8
<i>Haemophilus influenzae</i>	4.2	3.1
Bacteroides species	3.0	3.6
Other gram-negative	10.2	9.5
Fungus		
<i>Candida albicans</i>	1.7	2.0
Other candida species	5.0	4.5
Yeast	1.1	1.1
Other fungus	0.8	0.8

\*The site of infection was either documented or presumed on the basis of clinical findings.

†Other sites of infection included the blood, skin, central nervous system, bones and joints, cardiac system, and reproductive organs.

‡Patients may have had more than one organism cultured.

ference in survival between the two groups was evident within days after the initiation of the infusion and continued to increase throughout the remainder of the study period.

Prospectively defined subgroup analyses were performed for a number of base-line characteristics, including the APACHE II score, the number of dysfunctional organs or systems, other indicators of the severity of disease, sex, age, the site of infection, the type of infection (gram-positive, gram-negative, or mixed), and presence or absence of protein C deficiency. A consistent effect of treatment with drotrecogin alfa activated was observed among the subgroups (data not shown), including the subgroup with protein C deficiency and the subgroup with normal protein C levels.

**TABLE 3. BASE-LINE LEVELS OF INDICATORS OF COAGULATION AND INFLAMMATION.\***

VARIABLE	PLACEBO GROUP	DROTRECOGIN ALFA ACTIVATED GROUP
Plasma D-dimer		
No. of patients	758	792
Median level ( $\mu\text{g}/\text{ml}$ )	4.15	4.22
Interquartile range ( $\mu\text{g}/\text{ml}$ )	2.18–8.65	2.28–8.11
Serum interleukin-6		
No. of patients	808	827
Median level (pg/ml)	484	497
Interquartile range (pg/ml)	129–2540	153–2701
Plasma protein C activity		
No. of patients	775	799
Median level (%)	50	47
Interquartile range (%)	33–68	30–63
Protein C deficiency (% of patients)		
Yes	79.8	83.4
No	12.5	10.6
Unknown	7.7	6.0

\*The normal range of D-dimer levels is 0.0 to 0.39  $\mu\text{g}$  per milliliter. The normal range of interleukin-6 levels is 0.38 to 10.09 pg per milliliter. The normal range of protein C activity is 81 percent to 173 percent. A deficiency of protein C was defined as an activity level of less than 81 percent.

#### Levels of D-Dimer and Interleukin-6

Plasma D-dimer levels were significantly lower in patients in the drotrecogin alfa activated group than in patients in the placebo group on days 1 through 7 after the start of the infusion (Fig. 3). Decreases in serum interleukin-6 levels were significantly greater in the patients in the drotrecogin alfa activated group than in the patients in the placebo group on day 1 ( $P=0.009$ ) and on days 4, 5, 6, and 7 ( $P=0.025$ ,  $P=0.017$ ,  $P=0.016$ , and  $P=0.022$ , respectively).

#### Complications

The percentage of patients who had at least one serious adverse event was similar in the two groups (Table 5). The incidence of serious bleeding was higher in the drotrecogin alfa activated group than in the placebo group (3.5 percent vs. 2.0 percent) ( $P=0.06$ ). This difference in the incidence of serious bleeding was observed only during the infusion period; thereafter, the incidence was similar in the two groups. Among the patients who received drotrecogin alfa activated, the incidence of serious bleeding was similar for those who received drotrecogin alfa activated alone and those who also received heparin (3.7 percent and 3.5 percent). In both the drotrecogin alfa activated group and the placebo group, serious bleeding occurred primarily in patients with an identifiable predisposition to bleeding, such as gastrointestinal ulceration, an activated partial-thromboplastin time of more than 120 seconds, a prolonged prothrombin time (an international normalized ratio of more than 3.0), a platelet count that decreased to less than

**TABLE 4.** ANALYSIS OF THE RATES AND RISKS OF DEATH FROM ANY CAUSE AT 28 DAYS.\*

VARIABLE	PLACEBO GROUP	DROTRECUGIN ALFA ACTIVATED GROUP	P VALUE†	RELATIVE RISK OF DEATH (95% CI)‡	ABSOLUTE REDUCTION IN RISK (95% CI)§
	no./total no. (%)				
<b>Treated patients</b>					
Nonstratified analysis	259/840 (30.8)	210/850 (24.7)	0.005	0.80 (0.69 to 0.94)	6.1 (1.9 to 10.4)
Stratified analysis¶			0.005	0.81 (0.70 to 0.93)	6.2 (1.6 to 10.8)
Protein C deficiency					
Yes	215/670 (32.1)	182/709 (25.7)	0.009	0.80 (0.68 to 0.95)	6.4 (1.6 to 11.2)
No	28/105 (26.7)	14/90 (15.6)	0.06	0.58 (0.33 to 1.04)	11.1 (-0.4 to 22.6)
Unknown	16/65 (24.6)	14/51 (27.5)	0.73	1.12 (0.60 to 2.07)	-2.8 (-19.0 to 13.4)
<b>Randomized patients  </b>					
Nonstratified analysis	268/857 (31.3)	216/871 (24.8)	0.003	0.79 (0.68 to 0.92)	6.5 (2.2 to 10.7)

\*Patients were analyzed in the treatment group to which they were assigned at randomization. CI denotes confidence interval.

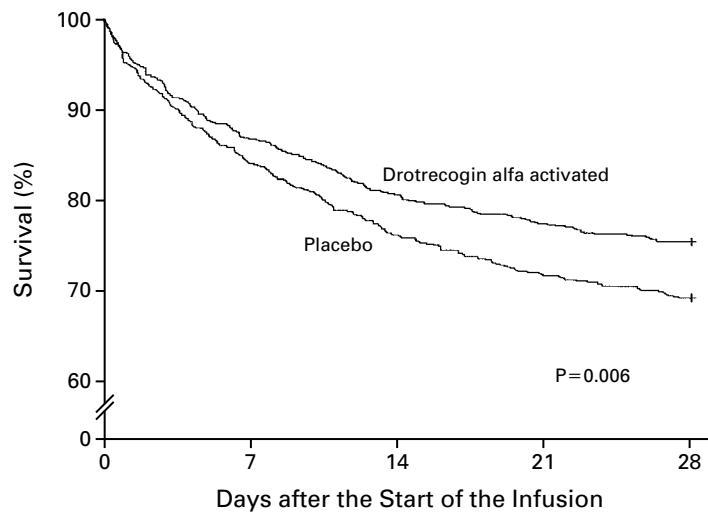
†Two-sided P values for the nonstratified and subgroup analyses are based on Pearson's chi-square tests, and the P value for the primary stratified analysis is based on the Cochran-Mantel-Haenszel test.

‡The relative risk of death is calculated as the mortality rate in the drotrecogin alfa activated group divided by the mortality rate in the placebo group.

§For the stratified analysis, the absolute reduction in risk was estimated with use of the average of the absolute reduction in risk within strata.

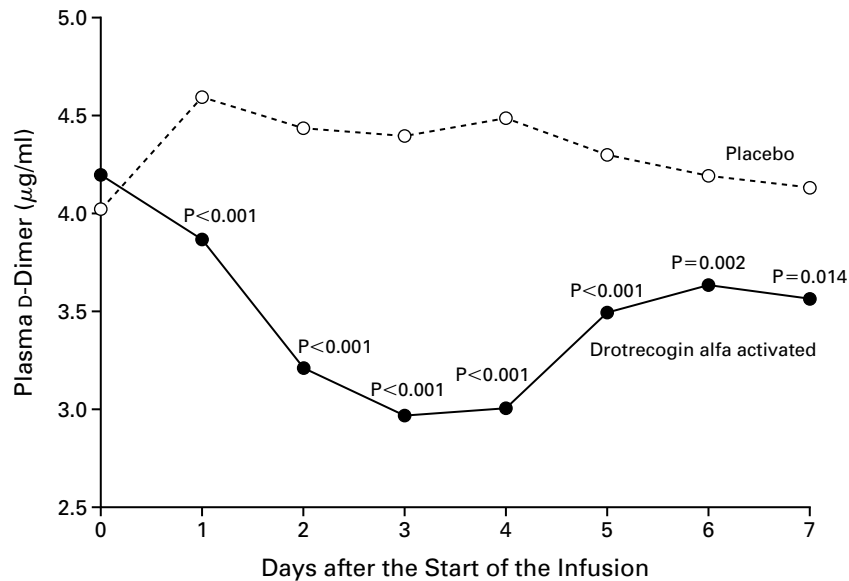
¶In the prospectively defined stratified analysis, the relative risk of death was calculated after an adjustment for the base-line APACHE II quartile, age, and protein C activity.

||This analysis included 38 patients who were randomly assigned to treatment but who never received the study drug.



No. AT RISK	0	7	14	21	28
Drotrecogin alfa activated	850	737	684	657	640
Placebo	840	705	639	602	581

**Figure 2.** Kaplan-Meier Estimates of Survival among 850 Patients with Severe Sepsis in the Drotrecogin Alfa Activated Group and 840 Patients with Severe Sepsis in the Placebo Group. Treatment with drotrecogin alfa activated was associated with a significantly higher rate of survival (P=0.006 by the stratified log-rank test).



**Figure 3.** Changes in Median Plasma D-Dimer Levels in 770 Patients with Severe Sepsis in the Drotrecogin Alfa Activated Group and 729 Patients in the Placebo Group. Only patients with base-line values and at least one subsequent value were included in the analysis. The P values are for the comparison with the placebo group.

30,000 per cubic millimeter and remained at that level despite standard therapy, traumatic injury of a blood vessel, or traumatic injury of a highly vascular organ. There was a fatal intracranial hemorrhage in two patients in the drotrecogin alfa activated group during the infusion (on day 1 and day 4) and in one patient in the placebo group six days after the end of the infusion. After adjustment for the duration of survival, blood-transfusion requirements were similar in the two groups ( $P=0.90$ ).

There were no other safety concerns associated with treatment with drotrecogin alfa activated on the basis of assessments of organ dysfunction, vital signs, serum chemical data, or hematologic data. The incidence of thrombotic events was similar in the two groups (Table 5). New infections occurred in 25.5 percent of the patients in the drotrecogin alfa activated group and 25.1 percent of the patients in the placebo group ( $P=0.85$ ). Neutralizing antibodies against activated protein C were not detected in any patient.

**DISCUSSION**

In this study, the administration of drotrecogin alfa activated reduced the rate of death from any cause at 28 days in patients with a clinical diagnosis of severe sepsis, resulting in a 19.4 percent reduction in the relative risk of death and an absolute reduction of 6.1 percent. A survival benefit was evident throughout

**TABLE 5.** INCIDENCE OF SERIOUS ADVERSE EVENTS.

VARIABLE	PLACEBO GROUP (N=840)	DROTRECOGIN ALFA ACTIVATED GROUP (N=850)	P VALUE
	no. of patients (%)		
At least one serious adverse event	102 (12.1)	106 (12.5)	0.84
Serious bleeding event*	17 (2.0)	30 (3.5)	0.06
Gastrointestinal	9 (1.1)	9 (1.1)	
Intraabdominal	4 (0.5)	3 (0.4)	
Intrathoracic	1 (0.1)	6 (0.7)	
Retroperitoneal	0	4 (0.5)	
Intracranial	1 (0.1)	2 (0.2)	
Skin or soft tissue	0	2 (0.2)	
Genitourinary	0	2 (0.2)	
Source unidentified†	2 (0.2)	2 (0.2)	
Thrombotic events	25 (3.0)	17 (2.0)	0.20

\*A serious bleeding event was defined as any intracranial hemorrhage, any life-threatening bleeding, any bleeding event classified as serious by the investigator, or any bleeding that required the administration of 3 units of packed red cells on two consecutive days.

†These patients received 3 units of packed red cells on two consecutive days but had no identifiable source of bleeding.

the 28-day study period, whether or not the groups were stratified according to the severity of disease. Our results indicate that in this population, 1 additional life would be saved for every 16 patients treated with drotrecogin alfa activated.

Though the study population was heterogeneous with respect to clinical features, it was homogeneous with respect to the biochemical evidence of systemic inflammation and coagulopathy. In these patients, the benefit of drotrecogin alfa activated is most likely explained by the drug's biologic activity. Activated protein C inhibits the generation of thrombin by inactivating factor Va and factor VIIIa.<sup>30,31</sup> As compared with the patients who received placebo, patients who received drotrecogin alfa activated had greater decreases in plasma D-dimer levels during the first seven days after the infusion was initiated, indicating a reduction in the generation of thrombin. The rise in D-dimer levels after the completion of the 96-hour infusion of drotrecogin alfa activated indicates incomplete resolution of the procoagulant state seen in patients with sepsis. An evaluation of longer periods of infusion of drotrecogin alfa activated may be warranted.

Treatment with drotrecogin alfa activated decreased inflammation, as indicated by decreases in interleukin-6 levels, a finding consistent with the known antiinflammatory activity of activated protein C. The antiinflammatory activity of drotrecogin alfa activated may be mediated indirectly through the inhibition of the generation of thrombin, which leads to decreased activation of platelets, recruitment of neutrophils, and degranulation of mast cells.<sup>8</sup> Furthermore, preclinical studies demonstrated that activated protein C has direct antiinflammatory properties, including the inhibition of neutrophil activation, the production of cytokines by lipopolysaccharide-challenged monocytes, and E-selectin-mediated adhesion of cells to vascular endothelium.<sup>32-34</sup>

A consistent effect of treatment with drotrecogin alfa activated was seen among the subgroups examined, including those stratified according to age, APACHE II score, sex, number of dysfunctional organs or systems, type of infection (gram-positive, gram-negative, or mixed), site of infection, and presence or absence of protein C deficiency at study entry. Reductions in the relative risk of death were observed regardless of whether the patients had a deficiency of

protein C at base line, suggesting that drotrecogin alfa activated has pharmacologic effects that go beyond simple physiologic replacement of activated protein C. This observation further suggests that measurements of protein C are not necessary to identify which patients would benefit from treatment with drotrecogin alfa activated. A consistent treatment effect was also observed regardless of the site of infection or the type of infection.

It was consistent with the antithrombotic activity of drotrecogin alfa activated that bleeding was the most common adverse event associated with the administration of the drug. The incidence of serious bleeding suggests that 1 additional serious bleeding event would occur for every 66 patients treated with drotrecogin alfa activated. Serious bleeding tended to occur in patients with predisposing conditions, such as gastrointestinal ulceration, traumatic injury of a blood vessel or highly vascular organ injury, or markedly abnormal values for indicators of coagulation (e.g., the platelet count, the activated partial-thromboplastin time, and the prothrombin time). The incidence of thrombotic events was not increased by treatment with drotrecogin alfa activated, and the antiinflammatory effect was not associated with an increased incidence of new infections. Treatment with drotrecogin alfa activated was not associated with the development of neutralizing antibodies against activated protein C.

In summary, the biologic activity of drotrecogin alfa activated was demonstrated by the finding of greater decreases in D-dimer and interleukin-6 levels in patients who received drotrecogin alfa activated than in those who received placebo. The higher incidence of serious bleeding during infusion in the drotrecogin alfa activated group is consistent with the antithrombotic activity of the drug and occurs predominantly in patients with a predisposition to bleeding. In patients with severe sepsis, an intravenous infusion of drotrecogin alfa activated at a dose of 24  $\mu\text{g}$  per kilogram per hour for 96 hours was associated with a significant reduction in mortality and a safety profile that was acceptable within the context of this clinical trial.

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APPENDIX 1. SUMMARY OF INCLUSION CRITERIA.\*

INFECTION CRITERIA	MODIFIED SIRS CRITERIA	CRITERIA FOR DYSFUNCTIONAL ORGANS OR SYSTEMS†
<p>Patients had to have a known infection or a suspected infection, as evidenced by one or more of the following: white cells in a normally sterile body fluid; perforated viscus; radiographic evidence of pneumonia in association with the production of purulent sputum; a syndrome associated with a high risk of infection (e.g., ascending cholangitis).</p>	<p>Patients had to meet at least three of the following four criteria: a core temperature of <math>\geq 38^{\circ}\text{C}</math> (<math>100.4^{\circ}\text{F}</math>) or <math>\leq 36^{\circ}\text{C}</math> (<math>96.8^{\circ}\text{F}</math>); a heart rate of <math>\geq 90</math> beats/min, except in patients with a medical condition known to increase the heart rate or those receiving treatment that would prevent tachycardia; a respiratory rate of <math>\geq 20</math> breaths/min or a <math>\text{PaCO}_2</math> of <math>\leq 32</math> mm Hg or the use of mechanical ventilation for an acute respiratory process; a white-cell count of <math>\geq 12,000/\text{mm}^3</math> or <math>\leq 4,000/\text{mm}^3</math> or a differential count showing <math>&gt;10</math> percent immature neutrophils.</p>	<p>Patients had to meet at least one of the following five criteria: for cardiovascular-system dysfunction, the arterial systolic blood pressure had to be <math>\leq 90</math> mm Hg or the mean arterial pressure <math>\leq 70</math> mm Hg for at least 1 hour despite adequate fluid resuscitation, adequate intravascular volume status or the use of vasopressors in an attempt to maintain a systolic blood pressure of <math>\geq 90</math> mm Hg or a mean arterial pressure of <math>\geq 70</math> mm Hg; for kidney dysfunction, urine output had to be <math>&lt;0.5</math> ml/kg of body weight/hr for 1 hour, despite adequate fluid resuscitation; for respiratory-system dysfunction, the ratio of <math>\text{PaO}_2</math> to <math>\text{FiO}_2</math> had to be <math>\leq 250</math> in the presence of other dysfunctional organs or systems or <math>\leq 200</math> if the lung was the only dysfunctional organ; for hematologic dysfunction, the platelet count had to be <math>&lt;80,000/\text{mm}^3</math> or to have decreased by 50 percent in the 3 days preceding enrollment; in the case of unexplained metabolic acidosis, the pH had to be <math>\leq 7.30</math> or the base deficit had to be <math>\geq 5.0</math> mmol/liter in association with a plasma lactate level that was <math>&gt;1.5</math> times the upper limit of the normal value for the reporting laboratory.</p>

\*SIRS denotes systemic inflammatory response syndrome,  $\text{PaCO}_2$  partial pressure of arterial carbon dioxide,  $\text{PaO}_2$  partial pressure of arterial oxygen, and  $\text{FiO}_2$  fraction of inspired oxygen.

†The first sepsis-induced organ or system dysfunction had to develop within 24 hours before study enrollment.

APPENDIX 2. SUMMARY OF EXCLUSION CRITERIA.

Pregnancy or breast-feeding  
 Age  $<18$  yr or weight  $>135$  kg  
 Platelet count  $<30,000/\text{mm}^3$   
 Conditions that increased the risk of bleeding: surgery requiring general or spinal anesthesia within 12 hours before the infusion, the potential need for such surgery during the infusion, or evidence of active bleeding postoperatively; a history of severe head trauma requiring hospitalization, intracranial surgery, or stroke within 3 months before the study or any history of intracerebral arteriovenous malformation, cerebral aneurysm, or mass lesions of the central nervous system; a history of congenital bleeding diatheses; gastrointestinal bleeding within 6 weeks before the study unless corrective surgery had been performed; and trauma considered to increase the risk of bleeding  
 Known hypercoagulable condition, including resistance to activated protein C; hereditary deficiency of protein C, protein S, or antithrombin III; presence of anticardiolipin antibody, antiphospholipid antibody, lupus anticoagulant, or homocysteinemia; or recently documented (within 3 months before the study) or highly suspected deep-vein thrombosis or pulmonary embolism  
 Patient's family, physician, or both not in favor of aggressive treatment of patient or presence of an advanced directive to withhold life-sustaining treatment  
 Patient not expected to survive 28 days because of uncorrectable medical condition, such as poorly controlled neoplasm or other end-stage disease  
 Moribund state in which death was perceived to be imminent  
 Human immunodeficiency virus infection in association with a last known CD4 count of  $\leq 50/\text{mm}^3$   
 History of bone marrow, lung, liver, pancreas, or small-bowel transplantation  
 Chronic renal failure requiring hemodialysis or peritoneal dialysis\*  
 Known or suspected portosystemic hypertension, chronic jaundice, cirrhosis, or chronic ascites  
 Acute pancreatitis with no established source of infection  
 Participation in another investigational study within 30 days before the current study  
 Use of any of the following medications or treatment regimens: unfractionated heparin to treat an active thrombotic event within 8 hours before the infusion†; low-molecular-weight heparin at a higher dose than recommended for prophylactic use (as specified in the package insert) within 12 hours before the infusion; warfarin (if used within 7 days before study entry and if the prothrombin time exceeded the upper limit of the normal range for the institution); acetylsalicylic acid at a dose of more than 650 mg/day within 3 days before the study; thrombolytic therapy within 3 days before the study‡; glycoprotein IIb/IIIa antagonists within 7 days before study entry; antithrombin III at a dose of more than 10,000 U within 12 hours before the study; or protein C within 24 hours before the study

\*Acute renal failure was not an exclusion criterion.

†Prophylactic treatment with a dose of unfractionated heparin of up to 15,000 U per day was permitted.

‡Thrombolytic agents were permitted for the treatment of thromboses within a catheter.

## APPENDIX 3

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## REFERENCES

1. Bone RC, Grodzin CJ, Balk RA. Sepsis: a new hypothesis for pathogenesis of the disease process. *Chest* 1997;112:235-43.
2. Angus DC, Birmingham MC, Balk RA, et al. E5 murine monoclonal antiendotoxin antibody in gram-negative sepsis: a randomized controlled trial. *JAMA* 2000;283:1723-30.
3. Rangel-Frausto MS, Pittet D, Costigan M, Hwang T, Davis CS, Wenzel RP. The natural history of the systemic inflammatory response syndrome (SIRS): a prospective study. *JAMA* 1995;273:117-23.
4. Annane D, Sebille V, Troche G, Raphael JC, Gajdos P, Bellissant E. A 3-level prognostic classification in septic shock based on cortisol levels and cortisol response to corticotropin. *JAMA* 2000;283:1038-45.
5. Increase in National Hospital Discharge Survey rates for septicemia — United States, 1979-1987. *JAMA* 1990;263:937-8.
6. Linde-Zwirble WT, Angus DC, Carcillo J, Lidicker J, Clermont G, Pinsky MR. Age-specific incidence and outcome of sepsis in the US. *Crit Care Med* 1999;27:Suppl 1:A33. abstract.
7. Esmon CT, Taylor FB Jr, Snow TR. Inflammation and coagulation: linked processes potentially regulated through a common pathway mediated by protein C. *Thromb Haemost* 1991;66:160-5.
8. Yan SB, Grinnell BW. Recombinant human protein C, protein S, and thrombomodulin as antithrombotics. *Perspect Drug Discovery Des* 1993;1:503-20.
9. Stouthard JM, Levi M, Hack CE, et al. Interleukin-6 stimulates coagulation, not fibrinolysis, in humans. *Thromb Haemost* 1996;76:738-42.
10. Conkling PR, Greenberg CS, Weinberg JB. Tumor necrosis factor induces tissue factor-like activity in human leukemia cell line U937 and peripheral blood monocytes. *Blood* 1988;72:128-33.
11. Bevilacqua MP, Pober JS, Majeau GR, Fiers W, Cotran RS, Gimbrone MA Jr. Recombinant tumor necrosis induces procoagulant activity in cultured human vascular endothelium: characterization and comparison with the actions of interleukin 1. *Proc Natl Acad Sci U S A* 1986;12:4533-7.
12. Vervloet MG, Thijs LG, Hack CE. Derangements of coagulation and fibrinolysis in critically ill patients with sepsis and septic shock. *Semin Thromb Hemost* 1998;24:33-44.
13. Levi M, ten Cate B. Disseminated intravascular coagulation. *N Engl J Med* 1999;341:586-92.
14. Iba T, Kidokoro A, Yagi Y. The role of the endothelium in changes in procoagulant activity in sepsis. *J Am Coll Surg* 1998;187:321-9.
15. Neshelm M, Wang B, Boffa M, Nagashima M, Morser J, Bajzar L. Thrombin, thrombomodulin and TAFI in the molecular link between coagulation and fibrinolysis. *Thromb Haemost* 1997;78:386-91.
16. Esmon CT. Inflammation and thrombosis: mutual regulation by protein C. *Immunologist* 1998;6:84-9.
17. Bajzar L, Neshelm ME, Tracy PB. The profibrinolytic effect of activated protein C in clots formed from plasma is TAFI-dependent. *Blood* 1996;88:2093-100.
18. Esmon CT. The protein C anticoagulant pathway. *Arterioscler Thromb* 1992;12:135-45.
19. Boehme MW, Deng Y, Raeth U, et al. Release of thrombomodulin from endothelial cells by concerted action of TNF-alpha and neutrophils: in vivo and in vitro studies. *Immunology* 1996;87:134-40.
20. Fourrier F, Chopin C, Goudemand J, et al. Septic shock, multiple organ failure, and disseminated intravascular coagulation: compared patterns of antithrombin III, protein C, and protein S deficiencies. *Chest* 1992;101:816-23.

21. Lorente JA, Garcia-Frade LJ, Landin L, et al. Time course of hemostatic abnormalities in sepsis and its relation to outcome. *Chest* 1993;103:1536-42.
22. Boldt J, Papsdorf M, Rothe A, Kumle B, Piper S. Changes of the hemostatic network in critically ill patients — is there a difference between sepsis, trauma, and neurosurgery patients? *Crit Care Med* 2000;28:445-50.
23. Powars D, Larsen R, Johnson J, et al. Epidemic meningococemia and purpura fulminans with induced protein C deficiency. *Clin Infect Dis* 1993;17:254-61.
24. Taylor FB Jr, Chang A, Esmon CT, D'Angelo A, Vigano-D'Angelo S, Blick KE. Protein C prevents the coagulopathic and lethal effects of *Escherichia coli* infusion in the baboon. *J Clin Invest* 1987;79:918-25.
25. Hartman DL, Bernard GR, Helterbrand JD, Yan SB, Fisher CJ. Recombinant human activated protein C (rhAPC) improves coagulation abnormalities associated with severe sepsis. *Intensive Care Med* 1998;24: Suppl 1:S77. abstract.
26. Bone RC, Balk RA, Cerra FB, et al. Definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. *Chest* 1992;101:1644-55.
27. Yan SC, Razzano P, Chao YB, et al. Characterization and novel purification of recombinant human protein C from three mammalian cell lines. *Biotechnology (N Y)* 1990;8:655-61.
28. Lan KKG, DeMets DL. Discrete sequential boundaries for clinical trials. *Biometrika* 1983;70:659-63.
29. Knaus WA, Draper EA, Wagner DP, Zimmerman JE. APACHE II: a severity of disease classification system. *Crit Care Med* 1985;13:818-29.
30. Walker FJ, Sexton PW, Esmon CT. The inhibition of blood coagulation by activated protein C through the selective inactivation of activated factor V. *Biochim Biophys Acta* 1979;571:333-42.
31. Fulcher CA, Gardiner JE, Griffin JH, Zimmerman TS. Proteolytic inactivation of human factor VIII procoagulant protein by activated human protein C and its analogy with factor V. *Blood* 1984;63:486-9.
32. Grey ST, Tsuchida A, Hau H, Orthner CL, Salem HH, Hancock WW. Selective inhibitory effects of the anticoagulant activated protein C on the responses of human mononuclear phagocytes to LPS, IFN-gamma, or phorbol ester. *J Immunol* 1994;153:3664-72.
33. Hirose K, Okajima K, Taoka Y, et al. Activated protein C reduces the ischemia/reperfusion-induced spinal cord injury in rats by inhibiting neutrophil activation. *Ann Surg* 2000;232:272-80.
34. Grinnell BW, Hermann RB, Yan SB. Human protein C inhibits selectin-mediated cell adhesion: role of unique fucosylated oligosaccharide. *Glycobiology* 1994;4:221-5.

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