Thromboelastometry (TEM®) Findings in Disseminated Intravascular Coagulation in a Pig Model of Endotoxinemia

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Standard coagulation tests have a low specificity and sensitivity for diagnosing disseminated intravascular coagulation. The aim of this study was to determine whether whole blood thromboelastometry (TEM) detects lipopolysaccharide (LPS)-induced changes in coagulation. Blood samples from 10 pigs were drawn at baseline, before and at the end of LPS infusion and 2, 3, 4 and 5 h after the start of endotoxinemia. Simultaneous to TEM, standard coagulation tests and extended coagulation analysis including tissue plasminogen activator (t-PA) and plasminogen activator inhibitor 1 (PAI-1) were performed. Endotoxinemia resulted in a significant acceleration of the nonactivated TEM (NATEM) clotting time 2 h after the end of LPS infusion; in contrast, the changes in international normalized ratio and activated partial thromboplastin time suggested delayed initiation of coagulation. NATEM maximum clot firmness (MCF) and fibrin-based thromboelastometry test (FIBTEM®)-MCF decreased significantly from baseline until the last time point (from 64.6 ± 7.8 and 35.1 ± 12.8 mm to 52.8 ± 4.6 and 21.4 ± 11.8 mm, respectively; $P = 0.01$ for both parameters). A sharp, transient increase of t-PA had no effect on maximum lysis in the NATEM test. PAI-1 increased significantly 3 h after the start of LPS infusion, paralleled by a decrease in maximum lysis. In conclusion, TEM was superior to standard coagulation tests in reflecting initial activation of coagulation during endotoxinemia. TEM further suggested consumption of coagulation substrate; at the same time, inhibition of plasminogen activation was accompanied by improved clot stability. Further investigations are necessary to establish the clinical relevance of these findings.
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INTRODUCTION

Massive infection activates the procoagulant pathway, resulting in disseminated intravascular coagulation (DIC), microthrombosis and organ failure (1–3). DIC occurs in approximately 30% of patients with severe sepsis (4). Diagnosis of DIC is complex because of the lack of specific tests (5). Routine coagulation analysis such as prothrombin time (PT) and activated partial thromboplastin time (aPTT) are available in most laboratories but have a low specificity and sensitivity for diagnosing DIC (6). Furthermore, increased activation of the procoagulant pathway and activation or inhibition of the fibrinolytic system cannot be portrayed accurately by these standard assays (7–10).

In contrast to routine coagulation tests, thromboelastometry not only provides an assessment of the initiation of coagulation, but also assesses the clot formation process and the clot quality and stability. Diverse reagents help to evaluate different aspects of the coagulation and fibrinolytic system (11). Thromboelastometry can be conducted at the patient's bedside, making it suitable as a point of care monitoring tool (12). The measurements are performed in whole blood and not in plasma; thus, platelets and activated monocytes expressing tissue factor, not removed by a centrifugation process, are also taken into consideration.

The aim of the study was to determine whether whole blood coagulation thromboelastometry is able to identify lipopolysaccharide (LPS)-induced DIC in a porcine model. Thromboelastometry was performed in parallel with standard coagulation analysis and extended coagulation profiles including antithrombin (AT) III, protein C, thrombin antithrombin complex (TAT), D-dimers, tissue plasminogen activator (t-PA) and plasminogen activator inhibitor 1 (PAI-1).

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The hypothesis was that coagulation monitoring by thromboelastometry offers a useful alternative to routine coagulation tests for assessing DIC.

MATERIALS AND METHODS

Animals

Ten healthy male pigs, between 27 and 43 kg body weight, were used for the study. The experimental protocol was approved by the Animal Protocol Review Board of the city government of Vienna, Austria. All experiments were performed under the conditions described in the *Guide for the Care and Use of Laboratory Animals*, as defined by the National Institutes of Health.

Anesthesia, Surgical Preparations and Measurements

Anesthesia was induced with ketamine (0.83 mg/kg; Pfizer, Vienna, Austria) and xylazine (1.0 mg/kg; Bayer HealthCare, Wuppertal, Germany) plus butorphanol (0.17 mg/kg; Alvetra and Werfft AG, Vienna, Austria) and Tiletamin (0.33 mg/kg) combined with Zolazepam (0.33 mg/kg; Virbac, Glattburg, Switzerland).

Anesthesia was maintained with midazolam (0.8 mg/kg/h; Nycomed, Linz, Austria), sufentanil (80 μg/kg/h; Janssen, Vienna, Austria) and rocuronium (1 mg/kg/h; Organon, Oss, the Netherlands). Catheters were placed by direct incision in the right femoral artery for continuous arterial pressure monitoring and the right femoral vein for fluid therapy. For measurement of cardiac output, a 7.5-Fr Swan-Ganz flow-directed thermodilution tip catheter (93A-431H7.5; Baxter Healthcare, Irvine, CA) was positioned in the proximal pulmonary artery via the right jugular vein. All catheters were flushed with heparinized saline (8 U/mL) to prevent clotting. A 14-Fr silastic catheter was inserted suprapubically into the bladder.

Experiment Protocol

After induction of anesthesia and placement of an arterial and venous line, baseline hemodynamic status was measured and blood samples were collected for coagulation parameters (baseline). After 1 h, a second blood sample was drawn and the animals received an increasing infusion of endotoxin starting at 0.03 μg/kg/min rising up to 0.33 μg/kg/min, accounting for a total dose of 10 μg/kg i.v. endotoxin (*Escherichia coli* LPS O26:B6; Difco Laboratories, Detroit, MI, USA) over a period of 60 min. If mean pulmonary artery pressure exceeded 40 mmHg, the LPS infusion was stopped and restarted again after mean pulmonary artery pressure decreased below this level. The dosing regime was chosen to avoid immediate pulmonary hypertension induced by experimental endotoxinemia, as previously described (12). The animals were followed thereafter for 5 h and then were euthanized with an intravenous bolus injection of 100 mg/kg pentobarbital followed by 10 mL potassium chloride (KCl).

Ringer solution was administered at a rate of 7 mL/kg/h i.v. to achieve adequate fluid resuscitation. The infusion rate was increased to 10 mL/kg/h, if mean arterial pressure decreased below 70 mmHg and to 15 mL/kg/h if mean arterial pressure decreased below 50 mmHg. If pulmonary artery occlusion pressure exceeded 10 mmHg, the rate of infusion was lowered to 7 mL/kg/h. Body temperature was kept constant around 38–39°C by using a heating blanket. Mean arterial pressure, mean pulmonary artery pressure and heart rate were monitored continuously.

Measurements, Blood Sampling and Analytical Methods

Thromboelastometry parameters (TEM; TEM International, Munich, Germany), PT (%), international normalized ratio (INR), aPTT, fibrinogen, AT, protein C, t-PA, PAI-1, blood cell count (hematocrit, hemoglobin concentration, white blood cell count, platelet count), blood gas analysis (pH, base deficit) and hemodynamic status (heart rate, mean arterial pressure, mean pulmonary artery pressure, pulmonary artery occlusion pressure, cardiac output) were measured at baseline; immediately before LPS infusion; at the end of LPS infusion; and 2, 3, 4 and 5 h after the start of endotoxin infusion.

Coagulation analyses were performed in blood collected in 3-mL tubes containing 0.3 mL buffered 3.2% trisodium citrate, giving a volume ratio of 1:10 (Vacuette; Greiner Bio-One, Linz, Austria). Furthermore, PT (%), INR (Thromborel S; Siemens, Marburg, Germany), aPTT (Dade Actin FS; Siemens), fibrinogen concentration (according to Clauss [Dade Thrombin Reagent; Siemens]), AT (Berichrom Antithrombin III; Siemens), Protein C (Berichrom Protein C; Siemens) and D-dimer (Innovance D-Dimer; Siemens) were analyzed. The tests were run on the Sysmex CA 1500 (Siemens).

Measurement of hematocrit, hemoglobin concentration, white blood cell count and platelet count was performed in blood collected in 3-mL K_3EDTA tubes containing 1.6 mg/mL ethylenediaminetetraacetic acid (EDTA; Vacuette). Cell counts were measured with a CELL-DYN 3700 instrument (Abbott, Vienna, Austria).

EDTA blood and citrate blood was collected and centrifuged immediately at 2,000*g* for 10 min to obtain plasma. Plasma samples were stored at –80°C until analysis of t-PA activity (enzyme-linked immunosorbent assay [ELISA], Technoclone TC16000), PAI-1 (Porcine PAI-1 Activity Assay; Dunn Labortechnik, Aspach, Germany) and TAT (Enzygnost TAT micro ELISA, Behring, Germany). Normal values for pigs have been published previously (13).

Rotational Thromboelastometry (ROTEM®)

TEM was recorded in parallel to coagulation analysis. Details about the function of the analyzer and the assays routinely used are published elsewhere (10).

In the present study, blood samples were recalcified by the addition of 20 μL start-TEM® ($CaCl_2$, 200 mmol/L) but not activated (NATEM). This approach increases the sensitivity of the test to endogenous tissue factor. The final volume in the TEM cup was 340 μL in all cases.

The FIBTEM assay is an extrinsically activated test (EXTEM®) with the addition of cytochalasin D. Cytochalasin D inhibits platelet function by inactivation of the cytoskeleton of platelets. This test provides information on the fibrin component of the clot separately. The FIBTEM assay contains 20 μL $CaCl_2$ (0.2 mol/L), cytochalasin D, 20 μL tissue factor and 300 mL whole blood.

The following TEM variables were measured. Clotting time (CT [s]) was determined by the time from the start of the measurement until formation of a clot 2 mm in amplitude. Clot formation time (CFT [s]) was the time from the end of the CT (amplitude of 2 mm) until a clot firmness of 20 mm was achieved. The α angle (α) was the angle between the central line (x axis) and the tangent of the TEM tracing at the amplitude point of 2 mm describing the kinetics of clot formation. The maximum clot firmness (MCF [mm]) reflects the final strength of the clot and results from the interaction of fibrin, activated platelets and factor XIII (FXIII). The lysis index at 60 min (LI 60 [%]) is defined as the percentage of remaining clot stability in relation to the MCF after the 60-min observation period after CT and indicates the speed of fibrinolysis. Maximum lysis (ML [%]) represents the reduction of the MCF in percentages during the analysis.

All reagents were purchased from TEM International. The TEM device was checked for proper functioning according to the manufacturer's recommendation by using a control serum (ROTROL N).

Statistical Analysis

Data were expressed as the mean and standard deviation. Normal distribution of the data was confirmed using the Kolmogorov-Smirnov test. One-way repeated measures analysis of variance was used to detect differences between time points. In case of significant differences between time points, a Newman-Keuls *post hoc* correction for multiple comparisons was applied. A two-tailed P value of <0.05 was considered significant. All statistical calculations were performed using commercially available statistical software (GraphPad Prism 5.03).

RESULTS

All animals survived to the end of the study. All TEM measurements were performed by the same investigator (HS). Baseline coagulation parameters, TEM test results and blood cell counts are summarized in Table 1.

Coagulation Tests

Endotoxinemia resulted in a significant shortening of the CT 2 h after the end of LPS infusion ($P = 0.037$ compared with baseline). After this time point, CT became increasingly prolonged and exceeded the preoperative value. At the end of the experiment, CT was comparable to baseline (P = not significant) and was significantly longer compared with 2 h after the start of LPS infusion ($P < 0.001$; Figure 1). CFT increased significantly from baseline to the end of the experiment ($P = 0.05$), whereas α angle decreased over the same period ($P = 0.0001$).

Unlike CT, standard coagulation times became increasingly prolonged from baseline until the end of the 5-h observation time (Table 1). During the same examination period, TAT increased significantly from baseline until 3 h after the end of LPS infusion ($P = 0.0002$). D-dimers nearly doubled from baseline to the end of the experiment, with a maximum concentration 3 h after the start of LPS infusion ($P = 0.0001$). The major inhibitors of the coagulation system, AT and protein C, decreased significantly from baseline values to the end of the study ($P < 0.01$ and $P < 0.0001$, respectively; Table 1).

Effects of LPS Infusion on Maximum Clot Firmness and Fibrinolysis

The NATEM-MCF decreased significantly over the whole observation period ($P = 0.01$). The same was observed for the FIBTEM-MCF, which decreased significantly ($P = 0.01$) from baseline to the end of the experiment (Figure 2). As expected from the FIBTEM measurements, fibrinogen concentration also decreased significantly ($P = 0.0075$) until the end of the observation period (Table 1).

Surprisingly, ML decreased constantly throughout the experiment. Two h after beginning LPS infusion and at all following time points, ML was significantly lower than at baseline (Figure 3). This evolution was mirrored by a constant increase in LI 60 from baseline to the 5-h observation time (Table 1).

An abrupt but transient increase in t-PA was observed at the end of LPS infusion ($P = 0.001$ compared with baseline). t-PA values returned to baseline levels 3 h after induction of endotoxinemia (Figure 4). Furthermore, 3 h after the start of LPS infusion, PAI-1 concentration began to increase abruptly, and the maximum concentration was reached at the end of the observation time ($P = 0.0001$; Figure 5).

Blood Cell Count

LPS infusion resulted in a significant decrease in white blood cell count with a nadir 3 h after induction of endotoxinemia ($P = 0.001$; data not shown). Platelet count also decreased significantly until the end of the observation period ($P = 0.005$; Table 1). Hemoglobin and hematocrit also decreased significantly from baseline until the end of the study ($P = 0.01$; data not shown).

DISCUSSION

In this porcine model, LPS infusion resulted in significant changes of whole blood viscoelastic variables. This study revealed that, in contrast to standard coagulation tests, thromboelastometry is able to detect the early endotoxin-related activation of coagulation and the consecutive consumption of coagulation factors and platelets.

An accurate diagnosis of DIC is complex; there is no single diagnostic test offering the capacity to confirm or reject the diagnosis of DIC (14). The International Society on Thrombosis and Haemostasis established a scoring system to diagnose DIC based on prothrombin time, platelet count, fibrinogen concentration and fibrin-related markers

Table 1. Coagulation test results.

	Baseline	Start of LPS infusion	End of LPS infusion	2 h after end of LPS infusion	3 h after end of LPS infusion	4 h after end of LPS infusion	5 h after end of LPS infusion
Standard coagulation analyses							
INR	0.9 ± 0.1[b,c]	0.9 ± 0.1[c,d]	1 ± 0.1[e]	1.1 ± 0.1[e]	1.2 ± 0.2[b,c,e]	1.2 ± 0.2[b,c,e]	1.2 ± 0.2[b,c,e]
Prothrombin time (%)	124 ± 13[b,c]	114 ± 10[b,c,f]	98 ± 20[e]	94 ± 19[e]	78 ± 16[b,c,e]	74 ± 13[b,c,e]	75 ± 15[b,c,e]
aPTT (s)	13.3 ± 0.8[b,c]	14.0 ± 0.6[f]	14.5 ± 1[e]	14.4 ± 0.9[e]	15.2 ± 1.4[d,e,g]	15.7 ± 1.2[b,c,e]	15.9 ± 1.2[b,c,e]
Fibrinogen (g/L)	1.5 ± 0.2	1.5 ± 0.2	1.4 ± 0.2	1.3 ± 0.2[e]	1.3 ± 0.3[e]	1.2 ± 0.3[b,e]	1.3 ± 0.3[e]
Platelet count (1,000/μL[f])	436 ± 115[b,c]	377 ± 83[b,e]	293 ± 73[e]	267 ± 96[e]	232 ± 101[d,e]	222 ± 84[b,e]	249 ± 78[e]
Extended coagulation analyses							
TAT (μg/L)	31.4 ± 15.1[c,d]	15.7 ± 6.3[c,d,f]	52.8 ± 21.3	89.7 ± 23.7[d,e]	149.4 ± 67.3[b,c,e]	137.6 ± 59.2[b,c,e]	107.2 ± 35.8[b,e]
D-dimers (μg/L)	0.3 ± 0.1[c]	0.3 ± 0.2[e]	0.5 ± 0.1	0.9 ± 0.4[b,e]	1.4 ± 0.3[b,c,e]	1.2 ± 0.2[b,c,e]	0.9 ± 0.2[b,e]
AT III (%)	128 ± 9[b]	121 ± 9	120 ± 12	115 ± 18[e]	111 ± 20[e]	106 ± 22[b,e,g]	102 ± 18[b,c,e]
Protein C	88 ± 25	94 ± 26	89 ± 25	87 ± 25	81 ± 25	75 ± 24[b,c,e]	68 ± 22[b,c,e]
t-PA (U/mL)	0.039 ± 0.03[b,c]	0.04 ± 0.02[b,c]	0.808 ± 0.44[c,e]	0.398 ± 0.37[b,e]	0.035 ± 0.02[b,c]	0.030 ± 0.02[b,c]	0.032 ± 0.02[b,c]
PAI-1 (ng/mL)	7.3 ± 1.7	7.3 ± 2.2	4.9 ± 9.1	6 ± 11.3	196.2 ± 112.4[b,c,e]	424.8 ± 91.4[b,c,e]	405.3 ± 119.5[b,c,e]
TEM parameters							
NATEM-CT (s)	525 ± 100[g]	513 ± 68	513 ± 75	429 ± 66[f]	457 ± 104	567 ± 116[c]	571 ± 50[c]
NATEM-CFT (s)	137 ± 49	120 ± 23[c]	153 ± 49	116 ± 23	168 ± 80	196 ± 72[c,f]	202 ± 44[c,f]
NATEM-α (°)	66 ± 8	68 ± 4	62 ± 8	68 ± 4	61 ± 11	56 ± 11[c,e]	55 ± 6[c,e]
NATEM-MCF (mm)	65 ± 8[b]	64 ± 6[b]	55 ± 7[e]	59 ± 4	56 ± 6[e]	53 ± 9[e,g]	53 ± 5[e,g]
NATEM-ML (%)	14.7 ± 3.6[c]	14.2 ± 3.7	13.2 ± 4.7[c]	9.8 ± 4.2[b,e]	8.2 ± 4.2[b,e]	7.7 ± 5[b,e]	8.4 ± 5.2[b,e]
NATEM-LI (%)	89.3 ± 3.4[g]	89.7 ± 3.7[g]	90.5 ± 4.6	92.1 ± 3.8[f]	94 ± 3.1[b,e,g]	95.1 ± 3.6[b,c,e]	95.1 ± 3.2[b,c,e]
FIBTEM-MCF (mm)	35 ± 12[b,c]	31 ± 13[g]	24 ± 13[e]	25 ± 13[e]	25 ± 12[f]	23 ± 12[b,e]	22 ± 12[e]

[a]Data are presented as means ± standard deviation.
[b]$P < 0.01$ compared with end of LPS infusion.
[c]$P < 0.01$ compared with 2 h after end of LPS infusion.
[d]$P < 0.05$ compared with end of LPS infusion.
[e]$P < 0.01$ compared with baseline.
[f]$P < 0.05$ compared with baseline.
[g]$P < 0.05$ compared with 2 h after end of LPS infusion.

(D-dimers, fibrin degradation products) (15). This scoring system has been proven to be helpful in the accurate diagnosis of DIC (5).

Sepsis-induced coagulopathy is in part the result of tissue factor expression on monocytes. Using standard coagulation tests, these cells are removed by the centrifugation process. A cell-based coagulation monitoring system, like thromboelastometry, could therefore be an interesting alternative to routine coagulation

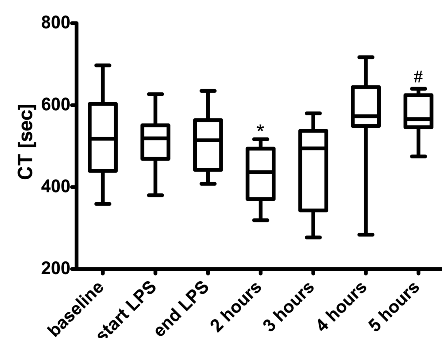

Figure 1. CT in NATEM: $*P < 0.05$ versus baseline; $^{\#}P < 0.01$ versus 2 h.

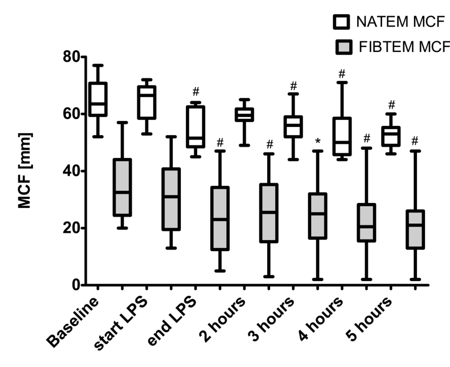

Figure 2. NATEM MCF: $*P < 0.05$; $^{\#}P < 0.01$ versus baseline. FIBTEM MCF: $^{\#}P < 0.01$ versus baseline.

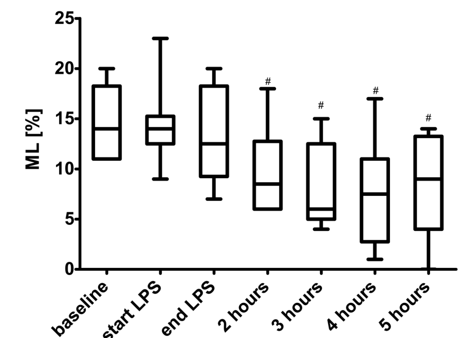

Figure 3. ML in NATEM: $^{\#}P < 0.01$ versus baseline.

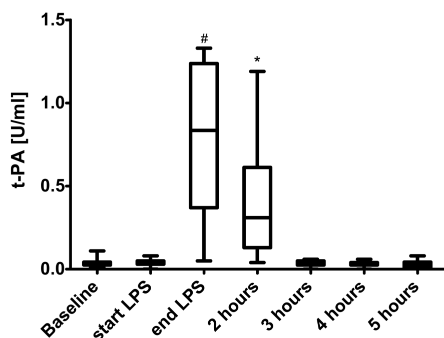

Figure 4. t-PA: *$P < 0.01$; #$P < 0.0001$ versus baseline.

tests for an early detection of DIC-induced activation of coagulation (16). Furthermore, thromboelastometry (TEM) and thrombelastography (TEG®) represent the only bedside methods, which also provide information about the changes in the fibrinolytic system (17,18). However, reports on the use of TEM/TEG as a point-of-care monitoring device in sepsis and DIC are rare (16,19,20,21). Collins *et al.* (16) observed that compared with normal controls, critically ill patients with sepsis syndrome had a prolonged CT, increased α angle and increased MCF. Sivula *et al.* (19) reported that septic patients with overt DIC had lower MCF, prolonged CFT and decreased α angle, indicating a hypocoagulable state. Conversely, Daudel *et al.* (20) found that variables of TEM remained within the reference in patients with severe sepsis and septic shock. The conflicting results were most likely caused by a variable time course of sepsis in patients. In contrast, our data indicating an early hypercoagulable state after onset of endotoxinemia were obtained under controlled and standardized conditions. Recently, Adamzik *et al.* (21) reported in an observational cohort study that, in critically ill adults, thromboelastometry lysis index was a more reliable biomarker of severe sepsis than procalcitonin, interleukin 6 and C-reactive protein.

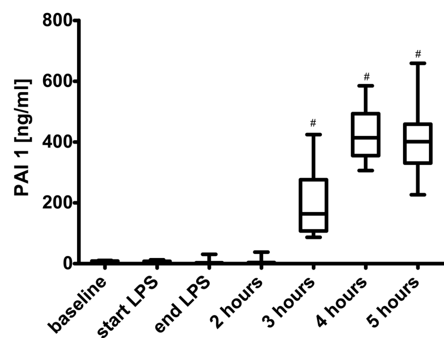

Figure 5. PAI-1: #$P < 0.01$ versus baseline.

In the present study, clotting time shortened immediately after endotoxin infusion, suggesting an early activation of the coagulation system. A nadir was reached 2 h after the start of the LPS infusion, followed by a significant increase in CT until the end of the experiment. Furthermore, TAT complex levels and D-dimer levels increased significantly after LPS infusion, indicating a massive thrombin activation and fibrin formation. In contrast, during the same examination period, standard coagulation tests such as INR and aPTT prolonged steadily until the end of the experiment. Thus, increased LPS-induced activation of the coagulation process was not mirrored at the early stage by the routine coagulation test results. A similar CT reduction in an *in vitro* model was reported by Zacharowski *et al.* (22), who incubated whole blood with different concentrations of LPS (0.005–1 mg/mL) for 4 h. Increasing amounts of LPS resulted in a substantial shortening in CT, and the half-maximal effective concentration (ED50) of LPS effect on CT was recorded at 18 μg/mL (22). The same study also showed that *ex vivo* addition of 10 ng/mL LPS to platelet-poor plasma did not change CT. However, a reduction in CT was noted in platelet-poor plasma obtained after whole blood stimulation with LPS. These findings suggest that the decrease in CT is the result of a cellular/humoral-based mechanism, most likely, by cell-derived soluble factor(s), which leads to further DIC-like changes *in vivo*. In our *in vivo* study, this result was reflected in acceleration of coagulation, consumption of platelets and fibrinogen and increase of the TAT complexes as a sign of increased thrombin generation.

A significant, transient shortening of CT was also observed by Spiel *et al.* (23) after low-dose LPS infusion in human volunteers, suggesting hypercoagulability. In burn patients, Park *et al.* (24) reported that only thrombelastography analysis of whole blood revealed that patients were in a hypercoagulable state. Like in our experiment, hypercoagulability was not detectable by plasma-based clotting assays (PT and aPTT). In the present study, AT and protein C, both important inhibitors of the coagulation system, decreased only slightly over the same period of time. Therefore, the prolongation of the CT to the end of the study period seems to be a consequence of a consumptive process and not a result of a depletion of inhibitors.

The propagation phase of clot formation is reflected by the α angle and the CFT. The α angle and CFT characterize thrombin generation and early fibrin formation. The α angle showed a significant and continuous reduction, whereas the CFT demonstrated a continuous increase over the whole study period. As fibrinogen concentration and platelets also decreased over the whole observation phase, the increase in CFT and the decrease of the α angle could also be assumed to be a consumptive process. This observation is in contrast to the report of Spiel *et al.* (23), who did not observe any changes in CFT. This difference in findings might be explained by a much higher dose of LPS infused in our porcine model compared with low-dose endotoxin used in the human volunteer study. However, the high dose used in the present study reflects more accurately the situation occurring during sepsis. Zacharowski *et al.* (22) also reported that CFT was unaffected by *in vitro* incubation of blood samples with LPS. They used higher concentrations of LPS and incubated the blood samples *ex vivo*, thus, it did not result in a consumptive state.

MCF in NATEM and FIBTEM-MCF reduced constantly until the end of the experiment. This result can be interpreted

as a consequence of DIC-associated consumption of platelets and fibrinogen.

In experimental endotoxinemia, a short-acting increase in fibrinolytic activity due to the release of t-PA from the endothelial cells was reported (9,25). In the present study, this transient and highly significant increase in t-PA activity was also observed. Surprisingly enough, this was not accompanied by an increase in ML. In contrast, ML decreased significantly from 2 h after LPS infusion until the end of the experiment, whereas LI 60 increased. This finding does not support the results published by Spiel *et al.* (23), who reported an increase of ML in parallel with the t-PA peak. This result could be because of a significant increase of PAI-1 3 h after LPS infusion, since PAI-1 is the major antagonist of t-PA resulting in a complete inhibition of the fibrinolytic system (1).

The results of the present study suggest that TEM may support an earlier diagnosis of DIC-related coagulopathy than standard laboratory tests. In the majority of published human studies, TEM/TEG was used as a coagulation monitoring device at a later, more advanced phase of sepsis. No clear benefit could be identified from the use of TEM at this advanced stage. One explanation for the lack of benefit may be that these studies investigated TEM/TEG at a time point when standard laboratory parameters also reflected impairment of coagulation. In contrast, our findings suggest that TEM may specifically help the detection of LPS (sepsis)-induced changes of the coagulation system when performed at an early stage of sepsis. If this proves correct, TEM could be a valuable instrument for early detection of DIC in conditions with fulminant evolution (for example, meningococcal sepsis).

Several limitations of this study require discussion. The extent to which results obtained in our experiments on swine are transferrable to humans is unclear. No attempt was made to correlate a calculated International Society on Thrombosis and Haemostasis DIC score in swine with the TEM parameters. In fact, the calculation of this score in swine is not recommended, since the cutoff values used in humans for the standard laboratory tests do not apply for swine. However, the identification of a coagulopathic profile using TEM analysis at such an early stage in sepsis could potentially provide an early alarm signal. Further clinical studies to evaluate and compare the coagulation profiles provided by TEM and standard laboratory tests at early stages of sepsis are warranted to confirm this hypothesis. In addition to the limitations of this study, it is unclear whether this porcine model reflects the human response to LPS with respect to the activation of the coagulation process and fibrinolysis. t-PA peak combined with the lack of a corresponding increase in ML was different to that seen in human volunteers (23), but this may also be due to variation in LPS concentration.

Conclusions

In this model of endotoxin-induced DIC, CT shortened immediately after LPS infusion—a finding not reflected by routine coagulation tests, including INR or aPTT. The decrease in maximum clot firmness in NATEM and FIBTEM further suggested consumption of a coagulation substrate. ML decreased over the whole observation period, which could be a result of increased concentrations of PAI-1. Interestingly, the substantial increase in t-PA at the end of LPS infusion had no effect on ML. All these changes in coagulation were identified by a single test method. From this point of view, TEM is an interesting monitoring alternative in the detection of LPS-induced changes of the coagulation system. However, further investigation is necessary to establish the clinical relevance of these findings and the importance of thromboelastometry for describing the coagulation disturbances observed in DIC.

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H Schöchl contributed to the design of the study, analysis of the results and interpretation of the data and drafted the manuscript. C Solomon, A Schulz, W Voelckel, A Hanke and M van Griensven were involved in drafting the manuscript. H Redl and S Bahrami contributed to the design of the study, drafting and critical revision of the manuscript. All authors read and approved the final manuscript.

DISCLOSURE

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