Effects of hypothermia on thrombelastography in patients undergoing cardiopulmonary bypass


Summary
Thrombelastography (TEG) correlates with postoperative chest drain output in patients undergoing cardiopulmonary bypass (CPB). In vitro incubation with heparinase allows TEG monitoring during CPB, despite heparin anticoagulation. Hypothermia impairs coagulation, but these effects cannot be assessed by standard coagulation tests performed at 37 °C. The aim of this study was to assess the effects of hypothermia on TEG. Therefore, we have compared normothermic and temperature-adapted TEG in 30 patients undergoing CPB. Our data showed significantly impaired reaction time (r), kinetic time (k), and angle α (α) in temperature-adapted compared with normothermic TEG. Maximum amplitude (MA), reflecting absolute clot strength, was not affected at temperatures of 33–37 °C. These findings indicate a decrease in the speed of clot formation, but not absolute deterioration in clot quality. Furthermore, heparinase-modified TEG indicated that there were nine cases in which hypothermic effects persisted after heparin reversal with protamine, providing a rational guide to protamine therapy. (Br. J. Anaesth. 1998; 80: 313–317)

Keywords: measurement techniques, thrombelastography; heart, cardiopulmonary bypass; surgery, cardiovascular; hypothermia; blood, anticoagulants, heparin

Severe bleeding is a major complication of cardiopulmonary bypass (CPB) for open heart surgery. Various causes are known and have been investigated, for example platelet dysfunction, haemodilution, contact activation by the artificial surface of the CPB, heparin excess and inadequate heparin reversal.1 Hypothermia induced during CPB to reduce neuronal damage may also impair coagulation. Patients undergoing hypothermic CPB show a tendency towards increased intraoperative and postoperative blood loss and an increased rate of re-operation for bleeding.2–4

However, the influence of hypothermia on the haemostatic system cannot be assessed by standard coagulation tests, as these tests are performed at 37 °C. Thrombelastography (TEG) is an easy and reliable coagulation monitoring system which can be performed at different temperatures.5 In patients undergoing CPB, TEG performed at 37 °C correlates with postoperative chest tube drainage and predicts postoperative bleeding better than activated clotting time (ACT), routine coagulation screen tests or Sonoclot measurements.6,7

To prevent clotting in the extracorporeal oxygenator, unfractionated heparin is given before CPB, making it impossible to perform TEG with native whole blood. In vitro incubation with heparin-utilizing substances allows TEG monitoring during bypass, despite anticoagulation with heparin.8

Protamine is used for antagonism of heparin. In the absence of heparin, protamine per se may affect coagulation tests.9 Thus the heparin-to-protamine ratio has to be adequate to avoid either heparin or protamine excess leading to prolongation of coagulation tests. Heparinase, an enzyme isolated from Flavobacterium heparinum, uses heparin without affecting coagulation tests in the absence of heparin.10 For TEG, heparinase neutralizes high doses of heparin, enabling TEG monitoring during CPB.9 The use of heparinase-modified blood, temperature-adapted TEG permits assessment of the effects of hypothermia on TEG during CPB.

Only one study has investigated the influence of hypothermia on TEG monitoring in patients undergoing liver transplantation.11 The aetiology of coagulopathy is different during liver transplantation and open heart surgery with the use of CPB. During CPB, coagulation is affected by haemodilution and contact activation, and blood is anticoagulated to prevent clotting in the extracorporeal oxygenator. The aetiologies of hypothermia are also different. Whereas it is therapeutically indicated during CPB, hypothermia occurs accidentally during liver transplantation and can be avoided by surface warming.12

We have assessed the effects of hypothermia on TEG monitoring in patients undergoing CPB. We compared TEG performed at 37 °C and at body core temperature before, during and after CPB. To allow TEG during anticoagulation with heparin, we performed native and heparinase-modified measurements. Moreover, comparison of native and heparinase-modified TEG after protamine adminis-

S. C. KETTNER*, MD, S. A. KOZEK, MD, J. P. GROETZNER, MD, C. GONANO, MD, A. SCHELLONGOWSKI, MD, ZIMPFER, MD, Department of Anaesthesiology and General Intensive Care, University of Vienna, Austria and Ludwig Boltzmann Institute of Clinical Anaesthesiology and Intensive Care, Vienna, Austria. M. KUCERA, MRC, Department of Computer Science, Technical University of Vienna, Treitsstr. 3/3/182, A-1040 Vienna, Austria. Accepted for publication: October 13, 1997.

*Address for correspondence: Department of Anaesthesiology and General Intensive Care, General Hospital Vienna, 18–20 Währinger Gürtel, A-1090 Vienna, Austria.
tration may indicate the incidence of inadequate heparin reversal after CPB.

**Patients and methods**

After obtaining local Ethics Committee approval and informed consent, we studied 30 consecutive patients. Inclusion criteria were age > 18 yr and elective primary coronary revascularisation with the use of CPB under mild hypothermia. Exclusion criteria were the use of platelet altering medications within 1 week of surgery, pre-existing bleeding disorders or diabetes with severe peripheral vascular complications.

The extracorporeal circuit consisted of a membrane oxygenator with polyvinyl chloride tubing, non-occlusive roller pumps and a cardiotomy reservoir (William Harvey, USA). The oxygenator was primed with crystalloid solution 2000 ml and 20% mannitol 100 ml, containing unfractionated heparin 8000 u. and aprotinin 1 × 10^4 ku. (Trasylol, Bayer, Austria). CPB was performed with a flow rate of 2.5 litre min\(^{-1}\) and moderate hypothermia. Blood was cooled to 32 °C during the hypothermic phase of CPB and warmed to 37 °C during rewarming; the rate of cooling and warming was controlled to approximately 1 °C min\(^{-1}\). We monitored blood temperature using a pulmonary artery flotation catheter, oesophageal temperature and bladder temperature. As oesophageal temperature may be influenced by cold cardioplegia solution\(^3\) and as we wished to avoid overestimation of hypothermia, we considered bladder temperature as body core temperature. We did not use the temperature recorded with the pulmonary artery flotation catheter because of reduced lung perfusion during CPB.

Patients were given unfractionated heparin 300 u. kg\(^{-1}\) (Heparin Immuno, Immuno, Austria) before aortic cannulation to achieve an activated clotting time (ACT) of >450 s (Hemochron 800, Intern. Technidine Corp., USA). If ACT was < 450 s after administration of this initial dose, an additional bolus dose of heparin 100 u. kg\(^{-1}\) was given. ACT was monitored every 30 min until the end of CPB. If ACT was less than 400 s during CPB, additional boluses of heparin 100 u. kg\(^{-1}\) were given. The initial heparin dose necessary to achieve an ACT > 450 s was neutralized after the end of CPB by protamine (Protamin Roche, Hoffmann-La Roche, Switzerland) in a ratio of 1:1.

Blood samples were obtained from an arterial cannula (or from the port of the oxygenator during CPB) before skin incision; after heparin administration; 10 min after the start of CPB; at the lowest body core temperature during CPB; immediately before protamine administration; and at the end of surgery. Samples were collected in silicon-coated glass tubes containing buffered sodium citrate 0.129 mol litre\(^{-1}\) (Vacutainer, Becton Dickinson, France).

We performed four TEG measurements simultaneously on each blood sample at 37 °C and at core temperature, using native and heparinase-modified blood. Temperature-adapted measurements were not performed when body temperature was 36.5 °C or higher. TEG was performed with 300 μl of citrated whole blood and 60 μl of 0.645% CaCl\(_2\). For heparinase-modified measurements, blood was incubated in heparinase vials (Hemoscope, USA) containing heparinase 4 u./ml of blood. Four commercially available temperature-adaptable Thrombelastographs D (Hemoscope, USA) were used for this investigation.

TEG reaction time (r) is the time from the start of measurement until initial fibrin formation. Clot formation time (k) measures the time necessary to reach 20 mm of clot strength. Coagulation time (r + k) is the sum of r and k (fig. 1). Values for r, k and r + k are expressed in millimetres (mm), as the chart speed is 2 mm min\(^{-1}\) and the time in minutes is equal to the distance in mm divided by 2 (normal ranges: r: 10–19 mm; k: 4–10 mm). The angle α measures the rapidity of fibrin build-up and cross-linking, which is the speed of clot strengthening (normal range 44–56°).

Maximum amplitude (MA) measures maximum clot strength which is dependent on platelet function and, to a lesser extent, on fibrinogen level (normal range 50–64 mm) (fig. 1).\(^5\)

The accuracy of temperature measurement in the TEG was verified by measuring the temperature of the thrombelastograph cuvettes filled with normal saline before the investigation. The accuracy of temperature measurement between 28 °C and 37 °C was within ±0.2 °C. Measurements were performed with prewarmed (28 °C) disposable plastic pins and cups (Haemoscope, USA) which were inserted at least 10 min before measurement to confirm the exact temperature.

**Statistical analysis**

After testing for normal distribution of the data, we performed ANOVA for repeated measurements to test the effects on TEG variables. The Newman–Keuls test was used to test for differences between normothermic and temperature-adapted TEG. To compare native and heparinase-modified measurements at the end of surgery, a t test for dependent samples was used. Statistical significance was considered at \(P < 0.05\). All values are expressed as mean (SD).

**Results**

Thirty patients (24 males, six females; mean age 68.1 yr (range 40–85 yr); mean height 171.5 (SD 10.8) cm; mean weight 75.4 (12.8) kg) underwent surgery for 3.5 (0.7) aortocoronary bypass procedures. Extracorporeal circulation time was 94.6 (17.6) min, with an aortic clamping time of 52.7 (12.5) min. To achieve adequate anticoagulation, heparin 29 056 (11 880) u. were given. To reverse heparin after CPB,
Temperature-adapted thrombelastography during CPB

We have demonstrated that the heparinase-modified TEG showed significantly impaired \( r \), \( k \) and \( \alpha \) values in temperature-adapted measurements compared with normothermic values.

At the time of skin incision, before administration of heparin, native and heparinase-modified measurements did not differ. After administration of heparin, native TEG measurements resulted in straight lines after 60 min in all cases. At the end of surgery, after heparin reversal with protamine, native and heparinase-modified TEG were different (table 2). In nine cases heparinase-modified measurements were improved compared with native measurements (table 2). Coagulation time was the best indicator for remaining heparin effects (fig. 7).

**Discussion**

We have demonstrated that the heparinase-modified TEG showed significantly impaired \( r \), \( k \) and \( \alpha \) values in temperature-adapted measurements compared with normothermic values.
with normothermic measurements during CPB. Similar results have been shown in hypothermic patients during liver transplantation. Reaction time \((r)\) represents the time necessary for building the first fibrin strands, \(k\) the time for reaching a certain clot strength and \(\alpha\) represents the formation rate of the clot. Reaction time is dependent mainly on coagulation factor activity, whereas \(k\) and \(\alpha\) depend on plasma coagulation and the interaction of platelets with fibrin, increasing clot stability. Impairment of these variables by temperature reduction indicates a reduction in activity of both coagulation factors and platelet function. In patients undergoing CPB, \(r\), \(k\) and \(\alpha\) are affected by a temperature reduction of 3.7 °C leading to an approximate 50% reduction in coagulation, as assessed by TEG. Performing heparinase-modified TEG at 37 °C in hypothermic patients may lead to underestimation of coagulopathy during CPB when hypothermia is not considered.

However, temperature-adapted TEG could lead to inappropriate therapeutic interventions if prolongation in TEG is caused by hypothermia and treated with administration of coagulation factor. Therefore, it seems appropriate to treat patients with coagulation factors or platelets only when results of normothermic TEG are abnormal. In the treatment of hypothermic patients with bleeding disorders, comparison of temperature-adapted and normothermic TEG can help to guide therapeutic interventions, for example administration of coagulation factor, transfusion of platelets, active warming or surgical re-exploration. As hypothermia impairs coagulation, it should be treated. The literature shows a decrease in blood loss in patients undergoing normothermic CPB compared with those undergoing hypothermic CPB, without an increase in adverse systemic effects.

Maximum amplitude (MA) reflects absolute clot strength and not coagulation time, unlike \(r\) and \(k\). Temperatures between 33 °C and 37 °C do not affect MA. This finding, together with prolonged \(r\), \(k\) and a smaller \(\alpha\) value, indicates a decrease in the speed of clot formation, but not absolute deterioration in clot quality, which is represented by MA. In the course of surgery MA decreased for normothermic and temperature-adapted measurements in the same manner. This indicates that TEG is able to detect impairment of platelet function caused by surgery and the extracorporeal circulation.

Two investigations showed prolongation in plasma coagulation tests caused by in vitro reduction of the assay temperature. Thrombin time (TT) was less prolonged than prothrombin time (PT) and activated partial thromboplastin time (APTT). It was concluded that there may be a cumulative effect in multi-step assays, such as PT or APTT, leading to greater prolongation than in one-step assays, such as TT. Prolongation of approximately 50–60% in PT and APTT was found at 29 °C or 28 °C, respectively. Our data showed 50–60% prolongation of TEG variables at 33.3 °C. TEG is a global assessment of haemostatic function, documenting the interaction of platelets with the protein coagulation cascade from the time of initial fibrin formation through platelet aggregation, clot strengthening and fibrin cross linkage to eventual clot lysis. It is a multi-step assay of many more stages than plasma coagulation tests such as PT or APTT. The high sensitivity of TEG to temperature may be caused by cumulative slowing of the enzymatic steps of coagulation, as assessed by TEG. Alternatively, either interaction of platelets with coagulation proteins or platelet function per se, might be especially sensitive to the effects of hypothermia. Hypothermia-induced reversible platelet dysfunction was shown in an animal study by Valeri and colleagues. In patients undergoing CPB, hypothermia reduced intraoperative platelet function and platelet recovery in the post-bypass period. In that study, aprotinin reduced the negative effects of hypothermic CPB on platelet function. Although our patients received aprotinin in a much lower dose, impairment of TEG variables by temperature may be even higher when no aprotinin is given. Furthermore, aprotinin reduces clot lysis, as assessed by TEG. In fact, we found no differences in TEG clot lysis in normothermic compared with temperature-adapted measurements in our study. Only further study of the enzymatic activity of coagulation proteins and platelet activity under conditions of hypothermia will determine the extent to which hypothermia impairs platelet function.
which different mechanisms contribute to hypothermia-induced coagulopathy.

Data in figures 3–6 are from heparinase-modified measurements. The heparinase-modified measurements, whether temperature-adapted or not, were not different compared with the corresponding native measurements before heparin administration. This shows that heparinase per se does not affect TEG and correlates with the findings of other investigations.8,17

The heparinase-modified TEG provides useful additional information compared with native TEG. At the end of surgery, after heparin reversal with protamine, nine cases of heparinase-modified TEG were improved compared with native TEG (fig. 7, table 2). These TEG measurements showed remaining heparin effects, which can increase bleeding after CPB. Comparison of native with heparinase-modified TEG is a useful guide when reconsidering protamine therapy after CPB.

In summary, our data showed reductions in coagulation caused by hypothermia, indicating the high sensitivity of TEG to temperature. Heparinase-modified TEG detected heparin effects remaining after heparin reversal with protamine. This could provide a rational guide to protamine therapy after CPB.

References