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Prothrombin complex concentrate mitigates diffuse bleeding after cardiopulmonary bypass in a porcine model

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Key points

- Patients undergoing cardiopulmonary bypass (CPB) may develop dilutional coagulopathy.
- The value of prothrombin complex concentrate in reducing suture hole bleeding from the carotid artery was assessed in pigs after CPB.
- The prothrombin complex concentrate reduced bleeding.
- This agent may be useful in the clinical setting.

Background. Extracorporeal circuit priming and intravascular volume expansion during cardiopulmonary bypass (CPB) may lead to dilutional coagulopathy and excessive diffuse postoperative bleeding. Prothrombin complex concentrate (PCC) containing clotting factors II (FII), VII (FVII), IX (FIX), and X (FX) could be of potential value in correcting dilutional coagulopathy and reducing blood loss.

Methods. Anaesthetized pigs underwent CPB with hypothermia for 2 h at 25°C followed by 1 h of normothermia. Approximately 1 h after CPB, animals randomly received either isotonic saline 1 ml kg^{-1} or PCC 30 IU kg^{-1} in a volume of 1 ml kg^{-1}. Diffuse coagulopathic bleeding was assessed as suture hole blood loss from a Gore-Tex patch placed over a full-thickness incision in the left carotid artery.

Results. After CPB, levels of FII, FVII, FIX, and FX declined from baseline by 32% to 48%, and PCC fully or partially reversed those deficits. Median suture hole blood loss after administration of saline placebo was 74 ml. PCC reduced suture hole bleeding by a median of 54 ml with a 95% confidence interval of 6–112 ml (P=0.026) compared with saline. PCC, but not saline, normalized skin bleeding time. Peak thrombin generation markedly decreased after CPB, but then returned in PCC-treated animals to a level higher than baseline by 28.7 nM (14.5–41.1 nM; P=0.031).

Conclusions. PCC was effective in correcting dilutional coagulopathy and reducing diffuse bleeding in an *in vivo* large-animal CPB model. Further research is warranted on PCC as a haemostatic agent in CPB.

Keywords: blood coagulation disorders; cardiopulmonary bypass; haemodilution; haemorrhage; prothrombin complex concentrates

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Excessive postoperative bleeding remains a frequent, serious, and unpredictable complication of cardiac surgery with cardiopulmonary bypass (CPB).[1–4] Such bleeding is often non-surgical. For instance, diffuse oozing with no identifiable surgical site of bleeding was present in 42 of 191 consecutive CPB patients (22%) undergoing re-exploration for excessive blood loss.[2]

Dilution of coagulation factors may contribute to excessive haemorrhage post-CPB.[5] Commencement of CPB precipitates an abrupt major haemodilution due to extracorporeal circuit priming with fluid volumes typically of ~2000 ml or more. More than 2000 ml of additional fluids may be administered during surgery, further exacerbating haemodilution. As a consequence, the estimated dilution of plasma clotting factors resulting from CPB surgery is ~50%.[5]

Prothrombin complex concentrate (PCC) could be of potential utility in correcting dilutional coagulopathy arising during CPB. The ability of PCC to ameliorate diffuse coagulopathic bleeding has already been demonstrated in a study of general surgery patients.[6] In that study, 27 patients with life-threatening diffuse bleeding and an international normalized ratio (INR) above 1.1 received Beriplex P/N, a biochemically well-characterized balanced PCC containing coagulation factors II (FII), VII (FVII), IX (FIX), and X (FX), and the anticoagulant proteins C and S.[7] Cessation of bleeding was attained in 26 patients (96%) within 3 h of PCC administration. Clinical evidence on the use of Beriplex P/N in cardiac surgery has also been provided by case reports, a retrospective study, and a randomized trial.[8–10]

A conventional option for correcting haemodilution-induced coagulation factor deficiencies after CPB is transfusion of fresh-frozen plasma (FFP), which contains all the coagulation factors. However, since FII, FVII, FIX, and FX are present in Beriplex P/N at ~25 times the corresponding plasma concentrations, smaller volumes and less time are needed to correct dilutional coagulopathy compared with FFP. Also, FFP requires thawing, and blood type matching may also be needed. Furthermore, most FFP preparations have not been virally inactivated. In the purification of Beriplex P/N, such inactivation is accomplished by pasteurization and nanofiltration of plasma screened by polymerase chain reaction.[7 11] There has been no evidence in clinical trials of viral transmission related to Beriplex P/N infusion.[12 13] The present study evaluated the ability of Beriplex P/N to attenuate diffuse post-CPB bleeding in a porcine model.

Methods

Animals

Seventeen castrated male pigs (large white × German noble) weighing 24–40 kg were procured from a local breeding farm (Willi Schlosser, Schwalmtal, Germany) at age 3–4 months. The animals were housed at 18–21°C in stables with straw bedding under ambient day–night cycles and fed *ad libitum* with Deuka V pig chow (Deutsche Tiernahrung Cremer GmbH & Co., KG, Düsseldorf, Germany). Tap water was supplied *ad libitum*. Animal husbandry and study procedures complied with the German Animal Welfare law and European Union regulations. The study was approved by the regional government authorities.

Anaesthesia

Anaesthetic procedures have been previously described.[14] Briefly, after an overnight fast with unrestricted access to water and i.m. premedication using a mixture of azaperone 2 mg kg^{-1} (Stresnil®, Janssen-Cilag GmbH, Neuss, Germany), ketamine 15 mg kg^{-1} (Ketavet, Pharmacia & Upjohn, Erlangen, Germany), and atropine sulphate 0.02 mg kg^{-1} (Atropinsulfate, B. Braun Melsungen AG, Melsungen, Germany), the pigs were anaesthetized with thiopental sodium 10 mg kg^{-1} *via* an ear vein. After tracheal intubation, respiration was supported *via* a Heyer Access ventilator. Inhaled anaesthesia was maintained with isoflurane 1–2% (Isofluran CP®, CP Pharma GmbH, Burgdorf, Germany). Attainment and maintenance of deep anaesthesia were confirmed by an absent pedal withdrawal reflex and lack of any response to surgery. A 1.4 × 2.1 mm catheter was advanced into a carotid artery for collection of blood samples and a 0.5 × 0.9 mm catheter into a femoral artery for continuous arterial pressure measurements. Ringer's solution at 4 ml kg^{-1} h^{-1} to satisfy basal fluid requirements and test fluids were infused *via* an indwelling 1.4 × 2.1 mm catheter in an external jugular vein. Body temperature was monitored by rectal thermometry.

Cardiopulmonary bypass

The experimental design is outlined in Figure 1. Sternotomy was performed with an oscillating saw, and the heart was exposed. The pericardium was opened longitudinally and secured to the chest wall with four sutures. Two purse-string sutures each were placed in the ascending aorta and right atrium. An i.v. bolus of heparin 300 U kg^{-1} was administered. After 10 min, a 5.2 mm diameter arterial catheter and a 32 Fr venous catheter were placed and secured with tourniquets. Both catheters were connected to a small adult hollow fibre oxygenator with a hard shell venous reservoir (D905 EOS, Sorin SpA, Milan, Italy). The extracorporeal circuit was primed with a solution consisting of isotonic saline 500 ml, 6% hydroxyethyl starch 200/0.5 1000 ml (Infukoll, Schwarz Pharma AG, Mannheim, Germany), 15% mannitol 2 ml kg^{-1} (Osmofundin®, B. Braun) and heparin 1000 units. Hydroxyethyl starch has been extensively investigated for priming in cardiac surgery randomized trials[15] and is a common choice in clinical

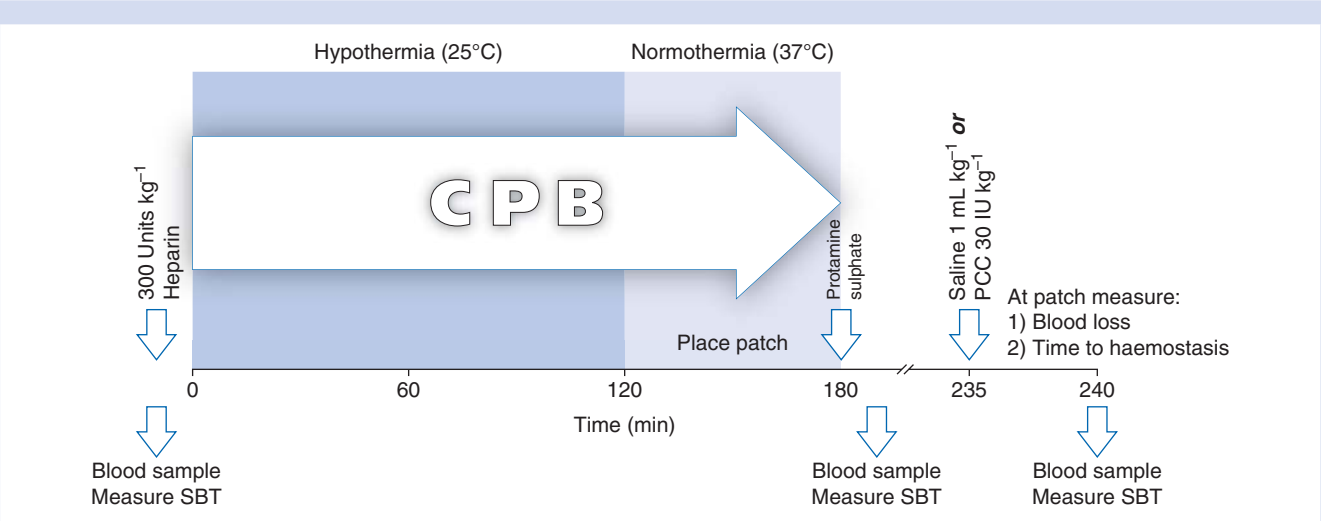

Fig 1 Study design. CPB, cardiopulmonary bypass; PCC, prothrombin complex concentrate; SBT, skin bleeding time.