Original Article

The Influence of Heparin-Coated and Uncoated Extracorporeal Circuits on Blood Rheology During Cardiac Surgery

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ABSTRACT

The effect of heparin-coated perfusion circuits on blood trauma during clinical cardiopulmonary bypass (CPB) was studied in order to find out if traumatic changes in the blood could be minimized. Twenty-four patients undergoing coronary artery bypass surgery were randomized prospectively to CPB with heparin-coated circuits (HCC) or non-coated circuits (NCC). The trauma to blood was assessed by measuring damage to blood cells by estimating red and white cell rheology changes. These were measured as red cell filtration rate (RFR) and white cell filtration rate (WFR) using standard microfiltration methods. Furthermore, changes in plasma hemoglobin (P-Hb), whole blood and plasma viscosity were simultaneously assessed. The RFR was significantly reduced in both groups during CPB by 10% in the HCC and 32% in the NCC groups ($p<0.01$). When comparing the HCC and NCC groups, a significant difference was first seen after 30 minutes of bypass ($p<0.05$) and increased at the end of CPB ($p<0.01$). Similar results were seen regarding WFR (15% and 36%, $p<0.01$). After 30 minutes of bypass, a significant difference was seen between HCC and NCC groups ($p<0.05$). Furthermore, a significant increase in P-Hb levels were seen during CPB in both patient groups. At the end of CPB, there was a significant difference in P-Hb levels (HCC 305±90 mg/L; NCC 455±78 mg/L, $p<0.01$) when comparing the two groups.

When comparing HCC and NCC regarding corrected blood viscosity at shear rate 92 s^{-1}, there were significantly higher viscosity values in the NCC group starting at 30 minutes of bypass ($p<0.05$). Plasma viscosity (PV) at shear rate 583 s^{-1}, showed similar results ($p<0.05$).

This study suggests that heparin-coated extracorporeal circuits are less damaging to the rheological properties of blood and would therefore be better suited for clinical use during cardiac surgery using CPB.

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INTRODUCTION

During open heart surgery, blood is in contact with artificial materials which constitute the non-physiological surfaces of the various devices routinely used for extracorporeal circulation (ECC). These nonbiological surfaces are in direct contact with blood components resulting in the deterioration of blood which can be measured by rheological changes and cellular and plasma component activations. (1-6)

Blood damage in turn leads to increased postoperative morbidity (7) such as bleeding, organ dysfunction and sometimes organ failure. Therefore efforts have been made to reduce the damaging effect of ECC on blood components. Improved biocompatibility of the artificial surfaces used in ECC is needed to reduce the side effects. The presence of heparin-coated circuits has been reported to be favorable in prolonging the use of ECC (8).

In the present study we investigated if heparin-coated circuits protected blood during CPB in patients undergoing coronary artery bypass surgery.

PATIENTS AND METHODS

Twenty four patients were electively operated on for coronary artery bypass surgery by either using vein grafts, the left internal mammary artery or a combination thereof. All patients had normal preoperative coagulation parameters. Patients with previous cerebrovascular accidents, diabetes with peripheral vascular complications, bleeding disorders, anticoagulation therapy, intermittent claudication, pulmonary, renal or hepatic diseases were excluded from the study. The study protocol conformed to the rules of the Helsinki declaration and was approved by the Ethics Committee of the University of Gothenbourg. Informed consent was obtained from patients participating in the study.

Patients were prospectively randomized into 1 of 2 groups:

Heparin-coated circuit group (HCC): 12 patients (11 males and 1 female) were operated on with the use of heparin-coated CPB circuits. The mean age was 65±8.4 years (mean±SD). Two patients were in NYHA-class II and 10 patients in NYHA-class III .

Non-coated circuit group (NCC): 12 male patients were operated on with non-coated CPB circuits. The mean age was 64±7 years (mean±SD). Three patients were in NYHA-class II and 9 patients were in NYHA-class III. Patient data are given in Table 1.

Anesthesia: Premedication consisted of morphine 0.1 mg/kg i.m. and scopolamine 0.4 mg. Anesthesia was induced with thiopental 3-5 mg/kg, followed by pancuronium 0.1 mg/kg. Fentanyl was given in incremental doses during induction of anesthesia and after intubation up to a total amount of 15 ug/kg. All operations were performed through a midline sternotomy using ECC with cannulation of the ascending aorta and right atrium.

Cardiopulmonary Bypass: A heart-lung machine with a roller pump[a] was used. The CPB circuit consisted of a Maxima membrane oxygenator[b] connected to a collapsible soft venous reservoir[b]. The CPB circuit was primed with 2000 ml Ringerdex[c]. A modified St.Thomas solution[c] was used to achieve cardioplegia and general hypothermia (28-30°C) was instituted. Patients were given the standard dose of heparin (300 IU/kg). CPB was started when the activated coagulation time (ACT) was above 450 sec. Additional heparin was given if the ACT fell below 450 sec. At the end of CPB, the effect of heparin was reversed by protamine. The blood flow rate at 37°C and during rewarming was 3-4 L/min/m^2 and 2 L/min/m^2 during hypothermia at 28°C. The gas flow-blood flow ratio was started at 1:1. The gas flow was then reduced according to the arterial blood gases where a pO_2 of 75-113 mmHg and pCO_2 of 30-45 mmHg were aimed for. The hematocrit was maintained between 18-25%. A cardiotomy suction was used to return the blood from the chest to the heart-lung machine. After CPB, the remaining blood was collected by chest drains into the cardiotomy reservoir and retransfused back to the patient. In the HCC group, the extracorporeal circuits were heparin-coated from cannula to cannula except the reservoirs (Carmeda Bioactive Surface[b]).

Measurement of routine hematological parameters. Blood samples for plasma hemoglobin (P-Hb) and hematocrit (HCT) were taken at the induction of anesthesia, beginning of CPB, every 30 minutes thereafter, and after CPB was terminated. Total hemoglobin (Hb), white cell count (WCC), and platelet count (PC) were also taken at these same times.

Red cell filtration rate (RFR) measurement technique. Blood samples for microfiltration studies were taken during induction of anesthesia, start of CPB, and every 30 minutes thereafter, and after termination of CPB. A microfiltration method previously described was used (9). Micropore filters with a 5um pore diameter and 4 x 10^5/cm^2 pore density were obtained from Nucleopore[d]. Filterability was expressed as the filtration rate of a red cell suspension in microliters per second (ul/s). Ten ml of venous blood was drawn into a syringe containing ethylenediaminetetraacetic acid (EDTA). All samples were analyzed within 1 hour. The sampled blood was centrifuged at 4000 rpm for 5 minutes, the supernatant fluid representing the plasma and the buffy coat of white cells was discarded and used for white cell filtration. The red cells were washed twice in normal saline at room temperature (22°C) and suspended in saline in an amount giving a 20% red cell suspension. 0.5 ml was allowed to pass by gravity through the filter. The height of the suspension above the filter was allowed to decrease from 11.5 to 8.5 cm and the time required for this decrease was recorded. Measurements were performed at room temperature. For each value, a mean of three readings was calculated. Each filter was initially standardized

a Gambro, Lund, Sweden
b Medtronic Cardiopulmonary Division, Anaheim, CA
c Pharmacia, Uppsala, Sweden
d Nucleopore Corp., CA

Figure 1
% RFR Reduction During ECC
(HCC = coated; NCC = control). START = start of ECC; STOP = end of ECC. RFR = red cell filtration rate.

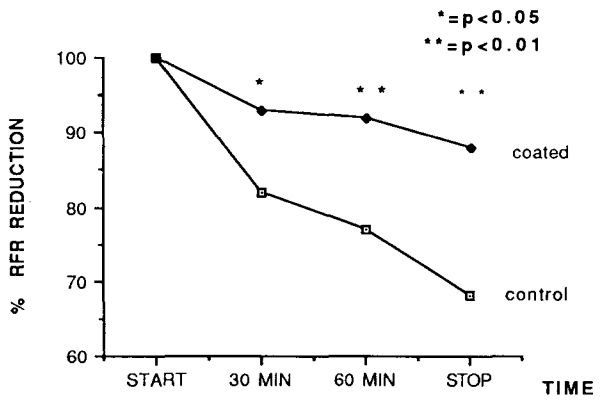
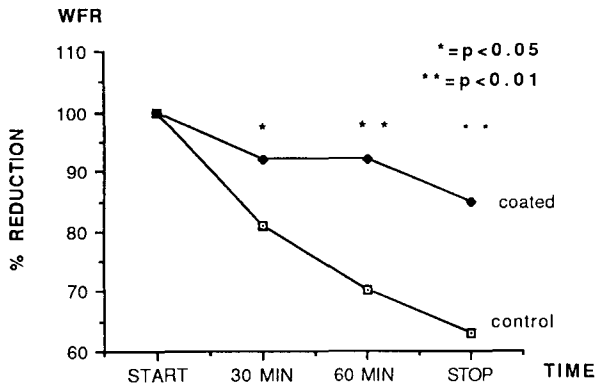


Figure 2
% WFR Reduction During ECC
(HCC = coated; NCC = control). START = start of ECC; STOP = end of ECC. RFR = red cell filtration rate.



with buffered glucose and only filters with a mean flow time of 2±0.2 seconds were used. The RFR was calculated by a standard formula (9).

White cell filtration rate (WFR) measurement technique. The supernatant plasma was discarded and the buffy coat then diluted with isotonic saline giving a count of 4 x 10^9/L ±400 WBC; the WBC suspension (0.5 ml) was then filtered as above.

Viscosity Measurements. The Bohlin rheometer (10,11) is a couvette viscometer with rotation speeds (0.0019-1470 rpm) that are computer controlled, with an integrated temperature control unit. The system can construct temperature or time-programmed flow curves at different sweep speeds.

Table 1
Patient Data

		Heparin-coated	Control
Number	M	11	12
	F	1	0
Age (yrs)		65±8.4	64±7.0
Ejection Fraction (%)		54±12	57±10
No. of Grafts		4.4±1.4	3.3±1.1

Table 2
Perfusion Data

	Heparin-coated	Control
ECC-time (min)	113±22	96±31
Aortic cross clamp time (min)	69±19	54±24
Rectal Temp (°C)	31.6±0.5	31.9±0.9
Gas flow (L/min)	3.3±0.5	3.6±1.5
Blood flow (L/min)	3.5±1.6	4.1±1.6
Hematocrit (%)	21±4	23±3
Heparin (5000 U/ml)	6.9±1.8	6.8±1.2
Protamine (mg)	325±50	329±40

Table 3
Plasma Hemoglobin (mg/l) During ECC

Time (min)	HCC (n=12)	NCC (n=12)	p
30	168±42	205±59	<0.05
60	218±95	309±89	<0.02
Stop	305±90	443±67	<0.01

The coefficient of variation (CV) was determined by 10 consecutive measurements of the same blood and plasma. For native blood viscosity (NBV) at shear rate 92 s^{-1} the CV was 2%. For corrected blood viscosity (CBV) at shear rate 92 s^{-1} adjusted before measurement the CV was 3%. The CV for plasma viscos-

Table 4
Corrected Blood Viscosity (mPas) at a Shear Rate 92 s^{-1}

Time(min)	HCC (n=12) mean (SD)	NCC (n=12) mean (SD)	p
30	6.19±(0.36)	6.68±(0.39)	<0.05
60	6.57±(0.34)	7.18±(0.42)	<0.01
Stop	6.87±(0.44)	7.72±(0.48)	<0.001

Table 5
Plasma Viscosity (mPas) at a Shear Rate 583 s^{-1}

Time(min)	HCC (n=12) mean (SD)	NCC (n=12) mean (SD)	p
30	1.78±(0.058)	2.05±(0.072)	<0.05
60	1.97±(0.110)	2.49±(0.090)	<0.01
Stop	2.14±(0.100)	2.84±(0.121)	<0.001

ity (PV) at shear rate 583 s^{-1} was 1.4%. A standard silicon oil with a known viscosity of 9.5 mPas at 23°C was used for calibration (12). NBV and PV were measured at a constant temperature of 27°C and at shear rates of 92 s^{-1} and 583 s^{-1} respectively.

STATISTICS

Means are expressed with one standard deviation (SD). Comparisons between two means were performed using the appropriate t-test. For comparison between percentages, the percentage test was used (13). A p value of less than 0.05 was considered statistically significant. The Statview statistical program was used with a Macintosh computer[e].

RESULTS

There were no significant differences in the mean values for perfusion time, aortic cross clamp time, body temperature, gas and blood flow rates between the two groups (Table 2). No significant differences were found preoperatively regarding red cell, white cell or platelet count between groups. All patients survived the operative procedure and were discharged from the hospital.

The RFR in both HCC and NCC groups was significantly reduced during CPB but the reduction was greater in the NCC group, reaching a significant difference after thirty minutes ($p<0.05$) (Figure 1). The differences in RFR values remained statistically significant throughout CPB, and at the end of CPB the HCC group had a total reduction of 10%, compared to 32% in the NCC group ($p<0.01$) (Figure1).

The WFR in both HCC and NCC groups was significantly reduced during CPB, with the NCC group showing a significantly greater reduction than the ICC group ($p<0.05$) (Figure 2). At the end of CPB the WFR in the HCC group was reduced by 15% compared to 36% in the NCC group ($p<0.01$) (Figure 2).

The mean P-Hb increased significantly with the duration of CPB in both groups. At all measurements the P-Hb in the HCC group was significantly less elevated when compared to the NCC group (Table 3).

No difference was found when comparing the NBV of HCC and NCC groups at a shear rate of 92 s^{-1} (5.29±0.245 and 5.16±0.339, respectively).

At a shear rate 92 s^{-1} and a constant hematocrit of 25%, a significant difference was seen in CBV when comparing HCC and NCC groups throughout CPB ($p<0.05$) (Table 4).

At a shear rate 583 s^{-1}, PV was significantly elevated in the NCC group when compared to the HCC patients throughout CPB (Table 5).

DISCUSSION

The results of this study showed that the heparin-coated surface appears to have a protective function to blood cells during CPB when using blood cell rheology to assess blood trauma. Blood cell filtration tests offer a rapid and relatively cheap and simple method of assessing the flow properties of blood cells, in a model simulating the microcirculation in the capillaries. The use of filterability measurements is widespread (14-21). The method, in this study, used a suspension of cells of fixed hematocrit or cell number, which was allowed to flow through a filter with a fixed number of pores ($4x10^{-5}$ pores/cm^2). In general, for red cells, and in the presence of a 5um pore filter, there is a relatively lesser degree of pore blockage while for white cells, the cell suspension, although more diluted, the blockage is generally more marked. Thus, filtration rates for RBC are much greater than white cells as seen in the various studies and has been observed by other authors (21). Abnormal red cell filterability is present in patients with chronic circulatory disease (22-24), organ failure (25-27), inherited disorders (28,29) and infection (30). Filterability measurements of white cells may reflect any process which stimulates circulating white cells (29,31).

During open heart surgery, using extracorporeal circulation, the damage to blood cells is associated with the contact with artificial surfaces, gas and blood flow rates, use of suction, priming fluids, and activated blood enzyme systems on the blood cell membranes (32-38). Several studies have shown changes in blood cell rheology during CPB (39-45). A number of changes in the blood have been described during CPB, such as activation of the complement system (41), a deposition of complement complexes on erythrocytes (46), hemolysis (36), thrombocytopenia (47), leukocytopenia (48), platelet damage (4,49-52), and changes

e Apple Computer Inc., Cupertino, CA 95014

in blood clotting (53). All the phenomena described above could contribute to the changes of filterability found during cardiopulmonary bypass. Thus, the trauma to blood cells probably led to reductions in RFR and WFR.

The protective effect of heparin-coated surfaces seems to reduce the usual loss of rheologic function of red and white blood cells. Previous studies have confirmed blood cell rheologic deterioration during CPB. (2) Factors known to reduce blood cell filterability are CPB time, high oxygenator gas and flow rates, bubble oxygenators, complement activation and clinical state of the patient (54-57). The CPB time and gas flow rates in both groups were comparable. The same type of oxygenator was also used in all patients in this study. The patient data in HCC and NCC may also be considered as comparable. Thus, the only difference between the groups is the heparin-coating, which strongly suggests a positive influence in reducing blood trauma. It can be inferred that protection of blood cell rheology would have a sparing action on the microcirculation disturbances seen during and after CPB. Thus, one would expect a potential decrease in postoperative morbidity and complications resulting from rheologic protection (7).

The exact mechanism of this beneficial effect on blood rheology has not been studied here. It has been suggested through in vitro and in vivo studies that a reduction in complement activation and white blood cell damage may be responsible for the protective action of heparin-coated surfaces during ECC (58,59).

The activation of white blood cells would lead to the release of free radicals which are known to damage red blood cells*. The increase in plasma hemoglobin in both groups suggests that the direct red blood cell trauma due to shear stress is comparable during the early stages of CPB. However, at the end of CPB, the significant difference in plasma hemoglobin between the two groups suggests that other factors, besides mechanical stress, appear to be responsible for the increased hemolysis in the NCC group. It is known that the ADP release from hemolyzed red blood cells activates platelet aggregation, which in turn could induce free radical generation from white blood cells, thus leading to further red cell damage.

These results should be complemented by further studies examining platelet and white cell function to clarify the beneficial effects of heparin coating.

In conclusion, heparin-coated cardiopulmonary bypass circuits were shown to protect blood rheology, and thereby improve biocompatibility of the surfaces in these circuits. We suggest that heparin-coated circuits may be used for routine cardiac surgery using cardiopulmonary bypass for either short or prolonged procedures.

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