Original Article

Biocompatibility of Poly2methoxyethylacrylate Coating for Cardiopulmonary Bypass

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The systemic inflammatory response to cardiopulmonary bypass (CPB) may contribute to the development of postoperative complications. Heparin-coated circuits and poly2methoxyethylacrylate (PMEA)-coated circuits have been developed to reduce the risk of such complications. We compared the biocompatibility of these circuits. Twelve patients scheduled to undergo elective coronary artery bypass grafting (CABG) with CPB were assigned to CPB with a PMEA-coated circuit (PMEA-coated group, n=6) or a heparin-coated circuit (heparin-coated group, n=6). The plasma concentrations of the following inflammatory markers were measured before CPB and just after, 4 hours after, and 24 hours after the termination of CPB: cytokines (interleukin [IL]-6, IL-8, IL-10), complement factor (C3a), polymorphonuclear elastase (PMNE), and coagulofibrinolytic factors (thrombin-antithrombin III complex [TAT], D-dimer). Postoperative clinical response was evaluated on the basis of respiratory index, blood loss, and the postoperative and preoperative body-weight percent ratio. There were no significant differences between the groups in the plasma concentrations of IL-6, IL-10, C3a, PMNE, TAT, or D-dimer. Plasma IL-8 concentrations were below the assay detection limits at all time points in both groups. Clinical variables did not differ significantly between the groups. In conclusion, PMEA-coated CPB circuits are as biocompatible as heparin-coated CPB circuits and prevent postoperative organ dysfunction in patients undergoing elective CABG with CPB. (Ann Thorac Cardiovasc Surg 2003; 9: 22–8)

Key words: poly2methoxyethylacrylate (PMEA), cardiopulmonary bypass (CPB), coronary artery bypass grafting (CABG)

Introduction

The systemic inflammatory response to cardiopulmonary bypass (CPB) is characterized by activation of the coagulation, fibrinolytic, and complement cascades, resulting in the production and release of proinflammatory cytokines and the stimulation of neutrophil adhesion and degranulation.[1)] This inflammatory response may contribute to the development of postoperative complications, including respiratory failure, renal dysfunction, bleeding disorders, and multiple organ failure.[2)] During the past few decades, heparin-coated circuits for CPB have been developed and their biocompatibility has improved. Heparin-coated circuits can reduce complement activation and consequent release of cytokines.[3)] Recently, new polymer-coated (Xcoating™) extracorporeal circuits, the blood-contacting surfaces of which are coated with poly2-methoxyethylacrylate (PMEA), have been developed by Terumo Corporation.[4)] We clinically compared the biocompatibility of PMEA-coated circuits with that of heparin-coated circuits in patients undergoing coronary artery bypass grafting (CABG) with CPB.

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Patients and Methods

Between March 2000 and February 2001, 12 patients undergoing elective CABG with CPB were assigned to two groups. Informed consent was obtained from all the patients studied. The heparin-coated group consisted of the initial six serial patients in whom a heparin-coated CPB circuit (CAPIOX-SX(HP), Terumo Corporation, Tokyo, Japan) was used. The PMEA-coated group consisted of the following six serial patients in whom a PMEA-coated CPB circuit (CAPIOX-RX, Terumo Corporation) was used. All components of the circuits were coated with either heparin or PMEA respectively except for aortic and venous cannulae. Patients scheduled to undergo single CABG and those with malignancy, autoimmune disease, or coagulopathy were excluded from this study.

Standardized protocols were followed for anesthesia and CPB. After premedication, general anesthesia was induced by modified neurolepto-anesthesia and analgesia. Anesthesia was maintained with fentanyl, propofol, sevoflurane, and vecuronium bromide. The subjects received 100 mg of betamethasone sodium phosphate intravenously before the initiation of CPB. The extracorporeal circuit and oxygenator were primed with 1.8 L of 20% D-mannitol (3 mL/kg), 6% hydroxyethylated starch (10 mL/kg), and Ringer's lactate solution without blood. Nonpulsatile extracorporeal circulation was initiated at a perfusion index stratified according to body surface area. A roller pump (Sarns 9000: Terumo Cardiovascular Systems, Ann Arbor, MI) was used. The subject was given an initial pre-bypass bolus dose of heparin (150 IU/kg). Intravenous doses of heparin were given as needed to maintain the whole blood activated clotting time at greater than 300 seconds for the entire duration of CPB. The operative procedures were performed under moderate whole body hypothermia, with the rectal temperature maintained between 28°C and 32°C. Myocardial protection was provided by injecting cold cardioplegic solution (4°C). Prostaglandin E1 was used to maintain an adequate perfusion pressure (40 to 60 mmHg) during CPB. After CPB, protamine sulfate (150 IU/kg) was administered.

Blood was withdrawn from an indwelling arterial cannula at the following times: before CPB; just after the termination of CPB; four hours after the termination of CPB; and 24 hours after the termination of CPB. Plasma samples were separated immediately by centrifugation (3,000 rpm) for 15 minutes at 4°C. The samples were stored at –80°C until analysis by enzyme-1inked immunosorbent assay kits (interleukin [IL]-6 {QuantiGlo human IL-6 Immunoassay, R&D systems, Minneapolis, MN}, IL-8 {HUMAN INTERLEUKIN-8 ELISA (enzyme-linked immunosorbent assay) KIT, TFB, Tokyo, Japan}, IL-10 {CytoscreenUS hIL-10 ULTRA SENSITIVE, BIOSOURCE INTERNATIONAL, Camarillo, CA}, thrombin-antithrombin III complex [TAT] {Enzygnost TAT micro, DADE BEHRING, Liederbach, Germany}), latex agglutination (polymorphonuclear elastase [PMN-E] {Ecoline PMN elastase, Diagnostica MERCK, Darmstadt, Germany}, D-dimer {LPIA ACE D-D DIMER, DIA-IATRON, Tokyo, Japan}), and radioimmunoassay (complement factor [C3a] {Human Complement C3a des Arg (^{125}I) assay system, Amersham, Buckinghamshire, U.K.}). The limits of sensitivity were 0.15 pg/ml (IL-6), 10 pg/ml (IL-8), 0.5 pg/ml (IL-10), 20 ng/ml (C3a), 14.3 μg/l (PMNE), 0.5 ng/ml (TAT), and 0.5 ng/ml (D-dimer).

Respiratory index (RI) is an indicator of oxygenation that reflects the presence of various pulmonary complications. To standardize alveolar-arterial oxygen gradients to the inspired fraction of oxygen during ventilation, the RI was calculated as follows: RI=alveolar-arterial oxygen tension gradient/arterial oxygen tension. The index was calculated before the initiation of CPB, just after the end of CPB, and just after admission to the intensive care unit (ICU). To examine differences in postoperative clinical status, blood loss through chest drainage tubes was measured for 24 hours after surgery and compared between the groups. Postoperative blood loss was expressed in mL/kg of body weight. The body-weight percent ratio just after admission to the ICU and 24 hours after CPB was determined as an index of postoperative body weight gain.

All variables are expressed as mean±standard error. Repeated-measures analysis of variance was performed to examine differences between the groups over time. The Wilcoxon signed-rank test was used to evaluate differences as compared with baseline values in each group. A p value of less than 0.05 was considered to indicate statistical significance.

Results

There were no statistically significant differences between the groups with respect to age, sex, operative time, CPB time, and aortic cross-clamp (AXC) time (Table 1). There were no deaths or postoperative complications in this series.

Table 1. Clinical characteristics

	PMEA-coated group (n=6)	Heparin-coated group (n=6)
Age (years)	70.0±2.3	68.7±3.2
Sex (male/female)	4/2	2/4
No. of grafts (branch)	3.2±0.3	2.8±0.3
Operative time (min)	293±21	297±27
CPB time (min)	119±15	121±11
AXC time (min)	78±12	76±9

CPB, cardiopulmonary bypass; AXC, aortic cross-clamp

Cytokines

IL-6 (Fig. 1)

In the PMEA-coated group, IL-6 gradually increased and reached peak concentration (74.5±9.5 pg/ml) just after the termination of CPB. Thereafter, the concentration decreased. In the heparin-coated group, IL-6 reached peak concentration (108.3±20.4 pg/ml) four hours after the termination of CPB and decreased subsequently. In both groups, IL-6 concentrations 24 hours after the termination of CPB remained significantly higher than their respective values before CPB ($p<0.05$). There were no significant differences between the groups at any time.

IL-8

In both groups, the IL-8 concentrations were under the detection limit of the assay at all time points.

IL-10 (Fig. 2)

In both groups, the IL-10 concentrations reached peak values (PMEA-coated group, 293±53 pg/ml; heparin-coated group, 486±114 pg/ml) just after the termination of CPB and decreased gradually. Twenty-four hours after the end of CPB, the IL-10 concentration returned to the preoperative level in the PMEA-coated group, but not in the heparin-coated group. There were no significant differences between the groups at any time.

C3a (Fig. 3)

The C3a concentration just after the termination of CPB was sevenfold higher than the pre-bypass value in both groups (PMEA-coated group, 2,228±401 ng/ml; heparin-coated group, 1,895±282 ng/ml). The C3a concentration rapidly returned to the baseline value and remained within normal range 24 hours after the termination of CPB. The

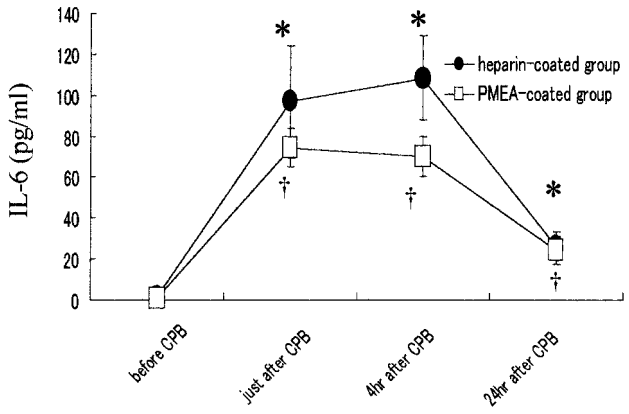

Fig. 1. Changes in plasma IL-6 concentrations before, just after, four hours after, and 24 hours after CPB.
*$p<0.05$ vs. baseline value in the heparin-coated group, †$p<0.05$ vs. baseline value in the PMEA-coated group

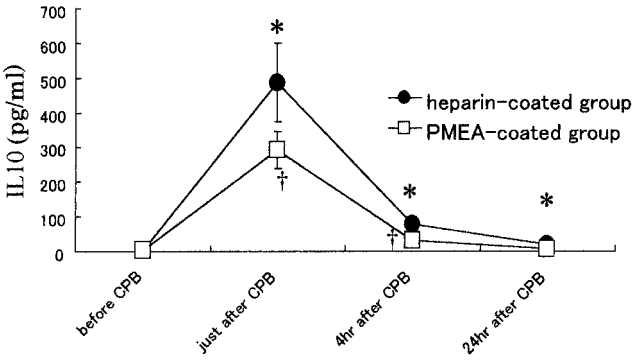

Fig. 2. Changes in plasma IL-10 concentrations before, just after, four hours after, and 24 hours after CPB.
*$p<0.05$ vs. baseline value in the heparin-coated group, †$p<0.05$ vs. baseline value in the PMEA-coated group

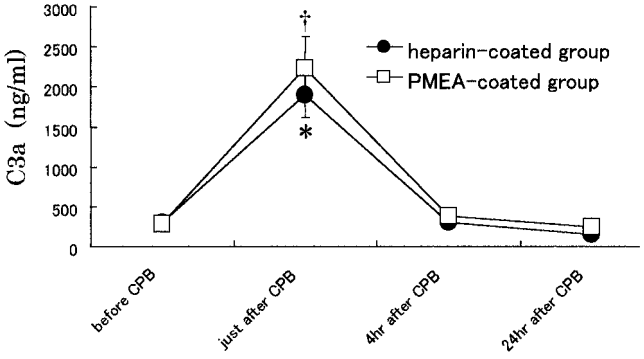

Fig. 3. Changes in plasma complement factor (C3a) concentrations before, just after, four hours after, and 24 hours after CPB. *p<0.05 vs. baseline value in the heparin-coated group, †p<0.05 vs. baseline value in the PMEA-coated group

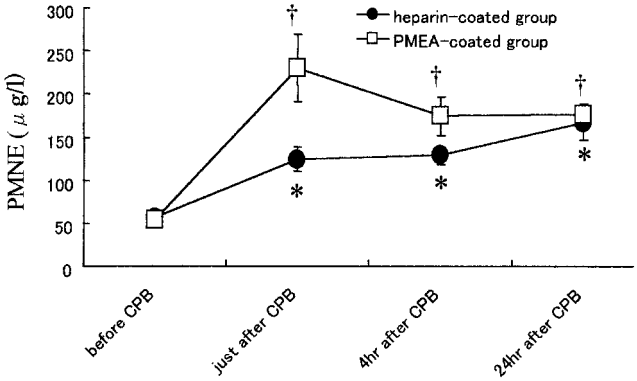

Fig. 4. Changes in plasma PMNE concentrations before, just after, four hours after, and 24 hours after CPB. *p<0.05 vs. baseline value in the heparin-coated group, †p<0.05 vs. baseline value in the PMEA-coated group

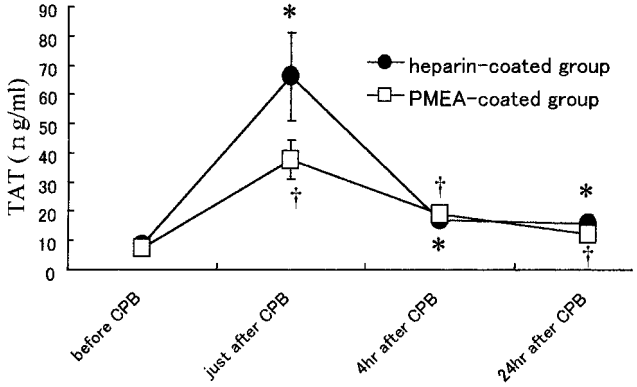

Fig. 5. Changes in plasma TAT concentrations before, just after, four hours after, and 24 hours after CPB. *p<0.05 vs. baseline value in the heparin-coated group, †p<0.05 vs. baseline value in the PMEA-coated group

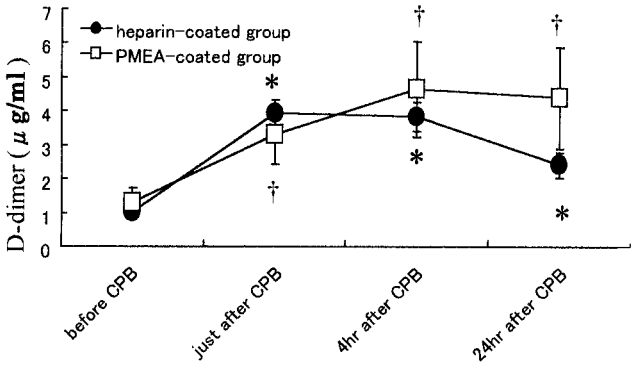

Fig. 6. Changes in plasma D-dimer concentrations before, just after, four hours after, and 24 hours after CPB. *p<0.05 vs. baseline value in the heparin-coated group, †p<0.05 vs. baseline value in the PMEA-coated group

C3a concentrations did not differ significantly between the groups at any time.

PMNE (Fig. 4)

In the PMEA-coated group, the PMNE concentration gradually increased and reached peak value (230±39 μg/l) just after the termination of CPB. The concentration remained high until 24 hours after the termination of CPB. In the heparin-coated group, the PMNE concentration began to increase steadily and reached peak value (167±21 μg/l) 24 hours after the termination of CPB. There were no significant differences between the groups at any time.

TAT (Fig. 5)

In both groups, the TAT concentrations reached peak values (PMEA-coated group, 37.7±6.6 ng/ml; heparin-coated group, 66.1±15.1 ng/ml) just after the termination of CPB and then decreased gradually. The TAT concentration 24 hours after the termination of CPB remained significantly higher than that before CPB in both groups ($p<0.05$). There were no significant differences between the groups at any time.

D-dimer (Fig. 6)

As compared with the preoperative value, the D-dimer concentration in the PMEA-coated group increased significantly just after the termination of CPB, reached peak value (4.64±1.41 μg/ml) four hours after the termination of CPB, and decreased slightly thereafter. In the heparin-coated group, the D-dimer concentration reached peak value just after the termination of CPB (3.95±0.42 μg/ml) and then decreased gradually. There were no signifi-

Table 2. Postoperative patient data

	PMEA-coated group (n=6)	Heparin-coated group (n=6)
Blood loss (ml/kg)[a]	8.4±1.1	13.7±3.8
Respiratory index		
Before CPB	0.27±0.05[b]	0.85±0.12
Just after CPB	1.49±0.48	2.01±0.58
Just after ICU admission	1.68±0.42	1.84±0.28
%R-BW		
Just after ICU admission	104.5±0.4	104.4±0.7
24 hours after CPB	102.5±0.5	101.6±0.6
Intubation time (hours)	9.0±2.7	6.5±1.7

[a]from ICU admission to 24 hours thereafter, [b]$p<0.05$ vs. baseline of the heparin-coated group
CPB, cardiopulmonary bypass; ICU, intensive care unit; %R-BW, body-weight percent ratio as compared with preoperative value

cant differences between the groups at any time.

Patients and postoperative clinical variables

Intraoperative clinical data are summarized according to group in Table 1. There were no significant differences between the groups in the CPB or AXC time, the administered doses of heparin or protamine, and the volume of transfused blood.

The postoperative clinical data are summarized in Table 2. There were no operative deaths or early re-explorations for bleeding in either group. There were also no significant differences between the groups in postoperative blood loss during the first 24 hours and in intubation time. The PMEA-coated group had a significantly lower RI than the heparin-coated group before CPB, but there were no other significant differences between the groups. The postoperative and preoperative body-weight percent ratios just after operation and one day after operation did not differ between the groups.

Comment

Our study demonstrated that the systemic inflammatory responses associated with CPB were similar for PMEA-coated circuits and heparin-coated CPB circuits in patients who underwent elective CABG. Although both types of circuits have been clinically shown to prevent morbidity and mortality, a number of unfavorable reactions related to CPB have been reported during the past 10 years.[1,5] The so-called post-pump inflammatory response is attributed to exposure of blood to foreign materials and abnormal shear forces.[5] Preventing these adverse inflammatory responses remains an important concern.

The use of PMEA-coated circuits is based on the hypothesis that minimizing surface protein adsorption would decrease surface-biocomponent interactions and thereby enhance biocompatibility. The amount of protein adsorbed on PMEA-coated circuits is significantly lower than that on uncoated circuits. Although immunogloblin G (IgG) and immunogloblin M (IgM) are the major proteins adsorbed on uncoated circuits, they are not detected among the proteins adsorbed on PMEA-coated circuits.[4] Tanaka et al. have demonstrated in vitro that the high blood compatibility of PMEA is closely related to the low denaturation and high dissociation rate constant of proteins adsorbed onto its surface.[6]

IL-6 is involved in the modulation of acute phase response and is synthesized by a variety of activated cell types, including monocytes, macrophages, endothelial cells, and fibroblasts, in response to stimulation by tumor necrosis factor (TNF)-α and IL-1.[7] We found no significant difference in IL-6 release between the PMEA-coated and heparin-coated groups, although the absolute level of IL-6 in the PMEA-coated group was consistently lower than that in the heparin-coated group. These findings suggest a similar degree of tissue trauma in both groups.

IL-8 is an extremely potent chemoattractant for neutrophils and induces pulmonary sequestration of neutrophils similar to that in the adult respiratory distress syndrome.[8] In our study, IL-8 concentrations were consis-

tently below the assay detection limits in both groups. Our findings are not supported by the result of other studies, which have reported elevated levels of IL-8 during CPB.[3,9,10] This difference may be explained by the biocompatibility of both PMEA-coated and heparin-coated circuits.

The systemic inflammatory response is self-limiting in most patients. Endogenous factors limiting this response have been identified. IL-10 exerts a number of anti-inflammatory effects, including inhibition of pro-inflammatory cytokine synthesis. Increased levels of IL-10 have been confirmed to follow rises in pro-inflammatory cytokine levels during CPB and may represent an endogenous response designed to limit the inflammatory response.[11] In our study, the IL-10 concentration had a lower peak value in the PMEA-coated group than in the heparin-coated group just after CPB. Subsequently, the IL-10 concentration returned to the baseline level in the PMEA-coated group, but not in the heparin-coated group. These results provide indirect evidence suggesting milder inflammatory reactions in the PMEA-coated group than in the heparin-coated group.

Activation of the complement system due to exposure of blood to artificial surfaces is considered the initial step in CPB-induced inflammatory response.[5] PMEA coating reduces complement activation by suppressing protein adsorption, especially the adsorption of IgG and IgM, which combine with Clq and consequently activate C3 via the classical pathway. This mechanism differs considerably from the proposed mechanism by which heparin coating reduces complement activation. Suppression of complement activation by heparin is ascribed directly to the heparin molecule, particularly its inhibition of the alternative pathway of complement activation.[12,13] In our study, the C3a concentration reached peak value just after the termination of CPB and then promptly returned to baseline level in both groups. Our results contrast with those of a previous study in patients undergoing CPB with non-coated circuits. That study found that the C3a concentration just after CABG was fivefold higher than the preoperative value.[14] Our findings thus indicate that both PMEA and heparin coatings effectively reduced activation of the complement system.

PMNE is released from activated neutrophils induced by IL-8 or complement.[8,15] We obtained no evidence of reduced granulocyte activation with either circuit, although IL-8 remained below the assay detection limit and C3a returned to the baseline level four hours after the termination of CPB. We are somewhat confused by these inconsistent results, which probably arose from factors not directly related to CPB, such as surgical procedure, bleeding, anesthesia, or mechanical ventilation.

Heparin has no inherent anticoagulant activity. Rather, it accelerates TAT formation by binding the serine protease inhibitor antithrombin III to thrombin. In our study, the TAT concentration just after the termination of CPB was about twofold higher in the heparin-coated group than in the PMEA-coated group. This finding suggests that the anticoagulant activity of PMEA-coating is not mediated by antithrombin III.

Increased bleeding after cardiac operations might result from accelerated fibrinolytic activity after CPB. It has been reported that postoperative bleeding is strongly affected by postoperative fibrinolytic activity.[16] In the present study, although TAT increased rapidly just after the termination of CPB and then promptly decreased, both groups showed persistently high concentrations of D-dimer after CPB in accordance with the results of previous studies. Clinically, the bleeding volume was slightly but not significantly lower in the PMEA-coated group than in the heparin-coated group.

Numerous studies have demonstrated postoperative clinical benefits of heparin-treated CPB. However, the postoperative clinical benefits of PMEA-coated CPB remain incompletely understood. We found no major complications with PMEA-coated CPB or any significant difference in postoperative clinical variables as compared with heparin-coated CPB. In terms of safety, PMEA coating thus appears to be similar to heparin coating in the clinical setting of our study. We conclude that PMEA-coated CPB circuits are as biocompatible as heparin-coated CPB circuits and prevent postoperative organ dysfunction.

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