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A baboon model for hematologic studies of cardiopulmonary bypass

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Objective investigation of new inhibitors of blood protein or cellular systems that are activated during cardiopulmonary bypass (CPB) is impeded by the absence of a satisfactory animal model. Because most baboon hematologic proteins immunologically cross-react with those used for human assays, we developed a robust, reusable baboon model of CPB. Blood samples were obtained from adult baboons at six time intervals before, during, and after 60 minutes of partial CPB at 37° C with peripheral cannulas. Both membrane (n = 7) and bubble oxygenators (n = 7) were investigated. We measured platelet and white blood cell counts; platelet response to adenosine diphosphate and release of β -thromboglobulin; fibrinopeptide A, prothrombin fragment F_{1,2}, thrombin-antithrombin complex, D-dimer, and plasmin-antiplasmin complex; activated complement (C3b/c and C4b/c); elastase- α_1 proteinase inhibitor complex; and bleeding times. Adherent glycoprotein III_a antigen in Triton X-100 washes of the perfusion circuit was also measured. Markers of baboon platelet, complement, and neutrophil activation and thrombosis significantly increased during CPB with bubble oxygenator systems but did not change appreciably in membrane oxygenator circuits. Markers of fibrinolysis, D-dimer, and plasmin-antiplasmin complex did not change with either oxygenator. The baboon model of CPB, when a bubble oxygenator is used, is a robust, reusable animal model for evaluating inhibitors of platelet, complement, and neutrophil activation and thrombosis during and after CPB. (J Lab Clin Med 1997;130:412-20)

Abbreviations: ACD = acid citrate dextrose; ADP = adenosine diphosphate; ANOVA = analysis of variance; CPB = cardiopulmonary bypass; elastase- α_1 PI = elastase- α_1 proteinase inhibitor complex; ELISA = enzyme-linked immunosorbent assay; FPA = fibrinopeptide A; GP = glycoprotein; PAP = plasmin-antiplasmin complex; PBS phosphate-buffered saline solution; PPP = platelet-poor plasma; PRP = platelet-rich plasma; TAT = thrombin-antithrombin complex; β TG = β -thromboglobulin; WBC = white blood cell

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Cardiopulmonary bypass activates blood elements to cause bleeding, thrombotic emboli, massive fluid accumulation, and temporary organ dysfunction that produces much of

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the morbidity associated with open heart surgery.¹⁻⁵ During CPB, activation of blood elements produces a host of vasoactive substances and coagulation and inflammatory proteases that circulate.⁶⁻⁸ Efforts to inhibit activation of blood elements during CPB are impeded by the absence of a large animal model whose blood cross-reacts with antibodies developed for biochemical studies of human blood proteins and cells. In other animal models of CPB, the inability to use antibodies raised against human proteins and cellular receptors and the need to rely on functional assays of coagulation seriously impedes the evaluation of potential inhibitors.

Baboon blood has high immunologic cross-reactivity with most assays for human plasma proteins and cellular elements, and the baboon is large enough to allow extracorporeal perfusion through peripheral cannulas without sternotomy.⁹ We compared membrane and bubble oxygenators in this model to determine which system produced the most robust model for evaluation of potential inhibitors of activated blood elements.

METHODS

Juvenile female baboons (*Papio anubis*) weighing between 12 and 23 kg were divided into two groups: one group underwent CPB with a bubble oxygenator ($n = 7$) and a second group underwent CPB with a membrane oxygenator ($n = 7$). After a baboon was constrained with a squeeze cage, it received glycopyrrolate (0.4 mg IM), ketamine hydrochloride (10 mg/kg IM) for sedation, and finally thiopental sodium (5 mg/kg IV) for induction of anesthesia. The animal was endotracheally intubated, and general inhalational anesthesia was maintained with isoflurane. The right or left neck and both groins were prepared and draped appropriately for sterile cutdown and cannulation of vessels. Hemodynamic monitoring was accomplished by using an arterial line placed in the femoral artery and a Swan-Ganz catheter placed transvenously in a femoral vein. After initial anticoagulation with porcine sodium heparin (300 U/kg; Elkins-Sinn Inc., Cherry Hill, N.J.), a 10-14 French wire-reinforced, polyurethane catheter (BioMedicus; Medtronic Inc., Eden Prairie, Minn.) was introduced into the jugular vein and advanced into the right atrium. A similar, shorter 8 French arterial catheter was used for reinfusion by cannulation of the other femoral artery.

Each bypass circuit was assembled with Silastic tubing (Dow Corning Corp., Midland, Mich.), incorporating a bubble oxygenator (Bentley 5/Pediatric; Baxter Healthcare Inc., Irvine, Calif.) or a spiral coil membrane oxygenator (model 800-2A, 0.8 M²; Avecor Inc., Plymouth, Minn.), an arterial filter (Intersept Pediatric; Medtronic Inc., Anaheim, Calif.), and a roller pump (model 13400; Sarns 3M, Ann Arbor, Mich.). The circuit was primed with approximately 500 ml of Normosol (Abbott Laboratories,

North Chicago, Ill.). Normothermic CPB began at a flow rate of 50 ml/kg/min (approximately one half of the baboon's resting cardiac output), and perfusion was maintained for 60 minutes. Six blood samples (20 ml each) were obtained at baseline before heparin (Fig. 1, time point labeled *BASELINE*), 5 minutes after heparin (*HEPARIN*), 5 minutes after starting CPB (*START*), 5 minutes before stopping CPB (*END*), 10 minutes after the administration of protamine sulfate (3 mg/kg; Elkins-Sinn)(*PROTAMINE-10*), and 60 minutes after the time point *PROTAMINE-10* (*PROTAMINE-60*).

We monitored heart rate by electrocardiogram, systemic (systolic, diastolic, mean) arterial blood pressures, central venous pressure, pulmonary arterial pressure, and pulmonary capillary wedge pressure before, during, and after CPB. Intermittent thermodilution cardiac outputs were measured in duplicate before and after CPB. The total amount of blood withdrawn was limited to less than 10% of the animal's body weight, and blood remaining in the extracorporeal circuit was reinfused at the end of CPB. At least 6 weeks of recovery for each baboon was allowed before the next experiment.

After CPB the circuit was rinsed with 500 ml normal saline solution followed by 500 ml of 0.1% Triton X-100 (Sigma Chemical Co., St. Louis, Mo.). An aliquot of the Triton X-100 wash was taken for measurement of surface adherent platelet GP III_a antigen. Plasma and Triton X-100 wash samples were frozen at -70° C until analysis.

Hematocrit, WBC count, and platelet count assays were performed on whole blood. Platelets were counted by phase microscopy or by Coulter Counter (model STKR; Coulter Electronics Inc., Hialeah, Fla.), in triplicate. Platelet aggregation in response to ADP was studied by using a Payton aggregometer (model 440; Chrono-Log Inc., Havertown, Pa.). PRP and PPP were prepared from citrated blood (10 ml with 10% by volume of 3.8% citrate) by differential centrifugation at 150 g for 10 minutes and 13,600 g for 5 minutes, respectively. Before aggregation studies, PRP platelet count was adjusted to 150,000/ μ l by dilution with PPP. The concentration of ADP required to produce complete second wave aggregation was measured, with complete second wave aggregation assumed when light transmission was 61% or greater within 5 minutes. Platelet aggregation results are reported as a percentage, normalized to aggregation of the baseline sample.

Bleeding time was measured in duplicate with a blood pressure cuff inflated to 40 mm Hg on the forearm at the same time points as blood samples were drawn (bleeding times were not measured during CPB). The Simplate II (Organon Teknika Corp., Durham, N.C.) lancet was used to create reproducible skin incisions for bleeding time determination.

For plasma β TG analysis, blood was withdrawn into centrifugation tubes containing 10% (by volume) of 3.8% ACD and prostaglandin E₁ solution at 0° C. β TG was measured by radioimmunoassay.¹⁰

For the measurement of platelet GPII_a antigen eluted

Fig. 1. Heart rate, cardiac output, and mean arterial blood pressure before, during, and after CPB when using a membrane or bubble oxygenator. ■, Membrane oxygenator group. ○, Bubble oxygenator group. Values are expressed as mean $\pm$ SEM. *Significant at $p < 0.05$ when using t test with Bonferroni correction for comparison of values within membrane or bubble groups, compared with BASELINE. †Significant at $p < 0.05$ when using Student's unpaired t test for comparison of values between membrane and bubble oxygenator groups. A significant difference was found between membrane and bubble groups for heart rate ($p = 0.0003$) and mean arterial blood pressure ($p = 0.001$) when compared by ANOVA.

in the Triton X-100 wash, the ELISA plates were coated with a 400 nmol/L solution of the disintegrin eristostatine (200 μ mol/well) in 0.05 mol/L carbonate/bicarbonate buffer (pH 9.2) and incubated at 4° C overnight. Eristosta-

tin was prepared as described earlier.¹¹ To block nonreacted surfaces, the plates were incubated for 60 minutes at 37° C with 250 μ l/well PBS/0.5% Tween 20 containing 5% nonfat milk. After being washed (three times) with PBS/Tween, the samples were added into wells in 1:2 dilution in buffer containing 20 mmol/L Tris-HCl (pH 7.4), 150 mmol/L NaCl, 2 mmol/L CaCl_2 , 2 mmol/L MgCl_2 , and 1% bovine serum albumin and incubated for 60 minutes at 37° C. Subsequently, known concentrations of purified GPIIb/IIIa were added to the wells on the same plate to obtain the standard curve. The plate was washed three times with PBS/Tween buffer, and the aliquots of 400 ng of monoclonal antibody AP3 (recognizing GPIIIa) in PBS/Tween buffer containing 3% nonfat milk were added to each well. Plates were incubated for 60 minutes at 37° C, then washed three times with PBS/Tween buffer. After washing, the binding of AP3 was detected by using alkaline phosphatase-conjugated goat anti-mouse immunoglobulin G as has been described previously.¹²

Plasma levels of F_{1,2} (Behring Diagnostics Inc., Westwood, Mass.), FPA (American Bioproducts, Parsippany, N.J.), TAT (Behring Diagnostics Inc.) and d-dimer (American Diagnostica Inc., Greenwich, Conn.) were measured by ELISA with commercial assay kits.¹³⁻¹⁶ PAP levels were measured with a baboon-specific ELISA with urokinase-activated normal baboon plasma.¹⁷

C3b/c levels were assessed by radioimmunoassay with a monoclonal anti-C3-28 antibody and a polyclonal iodine 125-labeled anti-C3c antibody.¹⁸⁻²⁰ C4b/c was measured by a radioimmunoassay similar to that used for the determination of C3b/c, except that a monoclonal anti-C4-1 antibody and a polyclonal ¹²⁵I-labeled anti-C4 antibody were used.^{20,21} Both C3b/c and C4b/c results were expressed as a percentage of the amount of C3b/c or C4b/c present in normal baboon serum aged.^{18,20}

Elastase- α_1 PI levels were measured by two different ELISA methods. The first assay (elastase- α_1 PI polyclonal method) was performed with a commercial kit that used a polyclonal antibody to elastase as a capture antibody and conjugation with an antibody to α_1 PI complexed with alkaline phosphatase (MERCK Immunoassay; Merck Inc., West Point, Pa.). The second assay (elastase- α_1 PI antitrypsin method) used two monoclonal antibodies against elastase complexed with Sephadex beads and against α_1 -antitrypsin labeled by ¹²⁵I.²²

Data points represent the mean $\pm$ SEM of measurements. All measurements of blood elements were corrected for dilution by using the hematocrit. The unpaired Student t test and ANOVA for repeated measures were used for statistical analysis of differences between the bubble and membrane oxygenator groups (SPSS for Windows 6.1; SPSS Inc., Chicago, Ill.), and the paired Student t test with Bonferroni correction was used for analysis of differences within the bubble and membrane oxygenator groups. Differences were considered to be statistically significant at the $p < 0.05$ level. This study was approved by the University of Pennsylvania Committee on Animal Care and Utilization.

Table I. Measured blood and plasma constituents during experiments

Parameter	Baseline	Heparin	Start	End	Protamine-10	Protamine-60	ANOVA (p) (membrane vs bubble)
Hematocrit (%)							
Membrane	34.2 ± 1.6	33.0 ± 1.7	20.0 ± 0.9*	24.6 ± 1.2*	24.7 ± 1.7*	26.3 ± 1.4*	0.918
Bubble	36.3 ± 0.8	35.1 ± 0.7	14.7 ± 0.6*†	20.7 ± 1.2*	28.6 ± 0.5*†	28.3 ± 0.5*	
Platelet count (% of baseline)							
Membrane	100	93.0 ± 2.7	89.1 ± 7.3	97.9 ± 4.3	90.9 ± 8.2	93.5 ± 5.9	0.00001
Bubble	100	82.3 ± 5.6*	86.2 ± 3.0*	73.9 ± 7.4*	58.1 ± 4.6*†	63.0 ± 4.4*†	
ADP aggregation (% of baseline)							
Membrane	100	106 ± 4.5	103.1 ± 4.8	104.1 ± 6.1	78.7 ± 6.8*	87.0 ± 6.1	0.011
Bubble	100	109.0 ± 9.1	67.2 ± 12.5*†	81.3 ± 8.8	68.2 ± 7.5*	75.2 ± 4.8*†	
βTG (ng/ml)							
Membrane	—	35.3 ± 8.6	39.4 ± 16.4	40.8 ± 8.9	41.7 ± 10.4	15.1 ± 4.1	0.000001
Bubble	—	28.1 ± 8.9	108.3 ± 19.2*†	189.1 ± 8.8*†	43.4 ± 7.1	8.9 ± 3.4	
Bleeding times (minutes)							
Membrane	3.5 ± 0.1	5.7 ± 0.5*			8.3 ± 0.3*	7.5 ± 0.4*	0.082
Bubble	3.6 ± 0.1	5.7 ± 0.5*			13.0 ± 1.4*†	10.7 ± 1.4*	
FPA (ng/ml)							
Membrane	—	6.7 ± 1.9	2.7 ± 0.5*	4.0 ± 1.4	4.6 ± 1.2	4.0 ± 0.9	0.444
Bubble	—	4.8 ± 0.8	5.7 ± 1.8	8.2 ± 1.1*	3.9 ± 0.4	2.2 ± 0.2	
F _{1,2} (nmol/L)							
Membrane	1.24 ± 0.15	1.26 ± 0.10	1.05 ± 0.06	1.20 ± 0.16	1.15 ± 0.10	1.37 ± 0.12	0.00001
Bubble	1.33 ± 0.12	1.46 ± 0.13	1.63 ± 0.26†	3.33 ± 0.22*†	2.26 ± 0.14*†	2.02 ± 0.14*†	
TAT (μg/L)							
Membrane	14.0 ± 2.2	17.9 ± 4.9	8.9 ± 2.1	16.5 ± 5.4	11.5 ± 2.1	23.5 ± 4.4	0.026
Bubble	19.2 ± 3.9	21.5 ± 3.9	14.2 ± 1.5†	87.0 ± 11.0*†	26.9 ± 4.6†	24.5 ± 4.8	
D-dimer (mg/L)							
Membrane	0.28 ± 0.08	0.29 ± 0.06	0.35 ± 0.06	0.33 ± 0.06	0.30 ± 0.06	0.30 ± 0.06	0.204
Bubble	0.23 ± 0.05	0.24 ± 0.05	0.30 ± 0.05	0.32 ± 0.04	0.28 ± 0.03	0.29 ± 0.03	
PAP (%)							
Membrane	—	0.015 ± 0.005	0.031 ± 0.005	0.080 ± 0.044	0.034 ± 0.011	0.027 ± 0.012	0.45
Bubble	—	0.027 ± 0.006	0.117 ± 0.055	0.041 ± 0.009	0.019 ± 0.007	0.031 ± 0.011	
C3b/c (% of NBA)							
Membrane	—	1.8 ± 0.8	7.0 ± 1.2*	10.9 ± 2.4*	9.0 ± 1.3*	6.9 ± 1.2*	0.000001
Bubble	—	1.4 ± 0.3	6.7 ± 1.5	31.6 ± 4.2*†	24.0 ± 3.5*†	18.3 ± 2.5*†	
C4b/c (% of NBA)							
Membrane	—	2.9 ± 0.9	2.3 ± 0.9	4.0 ± 1.2	8.3 ± 2.0*	6.3 ± 1.4	0.714
Bubble	—	3.1 ± 1.0	3.0 ± 0.4	5.5 ± 0.9	8.1 ± 1.4*	6.0 ± 0.8*	
WBC (% of baseline)							
Membrane	100	112.4 ± 6.0	85.6 ± 13.4	132.6 ± 14.4	108.0 ± 13.2	180.6 ± 15.2*	0.097
Bubble	100	118.8 ± 6.2	103.4 ± 12.5	195.1 ± 44.2*	141.8 ± 28.2	212.0 ± 39.3*	
Elastase-α ₁ PI (polyclonal method, μg/L)							
Membrane	—	25.5 ± 4.3	40.0 ± 4.1	36.5 ± 7.7	55.9 ± 20.8	32.4 ± 6.2	0.118
Bubble	—	26.8 ± 3.8	55.6 ± 9.4*	63.0 ± 9.2*	46.7 ± 7.0	43.9 ± 5.7	
Elastase-α ₁ PI (monoclonal method, μg/L)							
Membrane	—	45.2 ± 4.6	86.7 ± 14.8	93.5 ± 16.6	118.5 ± 11.5*	142.0 ± 31.9*	0.0001
Bubble	—	58.8 ± 6.2	95.2 ± 14.6	275.7 ± 27.7*†	241.4 ± 13.1*†	218.3 ± 12.3*	

Values are expressed as mean ± SEM. ANOVA computed for CPB when using membrane oxygenator versus bubble oxygenator (statistical significance identified at $p < 0.05$, for p value of F statistic by ANOVA for repeated measures).

F_{1,2}, plasma prothrombin fragment F_{1,2}; D-dimer, fibrin fragment, D-dimer; C3b/c, complement fragment C3b/c; C4b/c, complement fragment C4b/c; NBA, normal baboon serum aged.

*Significant difference ($p < 0.05$, t test with Bonferroni correction) within membrane or bubble group as compared with baseline (or heparin).

†Significant difference ($p < 0.05$, by unpaired Student's t test) between means of membrane and bubble oxygenator groups.

Fig. 2. Hematocrit-corrected platelet counts before, during, and after CPB when using a membrane or bubble oxygenator. Platelet counts are standardized as a percentage of platelet count at BASELINE for each time point of the group. Symbols as in Fig. 1.

RESULTS

Hemodynamic measurements of heart rate, cardiac output, and mean arterial blood pressure are depicted in Fig. 1. In the membrane oxygenator group, heart rate and mean arterial blood pressure were lower for nearly all time points; a significant difference between the two groups was determined by ANOVA for heart rate ($p = 0.0003$) and mean arterial blood pressure ($p = 0.001$) but not for cardiac output. After protamine, heart rate (in both groups) and mean arterial blood pressure (in the bubble oxygenator group) decreased significantly. Although cardiac output decreased after CPB, the change was not significant in either group.

Changes in measured blood and plasma constituents during the experiments are listed in Table I. The circuit priming volume diluted hematocrit during and after CPB. All values in Table I (except platelet aggregation to ADP) were corrected for hemodilution. In the bubble oxygenator group, platelet counts decreased to a nadir of 58.1% of the initial level (time point PROTAMINE-10), whereas the membrane oxygenator group showed no significant change during and after CPB (Fig. 2). When platelet counts of both groups were compared by

Fig. 3. A, Platelet aggregation to ADP before, during, and after CPB when using a membrane or bubble oxygenator. Data points are standardized as a percentage of BASELINE for each time point of the group. B, β TG before, during, and after CPB when using a membrane or bubble oxygenator. Symbols as in Fig. 1.

ANOVA, the difference was statistically significant ($p = 0.00001$).

In the bubble oxygenator group, platelet aggregation to ADP was significantly attenuated soon after CPB was started and remained more attenuated

Fig. 4. Template bleeding times before, during, and after CPB when using a membrane or bubble oxygenator. White bars indicate the membrane oxygenator group, black bars indicate the bubble oxygenator group. Other symbols as in Fig. 1.

than the membrane oxygenator group (Fig. 3, A). β TG release increased to a level six times that of the initial HEPARIN time point with the bubble oxygenator at the conclusion of CPB (END); no significant change was found with the membrane oxygenator (Fig. 3, B). Bleeding time was significantly prolonged after systemic heparinization in both groups, and it remained more prolonged in the bubble oxygenator group after protamine administration (PROTAMINE-10) (Fig. 4). There was no significant difference between the two oxygenator systems by ANOVA. The average amount of surface GPIII_a antigen eluted in the Triton X-100 wash was higher in the bubble oxygenator group (3.22 ± 0.46 ng/ μ l) as compared with that in the membrane oxygenator group (2.33 ± 0.11 ng/ μ l). This difference was not significant (Student's unpaired *t* test, *p* = 0.188).

Plasma FPA levels showed significant elevation only at the conclusion of CPB (END) in the bubble oxygenator group. No significant elevations were noted in the membrane group. A significant elevation in $F_{1,2}$ and TAT levels was noted in the bubble oxygenator group at the conclusion of CPB (END), and $F_{1,2}$ remained significantly higher after prota-

Fig. 5. A, $F_{1,2}$ before, during, and after CPB when using a membrane or bubble oxygenator. **B,** TAT before, during, and after CPB when using a membrane or bubble oxygenator. Symbols as in Fig. 1.

mine administration. $F_{1,2}$ (*p* = 0.00001) and TAT (*p* = 0.026) levels were consistently higher in the bubble group. Both markers did not change throughout the experiment with the membrane oxygenator (Fig. 5, A and B). d-dimer and PAP, which

Fig. 6. C3b/c before, during, and after CPB when using a membrane or bubble oxygenator. Symbols as in Fig. 1.

are markers of fibrinolysis, showed no significant change throughout the duration of the experiment with either oxygenator type.

C3b/c level increased significantly during and after CPB in both groups and was greatest in the bubble oxygenator group at the conclusion of CPB (END) (Fig. 6). C4b/c level was elevated significantly in both membrane and bubble oxygenator groups only after protamine administration.

White blood cell count increased significantly during and after CPB in both groups. Elastase- α_1 PI (polyclonal method) increased significantly with the bubble oxygenator at the conclusion of CPB (END). Elastase- α_1 PI (monoclonal method) increased significantly during and after CPB in the bubble oxygenator groups (Fig. 7). There was a significant difference between the two oxygenator systems when compared by ANOVA.

DISCUSSION

Many enzymes and chemicals produced by activation of blood constituents are procoagulant or vasoactive and mediate the inflammatory, embolic, and bleeding complications associated with CPB and open heart surgery.⁶ CPB is not possible without heparin, but heparin blocks thrombin formation at the end of the coagulation cascade instead of at the beginning. Contact with the biomaterials of the per-

Fig. 7. Elastase- α_1 PI complex (monoclonal method) before, during, and after CPB when using a membrane or bubble oxygenator. Symbols as in Fig. 1.

fusion circuit and the wound activate both the intrinsic and extrinsic coagulation pathways,²³ contact proteins and fibrinolytic plasma proteins, complement, platelets, neutrophils, endothelial cells and monocytes² to produce chemicals that cause vascular smooth muscle and endothelial cells to contract or relax and to depress cardiac myocyte contractility.²⁴ Because all non-endothelial cell surfaces activate blood elements, selective inhibition of these reactions is an attractive strategy to control blood-mediated complications of CPB.²

Scores of antibodies against human blood proteases and cellular receptors have been developed, but few cross-react with blood proteins and cells of large, non-primate laboratory animals. Only a few antibodies are available against non-primate blood proteins.²⁵⁻²⁹ Consequently, promising inhibitors of target reactions in vitro are not easily evaluated in vivo. We developed this baboon model to provide an efficient animal model for evaluating specific inhibitors of blood reactions that produce the inflammatory, thrombotic, and bleeding complications associated with CPB.

Within the perfusion circuit, the oxygenator contains the largest surface areas in contact with blood, and it is the site of maximal blood activation. Bubble oxygenators, which expose blood constituents to gas surfaces of numerous bubbles, more strongly acti-

vate blood elements than do microporous membrane oxygenators, but the mechanism of activation is similar and differs only in magnitude.² Baboon platelets are less sensitive to activation than are human platelets,³⁰ and other blood proteins and cells may also have higher thresholds of stimulation. In both this and a previous study,³⁰ perfusion of baboon blood through a membrane oxygenator system did not significantly activate platelets or stimulate neutrophil release of elastase. In this study the membrane oxygenator system did not significantly increase fibrinopeptide A, F_{1,2}, TAT, or C3b/c during 1 hour of perfusion. Although silicon rubber membrane oxygenators did not activate these complexes and proteases in baboons, membrane oxygenators activate all of these blood elements in patients.² With a bubble oxygenator system, all of these markers and those related to platelets and neutrophils significantly increase during CPB.

Neither D-dimer nor plasmin-antiplasmin complex increased during perfusion with either oxygenator in the baboon. Fibrinolysis does not occur in this baboon model, whereas both tissue plasminogen activator and D-dimer increase dramatically in patients during open heart surgery.³¹ Absence of a large wound in this model probably best explains these findings.^{23,32}

This study establishes the baboon perfused with the bubble oxygenator as a robust, reusable, cost-effective animal model for evaluation of inhibitors of platelet, neutrophil, and complement activation and of thrombin formation during CPB. The model has been used successfully to evaluate a platelet inhibitor.³³ Animals have been perfused up to 10 times with 6-week intervals between perfusions. At present the model does not provide a means to study inhibitors of fibrinolysis. Nevertheless, quantitative measurements of most of the important markers of the inflammatory, thrombotic, and bleeding complications associated with CPB provide a means to critically evaluate potential inhibitors of these reactions and speed progression of promising drugs to phase I trials.

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