

Original Article

Safety of Heparin-Coated Circuits in Primates During Deep Hypothermic Cardiopulmonary Bypass

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ABSTRACT

The purpose of this study is to evaluate the biologic impact of heparin-coated circuits without systemic heparinization during deep hypothermia. Baboons (n=6) were placed on a heparin-coated pediatric closed-circuit cardiopulmonary bypass (CPB) system and cooled to 18°C. A control group (n=7) underwent similar protocol with a non heparin-coated circuit and received systemic heparin. Either low flow at 0.5 L/min/m² (n=8; 4 in each group) or circulatory arrest (n=5; 2 in experimental group and 3 in control group) was used during deep hypothermia. Samples for complete blood count (CBC), hepatic and renal function tests, activated clotting time (ACT) and thrombelastogram (TEG) were obtained before, during, and after bypass. Cerebral blood flow was measured using Xenon-133 and autopsies were performed to assess end-organ damage.

The ACT returned to baseline in both groups, and renal and hepatic function were within normal limits. There was no significant difference between the TEG values between the groups post bypass. Fibrin split products were absent and fibrinogen levels were normal in both groups following bypass. Cerebral blood flows were equivalent in both groups before and after bypass, although in the heparin-coated group cerebral blood flows were significantly higher during CPB. There were no brain histologic changes in the heparin-coated group and one focal cortical infarct in the control group.

This study suggests that hypothermia induced a state of anticoagulation that did not result in thrombus formation or end organ dysfunction during CPB with a heparin-coated circuit. Safe use of these circuits without any systemic anticoagulation should be limited to conditions where the patient's coagulation time is prolonged such as hypothermia.

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INTRODUCTION

Heparin coated cardiopulmonary bypass circuits have generally been used with some degree of systemic heparinization (1). This study was undertaken to evaluate the effects of a heparin-coated CPB circuit on a non-heparinized baboon and to uncover the safety issues related to its clinical use. Several authors have compared heparin-coated circuits in animals without exogenous heparin, but conclusions in non-primate species may not be valid in humans because of differences in hemostatic mechanisms (1-3). In humans, a stepwise reduction in heparinization with heparin-coated circuits has been carried out, maintaining ACT values of >180 seconds (4). Attempts made to further decrease the ACT during experimentation will decide the ACT range used by clinicians (5, 6).

One major limitation of present clinical techniques using heparin-coated systems with reduced ACT is the inability to return the cardiotomy suction blood to the extracorporeal circuit without washing it first in a cell washer device. The use of a cell washer instead of the cardiotomy suction promotes the loss of plasma proteins. This may not be significant in an uncomplicated coronary artery bypass procedure, but is unacceptable for a difficult case with significant obligatory blood loss into the operative field. Even if the cardiotomy reservoir is heparin-coated, coagulation within the reservoir may occur due to stagnation of blood in the device. At the time this study was completed, heparin-coated cardiotomy reservoirs were not available.

The use of hypothermia and hemodilution have long been mainstays of the perfusion armamentarium. Their effects on prolonging anticoagulation have been recognized, for instance the ACT value increases with the onset of bypass due to hemodilution and cooling, with a concurrent decrease in the heparin concentration. Additional heparin is often not administered until the ACT begins to decrease during the rewarming phase of the operation.

We were interested in combining these two modalities to test the feasibility of using heparin-coated bypass systems without systemic heparinization during low flow CPB and circulatory arrest. These two states of CPB could be defined as periods of blood stagnation, which places the animal at a higher risk of thrombus formation (7).

Potential clinical applications for heparin-coated CPB without systemic heparinization include thoracic aortic aneurysm repair, Type B aortic dissection repair, and clipping of cerebral aneurysms. These procedures routinely incorporate deep hypothermia and circulatory arrest. The ability to conduct these surgical procedures without systemic heparinization offers the possibility of reducing complications related to blood loss.

MATERIALS AND METHODS

All animals used in the study received humane care in compliance with the "Guide for the Care and Use of Laboratory

Animals" published by the National Institutes of Health (NIH Publication No.85-23 revised 1985). The animals were randomly divided into an experimental group (heparin-coated) and a control group (non-heparin coated). Due to the similarities between non-human and human primates in their hemostatic responses to contact with extracorporeal surfaces, baboons (5-15 kg) were chosen for this study. In the experimental group, baboons (n=6) were placed on a heparin-coated pediatric closed-circuit bypass without any systemic heparin. In the control group (n=7), a non heparin-coated system was used with a systemic dose (300 units per kilogram) of porcine intestinal mucosa heparin. In the control group the ACT was maintained greater than 480 seconds. Heparin reversal in the control group following bypass was accomplished with a 1.3:1 ratio of protamine:heparin. In the experimental group, neither heparin or protamine were administered. Both groups were placed on bypass following placement of a catheter in the right common carotid artery for injection of Xenon-133, and a radial artery catheter for arterial monitoring and sampling.

The bypass circuit consisted of a Medtronic-Minimax^a oxygenator, a BP-50 Biopump^b and 1/4" tubing. The system was primed with 175 ml of Normosol-R^c and 175 ml of 6% Hespand^d. All surgical blood loss was collected and processed with a Cell Saver 1^e and then added to the circuit. Blood flow was maintained at 2.4 L/min/m² during cooling and rewarming; low flow was at 0.5L/min/m². Circulatory arrest or low flow was maintained for 1 hour at a rectal and tympanic temperature of 18°C. Crystalloid cardioplegia, 10 ml/kg, was administered following aortic crossclamping. Venous cannulation was either single atrial for circulatory arrest or bicaval for low flow. The aorta was transected and reanastomosed to simulate a surgical procedure. To prevent blood stagnation during periods of circulatory arrest and after termination of bypass, blood was continuously recirculated through an arterial-venous bridge just proximal to the cannulas. During circulatory arrest, the arterial and venous cannulas were removed from the animal and flushed with saline to prevent thrombus formation in them. Following decannulation, volume remaining in the circuit was drained by gravity into CPD transfer bags for reinfusion to the animal via a peripheral intravenous line.

Laboratory studies were conducted pre, during and post bypass and these included CBC, liver and renal function studies, ACT, thrombelastogram (TEG), and blood gases. Intraoperative cerebral blood flow studies were performed using Xenon-133. The radioactive xenon was injected into the right internal carotid artery with the external carotid artery occluded. The initial slope method was used and all values were corrected for hematocrit and temperature. Cerebral blood flow was reported in ml/min/100 gm of brain tissue. An autopsy was performed approximately one

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	HEPARIN-COATED		NON-COATED		p - VALUE * < .05
	mean	S.D.	mean	S.D.	
weight(kg)	12.75	3.5	9.57	2.3	.13
BSA (m ²)	.56	.08	.47	.1	.13
CBP time(min)	177.6	26.1	173.1	.76	.89
cool time(min)	37.5	17.0	34	11.2	.73
warm time(min)	54.6	14.8	53.4	17.9	.90
low flow time(min)	67.25	3.4	54.4	22.8	.30
circ. arrest(min)	61.5	2.12	67.6	2.9	.96

Table 1

Heparin-coated vs. non-coated mean and standard deviation comparison in both groups with respect to weight, BSA (m²), CBP time (min), cooling time (min), warming time (min), low flow time (min), and circulatory arrest time (min).

week following the procedure to determine the presence of end-organ damage by histopathological examination.

Results from the experimental and control groups were compared using the Students t-test to test for significance (p<.05)-

RESULTS

Both groups were subjected to an equivalent time on bypass and either circulatory arrest or low flow. Cooling and

rewarming times were not significantly different between the two groups (Table 1). Although the ACT increased during hypothermic bypass in the heparin-coated group (Figure 1), it declined during rewarming and there was no significant difference post bypass. Coagulation returned to normal in both groups as evidenced by the normal post bypass TEG (Table 2) and ACT. The TEG did not reveal any evidence of fibrinolysis. The A60 was normal in both groups. This lack of fibrinolysis was confirmed by measurement of fibrin split products (negative in both groups). There was no significant difference between the two groups in post bypass platelet counts or fibrinogen (Figure 2 and 3).

Surgical bleeding in the heparin-coated group was noticeably less than in the experimental group and it was necessary to use a cardiomy suction and reservoir with the control group in order to return blood rapidly to the perfusion circuit. In the experimental group, the Cell Saver was capable of handling the minimal blood loss.

Renal and hepatic function was normal in both groups (Tables 3 and 4). Histopathological examinations of the brain in both groups were normal with the exception of one cortical infarct in the heparinized control group. Normal cerebral architecture was preserved in all the other specimens examined from both groups.

Cerebral blood flow was measured with Xenon-133 using a Nova Cerebrograph-10af. There was no significant difference

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Mean ACT Values

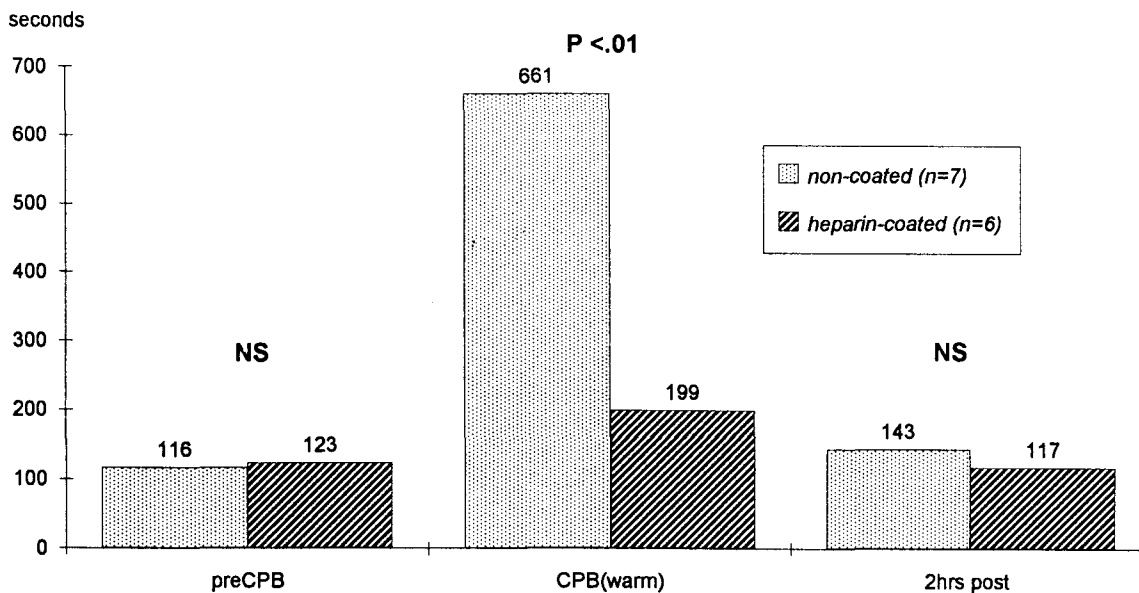
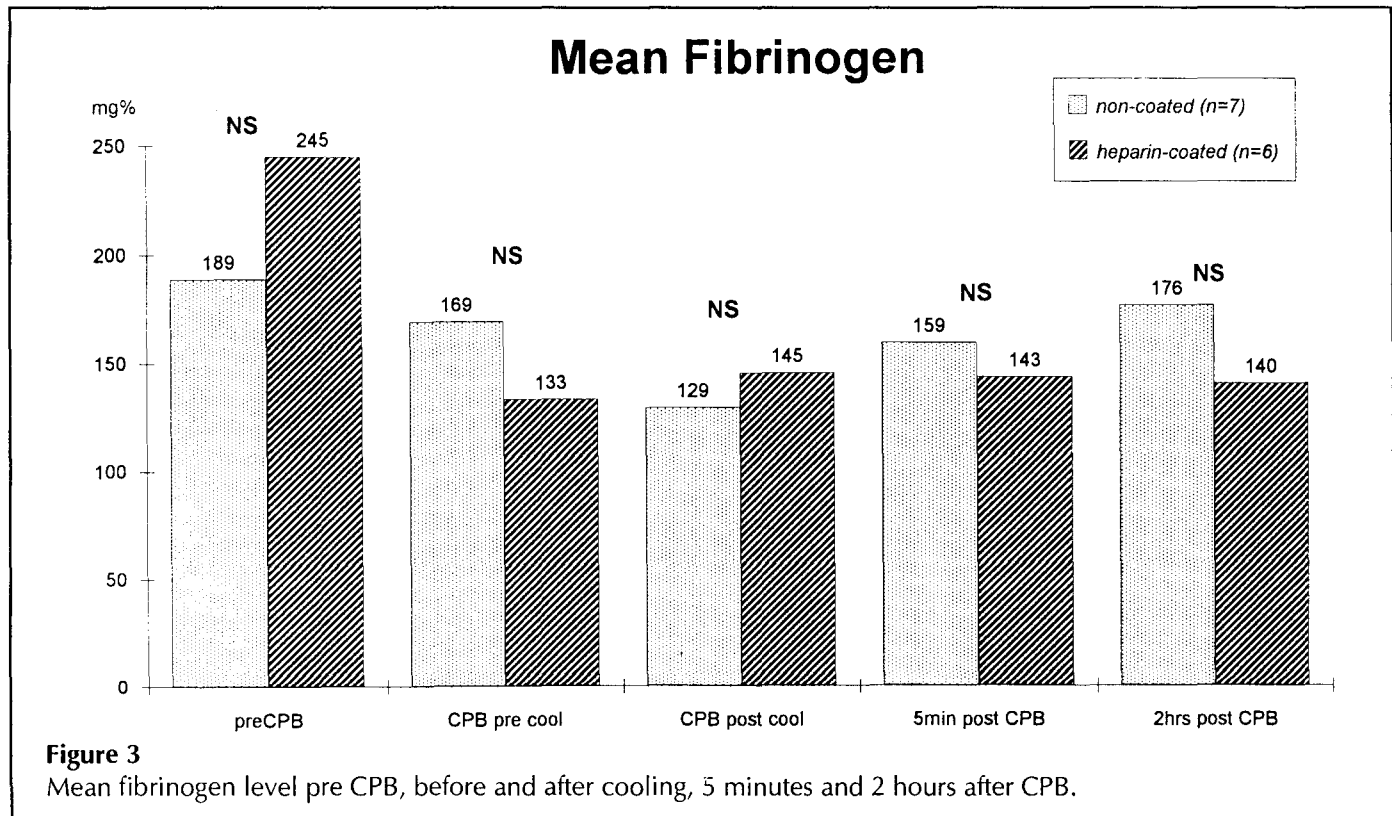
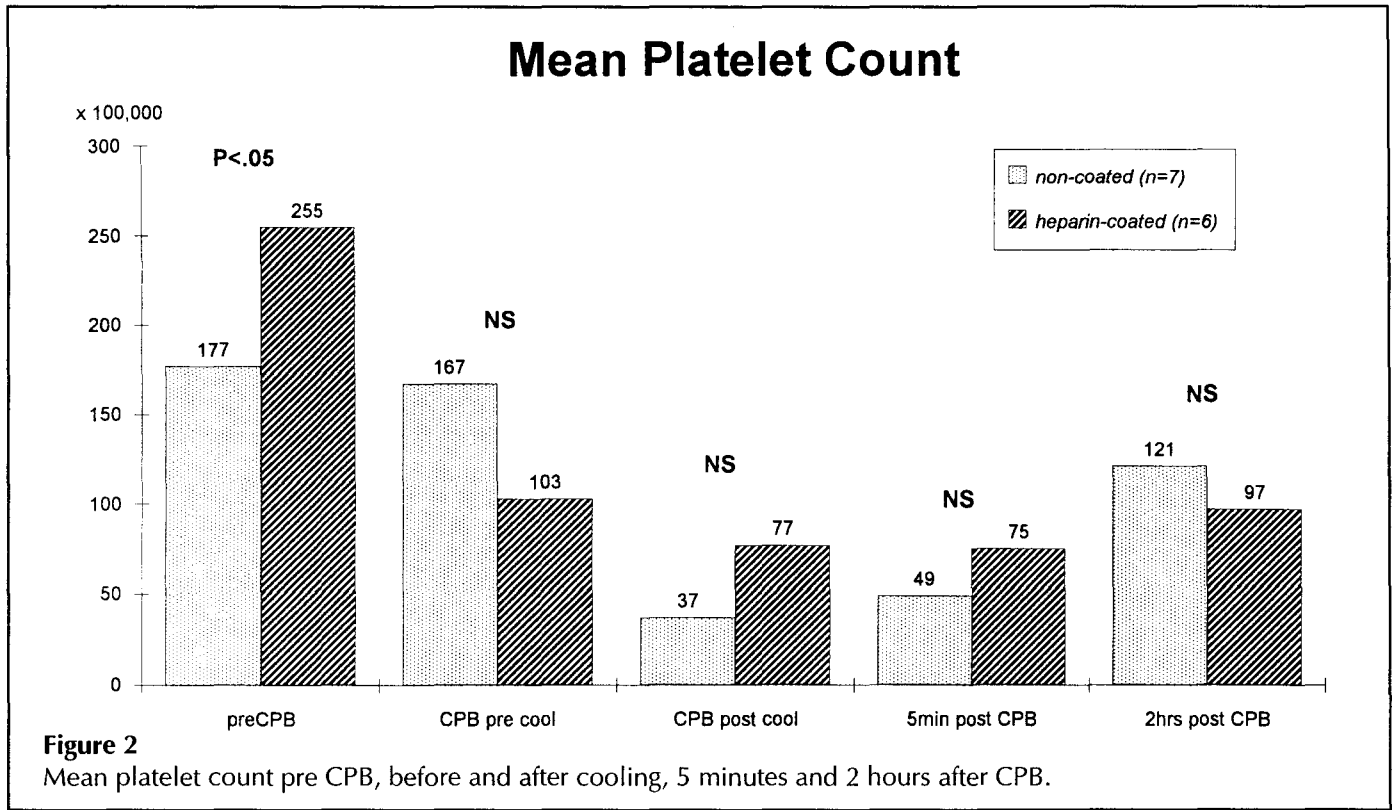


Figure 1

Mean ACT values prebypass, during bypass (37°C), and post bypass in heparin-coated and non-coated groups.



	HEPARIN-COATED		NON-COATED		p - Value * < .05
	mean	S.D.	mean	S.D.	
R prebypass (min)	8.16	2.04	7.6	3.8	.81
R postbypass (min)	25.8	39.17	13.2	.1	.47
K prebypass (min)	3.5	1.09	6.8	4.0	.25
K post bypass (min)	19.1	21.2	10.3	6.3	.45
alpha angle (degrees) prebypass	62	7.5	44.8	14.9	.06 *
alpha angle (degrees) postbypass	31.8	18.6	37.7	14.8	.62
MA prebypass (mm)	75	6.7	65.2	5.5	.04 *
MA post bypass (mm)	58.3	11.1	47.5	13.6	.23
A60 pre bypass (mm)	74	6.8	62.8	2.5	.008 *
A60 post bypass (mm)	55.5	9.9	47.5	13.9	.37

Table 2
TEG analysis in heparin-coated vs non-coated during pre bypass and post bypass periods. The R, K, alpha angle, MA and A₆₀.

in cerebral blood flow between the two groups in the pre and post bypass periods. However, during bypass there was a significant difference (p<.05) in cerebral blood flow between the two groups (Table 5). Following termination of bypass, blood was recirculated through a shunt line until the animal was hemodynamically

stable. The remaining blood in the circuit was drained into CPD transfer bags for reinfusion. Whenever the final ACT on bypass was below baseline (in the heparin-coated group), difficulty in draining blood from the circuit was encountered due to rapid thrombus formation soon after the pump was turned off. In those cases some blood remained in the extracorporeal circuit and could not be returned to the animal following bypass. This is reflected in the significantly lower hematocrit in the heparin-coated group 2 hours after bypass (Figure 4).

DISCUSSION

Extracorporeal circulation requires systemic heparinization in order to prevent circuit thrombosis (1). The development of a biocompatible circuit is highly desirable since it could eliminate the risks associated with systemic anticoagulation. Heparin coating of the extracorporeal circuit is one potential technology (6). The use of deep hypothermia and hemodilution extends the coagulation cascade and heparin-coated circuits can be used without fear of subclinical coagulation (8).

A characteristic thrombelastograph (TEG) tracing represents the shear elasticity over time of a blood clot as it develops and its eventual lysis. The A₆₀ is the amplitude of the tracing measured 60 minutes after the mean amplitude and signifies clot retraction or lysis (9). In our study, based upon TEG, ACT, and fibrin split product analyses, coagulation returned to normal in both groups suggesting hypothermia plus heparin-coated circuits requires no systemic heparinization.

Mean Hematocrit

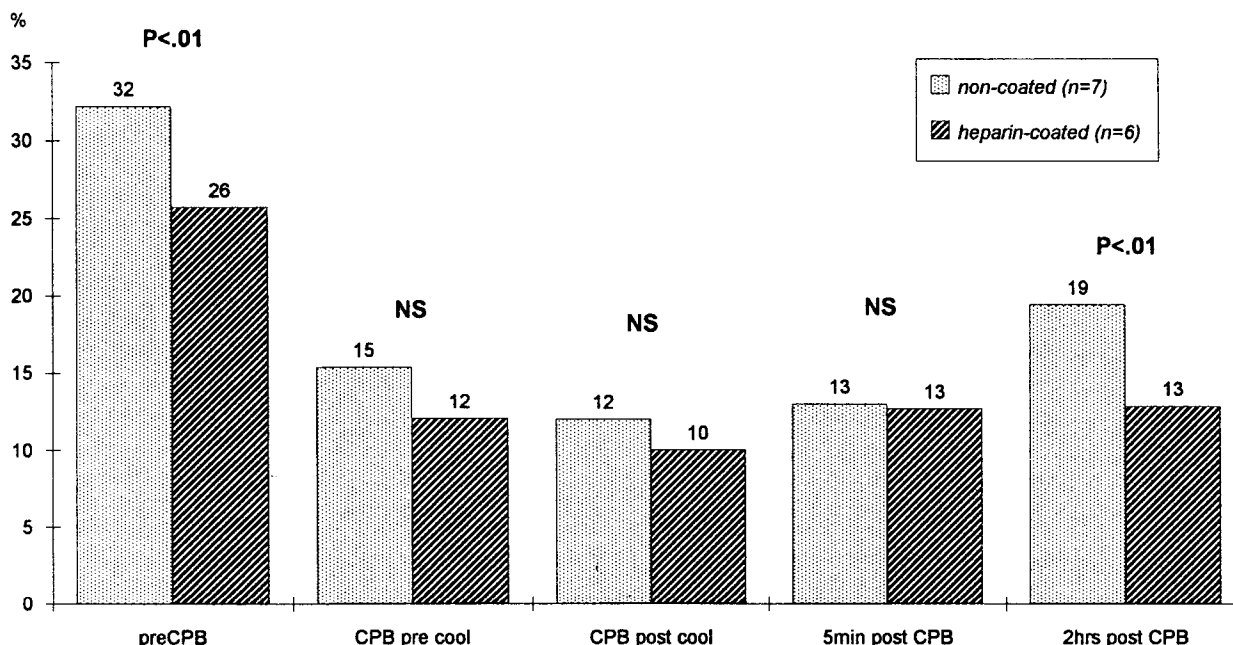


Figure 4
Mean hematocrit pre CPB, before and after cooling, 5 minutes and 2 hours after CPB.

	HEPARIN-COATED		NON-COATED		p - value * < .05
	mean	S.D.	mean	S.D.	
BUN prebypass	15.1	4.1	19.7	3.6	.08 *
BUN postbypass	12.8	3.3	16.1	4.0	.19
creatinine prebypass	.76	.13	.75	.18	.92
creatinine postbypass	.71	.16	1.18	1.07	.32

Table 3
Renal function in heparin-coated vs. non-coated groups.

	HEPARIN-COATED		NON-COATED		p - value * < .05
	mean	S.D.	mean	S.D.	
LDH prebypass	562	191	601	127	.71
LDH postbypass	465	160	902	532	.12
SGOT prebypass	45.3	.6	60.7	12	.03 *
SGOT postbypass	66	21.7	134	48	.014 *
SGPT prebypass	30.8	12.4	29.1	11	.81
SGPT postbypass	26.1	10	68.6	74	.19

Table 4
Hepatic function in heparin-coated vs. non-coated measuring LDH, SGOT, and SGPT pre and post CPB.

	pre	pre	during	during	post	post
	mean	SD	mean	SD	mean	SD
control group	26.88	5.85	28	5.91	24.4	8.02
experimental group	28.1	7.12	50.9	17.3	40.18	0.59

p = .78 p = .02* p = .14

Table 5
Cerebral blood flow measurement prebypass, during bypass, and postbypass in heparin-coated and non-coated groups.

The significant difference in cerebral blood flow during bypass may be related to differences in hematocrit values on bypass in the experimental group. Auto-regulation of cerebral blood flow to compensate for reduced oxygen content and maintain oxygen delivery may account for the blood flow differences. Hypoperfusion of the brain and micro/macroembolism during cardiopulmonary bypass are common and considered to cause deterioration in neuropsychological function. Heparin-coated circuits have been shown to be more biocompatible with regard to neurological impairment than conventional circuits.

The lack of decrease in cerebral blood flow following circulatory arrest or low flow in the experimental group implies maintenance of cerebral vascular integrity. Brain histological changes as well as renal and hepatic function proves preservation of organs. The cortical infarct in one of the control animals may be related to a low flow period of 68 minutes and an additional period of circulatory arrest required to control bleeding during this experiment.

At normothermia, sudden interruption of blood flow could become a life threatening event due to activation of the coagulation process(6,10). The importance of blood flow in preventing thrombus formation was dramatically shown during this study while recovering blood from the perfusion circuit following bypass. Instead of draining blood from the circuit into CPD bags, an alternate procedure might be to add heparin to the perfusion circuit and then wash the cells with a Cell Saver. Problems with recovery of blood from the circuit emphasize the requirement for some systemic anticoagulation when using a heparin-coated extracorporeal circuit at normothermia. If blood flow interruption (such as a pump failure) occurs in the patient with an intact coagulation system, immediate and life threatening clot formation may develop.

This study suggests that during hypothermic induced anticoagulation, thrombus formation and end-organ dysfunction do not occur with a heparin-coated CPB circuit. Safe use of these circuits without any systemic anticoagulation should be limited to conditions where the patient's coagulation time is prolonged such as hypothermia.

REFERENCES

1. Toomasian JM, Hsu LC, Hirschl LB, Heiss KF, Hultquist KA, Bartlett RH. Evaluation of Duraflo II heparin coating in prolonged extracorporeal membrane oxygenation. *Trans Am Soc Artif Intern Organs*. 1988;34:410-414.
2. von Segesser LK, Turina MI. Cardiopulmonary bypass without systemic heparinization. *J Thorac Cardiovasc Surg*. 1989;98:386-396.
3. Matsuwaka R, Matsuda H, Kaneko, et al. Experimental evaluation of a heparin coated ECMO system simplified with a centrifugal pump. *Trans Am Soc Artif Intern Organs*. 1990;36:473-475.
4. von Segesser LK, Weiss BM, Garcia E, Turina MI. Clinical application of heparin-coated perfusion equipment with special emphasis on patients refusing homologous transfusions. *Perfusion*. 1991;6:227-233.
5. Gu YJ, van Oeveren W, van der Kamp K, Akkerman C, Boonstra PW, Wildevuur, CHR. Heparin-coating of extracorporeal circuits reduces thrombin formation in patients undergoing cardiopulmonary bypass. *Perfusion* 1991;6:221-225.
6. Hsu LC, Principles of heparin-coating techniques. *Perfusion*. 1991; 6:209-219.
7. Borowiec J, Thelin S, Bagge L, Heparin coated cardiopul-

- monary bypass circuits in coronary artery surgery - a clinical study. *AmSECT Proceedings*; 1990;86-90.
8. von Segesser LK, Weiss BM, Garcia E, von Fetten E, Turina MI. Coagulation patterns in perfusion with low systemic heparinization. *AmSECT Proceedings*. 1992;23:80-85.
 9. Bennett J, Hill J, Long B, et al. Biocompatible Circuits: An adjunct to non-cardiac extracorporeal cardiopulmonary support. *J Extra-Corp Technol*. 1992;24:6-10.
 10. Spiess BD, Tuman KJ, McCarthy RJ, et al. Thrombelastography as an indicator of post-cardiopulmonary bypass coagulopathies. *J Clin Monitor*. 1987;3:25-30.
 11. von Segesser LK, Garcia E, Turina MI. Perfusion without systemic heparinization for rewarming in accidental hypothermia. *Ann Thorac Surg*. 1991;52:560-561.
 12. Hoxmeier K, Goddard D, Morton R, Chapin J, Landers D, Fleming W. Thrombelastography (TEG): a graphic profile analyzing coagulation after cardiopulmonary bypass. *AmSECT Proceedings*. 1992;23:152-3.