Complement, leukocytes, and leukocyte elastase in full-term neonates undergoing cardiac operation

In 13 neonates undergoing cardiac operations for congenital cardiac defects, complement, leukocytes, and leukocyte elastase were studied during and after cardiopulmonary bypass. All but two neonates received prostaglandin E_1 before the operation. The C3d/C3 ratio rose significantly during cardiopulmonary bypass from 0.86 ± 0.55 to 1.40 ± 0.56 (mean $\pm$ standard deviation; $p < 0.0001$). Abnormally elevated C5a levels ($18.6 \pm 7.3 \mu\text{g/L}$) were measured at the end of cardiopulmonary bypass. C4 was not overtly consumed during the procedure. Leukocytes fell from a preoperative value of $10.06 \pm 3.15 \times 10^9/\text{L}$ to $3.21 \pm 0.64 \times 10^9/\text{L}$ after beginning of cardiopulmonary bypass ($p < 0.0001$) and rose at the end of the procedure from $2.33 \pm 0.67 \times 10^9/\text{L}$ to $7.19 \pm 1.84 \times 10^9/\text{L}$, after protamine administration ($p < 0.0001$). Neutrophils fell from a preoperative value of $5.14 \pm 1.18 \times 10^9/\text{L}$ to $1.46 \pm 0.35 \times 10^9/\text{L}$ after beginning of cardiopulmonary bypass and rose at the end of extracorporeal circulation from $1.00 \pm 0.31 \times 10^9/\text{L}$ to $4.10 \pm 1.18 \times 10^9/\text{L}$, after protamine administration ($p < 0.005$). Elastase release occurred in all neonates during cardiopulmonary bypass and averaged $331.5 \pm 175.7 \mu\text{g/L}$. Complement activation and leukocyte stimulation did not correlate with postoperative complications or outcome. This study demonstrates complement activation and leukocyte stimulation in neonates undergoing cardiac operation. (J THORAC CARDIOVASC SURG 1994;108:29-36)

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Cardiopulmonary bypass (CPB) in adults and children may be responsible for several postoperative complications that are basically due to activated complement resulting in anaphylatoxin liberation and leukocyte stimulation.^{1,2} Because full-term neonates are reported to have subnormal activity of the alternative pathway of the complement system compared with that of adults^{3,4} and immaturity of complement receptors on neutrophils,⁵ we investigated their response to CPB with regard to com-

plement activation and leukocyte stimulation and considered a possible correlation between inflammatory response and postoperative outcome.

Patients and methods

The study included 13 full-term consecutive neonates (10 boys, 3 girls) aged 4 to 8 days (median 6 days) undergoing cardiac operations for congenital heart disease in our institution. Diagnosis, age, and weight at time of operation are shown in Table I. All but two neonates were treated with prostaglandin E_1 (PGE_1) before operation. Cardiac catheterization with angiocardiology was performed in all patients 1 to 7 days (median 3 days) before the operation. The lungs of eight neonates were mechanically ventilated before the operation. Routine laboratory investigations were in normal range before the operation in all patients. Materno-fetal infection was excluded in all cases.

Anesthesia and antibiotic regimen. General anesthesia and myoplegia were performed with diazepam, fentanyl, and pancuronium. As we do for infants and children, we administered dexamethasone to all neonates on the first postoperative day (10 mg/m^2 per day, administration was begun before sternotomy) and cefotiam hydrochloride (75 mg/kg per day). PGE_1 administration was continued until CPB was begun.

CPB. The extracorporeal circuit was identical for the 13

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Table I. Clinical and perioperative data

Patient No.	Age (days)	Weight (gm)	Diagnosis	Operative procedure	CPB (min)	CCA (min)	AXT (min)	Complications	Outcome
1	6	3895	HypoRV, critical PS	CoPV + RVOT enlargement	49	40	45	Hypoxia	Died
2	4	3200	TAPVR(IC)	Correction	35	26	29	None	Well
3	8	2800	TAPVR(C)	Correction	47	27	29	None	Well
4	7	3930	D-TGA	ASO	90	59	63	Digoxin overdose	Died
5	5	3450	D-TGA	ASO	84	60	62	Anasarca	Well
6	4	3000	D-TGA	ASO	67	68	65	None	Well
7	8	3250	D-TGA	ASO	73	63	88	None	Well
8	5	3450	D-TGA + SCO	Senning	35	59	61	AF	Well
9	4	3050	D-TGA VSD	ASO	98	68	67	Anasarca	Well
10	5	3485	D-TGA	ASO	81	66	81	None	Well
11	5	3700	D-TGA	ASO	63	51	53	None	Well
12	6	3100	D-TGA	ASO	58	55	56	Anasarca	Well
13	6	2885	D-TGA	ASO + AMI to RCA bypass	116	88	87	MI, shock, chylothorax, anasarca	Well

CCA, Cardiocirculatory arrest; AXT, aortic crossclamping time; *HypoRV*, hypoplastic right ventricle; PS, pulmonary stenosis; *CoPV*, commissurotomy pulmonary valve; *RVOT*, right ventricular outflow tract; *TAPVR*, Total anomalous pulmonary venous return; *IC*, infracardial; *C*, cardial; *D-TGA*, D-transposition of the great arteries; *ASO*, arterial switch operation; *SCO*, single coronary ostium; *AF*, atrial flutter; *VSD*, ventricular septal defect; *AMI*, arteria mammaria interna; *RCA*, right coronary artery; *MI*, myocardial infarction.

operations and consisted of a roller pump inducing a nonpulsatile flow (Stöckert Instrumente GmbH, Munich, Germany) and a disposable membrane oxygenator (VPCML; Cobe Laboratories, Inc., Lakewood, Colo.) with a heat exchanger and an arterial filter (Pall Corp., Glen Cove, N.Y.). The priming solution consisted of a crystalloid solution (Tutofusin Päd, Kabi Pharmacia, Erlangen, Germany), mannitol (3 ml/kg), and compatible fresh whole blood. The hematocrit value of the circulating blood averaged 25%. After median sternotomy and thymectomy, heparin sulfate was administered (3 mg/kg) and CPB was instituted with a perfusion index of 2.7 L/m² per minute. Deep hypothermia (nasopharyngeal temperature averaging 15° C) was induced by cooling the priming solution in the extracorporeal circuit and the circulating blood with the heat exchanger. For vasodilatation during the cooling period, seven patients received phentolamine (0.3 to 0.5 mg/kg), and six received sodium nitroprussid (0.5 to 1 µg/kg per minute). After deep hypothermia was reached, cardiocirculatory arrest was established after aortic crossclamping, and cardioplegia was induced by a single intraaortic injection of a 4° C cold Bretschneider solution (30 ml/kg). Circulatory arrest was maintained up to a maximum of 60 minutes. If necessary, the surgical procedure was continued under low-flow perfusion (about 25% of the initial flow rate). At the end of the procedure, normal flow was reestablished and the patient was rewarmed. One patient received phentolamine and four received sodium nitroprusside for vasodilatation in the rewarming period. Lungs were reventilated when core temperature reached 30° C and neutralization of heparin was achieved by protamine sulfate (6 mg/kg). If necessary, dopamine or epinephrine was infused before the patient was weaned off bypass.

Postoperative care. The lungs of all neonates were mechanically ventilated after the operation. Cardiac output was estimated clinically. Catecholamines (dopamine, epinephrine) and diuretics (furosemid) were infused to optimize blood pressure

and diuresis. Laboratory studies including blood status, blood gases, electrolytes, renal and liver function tests, coagulation tests, and creatin phosphokinase were performed immediately after the operation and at least at 4 and 16 hours after the operation, as well as daily for the first postoperative week. Electrocardiography, echocardiography, chest roentgenography, and sonography of the pleural space, abdomen, and cerebrum were performed as often as necessary.

Collection of samples. Samples of venous blood were collected in the preoperative period from a peripheral vein, in the intraoperative and postoperative periods from a central venous catheter, and during CPB from the arterial line of the oxygenator. Sample collecting times are listed in Table II. For each sample 1.5 ml of blood taken in tubes containing ethylenediaminetetraacetic acid were necessary for study of complement factors and leukocyte elastase, and, during CPB, a supplement of 0.3 ml was required for leukocyte count and differentiation.

Determination of complement factors. Blood samples were immediately centrifuged for 3 minutes at 3000 rpm, and plasma was stored at 70° C. C3 and C4 were determined by standard turbidimetry with the use of the immunoglobulin G fraction of a rabbit anti-human C3 or C4 antiserum with an automated procedure on an RA 1000 turbidimeter (Technicon, Brussels, Belgium). Normal values for neonates were considered to range between 80% and 100% of normal values of healthy adults.⁶ For C3d determination, the sample was first incubated (vol/vol) with borate buffer containing 22% polyethylene glycol (molecular weight 6000) for 1 night at 4° C. After centrifugation to discard the native C3 molecules, the supernatant containing the C3d was measured by turbidimetry (as was done for C3 and C4) with the following modifications: the sample diluant was supplemented with 20% distilled water to allow the reduction of polyethylene glycol to a maximum of 3.5% in the reaction mixture during immunoprecipitation.

C5a was determined by enzyme immunoassay (Enzygnost

C5a, Behring; Hoechst, Brussels, Belgium). Normal values for healthy adults range between 0.15 and 0.45 $\mu\text{g/L}$. Elastase was measured with immunoactivation enzyme immunoassay (Merck, Darmstadt, Germany). The normal range for healthy adults is $22 \pm 10 \mu\text{g/L}$.

Leukocyte count and differentiation was performed with a Coulter counter (Coulter Electronics, Inc. Hialeah, Fla.). Leukocyte and neutrophil counts during CPB were corrected for hemodilution: determined values were multiplied by the ratio of preoperative hematocrit value (sample time 1) to hematocrit value at sampling. Complement fractions and elastase measured during CPB were not corrected for hemodilution. C3d/C3 was used as index of C3 conversion independent of C3 concentration. The other values were analyzed under equal conditions of hemodilution, namely between the beginning (sample time 3) and the end of CPB (sample time 8) and between the first (sample time 9) and the seventh (sample time 13) postoperative days.

Statistical analysis. Results are expressed as the mean value $\pm$ standard deviation (SD) or as the mean value $\pm$ standard error of the mean (SEM). For statistical analysis parametric *t* tests (paired and not paired) and nonparametric tests (Kruskal-Wallis test, Wilcoxon test) were used. For analysis of χ^2 , the Yates correction was always used. The data were computerized and analyzed with SPSS (SPSS Inc., Chicago, Ill.) and SAS (SAS Institute, Inc., Cary, N.C.) software, and *p* values less than 0.05 were considered significant.

Results

Clinical results. Type of surgical correction and duration of CPB, cardiocirculatory arrest, and aortic cross-clamping are listed in Table I. All neonates received catecholamines for coming off bypass (epinephrine 0.14 to 1.2 $\mu\text{g/kg}$ per minute, median 0.4, *n* = 12; dopamine 2.9 to 8 $\mu\text{g/kg}$ per minute, median 5.4, *n* = 13). Two patients (Nos. 1 and 4) died 48 and 24 hours after the operation from residual intracardiac shunt and digoxine overdose, respectively. One patient (No. 13) had myocardial infarction after arterial switch operation with consecutive low cardiac output and significant capillary leak syndrome. Three other neonates (Nos. 5, 9, and 12) had significant capillary leak syndrome with anasarca but without renal failure after arterial switch operation. Duration of mechanical ventilation in the survivors ranged between 3 and 21 days (median 7 days). Postoperative complications and outcome are summarized in Table I.

Hematocrit value. In the 13 patients, the hematocrit level fell from a preoperative value of $36.3\% \pm 3.4\%$ (mean $\pm$ SD) to $27.5\% \pm 2.2\%$ 10 minutes after beginning of CPB (sample time 3), due to hemodilution. The hematocrit level remained stable during CPB and increased slightly at sample time 8 ($32.6\% \pm 4.5\%$). From the first postoperative day on, the hematocrit level was normal for age and remained stable until the end of the first postoperative week.

Table II. Sample time schedule

Sample time	
1	Preoperative
2	After heparin administration
3	10 minutes after onset of CPB
4	After CCA
5	10 minutes after recirculation
6	Lungs reventilated
7	Core temperature $> 30^\circ \text{C}$
8	After protamine administration
9	First postoperative day
10	Second postoperative day
11	Third postoperative day
12	Fifth postoperative day
13	Seventh postoperative day

CCA, Cardiopulmonary arrest.

Complement factors

C3. After institution of CPB (sample time 3), C3 levels fell in all neonates from a preoperative value of $85.9 \pm 27.4 \text{ mg/dl}$ (mean $\pm$ SD) to $44.9 \pm 14.3 \text{ mg/dl}$. C3 levels remained stable during CPB and after cardiocirculatory arrest, rising significantly between the end of the rewarming period (sample time 7) and protamine administration (sample time 8) (*p* < 0.05). From the third postoperative day on, C3 levels reached normal preoperative values (Fig. 1).

C3d/C3. Nine neonates showed normal preoperative values of C3d/C3 ($\text{C3d/C3} = 0.50 \pm 0.13$) (mean $\pm$ SD). In contrast, four other neonates had abnormally elevated C3d/C3 ratio before the operation ($\text{C3d/C3} = 1.59 \pm 0.11$). Age at operation and at catheterization or delay between catheterization and operation, PGE₁ dosage, preoperative leukocyte count, and CRP were identical in patients with normal and abnormally elevated preoperative values of C3d/C3 (data not shown).

In these 13 neonates, C3d/C3 levels increased significantly between the beginning of CPB (sample time 3) ($\text{C3d/C3} = 0.86 \pm 0.55$) and the end of CPB, after protamine administration (sample time 8) ($\text{C3d/C3} = 1.40 \pm 0.56$) (*p* < 0.0001). C3 conversion was already significant from the beginning of the rewarming period on (sample time 6), compared with the beginning of CPB (sample time 3) (*p* < 0.05). In all neonates, C3d/C3 measured at sample time 8 correlated positively with the value measured at sample time 3 (*p* < 0.0005), as well as with the preoperative value (*p* < 0.05). Fig. 2 shows the course of C3d/C3 in the 13 neonates, and Fig. 3 shows the course of C3d/C3 in neonates with normal and with abnormally elevated preoperative C3d/C3 values. In all patients, no correlation between duration of CPB and

Fig. 1. Course of C3 before, during, and after CPB in 13 neonates (up to sample time 9, $n = 13$, and for sample times 10 to 13, $n = 11$). *mg*%, Milligrams per deciliter.

Fig. 2. Course of C3d/C3 before, during, and after CPB (up to sample time 9, $n = 13$, and for sample times 10 to 13, $n = 11$).

C3d/C3 value at sample time 8 or increase in C3d/C3 levels during CPB (C3d/C3 at sample time 8 – C3d/C3 at sample time 3) was found. C3d/C3 ratios at the end of CPB and within the 3 first postoperative days did not correlate with catecholamine requirement at the end of operation or with outcome.

C4. In the 13 neonates, C4 levels fell from a preoperative value of 14.6 ± 4.7 mg/dl (mean $\pm$ SD) to a value of 8.5 ± 4.0 mg/dl 10 minutes after institution of CPB (sample time 3). C4 levels remained stable during CPB and increased slowly from the end of CPB on (sample time 8) within the 5 first postoperative days, reaching preoperative values (sample time 12). C4 was not significantly consumed after protamine administration (sample time 8) or on the first postoperative day (sample time 9). The four patients with high C3d/C3 ratios before the

operation had significantly lower preoperative values of C4 (10.2 ± 2.6 mg/dl) than did the nine patients with normal preoperative C3d/C3 ratios (17.0 ± 4.6 mg/dl) ($p < 0.001$). This difference persisted during CPB. After protamine administration, C4 levels were significantly lower in patients with high preoperative C3d/C3 levels ($C4 = 7.5 \pm 3.1$ mg/dl) than in patients with normal preoperative C3d/C3 ($C4 = 11.6 \pm 2.9$ mg/dl) ($p < 0.05$). Fig. 4 shows the course of C4 in the neonates with normal and abnormally elevated preoperative C3d/C3 values.

C4 levels measured after protamine administration and during the first 3 postoperative days did not correlate with catecholamine requirement after the operation or with outcome.

C5a. C5a levels were abnormally elevated at the end of

Fig. 3. Course of C3d/C3 before, during, and after CPB in neonates with normal preoperative C3d/C3 (dotted line, $n = 9$) and abnormally elevated preoperative C3d/C3 (solid line, $n = 4$) (from sample time 10 on, $n = 7$ for neonates with normal preoperative C3d/C3 levels).

Fig. 4. Course of C4 before, during, and after CPB in neonates with normal preoperative C3d/C3 (dotted line, $n = 9$) and abnormally elevated preoperative C3d/C3 (solid line, $n = 4$) (from sample time 10 on, $n = 7$ for neonates with normal preoperative C3d/C3 levels). mg%, Milligrams per deciliter.

CPB in the 13 neonates ($C5a = 18.6 \pm 7.3 \mu\text{g/L}$) (mean $\pm$ SD). $C5a$ measured at the end of CPB did not correlate with the duration of CPB. No difference in $C5a$ was observed between patients with normal or elevated preoperative C3d/C3. $C5a$ did not correlate with catecholamine requirement immediately after the operation or with outcome. Values of C3d/C3, C4, and $C5a$ before, during, and after CPB are summarized in Table III.

Leukocytes and neutrophils. In the 10 neonates studied in this respect, leukocyte levels fell from a preoperative value of $10.06 \pm 3.15 \times 10^9/\text{L}$ (mean $\pm$ SD) to $3.21 \pm 0.64 \times 10^9/\text{L}$ 10 minutes, after institution of CPB (sample time 3) ($p < 0.0001$). In addition, leukocyte levels fell significantly immediately after extracorporeal

circulation was restored after cardiocirculatory arrest (sample time 4) (leukocytes = $2.47 \pm 0.46 \times 10^9/\text{L}$) ($p < 0.0001$) and again 10 minutes after recirculation (sample time 5) (leukocytes = $2.23 \pm 0.45 \times 10^9/\text{L}$) ($p < 0.05$). Leukocyte levels remained stable during the rewarming period. Between the end of the rewarming period (sample time 7) and a few minutes after protamine administration (sample time 8), leukocyte levels rose significantly from a value of $2.33 \pm 0.67 \times 10^9/\text{L}$ to a value of $7.19 \pm 1.84 \times 10^9/\text{L}$ ($p < 0.0001$). Leukocyte levels rose further to a value of $13.19 \pm 3.53 \times 10^9/\text{L}$ 4 hours after the operation and reached a value of $17.63 \pm 5.81 \times 10^9/\text{L}$ on the first postoperative day.

The course of neutrophils was parallel to that of leuko-

Table III. Measured values of C3d/C3, C4, C5a, leukocytes, neutrophils, and leukocyte elastase before, during, and after CPB.

	Sample time						
	1	3	4	5	6	7	8
C3d/C3	0.78 ± 0.48	0.86 ± 0.55	0.96 ± 0.41	1.01 ± 0.41	1.19 ± 0.62	1.19 ± 0.54	1.40 ± 0.56
C4 (mg/dl)	14.6 ± 4.7	8.5 ± 4.0		8.30 ± 3.3	8.95 ± 3.5	8.9 ± 3.5	10.4 ± 3.5
C5a (μg/L)							18.6 ± 7.3
Leukocytes (×10 ⁹ /L)	10.06 ± 3.15	3.21 ± 0.64	2.47 ± 0.46	2.23 ± 0.45	2.38 ± 0.82	2.33 ± 0.67	7.19 ± 1.84
Neutrophils (×10 ⁹ /L)	5.14 ± 1.18	1.46 ± 0.35	1.15 ± 0.27	0.95 ± 0.31	0.99 ± 0.44	1.00 ± 0.31	4.10 ± 1.18
Elastase (μg/L)		107 ± 39					438 ± 198

Sample times are listed in Table II. Values are given as mean ± SD.

cytes. Neutrophil levels fell significantly from a preoperative value of $5.14 \pm 1.18 \times 10^9/\text{L}$ to $1.46 \pm 0.35 \times 10^9/\text{L}$ 10 minutes after beginning of extracorporeal circulation ($p < 0.005$). Neutrophils were at a minimal level after extracorporeal circulation was restored immediately after cardiocirculatory arrest (sample time 5) (neutrophils = $0.95 \pm 0.31 \times 10^9/\text{L}$) and remained stable until termination of bypass, before protamine administration. Neutrophil levels rose from a value of $1.00 \pm 0.31 \times 10^9/\text{L}$ at the end of the rewarming period (sample time 7) to a value of $4.10 \pm 1.18 \times 10^9/\text{L}$ immediately after protamine administration (sample time 8) ($p < 0.005$).

No difference in leukocyte and neutrophil count was found before, during, or immediately after CPB between patients with normal or elevated preoperative C3d/C3 levels. Fig. 5 shows the course of leukocytes and neutrophils during and after CPB in 10 neonates. Values of leukocytes and neutrophils before, during, and after CPB are summarized in Table III.

Elastase. In the 13 neonates, elastase averaged $106.6 \pm 39.3 \mu\text{g}/\text{L}$ (mean ± SD) after institution of CPB (sample time 3) and rose to a value of $438.0 \pm 198.0 \mu\text{g}/\text{L}$ at the end of CPB (sample time 8) (Table III). Elastase release during CPB (elastase at sample time 8 – elastase at sample time 3) averaged $331.5 \pm 175.7 \mu\text{g}/\text{L}$. Elastase release and elastase level at sample time did not correlate with leukocyte or neutrophil count after protamine administration. Elastase release did not correlate with duration of CPB, with catecholamine requirement immediately after CPB, or with outcome.

Discussion

The inflammatory response during cardiac operations has not yet been studied in neonates. We report here that

full-term neonates show significant complement activation and leukocyte stimulation when undergoing CPB. C3 conversion in this age group is mainly due to the activation of the alternative pathway that occurs primarily in the rewarming period and shows its peak after protamine administration, which is similar to the process in infants and children.⁷ Late phase of complement activation is also involved as shown by the elevated C5a values measured in all patients at the end of extracorporeal circulation, with consecutive leukocyte elastase release caused by leukocyte stimulation. The course of leukocytes and neutrophils differs in part from that previously described in older children.⁷ After institution of extracorporeal circulation, an initial marked leukopenia and neutropenia occurs, as classically described.⁷⁻⁹ Using C3d/C3 determination, we could not detect complement activation at this time. Hence, we suggest that early leukopenia may be independent of complement activation and due to physical factors such as hypothermia, increasing leukocyte membrane rigidity, and favoring leukocyte trapping in the lungs and in the extracorporeal circuit. Circulating leukocyte and neutrophil levels decrease further during low-flow CPB most probably in response to activated complement.⁸ In contrast to what is observed in children,⁷ leukocyte and neutrophil counts of neonates remain low during the whole rewarming period; also, the peak value of leukocytes and neutrophils after protamine administration is significantly lower in this age group, not reaching preoperative values. This observation might be explained by a limited mobilization of leukocytes from the lungs, bone marrow, and perhaps other hematopoietic organs such as the liver and spleen, probably as a result of an impaired motility of neonatal leukocytes and neutrophils.¹¹

Elastase release during CPB is not different from that

Fig. 5. Leukocyte (dotted line) and neutrophil count (solid line) before, during, and after CPB in 10 neonates.

observed in children,⁷ indicating that leukocytes of neonates can be stimulated in this regard to the same extent as those of children. The lack of correlation between elastase release and the number of concomitant circulating neutrophils could be due to a nonparallel relation between the effect of CPB on neutrophil stimulation with regard to mobilization or aggregation and degranulation.

Considering complement activation and leukocyte stimulation, neonates, when subjected to the stress of cardiac operation, show a similar inflammatory response to those of older children and adults. Although, in infants and children enrolled in a similar study over the same time period, CPB-related complications have been frequent and a relationship could be shown between postoperative complications and complement activation,⁷ neonates studied in this series had no or only mild CPB-related complications. The complicated postoperative course of patient 13 was due to myocardial ischemia, and the two neonates who died had inadequate cardiac correction and digoxin overdose. Four of the surviving neonates had important capillary leak syndrome that was considered to be CPB-related.¹² Actually, in our series, no correlation could be found between this complication and the inflammatory reaction during CPB, with regard to complement activation and leukocyte stimulation. The reasons why CPB-related complications appear to be milder in neonates compared with those of older infants and children, in spite of a similar inflammatory response during CPB, are unclear. Neonates have immature complement receptors on neutrophils⁵ and decreased complement-mediated opsonization¹⁰ impairing neutrophil migration and phagocytosis,¹¹ both possibly limiting the tissue destruction by various inflammatory stimuli occurring during cardiac operation. Nevertheless, in our series of neonates,

neutrophils were stimulated during CPB and liberated proteolytic enzymes as shown by leukocyte elastase release, suggesting complement-induced leukocyte activation. It is noteworthy that all but two patients were treated with PGE₁ before the operation for at least 24 hours. Because PGE₁ is known to have cytoprotective effects and inhibit leukocyte adherence,^{13,14} pretreatment with PGE₁ together with naturally impaired neutrophil migration could have been of benefit in the particular situation of neonates undergoing cardiac operation.

Four patients had abnormally elevated preoperative values of C3d/C3 and lower preoperative values of C4 compared with those of the nine other patients, in the absence of other signs of inflammation or infection. A rational explanation for this observation is actually lacking. Bacterial fetomaternal infection was excluded in all cases. Because our patients were subjected to several manipulations that were able to induce complement activation (delivery, cardiac catheterization with angiography, etc.)^{15,16}, the high levels of C3d/C3 and low values of C4 observed in the four neonates might be discussed with regard to interindividual variations of postnatal complement activation and intrahepatic clearance of C3d and synthesis of C4. In spite of the lack of clinical and biologic evidence of impaired liver function in the four neonates with high preoperative C3d/C3 levels, the rapid normalization of C3d/C3 after the operation seems to support the hypothesis of impaired C3d hepatic clearance in the preoperative period.

In conclusion, neonates show important inflammatory response when undergoing CPB with complement activation and leukocyte stimulation. In our series, CPB-related complications were not dramatic; the physiologic immaturity of complement receptors on the neutrophils of the neonates, maturational defects in phagocytic cells,

and pretreatment with PGE₁ possibly exert a beneficial cytoprotective effect that must be investigated further.

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REFERENCES

1. Chenoweth DE, Cooper SW, Hugli TE, et al. Complement activation during cardiopulmonary bypass: evidence for generation of C3a and C5a anaphylatoxins. *N Engl J Med* 1981;304:497-503.
2. Kirklin JK, Westaby S, Blackstone EH, et al. Complement and the damaging effects of cardiopulmonary bypass. *J THORAC CARDIOVASC SURG* 1983;86:845-57.
3. Strunk RC, Fenton LJ, Gaines JA. Alternative pathway of complement activation in full-term and premature infants. *Pediatr Res* 1979;13:641-3.
4. Berger M. Complement deficiency and neutrophil dysfunction as risk factors for bacterial infection in newborns and the role of granulocyte transfusion in therapy. *Rev Infect Dis* 1990;12(Suppl 4):401-9.
5. Bruce MC, Baley JE, Medvik KA, et al. Impaired surface membrane expression of C3bi but not C3b receptors on neonatal neutrophils. *Pediatr Res* 1987;21:306-11.
6. Johnston RB, Altenburger KM, Atkinson AW, et al. Complement in the newborn infant. *Pediatrics* 1979;65 (Suppl 5):781-6.
7. Seghaye MC, Duchateau J, Grabitz RG, et al. Complement activation during cardiopulmonary bypass in infants and children: relation to the postoperative multiple system organ failure. *J THORAC CARDIOVASC SURG* 1993;106: 978-87.
8. Hammerschmidt DE, Stroncek DF, Bowers TK, et al. Complement activation and neutropenia occurring during cardiopulmonary bypass. *J THORAC CARDIOVASC SURG* 1981;81:370-7.
9. Quiroga MM, Miyagishima R, Haendschen LC, et al. The effect of body temperature on leukocyte kinetics during cardiopulmonary bypass. *J THORAC CARDIOVASC SURG* 1985;90:91-6.
10. Zach TL, Hostetter MK. Biochemical abnormalities of the third component of complement in neonates. *Pediatr Res* 1989;26:116-20.
11. Klein RB, Fischer TJ, Gard SE, et al. Decreased mononuclear and polymorphonuclear chemotaxis in human newborns, infants and young children. *Pediatrics* 1977;60:467-72.
12. Marath A, Man W, Taylor KM. Histamine release in pediatric cardiopulmonary bypass: a possible role in the capillary leak syndrome. *Agents Actions* 1987;20:299-302.
13. Bihari DJ, Tinker J. The therapeutic value of vasodilator prostaglandins in multiple organ failure associated with sepsis. *Intensive Care Med* 1988;15:2-7.
14. Chopra J, Webster RO. PGE₁ inhibits neutrophil adherence and neutrophil-mediated injury to cultured endothelial cells. *Am Rev Respir Dis* 1988;138:915-20.
15. Herson VC, Block C, Eisenfeld LI, et al. Effect of labor and delivery on neonatal polymorphonuclear leukocyte number and function. *Am J Perinatol* 1992;9:285-8.
16. Eaton S, Tsay HM, Yost F, et al. Assays for plasma complement activation by x-ray contrast media. *Invest Radiol* 1990;25:789-92.