CLINICAL STUDY

Procalcitonin, neopterin and C-reactive protein after pediatric cardiac surgery with cardiopulmonary bypass

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Abstract

Background: **Kinetics of activation of newly introduced inflammatory markers is of particular value in their use in postoperative setting after pediatric cardiac surgery.**
Objective: **To assess the influence of cardiopulmonary bypass (CPB) on activation of inflammatory markers.**
Methods: **Procalcitonin, neopterin and C-reactive protein (CRP) blood levels were measured before, on day 1 and day 2 after surgery in 152 pediatric patients undergoing cardiac surgery with CPB. All patients had infection-free postoperative course.**
Results: **Procalcitonin blood levels increased from 0.08 (0.001–0.19) ng/mL before surgery to 0.79 (0.13–3.5) ng/mL on day 1 (p<0.001) and 0.52 (0.07–2.7) ng/mL on day 2 (p<0.001) after surgery. Procalcitonin levels on day 1 correlated with CPB duration, cross-clamping time and use of aprotinin. Neopterin values increased from 8.4 (4.4–32) nmol/L before surgery to 16.1 (6.8–37.6) nmol/L on day 1 (p<0.001) and 10.9 (5.3–31.1) nmol/L on day 2 (p<0.001) after surgery. Neopterin levels on day 1 correlated negatively with age, lowest esophageal temperature and use of aprotinin; and positively with circulatory arrest, length of stay and use of modified ultrafiltration. CRP values increased from 4.7 (2.7–9.6) mg/L before surgery to 38.8 (13–73.5) mg/L (p<0.001) on day 1 and 38.3 (15–88) mg/L (p<0.001) on day 2 after surgery. CRP levels correlated positively with age at surgery, use of aprotinin and lowest esophageal temperature; and negatively with circulatory arrest and modified ultrafiltration.**
Conclusions: **Procalcitonin and neopterin levels peaked on day 1, CRP levels remained elevated during first two postoperative days. Neopterin and CRP had age dependent kinetics *(Tab. 2, Fig. 2, Ref. 18)*. Full Text (Free, PDF) *www.bmj.sk*.**
Key words: procalcitonin, neopterin, C-reactive protein, children, cardiac surgery.

Surgery and cardiopulmonary bypass (CPB) induce inflammatory cascade (1) and prolonged activation of inflammatory markers such as C-reactive protein (CRP). This activation limits the use of inflammatory markers for diagnosis of postoperative infection. Knowledge on activation of newly introduced markers is warranted. Procalcitonin (PCT), the precursor of calcitonin, is a marker of severe bacterial infection (2) and now an established sepsis marker (3). An increase in PCT blood levels has been described in adult patients after cardiac surgery (4–6) and more recently, also in children (7, 8). Neopterin, a pteridin derivate synthesized from guanosine, is produced by activated monocytes and macrophages. It is production is mainly induced by interferon-gamma from T lymphocytes (9). Neopterin monitoring is helpful in the follow-up of pathological conditions associated with the activation of cell-mediated immunity (10). Also, increased neopterin levels were described in patients with Down syndrome (11). Data on neopterin kinetics in children after cardiac surgery are limited.

Activation of inflammatory process associated with cardiac surgery can be reduced by using anti-inflammatory strategies. Aprotinin, serine protease inhibitor used to reduce blood losses from generalized activation of fibrinolysis, has also anti-inflammatory properties (12). Modified ultrafiltration (MUF) is an elimi-

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nation technique now used by pediatric cardiac surgeons to reduce the volume loading after CPB (13). Little is known about influence of aprotinin and MUF on PCT, neopterin and CRP activation.

The objective of our study was to assess the kinetics of PCT, neopterin, and CRP in children after cardiovascular surgery performed with CPB and the influence of CPB parameters on marker's activation.

Methods

The prospective study included a total of 152 children surgically treated with cardiopulmonary bypass for congenital heart defects from December 1999 to February 2001. Anesthesia was induced with 0.2 mg/kg midazolam, 25 mcg/kg fentanyl and 0.1 mg/kg vecuronium. Fentanyl and midazolam boluses were used to maintain anesthesia, supplemented with volatile agent (1 % sevofluran). Patients were mechanically ventilated. CPB was initiated after aorta-bicaval cannulation. The circuit was primed with crystaloid solution (Normosol-R), 5 % albumin and packed red blood cells. Methylprednisolon, 30 mg/kg, was administered to pump prime to all patients. Aprotinin, 10–20.000 KIU/kg, was administered to patients with history of previous cardiac surgery (n=23). Moderate to deep hypothermia with esophageal temperature 12–34 °C was used during CPB. Pump flows were 50–200 ml/kg/min. A period of circulatory arrest was necessary to complete the surgery in selected patients. Arteriovenous MUF was initiated after the separation from CPB at the surgeon's discretion. Infusion rates were adjusted to maintain appropriate filling pressures.

Patients received prophylactic cephazolin 20 mg/kg before surgery and thereafter, 10 mg/kg three times a day at cardiac intensive care unit. Postoperative care included mechanical ventilation, inotropic support and continuous analgesia with 20 mcg/kg/hr of morphine. Muscle paralysis with atracurium 10 mcg/kg/min and analgesia with fentanyl infusion, 5 mcg/kg/hr, was administered to patients at risk of pulmonary hypertension crisis. No clinical sign of infection was detected in any patient in the early postoperative period.

PCT, neopterin and CRP blood values were measured immediately before the surgery and postoperatively on days 1 and 2. Serum concentration of PCT was measured with immunoluminometric assay LUMItest® PCT (BRAHMS Diagnostica; normal values: <0.5 ng/mL), serum neopterin concentration was determined with ELISA (BRAHMS Diagnostica; normal values in neonates: <65 nmol/L, in infants and children <10 nmol/L), and serum CRP concentration were determined with immunoturbidimetric method (ORTHO CLINICAL DIAGNOSTICS, Inc., normal values: <10 mg/L). Collected data included age, cardiac diagnosis, presence of morbus Down, type of surgery, Aristotle Basic Complexity Score (ABCS) (14), length of stay at cardiac intensive care unit, CPB and cross-clamping time, use of circulatory arrest, lowest esophageal temperature during CPB, and use of aprotinin and MUF. The study protocol was approved by the Ethical Committee of Children's University Hospital.

Data are presented as median (10th and 90th percentile) in text and as a box plot (10th, 25th, 50th, 75th and 90th percentile) in figures. Bivariate analysis was used to assess the influence of continuous parameters on PCT, Neo and CRP values. Analysis of variance and t-test was used to assess the influence of nominal parameters on study markers. Level of significance was set at P<0.05. JMP discovery software version 4.0.2 (SAS Institute Inc., Cary, NC, USA) was used for statistical analysis.

Results

Demographic details are provided in Table 1.

Procalcitonin

Serum PCT values were within normal range in all patients before the surgery. PCT values increased from 0.08 (0.001–0.19) ng/mL before surgery to 0.79 (0.13–3.5) ng/mL on day 1 (p<0.001) and 0.52 (0.07–2.7) ng/mL on day 2 (p<0.001) after surgery (Fig. 1). The PCT value peaked on postoperative day 1 in 74 % of patients. There was a positive correlation between the CPB duration and PCT on day 1 after surgery (r=0.04; p=0.012). Duration of cross-clamping time with PCT levels on day 1 did not reach statistical difference (r=0.01; p=0.22), however patients with cross-clamping time longer than 80 minutes had higher PCT levels: 1.2 (0.4–4.6) versus 0.7 (0.13–2.7) ng/mL, p=0.03. There was no additional increase in PCT values in patients with circulatory arrest. Patients who received aprotinin had higher PCT levels 1.69 (0.12–5.4) versus 0.7 (0.13–2.6) ng/ml (p=0.0075) on day 1. There was no correlation between PCT on

Tab. 1. Perioperative characteristics of patients.

Patient characteristics	n=152
Age (median, range; years)	1y (11d–13y)
Weight (median, range; kg)	8.4 (3.3–44.0)
CPB time (median, range; min)	96 (48–149)
Cross-clamping time (median, range, min)	53 (20–95)
Arrest (n)	30
Arrest time (median, range, min)	13 (1–44)
ICU stay (median, range; days)	2 (1–7)
ABCS (median, range)	8 (4–14)
Aprotinin (n)	23
Modified ultrafiltration (n)	70
Down syndrome (n)	20
Cardiac diagnosis:	
Aortic stenosis/insufficiency	11
Atrial septal defect	21
Ventricular septal defect	30
Atrioventricular septal defetc complete	11
Double outlet right ventricle	5
D-transposition of great arteries	19
Hypoplastic left heart syndrome	8
Tricuspid atresia	9
Tetralogy of Fallot	19
Other	19

d – day, m – month, y – year, ABCS – Aristotle Basic Complexity Score, ICU – intensive care unit

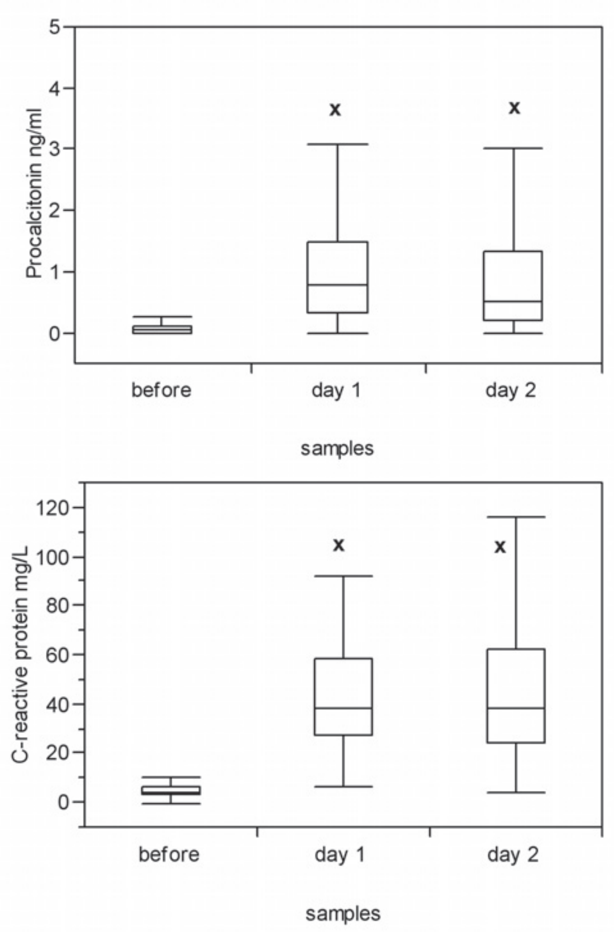

Fig. 1. Procalcitonin and CRP values in patients after pediatric cardiac surgery with cardiopulmonary bypass. (The boxes extend from the 25th to the 75 th percentile, with a horizontal line at the median. Whiskers extend down to 10 th and up to 90 th percentile. Before surgery, day 1 – the first postoperative day; day 2 – the second postoperative day; x – p<0.05).

day 1 and 2 and age, lowest esophageal temperature, ABCS, use of MUF, and length of stay.

Neopterin

Because of age-depended normal values, results are presented in Figure 2 separately for neonates and for older children. All neonates had baseline neopterin values in normal range. We observed abnormal baseline neopterin values (11.2–32 nmol/L) in 24 older children. Of those, 18 were infants and six older children. Results in text are for the whole group (neonates, infants and older children). In whole studied group of patients neopterin values increased from 8.4 (4.4–32.0) nmol/L before surgery to 16.1 (6.8–37.6) nmol/L on day 1 ($p<0.001$) and 10.9 (5.3–31.1) nmol/L on day 2 ($p<0.001$) after surgery. Neopterin value peaked on day 1 in 79 % of patients. There was a negative correlation between the age at surgery and the value of neopterin on both days, 1 ($r=0.19$, $p<0.001$) and 2 ($r=0.12$, $p<0.001$). CPB duration and cross-clamping time did not correlate with neopterin levels. Patients with circulatory arrest had higher neopterin levels compared to patients without circulatory arrest: 28.3 (15.0–70.0) nmol/L versus 14 (6.4–31.9) nmol/L ($p<0.001$) on day 1; and 24.8 (8.8–47.5) nmol/L versus 9.5 (5.1–23.0) nmol/L ($p<0.001$) on day 2. There was a positive correlation between the length of stay at ICU and neopterin level on days 1 ($r=0.03$, $p=0.023$) and 2 ($r=0.07$, $p=0.001$). Patients who received aprotinin had lower neopterin levels on day 1: 11.2 (5.4–23) nmol/L versus 17.2 (7.2–42.6) nmol/L, $p=0.008$; and on day 2: 7.5 (4.8–17.4) nmol/L versus 11.9 (5.5–33.5) nmol/L, $p=0.017$. Patients with MUF had higher neopterin levels on day 1: 18.5 (9.5–50) versus 13.1 (6.1–30) nmol/L, $p=0.0006$, and on day 2: 16.1 (5.6–42) versus 8.9 (5.1–23.6) nmol/L, $p<0.001$. There was a positive correlation between ABCS and neopterin on days 1

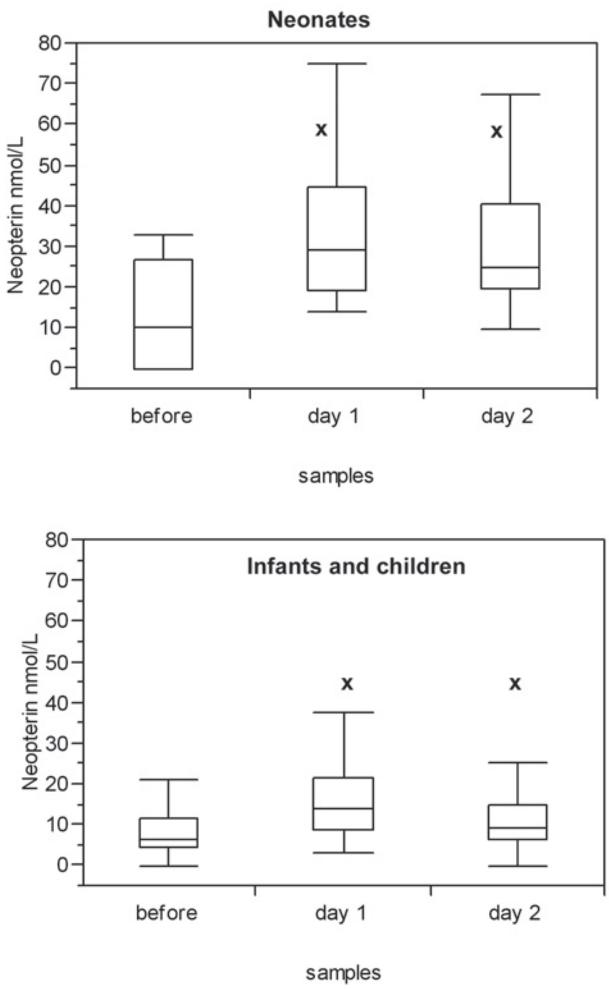

Fig. 2. Neopterin values in neonates (above) and infants and older children (below) after pediatric cardiac surgery with cardiopulmonary bypas. (The boxes extend from the 25th to the 75 th percentile, with a horizontal line at the median. Whiskers extend down to 10 th and up to 90 th percentile. Before surgery, day 1 – the first postoperative day; day 2 – the second postoperative day; x – p<0.05).

(r=0.12, p<0.001) and 2 (r=0.21, p<0.001). There was a negative correlation between the lowest temperature on CPB and neopterin on days 1 (r=0.18, p<0.001) and 2 (r=0.22, p<0.001).

C-reactive protein

Serum CRP values were within normal range in all patients before the surgery. CRP values increased from 4.7 (2.7–9.6) mg/L before surgery to 38.8 (13.0–73.5) mg/L (p<0.001) on day 1 (p<0.001) and 38.3 (15–88) mg/L (p<0.001) on day 2 after surgery (Fig. 1). In 55 % of patients the peak value of CRP was observed on day 1. CRP values on day 1 positively correlated with patient's age at surgery (r=0.16, p<0.001). There was no correlation between the CPB duration, cross-clamping time and CRP levels on days 1 and 2 after surgery. Patients with circulatory arrest had lower CRP levels on day 1 21.3 (8.1–63.6) mg/L versus 43.0 (22.6–74.5) mg/L (p<0.001). There was no correlation of CRP values with length of stay, there was a positive correlation between the lowest temperature on CPB and CRP values on days 1 (r=0.11, p<0.001) and 2 (r=0.04, p=0.01). Patients with MUF had significantly lower CRP values on day 1: 35.0 (10.8–68) versus 44.7 (22.09–74.9) mg/L, p=0.035. Patients who received aprotinin have higher CRP values on day 1: 56 (29.5–77.4) versus 36.1 (12.5–72) mg/L, p=0.001. There was a negative correlation between ABCS and CRP values on day 1 (r=0.05, p=0.004).

Presence of Down syndrome has no influence on PCT, Neopterin and CRP levels pre- and postoperatively. Patients with MUF and lower esophageal temperature were significantly younger than patients without MUF and higher esophageal temperature. Patients who received aprotinin were significantly older than those without aprotinin. There was no correlation between ABCS and age. The impact of parameters' influence on inflammatory markers activation is summarized in Table 2.

Tab. 2. Influence of perioperative characteristics on inflammatory marker activation.

	Procalcitonin		Neopterin		C-reactive protein	
Parameter	day 1	day 2	day 1	day 2	day 1	day 2
Age	–	–	↓	↓	↑	–
CPB time	↑	–	–	–	–	–
Cross-clamping time	↑	–	–	–	–	–
Circulatory arrest	–	–	↑	↑	↓	–
Temperature	–	–	↓	↓	↑	↑
Aprotinin	↑	–	↓	↓	↑	–
MUF	–	–	↑	↑	↓	–
ABCS	–	–	↑	↑	↓	–
Length of ICU stay	–	–	↑	↑	–	–

ABCS – Aristotle basic complexity score, CPB – cardiopulmonary bypass, ICU – intensive care unit, MUF – modified – ultrafiltration, positive correlation or increased level of marker in the presence of parameter, – negative correlation or decreased level of marker in the presence of parameter

Discussion

Our study showed that serum concentrations of PCT, neopterin and CRP increased over normal values after cardiac surgery with the use of CPB. PCT values peaked on day 1 in almost three-quarters of the patients. These results are in concordance with results of previous studies in children (8, 15). Arkader observed all PCT values below 2 ng/mL, the fact of which is considered to be the upper limit for systemic inflammatory response. Beghetti reported 75th percentile at 2.25 ng/mL. This correlates with our results, 18 % of patients had PCT values higher than 2 ng/mL and all values were below 5 ng/mL. Hammer (7) observed significantly higher levels of PCT in patients with cross-clamping time of more than 80 minutes, as we did in our study. In addition to this, a positive correlation between CPB time and PCT value on day 1 after surgery was found. This correlation can be explained by bacterial translocation in intestinal tract. The longer the period of non-pulsatile flow during cardiopulmonary bypass, the greater the possibility of intestinal barrier disruption and bacterial translocation that can be measured by endotoxemia (16). Endotoxin is a potent stimulator of PCT production. Although aprotinin has anti-inflammatory properties, PCT levels in patients who received aprotinin into pump prime were higher postoperatively. This can be explained by longer CPB time in these patients. PCT values did not correlate with age at surgery, lowest esophageal temperature, use of circulatory arrest and length of stay.

Eighteen infants and six older children had abnormal baseline neopterin values in our study. This may represent high inter-individual variability in an age group of neonates and infants. Neopterin value peaked in 79 % of patients on day 1 after surgery. This finding is in accordance with published data for adult patients. Neopterin levels were age-dependent in our study; higher neopterin levels were recorded in younger patients. In patients with circulatory arrest we observed significantly higher neopterin levels but this can be explained by lower age of patients with circulatory arrest. Similarly the influence of esophageal temperature, MUF, and use of aprotinin can be explained by age-dependency of neopterin. Brkic (17) observed a positive correlation of baseline and postoperative day 1 values of neopterin with length of stay at ICU. We observed the same correlation, but only postoperatively. Neopterin was described as prognostic marker of postoperative complications after cardiac surgery (18). However, all our patients had no complications during the postoperative period.

Begheti (8) observed peak CRP levels in children on postoperative day 3 with the levels remaining abnormal on postoperative day 5. In adult patients, CRP is reported to peak on day 2 or day 3 after cardiac surgery, with the median level of 200 mg/L (4–6). In majority of our patients CRP levels were below 100 mg/L and remained unchanged during postoperative days 1 and 2. We observed positive correlation of CRP levels on day 1 with age. Increased CRP values were recorded in patients with higher esophageal temperature and use of aprotinin. Conversely lower CRP values in patients with circulatory arrest and MUF. The exact cause of these findings is not known. However; the effect of age may be more important than are effects of other factors.

Aristotle basic complexity score (ABCS, range 3–15) is a parameter for assessing the intricacy of congenital cardiac surgery. There was a positive correlation with neopterin values on days 1 and 2 after surgery and a negative correlation with CRP values on day 1. These findings remain to be elucidated.

The limitation of our study is the short period of postoperative inflammatory marker monitoring. Since arterial and central venous catheters are usually removed on the second postoperative day, extended monitoring would require venepuncture in most of our patients with no complications during the postoperative period.

To our knowledge, this is the largest study evaluating CRP, PCT and neopterin levels in children undergoing CPB. We believe that our findings will contribute to the general knowledge of inflammatory markers kinetics and thus help clinicians to evaluate appropriately the patient's inflammatory status after cardiac surgery. However, the influence of postoperative complications such as severe low cardiac output and multiorgan dysfunction remains to be assessed.

In conclusions, our study showed that Procalcitonin, neopterin and C-reactive protein levels increase above normal in children after cardiac surgery with CPB. PCT and neopterin reach their maximum on postoperative day 1 and then decrease, whereas CRP values remain elevated during postoperative days 1 and 2. Due to rapid kinetics, PCT and neopterin appear to be promising inflammatory markers in postoperative period. Age dependency of neopterin and CRP should be taken into consideration.

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