Quantitative Analysis of Procalcitonin After Pulmonary Endarterectomy in Relation to Cytokines and C-Reactive Protein

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Received April 10, 2008; Accepted December 8, 2008.

Keywords: C-reactive protein – Interleukin-6 – Procalcitonin – Pulmonary endarterectomy – Tumor necrosis factor-α

This study was supported by grants IGA MZ NR9223-3 of the Ministry of Health and MSM0021620819 of the Ministry of Education, Czech Republic.

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Abstract

Background. Endarterectomy of the pulmonary artery (PEA) is a potential curative treatment method for selected patients with chronic thromboembolic pulmonary hypertension (CTEPH). The postoperative course after PEA is associated with high rate of early postoperative mortality, which ranges from 5 to 23%. The aim of this prospective clinical study was to assess the time course of procalcitonin (PCT) after uncomplicated PEA in relation to other inflammatory parameters.

Patients and methods. 32 patients with CTEPH treated with PEA using cardiopulmonary bypass and deep hypothermic circulatory arrest were included into study. PCT, C-reactive protein (CRP), interleukin (IL)-1β, IL-6, IL-8, and sTNFRI arterial concentrations were measured before/after sternotomy, last DHCA, separation from bypass, and repeatedly to 48 h after sternotomy.

Results. Mean duration of CPB was 338.2 min.; mean circulatory arrest time 39.9 min. The initial decline of PCT, IL-6, and IL-8 was followed by an increase culminated 6–24 h after sternotomy. PCT peak was detected +24 h (1.97 ng/ml, 1.70–2.54). IL-6 culminated after separation from CPB, IL-8 was highest 12 h after sternotomy. PCT levels correlated with IL-6 ($r = 0.81$), CRP ($r = 0.72$), and sTNFRI ($r = 0{,}58$).

Conclusion. Postoperative PCT culmination was delayed in alignment to pro-inflammatory mediators as IL-6 and IL-8. Positive correlation between PCT and IL-6 corresponded with the role of IL-6 in PCT induction in perioperative phase. Plasma PCT estimation extended to a measurement of CRP and IL-6 may become a useful complementary examination in early postoperative period after PEA.

Introduction

Endarterectomy of the pulmonary artery (PEA) is a potential curative treatment method for selected patients with chronic thromboembolic pulmonary hypertension (CTEPH), whose prognosis would be otherwise very poor. After successful endarterectomy, pulmonary pressure and pulmonary vascular resistance drop and the cardiac output increases. PEA provides a significant survival advantage, compared to the natural prognosis of CTEPH, medical treatment or transplant. However the postoperative course after PEA is accompanied by a number of specific complications, which need the prolonged care in ICU, but also contributes to the high rate of early postoperative mortality, which ranges from 5 to 23%. Common causes of death are massive hemoptysis, pulmonary reperfusion edema, right ventricular failure, and multi system organ failure. In addition to postoperative complications, PEA is also associated with hemodynamic instability in the perioperative course, suggesting the involvement of circulating mediators and cytokines as interleukin-1β (IL-1β) and IL-6 [1, 2]. The lungs, specifically the endothelial lining of the pulmonary circulation, play an active role in hemodynamic and immune processes, including the production and action of various inflammatory mediators [3].

Procalcitonin (PCT) is a highly specific marker for the diagnosis of clinically relevant bacterial infections and sepsis. The role of PCT in inflammatory conditions, such as sepsis, was first described by Assicot et al. [4] who observed a rise in serum PCT levels 3–4 h after a single injection of endotoxin. Over the last decade, PCT has become useful as a novel marker of infection. One major advantage of PCT compared to both inflammatory cytokines and acute phase proteins is its early and specific increase in response to severe systemic bacterial infections and sepsis. PCT is currently discussed as an indicator of postoperative complications following cardiac surgery [1, 5] and some recent studies refer to limited specificity of PCT in detection of infection complications in early postoperative period [6].

The aims of this prospective study were to identify the time course of plasma PCT levels in first 48 h after uncomplicated PEA and to characterize the possible differences in serum PCT dynamics in relation to main inflammatory cytokines, which are involved in PCT induction during immune response. In this study, the plasma levels of PCT, IL-1β, IL-6, IL-8, soluble tumor necrosis factor receptor type I (sTNFRI), and C-reactive protein (CRP) were measured in patients during first 48 h after the PEA procedure.

Material and Methods

The prospective study was realized on the Second Department of Surgery – Department of Cardiovascular Surgery of the First Faculty of Medicine in Prague from January 2006 to December 2007. The ethical committee of the institution approved a study protocol and informed consent was obtained from the subjects.

Patients

32 patients with CTEPH (consecutive series of patients, 21 males and 11 females, mean age being 52.2 ± 11.9 yr.) with New York Heart Association (NYHA) class 3.5 ± 0.4 (mean ± SD), were follow with PEA. Their mean pressure in the main pulmonary artery was 54 mm Hg. The control group for estimation of reference laboratory values consisted of 30 healthy persons (19 males and 11 females, the average age 46.9 ± 8.1 yr.).

Surgical procedures

Following median sternotomy, cardiopulmonary bypass (CPB) was established with cannulation of the ascending aorta and the inferior and superior vena cava. Cooling began immediately using CPB cooling blankets, cooled to a bladder temperature of 18–20 °C. Cardiac arrest was induced after aortic cross-clamping by infusion of cardioplegic solution (mostly St. Thomas).

Approach to the pulmonary artery had to be bilateral; both pulmonary arteries had to be substantially involved. Pulmonary artery was open; a correct dissection plane was made and pursued to the segmental branches of pulmonary artery. For

precision visualization during peripheral dissection, repeated periods of deep hypothermic circulatory arrest (DHCA) with reestablishment of CPB between them were necessary. If other cardiac procedures were required, these were performed during the systemic rewarming. After rewarming period patient was weaned from CPB by the stepwise reduction of pump flow. Before the end of CPB, we used an ultrafiltration of diluted blood for hemoconcentration.

Arterial blood pressure was continuously recorded after catheterization of a femoral artery. Hemodynamic monitoring included a surgically placed left atrial catheter in all patients and flow–directed Swan-Ganz-catheter in the PEA patients.

PCT, Cytokine and CRP Analysis

Arterial blood samples were drawn from femoral artery catheter before operation, after sternotomy, after the last DHCA, after separation from bypass, 12, 18, 24, 36, and 48 h after start of surgery. Blood samples of control group were drawn only at baseline. For all measurements, 5-ml of arterial blood was taken into a vacutainer tube and immediately centrifuged at 5000 rpm for 15 min. Plasma was stored at –80 °C until analysis. Plasma levels of PCT were detected by Kryptor test (B.R.A.H.M.S. AG, Henningsdorf, Germany) in duplicates. Plasma concentrations of IL-1β, IL-6, IL-8, sTNFRI (ELISA, Immunotech, Paris, France), and CRP (Kryptor – TRACE technology, ultrasensitive analysis, B.R.A.H.M.S. AG) were measured in duplicates. The intra- and inter- assay coefficients of variation were below 5%.

Statistical Analysis

The statistical analyses using SPSS software (version 12.0) for Windows (SPSS, Chicago, ILL) were performed. Analysis of covariance (ANCOVA) was used for statistical evaluation. The normal distribution of all data was examined using the Kolmogorov-Smirnov normality test to determine subsequent use of tests for statistical comparison. As variables were not normally distributed, the data were reported as means and interquartile range. Relations between the monitored indicators were evaluated by the Pearson's correlation coefficient and the Spearman's rank correlation. For all the tests, $p < 0.05$ was defined as statistically significant.

Results

Mean duration of CPB was 338.2 ± 44.4 min.; mean duration of cross-clamping time was 126.5 ± 20.5 min. and circulatory arrest time 39.9 ± 7.8 min. Extracorporeal circulation time was 338.2 ± 44.4 min.; duration of mechanical ventilation was 51.3 ± 36.3 h. PEA significantly decreased the mean pressure in the main pulmonary artery (mPAP) and pulmonary vascular resistance (PVR) and increased cardiac index (CI) within first 24 h after surgery (Table 1).

The mean preoperative PCT plasma levels were 0.24 ng/ml (interquartile range, 0.18–0.31), and PCT of all tested patients ranged within normal limit 24 h before

surgery. The mean preoperative IL-6 plasma concentrations were 16.4 ng/ml (10.9–30.1), the plasma levels of IL-6 were elevated in 6 of 32 patients 24 h before surgery. There was no correlation between preoperative plasma levels and hemodynamic parameters as well as between IL-6 and hemodynamic status.

Postoperative course of all tested patients was uncomplicated within 48 h after surgery. One patient died 9th day after surgery with the diagnosis of bronchopneumonia. The first clinical signs of inflammation were found 8th day after PEA. Following section proved this diagnosis. The only patient with poor prognosis didn't distinguish from uncomplicated course in PCT, cytokine and CRP dynamics in evaluated 48-h period after sternotomy.

Arterial blood samples analysis documents a transient initial decline of PCT (minimum 3 h after sternotomy) with subsequent elevation (Figure 1). Transient initial decline of PCT correlates significantly with decrease of hematocrit during hemodilution ($r = 0.78$, $p < 0.01$). Serum PCT levels increase postoperatively from 3 h reaching a peak level 24 h after sternotomy (2.04 ng/ml, 1.70–2.54). 'PCT levels were elevated in all 32 patients 24 h after sternotomy in comparison to preoperative levels. Peak levels of PCT were statistically significant higher in relation to both preoperative levels and control group ($p < 0.01$).

The similar course with initial decrease was revealed for IL-6, IL-8, sTNFRI, and CRP, too (Figures 2–5). Peak level of IL-6 was 6 h after CPB (521.9 ng/l,

Table 1 – Hemodynamic status early after PEA

	Preoperative	Postoperative (24 h)	P value
mPAP (mm Hg)	58.0 ± 11.7	25.7 ± 7.27	< 0.001
CI ($l.min^{-1}m^{-2}$)	1.8 ± 0.25	2.99 ± 0.43	< 0.001
PVR ($dynes.s.cm^{-5}$)	1161.5 ± 306.5	201.9 ± 99.8	< 0.001

Abbreviations: mPAP – mean pressure in the main pulmonary artery; CI – cardiac index; PVR – pulmonary vascular resistance.

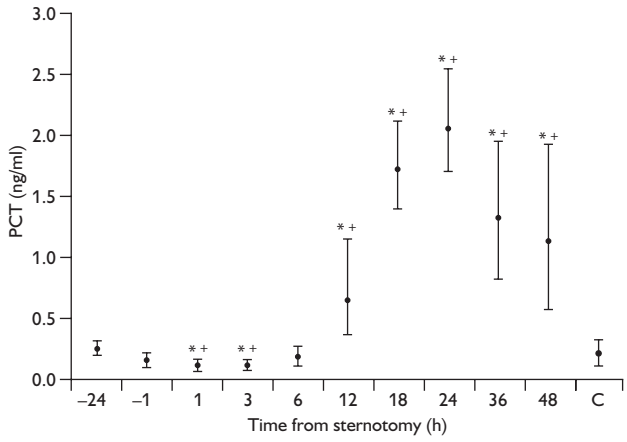

*Figure 1 – PCT evolution in perioperative period (mean values, interquartile range). C = Control group, * = Statistically significant differences to control group on $p < 0,05$. + = Statistically significant differences to preoperative values on $p < 0,05$. The same arrangement used for Figures 1–5.*

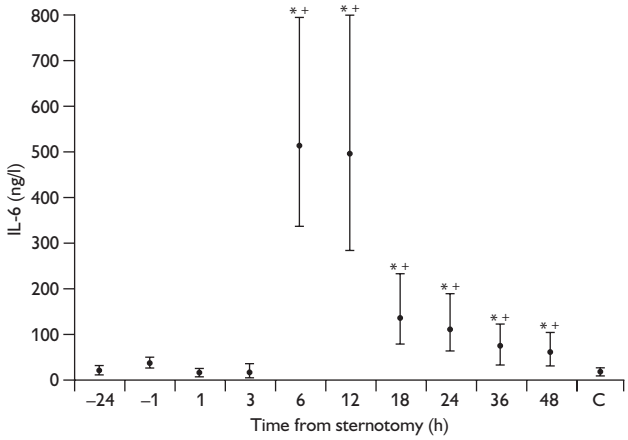

Figure 2 – IL-6 plasma concentrations in perioperative period (mean values, interquartile range)

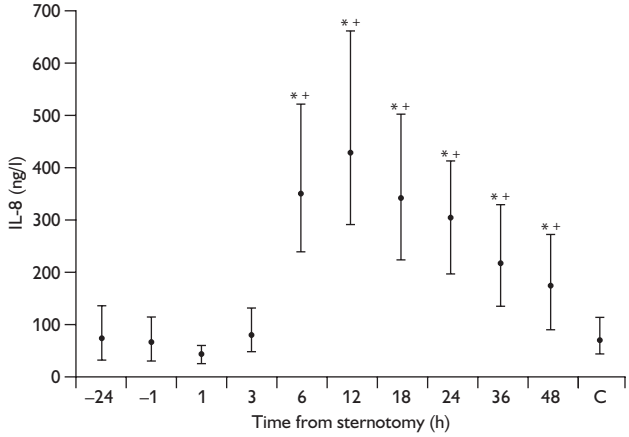

Figure 3 – IL-8 plasma concentrations in perioperative period (mean values, interquartile range)

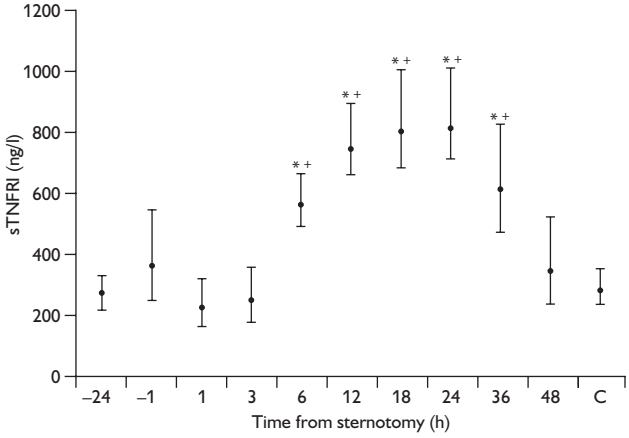

Figure 4 – sTNFRI plasma concentrations in perioperative period (mean values, interquartile range)

335.4–794.2). IL-8 culminated later – 12 h after sternotomy (431.2 ng/l, 289.9–659.1). The dynamics of sTNFRI were qualitatively similar but delayed with maximum 24 h after sternotomy (809.4, 711.2–1010.2). IL-1β elevation with maximum 6 h after start of surgery wasn't statistically significant. CRP showed prolonged elevation with a peak level in last sample (74.6 mg/l, 41.5–115.3).

Postoperative peak values of PCT and IL-6 closely correlated ($r = 0.81$, $p < 0.01$) (Figure 6), as well as peak values of PCT and CRP ($r = 0.72$, $p < 0.01$), and peak values of PCT and sTNFRI ($r = 0.58$, $p < 0.05$). Correlation between PCT and IL-1β wasn't significant on $p < 0.05$.

Discussion

Uncomplicated cardiac surgery induced a postoperative increase in serum PCT levels. Peak PCT levels were reached 24 h postoperatively. Maximal PCT values around 2.1 ng/ml are in agreement with other authors evaluated post-surgical

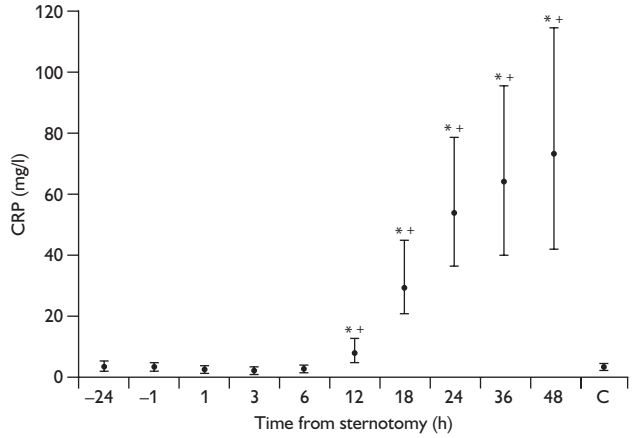

Figure 5 – CRP plasma concentrations in perioperative period (mean values, interquartile range)

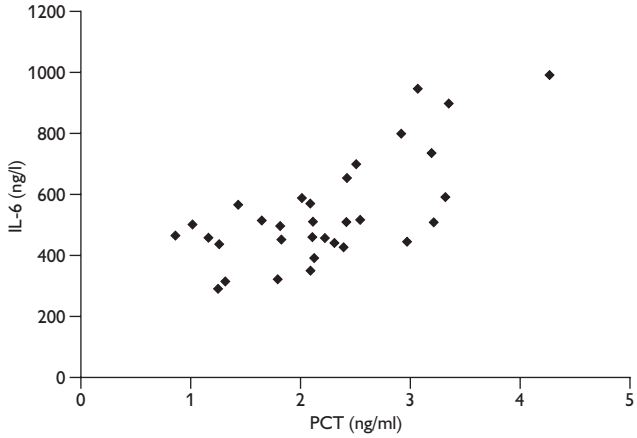

Figure 6 – Correlation of PCT and IL-6 peak values in post-surgical period ($r = 0.81$, $p < 0.01$)

course of PCT (range from 0.5 to 7.0 ng/ml) [7, 8, 9, 10]. Levels of proinflammatory cytokines as IL-1β, IL-6 and IL-8 also increase postoperatively, peaking at 6 to 12 h after PEA. CRP levels increase with extended course in comparison with both PCT and cytokines. The highest levels of CRP were detected in last blood samples 48 h after sternotomy.

PCT, a protein of 116 amino-acids with molecular weight of 13 kD, was discovered 32 years ago as a prohormone of calcitonin produced by C-cells of the thyroid gland. Since 1993 when its elevated level was found in patients with bacterial infection (4), PCT became an important marker in the detection and differential diagnostics of inflammatory states [11]. The highest plasma levels of PCT are achieved in acute bacterial infections and sepsis. Plasma levels are enhanced by the presence of a systemic inflammatory response. Local bacterial infections as well as abscesses do not raise plasma PCT significantly [12].

Surgical patients, especially those after cardiac surgery, represent a major diagnostic challenge in terms of identification of infectious complications. Meisner et al. [13] reported that PCT concentrations were increased moderately above the normal range in 32% of patients after minor and aseptic surgery, and these results were supported with other researchers including authors of this study [14]. Cardiac surgery leads to a more pronounced activation of cytokines than that of some other surgical procedure [6]. This cytokine 'burst' mediates a systemic response by the body's inflammatory system, well known as the systemic inflammatory response syndrome (SIRS) [15].

Several factors may influence the evolution of serum PCT levels after cardiac surgery in the absence of postoperative complications. The increase of PCT seems to be dependent on the surgical procedure, with more invasive procedures associated with higher PCT levels [16]. The source of PCT production in these conditions could be explained by non-specific cytokine liberation from the injured tissue [17]. The following decline of PCT levels to normal within a few days after surgery after an uncomplicated postoperative course corresponds to the half-life of PCT (18 to 24 h) in the absence of a further insult that may induce more PCT production.

Transient initial decline of PCT after surgery – statistically significant on $p < 0.05$ – is explained mostly by hemodilution. Significant correlation between PCT and hematocrit demonstrated these phenomena. We suspect the role of hemofiltration as another factor affecting PCT course in perioperative phase, too. Hemofiltration is used for hemoconcentration at the end of the operation. The tendency to transitory decrease after start of surgery was revealed in all tested parameters, but without statistical significance on $p < 0.05$.

Early differentiation between SIRS after PEA and the development of perioperative infection are crucial to enable appropriate antibiotic therapy to be started and to prevent subsequent complications. PCT values reported in infected patients are generally higher than in non-infected patients after PEA. However, it is

difficult to recommend cut-off points for discriminating patients according to the risk of sepsis syndrome after PAE. The dynamics of PCT levels over time may be more important than absolute values. Presented study was limited to uncomplicated surgical patients and these patients were compared to healthy controls. The comparisons to infected patients with the same surgical procedure will be useful for an assessment of the cut-off point for discriminating infection from uncomplicated course.

Conclusion

PEA induced systemic inflammatory response was accompanied with an elevation of plasma PCT levels. The highest concentrations of PCT were documented in blood samples 24 h after sternotomy. PCT culmination was delayed in comparison to IL-1β, IL-6, and IL-8 elevation. Positive correlation between PCT and IL-6 corresponded with the role of IL-6 in PCT induction in perioperative phase.

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