ORIGINAL ARTICLE

# Procalcitonin in preterm infants during the first few days of life: introducing an age related nomogram

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*Arch Dis Child Fetal Neonatal Ed* 2006;**91**:F283–F286. doi: 10.1136/adc.2005.085449

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Accepted 22 February 2006
**Published Online First 17 March 2006**

**Objective:** To determine normal concentrations of procalcitonin in preterm infants shortly after birth and to assess its accuracy in detecting bacterial infection.
**Methods:** Blood samples of 100 preterm infants were prospectively drawn during the first 4 days of life for determination of procalcitonin concentration. Infants were classified into four groups according to their sepsis status.
**Results:** Mean (SD) gestational age and birth weight were 32 (2.9) weeks and 1682 (500) g respectively. A total of 283 procalcitonin concentrations from healthy infants were plotted to construct nomograms of physiologically raised procalcitonin concentration after birth, stratified by two groups to 24–30 and 31–36 weeks gestation. The peak 95th centile procalcitonin concentration was plotted at 28 hours of age; values return to normal after 4 days of life. Only 12 infants were infected, and 13 of their 16 procalcitonin concentrations after birth were higher than the 95th centile, whereas samples taken at birth were lower. In a multivariable analysis, gestational age, premature rupture of membrane, and sepsis status influenced procalcitonin concentration independently, but maternal infection status did not.
**Conclusions:** The suggested neonatal nomograms of preterm infants are different from those of term infants. Procalcitonin concentrations exceeding the 95th centile may be helpful in detecting congenital infection, but not at birth.

Bacterial infection is one of the leading causes of morbidity and mortality in preterm infants.[1] As bacterial infection is one of the causes of preterm labour, many infants are evaluated to rule out systemic infection at birth and are treated with antibiotics, often unnecessarily. The decision whether to start and when to stop antibiotic treatment is not always clear, as many clinical signs of infection are seen in non-infectious conditions, such as respiratory distress syndrome and hypoglycaemia.

Procalcitonin (PCT) has been reported to be an effective laboratory tool for detecting disseminated bacterial infection in both adults and children.[2–5] It is a 116 amino acid prohormone, the serum concentration of which increases significantly during bacterial and fungal systemic infections. It is presumably secreted from the liver and circulating macrophages in response to endotoxins.[6] PCT concentration increases in the serum within two to three hours of the beginning of infection, peaking by 6–12 hours, and returning to normal concentrations within two days.[7] The short half life of PCT (20–24 hours)[8] enables not only rapid detection of infection but also titration of response to the treatment.

Various studies in children have found normal PCT concentrations to be <0.5 ng/ml, with a slight increase (0.5–2 ng/ml) during viral infections, non-infectious inflammation, stress situations, and focal bacterial infections.[4] In systemic bacterial and fungal infection, PCT increases to above 2 ng/ml or even over 50–100 ng/ml.[9] These values refer to infants and children,[4 9 10] but not to newborns during the first days of life when high physiological PCT concentrations are also measured in healthy newborns. Chiesa *et al*[9] proposed an age related 95th centile nomogram for term infants during the first 48 hours of life. No similar nomogram exists for preterm infants even though they potentially have different pharmacokinetics from term infants.

The objective of this study was to create an age related 95th centile nomogram of PCT concentration based on a sizeable population of preterm infants during the first 4 days of life.

## MATERIALS AND METHODS

### Population

The study was performed in a 30 bed neonatal intensive care unit at Shaare Zedek Medical Center, a university affiliated hospital in Jerusalem, Israel. The hospital deals with about 10 000 births a year; in 7.8% the babies weigh less than 2500 g. Most of the apparently well infants weighing over 2000 g are treated in the well baby nursery.

### Inclusion criteria

All preterm infants (36 weeks or less) admitted to the neonatal intensive care unit between September 2003 and January 2004 were eligible for enrolment. Parent's written informed consent was obtained according to the instructions of the local research ethics board.

### Sample collection and definition of sepsis groups

During the first 4 days of life, three to four blood samples for PCT determination were obtained from the enrolled infants, ideally, at birth and one sample on each of the subsequent three days. However, the exact time of sample collection varied according to the schedule of the routine tests. Infants were classified into sepsis groups at each sample time according to their clinical status (table 1): group1, proven sepsis; group 2, suspected sepsis; group 3, uncertain; group 4, no infection.[9 11] To minimise measurement bias, the rules for classification were stringent.

### PCT analysis

A 0.5 ml sample of blood was drawn into a tube containing EDTA. The samples were refrigerated for up to 24 hours, separated, and the plasma was frozen at −70°C.[13] The plasma was analysed for PCT concentration at the end of the study period, to ensure blinding of the doctors who classified the groups and managed the infants. The immunoluminometric assay, LUMItest (BRAHMS Diagnostica, Berlin, Germany), was used, as described in detail elsewhere.[9]

**Table 1** Definition criteria of four sepsis groups

| | |
|---|---|
| **Clinical signs of infection (positive if two or more of the following)** | |
| Respiratory | Any of: tachypnoea/hypopnoea/apnoea/cyanosis |
| Cardiovascular | Any of: bradycardia/tachycardia |
| Shock | Any of: poor perfusion/low blood pressure |
| Consciousness | Any of: irritability/lethargy/poor feeding/hypotonia/seizures |
| Others | Any of: hepatosplenomegaly/jaundice/fever |
| **Sepsis screen (positive if two or more of the following)** | |
| WBC | Any of: leucocytosis/leucocytopenia |
| ANC | Any of: neutrophilia/neutropenia |
| CRP | ⩾10 mg/l |
| Thrombocytes | Any of: thrombocytopenia/thrombocytosis |
| CSF | Any of: pleocytosis/positive Gram stain |
| Glucose | Any of: 40 mg/100 ml>Glucose>180 mg/100 ml |
| **Sepsis groups** | |
| 1: Confirmed sepsis | Positive blood culture/urine culture/pneumonia/cellulitis and clinical signs of infection |
| 2: Suspected sepsis | Clinical signs of infection and positive sepsis screen |
| 3: Uncertain | No fulfilment of the above criteria and no change in status after three days, requiring longer antibiotic therapy |
| 4: No infection | No fulfilment of the above criteria and clinical improvement within three days |

Reference values for laboratory results were determined as described elsewhere[8 12]. Poor perfusion was defined as corpus capillary refill >3 seconds.
WBC, White blood cells; ANC, absolute neutrophil count; CSF, cerebrospinal fluid; CRP, C reactive protein.

### Data collection

Basic information and data on pregnancy, delivery, and maternal infections were collected shortly after birth. The presence of maternal infection at the time of the delivery (dichotomous variable) was defined as fever or documented chorioamnionitis or positive placental culture. Neonatal data were collected prospectively, including clinical condition, laboratory results, treatment, and associated illness.

### Statistical analysis

A sepsis group was assigned for each infant at the time of each PCT sampling. Data are presented as mean (SD) or median (interquartile range) and compared using unpaired Student's *t* test or Wilcoxon rank sum test, as appropriate for the distribution normality. A multivariable regression analysis for repeated measures was used to determine the relative contributions of several explanatory variables on the PCT concentrations at 0–96 hours after birth, while controlling for time. The following explanatory variables were initially entered and eliminated using a backward stepwise approach: percentage of oxygen dependency, sepsis group (four point ordinal scale), maternal intravenous antibiotic therapy (dichotomous), maternal infection (dichotomous), hours of premature rupture of membranes, birth weight, and gestational age. PCT concentrations were log transformed to meet the assumptions of the model. Significance was defined as $p<0.05$. To construct the age related 95th centile nomogram for PCT concentration, samples were stratified to nine time intervals (at birth and every 12 hours thereafter). For each interval, the 95th and 50th centiles of the serum PCT concentrations taken during that time interval were calculated, and smoothed centile curves were drawn.[14–16] This method was used to ensure that infants are not represented more than once in each time window, thereby contributing only once to each of the nine centile values. We believe that this time frame is sensitive enough to detect PCT changes over time. Statistical analysis was performed using SAS for Windows version 9.

## RESULTS

A total of 332 blood samples were drawn during the first 4 days of life from 100 preterm infants, and PCT concentrations determined. These infants represent 90% of the total admissions of preterm infants to the neonatal intensive care unit during that time. Mean (SD) gestational age and birth weight was 32 (2.9) weeks (range 24–36) and 1682 (500) g (479–2615) respectively. There were 30 and 70 infants of 24–30 and 31–36 weeks of gestation respectively. The mean number of samples taken from each infant in the 24–30 and 31–36 weeks of gestation was 3.5 and 3.7 respectively (Student's *t* test, $p>0.05$). No deaths were recorded during the first week of life in any of the sepsis groups.

Five (2%) of the 332 samples were obtained from infants classified as septic group 1, 15 (4%) as group 2, 29 (9%) as group 3, and 283 (85%) as group 4. The 283 PCT values for group 4 were plotted to construct an age related nomogram from birth to 96 hours of age, stratified by gestational age (fig 1). The peak 95th centile of PCT concentration was plotted at 28 hours of age. Three of the 283 samples were excluded from the plot because they were exceedingly high and diverted the curve: PCT concentration of 36, 27, and 28 ng/ml at 28, 32, and 51 hours of life respectively. The infant with the PCT concentration of 36 ng/ml developed *Klebsiella pneumonia* sepsis two days later and was treated successfully. The other two infants with the outlying values were otherwise healthy.

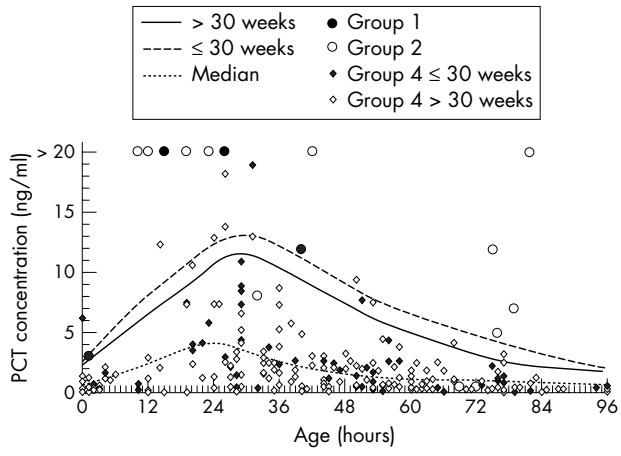


**Figure 1** Age related nomogram for procalcitonin (PCT) concentrations during the first 4 days of life in preterm infants. PCT concentrations of 283 samples drawn from healthy preterm infants (group 4), four samples from infants with confirmed sepsis (group 1), and 12 from infants with suspected sepsis (group 2) were plotted from birth to 96 hours of life. The continuous and dashed lines represent the 95th centile of infants 24–30 and 31–36 weeks gestation respectively. The dotted line represents the median of all group 4 samples. For simplicity, eight values for groups 1 and 2 that exceeded 20 ng/ml were plotted as >20 ng/ml (26 and 34 ng/ml in group 1 and 24, 36, 110, 58, 202, and 56 ng/ml in group 2).

In this cohort, only three infants (3%) were confirmed with sepsis (group 1), contributing to five samples of PCT, and a further nine (9%) with suspected sepsis (group 2), contributing to 15 samples. Four of the 20 samples (groups 1 and 2) were drawn at birth, all of which had normal PCT concentration (<1 ng/ml). The other 16 samples are plotted on the curve (fig 1). Only three of the 29 samples of group 3 were higher than the 95th centile (median 1.02 ng/ml (interquartile range 4.56)). Although this group represents cases of uncertainty, their PCT concentrations are similar to the non-infected group.

Median (interquartile range) PCT concentrations of the septic groups on the first and second day of life were significantly higher (Wilcoxon rank sum test) than those of the non-septic groups (42 ng/ml (24–110) *v* 1.2 ng/ml (0.3–4.1) for day 1, $p<0.01$; and 22 ng/ml (9–45) *v* 2.5 ng/ml (1.2–5.3) for day 2, $p<0.01$). However, the small number of samples drawn from infected infants prevented systematic comparisons between the septic and the non-septic infants and stratification by gestational age.

In the multivariable regression for repeated measures, the total PCT concentrations during the first 96 hours of life were independently affected by the sepsis group ($p<0.0001$ for the adjusted coefficient), hours of premature rupture of membranes ($p = 0.038$), and gestational age ($p = 0.013$). PCT concentrations were not affected by maternal infection ($n = 6$), maternal antibiotic treatment ($n = 30$), birth weight, or percentage of oxygen consumption (all $p>0.05$).

## DISCUSSION

In this study we constructed an age related nomogram for PCT concentration in a large cohort of non-infected preterm infants. As gestational age, and not birth weight, independently affected PCT concentration in the multivariable analysis, we stratified the nomogram according to gestational age. Other studies failed to show similar differences in PCT concentration according to gestational age,[9–11 17] possibly because the study populations were composed of predominantly larger infants with only small numbers of very low birthweight infants.

Studies in term infants[9 11 17–20] reported a physiological rise in PCT concentration during the first 48 hours of life, with a peak of 20 ng/ml at 24 hours. A preliminary report on a small number of preterm infants (<1500 g) suggested that the physiological rise in PCT concentration of preterm infants may be longer than in term infants.[21] In our cohort, the peak value was measured at 28 hours after birth but was lower than in term infants. The results from our stratified cohort and the data from term infants suggest that PCT concentrations decrease with prematurity possibly because of the immature immune system. The values in our study returned to normal only after 72–96 hours of life rather than 48–72 hours in term infants as described by Chiesa *et al*.[9] This is understandable, as the excretion mechanisms in preterm infants are less mature than in term infants, resulting in different kinetics.

As reported by others,[21] maternal infectious status (reflected by fever, chorioamnionitis, positive placental culture, and intravenous antibiotic therapy) had no effect on PCT concentration, suggesting that PCT does not cross the placenta. This is in contradiction with a report that showed increased PCT in response to maternal PCT concentrations.[18] Hours of premature rupture of membranes, however, did influence PCT concentration in our cohort, independently of the sepsis group.

Most studies that looked at the kinetics of PCT during the first few days of life (predominantly term infants) failed to show high PCT concentration at birth in septic newborns.[9 17–20 22] Similarly, all six samples that were drawn at birth from our septic infants (groups 1 and 2) were normal (PCT<1 ng/ml), suggesting that PCT may be useful in diagnosing sepsis only after a few hours of life, allowing the production of PCT. The clinical decision as to when to discontinue antibiotic therapy in a neonate with negative cultures who has become asymptomatic is often a difficult one. Therefore, although our results preclude the use of PCT concentration in the decision of whether to start antibiotic therapy at birth, it may prove useful in the decision as to when to discontinue antibiotics in questionable cases. However, the small number of infected infants limits our discussion to descriptive analysis only.

Conflicting results on the sensitivity and specificity of PCT as a useful marker of sepsis in preterm infants have been reported.[19 21 23] Janota *et al*[21] found the specificity and sensitivity of PCT concentration in detecting early sepsis in 37 preterm infants (<1500 g) to be only 75%. Similar reports of low sensitivity were found in 162 infants after birth (term and preterm together).[19] PCT concentrations higher than 2.5 ng/ml have been found to have a negative predictive value of 93% in detecting sepsis in 120 preterm and term infants during the first 3 days of life.[17] The low specificity could be attributed to several factors. As we have shown, PCT concentrations are raised during the first 4 days of life even in normal premature neonates. To eliminate this bias, it is important to adhere to the nomogram proposed in this study for preterm infants. From the small number of infected infants in our study, all samples of group 1 and nine of 12 samples of group 2, taken after birth, were higher than the proposed nomograms. In contrast with other studies,[20 23 24] oxygen dependency was not found to influence PCT concentrations independently. However, as only six infants in our cohort had severe hypoxia ($FIO_2 >0.5$), our results may be flawed, and therefore high concentrations of PCT in severely hypoxic preterm infants should be analysed with caution because of the probable influence of hypoxia previously found.

A major limitation of this study is the small number of infants who were diagnosed with congenital sepsis. This prevented systematic measurement of the sensitivity and specificity of the nomogram. However, the description of the several samples obtained from the infected groups after birth shows promising results for further studies. On the other hand, the strengths of the study lie in its prospective nature, in the large sample size of preterm infants, and in the

### What is already known on this topic

- Procalcitonin is an effective laboratory tool for detecting disseminated bacterial infection in both adults and children; normal concentrations in children are <0.5 ng/ml, but these values are not valid after birth, when procalcitonin is physiologically increased
- An age related nomogram is available for term infants during the first 3 days of life but not for preterm infants

### What this study adds

- This study introduces an age related nomogram for procalcitonin concentrations during the first 4 days of life of preterm infants, stratified by gestational age
- Preliminary results for several infected newborns from our cohort suggest that the nomogram is effective, but not at birth

stringent criteria used to differentiate septic from non-septic infants.

In conclusion, we developed a nomogram for PCT concentrations for preterm newborns during the first 4 days of life. It would seem to be important to use the suggested preterm nomogram, as values differ from the published reference curve for term infants. Preliminary data on PCT concentrations in our infected infants are encouraging with respect to their ability to detect sepsis. Further studies are needed to confirm the accuracy of the nomogram.

## ACKNOWLEDGEMENTS

BRAHMS Diagnostica provided the testing kits for PCT determination but was not involved in any part of the data collection, analysis, or the preparation of the manuscript. We thank the nurses of the neonatal intensive care unit who meticulously collected the blood samples, and Professor Arthur Eidelman for his help in preparing the manuscript.

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Financial support: BRAHMS Diagnostica supplied the testing kits for PCT value determination but was not involved in any aspects of this study and the manuscript preparation.

Competing interests: none declared

## REFERENCES

1 **Kaufman D**, Fairchild KD. Clinical microbiology of bacterial and fungal sepsis in very-low-birth-weight infants. *Clin Microbiol Rev* 2004;**17**:638–80.
2 **Chiesa C**, Pacifico L, Mancuso G, *et al.* PCT in pediatrics; overview and challenge. *Infection* 1998;**26**:236–41.
3 **Assicot M**, Gendral D, Carsin H, *et al.* High serum PCT concentration in patients with sepsis and infection. *Lancet* 1993;**341**:515–18.
4 **van Rossum AM**, Wulkan RW, Oudesluys-Murphy AM. Procalcitonin as an early marker of infection in neonates and children. *Lancet Infect Dis* 2004;**4**:620–30.
5 **Vazzalwar R**, Pina-Rodrigues E, Puppala BL, *et al.* Procalcitonin as a screening test for late-onset sepsis in preterm very low birth weight infants. *J Perinatol* 2005;**25**:397–402.
6 **Dandona P**, Nix D, Wilson MF, *et al.* Procalcitonin increase after endotoxin injection in normal subjects. *J Clin Endocrinol Metab* 1994;**79**:1605–8.
7 **Meisner M**, Schmidt J, Huettner H, *et al.* The natural elimination rate of procalcitonin in patients with normal and impaired renal function. *Intensive Care Med* 2000;**26**(suppl 2):212–16.
8 **Oski F**, Naiman J. *Hematologic problems in the newborn.* Philadelphia: WB Saunders, 1982:1380–1.
9 **Chiesa C**, Panero A, Rossi N, *et al.* Reliability of procalcitonin concentrations for the diagnosis of sepsis in critically ill neonates. *Clin Infect Dis* 1998;**26**:664–72.
10 **Gendrel D**, Assicot M, Raymond J, *et al.* Procalcitonin as a marker for the early diagnosis of neonatal infection. *J Pediatrics* 1996;**128**:570–3.
11 **Monneret G**, Labaune JM, Isaac C, *et al.* Procalcitonin and C-reactive protein levels in neonatal infections. *Acta Paediatr* 1997;**86**:209–12.
12 **Manroe BL**, Weinberg AG, Rosenfeld CR, *et al.* The neonatal blood count in health and disease-reference values for neutrophilic cells. *J Pediatr* 1979;**95**:89–98.
13 **Meisner M**, Tschaikowsky K, Schnabel S, *et al.* Procalcitonin-influence of temperature, storage, anticoagulation and arterial or venous asservation of blood samples on procalcitonin concentration. *Eur J Clin Chem Clin Biochem* 1997;**35**:597–601.
14 **Kaplan M**, Hammerman C, Feldman R, *et al.* Predischarge bilirubin screening in glucose-6-phosphate dehydrogenase-deficient neonates. *Pediatrics* 2000;**105**:533–7.
15 **Sarici SU**, Yurdakok M, Serdar MA, *et al.* An early (sixth-hour) serum bilirubin measurement is useful in predicting the development of significant hyperbilirubinemia and severe ABO hemolytic disease in a selective high-risk population of newborns with ABO incompatibility. *Pediatrics* 2002;**109**:e53.
16 **Schwahn C**, Meisel P, Biffar R, *et al.* Dental nomograms for benchmarking based on the study of health in Pomerania data set. *J Clin Periodontol* 2004;**31**:1099–105.
17 **Guibourdenche J**, Bedu A, Petzold L, *et al.* Biochemical markers of neonatal sepsis: value of PCT in the emergency setting. *Ann Clin Biochem* 2000;**39**:130–5.
18 **Assumma M**, Signore F, Pacifico L, *et al.* Serum PCT concentration in term delivering mothers and their healthy offspring: a longitudinal study. *Clin Chem* 2000;**46**:1583–7.
19 **Franz AR**, Kron M, Pohlandt F, *et al.* Comparison of PCT with IL-8, CRP and differential WBC count for the early diagnosis of bacterial infections in newborn infants. *Pediatr Infect Dis J* 1999;**18**:666–71.
20 **Koskenvuo MM**, Irjala K, Kinnala A, *et al.* Value of monitoring serum PCT in neonates at risk of infection. *Eur J Clin Microbiol Infect Dis* 2003;**22**:377–8.
21 **Janota J**, Stranak Z, Belohlavkova S, *et al.* Postnatal increase of PCT in premature newborns is enhanced by chorioamnionitis and neonatal sepsis. *Eur J Clin Invest* 2000;**31**:978–83.
22 **Marchini G**, Berggren V, Djilali-merzoug R, *et al.* The birth process initiates an acute phase reaction in the fetus newborn infant. *Acta Paediatr* 2000;**89**:1082–6.
23 **Monneret G**, Labaune JM, Isaac C, *et al.* Increased serum PCT levels are not specific to sepsis in neonates. *Clin Infect Dis* 1998;**27**:1559–61.
24 **Lapillonne A**, Basson E, Monneret G, *et al.* Lack of specificity of PCT for sepsis diagnosis in premature infants. *Lancet* 1998;**351**:1211–12.