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Procalcitonin, IL-6, IL-8, IL-1 receptor antagonist and C-reactive protein as identificators of serious bacterial infections in children with fever without localising signs

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Abstract Fever without localising signs in very young children remains a diagnostic problem. Until present, a clinical scoring system combined with leucocyte count, urine analysis and determination of CRP are recognised as being helpful to identify patients at risk of serious bacterial illness. In this study we asked the question whether the determination of procalcitonin (PCT), interleukin (IL)-6, IL-8 and interleukin-1 receptor antagonist (IL-1Ra) was superior to these commonly used markers for the prediction of a serious bacterial infection (SBI). Children, 7 days to 36 months of age, with a rectal temperature above 38 °C and without localising signs of infection were prospectively enrolled. For each infant, we performed a physical examination, a clinical score according to McCarthy, a complete white cell count, an urine analysis and a determination of CRP. We further determined PCT, IL-6, IL-8, and IL-1Ra concentrations and compared their predictive value with those of the usual management of fever without localising signs. Each infant at risk of SBI had blood culture, urine and cerebrospinal fluid cultures when indicated, and received antibiotics until culture results were available. A total of 124 children were included of whom 28 (23%) had SBI. Concentrations of PCT, CRP and IL-6 were significantly higher in the group of children with SBI but IL-8 and IL-1Ra were comparable between both groups. PCT showed a sensitivity of 93% and a specificity of 78% for detection of SBI and CRP had a sensitivity of 89% and a specificity of 75%.

Conclusion Compared to commonly used screening methods such as the McCarthy score, leucocyte count and other inflammatory markers such as interleukin-6, interleukin-8 and interleukin-1 receptor antagonist, procalcitonin and C-reactive protein offer a better sensitivity and specificity in predicting serious bacterial infection in children with fever without localising signs.

Key words Bacterial infection · C-reactive protein · Interleukin-6 · Paediatrics · Procalcitonin

Abbreviations *DMSA* dimercaptosuccinic acid · *IL* interleukin · *IL-1Ra* interleukin-1 receptor antagonist · *PCT* procalcitonin · *ROC* receiver operating characteristics · *SBI* serious bacterial infection

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Introduction

Fever without localising signs in young children remains a difficult diagnostic problem, since clinical signs and symptoms are often unreliable predictors of a serious bacterial infection (SBI) which requires rapid therapeutic intervention with intravenous antibiotic therapy. Many clinical studies [3, 10, 14, 18] have addressed this problem and the combination of a clinical scoring system such as that of McCarthy [19] combined with a total and differential leucocyte count and a determination of the CRP concentration are commonly used screening methods. More recently, the concentration of procalcitonin (PCT), a prohormone of calcitonin [2, 5, 15] and of cytokines such as interleukin-6 (IL-6), interleukin-8 (IL-8), as well as the cytokine antagonist interleukin-1 receptor antagonist (IL-1Ra) [8, 11, 16, 22] have been reported to lead to a better diagnostic accuracy when used in specific clinical situations. For instance, IL-1Ra and IL-6 were superior to circulating intercellular adhesion molecule 1 and CRP in predicting neonatal sepsis [16], whereas PCT was the best predictor in other clinical situations such as bacteraemic infections in adults or children [1, 6, 9, 12, 13]. The time course of changes among these various parameters is very different: IL-6 reaches peak concentrations in bacteraemic patients several hours before the rise in CRP concentration occurs [4, 7]. PCT starts to rise 2 h after experimental endotoxin administration and reaches peak levels within 6–8 h [4, 7]. From many studies [14, 25], it is known that CRP becomes a relatively reliable predictor of SBIs in fever of more than 12 h duration. Therefore it is to be expected that the predictive value of each of these markers depends on the duration between the invasion by the infectious agent and the concentration of the inflammatory marker. Furthermore, the spectrum of infectious agents possibly involved in the generation of fever varies from newborn to children below 3 years of age or even older children and adults. The diagnostic accuracy of these parameters may therefore well vary in these different clinical situations.

Until present, we used the clinical scoring system of McCarthy [19] combined with a total and differential leucocyte count and CRP concentration to decide whether a patient with fever without localising signs below 3 years of age (but beyond 7 days of age) required further diagnostic work-up and antimicrobial therapy. In this study, we asked the question, whether the determination, in addition to the previously used parameters, of PCT, IL-6, IL-8 or IL-1Ra offered an advantage in terms of sensitivity and specificity, with which a SBI could be predicted.

Patients and methods

The study protocol was approved by the Ethical Committee of the Department of Paediatrics, University Hospitals of Geneva.

Children aged 7 days to 36 months of age consulting the Emergency Department of the University Children's Hospital of Geneva with a rectal temperature above 38 °C and without localising signs of infection in their history or at physical examination were prospectively enrolled. Each infant was examined by a paediatric resident who took a complete history, performed a physical examination, recorded the degree and duration of fever and determined a clinical score according to McCarthy [19]. This scoring system allows to identify seriously ill children. Children with fever lasting longer than 7 days, neonates of less than 1 week and all children treated with antibiotics during the 2 previous days as well as those with known immunodeficiencies (like neutropenia due to chemotherapy or HIV-infected children) were excluded. All children had a urine analysis and blood drawn for a white blood cell count and for determination of CRP, PCT, IL-6, IL-8 and IL-1Ra concentrations.

White blood cell count and CRP determination were performed in blood samples mixed with EDTA. CRP was determined by a rapid immunometric kit method (Nycocard CRP). For the determination of PCT, IL-8, IL-6 and IL-1Ra, the blood sample was centrifuged to sediment red cells and serum frozen at −20 °C within 1 h. PCT was measured in a blinded manner by an immunoluminometric assay (Lumitest PCT, Brahms Diagnostica, Berlin). This assay requires 20 µl of sample and can be performed within 2 h. IL-6, IL-8 and IL-1Ra were measured using commercially available quantitative sandwich enzyme immunoassays (R&D Systems Europe, UK). Due to the limited amount of plasma available, samples were diluted in sample buffer prior to the assays. IL-6 was measured using a highly sensitive immunoassay specific for IL-6 whose sensitivity is 0.1 pg/ml (Quantikine HS human IL-6). Serum samples were assayed at a 1/40 dilution and therefore the limit of detection was 4 pg/ml. Coefficients of variation are less than 12% and 30% for intra-assay and interassay variation respectively. IL-8 was measured using an immunoassay specific for IL-8 (Quantikine human IL-8). The sensitivity of the assay is 8 pg/ml. Serum samples were assayed at a 1/5 dilution and the limit of detection was then 40 pg/ml. Coefficients of variation are less than 7% and 10% for intra-assay and interassay variations respectively. IL-1Ra was measured using an immunoassay specific for IL-1Ra. The sensitivity of the assay is 14 pg/ml. Serum samples were assayed at a 1/20 dilution and the limit of detection was then 280 pg/ml. Coefficients of variation are less than 7% for intra-assay and interassay variations.

Children with leucocytes $>15{,}000/mm^3$, band counts $>1500/mm^3$, leucocyturia or CRP >40 mg/l had a blood culture, an urine culture, and a spinal tap when meningitis was suspected. They also received antibiotics for 48–72 h until the results of the cultures were known. All children had a clinical follow-up with physical examination by a paediatrician within the following 48 h or by telephone contact. The diagnosis was registered at the end of the clinical follow-up. Infections requiring intravenous antibiotic therapy, such as bacteraemia (positive blood culture), pyelonephritis (positive urine culture with $>10^4$ colonies/ml and a positive technetium 99M-dimercaptosuccinic acid (DMSA) renal scintigraphy at 4 days with a reversible cortical defect on the control scintigraphy at 90 days), lobar pneumonia (radiological diagnosis of lobar infiltrate by the radiologist in a blinded manner), meningitis (pleocytosis of >5 cells/µl and a positive culture of cerebrospinal fluid) or osteo-arthritis were defined as SBI. The remaining patients suffered from infections classified as benign for the purpose of this study on the basis that they did neither require oral antibiotic therapy at follow-up (probable viral infections) nor parenteral therapy for infections such as acute otitis media, lower urinary tract infection (negative renal DMSA scintigraphy), gastroenteritis or adenitis (focal infections).

Statistics

Demographic characteristics and laboratory values of children with and without SBI were compared using the Fischer exact test for frequencies, the Student *t*-test for normally distributed continuous variables and the Mann-Whitney U test otherwise. The sensitivity,

specificity, negative and positive predictive values for the detection of a SBI were determined for the McCarthy score and the different laboratory parameters. Binominal exact 95% confidence intervals were calculated for sensitivity and specificity. The diagnostic accuracy of the different parameters and the best cut off points were determined with a receiver operating characteristics (ROC) curve. For PCT and CRP, likelihood ratios were determined. The likelihood ratio for a positive test expresses the odds that a positive test result would be expected in a patient with (as opposed to one without) a SBI, and is calculated as sensitivity/(1 – specificity) [24]. In order to calculate 95% confidence intervals around the likelihood ratio, a Taylor series expansion was used to determine the variance of this ratio [21].

Results

A total of 133 children were included from March 1998 to August 1999. Nine children were excluded because they did not present at clinical follow-up or suffered from immunodeficiencies and the data of 124 children were analysed. Patients with and without serious infections were comparable for the median age, the height of fever and the McCarthy score [19] with a slight increase in the median duration of fever for patients with SBI (Table 1). In children with fever above 40 °C recorded during this febrile episode, there was a tendency to an increased percentage of SBI (43% versus 23%, $P = 0.06$). Of 62 (50%) children who were hospitalised, 57 (92%) were treated with antibiotics (54 i.v. and 3 i.m.) and among those sent home, antibiotics were prescribed for 20 (32%) (12 oral, 4 i.m. and 4 i.v.). A blood culture was performed in 91 (73%) children, an urine culture specimen in 103 (83%) and a cerebrospinal fluid culture in 13 (11%).

There was a comparable incidence of SBI in infants <3 months ($8/31 = 26\%$), in those 3–12 months old ($10/49 = 20\%$) and >12 months ($10/44 = 23\%$) ($P = 0.8$). The final diagnosis was: SBI in 28 children (23%) (4 bacteraemia, 19 pyelonephritis, 5 lobar pulmonary condensation), focal bacterial infection in 13 children (10%) (7 cystitis, 4 otitis, 1 adenitis, 1 *Campylobacter* gastro-enteritis) and probable viral infection in 83 children (67%) (negative culture and no signs for focal infection at clinical follow-up).

The concentrations of PCT, CRP and IL-6 were significantly higher in the group of children with SBI ($P < 0.001$). IL-8 and IL-1Ra values were comparable between both groups (Table 1). PCT concentrations were comparable in the group of probable viral compared to focal infection; median (range): 0.40 ng/ml (0.11–43.30 ng/ml) versus 0.44 ng/ml (0.16–1.00 ng/ml), respectively.

Box plots of the distribution of PCT, CRP and IL-6 concentrations are shown in Fig. 1. PCT and CRP effectively discriminated between benign infections and SBI with a slight advantage for PCT (area under ROC curve 0.8824 and 0.8767, respectively) and both were superior to IL-6 (area under the ROC curve: 0.7781), IL1-Ra (0.5327) and IL-8 (0.4404). The likelihood ratio for a positive PCT was 4.24 (95% CI: 2.58–5.90) and for a positive CRP 3.57 (95% CI: 2.25–4.89). In Table 2, the sensitivity, specificity and predictive values of other parameters routinely used in the management of children with SBI are compared with those of PCT and cytokines. The PCT concentration had the best sensitivity (93%) and specificity (78%). The four bacteraemic patients had PCT values superior to 3 ng/ml with a value of 360 ng/nl for a patient with an *Escherichia coli* bacteraemia. Among the 28 children with SBI, 2 had a PCT concentration below the cut-off level (0.9 ng/ml). CRP had a sensitivity of 89% and a specificity of 75%. The other parameters used routinely (total and differential leucocyte count, McCarthy score) had a lower sensitivity ranging from 20%–68%. When PCT and CRP were associated, the sensitivity of the combination increased but the specificity decreased (Table 2). Table 3 compares the predictive value of PCT and CRP between subjects below or above 12 months of age, both parameters had a trend towards poorer specificity in older children.

Discussion

Among our population of 124 children below the age of 3 years presenting at the emergency room with fever without localising signs, 28 suffered from a SBI such as bacteraemia, pyelonephritis or lobar pneumonia. In terms of age and level of the initial temperature, the groups were comparable (Table 1). The sensitivity and the specificity for predicting a SBI were calculated for the various inflammatory markers measured, the PCT values were better than CRP. However, in this study, both tests were not significantly different, this could be

Table 1 Comparison of different parameters and of the mean concentrations of PCT, CRP, IL-6, IL-8 and IL-1Ra (as mean + standard error or median and range) between children with benign infections and SBI. (*ND* non detectable, *NS* not significant)

	Benign infection ($n = 96$)	SBI ($n = 28$)	*P*
Age (months)	10.9 ± 0.9	11.2 ± 1.8	NS
Fever duration (h)	24 (1–240)	27 (2–140)	0.02
Temperature (C)	39.0 ± 0.1	39.1 ± 0.2	NS
PCT (ng/ml)	0.40 (0.11–43.3)	3.6 (0.25–364)	<0.01
CRP (mg/l)	20 (10–200)	108 (10–200)	<0.01
IL-6 (pg/l)	14.7 (1.5–801)	69 (10–801)	<0.01
[a]IL-8 (pg/l)	ND (ND-3869)	43.5 (ND-145)	NS
Il-1Ra (pg/l)	5173 (435–74868)	8381 (689–49917)	NS

[a] IL-8 values were below the detection level (40 pg/ml) in 50 subjects with a benign infection and in seven subjects with a serious infection

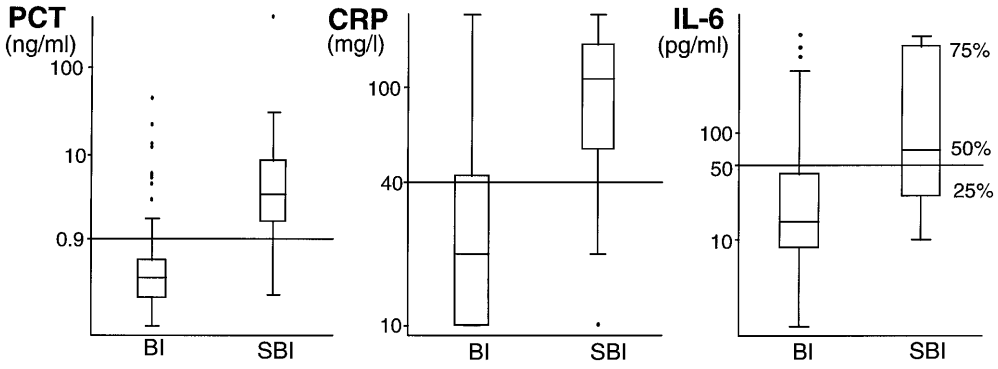

Fig. 1 Logarithm of PCT, CRP, IL-6 concentrations in children with SBI or benign infections (*BI*). The box plots show the 25th, 50th and 75th percentiles of the distribution. The upper and lower limits correspond to the highest and lowest value within 1.5 times the interquartile range above the 75th percentile and below the 25th percentile respectively. Points outside these limits are outliers and are graphically represented

Table 2 Sensitivity, specificity and predictive values of the different markers for the prediction of a SBI

Parameter	Sensitivity (95% CI)	Specificity (95% CI)	Negative predictive value (%)	Positive predictive value (%)
PCT (0.9 ng/ml)[a]	93 (77–99)	78 (69–86)	97	55
CRP (40 mg/l)[a]	89 (72–98)	75 (65–83)	96	51
Leucocytes $>15{,}000/mm^3$	68 (48–84)	77 (67–85)	89	46
Band $>1500/mm^3$	29 (13–49)	91 (83–96)	81	46
McCarthy score >10	20 (3–56)	86 (76–93)	79	29
IL-6 (50 pg/l)[a]	79 (59–92)	66 (55–75)	91	40
IL-1Ra (9500 pg/l)[a]	71 (51–87)	63 (52–72)	88	36
IL-8 (70 pg/l)[a]	38 (15–65)	79 (69–87)	81	34
PCT (0.9 ng/ml)[a] or CRP (40 mg/l)[a]	96 (82–100)	67 (56–76)	98	46
PCT (0.9 ng/ml)[a] or Leucocytes $>15{,}000/mm^3$	100 (88–100)	62 (51–71)	100	43

[a] Cut off level

Table 3 Sensitivity, specificity and predictive value (%) for a SBI of PCT and CRP in relation to age

Parameter	Age (months)	Sensitivity	Specificity	Negative predictive value (%)	Positive predictive value (%)
PCT	<12 (n = 80)	94	87	98	68
(0.9 ng/ml)[a]	>12 (n = 44)	90	62	96	41
CRP	<12 (n = 80)	94	84	98	63
(40 mg/l)[a]	>12 (n = 44)	80	59	91	36

[a] Cut off level

due to the small size of the study and larger cohorts of patients would be necessary to identify a statistically significant advantage of PCT over CRP. The other markers IL-6, IL-8 and IL-1Ra as well as the total and differential leucocyte count, were all below PCT and CRP. Only 2 infants among the 28 with a SBI had a PCT concentration below the cut-off value of 0.9 ng/ml. At follow-up examination, one had pyelonephritis with a minimal renal lesion on DMSA scintigraphy and another a classical pyelonephritis. For the former patient, it is possible that the inflammatory reaction at the moment when PCT was determined was too weak. In children below the age of 1 year, in whom clinical signs and symptoms are often unreliable, PCT and CRP were more predictive than in older children (Table 3).

Interestingly, IL-1Ra, IL-6 or IL-8 concentrations did not offer any advantage over PCT and performed less well than CRP. In newborn children, Küster et al. [16] showed that IL-1Ra and IL-6 increased significantly earlier than CRP, but no data on PCT were obtained, whereas in a recent study in newborns, PCT determination detected early-onset sepsis with a sensitivity of 93% and a specificity of 98% [6]. These excellent results were not confirmed by another group reporting on newborns [17]. However, a population of hospitalised newborns, in whom clinical signs and symptoms as well as inflammatory markers were studied prospectively, is not comparable to our study population in which the clinician saw the child only at the time when it was brought in by the parents and after onset of fever. Furthermore, hospitalised newborn infants very rarely suffer from post-natally acquired viral diseases in contrast to older children presenting with fever without localising signs. Our study clearly does not address the

question in newborn infants, but it is the first one in children with fever without localising signs in which PCT, IL-6 and IL-1Ra, are measured simultaneously.

How can we explain the striking difference between PCT (sensitivity 93%, specificity 78%) and CRP (sensitivity 89%, specificity 75%) on the one hand and IL-6 (sensitivity 79%, specificity 66%) and IL-1Ra (sensitivity 71%, specificity 63%) on the other ? First, for IL-6 and IL-1Ra, their concentrations indeed increase early, i.e. within 6 h, but it may already have dropped when our patients presented at the emergency room, whereas PCT remains longer in the circulation [7]. Second, it is possible that PCT, more than IL-1Ra and IL-6, allows to distinguish between bacterial and viral infections, which are frequent in groups of children such as the one we studied. IL-6, IL-8 and IL-1Ra are indeed activated by various viruses [23, 26, 27, 28]. Finally, PCT is a molecule with a remarkable stability without significant influence of the blood sampling technique and of repeated freezing and thawing cycles [20] which is less the case for the cytokines.

On the basis of our data, PCT offers only a modest advantage over CRP, which at present is more easily measurable in an outpatient setting. If CRP is used by taking into account the kinetics of parameters of inflammation, it still offers a good prediction of a SBI. However, PCT determination has recently been simplified and, after careful testing with regard to accuracy of the rapid test, it may offer an advantage over CRP in children with fever without localising signs, especially in those of younger age. Since the number of children below the age of 1 year tested in our study was rather small, this problem should be further addressed. Moreover, because PCT rises earlier than CRP after a bacterial stimulus, this test may prove to be more accurate at the beginning of an infection. Total and differential leucocyte count, which is commonly used in the decision algorithm, performed poorly compared to PCT and CRP raising the question of its utility. We therefore suggest that it may be abandoned as a routine first screening method.

Our study showed that in comparison to commonly used screening methods, such as the clinical score of McCarthy [19], the total and differential leucocyte count and CRP as well as to other inflammatory markers such as IL-6, IL-1Ra and IL-8, PCT offers a slightly better sensitivity (93%) and specificity (78%) in predicting a SBI in children with fever without localising signs and may be used alone in the initial screening.

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References

1. Benador N, Siegrist CA, Gendrel D, Greder C, Benador D, Assicot M, Bohuon C, Girardin E (1998) Procalcitonin is a marker of severity of renal lesions in pyelonephritis. Pediatrics 102: 1422–1425
2. Bohuon C, Gendrel D (1999) La procalcitonine: nouvel indicateur d'infection bactérienne. Intérêt et perspectives. Arch Pédiatr 6: 141–144
3. Bonadio W (1998) The history and physical assessments of the febrile infant. Pediatr Clin North Am 45: 65–77
4. Brunkhorst F, Heinz U, Forycki Z (1998) Kinetics of procalcitonin in iatrogenic sepsis. Intensive Care Med 24: 888–892
5. Chiesa C, Pacifico L, Mancuso G, Panero A (1998) Procalcitonin in pediatrics: overview and challenge. Infection 26: 236–241
6. Chiesa C, Panero A, Rossi N, Stegagno M, De Giusti M, Osborn JF, Pacifico L (1998) Reliability of procalcitonin concentrations for the diagnosis of sepsis in critically ill neonates. Clin Infect Dis 26: 664–672
7. Dandona P, Nix D, Wilson MF, Aljada A, Love J, Assicot M, Bohuon C (1994) Procalcitonin increase after endotoxin injection in normal subjects. J Clin Endocrinol Metab 79: 1605–1608
8. De Bont E, De Leij L, Okken A, Baarsma R, Kimpen J (1995) Increased plasma concentrations of interleukin-1 receptor antagonist in neonatal sepsis. Pediatr Res 37: 626–629
9. De Werra I, Jaccard C, Corradin SB, Chiolero R, Yersin B, Gallati H, Assicot M, Bohuon C, Baumgartner JD, Glauser M, Heumann D (1997) Cytokines, nitrite/nitrate, soluble tumor necrosis factor receptors, and procalcitonin concentrations: comparisons in patients with septic shock, cardiogenic shock, and bacterial pneumonia. Crit Care Med 25: 607–612
10. Downs S, McNutt R, Margolis P (1991) Management of infants at risk for occult bacteremia: a decision analysis. J Pediatr 118: 11–20
11. Franz AR, Kron M, Pohlandt F, Steinbach G (1999) Comparison of procalcitonin with interleukin 8, C-reactive protein and differential white blood cell count for the early diagnosis of bacterial infections in newborn infants. Pediatr Infec Dis J 18: 666–671
12. Gendrel D, Raymond J, Coste J, Moulin F, Lorrot M, Guérin S, Ravilly S, Lefèvre H, Royer C, Lacombe C, Palmer P, Bohuon C (1999) Comparison of procalcitonin with C-reactive protein, interleukin-6 and interferon-alpha for differentiation of bacterial versus viral infections. Pediatr Infect Dis J 18: 875–881
13. Hatherill M, Tibby S, Sykes K, Turner C, Murdoch I (1999) Diagnostic markers of infection: comparison of procalcitonin with C reactive protein and leucocyte count. Arch Dis Child 81: 417–421
14. Jaye D, Waites K (1997) Clinical applications of C-reactive protein in pediatrics. Pediatr Infect Dis J 16: 735–747
15. Karzai W, Oberhoffer M, Meier-Hellmann A, Reinhart K (1997) Procalcitonin – a new indicator of the systemic response to severe infections. Infection 25: 329–334
16. Küster H, Weiss M, Willeitner A, Detlefsen S, Jeremias I, Zbojan J, Geiger R, Lipowsky G, Simbruner G (1998) Interleukin-1 receptor antagonist and interleukin-6 for early diagnosis of neonatal sepsis 2 days before clinical manifestation. Lancet 352: 1271–1277
17. Lapillonne A, Basson E, Monneret G, Bienvenu J, Salle B (1998) Lack of specificity of procalcitonin for sepsis diagnosis in premature infants. Lancet 351: 1211–1212
18. McCarthy P (1998) Fever. Pediatr Rev 19: 401–406
19. McCarthy P, Sharpe M, Spiesel S, Dolan T, Forsyth B, DeWitt T, Fink H, Baron M, Cicchetti D (1982) Observation scales to identify serious illness in febrile children. Pediatrics 70: 802–809
20. Meisner M, Tchaikowsky K, Schnabel S, Schmidt J, Katalinic A, Schüttler J (1982) Procalcitonin – influence of temperature, storage, anticoagulation and arterial or venous asservation of blood samples on procalcitonin concentrations. Eur J Clin Chem Biochem 35: 597–601
21. Mood AM, Graybill FA, Boes DC (1974) Introduction to the theory of statistics, 3d edn. McGraw-Hill, New York, pp 181
22. Panero S, Pacifico L, Rossi N, Mancuso G, Stegagno E, Chiesa C (1997) Interleukin 6 in neonates with early and late onset infection. Pediatr Infec Dis J 16: 370–375
23. Roberge CJ, Poubelle PE, Beaulieu AD, Heitz D, Gosselin J (1996) The IL-1 and IL-1 receptor antagonist (IL-1Ra)

response of human neutrophils to EBV stimulation. Preponderance of IL-1Ra detection. J Immunol 156: 4884–4891
24. Sackett DL, Haynes RB, Guyatt GH, Tugwell P (1991) Clinical epidemiology: a basic science for clinical medicine, 2nd edn. Little Brown, Boston, pp 119–139
25. Sormunen P, Kallio M, Kilpi T, Peltola H (1999) C-reactive protein is useful in distinguishing Gram stain-negative bacterial meningitis from viral meningitis in children. J Pediatr 134: 725–729
26. Yoon HJ, Zhu Z, Gwaltney JM, Elias JA (1999) Rhinovirus regulation of IL-1 receptor antagonist in vivo and in vitro: a potential mechanism of symptom resolution. J Immunol 162: 7461–7469
27. Zhu Z, Tang W, Ray A, Wu Y, Einarsson O, Landry ML, Gwaltney J, Elias JA (1996) Rhinovirus stimulation of interleukin-6 in vivo and in vitro. Evidence for nuclear factor kappa B-dependant transcriptional activation. J Clin Invest 97: 421–430
28. Zhu Z, Tang W, Gwaltney JM, Wu Y, Elias JA (1997) Rhinovirus stimulation of interleukin-8 in vivo and in vitro: role of NF-kappa B. Am J Physiol 273: L814–L824