ORIGINAL ARTICLE 10.1111/j.1469-0691.2004.00883.x

Potential use of procalcitonin as a diagnostic criterion in febrile neutropenia: experience from a multicentre study

H. Giamarellou[1], E. J. Giamarellos-Bourboulis[1], P. Repoussis[2], L. Galani[1], N. Anagnostopoulos[3], P. Grecka[1], D. Lubos[4], M. Aoun[5], K. Athanassiou[1], E. Bouza[6], E. Devigili[7], V. Krçmery[4], F. Menichetti[8], E. Panaretou[3], E. Papageorgiou[9] and D. Plachouras[1]

[1]4th Department of Internal Medicine, Athens Medical School, [2]Department of Haematology, Pireas Metaxa Hospital, [3]Department of Haematology, Athens General Hospital G. Gennimatas, Athens, Greece, [4]Department of Internal Medicine, National Cancer Institute, Bratislava, Slovakia, [5]Department of Infectious Diseases, Institut J. Bordet, Bruxelles, Belgium, [6]Hospital 'Gregorio Maranon', Madrid, Spain, [7]Ospedale Generale Regionale, Centro Trapianti Di Midollo Osseo, Bolzano, [8]Azienda Ospedaliera Pisana, UO Malattie Cisanelo, Pisa, Italy and [9]Department of Haematology, Athens Hospital Evangelismos, Athens, Greece

ABSTRACT

In order to assess the diagnostic value of procalcitonin, 158 patients with febrile neutropenia from centres across Europe were studied. Patients with fever were diagnosed on the basis of either: (1) clinical, radiological and microbiological criteria; or (2) the procalcitonin value. In the latter case, concentrations of 0.5–1.0 ng/mL were considered diagnostic of localised infection, concentrations of 1.0–5.0 ng/mL of bacteraemia, and concentrations of >5.0 ng/mL of severe sepsis. Procalcitonin and C-reactive protein were estimated daily in serum by immunochemiluminescence and nephelometry, respectively. Overall, the sensitivity (specificity) of procalcitonin for bacteraemia was 44.2% (64.3%) at concentrations of 1.0–5.0 ng/mL, and 83.3% (100%) for severe sepsis at concentrations of >5.0 ng/mL. It was concluded that procalcitonin is a marker strongly suggestive of severe sepsis at concentrations of >5.0 ng/mL. Estimated concentrations of <0.5 ng/mL indicate that infection is unlikely, but it was observed that bacteraemia associated with coagulase-negative staphylococci may fail to elevate serum procalcitonin levels.

Keywords Bacteraemia, C-reactive protein, diagnosis, fever, procalcitonin

Original Submission: 25 May 2003; **Revised Submission:** 19 July 2003; **Accepted:** 13 October 2003

Clin Microbiol Infect 2004; 10: 628–633

INTRODUCTION

Febrile neutropenia is a common syndrome in patients with haematological malignancies. Subtle clinical findings and lack of microbiological confirmation are often associated with these patients. Moreover, clinical criteria for diagnosis of the systemic inflammatory response syndrome and sepsis [1] are difficult to apply because of the presence of neutropenia. Thus, there is a need for an indicator to unmask the cause of fever. Procalcitonin (PCT) has been proposed as such an indicator of systemic infection, as its concentration is elevated in septic patients, even in the presence of immunosuppression [2–4]. In order to elucidate the value of PCT in diagnosing febrile neutropenia, the present study investigated 158 patients from centres across Europe. The aim of the study was to clarify the sensitivity of PCT as a diagnostic marker, and to identify how the cause of fever in neutropenia would be considered if measuring PCT was the only available diagnostic approach. As a consequence, its diagnostic value could be evaluated for cases when clinical indices of infection were lacking.

Corresponding author and reprint requests: H. Giamarellou, 4th Department of Internal Medicine, Attikon University General Hospital, Athens 124 64, Greece
E-mail: giamarel@internet.gr

PATIENTS AND METHODS

Initially, 181 patients were enrolled in a multicentre prospective clinical trial between June 1999 and December 2001. The participating centres were: Institut J. Bordet, Belgium; Athens General Hospital G. Gennimatas, Evangelismos General Hospital, Metaxa Anticancer Centre, and Sismanoglion General Hospital, Greece; Hospitals of Bolzano and Cisanello-Pisa, Italy; Bratislava General Hospital, Slovakia; and Hospital G. Maranon, Madrid, Spain. Complete data were available for 158 of the 181 patients (Table 1). All suffered from haematological malignancies and presented with neutropenia (polymorphs $<500/\mu L$) after anti-neoplastic chemotherapy, followed by the development of fever. Following diagnosis of neutropenia, blood was collected from one forearm vein; subsequent samples were taken on a daily basis following onset of fever until its resolution. Fever was considered as any single spike of body temperature of >38.5 °C, or as any two or more spikes of >38 °C within 12 h. Fever resolution was considered to have occurred whenever body temperature decreased to ≤ 37.6 °C [5].

Blood was collected into pyrogen-free tubes and centrifuged briefly to separate the serum, which was then stored at −70 °C until processed. Serum PCT was estimated with an assay based on immunochemiluminescence (BRAHMS Diagnostica, Berlin, Germany; lower detection limit of 0.08 ng/mL), and C-reactive protein (CRP) was measured by nephelometry (Boehring, Mannhein, Germany; lower detection limit of 3.2 mg/L).

In order to facilitate statistical analysis, diagnosis in any patient was based on either: (1) clinical observations combined with radiological and microbiological findings; or (2) single estimations of PCT levels following advent of fever.

For (1), patients were divided into six main categories comparable to those used in previous studies [2]: i.e., patients with bacteraemia without severe sepsis; patients with a microbiologically documented localised bacterial infection; patients with severe sepsis; patients with a clinically documented localised infection; patients with systemic mycosis; and patients with fever of unknown origin (FUO). In order to establish the diagnosis, a complete examination of the patient was performed following advent of fever. This comprised cultures of blood, urine and sputum, blood gas analysis, lung X-rays, and computed tomography of lungs and upper or lower abdomen, whenever considered necessary. Diagnosis of bacteraemia associated with coagulase-negative staphylococci (CNS) was based on the isolation of strains with identical antibiograms from at least two separate blood cultures taken at different times. Severe sepsis was recorded whenever fever was accompanied by signs of tissue hypoperfusion, such as decreased urine output, metabolic acidosis or deterioration of mental status [1]. FUO was recorded when no cause of fever could be established after three febrile days [5].

For (2), patients were divided into four main diagnostic categories by a single measurement of the PCT level on presentation according to the diagnostic cut-offs proposed previously [6], namely:

- Bacteraemia: PCT level of 1.0–5.0 ng/mL
- Severe sepsis: PCT level of >5.0 ng/mL
- Localised infection: PCT level of 0.5–1.0 ng/mL
- FUO: PCT level of <0.5 ng/mL

For all patients categorised by criterion (1), PCT and CRP levels on sequential days were expressed by their mean (± SE). Statistical comparisons between groups of patients on the same day of sampling were performed by Tukey's test. PCT levels on the day of presentation of fever were expressed by their mean (± SE), separately for patients who eventually died and for those who were resuscitated. The same analysis was performed for patients with CNS bacteraemia, for bacteraemia caused by other Gram-positive cocci, and for bacteraemia caused by Gram-negative bacteria. Comparisons were performed by ANOVA. Any p value ≤ 0.05 was considered to be significant.

Table 1. Demographic data for patients enrolled in the study

	Category of infection					
	Bacteraemia	**Localised bacterial infection**	**Severe sepsis**	**Clinically localised infection**	**Systemic mycosis**	**Fever of unknown origin**
Number of patients	52 (32.1%)	14 (8.8%)	12(7.6%)	20 (12.7%)	5 (3.1%)	55 (34.0%)
Age in years (mean ± SD)	50.4 ± 18.2	53.3 ± 20.5	53.6 ± 15.0	52.3 ± 19.6	50.0 ± 20.9	49.1 ± 19.3
Male:female ratio	22:30	8:6	8:4	9:11	4:1	39:16
Underlying malignancy (number of patients)						
AML	32	5	7	11	2	30
NHL	7	4	2	2	–	11
ALL	11	3	1	4	2	11
Other	2	2	2	3	1	3
Underlying infection						
Primary bacteraemia	31	–	–	–	1	
UTI	1	7	3	–	–	
LRTI	3	3	5	9	3	
Central venous catheter	17	1	1	1	–	
Other	–	3	3	10	1	
Isolated pathogen						
CNS	26	1				
Streptococcus spp.	5	1				
Escherichia coli	6	7				
Pseudomonas aeruginosa	3	–				
Klebsiella pneumoniae	2	2				
Aspergillus spp.	–	–			4	
Other	10	1	3		1	

AML, acute myelogenous leukaemia; NHL, non-Hodgkin's lymphoma; ALL, acute lymphoblastic leukaemia; LRTI, lower respiratory tract infection; UTI, urinary tract infection; CNS, coagulase-negative staphylococci.

Recipient operating characteristic (ROC) curves of the sensitivity and specificity of PCT and CRP levels for the diagnosis of bacteraemia and severe sepsis were constructed according to (1). Patients with localised bacterial infections were used as controls when designing the ROC curve for bacteraemia, while patients with a localised clinical infection were used as controls when designing the ROC curve for severe sepsis.

RESULTS

For patients diagnosed by (1), the mean (± SE) concentrations of PCT on advent of fever were 2.98 ± 1.03 ng/mL in patients with bacteraemia, 0.98 ± 0.24 ng/mL in patients with microbiologically documented localised infection, 14.54 ± 5.05 ng/mL in patients with severe sepsis, 0.87 ± 0.23 ng/mL in patients with clinically documented localised infection, and 1.11 ± 0.27 ng/mL in patients with FUO. Follow-up of these patients is shown in Table 2. The mean PCT levels (ng/mL) of five patients with systemic mycosis were 1.17 ± 0.44, 0.42 ± 0.19, 0.86 ± 0.36 and 0.39 ± 0.18 on the first, second, third and fourth days of fever, respectively. Follow-up of CRP levels is shown in Table 3.

Twelve (7.6%) patients died while febrile. Their mean PCT level on the first day of fever was 20.45 ± 4.48 ng/mL, compared to 9.79 ± 1.29 ng/mL for patients surviving after the febrile episode ($p < 0.0001$). Among patients with bacteraemia caused by CNS, other Gram-positive cocci and Gram-negative bacteria, the mean PCT levels (ng/mL) on advent of fever were 0.83 ± 0.21, 3.22 ± 1.62 and 6.22 ± 2.49, respectively. The distribution of PCT levels for these patients, and their statistical comparisons, are shown in Fig. 1.

Among 52 patients with bacteraemia diagnosed by (1), 23 had PCT levels of 1.0–5.0 ng/mL. At that cut-off level, the sensitivity, specificity, positive and negative predictive values of PCT for bacteraemia were 44.2%, 64.3%, 82.1% and 18.8%, respectively. The corresponding CRP values at a cut-off of ≥3.2 mg/L were 34.6%, 21.4%, 62.1%, and 8.3%, respectively. Among 12 patients with severe sepsis diagnosed by (1), ten had PCT levels of >5.0 ng/mL. At that cut-off level, the sensitivity, specificity, positive and negative predictive values of PCT for severe sepsis were 83.3%, 100%, 100%, and 90.9%, respectively. The corresponding CRP values at a cut-off of ≥3.2 mg/L were 100%, 5.0%, 38.7% and 100%, respectively. Comparative ROC curves for the diagnostic accuracy of PCT and CRP levels for the diagnosis of bacteraemia and severe sepsis are shown in Fig. 2.

If diagnosis was based on (2), 45 patients would have been diagnosed with bacteraemia, 25 with a localised infection, 17 with severe sepsis, and 71 with FUO. Mean PCT levels (ng/mL) for these patients were 2.32 ± 0.14, 0.71 ± 0.07, 17.53 ± 3.99, and 0.23 ± 0.02, respectively. The correlation between diagnosis by (1) and (2) is shown in Table 4.

DISCUSSION

Febrile neutropenia is a medical emergency requiring prompt diagnosis of any underlying

Table 2. Daily follow-up of procalcitonin (PCT) levels of patients enrolled in the study, correlated with the category of infection

	Mean (± SE) PCT level (ng/mL)				
	Bacteraemia	**Localised bacterial infection**	**Severe sepsis**	**Clinically localised infection**	**Fever of unknown origin**
Afebrile neutropenia	0.85 ± 0.37	0.36 ± 0.10	0.47 ± 0.12	0.24 ± 0.02	0.59 ± 0.17
Advent of fever					
1st day	2.98 ± 1.03[a,b]	0.98 ± 0.24[c]	14.54 ± 5.05[d,e]	0.87 ± 0.23[g]	1.11 ± 0.27
2nd day	2.33 ± 0.54[a,b]	0.74 ± 0.17[c]	14.48 ± 6.08[d,f]	1.99 ± 1.59[g]	0.91 ± 0.32
3rd day	1.57 ± 0.37[a,b]	0.81 ± 0.34[c]	12.76 ± 6.15[d,f]	1.02 ± 0.31[g]	1.14 ± 0.49
4th day	1.04 ± 0.26[a,b]	0.53 ± 0.17[c]	8.91 ± 2.31[d,f]	0.47 ± 0.15[g]	1.17 ± 0.52
Afebrile	1.30 ± 0.37	0.39 ± 0.20	8.67 ± 5.65	0.53 ± 0.15	0.72 ± 0.16

[a]p NS (non-significant) when comparing the PCT levels of patients with bacteraemia to those of patients with fever of unknown origin (FUO).
[b]p NS when comparing the PCT levels of patients with bacteraemia to those of patients with localised bacterial infections.
[c]p NS when comparing the PCT levels of patients with localised bacterial infections to those of patients with FUO.
[d]$p < 0.0001$ when comparing the PCT levels of patients with severe sepsis to those of patients with localised infection.
[e]p 0.001 or [f]$p < 0.0001$ when comparing the PCT levels of patients with severe sepsis to those of patients with FUO.
[g]p NS when comparing the PCT levels of patients with clinically localised infections to those of patients with FUO.

Table 3. Daily follow-up of C-reactive protein (CRP) levels of patients enrolled in the study, correlated with the category of infection

	Mean (± SE) CRP level (mg/L)				
	Bacteraemia	Localised bacterial infection	Severe sepsis	Clinically localised infection	Fever of unknown origin
Afebrile neutropenia	54.8 ± 20.9	50.6 ± 39.9	96.3 ± 26.9	24.0 ± 23.2	29.6 ± 8.2
Advent of fever					
1st day	89.7 ± 19.2[a,b]	94.9 ± 52.0[c]	228.6 ± 28.5[d,f]	51.1 ± 21.4[l]	82.5 ± 17.9
2nd day	123.9 ± 12.4[a,b]	90.6 ± 35.2[c]	214.2 ± 28.0[e,g]	129.0 ± 7.0[l]	76.1 ± 15.6
3rd day	123.8 ± 13.1[a,b]	90.1 ± 34.5[c]	205.0 ± 51.6[e,h]	102.0 ± 9.3[l]	87.3 ± 16.9
4th day	118.5 ± 26.5[a,b]	166.3 ± 69.9[c]	74.0 ± 4.0[e,h]	98.0 ± 15.0[l]	82.5 ± 20.0
Afebrile	77.7 ± 16.9	122.6 ± 33.9	86.7 ± 50.6	146.8 ± 53.2	53.2 ± 11.6

[a]p NS (non-significant) when comparing the CRP levels of patients with bacteraemia to those of patients with fever of unknown origin (FUO).
[b]p NS when comparing the CRP levels of patients with bacteraemia to those of patients with localised bacterial infections.
[c]p NS when comparing the CRP levels of patients with localised bacterial infections to those of patients with FUO.
[d]p 0.005 or [e]NS when comparing the CRP levels of patients with severe sepsis to those of patients with localised infection.
[f]p 0.039, [g]0.001 or [h]NS when comparing the CRP levels of patients with severe sepsis to those of patients with FUO.
[l]p NS when comparing the CRP levels of patients with clinically localised bacterial to those of patients with FUO.

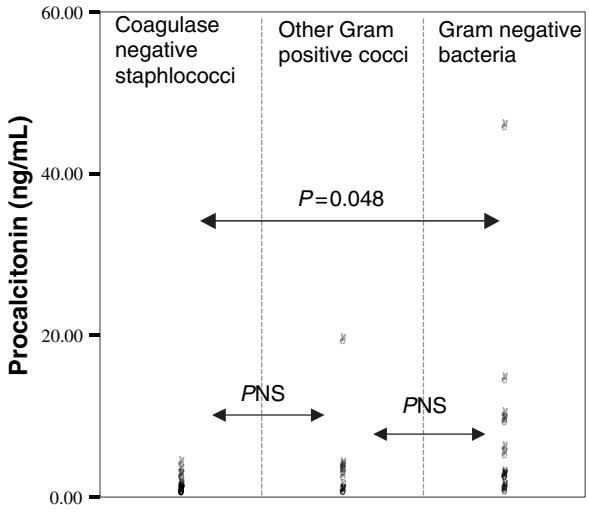

Fig. 1. Distribution of concentrations of procalcitonin at the time of fever manifestation among patients with bacteraemia caused by coagulase-negative staphylococci, other Gram-positive cocci and Gram-negative bacteria.

infection and early administration of antimicrobial agents. Clinical, microbiological and radiological assessment of the patient often fails to reveal the cause of fever. There has been much debate as to whether PCT levels (elevated in systemic infections) might be used to assess the febrile neutropenic host [2–4]. The present multicentre study investigated 158 patients with febrile neutropenia, and clearly showed that PCT levels were elevated significantly in patients with severe sepsis compared to other patients (Table 2). In contrast, although mean PCT levels on the first day of fever were higher in patients with bacteraemia than in those with localised infections, this difference was not statistically significant. This lack of statistical significance can probably be attributed to the fact that patients with bacteraemia caused by CNS and other isolates were grouped together for analysis; the increase in PCT levels was significantly higher in bacteraemia caused by Gram-negative bacteria than in that caused by CNS (Fig. 1). Unlike that of PCT, the CRP level was elevated in all febrile neutropenic patients; however, the increase for severe sepsis was remarkably higher than for the other diagnostic categories (Table 3).

Conflicting results have been reported regarding the diagnostic significance of PCT for systemic mycosis, but only for a limited number of patients [7]. The results in the present study indicated that PCT levels in systemic mycosis were comparable to or even lower than those of patients with localised infections. Four of the five cases of mycosis in the present study were caused by *Aspergillus* spp.

The above results were in general agreement with previous studies describing higher PCT levels in patients with febrile neutropenia and bactaeremia or severe sepsis than in patients with localised infections [2–4]. However, a major discrepancy with these studies concerns the diagnostic significance of PCT. Contrary to previous studies with immunocompetent hosts [8], in which low PCT levels excluded the presence of bacteraemia, the present study indicated that low or normal PCT levels did not exclude bacteraemia associated with CNS. Elevated PCT levels were associated mostly with bacteraemia caused by other species of Gram-positive cocci or Gram-negative bacteria. This is

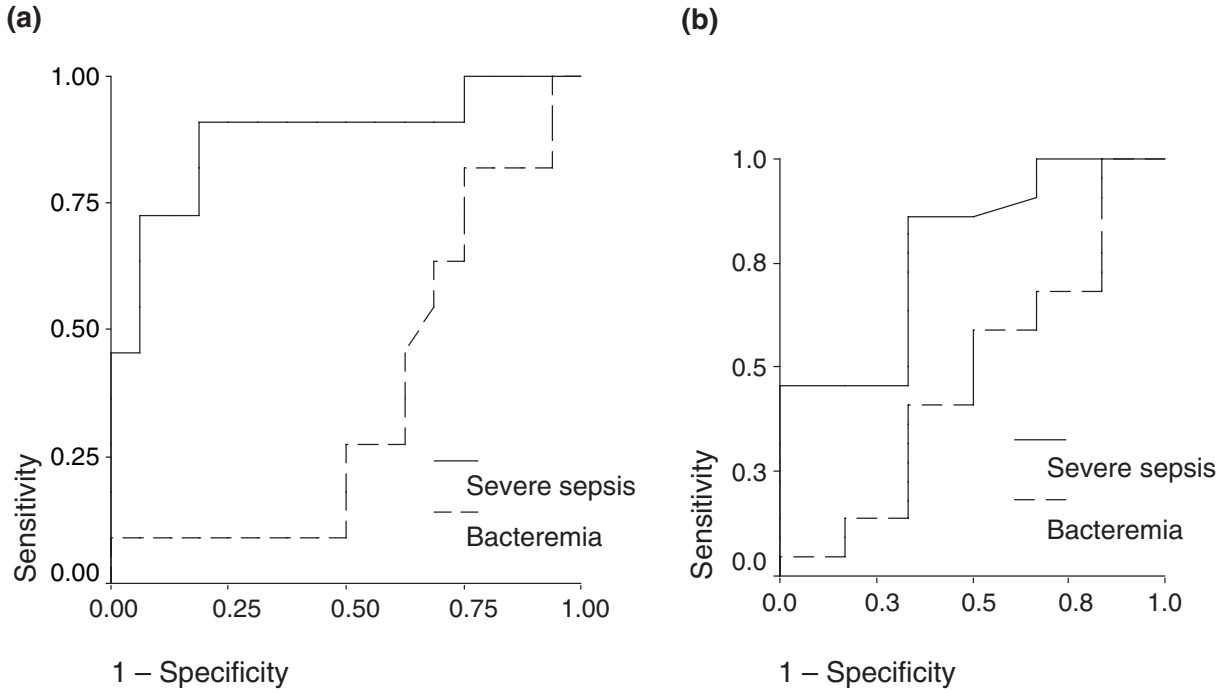

Fig. 2. Comparative ROC curves (a) of procalcitonin and (b) of C-reactive protein for the diagnosis of bacteraemia and severe sepsis. Diagnosis of bacteraemia and severe sepsis was based on clinical, radiological and microbiological criteria (see Methods).

also the main reason for the limited sensitivity of a PCT cut-off value of 1.0 ng/mL in diagnosing bacteraemia with safety (Fig. 2). However, PCT was found to indicate cases of severe sepsis reliably whenever its level was > 5.0 ng/mL.

The present study was the first to attempt to diagnose clinical syndromes in the context of febrile neutropenia on the sole basis of PCT levels. The correlation between diagnosis on the basis of clinical criteria or PCT levels revealed that PCT could predict cases of FUO and cases of localised infections safely (Table 4). The results also indicated that PCT levels could detect patients with severe sepsis accurately, with a high sensitivity and positive predictive value. Although the sensitivity of CRP for diagnosing severe sepsis was also high, its specificity and positive predictive value were very low. The strong diagnostic association between PCT levels and severe sepsis has already been shown elsewhere [9]. Although the diagnostic accuracy of PCT for bacteraemia was lower than for severe sepsis, PCT could discriminate between systemic and localised bacterial infections, a result confirming those of other studies [10,11]. CRP levels could not distinguish between these conditions, irrespective of the possible effect of the underlying malignancy on its production [12].

Finally, it should be emphasised that PCT also had a considerable prognostic value, since the

Table 4. Correlation between diagnosis of the cause of febrile neutropenia by either (1) clinical criteria plus radiological and microbiological findings, or (2) procalcitonin levels

	Diagnosis by (2)[a]				
Diagnosis by (1)[b]	Bacteraemia	Localised infection	Severe sepsis	Fever of unknown origin	Total
Bacteraemia	19	7	6	20	52
Localised bacterial infection	4	4		6	14
Severe sepsis	3		8	1	12
Localised clinical infection	5	6		9	20
Fever of unknown origin	10	8	3	34	55
Systemic fungosis	4			1	5
Total	45	25	17	71	158

[a]Clinical critieria plus radiological and microbiological findings.
[b]Procalcitonin levels.

final patient outcome correlated with the PCT level on the first day of fever. The prognostic significance of PCT has been shown already for immunocompetent hosts [13] and children with febrile neutropenia [14]. There is no doubt that evaluation of a patient should rely always on clinical findings. However, febrile neutropenia is unique in that the clinical picture is often misleading. The findings in the present study revealed that PCT is a marker that might help the clinician: i.e., concentrations of >5.0 ng/mL are strongly suggestive of severe sepsis, levels of 1.0–5.0 ng/mL of bacteremia, and levels of 0.5–1.0 ng/mL of localised infection. Although estimated concentrations of <0.5 ng/mL may indicate that an infection is unlikely, it should always be considered that bacteraemia caused by CNS may fail to elevate serum PCT levels.

REFERENCES

1. American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference Committee. Definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. *Crit Care Med* 1992; **20**: 864–874.
2. Giamarellos-Bourboulis EJ, Grecka P, Poulakou G, Anargyrou K, Katsilambros N, Giamarellou H. Assessment of procalcitonin as a diagnostic marker of underlying infection in patients with febrile neutropenia. *Clin Infect Dis* 2001; **32**: 1718–1728.
3. Fleischhack G, Kambeck I, Cipic D, Hasan C, Bode U. Procalcitonin in paediatric cancer patients: its diagnostic relevance is superior to that of C-reactive protein, interleukin 6, interleukin 8, soluble interleukin 2 receptor and soluble tumour necrosis factor receptor II. *Br J Haematol* 2000; **111**: 1093–1102.
4. Ruokonen E, Nousianen T, Pulkki K, Takala J. Procalcitonin concentrations in patients with neutropenic fever. *Eur J Clin Microbiol Infect Dis* 1999; **18**: 283–285.
5. Giamarellou H. Infections in febrile neutropenia. In: Cunha BA, ed. *Infectious diseases in critical care medicine*. New York: Marcel Dekker, 1998; 563–598.
6. Zahorec R. Definition for septic syndrome should be re-evaluated. *Intensive Care Med* 2000; **26**: 1870.
7. Christofilopoulou S, Charvalos E, Petrikkos G. Could procalcitonin be a predictive marker in systemic fungal infections? Study of 14 cases. *Eur J Intern Med* 2002; **13**: 493–495.
8. Chirouze C, Schuhmacher H, Rabaud C *et al*. Low serum procalcitonin level accurately predicts the absence of bacteremia in adult patients with acute fever. *Clin Infect Dis* 2002; **35**: 156–161.
9. Hambach L, Eder M, Dammann E *et al*. Diagnostic value of procalcitonin serum levels in comparison with C-reactive protein in allogeneic stem cell transplantation. *Haematologica* 2002; **87**: 643–651.
10. Kallio R, Surcel HM, Bloigu A, Syrjala H. C-reactive protein, procalcitonin and interleukin-8 in the primary diagnosis of infections in cancer patients. *Eur J Cancer* 2000; **36**: 889–894.
11. Engel A, Steinbach G, Kern P, Kern W. Diagnostic value of procalcitonin serum levels in neutropenic patients with fever: comparison with interleukin-8. *Scand J Infect Dis* 1999; **31**: 185–189.
12. Sudhoff T, Giagounidis A, Karthaus M. Serum and plasma parameters in clinical evaluation of neutropenic fever. *Antibiot Chemother* 2000; **50**: 10–19.
13. Giamarellos-Bourboulis EJ, Mega A, Grecka P *et al*. Procalcitonin: a marker to clearly differentiate systemic inflammatory response syndrome and sepsis in the critically ill patient? *Intensive Care Med* 2002; **28**: 1351–1356.
14. Barnes C, Ignjatovic V, Newall F *et al*. Change in serum procalcitonin (delta PCT) predicts the clinical outcome of children admitted with febrile neutropenia. *Br J Haematol* 2002; **118**: 1197–1198.