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Accuracy of Procalcitonin in the Diagnosis of Bacteremia and Discrimination from Contamination

Nuket Hayirlioglu [1], Ayse Demet Kaya [2], Deniz Sertel Selale [3], Mine Aydin Kurc [4], Gamze Varol [5]

[1] Darica Farabi Training and Research Hospital Microbiology Laboratory, Kocaeli, Turkiye
[2] Department of Medical Microbiology, Faculty of Medicine, Istanbul Okan University, Istanbul, Turkiye
[3] Department of Microbiology and Clinical Microbiology, Faculty of Medicine, Istinye University, Istanbul, Turkiye
[4] Department of Medical Microbiology, Faculty of Medicine, Namik Kemal University, Tekirdag, Turkiye
[5] Department of Public Health, Faculty of Medicine, Namik Kemal University, Tekirdag, Turkiye

Abstract

Objective: In this study, we aimed to evaluate the concordance of blood culture with procalcitonin (PCT) alone and together with C-reactive protein (CRP) in detecting bacteremia and the diagnostic performance of these biomarkers to differentiate contamination from true bacteremia.

Materials and Methods: The medical records of 310 patients were analysed retrospectively. Advia Centaur XP immunoassay system and Au analysers were used to determine PCT and CRP levels, respectively. BacT/Alert 3D60 hemoculture system was used to incubate blood specimens, and VITEK 2 compact was used to identify isolated strains.

Results: The accuracy of PCT and CRP in detecting bacteremia were found to be 68.1% and 36.4%, respectively, and combining PCT and CRP had no added value. In analysis of receiver operating characteristic (ROC), the area under the ROC curve (AUROC) values of PCT and CRP were found to be 0.889 and 0.779 in discriminating the culture-negative group from the culture-positive group, and 0.645 and 0.502 in discriminating bacteremia from contamination, respectively.

Conclusion: PCT is a reliable marker that can be used to detect bacteremia. However, its discriminative power was low in differentiating true bacteremia from contamination. Therefore, PCT levels alone should not be used to rule out blood culture contamination.

Keywords Procalcitonin · CRP · Blood cultures · Bacteremia · Contamination

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Corresponding author: Deniz Sertel Selale deniz.sertel@istinye.edu.tr

INTRODUCTION

Bacteremia, if treatment is delayed, may progress to sepsis, septic shock, and death. Therefore, prompt diagnosis is of great importance. In diagnosis, blood culture is considered as the gold standard, but long incubation times (>24 hours), false-positive results due to contamination and unacceptably high false-negative results have created the need for the search for rapid and accurate tests (1-3).

Acute-phase reactants (APRs) have been commonly used as biomarkers indicating infections as they are present in the serum in case of injury and inflammation (1, 4). However, very few can discriminate bacterial infection from inflammation due to non-infectious causes, and information regarding their ability to differentiate true bacteremia from contamination is either unavailable or limited. One of the most commonly used APRs is C-reactive protein (CRP). CRP is predominantly produced by the liver and its concentration in the bloodstream increases gradually in response to pro-inflammatory cytokines, principally interleukin-6 (IL-6), which is produced in case of inflammation due to infection or other causes (4, 5).

Procalcitonin (PCT), the prohormone of calcitonin, is synthesized ubiquitously in response to systemic inflammatory stimuli, particularly bacterial infection (4-7). In detecting bacterial infections, PCT has a number of advantages over other commonly used biomarkers such that it increases rapidly after bacterial infections (rises up to detectable levels in serum within 2–6 hours following stimulation), it has a long half-life (can be detected for up to 7 days), and viral infections or autoimmune diseases do not increase its levels. In addition, PCT levels in the serum coincide with the infection's severity (PCT levels increases parallel to the severity of the infection) and production is not affected by anti-inflammatory and immunosuppressive states (5, 8, 9). These properties of PCT make it a plausible choice as an indicator of severe bacterial infections and bacteremia; hence, PCT is a commonly used biomarker for predicting sepsis due to bacterial infection (4-7). Serum PCT levels have also been considered for guiding antibiotic therapy and for monitoring the prognosis of the disease and the efficacy of the treatment (10, 11). Although there are a few studies that have shown that PCT can also be used in differentiating bacteremia from contamination, this feature should be verified with additional studies before using it in clinical practice.

Besides the clear benefits of PCT over the other APRs including CRP, PCT levels alone are not sufficient to make critical decisions. Therefore, we examined and compared the diagnostic accuracy of PCT alone and together with CRP in detecting bacteremia and their performance in distinguishing bacteremia from contamination.

MATERIALS and METHODS

Study Design and Settings

The Kocaeli University Ethical Committee of Non-Invasive Clinical Research reviewed and approved this study (Approval no: GOKAEK-2017/4.09) and the ethical standards of the Helsinki were followed.

The study was carried out at Darica Farabi Training and Research Hospital throughout a six-month study period. The medical records of patients with suspected blood stream infection were retrospectively reviewed provided that the levels of PCT and CRP were investigated and blood culture had been performed simultaneously. Only data of the patients in whom PCT, CRP, and blood cultures were assayed simultaneously were included. Patients were excluded if the results of any of the tests were not available.

Blood Culture, PCT, and CRP Analysis

Blood samples were inoculated in hemoculture vials of the automated hemoculture system BacT/Alert3D60 (Biomerieux) and incubated for up to seven days. Bacteria isolated from positive cultures were identified using the VITEK2 compact (Biomerieux) system. Blood cultures were considered negative if no growth was observed at the end of the seventh day. Isolation of skin microbiota members (coagulase-negative *Staphylococcus* [CoNS], *Corynebacterium* spp, *Propionibacterium acnes*, etc) were considered as contamination unless isolates that had similar antibiograms were isolated from more than one blood culture taken at two different times (12-14).

Advia Centaur XP immunoassay system (Siemens) was used for detecting PCT levels and Au 680 and Au 5800 analysers (Beckman Coulter) were used for detecting CRP levels. If the levels of CRP or PCT were undetectable, a value equal to the threshold of detection was assigned. For CRP, the cut-off value was accepted as 5mg/dL. The PCT cut-offs used was 5ng/mL, 2ng/mL, and 10ng/mL.

Blood cultures were considered as the reference test, and the accuracy of PCT and CRP was determined by comparing the results of these tests with those of the blood culture.

To assess whether including CRP test has any added value, PCT and CRP results were analysed in combination. If both PCT and CRP tests were positive, the result was considered positive, but if any of the tests were negative, the result was accepted as negative.

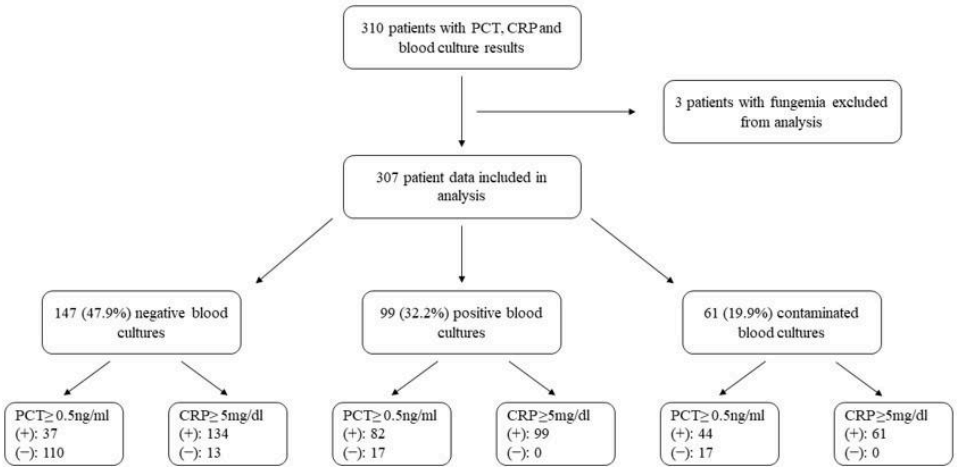

Figure 1. Flow diagram of the included cases. PCT: Procalcitonin; CRP: C-reactive protein

Statistical Analyses

The SPSS 20.0 (SPSS Inc., Chicago, IL, USA) package program was used to evaluate the data. The sensitivity, specificity, and positive- and negative-predictive values were calculated in the methodological analysis. Categorical variables were compared using Pearson Chi-square test and Fisher's exact test. One-way analysis of variance (ANOVA) was performed, and Post Hoc Bonferroni correction was applied to compare the measurement values of more than two groups. The accuracy of PCT and CRP in discriminating culture positive from culture negative and culture positive from contamination were determined by receiver operating characteristic (ROC) analysis. p value was accepted below 0.05 as significant.

RESULTS

During the six-month study period, PCT, CRP, and blood cultures were concomitantly assayed in 310 patients. Data of three patients were excluded because *Candida* spp. were grown in the blood cultures. The mean age was 66.60 ± 16.61 (range: 18-96), and the gender distribution was 48.5% female and 51.5% male. Patients were categorised into three groups regarding the blood culture results: culture positive (n=99, 32.2%), culture negative (n=147, 47.9%) and contamination (n=61, 19.9%) (Figure 1). Culture-positive group was again divided into 2 groups as Gram positive bacteremia (n=55) and Gram negative bacteremia (n=44). Among Gram positive bacteria isolated from blood cultures, *Staphylococcus aureus* (n=21) was the predominant species, followed by *Enterococcus faecalis* (n=20), *Enterococcus faecium* (n=11) and *Enterococcus cloacae* (n=1). Among Gram negative bacteria, *Klebsiella pneumoniae* (n=18) ranked first, followed by *Escherichia coli* (n=16), *Acinetobacter baumannii* (n=8), *Pseudomonas aeruginosa* (n=1), and *Serratia marcescens* (n=1). All bacteria in the contamination group were CoNS.

The mean, minimum and maximum PCT and CRP levels were assessed and compared among the groups (Table 1). In the culture-negative group, PCT and CRP levels were significantly lower (p<0.001) than in the culture-positive and contamination groups. Although the mean PCT and CRP concentrations were lower in the contamination group in comparison to the culture-positive group, Gram positive and Gram negative bacteremia group, the difference was not significant statistically (p=1.00). The PCT and CRP levels in the Gram negative bacteremia group were slightly above the levels detected in the Gram positive bacteremia group, but the difference was not statistically meaningful (p=1.00).

Table 1. Mean, minimum and maximum PCT and CRP values among the groups.

Groups	Mean ± Standard Deviation (min-med-max)	
	PCT (ng/mL)	CRP (mg/dL)
Contamination (n=61)	9.87 ± 23.36	177.07 ± 86.21
	(0.01-1.10-101.00)	(9.20-177.20-347.70)
Culture positive (n=99)	13.23 ± 20.76	178.71 ± 88.97
	(0.10-4.68-101.00)	(9.80-178.30-411.90)
Gram-positive bacteremia (n=56)	12.80 ± 21.85	173.43 ± 88.33
	(0.12-3.98-101.00)	(9.80-179.25-411.90)
Gram-negative bacteremia (n=43)	13.78 ± 19.50	185.59 ± 90.36
	(0.10-6.43-90.60)	(44.00-177.90-368.97)
Culture-negative (n=147)	1.06 ± 3.26	87.95 ± 86.85
	(0.00-0.14-23.04)	(0.40-63.00-421.60)
p value*	<0.001	<0.001

* One-way analysis of variance with Post Hoc Bonferroni correction was applied. Statistically significant difference was present only between culture-negative group vs culture positive, contamination, Gram-positive bacteremia and Gram-negative bacteremia groups. PCT: Procalcitonin; CRP: C-reactive protein.

In comparison to the reference blood culture, PCT was found to be 82.8% sensitive and 61.1% specific when 0.5 ng/mL was used as the cut-off. As for CRP, sensitivity was 100% and specificity was 6.2%. The specificity of PCT increased markedly when higher cut-off values were used, but the sensitivity declined. When 2 ng/mL was used as the cut-off, sensitivity decreased to 64.7% and specificity increased to 83.2%. When 10 ng/mL was used as cutt-of, sensitivity decreased dramatically to 36.4% and specificity increased further to 91.8% (Table 2). On the basis of these results, the optimal PCT threshold for detecting bacteremia was considered to be 0.5 ng/mL.

Table 2. Diagnostic accuracy of PCT and CRP in detecting bacteremia

	PCT		CRP	
Cut-off value	0.5 ng/mL	2 ng/mL	10 ng/mL	5 mg/dL
Sensitivity, (95% CI)	82.8 (75-90)	64.7 (55-74)	36.4 (27-46)	100 (100-100)
Specificity, (95% CI)	61.1 (54-67)	83.2 (78-88)	91.8 (88-95)	6.2 (3-10)
PPV, (95% CI)	50.3 (43-58)	64.7 (55-74)	67.9 (55-80)	33.6 (28-39)
NPV, (95% CI)	88.2 (83-93)	83.2 (78-88)	75.2 (70-81)	100 (100-100)
Accuracy, (95% CI)	68.1 (63-73)	77.2 (73-82)	73.4 (69-79)	36.4 (31-42)

PCT: Procalcitonin; CRP: C-reactive protein; CI: Confidence interval; PPV: Positive predictive value; NPV: Negative predictive value.

The diagnostic performance of PCT (0.5 ng/mL) and CRP (5 mg/dL) was compared between Gram positive bacteremia group and Gram negative bacteremia group. The diagnostic accuracy of PCT in detecting Gram positive and Gram negative bacteremia was 65.8% (95% CI=60-72) and 64.7% (95% CI=59-71). CRP detected Gram positive and Gram negative bacteremia with 25.9% (95% CI=21-31) and 22.6% (95% CI=17-28) accuracy, respectively. A statistically significant difference was not found for both tests.

The diagnostic accuracy of the PCT-CRP combination (accuracy 68.1%) was analysed but the results did not differ from the PCT test alone (accuracy 68.1%); hence, including the CRP test had no added value.

In the ROC analysis, the area under the ROC curve (AUROC) values of PCT and CRP in discriminating the culture-positive group from the culture-negative group were 0.889 and 0.779, respectively (Figure 2). In distinguishing the culture-positive from the contamination group, PCT had an AUROC of 0.645, and CRP had an AUROC of 0.502 (Figure 3).

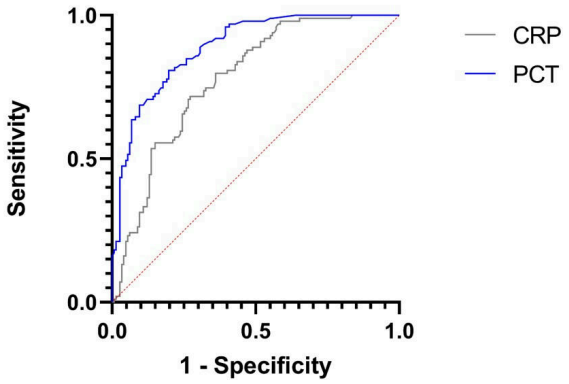

Figure 2. ROC analysis of the PCT and CRP for distinguishing culture-positive from culture-negative. ROC: Receiver operating characteristic; PCT: Procalcitonin; CRP: C-reactive protein

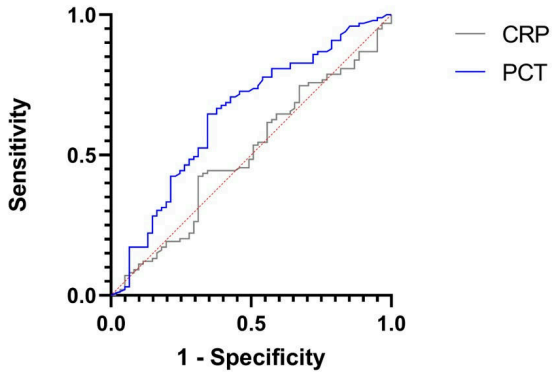

Figure 3. ROC analysis of PCT and CRP for distinguishing culture-positive from contamination. ROC: Receiver operating characteristic; PCT: Procalcitonin; CRP: C-reactive protein

DISCUSSION

Our results have shown that PCT can be used as a reliable biomarker for detecting bacteremia, but its discriminative power is low in distinguishing true bacteremia from contamination.

Multiple studies have investigated the diagnostic performance of PCT and CRP, but relatively few have investigated their ability to distinguish between true bacteremia and contamination (15-18). Schuetz and colleagues were the first to investigate the diagnostic accuracy of PCT to discriminate blood culture contamination from bloodstream infection. In this prospective study conducted in a small cohort (n=40), PCT was found to be an early and accurate biomarker that can aid in distinguishing bloodstream infection from contamination due to CoNS (15). Following this research, a limited number of studies were conducted analysing the capability of PCT and CRP to distinguish bacteremia and contamination (16-18). These studies have found PCT to be superior to CRP and a reliable marker that

can accurately differentiate bacteremia from contamination (16-18). Interestingly, in these studies, PCT and CRP performed better at differentiating true bacteraemia from contamination than discriminating culture-positive from culture-negative (Table 3) (16-18). In a recent study, the usefulness of PCT in diagnosing blood culture contamination was evaluated (19). This study used 0.1 ng/mL as the threshold and concluded that PCT levels below 0.1 ng/mL was a reliable biomarker in determining contamination of blood cultures in hospitalised patients (19).

Table 3. Diagnostic performance of PCT and CRP to discriminate culture positive (CP) from culture negative (CN) and CP from contamination

Study	CP/Cont (% Cont)	CP vs CN		CP vs. Cont	
		AUROC PCT	AUROC CRP	AUROC PCT	AUROC CRP
Present study	99/61 (38.1)	0.889	0.779	0.645	0.502
Oksuz et al.[16]	88/49 (35.8)	0.755	0.601	0.864	0.744
Iqbal-Mirza et al.[17]	154/112 (42.1)	NA	NA	0.983	0.639
Jeong S et al.[18]	331/156 (40.3)	0.76	0.64	0.86	0.65

PCT: Procalcitonin; CRP: C-reactive protein; CP: Culture positive; CN: Culture negative; Cont: Contamination; ROC: Receiver operating characteristic; AUROC: Area under the ROC curve; NA: Not available.

In our study, PCT performed well in discriminating culture negative from culture positive, but its performance was poor in discriminating true bacteremia from contamination, and the performance of CRP was lower than that of PCT in both regards. The AUROC of PCT and CRP in distinguishing culture-positive from contamination was lower than that in other studies with a similar design. Conversely, in discriminating culture-positive from culture-negative, we found the performance of PCT and CRP to be better than reported in the aforementioned studies (Table 3). The reason behind this difference is not clear. It is possible that in certain patients, isolated CoNS might be misidentified as contaminants. To implicate CoNS and other common contaminants as the cause of bacteremia, two or more positive blood cultures were required, but in 17 out of 61 (27.9%) patients, from whom CoNS were isolated, only one sample of blood was drawn for culture; therefore, according to our criteria these were classified as contaminants.

A number of studies have investigated the efficiency of using PCT concentration to discriminate bacteremia or sepsis due to Gram positive and Gram negative pathogens. In our study, the PCT concentrations did not significantly vary among bacteremia caused by Gram negative and Gram positive bacteria. This finding is in line with studies of Oksuz et al. (16) and Kim et al. (20) but discordant with the studies of Engel et al. (21) Svaldi et al. (22), Jeong et al. (18), Leli et al. (23), Yan et al. (24), Ogawa et al. (25) and Dincer et al. (26). In some studies, statistically significant differences were observed even in between bacteremia due to different species of Gram negative bacteria (23, 24).

Although PCT is explicitly more specific for infections caused by bacteria compared to most other APRs, it is still far from being perfect, therefore we also analysed the diagnostic accuracy of the PCT-CRP combination. However, combining PCT with CRP did not have any added value in regards of specificity. Thus, according to our findings, if sepsis due to bacterial infection is suspected, the PCT test alone will be sufficient for the presumptive diagnosis.

Due to its retrospective nature, information regarding certain factors that can influence the PCT levels like baseline characteristics and comorbidities of, and the treatment applied to the patients were not available and therefore could not be assessed. Furthermore, all the data were obtained from inpatients and antibiotic therapy is frequently applied to hospitalised patients which may result in lower PCT concentrations either by direct effect or by lowering the microbial load, also it may cause false negative results in blood culture (27). Although the impact of underlying diseases and treatments applied could not be assessed, the factors that influence PCT and CRP concentrations have been well established in numerous studies conducted previously, and the purpose of this study was to assert the accuracy of PCT and CRP as independent parameters in diagnosing bacteremia and discrimination from contamination.

An important challenge that physicians face in patients with sepsis is to decide whether the cause is a bacterial agent, or not. Guidelines and algorithms regarding the use of PCT levels to initiate and terminate antibiotic treatment in patients with sepsis have been established, and in 2017, the Food and Drug Administration cleared the use of procalcitonin tests in determining whether to initiate or terminate antibiotic treatment in patients with sepsis and lower respiratory tract infections (27-29). Although few studies have shown that PCT can effectively distinguish true bacteremia from contamination, no algorithms have been proposed. This may be due to the fact that the number of studies that have been conducted on this topic is insufficient. To contribute to the literature, we conducted this retrospective study in a training and research hospital in Turkiye.

Our study results demonstrate that PCT can be used in the presumptive diagnosis of bacteremia as a reliable biomarker and is clearly a better option than CRP. To differentiate Gram positive and -negative pathogens, we cannot propose a cut-off value that can be used because no significant difference was found between the two groups. Discriminating true

bacteremia from contamination promptly is vital, particularly for hospital-acquired infections, in which bacteria frequently regarded as contaminants may be the cause of the infection. The diagnostic performance of PCT was poor in differentiating true bacteremia from contamination; hence, PCT levels alone cannot be used in deciding whether the isolate is the cause or a contaminant. Biomarkers, including PCT, can contribute, but the available data are not sufficient. Therefore, prospective studies with large cohorts should be conducted to establish a reliable diagnostic algorithm.

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Ethics Committee Approval: Kocaeli University Ethical Committee of Non-Invasive Clinical Research reviewed and approved this study (Approval no: GOKAEK-2017/4.09) and the ethical standards of the Helsinki were followed.

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Author Details

Nuket Hayirlioglu
[1] Darica Farabi Training and Research Hospital Microbiology Laboratory, Kocaeli, Turkiye
0000-0003-1975-4884

Ayse Demet Kaya
[2] Department of Medical Microbiology, Faculty of Medicine, Istanbul Okan University, Istanbul, Turkiye
0000-0001-8224-8242

Deniz Sertel Selale
[3] Department of Microbiology and Clinical Microbiology, Faculty of Medicine, Istinye University, Istanbul, Turkiye
0000-0001-8437-3557 ✉ deniz.sertel@istinye.edu.tr

Mine Aydin Kurc
[4] Department of Medical Microbiology, Faculty of Medicine, Namik Kemal University, Tekirdag, Turkiye
0000-0002-5053-4276

Gamze Varol
[5] Department of Public Health, Faculty of Medicine, Namik Kemal University, Tekirdag, Turkiye
0000-0002-3490-3406

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