Available at https://www.iasj.net/iasj
Iraqi Academic Scientific Journals
Journal homepage:
https://journals.uokerbala.edu.iq/index.php/UOKJ

Research Article

The Procalcitonin Paradox in COVID-19: A Case-Control Study from the 2025 Arbaeen Pilgrimage

[1,]Ali Jabbar Abd Al-Hussain Alkawaz [2,] Maryam Sabah Naser [3,] Ali Jalil Obaid

[1,]Department of Biology, College of Science, University of Kerbala: Karbala, Iraq
[2,3]Department of Applied Biotechnology, College of Biotechnology, AL-Qasim Green University, Babylon, Iraq

Article Info
Article history:
Received 20-12-2025
Received in revised form 8-2-2026
Accepted 5-4-2026
Available online 31 - 3-2026
Keywords: COVID-19; procalcitonin; interleukin-6; bacterial co-infection; antibiotic stewardship; Arbaeen pilgrimage

Abstract:

Background: Procalcitonin (PCT) is used to differentiate between bacterial and viral infections; nevertheless, its effectiveness in the context of COVID-19 is unknown. The inflammatory response associated with COVID-19, mediated by IL-6 cytokine activation, may contribute to increased levels of PCT regardless of bacterial coinfections.

Methods: This prospective case-control study recruited 200 patients during the 2025 Arbaeen pilgrimage. Serum levels of PCT, IL-6, CRP, WBC count, and bacterial cultures were used to assess their value in detecting bacterial coinfections among cases with or without COVID-19.

Results: Significantly, PCT levels were not increased in cases with or without bacterial coinfections. The incidence of bacterial coinfections was significantly reduced in cases with COVID-19 compared with cases without COVID-19. On the other hand, levels of IL-6 were substantially increased in cases with COVID-19 compared with cases without COVID-19. The ROC curve showed that PCT was of limited value in detecting bacterial coinfections (AUC = 0.72; 95% CI = 0.65 to 0.79). The sensitivity of PCT at an optimal cutoff of 0.12 ng/mL was 71%, with specificity of 68%. Multivariable logistic regression identified IL-6 and WBC count as independent predictors of bacterial coinfections.

Conclusion: Procalcitonin is an inaccurate marker for detecting bacterial coinfections in the context of COVID-19 because of its relationship with IL-6 levels. The effectiveness of IL-6 levels and WBC count in detecting bacterial coinfections makes them more useful than PCT levels.

Corresponding Author E-mail: ali.abdulhussein@uokerbala.edu.iq
Peer review under responsibility of Iraqi Academic Scientific Journal and University of Kerbala.

Highlights

- Procalcitonin showed limited to moderate diagnostic performance for detecting bacterial coinfection in COVID-19.
- IL-6 elevation may increase PCT independent of bacteria.
- Bacterial infections were less frequent in COVID-19–positive patients.
- Diagnostic performance was limited (AUC ≈0.72).
- Antibiotic use should be guided by cultures and clinical judgment, not PCT.

Introduction

Antibiotics are frequently used for the treatment of patients with COVID-19, yet according to global statistics, only 5-8% of patients with COVID-19 and bacterial coinfections have been documented, and there are growing concerns regarding the misuse of antimicrobial agents worldwide [1-4]. Procalcitonin (PCT), a peptide biomarker, increases in response to bacterial endotoxins and proinflammatory cytokines such as IL-6 and TNF-α and is commonly used for the differential diagnosis of bacterial and viral infections [5,6]. In the case of COVID-19, interpreting PCT levels is complicated due to the proinflammatory environment, and elevated levels of IL-6 stimulate PCT release in the absence of bacterial pathogens, reducing the specificity of PCT for bacterial coinfections in patients with COVID-19 [7]. Several studies have investigated the clinical utility of PCT in patients with COVID-19 and found that there are only minor differences between patients with and without bacterial coinfections, and the ability of PCT to diagnose bacterial coinfections in patients with COVID-19 is poor (AUC=0.56) [8,9]. In spite of these growing concerns, the misuse of antibiotics for the treatment of patients with COVID-19 and bacterial coinfections is widespread [3,10]. This study aimed to assess the reliability of PCT for the diagnosis of bacterial coinfections in patients with COVID-19, in comparison with other biomarkers and microbiological culture, in a prospective case-control study conducted during the Arbaeen pilgrimage in Iraq.

Methods

This prospective case-control study took place in Hillah, Iraq, during the Arbaeen pilgrimage of Imam Hussein in August 2025. We recruited 200 adults suspected of having an infection. Of these, 100 confirmed cases of COVID-19 through RT-PCR and antigen tests were recruited along with another 100 who tested negative. The sample size is dependent on the number of patients we had during the period of the study. This limits the ability to detect smaller effects, but the trends in the results are consistent and support our results in the context of a mass-gathering event. The article explains how the patients are recruited and allocated to the groups. We used all the results we had in the final analysis.

We took our samples within 24-48 hours of admission to assess the levels of procalcitonin (PCT), interleukin-6 (IL-6), C-reactive protein (CRP), and white blood cells (WBC) in our patients [5,6,7]. We also did bacterial cultures on our samples when clinically indicated and excluded samples with fungal contamination. We recorded sex as male or female and did not assess gender identity.

The study has been approved by the Research Ethics Committee at the College of Science, University of Karbala, and we have obtained informed consent. Continuous variables such as PCT, IL-6, CRP, WBC are expressed as the "mean ± SD" along with 95% confidence intervals. For comparison of COVID-19 positive and negative patients, simple statistical tests such as t-tests or Mann-Whitney tests were employed. For comparison of culture results, we have employed the chi-square test or Fisher's test along with odds ratio calculation.

For assessing bacterial coinfection, a regression analysis has been employed with PCT as the main outcome variable while considering age, sex, COVID-19 positive or negative, IL-6, WBC, CRP, and history of using antibiotics as confounding variables.

For assessing the accuracy of PCT, ROC analysis has been employed along with

calculation of AUC values. For assessing correlations among variables, Spearman's test has been employed.

The statistical analysis has been conducted using SPSS version 27 software with $p < 0.05$ for two-tailed tests.

Results Demographic and Biomarker Comparisons

Among the 200 patients, demographic factors were found to be comparable between COVID-19 positive and negative patients. COVID-19 positive patients had higher levels of Interleukin-6 (IL-6) and lower white blood cell (WBC) counts. For C-reactive protein (CRP) and procalcitonin (PCT), no significant difference was found between COVID-19 positive and negative patients. However, after adjusting for analysis of covariance, CRP levels were found to be higher among COVID-19 positive patients by +16.8 mg/L (95% CI, 13.9 to 19.7; $p < 0.001$), suggesting that confounding effects were present. PCT, however, remained non-significant after adjusting for ANCOVA, while IL-6 and WBC counts showed strong and consistent associations.

Bacterial Coinfection and Predictive Modeling

Bacterial coinfection was identified in 40% of COVID-19–positive patients compared with 75% of COVID-19–negative patients. Multivariable logistic regression analysis was adjusted for age, sex, COVID-19 status, IL-6 levels, WBC count, CRP levels, and prior antibiotic use. Within this model, PCT was not an independent predictor of bacterial coinfection, whereas IL-6 and WBC counts remained statistically significant predictors. Receiver operating characteristic (ROC) analysis demonstrated limited diagnostic performance of PCT; at a cutoff value of 0.12 ng/mL, sensitivity was 71% and specificity was 68%.

Table 1: Demographic Characteristics, Biomarkers, and Predictors of Bacterial Coinfection in COVID-19–Positive and COVID-19–Negative Patients

Demographics			
Variable	**COVID-19 Positive (n=100)**	**COVID-19 Negative (n=100)**	**p-value**
Age (years), mean ± SD	44.2 ± 12.5	43.6 ± 11.8	0.73
Male sex, n (%)	58 (58)	55 (55)	—
Female sex, n (%)	42 (42)	45 (45)	0.78
Iraqi nationality, n (%)	72 (72)	70 (70)	—
Iranian nationality, n (%)	20 (20)	22 (22)	—
Other nationality, n (%)	8 (8)	8 (8)	0.94

Biomarkers and Microbiological Findings (Unadjusted)				
Variable	**COVID-19 Positive**	**COVID-19 Negative**	**p-value**	**Interpretation**
IL-6 (pg/mL)	55 ± 15	12 ± 4	0.0005	Significantly higher in COVID-19–positive patients
WBC (×10⁹/L)	5.7 ± 1.5	7.6 ± 1.5	0.012	Significantly lower in COVID-19–positive patients
Bacterial culture positive (%)	40	75	<0.0001	Less frequent in COVID-19–positive patients
CRP (mg/L)	63 ± 15	46 ± 10	0.075	Not significant
Procalcitonin (ng/mL)	0.11 ± 0.03	0.14 ± 0.03	0.23	Not significant

Multivariable Logistic Regression (Outcome: Bacterial Coinfection)			
Predictor	**aOR (95% CI)**	**p-value**	**Interpretation**
PCT (per 1 SD increase)	1.16 (0.82–1.63)	0.395	Not significant

IL-6 (per 10 pg/mL increase)	1.45 (1.25–1.72)	<0.001	Significant predictor
WBC (per 1 ×10⁹/L increase)	0.72 (0.58–0.87)	0.001	Protective
CRP (per 10 mg/L increase)	1.08 (0.95–1.23)	0.210	Not significant
COVID-19 positive (vs. negative)	0.35 (0.22–0.54)	<0.001	Lower odds of coinfection

Adjusted Analyses (ANCOVA)			
Variable	**Adjusted mean difference (95% CI)**	**p-value**	**Interpretation**
IL-6 (pg/mL)	+42.2 (36.8–47.5)	<0.001	Higher in COVID-19–positive patients
WBC (×10⁹/L)	−1.83 (−2.10 to −1.56)	<0.001	Lower in COVID-19–positive patients
CRP (mg/L)	+16.8 (13.9–19.7)	<0.001	Higher in COVID-19–positive patients
PCT (ng/mL)	−0.030 (−0.036 to −0.025)	<0.001	Slightly lower in COVID-19–positive patients

Abbreviations: IL-6, interleukin-6; WBC, white blood cell count; CRP, C-reactive protein; PCT, procalcitonin; SD, standard deviation; CI, confidence interval; aOR, adjusted odds ratio. Unadjusted comparisons are presented for descriptive variables. Logistic regression models report adjusted odds ratios for predictors of bacterial coinfection. ANCOVA results represent covariate-adjusted mean differences between COVID-19–positive and COVID-19–negative groups.

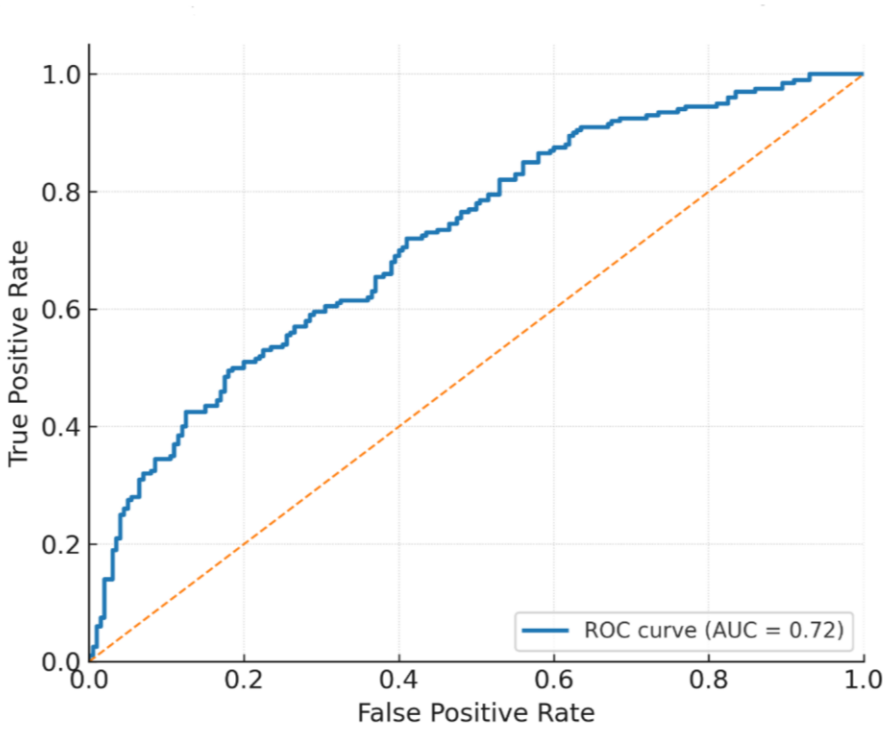

Figure 1: ROC curve of PCT predicting bacterial coinfection in COVID-19: AUC 0.72 (95% CI: 0.65–0.79); cutoff 0.12 ng/mL yielded 71% sensitivity, 68% specificity.

Discussion

Our findings demonstrate that serum procalcitonin (PCT) is not a reliable biomarker for identifying secondary bacterial infections in patients with COVID-19. To our knowledge, this is the first study to specifically evaluate the diagnostic limitations of PCT within the context of a large mass-gathering event. Such settings—including religious pilgrimages, Hajj, and international sporting events—are characterized by heightened infection risk and diagnostic complexity due to overlapping inflammatory and infectious processes.

Despite the presence of bacterial infections in COVID-19-negative patients, the levels of PCT were similar in both COVID-19-positive and COVID-19-negative patients. This forms the basis for what we call the "procalcitonin paradox," which, for the purpose of this paper, refers to a disconnect between bacterial load and PCT levels in the presence of overwhelming virus-induced inflammatory processes. In such cases, PCT levels are not elevated proportionally with the presence of bacterial coinfections, nor are they elevated independently of bacterial coinfections. The results of the relatively low sensitivity and specificity at the optimal cutoff value further reinforce the fact that PCT levels are not useful in distinguishing between bacterial coinfections and COVID-19 infection. Thus, IL-6 levels and microbial cultures would provide a clearer direction for antimicrobial therapy decisions. This might be a feature of a number of disease processes that are associated with overwhelming inflammatory processes, such as influenza and severe viral pneumonia, as previously described in the literature [7].

From a mechanistic perspective, it appears that elevated levels of PCT in COVID-19 infection are not due to bacterial stimulation but are instead a result of IL-6-induced inflammatory processes, which, in turn, stimulate the NF-κB and AP-1 pathways, leading to PCT gene expression, even in the absence of bacterial infection and toxin stimulation. These limitations of PCT levels in distinguishing between bacterial and viral infections, including COVID-19, have been previously documented, where Relph et al. demonstrated a lack of discriminatory value and poor diagnostic accuracy of PCT levels in hospitalized COVID-19 patients, with an AUC of 0.56, with a cutoff value of 0.25, as previously demonstrated in the literature [8], and Xu et al., who highlighted the poor ability of PCT levels to differentiate between bacterial and viral infections.

While CRP levels were significantly increased with adjustment, CRP is a nonspecific acute-phase protein that increases with bacterial infections and severe viral infections, limiting the discriminative power of this test in this context. In contrast, IL-6 levels were more consistent with an independent association with bacterial co-infection risk, providing support for the utility of IL-6 as a complementary test result when interpreted with WBC counts and microbiological data.

Overall, these findings support the larger body of literature showing that antibiotic therapy is common in COVID-19 patients despite the low prevalence of confirmed bacterial co-infections [2,3]. Our data suggest that antibiotic stewardship strategies, such as culture-based testing and integration with clinical data, rather than reliance on PCT alone, are warranted. The inverse association with PCT, despite adjustment, likely represents confounding by inflammation but does not suggest a true biological effect of reduced PCT levels. Although no formal calculation of a priori power was performed, the consistency of effect sizes with these data and international literature support the internal validity of these findings.

Limitations

There are a few limitations of the study to be considered while interpreting the results. The study has a few limitations. There was no formal sample size calculation done a priori, so we may have missed detecting small effects. The convenience sample from a big mass-gathering event may cause selection bias. The results may not be applicable to other clinical settings. Bacterial cultures were performed only if

indicated clinically. This may cause verification bias and misclassification of coinfection status. The biomarkers were measured 24-48 hours after presentation. This may not show the full dynamics of procalcitonin. We do not have detailed clinical severity scores and timing of antibiotic administration. This may have affected the biomarkers. We have adjusted for several important confounders in the regression analysis.

Conclusion

In this case-control study, procalcitonin isn't that accurate in differentiating bacterial coinfections among COVID-19 patients. The procalcitonin levels observed may be more related to IL-6-induced inflammation rather than bacterial coinfections. Thus, procalcitonin levels may not be used to make clinical decisions regarding antibiotic therapy in inflammatory viral diseases. A better approach would be to consider both microbiologic culture and inflammatory markers such as IL-6, and some useful blood work, to give a better clinical picture. This integrated approach, added to existing strategies, especially in risky situations such as large events, may improve antibiotic stewardship and reduce antibiotic overuse. However, further multicenter studies are necessary to validate this approach among different populations and clinical settings.

Funding: None.

Conflict of Interest: The authors declare no conflicts of interest.

AI Use: No AI-assisted technologies were used in the writing of this manuscript.

Ethics: The study protocol was reviewed and approved by the Research Ethics Committee, College of Science, University of Karbala (Ethical Approval ID No. [0021CSE] on [31-8-2025]). Written informed consent was obtained from all participants prior to their enrollment in the study.

Data Availability: De-identified data are available from the corresponding author upon reasonable request.

Acknowledgement: We thank Jood Laboratory for providing the facilities needed for this research. We also acknowledge the scientific and technical support from the University of Kerbala, College of Science, Department of Biology, and Al-Qasim Green University, College of Biotechnology. Their support was important for completing this study.

All procedures were conducted in accordance with institutional and international ethical standards, including data confidentiality, informed consent, and the Declaration of Helsinki (2013 revision).

References

1. Chen S, Zhu Q, Xiao Y, et al. Clinical and etiological analysis of coinfections and secondary infections in COVID-19 patients: an observational study. *Clin Respir J.* 2021;15(7):815-825. doi:10.1111/crj.13342
2. World Health Organization. WHO reports widespread overuse of antibiotics in patients hospitalized with COVID-19. Published April 26, 2024. Accessed August 23, 2025. Available from: https://www.who.int/news/item/26-04-2024-who-reports-widespread-overuse-of-antibiotics-in-patients--hospitalized-with-covid-19
3. Langford BJ, So M, Raybardhan S, et al. Antibiotic prescribing in patients with COVID-19: rapid review and meta-analysis. *Clin Microbiol Infect.* 2021;27(4):520-531. doi: 10.1016/j.cmi.2020.12.018
4. Russell CD, Fairfield CJ, Drake TM, et al. Coinfections, secondary infections, and antimicrobial use in patients hospitalised with COVID-19 during the first pandemic wave from the ISARIC WHO CCP-UK study: a multicenter, prospective cohort study. *Lancet Microbe.* 2021;2(8): e354-e365. doi:10.1016/S2666-5247(21)00090-2
5. Farrokhpour M, Kiani A, Mortaz E, et al. Procalcitonin and proinflammatory cytokines in early diagnosis of bacterial infections after bronchoscopy. *Open Access Maced J Med Sci.* 2019;7(6):913-919. doi:10.3889/oamjms.2019.183
6. Xu HG, Tian M, Pan SY. Clinical utility of procalcitonin and its association with pathogenic microorganisms. *Crit Rev Clin Lab Sci.* 2022;59(6):401-414. doi:10.1080/10408363.2022.2067425
7. Thwaites RS, Sanchez Sevilla Uruchurtu A, Siggins MK, et al. Inflammatory profiles across the spectrum of disease reveal a distinct role for GM-CSF in severe COVID-19. *Sci Immunol.* 2021;6(59): eabg9873. doi:10.1126/sciimmunol. abg9873
8. Relph KA, Russell CD, Fairfield CJ, et al. Procalcitonin is not a reliable biomarker of bacterial coinfection in people with coronavirus disease 2019 undergoing microbiological investigation at the time of hospital admission. *Open Forum Infect Dis.* 2022;9(5): ofac179. doi:10.1093/ofid/ofac179
9. Powell N, Howard P, Llewelyn MJ, et al. Use of procalcitonin during the first wave of COVID-19 in the acute NHS hospitals: a retrospective observational study. *Antibiotics (Basel).* 2021;10(5):516. doi:10.3390/antibiotics10050516
10. Chalmers JD, Crichton ML, Goeminne PC, et al. Management of hospitalised adults with coronavirus disease 2019 (COVID-19): a European Respiratory Society living guideline. *Eur Respir J.* 2021;57(4):2100048. doi:10.1183/13993003.00048-2021