

DOI: 10.4274/mjima.galenos.2026.26791
Mediterr J Infect Microb Antimicrob 2026;15:26791
Erişim: <http://dx.doi.org/10.4274/mjima.galenos.2026.26791>

Diagnostic Performance of Combined Routine Inflammatory Biomarkers for the Identification of Active Tuberculosis

Aktif Tüberkülozun Belirlenmesinde Kombine Rutin Enflamatuvar Biyobelirteçlerin Tanısal Performansı

● Kevser Elçi¹, ● Taylan Bozok¹, ● Lülüfer Tamer², ● Necmiye Canacankatan³, ● Eylem Sercan Özgür⁴, ● Gönül Aslan¹

¹Mersin University Faculty of Medicine, Department of Medical Microbiology, Mersin, Türkiye

²Mersin University Faculty of Medicine, Department of Medical Biochemistry, Mersin, Türkiye

³Department of Biochemistry, Faculty of Pharmacy, Mersin University, Mersin, Türkiye

⁴Mersin University Faculty of Medicine, Department of Chest Diseases, Mersin, Türkiye

Abstract

Introduction: Conventional diagnostic approaches for tuberculosis (TB) often require prolonged periods to yield definitive results, which may delay timely clinical decision-making. Host-derived inflammatory biomarkers reflecting innate immune responses have gained increasing attention; however, the diagnostic value of integrating the long pentraxin 3 (PTX3) with routinely available acute-phase proteins and hematological parameters remains insufficiently explored. We hypothesized that a multiparametric inflammatory panel reflecting systemic inflammation and innate immune responses could improve the diagnostic performance for TB. Therefore, this study aimed to evaluate the diagnostic performance of infection-related biomarkers, including PTX3, C-reactive protein (CRP), and serum amyloid A (SAA), together with hematological parameters (lymphocytes, monocytes, neutrophils, eosinophils, basophils, and platelets), and to investigate whether different biomarker combinations improve the identification of active TB.

Materials and Methods: In this cross-sectional study, peripheral blood samples were analyzed using a Sysmex XN-1000 hematology analyzer. CRP and SAA levels were measured using chemiluminescence, and PTX3 levels were measured using enzyme-linked immunosorbent assay. Diagnostic performance was assessed using receiver operating characteristic curve analysis.

Results: In the TB group, CRP, SAA, WBC, monocyte count (MONO), and NEU levels were significantly higher, whereas LYM and BASO levels were significantly lower than those in the control group ($p < 0.05$). Among the evaluated biomarker combinations, CRP-MONO yielded the numerically highest area under the curve (AUC; 0.943), followed by CRP-LYM (AUC, 0.934).

Conclusion: Our findings suggest that combining routinely available inflammatory biomarkers with hematological parameters may have potential utility in distinguishing patients with active TB from healthy controls. These exploratory findings require validation in larger, independent, multicenter cohorts before clinical application.

Keywords: C-reactive protein, infection, pentraxin 3, serum amyloid A, white blood cells, tuberculosis



Address for Correspondence/Yazışma Adresi: Kevser Elçi, PhD, Mersin University Faculty of Medicine, Department of Medical Microbiology, Mersin, Türkiye

E-mail: kvsrelci@gmail.com, kevserelci@mersin.edu.tr **ORCID ID:** orcid.org/0000-0002-2709-302X

Received/Geliş Tarihi: 16.03.2026 **Accepted/Kabul Tarihi:** 07.09.2026 **Published/Yayınlanma tarihi:** 05.10.2026

Cite this article as/Atıf: Elçi K, Bozok T, Tamer L, Canacankatan N, Sercan Özgür E, Aslan G. Diagnostic performance of combined routine inflammatory biomarkers for the identification of active tuberculosis. *Mediterr J Infect Microb Antimicrob*. 2026;15:245-254



©Copyright 2026 The Author(s). Published by Galenos Publishing House on behalf of Infectious Diseases and Clinical Microbiology Specialty Society of Turkey. Licensed under a Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License (CC BY-NC-ND 4.0)

Öz

Giriş: Tüberküloz (TB) için kullanılan geleneksel tanı yaklaşımları çoğu zaman kesin sonuç elde etmek için uzun süre gerektirmekte ve bu durum zamanında klinik karar verilmesini geciktirebilmektedir. Doğal bağışıklık yanıtını yansıtan konak kaynaklı inflamatuvar biyobelirteçler son yıllarda giderek daha fazla ilgi görmektedir. Ancak uzun pentraxin ailesinden olan PTX3'ün rutin olarak kullanılan akut faz proteinleri ve hematolojik parametrelerle birlikte değerlendirilmesinin tanısal değeri henüz yeterince araştırılmamıştır. Bu çalışmada, sistemik inflamasyonu ve doğal bağışıklık dinamiklerini yansıtan çok parametrelili bir inflamatuvar panelin TB tanısındaki performansı artırabileceği hipotez edilmiştir. Bu nedenle çalışmanın amacı, PTX3, C-reaktif protein (CRP) ve serum amiloid A (SAA) gibi enfeksiyon ilişkili biyobelirteçlerin; lenfosit, monosit, nötrofil, eozinofil, bazofil ve trombosit gibi hematolojik parametrelerle birlikte tanısal performansını değerlendirmek ve farklı biyobelirteç kombinasyonlarının TB hastalarının belirlenmesine katkısını araştırmaktır.

Gereç ve Yöntem: Bu kesitsel çalışmada periferik kan örnekleri Sysmex XN-1000 hematoloji analizörü kullanılarak analiz edilmiştir. CRP ve SAA düzeyleri kemilüminesans yöntemiyle, PTX3 düzeyleri ise ELISA yöntemi ile ölçülmüştür. Tanısal performans, alıcı işletim karakteristiği eğrisi analizi kullanılarak değerlendirilmiştir.

Bulgular: TB grubunda CRP, SAA, WBC, MONO ve NEU düzeyleri kontrol grubuna göre anlamlı derecede yüksek, LYM ve BASO düzeyleri ise anlamlı derecede düşük bulunmuştur ($p < 0,05$). Değerlendirilen biyobelirteç kombinasyonları arasında sayısal olarak en yüksek tanısal performans CRP–MONO modelinde (AUC: 0.943), ardından CRP-LYM modelinde (AUC: 0.934) elde edilmiştir.

Sonuç: Bulgularımız, rutin olarak elde edilebilen inflamatuvar biyobelirteçlerin hematolojik parametrelerle birleştirilmesinin, aktif TB'li hastaları sağlıklı kontrollerden ayırt etmede potansiyel bir fayda sağlayabileceğini düşündürmektedir. Bu keşif niteliğindeki bulguların klinik uygulamaya geçmeden önce daha büyük, bağımsız, çok merkezli kohortlarda doğrulanması gerekmektedir.

Anahtar Kelimeler: C-reaktif protein, enfeksiyon, pentraxin 3, serum amiloid A, beyaz kan hücreleri, tüberküloz

Introduction

Tuberculosis (TB) is one of the oldest known chronic infectious diseases in human history and remains a major cause of morbidity and mortality caused by *Mycobacterium tuberculosis*^[1]. Despite being treatable, it has remained a major global health problem for decades and was the leading cause of death from a single infectious pathogen until 2019^[2,3]. According to the World Health Organization (WHO), approximately one-quarter of the global population has been infected with *Mycobacterium tuberculosis* and is estimated to have latent TB infection^[4].

Inflammation plays an important role in the pathogenesis of TB^[5]. C-reactive protein (CRP), serum amyloid A (SAA), and pentraxin 3 (PTX3) are acute-phase reactants released in response to cytokines, particularly interleukin-6, during infectious diseases. These proteins contribute to the host response to infection and inflammation^[6,7]. PTX3 is a key component of innate immunity and functions as a soluble pattern recognition molecule. CRP is released from the liver, indicating a systemic response to local inflammation, whereas PTX3 is directly released by damaged tissues, reflecting the local inflammatory status^[8,9]. SAA also plays a significant role in acute and chronic inflammation and is used as an indicator of inflammation in clinical laboratories^[10]. Additionally, severe TB cases frequently exhibit hematological abnormalities^[11]. In TB, complete blood count indices, including neutrophil (NEU) count, platelet (PLT) count, and CRP levels, may be useful for determining disease severity and monitoring treatment response^[5]. Various studies have suggested that CRP and PLT distribution width could serve as potential biomarkers of TB severity^[12-14].

Despite advances in diagnostic technologies, current tools for the detection of *Mycobacterium tuberculosis* infection remain limited by delayed turnaround times and suboptimal sensitivity, leading to missed or delayed diagnoses worldwide^[15,16]. Although culture remains the gold standard for TB diagnosis, the time required to obtain results often extends over several weeks^[17]. Consequently, increasing attention has been directed toward host-derived inflammatory biomarkers that reflect the innate immune response during infection. While acute-phase proteins have been widely investigated, the diagnostic value of integrating the long pentraxin PTX3 with routinely available inflammatory and hematological parameters has not been sufficiently explored. We hypothesized that a multiparametric inflammatory panel reflecting both systemic inflammation and innate immune dynamics could improve diagnostic performance in TB. The novelty of the present study lies in the simultaneous evaluation of PTX3, conventional acute-phase reactants, and routinely available hematological parameters, as well as their two- and three-marker combinations, within the same cohort. Rather than focusing on a single biomarker class, this exploratory approach integrates markers reflecting different components of the inflammatory response and evaluates their diagnostic performance both individually and in combination for discriminating active TB from healthy controls. Therefore, the present study aimed to evaluate PTX3, CRP, and SAA together with peripheral blood cell counts and to investigate whether specific biomarker combinations enhance the identification of patients with active TB using a cost-effective and clinically applicable diagnostic strategy. Given the relatively small sample size and exploratory design, this study should be considered a pilot investigation intended to assess the potential diagnostic utility of combined inflammatory biomarkers in active TB.

Materials and Methods

This study was conducted in accordance with the ethical principles outlined in the 1975 Declaration of Helsinki. Ethical approval was obtained from the Mersin University Clinical Research Ethics Committee (decision number: 2023/359, dated May 24, 2023). Because the analyses were performed using blood samples collected as part of routine clinical testing, the Ethics Committee determined that additional written informed consent was not required. This was a single-center, cross-sectional study. Participants were consecutively enrolled during the study period, and blood samples collected during routine clinical evaluation were analyzed for the study biomarkers.

Study Groups

The target sample size was determined pragmatically based on the estimated number of eligible patients presenting to the Chest Diseases Outpatient Clinic. Hospital records from the preceding 12-month period indicated that approximately 50 patients who met the eligibility criteria had been diagnosed with TB. Considering the anticipated number of eligible patients during the study period, recruitment feasibility, and the potential for data loss, a minimum target of 20 patients with active TB and 20 healthy controls was established. No formal a priori sample-size or power calculation was performed. Participants were consecutively selected from individuals presenting to the Chest Diseases Outpatient Clinic between March 1, 2022, and March 1, 2023. TB was diagnosed based on clinical evaluation and confirmed by microbiological culture positivity for *Mycobacterium tuberculosis*. Patients with culture-confirmed active pulmonary TB (PTB) were included in the study. The patient group comprised 24 individuals with newly diagnosed active TB who had not previously received TB treatment and had no concomitant infectious or chronic diseases. The control group comprised 24 healthy volunteers recruited from individuals presenting to the hospital laboratory for routine laboratory testing during the same study period. Controls were not recruited from the Chest Diseases Outpatient Clinic. Individuals with a history of infectious or chronic disease or clinical symptoms suggestive of active infection were excluded from the control group.

Sample Collection

For PTX3, CRP, and SAA analyses, 7–10 mL of peripheral blood was collected into vacuum gel tubes without anticoagulant (Vacusera, Türkiye). For WBC, LYM, monocyte count (MONO), NEU, eosinophil (EOS), basophil (BASO), and PLT counts, 3–5 mL of peripheral blood was collected into hemogram tubes containing Na₂EDTA (Vacusera, Türkiye). Blood samples collected in vacuum gel tubes without anticoagulant were

centrifuged at 4,000 rpm for 10 minutes to obtain serum. Subsequently, approximately 1–1.5 mL of serum was carefully transferred to a sterile Eppendorf tube and stored at –20°C until analysis. All samples were analyzed within 6 months of storage at –20°C to maintain analytical integrity. Blood samples collected in Na₂EDTA tubes were analyzed within a maximum of 2 hours after collection.

Analysis Process

WBC, LYM, MONO, NEU, EOS, BASO, and PLT were analyzed within 1–2 hours after blood collection using an automated hematology analyzer (XN-1000, Sysmex Corp., Japan) based on electrical impedance and flow cytometry. Serum samples stored at –20°C until the day of analysis were thawed at room temperature before CRP, PTX3, and SAA measurements. Quantitative CRP and SAA measurements were performed using the chemiluminescence-based Maglumi X3 Automated System (Snibe Co. Ltd., Shenzhen, China), whereas PTX3 was measured using the Human Ptx3 Enzyme-Linked Immunosorbent Assay (ELISA) kit (Lot No. 201609, SunRedBio, Shanghai) according to the manufacturer's instructions.

Statistical Analysis

Quantitative data were analyzed using SPSS version 20.0 (IBM Corporation, Armonk, New York, USA). The normality of continuous variables was assessed using the Shapiro-Wilk test. Comparisons of continuous variables between the 2 groups were performed using the Mann-Whitney U test. Spearman rank correlation coefficients were calculated to assess correlations between the measured variables. Receiver operating characteristic (ROC) curve analysis was performed to evaluate diagnostic performance, and the area under the curve (AUC) was calculated. Optimal cut-off values were determined using Youden's index, defined as sensitivity + specificity – 1, with the threshold that maximized the Youden index selected as the optimal cut-off value. The AUCs of CRP alone and the CRP-MONO combination were compared using DeLong's test for correlated ROC curves. Binary logistic regression analysis was used to evaluate the combined diagnostic performance of the biomarkers, with TB status as the dependent variable. Regression coefficients (β), odds ratios (ORs), 95% confidence intervals (CIs), and p values were estimated for each model. The predicted probability of active TB was derived from the logistic regression equation, where $\text{logit}(p) = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \dots + \beta_k X_k$ and $p = 1/[1 + \exp(-\text{logit}(p))]$. Detailed regression coefficients and model-specific equations are provided in the Supplementary Material. For the CRP-MONO threshold-based analysis, the two biomarker thresholds were jointly evaluated using an OR classification rule. A participant was classified as positive when CRP was ≥ 5.44 mg/L or MONO was $\geq 1.04 \times 10^3/\mu\text{L}$. Sensitivity, specificity, and the Youden index

were calculated for this combined binary classification rule. This threshold-based analysis was distinct from the continuous combined biomarker analysis. Analyses with a p value <0.05 were considered statistically significant.

Results

A total of 48 individuals, including 24 patients with active TB and 24 healthy controls, were included in the study. In the TB group, 16 participants (66.7%) were male and 8 (33.3%) were female, whereas the control group consisted of 19 males (79.2%) and 5 females (20.8%). The mean age was 47.3 ± 20.8 years in the TB group and 41.7 ± 9.0 years in the control group, with no statistically significant difference between the groups ($p = 0.240$). The median (IQR) ages were 51 (28-61) years and 43 (37.8-48) years in the TB and control groups, respectively. All patients in the TB group had PTB. Nine patients (37.5%) with PTB had cavitory lesions on radiologic imaging. The acid-fast bacilli (AFB) smear positivity rate among TB patients was 83.3% (20/24), and at least one of the three *Mycobacterium tuberculosis* strains (12.5%) was resistant to a first-line anti-TB drug.

In the TB group, CRP, SAA, WBC, MONO, and NEU levels were significantly higher, whereas LYM and BASO levels were significantly lower than those in the control group ($p < 0.05$). No significant differences were observed between the groups in PTX3, PLT, or EOS levels ($p = 0.853$, $p = 0.170$, and $p = 0.116$, respectively) (Table 1). When the patient group was evaluated according to AFB smear positivity, only CRP levels differed significantly between AFB smear-positive and AFB smear-negative patients ($p = 0.042$).

When correlations between the parameters were analyzed, moderate positive correlations were observed between CRP and SAA, WBC, MONO, and NEU ($r = 0.597$, 0.308 , 0.395 , and 0.530 , respectively); between MONO and WBC, NEU, and EOS ($r = 0.674$, 0.608 , and 0.315 , respectively); and between LYM and WBC and BASO ($r = 0.315$ and 0.398 , respectively). A strong positive correlation was observed between NEU and WBC ($r = 0.894$) ($p < 0.05$). In addition, moderate negative correlations were observed between LYM and CRP ($r = -0.511$), LYM and SAA ($r = -0.443$), and CRP and BASO ($r = -0.407$) (Table 2).

The diagnostic performance analysis of different combinations of parameters revealed that the CRP-LYM (AUC, 0.934), CRP-NEU (AUC, 0.932), and PTX3-CRP (AUC, 0.905) pairs, as well as the CRP-MONO-LYM (AUC, 0.939), CRP-MONO-NEU (AUC, 0.934), CRP-WBC-NEU (AUC, 0.929), and CRP-PTX3-MONO (AUC, 0.917) triple combinations, yielded relatively high AUC values. However, given the non-significant diagnostic performance of PTX3 alone, the AUC values observed for PTX3-containing combinations should not be interpreted as evidence of an incremental diagnostic contribution from PTX3. Among the evaluated combinations, CRP-MONO yielded the numerically highest AUC (0.943) (Table 3, Figure 1). However, the difference between the AUC of CRP alone (0.936) and that of the CRP-MONO combination (0.943) was not statistically significant (DeLong test, $p = 0.307$).

When the diagnostic performance of the parameters was analyzed individually, CRP (AUC, 0.936), MONO (AUC, 0.828), and NEU (AUC, 0.795) demonstrated the best diagnostic performance as individual parameters for TB diagnosis. For differentiating patients with active TB from healthy controls, the optimal cut-off value for CRP was 4.97 mg/L, with a Youden index of 0.875, sensitivity of 91%, and specificity of 95%. The

Table 1. Median, minimum-maximum(min-max), and p values of PTX3, CRP, SAA, WBC, LYM, MONO, NEU, EOS, BASO, and PLT parameters in the tuberculosis and control groups.

Laboratory parameters	Tuberculosis group (n = 24) Median (min-max)	Control group (n = 24) Median (min-max)	p value
PTX3 ng/mL	1.509 (0.432–19.02)	1.451 (1.106–6.868)	0.853
CRP mg/L	47.470 (0.27–242.83)	2.100 (0.20–15)	< 0.001
SAA µg/mL	235.000 (0.57–300)	12.400 (0.10–300)	0.004
WBC $\times 10^3/\mu\text{L}$	9.595 (4.41–14.88)	7.780 (5.35–9.42)	0.030
LYM $\times 10^3/\mu\text{L}$	1.835 (0.58–3.12)	2.580 (1.15–3.49)	0.004
MONO $\times 10^3/\mu\text{L}$	0.805 (0.48–1.86)	0.570 (0.41–0.83)	< 0.001
NEU $\times 10^3/\mu\text{L}$	6.045 (2.82–10.78)	4.415 (2.46–5.88)	< 0.001
EOS $\times 10^3/\mu\text{L}$	0.150 (0.0–0.76)	0.060 (0.02–0.34)	0.116
BASO $\times 10^3/\mu\text{L}$	0.055 (0.010–0.55)	0.105 (0.020–1.26)	0.004
PLT $\times 10^3/\mu\text{L}$	293.500 (10.5–463)	263.000 (195–380)	0.170

n, Number of samples; p, values of significance with difference for each group. PTX3, pentraxin 3; CRP, C-reactive protein; SAA, serum amyloid A; WBC, white blood cell count; LYM, lymphocyte count; MONO, monocyte count; NEU, neutrophil count; EOS, eosinophil; BASO, basophils; PLT, platelet.

optimal cut-off value for MONO was $0.630 \times 10^3/\mu\text{L}$, with a Youden index of 0.542, sensitivity of 83%, and specificity of 71%. The optimal cut-off value for NEU was $5.35 \times 10^3/\mu\text{L}$, with a Youden index of 0.667, sensitivity of 75%, and specificity of 91% (Table 4). Furthermore, the CRP–MONO combination

yielded the numerically highest AUC (0.943), with cut-off values of 5.44 mg/L for CRP and $1.04 \times 10^3/\mu\text{L}$ for MONO. The Youden index was 0.875, with a sensitivity of 91% and specificity of 95% (Table 4).

Table 2. Spearman correlation coefficients (r values) among PTX3, CRP, SAA, WBC, LYM, MONO, NEU, EOS, BASO, and PLT.

	PTX3	CRP	SAA	WBC	LYM	MONO	NEU	EOS	BASO	PLT
PTX3	1.000	0.155	0.004	−0.064	0.038	−0.033	−0.097	0.127	0.039	−0.120
CRP	0.155	1.000	0.597**	0.308*	−0.511**	0.395**	0.530**	0.038	−0.407**	0.057
SAA	0.004	0.597**	1.000	0.078	−0.443**	0.275	0.244	−0.057	−0.219	0.109
WBC	−0.064	0.308*	0.078	1.000	0.315*	0.674**	0.894**	0.137	0.025	0.162
LYM	0.038	−0.511**	−0.443**	0.315*	1.000	0.150	−0.064	0.125	0.398**	−0.163
MONO	−0.033	0.395**	0.275	0.674**	0.150	1.000	0.608**	0.315*	−0.185	0.237
NEU	−0.097	0.530**	0.244	0.894**	−0.064	0.608**	1.000	0.101	−0.198	0.210
EOS	0.127	0.038	−0.057	0.137	0.125	0.315*	0.101	1.000	−0.198	0.014
BASO	0.039	−0.407**	−0.219	0.025	0.398**	−0.185	−0.198	−0.198	1.000	0.023
PLT	−0.120	0.057	0.109	0.162	−0.163	0.237	0.210	0.014	0.023	1.000

**Correlation is significant at the 0.01 level (2-tailed). *Correlation is significant at the 0.05 level (2-tailed). Correlation coefficients were interpreted as follows: $|r| \leq 0.30$, weak; $0.30 < |r| \leq 0.70$, moderate; and $|r| > 0.70$, strong. PTX3, pentraxin 3; CRP, C-reactive protein; SAA, serum amyloid A; WBC, white blood cell count; LYM, lymphocyte count; MONO, monocyte count; NEU, neutrophil count; EOS, eosinophil; BASO, basophils; PLT, platelet.

Table 3. PTX3, CRP, SAA, WBC, LYM, MONO, NEU, EOS, BASO, PLT, and the diagnostic performance values (AUC: Area Under the Curve).

Tests	AUC	95 % CI	p value
PTX3	0.516	0.346–0.686	0.853
CRP	0.936	0.851–1.000	<0.001
SAA	0.736	0.592–0.880	0.005
WBC	0.683	0.521–0.846	0.030
LYM	0.746	0.604–0.887	0.004
MONO	0.828	0.711–0.945	<0.001
NEU	0.795	0.651–0.939	<0.001
EOS	0.632	0.458–0.806	0.117
BASO	0.739	0.598–0.879	0.005
PLT	0.615	0.454–0.777	0.170
CRP–MONO	0.943	0.862–1.000	<0.001
CRP–LYM	0.934	0.849–1.000	<0.001
CRP–NEU	0.932	0.842–1.000	<0.001
PTX3–CRP	0.905	0.807–1.000	<0.001
CRP–MONO–LYM	0.939	0.857–1.000	<0.001
CRP–MONO–NEU	0.934	0.846–1.000	<0.001
CRP–WBC–NEU	0.929	0.835–1.000	<0.001
CRP–MONO–PTX3	0.917	0.824–1.000	<0.001

p, values of significance with difference in each group; CI, confidence interval, AUC, area under the curve; PTX3, pentraxin 3; CRP, C-reactive protein; SAA, serum amyloid A; WBC, white blood cell count; LYM, lymphocyte count; MONO, monocyte count; NEU, neutrophil count; EOS, eosinophil; BASO, basophils; PLT, platelet.

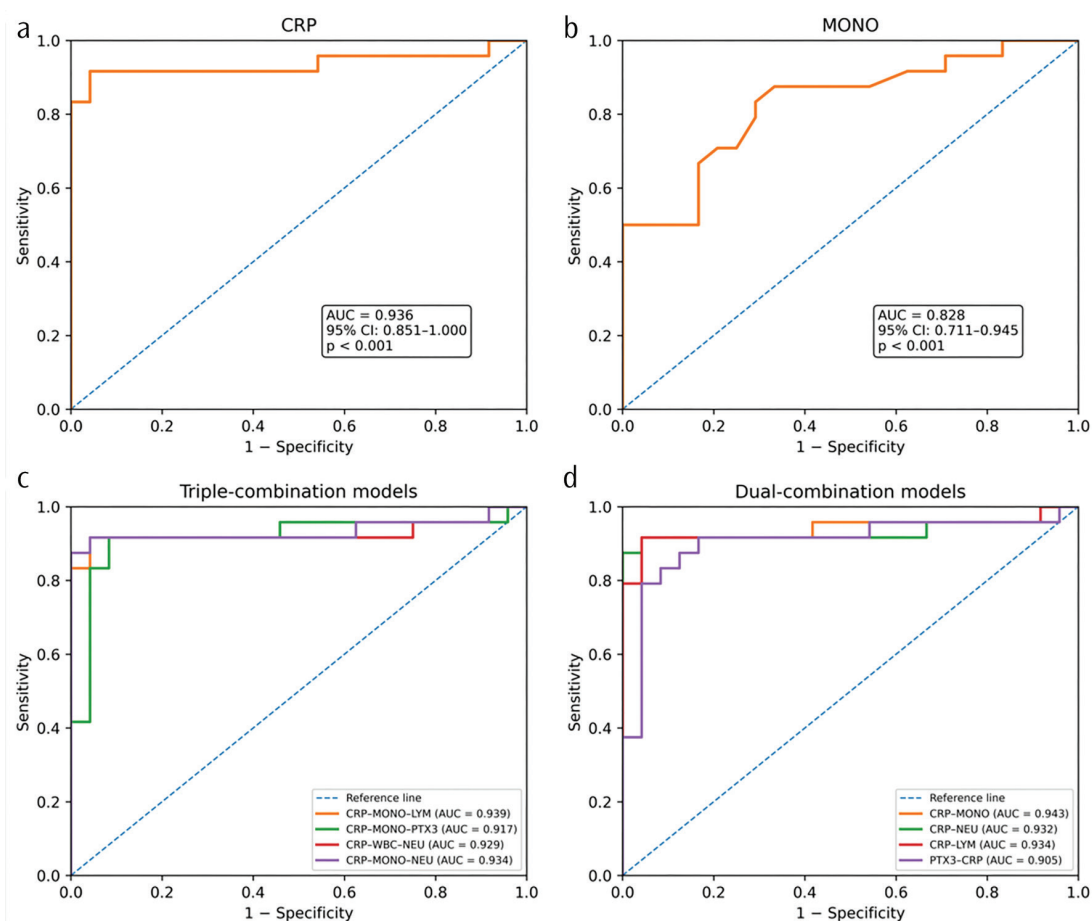


Figure 1. Receiver operating characteristic (ROC) curve analyses of inflammatory biomarkers and combined biomarker scores for the diagnosis of active TB. (A) ROC curve for CRP. (B) ROC curve for MONO. (C) ROC curves for selected triple-marker combinations. (D) ROC curves for selected dual-marker combinations. AUC, area under the curve; CI, confidence interval; CRP, C-reactive protein; LYM, lymphocyte count; MONO, monocyte count; NEU, neutrophil count; PTX3, pentraxin 3; WBC, white blood cell count.

Table 4. The cut-off values, sensitivity, specificity, and Youden indices for CRP, SAA, WBC, MONO, NEU, LYM, and BASO parameters, which exhibit significant differences between the TB and control groups.

	Cut-off point	Sensitivity	Specificity	Youden index
CRP	4.97 mg/L	0.917	0.958	0.875
SAA	41.75 µg/mL	0.667	0.833	0.5
WBC	$9.415 \times 10^3/\mu\text{L}$	0.542	0.958	0.5
MONO	$0.63 \times 10^3/\mu\text{L}$	0.833	0.708	0.542
NEU	$5.355 \times 10^3/\mu\text{L}$	0.75	0.917	0.667
LYM	$2.165 \times 10^3/\mu\text{L}$	0.667	0.833	0.5
BASO	$0.11 \times 10^3/\mu\text{L}$	0.875	0.5	0.375
CRP;MONO	5.44 mg/L; $1.04 \times 10^3/\mu\text{L}$	0.917	0.958	0.875

For the CRP-MONO combination, positivity was defined as CRP ≥ 5.44 mg/L or MONO $\geq 1.04 \times 10^3/\mu\text{L}$. Sensitivity, specificity, and the Youden index were calculated according to this combined binary classification rule. CRP, C-reactive protein; SAA, serum amyloid A; WBC, white blood cell count; LYM, lymphocyte count; MONO, monocyte count; NEU, neutrophil count; BASO, basophils; TB, tuberculosis.

Discussion

Rapid and accurate differentiation of patients with TB from healthy controls is crucial for effective disease management. However, traditional TB diagnostic methods can be costly and may require considerable time to yield definitive results. There remains an ongoing need for low-cost, accurate, and rapid methods, including triage tests, that are easily accessible and practical for use in clinical settings^[18,19]. Blood tests and biochemical analyses routinely performed in clinical settings can provide rapidly accessible indicators of disease^[20].

The importance of hematological parameters has been emphasized for predicting the risk of TB development, identifying active TB infection, and monitoring the response to antimicrobial therapy. Complete blood count parameters have been proposed as potential discriminatory biomarkers for inflammatory diseases^[21]. Acute-phase proteins such as PTX3, CRP, and SAA are routinely measurable and have been recommended as supportive discriminatory biomarkers in infectious diseases^[6,7]. The combined use of acute-phase proteins in bacterial infections has also been suggested to provide additional information and improve sensitivity for evaluating inflammatory status and supporting diagnosis^[10]. Therefore, the present study evaluated the diagnostic performance of whole blood indices (WBC, LYM, MONO, NEU, EOS, BASO, and PLT) and blood proteins (PTX3, CRP, and SAA), both individually and in combination, which can be readily analyzed from blood and serum samples in patients with active TB.

CRP is an acute-phase protein and a member of the pentraxin family that is produced in response to proinflammatory stimuli. CRP, which has been extensively studied in TB, has emerged as a promising marker for both diagnosis and monitoring of treatment response^[22,23]. Similarly, SAA, another acute-phase protein, has multiple immunological functions^[6]. SAA has been identified as a potential marker of TB and may play an important immunological role in reinforcing granulomatous responses to mycobacterial antigens^[24]. Several studies have reported significantly elevated levels of SAA and CRP in patients with TB^[25-27]. In the present study, both SAA and CRP levels were significantly higher in the TB group than in healthy controls ($p = 0.004$ and $p < 0.001$, respectively).

Despite being classified as a non-specific marker of infection, CRP has been shown in systematic reviews and meta-analyses to be superior to symptom-based approaches for identifying TB. Because of its sensitivity and specificity, the WHO has recommended including CRP in diagnostic algorithms for HIV-infected individuals^[28]. In a study by Samuels et al. involving 765 samples, a CRP cut-off value of 8 mg/L yielded a sensitivity

of 79.8% and a specificity of 62.8% for differentiating patients with TB from healthy controls^[22]. Recent WHO guidance highlights the importance of systematic screening and triage approaches for TB and emphasizes the need for tools that can identify individuals who require confirmatory diagnostic testing^[29]. In the present study, a CRP cut-off value of 4.97 mg/L yielded a sensitivity of 91% and a specificity of 95%. Although these sensitivity and specificity values were encouraging, they should be interpreted cautiously because the study compared patients with active TB with healthy controls rather than clinically relevant disease-control groups. Although CRP is a widely available and routinely measured inflammatory marker, the CRP cut-off value identified in the present study (4.97 mg/L) should also be interpreted cautiously, as biomarker thresholds may vary according to laboratory methods, assay characteristics, patient populations, and clinical settings. Therefore, this cut-off should not be considered a universally applicable diagnostic threshold and requires validation in independent and clinically representative cohorts before routine clinical use.

PTX3 plays a role in the acute-phase response to injury, trauma, and infection^[30]. PTX3, like CRP, is a member of the pentraxin family but is synthesized extrahepatically^[31]. Whereas CRP is produced almost exclusively in the liver, PTX3 is locally expressed by various cells at sites of inflammation and is stored in NEU-specific granules^[32,33]. In this study, although serum PTX3 levels were higher in patients with TB than in the control group, the difference was not statistically significant. Azzurri et al.^[31] reported that plasma PTX3 levels were significantly higher in patients with TB than in healthy controls. The discrepancy between their findings and those of the present study may be attributable to differences in the local synthesis of PTX3 or sample size. Although PTX3 alone did not demonstrate significant discriminatory performance, some PTX3-containing combinations yielded relatively high AUC values. However, these findings do not establish an incremental diagnostic contribution of PTX3 and should therefore be considered exploratory and validated in larger, independent cohorts.

In some diseases, specific types of WBCs increase in response to immune system activation, which may provide insight into the underlying cause of inflammation. WBC differentials (NEU, LYM, MONO, EOS, and BASO) are commonly used in the diagnosis of diseases affecting specific WBC subtypes^[34]. Hematological abnormalities have been reported to occur commonly in patients with TB, in whom the immune response plays a significant role in disease pathogenesis^[11]. Monocytes play a critical role in host immunity during *Mycobacterium tuberculosis* infection. NEUs also have multifaceted roles in host defense and are activated in response to bacterial

exposure during the early stages of infection. Both monocyte and NEU counts increase in TB and decrease significantly after treatment^[21]. Consistent with these findings, the present study observed elevated monocyte and NEU counts in patients with TB. Moreover, moderate correlations were observed between monocytes, NEUs, and CRP. Among the individual parameters, CRP, MONO, and NEU demonstrated the highest diagnostic performance, with AUCs of 0.936, 0.828, and 0.795, respectively. A formal comparison of the ROC curves was performed using DeLong's test. Although the CRP-MONO combination yielded a numerically higher AUC than CRP alone, the difference between the 2 ROC curves was not statistically significant (DeLong test, $p = 0.307$).

Specifically, lymphopenia has been observed in 46% of untreated patients with PTB and 76.92% of patients with miliary TB^[35]. Kang et al.^[36] demonstrated that lymphocyte counts were significantly lower in patients with active TB than in healthy controls. Similarly, in the present study, lymphocyte count was positively correlated with WBC and basophil counts and negatively correlated with CRP and SAA levels ($p = 0.004$).

PLT values have been associated with inflammatory reactions and immune responses. Research indicates that PLTs play an important role in the immune system^[21]. Tozkoparan et al.^[12] reported that PLT counts were significantly higher in patients with active TB. However, in the present study, no significant difference in PLT counts was observed between patients with active TB and controls. Conversely, other studies have associated thrombocytopenia with TB. For example, thrombocytopenia was reported in 37.5% of 128 Indian patients^[37]. Therefore, differences in sample size and participant selection may have contributed to the variation in findings.

Few studies have examined the combined diagnostic performance of multiple biomarkers in TB^[38]. This study comprehensively evaluated the diagnostic performance of combinations of PTX3, CRP, SAA, WBC, LYM, MONO, NEU, EOS, BASO, and PLT for distinguishing patients with active TB from healthy controls. An important feature of the present study was the integrated evaluation of biomarkers representing different components of the inflammatory response within the same exploratory framework. Specifically, PTX3 and conventional acute-phase reactants were evaluated together with routinely available hematological parameters, both individually and in multiple 2- and 3-marker combinations. This multiparametric approach extends the assessment beyond a single biomarker class and enables comparative evaluation of different inflammatory marker combinations within the same cohort.

Given the relatively small sample size and the evaluation of multiple biomarker combinations, the models may be susceptible to overfitting. Therefore, the reported diagnostic performance should be considered exploratory rather than definitive, and these findings should be regarded as hypothesis-generating until validated in larger, independent cohorts.

Although PTX3 levels were not significantly different between groups in this study, combinations containing PTX3 yielded relatively high AUC values. However, no incremental diagnostic contribution of PTX3 beyond CRP-based models was demonstrated. Therefore, the findings regarding PTX3 should be interpreted cautiously and considered exploratory. These findings suggest that selected biomarker combinations may warrant further investigation as practical and accessible diagnostic approaches, particularly in resource-limited settings. Larger, multicenter studies that include clinically relevant disease-control groups are needed to validate these findings before their clinical applicability can be established.

Study Limitations

The relatively small sample size and single-center design may restrict the generalizability of the findings. Furthermore, the study was limited to the pretreatment phase of TB, precluding assessment of biomarker changes during or after therapy. Internal validation techniques, such as bootstrap resampling or cross-validation, were not performed in this exploratory pilot study. Consequently, the reported model performance may be optimistic, and the robustness and generalizability of the prediction models remain uncertain. Therefore, these findings require validation in larger, independent, preferably multicenter cohorts. Future studies should also incorporate longitudinal follow-up to validate and extend these results for broader clinical application. Another limitation is the lack of detailed evaluation of potential confounding factors, such as smoking status, comorbid diseases, and subclinical inflammatory conditions, all of which may influence inflammatory biomarker levels. Therefore, the observed biomarker differences cannot be considered entirely specific to TB.

Another important limitation is the absence of clinically relevant disease-control groups. Consequently, the diagnostic performance reported in this study reflects discrimination between active TB and healthy individuals rather than the differential diagnosis encountered in routine clinical practice. Future studies should include patients with pneumonia, sarcoidosis, malignancy, and other inflammatory lung diseases. Because the control group consisted of healthy individuals, the specificity values observed in this study may be overestimated compared with those in real-world clinical settings, where patients with TB-like symptoms are evaluated.

Conclusion

This exploratory pilot study suggests that combinations of routinely available inflammatory and hematological parameters may have potential utility for distinguishing patients with active TB from healthy controls. Among the evaluated combinations, CRP–MONO yielded the numerically highest AUC. PTX3 alone showed limited discriminatory performance, and its incremental diagnostic contribution within combined models was not demonstrated. These findings should be considered hypothesis-generating rather than confirmatory and should not be interpreted as establishing clinically applicable diagnostic models. Larger, independent, multicenter studies that include clinically relevant disease-control groups are required to validate these findings before their potential clinical application can be established.

Ethics

Ethics Committee Approval: Ethical approval was obtained from the Mersin University Clinical Research Ethics Committee (decision number: 2023/359, dated May 24, 2023).

Informed Consent: Because the analyses were performed using blood samples collected as part of routine clinical testing, the Ethics Committee determined that additional written informed consent was not required.

Footnotes

Authorship Contributions

Surgical and Medical Practices: K.E., E.S.Ö., Concept: K.E., T.B., E.S.Ö., L.T., Design: K.E., T.B., N.C., Data Collection or Processing: K.E., N.C., G.A., Analysis or Interpretation: K.E., T.B., L.T., G.A., Literature Search: K.E., T.B., L.T., N.C., E.S.Ö., G.A., Writing: K.E.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: The authors declared that this study received no financial support.

References

- Gagneux S. Host-pathogen coevolution in human tuberculosis. *Philos Trans R Soc Lond B Biol Sci.* 2012;367:850-859.
- Saravanan M, Niguse S, Abdulkader M, Tsegay E, Hailekiros H, Gebrekidan A, Araya T, Pugazhendhi A. Review on emergence of drug-resistant tuberculosis (MDR & XDR-TB) and its molecular diagnosis in Ethiopia. *Microb Pathog.* 2018;117:237-242.
- Chakaya J, Khan M, Ntoumi F, Aklillu E, Fatima R, Mwaba P, Kapata N, Mfinanga S, Hasnain SE, Katoto PDMC, Bulabula ANH, Sam-Agudu NA, Nachega JB, Tiberi S, McHugh TD, Abubakar I, Zumla A. Global Tuberculosis Report 2020 - Reflections on the Global TB burden, treatment and prevention efforts. *Int J Infect Dis.* 2021;113 Suppl 1:S7-S12.
- World Health Organization. Global tuberculosis report 2024 [Internet]. Geneva: World Health Organization; 2024 [cited 2026 Jun 1]. Available from: <https://www.who.int/teams/global-programme-on-tuberculosis-and-lung-health/tb-reports/global-tuberculosis-report-2024>.
- Abakay O, Abakay A, Sen HS, Tanrikulu AC. The relationship between inflammatory marker levels and pulmonary tuberculosis severity. *Inflammation.* 2015;38:691-696.
- Vietri L, Fui A, Bergantini L, d'Alessandro M, Cameli P, Sestini P, Rottoli P, Bargagli E. Serum amyloid A: A potential biomarker of lung disorders. *Respir Investig.* 2020;58:21-27.
- Hacimustafaoğlu M. Akut Faz Reaktanları (Eritrosit Sedimentasyon Hızı, CRP). *J Pediatr Inf.* 2017;11:53-55.
- Mantovani A, Garlanda C, Doni A, Bottazzi B. Pentraxins in innate immunity: from C-reactive protein to the long pentraxin PTX3. *J Clin Immunol.* 2008;28:1-13.
- Ortega-Hernandez OD, Bassi N, Shoenfeld Y, Anaya JM. The long pentraxin 3 and its role in autoimmunity. *Semin Arthritis Rheum.* 2009;39:38-54.
- Zhang Y, Zhang J, Sheng H, Li H, Wang R. Acute phase reactant serum amyloid A in inflammation and other diseases. *Adv Clin Chem.* 2019;90:25-80.
- Atomsa D, Abebe G, Sewunet T. Immunological markers and hematological parameters among newly diagnosed tuberculosis patients at Jimma University Specialized Hospital. *Ethiop J Health Sci.* 2014;24:311-318.
- Tozkoparan E, Deniz O, Ucar E, Bilgic H, Ekiz K. Changes in platelet count and indices in pulmonary tuberculosis. *Clin Chem Lab Med.* 2007;45:1009-1013.
- Unsal E, Aksaray S, Köksal D, Sipit T. Potential role of interleukin 6 in reactive thrombocytosis and acute phase response in pulmonary tuberculosis. *Postgrad Med J.* 2005;81:604-607.
- Sahin F, Yazar E, Yıldız P. Prominent features of platelet count, plateletcrit, mean platelet volume and platelet distribution width in pulmonary tuberculosis. *Multidiscip Respir Med.* 2012;7:38.
- Druszczyńska M, Seweryn M, Wawrocki S, Kowalewska-Pietrzak M, Pankowska A, Rudnicka W. Cytokine Biosignature of Active and Latent Mycobacterium Tuberculosis Infection in Children. *Pathogens.* 2021;10:517.
- Escalante P, Theel ES. Progress Towards Developing a Rapid Triage/Referral Test for Tuberculosis. *Clin Chem.* 2020;66:995-997.
- Babu S. Biomarkers for Treatment Monitoring in Tuberculosis: A New Hope. *EBioMedicine.* 2017;26:13-14.
- Al-Zamel FA. Detection and diagnosis of Mycobacterium tuberculosis. *Expert Rev Anti Infect Ther.* 2009;7:1099-1108.
- Ahmad R, Xie L, Pyle M, Suarez MF, Broger T, Steinberg D, Ame SM, Lucero MG, Szucs MJ, MacMullan M, Berven FS, Dutta A, Sanvictores DM, Tallo VL, Bencher R, Eisinger DP, Dhingra U, Deb S, Ali SM, Mehta S, Fawzi WW, Riley ID, Sazawal A, Premji Z, Black R, Murray CJL, Rodriguez B, Carr SA, Walt DR, Gillette MA. A rapid triage test for active pulmonary tuberculosis in adult patients with persistent cough. *Sci Transl Med.* 2019;11:eaaw8287.
- Zhan M, Xue H, Wang Y, Wu Z, Wen Q, Shi X, Wang J. A clinical indicator-based prognostic model predicting treatment outcomes of pulmonary tuberculosis: a prospective cohort study. *BMC Infect Dis.* 2023;23:101.
- Ştefanescu S, Cocoş R, Turcu-Stiolica A, Mahler B, Meca AD, Giura AMC, Bogdan M, Shelby ES, Zamfirescu G, Pisoschi CG. Evaluation of prognostic significance of hematological profiles after the intensive phase treatment in pulmonary tuberculosis patients from Romania. *PLoS One.* 2021;16:e0249301.
- Samuels THA, Wyss R, Ongarello S, Moore DAJ, Schumacher SG, Denkinge CM. Evaluation of the diagnostic performance of laboratory-based c-reactive protein as a triage test for active pulmonary tuberculosis. *PLoS One.* 2021;16:e0254002.
- Rao S, Bernhardt V. Serum C-Reactive Protein in Pulmonary Tuberculosis. *Infect Dis Clin Pract (Baltim Md).* 2009;17:314-316.
- Chen ES, Song Z, Willett MH, Heine S, Yung RC, Liu MC, Groshong SD, Zhang Y, Tudor RM, Moller DR. Serum amyloid A regulates granulomatous inflammation in sarcoidosis through Toll-like receptor-2. *Am J Respir Crit Care Med.* 2010;181:360-373.

25. Jiang TT, Shi LY, Wei LL, Li X, Yang S, Wang C, Liu CM, Chen ZL, Tu HH, Li ZJ, Li JC. Serum amyloid A, protein Z, and C4b-binding protein β chain as new potential biomarkers for pulmonary tuberculosis. *PLoS One*. 2017;12:e0173304.
26. de Beer FC, Nel AE, Gie RP, Donald PR, Strachan AF. Serum amyloid A protein and C-reactive protein levels in pulmonary tuberculosis: relationship to amyloidosis. *Thorax*. 1984;39:196-200.
27. Jacobs R, Malherbe S, Loxton AG, Stanley K, van der Spuy G, Walzl G, Chegou NN. Identification of novel host biomarkers in plasma as candidates for the immunodiagnosis of tuberculosis disease and monitoring of tuberculosis treatment response. *Oncotarget*. 2016;7:57581-57592.
28. Abdulgader SM, Okunola AO, Ndlangalavu G, Reeve BWP, Allwood BW, Koegelenberg CFN, Warren RM, Theron G. Diagnosing Tuberculosis: What Do New Technologies Allow Us to (Not) Do?. *Respiration*. 2022;101:797-813.
29. World Health Organization. Operational handbook on tuberculosis. Module 2: Screening – systematic screening for tuberculosis disease. Geneva: World Health Organization; 2021. Accessed June 01, 2026. Available from: <https://www.who.int/publications/i/item/9789240022614>.
30. Mortaz E, Adcock IM, Tabarsi P, Masjedi MR, Mansouri D, Velayati AA, Casanova JL, Barnes PJ. Interaction of Pattern Recognition Receptors with Mycobacterium Tuberculosis. *J Clin Immunol*. 2015;35:1-10.
31. Azzurri A, Sow OY, Amedei A, Bah B, Diallo S, Peri G, Benagiano M, D'Elis MM, Mantovani A, Del Prete G. IFN-gamma-inducible protein 10 and pentraxin 3 plasma levels are tools for monitoring inflammation and disease activity in Mycobacterium tuberculosis infection. *Microbes Infect*. 2005;7:1-8.
32. Garlanda C, Bottazzi B, Magrini E, Inforzato A, Mantovani A. PTX3, a Humoral Pattern Recognition Molecule, in Innate Immunity, Tissue Repair, and Cancer. *Physiol Rev*. 2018;98:623-639.
33. Liu S, Qu X, Liu F, Wang C. Pentraxin 3 as a prognostic biomarker in patients with systemic inflammation or infection. *Mediators Inflamm*. 2014;2014:421429.
34. Haffor ASA, Dahan T, Ismaeel M. Reliable quantitative score for grading chest X-ray using the dynamic of blood cell count in adult asthma. *Egypt J Chest Dis Tuberc*. 2015;64:325-334.
35. Li F, Chen D, Zeng Q, Du Y. Possible Mechanisms of Lymphopenia in Severe Tuberculosis. *Microorganisms*. 2023;11:2640.
36. Kang YJ, Park H, Park SB, Kim J, Lee J, Kim J, Park S, Lee YS, Kim S. Combined analysis of whole blood interferon gamma release assay and complete blood count analysis for rapid discrimination of active tuberculosis and latent tuberculosis infection. *J Clin Tuberc Other Mycobact Dis*. 2021;24:100253.
37. Kutiya AS, Gupta N, Garg S, Hira HS. A Study of Haematological and Haemostasis Parameters and Hypercoagulable State in Tuberculosis Patients in Northern India and the Outcome with Anti-Tubercular Therapy. *J Clin Diagn Res*. 2017;11:OC09-OC13.
38. Garay-Baquero DJ, White CH, Walker NF, Tebruegge M, Schiff HF, Ugarte-Gil C, Morris-Jones S, Marshall BG, Manousopoulou A, Adamson J, Vallejo AF, Bielecka MK, Wilkinson RJ, Tezera LB, Woelk CH, Garbis SD, Elkington P. Comprehensive plasma proteomic profiling reveals biomarkers for active tuberculosis. *JCI Insight*. 2020;5:137427.