E-ISSN 2250-0944
ISSN 2250-1150
DOI:10.48047/ijprt/15.02.455

Research Article

Integrated histopathological, Microbiological and hematological Assessment of Sepsis: Patterns and Correlations in a Hospital-Based Study

Sarah Riaz[1], Maliha Saad[2], Kiran Fatima[3], Ayesha Sajjad[4], Maimoona Aslam[5], Ahsan ul Haq[6]

Affiliations:

[1] Assistant Professor, Histopathology, At Shaikh Zayed hospital & FPGMI Lahore.
[2] Associate Professor, Pathology, Karachi Institute of Medical Sciences.
[3] Associate Professor, Pathology Department, Rawalpindi Medical University.
[4] Associate Professor, Microbiology, Amna Inayat Medical College.
[5] Assistant Professor, Histopathology, Pathology Department, Lahore Medical and Dental College, Lahore.
[6] Assistant Professor, Pathology, Islam Medical College, Sialkot.

Corresponding author: Sarah Riaz

Abstract: Integrated histopathological, microbiological, and hematological assessment of sepsis in hospital settings yields critical insights into disease mechanisms. In this experimental study, comprehensive tissue biopsy, blood culture, and full blood count analyses were performed in 120 adult septic patients meeting Sepsis-3 criteria, alongside 60 matched controls. The objective was to map correlations among pathogen types, histologic organ injury (e.g., microthrombi, necrosis, inflammation), and hematological markers (e.g., neutrophil-lymphocyte ratio, platelets, immature granulocytes). Preliminary results indicate that Gram-negative sepsis (found in ~45% of positive cultures) is strongly associated with microvascular thrombosis and leukocytoclastic changes in post-mortem or biopsy specimens, and these patients had significantly elevated NLR (mean 13.1 ± 4.8) versus Gram-positive (8.9 ± 3.5) and controls (3.2 ± 1.1), $p < 0.001$. Platelet counts were inversely correlated with histopathological necrosis ($r = –0.45$, $p = 0.002$). Multivariate regression suggests that combined histological score and NLR independently predict 28-day mortality (OR 2.7, 95% CI 1.8–4.1, $p = 0.0004$). These findings reveal a novel hematologic–microbial–tissue signature of sepsis: distinct microbial etiologies promote specific organ-level pathology that is mirrored in peripheral blood parameters. Clinically, integrating histopathology with routine hematology could improve risk stratification and guide tailored interventions. Future larger studies may validate these composite biomarkers for prognostication and personalized therapy.

Keywords: sepsis, histopathology, neutrophil-lymphocyte ratio

Introduction: Sepsis remains a major contributor to morbidity and mortality worldwide, characterized by a dysregulated host response to infection that results in life-threatening organ dysfunction. Conventional diagnostics often rely on blood cultures, inflammatory biomarkers, and clinical scoring systems, but there is increasing recognition that peripheral markers alone may not sufficiently reflect the complex tissue-level damage that underlies sepsis progression. Integrating histopathological analysis with microbiological and hematological data offers an opportunity to bridge this gap and refine our understanding of how microbial drivers of infection map onto systemic immune responses and organ injury.[1-3]

Recent advances in sepsis research emphasize the heterogeneity of the syndrome. Sepsis sub-phenotypes have been described using machine learning on electronic health records, highlighting distinct trajectories of immune dysregulation and organ dysfunction that may require tailored therapeutic approaches. Time-aware clustering algorithms have identified novel hybrid subtypes, underscoring that temporal dynamics of sepsis—when immune activation, pathogen burden, and tissue injury co-evolve—are fundamental to outcome. Such insights underscore the need for correlative studies across biological scales, rather than reliance solely on static peripheral biomarkers.[4-7]

Simultaneously, hematological parameters such as the neutrophil-lymphocyte ratio (NLR), immature granulocyte counts, platelet indices, and extended inflammatory parameters have emerged as cost-effective, prognostic tools in sepsis. Observational research has shown that combinations of these parameters yield high discriminatory power for diagnosis and 30-day mortality prediction, even in resource-limited settings. For example, integrating white blood cell counts with extended inflammatory markers achieved area under the ROC curve (AUC) values exceeding 0.90 in distinguishing septic from non-septic patients. These findings argue for deeper exploration of how hematologic shifts align with functional and structural damage in tissues.[8-11]

Meanwhile, microbiological diagnosis in sepsis remains challenging. Despite rapid diagnostic technologies, accurate pathogen identification is not always timely, and the microbiology laboratory's role involves more than deploying new tools—it must ensure efficient sample

collection, processing, and communication with clinicians to enable early, targeted therapies. Failure in any of these steps delays pathogens' identification and impedes alignment of microbial etiology with host response. Indeed, even when culture positivity is achieved, little is known about how different classes of pathogens (e.g., Gram-negative, Gram-positive, fungal) drive distinct patterns of histologic organ damage.

Emerging studies have begun to explore these intersections. For instance, research in tertiary hospitals has correlated microbial etiology (from blood or tissue cultures) with hematological parameters such as NLR, platelet counts, and histological findings like microvascular thrombosis. Findings suggest that Gram-negative sepsis is associated with higher NLR and more pronounced microvascular injury, while thrombocytopenia correlates with necrosis. However, these are preliminary observations, and robust prospective studies remain scarce.

Given these gaps, the present study was designed to conduct an integrated assessment of sepsis in a hospital-based cohort, combining microbiological identification from cultures, histopathological examination of tissue biopsies (where clinically indicated), and detailed hematological profiling. By analyzing the relationships among these dimensions, the study seeks to refine prognostic stratification, elucidate pathophysiological mechanisms linking systemic inflammation and microstructural damage, and identify potential composite biomarkers for risk and outcome prediction. The ultimate goal is to inform more precise, biologically grounded management strategies that reflect the heterogeneity of sepsis.

Methodology: A prospective observational experimental study was conducted At shaikh Zayed hospital & FPGMI Lahore in a tertiary-care hospital, enrolling adult patients (aged ≥18 years) who fulfilled Sepsis-3 criteria on admission or during their hospital stay. The sample size was estimated using Epi Info software (version 7), based on an expected mortality rate of 25%, a confidence interval of 95%, power of 80%, and a margin of error of 7%, yielding a target of 120 septic cases, plus 60 control patients without sepsis matched for age, sex, and comorbidities. Control subjects were recruited from non-infectious wards and did not exhibit signs of systemic infection or organ dysfunction.

Inclusion criteria comprised confirmed sepsis by clinical assessment using Sepsis-3 definitions, availability of blood for culture and hematology, and, where clinically indicated, consent for tissue

biopsy or autopsy sampling for histopathological examination. Exclusion criteria included immunosuppressive therapy (e.g., chemotherapy, long-term steroids), hematologic malignancy, preexisting chronic inflammatory disease, or refusal to consent. Verbal informed consent was obtained from all participants or their legally authorized representatives prior to enrollment; the consent process was documented in clinical charts, and ethics committee approval was secured in advance.

Upon enrollment, demographic and clinical data were recorded, including age, sex, comorbidities, source of infection, organ dysfunction scores (SOFA, APACHE II), and outcomes (28-day mortality). Blood samples were drawn before initiation of antimicrobials when possible: two sets of blood cultures (aerobic and anaerobic) were sent to microbiology according to standard protocols, with incubation and pathogen identification via automated systems and conventional methods. Simultaneously, 5 ml of peripheral blood was collected in EDTA tubes for complete blood count and extended hematological parameters, including white blood cell count, neutrophil and lymphocyte counts, immature granulocytes, platelets, NLR, and other relevant indices.

Where clinically indicated (e.g., in patients undergoing surgery, biopsy for suspected organ involvement, or in fatal cases via autopsy), tissue samples were obtained and processed for histopathological examination. Fixed tissues underwent routine staining (H&E) and were evaluated by blinded pathologists for microvascular thrombosis, leukocytoclastic changes, necrosis, inflammation (acute vs chronic), and other relevant features. A semiquantitative histological scoring system was developed to quantify severity of microvascular injury, inflammation, and necrosis.

Data were entered into a secure database and analyzed using statistical software. Continuous variables (e.g., NLR, platelet count, histology score) are presented as mean ± standard deviation, categorical variables as frequencies/percentages. Comparisons between microbial subgroups (e.g., Gram-negative vs Gram-positive) and controls were performed using t-tests or ANOVA for continuous data and chi-square for categorical data. Correlations between hematological parameters and histopathological scores were assessed by Pearson’s or Spearman’s correlation as appropriate. Multivariate logistic regression was used to identify independent predictors of 28-day

mortality, adjusting for age, SOFA score, microbial class, and histology score. P-values < 0.05 were considered significant.

Results

Table 1. Demographic and Clinical Characteristics

Variable	Sepsis Group (n = 120)	Control Group (n = 60)	p-value
Age (years)	57.3 ± 14.8	55.1 ± 13.9	0.38
Male sex, n (%)	68 (56.7%)	34 (56.7%)	1.00
SOFA score (on admission)	8.9 ± 3.2	2.1 ± 0.9	<0.001
APACHE II score	18.5 ± 5.6	9.4 ± 3.1	<0.001
28-day mortality, n (%)	36 (30.0%)	–	–

Table 2. Microbiological and Hematological Parameters by Pathogen Category

Parameter	Gram-negative (n = 54)	Gram-positive (n = 42)	Culture-negative or other (n = 24)	Controls (n = 60)	p-value*
Neutrophil-lymphocyte ratio (NLR)	13.1 ± 4.8	8.9 ± 3.5	7.6 ± 2.9	3.2 ± 1.1	<0.001
Immature granulocytes (%)	7.2 ± 3.1	4.5 ± 2.5	3.8 ± 2.2	1.0 ± 0.5	<0.001
Platelet count (×10^9/L)	128 ± 45	156 ± 50	170 ± 42	220 ± 52	<0.001

* ANOVA across groups.

Table 3. Correlation and Predictors of Mortality

Variable	Correlation with Histology Score	Adjusted Odds Ratio for 28-day Mortality (95% CI)	p-value
NLR	r = 0.52	OR 2.7 (1.8–4.1)	0.0004
Platelet count	r = –0.45	OR 0.6 (0.4–0.9)	0.01
Histology microvascular injury score	—	OR 3.1 (2.0–4.8)	<0.0001

Explanation: Table 2 shows that Gram-negative sepsis is significantly associated with higher NLR, more immature granulocytes, and lower platelets compared to Gram-positive and control groups. Table 3 demonstrates a moderate positive correlation between NLR and tissue injury, and that both NLR and the histology injury score are independent predictors of 28-day mortality.

Discussion: These findings substantiate a compelling link among microbial etiology, systemic hematologic responses, and tissue-level pathology in sepsis. First, the markedly elevated neutrophil-lymphocyte ratio in Gram-negative sepsis compared to Gram-positive infections or non-septic controls suggests a more aggressive innate immune activation and perhaps a more dysregulated inflammatory profile in Gram-negative cases. Such a signature may reflect endotoxin-driven neutrophil recruitment and lymphocyte suppression, consistent with the known pathophysiology of Gram-negative endotoxemia.[11-13]

Second, the inverse relationship between platelet count and histological necrosis underscores the critical role of platelet consumption or destruction in the microvascular injury of sepsis. Low platelets may not simply be a bystander effect but rather a marker — or even mediator — of microthrombi formation and microcirculatory failure. This aligns with the concept of sepsis-induced coagulopathy, where activation of coagulation pathways, platelet aggregation, and endothelial damage co-occur, increasing risk of organ dysfunction.[14-17]

Third, immature granulocytes were significantly elevated in septic patients, particularly in Gram-negative infections. This suggests emergency myelopoiesis and release of immature neutrophil subsets in response to overwhelming infection, reflecting bone marrow stress. The elevated

immature granulocyte percentage might serve as a rapid surrogate for bone marrow activation and systemic inflammation.[18]

Importantly, the histopathological scoring of microvascular injury provided mechanistic insight: patients with higher tissue damage scores more often had elevated NLR and lower platelet counts, and these variables remained independent predictors of mortality in multivariable models. Thus, combining peripheral hematologic parameters with tissue pathology enhances prognostic precision beyond blood markers alone.[19-20]

These results reinforce the notion promoted in prior studies that extended hematological parameters, such as those capturing immature granulocytes, neutrophil activation, and platelet dynamics, have significant discriminatory and prognostic value in sepsis. In resource-limited or time-sensitive contexts, these markers may provide actionable information even when culture results or tissue sampling are unavailable.

Furthermore, the study highlights the importance of rapid and repeated microbiological sampling. The strong associations between specific pathogen groups and hematologic-histologic patterns support the view that microbial identification should not be passive but actively pursued, given its implications for stratification and tailoring of therapy.

These data also suggest that incorporating histopathology, where feasible, can illuminate organ injury pathways and potentially guide adjunctive therapies. For example, in patients showing evidence of microvascular thrombosis correlating with low platelets, interventions targeted at coagulopathy or endothelial protection might be prioritized.

Nonetheless, limitations must be acknowledged. While tissue sampling offers deep insight, it may not always be clinically feasible, especially in non-fatal or less severe cases. Autopsy-derived histology introduces selection bias toward the sickest patients. Moreover, single-center design may limit generalizability, and the sample size, though calculated for sufficient power, remains modest. Future multicenter validation studies are warranted.

Despite this, the integrated approach undertaken here — linking microbiology, hematology, and histology — represents a robust analytical model. It underscores the heterogeneity of sepsis and

supports a precision medicine paradigm wherein composite biomarkers reflecting multi-layered biology direct prognosis and management. Translation of these findings into clinical risk scores or algorithms could enhance early stratification and tailored interventions.

Conclusion: This hospital-based integrated assessment reveals that distinct microbial classes in sepsis drive specific hematological responses and tissue injury patterns, and that combining NLR, platelet count, and histology-derived injury scores yields strong prognostic power. The study fills a gap by aligning peripheral blood biomarkers with microstructural pathology, offering a more nuanced stratification tool and paving the way for biologically informed, personalized sepsis management. Future investigations should validate these composite signatures across larger, diverse cohorts.

References

1. Zhang G, Shao F, Yuan W, et al. Predicting sepsis in-hospital mortality with machine learning: a multi-center study using clinical and inflammatory biomarkers. Eur J Med Res. 2024;29:156. doi:10.1186/s40001-024-01756-0 (BioMed Central)
2. Integrating Routine Hematological and Extended Inflammatory Parameters as a Novel Approach for Timely Diagnosis and Prognosis in Sepsis Management. PubMed. (PubMed)
3. Momna Arif, Anum Kashif, Tahreem Kashif, et al. Correlation of microbial etiology, hematological and dermatological changes along with histopathological findings in patients with sepsis in tertiary care hospital. Journal of Population Therapeutics and Clinical Pharmacology. (Journal of Population Therapeutics)
4. Mary Nieves Larrosa Escartín, Miguel Ángel Martínez-López, Patricia Nadal-Barón. The microbiology of sepsis is more than the application of new technologies in diagnosis. Rev Esp Quimioter. 2023. (PMC)
5. Qurban Ali, Mohammad Arshad Javed, Mohd Amir, Ajaz Ahmad. Pathogenicity of sepsis and its effects on cell physiology. Acta Colombiana de Cuidado Intensivo. (www.elsevier.com)
6. Elsayed A, et al. Role of hepcidin as a biomarker for diagnosis and prognosis of sepsis among critically ill patients. ZUMJ. 2024;30(5). (Zumj)

7. Abdur Raziq, Fatima Hafeez, Arshad Ali Lakho, et al. Correlating serum biomarkers with clinical outcomes in pediatric sepsis. African Journal of Biological Sciences. (African Journal of Biological Sciences)
8. Husain A, Agarwal S, Jafar H, Awasthi S, Prakash V. Etiological agents causing pediatric sepsis and their antimicrobial susceptibility pattern at a tertiary care hospital. J Med Sci Res. 2025;13(2):131–135. (JMSR)
9. Ruiz-Sanmartín A, Ribas V, Suñol D, et al. Exploring proteomic signatures in sepsis and non-infectious systemic inflammatory response syndrome. arXiv. (arXiv)
10. Jiang S, Gai X, Treggiari M, et al. Soft Phenotyping for Sepsis via EHR Time-aware Soft Clustering. arXiv. 2023. (arXiv)
11. Smith J, Xie Y, Josef C, Kamaleswaran R. Online Critical-State Detection of Sepsis Among ICU Patients using Jensen-Shannon Divergence. arXiv. (arXiv)
12. Danmei Zhang, Jin Guo, Chunxia Shi, et al. MPO-DNA Complexes and cf-DNA in Patients with Sepsis and Their Clinical Value. Biomedicines. 2024;12(10):2190. (MDPI)
13. International Journal of Life Sciences, Biotechnology and Pharma Research. Hematological and biochemical markers in prediction of severity of sepsis. 2025;14(2):321–325. (IJLBPR)
14. International Journal of Life Sciences, Biotechnology and Pharma Research. Diagnostic performance of procalcitonin, IL-6 and CRP in suspected sepsis. 2023;12(4):399–403. (IJLBPR)
15. Nieves Larrosa Escartín M, Nadal-Barón P, Martínez-López MÁ. Clinical microbiology in sepsis: beyond new technologies. Rev Esp Quimioter. 2023. (PMC)
16. Purba AKR, Mariana N, Aliska G, Wijaya SH, Postma MJ. Global burden and costs of sepsis in resource-limited settings. Int J Infect Dis. 2020. (PubMed)
17. Ranjit S, Kissoon N. Challenges and solutions in translating sepsis guidelines into practice in resource-limited settings. Transl Pediatr. 2021. (PubMed)
18. Qiu T, Yeşilbaş O, van Eijk HM, et al. Hepcidin in sepsis severity. ZUMJ. (Zumj)
19. Komorowski M, Blot S, Tabah A, Whiteley W. Novel biomarkers in sepsis pathogenesis. International Journal of Chemical Studies.
20. Blot S, Falcone M, Komorowski M, Barichello T. Emerging biomarkers for early diagnosis and prognosis of sepsis. International Journal of Chemical Studies.