Journal of Karnali Academy of Health Sciences
An Official Journal of Karnali Academy of Health Sciences

Comparative study of serum bilirubin and total leukocyte count in assessing the severity of acute appendicitis

Dinesh Shahi[1], **Ananda Neupane**[2], **Atmanand Verma**[1], **Sushil Khaniya**[2], **Suryaman Menyangbo**[3]

[1]Lecturer, Department General Surgery, Rapti Academy of Health Sciences, Ghorahi, Dang, Nepal
[2]Assistant Professor, Department General Surgery, Rapti Academy of Health Sciences, Ghorahi, Dang, Nepal
[3]Professor, Department General Surgery, Rapti Academy of Health Sciences, Ghorahi, Dang, Nepal

ABSTRACT

Introduction: Appendicitis is diagnosed based on the combination of clinical assessment, laboratory investigation, and radiological test. Distinguishing between uncomplicated and complicated appendicitis remains a frequent challenge in clinical practice. This study is designed to evaluate and compare the diagnostic utility of serum bilirubin (TB) and total leucocyte count (TLC) in severity of appendicitis.

Methods: This is a retrospective cross sectional analytical study with a sample size of 167 undergoing open appendectomy from January 2025 to January 2026 Preoperatively TLC with TB was done. Severity of appendicitis was divided in uncomplicated and complicated. Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and diagnostic accuracy were calculated. The optimal cut off value for each parameter and area under the curve calculated using ROC curve.

Results: A total of 167 patients were included, with mean age± SD was 34.38 ± 17.80 years ,most common age group10-20 years. The sensitivity, specificity, PPV, NPV and accuracy of TLC with cut off value >11000 were 64.28%, 32.99%, 40.90%, 56.14% and 46.10%, for TB with cut off value >1.4mg/dl were 41.42%, 68.04%, 48.33%, 61.68% and 56.88%. The area under the curve (AUC) of TLC, TB and N/L ratio were 0.601, 0.573 and 0.745. The diagnostic accuracy of TB over TLC is statistically significant (p value <0.001).

Conclusion: Increased bilirubin level has higher diagnostic accuracy than leukocytosis for severity of appendicitis. Combining inflammatory biomarkers such as TLC and TB improves diagnostic accuracy in appendicitis.

Keywords: Appendicitis, Bilirubin, Complicated, Leucocyte Count, Uncomplicated

INTRODUCTION

Appendicitis is one of the most common causes of acute abdominal pain requiring urgent surgical intervention.[1] About 7-8% of the population will experience appendicitis in their lifetime, with the highest prevalence rates occurring between the 2nd and 3rd decades of life, and the age-adjusted mortality rate is 1.00 per 100,000 population .[2,3,4]

Diagnosis of appendicitis is based on the combination of clinical assessment, laboratory investigation, and radiological investigation. In many cases, the complete blood count is a commonly performed laboratory investigation. Because the specificity of leukocytosis is low and it is a common feature of the systemic inflammatory response, it is less useful as a single parameter.[5] Recently, bilirubin levels have been proposed as an auxiliary marker to evaluate the severity of appendicitis.[6] Increased bilirubin may indicate a more common pathway of bacterial translocation and endotoxemia, which is more frequent in severe presentations, such as perforated or gangrenous appendicitis, and can serve as a surrogate marker.[7,8]

Despite the significant advances in imaββging and laboratory diagnostics, distinguishing between uncomplicated and complicated appendicitis, such as gangrenous, perforated, or abscess, remains a frequent challenge in clinical practice. Existing studies provide mixed results: increased bilirubin levels are a better predictor of appendicular perforation than leukocytosis.[9,10,11] Similar results were obtained from a study done in India and Nepal too.[12,13] The diagnostic yield of TLC and serum bilirubin is modest when used alone; however, when combinations of these variables are used, the ability to discriminate between uncomplicated and complicated appendicitis is significantly increased.[14]

This study is designed to evaluate and compare the diagnostic accuracy of serum bilirubin and TLC in differentiating uncomplicated from complicated acute appendicitis. The ultimate aim is to improve preoperative risk assessment and support timely clinical decision-making, potentially reducing morbidity associated with delayed or missed diagnosis of severe appendiceal disease.

METHODS

This is a retrospective cross sectional analytical study done at tertiary hospital. Case was complicated appendicitis and control was uncomplicated appendicitis. Ethical approval was taken from institutional review committee (IRC-RAHS). Considering the sensitivity and specificity of total bilirubin is 0.88 and 0.26 and for leukocytosis is 0.83 and 0.62.[7] Sample size(n)= $((Z_{1-\alpha/2}+ Z_{1-\beta})^2 \cdot (1 - \rho)) / (\Delta AUC)^2$. Where, $Z_{1-\alpha/2}$=1.96 (for 95% confidence), $Z_{1-\beta}$ =0.84 (for 80% power), ρ=0.6 (assumed correlation), and ΔAUC=0.725−0.57=0.155.

The calculated sample size was 167. The patients who underwent emergency open appendectomy from January 2025 to January 2026 however case of interval appendectomy, appendicitis with known liver disease and age less than 10 years were excluded from study. Patients who presented to the emergency department with symptoms suggestive of acute appendicitis underwent routine preoperative investigations, including serum total bilirubin and total leukocyte count (TLC) along with other preoperative test for surgery. Open appendectomy was doe as per standard procedure by consultant general surgeons. The

Corresponding Author:
Dr. Suryaman Menyangbo
Email: suryaman77@gmail.com

Date of submission: March 17, 2026
Date of Acceptance: April 28, 2026
Date of publication: May 10, 2026

DOI: https://doi.org/ 10.61814/jkahs.v9i1.1177

morphology of appendix was noted as normal looking appendix, inflamed, gangrenous or perforated appendicitis. These values were recorded following surgery, patients are classified into one of two groups, uncomplicated appendicitis or complicated appendicitis, based on what was found during the operation and confirmed by histopathological examination. Those with perforation, gangrene, or abscess formation are included as complicated, whereas only inflammation of the appendix is counted as uncomplicated appendicitis.

Data was collected in a preformed proforma and entered in a Microsoft Excel sheet and analyzed using Statistical package for social science (SPSS) version 16. The central tendency for continuous data was measured with the mean±standard deviation for the data being normally distributed. For comparison of bilirubin and total count and neutrophil to lymphocyte ratio across uncomplicated and complicated appendicitis group, unpaired t- test was applied. Sensitivity, specificity, positive predictive value, negative predictive value and diagnostic accuracy were calculated for increased bilirubin and leucocyte and neutrophil to lymphocyte count. The cut off value for each parameter (TLC, TB, N/L ratio) and area under the curve (AUC) were defined using receiver operating characteristic (ROC) curve with Youden index.[15] A 95 % confidence interval was taken and a p-value less than 0.05 was termed as statistically significant. This article has been reported in line with STROBE guideline.[16]

RESULTS

A total 167 patients were included in this study. The mean age± SD was 34.38 ± 17.80 years old and the most common age group is 11-20 years of age (29.20%). Out of 167 patient male to female ratio was 47.3 to 52.7 with decreasing trend of male patient after 20 years of age (table 1)

Table 1: Age wise distribution of patient (n= 167)

Age (Years)	No of patient		Percentage (%)	Mean age ±SD
	Male	Female		
11-20	33	17	29.94	34.38 ± 17.80
21-30	11	19	17.96	
31-40	7	21	16.77	
41-50	12	17	17.37	
51-60	5	5	5.98	
60+	11	8	11.37	
Total	79	87	167 (100%)	

Morphologically, the most common finding was inflamed appendicitis followed by perforated appendicitis (26.95%). Out of 97 case one case histopathological was found to be fibrous obliteration. (Table 2)

Table 2: Distribution of patient as per morphology of appendix (n= 167)

Morphology of appendix	Number (N=167)	Percent (%)
Inflamed appendicitis	97	58.08
Gangrenous appendicitis	25	14.97
Perforated appendicitis	45	26.95

Comparing the two group of appendicitis, uncomplicated versus complicated appendicitis) the age distribution was similar in two group. The mean ± SD for age, total leucocyte count, total bilirubin, and neutrophil to lymphocyte (N/L ratio) as per table 3. The mean of TLC and TB and Neutrophil to lymphocytes ratio statistically significant between two group with p value < 0.05 (Table 3)

The sensitivity, specificity positive, PPV, NPV and accuracy of TLC with cut off value >11000 were 64.28%, 32.99%, 40.90%, 56.14% and 46.10%. similarly, sensitivity, specificity positive, PPV, NPV and accuracy of TB with cut off value >1.4mg/dl were 41.42%, 68.04%, 48.33%, 61.68% and 56.88%. The diagnostic performance of serum bilirubin and leucocyte count is statistically significantly with p value <0.001.(Table 4)

Table 3: Comparison of Age, TLC, TB, N/L ratio between uncomplicated and complicated appendicitis (n=167)

Parameter	Morphology of appendix (mean ± SD)		P value (t -test)
	Uncomplicated (n=97)	Complicated (n=70)	
Age	32.03 ± 15.96	37.62 ± 19.74	0.053
Total leucocyte count (cells/ mm3)	11739 ± 3554.78	13407 ± 4495.32	0.011
Neutrophil to lymphocyte (N/L) ratio	6.67 ± 4.74	13.07 ± 9.92	<0.001
Total bilirubin	1.27 ± 0.82	1.59 ± 1.15	0.049

Table 4: Predictive marker of severity of appendicitis with leukocytosis (TLC>11000) and increased bilirubin (TB>1.4mg/dl) (n=167)

Parameters	Morphology of appendix		
	Complicated	Un-complicated	
Total leucocyte count (TLC) cells/mm3 Total Leucocyte count			
Increased (>11000)	45	65	Sensitivity =64.28% Specificity =32.99% PPV= 40.90%, NPV= 56.14% Accuracy = 46.10%
Normal (≤11,000)	25	32	
Total bilirubin (mg/dl)			
Increased (>1.4)	29	31	Sensitivity =41.42% Specificity =68.04% PPV= 48.33%, NPV= 61.68% Accuracy = 56.88%
Normal (≤1.4)	41	66	

***PPV: Positive predictive value, NPV: negative predictive value, McNemar χ^2 = 16.10, p < 0.001 for complicated appendicitis**

Analyzing the receiver operating curve (ROC) curves, the area under the curve (AUC) of total leucocyte count (TLC), total bilirubin (TB) and neutrophil lymphocyte ratio (N/L) were 0.601, 0.573 and 0.745. Regarding the cutoff value of the Youden index for TLC was 14275 with sensitivity and specificity was 41.4% and 82%, for total bilirubin 1.63mg/dl with sensitivity and specificity was 32% and 80.4% and N/L ratio of 6.7 was sensitivity and specificity are was 80% and 62% (Figure 1)

DISCUSSION

Acute appendicitis is among the most prevalent surgical emergencies across the globe, and prompt diagnosis of complicated appendicitis is key factor early intervention and decrease the morbidity. Appendicitis is mainly diagnosed by clinical, laboratory and radiological investigation. Several parameters in laboratories have been explored as possible predictors of disease severity. The present study aimed to determine the diagnostic usefulness of total leukocyte count

(TLC), total bilirubin (TB), and neutrophil-to-lymphocyte (N/L) ratio in distinguishing between uncomplicated and complicated appendicitis.

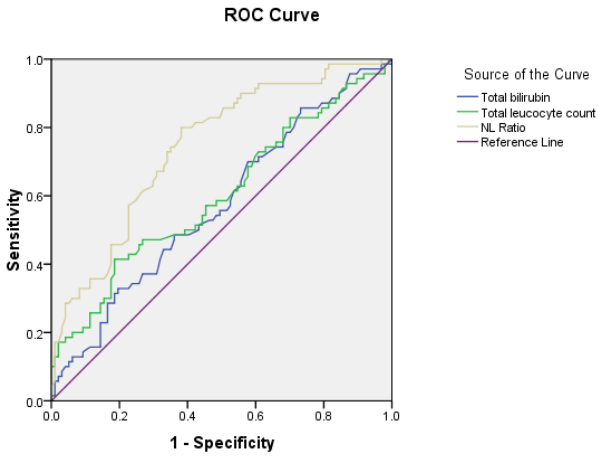

Figure 1: Receiver operating characteristic (ROC) curve (TLC, TB, N/L ratio)

In this study, the mean age of 34.38 ± 17.80 years, with 11-20 years being the most affected group (29.2%). This observation coincides with the previous literature indicating that acute appendicitis is more common among adolescents and young adults. [3,17,18] The male-to-female ratio in the current study exhibited a mild female predisposition (47.3 to 52.7). The number of male patients decreasing after 20 years of age which could be due to the increased number of males going out for education, job opportunity.

In this study, the mean ± SD for TLC count in complicated and uncomplicated appendicitis is 13407 ± 4495.32cell/mm^3 and 11739 ± 3554.78 cell/mm^3) higher than uncomplicated appendicitis. Similar finding with other study[19,20,21] Leukocytosis is a marker of inflammation Nonetheless, its diagnostic specificity in predicting complications is limited. The current investigation revealed that TLC at a cut-off of >11,000 cells/mm3 had a sensitivity of 64.28% but a low specificity of 32.99%, However the sensitivity and specificity was 41.4% and 82% with the cutoff value of the Youden index for TLC was 14275. The area under the curve is 0.573 indicating poor predictor of severity of appendicitis. Similar to studied done by coleman et al.[22] Regarding neutrophil to lymphocyte ratio is significantly high (uncomplicated 6.67 ± 4.74 and complicated 13.07 ± 9.92) similar to multiple studies[23,24] but have poor severity prediction and lack accurate estimation of postoperative complications or total length of hospital stay (LOS)[25]some healthcare settings cannot offer imaging to all patients. Resource-limited hospitals have to make a diagnosis of acute appendicitis on a combination of physical exam and laboratory results. The neutrophil-to-lymphocyte ratio (NLR).

Our study shows the mean ± SD total bilirubin level in complicated appendicitis is significantly high than uncomplicated appendicitis (1.59 ± 1.15 and 1.27 ± 0.82). Total bilirubin with a cut-off of exceeding 1.4 mg/dL in this study had a sensitivity of 41.42% and specificity of 68.04%. However, the sensitivity and specificity is 32% and 80.4% with the cutoff value of the Youden index for total bilirubin 1.63mg/dl. The specificity is somewhat higher, indicating that hyperbilirubinemia could be more effective in diagnosing complicated appendicitis rather than diagnosing it. Pathophysiological processes of hyperbilirubinemia in appendicitis are considered to be associated with bacterial endotoxemia and hepatic dysfunction due to systemic inflammatory response, which disrupts bilirubin excretion. Serum bilirubin has also been studied as a predictor of complicated appendicitis. A number of studies have shown that elevated bilirubin levels are significantly associated with perforated or gangrenous appendicitis.[7,8,26]

In this study, total leucocyte count, total bilirubin, and neutrophil to lymphocyte ratio were significantly higher among patients with complicated appendicitis than uncomplicated appendicitis ($p < 0.05$). Comparing the total leucocyte count, total bilirubin the sensitivity, specificity, PPV, NPV and accuracy for leukocytosis (>11000cells/mm3) are 64.28%, 32.99%, 40.90%, 56.14% and 46.10% and for increased bilirubin level (1.4mg/dl) are 41.42%, 68.04%, 48.33%, 61.68%, 56.88% showing bilirubin is less sensitive, more specific and has higher accuracy to predict the severity of appendicitis with p value <0.001 for complicated appendicitis which is similar to other studies results.[10,11,12,13,14] however, Comparing the area under curve for TLC and bilirubin shows 0.601, 0.573 not statistically significant (p value 0.46) In the current study, the combination of leukocytosis and hyperbilirubinemia enhanced diagnostics performance. In parallel analysis, sensitivity rose to 84.94%, better detecting complicated appendicitis, but specificity dropped. On the other hand, the specificity of series testing was 78.56% indicating that combined lab values could be more diagnostic than any single marker alone. The same results have been documented in research that has shown that a combination of inflammatory biomarkers improves diagnostic accuracy in appendicitis.[27,28]

CONCLUSION

The outcome of this study shows that total leucocyte count and total bilirubin and neutrophil to lymphocyte ratio are a good diagnostic marker of severe appendicitis. Comparing between these tests increased bilirubin level has more diagnostic accuracy than leukocytosis for severity of appendicitis. Using the combination of inflammatory biomarkers TLC, TB and neutrophil lymphocyte ratio improves diagnostic accuracy in appendicitis.

DECLARATIONS

Author Contributions

DS reviewed the literature, conceptualized and designed the research, proposal development, contributed to data acquisition, analysis, statistical work, and manuscript preparation and review; AN reviewed the literature, manuscript preparation; AV conceptualized and designed the research, contributed to data acquisition; SK analysis, statistical work, and manuscript preparation and review; SM conceptualized and designed the research, contributed to data acquisition, analysis, statistical work, and manuscript preparation and review; All authors approved the final version of the manuscript and agreed to be accountable for all aspects of the research work

Acknowledgements

The author would like to acknowledge the medical officers and nursing staff for maintaining clinical records and facilitating data retrieval for the study

Ethical Approval

Approval for study was taken from administration (reference no 239, 2082/083) and ethical clearance was taken from institutional review committee (IRC- RAHS)with reference number 706 on November 2, 2025.

Consent/Assent

Since this study is retrospective study so consent/assent is not applicable

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Conflicts of Interest

Author(s) declare no conflict of interest

Source of Funding

The author(s) received no external fund for this research

REFERENCES

1. Lotfollahzadeh S, Lopez RA, Deppen JG. Appendicitis. In: StatPearls. Treasure Island (FL): StatPearls | PubMed |

2. Addiss, D. G., Shaffer, N., Fowler, B. S., & Tauxe, R. V. (1990). The epidemiology of appendicitis and appendectomy in the United States. American journal of epidemiology, 132(5), 910–925. |PubMed |

3. Panthi J, Shahi D, Pokhrel B. Epidemiological Profile and Seasonal Variation of Acute Appendicitis. J Rapti Academy Health Science. 2024;1(2):58–62. |DOI |

4. World Life Expectancy. Appendicitis in Nepal.https://www.worldlifeexpectancy.com/nepal-appendicitis |Weblink |

5. Baddam S, Burns B. Systemic Inflammatory Response Syndrome. StatPearls Publishing; 2025. |PubMed |

6. Motie MR, Nik MM, Gharaee M. Evaluation of the diagnostic value of serum level of total bilirubin in patients with suspected acute appendicitis. Electron Physician. 2017 Apr 25;9(4):4048. |PubMed |

7. M S, Fg B, T HL, D S, G M, B M. Diagnostic value of hyperbilirubinemia as a predictive factor for appendiceal perforation in acute appendicitis. Am J Surg. 2009 Aug;198(2). |DOI |

8. Akai, M., Iwakawa, K., Yasui, Y., Yoshida, Y., Kato, T., Kitada, K., Hamano, R., Tokunaga, N., Miyaso, H., Tsunemitsu, Y., Otsuka, S., Inagaki, M., & Iwagaki, H. (2019). Hyperbilirubinemia as a predictor of severity of acute appendicitis. The Journal of international medical research, 47(8), 3663–3669. |PubMed |

9. Eren, T., Tombalak, E., Ozemir, I. A., Leblebici, M., Ziyade, S., Ekinci, O., & Alimoglu, O. (2016). Hyperbilirubinemia as a predictive factor in acute appendicitis. European journal of trauma and emergency surgery : official publication of the European Trauma Society, 42(4), 471–476. |DOI |

10. Kar S, Behera TK, Jena K, Sahoo AK. Hyperbilirubinemia as a Possible Predictor of Appendiceal Perforation in Acute Appendicitis: A Prospective Study. Cureus. 2022 Feb 2;14(2):e21851. |PubMed |

11. Judal H, Ganatra V, Choudhary P ram. The diagnostic value of total leucocytes count, C-reactive protein and total bilirubin in acute appendicitis: a prospective study. Int Surg J. 2022 Jan 29;9(2):372–7. |DOI |

12. Sevinç MM, Kınacı E, Çakar E, Bayrak S, Özakay A, Aren A, et al. Diagnostic value of basic laboratory parameters for simple and perforated acute appendicitis: an analysis of 3392 cases. Ulus Travma Ve Acil Cerrahi Derg Turk J Trauma Emerg Surg TJTES. 2016 Mar;22(2):155–62. |DOI |

13. Ghimire P, Thapa P, Yogi N, Ghimire P. Role of serum bilirubin as a marker of acute gangrenous appendicitis. Nepal J Med Sci. 2012 Aug 2;1(2):89–92. |DOI |

14. Andersson REB. Meta-analysis of the clinical and laboratory diagnosis of appendicitis. Br J Surg. 2003 Dec 29;91(1):28–37. doi:10.1002/bjs.4464 |Fulltext |

15. Youden Index - an overview | ScienceDirect Topics. |Fulltext |

16. Strengthening the Reporting of Observational Studies in Epidemiology (STROBE): explanation and elaboration. PLoS Med. 2007 Oct 16;4(10):e297. PMID: 17941715 |Fulltext |

17. Addiss, D. G., Shaffer, N., Fowler, B. S., & Tauxe, R. V. (1990). The epidemiology of appendicitis and appendectomy in the United States. American journal of epidemiology, 132(5), 910–925. |DOI |

18. Lotfollahzadeh S, Lopez RA, Deppen JG. Appendicitis. In: StatPearls. Treasure Island (FL): StatPearls | PubMed |

19. Ribeiro AM, Romero I, Pereira CC, Soares F, Gonçalves Á, Costa S, et al. Inflammatory parameters as predictive factors for complicated appendicitis: A retrospective cohort study. Ann Med Surg. 2022 Feb 1;74:103266. |DOI |

20. Puputti J, Pakarinen MP, Suominen JS. Differentiating Uncomplicated From Complicated Acute Appendicitis in Paediatric Patients With C-Reactive Protein and Imaging Findings-A Retrospective Single Centre Study in 1149 Patients. Acta Paediatr. 2026 Feb;115(2):399-407 |PubMed |

21. Guraya SY, Al-Tuwaijri TA, Khairy GA, Murshid KR. Validity of leukocyte count to predict the severity of acute appendicitis. Saudi Med J. 2005 Dec;26(12):1945–7. PubMed PMID: 16380778. |PubMed |

22. Coleman C, Thompson JE, Bennion RS, Schmit PJ. White blood cell count is a poor predictor of severity of disease in the diagnosis of appendicitis. Am Surg. 1998 Oct;64(10):983–5. PubMed PMID: 9764707. |PubMed |

23. Neupane J, Aacharya RP, Gupta SK, Mishra A, Mahat P, Kc B. Role of neutrophil to lymphocyte ratio in predicting severity of acute appendicitis. J Gen Pract Emerg Med Nepal [Internet]. 2024 [cited 2026 Mar 12];11(18):56–9. Located |Fulltext |

24. Kelly, M. E., Khan, A., Riaz, M., Bolger, J. C., Bennani, F., Khan, W., Waldron, R., Khan, I. Z., & Barry, K. (2015). The Utility of Neutrophil-to-Lymphocyte Ratio as a Severity Predictor of Acute Appendicitis, Length of Hospital Stay and Postoperative Complication Rates. Digestive surgery, 32(6), 459–463. | DOI |

25. Morales-Morales CA, Llamas-Ostos AN, Maciel-Garcia PA, Zambrano-Lara M, Fukumoto-Inukai KA, Quevedo-Fernandez E, et al. Neutrophil-lymphocyte ratio as a severity predictor in acute appendicitis. Int Surg J. 2026;13(1):14–8. doi:10.18203/2349-2902.isj20254322| Fulltext|

26. Sandstrom, A., & Grieve, D. A. (2017). Hyperbilirubinaemia: its utility in non-perforated appendicitis. ANZ journal of surgery, 87(7-8), 587–590. |DOI |

27. Dinç T, Sapmaz A, Erkus Y, Yavuz Z. Complicated or non-complicated appendicitis? That is the question. Turk J Trauma Emerg Surg. 2022 Mar 1;28(3):390 |PubMed |

28. Patmano M, Çetin DA, Gümüs T. Laboratory markers used in the prediction of perforation in acute appendicitis. Turk J Trauma Emerg Surg. 2022 Jul 1;28(7):960–6. |PubMed |