ORIGINAL ARTICLE

The role of ischemia-modified albumin, presepsin, delta neutrophil index, and inflammatory markers in diagnosing acute cholecystitis

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ABSTRACT

BACKGROUND: The purpose of this study is to determine the significance of markers such as C-reactive protein, procalcitonin, complete blood count parameters, delta neutrophil index, ischemia-modified albumin, presepsin, and oxidative stress indicators, which are associated with inflammation, oxidative stress, and ischemia in the pathology and diagnosis of acute cholecystitis in adults.

METHODS: Patients diagnosed with acute cholecystitis in the emergency department and healthy individuals in the control group were included in the study. Routine blood count and biochemistry analyses were performed on the participants. Blood serum was used to measure ischemia-modified albumin, presepsin, and oxidative stress indicators.

RESULTS: White blood cell count, neutrophil-to-lymphocyte ratio, platelet-to-lymphocyte ratio, delta neutrophil index, C-reactive protein, procalcitonin, ischemia-modified albumin, ischemia-modified albumin to albumin ratio, presepsin, and oxidative stress indicators were significantly higher in patients with cholecystitis compared to the control group. Measurements of white blood cell count, neutrophil-to-lymphocyte ratio, platelet-to-lymphocyte ratio, and delta neutrophil index can be included as part of the complete blood count. The complete blood count parameters are readily available and do not incur additional costs to the healthcare system.

CONCLUSION: The authors believe that the neutrophil-to-lymphocyte ratio, delta neutrophil index, ischemia-modified albumin, ischemia-modified albumin to albumin ratio, and presepsin values can be used as new markers in the diagnosis of acute cholecystitis due to their high sensitivity, specificity, and low negative likelihood ratio.

Keywords: Acute cholecystitis; delta neutrophil index; ischemia-modified albumin; neutrophil-to-lymphocyte ratio; presepsin.

INTRODUCTION

Acute cholecystitis (AC) is one of the most common causes of acute abdomen that necessitates emergency surgical intervention.[1,2,3,4] Biochemical markers such as white blood cell (WBC) count, C-reactive protein (CRP), procalcitonin (PCT), and the neutrophil/lymphocyte ratio (NLR) have been identified as potential predictors of cholecystitis.[1,2,3,4] Recent studies have highlighted the role of oxidative stress and imbalances in the oxidative defense system in the pathogenesis of several ischemia-related diseases.[1,5,6,7,8]

Ischemia-modified albumin (IMA), a variant of albumin produced during ischemia and oxidation, serves as an indicator of hypoperfusion and oxidative stress.[1,2,3] This marker is recognized for its relevance in ischemia-related conditions, including stroke, acute mesenteric ischemia, acute appendicitis, acute cholecystitis, and acute coronary syndrome.[9,10,11,12,13]

WBC, neutrophil, lymphocyte, and platelet (PLT) parameters from the complete blood count, along with their ratios-NLR and platelet-to-lymphocyte ratio (PLR)-as well as CRP and PCT, are utilized to evaluate the inflammatory process in the pathology and progression of acute cholecystitis.

The delta neutrophil index (DNI), a marker indicating the presence of immature granulocytes in peripheral circulation, has been shown to be elevated in inflammatory conditions.[14]

Presepsin is reported to possess high sensitivity and specificity for the early diagnosis and prognosis prediction of sepsis. Numerous studies have highlighted its effectiveness in diagnosing sepsis and assessing inflammation, prognosis, and mortality.[15]

The degree of oxidative stress and the efficacy of the antioxidant defense system can be determined through laboratory tests, including total antioxidant status (TAS), total oxidant status (TOS), and the oxidative stress index (OSI).[9,16]

In this study, markers such as CRP, PCT, complete blood count parameters, DNI, IMA, presepsin, TAS/TOS, and OSI, which are associated with inflammation, oxidative stress, and ischemia and play roles in the pathology and disease progression of AC, were investigated in adults with acute cholecystitis. The aim is to elucidate the disease mechanisms and the relationships among these markers. Many of these markers are tested automatically as part of a standard complete blood count and are readily available blood parameters that do not incur additional costs to the healthcare system. We believe that the results of the new biomarkers examined in this study will assist clinicians in diagnosing acute cholecystitis.

MATERIALS AND METHODS

This study is a methodological epidemiological investigation aimed at diagnosing acute cholecystitis. It was conducted prospectively, examining new biomarkers such as Presepsin, IMA, and Immunoglobulins (IG) for diagnosing AC. The sensitivity and specificity values of these new biomarkers were analyzed. The study included 53 individuals who presented at the emergency department between February 2022 and May 2022. Participants were over 18 years of age, had no other significant comorbidities, and were diagnosed with acute cholecystitis. The control group consisted of 48 healthy individuals matched by age and sex. Inclusion criteria for the study were adult patients who presented to the emergency department with abdominal pain and were diagnosed with acute cholecystitis. Exclusion criteria were as follows: patients not diagnosed with acute cholecystitis; patients under the age of 18 (non-adults);

patients with chronic diseases; patients with data errors or incompleteness; patients who voluntarily left the clinic before completing clinical procedures; patients with a history of continuous drug use; and patients who declined to participate in the study. Patient selection was based on the diagnostic criteria for acute cholecystitis. According to the Acute Cholecystitis Diagnostic Criteria,[4,17] patients showing one of the local signs of inflammation, such as Murphy's sign or a mass, pain, or tenderness in the right upper quadrant, along with one of the systemic signs of inflammation, such as fever, elevated white blood cell count, and elevated C-reactive protein level, were diagnosed with acute cholecystitis. Patients in whom suspected clinical findings were confirmed by diagnostic imaging were also diagnosed with acute cholecystitis.

Based on the physical examination, patients admitted to the emergency department suspected of having acute cholecystitis underwent routine diagnostic procedures, including a full blood count, CRP evaluation, and medical imaging. Subsequently, serum levels of IMA, TAS, TOS, OSI, and presepsin were analyzed using the spectrophotometric method. The study received ethics committee approval from the KSU Faculty of Medicine's Non-Interventional Clinical Research Ethics Committee, with the resolution number 02 in session 2022/07 on 15. 02. 2022.

Statistical Analysis

The Statistical Package for the Social Sciences (SPSS) version 22.0 (SPSS Inc., Chicago, Illinois, USA) was utilized for the statistical analysis of the data obtained from the study. Descriptive statistics for qualitative data were reported as frequency and percentage, and for quantitative data as frequency, mean, and standard deviation (SD). For comparing quantitative data with a known normal distribution, a T-test on a sample was applied if parametric assumptions were met. If parametric assumptions could not be met, a Wilcoxon signed-rank test was used. The t-test was employed for comparing quantitative data when parametric assumptions were met, and the Mann-Whitney U test was used when they were not. The chi-square (χ2) test was applied for comparing qualitative data. The diagnostic decision-making capabilities of biomarkers in predicting the diagnosis of acute cholecystitis were assessed by analyzing the receiver operating characteristics (ROC) curve. The type 1 error level for the study was set at 5%, and estimates made at a 95% confidence interval. Cases where the p-value was less than 0.05 were considered statistically significant.

RESULTS

Table 1 presents the means and standard deviations of WBC, platelet count, NLR, PLR, DNI, CRP, PCT, albumin, IMA, IMA/albumin, TAS, TOS, OSI, and presepsin values in the acute cholecystitis and control groups.

Table 2 displays the cutoff, sensitivity, specificity, positive likelihood ratio (LR), negative LR, area under the curve (AUC),

Table 1. Comparison of WBC, platelet count, NLR, PLR, DNI, CRP, PCT, albumin, IMA, IMA/albumin, TAS, TOS, OSI, and presepsin values in acute cholecystitis and control groups

	Acute Cholecystitis (n=53) Mean±SD	Control (n=48) Mean±SD	p
WBC (10^3/μL)	11.2±4.7	7.4±1.6	<0.001[a]
Platelet (10^3/μL)	240.7±90.0	267.3±61.3	0.083[b]
NLR	8.1±8.1	1.8±0.6	<0.001[a]
PLR	192.5±129.0	116.4±38.2	<0.001[a]
DNI	71.1±60.6	17.9±10.1	<0.001[a]
CRP (mg/dL)*	74.9±108.4	4.0±0.0	<0.001[c]
PCT (μg/L)*	3.0±6.4	0.046±0.0	0.001[c]
Albumin (g/dL)*	3.6±0.6	4.0±0.0	<0.001[d]
IMA (AbsU)	0.37±0.08	0.26±0.05	<0.001[b]
IMA/Albumin	0.10±0.03	0.06±0.01	<0.001[a]
TAS (mmol/L)	0.92±0.31	0.67±0.20	<0.001[a]
TOS (mmol/L)	8.9±4.5	5.7±2.4	<0.001[a]
OSI	1.3±1.3	1.0±0.6	0.640[a]
Presepsin (pg/mL)	102.6±32.2	187.3±153.2	<0.001[a]

CRP: C-Reactive Protein; DNI: Delta Neutrophil Index; IMA: Ischemia-Modified Albumin; NLR: Neutrophil-to-Lymphocyte Ratio; OSI: Oxidative Stress Index; PCT: Procalcitonin; PLR: Platelet-to-Lymphocyte Ratio; SD: Standard Deviation; TAS: Total Antioxidant Status; TOS: Total Oxidant Status; WBC: White Blood Cell Count. *The relevant variables were not studied in the control group and were compared to normal values in the literature (Albumin: 3.97-4.94, Procalcitonin: <0.046, CRP: <4.0). a Mann-Whitney U test; b T-test; c Wilcoxon Signed Rank Test; d One-sample t-test.

95% confidence interval, and p-values for the predictive power of the WBC, NLR, PLR, DNI, CRP, PCT, albumin, IMA, IMA/albumin, TAS, TOS, and presepsin parameters in predicting AC. It was observed that the parameters with positive LR results were NLR, DNI, CRP, PCT, albumin, IMA/albumin, and IMA (positive LR respectively: 9.8, 5.0, undefined, undefined, undefined, 6.7, 6.3). The parameters with negative LR results were NLR, DNI, CRP, PCT, IMA/albumin, IMA, and presepsin (negative LR respectively: 0.2, 0.2, 0.2, 0.1, 0.2, 0.2, 0.2).

ROC analysis was conducted for inflammatory markers. The results of the ROC analysis for inflammatory markers (WBC, NLR, PLR, DNI, CRP, PCT, albumin, IMA, IMA/albumin, TAS, TOS and presepsin) are illustrated in Figure 1.

Table 2. Cut-off, positive LR, negative LR, AUC, 95% confidence interval, and p-values for the predictive power of WBC, NLR, PLR, DNI, CRP, PCT, albumin, IMA, IMA/albumin, TAS, TOS, OSI, and presepsin parameters in the prediction of AC

	Cut-off	Sensitivity	Specificity	Positive LR	Negative LR	AUC	95% CI	pa
WBC (10^3/μL)	7.5	77.4	66.7	2.3	0.4	0.779	68.7-87.2	<0.001
NLR	2.5	81.1	91.7	9.8	0.2	0.901	83.8-96.4	<0.001
PLR	107	84.9	54.2	1.8	0.3	0.725	62.7-82.4	<0.001
DNI	25	83.0	83.3	5.0	0.2	0.884	82.0-94.9	<0.001
CRP (mg/dL)	4	78.4	100.0	b	0.2	0.804	69.7-91.1	<0.001
PCT (μg/L)	0.053	93.8	100.0	b	0.1	0.938	81.9-100.0	<0.001
Albumin (g/dL)	3.985	68.2	100.0	b	0.3	0.682	54.4-81.9	0.003
IMA/Albumin	0.077	84.1	87.5	6.7	0.2	0.888	81.3-96.3	<0.001
IMA (AbsU)	0.31	79.2	87.5	6.3	0.2	0.892	83.1-95.3	<0.001
TAS (mmol/L)	0.83	67.9	77.1	3.0	0.4	0.762	66.8-85.7	<0.001
TOS (mmol/L)	6.76	72.2	76.1	3.0	0.4	0.754	64.8-86.1	<0.001
Presepsin	134.5	88.7	75.0	3.6	0.2	0.859	78.8-93.0	<0.001

AUC: Area Under the Curve; CI: Confidence Interval; LR: Likelihood Ratio. a Values in groups were calculated using the ROC curve. b It was found to be undefined due to a specificity of 100%.
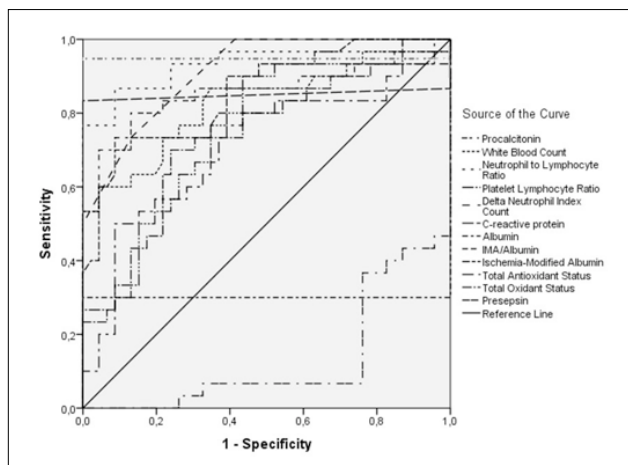
Figure 1. ROC curves for inflammatory markers.

DISCUSSION

Acute cholecystitis is one of the most common causes of acute abdominal pain. The diagnosis of acute cholecystitis should always be based on a comprehensive evaluation that includes patient history, physical examination findings, laboratory test results, and, when necessary, diagnostic imaging. [1,2,3,4] Ultrasound and computed tomography are commonly used to diagnose AC and identify its complications. However, the limited availability of ultrasound and computed tomography in some hospitals and the subjective nature of radiological interpretations can complicate the diagnosis process. As a result, certain serological markers are often utilized to assist in diagnosing AC. Biochemical markers such as white blood cell count, C-reactive protein, procalcitonin, neutrophil-to-lymphocyte ratio, and platelet-to-lymphocyte ratio have been identified as potential predictors of AC. Given the roles of inflammation, oxidative stress, and ischemia in the disease's pathophysiology, research into the diagnostic value of markers related to these processes is crucial. [1,2,3,4]

In the study conducted by Korkut et al., [17] the delta neutrophil index and delta neutrophil count were found to be significantly higher in patients presenting to the emergency department and diagnosed with AC compared to those not diagnosed with AC. The findings included a WBC count of 12.84±5.42×10³/mm3, an NLR of 8.81±1.11, a PLR of 205.97±163.05, a DNI of 0.5% (0.32), a delta neutrophil count of 60, and a CRP level of 47 mg/dL.

In this study, consistent with the literature, acute phase reactants and inflammatory markers such as WBC (11.2×10³/mm3±4.7), NLR (8.1±8.1), PLR (192.5±129.0), DNI (71.1±60.6), CRP (74.9 mg/dL±108.4), and PCT (3.0 µg/L±6.4) were elevated.

This research determined the cutoff values, sensitivity, specificity, positive LR, negative LR, and hemogram values of inflammatory markers in patients diagnosed with AC, presenting these findings in the results section.

The levels of leukocytes, NLR, PLR, DNI, CRP, and PCT at the time of diagnosis of acute cholecystitis were significantly higher in patients with cholecystitis compared to the control group. The authors believe that due to their high sensitivity, specificity, and low negative LR, NLR and DNI could serve as novel markers for diagnosing acute cholecystitis. Leukocytes, delta neutrophil index, NLR, and PLR are readily accessible blood parameters automatically tested as part of the complete blood count, thus, not incurring additional costs to the healthcare system. Therefore, these parameters could be deemed the most suitable for diagnosing AC.

In the study conducted by Gül et al., it was demonstrated that levels of leukocytes, CRP, and IMA were elevated in patients with acute cholecystitis compared to the control group. Moreover, the authors identified that with a cutoff value of IMA at 84 ng/mL, the sensitivity was 76%, and the specificity was 40% (AUC: 0.665, p=0.017, 95% confidence interval). The findings by Gül et al. concluded that ischemia-modified albumin could be beneficial in diagnosing acute cholecystitis, hence its potential utility for early diagnosis and treatment by clinicians. [13]

In this study, the cutoff value of the IMA/albumin ratio was determined to be 0.077, with a sensitivity of 84.1, specificity of 87.5, positive LR of 6.7, negative LR of 0.2, AUC of 0.888, and a 95% confidence interval (CI) of 81.3-96.3; p<0.001. It is suggested that due to its high sensitivity and specificity, the IMA/albumin ratio could serve as a parameter for diagnosing acute cholecystitis.

The cutoff value of IMA ((in Absorbance Units, AbsU) was set at 0.31, with a sensitivity of 79.2, specificity of 87.5, positive LR of 6.3, negative LR of 0.2, AUC of 0.892, and 95% CI of 83.1-95.3; p<0.001. It is proposed that due to its high sensitivity and specificity, IMA values could be a useful marker for the early diagnosis and treatment of acute cholecystitis.

In the studies conducted by Bösch et al. and Mugazov et al., presepsin was identified as a new determinant of mortality in patients undergoing surgery for intra-abdominal infections, acute surgical diseases of the abdominal cavity, and sepsis. These studies demonstrated that presepsin was a sensitive and specific predictor for the development of sepsis in abdominal organs, leading the authors to conclude that presepsin could be utilized as a diagnostic marker. [18,19]

In this study, the cutoff value for presepsin in patients with acute cholecystitis was found to be 134.5, with a sensitivity of 88.7, specificity of 75.0, positive LR of 3.6, negative LR of 0.2, AUC of 0.859, and a 95% CI of 78.8.1-93.0; p<0.001. It is contended that due to its high sensitivity and low negative LR, presepsin could emerge as a new marker for the early diagnosis and treatment of acute cholecystitis.

Oxidative stress responses, marked by a balance between pro-oxidants and antioxidants, are crucial aspects of the body's response to surgical stress and the activation of inflammatory

cells. The state of inflammation observed in acute cholecystitis amplifies oxidative stress. In the study conducted by Arslan Onuk et al., values of oxidative stress markers, including TAS, TOS, and OSI, were found to be elevated in patients with acute cholecystitis compared to those in the control group.[20] Similarly, in our study, oxidative stress markers (TAS, TOS, OSI) were found to be higher in patients with acute cholecystitis than in the control group. The data examining the relationship between TAS/TOS/OSI and acute cholecystitis presented in this study is rare in the literature, providing detailed insights into this relationship. In this study specifically, the TAS cutoff value was determined to be 0.83 with a sensitivity of 67.9, specificity of 77.1, positive LR of 3, negative LR of 0.4, AUC of 0.762, and a 95% CI of 66.8-85.7 (p<0.001) in patients with AC. The TOS cutoff value was 6.76 with a sensitivity of 72.2, specificity of 76.1, positive LR of 3, negative LR of 0.4, AUC of 0.754, and a 95% CI of 64.8-86.1 (p<0.001).

CONCLUSION

This study found elevated levels of inflammatory markers in patients diagnosed with acute cholecystitis: WBC at 11.2×10^3/mm3 (±4.7), NLR at 8.1 (±8.1), PLR at 192.5 (±129.0), DNI at 71.1 (±60.6), CRP at 74.9 mg/dL (±108.4), and PCT at 3.0 µg/L (±6.4). In patients diagnosed with acute cholecystitis, the mean value of IMA (AbsU) was found to be 0.37±0.08, the mean IMA/albumin ratio was 0.10±0.03, and the mean presepsin level (pg/mL) was 102.6±32.2. The levels of leukocytes, NLR, PLR, DNI, CRP, PCT, IMA, IMA/albumin, presepsin, and the TAS/TOS ratio were significantly higher in patients with cholecystitis compared to the control group. White blood cell count, neutrophil-to-lymphocyte ratio, platelet-to-lymphocyte ratio, and delta neutrophil index measurements can be tested as part of the complete blood count. The complete blood count parameters are readily available blood tests that do not incur additional costs to the healthcare system. The results obtained in this study will assist clinicians in diagnosing acute cholecystitis. The authors believe that due to their high sensitivity, specificity, and low negative likelihood ratios, NLR, PLR, DNI, IMA, IMA/albumin ratio, and presepsin levels could serve as new markers in the diagnosis of acute cholecystitis.

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ORİJİNAL ÇALIŞMA - ÖZ

Akut kolesistit tanısında iskemik modifiye albümin, presepsin, delta nötrofil indeksi ve inflamatuar belirteçlerin rolü

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AMAÇ: Erişkinlerde akut kolesistit tanısında C-reaktif protein, prokalsitonin, tam kan sayımı parametreleri, delta nötrofil indeksi, iskemik modifiye albümin, presepsin, oksidatif stres göstergeleri gibi inflamasyon, oksidatif stres ve iskemi ile ilişkili belirteçlerin patolojideki yerini belirlemektir.

GEREÇ VE YÖNTEM: Çalışmaya acil serviste akut kolesistit tanısı alan hastalar ve sağlıklı kontrol grubu dahil edildi. Çalışmaya alınan hastalara rutin kan sayımı ve biyokimya tetkikleri yapıldı. İskemik modifiye albümin, presepsin ve oksidatif stres göstergelerini ölçmek için kan serumu kullanıldı.

BULGULAR: Lökosit sayısı, nötrofil/lenfosit oranı, trombosit/lenfosit oranı, delta nötrofil indeksi, C-reaktif protein, prokalsitonin, iskemik modifiye albümin, iskemik modifiye albümin/albümin oranı, presepsin ve oksidatif stres göstergeleri değerleri akut kolesistit tanılı hastalar da kontrol grubu ile karşılaştırıldığında anlamlı olarak yüksek saptandı.

SONUÇ: Nötrofil/lenfosit oranı, delta nötrofil indeksi, iskemik modifiye albümin, iskemik modifiye albümin/albümin oranı ve presepsin değerlerinin yüksek duyarlılık, özgüllük ve düşük negatiflik olasılığı oranları nedeniyle akut kolesistit tanısında yeni belirteçler olarak kullanılabileceğine inanıyoruz.

Anahtar sözcükler: Akut kolesistit, İskemik modifiye albümin, Presepsin, Nötrofil/lenfosit oranı, Delta nötrofil indeksi