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Can immature granulocytes be used as a predictive new marker in the diagnosis of acute cholecystitis?

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Abstract

Aim: Acute cholecystitis (AC) is one of the most common acute surgical diseases in the emergency department (ED). The aim of this study was to investigate the efficacy of inflammatory parameters as immature granulocyte count (IGC) and immature granulocyte percentage (IG%) in the diagnosis of AC.

Material and methods: This retrospective and observational study consisted of patients, diagnosed with AC, who were admitted to a tertiary ED with abdominal pain between March 2019 and April 2021. The effectiveness of IGC and IG% in the diagnosis of AC was examined by comparing the results with the control group (CG).

Results: A total of 493 patients were included in the study. 270 patients were in the AC group, 223 patients were in the CG. IG% and IGC were found to be significantly higher in the AC group than in the CG (0.5 (0.32) vs. 0.4 (0.2); 0.06 (0.08) vs. 0.03 (0.03); p<0.001, p<0.001 respectively). It has been shown that IGC, at a cut-off value of 0.03, predicts the diagnosis of AC with 72.1% sensitivity and 55.5% specificity. On the other hand, IG%, at a cut-off value of 0.45, predicts the diagnosis of AC with 53.2% sensitivity and 72.7% specificity (AUC [0.717 (0.672-0.762); 0.692 (0.645-0.738)], respectively p<0.001, p<0.001).

Conclusions: In conclusion, IGC and IG% can be used as a useful inflammatory parameter in the diagnosis of AC in patients admitted to the emergency department.

Key words: immature granulocyte, acute cholecystitis, emergency department

Introduction

Abdominal pain is one of the most common complaints in the emergency department (ED). Many abdominal pain generally to have a benign etiology [1]. However a sign of local inflammation (Murphy sign or right upper quadrant pain, tenderness) should reveal the suspicion of acute cholecystitis.

Acute cholecystitis (AC) is a common complication of gall bladder stones and is one of the most common acute surgical diseases [2]. Although the general prevalence of gallstones varies between countries, it is estimated to be in 10%-15% of the general population. As a complication related to gallstones, calculosis AC occurs with an annual incidence of 1%-3% [3]. Cystic duct obstruction generally develops in these patients, due to gall bladder stones, which results with edema and inflammation in the gallbladder. The disease is an emergency surgical situation and mortality and morbidity increase in delayed surgical cases due to complications accompanied by gangrenous cholecystitis and gallbladder perforation [4]. Therefore, a detailed physical examination and history, laboratory tests, and extensive radiological imaging is needed in patients with suspected AC. One of the radiological imaging methods, such as USG/CT or Magnetic resonance imaging (MRI), is needed for definitive diagnosis in the case of AC clinical suspicion [1]. Clinical classification that includes radiological imaging findings of inflammation together with clinical markers is often useful, for the diagnosis of AC as suggested by The Tokyo Guidelines (TG 18) [5]. However, in cases where radiological imaging is not possible; laboratory tests for new inflammatory markers such as bedside Neutrophil-to-lymphocyte ratio (NLR) may be useful [6]. Immature granulocyte, one of these new inflammatory markers, is an indicator of increased myeloid cell production and it has been shown to increase in inflammatory conditions [7, 8]. In recent studies in the literature, it has been shown that immature granulocyte count (IGC) and immature granulocyte percentage (IG%)

are useful as inflammatory markers in many diseases such as acute pancreatitis accompanied by inflammation, acute appendicitis, acute gastrointestinal hemorrhage and intracerebral hemorrhage [9-12]. There is no study yet showing the predictivity of IGC and IG% in the diagnosis of AC. Therefore, the purpose of the study was to investigate the effectiveness of IGC and IG% as a diagnostic marker in AC patients.

Material and methods

This retrospective and observational study consisted of patients, diagnosed with acute cholecystitis (AC), who were admitted to a tertiary emergency department (ED) with abdominal pain between March 2019 and April 2021. The Control group (CG) had patients who were admitted to the ED with non-specific abdominal pain. CG was randomly determined and was similar in age and gender to AC patients. Approval with the date and decision number as 24/06/2021-9/16 was obtained from the local ethics committee.

Study design and participants

All patients who applied to the emergency department with abdominal pain during the study were retrospectively screened. Radiological imaging was performed using abdominal ultrasonography (USG) or abdominal computed tomography (CT) to patients with clinically suspected AC by an emergency specialist in the emergency department. Laboratory parameters were studied at the time of admission to the emergency department. The diagnosis of AC was made after consulting a general surgeon by following the criteria below: A. A sign of local inflammation (Murphy sign or right upper quadrant pain, tenderness) B. Signs of systemic inflammation (fever, elevated CRP, elevated leukocyte count) C. Imaging findings (characteristic USG or CT finding for AC). The diagnosis of AC was confirmed by the presence of at least one of the B or C criteria along with an A criterion [5]. All patients <18 years of age, pregnant patients, patients with hematological malignancies that may change hematological parameters, patients using granulocyte colony stimulating factor, immunosuppressive agents or steroids, patients with post-traumatic abdominal pain and patients with incomplete data were excluded from the study. The CG consisted of patients without a diagnosis of AC. Patients were categorized into two groups as AC and CG.

Data collection and measurements of variable

The records of the patients were scanned through the electronic data-based hospital information system. Complete blood count (CBC) analysis was performed with the Sysmex XN-1000 (Sysmex Corp., Kobe, Japan) device at the time of patients' admission to the emergency department. WBC, neutrophil, platelet, lymphocyte, IGC, IG%, CRP, and biochemistry [aspartate aminotransferase (AST), alanine transaminase (ALT), gamma glutamyl transferase (GGT), amylase, lipase, total bilirubin, direct bilirubin, glucose, blood urea nitrogen (BUN)] platelet distribution width (PDW) and Red cell distribution width (RDW) were recorded. Neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) were calculated from CBC results.

Outcomes

As a primary outcome, the predictivity of IGC and IG% in the diagnosis of AC was evaluated by comparing it with the control group.

Statistical analysis

All variables were evaluated using descriptive statistics. Standard deviation and mean values were calculated for continuous variables, and median and interquartile ranges were given. Each independent variable was compared using the chi-square test and the independent t test were used for continuous variables. Descriptive statistical analysis of all variables was made using the SPSS 20.0 program. Optimal cut-off value of IGC and IG % parameters as a diagnostic biomarker in the diagnosis of acute cholecystitis was analyzed by Receiver operating characteristic (ROC) analysis.

Results

A total of 493 patients were included in the study. 270 patients were in the AC group, 223 patients were in the CG. The mean age of the patients was 58.49±16.48 years in the AC group and 39.94±14.25 years in the CG. The mean age of the patients was significantly higher in the AC group ($p<0.001$). Among the male patients, 133 (49.3%) were in the AC group and 74 (33.2%) were in the CG. There was a significant difference between the groups in terms of gender ($p<0.001$). The mean WBC count was found to be significantly higher in the AC group than in the CG (12.84±5.42 vs. 9.41±3.05; $p<0.001$). The mean CRP level of the patients was found to be significantly higher in the AC group than in the CG (47 (139) vs. 4 (10); $p <0.001$).

Figure 1 - Comparision of immature granulocyte count (IGC) levels between healthy control and acute cholecystitis groups

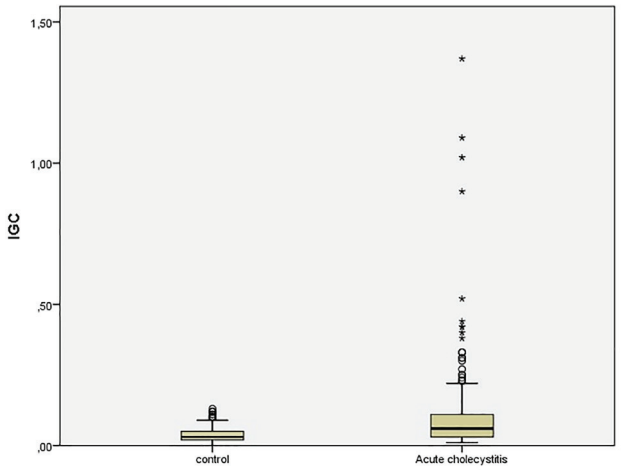


Figure 2 - Comparision of immature granulocyte percentage (IG%) levels between healthy control and acute cholecystitis groups

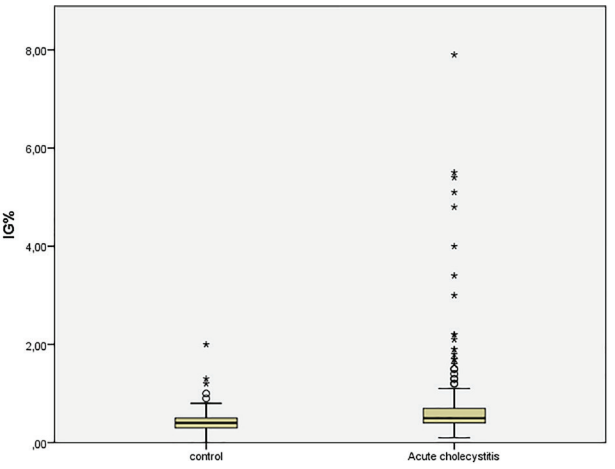


Table 1 Baseline of acute cholecystitis (AC) patient and healthy control groups

	Patient (n=270)	Control (n=223)	*P* value
Age (years)(mean±SD)	58.49±16.48	39.94±14.25	<0.001
Male, n (%)	133 (49.3)	74 (33.2)	<0.001
Laboratory tests(mean±SD)			
WBC count (×103/mm3)	12.84±5.42	9.41±3.05	<0.001
Hemoglobin (mg/dL)	13.02±2.01	13.22±2.06	0.221
PDW(fL; mean ± SD)	12.51±2.25	11.41±2.80	<0.001
RDW(fL; mean ± SD)	14.07±1.93	13.43±2.03	<0.001
PLT (×103/mm3)	261.96±86.57	274.89±77.57	0.59
Neutrophil, (×103/mm3)	9.97±5.26	6.20±3.04	<0.001
Lymphocyte, (×103/mm3)	1.73±0.98	2.31±1.01	<0.001
NLR	8.81±1.11	3.83±1.29	<0.001
PLR	205.97±163.05	150.64±121.97	<0.001
IG% (IQR)	0.5 (0.32)	0.4 (0.2)	<0.001
IG count (×103/mm3 (IQR)	0.06 (0.08)	0.03 (0.03)	<0.001
Glucose (mg/dl)	138.62±65.05	105.45±35.14	<0.001
BUN(mg/dL)	18.06±12.00	13.41±5.24	<0.001
Creatinine (mg/dl) (IQR)	1.09±0.89	0.88±0.26	0.31
CRP (mg/dL) (IQR)	47 (139)	4 (10)	<0.001
ALT(IU/L) (IQR)	31 (75)	18 (13)	<0.001
AST(IU/L) (IQR)	34 (84)	23 (9)	<0.001
GGT(IU/L) (IQR)	51 (173)	18 (13)	<0.001
Total bilirubin (mg/dl) (IQR)	0.94 (1.36)	0.45 (0.38)	<0.001
Direct bilirubin (mg/dl) (IQR)	0.20 (0.68)	0.08 (0.05)	<0.001
Lipase (IU/L) (IQR)	21 (27)	20 (18)	0.186

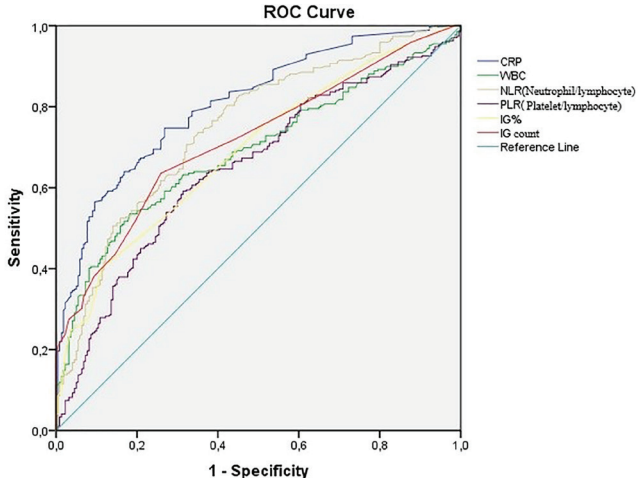
WBC: White blood cell; **CRP:** C-reactive protein; **IG:** Immature granulocytes **BUN:** Blood urea nitrogen
PLT: Platelet count **NLR:** Neutrophil-to-lymphocyte ratio; **PLR:** Platelet-to-lymphocyte ratio;
PDW: Platelet distribution width; **RDW:** Red cell distribution width
ALT: Alanine transaminase **AST:**Aspartate aminotransferase **GGT:** Gamma Glutamyltransferase

Table 2 Diagnostic predictors of acute cholecystitis

	AUC	95% CI	CUT-OFF	SEN (%)	SPE (%)	p
WBC	0.690	0.644-0.737	10.71	57.2	73.2	<0.001
CRP	0.808	0.770-0.845	10.25	71.7	74.1	<0.001
NLR	0.747	0.704-0.791	3.49	66.9	69.2	<0.001
PLR	0.650	0.602-0.699	160.59	53.9	72.7	<0.001
IG%	0.692	0.645-0.738	0.45	53.2	72.7	<0.001
IGC	0.717	0.672-0.762	0.03	72.1	55.5	<0.001

AUC: Area under the curve; **CI:** Confidence interval; **SEN:** Sensitivity; **SPE:** Specificity; **WBC:** White blood cell; **CRP:** C-reactive protein; **IGC:** Immature granulocytes count **IG:** Immature granulocytes **NLR:** Neutrophil-to-lymphocyte ratio; **PLR:** Platelet-to-lymphocyte ratio

Figure 3 - Receiver operating characteristic (ROC) curve of immature granulocyte and other inflammation parameters for predicting acute cholecystitis.



Besides, the mean IG% and IGC was found to be significantly higher in the AC group when compared with CG (0.5 (0.32) vs. 0.4 (0.2); 0.06 (0.08) vs. 0.03 (0.03); p <0.001, p <0.001 respectively) (Figure 1, Figure 2). The mean value of NLR and PLR were found to be significantly higher in the AC group than in the CG (8.81±1.11vs. 3.83±1.29; 205.97±163.05 vs. 150.64±121.97, respectively p<0.001). Table 1 shows the relationship between the baseline characteristics and other laboratory parameters of AC patients with the CG. ROC analysis was performed to show the predictive efficacy of IGC, IG%, WBC, CRP, NLR and PLR in the diagnosis of AC. Area under the curve (AUC) was calculated for each parameter and was found to be statistically significant in the diagnosis of AC (Figure 3). CRP had the highest AUC (0.808; p<0.001). AUC value was found to be statistically significant for NLR, IGC, IG%, WBC and PLR following CRP (AUC=0.747, 0.717, 0.692, 0.690 and 0.650). It has been shown that IGC, at a cut-off value of 0.03, predicts the diagnosis of AC with 72.1% sensitivity and 55.5%

specificity. On the other hand, IG%, at a cut-off value of 0.45, predicts the diagnosis of AC with 53.2 % sensitivity and 72.7% specificity (AUC: 0.717 (0.672-0.762); 0.692 (0.645-0.738), respectively p<0.001, p<0.001) (Table 2).

Discussion

The results of our study showed that IGC and IG% are useful in the diagnosis of AC compared to other inflammation parameters. The AC prediction ability of IGC and IG% was revealed with similar sensitivity and lower specificity compared to CRP, which has the highest sensitivity and specificity. Therefore, this study was found to be important in guiding the clinician in terms of accepting IGC and IG% as a new inflammatory parameter in the diagnosis of AC.

Recently, IG can be measured as an infection marker with an easy, cheap and fast method with new generation automatic hemogram devices [13]. IG cells are not physiologically found in the circulation. However, studies have shown that immature granulocyte is a useful marker in diseases accompanied by inflammation [14, 15] Since there is no feature strong enough to diagnose or exclude AC; evaluation of clinical or laboratory results alone is not recommended by current guidelines. Combination with detailed history, physical examination and laboratory tests and imaging is recommended [3]. USG imaging has been the first preferred diagnostic imaging method in many guidelines [16]. In fact, its diagnostic efficiency has been demonstrated with a high sonographic AC score in a prospective bedside USG validation study [17]. However, USG imaging is operator-dependent, and it can be difficult to diagnose AC in the emergency department because it requires patient compliance. Other laboratory tests are needed along with the clinic. Therefore, researchers have focused on new inflammatory parameters in the diagnosis of AC since there is no specific laboratory parameter. Studies have shown that NLR and PLR are as useful as CRP in the diagnosis of AC. In a study of Bedel C. [6], it was shown that NLR, with the highest sensitivity 94.4% and AUC (0.846; p<0.001), followed by PLR with 77.9% sensitivity and AUC (0.768; p<0.001) are diagnostically effective. Similar to Bedel's study, NLR and PLR were found to be statistically significant in the diagnosis of AC in present study. In another study on the role of NLR in the differentiation of AC and complicated AC; NLR was found as a diagnostic parameter with 80.9% positive predictive value and AUC (0.736; p<0.001) [18]. However, when we searched the literature, there is no study on IG in the diagnosis of AC yet. In many studies, the prognostic and diagnostic relationship of IG in diseases accompanied by inflammatory processes has been examined. Recent studies have also demonstrated the prognostic value of IG. In a study of Barut et al. [19], in which the prognostic value of IG was evaluated in gall bladder cancer, IGC was found as significant with 83.6% sensitivity and 84.2% specificity at 0.08 cut-off value and AUC: 0.910 [95% CI 0.858-0.962]. In a study of Unal et al. [11], it was determined that IG could distinguish complicated appendicitis from simple appendicitis at a cut-off value of 0.1. In another study of Güngör et al. [13], IG has been shown to be effective in differentiating acute complicated appendicitis from simple appendicitis, with a sensitivity of 85.4% at a cut-off value of 0.35% and AUC: 0.82 [95% confidence interval (CI) 0.77–0.87]. In present study, IGC and IG% was found to be effective in the diagnosis of AC at cut-off value of 0.03 and 0.45, respectively.

The present study has some limitations. First, this study was designed in a retrospective nature at a single center with a small number of patients. The second important limitation is that IG was measured only during admission. Serial measurements were not made at certain time intervals starting from the onset of symptoms until the diagnosis. Our recommendation to the researchers is to carry out further studies in a prospective manner with wide-ranging population at multi centers.

Conclusion

In conclusion, IGC and IG% can be used as a useful inflammatory parameter in the diagnosis of AC in patients admitted to the emergency department.

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