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Platelet-to-lymphocyte ratio and neutrophil-to-lymphocyte ratio as predictors of refractory anaphylaxis

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ABSTRACT

Background: Refractory anaphylaxis poses an ongoing, lethal hypersensitivity response that unpredictably involves multiple organs despite appropriate intramuscular (IM) adrenaline injections. Studies on the association of the platelet-to-lymphocyte ratio (PLR) and neutrophil-to-lymphocyte ratio (NLR) concerning anaphylactic severity have yet to be carried out. The study aimed to evaluate the association between blood PLR and NLR levels and refractory anaphylaxis.

Methods: We carried out a retrospective cross-sectional study in which medical records of patients with anaphylaxis who sought urgent care at the Emergency Department (ED) of Tertiary Hospital in Hanoi, Vietnam, were evaluated. Based on the United Kingdom Resuscitation Council guidelines in 2021, patients were classified as refractory anaphylaxis if they needed more than two appropriate doses of intramuscular adrenaline for anaphylactic symptoms resolution. Clinical data and laboratory results were obtained in the medical records. Logistic regression analysis determined the association between contributing factors and refractory anaphylaxis.

Results: One-hundred eighteen adults (age 51.80 $\pm$ 18.25 years) were analyzed, including 38 refractory anaphylaxis patients (32.2%). Refractory anaphylaxis patients exhibited notably elevated platelet-to-lymphocyte ratio (PLR) ($P = 0.006$) and increased neutrophil-to-lymphocyte ratio (NLR) ($P < 0.001$) in comparison to non-refractory anaphylaxis patients. Receiver operating characteristic curve (ROC) analysis demonstrated an optimal PLR cutoff value of 129.5 (area under the ROC curve [AUC] 0.658, sensitivity 73.68%, specificity 61.25%, $P = 0.004$) and an optimal NLR cutoff value of 4 (AUC 0.736, sensitivity 65.79%, specificity 73.75%, $P < 0.001$) for refractory anaphylaxis. Multivariate logistic regression analysis revealed a PLR$\geq$129.5 (OR = 4.83, 95% CI: 1.87–12.48) and an NLR$\geq$4 (OR = 4.60, 95% CI: 1.86–11.41) were independently associated with refractory anaphylaxis.

Conclusion: Elevated PLR and NLR serve as independent indicators significantly associated with refractory anaphylaxis.

Keywords: Anaphylaxis, Refractory anaphylaxis, Adrenaline, Platelet-to-lymphocyte ratio, Neutrophil-to-lymphocyte ratio

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INTRODUCTION

Anaphylaxis stands as a significant, multifactorial, potentially fatal type I allergic response. The condition unpredictably manifests through systemic hypersensitivity reactions, exhibiting a spectrum of symptoms from mild such as urticaria, to severe such as airway obstruction and distributive shock. These responses occur immediately within minutes or hours following the allergen exposure, requiring prompt and appropriate treatment.[1] Previous research has documented a worldwide prevalence of anaphylaxis spanning from 50 to 112 episodes per 100,000 person-years.[2] The mortality rates were around 0.5 to 1 fatality per million.[3] The mainstay of anaphylaxis management is intramuscular (IM) adrenaline injection,[4] and most patients responded well after 1 or 2 IM adrenaline doses.[3] However, some patients, unfortunately, suffered persistent anaphylactic symptoms despite administering 2 appropriate IM adrenaline injections, facing a higher risk of mortality condition, called refractory anaphylaxis.[4]

Refractory anaphylaxis, according to the United Kingdom Resuscitation Council (UKRC) guidelines in 2021, is characterized as a persistent anaphylactic state necessitating further intervention because of continuous respiratory or cardiovascular symptoms, even after the administration of two appropriate doses of IM adrenaline. The condition demands immediate treatment modalities for life-threatening conditions such as respiratory failure or shock.[4] Currently, the epidemiology of refractory anaphylaxis lacks clarity due to the need for additional data. Given the unpredictable progression of anaphylaxis, early prediction and clinical assessment may be paramount to recognize these adrenaline-refractory cases.

However, no tests are currently available to predict the probability of refractory anaphylaxis.[5] Contrastly, other allergic pathologies such as asthma,[6] allergic rhinitis,[7] and nonsteroidal anti-inflammatory drugs hypersensitivity[8] have reported cost-effective indices, including platelet-to-lymphocyte ratio (PLR) and neutrophil-to-lymphocyte ratio (NLR), acquired from the complete blood count (CBC) results, to have the association with the severity of the reaction. Hence, our study aimed to evaluate the association between blood PLR and NLR levels and refractory anaphylaxis.

METHODS

This retrospective cross-sectional study recruited anaphylaxis patients visiting Emergency Department (ED) of Tertiary Hospital in Hanoi, Vietnam, between October 2017 and October 2023. We collected the patient characteristics data via electronic medical records and maintained confidentiality. The inclusion criteria comprised adult patients over 18 years old who were diagnosed with anaphylaxis based on the World Allergy Organization (WAO) amended diagnostic criteria[9] and graded following the generalized hypersensitivity reaction grading system suggested by Brown in 2004,[10] either arrived directly or were transferred from other hospitals within 24 h since the beginning of symptoms. We excluded the patients who suffered prehospital cardiac arrest because these patients were deemed to be at high risk of mortality, or who had been diagnosed with malignancies, myelosuppression conditions or autoimmune disorders that induce lymphopenia and/or lymphocytosis, or who got chemotherapy treatment within the last month before admision, or suffered liver cirrhosis with Child-Pugh B or C, or who had no laboratory data available.

Our research adhered to ethical principles stated in the Declaration of Helsinki.[11] Informed consent was not necessary because of a retrospective study.

Definition of anaphylaxis diagnosis criteria and grading system

The diagnosis criteria were followed by the World Allergy Organization (WAO) Anaphylaxis Committee amended diagnostic criteria.[9] Anaphylaxis was diagnosed when any of the 2 criteria was met. First, anaphylaxis manifestations happened acutely (within minutes to a few hours) involving cutaneous (systemic urticaria, pruritus, lip-tongue-uvula oedema), and either manifestation occurred from respiratory (dyspnea, wheeze-bronchospasm, stridor, hypoxemia) or cardiovascular (collapse, incontinence, chest discomfort, reduced blood pressure [decreased systolic blood pressure >30% from that patient's baseline or <90 mmHg]) or gastrointestinal system (crampy abdominal pain, repetitive vomiting, diarrhea).

Second, individuals had hypotension, wheeze-bronchospasm, or laryngeal involvement characterized by stridor, vocal changes, and odynophagia, which ensued within minutes to several hours after a confirmed or highly possible allergen exposure for each individual, even in the absence of cutaneous symptoms.

The grading system followed the simplified generalized hypersensitivity reactions suggested by Brown in 2004.[10] Anaphylaxis reaction severity was graded as follows. Mild reaction was characterized as limited cutaneous symptoms. Moderate reaction was defined as mild symptoms plus the presence of either respiratory, cardiovascular or gastrointestinal symptoms. Severe reaction was defined as anaphylaxis involving either cyanosis, oxygen saturation <92%, hypotension, or neurological symptoms (confusion, collapse, incontinence).

Definition of refractory anaphylaxis

According to UKRC guidelines, refractory anaphylaxis is characterized as a persistent anaphylactic state necessitating further intervention because of continuous respiratory or cardiovascular symptoms, even after the administration of two appropriate doses of IM adrenaline.[4]

Anaphylaxis management

Anaphylaxis patients were managed by strictly following the established guidelines in 2017 by the Vietnam Ministry of Health.[12] Besides taking blood samples for laboratory tests, intramuscular adrenaline was injected with a dose of 0.5 mg (half of the commonly used vial). Each repeat IM adrenaline injection was ordered if the patient's symptoms did not improve after 3 to 5 min since the prior injection, with a maximum of 3 doses. Suppose the symptoms continued after 2 IM adrenaline injections (refractory anaphylaxis); intravenous (IV) adrenaline or dopamine, dobutamine, and norepinephrine would be administered. Additional medications such as antihistamines, glucocorticosteroids, and intensively resuscitative therapies such as intubation, emergent tracheostomy, and IV fluid were taken as needed for respiratory and cardiovascular support. Admitting to the intensive care unit was considered for the patients with persisting anaphylactic symptoms, managed with IV vasopressor or intubated.

Data collection

Based on the UKRC definition,[4] the analyzed records were split into 2 groups: refractory and non-refractory anaphylaxis patients.

The patient's data were collected: sex, age, history of medical diseases, history of allergic diseases, present anaphylaxis elicitors; anaphylactic symptoms (skin and mucous, cardiovascular, respiratory, and gastrointestinal systems);[9] vital signs; number of IM adrenaline injections, use of IV vasopressors, antihistamines, and glucocorticosteroids; whether the patient was admitted and their admission length of stay.

Laboratory tests were obtained during ED arrival: complete blood count (CBC) and comprehensive metabolic panel (CMP), including glucose, blood urea, creatinine, AST, ALT, sodium, potassium, and chloride. The CBC and CMP were analyzed by using standard automatic systems.

PLR and NLR calculations

PLR and NLR were determined by dividing the absolute number of platelet or neutrophil to the lymphocyte count, with the same measured unit as thousand per cubic millimeter, which were all obtained from the CBC result, respectively.

Statistical analysis

Clinical and laboratory characteristics were observed in refractory and non-refractory groups. The results were presented as the absolute numbers and percentages for categorical variables, tested with Chi-square and Fisher's exact test. For continuous variables, normally distributed data were described by the means and standard deviation, tested with Student's t-test and ANOVA, while the median value and interquartile range demonstrated skewed distribution data, tested with Mann-Whitney U. Performing the receiver operating characteristic curve (ROC) analysis and calculating the area under the curve (AUC) to identify the predictors of refractory anaphylaxis. Identifying the refractory anaphylaxis risk factors by performing the multivariate logistic regression analysis to determine the potential predictors of refractory anaphylaxis, presented as odds ratio

(OR) and 95% confidence interval (CI), any variables demonstrating statistical significance with a *P*-value<0.05 in the univariate logistic regression analysis were selectively integrated into the multivariate logistic regression analysis.

All clinical and laboratory comparisons were calculated using Epi Info version 7.2.6.0 for Windows (Centers for Disease Control and Prevention, Georgia, USA). Box plots within the R package of "ggplot2", ROC curve, AUC, and 95% CI were determined by using the "roc", "auc" and "ci" functions within the R package of "pROC" in RStudio Desktop version 2023.09.1 + 494 (Posit, Boston, Massachusetts, USA). A $P < 0.05$ was considered statistically significant.

RESULTS

One hundred thirty patients were diagnosed with anaphylaxis admitted to the ED. Twelve patients were excluded (Fig. 1). One hundred eighteen patients diagnosed with anaphylaxis with a mean age of 51.80 ± 18.25 years were enrolled in our research. Of these patients, 57 (48.31%) were male, and 38 (32.2%) were classified as refractory anaphylaxis (Table 1). Notably, an association between gender and refractory anaphylaxis was observed in men, as male patients exhibited a significantly higher prevalence of refractory events compared to females (24/57 vs 14/61, respectively, $P = 0.027$). Between refractory and non-refractory patients, the history of allergic and medical diseases was insignificant ($P = 0.565$, $P = 0.135$; respectively). Compared to the non-refractory group, refractory anaphylaxis patients required significantly more IM adrenaline injections (3 doses vs 1 dose, $P < 0.001$). Moreover, 37 cases received continuous IV adrenaline, significantly higher than the non-refractory group (97.37% vs 0%, $P < 0.001$). Also, refractory patients stayed in the hospital significantly longer than non-refractory patients (3 days vs 2 days, $P < 0.001$).

White blood cell (WBC) count with differentials

Refractory anaphylaxis patients had significantly higher median white blood cell (WBC) counts than non-refractory patients ($P = 0.004$). Additionally, the absolute numbers of neutrophils and lymphocytes were also statistically significant, with a higher mean neutrophil count ($P < 0.001$) and lower median lymphocyte count ($P = 0.006$)

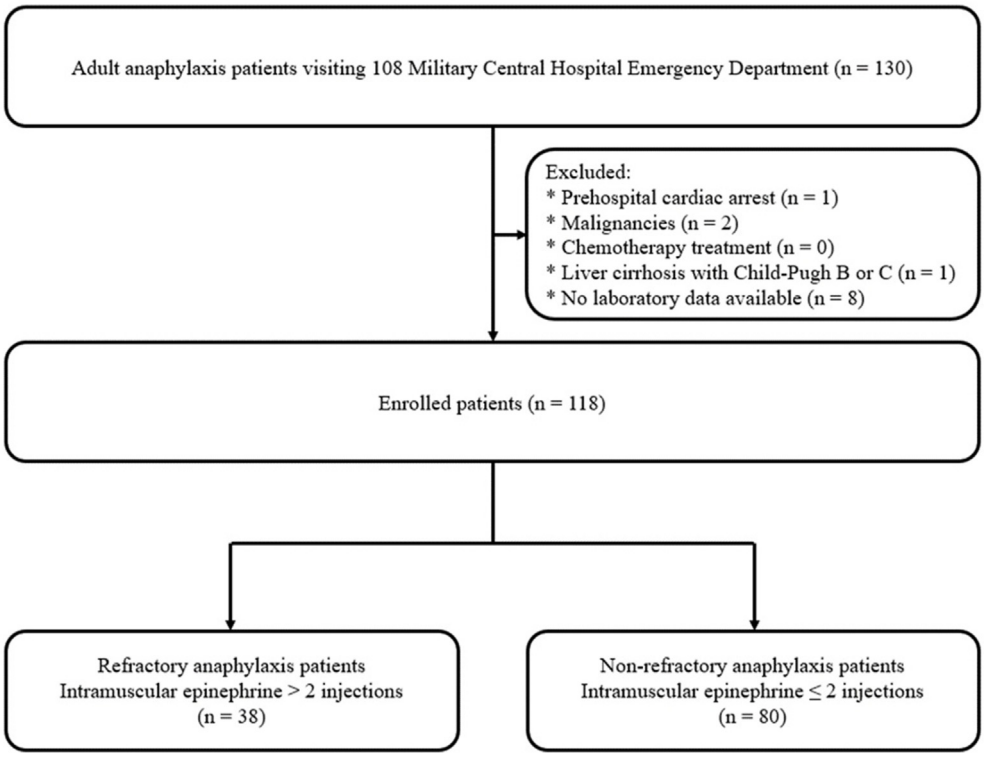

Fig. 1 Flow chart of the study design

	All (n = 118)	Refractory (n = 38)	Non-refractory (n = 80)	*P*-value
Male, n (%)	57 (48.31)	24 (63.16)	33 (41.25)	0.027
Age (years)	51.80 ± 18.25	55.53 ± 16.08	50.02 ± 19.03	0.106
History of allergic diseases				
Asthma, n (%)	6 (5.08)	1 (2.63)	5 (6.25)	0.662
Allergic rhinitis, n (%)	1 (0.80)	0 (0.00)	1 (1.25)	1.000
Food allergy, n (%)	16 (13.56)	5 (13.16)	11 (13.75)	1.000
Drug allergy, n (%)	25 (21.19)	7 (18.42)	18 (22.50)	0.791
History of medical diseases				
Hypertension, n (%)	30 (25.42)	12 (31.58)	18 (22.50)	0.405
Diabetes mellitus, n (%)	12 (10.17)	5 (13.16)	7 (8.75)	0.520
Cardiovascular disease, n (%)	8 (6.78)	1 (2.63)	7 (8.75)	0.434
Pulmonary disease, n (%)	4 (3.39)	2 (5.26)	2 (2.50)	0.593
Renal disease, n (%)	3 (2.54)	1 (2.63)	2 (2.50)	1.000
Elicitors				
Foods, n (%)	28 (23.73)	8 (21.05)	20 (25.00)	0.811
Medications, n (%)	62 (52.54)	15 (39.47)	47 (58.75)	0.078
Contrast media, n (%)	19 (16.10)	10 (26.31)	9 (11.25)	0.070
Insect venom, n (%)	4 (3.39)	2 (5.26)	2 (2.50)	0.593
Others, n (%)	3 (2.55)	1 (2.63)	2 (2.50)	1.000
Idiopathic, n (%)	2 (1.69)	2 (5.26)	0 (0.00)	0.102
Symptoms of anaphylaxis				
Cardiovascular, n (%)	78 (66.10)	33 (86.84)	45 (56.25)	0.002
Hypotension, n (%)	63 (53.39)	33 (86.84)	30 (37.50)	<0.001
Syncope, n (%)	20 (16.95)	11 (28.95)	9 (11.25)	0.033
Chest discomfort, n (%)	30 (25.42)	11 (28.95)	19 (23.75)	0.704
Respiratory, n (%)	91 (77.12)	26 (68.42)	65 (81.25)	0.188
Wheezing, n (%)	9 (7.63)	5 (13.16)	4 (5.00)	0.145
Dyspnea, n (%)	90 (76.27)	26 (68.42)	64 (80.00)	0.250
Gastrointestinal, n (%)	39 (33.05)	15 (39.47)	24 (30.00)	0.416
Abdominal pain, n (%)	21 (17.80)	8 (21.05)	13 (16.25)	0.704
Vomiting, n (%)	25 (21.19)	11 (28.95)	14 (17.50)	0.238
Diarrhea, n (%)	15 (12.71)	5 (13.16)	10 (12.50)	1.000
Skin, n (%)	88 (74.58)	21 (55.26)	67 (83.75)	0.002
Urticaria, n (%)	85 (72.03)	21 (55.26)	64 (80.00)	0.010
Angioedema, n (%)	17 (14.41)	2 (5.26)	15 (18.75)	0.095

(continued)

	All (n = 118)	Refractory (n = 38)	Non-refractory (n = 80)	*P*-value
Generalized swelling, n (%)	86 (72.88)	21 (55.26)	65 (81.25)	0.006
Pruritus, n (%)	84 (71.19)	19 (50.00)	65 (81.25)	0.001
Treatment				
Number of IM adrenaline injections	1 (1–3)	3 (3–3)	1 (0–1)	<0.001
Intravenous adrenaline use, n (%)	37 (31.36)	37 (97.37)	0 (0.00)	<0.001
H1 blocker, n (%)	89 (75.42)	24 (63.16)	65 (81.25)	0.057
H2 blocker, n (%)	3 (2.54)	3 (7.89)	0 (0.00)	0.032
Glucocorticosteroids, n (%)	105 (88.98)	33 (86.84)	72 (90.00)	0.754
Disposition				
Hospital admission, n (%)	86 (72.88)	37 (97.37)	49 (61.25)	<0.001
Hospital length of stay (days)	3 (2–4)	3 (2–6)	2 (0–3)	<0.001

Table 1. (Continued) Clinical characteristics: refractory vs. non-refractory anaphylaxis patients. *The results are presented as follows: categorical variables (numbers and percentages), continuous variables: normally distribution (means and standard deviation), non-normally distribution (medians and interquartile range).*

among refractory anaphylaxis patients, compared to non-refractory group (Table 2).

Platelet-to-lymphocyte ratio

Between refractory and non-refractory groups, refractory anaphylaxis patient's median PLR was significantly higher (181.74 vs 110.63; $P = 0.006$) (Table 2), (Fig. 3). The ROC curve analysis demonstrated an ideal PLR cutoff value of 129.5 for predicting refractory anaphylaxis (AUC 0.658, sensitivity 73.68%, specificity 61.25%, $P = 0.004$), (Fig. 4).

Neutrophil-to-lymphocyte ratio

Between refractory and non-refractory groups, refractory anaphylaxis patient's median NLR was significantly higher (6.41 vs 2.20; $P < 0.001$) (Table 2), (Fig. 2). The ROC curve analysis demonstrated an ideal NLR cutoff value of 4 for predicting refractory anaphylaxis (AUC 0.736, sensitivity 65.79%, specificity 73.75%, $P < 0.001$) (Fig. 4).

Independent predictors of refractory anaphylaxis

Results of univariate logistic regression indicated that male, exposure to contrast media, blood glucose level, PLR≥129.5, and NLR≥4 were all significantly associated with refractory anaphylaxis. The subsequent multivariate logistic regression revealed contrast media exposure (Model 1: OR = 4.68, 95% CI: 1.48–14.73; $P = 0.008$; Model 2: OR = 4.88, 95% CI: 1.55–15.37; $P = 0.007$), blood glucose level (Model 1: OR = 1.14, 95% CI: 1.02–1.27; $P = 0.017$; Model 2: OR = 1.17, 95% CI: 1.05–1.31; $P = 0.005$), PLR≥129.5 (OR = 4.83, 95% CI: 1.87–12.48; $P = 0.001$) and NLR≥4 (OR = 4.60, 95% CI: 1.86–11.41; $P = 0.001$) emerged as significant potential factors for predicting refractory anaphylaxis (Table 3).

DISCUSSION

Current research on the association between PLR, NLR and refractory anaphylaxis has been seldom documented.[5] This is one of the first studies about the association between PLR, NLR, and refractory anaphylaxis. In our research, we discerned that a PLR≥129.5 and an NLR≥4 stood independently associated with refractory anaphylaxis and may be used to predict refractory anaphylaxis in patients, and refractory anaphylaxis prevalence was 32.2%. Therefore, our findings provide novel insights and merit further elaboration.

Refractory anaphylaxis: Prevalence

In our study, the prevalence of refractory anaphylaxis exhibited a notably higher frequency compared to findings reported in existing literature. A long-term study in the United States by Korenblat showed that 16.2% of anaphylaxis

	All (n = 118)	Refractory (n = 38)	Non-refractory (n = 80)	*P*-value
WBC (10^9/L)	11.5 (7.8–6.2)	14.2 (9.7–23.1)	10.0 (9.5–14.9)	0.004
Neutrophils (%)	69.70 (53.20–83.60)	80.40 (67.00–87.70)	61.75 (45.75–75.95)	<0.001
Lymphocytes (%)	22.30 (9.80–38.70)	12.25 (7.00–23.90)	28.50 (14.30–44.60)	<0.001
Neutrophils (10^9/L)	7.37 (4.35–13.08)	10.63 (6.43–19.28)	5.76 (3.68–10.98)	<0.001
Lymphocytes (10^9/L)	2.15 (1.27–3.62)	1.44 (0.91–2.43)	2.41 (1.78–3.71)	0.006
RBC (10^{12}/L)	4.58 (4.20–4.98)	4.49 (4.17–4.90)	4.61 (4.25–5.09)	0.258
Hemoglobin (g/dL)	13.50 (12.80–15.30)	13.40 (12.60–14.60)	13.50 (12.80–15.40)	0.436
Hematocrit (%)	41.30 (38.40–45.20)	41.20 (38.60–45.20)	41.30 (38.30–45.70)	0.750
MCV (fL)	91.00 (87.80–93.40)	91.80 (89.50–95.30)	90.80 (87.10–92.50)	0.120
MCH (pg)	30.10 (29.10–31.30)	30.50 (29.10–31.30)	29.90 (21.10–31.40)	0.739
Platelet (10^9/L)	279 (226–331)	259 (206–331)	282 (239–333)	0.247
NLR	3.09 (1.40–8.62)	6.41 (2.82–12.42)	2.20 (1.03–5.31)	<0.001
ELR	0.05 (0.03–0.10)	0.05 (0.03–0.10)	0.04 (0.02–0.09)	0.479
PLR	133.42 (81.23–223.53)	181.74 (100.94–291.89)	110.63 (74.45–190.84)	0.006
Glucose (mmol/L)	7.76 (6.09–10.79)	9.16 (6.94–12.37)	7.56 (6.03–9.56)	0.028
Urea (mmol/L)	5.57 (4.61–6.64)	5.94 (5.00–7.37)	5.34 (4.56–6.33)	0.066
Creatinine (umol/L)	81 (64–100)	92 (75–113)	74 (62–96)	0.004
AST (U/L)	26.40 (20.10–36.40)	29.40 (21.70–43.40)	24.75 (19.65–34.60)	0.196
ALT (U/L)	22.30 (14.80–35.00)	25.00 (17.40–41.00)	20.40 (13.40–33.40)	0.211
Sodium (mmol/L)	137 (135–139)	137 (135–140)	137 (135–139)	0.899
Potassium (mmol/L)	3.40 (3.10–3.80)	3.30 (3.00–4.00)	3.45 (3.20–3.80)	0.223

(continued)

	All (n = 118)	Refractory (n = 38)	Non-refractory (n = 80)	*P*-value
Chloride (mmol/L)	105 (103–106)	104 (102–106)	105 (102–106)	0.589

Table 2. (Continued) Laboratory results: refractory vs. non-refractory anaphylaxis patients. *The results are presented as follows: categorical variables (numbers and percentages), continuous variables: normally distribution (means and standard deviation), non-normally distribution (medians and interquartile range). Abbreviations: NLR, Neutrophil-to-lymphocyte ratio; PLR, Platelet-to-lymphocyte ratio; ELR, Eosinophil-to-lymphocyte ratio; MCH, Mean corpuscular hemoglobin; MCV, Mean corpuscular volume; RBC, Red blood cell; WBC, White blood cell.*

patients required more than 3 IM adrenaline injections.[13] In Korea, a cross-sectional study conducted by Kim et al revealed the prevalence of refractory anaphylaxis was around 25%.[5] The variation in refractory anaphylaxis prevalence may result from pre-hospital treatment factors. Because adrenaline autoinjector is not available in Vietnam, every anaphylaxis patient received the IM adrenaline doses only after arriving at the ED. Since the condition may worsen unpredictably, severe and fatal anaphylaxis outcomes in some cases have been associated with administering adrenaline postponement.[1] Such instances may have contributed to the increased occurrence of refractory anaphylactic events observed in our study compared to prior research.

Refractory anaphylaxis: Platelet-to-lymphocyte ratio

Our study exhibited a noteworthy finding: the PLR ≥129.5 was associated with refractory

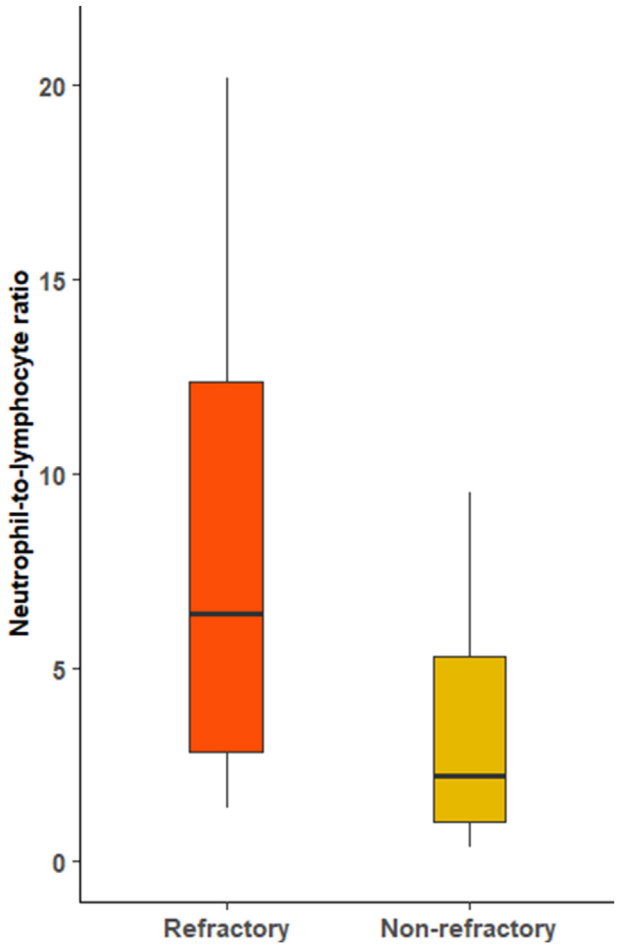

Fig. 2 Neutrophil-to-lymphocyte ratio in patients with refractory and non-refractory anaphylaxis, displayed in box plot

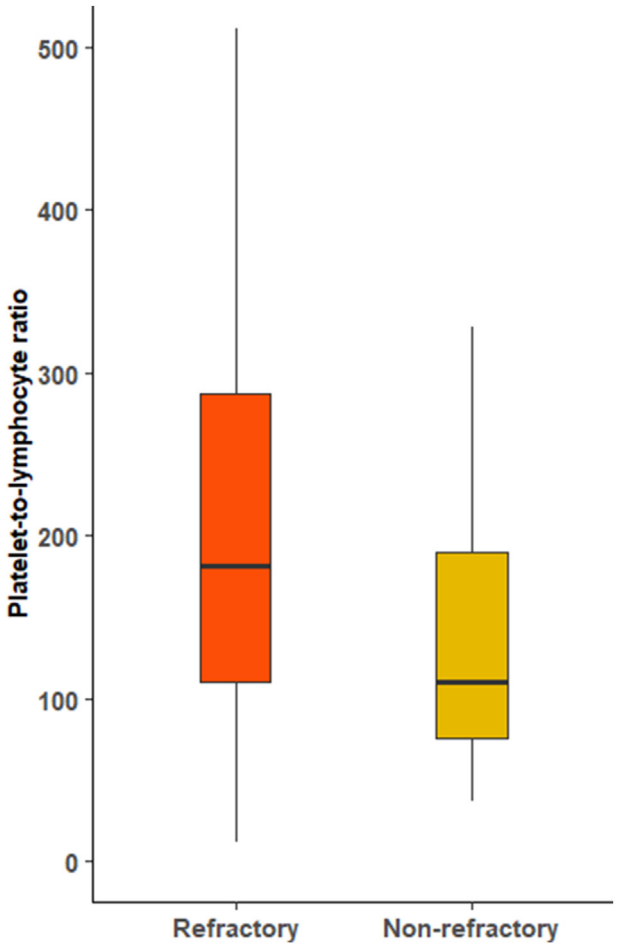

Fig. 3 Platelet-to-lymphocyte ratio in patients with refractory and non-refractory anaphylaxis, displayed in box plot

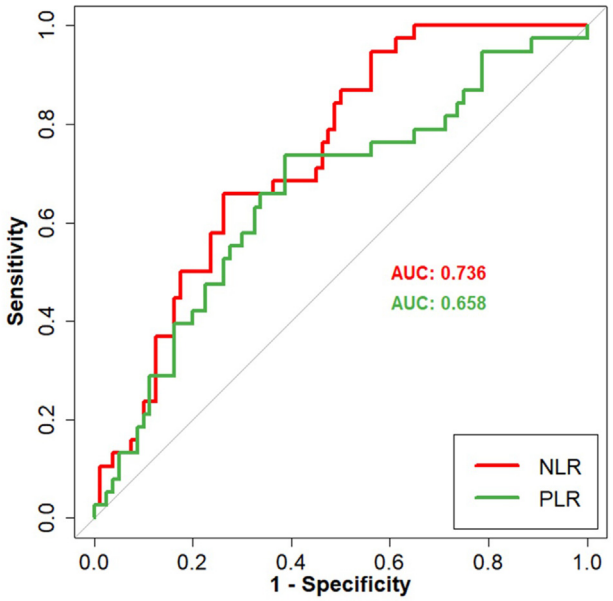

Fig. 4 ROC curve analysis of neutrophil-to-lymphocyte ratio and platelet-to-lymphocyte ratio to predict refractory anaphylaxis

anaphylaxis. This is a new finding since there has been no PLR research on anaphylaxis, especially refractory anaphylaxis. To explain the increase of PLR in our study, which can be a discrepancy between our result and the pathophysiology as platelets are found to aggregate in anaphylaxis, resulting in lower platelet levels,[14] it may be attributed to the relatively decreased level of lymphocyte. In our research, the difference of median platelet count of the refractory and non-refractory anaphylaxis groups was not statistically significant but did show the absolute median platelet in the refractory patients was lower than the non-refractory group, which was consistent to the pathophysiology of platelet aggregation. Besides, the level of lymphocyte in refractory anaphylaxis patients was statistically lower than the non-refractory group, which might eventually lead to the increase in PLR. The lymphopenia pathophysiology in anaphylaxis remains unknown, but it was considered to occur from the lymphocyte apoptosis process during the reaction.[15,16]

Prior research has investigated the activation of platelet aggregation in the context of anaphylaxis severity. Remarkably, platelet-activating factor (PAF) has been established as a noteworthy

Dependent variables	Univariate		Multivariate			
			Model 1		Model 2	
	Odds Ratio (95% CI)	*P*-value	Odds Ratio (95% CI)	*P*-value	Odds Ratio (95% CI)	*P*-value
Age	1.02 (1.00–1.04)	0.127				
Male	2.44 (1.10–5.41)	0.028	1.99 (0.81–4.91)	0.133	2.20 (0.90–5.38)	0.086
Elicitor: Food	0.80 (0.32–2.03)	0.638				
Elicitor: Medications	0.46 (0.21–1.01)	0.052				
Elicitor: Contrast media	2.82 (1.04–7.66)	0.043	4.68 (1.48–14.73)	0.008	4.88 (1.55–15.37)	0.007
Glucose	1.13 (1.03–1.23)	0.009	1.14 (1.02–1.27)	0.017	1.17 (1.05–1.31)	0.005
NLR ≥4 vs. <4	5.07 (2.21–11.63)	<0.001	4.60 (1.86–11.41)	0.001		
PLR ≥129.5 vs. <129.5	4.20 (1.80–9.82)	<0.001			4.83 (1.87–12.48)	0.001

Table 3. Independent predictors of refractory anaphylaxis among 118 patients: Logistic regression analysis. *Variables in the univariate logistic regression analysis with the P-value <0.05 are manually inserted in the multivariate logistic regression analysis. Abbreviations: CI: Confidence interval; NLR: Neutrophil-to-lymphocyte ratio; PLR: Platelet-to-lymphocyte ratio.*

contributor toward the progression of severe manifestations of anaphylaxis, including the onset of anaphylactic shock, has emerged as the most common focused subject for research, explaining the decline in platelet count during the attack.[14] PAF, which causes platelets to aggregate, is synthesized from various sources and triggered by inflammatory mediators.[17,18] Neutrophil has a role in releasing PAF as a potent mediator of severe allergic reactions. A study by Vadas indicated that PAF significantly increased during the anaphylactic reaction.[19]

Refractory anaphylaxis: Neutrophil-to-lymphocyte ratio

Our result supports the hypothesis that higher neutrophil and lower lymphocyte counts leading to higher NLR were independently associated with more severe anaphylaxis. Indeed, our NLR in refractory anaphylaxis is consistent with previous allergic condition studies, such as allergic rhinitis and asthma, which also found NLR was significantly higher in severe patients.[7,20] However, our finding is incompatible with studies that found the opposite results. To date, 3 studies have involved CBC comparison in anaphylaxis. An investigation by Tang et al in 2015 compared laboratory results between 2 groups of anaphylaxis patients based on the number of organ systems involved and showed an increase in WBC and neutrophil, but there was no NLR comparison.[21] A previous study identified the difference in anaphylaxis clinical characteristics due to various elicitors and reported laboratory results comparison between mild-to-moderate anaphylaxis and severe anaphylaxis patients did not show a statistically significant WBC difference.[22] NLR and refractory anaphylaxis association has been studied in a recent observational study, which revealed the NLR <0.68 to predict refractory anaphylaxis.[5] It is not apparent why there were differences in findings between studies. Still, it may be due to the differences in characteristics of participants, such as race, age, and allergens.

The intricate pathophysiology of anaphylaxis and the potential roles of neutrophils have been thoroughly addressed.[23,24] Neutrophils have been identified as a component that initiates and contributes to systemic anaphylaxis.[24] The condition was previously known as an immunoglobulin E-dependent allergy.[25] However, recent studies have shown alternative pathways, such as Fc gamma receptors as an immunoglobulin G subset receptor expressed by macrophages, monocytes and neutrophils as exposing immediate effects during anaphylactic reactions by directly encountering immune complex. In contrast, myeloperoxidase is an enzyme predominantly expressed in neutrophils and released upon activation in specific settings, including anaphylaxis.[24,26] However, the pathophysiology of the increased NLR and refractory anaphylaxis association has not been studied well.

Refractory anaphylaxis: Clinical implications of PLR and NLR as predicting indexes

The novel finding from our study was the association between PLR, NLR, and refractory anaphylaxis. In our study, PLR$\geq$129.5 and NLR$\geq$4 were found to be associated with the occurrence of refractory anaphylaxis. PLR and NLR are both easily calculated using the CBC result, a widespread laboratory test indicated worldwide. Therefore, our results suggest that the clinical implications of both figures may be used to predict refractory anaphylaxis. Higher NLR has been studied well in predicting asthma hospitalization,[27] whereas higher PLR has been investigated in more severe allergic rhinitis.[7] However, our study held particular significance for comprehensively analysing clinical and laboratory data in refractory anaphylaxis. Because the association between CBC and the anaphylaxis severity in Vietnam and worldwide has been rarely documented, our research results may contribute to the anaphylaxis grey area for epidemiological data and severity prediction.

Furthermore, in our research, the association between gender and refractory anaphylaxis was observed in men. The mechanism for this different association is unknown.[23] Whereas the epide miology of anaphylaxis was discussed previously demonstrated the higher occurrence of anaphylaxis happened in women,[25,28] a recently published study in an anaphylaxis registry in 2015 by Francuzik et al revealed that males from 13 to 56 years old suffered a modestly elevated risk of anaphylactic severe responses in comparison to females.[29] Also, an anaphylaxis study by Kim et al

in 2018 reported a significantly higher in percentage of severe anaphylactic male patients than the mild-to-moderate group.[22] Beside anaphylaxis, NLR and PLR were also studied in other conditions as part of the acute phase response. Different prognostic values of NLR and PLR were studied in other forms of shock like cardiogenic shock[30–32] or septic shock[33–35] by well-defined research, of which the most important pathophysiology reported was the systemic inflammation process where a significant reduction in lymphocyte count occurred during the acute response. Therefore, the hypothesis in refractory anaphylaxis needs to be tested further by well-characterized longitudinal studies to investigate predictive factors.

Limitations

Our study encountered several limitations. Although the patient data were collected retrospectively over 6 years, the pool of patients might still be small. Based on medical records alone, the presented histories about the anaphylaxis elicitors and clinical symptoms might involve memorizing bias or not fully addressed. We were unable to fully exclude every individual who had records of systemic corticosteroid or antibiotic use. The laboratory tests were drawn when the patient was admitted to the emergency room, but we could not verify the time of blood sample collection and medications administration. Also, the single-center study design was exposed to confoundings and biases. Additionally, because this was an observational study, no causal inference could be made on the association between PLR, NLR, and refractory anaphylaxis as well as delving into the pathophysiology of the elicitors, such as contrast media, to refractory anaphylaxis. Therefore, future research will emphasize nationwide multicenter cohort studies to explore the pathophysiology, prevalence and clinical characteristics as well as laboratory data on anaphylaxis in Vietnam.

CONCLUSION

Elevated PLR and NLR levels are significantly associated with refractory anaphylaxis. These findings suggest that PLR and NLR could serve as useful biomarkers for predicting the severity of anaphylactic reactions. Nevertheless, further research is imperative to validate and consolidate these observations.

Abbreviations

CI, Confidence interval; CMP, comprehensive metabolic panel; NLR, Neutrophil-to-lymphocyte ratio; PLR, Platelet-to-lymphocyte ratio; ELR, Eosinophil-to-lymphocyte ratio; IM, intramuscular; MCH, Mean corpuscular hemoglobin; MCV, Mean corpuscular volume; RBC, Red blood cell; WBC, White blood cell.

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Availability of data and material

The data used or analyzed during the current study are available from the corresponding author on reasonable request.

Author contributions

Conceived and designed the study: Le Vinh Nghi and Pham Dang Hai.
Performed the experiments and data collection: Le Vinh Nghi and Nguyen Hoang Phuc. Analyzed the data results: Le Vinh Nghi and Pham Dang Hai. Wrote the paper and interpretation of data: Le Vinh Nghi and Nguyen Hoang Phuc. Guided the research and revised the manuscript: Pham Dang Hai.

Statement of ethics

Our study was approved by The Institutional Review Board at 108 Military Central Hospital (number 6887/CN-HDDD BV).

Consent for publication

All authors have seen and approved the last version and agreed to publication of the work.

Declaration of competing interest

There are no conflicts of interest between authors.

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