ORIGINAL RESEARCH

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Predictivity of Platelet to Lymphocyte and Neutrophil to Lymphocyte Ratios in the First Trimester Missed Abortion: A Retrospective Case-Control Study

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ABSTRACT Objective: The aim of the study is to investigate the predictive value of neutrophil to lymphocyte ratio (NLR) and platelet to lymphocyte ratio (PLR) in predicting the outcome of missed abortion (MA). **Material and Methods:** In this retrospective case-control study, 50 MA patients and 50 matched healthy pregnant women admitted to our clinic, between January 2018 and November 2021, were included. The preoperative complete blood count parameter of the MA and control group, which were evaluated at the week of gestation in the fetal heart rate was detected in the first trimester were compared. The receiver operating characteristic (ROC) curve was used to calculate the predictive values. **Results:** PLR was higher in the MA group, but this value was not statistically significant (p=0.3). NLR was higher in the MA group and there was a significant difference between the groups (p=0.02). The multivariate analysis also revealed that NLR was the prognostic factor of MA (p=0.04). ROC analysis showed that the best predictive values for MA based on the area under the curve (AUC) was NLR with the optimal cutoff value of 4.56, and the AUC was 0.62, the specificity and sensitivity were 52% and 86% respectively (p=0.02). **Conclusion:** NLR is a suitable predictor of MA in the first trimester. The results of the study may help predict a defective placentation and inflammation in the pathogenesis of MA.

Keywords: Missed abortion; neutrophils; platelet count

Missed abortion (MA) is defined as the part of the embryo losing its viability that remains in the uterus.[1] MA is detected in 15% of clinical pregnancies.[1] Chromosomal abnormalities, uterine anomalies, infections, immunological, and endocrine causes are in the etiopathogenesis of MA.[2] However, the exact cause of its etiopathogenesis remains unknown.[2]

The imperfect placentation may cause a maternal systemic inflammatory response.[3,4] It is known that the levels of many serum inflammatory cytokines and interleukins are increased in MA patients.[5,6] However, these markers are not cheap and accessible enough to be routinely examined. Simple and easily convenient complete blood count (CBC) parameters can be used as indicators of systemic inflammation and stress.[7] Parameters such as neutrophils, platelets and lymphocytes in the report of the CBC can be accessed.[7] The neutrophil to lymphocyte ratio (NLR) and platelet to lymphocyte ratio (PLR) results can be obtained by calculating the ratio of these values to each other.[7] These markers are thought to effectively reflect systemic inflammation and immunity.[7] In this respect, the predictivity of mean platelet volume (MPV) and monocyte lymphocyte ratio (MLR) is considered analogous to the above mentioned markers.[8,9] It has been revealed that these markers and easily calculable rates may be predictive in systemic inflammatory diseases.[10-12]

The common point in the etiopathogenesis of preeclampsia and MA is thought to be placental dysfunction.[13] Inflammatory markers increase in preeclampsia.[14] Based on this hypothesis, some studies have investigated markers such as NLR, PLR, and MLR in the first trimester MA cases.[2,7,15,16]

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There are few studies in the literature evaluating the prediction of MA cases according to CBC parameters. There are controversial cut-off values for proportional calculations. Therefore, in our study, we predicted the outcome of MA cases with these parameters and proportional calculations between them.

MATERIAL AND METHODS

SUBJECTS

This is a retrospective case-control study. Ethical approval was obtained from Dokuz Eylül University Non-Invasive Researches Ethics Committee (date: December 15, 2021; number: 2021/37-11). Informed consent of the patients was obtained. The study was conducted in accordance with the Helsinki Declaration principles. A total of 50 MA patients and 50 matched healthy pregnant women were included. The data of the cases between January 2018 and November 2021 were evaluated. The gestational week of patients was between 6^{0-7} and 13^{+6}. The age of the patients was between 18 and 42.

Gestational week was determined according to the last menstrual period. It was verified by crown rump length measurement on abdominal or transvaginal ultrasound. The study group consisted of MA patients. Miscarriage was defined according to the ultrasound criteria specified in the guidelines.[17,18] Patients with insufficient data, chromosomal anomaly, uterine structural anomaly, acute or chronic infectious disease, cancer, comorbidity under medical treatment, smoking during pregnancy, and followed up pregnancy at another center were excluded from the study.

SAMPLE COLLECTION

Demographic and laboratory characteristics of the patients were obtained from the medical records. CBC parameters, including hemoglobin, hematocrit, platelet, leukocyte, neutrophil, monocyte, lymphocyte, and MPV were determined. The preoperative CBC parameter of the MA and the CBC parameters of the control group, which were evaluated at the week of gestation in the fetal heart rate was detected in the first trimester, were compared.

STATISTICAL ANALYSIS

Analyzes were performed with SPSS version 25.0 (IBM Inc., Chicago, IL, USA). Normality analysis was performed according to the Kolmogorov-Smirnov test. Non-normally distributed hematological parameters were analyzed with the Mann-Whitney U test. Results were expressed as median (minimum-maximum) values. The logistic regression model was performed to compare the multiple effects of important independent factors that may be effective in predicting MA. The specificity and sensitivity analysis of each marker was performed with the receiver operating characteristic (ROC). ROC analysis was performed to calculate the area under the curve (AUC), which indicates the average sensitivity of a marker. The results were 95% confidence interval. The appropriate cut-off value indicating the sum of the highest sensitivity and specificity was calculated for the most predictive marker. The p value considered statistically significant was <0.05.

RESULTS

There was no statistical difference between the groups in terms of gravity, parity and abortion (Table 1). It was determined that the maternal age and gestational age in the MA group were statistically significantly higher (p<0.001, p=0.006, respectively) (Table 1).

Laboratory values of the groups are shown in Table 2. Leukocyte, neutrophil and NLR values were found to be statistically significantly higher in the MA group (respectively; p=0.001, p=0.003, p=0.02). Other laboratory results in the table were similar between the groups.

Logistic regression analyzes are shown in Table 3. Maternal age, gestational week, PLR and NLR

TABLE 1: Demographic and clinical features of the groups.

Variables	Missed abortion (n=50)	Control group (n=50)	p value
Age (years)	31 (19-42)	27.5 (19-40)	<0.001
Gestational weeks*	12 (7-14)	10 (8-14)	0.006
Gravida	3 (1-8)	2 (1-7)	0.1
Parity	1 (0-4)	1 (0-9)	0.9
Abortus	0 (0-3)	0 (0-9)	0.1

*Based on menstrual dates.

TABLE 2: Laboratory values of the groups.

Variables	Missed abortion (n=50)	Control group (n=50)	p value
Hemoglobin (g/dL)	12.1 (7.2-14.5)	12.3 (9.7-14.3)	0.4
Hematocrit (%)	35.7 (19.7-42.9)	36.6 (29.1-42.1)	0.7
Platelet (10^3/μL)	266 (158-672)	245 (146-418)	0.3
Leukocyte (WBC) (10^3/μL)	10.9 (5.8-17.9)	9 (5.2-16.1)	0.001
Neutrophil (10^3/μL)	8 (3.5-16.7)	6.1 (3.2-14)	0.003
Monocyte (10^3/μL)	0.6 (0.2-1.2)	0.5 (0.3-1)	0.4
Lymphocyte (10^3/μL)	1.8 (3.5-1)	1.7 (0.9-4)	0.8
Mean platelet volume (fL)	8.4 (6.8-9.9)	8.5 (6.8-11)	0.5
Neutrophil to lymphocyte ratio	4.6 (1.8-10)	3.3 (1.2-11.6)	0.02
Monocyte to lymphocyte ratio	0.3 (0.1-0.6)	0.3 (0.1-0.6)	0.8
Platelet to lymphocyte ratio	156 (73-280)	135.6 (84-270)	0.3

WBC: White blood cell.

TABLE 3: Logistic regression analysis of independent variables in predicting missed abortion.

	Univariable logistic regression				Multivariable logistic regression*			
Variables	Wald	SE	OR (95% CI)	p value	Wald	SE	OR (95% CI)	p value
Age (years)	12.2	0.49	1.188 (1.079-1.308)	<0.0001				
Gestational weeks[a]	6.7	0.117	1.355 (1.077-1.704)	0.009				
NLR	5.4	0.112	1.29 (1.042-1.613)	0.02	4.1	0.121	1.279 (1.009-1.620)	0.04
PLR	0.8	0.004	1 (0.995-1.013)	0.3				

[a]Based on menstrual dates; *Adjusted by age (days) and gestational weeks; SE: Standard error; OR: Odds ratio; CI: Confidence interval; NLR: Neutrophil lymphocyte ratio; PLR: Platelet lymphocyte ratio.

were included in the univariate analyses. NLR was found as a significant independent factor (p=0.02). PLR was not found as a significant independent factor (p=0.3). The multivariate analysis also revealed that NLR was the prognostic factor of MA (p=0.04).

ROC analysis was performed to determine the diagnostic NLR value for MA (Figure 1). It was concluded that the best predicted value for MA based on AUC was NLR with an optimal cut-off value of 4.56 and AUC of 0.62, with specificity and sensitivity of 52% and 86%, respectively.

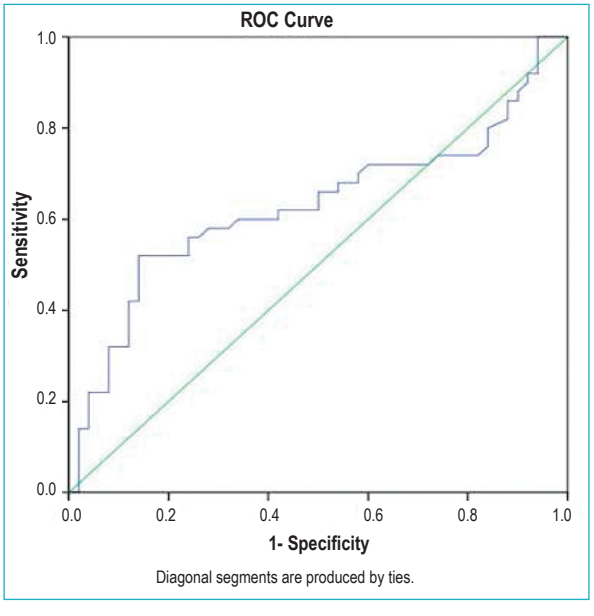

FIGURE 1: The ROC curve analysis for the NLR. The area under the curve for the NLR was 0.629 (95% confidence interval, 0.515-0.742; p=0.02).
ROC: Receiver operating characteristic; NLR: Neutrophil lymphocyte ratio.

DISCUSSION

As the maternal age increases above 35, the risk of MA gradually increases.[19] There was a significant difference in the MA group in terms of maternal age. The mean gestational week of the MA group was higher. Further analysis was performed to determine

that the statistically significant difference between the groups did not affect our main result. In the logistic regression analysis, we revealed that NLR can predict MA cases, independent of the age and gestational week. Recurrent abortion patients are more likely to have abortion compared to the normal population.[19] Recurrent miscarriage group was not excluded from our study. However, the groups were similar in terms of abortion history.

Progesterone has a significant effect on every step of normal pregnancy.[20] Progesterone is extensively prescribed for treating recurrent miscarriages and threatened abortion.[20] The levels of progesterone are crucial factors in regulating the white blood cell (WBC) counts.[21] WBC and granulocyte counts increase in the luteal phase compared with the follicular phase. This is related to progesterone, which is more effective in the luteal phase.[22] In our study, hemoglobin, hematocrit, platelet, monocyte, lymphocyte and MLR values were similar between the groups. WBC value was higher in the MA group with a statistically significant difference. Wang et al. analyzed MA cases at week 7 in terms of WBC value.[16] Contrary to our data, they reported that the risk of MA increased when this value was below $8x10^3/\mu L$ in MA cases (84% sensitivity and 54% specificity). Wang et al. further explored the effects of progesterone on the total or differential WBC value, NLR and MLR in MA cases. However, it was revealed that progesterone treatment did not create a significant difference between the groups in terms of these parameters.[16]

MPV is a specific parameter of the platelet function. Larger platelets are hemostatically more active. These factors have a significant impact on the development of intracoronary thrombus and acute myocardial infarction (AMI).[23] The high MPV value of patients with AMI also increases the risk of mortality and the need for revascularization.[23] MPV continues to increase after AMI returns to normal levels within the recovery period.[23] MPV is a parameter detected during routine blood counts to which clinicians do not often pay attention.[24] Increase of MPV is due to the synthesis of prothrombotic and proinflammatory agents in platelets and release of reactive platelets.[25] Increased MPV and increased platelet aggregation are also associated with obstetric causes and outcomes. It has been theoretically suggested that preeclampsia is involved in the etiopathogenesis. Kim et al. determined that MPV was normal in spontaneous vaginal delivery. MPV value is found to be higher in abortion, spontaneous premature rupture of membranes, and preeclampsia.[26,27] MPV was also greater in abnormal pregnancies compared with normal Doppler examination results.[28] Recently, the ability of markers such as NLR and PLR to predict the prognosis of inflammation related diseases has been studied.[29] Additionally, it has been reported that these markers may be independent prognostic factors evaluating mortality and morbidity in cancer and cardiovascular diseases.[30] Liu et al. found the PLR value to be similar between the MA and control groups. MPV value was significantly lower in the MA group.[7] Biyik et al. reported that in MA group, the rates of PLR and MPV were statistically significant higher.[2] Interestingly, MPV value was found to be lower in the MA group in our study. PLR value was higher in the MA group. The results of the Eroglu et al.'s study are parallel to our results in terms of MPV.[15] The functionality of platelets changes in normal pregnancy. Tygart et al. draws attention to the sensitivity of this functionality.[31] Thus, in pregnancies, as a sign of platelet function, MPV is more important than platelet number. In our study, platelet counts in MA group were not significantly different from the control group. We observed that platelet counts in MA were slightly higher in relation to the normal pregnancies.

Neutrophil value and NLR were statistically significantly higher in the MA group. Liu et al. reported higher neutrophil values and NLR in the control group.[7] However, they found no statistical difference between the groups. In contrast, Biyik et al. reported that NLR was statistically higher in the MA group.[2] However, Wang et al. stated that, contrary to the abovementioned data, the NLR value in the MA group was low, which showed a statistically significant difference.[16] According to the ROC analysis, the best predictive marker for MA was found to be NLR. The optimal cutoff value was 4.56. It was shown that the AUC was 0.62, the specificity and sensitivity were 52% and 86%, respectively. Considering other proportional calculations in the literature, the sensitivity value in our study is similar to that of others.

The specificity value is higher than literature.[2,7,29] The NLR threshold value calculated to predict MA was reported to be below 2.4 in Wang et al.'s study and below 5.72 in Kim's study.[16,29] The cut-off value in these studies is in the opposite direction with our results. The difference between these studies in the literature is due to 2 reasons. First, the progesterone admission status of the patients may affect the outcome. Secondly, it is not possible to calculate the change in serum biomarkers between the time when the patients' MA status occurs and the time the diagnosis is detected by ultrasound.

The type of research is retrospective. This is the weakness of our study. The strengths of our study are that it was conducted in a tertiary hospital, the reliability of the data records, and the independent factors predicting MA were well analyzed.

CONCLUSION

We were relevelanced about infectious agents causing MAs in the first trimester. Therefore, we focused on infection markers such as PLR and NLR. These are simple and rapid markers. According to our study, PLR was not detected as a prognostic marker. Statistically, NLR was the only prognostic value for this study and the threshold value of NLR was 4.56. This means that a level above 4.56 increases the rate of MA. Further large-sample sized, multicenter, and prospective studies are needed to elucidate the effect of infectious markers on MA.

Source of Finance

During this study, no financial or spiritual support was received neither from any pharmaceutical company that has a direct connection with the research subject, nor from a company that provides or produces medical instruments and materials which may negatively affect the evaluation process of this study.

Conflict of Interest

No conflicts of interest between the authors and / or family members of the scientific and medical committee members or members of the potential conflicts of interest, counseling, expertise, working conditions, share holding and similar situations in any firm.

Authorship Contributions

Idea/Concept: *Onur Yavuz;* ***Design:*** *Onur Yavuz, Aslı Akdöner;* ***Control/Supervision:*** *Onur Yavuz, Ömer Erbil Doğan;* ***Data Collection and/or Processing:*** *Onur Yavuz, Mehmet Eyüphan Ögözen;* ***Analysis and/or Interpretation:*** *Onur Yavuz, Mehmet Eyüphan Ögözen;* ***Literature Review:*** *Aslı Akdöner;* ***Writing the Article:*** *Onur Yavuz, Ömer Erbil Doğan;* ***Critical Review:*** *Onur Yavuz, Ömer Erbil Doğan.*

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