The Association between Platelet Indices and Unexplained Recurrent Miscarriage

Original Article

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ABSTRACT

Introduction: Recurrent miscarriage is an important health issue. It is defined loss of three or more consecutive pregnancies. It has multifactorial causes. Platelets (PLT) are generally known as a marker for inflammation and thrombosis.

Aim: This work aimed to find the association between platelet indices and recurrent unexplained miscarriage.

Materials and Methods: This study was a case control study conducted at Ain Shams University Maternity Hospital (ASUMH) recruiting women from the outpatient clinic and the recurrent pregnancy loss clinic over the period from December 2018 to August 2019. Group A included 35 women with recurrent miscarriage (RM), Group B included 35 women without history of miscarriage and delivered at least once. Platelet count and indices were measured in both groups and statistical analysis was done.

Results: In the current study, there was no statistically significant difference between the cases and controls as regard PLT count, plateletcrit (PCT), mean platelet volume (MPV), and platelet distribution width (PDW).

Conclusion: Platelet indices are not useful as predictor for recurrent miscarriage.

Key Words: Mean platelet volume, platelet distribution width, plateletcrit, recurrent miscarriage

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INTRODUCTION

Recurrent pregnancy loss (RPL) is defined as two or more failed clinical pregnancies as documented by ultrasonography or histopathologic examination. According to the Royal College of Obstetricians and Gynaecologists; spontaneous miscarriage is defined as spontaneous loss of pregnancy before foetus reaches viability; while recurrent miscarriage is defined as loss of three or more consecutive pregnancies[1].

Also, miscarriage is defined as the loss of a clinical pregnancy before 20 completed weeks of gestational age (18 weeks after fertilization). If gestational age is unknown, it will be considered according to the weight (the loss of an embryo/foetus<400 gram)[2]. Ectopic, molar and biochemical pregnancies are thus not included(3). Spontaneous miscarriage is a relatively common event, occurring approximately in 15%-25% of pregnancies, and increasing in prevalence with maternal age[3].

Thus, the incidence of miscarriage is seen in 5 % of couples with two or more losses and in 1–2 % of those with three or more losses. These findings suggest that most RPL is not due to chance alone and should be investigated clinically[4]. Although the aetiology of recurrent miscarriage is multifactorial; the cause of recurrent miscarriage can't be clarified in 50%-75% of cases, which causes psychological problems for the couples with unexplained recurrent miscarriage[1].

There is a natural pregnancy induced hypercoagulability which serves as a physiologically adaptive mechanism to prepare for haemostasis postpartum[6]. The increase in prothrombotic factors and the decrease in antithrombotic factors result in modification of the haemostatic balance in the placental vessels and cause placental perfusion and fetomaternal circulation insufficiency[7].

Successful implantation of the embryo in normal physiologically pregnancy requires the presence of sterile inflammation[7]. So, persistent and uncontrolled inflammatory responses can damage placental growth and cause miscarriage[8]. As the polymorphism of cytokines such as IL-6, IL-8 and TNF-α triggers thrombotic and inflammatory processes in maternal blood vessels causing miscarriage[9].

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Platelets are a component of blood whose function along with the coagulation factors is to react to bleeding from blood vessel injury by clumping, thereby initiating a blood clot. They are activated rapidly to provide the initial response to the vascular injury[10]. So they are also important for the process of chronic inflammation that is associated with disease pathology[11]. Platelet indices such as mean platelets volume (MPV), platelets distributed width (PDW), and plateletcrit (PCT) are useful as cheap non-invasive biomarkers for assessing the diseased states[12]. They are related to platelet's morphology and proliferation kinetics. Mean platelet volume (MPV) is an analyser-calculated measure of thrombocyte volume. Platelet distribution width (PDW) is an indicator of volume variability in platelets size. Plateletcrit (PCT) is the volume occupied by platelets in the blood[13].

These parameters have been investigated as the markers of platelet activation and predictors of many disorders as thrombosis, and inflammation[14]. As RM can be correlated with the inflammatory processes, so platelet indices may be used for prediction of RM.

AIM OF THE WORK

The aim of this study is to find the association between platelet indices and recurrent unexplained miscarriage.

PATIENTS AND METHODS

This study was a case control study conducted at Ain Shams University Maternity Hospital (ASUMH) recruiting women from the outpatient clinic and the recurrent pregnancy loss clinic over the period from December 2018 to August 2019. Women were included according to ASRM criteria as cases: Group A 35 women with history of recurrent miscarriage (two or more consecutive miscarriages); controls: Group B 35 women without previous miscarriages and delivered at least once.

Women were excluded from the study recruitment if they were pregnant women, chromosomal abnormalities detected by peripheral karyotypic analysis of the parents, women with antiphospholipid syndrome as screened by lupus anticoagulant, anticardiolipin antibodies and anti-b2 glycoprotein I, women with hormonal and metabolic factors (e. g. thyroid disorders as screened by TSH level, hyperprolactinemia as screened by serum prolactin level, and uncontrolled diabetes mellitus), women with known uterine anomalies as detected by sonohysterogram, hystero-salpingogram, and/or hysteroscopy, obesity (BMI >30%), smokers, alcohol consumption (3 to 5 drinks per week) and cocaine use.

The study was approved by the local institutional ethical committee and an informed consent was taken from all participants after full explanation of the steps and significance of the study.

All patients were subjected to detailed history including personal and demographic data and general examination for BMI, blood pressure. transvaginal ultrasound: Women with uterine anomalies were excluded. All other enrolled women were tested by CBC by XN-1000 (SA-01) machine. The blood sample was preserved by EDTA and was processed within maximum 2 hours. The following platelet indices were recorded (measured by the automatic analyzer): Mean Platelet Volume (MPV), Platelet Distribution Width (PDW), Plateletcrit (PCT).

Sample size justification: The means of the platelet indices were used to calculate the sample size. Group sample sizes of 35 per group achieve 80% power to reject the null hypothesis of zero effect size when the population effect size is 0. 70 (moderate to large) and the significance level (alpha) is 0. 050 using a two-sided two-sample equal-variance t-test (Machin *et al*.,1997).

Data collection and recording: Data of each patient (age, parity, number of previous miscarriages, BMI, platelet indices) were recorded in a Case Record Form (CRF).

STATISTICAL ANALYSIS

Descriptive statistics for measured variables were expressed as mean and standard deviation (for metric data); median and interquartile range (for parametric data); and number and percentage (for categorical data). The platelet indices in each clinic were compared using the t-test or Mann-Whitney U test according whether data are normally distributed or skewed. A P-value of 0. 05 or less were considered statistically significant. To determine the best cut-off value for each of the platelet indices for prediction of recurrent miscarriage, a receiver-operator characteristic curve (ROC) curve were plotted for each platelet index, and the value associated with the highest Youden index ([sensitivity + specificity] – 1) will be taken as the best cut-off value. The following quality indices will be calculated to examine the predictive performance of the different platelet indices: sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and diagnostic odds ratio (DOR).

RESULTS

This study was a case control study conducted at Ain Shams University Maternity Hospital (ASUMH) recruiting women from the outpatient clinic and the recurrent pregnancy loss clinic over the period from December 2018 to August 2019. One hundred forty women were enrolled. Fifty-eight women were excluded (consent wasn't given in 13 women, uterine anomalies were found in 8 women,

antiphospholipid syndrome was diagnosed in 15 women, abnormal TSH found in 3 women, abnormal HbA1c found in 9 women and BMI was above 30% in 10 women). Eighty-two participants were eligible for the study. Complete blood picture for platelet indices test was done. Inconclusive test due to technical problem in the sample provided was found in 12 samples. Seventy women were eligible. Group (A) included 35 women with unexplained recurrent miscarriage (RM) according to ASRM criteria, Group (B) included 35 women without history of miscarriage and delivered at least once.

This table shows descriptive analysis of the whole population as regard age and BMI. There was insignificant difference as regard the BMI (Table 1).

There is significant difference as regard gravidity between cases and controls because the cases had multiple trials to conceive. On the other hand, there is significant difference as regard parity between cases and controls because the eligibility criteria recommend that controls should have delivered once or more as in Table 2.

Indeed, there is no value of comparing miscarriage rate between cases and controls, because the eligibility criteria recommend that controls don't have any miscarriages at all. On comparing the two studied groups as regard platelet count, there was no significant difference between the two studied groups with *p value 0. 888*. This is shown in Table 3.

On comparing the two studied groups for platelet indices, there was no significant difference between them as regards MPV, PDW and PCT (Table 4).

As regard the cases, we explored the possibility of finding any correlation between the platelet count, platelet indices and the timing of miscarriage. We subgrouped the cases into group (X) with first trimesteric miscarriage and group (Y) with second trimesteric miscarriage. It was found that there is no significant difference in subgroups of cases (first and second trimesteric miscarriages) as regard PLT indices namely (PCT, MPV, PDW) and platelet count as in Table 5.

Table 1: Comparison between the two studied groups according to demographic data:

	Cases (n = 35)	Control (n = 35)	*p*
Age (years)			
Mean ± SD.	26. 89 ± 5. 69	31. 26 ± 5. 30	0. 001*
BMI (kg/m^2)			
Mean ± SD.	25. 44 ± 2. 77	25. 26 ± 2. 44	0. 776

p: p value for comparing between the studied groups

*: Statistically significant at $p \leq 0.05$

Table 2: Comparison between the two studied groups according to obstetric history:

	Cases (n = 35)	Control (n = 35)	*p*
Gravidity			
Min. – Max.	2. 0 – 9. 0	1. 0 – 5. 0	<0. 001*
Median (IQR)	4. 0	3. 0 (2. 0 – 3. 0)	
Parity			
Min. – Max.	0. 0 – 5. 0	1. 0 – 5. 0	<0. 001*
Median (IQR)	0. 0 (0. 0 – 2. 0)	3. 0 (2. 0 – 3. 0)	

p: p value for comparing between the studied groups

Table 3: Comparison between the two studied groups according to Platelet count:

PLT count	Cases (n = 35)	Control (n = 35)	*p*
Median (IQR)	284. 0 (238 – 329)	287 (249 – 340)	0. 888

p: p value for comparing between the studied groups

Table 4: Comparison between the two studied groups according to PLT indices:

PLT indices	Cases (n = 35)	Control (n = 35)	*p*
MPV			
Mean ± SD.	11. 04 ± 0. 96	10. 65 ± 0. 80	0. 069
PDW			
Mean ± SD.	13. 37 ± 2. 21	12. 46 ± 1. 90	0. 070
PCT			
Mean ± SD.	0. 31 ± 0. 07	0. 31± 0. 08	0. 755

p: p value for comparing between the studied groups

Table 5: Comparison between the subgroups of cases according to platelet indices:

Platelet indices	1st trimester (n = 21)	2nd trimester (n = 14)	*p*
PCT Mean ± SD.	0. 30 ± 0. 08	0. 32 ± 0. 06	0. 468
MPV			
Mean ± SD.	11. 08 ± 1. 12	10. 98 ± 0. 69	0. 773
PDW			
Mean ± SD.	13. 59 ± 2. 65	13. 04 ± 1. 33	0. 426
Platelet count			
Median (IQR)	290. 0 (231. 5 – 328. 5)	278. 5 (247. 0 – 333. 5)	0. 933

p: p value for comparing between the studied groups.

DISCUSSION

Recurrent miscarriage is a heterogeneous multifactorial condition. Its incidence has been estimated about 1% among fertile couples of reproductive age[3]. However, while there are various possible causes, in more than 50%-60% of cases, no clear cause can be identified[1]. Pregnancy is a state of hypercoagulability due to alterations of coagulation proteins. The increase in prothrombotic factors and decrease in antithrombotic factors result in modification of the haemostatic balance in placental vessels, causing insufficient placental perfusion and feto-maternal circulation insufficiency[15].

Evidence shows that impaired inflammatory response is implicated in numerous female reproductive tract pathologies including recurrent miscarriage[16]. In the pathogenesis of RM, inflammation and coagulation disorders are proposed to have important role, since immunopathological evaluation of the abortus material at the placental implantation site reveals inflammation and fibrin deposition in the decidua and thromboembolism in decidual vessels.

Several physiological and pathological conditions have impact on platelet activation and change. The number and activities of platelets in complicated pregnancies have been evaluated in many studies. Preeclampsia, premature rupture of membranes (PROM), and intra uterine growth restriction (IUGR) are the most commonly studied pregnancy complications[17]. These studies have reported that increased MPV (and therefore increased PLT activity) increases placental PLT aggregation, causing thrombosis and placental deficiency[18].

This study aimed at exploring the relationship between unexplained recurrent miscarriage and PLT indices, which are thought to serve as inflammatory markers for various diseases. Measurement of PLT count and other platelet indices may have a predictive value for RM. Increased levels of platelet activating factor were observed in serum samples from women who have suffered two or more RM. This could have implications for placental function and fetal growth[19]. However, the role of platelet indices in these pathways still is not known completely. The present study aimed to compare changes in platelet indices namely: PLT count, MPV, PDW, and PCT, between women with unexplained recurrent miscarriage and women without miscarriage and had delivered at least once.

This study was a case control study conducted at Ain Shams University Maternity Hospital (ASUMH) recruiting women from the outpatient clinic and the recurrent pregnancy loss clinic over the period from December 2018 to August 2019. One hundred forty women were enrolled. Fifty-eight women were excluded (consent wasn't given in 13 women, uterine anomalies were found in 8 women, antiphospholipid syndrome was diagnosed in 15 women, abnormal TSH found in 3 women, abnormal HbA1c found in 9 women and BMI was above 30% in 10 women). Eighty-two participants were eligible for the study. Complete blood picture for platelet indices test was done on them. Inconclusive test due to technical problem in the sample provided was found in 12 samples. Seventy women were eligible. Group (A) included 35 women with unexplained recurrent miscarriage (RM), Group (B) included 35 women without history of miscarriage and delivered at least once.

Recurrent miscarriage was defined as two or more consecutive pregnancy losses (<20 weeks). Women who had experienced RM due to any identifiable were excluded.

The demographic characteristics of the present study revealed that the two study groups were not statistically different in terms of body mass index. Women with RM had significantly higher gravidity (with median 4 for cases versus 2 for controls) and significantly lower parity as they have multiple trials to conceive.

Furthermore, there was no need to compare between cases and controls as regard number of miscarriages as the eligibility criteria recommend that the controls should have delivered at least once and don't have any miscarriages at all.

Platelet count is considered generally marker for inflammation[20]. The present study stated that there was no statistically significant difference for PLT count between the two groups. This agrees with Dundar *et al.* study that was retrospective evaluation of 60 women who had a history of recurrent pregnancy loss, 60 healthy women who had a first trimester pregnancy and 60 healthy parous women; found that there is no significant difference in PLT count in women with RM[21]. Also, Ural *et al.* conducted a prospective study based on the comparison of 74 patients with unexplained recurrent first-trimester pregnancy loss with 208 control subjects matched for age. The two groups were compared in terms of platelet indices and found that there was no significant difference for PLT count between the two groups[6]. In contrast, a study by Aynıoglu *et al* .as conducted on 208 patients who experienced 2 or more first trimester spontaneous abortions and 95 controls who had no abortions showed that the PLT count and PCT were significantly higher in patients with RPL than in controls[8]. Also Alaghbary *et al.* study which was retrospectively conducted on 45 women with a history of RPL and 45 women who gave birth without RPL disagrees with our study and found that there was significant difference in PLT count in women with RPL[5].

Plateletcrit (PCT) is an indicator of circulating PLTs in a unit volume of blood[22]. In the present study when compared with control women, the women with a history

of RM, PCT values were not significantly different with a *P value 0.755* (mean PCT for cases 0.31± 0.07 Vs 0.31± 0.08 for controls), and no cut-off value could be used to predict cases of RM. This agrees with Dundar *et al.* and Ural *et al.* studies which reported that there was no significant difference in PCT values between the studied groups[21]. While study by Aynıoglu *et al.* and Alaghbary *et al.* showed that the PCT values were highly significant different in RPL patients[8].

Platelet functions and aging are indicated by the platelet mass and examined with the use of MPV. It has been reported that PLT aggregation and thrombosis during early placental development can cause pregnancy loss, and this can be associated with high MPV values[23]. The present study showed that there is no significant difference in MPV between the two study groups with a *P value 0.069* (mean of MPV for cases 11.04 ± 0.96 Vs 10.56 ± 0.80 for controls) and no cut-off value could predict cases of RM. This agrees with Ural et al study which found that there is no significant difference in MPV in women with RM[6].

On the other hand, a preliminary study by Yilmaz *et al.* retrospectively investigated the data of 120 patients with unexplained recurrent miscarriage and compared them with the data of 120 match-paired controls to detect the relationship between MPV and RPL and reported a significantly higher MPV values in patients with RPL[24]. This was similar to the study by Aynıoglu *et al.* which showed a significant increase in the MPV in RPL patients[8].

Platelet distribution width (PDW) is an indicator of volume variability in platelet size. It has been implicated as a specific marker of platelet activation. Our study did not find a significant difference in PDW between cases and controls with a *P value 0.070* (mean of PDW for cases 13.37± 2.21 Vs 12.46±1.90 for controls).

However; Ural *et al.* study stated that the PDW increased with RPL[6]. Also, Dundar *et al.* reported that there is elevation in PDW values in women with RPL. Alaghbary *et al.* study found that there was significant difference in PDW in women with RPL[5].

As regard the cases, we explored the possibility of finding any correlation between the platelet count, platelet indices and the timing of miscarriage. We subgrouped the cases into group (X) with first trimesteric miscarriage and group (Y) with second trimesteric miscarriage. It was found that there is no significant difference in subgroups of cases (first and second trimesteric miscarriages) as regard PLT indices namely (PCT, MPV, PDW) and platelet count.

DISCUSSION

Neither PLT count nor platelet indices are useful as predictor of cases with recurrent miscarriage.

RECOMMENDATIONS

Platelet indices test isn't recommended for screening for recurrent miscarriage. Further studies with larger groups with different trimesteric miscarriages are needed for confirmation of the test of platelet indices as predictor for RM.

REFERENCES

1. Green Top Guidelines No.17; The Investigation and Treatment of Couples with recurrent First-trimester and Second-trimester Miscarriage. Royal College of Obstetrics and Gynecology 2011. Last accessed at October 2018. https:/www.rcog.org.uk.gtg17.

2. El Hachem H, Vincent C, Pascale MP, Philippe D, Guillaume L and Pierre EB. Recurrent pregnancy loss: current perspectives. Int J Women's Health. 2017; 9: 331–345.

3. American Society for Reproductive Medicine practice committee. Evaluation and treatment of recurrent pregnancy loss: a committee opinion. Fertil Steril. 2012; 98(5):1103–1111.

4. Bashiri A, Ratzon R, Amar S, Serjienko R, Mazor M and Shoham-Vardi I. Two vs. three or more primary recurrent pregnancy losses- are there any differences in epidemiologic characteristics and index pregnancy outcome? J Perinat Med. 2012; 40:365–71.

5. Al Aghbary A, Mohammed AA, Dikra WM, Waiel MA. Platelet indices in evaluation of patients with recurrent pregnancy loss. Asian Pacific Journal of Reproduction. 2018; 7: 15-18.

6. Ural Mete Ü, Bayoğlu TY, Balik G, Kir ŞF and Çolak S. Could platelet distribution width be a predictive marker for unexplained recurrent miscarriage? Archives of Gynecology and Obstetrics. 2014; 290 (2): 233–236.

7. Ramsay M. Normal haematological changes during pregnancy and the puerperium. In Pavord S, Hunt B (ed). The Obstetric Haematology Manual. 2010: 3–12.

8. Aynıoglu O, Isık H, Sahbaz A, Harma M I, Isık M, Kokturk F. Can Plateletcrit be a Marker for Recurrent Pregnancy Loss? Clinical and Applied Thrombosis/ Hemostasis. 2014; 22 (5), 447–452.

9. Vallée N M, Obari D, Palacios J, Brien MÈ, Duval C, Chemtob S,Girard, S. Sterile inflammation and pregnancy complications: a review. Reproduction. 2016: 152(6), R277–R292.

10. Machlus KR, Thon JN and Italiano JE. Interpreting the developmental dance of the megakaryocyte: a review of the cellular and molecular processes mediating platelet formation. British Journal of Haematology. 2014,165 (2): 227–36.

11. Norris LA. Blood Coagulation. Best Practice and Research Gynecology. 2003; 17:369-383.

12. Tamburrelli C, Gianfagna F, D'Imperio M, De Curtis A, Rotilio D, Iacoviello L and Cerletti C. Postprandial cell inflammatory response to a standardised fatty meal in subjects at different degree of cardiovascular risk. Thrombosis and Haemostasis. 2012; 107 (03), 530–537.

13. Kakali G and Bhatta C. Overview of Platelet Physiology: Its Haemostatic and Nonhemostatic role in disease pathogenesis. The scientific World Journal. 2014;1-16.

14. Ustundag BY, Murat P and Kagan H. The use of platelet indices, plateletcrit, mean platelet volume and platelet distribution width in emergency non-traumatic abdominal surgery: a systematic review: Biochemia Medica. 2016; 26 (2):178–193.

15. Thornton P and Douglas J. Coagulation with pregnancy. Best practice & Research Clinical Obstetrics and Gynecology 24 (2010); 339-352.

16. Laird SM, Tuckerman EM, Cork BA, Linjawi S, Blakemore AI and Li TC. A review of immune cells and molecules in women with recurrent miscarriage. Hum Reprod Update 9,2003.

17. Howard JA. Recurrent Pregnancy Loss; Causes, Controversies and Treatment. Journal of Maternal-Fetal & Neonatal Medicine, 2015.

18. Kaplanoglu M, Yuce T and Bulbul M. Decreased mean platelet volume is associated with the developing stage of fetoplacental unit in spontaneous abortion. Int J Clin Exp Med 2015;8:11301-6.

19. Eser A, Inegol GI, Erdamar H, Kaygusuz I, Yildirim M and Usluogullari B. Levels ofthrombin-activatable fibrinolysis inhibitor and platelet-activating factor in recurrent pregnancy loss patients. Taiwan J Obstet Gynecol. 2016; 55:60-3

20. Zareifar S, Farahmand Far MR, Golfeshan F and Cohan N. Changes in Platelet Count and Mean Platelet Volume. During Infectious and Inflammatory Disease and Their Correlation With ESR and CRP. Journal of Clinical Laboratory Analysis. 2014; 28 (3), 245–248.

21. Dundar o, Mıne Kanat Pektas, Serkan Bodur, Lale Vuslat Bakır and Ahmet Cetin. Recurrent pregnancy loss is associated with increased red cell distribution width and platelet distribution width. J. Obstet. Gynaecol. Res. 2014.

22. Bain BJ, Dacie JV, Lewis SM. Dacie and Lewis. Practical haematology. 11th ed. Edinburgh:Churchill Livingstone; 2012

23. Rappaport VJ, Velazquez M, Williams K. Hemoglobinopathies in pregnancy. Obstet Gynecol Clin North Am 2004;31:287-317.

24. Yilmaz M, Delibas IB, Isaoglu U, Ingec M, Borekci B and Ulug P. Relationship between mean platelet volume and recurrent miscarriage: a preliminary study. Arch Med Sci. 2015; 11: 989-93.