ORIGINAL ARTICLE

Pseudothrombocytopenia Unveiled: Navigating Frequency and Causes in a Tertiary Care Hospital

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ABSTRACT

Objective: To determine the proportion and laboratory-identified mechanisms of pseudothrombocytopenia among patients with automated platelet counts <100 × 10^9/L in a tertiary care hospital.

Study Design: Descriptive Cross-sectional study

Place and Duration of Study: This study was conducted in the Department of Pathology, Fauji Foundation Hospital Rawalpindi, from January 01, 2025, to June 30, 2025.

Materials and Methods: All patients undergoing complete blood count analysis during the study period with an automated platelet count below 100 × 10^9/L were included. Peripheral blood smears were examined to classify the subtypes of thrombocytopenia into true thrombocytopenia and pseudothrombocytopenia, including platelet clumping, giant platelets, and abnormal platelet distribution. Data was analyzed using SPSS Statistics version 29.0. Chi-square test was applied for categorical variables, and p-value *< 0.05* which is considered statistically significant.

Results: Out of total 755 blood samples**,** morphologically consistent with true thrombocytopenia (absence of smear artefacts) accounted for the majority of cases (70.7%). Among the 221 cases of pseudo-thrombocytopenia (29.3%), platelet clumping was the most frequent cause (19.7%), followed by giant platelets (7.9%) and abnormal platelet distribution (1.6%).

Conclusion: Pseudothrombocytopenia remains a frequent laboratory artefact in patients with low automated platelet counts. Accurate classification through systematic peripheral blood smear evaluation is essential to differentiate pseudothrombocytopenia from true thrombocytopenia. Implementation of standardized laboratory verification practices supports reliable platelet count reporting and enhances patient safety.

Key Words*: Automated Hematology Analyzers, EDTA, Peripheral Blood Smear, Platelet Count, Pseudothrombocytopenia.*

Introduction

Pseudothrombocytopenia (PTCP) is well-known in vitro diagnostic artefact characterized by spuriously low automated platelet counts despite adequate in vivo levels. [1] Platelet clumping is most commonly observed in ethylenediaminetetraacetic acid (EDTA) anticoagulated samples, when cryptic glycoprotein epitopes are exposed and subsequently clumped by autoantibodies which cause in vitro platelet clumping and underestimation by automated analyzers, which fail to count as individual cells. Although EDTA induced clumping is common mechanism spuriously low automated platelet counts may also arise from additional analytical and pre-analytical factors, indicating that the phenomenon is multifactorial.[2]

This phenomenon poses a significant diagnostic challenge and laboratory reporting challenge. Failure to recognize PTCP cases might result in misdiagnosis, unnecessary delayed therapeutic interventions, additional costly testing, unnecessary transfusions, bone marrow examination, and patient anxiety. [3] According to recent data, the prevalence in routine laboratory practice is between 0.03 to 0.3%; however, hospitalized and hematology-oncology populations may have higher frequencies.[4]

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EDTA-induced platelet clumping is the most common cause of spuriously low automated platelet counts, while true thrombocytopenia may coexist with artefactual mechanisms, further complicating interpretation. [5] Furthermore, the presence of giant platelets can be misclassified by analyzers based on size thresholds, often being counted as other blood cells. A less common but notable cause is abnormal platelet distribution, often due to partial clotting or platelet satellitism around neutrophils, which also leads to erroneous underestimation.[6]

Although detection has been improved by advances in laboratory diagnostics, such as digital image analysis, alternate anticoagulant tubes (citrate, magnesium, or heparin), and improved analyzer flagging, standardization remains a concern due to variations in laboratory methods. [7] Recent reports have described transient PTCP associated with infections and immunologic stimuli, including SARS-CoV-2 infection and post-vaccination states, highlighting the dynamic nature of this artefact.[8]

The absence of systematic studies classifying PTCP according to its underlying mechanisms in tertiary care laboratories restricts evidence-based reporting practicesA comprehensive knowledge of the prevalence and implications of PTCP in our tertiary care setting would be possible via systematic study of the condition in a focus on its main causative categories: giant platelets, platelet clumping, genuine thrombocytopenia and abnormal distribution. Such studies are crucial to establish laboratory algorithms that ensure accurate reporting, avoid unnecessary treatments and enhance patient safety.[9] Given the diagnostic and clinical implications of pseudothrombocytopenia and the lack of standardized laboratory data; the objective of this study was to determine the proportion and laboratory-identified mechanisms of pseudothrombocytopenia among patients with automated platelet counts <100 × 10^9/L in a tertiary care hospital.

Materials and Methods

The study was designed as a descriptive cross-sectional observational study and was conducted in the Department of Pathology, Fauji Foundation Hospital, Rawalpindi, from January 01, 2025 to June 30, 2025. with ethical approval obtained from the Institutional Review Committee (Application Ref Number: 780/RC/FFH/RWP, dated January 4th, 2024). Patient confidentiality was maintained throughout the study. No additional blood sampling was performed other than specified in this study protocol. All eligible patients meeting the inclusion criteria during the study duration were consecutively included. Both outpatient and admitted patients were considered, and those with an automated platelet count below 100 ×10^9/L, with samples received in EDTA-anticoagulated tubes ensuring adequate sample volume and proper labeling were included. Patients were excluded if they had a platelet count above 150 × 10^9/L, known cause of thrombocytopenia(documented clinical diagnosis in hospital records (e.g., ITP, aplastic anemia, chemotherapy-induced thrombocytopenia) prior to current sample evaluation), who were taking drugs known to induce thrombocytopenia or had clotted and hemolyzed samples. Venous blood (2 mL) samples were collected by trained phlebotomy staff under aseptic conditions in tubes containing anticoagulant Ethylenediaminetetraacetic acid (EDTA) and analyzed within two hours using the Sysmex XN-1000 Automated Hematology Analyzer. All procedures adhered to the manufacturer's instructions and departmental standard operating procedures. underwent peripheral. Smears were prepared and stained with Leishman stain and manual platelet counts were performed on samples with platelet counts below 100 × 10^9/L. A third-year hematology resident examined the smears for giant platelets, platelet clumps, platelet satellitism and abnormal platelet distribution, with findings subsequently verified by senior consultant hematologists. Platelet clumping was defined by the presence of two or more discrete platelet aggregates per low-power field (10×) or readily visible clumps on smear scanning, associated with spuriously low automated platelet counts. Giant platelets were identified when platelet size equaled or exceeded that of a normal red blood cell (>7 µm), with individual platelets clearly recognizable on oil immersion. Abnormal platelet distribution was diagnosed when platelets showed uneven peripheral smear dispersion, including satellitism around leukocytes, in the absence of fibrin strands or widespread clot formation. Confirmatory testing with alternative anticoagulants was not routinely

performed. Data analysis was performed using SPSS version 29.0. Descriptive statistics were used to summarize continuous variables as mean ± standard deviation and categorical variables as frequencies and percentages. Cross tabulations were performed to examine the distribution of thrombocytopenia causes across gender, patient setting, age groups and hospital departments. Associations between categorical variables were assessed using the chi-square test. Data analysis was carried out using IBM SPSS Statistics version 29.0, with exact p-values and 95% confidence intervals calculated. The p-value was statistically significant (*P < 0.05)* and all percentages were reported together with their corresponding absolute numbers.

Results

A total of 755 patients were included in the final analysis. The findings were presented in sequential order, beginning with the demographic characteristics of the study population, followed by the distribution of thrombocytopenia causes. Cross tabulations were performed to explore the relationship of causes with gender, patient setting, age groups, and hospital departments. Chi-square tests were applied to assess statistical associations, and results were summarized in both descriptive and analytical tables.

As shown in Table I, the study population had a mean age of 46.3 ± 23.3 years. Females constituted (603/755; 79.9%) and the admitted patients (484/755; 64.1%).

The distribution of causes of thrombocytopenia is presented in Table II. Morphologically consistent with true thrombocytopenia accounted for most cases (70.7%). Among the 221 cases of pseudo-thrombocytopenia (29.3%), platelet clumping was the most frequent cause (19.7%), followed by giant platelets (7.9%) and abnormal platelet distribution (1.6%).

Table III. No statistically significant association was observed between thrombocytopenia cause and gender (p = 0.560). Statistically significant associations were identified with patient setting (p = 0.013), age group (p = 0.001), and department (p < 0.001).

The distribution of thrombocytopenia causes across demographic and clinical variables is shown in **Table IV**. True thrombocytopenia remained the most frequent finding across both genders and patient settings. A higher proportion of true thrombocytopenia was observed among inpatients compared with outpatients. Across age groups, true thrombocytopenia was most frequent in patients aged ≥61 years. Departmental distribution demonstrated variability in the pattern of causes, with pseudothrombocytopenia observed across multiple clinical services.

The results of chi-square analysis are presented in

Table I: Demographic characteristics of study population (n=755)

Variable	Category	n	%
Age (years)	Mean 46.3 ± 23.3; Range 1–101 years		–
Gender	Male	152	20.1
	Female	603	79.9
Setting	Outpatient	271	35.9
	Inpatient	484	64.1

n = 755

Table II: Laboratory-based distribution of causes of thrombocytopenia among the study population (n = 755)

Cause	n	%
True thrombocytopenia	534	70.73
Platelet clumps	149	19.74
Giant platelets	60	7.94
Abnormal platelet distribution	12	1.59

n = 755

Table III: Chi-square analysis of associations between thrombocytopenia causes and demographic and clinical variables (n = 755)

Variable	χ^2	Df	*p-value*
Gender	2.06	3	*0.560*
Setting	10.85	3	*0.013*
Age group	27.56	9	*0.001*
Department	132.12	57	*<0.001**

n = 755

A p-value of <0.05 was considered statistically significant.

Discussion

Several factors contribute to pseudothrombocytopenia; however, EDTA, the most commonly used anticoagulant for complete blood count analysis, remains the leading cause of spuriously low platelet counts. The EDTA dependent phenomenon is largely reversible using alternative anticoagulants such as citrate, oxalate, or heparin, which prevent antibody-mediated platelet agglutination. [10,11] In our study the most common cause of PTCP was EDTA

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Table IV: Distribution of laboratory-identified causes of thrombocytopenia across demographic and clinical variables (n = 755)

	Category	True thrombocytopenia n (%)	Platelet clumping n (%)	Giant platelets n (%)	Abnormal platelet distribution n (%)	Total (n)
Gender	Male	113 (74.34)	26 (17.11)	12 (7.89)	1 (0.66)	152
	Female	421 (69.8)	123 (20.4)	48 (8)	11 (1.8)	603
Patient setting	Outpatient	173 (63.8)	64 (23.6)	27 (10.0)	7 (2.6)	271
	Inpatient	361 (74.6)	85 (17.6)	33 (6.8)	5 (1.0)	484
Age group (years)	≤20	105 (74)	29 (20.4)	8 (5.6)	0 (0.0)	142
	21–40	82 (63.57)	21 (16.28)	22 (17.05)	4 (3.1)	129
	41–60	170 (68.55)	58 (23.39)	14 (5.64)	6 (2.42)	248
	≥61	177 (75.0)	41 (17.4)	16 (6.8)	2 (0.8)	236
Department	Medicine	245 (76.1)	63 (19.6)	11 (3.4)	3 (0.9)	322
	Pediatrics	69 (75.8)	18 (19.8)	4 (4.4)	0 (0.0)	91
	Gynecology	23 (50.0)	9 (19.6)	12 (26.1)	2 (4.3)	46
	General Surgery	21 (55.3)	14 (36.8)	2 (5.3)	1 (2.6)	38
	ICU	55 (85.9)	7 (10.9)	1 (1.6)	1 (1.6)	64
	Emergency Dept.	49 (76.56)	10 (15.63)	4 (6.25)	1 (1.56)	64
	Oncology	42 (93.3)	1 (2.2)	2 (4.4)	0 (0.0)	45
	Nephrology	7 (50)	3 (21.43)	3 (21.43)	1 (7.14)	14
	Others	23 (32.4)	28 (39.4)	20 (28.2)	0 (0.0)	71

n = 755

induced PTCP (which accounts for approximately 29% of total cases). Gowland et. al first reported PTCP induced by EDTA establishing its antibody-mediated in-vitro nature. [7]. Lixia Zhang et al reported the rate of EDTA induced PTCP to be 49.1% which is closely in accordance with our study findings. [9] Incidence of EDTA dependent PTCP is approximately between 0.07% and 0.20% in general hospital practice. [12] A recent national study conducted by Noureen A found that the incidence of EDTA induced psuedothrombocyopenia to be around 45% which is in strong agreement to our study.[13]

Kausar F reported the incidence of EDTA PTCP to be approximately 75%. This frequency of EDTA PTCP was quite high compared to our study. which may reflect differences in study population, laboratory protocols, or inclusion criteria. [14]

In our current study, the satellitism pattern of platelets resulting from platelet adherence to leukocytes in anticoagulated blood samples are also a cause of pseudo thrombocytopenia. Naureen A reported platelet satellitism to be around 09% while kausar F reported platelet satellitism to be approximately around 10.9% which are comparable to our study. [13,14]

Bizzaro N et al showed platelet satellitisms around polymorphonuclears. Suri et al observed Spurious thrombocytopenia due to platelet satellitisms around neutrophils, with platelet phagocytosis. [15] Platelet satellitism was also reported around eosinophils and rarely basophils. Seven patients had pseudo thrombocytopenia due to clumps around neutrophils which make it a total of (8%). This in vitro phenomenon involves platelets adhering to the plasma membrane of white blood cells in anticoagulated blood. In our study, the occurrence of

platelet satellitism was the lowest. It was observed in only one patient among PTCP. Tangella et al. has a similar finding of a very low occurrence of platelet satellitism, which was 0.1% for PTCP due to platelet satellitism describing its rare occurrence.[8]

Electronic counters may underestimate platelet counts in patients with large or giant platelets because they are not recognized as such due to their size (1.5-3 µm diameter) due to size-based misclassification, resulting in falsely low automated counts compared with manual estimation. [2] Consequently, manual counts often show higher values than automated counts in these cases. In our study, giant platelets were another cause of PTCP, Gogoi G et al reported giant platelets as a cause of PTCP around 11.5% [16] while Naureen A reported to be around 18% which is slightly higher[13].

Management of PTP involves all three steps including identification, confirmation and prevention. [17,18] Automated counters tend to miss psuedothrombocytopenia. [19] Due to inconsistent flagging of automated counters it is sometimes hard to identify PTCP.[20] Confirmation and prevention are two further crucial steps in the management of PTCP which when applied in routine blood complete picture reporting will ascertain accurate reporting.[21, 22]

This study was conducted at a single tertiary care center, which may limit the wide applicability of the findings. The analysis was restricted to patients with platelet counts below 100×10^9/L, and confirmatory testing using alternative anticoagulants or advanced platelet counting methods could not be performed in all cases due to limited resources; therefore, definitive resolution of pseudo thrombocytopenia was not possible in every patient. Future multicenter studies with larger sample sizes are recommended to better define the true burden of pseudo thrombocytopenia. The use of repeat sampling with non-EDTA anticoagulants, standardized peripheral smear review protocols and advanced platelet counting techniques should be incorporated in future studies to allow complete resolution and improve diagnostic accuracy.[17] Considering the diagnostic challenges and associated clinical risks of pseudothrombocytopenia, a structured evaluation of its prevalence and causative mechanisms in a tertiary care laboratory was undertaken. This study was conducted to support evidence-based laboratory algorithms that improve platelet count interpretation, prevent misdiagnosis, enhance patient safety.

Conclusion

Pseudothrombocytopenia represents a considerable proportion among patients with low platelet counts and requires systematic recognition in routine hematology practice to prevent misinterpretation and inappropriate clinical decisions. Automated analyzers may underestimate platelet counts; therefore, manual verification with peripheral blood smear examination is essential for accurate diagnosis. Accurate identification and classification of its underlying causes are essential to distinguish spurious thrombocytopenia from true thrombocytopenia. A structured laboratory approach incorporating peripheral blood smear review and appropriate verification strategies, supports reliable platelet count reporting. Standardization of diagnostic algorithms in tertiary care laboratories contributes to improved result interpretation and patient safety.

REFERENCES

1. Markewitz R, Sharifzadeh SR, Junker R, Wandinger KP. EDTA-associated pseudothrombocytopenia: definition and real-world occurrence. J Lab Med. 2023;47(3):105–14. doi:10.1515/labmed-2023-0032.
2. Scridon A. Platelets and their role in hemostasis and thrombosis—From physiology to pathophysiology and therapeutic implications. Int J Mol Sci. 2022;23:112772. doi:10.3390/ijms232112772.
3. Lee ACW. Pseudothrombocytopenia: what every clinician should know. Pediatr Neonatol. 2021;62(2):218–9. doi:10.1016/j.pedneo.2020.12.002.
4. Schuff-Werner P, Mansour J, Gropp A. Pseudo-thrombocytopenia (PTCP). A challenge in the daily laboratory routine? J Lab Med. 2020;44:295–304. doi:10.1515/labmed-2020-0099.
5. Lardinois B, Favresse J, Chatelain B, Lippi G, Mullier F. Pseudothrombocytopenia—a review on causes, occurrence and clinical implications. J Clin Med. 2021;10:594. doi:10.3390/jcm10040594.
6. Nagler M, Keller P, Siegrist D, Alberio L. A case of EDTA-dependent pseudothrombocytopenia: simple recognition of an underdiagnosed and misleading phenomenon. BMC Clin Pathol. 2014;14:19. doi:10.1186/1472-6890-14-19.
7. Amir E, Latif AQ, Anwar M, Rahat N, Khalid G, Anwar R. EDTA-related pseudothrombocytopenia (PTCP): incidence and resolution of clinically threatened artifact. Pak J Med Health Sci. 2023;17(4):166–9. doi:10.53350/pjmhs2023174166.
8. Tangella AV, Peta RK, Yadlapalli DC, Raghunadha Rao D, M

MS. Ethylene diamine tetra acetate-induced pseudo thrombocytopenia (EDTA-PTCP) in an adolescent: a case report. Cureus. 2023;15(5):e38545. doi:10.7759/cureus.38545.
9. Zhang L, Xu J, Gao L, Pan S. Spurious thrombocytopenia in automated platelet count. Lab Med. 2018;49(2):130–3. doi:10.1093/labmed/lmx081.
10. Tan GC, Stalling M, Dennis G, Nunez M, Kahwash SB. Pseudothrombocytopenia due to platelet clumping: a case report and brief review of the literature. Case Rep Hematol. 2016;2016:3036476. doi:10.1155/2016/3036476.
11. Bahri R, Aznag MA, Khayati S, Elfarssani F, Eddyb S, Yahyaoui H, et al. Pseudothrombocytopenia: automate-blood smear confrontation. Int J Recent Innov Med Clin Res. 2021;(1):21–9. Available from: https://ijrimcr.com/.
12. Isik A, Balcik OS, Akdeniz D, Cipil H, Uysal S, Kosar A. Relationship between some clinical situations, autoantibodies, and pseudothrombocytopenia. Clin Appl Thromb Hemost. 2012;18(6):645–9. doi:10.1177/1076029611434525.
13. Noureen A, Ali S, Faisal A, Zaman A, Shanker B, Aslam F. Pseudothrombocytopenia due to platelet clumping: a case report and brief review of the literature. Case Rep Hematol. 2022;12(2):149–54. Available from: https://jmmc.mmc.edu.pk/index.php/JMMC/article/view/230
14. Kausar S, Fateen T, Chaudary HT. Frequency of causes of spurious platelet count on routine complete blood count by an automated hematology cell analyser. LMRJ. 2021;3. doi:10.38106/LMRJ.2021.3.
15. Suri M. An isolated case of pseudothrombocytopenia due to platelet satellitism with review of literature. Ann Med Sci Res. 2024;3(1):62–5. doi:10.4103/amsr.amsr_29_23.
16. Gogoi G. The grey zone of thrombocytopenia: accuracy of automated analyser vs manual method. Int Clin Pathol J. 2018;6(2). doi:10.15406/icpjl.2018.06.00163.
17. Pujol-Moix N, Muñiz-Díaz E, Español I, et al. Pseudothrombocytopenia, beyond a laboratory phenomenon: study of 192 cases. Ann Hematol. 2023;102:1363–74. doi:10.1007/s00277-023-05192-8.
18. Santosh T, Patro MK. Ethylenediaminetetraacetic acid-induced pseudothrombocytopenia: The story of platelet clumps and report of three cases. J Hematol Allied Sci. 2023;3:143-7. doi: 10.25259/JHAS_23_2023.
19. Goyal, Snigdha; Goyal, Sunder. EDTA dependent pseudo thrombocytopenia- A diagnostic dilemma!. Indian J Path Microbiol,2023;66(1):227-8. doi 10.4103/IJPM.IJPM_1015_21
20. Cattaneo M. Pseudothrombocytopenia and other conditions associated with spuriously low platelet counts. Haematologica. 2025;;110(8):1677-92. doi: 10.3324/haematol.2024.286234.
21. Balcázar-Villarroel M, Carmine F, Torrens F, Birditt K, Sandoval C. First Report of Ethylenediaminetetraacetic Acid-Dependent Pseudo-Thrombocytopenia in Chile: Prevalence and Laboratory Insights. Diagnostics. 2025; 15(8):1050. https://doi.org/10.3390/diagnostics 15081050.
22. Ashworth I,Thielemans L, Chevassut, T. Thrombocytopenia: The good, the bad and the ugly. Clin. Med. 2022;22(3): 214–17. doi: 10.7861/clinmed.2022-0146.

CONFLICT OF INTEREST

Authors declared no conflicts of Interest.

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DATA SHARING STATEMENT

The data that support the findings of this study are available from the corresponding author upon request.