Open Access

Original Article

# Significance of Immature Platelet Fraction in Diagnosed Patients of the Immune Thrombocytopenic Purpura

**Mohammad Shabih Haider, Asad Mahmood*, Manzar Bozdar, Saima Zahir, Saadia Muzafar**, Maliha Batool Bukhari*****

Armed Forces Institute of Pathology/National University of Medical Sciences (NUMS) Rawalpindi Pakistan, *Armed Forces Institute of Transfusion/National University of Medical Sciences (NUMS) Rawalpindi Pakistan, **Watim Medical College, Rawalpindi Pakistan, ***Polyclinic Hospital Islamabad, Pakistan.

## ABSTRACT

***Objective:*** Determination of significance of immature platelet fraction (IPF%) in diagnosed patients of Immune thrombocytopenic purpura (ITP).

***Study Design:*** It was a cross-sectional study.

***Place and Study Duration:*** Haematology department, Armed forces Institute of Pathology (AFIP), Rawalpindi Pakistan June 2021 to December 2021.

***Methodology:*** Patients of all ages and genders diagnosed with ITP and platelets less than 100 x $10^9$/L after a review of peripheral blood film were included in the study. Detailed history and examination were made, complete blood count (CBC) test was done on Sysmex XN-3000 automated haematology analyzer. Peripheral blood was stained with Leishman stain and examined under a microscope to rule out pseudo thrombocytopenia and the presence of abnormal cells and dysplasia. After adequate quality and control, the immature platelet fraction was performed on Sysmex XN 3000.

***Results:*** Seventy-two diagnosed cases of immune thrombocytopenic purpura were part of the study. The mean age of the patients was 21.00±14.50 years. 34 (48%) patients were males, and 38 (52%) were females. The mean platelet count was 49.40±24.60. Mean IPF was 17.90±9.50 per microliter. Immature platelet fraction (IPF%) was raised in all the patients with immune thrombocytopenic purpura, confirming our hypothesis that IPF% was an independent predictor for the detection of ITP.

***Conclusion:*** Immature platelet fraction is a unique parameter that can pick patients having thrombocytopenia due to peripheral destruction, including immune thrombocytopenic purpura.

***Keywords:*** Immature platelet fraction, Immune thrombocytopenic purpura, Peripheral destruction of platelets.

***How to Cite This Article:*** *Haider MS, Mahmood A, Bozdar M, Zahir S, Muzafar S, Bukhari MB. Significance of Immature Platelet Fraction (IPF%) in Diagnosed Patients of the Immune Thrombocytopenic Purpura (ITP). Pak Armed Forces Med J 2022; 72(4): 1420-1423.*

*DOI: https://doi.org/10.51253/pafmj.v72i4.8376*



## INTRODUCTION

Immune thrombocytopenic purpura, also known as ITP, is a common autoimmune disorder in which there is isolated thrombocytopenia when other exogenous etiological factors are lacking.[1] Thrombocytopenia is due to autoantibody binding to specific platelet membrane glycoprotein and causing premature destruction of platelets.[2]

Diagnosis of ITP is made upon history, clinical examination, Blood CP, and peripheral blood film examination. Bone marrow biopsy is often done to look for other causes of thrombocytopenia. Bone marrow in ITP is usually normocellular with increased megakaryocytes. Bone marrow being an invasive procedure is often not done if a clinical diagnosis is established.[3]

With advances in the medical field, a new parameter has been introduced which can provide useful information about the peripheral destruction of platelets.[4] Immature Platelet Fraction (IPF%) is an automated parameter that tells us about the proportion of immature platelets to the total number of platelets in a patient's peripheral blood.[5,6] The Sysmex® XN- and XE-Series of Automated Hematology Analyzers can report the IPF% result by flowcytometry to help physicians determine the cause of thrombocytopenia.[7] IPF reference range varies from 1.6 - 7.1% in adults and 1.0-6.8% in children.[8]

Immature platelet fraction is raised in patients with peripheral destruction of platelets, whereas it is normal or decreased in cases of central thrombocytopenia.[9,10] This test is a noninvasive, cost-effective and reliable test to diagnose the cases of peripheral destruction of platelets along with the history, examination, Blood CP, and peripheral blood film review. This study was conducted to determine the significance of IPF % as a predictive marker for the diagnosis of peripheral destruction of platelets, including Immune Thrombocytopenic Purpura (ITP).

**Correspondence: Dr Muhammad Shabih Haider,** Department of Haematology, AFIP, Rawalpindi, Pakistan

*Received:12 Mar 2022; revision received: 08 May 2022; accepted: 11 May 2022*

## METHODOLOGY

This cross-sectional study was conducted at the Armed Forces Institute of Pathology from June 2021 to December 2021, over seven months after permission from the Institutional Review Board was taken (IRB no FC-HEM 20-21/READ-IRB/22/840). Keeping the confidence level at 95%, the margin of error at 5% and taking the population proportion to 95% based on a study conducted on NIBD,[11] in which the majority of patients with ITP had raised IPF. The WHO calculator was used to calculate the sample size of 73. Non-probability purposive sampling was employed.

**Inclusion criterion:** Patients in the study were of all ages and genders diagnosed with immune thrombocytopenic purpura based on clinical features and bone marrow biopsy in some cases, along with platelets less than 100 x $10^9$/L, confirmed and reviewed after the peripheral examination.

**Exclusion criteria:** Patients with pseudo thrombocytopenia after peripheral blood film review or presence of blast cell or dysplasia in the peripheral blood and patients who have already started treatment for their haematological disorders were exdcluded from the study.

Informed written consent was ensured. History and examination were made. In history, dental history, spontaneous bruising, bleeding after surgery, menstrual history and epistaxis history were taken. In physical examination, petechial bruises, ecchymosis, and hematoma were looked.

5ml of venous blood was drawn under aseptic measures and kept in an EDTA vial. After adequate quality control, a blood CP card was obtained from XN 3000 fully automated analyser. Slides were stained with Leishman stain, and peripheral blood examination was done under a microscope to rule out pseudo thrombocytopenia, dysplasia and the presence of blast cells. IPF command was given on XN 3000 to measure the value of the Immature platelet fraction.

Data was analysed on Statistical package for social sciences (SPPS) version 23.00. For analysis of qualitative variables, percentages and frequencies were calculated, whereas for quantitative variables, standard deviation and mean was calculated.

## RESULTS

A total of seventy-two diagnosed patients with ITP were included in the study. The mean age of patients was 21.0±14.5 years. 34 (48%) patients were males, and thirty-eight (52%) were females. The mean platelet count was 49.4±24.6 X $10^3$/ul. Mean IPF was 13.90±10.50). Details of descriptive statistics were shown in Table-I.

Mean platelet count and IPF in males and females were shown in Table-II. There was no statistical difference in Platelet count and IPF in both genders, $p$> 0.05. Mean platelet count and IPF Age wise were shown in Table-III.

**Table-I: Mean and Standard Deviation of Quantitative Vari-Ables (n=72)**

| Characteristics | Minimum | Maximum | Mean ± SD |
|---|---|---|---|
| Age | 1.0 | 57.0 | 21.0±14.5 years |
| Platelet count | 10.0 | 97.0 | 49.4±24.6 × 109/L |
| Immature Platelet Fraction | 5.7 | 56.0 | 17.9±9.5 per microliter |
| Total Leukocyte Count | 3.9 | 11.2 | 6.7±1.6 x 109 |
| Haemoglobin | 9.5 | 16.2 | 13.3±1.5 g/dl |

**Table-II: Gender wise Mean Platelet Count and Immature Platelet Fraction (IPF) levels (n=72)**

| Characteristics | Male (n=34) | Female (n=38) | $p$-value |
|---|---|---|---|
| Platelet count | 45.91±23.60 109/L | 52.63±25.47 109/L | 0.251 |
| Immature Platelet Fraction (IPF) | 18.43±10.29 per microliter | 17.60±8.99 per microliter | 0.714 |

**Table-III: Age wise Mean Platelet Count and Immature Platelet fraction (IPF) levels (n=72)**

| Characteristics | ≤ Mean Age (n=38) | >Mean Age (n=34) | $p$-value |
|---|---|---|---|
| Platelet count | 49.47±26.18 ×109/L | 49.44±3.38 ×109/L | 0.996 |
| Immature Platelet Fraction (IPF) | 17.33±9.12 per microliter | 18.74±10.06 per microliter | 0.536 |

## DISCUSSION

IPF possesses a high clinical utility in the laboratory diagnosis and management of thrombocytopenia because raised IPF levels reflect increased peripheral platelet destruction. In particular, it is utilized to diagnose immune thrombocytopenic purpura and differentiate it from bone marrow suppression or failure. In the latter, the IPF value would be low.[7]

Immune thrombocytopenic purpura (ITP) is an autoimmune disease with a drop in the platelet count in the peripheral blood. A decrease in platelet count causes bruising, gum bleeding or internal bleed.[8] ITP is an immune reaction against one's platelets.[9]

Diagnosis of ITP can be made based on clinical history and examination, but laboratory analysis is often required before commencing the treatment. In

most settings, bone marrow aspiration and trephine are done to confirm ITP.[10] Bone marrow biopsy is a procedure which can cause pain; therefore, the flow cytometric technique is now being utilized to reach the final diagnosis.[11]

In the present era, IPF can be used to differentiate the cases of ITP and central thrombocytopenia. Studies have shown that under circumstances of thrombocytopenia, the RNA content of platelets corresponds directly with megakaryopoiesis.[12,13] IPF% is a new parameter introduced on the Sysmex XN and XEseries automated analyzer expressed as IPF percent (IPF%) or absolute IPF count. IPF% is utilized as a noninvasive parameter to correlate the clinical features with thrombocytopenia for diagnosis of ITP. This test is noninvasive, simple and rapid.[12,14]

In our study population, nearly all patients with ITP had raised IPF. The rise in IPF was proportional to the degree of thrombocytopenia. Hence, we can say that IPF has the potential to replace bone marrow examination in the diagnosis of ITP. However, peripheral blood review is essential with IPF to rule out pseudo thrombocytopenia, dysplastic cells and the presence of blast cells.[15]

In a study conducted at the National Institute of Bone Marrow Diseases (NIBD) in 2016, 95% of patients had raised IPF in ITP patients, and the mean IPF % was 16.3.[16] This study is comparable to our results in which the mean IPF % is 17.9, and all patients with ITP have raised IPF levels. A similar study was conducted at King Edward Medical College Lahore, which showed raised IPF % in patients with peripheral destruction of platelets with a mean IPF % of 25.5.[17] The mean IPF in this study is more compared to the results of our study, however in congregation with our hypothesis that the IPF % is raised in ITP patients.

An international retrospective study was conducted in Korea on 568 patients with ITP. The mean IPF % was 8.7 %.[18] This value is again above the upper limit for the normal reference range of IPF. A study was carried out in Egypt on children with ITP who were tested, and the mean platelet count was found to be 55 and IPF% was 9.8%.[19] These results are also consistent with our findings of raised IPF in ITP patients.

## ACKNOWLEDGMENTS

All authors like to acknowledge the effort of laboratory staff that cooperated in obtaining the Blood CP card and helped in staining blood films.

## LIMITATION OF STUDY

The limitation of this study was that it was a single-centre study. After multiple centres become involved in this study, we can extend the results to represent the Pakistani population.

## CONCLUSION

Immature Platelet Fraction (IPF %) is a novel parameter and can be routinely used with CBC in patients with low platelet count. This unique parameter will guide the physician and help differentiate the aetiology of various causes of thrombocytopenia, including Immune thrombocytopenic purpura. This cost-effective test can be an alternative to bone marrow biopsy examination. Nevertheless, unfortunately, that is an invasive and quite painful procedure.

**Conflict of Interest:** None.

**Author's Contribution**

MSH: Collected the data, analyzed the statistical data and wrote the manuscript, AM:, MB:, SZ:, SM:,MB: Did the review and final proof reading of the manuscript.

## REFERENCES

1. Zainal A, Salama A, Alweis R. Immune thrombocytopenic purpura. J Community Hosp Intern Med Perspect 2019; 9(1): 59-61.
2. George JN, El-Harake MA, Raskob GE. Chronic idiopathic thrombocytopenic purpura. N Eng J Med 1994; 331(18): 1207-1211.
3. Cines DB, Bussel JB. How I treat idiopathic thrombocytopenic purpura (ITP). Blood 2005; 106(7): 2244-2251.
4. Benlachgar N, Doghmi K, Masrar A, Mahtat EM, Harmouche H, Mezalek ZT. Immature platelets: a review of the available evidence. Thromb Res 2020; 195(1): 43-50.
5. Ko YJ, Hur M, Kim H, Choi SG, Moon H-W, Yun Y-M. Reference interval for immature platelet fraction on Sysmex XN hematology analyzer: a comparison study with Sysmex XE-2100. Clin Chem Lab Med (CCLM) 2015; 53(7): 1091-1097.
6. Li J, Li Y, Ouyang J, Zhang F, Liang C, Ye Z, et al. Immature platelet fraction related parameters in the differential diagnosis of thrombocytopenia. Platelets 2020; 31(6): 771-776.
7. Jeon K, Kim M, Lee J, Lee J-S, Kim H-S, Kang HJ, et al. Immature platelet fraction: A useful marker for identifying the cause of thrombocytopenia and predicting platelet recovery. Medicine (Baltimore) 2020; 99(7): e19096.
8. Pietras N, Pearson-Shaver A. Immune Thrombocytopenic Purpura (ITP). StatPearls Publishing; 2022, [Internet] available at: https://www.ncbi.nlm.nih.gov/books/NBK562282/.
9. Rodeghiero F, Stasi R, Gernsheimer T, Michel M, Provan D, Arnold DM, et al. Standardization of terminology, definitions and outcome criteria in immune thrombocytopenic purpura of adults and children: report from an international working group. Blood, J Am Soc Hematol 2009; 113(11): 2386-2393.
10. Jubelirer SJ, Harpold R. The role of the bone marrow examination in the diagnosis of immune thrombocytopenic purpura: case series and literature review. Clin Appl Thromb Hemost 2002; 8(1): 73-76.
11. Goubran H, Hart C, Othman I, Seghatchian J. Flow cytometry and immune thrombocytopenic purpura. Transfus Apher Sci 2018; 57(6): 800-803.
12. Buttarello M, Mezzapelle G, Freguglia F, Plebani M. Reticulated platelets and immature platelet fraction: Clinical applications and method limitations. Int J Lab Hematol 2020; 42(4): 363-370.

13. Ali I, Graham C, Dempsey-Hibbert NC. Immature platelet fraction as a useful marker in the etiological determination of thrombocytopenia. Exp Hematol 2019; 78(1): 56-61.
14. Van-De Wyngaert Z, Fournier E, Bera E, Carrette M, Soenen V. Immature platelet fraction (IPF): A reliable tool to predict peripheral thrombocytopenia. Curr Res Trans Med 2020; 68(1): 37-42.
15. McDonnell A, Bride KL, Lim D, Paessler M, Witmer CM. Utility of the immature platelet fraction in pediatric immune thrombocytopenia: Differentiating from bone marrow failure and predicting bleeding risk. Pediatr Blood Cancer 2018; 65(2): e26812.
16. Naz A, Mukry SN, Shaikh MR, Bukhari AR, Shamsi TS. Importance of immature platelet fraction as predictor of immune thrombocytopenic purpura. Pak J Med Sci 2016; 32(3): 575.
17. Ashraf S, Rehman S, Asgher Z, Hamid A, Qamar S. Comparison of immature platelet fraction (IPF) in patients with central thrombocytopenia and peripheral thrombocytopenia. J Coll Physicians Surg Pak 2020; 30(8): 796-800.
18. Jeon MJ, Yu ES, Kang K-W, Lee B-H, Park Y. Imma-ture platelet fraction based diagnostic predictive scoring model for immune thrombocytopenia. Korean J Intern Med 2020; 35(4): 970-974.
19. Adly AAM, Ragab IA, Ismail EAR. Evaluation of the immature platelet fraction in the diagnosis and prognosis of childhood immune thrombocytopenia. Platelets 2015; 26(7): 645-650.
20. Goel G, Semwal S, Khare A, Joshi D, Amerneni CK, Pakhare A, et al. Immature Platelet Fraction: Its Clinical Utility in Thrombocytopenia Patients. J Lab Physicians 2021; 13(03): 214-218.