ORIGINAL ARTICLE

A SINGLE CENTER ANALYSIS OF PRIMARY AND SECONDARY IMMUNE THROMBOCYTOPENIC PURPURA (ITP) IN THE MALAYSIAN POPULATION.

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Abstract

Background: Immune thrombocytopenic purpura (ITP) is characterized by destruction of platelets by autoantibodies produced through immune-mediated mechanism. Various etiologies have been implicated for the development of ITP and there is geographical variation in the prevalence and incidence of this disorder. This retrospective study is to evaluate the prevalence of ITP in a tertiary referral center for hematology, and the spectrum of clinical manifestations.

Methodology: This is large cohort of ITP patients from a tertiary hematology clinic Hospital Raja Permaisuri Bainun, Ipoh, Perak, Malaysia. The study was performed in 2019 and included all patients with ITP seen in the clinic till the time this study was conducted. Patients' information were retrieved from the clinic records. Socio-demographic characteristics, platelet counts, serological markers and hematological complications were evaluated.

Results: A total of 241 patients with immune thrombocytopenic purpura (ITP) were included. The mean age was 46.37 ± 18.19 years. Higher prevalence of ITP was seen in Malays (44.4%) followed by Chinese (31%) and Indian (21%) with female to male ratio of 3:1. The prevalence of primary ITP (pITP) was substantially higher (69.7%) than secondary ITP (30.3%). ANA was negative in 61% and positive in 39%. SLE was diagnosed in 22% of patients, the majority (81%) of whom did not have any haemorrhagic manifestations. Only 7.9% of these patients had a positive anti- dsDNA antibody test. Mild haemorrhagic manifestations (grade1) was seen 53.9% and the average platelet count was 69.0 +- 73.35 x103/l.

Conclusion: Our study showed that primary ITP was more prevalent than secondary ITP. Anti-nuclear antibody negative (ANA) associated ITP accounted for the majority of cases. Hemorrhagic manifestations occur in almost half of the studied patients.

Keywords: ITP, primary, secondary, ANA, hemorrhagic manifestations.

Introduction

Immune thrombocytopenia (ITP) is an acquired immune mediated disorder, with destruction of platelet due to platelet autoantibodies and inhibition of their production. ITP is defined as a peripheral blood platelet (PLT) count less than $100x10^9$/L, most commonly idiopathic or primary immune thrombocytopenia (pITP).[1, 2] Several etiologies have been implicated in secondary immune thrombocytopenia (sITP) such as autoimmune conditions e.g. systemic lupus erythematosus (SLE), infectious disease e.g. human immunodeficiency virus (HIV), hepatitis C, drug-induced, etc.[3] The different spectrum of pathogenesis has been suggested for the diversity of clinical characteristics of ITP as patients did not meet the recommended criteria of any specific autoimmune disease.[4] This retrospective study was undertaken to analyze the clinical manifestations of both primary and secondary ITP in the Haematology clinic, Hospital Raja Permaisuri Bainun Ipoh, Malaysia.

Materials and Methods

This is a descriptive study conducted in March 2019, involving all patient diagnosed with ITP (in accordance with the American society of Hematology 2011 evidence-based practice guideline for immune thrombocytopenia [1, 5]) at the haematology clinic, Hospital Raja Permaisuri Bainun Ipoh, Malaysia. Patient's clinic records were reviewed and demographic information and laboratory results were obtained. Thrombocytopenia due to drugs such as anti-platelet and anti-thrombotic agents (aspirin, clopidogrel, and heparin), viral fever and malignancy were excluded. Thrombocytopenia was defined as having a platelet count of less than $100x10^9$/l. [2]

sITP was considered in patients with systemic lupus erythematosus (SLE) who fulfilled the American College Rheumatology Classification Revised Criteria for SLE 1982 and the Systemic Lupus International Collaboration Clinic classification criteria 2012.[6, 7] Secondary thrombocytopenia due to other autoimmune disorders includes mixed connective tissue disease, rheumatoid arthritis, HIV, hepatitis B and C and drug-induced were also evaluated. The presence of immunological markers anti-nuclear antibody (ANA) and anti-double stranded deoxyribonucleic antibody (dsDNA)) was also noted.

Severity of hemorrhagic manifestations in patients with thrombocytopenia was based on the criteria by Ziakas et al and the Massachusetts Medical Center. [8, 9] That is grade 0, no bleeding; grade 1, mucocutaneous hemorrhage; grade 2: obvious hemorrhage such as bleeding in the nose, urinary, and genital system or digestive system; grade 3: life-threatening intracranial hemorrhage and hemorrhage in the respiratory system.

Ethical Issues

Ethical clearance was obtained from Universiti Kuala Lumpur Royal College of Medicine Perak, Clinical Research Centre (CRC) Hospital Raja Permaisuri Bainun and Malaysian Research Ethic Committee (MREC).

Statistical analysis

The data was analyzed by using SPSS version 23.0. Descriptive statistical analysis was used to determine the significance of difference for the different categorical variables. Non-parametric test was performed for variables which were not normally distributed. Binary logistic regression was performed to determine the odd ratio (OR) to measure the association between gender and ethnicity with development of ITP and grades of hemorrhagic category. $P < 0.05$ was taken as statistically significant.

Results

Total of 241 ITP patients medical records were evaluated in this retrospective study. 55 (22.8%) were males and 184 (77.2%) were females

(female to male ratio 3: 1) with mean age of 46.37 ± 18.19 years. There was no significant different in prevalence of ITP in the 3 major ethnic groups although Malays was the majority (44.4%). 224(92.9%) patients had the disease for more than 2 years at the time of data collection. Bone marrow with trephine biopsy was performed in 45.6% of patients. The mean platelet count was $69.01 \pm 75.35 \times 10^3/\mu L$ with significant difference between primary and secondary ITP ($p < 0.02$).

ANA was negative in 61% and positive in 39% of these patients. ANA negative was more prevalent in pITP whilst ANA positive was substantially higher in secondary ITP. Anti-dsDNA was positive in only 3.7% 0f patients. This may have been due to the laboratory criteria of performing this test only if the ANA showed a homogenous pattern. Systemic lupus erythematosus (SLE) was the diagnosis in 22% of the patients (Table 1), 49 % of whom had s platelet count of less than $100x10^3/\mu L$ (Odd ratio [OR] = 1.734, 95% confidence interval [CI]: 1.314-2.289, P <0.05). Majority (81%) of SLE patients did not have any hemorrhagic manifestation (grade 0).

There was a significant lower occurrence of SLE among those with haemorrhagic manifestation grade I and II (OR= 0.112, 95% CI 0.038 - 0.331, P <0.001 and OR = 0.106, 95% CI 0.031- 0.363, P <0.001 respectively), but this was not the case with grade III where there was no statistical difference (OR= 0.349, 95% CI 0.095- 1.284, P >0.05) (Table 4).

Comparing different haemorrhagic severity, risk predictors for haemorrhagic manifestations among SLE patient showed that the risk was significantly higher in all categories of bleeding tendencies (OR=7.249, 95% CI 3.428-15.329, P < 0.01), mucosal bleeding (OR= 5.465, 95% CI 1.884-15.857, P < 0.01), and GIT bleeding (OR= 5.399, 95% CI 1.607-18.134, P < 0.01). However, There was significantly lower risk of having no bleeding (OR= 0.138, 95% CI 0.065 to 0.292, P < 0.01), and there was no significant difference in risk of having CNS or pulmonary bleeding among those with SLE (OR= 1.341, 95% CI 0.371-4.852, P > 0.05). (Table 4).

Hemorrhagic manifestations of grade I to III of severity more common in pITP (44.8%). However, 46.9% patients from both pITP and secondary ITP were asymptomatic. Patient developed hemorrhagic symptom grade I to III with platelet count of more than $41 \times 10^3/\mu L$ (Table 3).

Males and females had a significantly lower risk and higher risk of developing secondary ITP (P <0.05) respectively. As compared to Malay, Indians had a significantly higher risk to have primary ITP (P <0.05) and a significantly lower risk to have secondary ITP (P <0.05). Grade I and II haemorrhagic severity were significantly higher among primary ITP patients (P <0.05) and lower among secondary ITP patients (P <0.05). (Table 4)

81.3% patients were on oral prednisolone and most of them in combination with immunosuppressant. 2.5% of patients were given anti-CD20 antibody (rituximab). Only 1.2% of ITP patients underwent splenectomy (Table 5). There was no significant difference in treatment modalities between both primary and secondary ITPs.

Discussion

The demographic characteristics of ITP is distinctively shown in Asian regions compared to Western. Such manifestations is unique in Malaysia since it affects various major ethnic groups with difference in genetic background that would contribute to the disparity. Globally, ITP predominantly affects the female, with female to male ratio of 1.7 [10, 11] comparable to our study. Previous studies had shown that ITP is commonly found in third decade of life which is illustrated in this study.

The present study demonstrate higher prevalence of pITP (69.7%) relatively comparable with

Jordanian, United States and France studies [12 -16] but contrary with previous reports among Asian population. [4, 17] Majority of sITP has various underlying etiologies. However, in our cohort, the number of secondary causes was negligible being hepatitis C, HIV and drug-induced among the identified etiologies. Nevertheless, SLE, although the number was small in this study, showed a significant association with ITP for which thrombocytopenia as part of the clinical manifestation comparable to study done by Fayyaz *et al.* [18] SLE was diagnosed based more on clinical ground and not solely rely on immunological markers. In our laboratory setting, dsDNA test performed only when the ANA is in homogenous pattern. Thus, the results in this study is self-explanatory. ANA may not be detected at the onset of the diagnosis of ITP and found positive in 25 – 26% of ITP patients [19, 20] in contrast with present study. Nonetheless, ANA showed positivity regardless the type of ITPs.

Previous study had shown that mucosal bleeding is the commonest manifestation associated with platelets less than $30x10^3$ /µL. [21] In contrast, this current study, mucosal bleeding occurred with mean platelets of 41.51 ± 38.77 $x10^3$ µ/L. Nevertheless, statistically, the hemorrhagic events are comparable with the Intercontinental Cooperative Immune Thrombocytopenia Study Group [22] but lower than that of Pakistan study. [17]

Hemorrhagic manifestations often seen frequently in elderly patients with ITP above 60 years with platelets less than $30x10^3$ µ/L. [14] In contrary, our study showed substantially higher occurrence of hemorrhagic events with higher platelets count more than 55 x 10^3/µL among patients more than 60 years of age.

As shown in this study that SLE patients are at higher risk of bleeding tendencies with severe thrombocytopenia. This findings had been observed as a significant adverse prognostic factor of SLE in a study by Jie li *et al* and Jung JH *et al.* [23, 24]

Our study had several limitations and based on retrospective data collection. There were missing data or not available which may contribute to discrepancies in the analysis. Certain laboratory tests were not able to performed in particular etiology which may contribute to sITP such as antiphospholipid syndrome, hypothyroidism etc.

Conclusion

Our study shows that pITP has a higher prevalence in this region which is comparable with other western studies. SLE and ANA-positive ITP are the common causes of sITP although the numbers were small. Minor bleeding tendencies (grade I and II) occurred in both primary and secondary ITP. This study may serve as a platform for future clinical research which should include the spectrum of clinical manifestations and etiologies as well as genetics, treatment modalities and assessment of the prognosis.

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Nil

Conflict of Interest

There are no conflicts of interest.

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Table 1. Socio-demographic, and clinical characteristic of [£]ITP patients.

	Primary ITP n = 168 n(%)	Secondary ITP n = 73 n(%)	P value
Gender			
Male	50 (90.9%)	5 (9.1%)	<0.001
Female	118 (63.4%)	68 (36.6%)	
Ethnicity			
Malay	67 (62.6%)	40 (37.4%)	<0.05
Chinese	56 (74.7%)	19 (25.3%)	
Indian	40 (78.4%)	11 (21.6%)	
Punjabi	0 (0%)	2 (100%)	
Others	5 (83.3%)	1 (16.7%)	
Mean Age ± SD years	49.02 ± 18.57	40.33 ± 15.81	<0.001
Duration of disease (years)			
< 2 years	17 (7.1)		
>2 years	224 (92.9)		
Bone marrow performed	110 (45.6)		
Mean Platelet count ± SD x10^3/µL (not normally distributed in ITP categories)	57.78 ± 64.58	95.22 ± 91.14	<0.02 (Non-parametric test)
[β]Hemorrhagic severity grades:			
0	60 (53.1%)	53(46.9%)	<0.001
I	55 (88.7%)	7 (11.3%)	
II	41(83.7%)	8 (16.3%)	
III	12 (70.6%)	5 (29.4%)	
Immunological marker			
*ANA			
- Positive	39 (41.5%)	55(58.5%)	<0.001
- Negative	129 (87.8%)	18 (12.2%)	
[Σ]Anti ds-DNA			
-Positive	2 (22.2%)	7 (77.8%)	<0.001
-Negative	22 (73.3%)	8 (26.7%)	
Underlying [α]SLE		53 (22.0)	

[£]ITP = Immune Thrombocytopenic Purpura; *ANA = Anti-Nuclear Antibody; [Σ]Anti ds-DNA = double stranded DNA, [α]SLE = Systemic Lupus Erythematosus; [β]Haemorrhagic severity grades: Grades 0 = none; I = mucocutaneous bleeding; II = gastrointestinal / genitourinary tract bleeding; III = central nervous system/pulmonary haemorrhage.

Table 2. Haemorrhagic manifestations by age groups.

Age (year)	Platelet count (mean ± SD) $x10^3/\mu L$	Haemorrhagic manifestation severity, n(%)			
		0	I	II	III
13-20	64.2 ± 68.9	5 (2.1)	4 (1.7)	0 (0.0)	1 (0.4)
21-30	75.1 ± 88.9	22 (9.2)	12 (5.0)	9 (3.8)	1 (0.4)
31-40	82.5 ± 93.7	25(10.4)	12 (5.0)	10 (4.2)	3 (1.3)
41-50	76.6 ± 91.2	16 (6.7)	12 (5.0)	6 (2.5)	4 (1.7)
51-60	60.2 ± 47.4	17 (7.1)	5 (2.1)	9 (3.8)	2 (0.8)
>60	55.5 ± 47.2	28 (11.7)	15 (6.3)	15 (6.3)	6 (2.5)

Table 3. Haemorrhagic manifestation of ITP in relation to platelet count.

Grade of severity	Platelet count (mean ± SD) $x10^3/uL$	n(%)	P (Between group) using ANOVA
0	90.55 ± 88.07	113(46.9)	
I	41.51 ± 38.77	62(25.7)	<0.001
II	63.82 ± 72.70	49(20.3)	
III	42.35 ± 48.95	17(7.1)	

Table 4. Odd Ratio (OR) of risk predictors for Primary ITP (pITP) or Secondary ITP (sITP) and risk of SLE developing haemorrhagic manifestations.

		Binary logistic regression	
		OR (95% CI)	P
Gender	Male as reference: Female - to have sITP	5.763 (2.192 - 15.149)	<0.05
	Female as reference: Male - to have sITP	0.174 (0.066 - 0.456)	<0.05
Ethnicity	Malay as reference: Chinese - to have pITP	1.760 (0.917 - 3.375)	>0.05
	Indian - to have y pITP	2.171 (1.001 - 4.706)	<0.05
	Malay as reference Chinese - to have sITP	0.568 (0.296 - 1.090)	>0.05
	Indian - to have sITP	0.461 (0.212 - 0.999)	<0.05
Haemorraghic severity grades	0 as reference: I - to be associated with sITP	0.144 (0.060 - 0.344)	<0.05
	II - to be associated with sITP	0.221 (0.095 - 0.513)	<0.05
	III- to be associated with sITP	0.472 (0.156 - 1.427)	>0.05
	0 as reference I - to be associated with pITP	6.940 (2.911 - 16.549)	<0.05
	II - to be associated with pITP	4.527 (1.949 - 10.516)	<0.05
	III to be associated with pITP	2.120 (0.701 - 6.412)	>0.05
SLE	0 as reference: I	0.122 (0.038 – 0.331)	<0.001
	II	0.106 (0.031 – 0.363)	<0.001
	III	0.349 (0.095 – 1.284)	>0.05
	I,II,III as reference: 0	0.138 (0.065 – 0.292)	<0.01
	0 as reference: I,II,III	7.249 (3.428 – 15.329)	<0.01
	0,II,III as reference: I	5.465 (1.884 – 15.857)	<0.01
	0,I,III as reference: II	5.399 (1.607 – 18.134)	<0.01
	0,I,II as reference: III	1.341 (0.371 – 4.852)	>0.05

Table 5. Treatment modalities in ITP patients.

Treatment	n(%)
Hydroxychloroquine	42 (17.4)
Azathioprine	55 (22.8)
Methotrexate	2 (0.8)
Salazopyrine	1 (0.4)
Cyclosporine	1 (0.4)
Cyclosphosphamide	1 (0.4)
Mycophenolate mofetil	1 (0.4)
Corticosteroid	196 (81.3)
Rituximab	6 (2.5)
Splenectomy	3 (1.2)

References

1. Neunert C, Lim W, Crowther M, Cohen A, Solberg L Jr, Crowther MA; American Society of Hematology. The American Society of Hematology 2011 evidence-based practice guideline for immune thrombocytopenia. Blood. 2011; 117(16):4190-207. doi: 10.1182/blood-2010-08-302984.
2. Provan D, Stasi R, Newland AC, Blanchette VS, Bolton-Maggs P, Bussel JB, et al. International consensus report on the investigation and management of primary immune thrombocytopenia. Blood. 2010; 115(2):168-86. doi: 10.1182/blood-2009-06-225565.
3. Frederiksen H, Schmidt K. The incidence of idiopathic thrombocytopenic purpura in adults increases with age. *Blood* 1999; 94(3): 909-13.
4. Luo J, Qian H, Duan L, Shi G. Clinical characteristics of immune thrombocytopenia associated with autoimmune disease: A retrospective study. Medicine (Baltimore). 2016; 95(50):e5565. doi: 10.1097/MD.0000000000005565.
5. Lo E, Deane S. Diagnosis and classification of immune-mediated thrombocytopenia. Autoimmunity Reviews. 2014; 13(4-5):577–83.
6. Tan EM, Cohen AS, Fries JF, Masi AT, McShane DJ, Rothfield NF, et al. The 1982 revised criteria for the classification of systemic lupus erythematosus. Arthritis Rheum. 1982; 25(11):1271-7. doi: 10.1002/art.1780251101.

7. Petri M, Orbai AM, Alarcón GS, Gordon C, Merrill JT, Fortin PR, et al. Derivation and validation of the Systemic Lupus International Collaborating Clinics classification criteria for systemic lupus erythematosus. Arthritis Rheum. 2012; 64(8):2677-86. doi: 10.1002/art.34473.
8. Ziakas PD, Giannouli S, Zintzaras E, Tzioufas AG, Voulgarelis M. Lupus thrombocytopenia: clinical implications and prognostic significance. Ann Rheum Dis. 2005; 64(9):1366-9. doi: 10.1136/ard.2004.033100.
9. Friedmann AM, Sengul H, Lehmann H, Schwartz C, Goodman S. Do basic laboratory tests or clinical observations predict bleeding in thrombocytopenic oncology patients? A reevaluation of phrophylactic platelet transfusion. Transfusion medicine reviews. 2002; 16(1):34-45. doi.org/10.1053/tmrv.2002.29403.
10. Farid J, Gul N, Qureshi WU, Idris M. Clinical presentations in immune thrombocytopenic purpura. J Ayub Med Coll Abbottabad 2012; 24(2): 3940.
11. Kistangari G, McCrae KR. Immune thrombocytopenia. Hematol Oncol Clin North Am 2013; 27(3): 495-520.
12. Cines DB, Bussel JB, Liebman HA, Luning Prak ET. The ITP syndrome: pathogenic and clinical diversity. Blood. 2009; 113(26):6511-21. doi: 10.1182/blood-2009-01-129155.
13. Cirasino L, Robino AM, Cattaneo M, Pioltelli PE, Pogliani EM, Morra E, et al. Reviewed diagnosis of primary and secondary immune thrombocytopenic purpura in 79 adult patients hospitalized in 2000-2002. Blood Coagul Fibrinolysis. 2011; 22(1):1-6. doi: 10.1097/MBC.0b013e3283401286.
14. Ayesh MH, Alawneh K, Khassawneh B, Khader Y, Kasasbeh A. Adult primary and secondary immune thrombocytopenic purpura: a comparative analysis of characteristics and clinical course. Clin Appl Thromb Hemost. 2013; 19(3):327-30. doi: 10.1177/1076029611433641
15. Moulis G, Palmaro A, Montastruc JL, Godeau B, Lapeyre-Mestre M, Sailler L. Epidemiology of incident immune thrombocytopenia: a nationwide population-based study in France. Blood. 2014; 124(22):3308-15. doi: 10.1182/blood-2014-05-578336.
16. Lambert MP, Gernsheimer TB. Clinical updates in adult immune thrombocytopenia. Blood. 2017; 129(21):2829–35.

17. Sultan S, Ahmed SI, Murad S, Irfan SM. Primary versus secondary immune thrombocytopenia in adults; a comparative analysis of clinical and laboratory attributes in newly diagnosed patients in Southern Pakistan. Med J Malaysia. 2016; 71(5):269-274.
18. Fayyaz A, Igoe A, Kurien BT, Danda D, James JA, Stafford HA, Scofield RH. Haematological manifestations of lupus. Lupus Sci Med. 2015 Mar 3;2(1):e000078. doi: 10.1136/lupus-2014-000078.
19. Cheng YS, Kuang LP, Zhuang CL, Jiang JD, Shi M. Effects of cytotoxin-associated gene A (CagA) positive Helicobacter pylori infection on anti-platelet glycoprotein antibody producing B cells in patients with primary idiopathic thrombocytopenic purpura (ITP). Pak J Med Sci. 2015; 31(1):121-6. doi: 10.12669/pjms.311.6409.
20. Grimaldi-Bensouda L, Nordon C, Michel M, Viallard JF, Adoue D, Magy-Bertrand N, et al.; Group for the PGRx-ITP Study. Immune thrombocytopenia in adults: a prospective cohort study of clinical features and predictors of outcome. Haematologica. 2016; 101(9):1039-45. doi: 10.3324/haematol.2016.146373.
21. Fogarty PF. Chronic immune thrombocytopenia in adults: epidemiology and clinical presentation. Hematol Oncol Clin North Am. 2009; 23(6):1213-21. doi: 10.1016/j.hoc.2009.08.004.
22. Kühne T, Berchtold W, Michaels LA, Wu R, Donato H, Espina B, et al.; Intercontinental Cooperative ITP Study Group. Newly diagnosed immune thrombocytopenia in children and adults: a comparative prospective observational registry of the Intercontinental Cooperative Immune Thrombocytopenia Study Group. Haematologica. 2011; 96(12):1831-7. doi: 10.3324/haematol.2011.050799.
23. Li J, Pan Z, Liu H, Ding F, Shu Q, Li X. Retrospective analysis of the risk of hemorrhage associated with moderate and severe thrombocytopenia of 173 patients with systemic lupus erythematosus. Medicine (Baltimore). 2018; 97(27):e11356. doi: 10.1097/MD.0000000000011356.
24. Jung JH, Soh MS, Ahn YH, Um YJ, Jung JY, Suh CH, et al. Thrombocytopenia in Systemic Lupus Erythematosus: Clinical Manifestations, Treatment, and Prognosis in 230 Patients. Medicine (Baltimore). 2016; 95(6):e2818. doi: 10.1097/MD.0000000000002818.