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Original Article

Clinical spectrum of thrombocytopenia - A hospital based prospective study

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Keywords:

COVID-19;
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Thrombocytopenia

ABSTRACT

Background: Platelets play a vital role in coagulation. The normal platelet count ranges from 280+/-130 x 10^9/L. A reduction in platelet count below 150x10^9/L is defined as thrombocytopenia. The present study was carried out to evaluate the clinico-hematological profile of various patients with thrombocytopenia and to study age and gender-wise distribution, various etiological factors, severity of thrombocytopenia and its association with bleeding manifestations in patients.

Materials and Methods: This prospective study was conducted at the hematology section of the department of Pathology at a tertiary care centre, Ahmedabad, Gujarat, India for a period of two years including 300 patients. EDTA samples were analyzed in5-part automated hematology analyzer. Thrombocytopenia cases were confirmed by peripheral blood smear examination.

Results: Thrombocytopenia was predominant in males (n=178, 59.3%). Most common age group was 20-29 years (n=81, 27%). Major clinical presentation was fever (n= 222, 74%) while bleeding manifestations were observed in only n=25, 8.3% patients. Association of thrombocytopenia with anemia was n=204, (68%) followed by leucopenia (n= 133, 44.3%) and pancytopenia (n=104, 34.6%). Bone marrow examination was done only in 40 (13.3%) patients. The majority had moderate thrombocytopenia (n=124, 41.3%) followed by mild (n=108, 36%). The most common etiology observed was COVID-19 (21%) followed by dengue (17%) and malaria (12.7%).

Conclusions: Thrombocytopenia is a very commonly observed hematological entity. The clinical spectrum of thrombocytopenia is quite varied and ranges from infections to malignancies.

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INTRODUCTION

Blood platelets are essential for maintaining the integrity of blood vessel walls and stopping bleeding at the site of injury. Platelet counts typically range from 280+/-130 x 10^9/L.[1] The lower limit of normal, as determined by the third US National Health and Nutrition Examination Survey (NHANES III), is 150 x 10^9/L, and a decrease in platelet count below that level is referred to as thrombocytopenia.[2] Although it is a diagnosis that is frequently made in clinical practice, it is not a disease condition in and of itself.[3] The processes involved in the occurrence of thrombocytopenia are decreased bone marrow production of platelets, splenic

sequestration, increased platelet destruction, etc. Reviewing the peripheral blood smear (PBS) is a crucial step in the evaluation of patients with thrombocytopenia to rule out pseudothrombocytopenia, which can be brought on by antibodies against ethylene diamine tetraacetic acid (EDTA), which can result in platelet clumps, particularly in a patient without an apparent cause for the thrombocytopenia.[4]

Clinical manifestations range from asymptomatic to mucocutaneous bleeding, simple bruises, purpura, petechiae, nasal bleeding, gum bleeds, and life-threatening bleeding in the central nervous system or gastrointestinal (GI) and genitourinary (GU) tracts. The relevance of thrombocytopenia varies from patient to patient and their clinical presentation. Although clinically significant spontaneous bleeding does not usually occur until severe thrombocytopenia, it can aggravate surgical or traumatic bleeding or prevent the administration of certain treatments (e.g., cancer chemotherapy). Sometimes, a low platelet count is the only initial manifestation of an underlying disorder, e.g., myelodysplastic syndromes (MDS), or is an important marker of disease activity (e.g., thrombotic microangiopathies). Hence, establishing the cause of thrombocytopenia, although quite challenging, is crucial.[5] An approach to diagnosing cause of thrombocytopenia is an integration of patient history, family history, the clinical profile of thrombocytopenia, bleeding manifestations (new onset, chronic, or relapsing), pregnancy status, drug history, travel history, recent transfusions, organomegaly, etc., and relevant laboratory investigations like complete blood count (CBC), examination of PBS with assessment of all three blood cell lineages, preliminary laboratory investigations like liver function tests (LFT) and renal function tests (RFT), coagulation profile, etc., as well as second-line investigations based on clinical findings and PBS. There is no single hematologic or biochemical test that is conclusive for a given mechanism of thrombocytopenia. If the etiology of the thrombocytopenia is unclear, a bone marrow examination should also be carried out.[5] The aim and objectives of the present study were to evaluate the clinico-hematological profile of various patients with thrombocytopenia and to study age and gender-wise distribution, various etiological factors, the severity of thrombocytopenia, and its association with bleeding manifestations in patients.

MATERIALS AND METHODS

This prospective study was carried out in the hematology section of the Department of Pathology at a tertiary care centre, Ahmedabad, Gujarat, India for period of two years from August 2020 to August 2022. A total of 300 patients were considered for this study. Guidelines of The World Medical Association Declaration of Helsinki were followed while conducting the study. Approval from the institutional research committee (2020/242) was obtained.

2 ml of blood was collected in EDTA vaccutainers and analyzed using a 5-part automated hematology analyzer-Sysmex XT-2000i for CBC. Platelet counts were enumerated as a part of CBC. The low platelets were also confirmed by examination of the PBS stained with field stain.

Inclusion Criteria

All indoor patients, both male and female, of all the age groups whose platelet counts were less than 1,50,000/µl were included in the study.

Exclusion Criteria

Patients whose clinical details were not available, patients with a recent history of blood transfusion, hemolyzed, and clotted samples.

Detailed history and clinical evaluation were done. Laboratory tests included: prothrombin (PT), partial thromboplastin time (PTT), C-reactive proteins (CRP), LFT, and serum lactate dehydrogenase (LDH). Patients presenting with fever were also investigated for malaria, typhoid, dengue fever by doing malaria rapid antigen test (MALCARD), widal test, (NS1 antigen, dengue IgG and IgM antibodies) respectively. Enzyme-linked immunosorbent assay (ELISA) was done for human immunodeficiency virus (HIV) when indicated by 4th generation kit, for hepatitis B surface antigen (HBsAg) and hepatitis C virus (HCV) using 3rd generation kits. As the study period was during the global pandemic of COVID-19 infection, a Covid real-time Reverse Transcription – Polymerase Chain (RT-PCR) was carried out in patients when indicated. Bone marrow aspiration and biopsy examination were performed whenever indicated. A panel of these investigations was viewed and assessed to reach the likely etiology of thrombocytopenia amongst a vast spectrum.[6] Thrombocytopenia was graded as follows based on the platelet count, mild: 100-150 × 10^3 / µL, moderate: 50-100 × 10^3 /µL, severe: <50 × 10^3 /µL, very severe: <20 × 10^3 /µL.

RESULTS

In the present study, thrombocytopenia cases were more predominant in males (n=178, 59.3%). The male-to-female ratio was 1.5:1. Out of 300 patients, the majority of patients (n=81, 27.0%) belonged to an age group of 20-29 years, followed by 30-39 years of age (n=47, 15.7%). The lowest frequency was observed between 0-9 years of age (n=5, 1.7%). (Table1)

Table 1: Age-wise distribution of patients with thrombocytopenia.

Age (years)	Number of patients (n=300)	Percentage %
0-9	5	1.7%
10-19	35	11.7%
20-29	81	27.0%
30-39	47	15.7%
40-49	34	11.3%
50-59	29	9.7%
60-69	33	11.0%
70-79	27	9.0%
>=80	9	3.0%
Total	300	100%

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The majority (n=124, 41.3%) had moderate thrombocytopenia followed by mild thrombocytopenia (n=108, 36.0%). Only 22 patients (7.3%) had severe thrombocytopenia. (fig.1)

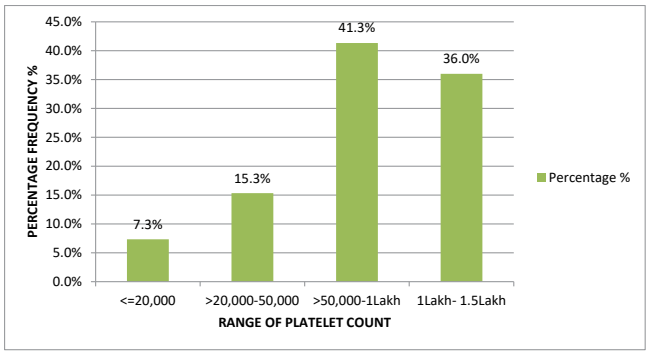

Figure 1: *The bar graph depicts the range of platelet counts of patients studied.*

The most common clinical complaint was fever (n=222, 74%), followed by generalized weakness (n=177, 59%) and bodyache (n=114, 38%). A minimum of n=4, 1.3% and n=5, 1.7% patients had splenomegaly and hepatomegaly respectively. (fig.2)

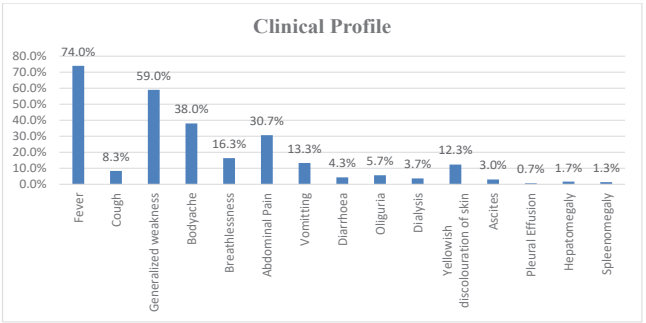

Figure 2: *Clinical profile of patients with thrombocytopenia.*

Figure 3 shows the most common diseases noted in present study. It was COVID-19 (n=63, 21.0%) followed by dengue (n=51, 17%) and malaria (n=38, 12.7%).Liver disease (n=31, 10.3%) and megaloblatic anemia (n=29, 9.7%) were other major diseases accounted for.

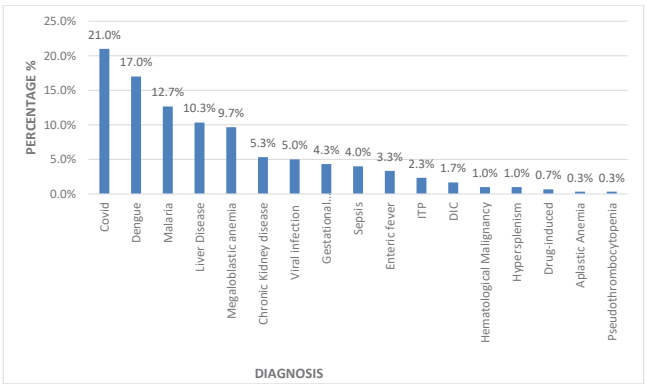

Figure 3: *The bar graph depicts the frequency distribution of the diagnosis.*

Table 2 shows platelet counts and morphology on PBS examination. Giant platelets were seen in ITP and platelet clumps were appreciated in pseudothrombocytopenia. There was a significant association between various disease categories and low platelet counts (p-value < 0.05). COVID-19 infection, malaria, dengue, enteric fever, gestational thrombocytopenia, megaloblastic anemia, and liver diseases were associated with mild to moderate thrombocytopenia. Very severe thrombocytopenia was noticed in dengue, ITP, and megaloblastic anemia.

Table 2: Association of disease categories and the degree of thrombocytopenia.

Disease categories	Platelets on peripheral smear	Platelet Range				
		<20000	20000-50000	50000-100000	>100,000-150,000	Chi-square test value
Aplastic Anemia	Reduced	1	0	0	0	
Chronic kidney disease	Reduced	1	1	4	10	
Covid	Reduced, Anisocytosis, Few Giant platelets	2	11	24	26	
Dengue	Reduced, Anisocytosis, Few Giant platelets	7	8	23	13	
Disseminated Intravascular Coagulation	Reduced	0	1	2	2	
Drug induced thrombocytopenia	Reduced	1	1	0	0	
Enteric fever	Reduced	0	2	1	7	
Gestational Thrombocytopenia	Reduced, Anisocytosis, Few Giant platelets	1	0	5	7	
Hematological Malignancy	Reduced, anisocytosis, Giant and hypogranular platelet	1	0	2	0	**p- value-0.023**
Hypersplenism	Reduced	0	1	1	1	
Immune Thrombocytopenia	Reduced,anisocytosis, Giant platelets	2	3	2	0	
Liver disease	Reduced, Anisocytosis, Few Giant platelets	1	5	18	7	
Malaria	Reduced	2	7	16	13	
Megaloblastic anemia	Reduced	3	3	13	10	
Pseudo thrombocytopenia	Many Platelet clumps	0	0	1	0	
Sepsis	Reduced	0	1	6	5	
Viral infection	Reduced	0	2	6	7	
Total		22	46	124	108	

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Out of 300 patients with thrombocytopenia, 204 (68%) patients also had anemia. 133 (44.3%) patients had leucopenia along with thrombocytopenia while 104 (34.6%) patients had pancytopenia.

In this study, 40 (13.3%) patients required bone marrow examination for evaluation of the cause of thrombocytopenia. The majority of patients were diagnosed as megaloblastic anemia (n=29, 72.5%), followed by ITP (n=7, 17.5%), plasma cell myeloma (n=2, 5.0%), MDS (n=1, 2.5%) and aplastic anemia (n=1, 2.5%).

It was observed that bleeding manifestations were present in only 25 (8.3%) out of 300 patients. The most common clinical manifestation observed in the study was petechiae (n=8, 32%), followed by gum bleeding (n=5, 20%) and easy bruising (n=3, 12%). Epistaxis and hematemesis were seen in similar frequency (n=2, 8%). It was observed that gastrointestinal (GI) and genitourinary (GU) bleeding like melena, hematuria, menorrhagia, hematemesis, as well as a combination of such manifestations occurred at the levels of very low platelet counts with counts being <20,000/µl in most cases (Table 3). Patients who presented with easy bruising in the present study also had platelet counts below 20,000/ µl. However, these findings fluctuated from patient to patient based on the clinical severity of the underlying disease and associated co-existing conditions.

Table 3: Frequency distribution of various types of bleeding manifestations with platelet counts

Type of bleeding manifestation	Number of patients with bleeding manifestation (n=25)	%	Platelet counts Mean ± SD
Petechiae	8	32.0	18966.7±3225.4
Easy bruising	5	20.0	13950.0±6434.7
Gum bleeding	3	12.0	45660.0±32310.0
Epistaxis	2	8.0	38200.0± -
Hematemesis	2	8.0	10000.00± -
Hematochezia	1	4.0	35200.0±6788.2
Hematuria, Gum Bleeding	1	4.0	11500.0± -
Hemoptysis, melena	1	4.0	6500.00± -
Melena	1	4.0	10000.0± -
Menorrhagia	1	4.0	26050.0±20967.3
Total	25	100.0	

**SD – Standard Deviation*

According to Table 4, significant association was found between low platelet counts and bleeding manifestations (p-value< 0.0001). The frequency of bleeding manifestations was more when platelet count was less than 20,000/µL (n=18, 72.0%). No bleeding manifestations were seen in cases of mild thrombocytopenia.

Table 4: Association of initial platelet count with bleeding manifestations.

Platelet counts / µL	Bleeding manifestations		Chi-square test value
	Yes [n (%)]	No [n (%)]	
<20000 (Very severe thrombocytopenia)	18 (72.0)	7 (2.5)	p-value <0.0001
20000-50000 (Severe thrombocytopenia)	7 (28.0)	40 (14.5)	
50000-100000 (Moderate thrombocytopenia)	0 (0.0)	120 (43.6)	
>100000 (Mild thrombocytopenia)	0 (0.0)	108 (39.3)	
Total	25 (100)	275 (100)	

DISCUSSION

Thrombocytopenia is one of the most common findings observed in various clinical conditions. It has an inverse relation to mortality and morbidity. The degree of thrombocytopenia in infections has prognostic value. The observation from the present study highlights the importance of platelet count in various diseases. The early appropriate treatment gives better results. In the present study, a total of 300 cases of thrombocytopenia were included to evaluate the clinical spectrum of thrombocytopenia.

Age

The mean and median age were 40 and 36 years respectively. The majority of the cases belonged to the age group of 20-29 years (n=81, 27.0%), followed by 30-39 years (n=47, 15.7%). These findings were similar to the study done by Paramjit E et al. in which the maximum number of patients were in the age group of 20-39 years (n=157, 52.3%).[6]

Gender

Thrombocytopenia cases were more predominant in males (n=178, 59.3%) which was similar to the findings of Paramjit E et al. (n=204, 68%), Bhalara et al. (57.2%) and Lakshmikumar et al. (n=115, 60.5%).[6-8]

In the present study, most of the patients (n=124, 41.3%) had platelet counts ranging from 50,000-1,00,000 which was concordant with the observations of Fawas MN et al. (n=134, 53%), Nakhale BD et al. (n=67, 54.91%) and Reddy YC et al.(n=40,40.0%).[9-11] The most common clinical presentations were fever (n=222, 74%), generalized weakness (n=177, 59%) and bodyache (n=114, 38%) which were comparable to the findings of other studies (Table 5).

Table 5: Comparison of clinical profile with other studies.

Clinical Profile	Lakshmikumar et al.8 (n=190)	Vimal M et al.12 (n=120)	Raman SD et al.13 (n=100)	Present study (n=300)
Fever	100.0%	45.0%	75.0%	74.0%
Cough	-	-	5.0%	8.3%
Generalized weakness	73.2%	-	-	59.0%
Bodyache	65.8%	17.5%	22%	38.0%
Breathlessness	26.8%	8.3%	-	16.3%
Abdominal Pain	49.5%	-	13.0%	30.7%
Vomitting	73.7%	-	10.0%	13.3%
Diarrhoea	16.3%	-	3.0%	4.3%
Oliguria	-	-	-	5.7%
Dialysis	-	-	-	3.7%
Yellowish discolouration of skin	17.4%	5.8%	1.0%	12.3%
Ascites	30.0%	10.0%	-	3.0%
Pleural Effusion	25.8%	-	11.0%	0.7%
Hepatomegaly	19.47%	5.0%	18.0%	1.7%
Splenomegaly	18.94%	9.2%	28.0%	1.3%

The most common etiology noticed was COVID-19 infection (n=63, 21.0%). This was not seen in studies by Bhalara et al. and Vimal M et al., as this study was carried out during the COVID-19 pandemic.[7, 12] The second most common etiology was dengue (n=51,17.0%), followed by malaria (n=38,12.7%) which was similar to the findings of Bhalara et al. (dengue 28.6%, malaria 22.8%).[7] However, in contrast to the present study, the second etiology in line found by Vimal M et al., was liver diseases (n=31,16.7%), this could be because of differences in the geographical area and period of study.[12] Other etiologies in the present study were more or less comparable to other studies. However, the consistency of findings was variable depending on various factors like place of study (dengue and malaria are endemic to certain regions), seasonal factors (other viral infections), etc.

The frequency of bleeding manifestations was less (n=25, 8.3%) in this study. This was disconcordant to the findings by Lakshmikumar et al. (n=83, 43.7%), Nakhale et al. (n=33, 27.06%), and Raman et al. (n= 36, 36%). [8, 10, 13] The most common bleeding manifestation in the study was petechiae (n=8, 32.0%). This finding was in contrast to the observation by Lakshmikumar et al. (n=17, 8.9%) but similar to the study carried out by Yadav BS et al. (n=13, 31.7%).[8, 14] However, the study carried out by Lakshmikumar et al. had gum bleeding as the most common manifestation (n=28, 14.7%).[8] Gum bleeding was the second most common manifestation in this study (n=5, 20.0%). The frequency of patients who presented with easy bruising was variable in our study due to the association of other conditions co-existing with thrombocytopenia. Major GI and GU bleeding occurred with very little frequency in this study (hematemesis, melena, hematuria each, n= 1, 4.0%) while it was variable in the study carried out by Lakshmikumar et al. (hematemesis and melena each, n=3, 1.6%, hematuria n=21, 11.1%), Yadav BS et al. (hematemesis n=11, 26.83%, melena n=10, 24.4 %, hematuria n=8, 19.5%) and Saini KC et al. (hematemesis 4.5%, melena 9.5%, hematuria1.5%)[8, 14, 15] The frequency of bleeding manifestations was n=18, 72.0% when platelet count was less than 20,000/µ, and n=7, 28.0% with platelet count between 20,000-50,000/µL. No Bleeding manifestations were seen when platelet counts were 50,000/µL. These findings were similar to a study carried out by Yadav BS et al. with a frequency of n=12, 27.27% with platelet count less than 20,000/µL, and n=17, 21.0% when 20,000-50,000/µL.[14] Yadav BS et al. noticed 12 cases (9.44%) with bleeding symptoms when platelet counts were between 50,000-100,000/ µL which was non-concordant with the present study. This may be attributed to the fact that this study was performed during the COVID-19 period.[14] In the present study, the degree of thrombocytopenia was mild to moderate in COVID-19 infection, however, the incidence of thrombocytopenia in patients with COVID-19 has been variable across studies. The majority of patients did not have any severe degree of thrombocytopenia similar to the present study (<20,000/µL or a sudden drop >50% over 24–48 h).[16]

Dengue was associated with moderate to severe thrombocytopenia in this study comprising of n=38, 74.5% which was comparable to the study carried out by Makroo RN et al. who noticed 191 cases (84.88%) having platelet count <100,000/µL.[17] The pattern of platelet counts over time in patients with dengue fever was typically lowest between the third and sixth days after the onset of illness, just before the fever began to subside.

In the study, malaria caused mild to moderate thrombocytopenia with counts remaining between 50,000-100,000/µL in most of the cases. Moreover, platelet count was lower in infections with p. falciparum compared to p.vivax. This was comparable to the study carried out by Pagaro PM et al.[18]

This study showed 31 (10.3%) cases of liver diseases which was concordant with the study done by Vimal M et al. (n=20,16.7%).[19] The degree of thrombocytopenia was however variable. The liver is the site of thrombopoietin production, reduction of functional liver cell mass in chronic liver diseases like cirrhosis, viral hepatitis, alcoholism, portal hypertension, carcinoma, etc. leads to suppressed thrombopoiesis and subsequent peripheral thrombocytopenia.[12]

In chronic kidney diseases, the present study showed mild thrombocytopenia. It is usually thought to be encountered due to reduced thrombopoietic activity. Platelet dysfunction and impaired platelet-vessel wall interaction resulted in complex hemostatic disorders in patients with end-stage

renal disease.[12]

In enteric fever, mild thrombocytopenia was observed in most of the patients which was comparable to findings by Shrivastava K et al.[20] Bicytopenia and subclinical Disseminated intravascular coagulation (DIC) were very common findings contributed by bone marrow suppression and hemophagocytosis. Pancytopenia and isolated thrombocytopenia in enteric fever were rare. This was because hematological findings in enteric fever may not follow a prototypic pattern in tropical regions. However, a differential diagnosis of enteric fever should also be considered when evaluating a patient with fever and thrombocytopenia.[12] In the present study, the degree of thrombocytopenia was varied in DIC, drug-induced thrombocytopenia, megaloblastic anemia, etc, depending upon the clinical severity of the disease.

In this study, three instances (1.0%) of hematological malignancies showed signs of thrombocytopenia. The fact that the current investigation was carried out during the COVID-19 pandemic may have contributed to the low frequency. Thrombocytopenia can occur in some hematological malignancies due to several factors, such as chemotherapy, tumor infiltration of the bone marrow, microangiopathic disorders, and secondary immunological thrombocytopenia.[12] In the present study, in patients with gestational thrombocytopenia, platelet counts were >50,000/μL showing mild to moderate thrombocytopenia in 12 (92.3%) cases and the remaining one case showed very severe thrombocytopenia. Gestational thrombocytopenia during the third trimester, with postpartum resolution was the most common cause of thrombocytopenia in pregnancy. Preeclampsia, HELLP syndrome, and ITP can pose life-threatening complications during pregnancy.[12]

In the rest of other etiologies, findings could not be correlated with other studies, in which, the range for mild, moderate, and severe thrombocytopenia were different as there was no uniformly accepted criteria for the grading of thrombocytopenia. Moreover, the degree of thrombocytopenia largely depended on disease severity and patient demographics.

CONCLUSIONS

Thrombocytopenia is an extremely common hematological condition. In present study, it was more prevalent in men. Most commonly affected age group was 20 to 29 years. The most prevalent clinical manifestations were fever and bodyaches, but bleeding signs were less common. Thrombocytopenia can occur in isolated forms and also with anemia, pancytopenia, and leucopenia. The most prevalent form was moderate thrombocytopenia. The most common etiologies were COVID-19, dengue fever and malaria in present study. As thrombocytopenia can have a variety of clinical presentations, including infections and malignancies, a battery of tests should be performed to discover the root cause. Only patients with unexplained severe or increasing thrombocytopenia should undergo bone marrow aspiration and biopsy.

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