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Original Article

Validation of international recommendations on bone marrow aspiration for Pediatric Immune Mediated Thrombocytopenic Purpura

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ABSTRACT

Introduction: Immune medicated thrombocytopenic purpura (ITP) is diagnosed on clinical basis with normal peripheral smear findings. Bone marrow examination is done to rule out leukemia or aplastic anemia. In most of cases with typical history and peripheral smear findings diagnosis is not changed after a bone marrow examination.

Objective: Objectives of this study were to study clinico-hematological findings in cases of immune medicated thrombocytopenic purpura and to validate findings of our study with international recommendations.

Methods: This study included fifty pediatric patients who presented with suspicion of ITP or with thrombocytopenia and turned out to be ITP. Their clinico-pathological signs and symptoms and peripheral blood smears and bone marrow findings were noted.

Results: Age range of patients in this study was from five months to twelve years. Female to male ratio was 54%: 46% respectively. All patients presented with history of cutaneous bleeding. None of patients had lymphadenopathy. Splenomegaly was present in 4% of cases. 50% of patients had platelet count of less than 20,000 $x10^9$/L and 16% had platelet count between 50,000 $x10^9$/L -100,000 $x10^9$/L. Bone marrow examination in all patients showed normal myeloid, erythroid and lymphoid series cells with prominent megakaryocytes. 94 % of patients showed increased number of megakaryocytes and 78% of patients showed immature forms of megakaryocytes on bone marrow aspiration.

Conclusion: Bone marrow examination is not required as a first line investigation for diagnosis in a typical clinical scenario. The routine of using bone marrow examination as a first line diagnostic tool in childhood ITP should be avoided unless clinically indicated.

Key Words: Pediatric Immune Mediated Thrombocytopenic Purpura, Immune thrombocytopenia, ITP, Bone marrow aspiration

Introduction

Immune thrombocytopenic purpura (ITP) is an auto-immune disorder which causes antibody mediated platelet destruction and cause decrease in production of platelets.[1] Patients with ITP have low platelet count (less than <100x10^9/L) without any other cause of thrombocytopenia. ITP affects patients of all age groups and both sexes are involved. There are no racial differences in its prevalence.

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Acute ITP is seen mostly in childhood and it recovers spontaneously in most of cases. Clinical presentation of ITP is highly varied. Patients will present with signs of low platelet count. But platelet count is often misleading, and patients may not have bleeding even when platelet count is very low. Mucocutaneous bleeding is most commonly observed sign.[2] Acute onset of illness is a main clue to diagnosis. In healthy children there is often a history of preceding viral infection. History of prolong fever, weight loss, bony aches and pains, visceromegaly and presence of lymph nodes suggests alternate diagnosis. If such atypical features are observed, then one should also keep in mind aplastic anemia and leukemia.

Assessment of complete blood counts followed by careful examination of peripheral blood smear is of utmost importance. Thrombocytopenia along with normal red blood cell (RBC) and white blood cell (WBC) morphology is suggestive of ITP. In a typical case of childhood ITP abnormalities in RBCs or WBCs are rare. Other laboratory tests like Rh typing and Coombs test may be helpful to rule out hemolysis. In some cases, antiphospholipid antibodies level, antinuclear antibodies, parvovirus and cytomegalovirus testing may be done. Detection of antiplatelet antibodies is not considered because it has low sensitivity and specificity. Immunoglobulin levels, screening for HIV, HCV and H pylori infection is also recommended in selected cases.[3] When bone marrow of patient with ITP is done, there will be normal bone marrow aspiration findings. Megakaryocytes are either adequate or increased in number. Morphologically megakaryocytes in ITP patient are either normal in appearance or immature forms can be observed.[4] In 2011 American Society of Hematology provided updates on its guidelines for diagnosis and management of ITP. This update is widely accepted and is followed worldwide. Other European countries have also developed various recommendations for diagnosis and management of ITP.[5] International recommendations suggest that proper history, physical examination, complete blood count and careful assessment of peripheral smear are enough to make a diagnosis of ITP. These guidelines recommend that if patient have typical history, physical examination findings, his complete blood count and peripheral smear are suggestive of ITP, then no further investigation is required, and bone marrow examination is not necessary in patients with typical features of disease.[6]

Methods

This study was conducted at pathology department of Pakistan Institute of Medical Sciences (P.I.M.S). Fifty cases were included. Children with suspicion of ITP and those with thrombocytopenia and turned out to be ITP on bone marrow examination were included in this study. Complete and detailed history and examination was carried out. All findings of history and examination were entered in preformed Performa. The findings of blood counts, differential leukocyte count, reticulocyte count, red blood cells and platelet morphology were also entered in another Performa. Bone marrow aspiration was done in all cases. Trephine biopsy was carried out in children more than two years of age. In children less than two years bone marrow clot was assessed. Bone marrow findings were noted in Performa. All data was entered and was analyzed by using SPSS version 16.

Results

Mean age was 3 years ± 1.8 SD. 36% of patients were in age group of 2 to 6 years. 28% of patients were up to two years of age. 54% of cases were females. Males in this study were 46%. Most common clinical features observed were bruises, epistaxis, petechial rash and gum bleeding. Bruises were present in 58% of cases. 50% of patients had petechial rash. 48 % of patients had epistaxis. Severe bleeding like hematuria, hematemesis and malena were present in 20% of patients. None of patients had lymphadenopathy. Splenomegaly was present in 4% of cases. Hepatomegaly was present in 6 % of cases. 28% of patients had a platelet count in range of 3,000x 10^9/L to 10,000 x10^9/L. 22% of patients had a platelet count of 10,000x10^9/L to 20,000 x10^9/L. Thus 50% of patients in our study had platelet count of less than 20,000 x10^9/L, 34% had platelet count between 20,000 x10^9/L -50,000 x10^9/L and 16% had platelet count between 50,000 x10^9/L -100,000 x10^9/L [Table 1]. Mean platelet count was 3.2 x10^9/L ±2.4 and Median platelet count was 2.5x10^9/L. In 40% of cases large platelets were seen on peripheral smear. On bone marrow aspiration 94 % of cases showed increased number of megakaryocytes while 6% cases showed adequate number of megakaryocytes [Table 2]. 78% of cases showed immature forms of megakaryocytes i.e. Left shift and 22% of cases showed normal maturation of megakaryocytes on bone marrow aspiration smear [Table 3]. 50% cases showed presence of iron in bone marrow stores while 32% of cases had no stored iron in bone marrow. Out of 50 cases that were sent for bone marrow aspiration, 37 cases i.e. 74% were sent with clinical suspicion of ITP. Bicytopenia (Anemia with thrombocytopenia) was an indication of bone marrow aspiration in 16% of cases. 68% of cases had a final diagnosis of ITP. 30% of ITP cases had associated iron deficiency anemia.

Table 1: Platelet count (n=50)

Platelet Count	Frequency	Percent
1000-10,000	14	28.0
10,000-20,000	11	22.0
20,000-30,000	10	20.0
30,000-40,000	4	8.0
40,000-50,000	3	6.0
50,000-60,000	2	4.0
60,000-70,000	3	6.0
80,000-90,000	1	2.0
90,000-100,000	2	4.0
Total	50	100.0

Table 2: Number of megakaryocytes on Bone Marrow Aspiration (n=50)

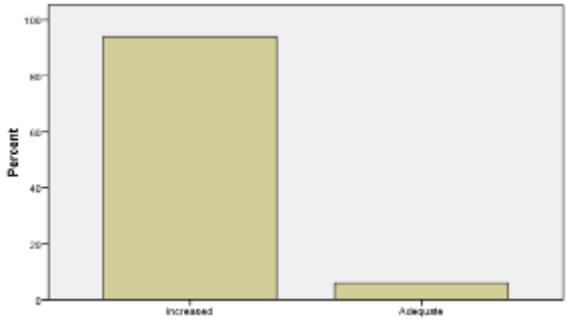
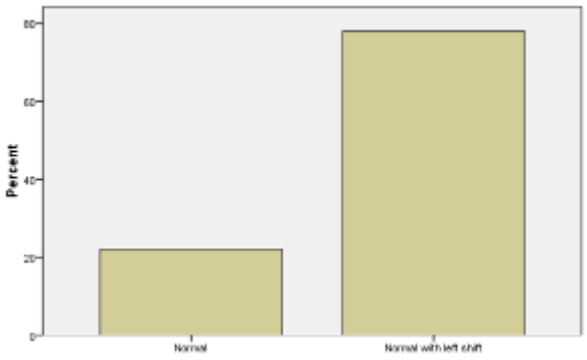


Table 3: Megakaryocyte maturation on Bone Marrow Aspiration (n=50) also seen (100X)



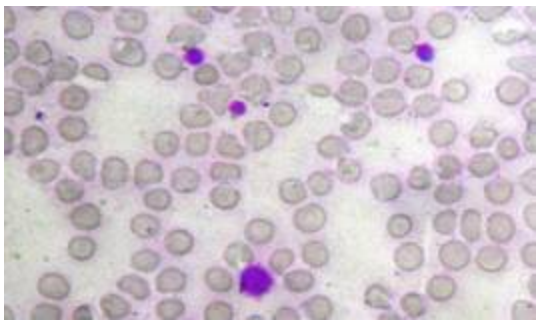


Fig 1: Peripheral smear: Inadequate platelets with Occasional large platelets (Wright Giemsa Stain X100)

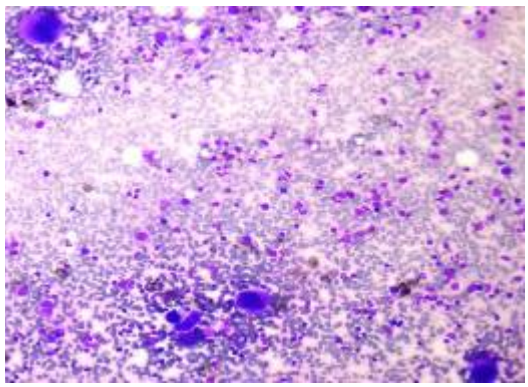


Fig 2: Hyper cellular bone marrow smear with increased megakaryocytes (Wright Giemsa Stain X100)

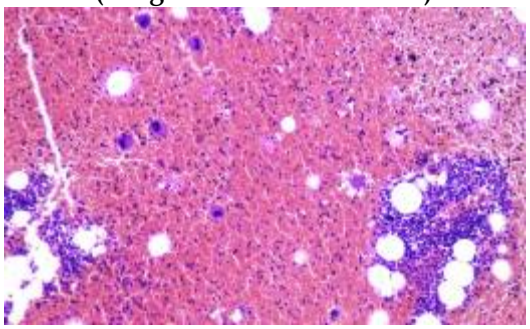


Fig 3: Bone marrow clot: increased megakaryocytes (Wright Giemsa Stain X100)

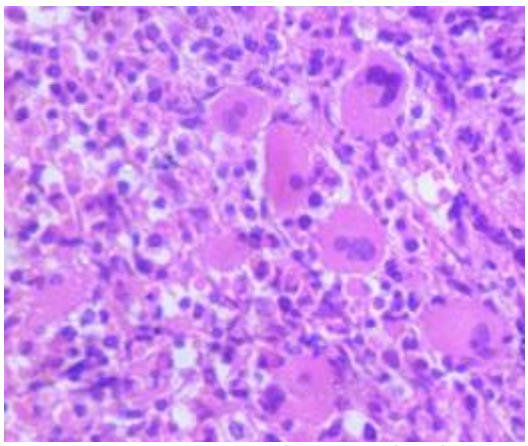


Fig 4: Bone marrow trephine showing increased megakaryocyte (Wright Giemsa Stain X400)

Discussion

Clinical features of ITP are associated with thrombocytopenia. Thus, most of patients present with some features of cutaneous bleeding like bruises or purpura as observed in our study. Study done in Agha

Khan University Hospital, Pakistan has also shown presence of cutaneous bleeding as most common clinical feature.[7] Physical examination is mostly normal in patients with ITP, except for signs of skin or mucosal bleeding. Presence of lymphadenopathy or hepatosplenomegaly is not a feature of ITP. 4 % of cases in our study presented with splenomegaly. Presence of splenomegaly in childhood ITP is reported in other studies. Study done by Saeidi S et al reported splenomegaly in 4.2% cases.[8] 50% of patients in our study had platelet count of less than 20,000 $x10^9/L$. Study by Keim-Malpass et al showed 62% of patients with platelet count less than 10,000 $x10^9/L$.[9] ITP is an immune mediated condition where exogenous platelet replacement has been historically considered to be ineffective as there is rapid immune mediated destruction of platelets. Initial treatment in patients with low platelet count accompanied by mild to moderate hemorrhagic symptoms is usually steroids or intravenous immunoglobulins and platelet transfusions are reserved for those patients who present with life-threatening bleeding. However, in our study 42% of patients received platelet transfusion. The decision of platelet transfusion was based on the low platelet count rather than severity of bleeding. Moreover, it was observed that after platelet transfusion there was no increment seen. Thus, validating the guideline regarding no platelet transfusion in ITP, except clinically significant bleeding and in that case use of platelet transfusion should be adequate to maintain hemostasis. As parenteral immunoglobulins take twenty-four hours and different forms of steroids due to different time of action can take up to many days, before a positive effect on the platelet count is observed. In the meantime, patients with severe bleeding or at risk of serious bleeding, may benefit from platelet transfusion.[10] 94 % of cases in our study showed increased number of megakaryocytes on bone marrow examination. This finding was consistent with findings of Vinayakamurthy s et al, Choudhary et al and Shi et al who found an increase in megakaryocytes in 80%, 91% and 98%, of ITP cases.[11] Our present study has proved American Society of Hematology (ASH) recommendations, as ASH recommends that if a patient has typical features of ITP, bone marrow examination is not required. It also recommends that bone marrow examination is not required in those children in whom intravenous immunoglobulin treatment fails. ASH does not recommend bone marrow examination before starting steroid therapy or before splencectomy.[6] International consensus report provides with recommendation that bone marrow examination may help when patient is old or has abnormal clinical features. They also recommend bone marrow examination in selected patients prior to splenectomy.[12] Bone-marrow aspiration in our setup is often performed in children with suspicion of acute ITP, to rule out leukemia, aplastic anemia or other hematologic diseases. Role of bone marrow aspiration in children with typical clinical and hematological features of acute ITP has been questioned. A study done by Mahabir et al, on bone marrow examination in patients of ITP and healthy controls, showed no difference in bone marrow findings of two groups. It was found that sensitivity of bone marrow examination in diagnosis of ITP was 24% and specificity was 90%.[13] Retrospective data analysis on bone marrow examination was done by Jubelirer SJ and Harpold R. Eighty six patients with isolated thrombocytopenia showed that eighty two of these patients had normal or increased megakaryocytes with normal erythroid and myeloid series on bone marrow examination. When these patients were followed for about twenty-two months, diagnosis of none of these patients changed. Ninety-nine patients of acute leukemia were also studied. All these patients had atypical features, which did not fit into ITP.[14] A study was performed by Muhsen AA and Abdurrahman KN, to determine whether bone marrow examination is indicated in acute childhood ITP. That study concluded that routine bone-marrow examination is not needed for children with typical features of acute ITP.[15] A similar study conducted by Ahmad Z et al, on need of performing bone-marrow for the diagnosis of ITP concluded that bone-marrow examination should not be a part of routine work-up for diagnosing ITP in children and should be reserved for those having atypical clinical and laboratory features.[16] This supports present study. If a patient has a typical history of ITP like acute onset of illness in otherwise healthy child with history of bruises and petechiae as manifestation of low platelet count and no abnormal findings on physical examination, isolated thrombocytopenia, normal red blood cell and leukocyte morphology then bone marrow examination should not be done for diagnosis of ITP. In children wait and watch policy should be practiced. Bone marrow examination should be carried out in those patients where alternative diagnosis is suspected or when patient does not respond to treatment. This practice will not only decrease unnecessary investigations but will also help in better management of patients. Presently no specific test is available for

the diagnosis of ITP, it is mostly based on diagnosis of exclusion. However, emergence of genetic biomarkers in future may help for early detection and confirmation of cases as well as they may help in prediction of prognosis.[17] With this detection of genetic component of ITP and availability of biomarkers will further reduce requirement of bone marrow examination in diagnosis of ITP.

Conclusion

ITP is a diagnosis of exclusion, so treatment should be started after excluding other causes of thrombocytopenia in children. Once the treatment is started with suspicion of ITP, to observe response to treatment is a better option than to requesting a bone marrow examination. Clinicians should wait for at least one week to see response of treatment, before requesting for a bone marrow examination. Spontaneous remission occurs in most cases of ITP in children, so clinicians should not carry out bone marrow examination as first line invasive test. Platelet transfusion in ITP should be reserved for clinically significant bleeding and practice of treating platelet counts should be avoided.

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Authors' Contributions:

- Hina Bilal: participated in conception/ study designing/ planning/ study conduction, analysis, interpretation, discussion and manuscript writing.
- Nazia Siddiqui: participated in study conduction, analysis, interpretation and discussion.
- Lubna Naseem: participated in conception/planning, analysis, interpretation and discussion.

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