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Clinicopathological Evaluation of Thrombocytopenia in Children Under 14 Years Based on Bone Marrow Aspiration Samples Submitted to the Pathology Department of Shahid Sadoughi Hospital, Yazd (2016–2022)

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A B S T R A C T

Introduction: Platelets play a crucial role in maintaining endothelial integrity and controlling bleeding in small vascular injuries by forming platelet plugs. Thrombocytopenia encompasses a wide range of clinical manifestations and pathological diagnoses. This study aimed to evaluate the clinicopathological features of thrombocytopenia in children under 14 years presenting to Shahid Sadoughi Hospital, Yazd.

Materials and Methods: Data were collected using a pre-designed checklist including age, gender, pathological diagnosis, complete blood count (CBC) findings, and clinical symptoms. All cases of thrombocytopenia in children under 14 years were extracted from the pathology records of Shahid Sadoughi Hospital. The data were analyzed using SPSS software, employing the Chi-square test. A p-value < 0.05 was considered statistically significant.

Results: Bone marrow aspirations from 375 children under 14 years with thrombocytopenia were analyzed, including 215 males and 160 females. The most affected age group was 1–5 years (50.7%), with males comprising 57.3% of cases. The most common pathological finding was acute leukemia (47.2%). Non-malignant diagnoses were overall more prevalent than malignant ones. The most frequent CBC finding was bicytopenia (low platelets and hemoglobin, 44.8%), and the most common clinical manifestation was bleeding manifestations (48.3%).

Conclusion: This study highlights that acute leukemia is the predominant pathological diagnosis in pediatric thrombocytopenia, with bleeding manifestations being the most common clinical presentation. These findings underscore the importance of thorough clinicopathological evaluation in effectively managing pediatric thrombocytopenia.

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Introduction

Thrombocytopenia, defined as a platelet count below the normal range, is a common hematological condition in children and may arise from a variety of benign and malignant disorders [1] .Platelets play a crucial role in maintaining vascular integrity and controlling bleeding by forming platelet plugs at sites of endothelial injury [2]. Consequently, thrombocytopenia can lead to significant clinical complications, particularly in pediatric patients [3]. The clinical manifestations of thrombocytopenia vary widely, ranging from mild bruising to severe hemorrhagic events, depending on the underlying cause and severity [4].

Bone marrow aspiration and biopsy are often essential diagnostic tools to identify the etiology, which may include conditions such as immune thrombocytopenia, infections, marrow failure syndromes, or malignancies like leukemia [3]. While thrombocytopenia is relatively common, its clinical and pathological patterns, as well as its underlying etiologies, can differ significantly based on demographic factors. Understanding the clinicopathological spectrum of thrombocytopenia in children is vital for timely diagnosis, appropriate treatment, and improved outcomes [5, 6].

Bone marrow aspiration is particularly important when peripheral blood findings are inconclusive. Studies have shown its value in distinguishing between primary bone marrow disorders (such as leukemia and aplastic anemia) and secondary causes including immune thrombocytopenia (ITP) and infections [7]. Acute leukemia is one of the most common malignant causes of thrombocytopenia in children [8], whereas non-malignant causes like ITP or viral infections often present with isolated thrombocytopenia and a more favorable prognosis. The age distribution and gender predisposition vary across studies [9].

Balasubramanian et al. (2022) emphasized the role of bone marrow examination in pediatric patients aged 12–18 years, with acute leukemia as the most frequent neoplastic diagnosis and infections the leading non-neoplastic cause [10]. Kumar reported that bicytopenia (86.13%) was more common than pancytopenia (13.86%), predominantly affecting boys aged 6-12, with infections as the leading cause [11] . Bahadure et al. found fever as the most common presenting symptom and noted megaloblastic anemia, aplastic anemia, and acute leukemia as key causes [12].

Waris identified acute leukemia (25%) and aplastic anemia (20%) as the leading etiologies of cytopenia, with leukemia being more commonly associated with bicytopenia and aplastic anemia with pancytopenia [13] . Rasheed reported pallor and fever as the most frequent symptoms, with hematological malignancies, infections, and megaloblastic anemia being common causes [14]. Bahal found anemia with thrombocytopenia to be the most frequent bicytopenia pattern, while pancytopenia was associated with increased bleeding [15]. Singh noted that although peripheral smears often showed dimorphic

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patterns, they lacked diagnostic correlation with marrow findings [16] .Dubey observed a 2.9% incidence of pancytopenia in children, with megaloblastic anemia, aplastic anemia, and leukemia as the top causes [17] .

Despite extensive research, gaps remain in understanding the specific clinicopathological patterns of thrombocytopenia in different populations. Regional data are essential for identifying unique epidemiological and pathological trends that can guide tailored diagnosis and treatment. This study adds value to previous research by providing region-specific data from Iran over a seven-year period (2016–2022), which has not been extensively reported in the literature. Unlike many earlier studies that mainly focused on pathological diagnoses, our work emphasizes the diagnostic significance of cytopenia patterns (isolated thrombocytopenia, bicytopenia, and pancytopenia) and their correlation with clinical manifestations. This clinicopathological approach provides a more comprehensive framework for differentiating between malignant and non-malignant causes in pediatric thrombocytopenia. This study aimed to evaluate the clinicopathological features of thrombocytopenia in children under 14 years based on bone marrow aspiration samples submitted to the pathology department of Shahid Sadoughi Hospital in Yazd from 2016 to 2022. The findings are expected to contribute to a deeper understanding of the most common underlying causes and clinical presentations of pediatric thrombocytopenia, thereby improving diagnostic and therapeutic strategies.

Materials and Methods

Study design

This retrospective descriptive study was conducted to evaluate the clinicopathological features of thrombocytopenia in children under 14 years of age.

Study setting and data collection

The study was conducted in the Pathology Department of Shahid Sadoughi Hospital, Yazd, Iran, and involved a review of bone marrow aspiration reports for pediatric patients diagnosed with thrombocytopenia between 2016 and 2022. Data were retrospectively extracted using a standardized checklist, capturing demographic information (age and gender), clinical findings (symptoms such as petechiae, purpura, mucosal bleeding, and any history of medications), laboratory data (CBC parameters, including hemoglobin level, platelet and white blood cell count), and pathology findings (megakaryocyte morphology and count, dysplasia, infiltration by malignant cells, and other significant observations). The diagnosis of malignancy was established based on bone marrow aspiration/biopsy findings, as well as peripheral blood smear evaluation, complemented by flow cytometry analysis when indicated. Patients with incomplete medical records, prior treatments affecting bone marrow function, or diluted/non-diagnostic bone marrow aspirates were excluded from the analysis. This was a retrospective study that included only pediatric patients with thrombocytopenia for whom bone marrow aspiration was deemed clinically necessary by the treating physician. Therefore, the findings

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may not be generalizable to all patients with thrombocytopenia, particularly those with milder or self-limiting forms of the condition.

Statistical analysis

The extracted data were entered into SPSS statistical software (v. 24). Descriptive statistics were used to summarize demographic, clinical, and laboratory characteristics. Continuous variables were reported as means ± standard deviations or medians with interquartile ranges, depending on distribution. Categorical variables were presented as frequencies and percentages. Comparisons between groups were performed using the Chi-square test for categorical variables and the independent t-test or Mann-Whitney U test for continuous variables, as appropriate. A p-value of less than 0.05 was considered statistically significant. No artificial intelligence tools were used for data collection or statistical analysis. AI-based assistance was applied only for minor language editing and improving the manuscript's readability.

Results

Demographics and pathological findings of patients

A total of 405 pediatric patients with thrombocytopenia underwent bone marrow aspiration during the study period (2016–2022). Of these, 30 cases were excluded due to inadequate sample quality, resulting in a final cohort of 375 patients. The mean age of the study population was 4.79 years (SD = 3.64), with a male predominance (215 cases, 57.3%) compared to females (160 cases, 42.7%). The highest prevalence of thrombocytopenia was observed in the 1 to ≤5-year age group (50.7%), followed by the 5 to ≤10-year age group (22.1%), and infants (<1 year, 15.2%). The lowest frequency was recorded in children older than 10 years (12.0%).

Bone marrow analysis revealed that acute leukemia was the most frequent diagnosis, accounting for 177 cases (47.2%), followed by other hematological and non-malignant disorders. Among the total cases, 196 (52.2%) were classified as non-malignant, whereas 179 (47.8%) were malignant. The distribution of pathological findings is summarized in Table 1. The most prevalent malignant disorder was acute leukemia (n=177, 47.2%), while chronic myeloid leukemia and metastatic disease were the least frequent diagnoses, each with only one reported case (0.3%). These findings underscore the significance of acute leukemia as the leading cause of thrombocytopenia in pediatric patients, with the highest burden observed in children aged 1 to ≤ 5 years. The results highlight the critical role of bone marrow aspiration in differentiating malignant from non-malignant causes, guiding timely diagnosis and appropriate clinical management.

Clinical manifestations of pediatric thrombocytopenia

The analysis of clinical manifestations among 375 pediatric patients with thrombocytopenia from 2016 to 2022 revealed that bleeding manifestations were the most prevalent symptom, observed in 181 cases (48.3%), followed by fever (124 cases, 33.1%), weakness and lethargy (46 cases, 12.3%), and bone pain (44 cases, 11.7%).

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Table 1. Demographics and pathological findings, and types of cytopenia in pediatric patients with thrombocytopenia (n = 375)

Parameter	Frequency	Percentage
Gender		
Male	215	57.3
Female	160	42.7
Age Groups		
<1 year	57	15.2
1 - 5 years	190	50.7
6 - 10 years	83	22.1
>10 years	45	12.1
Pathological findings		
Acute leukemia	177	47.2
Megakaryocytic hyperplasia	99	26.4
Erythroid hyperplasia	25	6.7
No pathological findings	21	5.6
Myelodysplastic changes	16	4.3
Mild excess of lymphoid cells	15	4.0
Eosinophilia	15	4.0
Hemophagocytosis	10	2.7
Myeloid hyperplasia	9	2.4
Monocytosis/ histiocytosis	7	1.9
Plasmacytosis	3	0.8
Gaucher's disease	1	0.3
Chronic myeloid leukemia	1	0.3
Metastasis	1	0.3
Malignancy classification		
Malignant cases	179	47.8
Non-malignant cases	196	52.2
Type of cytopenia		
Isolated thrombocytopenia	91	24.3
Bi-cytopenia (Thrombocytopenia and Anemia)	168	44.8
Bi-cytopenia (Thrombocytopenia and Leukopenia)	27	7.2
Pancytopenia	89	23.7

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Table 2. Frequency distribution of clinical manifestations in children with thrombocytopenia from 2016 to 2022

Clinical manifestation	Frequency	Percentage
Bleeding manifestations	181	44.7
Fever	124	30.6
Weakness and lethargy	46	11.4
Bone pain	44	10.9
Lymphadenopathy	30	8.0
Abdominal pain	25	6.2
Hepatosplenomegaly	24	6.0
Cold symptoms	24	6.0
Pallor	22	5.5
Nausea and vomiting	14	3.5
Loss of appetite	5	1.3
Jaundice	3	0.8
Growth retardation	2	0.5

Other notable findings included lymphadenopathy (8%), abdominal pain (6.7%), hepatosplenomegaly (6.4%), and cold symptoms (6.4%). Less frequent symptoms included pallor (5.9%), nausea and vomiting (3.7%), loss of appetite (1.3%), jaundice (0.8%), and growth retardation (0.5%), with growth retardation being the least common manifestation, occurring in only two cases (Table 2). These findings highlight the predominance of hemorrhagic symptoms in pediatric thrombocytopenia and underscore the diverse clinical presentations that may aid in early diagnosis and management.

Distribution of hematological findings and clinical manifestations in isolated thrombocytopenia

Among the 91 pediatric patients diagnosed with isolated thrombocytopenia, 59 (64.8%) were male, and 32 (35.2%) were female, with the highest prevalence observed in the 1 to 5-year age group. The majority of cases were classified as non-malignant (82 cases, 90.1%), while 9 cases (9.9%) were malignant. Megakaryocytic hyperplasia was the most frequently observed pathological finding, accounting for 62% of cases, and bleeding manifestations were the most common clinical presentation, occurring in 78% of patients. These findings highlight the predominance of benign etiologies in pediatric isolated thrombocytopenia and emphasize the significance of hematological evaluation in children presenting with bleeding symptoms.

Comparison of hematological parameters, clinical manifestations, and pathological findings in pediatric patients with bicytopenia and pancytopenia

In this study, a total of 284 pediatric patients with bicytopenia or pancytopenia were analyzed, revealing distinct hematological and clinical profiles. Among patients with bicytopenia involving low platelets & hemoglobin (n=168), 71.4% had malignant conditions, with acute leukemia as the most prevalent pathological finding, and bleeding symptoms (42.9%) as the most common clinical manifestation. In contrast, patients with bicytopenia involving low platelets & white blood cell (WBC) (n = 27)

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predominantly exhibited non-malignant conditions (77.8%), with megakaryocytic hyperplasia (40.7%) as the most frequent diagnosis and bleeding symptoms (51.9%) as the primary clinical presentation. Among pancytopenic patients (n = 89), malignancy and non-malignancy were nearly equally distributed (49.4% vs. 50.6%), with acute leukemia (49.4%) being the most common diagnosis and fever (44.9%) being the leading symptom. Notably, malignancy was most prevalent in bicytopenia, characterized by low platelets and hemoglobin, whereas non-malignant conditions were more common in isolated thrombocytopenia. These findings underscore the diagnostic significance of cytopenia patterns in distinguishing malignant from non-malignant hematological disorders in pediatric patients (Table 3).

Table 3. Comparison of Hematological Parameters, Clinical Manifestations, and Pathological Findings in Pediatric Patients with Bicytopenia and Pancytopenia

Group	Total cases	Male (%)	Female (%)	Most common age group	Non-malignant cases (%)	Malignant cases (%)	Most common pathological finding	Most common clinical manifestation
Bicytopenia (low platelets & hemoglobin)	168	93 (55.4)	75 (44.6)	1–5 years	48 (28.6)	120 (71.4)	Acute leukemia (71.4)	Bleeding symptoms (42.9)
Bicytopenia (low platelets & WBC)	27	14 (51.9)	13 (48.1)	1–5 years, 5–10 years	21 (77.8)	6 (22.2)	Megakaryocytic hyperplasia (40.7	Bleeding symptoms (51.9)
Pancytopenia	89	49 (55.1)	40 (44.9)	1–5 years	45 (50.6)	44 (49.4)	Acute leukemia (49.4)	Fever (44.9)

Table 4. Association Between Hematological Groups and Clinical Variables (Pearson Chi-Square Test)

Variable	Pancytopenia (P-value)	Bicytopenia (Low Platelets & Hemoglobin) (P-value)	Bicytopenia (Low Platelets & WBC) (P-value)	Isolated Thrombocytopenia (P-value)
Age	0.619	0.319	0.061	0.415
Gender	0.619	0.139	0.223	0.415
Malignancy	0.845	0.000	0.091	**0.000**
Clinical Manifestations				
Hemorrhagic Symptoms	0.000	0.000	N/A	N/A
Fever	0.006	0.001	N/A	N/A
Abdominal Pain	0.048	N/A	N/A	N/A
Pallor	0.014	0.046	N/A	N/A
Lymphadenopathy	0.029	0.006	N/A	N/A
Hepatosplenomegaly	0.033	0.005	N/A	N/A
Bone Pain	N/A	0.001	N/A	N/A

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Association between hematological groups and clinical variables

Statistical analysis revealed several significant associations between hematological groups and clinical variables. Malignancy showed a strong correlation with bicytopenia (low platelets & hemoglobin) ($p < 0.001$) and isolated thrombocytopenia ($p < 0.001$), indicating a potential role of underlying malignancies in these conditions. Hemorrhagic symptoms and fever were significantly associated with pancytopenia and bicytopenia (low platelets & hemoglobin) ($p < 0.001$), suggesting their diagnostic relevance in these hematological abnormalities. Additionally, pancytopenia was significantly correlated with pallor ($p = 0.014$), lymphadenopathy ($p = 0.029$), hepatosplenomegaly ($p = 0.033$), and abdominal pain ($p = 0.048$), while bicytopenia (low platelets & hemoglobin) showed strong associations with bone pain ($p = 0.001$) and hepatosplenomegaly ($p = 0.005$). These findings highlight the significance of clinical manifestations in distinguishing between different hematological conditions in pediatric patients (Table 4).

In contrast, no statistically significant associations were observed between age and any of the hematological groups, with p-values ranging from 0.061 to 0.619. Similarly, gender did not exhibit any significant correlation across pancytopenia ($p = 0.619$), bicytopenia (characterized by low platelets & hemoglobin) ($p = 0.139$), bicytopenia (characterized by low platelets & WBC) ($p = 0.223$), or isolated thrombocytopenia ($p = 0.415$). Malignancy was not significantly associated with pancytopenia ($p = 0.845$) or bicytopenia (low platelets & WBC) ($p = 0.091$), indicating variability in its hematological presentation. Furthermore, specific clinical manifestations, such as hemorrhagic symptoms, fever, and bone pain, lacked sufficient data to determine their associations with specific hematological subgroups (N/A). These findings suggest that while some variables are strongly linked to hematological abnormalities, others may require further investigation to clarify their clinical significance.

Discussion

This study highlights the critical role of bone marrow aspiration in diagnosing pediatric thrombocytopenia, particularly in differentiating between malignant and non-malignant conditions. Acute leukemia was the most prevalent malignant disorder, predominantly affecting children aged 1 to 5 years. Hemorrhagic symptoms and fever were the most common clinical presentations across hematological groups. Cytopenia patterns showed strong diagnostic value; malignancies were mainly associated with bicytopenia (low platelets and hemoglobin), while non-malignant conditions predominated in isolated thrombocytopenia and bicytopenia involving Low Platelets and WBC.

Significant associations between pancytopenia and systemic symptoms, including pallor, lymphadenopathy, and hepatosplenomegaly, confirm the diagnostic relevance of clinical features, consistent with

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findings by Bahadure et al. [12] and Sharif [18].

Studies by Waris [13], Rasheed [14], and Balasubramanian et al. [10] similarly identified acute leukemia as the leading malignancy associated with cytopenias. In contrast, Kumar [11], Singh [16], and Dubey [17] reported infections and megaloblastic anemia as more common causes, likely reflecting regional and nutritional differences. Our study's age distribution revealed that acute leukemia was most common in younger children (1–5 years), which differs from studies that found the peak incidence in older children (6–18 years) [10,11]. This difference may reflect regional variations in genetic predisposition, environmental exposures, referral patterns, or differences in study populations. A key aspect of our study is emphasizing cytopenia patterns as diagnostic markers; bicytopenia involving low platelets and hemoglobin was strongly linked to malignancies, whereas isolated thrombocytopenia or bicytopenia involving Low Platelets and WBC indicated non-malignant conditions. This contrasts with some previous studies in which infectious etiologies and megaloblastic anemia were more commonly reported. One possible explanation is the difference in referral patterns: in our center, bone marrow aspiration is generally performed for patients with more severe or persistent cytopenias, which may enrich the proportion of malignant cases. Additionally, regional variations in nutritional deficiencies, prevalence of infectious diseases, and diagnostic thresholds may account for the higher frequency of non-malignant causes in other reports. Differences in study design, sample size, and inclusion criteria may also contribute to this discrepancy .Our study found that isolated thrombocytopenia was predominantly associated with non-malignant conditions. Some previous reports have suggested a higher proportion of malignancy in cases of isolated thrombocytopenia. This discrepancy may be due to differences in study populations, referral patterns, or inclusion criteria. In our center, bone marrow aspiration is typically performed for children with persistent or severe cytopenias, which may select for cases more likely to reveal underlying malignancy, while milder isolated thrombocytopenia cases are often managed without invasive procedures. Regional variations in disease prevalence and diagnostic practices may also contribute to differences in reported malignancy rates. These findings align with those of Waris [13] and Bahal [15], who emphasized the importance of structured bone marrow evaluation in distinguishing between reversible conditions and hematological malignancies. Our results also support those of Bilal [19] and Neunert et al. [20], who reported that bone marrow biopsy is generally unnecessary in typical ITP cases, unless clinical red flags are present. This supports reserving invasive diagnostics for patients with a high clinical suspicion of malignancy.

The etiology of thrombocytopenia remains diverse, with studies such as Puri [21] and Choudhary [22] identifying infections, megaloblastic anemia, and dengue as major causes. The findings of this study are most applicable to pediatric patients with severe

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thrombocytopenia that prompts marrow evaluation and may not be generalizable to all thrombocytopenic children. While infectious causes were not the focus here, our findings suggest that cytopenia patterns, rather than isolated thrombocytopenia, should raise suspicion for malignancy. Emerging factors such as macrothrombocytopenia and Coronavirus disease 2019 (reported by Puri) [21] further demonstrate the evolving nature of hematological disorders and the need for updated diagnostic frameworks. Overall, this study contributes to the growing body of evidence supporting the diagnostic value of bone marrow aspiration in pediatric thrombocytopenia. By identifying specific cytopenia patterns linked to malignancy, our findings support earlier and more accurate differentiation between benign and malignant hematological conditions, ultimately improving patient outcomes. Future research should focus on integrating molecular and genetic biomarkers into routine diagnostics to enhance risk stratification, refine prognosis, and enable the development of personalized treatment strategies.

Conclusion

This study provides valuable insights into the clinicopathological spectrum of pediatric thrombocytopenia, emphasizing the diagnostic importance of bone marrow aspiration in distinguishing malignant from non-malignant causes. Acute leukemia was the most common malignancy, particularly in children aged 1–5 years, and was strongly associated with bicytopenia (low platelets and hemoglobin). In contrast, isolated thrombocytopenia and bicytopenia involving low platelets and WBC were mainly linked to non-malignant conditions. Pancytopenia correlated with systemic symptoms, serving as an important clinical indicator of malignancy. While the study is limited by its single-center, retrospective design and the lack of molecular analysis, it highlights the need for region-specific diagnostic protocols. Future multicenter studies incorporating molecular and non-invasive biomarkers are recommended to improve diagnostic accuracy and clinical outcomes in children with thrombocytopenia.

Ethical Considerations

The study adhered to ethical principles outlined in the Declaration of Helsinki. The study protocol was approved by the Pathology Department and received ethical approval from the Institutional Review Board (IRB) of Shahid Sadoughi University of Medical Sciences, and was conducted with the approval of the Ethics Committee at the Faculty of Medicine of Shahid Sadougi University of Medical Sciences, Yazd (IR.SSU.MEDICINE.REC.1401.162) .Informed consent was waived due to the retrospective nature of the study design. Patient confidentiality was ensured by anonymizing all data throughout the collection and analysis process.

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The authors have no funding to disclose.

Conflicts of Interest

The authors declare no conflict of interest.

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Data Availability Statement

The data presented in this study are available on request from the corresponding author.

Authors' Contributions

Conceptualization: S.E., and F.Z.M.; methodology: S.E., F.Z.M., and S.E.; formal analysis: S.E., and F.Z.M.; investigation: S.E., F.Z.M., and S.E.; resources: S.E., F.Z.M., and S.E.; data curation: S.E., S.E., and F.Z.M.; writing—original draft preparation: S.E., F.Z.M., and S.E.; writing—review and editing: S.E. and S.E.; supervision: S.E. All authors have read and agreed to the published version of the manuscript.

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