Original Research Paper PATHOLOGY

A STUDY OF ETIOLOGICAL, CLINICAL AND HEMATOLOGICAL CORRELATION IN PAEDIATRIC PATIENTS WITH PANCYTOPENIA/ BICYTOPENIA

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ABSTRACT

Background: The etiology of bicytopenia/pancytopenia varies widely in paediatric patients, ranging from transient marrow viral suppression to marrow infiltration by fatal malignancy. Depending on the etiology, the clinical presentation can be with fever, pallor or infection. Knowing the exact etiology is important for specific treatment and prognosis.

Aims: To study of etiological, clinical and hematological correlation in paediatric patients with pancytopenia/bicytopenia.

Materials and Methods: A review of bicytopenic and pancytopenic children referred for bone marrow examination from December 2011 to October 2013 at S. N Medical college Agra was done. Detailed history, clinical examination and hematological parameters at presentation were recorded.

Conclusion: During the study period, a total of 32 children were referred for bone marrow examination for different indications. Of these, 17(53%) had pancytopenia and 15 bicytopenia (47%). In patients with bicytopenia, Anemia and thrombocytopenia was the commonest combination (11 patients) followed by leucopenia and the thrombocytopenia and anemia and leucopenia (2 patients each). Among the children who presented with bicytopenia fever (93%), pallor (40%) generalized weakness (33%), bleeding (20%) were the commonest clinical findings. Fever (82%) was the commonest clinical finding in patients who presented with pancytopenia ,followed by pallor (64%) , splenomegaly (41%) , bleeding manifestations(29%) and hepatomegaly(29%).

KEYWORDS : Pancytopenia, Bicytopenia ,Bone marrow

INTRODUCTION

Peripheral cytopenia is defined as reduction in either of the cellular elements of the blood, i. e red blood cells, white blood cells or platelets. Bicytopenia is the reduction of any of the two cell lines and pancytopenia is reduction of all the three (haemoglobin<10g/dl, absolute neutrophil count 1.5×10^9/L and platelet count $<100 \times 10^9$/L)[1] . Commonest form of bicytopenia is anemia and thrombocytopenia followed by anemia and leucopenia.

The etiology of bicytopenia and pancytopenia varies widely in children, ranging from transient marrow viral suppression to marrow infiltration by life threatening malignancies. Pancytopenia can result from a failure of production of hematopoietic progenitors, their destruction, or replacement of the bone marrow by tumor or fibrosis. Although selective cytopenias are important clinical entities, pancytopenia is a loss of all marrow elements. Pancytopenia can be constitutional, arising as a consequence of an inherited genetic defect affecting hematopoietic progenitors, or can be acquired as a consequence of either direct destruction of progenitors, immune mediated damage to either hematopoietic progenitors or their nurturing microenvironment, or suppression of or crowding out of progenitors by tumor cells or fibrosis.

Although Fanconi anemia is the best-recognized constitutional pancytopenia, a number of other infrequent genetic disorders have also been implicated. These genetic syndromes include various modes of inheritance and may be associated with a number of congenital abnormalities, especially of the bones, kidneys, and heart. Because the hematologic manifestations of the congenital pancytopenias may not become manifested until the first years to even decades of life, a genetic predisposition to bone marrow failure should be considered in all cases of aplastic anemia in children. These disorders can be autosomal recessive (e. g., Fanconi anemia, dyskeratosis congenita), X linked, or autosomal dominant (e. g., dyskeratosis congenita).

Severeal of these genetic disorders may present initially with a single cytopenia and progress to pancytopenia (Swaschman-Diamond syndrome, amegakaryocytic thrombocytopenia, reticular dysgenesis). In addition, a number of inheritable familial marrow dysfunction syndromes have been associated with pancytopenia (which can also be autosomal recessive, autosomal dominant, or X linked), and aplastic anemia also occurs in association with other genetic disorders. Thus, pancytopenia can be either the primary disease manifestation or can emerge as a rare complication during the course of another illness. Because of the chromosomal fragility or defective repair mechanisms that may be associated, several of these disorders can also be complicated by cancer or other organ dysfunction(s)[2].

Most common non malignant cause of acquired pancytopenia is aplastic anemia followed by megaloblastic anemia. Among the malignant causes acute leukemia is most common. Pancytopenia caused by marrow replacement is seen to occur in leukemia. Pancytopenias can be caused by various drugs-chemotherapeutic agents, antiepileptics(hydantoin, carbamazepine) NSAIDS (phenylbutazone, ibuprofen, diclofenac. Prolonged cytopenias can occur after infection with many of the hepatitis viruses, Epstein Barr Virus, Cytomegalovirus. Pancytopenia is may occur after a single high dose of radiation.[3]

Non malignant conditions causing bicytopenia are Immune Thrombocytopenic Purpura, megaloblastic anemia, marrow hypocellularity and visceral leishmaniasis. Commonest malignant condition associated with bicytopenia is leukaemia, acute lymphoblastic leukemia being commoner.

The main presenting features in children with pancytopenia are fever and pallor. Other common symtoms are petechial rash, bleeding and bone pains. Dyspnea on exertion, easy bruising, epistaxis, gingival bleeding, headache may be the presenting features.[3] Oral infections in the form of gingivitis, tonsillitis and pharyngitis may be found.[4] Pancytopenia results in increased risk of fatigue, cardiac failure and infection. On clinical examination hepatomegaly and splenomegaly may be found.

Careful examination of peripheral blood smear for RBC, leukocyte and platelet morphology is important. A reticulocyte count should

be done to assess erythropoietic activity. Bone marrow examination should include bone marrow aspiration along with biopsy wherever indicated. Marrow should be carefully examined for morphology and cellularity.

Thus there are different etiologies causing pancytopenia / bicyto penia which cause similar clinical manifestations in the patients. Various studies have been done in this respect in adults but in paediatric patients this information is scarce. Therefore this study is being undertaken to study the etiological, clinical and haema tological correlation in paediatric patients with pancy topenia / bicytopenia

AIMS & OBJECTIVES

1. To study the etiological spectrum of pancytopenia or bicytopenia in children below 15 years of age.
2. To evaluate haematological parameters including bone marrow aspiration/ biopsy

Material & Methods

A prospective study was conducted in the Department of Pathology, S.N. Medical College, Agra. During the study period (December 2011 to October 2013) children below the age of 15 years presenting with the signs and symptoms of pancytopenia / bicytopenia were referred from the department of pediatrics for bone marrow examination.

The criteria for selection of patients were as follows;

1. Children below 15 years were only included.
2. Children were suffering from pancytopenia/bicytopenia as confirmed by hematological parameters.

Total number of cases in the study was 32. Detailed history, clinical examination and hematological parameters at presentation were recorded.

Hematological profile included hemoglobin, red cell indices, total and differential leucocyte counts, platelet count and peripheral blood smear morphology. Peripheral blood smear was drawn using blood from a finger prick or a venous sample. It was then stained with Leishman stain and then observed microscopically.

Bone marrow aspiration / biopsy was carried out as per clinical indication. Special stains like Perls stain for iron were done to assess the iron stores: Trephine biopsy of the bone marrow was carried out in cases where aspirate was not successful. Biopsy was carried out on the iliac crest, posterior approach was preferred. Chi-square test was used for statistical analysis. A P value of <0.01 was taken as statistically significant.

OBSERVATION

The present study was conducted at Pathology Department, S.N. Medical College,Agra. Bone marrow examination of children (<15 years) who presented with pancytopenia / bicytopenia was done in central clinical laboratory for hematological evaluation.

Total No. of cases = 32

No. of cases Pancytopenia	17	53%
No. of cases of bicytopenia	15	47%

Table 2: Distribution of cases according to age – panctopenia

Age (Yrs.)	No. of Cases	Male	Female
0-5	01 (6%)	01	0
6-10	03 (18%)	02	01
11-15	13 (76%)	06	07

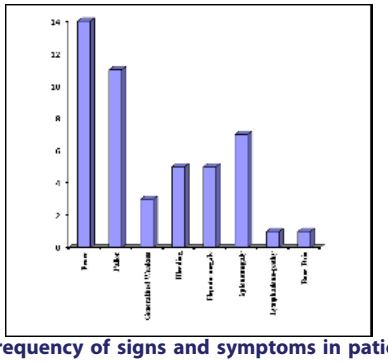
Table 3: Distribution of cases according to age – Bicytopenia

Age (Yrs.)	No. of Cases	Male	Female
0-5	3 (20%)	02	01
6-10	04 (27%)	-	04
11-15	08 (53%	04	04

Table 4: Peripheral blood findings in children with bicyto penia and pancytopenia

	Bicytopenia	Pancytopenia
Hemoglobin < 10g/dl	15 (100%)	18 (100%)
TLC < 4000/mm^3	06 (40%	17 (100%)
Platelet < 1,00,000/mm^3	14 (93%)	17 (100%)
Circulating blasts	01 (05%)	03 (20%)

Graph 1: Frequency of signs and symptoms in patients with pancytopenia



Graph 2: Frequency of signs and symptoms in patients with bicytopenia

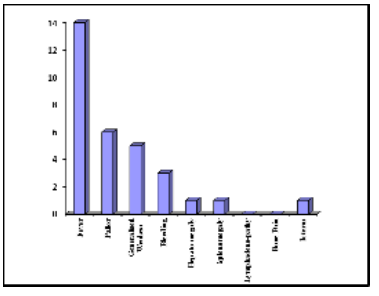
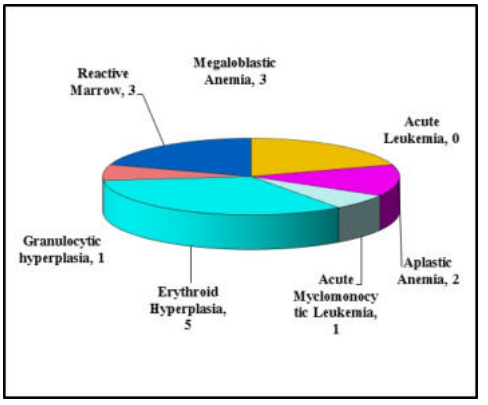


Table 5: Etiological profile of children with pancytopenia

Etiology	Cases	Percentage
Megaloblastic Anemia	6	35%
Aplastic Anemia	4	23%
Acute Leukemia	3	19%
Acute myelomonocytic leukemia	0	00%
Erythroid hyperplasia	04	23%
Granulocytic hyperplasia	0	00%
Reactive marrow	0	00%

Fig 1: Etiological profile of children with bicytopenia



DISCUSSION

A total of 32 cases were studied. These were children who presented with pancytopenia or bicytopenia. Of these 17 cases were those of pancytopenia while 15 were those of bicytopenia.

Age, gender-wise incidence, presenting complaints, peripheral blood pictures, bone marrow aspiration smears and various causes of pancytopenia / bicytopenia were studied in all cases and observations were compared with those studies published.
Male to female ratio in patient with pancytopenia :

Overall male to female ratio in our study was 1.1:1.

The common physical findings in pancytopenic patients in various studies. Fever (82%), was found to be the most common cause followed by pallor (64%), followed by splenomegaly (41%) and hepatomegaly (29%). This is fairly comparable with Naseem et al (2010)9 and Khan FS (2012)[8] et al where fever followed by pallor and organomegaly are the commonest physical findings.

Table 6: Comparison according to etiology in patient with panc ytopenia

	Bhatnagar et al [5](2005)	Jha et al [6](2008)	Gayatri et al [7](2008)	Khan et al [8](2012)	Present Study
Megaloblastic Anemia	31 (28.4%)	35 (23.6%)	77 (74%)	37 (12.2%)	06 (35%)
Aplastic Anemia	30 (21%)	43 (29%)	19 (18%)	86 (30.8)	04 (23%)
Acute Leukemia	23 (20%)	32 (21%)	04 (4%)	30 (32.2%)	3 (19%)
Normal Marrow	1 (1%)	5 (3.8%)	-	16 (5.7%)	-
Erythroid hyperplasia	11 (10%)	29 (19%)	-	15 (5.3%)	04 (2.3%)
Reactive bone marrow	-	-	-	10 (3.5%)	-
Non Hodgkin's lymphomia	-	-	-	1 (0.3%)	-
Metastatic Deposits	-	01 (0.5%)	-	2 (0.7%)	-
Non Specific	-	-	-	16 (5%)	-
Infections	23 (21%)	08 (5.4%)	04 (4%)	-	-
Hodgkin's disease	-	-	-	6 (2%)	-
Total number of cases	109	148	104	-	17

The present study is almost comparable with Bhatnagar et al (2005)5 and Gayatri et al (2008)7 where the most common cause of pancytopenia was Megaloblastic anemia, followed by aplastic anemia, followed by acute leukemia. Erythroid hyperplasia was a significant proportion in the present study.

Table 7: Comparison of physical findings in patients with bicytopenia :

Findings	Naseem et al [9](2010)	Present Study
Fever	69.2	93
Pallor	40.9	40
Bleeding	29.0	20
Bone pain	9.5	-
Hepatomegaly	69.2	6
Splenomegaly	60.5	06
Lymphadenopathy	41.8	-
Generalized Weakness	-	33
Icterus	-	06

In the above table present study is comparable with Naseem et al in the physical findings of fever. Pallor and bleeding. However organomegaly was found to be lesser. (Splenomegaly 6%, hepatomegaly – 6%). Lymphadenopathy was not observed in any case.

Table 8: Comparison of etiologies in bicytopenic patients

	Naseem et al [9](2010)	Present Study
Aplastic anemia	10 (2.95)	02 (13%)
Megaloblastic anemia	1`3 (3.7%)	03 (20%)
Erythroid hyperplsia	-	05 (33%)
Infection	8 (2.3%)	-
Storage Diseases	5 (1.4%)	-
Immune Thrombocytopenic purpura	18 (5.2%)	-
Acute Leukemia	232 (66.9%)	-
Acute myelomonocytic Leukemia	6 (1.7%)	1 (7%)
Reactive bone marrow	-	03 (20%)
Non Specific	55 (16%)	1 (7%)
Total Cases	347	15

Problems areas were:

- Sub-categorization of leukemia. In some cases there was difficulty in distinguishing between myeloblasts and lymphoblasts.
- In categorizing megaloblastic anemia from mixed reaction erythroid hyperplasia.
- Obtaining clinical history, hematological parameters of the patients.
- In cases of dilute and hemorrhagic aspirate.

Complicatons Of Procedure:

No significant complication of the procedure was noted except mild pain which was relieved by giving NSAIDS.

SUMMARY & CONCLUSION

The present work was done by bone marrow examination of 32 pediatric patients suffering from pancytopenia / bicytopenia during a two year period (2011-2013) in the central clinical laboratory of the Department Pathology in collaboration with the Department of Pediatrics, S.N. Medical College, Agra.

Pediatric patients (under 15 years of age) who filled the criteria for selection were subjected to detailed clinical history and physical examination. Hematological parameters at presentation were recorded. The patients were them subjected to bone marrow examination. Bone marrow aspiration was done in most cases after giving local anesthesia which was secured by 2% xylocaine. The smears were stained with May Grunwald Giemsa stain (and Perl's stain, if required). Only in difficult cases patient was subjected to bone marrow biopsy and H&E staining was done.

- Of the 32 patients evaluated, 17 presented with pancytopenia and 15 with bicytopenia.
- In the age group 5-15 years most patients were between 10-15 years.
- Male to female ratio in case of patients with pancyt openia was 1.1:1, while that in case of bicytopenia was 1:1.5.
- In patients with bicytopenia, 15 patients had anemia and 14 had thrombocytopenia, and 6 had leucopenia. Anemia and thrombocytopenia was the commonest combination (11 patients) followed by leucopenia and the thrombocytopenia and anemia and leucopenia (2 patients each).
- Among the children who presented with bicytopenia fever (93%), pallor (40%) generalized weakness (33%), bleeding (20%) were the commonest clinical findings.
- Fever (82%) was the commonest clinical finding in patients who presented with pancytopenia, followed by pallor (64%) ,

splenomegaly (41%), bleeding manifestations(29%) and hepatomegaly (29%).

- Hematological diagnosis of the 17 pancytopenic children: 6 cases were megaloblastic anemia, 4 aplastic anemia, 4 erythroid hyperplasia and 3 acute leukemia.
- Of the 15 bicytopenic patient 5 were erythroid hyperplasia, 3 megaloblastic anemia, 3 reactive marrow, 2 aplastic anemia, 1 acute myelomonocytic leukemia and 1 granulocytic hyper plasia.
- Overall finding were consistent with those reported in literature.
- The problems encountered were –
- Sub-categorization of acute leukemia. In some cases there was difficulty in sub-categorization of leukemia. Special stains could be useful in such cases.
- In differentiating megaloblastic anemia from mixed reaction erythroid hyperplasia. Perls stain is useful is such cases.
- Uncooperative patients were difficult to handle and expert aspiration were therefore required.

Complications were minimal and included only minimal discomfort or mild pain or slight bleeding from the site of bone marrow examination. Clinically the complications were of no consequence.

It can be concluded that etiology of pancytopenia and bicytopenia varies widely in children. It can range from transient marrow suppression to malignancy. Bone marrow examination is an important tool in the diagnosis and early initiation of treatment in vulnerable pediatric patients.

Conflict of interest: NONE

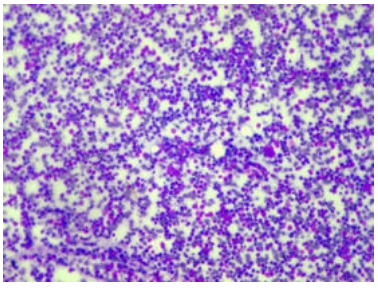


Photo1:Erythroid Hyperplasia in megaloblastic anemia(MGG 10X10).

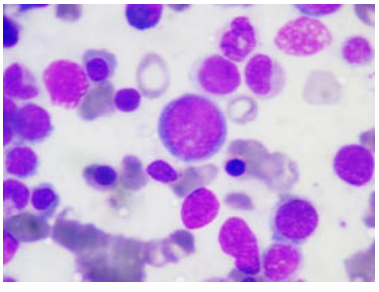


Photo2: Megaloblast (MGG 10X100)

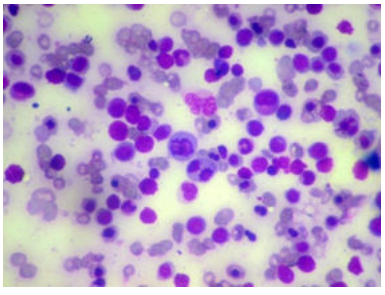


Photo 3: Dyserythropoiesis in megaloblastic anemia(MGG Stain 10 X 40)

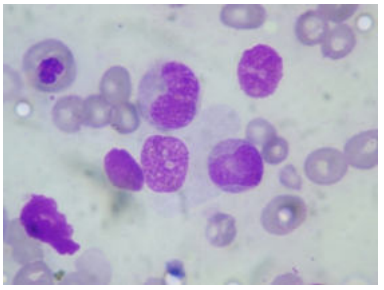


Photo 4: Giant metamyelocyte in megaloblastic anemia (MGG, 10 X 100)

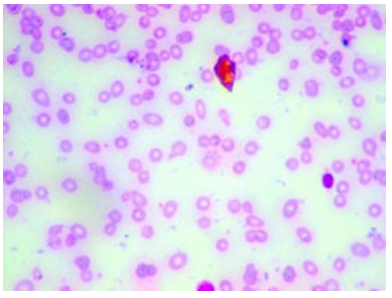


Photo5: Macrocytes,macrovalocytes,tear drop cells in megaloblastic anemia peripheral blood picture (Leishman , 10 X 40).

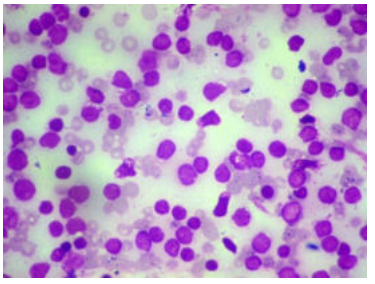


Photo 6: Lymphoblasts in Acute lymphoblastic leukemia(MGG, 10 X 40).

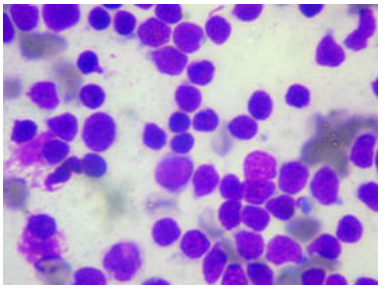


Photo 7: Blasts show scant cytoplasm,condensed chromatin and inconspicuous nucleoli . (MGG, 10 X 100)

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