

Original Research Article

Clinico-haematological profile and etiology of bi/pancytopenia in children aged six months to eighteen years

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ABSTRACT

Background: Bicytopenia/Pancytopenia is frequently found in our clinical practice having diverse etiologies, and still the optimal diagnostic approach remains undefined. Etiologies of bi/pancytopenia vary with genetic conditions, geographical area and the prevalence of infections and nutritional deficiencies in the community. The objective is to identify clinico-haematological profile and etiologies of bi/pancytopenia in children aged 6 months to 18 years from the region of western India.

Methods: A descriptive cross-sectional study was done during May 2016 to April 2018. All patients were investigated for identifying a cause of bi/ pancytopenia. Pancytopenia is defined as haemoglobin of <10 gm%, total leukocyte count <4000/mm³ with or without absolute neutrophil count of <1500/mm³, platelet count <1 lac/mm³. Bicytopenia is defined as any reduction in any two cell lineages.

Results: From 6109 hospitalized patients, 95 cases of bicytopenia and 79 of pancytopenia were evaluated. Fever, pallor, mucosal and skin bleeding were the predominant symptoms observed. Pallor was found in 90.8% cases followed by hepatomegaly in 64.8% and splenomegaly in 40.8% cases. From both bi/ pancytopenia groups, infective diseases were the common causes found in 87 (50%) cases, followed by malignancies in 28 (16%) cases and megaloblastic anemia in 22 (12.64%) cases. Aplastic anemia was found in 10 cases. Dengue fever and malaria were two common causes observed in bicytopenia group while megaloblastic anemia and acute lymphoblastic leukaemia were found in pancytopenia group.

Conclusions: Infective diseases were common causes giving rise to bi/pancytopenia. Pancytopenia is predominantly noticed in the conditions related to bone marrow production or due to infiltrative disorders.

Keywords: Bicytopenia, Pancytopenia, Bone marrow aspiration

INTRODUCTION

A definitive diagnosis is needed in all cases and clinical laboratory testing plays a crucial role in reaching to a diagnosis. Blood count is an integral part of most of the clinical diagnosis. Complete blood count is a basic screening test in both symptomatic and asymptomatic individuals who undergo for check-ups.

The word pancytopenia denotes a quantitative decrease of erythrocytes, leucocytes and platelets in circulating

blood. A word bicytopenia is defined as decrease in any two cellular elements in blood.

The value of haemoglobin (Hb) and total white blood cell count (WBC) varies with age in pediatric population. A value of Hb <10 gm/dl, WBC of less than 4000/mm³ or absolute neutrophil count (ANC) <1500/mm³ and platelet count (PC) of less than 1 lac/mm³ is reasonably considered an important hematological finding to investigate further in a case of pancytopenia for all age groups.¹⁻⁶

The true incidence is difficult to know as many cases are mild and transient type, which are treated on outdoor basis. As reported by Dubey et al the incidence was 2.9% amongst all admitted indoor patients. A study done by Singh et al found the incidence of 1.86%.^{6,7} Viral infections, megaloblastic anemia and cytotoxic drugs are some common causes of pancytopenia observed in children.

The etiology of bicytopenia and pancytopenia varies widely in children, ranging from transient marrow viral suppression to marrow infiltration by life-threatening malignancy. Pancytopenia results due to bone marrow failure or due to peripheral destruction and/or sequestration. Megaloblastic anemia was a common treatable cause found from India.^{5,7}

Present study is undertaken to identify clinico-haematological profile and etiologies of pancytopenia/bicytopenia in children aged 6 months to 18 years from the region of Western India where there is a surge of dengue fever cases and most people are vegetarian with their dietary habits.

METHODS

A hospital based cross sectional study was carried out at KGP Children hospital, Vadodara between May 2016-April 2018 where 174 patients who fulfilled the inclusion criteria were included after the approval from the Ethics committee and scientific committee.

Inclusion criteria

All children in the age group of 6 months to 18 years and who had pancytopenia defined as Hb<10 gm%, WBC<4000/mm³ with or without ANC<1500/mm³, platelet count<1 lac/mm³ or bicytopenia defined as any reduction in any two cell lineages, were included. Cases were included only if written consent for bone marrow aspiration was given.

Exclusion criteria

Patients who were diagnosed cases of malignancies and on chemotherapy, radiotherapy had been excluded.

Methodology

Patients were evaluated as per the history of presenting symptoms and examined completely as per the proforma after obtaining informed consent. Clinical details for presenting complaints were inquired and observed. A thorough physical examination was done to detect any presence of pallor, jaundice, hyperpigmentation of skin especially knuckles and extremities, hepatosplenomegaly, lymphadenopathy, bony tenderness, gum hypertrophy, skin rashes, petechiae, purpura, ecchymosis, mucosal bleeding, glossitis, joint swelling and oedema.

Basic haematological investigations like complete blood count and peripheral smear were done in all cases. Complete blood count was done through automated blood cell counter machine. Peripheral smear examination was done by using Wright-Giemsa stain. Bone marrow aspiration and biopsy was done wherever necessary and when no obvious cause was detected. Stained slides were examined for the bone marrow cellularity, structure, focal lesions and marrow fibrosis. Immunohistochemistry was done in selected cases as needed.

Statistical methods

Student’s unpaired t test is used to compare two groups of mean. P value is considered significant at 5% level for all the tests. ANOVA test is applied to analyse the mean values between two or more unmatched groups.

RESULTS

Out of 6109 admitted patients, 174 cases were found with bi/pancytopenia making an incidence of 2.84%. Out of 174 cases, bicytopenia was found in 95 (54.6%) cases and pancytopenia in 79(45.4%). As against 105 male cases, 69 were female cases.

From 95 cases of bicytopenia, little over one third 35 (36.8%) cases were found in the age group of 6 months to 2 years, and only 16 (16.8%) were found in the age group of 10-18 years. Among 79 cases of pancytopenia, the distribution of cases was more or less similar in the various age groups (Table 1).

Table 1: Age and gender distribution of cases of bicytopenia and pancytopenia.

Age group (in yrs)	Bicytopenia			Pancytopenia		
	Male (%)	Female (%)	Total (%)	Male (%)	Female (%)	Total (%)
6 months- 2 yrs	18 (32.1)	17 (43.6)	35 (36.8)	8 (16.3)	9 (30)	17 (21.6)
>2-5	18 (32.1)	8 (20.5)	26 (27.4)	15 (30.6)	4 (13.4)	19 (24)
>5-10	10 (17.9)	8 (20.5)	18 (19)	12 (24.5)	10 (33.3)	22 (27.8)
>10-18	10 (17.9)	6 (15.4)	16 (16.8)	14 (28.6)	7 (23.3)	21 (26.6)
Total (n=174)	56 (59)	39 (31)	95 (54.6)	49 (62)	30 (38)	79 (45.4)

Fever, pallor and mucosal/skin bleeds were predominant symptoms found along with other associated symptoms

in the cases of bi/pancytopenia. On examination pallor, jaundice, skin and mucosal bleeding, generalized

lymphadenopathy, hyper pigmented knuckles and extremities, hepatomegaly, splenomegaly were prominent signs observed (Figure 1).

Infective etiologies such as dengue fever and malaria were predominant causes observed in bicytopenia group, whereas megaloblastic anemia (MA) and acute lymphoblastic leukaemia (ALL) were two most common causes found in pancytopenia.

In both the groups, infective etiologies of various diseases were found in 62 (65.3) cases of bicytopenia and 25 (31.6) cases of pancytopenia (Table 2). One case from

pancytopenia group had co infection of dengue fever and malaria.

On comparing haemoglobin (Hb) values in the groups of bicytopenia and pancytopenia; Hb of <5 gm/dl was found more in pancytopenic group (30.4) than bicytopenia group (18.9) cases; total count (TC) of <1000/mm³ was not found in bicytopenia group. Platelet count (PC) <20,000/mm³ was found in 21 (26.5) cases of pancytopenia as compared to 15 (15.7) cases of bicytopenia. Corrected reticulocyte counts of <1 was significantly higher in pancytopenia (64.7) as against (35.3) in bicytopenia group.

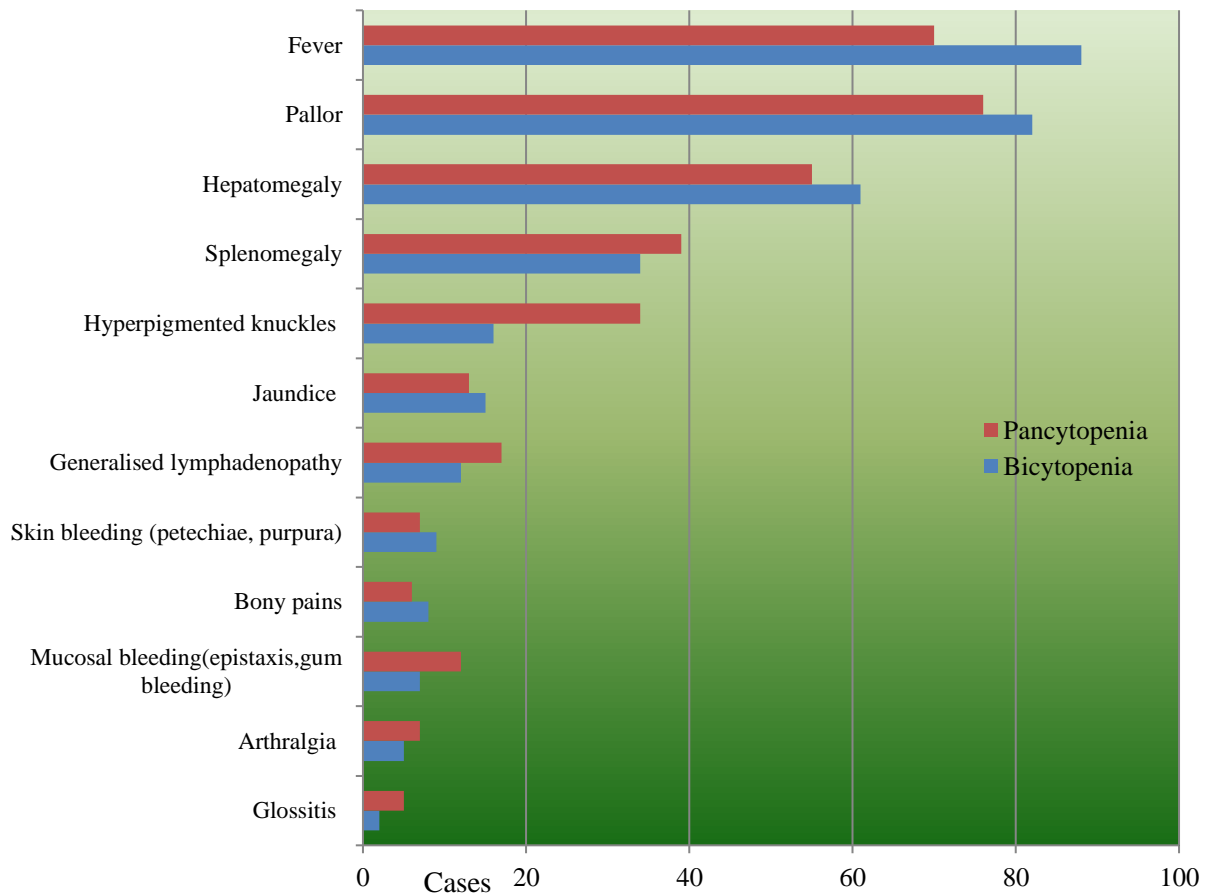


Figure 1: Clinical profile in bicytopenia and pancytopenia cases

Table 2: Distribution of various etiologies in cases of bicytopenia/pancytopenia.

Etiology	Bicytopenia n=95 (%)	Pancytopenia n=79 (%)	Total n=174 (%)
Malaria	18 (18.95)	11 (13.9)	29 (16.67)
Dengue fever	24 (25.26)	4 (5.06)	28 (16.09)
Acute lymphoblastic leukaemia	9 (9.47)	16 (20.25)	25 (14.37)
Megaloblastic anemia	5 (5.26)	17 (21.52)	22 (12.64)
Aplastic anemia	0 (0)	10 (12.65)	10 (5.75)
Septicaemia	6 (6.32)	4 (5.06)	10 (5.75)
Sickle cell anemia	8 (8.42)	1 (1.27)	9 (5.17)
Swine flu	7 (7.37)	0 (0)	7 (4.02)
IITP*	6 (6.32)	0 (0)	6 (3.45)

Continued.

Etiology	Bicytopenia n=95 (%)	Pancytopenia n=79 (%)	Total n=174 (%)
Enteric Fever	3 (3.16)	3 (3.8)	6 (3.45)
Viral fever	3 (3.16)	2 (2.54)	5 (2.87)
Hypersplenism	0	4 (5.06)	4 (2.08)
Nutritional (Dimorphic) anemia	4 (4.21)	0	4 (2.08)
Viral hepatitis- A	1 (1.05)	1 (1.27)	2 (1.04)
Kikuchi disease	1 (1.05)	0	1 (0.57)
Progressive familial intrahepatic cholestasis	0	1 (1.27)	1 (0.57)
Gaucher disease	0	1 (1.27)	1 (0.57)
Homocystinuria	0	1 (1.27)	1 (0.57)
Varicella	0	1 (1.27)	1 (0.57)
Acute promyelocytic leukaemia	0	1 (1.27)	1 (0.57)
Burkitt lymphoma	0	1 (1.27)	1 (0.57)
Non Hodgkin lymphoma	0	1 (1.27)	1 (0.57)

*ITP: immune thrombocytopenia

Table 3: Comparison of various laboratory parameters of bicytopenia with pancytopenia.

Variable	Cytopenia	N	Mean	SD	t-value	pvalue
Haemoglobin (gm/dl)	Bicytopenia	95	7.51	2.39	3.619	<0.001
	Pancytopenia	79	6.18	2.45		
MCV*(fl)	Bicytopenia	95	74.37	9.70	-3.045	0.003
	Pancytopenia	79	79.31	11.68		
Total count /mm ³	Bicytopenia	95	18587.26	45025.12	3.112	0.002
	Pancytopenia	79	2811.27	854.99		
Platelet count /mm ³	Bicytopenia	95	90558.21	112645.62	3.725	<0.001
	Pancytopenia	79	42091.06	28486.25		
Reticulocyte count %	Bicytopenia	30	0.72	0.56	2.154	0.034
	Pancytopenia	52	0.50	0.38		

*MCV: mean corpuscular volume; SD: standard deviation

Table 4: Mean and standard deviation of complete blood count in various subgroups of aetiologies of bi/pancytopenia.

Etiologies	N= 174	Mean Hb (gm/dl)	SD	Mean TC (cells/mm ³)	SD	Mean PC (cells/mm ³)	SD
Infections	87	8.21	2.04	7837.47	9012.20	84870.11	97594.84
Malignancy	28	5.74	2.50	34,948	80377.60	54145.35	95970.39
*MA	22	5.14	1.85	3847.27	1859.96	82037.72	125724.81
Aplastic anemia	10	4.49	2.12	2805.00	851.85	16993.00	19250.14
Others	27	6.22	2.07	7955.55	5921.17	64744.44	48544.59
Total	174	6.9036	2.49	11,425	34115.95	72543.85	93781.68

*MA: megaloblastic anemia; Hb: haemoglobin; TC: total count; PC: platelet count; SD: standard deviation

Hb, mean corpuscular volume (MCV), TC, corrected reticulocyte count and PC on admission were compared between bicytopenia and pancytopenia; the “p” value was statistically significant ($p < 0.05$) in all the variables as shown in (Table 3).

Etiologies of both bi/pancytopenia were divided into subgroups such as infective, malignancy, aplastic anemia, megaloblastic anemia (MA) and miscellaneous groups. Infective etiologies were the commonest cause found in 87(50) cases followed by malignancies in 28 cases and megaloblastic anemia in 22 cases. Aplastic anemia was

found in 10 cases only. The lowest mean age of 4.2 years was found in megaloblastic anemia. Hb, TC and PC were lowest in aplastic anemia and Hb was on higher side in infectious sub group (Table 4). On applying ANOVA Test to compare between the subgroups and within groups of various etiologies for the different variables, it was found that “p” value was significant in variables of Hb, MCV and total count.

In the major subgroups of etiologies of pancytopenia cases, out of 79 cases, infectious etiologies were found in 25(31.6), followed by malignancy in 19(24), MA in

17(21.6), AA in 10 (12.7) cases. MA was mainly presented in the age group from 6 months to 2 years. Hb <5 gm/dl was found mainly in cases of aplastic anemia (60) and megaloblastic anemia (53). Out of 10 cases of aplastic anemia, 7 (70) cases had platelet count of less than 20,000/mm³. Lower values of Hb was observed in AA and MA subgroups than infectious and malignancy groups.

Out of 42 bone marrow aspiration with biopsies, 27 cases had malignancies of which hyper cellular marrow was seen in 23 (85) cases and 4 (15) cases had normocellular marrow and none of the case had hypocellular marrow. Hypocellular marrow was seen in all cases of aplastic anaemia and 1 case with Gaucher disease.

DISCUSSION

The clinical profile and etiology leading to bi/pancytopenia might vary in different population and there could be differences in the incidence, etiology, clinical features, age of presentation, depending on geographical distribution, prevalence of diseases and nutritional deficiencies, dietary habits and genetic conditions.

Out of 6109 patients admitted, 79(45.4%) cases had pancytopenia and 95(54.6%) cases had bicytopenia making an incidence of 1.3% and 1.55% respectively. Incidence of pancytopenia among various studies done by Dubey et al, Memon et al, Jan et al, Singh G et al, Rathod et al were 2.9%, 3.57%, 1.4%, 1.86%, 2.02% respectively. These were all hospital based studies.^{1,6-9} A study done by Ejaz et al found the mean age of presentation of 7±3.3 years, while Sharif et al reported it to be 4.9±4.1 years. We have found a mean age of presentation of 5.28±4.36 years in bicytopenia group and 6.77±4.72 years in pancytopenia.^{10,11}

The most common symptom from bi/pancytopenia groups was fever, reported in 88 (92.6%) cases and 70 (88.6%) cases respectively which was followed by pallor. On examination, pallor, hyper pigmented knuckles, bleeding spots, hepatomegaly and splenomegaly were common signs detected from both the groups. Similar observations were reported from the studies done by Naseem et al Gupta et al and Sharif et al who also found fever as the most common symptom followed by pallor.^{2,11,12} Studies done by Dubey et al, Memon et al, Jan et al, Rathod GB et al, and Chhabra A et al found pallor as the commonest symptom followed by fever.^{1,7-9,5} A study done by Wu et al found the most common symptom as pallor followed by bleeding and fever.¹³

We had found the most common cause, leading to either bicytopenia or pancytopenia was malaria found in 29 (16.6%) cases followed by dengue fever in 28 (16.1%) cases, ALL in 25 (14.3%) cases, megaloblastic anemia in 22 (12.64%) cases. Unlike other studies, we have found lower frequency of 10 cases only of aplastic anemia.^{1,2,8,12,13}

In bicytopenia group, the most common cause was dengue fever seen in 24 (25.2%) cases followed by malaria in 18 (18.9%) cases, ALL in 9 (9.47%) cases. A study done by Naseem et al found the most common cause of bicytopenia was ALL followed by immune thrombocytopenia and megaloblastic anemia.²

Out of 79 cases of pancytopenia, most common cause was megaloblastic anemia seen in 17 (21.5%) cases, ALL in 16 (20.2%) cases, malaria in 11 (13.9%) cases and aplastic anemia in 10 (12.6%) cases. As shown in Table 5, various studies are summarized to show the clinical profile and two common causes of pancytopenia.

Table 2: Summary of various studies of pancytopenia.^{1,2,5-9,12-16}

Author	Place	Year	Number of cases	Most commonest cause	Second common cause	Most common clinical findings
Memom et al ¹	Hyderabad, Pakistan	2008	230	Aplastic anemia	Megaloblastic anemia	Pallor, fever, bruises
Naseem et al ²	Chandigarh, India	2010	175	Aplastic anemia	Acute leukaemia	Fever, pallor, hepatomegaly, splenomegaly
Chhabra et al ⁵	Uttarakand, India	2012	91	Megaloblastic anemia	Acute leukaemia	Bleeding, fever, hepatomegaly, splenomegaly
Singh et al ⁶	Rajasthan, India	2016	187	Severe acute malnutrition	Acute leukaemia	-
Dubey et al ⁷	Kanpur, India	2015	170	Megaloblastic anemia	Aplastic anemia	Pallor, fever, bleeding
Jan et al ⁸	Peshawar, Pakistan	2013	205	Aplastic anemia	Acute leukaemia	Pallor, fever, bruises
Rathod et al ⁹	Vadodara, India	2015	200	Megaloblastic anemia	Aplastic anemia	Pallor, fever, bruises

Continued.

Author	Place	Year	Number of cases	Most commonest cause	Second common cause	Most common clinical findings
Gupta et al ¹²	Banaras, India	2008	105	Aplastic anemia	Acute leukaemia	Fever, pallor, bleeding
Wu et al ¹³	China	2011	174	Aplastic anemia	Myelodysplastic syndrome	Pallor, bleeding, fever
Manzoor et al ¹⁴	Pune, India	2014	50	Megaloblastic anemia	Aplastic anemia	Pallor, fever, splenomegaly, hepatomegaly
Bhatnagar et al ¹⁵	India	2005	105	Megaloblastic anemia	Aplastic anemia and infections	Bleeding, hepatomegaly, splenomegaly
Savage et al ¹⁶	Zimbabwe	1999	134	Megaloblastic anemia	Aplastic anemia	-
Present study	Vadodara, India	2018	174	Megaloblastic anemia	Acute lymphoblastic leukaemia	Fever, pallor, hepatomegaly, splenomegaly,

In the present study, from 28 (16.09%) cases of malignancy, 9 cases had bicytopenia and 19 cases presented with pancytopenia. Out of which, 25 cases had ALL, rest each were diagnosed as acute promyelocytic leukaemia, Burkitt lymphoma and non-Hodgkin lymphoma (from the pancytopenia group). According to various studies, frequency of malignancy presenting with pancytopenia cases were 26.6% by Naseem et al (all cases had acute leukaemia); 25% by Gupta et al, 17.6% by Dubey et al, 17.4% by Memon et al (8.69% ALL and others AML, CML, lymphomas, neuroblastoma and nephroblastoma).^{1,2,7,12} From our study, we found only 9.5% cases of malignancies (all cases of ALL) from bicytopenia group and 24% from pancytopenia group, as against a study done by Naseem et al who found 69.5% cases (66.9% acute leukaemia) with bicytopenia and 26.6% with pancytopenia.²

From both the groups of bi/pancytopenia, infectious causes were leading etiologies, found in 87 (50%) cases. Out of 95 cases of bicytopenia, 62 (65.3%) cases were due to various infective causes where as in pancytopenia, it was found in 25 (31.6%) cases. A study done by Gupta et al found that kala azar (9.5%) was the most common cause among infections followed by malaria and enteric fever in 2.9% cases each.¹² Memon et al in their study found enteric fever (10.8%) as the most common cause followed by malaria and septicemia seen in 8.69% cases.¹ A study done by Chhabra et al found infections such as kala azar, malaria, enteric fever, bacterial septicaemia were common causes of pancytopenia found in 19.7% of the patients.⁵ As reported by Ejaz et al infections were responsible in 50% of pancytopenia cases, of which malaria in 22%, enteric fever in 12% were predominant.¹⁰ Present study has identified plasmodium vivax was an important cause for bi/pancytopenia (25 out of 29 malaria cases).

Aplastic anemia was found in 10 (12.6%) cases out of 79 cases of pancytopenia. Studies done by Naseem et al,

Gupta et al, Memon et al, Jan et al reported aplastic anemia as the most common cause of pancytopenia found in 33.8%, 43.8%, 23.9%, and 28.3% respectively.^{1,2,8,12} Immune thrombocytopenia (ITP) was found in 6 (3.4%) cases, all of which were found to have bicytopenia.

Hypersplenism leading to pancytopenia was found in 4 (5.06%) cases, of which one case was a diagnosed case of thalassemia major. Studies done by Ejaz et al Jan et al Singh et al, Rathod et al found it in 8%, 2.4%, 9.1%, 2.5% cases respectively.^{6-8,10} As our hospital is situated near by the area where the large concentration of tribal population is living; we also found sickle cell anaemia as one of the causes of low blood counts. Out of 9 (5.17%) cases, 8 (8.4%) cases had bicytopenia and 1 (1.27%) case had pancytopenia. Nutritional anemia leading to bicytopenia was found in 4 (2.08%) cases. A study done by Memon et al found that mixed nutritional deficiency anaemia was seen in 8.69% cases.¹ Singh et al reported the most common cause of pancytopenia was severe acute malnutrition found in 27.3% cases.⁶ We also found one case each of Gaucher disease and homocystinuria leading to pancytopenia. Present study also detected rare causes like Kikuchi disease leading to bicytopenia, progressive familial intrahepatic cholestasis leading to pancytopenia (Table 2).

From bicytopenia group, anemia and leucopenia (total count <4000/mm³) was found in 19 (20%) cases, anemia and thrombocytopenia in 62 (65.3%) cases, leucopenia and thrombocytopenia in 14 (14.7%) cases indicating that most common affected two cellular elements were haemoglobin and platelet count.

Mean haemoglobin was 6.1 gm/dl in pancytopenia group as against 7.5 gm/dl in bicytopenia. High mean of MCV was observed in pancytopenia than in bicytopenia due to more number of cases of megaloblastic anemia. Mean total count and platelet count were found to be on higher side in bicytopenia group than in pancytopenia (table-3).

On applying the independent “t” test, the “p” value observed was statistically significant ($p < 0.05$) in the variables of mean haemoglobin, total count, MCV, platelet count and corrected reticulocyte count.

An attempt had been made to compare different laboratory variables among the major subgroups of different etiologies in bi/pancytopenia. The division of cases were made into 5 subgroups (infections, malignancy, megaloblastic anemia, aplastic anemia and miscellaneous causes). From all the 174 cases, infections contributed to 50%, followed by malignancy in 16% and megaloblastic anemia in 12.64%. The mean age of presentation was found to be lowest at 4.2 years in megaloblastic anemia group. Mean Hb was found to be lowest (4.49 gm/dl) in aplastic anemia followed by 5.1 gm/dl in megaloblastic anemia. Mean TC was higher (34,948 cells/mm³) in malignancy group than other groups as majority cases were of acute leukemia. Mean Hb was found to be higher in infectious group (8.2 gm/dl). The mean Hb, TC and platelet count were lowest in cases of aplastic anemia in comparison to other subgroups (Table 4).

On examining peripheral smear, hypersegmented neutrophils were seen in 19 cases of megaloblastic anemia out of 22 cases, making a good clue for the diagnosis of MA. Out of 28 cases of malignancy, blast cells were seen in 21 (75%) cases, among which 25% cases were seen in bicytopenia group and 50% cases were in pancytopenia group. A study done by Naseem et al, found that blast cells were seen in 64.6% of bicytopenia group and 20.1% in pancytopenia group.²

On examination of bone marrow aspiration/biopsy that was done in 42 cases, bone marrow was hypocellular in 11 (26.1%) cases, normocellular in 8 (19%) cases and hypercellular in 23 (54.7%) cases. Among 27 cases of malignancies, 23 cases had hypercellular marrow and 4 cases had normocellular marrow out of which 25 cases had ALL. A study done by Chhabra et al on 91 cases of pancytopenia found cellular marrow in 71.4% cases, out of which 16.9% had ALL; hypocellular marrow was seen in 28.5% cases of which only 3.8% had ALL.⁵ We did not find a single case of malignancy with hypocellular bone marrow.

CONCLUSION

Infectious etiologies such as dengue fever, malaria were the common causes giving rise to bi/pancytopenia. Acute lymphoblastic leukaemia and megaloblastic anemia are two other common treatable causes of bi/pancytopenia. Severity of low values of complete blood count correlates with more serious conditions such as aplastic anemia, malignancies rather than infectious causes. Pancytopenia is more predominantly noticed with causes related to bone marrow production or due to bone marrow infiltrative disorders rather than peripheral destruction or sequestration.

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