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CLINICO-ETIOLOGICAL AND CLINICO-HAEMATOLOGICAL STUDY OF PATIENTS WITH PANCYTOPENIA IN RURAL AREA

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Abstract

Background: Pancytopenia is a manifestation of other underlying conditions, commonly associated with multiple benign and malignant conditions. Any patient presenting with pancytopenia requires a thorough evaluation to identify the underlying aetiology.

Aim: This current evaluates various hematological parameters including bone marrow aspiration (where ever feasible) in pancytopenia in adult group. Study also correlates clinico-haematological profile.

Method: Fifty patients with a hematological diagnosis of pancytopenia were studied during the period August 2020 to August 2022. The study included adult patients of both sexes having the age of 18 years and above. Tests for complete blood count, reticulocyte count, peripheral smear, bone marrow aspiration, and trephine biopsy were done.

Results: Various etiological factors were identified in which majority were of megaloblastic anaemia (48.9%), followed by mixed nutritional anaemia (22.2%), hypersplenism (13.3%), aplastic anaemia (8.9%), malignant conditions (6.7%), myelodysplastic syndromes (2%), and others (4%) respectively. Megaloblastic anaemia cases observed in the age group of 31–50 years with male preponderance. Hemoglobin, TLC, Platelet count, and Reticulocyte count ranged from 2 g% - 10g%, 500–4000 cells/cumm, 24,000–1.5 lakh cells/cumm, and 0.1% – 2%. MCV was higher than 100 fl in 57.5% of cases. Majority of the patients had macrocytic and dimorphic anaemia. Hypersegmented neutrophils were present in all the patients. Bone marrow of Megaloblastic anaemia was hypercellular. Megaloblastic erythropoiesis with giant meta- myelocytes and band forms were seen. Nutritional anaemia seen in the age group of 51-60 years. Haemoglobin, TLC, Platelet count, and Reticulocyte count ranges from 2.3 g%–7.8 g%, 1000–4000 cells/cumm, 5000–1.4 lakh cells/cumm, and 0.1–8%. Two cases had microcytic hypochromic anaemia in Nutritional anaemia. Bone marrow was hypercellular with a reversal of M:E ratio in 93.8% of cases. In hypersplenism seen in the age group of 51–60 years. Haemoglobin, TLC, Platelet count, and Reticulocyte count ranges from 3.8 g% – 10 g%, 1700–3800 cells/cumm, 26000-1.4 lakh cells/cumm, and 0.6–2% respectilvey in hypersplenism. 40% of hypersplenism patients had microcytic hypochromic anaemia. Bone marrow was hypercellular with a reversal of M:E ratio in 70% of hypersplenism cases. Aplastic anaemia seen in the age group of 41–50 years. Haemoglobin, TLC, Platelet count, and Reticulocyte count ranges from 3.1–10 g%, 1100–4000 cells/cumm, 51000–1.5lakh cells/cumm, and 0.2%–1.8%. Aplastic anaemia (35.8%) cases showed macrocytosis. Bone marrow was hypocellular with an increase in marrow fat and Lymphocytes and plasma cells were prominent in Aplastic anaemia cases. Leukaemia commonly seen in the age group of 31–40 years with male predominance. Hemoglobin, TLC, and Reticulocyte count ranges from 5.1–9.8%, 1100–4000 cells/cumm, and 0.6–2% respectively. Bone marrow was hypercellular with a reversal of M:E ratio in 80%.

Conclusion: Megaloblastic anaemia was the commonest cause of pancytopenia. Most other studies have reported aplastic anaemia as the commonest cause. This seems to reflect the higher prevalence of nutritional anaemia in the Indian subjects. The haematological parameters and bone marrow morphological features in patients with megaloblastic anaemia, aplastic anaemia, and malignant diseases including MDS in the present study were comparable to the findings by other authors. Uncommon etiological factors like dengue fever and hemolytic anaemia were identified in this study. A comprehensive clinical, haematological, and bone marrow study of patients with pancytopenia usually helps in identification of the underlying cause. However, in view of a wide array of etiological factors, pancytopenia continues to be a challenge for hematologists.

Keywords: Anaemia, Reticulocyte count, Platelet count, Pancytopenia.

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INTRODUCTION

Pancytopenia is characterized by a hemoglobin value of less than 12 g/dL in women and 13 g/dL in men, platelets of less than 150,000 per μL, and leukocytes of less than 4000 per ml (absolute neutrophil count of less than 1800 per ml).[1] Pancytopenia is a common clinico-hematological entity encountered in day-to-day clinical practice.[2,3] Pancytopenia refers to a disorder in which all three elements of the blood (RBCs, WBCs, and Platelets) are lower in counts than normal. Thus, it is not a disease entity by itself, but rather a triad of findings. It is a primarily or secondarily affecting bone marrow manifesting and lead to various hematological derangements, which is reflected in the peripheral blood smear as pancytopenia. There are varying trends in its aetiology, clinical pattern, treatment modalities, and outcome in different studies.[4-7]

Pancytopenia is defined as hemoglobin < 12 gm%, WBCs count < 4 x 10^9/L, and platelet count < 100 x 10^9/L. Anaemia defined as mild (Hb 9–12 gm%), moderate (Hb 5–9gm%), severe (Hb 3,000/ mm3), moderate (WBCs 1,000-3,000/mm3), and severe (WBCs 50,000/mm3), moderate (platelet count 20,000-50,000/mm3) and severe as (platelet count < 20,000/mm3). Most of the time pancytopenia is insidious in onset. The presenting symptoms are usually anaemia and thrombocytopenia, Leukopenia is an uncommon cause of initial presentation. There are many factors encompassing geographic distribution and genetic disturbances which cause pancytopenia according to various studies.[6,7]

To understand the aetiology of pancytopenia, bone marrow biopsy plays a significant role. In some other selected cases radiological, biochemical, and microbiological investigations are useful. The severity of pancytopenia and underlying aetiology determine management and prognosis. Thus, identification of correct cause will help in treatment. This study is therefore aimed to identify the frequent causes of pancytopenia in patients presenting to a rural area. There are blund to be variations in causes leading to pancytopenia. Similar studies are available in literature; an attempt is being made to correlate with regards to rural population.

Although it is a common clinical pattern with an extensive differential diagnosis, there is little discussion of this abnormality in major textbooks of internal medicine and hematology. Since the underlying pathology of pancytopenia determines the management and prognosis of patients, there is a definite need to study about pancytopenia.

The aim of the study was to evaluate various hematological parameters including bone marrow aspiration (where ever feasible) in pancytopenia in adult group. The study also correlates clinico-haematological profile.

MATERIALS AND METHODS

Fifty patients with a hematological diagnosis of pancytopenia were studied during the period, august 2020 to August 2022, in the Department of Pathology, SVS Medical College and hospital, Mahabubnagar.

Inclusion criteria: Patients within the age group of 18–60 with hematological evidence of pancytopenia.

Exclusion criteria: Patients below 18 years and above 60 years with pancytopenia. Patients not amenable for follow up were excluded.

Leishman's Stain was done by observing Cytoplasm in pink and Nucleus in blue color.

Periodic acid Schiff staining was done according to standard laboratory procedures

Counter stained with Harris' hematoxylin for 5–10 minutes. Nuclei in Blue color, substances showing positive reaction had Magenta Pink.

Clinical history recording and examination of all the identified cases of pancytopenia were done as per proforma. Two ml of anticoagulated blood was collected for complete hemogram.

Sudan black and Perls' stains were used wherever indicated.

Bleeding time measured by Ivy's method, and Clotting time by Tube method. While Hemoglobin percentage, Total leucocyte count, RBC Count, Platelet count, and Packed cell volume were measured by Sysmex cell counter.

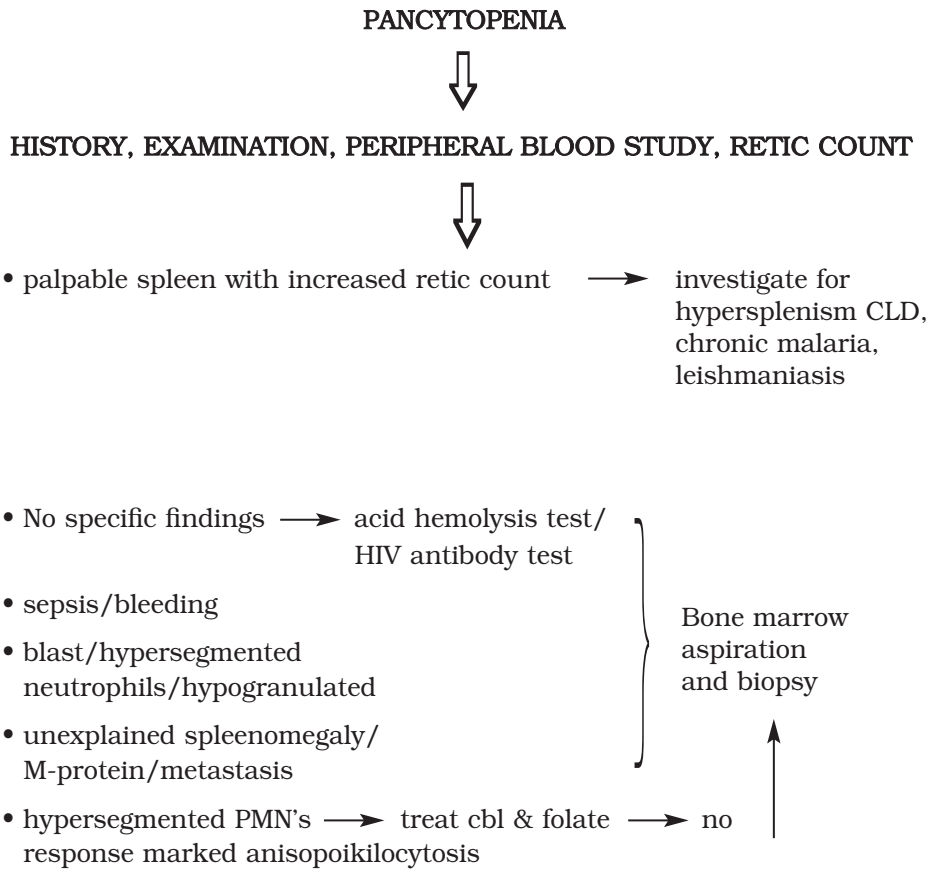
The peripheral smear was studied after staining with Leishman's stain. Special stains – Periodic acid Schiff reagent stain, Myeloperoxidase, Sudan black and Perls' stains were used wherever indicated.

Estimation of Serum Ferritin done by Chemiluminescence immunoassay (Lilac Acculite CLIA).

Bone marrow aspiration was done in all the patients to identify the aetiology. Slides were stained with Leishman's stain.

Hypochromicity on smear with low ferritin and MCV<75fL is called as Iron Deficiency Anaemia. Iron in the control subjects were 118.37 ± 20.79 µ/dl.

Iron deficiency anaemia is defined as Hb less than 11g/dl with the presence of a mean corpuscular volume (MCV) of less than 70 fl and serum ferritin below 30 ng/ml (in presence of infections).



Statistical analysis: The Crosstabs procedure forms two-way and multiway tables and provides a variety of tests and measures of association for two-way tables. The structure of the table and whether categories are ordered determine which test or measure to use. The data were tabulated in Microsoft Office Excel and analyzed using SPSS software version

18.0 (SPSS Inc, US). The chi-square test was used to analyze the association between two categorical variables. P value of <0.05 was statistically significant.

RESULTS

Most of the patients were in the age group of 41-60 years (53%). The sex distribution of pancytopenia showed a male preponderance. The male to female ratio was 2.1:1.

Generalised weakness (86%) was the commonest symptom in pancytopenic patients, followed by fever (39%), bleeding manifestations (5%), and pain abdomen (24%).

Totally 8% of the patients had Hb values between 1-3 g, followed by 22% were of 3.1 to 5 g of Hb levels, and 52% were of 5.1 to 7 g Hb levels.

Total leukocyte count was in the range of 500-4000 cells/mm^3. Most (40%) of the patients had values in the range of 3100-4000 cells/mm^3. 8% of the patients had values between 500 and 1000 cells/mm^3.

Most (36%) patients had platelet counts in the range of 51,000–75,000 cells/ mm^3. 24% of patients had platelet counts in the range of 4000– 50000 cells/mm^3.

The reticulocyte count varied from 0.1–20%. Majority (88%) of the patients had reticulocyte count in the range of 0.1–2%, and 4% of patients were between 6.1 to 20.

Bone marrow aspirate analysis shows various types of cellularity, such as 8% of hypocellularity, 80% of hypercellularity, and 12% of normocellular respectively.

a. **Pancytopenia associated with hypocellular marrow**: In the present study, 10 out of 50 patients had hypocellular marrow. The causes were varied, 40% being belonging to hypcellular marrow and Aplastic anaemia, which showed a peak incidence (60%, n=3) in the age group (41–50 years). The least incidence (20%, n=1) was seen in the age group of 31–40 years. Dengue fever (20%) and hypersplenism (20%) contributed to rest of the cases of hypocellular marrow. Aplastic anaemia (autoimmunity) was more common in males (60%, n=3). The male to female ratio of incidence was 1.5:1. The haemoglobin percentage varied from 3.1–10 gm%. Most patients (40%, n=2) had hemoglobin values in the range of 3.1-5 g%. 20% of patients were seen to have hemoglobin values in the range of 7.1–10 g%. The total leukocyte count ranged from 1100–4000 cells/mm^3. Most patients (40%, n=2) had a value in the range of 1100–2000 cells/ $mm^3$and 3100-4000 cells/mm^3. The reticulocyte count was in the range of 0.1–1.5%. Majority of the patients (80%,n=4) had values in the range of 0.1–1.0%. Most of the patients (64.2%) had normocytic normochromic erythrocytes. Some (35.8%) showed macrocytosis. 42.8% of them had relative neutrophilia while the others had relative lymphocytosis. The erythrocyte sedimentation rate was increased in most of the patients and ranged from 28–140 mm/hr. The bone marrow was hypocellular and the aspirate was mostly composed of fat cells. Other precursors appeared normal. There was a relative increase in the number of plasma cells and lymphocytes.

b. **Pancytopenia with hypercellular and normocellular marrow:** Hypercellular bone marrow was observed in 38 patients and it was normocellular in 7 patients. Most common etiology noted was megaloblastic anaemia (48.9%, n=22), followed by nutritional anaemia (22.2%, n=10), hypersplenism (13.3%, n=5), leukaemia (6.7%, n=4), myelodysplastic syndromes (2.2%, n=1), and others. Normocellular marrow was seen in (8%) megaloblastic anaemia, (4%) hypersplenism, and 2% in nutritional anaemia. There were 10 Hypocellular marrow, and 30 hypercellular marrows with pancytopenia were recorded.

Table 1 Depicting various categories and cellularity

CATEGORY	Number of cases	% of cases	Hypercellular marrow: 30 Percentage %	Hypocellular Marrow: 10 Percentage %	Normocellular Marrow: 10 Percentage %
Megaloblastic marrow	24	48	80	0	0
Mixed nutritional (dimorphic)	10	20	0	0	100
hypersplenism	2	4	0	20	0
MDS	1	2	3.3	0	0
MM	1	2	3.3	0	0
Acute leukaemia	3	6	10	0	0
Hemolytic anaemia	1	2	3.3	0	0
Dengue fever	2	4	0	20	0
aplastic	4	8	0	40	0
unclassifiable	2	4	0	20	0

Pancytopenia with megaloblastic anaemia: In the present study, 24 cases of megaloblastic anaemia were seen. It constituted 48% of cases of pancytopenia and 80% of all patients with cellular marrow. Megaloblastic anaemia was observed in the age group ranging from 21–60 years. Majority were seen in the age group of 31–50 years (63.6%, n=14). There were male preponderance and the male to female ratio was 2.4:1. Majority of the patients (68.2%, n=15) had values in the range of 5.1–7gm%. Majority of the patients (81.8%, n=20) had a leukocyte count in the range of 2100-4000 cells/mm^3. 4.6% of patients had values ranging from 500–1000 cells/mm^3. Most of the patients (40.9%) had a platelet count in the range of 51,000–75,000 cells/mm^3. 9.1% patients had values in the range of 26,000–50,000 cells/mm^3. The reticulocyte count varied from 0.1–6%. Most of them (77.3%) had reticulocyte count in the range of 0.1–2%.

Macro ovalocytosis with a considerable degree of anisopoikilocytosis were the main features. Mean corpuscular volume was more than 100 fl in 57.5% of patients. Dimorphic blood picture was seen in 10 patients (30%). Hypersegmented neutrophils were seen in most of the patients. Basophilic stippling and cabot rings were present. Platelets were reduced in number in all the cases.

The bone marrow was hypercellular with a reduction of fat cells in most of the patients (81.8%). Four patients (18.2%) had a normocellular marrow. Erythroid hyperplasia with megaloblastic maturation and reversal of M:E ratio was seen in all the patients. Megakaryopoiesis was normal in 63.6%, decreased in 18.2%, and increased in 18.2% of patients.

Pancytopenia with mixed nutritional anaemia

In the present study, 10 patients (20%) of pancytopenia were observed to have nutritional anaemia as the etiology, which contributed to all cases of normocellular marrow.

Nutritional anaemia was seen in the age group of 18–60 years. Majority of the patients (40%) were in the age group of 51–60 years. 20% of them were in the age group of 21–40 years. There was a male predominance and the male to female ratio of incidence was 2.3:1.

Hemoglobin percentage varied from 2.3–7.8%. Majority of the patients (50%) had haemoglobin in the range of 3.1–5 gm%.

The total leukocyte count ranged from 1000–4000 cells/ mm^3. Most of the patients (60%) had a value in the range of 3100–4000 cells/ mm^3. 10% of the patients had a count of 500–1000 cells/ mm^3. Most of the patients (40%) had a value between 51,000 and 75,000 cells/mm^3. The reticulocyte count varied from 0.1-8%. In majority of the patients (50%) the values ranged from 0.1–2%. Most of the patients had normocytic normochronic anaemia. In two patients had microcytic hypochromic anaemia, MCV was in the range of 65.6–108 fl.

MCHC was in the range of 26–34.7% and MCH was in the range of 18–38.9 pg. Nine patients (90%) had hypercellular marrow with a reversal of M:E ratio. Erythroid hyperplasia along with both megaloblastic and micronormoblastic maturation was observed in all the patients. Leucopoiesis was normal. Megakaryocytes were either normal or increased. Only two patients had decreased megakaryopoiesis.

Pancytopenia with hypersplenism

In the present study, 2 patients of hypersplenism were seen, which constituted to 4% of cases with pancytopenia and 20% of all hypocellular marrows. Hypersplenism was seen to occur in the age group ranging from 41–60 years. Majority of them were in the age group of 51–60 years. There was a male preponderance and male to female ratio of incidence was 2:1. Hemoglobin percentage varied from 3.8–10 gm%. Majority of the patients (50%) had values in the range of 7.1–10 gm%. The total leukocyte count ranged from 1700-3800 cells/mm^3. Majority of the patients (50%) had values between 1100–2000 cells/mm^3. The reticulocyte count ranged from 0.6–2%. Majority (50%) of the patients had values in the range of 0.6–1%. Most of the patients (60%) had normocytic normochromic anaemia. 40% of them had microcytic hypochromic anaemia with a severe degree of anisopoikilocytosis. MCV was in the range of 56.8 fl – 94.6 fl. 66.7% of patients had hypercellular marrow while the rest had normocellular (33.3%). Erythroid hyperplasia was observed in 90% of patients. Leukopoiesis was normal in 90% patients. Megakaryopoiesis was normal in 60% of patients, while it was increased in the rest.

Pancytopenia with malignant diseases, myelodysplastic syndromes, and others

In the present study, 10% of the patients with pancytopenia had the following etiologies of acute leukaemia (3, AML category), one case of multiple myeloma. They accounted to 10% of cases of pancytopenia with cellular marrow. The age ranged from 21–40 years. Majority of them (66.7%) were in the age group of 31–40 years. There was a male predominance. The male to female ratio of incidence was 2:1. The hemoglobin percentage ranged from 5.1–9.8%. 66.7% of the patients had values between 7.1 and 10 gm%. The total leukocyte count varied from 1100–4000 cells/mm^3. 66.7% of the patients had values between 3100–4000 cells/ mm^3. Platelet count ranged from 26,000-89,000 cells/mm^3. The reticulocyte count ranged from 0.6–2%. 66.7% of the patients had a value ranging from 0.6–1%. These patients presented with peripheral pancytopenia. The erythrocytes were normocytic normochromic. The leukocyte count was decreased with a presence of immature cells. Myeloblasts with fine chromatin, 2-3 nucleoli, and occasional Auer rods were seen. Platelets were decreased. Bone marrow was hypercellular in 100% of the patients. Erythroid series were decreased in 40% of the patients and increased in 60% patients. Myeloid hyperplasia was observed with more than 30% blasts in all of them. Immunocytochemistry demonstrated MPO positivity in them. Megakaryocytes were reduced.

One case of myelodysplastic syndrome was observed in the present study in a female patient aged 55 years who had peripheral pancytopenia with a macrocytic type of anaemia.

The aspirate was hypercellular with erythroid hyperplasia and features of dyserythropoiesis which included megaloblastic erythroblasts, nuclear fragmentation, budding multiple nuclei, and hyperlobulation. Dysmyelopoiesis with hypogranular neutrophils and blasts were also observed. Large hypolobulated megakaryocytes were seen.

Dengue fever

Two cases of dengue fever in an 18-year-old male and a 22 year old female presented with pancytopenia. The total leukocyte count was 800 cells/mm^3 and platelet count was 28,000 cells/cumm. Bone marrow was hypercellular with erythroid hyperplasia and a reversal of M:E ratio.

Hemolytic anaemia:

One case of hemolytic anaemia presented with pancytopenia. The patient was a 21-year-old female. The peripheral blood showed features of hemolysis. Erythrocytes showed a moderate degree of anisopoikilocytosis with increased polychromatophilic RBCs. The reticulocyte count was markedly increased. The bone marrow was hypercellular with a reversal of M:E ratio. Erythroid hyperplasia with normoblastic maturation was seen. Megakaryocytic hyperplasia was also noted.

Table 2 Association between Pancytopenia etiology and Haematological Parameters

	Hb % (gm/dl)	TLC (cells/mm^3)	Platelet (cells/mm^3)
Megaloblastic anaemia associated with pancytopenia	2–10	500–4000	26000–1,50,000
Nutritional anaemia (mixed)	2.3–7.8	1000–4000	5000–1,40,000
Aplastic anaemia associated with pancytopenia	3.1–10	1100–4000	5100–1,50,000
Hypersplenism	3.8–10	1700–3800	26,000–1,40,000
Leukaemia associated with pancytopenia	5.1–9.3	1100–4000	26000–89000

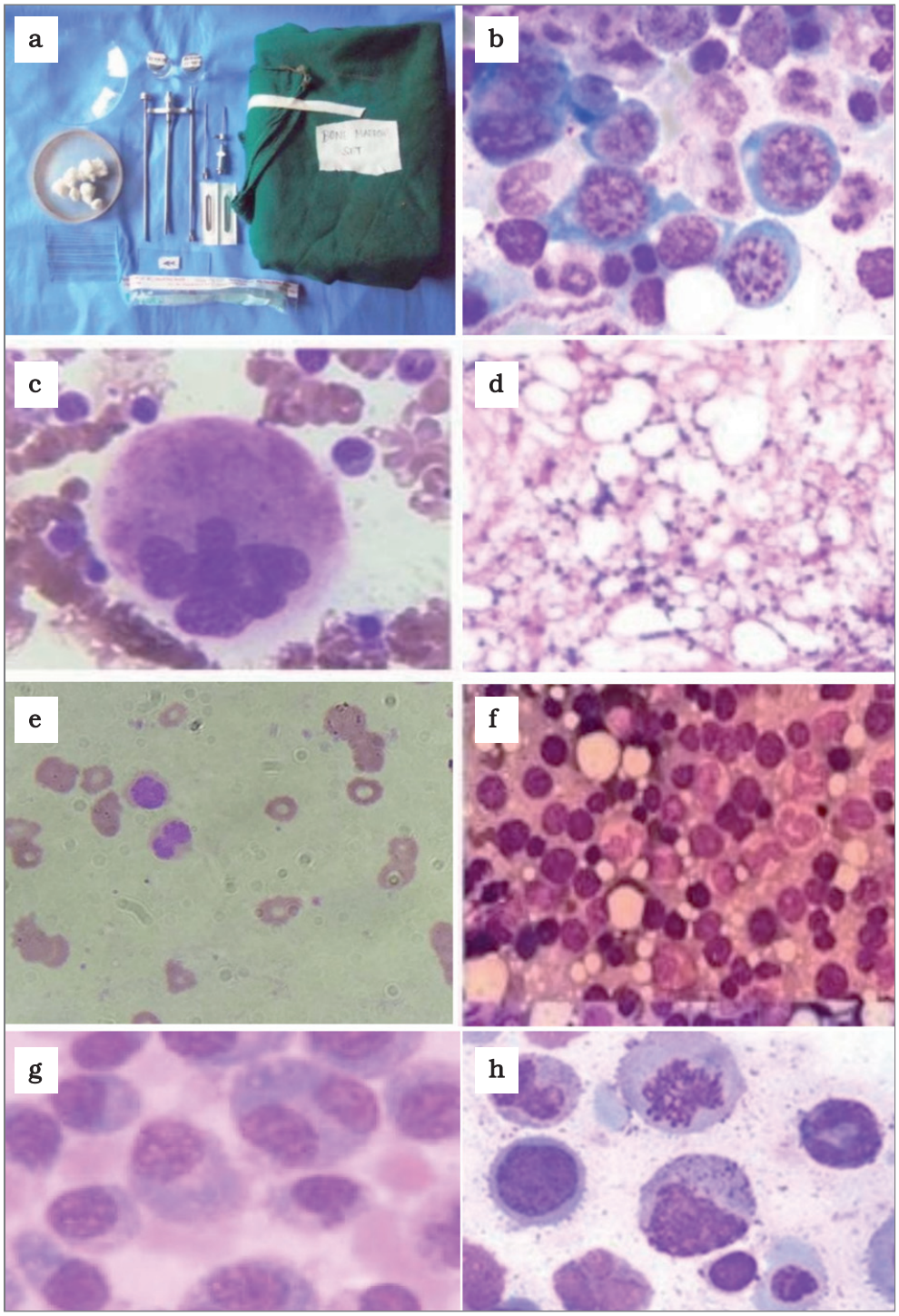


Fig. 1 a. Bone Marrow Set. b. Erythroid Hyperplasia (100x). c. Megakaryocytes (100x). d. Aplastic anaemia (100x). e. Microcytic Hypochromia with Neutrophils (100x). f. Erythroid Hyperplasia (100x). g. Plasma Cells (100x). h. Megaloblasts with Mitosis (100x).

DISCUSSION

In the present study, megaloblastic anaemia (48.9%) was the commonest cause of pancytopenia, followed by mixed nutritional anaemia (22.2%), hypersplenism (13.3%), aplastic anaemia (10%), malignant diseases (5%), myelodysplastic syndromes (0.6%), and other (4%) uncommon causes like Dengue fever (2%) and Hemolytic anaemia (2%).

The commonest cause of pancytopenia reported from various studies throughout the world has been aplastic anaemia (Table 3).

This is in contrast with the results of the present study where the commonest cause of pancytopenia was megaloblastic anaemia. This seems to reflect the higher prevalence of nutritional anaemia in Indian subjects as well as in developing countries. However, similar results have been reported in studies from other Indian centres.

Table 3 Causes of pancytopenia in various studies

Study	Country	Year	No. of cases	Commonest cause	Second most common cause
Retief FP, Heyns AD[8]	South Africa	1976	195	Bone marrow failure (67.7%)	Severe infection (9.7%)
Imbert M et al[9]	Europe	1989	213	Malignant myeloid disorders (42%)	Malignant lymphoid disorders (18%)
Varma N, Dash S[10]	India	1992	202	Aplastic anaemia (40.6%)	Megaloblastic anaemia (23.26%)
Tilak V, Jain R[5]	India	1998	77	Megaloblastic anaemia (68%)	Aplastic anaemia (7.7%)
Khodke et al.[11]	India	2001	166	Hypoplastic anaemia (29.51%)	Megaloblast ic anaemia (22.3%)
Naeem Khan M et al.[12]	Pakistan	2001	30	Aplastic anaemia (20%)	Megaloblastic anaemia (16.7%)
Kumar R et al.13	India	2001	166	Aplastic anaemia (29.5%)	Megaloblastic anaemia (22.3%)
Osama Ishtiaq et al.[14]	Pakistan	2002	100	Megaloblastic anaemia (39%)	Hypersplenism (19%)
Mobina et al.[15]	Pakistan	2005	392	Megaloblastic anaemia (35.95%)	Hypersplenism (16.3%)
Jha et al.[16]	Nepal	2008	148	Hypoplastic anaemia (29.5%)	Megaloblastic anaemia (23.64%)
Varma A et al [17]	India	2018	251	Megaloblastic anaemia (48.5%)	Dimorphic anaemia (17.8%)
Present study	India	2022	50	Megaloblastic anaemia (48.9%)	Nutritional anaemia (22.2%)

Megaloblastic anaemia associated with pancytopenia

Megaloblastic anaemia is common in India. This seems to reflect the higher prevalence of nutritional anaemia in Indian subjects. In a study of pancytopenia cases by Jha et al., the age range was 10–79 years (31 years). There was a male preponderance and male to female ratio was 1.5:1. In the study by Kumar et al.[13] the ages ranged from 14–73 years (39.5%). There was a female preponderance and the male to female ratio was 2:1. In the present study, the age ranged from 18–70 years. Majority of the patients were in the age group of 31–50 years (62%). There was a male preponderance and the male to female ratio was 2.4:1.

The principal hematologic manifestations are varying degrees of anaemia, leucopenia, thrombocytopenia, anisopoikilocytosis, macroovalocytosis and hyper-segmented neutrophils.

In a study by Kishore Khodke et al.[11] 20/22 cases showed anisocytosis, 10/22 cases showed dimorphic blood picture, and 20/22 cases showed hypersegmented neutrophils. In the study by Tilak et al.[5] 51/53 cases showed anisocytosis, 45/53 cases showed hypersegmented neutrophils, 13/53 showed circulating erythroblasts. Reticulocytes were seen in 5/53 and relative lymphocytosis was seen in 7/53 cases.

In the present study, macroovalocytes with considerable degree of anisopoikilocytosis were the main features in all the cases. MCV was more than 100 fl in 57.5% of cases and dimorphic blood picture was seen in 30% of cases (10 patients). Hypersegmented neutrophils were seen in most of the patients.

Bone marrow is usually hypercellular with predominantly megaloblastic erythropoiesis. Giant band forms, metamyelocytes, and giant megakaryocytes are also seen. In the present study, the bone marrow was hypercellular with a reduction of fat cells in most of the patients (81.8%). Four patients (18.2%) had normocellular marrow. Erythroid hyperplasia with megaloblastic maturation was seen in all the patients.

Nutritional anaemia (mixed)

Nutritional as a common etiological factor causing pancytopenia is well recognized and established. The nutritional deficiency of either B_{12} or folate results in megaloblastic anaemia. Other causes include mixed deficiency anaemia (microcytes and macrocytic). Mobina et al.[15] in their study of 392 cases of pancytopenia found 11.2% cases of mixed deficiency anaemia.

In the present study, mixed deficiency was seen in 22.2% of patients. This percentage is much lower than expected because 60–80% of world population is affected by iron deficiency anaemia which is the most common preventable nutritional deficiency in the world. The possible explanation is that majority of the cases present with anaemia rather than pancytopenia and are diagnosed on smear examination and treated as outpatients.

The age ranged in the present study from 18–60 years. There was a male preponderance and male to female ratio was 2.3:1. Most of the patients had dimorphic anaemia. Two patients had macrocytic and hypochromic anaemia. Bone marrow was hypercellular. Erythroid hyperplasia with both megaloblastic and micro normoblastic maturation was observed in all the patients. Leucopoiesis was normal. Megakaryopoeisis was either normal or increased.

Aplastic anaemia associated with pancytopenia

In the study by Kumar et al. the ages ranged from 12–63 years (29 years). There was a male preponderance and male to female ratio was 1.4:1. In the study by Jha et al.[16] the ages ranged from 1.5–70 years (17 years). There was a male preponderance with male to female ratio of 1.3:1.

In the present study, the ages ranged from 31-60 years. Majority of the patients were in the age group of 41–50 years. Aplastic anaemia was more common in males. The male to female ratio was 1.5:1.

In the study by Kishore Khodke, 3/7 patients showed anisocytosis and 1/7 patients showed relative lymphocytosis. In the study by Tilak et al. 2/6 patients had anisocytosis and 3/6 patients had relative lymphocytosis.

In the present study, 64.2% had normocytic normochromic erythrocytes. 35.8% of the patients had macrocytic anaemia and 56.3% of them had relative lymphocytosis.

Cellularity of bone marrow in aplastic anaemia[18] is very much reduced. It may be hypocellular or acellular. Lymphocytes and plasma cells are prominent. Daniel NM in their analysis of 50 cases reported 74% of patients with hypocellular marrow, 16% of patients with normocellular marrow which later became hypocellular and 10% with acellular marrow.

In the present study, bone marrow was mostly hypocellular and the aspirate was composed of fat cells in all the patients. There was a relative increase in plasma cells and lymphocytes. Bone marrow trephine biopsy revealed replacement of marrow by fat cells.

Hypersplenism

Hypersplenism is known to cause pancytopenia by sequestration of blood cells. In a study of 195 patients, Retief HP[8] found hypersplenism to be the cause of pancytopenia in 7.7% of the patients. Kumar et al. reported on incidence of hypersplenism in 19/166 cases in which ages ranged from 14–49 years. There was a male preponderance with the male to female ratio being 2:1.

In the study by Kumar et al. the Hb% ranged from 3.5–8.6 gm%. The TLC ranged from 1100–3600 cells/mm^3.

In the present study, the Hb% ranged from 3.8–10 g%, TLC ranged from 1700-3800 cells/mm^3.

Most of the patients (60%) had normocytic normochromic anaemia. 40% of them had microcytic hypochromic anaemia. In a study by Osama et al.[14] macrocytosis was seen in 63.1% cases and microcytosis in 36.8% cases. Bone marrow 66.7% had hypercellular marrow while the rest had normocellular.

Leukaemia associated with pancytopenia:

In a study by Jha et al., acute leukaemia alone constituted 90.62% of all the hematological malignancies. It accounted for 19.59% of total cases of pancytopenia. The age ranged from 2–75 years with a male to female ratio of 1.9:1. Khodke et al. and Tilak et al. reported one case of AML causing pancytopenia.

In the present study, majority (66.7%) of the pancytopenia cases were due to acute leukaemia. The ages varied from 21–40 years. There was a male preponderance with male to female ratio being 2:1.

In the study by Tilak Jain et al. one case of acute myeloid leukaemia with anisocytosis, circulating erythroblasts and immature cells was reported. Kishore Khodke et al. found one case of acute myeloid leukaemia with immature cells in the peripheral blood.

In the present study, all the patients had normocytic normochromic anaemia. Leukocyte count was reduced and circulating immature cells were seen. Platelet count was also reduced.

Myelodysplastic syndrome (MDS):

Pancytopenia is known to occur in MDS. It is the least common finding encountered in patients with MDS as compared to mono and bicytopenia.

In a study of 816 patients with MDS by Greenberg et al.[19] pancytopenia was found in 15% of the patients.

In a study of 118 patients with MDS by Juneja SK et al.[20] the age ranged from 48–95 years. In a study of 31 patients by Kini J et al.[21] the patients were in the age group of 4–7 years.

In the present study, one case presented with pancytopenia in a female patient aged 55 years.

Dengue fever

Naeem Khan et al studied 30 cases of pancytopenia and found 1 case of dengue fever. In the present study, 2 cases in 18 years (male) and 22 years (female) with dengue fever presented with pancytopenia. The total leukocyte count was 800 cells/cumm and platelet count was 28,000 cells/cumm. Bone marrow was hypercellular showing erythroid hyperplasia with a reversal of M:E ratio.

Hemolytic anaemia

Fazlur Rahim et al.[22] in their study found three cases of pancytopenia with hemolytic anaemia. Osama et al. in their study found two cases of pancytopenia with hemolytic anaemia.

In the present study, one patient of hemolytic anaemia presented with pancytopenia. Peripheral blood showed features of hemolysis. Erythrocytes showed a moderate degree of anisopoikilocytosis and an increase in number of polychromatophilic RBCs.

Reticulocyte count was markedly increased. Bone marrow was hypercellular with a reversal of M:E ratio. Erythroid hyperplasia with normoblastic maturation was seen. Megakaryocytic hyperplasia was noted in both the cases.

CONCLUSION

Megaloblastic anaemia was the commonest cause of pancytopenia in the present study. Most other studies have reported aplastic anaemia as the commonest cause. This seems to reflect the higher prevalence of nutritional anaemia, at the same time remembering the fact non nutrional causes contribute to megaloblastic change as in MDS or hemolytic anaemia. in the Indian subjects. The haematological parameters and bone marrow morphological features in patients with megaloblastic anaemia, aplastic anaemia, and malignant diseases including MDS in the present study were comparable to the findings by other authors. Uncommon etiological factors like dengue fever and hemolytic anaemia were identified in this study. A comprehensive clinical, haematological and bone marrow study of patients with pancytopenia usually helps in identification of the underlying cause. However, in view of a wide array of etiological factors, pancytopenia continues to be a challenge for hematologists. Therapeutic option for pancytopenia are drugs to stimulate blood cell production, blood transfusions, and antibiotics to treat an infection.

REFERENCES

1. Hoffman R, Benz EJ, Shattil SJ, Furie B, Cohen HJ, Siberstein LE. Hematology. Basic Principles and practice. 3rd ed. USA: Churchill Livingstone; 2005.
2. Hayat AS, Khan AH, Baloch GH, Shaikh N. Pancytopenia;: Study For Clinical Features And Etiological Pattern at Tertiary Care Settings in Abbottabad. The Professional Medical Journal. 2014 Feb 10;21(01):060-5.
3. Jain A, Naniwadekar M. An etiological reappraisal of pancytopenia-largest series reported to date from a single tertiary care teaching hospital. BMC Blood Disorders. 2013 Dec;13:1-9.
4. Alim M, Verma N, Kumar A, Pooniya V, Abdul Rahman R. Etio-Hematological Profile and Clinical Correlates of Outcome of Pancytopenia in Children: Experience From a Tertiary Care Center in North India. Cureus. 2021 Jun 2;13(6):e15382.
5. Tilak V, Jain R. Pancytopenia--a clinico-hematologic analysis of 77 cases. Indian journal of pathology & microbiology. 1999 Oct 1;42(4):399-404.
6. Dubey SR, Patel SK, Arya AK, Singh RP. Clinico-etiological spectrum of pancytopenia in hospitalized children. Int J Contemp Pediatr. 2016 Jan;3(1):169-72.
7. Vaidya S. Evaluation of bone marrow in cases of pancytopenia in a tertiary care hospital. Journal of Pathology of Nepal. 2015 Mar 27;5(9):691-5.
8. Retief FP, Heyns AD. Pancytopenia and aplastic anaemia : A retrospective study. South African Medical Journal= Suid-afrikaanse Tydskrif vir Geneeskunde. 1976 Aug 1;50(34):1318-22.
9. Imbert M, Scoazec JY, Mary JY, Jouzult H, Rochant H, Sultan C. Adult patients presenting with pancytopenia: a reappraisal of underlying pathology and diagnostic procedures in 213 cases. Hematologic pathology. 1989 Jan 1;3(4):159-67.
10. Varma N, Dash S. A reappraisal of underlying pathology in adult patients presenting with pancytopenia. Tropical and geographical medicine. 1992 Oct 1;44(4):322-7.

11. Khodke K, Marwah S, Buxi G, Yadav RB, Chaturvedi NK. Bone marrow examination in cases of pancytopenia. J Indian Acad Clin Med 2001;2:55-9.
12. Naeem Khan M, Ayyub M, Nawaz KH, Naeem Naqi, Hussain T, Shujaat H, et al. Pancytopenia: Clinico-pathological study of 30 cases at Military Hospital, Rawalpindi Pak J Pathol 2001 AprJun;12(2):37-41.
13. Kumar R, Kalra SP, Kumar H, Anand AC, Madan H. Pancytopenia- A six year Study. JAPI 2001; 49:1078-81.
14. Osama I, Baqai Hz, Anwar F, Hussain N. Patterns of Pancytopenia in a general medical ward and a proposed diagnostic approach JAMC 2002;16(1):8-13.
15. Dodhy MA, Bokhari N, Hayat A. Aetiology of Pancytopenia, A five-year experience. Ann Pak Inst Med Sci. 2005 Apr;1(2):92-5.
16. Jha A, Sayami G, Adhikari RC, Panta AD, Jha R. Bone marrow examination in cases of pancytopenia. J Nepal Med Assoc. 2008 Jan 1;47(169):12-7.
17. Varma A, Lokwani P, Malukani K, Gupta S, Maheshwari P. Study of hematological profile of adults presenting with pancytopenia in a tertiary care hospital of central India. Medical Journal of Dr. DY Patil Vidyapeeth. 2018 Nov 1;11(6):512.
18. Gnanaraj J, Parnes A, Francis CW, Go RS, Takemoto CM, Hashmi SK. Approach to pancytopenia: Diagnostic algorithm for clinical hematologists. Blood Rev. 2018 Sep;32(5):361-367.
19. Greenberg PL, Attar E, Bennett JM, Bloomfield CD, De Castro CM, Deeg HJ, Foran JM, Gaensler K, Garcia-Manero G, Gore SD, Head D. Myelodysplastic syndromes. Journal of the National Comprehensive Cancer Network. 2011 Jan 1;9(1):30-56.
20. Juneja SK, Imbert M, Jonault H, Swazec JY, Sigaux F, Sultan C. Hematological features of primary MDS at initial presentation: A study of 118 cases. J Clin Pathol. 1983;86:1129-35.
21. Kini J, Khandikar UN, Dayal JP. A study of the haematological spectrum of Myelo dysplastic syndrome. India J Pathol Microbiol. 2001;44(1):9- 12.
22. Rahim F, Irshad A, Saiful I, Muhammad H, Alikhan TK, Qudsia B. Spectrum of hematological disorders in children observed in 424 consecutive bone marrow aspiration/biopsies. Pak J Med Sci. 2005;21:433-6.

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