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Evaluation of Pancytopenia with Bone Marrow Study in a Tertiary Care Hospital

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Abstract: ***Introduction: Pancytopenia is reduction in all three major formed elements of blood: erythrocytes, leucocytes and platelets. It has been observed in Indian scenario that megaloblastosis –Vit.B-12 and Folate deficiency is the commonest cause of pancytopenia. However, the frequency with which each condition is associated with pancytopenia differs considerably depending upon various factors including geographic distribution. Aims & Objectives: 1. To study the frequency of pancytopenia in patients admitted in tertiary hospital. 2.To study the age wise and sex wise distribution of pancytopenia. 3.Clinicopathological correlation of pancytopenia from clinical findings, haemogram, bone marrow aspirate and / or bone marrow trephine biopsy. Material and Methods: This is a retrospective hospital based cross section study carried out over a period of Two years in the department of pathology at Santhiram medical college & general hospital from Jan 2023 to Dec.2024 through data collected from hospital records. In this period, a total of 60 bone marrow examinations that fulfilled the criteria for pancytopenia were studied. Results: Total 60 cases were studied in the given period. The most common cause of pancytopenia was Megaloblastic anemia seen in 30 patients (50%) followed by Normoblastic erythropoiesis (12%), transient hypoplasia (8%), Aplastic (5%)/Hypoplastic anemia (5%), leukemia (3.5), hypersplenism (3.5%), Mds(2%), Itp(2%) and miscellaneous (9%). Commonest age group of presentation of pancytopenia was between 21-30 years with 19(32%) out of 60 pateints. Females, 31 cases (51.6%) were most commonly affected than males,29 cases (48.4%). Pallor(86.6%) was the most common clinical feature. Conclusion: Among the conditions that causes Megaloblastic anemia due to Vit.B-12/Folate deficiency, i.e. nutritional in origin seems to reflect the higher prevalence of pancytopenia in Indian subjects-which is easily reversible.***

Keywords: Pancytopenia, bone marrow examination, Aplastic anemia

1. Introduction

Pancytopenia" is reduction in all three major classifications of formed elements of blood: erythrocytes, leucocytes and platelets. It is the simultaneous presence of anemia, leucopenia and thrombocytopenia. Thus, it is not an entity by itself, but a triad of finding that may result from number of disease processes[1,3].

Present study was carried out mainly with the twin aims of diagnosing the patients with Pancytopenia and finding out the common disease entities responsible for it.[6,9]

Pancytopenia develops due to decrease in hematopoietic cell production in bone marrow as a result of destruction of marrow tissue by toxins (aplastic/ hypoplastic marrow), replacement by abnormal or malignant tissue or suppression of normal marrow growth and differentiation. In other situations, however, the marrow may be normally cellular or even hypercellular and no abnormal cells may be present. The mechanism leading to pancytopenia in such patients are thought to include ineffective hematopoesis with cell death in marrow, formation of defective cells that are rapidly removed from the circulation, sequestration or destruction of cells by the action of antibodies and trapping normal cells in a hypertrophied and overactive reticuloendothelial system[3].

Criteria applied by people varied in different series. DeGruchy[1] gives the criteria as hemoglobin (Hb) level below 13.5 gm/dl for males and 11.5 gm/dl for females, total leucocytes count (TLC) below 4000/mm^3 and platelet count below 1.5 lakh /mm^3 [1]. However, criteria applied by "Kumar et al"[9] were Hb less than 9 gm/dl, TLC less than 4000/mm^3 and platelet count below 1,40,000/mm^3 [3].

2. Materials and methods

To evaluate the cases of pancytopenia and to their causes this retrospective hospital based cross sectional study was carried out in the department of Pathology. Santhiram medical college, Nandyal. All the patients referred to hematology section of pathology department for routine hematological investigations and peripheral blood smear (PBS) examination, were screened for pancytopenia and a total number of 60 cases were selected based on the following all three criteria-

- Hemoglobin less than 9 gm/dl
- Total leucocyte count less than 4000/mm3 and
- Platelet count less than 1,50,000/mm3

Patients who does not fulfill above criteria were excluded. Biochemical assays were not done.

Detailed hematological investigations were done on the blood as follows:

Hb, MCV, TLC, DLC, platelet count were done on electronic cell counter and also crosschecked by PBS. Informed consent was taken for doing marrow studies.

All PBS and bone marrow aspirates were stained with Leishman stain. Prussian blue stain was done on all bone marrow aspirates to assess bone marrow iron stores. Reticulocyte count by brilliant cresyl blue.

Bone marrow biopsies were processed as per standard method and stained with. Hematoxylin and Eosin and Reticulin stains. Additional stains like PAS etc. were done whenever necessary.

3. Results

The age range of patients in our study was 1 year to 80 years; with the commonest age group for presentation of Pancytopenia was between 21-30 years, a total of 19 cases (32%) belonged to this group. Out of the 60 cases, 29 were males and 31 were females – a slight female preponderance was seen.

Pallor was the most common clinical feature, found in 52 of 60 cases. Weakness/fatigability, breathlessness, fever were other common features. Fever and pedal edema were other common clinical findings. Splenomegaly in 22% and Hepatomegaly in 10% were other findings-both of which were common in Megaloblastic anemia.

Anisocytosis of varying severity was the commonest morphological type (78%). Hypersegmented neutrophilswas the commonest peripheral blood finding in Megaloblastic anemia. Immature cells in form of nucleated RBCs and immature myeloid cells were seen in 12% cases. Macrocytosis with ovalocytosis was seen predominantly in Megaloblastic anemia and in MDS

The Hemoglobin concentrations of patients in the study ranged from 1.7 to 9 gm/dl.

The total leucocyte counts of our patients ranged from 1000-3800/$mm^{3.}$

The platelet counts of patients in our study ranged from 10,000-1 lakh/$mm^{3.}$Majority i.e. 25 cases (42%) had platelet counts between 50,000-1 lakh/$mm^{3.}$10% of the patients had platelet counts below 20,000/mm^3 and majority belonged to Hypoplastic/Aplastic anemia group.

4. Discussion

All patients attending out-patient department and patients admitted, were screened for Pancytopenia as per the defined criteria and 60 such cases were selected.

In our study, the commonest cause of Pancytopenia was Megaloblastic anemia (50%) followed by Hypoplastic/Aplastic anemia (18%), sub-leukemic leukemia (3%) and Hypersplenism (3%).

Other cases were Myelodysplastic syndrome, Myelofibrosis, ITP, Gauchers disease. Cases included in miscellaneous group accounting for 18% comprised of Typhoid, malaria, SLE, and dimorphic anemia.

Comparison of common causes of Pancytopenia reported from various studies done in Indian sub- continent with those of present study is shown in following Table 4, 6, 9, 13, 14.

Table 1: Age and Sex distribution of cases studied

Sr. no.	Age in Years	Male	Female	Total no. of cases	Percentage (%)
1.	<1	--	--	--	0
2.	1-10	3	3	6	10
3.	11-20	7	7	14	24
4.	21-30	8	11	19	32
5.	31-40	4	7	11	18
6.	41-50	4	1	5	8
7.	51-60	1	1	2	3
8.	61-70	1	1	2	3
9.	71-80	1	--	1	2
	Total	29	31	60	100

Table 2: Age distribution in various disorders causing pancytopenia

Sr no.	Age (Inyrs)	Me g	Hyp/Apl	TH	ALeu	MDS	Hyper-spl	Gauc	Myelo fib	ITP	Misc	Total	%
1.	<1	--	--	--	--	--	--	--	--	--	--	--	--
2.	1-10	1	--	1	1	--	--	--	--	--	3	6	10
3.	11-20	5	3	3	1	--	--	--	--	--	2	14	24
4.	21-30	11	1	--	--	--	1	1	1	1	3	19	32
5.	31-40	7	1	1	--	--	1	--	--	--	1	11	18
6.	41-50	4	--	--	--	--	--	--	--	--	1	5	8
7.	51-60	1	--	--	--	--	--	--	--	--	1	2	3
8.	61-70	--	1	--	--	1	--	--	--	--	--	2	3
9.	71-80	1	--	--	--	--	--	--	--	--	--	1	2
	Total	30	6	5	2	1	2	1	1	1	11	60	100

[Meg = Megaloblastic anemia; Hyp/Apl = Hypoplasia/Aplasia; TH = Transient hypoplasia; A Leu = Acute leukemia; MDS = Myelodysplastic syndrome; Hyperspl = Hypersplenism; Gauch = Gaucher disease; ITP = Idiopathic thrombocytopenia purpura; Misc = Miscellaneous]

Table 3: Clinical features

Sr. No.	Clinical Features	Meg	Hyp/ Apl	ALeu	MDS	Hyper-spl	Gauc	Myelo-fib	ITP	SLE	Misc	Total
1.	Pallor	30	11	1	1	1	--	1	-	1	7	52
2.	Weakness/ Fatigability	15	3	1	--	--	1	--	-	-	4	25
3.	Breathlessness	16	6	1	1	--	--	1	-	-	7	34
4.	Fever	7	6	2	--	--	--	--	-	1	4	21
5.	Pedal Edema	9	2	--	1	--	--	--	-		1	14
6.	Icterus	4	1	--	--	--	--	--	-	-	1	6
7.	Bleeding tendencies	2	4	--	--	--	--	--	1	-	2	9
8.	Splenomegaly	6	1	--	--	2	1	1	-	-	2	13
9.	Hepatomegaly	5	1	--	--	--	--	--	-	-	1	7

[Meg = Megaloblastic anemia; Hyp/Apl = Hypoplasia/Aplasia; TH = Transient hypoplasia; A Leu = Acute leukemia; MDS = Myelodysplastic syndrome; Hyperspl = Hypersplenism; Gauch = Gaucher disease; ITP = Idiopathic thrombocytopenic purpura; Misc = Miscellaneous]

Table 4: Hemoglobin concentration in various conditions causing Pancytopenia

Hb (gm%)	Meg	Hyp/ Apl	Aleu	M D S	Hypersp l	Myel ofib	Gau ch	ITP	Misc	Total	%
9-7 (Mild)	1	1	--	--	1	--	1	--	--	4	7
<7-5 (Mod)	5	2	1	1	--	--	--	1	4	14	23
<5-3 (Sev)	13	5	1	--	1	1	--	--	5	26	43
<3	11	3	--	--	--	--	--	--	2	16	27
Total	30	11	2	1	2	1	1	1	9+2	60	

[Hb = hemoglobin; Meg = Megaloblastic anemia; Hyp/Apl = Hypoplasia/Aplasia; TH = Transient hypoplasia; A Leu = Acute leukemia; MDS = Myelodysplastic syndrome; Hyperspl = Hypersplenism; Gauch = Gaucher disease; ITP = Idiopathic thrombocytopenic purpura; Misc = Miscellaneous]

Table 5: Peripheral blood smear findings

S. No.	Etiology	Total Cases	A/P/C	Micro	Macro/ovalo	NN	Hyperseg Neutro	Imm / nRBCs	Rel. lymphocytosis
1.	Meg	30	29	1	29	--	18	2	4
2.	Hyp/Apl	11	6	1	2	1	--	1	4
3.	A leu	2	1	1	--	--	--	2	--
4.	MDS	1	1	--	1	--	--	1	--
5.	Hyperspl	2	2	--	--	--	--	--	--
6.	Myelofib	1	1	--	1	--	--	1	--
7.	ITP	1	1	--	--	--	--	--	--
8.	Gauch	1	--	--	--	1	--	--	--
9.	Misc	11	8	2	--	--	--	2	--
	Total	60	47	5	--	2	18	9	8
	%		78	8	55	3	30	15	13

[A/P/C=anisopoikilocytosis; N N = Normocytic Normochromic; hyperseg neutro = Hypersegmented neutrophils; Macro/ovalo=macrocytosis with ovalocytosis; imm = immature; nRBC = nucleated RBC Meg = Megaloblastic anemia; Hyp/Apl = Hypoplasia/Aplasia; TH = Transient hypoplasia; A Leu = Acute leukemia; MDS = Myelodysplastic syndrome; Hyperspl = Hypersplenism; Gauch = Gaucher disease; ITP = Idiopathic thrombocytopenic purpura; Misc = Miscellaneous]

Table 6: Total Leucocyte Count (TLC) distribution

TLC (In mm^3)	Meg	Hypo/ Apl	A leu	MDS	Hyper spl	Myelo fib	Gauch	ITP	Misc	Total	%
4000-3000	11	5	1	--	1	--	1	1	4	24	40
<3000-2000	10	4	1	1	--	--	--	-	4	20	33
<2000-1000	9	2	--	--	1	1	--	-	3	16	27
<1000	--	--	--	--	--	--	--	-	--	--	--
Total	30	11	2	1	2	1	1	1	9+2	60	

[Meg = Megaloblastic anemia; Hyp/Apl = Hypoplasia/Aplasia; TH = Transient hypoplasia; A Leu = Acute leukemia; MDS = Myelodysplastic syndrome; Hyperspl = Hypersplenism; Gauch = Gaucher disease; ITP = Idiopathic thrombocytopenic purpura; Misc = Miscellaneous]

Table 7: Platelet counts

Platelet count (In $mm^{3)}$	Meg	Hyp/ Apl	A leu	MDS	Hyper-spl	Myelo-fib	Gauch	ITP	Misc	Total	%
<1.5 –1 lakh	7 (23)	--	--	--	--	--	--	--	1	8	13
<1 lakh-50, 000	11 (37)	3 (27)	1 (50)	1 (100)	2 (100)	--	1 (100)	--	6	25	42
<50.000-20,000	11 (37)	6 (55)	1 (50)	--	--	1 (100)	--	--	2	21	35
<20,000	1 (3)	2 (18)	--	--	--	--	--	1 (100)	2	6	10
Total	30	11	2	1	2	1	1	1	9+2	50	

[Meg = Megaloblastic anemia; Hyp/Apl = Hypoplasia/Aplasia; TH = Transient hypoplasia; A Leu = Acute leukemia; MDS = Myelodysplastic syndrome; Hyperspl = Hypersplenism; Gauch = Gaucher disease; ITP = Idiopathic thrombocytopenic purpura; Misc = Miscellaneous] [**Fig. in bracket indicate percentage**

Table 8: Bone Marrow (B.M.) findings

Sr.no.	Bone marrow Findings	No. of cases	%
1.	Megaloblastic Anemia	30	50
2.	Normoblastic erythropoiesis	7	12
3.	Hypoplastic Anemia	3	5
4.	Transient Hypoplasia	5	8
5.	Aplastic Anemia	3	5
6.	AML Sub-leukemic phase	1	2
7.	ALL Sub-leukemic phase	1	2
8.	MDS	1	2
9.	Hypersplenism	2	3
10.	Myelofibrosis	1	2
11.	Gaucher Disease	1	2
12.	ITP	1	2
13.	Miscellaneous	4	7

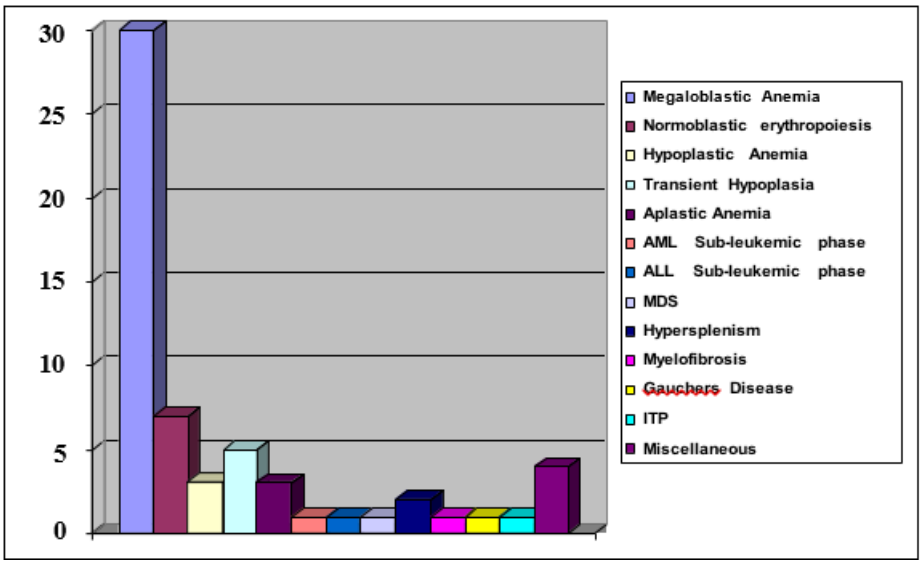

Figure 1: Various etiologies of pancytopenia

X-axis – Etiologies of pancytopenia in present study Y-axis – Number of cases

Table 9: Distribution of Megaloblastic erythropoiesis

Causes	Cases	%
Megaloblastic Anemia	16	53
Megaloblastic + Iron Deficiency anemia	14	47
Total	30	100

Table 10: Distribution of Hypoplastic/ Aplastic anemia

Causes	Cases	%
Hypoplastic Anemia	3	27
Transient Hypoplasia	5	46
Aplastic Anemia	3	27
Total	11	100

Table 11: Classification of various etiologies of Pancytopenia

Sr.no.	Causes	No. of Cases	Percentage (%)
1.	Megaloblastic anemia	30	50
2.	Normoblastic erythropoiesis	7	12
3.	Hypoplastic/ Aplastic anemia	6	10
4.	Transient marrow suppression (Hypoplasia)	5	8
5.	Acute leukemia (sub-leukemic phase)	2	3
6.	Myelodysplastic syndrome	1	2
7.	Hypersplenism	2	3

8.	Storage disorder (Gauchers disease)	1	2
9.	Myelofibrosis	1	2
10.	Idiopathic thrombocytopenic purpura (ITP)	1	2
11.	Miscellaneous	4	7
	Total	60	100

Table 12: Comparison of common causes of pancytopenia with other studies

	Tilak V,Jain R[6], (1999)	Khodke K.et al[4] (2001)	Kumar R.et al[9](2001)	Das R et al[29] (2016)	Present study (2025)
No of cases	77	50	166	64	60
Common causes	Megaloblastic anemia (68%)	Megaloblastic anemia (44%)	Aplastic anemia (29.51%)	Aplastic anemia (19%)	Megaloblastic anemia (50%)
	Aplastic anemia (7.70%)	Aplastic anemia (14%)	Megaloblastic anemia (22.28%)	Megaloblastic anemia (58%)	Hypoplastic/ Aplastic anemia (18%)
	Malaria (3.8%)	Kala azar (14%)	Aleukemic leukemia (12.04%)	Hypersplenism (12%)	Normoblastic Erythropoiesis (12%)
	Kalaazar (2.5%)	Normoblastic erythroid hyperplasia (14%)	Hypersplenism (11.44%)	Acute leukemia (11%)	Hypersple nism & acute leukemias (3% each)

The age range in our study was 1 year to 80 years. The commonest age group for presentation of Pancytopenia was between 21-30 years.

Ot of the 60 cases, 29 were males and 31 were females – a slight female preponderance was seen in contrast to other studies, which reported male preponderance.

Table 13: Age of presentation and commonest sub-type of marrow suppression

Authors	Commonest age group	Commonest sub-type of aplasia/hypoplasia
Young N.S.[27] (1991)	15-25 years	Idiopathic
Tilak V.[6] (1999)	20-30 years	Idiopathic
Das R [29] (2016)	20-30 years	Idiopathic
Present Study	10-20 years	Transient hypoplasia(secondary causes)

Our findings are comparable with above studies, indicating that Aplastic/Hypoplastic anemia is a disorder of younger age group with 83% cases occurring below 30 years of age. The age profiles in acute leukemias differ, but overlap completely. ALL is predominant in childhood and AML in adults. Out of 2 cases of sub-leukemic phase of acute leukemia, 1 was in first decade and of the Lymphoblastic type. The other belonged to second decade, and was Acute Myeloid Leukemia. Thus our findings are compatible with clinical age profiles of acute leukemias.

Laboratory Data and Clinical features: Patients with Pancytopenia presented with a variety of clinical features, but some common clinical features were considered.

Table 14: Common Clinical features in various studies – Compared

Authors	Commonest Presenting feature	2nd commonest feature	3rd commonest Feature	4th commonest Feature
Khodke K. et al[4] (2001)	Pallor	Fever	Weakness	Splenomegaly
Niazi M. et al[26] (2004)	Pallor	Weakness	Fever	Bleeding Tendencies
BN Gayathri et al[30](2005)	Pallor	Breathlessness	Fever	Weakness
Present study	Pallor	Breathlessness	Weakness	Fever

Table 15: Peripheral Blood Smear Findings

Authors	Anisocytosis	Hypersegmented Neutrophils	Immature cells (myeloid+ erythroid)	Relative lymphocytosis
Khodke K. et al[4] (2001)	60%	40%	12%	10%
Jha et al [28] (2008)	60%	45%	1.29%	14.28%
Present study	78%	30%	12%	12%

In our study, 18 cases (60%) of Megaloblastic anemia had hemoglobin levels ranging from 3 to 7 gm/dl suggesting moderate to severe degree of anemia

In the present study we had 1 case of MDS with hemoglobin <5gm/dL; TLC between 2000-3000mm^3 showing circulating immature cells upto myelocytes; platelets from 50,000-1,50,000/mm^3. Progressive pallor, breathlessness, pedal edema were commonest presenting features. All above studies are comparable with Koeffler HP et al, Foucar K et al, Linman JW et al, Pendry K et al cited in Wintrobe`s Clinical Hematology, 11th edition, 2004.

Bone Marrow examination: Out of 30 cases, 16 cases (53%) showed pure Megaloblastic anemia of varying severity whereas 14 cases (47%) showed combination of iron deficiency and Megaloblastic anemia in varying proportions. Sen R. et al (1996) too reported 75 cases of pure Megaloblastic anemia and 36 cases of combination of iron deficiency and Megaloblastic anemia (vit. B12/ folate).

This reflects the high prevalence of nutritional anemias in Indian subjects. As facilities for estimating vit.B_{12} / folate levels are not available in our institute, the exact deficiency could not be identified.

In addition we had one case of Gaucher disease where patient presented with abdominal pain, weakness and fatigability, with marginally reduced TLC, mild anemia and thrombocytopenia. Marrow aspiration revealed Gaucher cells- demonstrated by Periodic Acid Schiff stain (PAS).

One case of SLE, proven by serology, presenting with pancytopenia was identified. Marrow could not be done in this case, as platelet counts were very low.

Tilak V. et al[6] (1999) and Kumar R. et al[9] (2001) reported 3 and 5 cases of malaria in their respective studies. We had 2 cases of malaria – one was 19 years male with cerebral malaria presenting with pancytopenia, expired before marrow could be done. Other was 10 years female having mixed infection with typhoid and malaria on treatment. Whether the pancytopenia was due to infection or drugs could not be ascertained.

Since severity of pancytopenia and the underlying pathology determines the management and prognosis of these patients, treatment should commence at the earliest possible to prevent the unacceptable serious complications of pancytopenia. If all the elements of the complete blood count are considered in clinical context including physical examination, they can provide invaluable information to guiding the possible causes for pancytopenia and helping in planning the work up of tests needed for definite diagnosis.

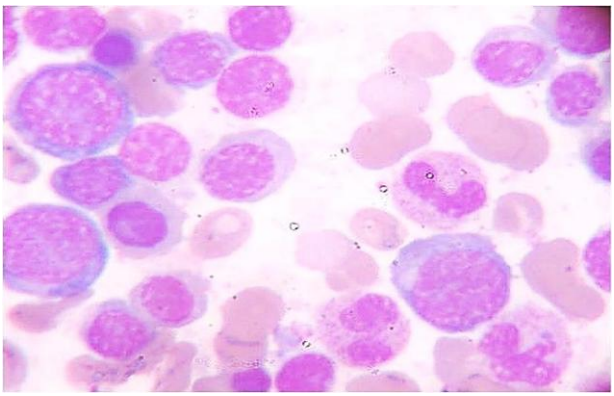

Figure 1: BMA showing Megaloblastic and normoblastic maturation, (Leishmann stain 100 ×).

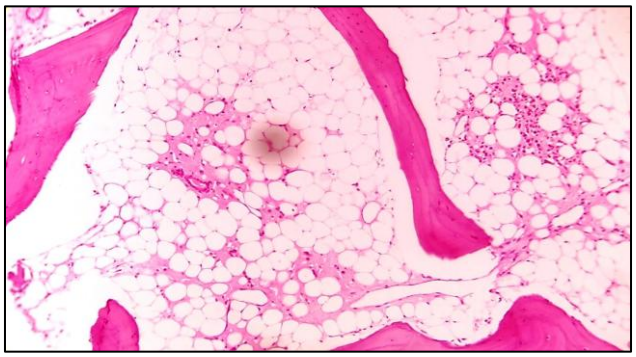

Figure 2: Bone marrow biopsy of aplastic anemia (10x, H&Estain)

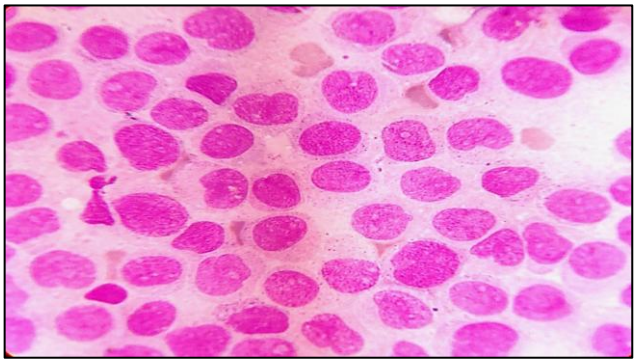

Figure 3: Acute myeloid leukemia.Bone marrow aspiration smear showing myeloblasts. (Leishmann stain 100 ×).

5. Conclusion

The common cause of pancytopenia was Megaloblastic anemia followed by aplastic/Hypoplastic anemia. Anisocytosis of varying degree was the most common morphological type.

Megaloblastic anemia due to Vit. B12/Folate deficiency, i.e. nutritional in origin seems to reflect the higher prevalence of pancytopenia in Indian subjects- which is easily reversible with treatment, thus putting Aplastic/Hypoplastic anemia in second position, which is the most common cause for pancytopenia in western countries.

However, other causes like hypersplenism, Myelofibrosis, MDS, Leukemia, post-infectious etc. should also be kept in mind while planning has treatable cause (Megaloblastic, Hypersplenism, Transient hypoplasia, and ITP) and so carried a better prognosis.

Thus, with the help of detailed clinical history, physical examination and hematological investigations, pancytopenia can be diagnosed and causes can be ascertained.

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