Journal of Population Therapeutics & Clinical Pharmacology

ORIGINAL RESEARCH ARTICLE
DOI: 10.53555/36gs1606

MEGALOBLASTIC ANAEMIA THE COMMONEST CAUSE OF PANCYTOPENIA IN ADULT POPULATION, A DESCRIPTIVE STUDY IN A TERTIARY CARE CENTER

Dr. Simi Sidharthan[1*], Dr. Shameem Ummer Ali[2], Dr. Prabhalakshmi[3]

[1*]Assistant Professor, Department of Pathology, Government Medical College, Kozhikode, Kerala, India.
[2]Professor & HOD, Department of Pathology, Al-Azhar Medical College and Super-specialty Hospital, Thodupuzha, Kerala, India.
[3]Professor & HOD, Department of Pathology, Government Medical College, Thrissur, Kerala, India.

***Corresponding Author :** Dr. Simi Sidharthan
Assistant Professor, Department of Pathology, Government Medical College, Kozhikode, Kerala, India.

ABSTRACT

Background

Pancytopenia is defined as reduction of all three formed elements blood like Rbcs,Wbcs and platelets below the normal reference ranges. It is the manifestation of a number of disease processes affecting the bone marrow. Aim to identify the various causes of pancytopenia in adult population by bone marrow examination, then the most common cause of pancytopenia and to find the frequency the most common cause of pancytopenia in relation to age and sex. This is two years description study.

Methods

Peripheral smears, buffy coat smears, marrow aspiration smears and trephine biopsies of thirty six cases were studied. Special stain done for suspicious cases of leukemia and lymphoma. Relevant clinical history and investigations were collected. Peadiatric cases and patients on chemotherapy were excluded from this study.

Results

Megaloblastic anaemia was the most common cause of Pancytopenia followed by Aplastic anaemia. Female are more common than male. Common among elderly population in their sixth decade. Peripheral smear showed characteristic features like macrocytes, macroovalocytes and hypersegmented neutrophils. Erythroid hyperplasia was seen in 18 cases of megaloblastic anaemia. Both peripheral smear and bone marrow findings were statistically significant.

Conclusion

Bone marrow aspiration and trephine biopsy are mandatory for the diagnosis of the various causes of pancytopenia. Macroovalocytes and hypersegmented neutrophils in peripheral smear, erthyroid hyperplasia with megaloblastic maturation helps in definitive diagnosis of Megaloblastic anaemia. Comprehensive clinical and hematological study on pancytopenia cases is needed to identify the cause with certainty.

Keywords: Megaloblastic anaemia, pancytopenia, peripheral smear, bone marrow aspiration, trephine biopsy.

INTRODUCTION

Megaloblastic anemia (MA), a common clinical disease, is characterized by weakness, palpitations, fatigue, light-headedness, and shortness of breath caused by low hematocrit.[1] Vitamin B12 or folic acid deficiency is the primary cause of malnutrition and is more prevalent in developing countries where malnutrition remains problematic. One study found a significantly greater prevalence in certain sub-populations of African and Asian countries, particularly Kenyan schoolchildren (70%), Indian preschool children (80%), and Indian adults (80%).[2] Pancytopenia is the reduction in three peripheral blood cell lines: erythrocytes, leukocytes, and platelets.[3] Pancytopenia is an important clinical-hematological entity in which the clinical pattern varies and thus treatment modalities and outcomes will also vary.[4] Pancytopenia is not a disease but a triad of anemia, leucopenia, and thrombocytopenia that may result from a number of disease processes.[5] Criteria for pancytopenia hemoglobin is less than 13.5g/dl in males, 11.5g/dl in females, the WBC count <4 x 109/L and platelet count <150x109/L.[3]

AIMS AND OBJECTIVE

To study the peripheral smear and bone marrow picture of megaloblastic anemia presenting as pancytopenia.

MATERIALS & METHODS

Peripheral smear, bone marrow aspirate, and bone marrow trephine of all pancytopenia cases during two years (2009 and 2011) were examined following ethical clearance. Among them the age, clinical features, peripheral smear, and bone marrow picture of megaloblastic anemia cases were studied.

Inclusion Criteria

All adult cases of megaloblastic anemia presenting as pancytopenia whose peripheral smear and bone marrow are available.

Exclusion Criteria

1. Cases with inadequate details
2. Paediatric cases.

Study Method

Clinical details and physical findings of the selected cases of megaloblastic anemia among pancytopenia were documented. Their peripheral and buffy coat smears were stained with Leishman stain and studied. Bone marrow study was done in these cases for further evaluation and confirmation.

Bone marrow aspiration and Trephine biopsies were done from the posterior superior iliac spine using a Jamshidi needle. Bone marrow aspirate and Imprint smears of the trephine biopsies were stained with Giemsa/Leishman stain. Bone marrow trephine biopsies were fixed in Bouin's fluid, decalcification was done in 5% nitric acid, and processed in the Histokinette. Trephine biopsies were stained in Haematoxylin and Eosin, Reticulin. Peripheral smears, bone marrow aspirates, and trephine biopsies of megaloblastic cases were examined under low power, high power, and oil immersion and were analyzed. Pancytopenic patients with macrocytic anemia whose bone marrow study was not available were excluded from the study. Our study was on megaloblastic anemia among 36 cases of pancytopenia where peripheral smears, bone marrow aspirate smears, and trephine biopsies were available.

Peripheral smear, buffy smear, marrow aspiration, and trephine biopsy of 36 cases of pancytopenia were studied and megaloblastic anemia constitute 19 cases which was the most common cause of pancytopenia. The characteristic peripheral smear and bone marrow picture of megaloblastic anaemia were studied.

Statistical Analysis

Data was entered in Excel 8. The analysis was done in Epi Info software 2008.

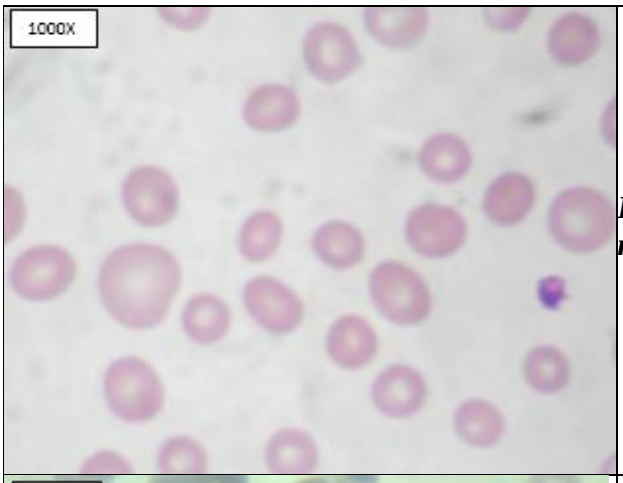

Image 1: Peripheral smear showing macrocytes and macroovalocytes

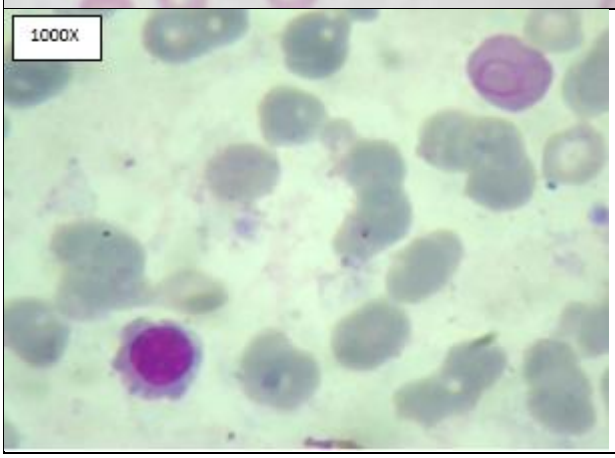

Image 2: Peripheral smear showing normoblast and cabot ring

RESULTS

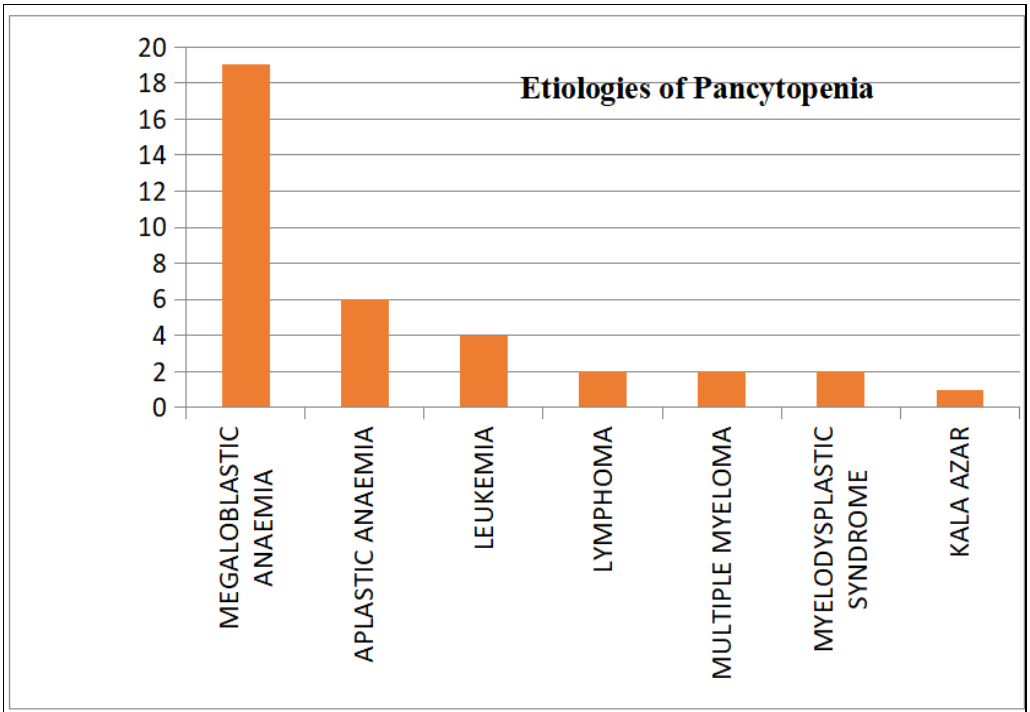

Figure 1 Etiology of pancytopenia after bone marrow examination

Present study of 36 cases of Pancytopenia showed Megaloblastic anaemia in 19 cases, Aplastic anaemia in 6 cases, Leukemia in 4 cases, Lymphoma in 2 cases, Multiple myeloma in 2 cases.

Sl. No	Study	Age in Years	Year of Study	Duration of Study	No of Cases	1st Common Cause	2nd Common Cause
	Tariq et al	>14 yrs	2008	10 months	50	Aplastic anaemia	Megaloblastic anaemia
2.	Gayathri et al	2 -80 yrs	2005 -2007	2 years	104	Megaloblastic anaemia	Aplastic anaemia
3.	Osama et al	12 – 82 yrs	2001	1 year	100	Megaloblastic anaemia	Aplastic anaemia
4.	Khodke et al	3 -69 yrs	1999	6 months	50	Megaloblastic anaemia	Aplastic anaemia
5.	Present study	18- 80 yrs	2009 -2011	2 years	36	Megaloblastic anaemia	Aplastic anaemia

Table 1 Comparison with various studies

Age	Cases	Percentage
11-20	1	5.6%
21-30	2	11.1%
31-40	4	21.0%
41-50	3	16.7%
51-60	3	11.1%
61-70	5	26.3%
71-80	1	11.1%
Total	19	100%
Distribution of cases according to age		
Gender	**Cases**	**Percentage**
Female	12	66.7%
Male	7	33.3%
Total	19	100%
Distribution of cases according to gender		
Table 2		

Symptoms	No of Cases	Sign	No of Cases
Pallor	19	Lymphadenopathy	1
Weakness	15	Splenomegaly	1
Breathlessness	6	Hepatomegaly	1
Skin pigment/bleeding	3		
Table 3: Sign and symptoms of megaloblastic anaemia			

Haemoglobin	Cases
< 5 gm%	10
5.1 - 9 gm%	9
>9.1 gm%	0
Total Count/cumm	**Cases**
0-1000	7
1001-3000	11
3001-4000	1
Platelet Count/cumm	**Cases**
< 20000	6

20000 – 50000	10
50000 - 100000	3

Table 4: Distribution of haemoglobin, Total WBC count and platelet count in megaloblastic anaemia cases

Peripheral Smear Findings	**Cases**
Anisocytosis	19(100%)
Poikilocytosis	16(61.1%)
Macrocytes	19(100%)
Macro Ovalocytes	14(74%)
Nrbc/100wbc's	5(26%)
Hypersegmented Neutrophils	8(42%)

Table 5: Peripheral smear findings observed in megaloblastic anemia

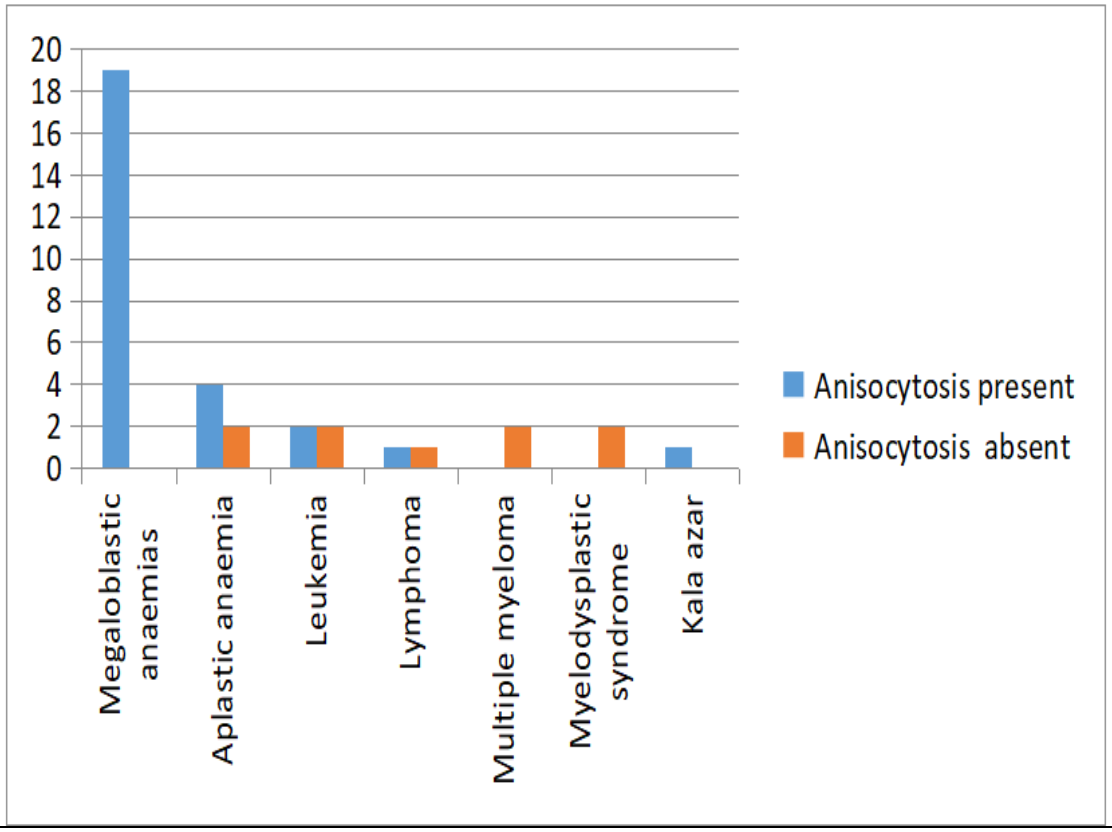

Figure 2: Comparison of Anisocytosis between megaloblastic anaemia and with rest of the etiologies of pancytopenia

In this study anisocytosis was noted in 100% of megaloblastic anaemia and 47.05% in other causes of pancytopenia 'p' value: 0.008 (significant) (***Figure 2)***

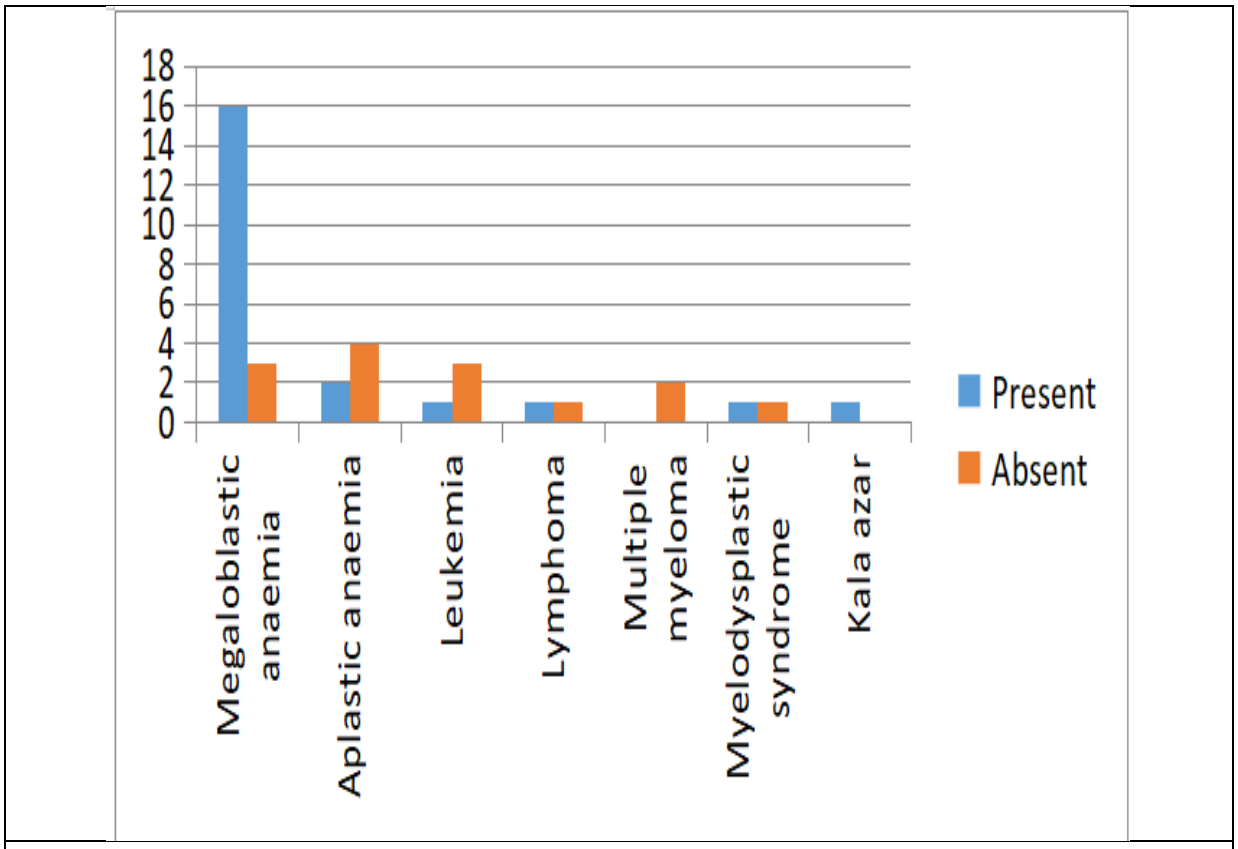

Figure 3: Comparison of poikilocytosis between megaloblastic anaemia and with rest of the etiologies of pancytopenia

Present study showed poikilocytosis in 84.21% of megaloblastic anaemia and rest of the disease group showed 47.05% of poikilocytosis.'p' value: 0.007 (significant)

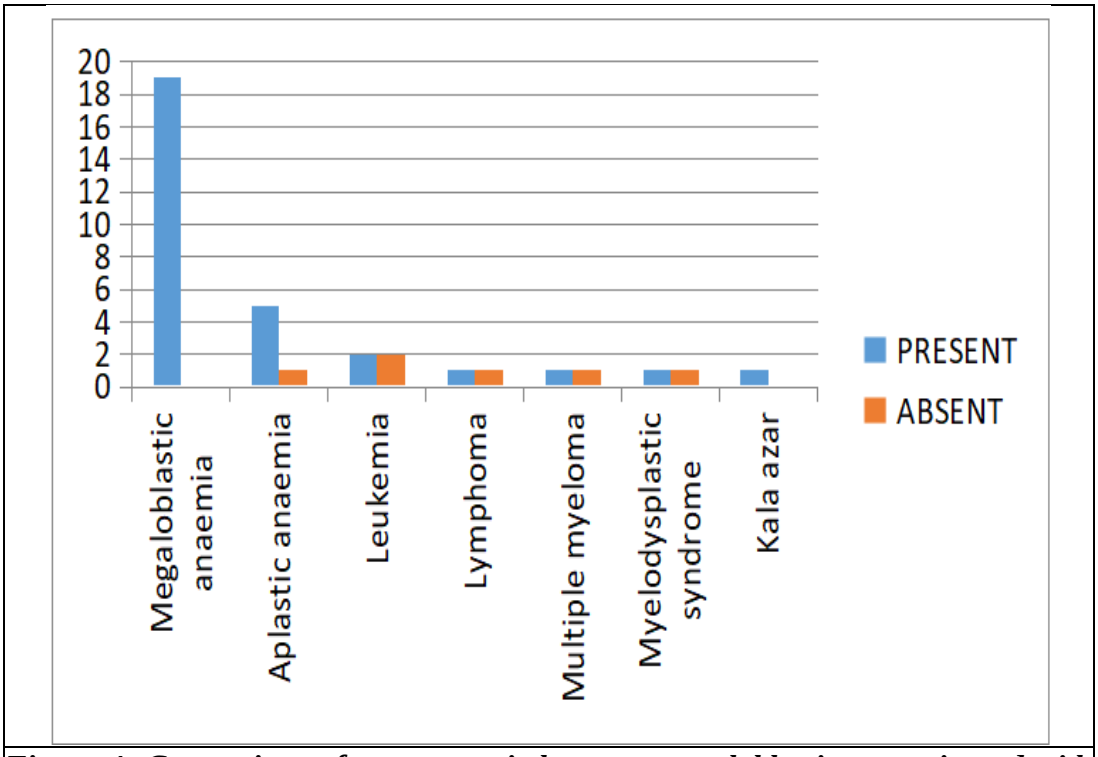

Figure 4: Comparison of macrocytosis between megaloblastic anaemia and with rest of the disease groups of pancytopenia

Present study showed presence of macrocytes in 100% of cases of megaloblastic anaemia and presence of macrocytes in 64.70 % of cases in rest of the disease groups. ‘p’ value: 0.006% (significant).

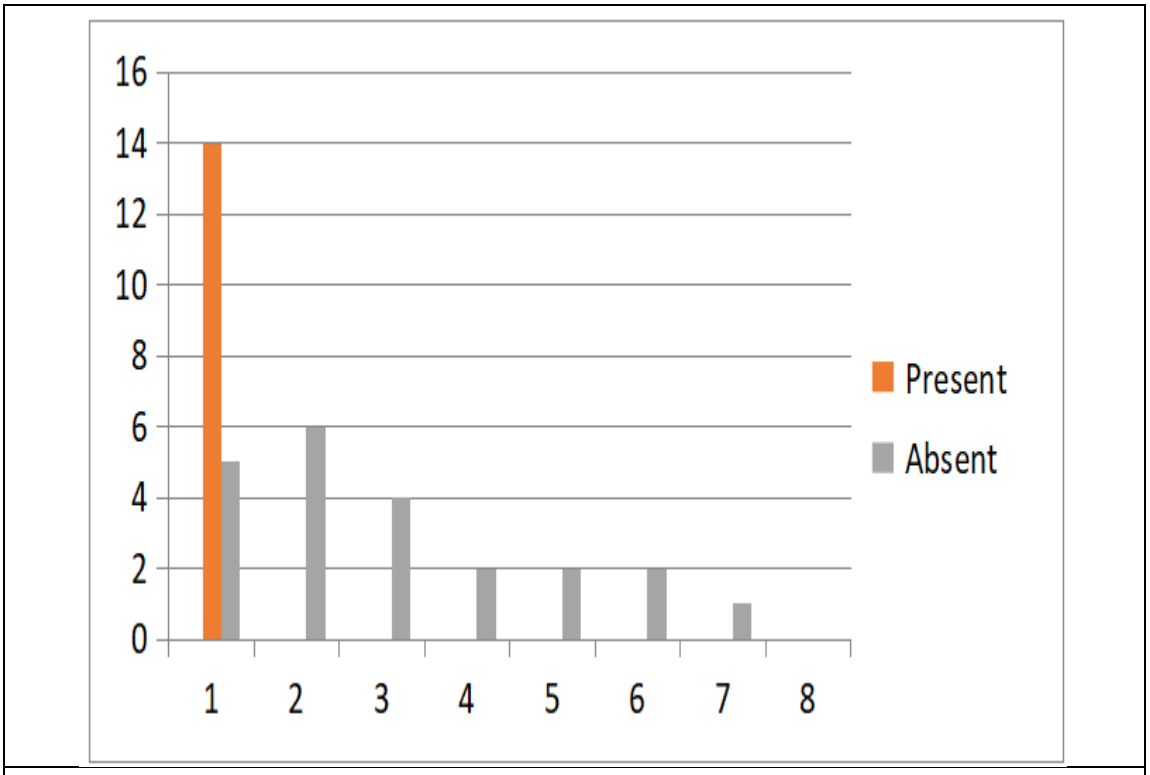

Figure 5: Comparison of macroovalocytes between megaloblastic anaemia and with rest of the disease groups of pancytopenia

Present study showed macro ovalocytes observed in 73.68% of megaloblastic anaemia cases ‘p’ vaue 0.000285.

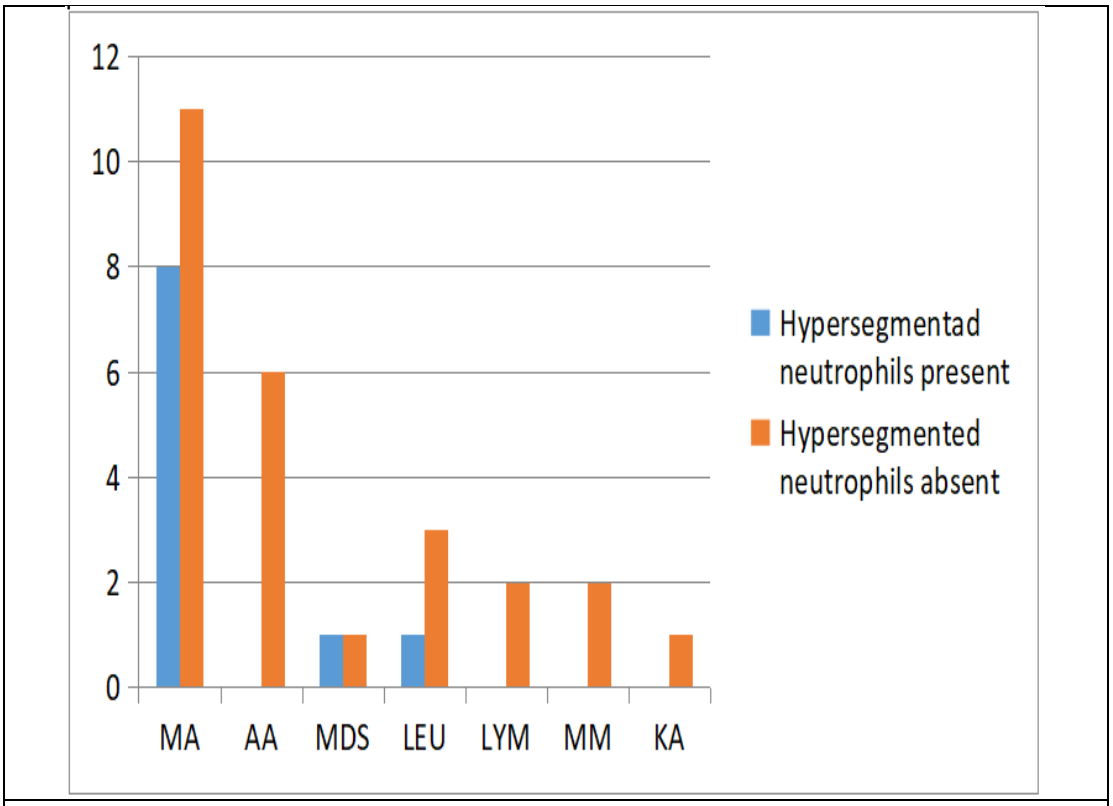

Figure 7: Comparison of hypersegmented neutrophils between megaloblastic anaemia and with rest of the disease groups of pancytopenia

Present study showed hypersegmented neutrophils in 42.10% of megaloblastic anaemia and rest of disease groups showed 11.76%. 'p' value: 0.046 (significant).

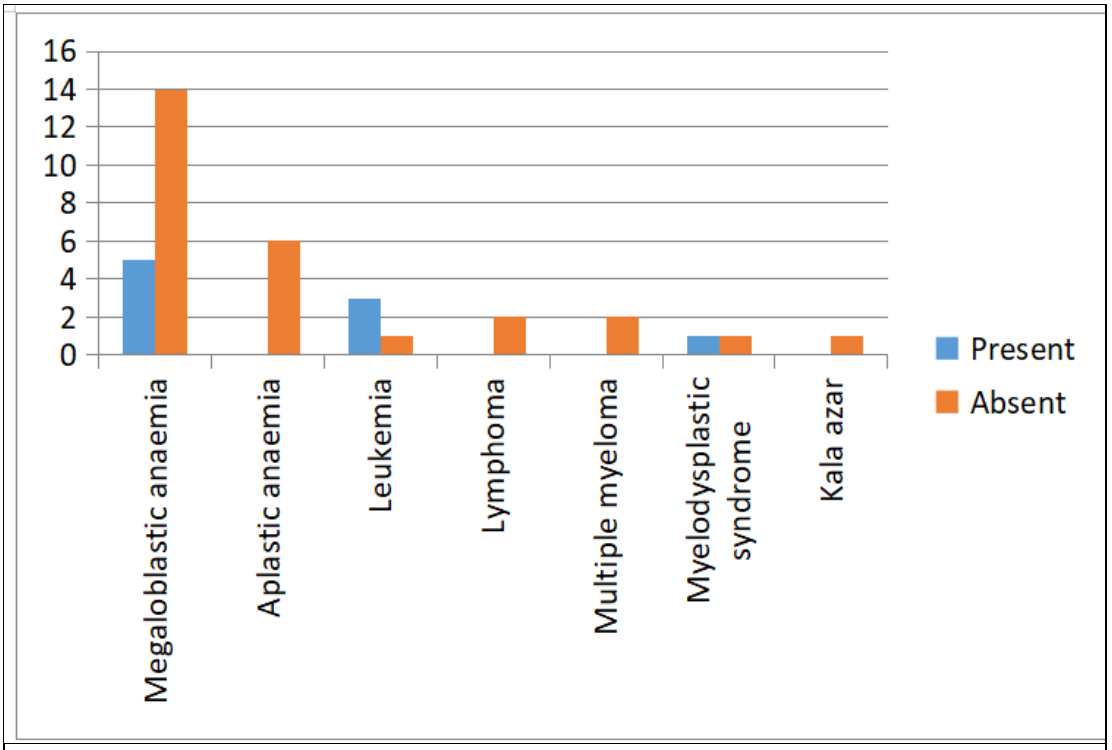

Figure 8: Comparison of presence normoblast between megaloblastic anaemia and with rest of the disease groups of pancytopenia

Normoblast were observed in 26.31% of Megaloblastic anaemia and rest of disease groups showed 23.53%. ‘p’ value: 0.577 (not significant).

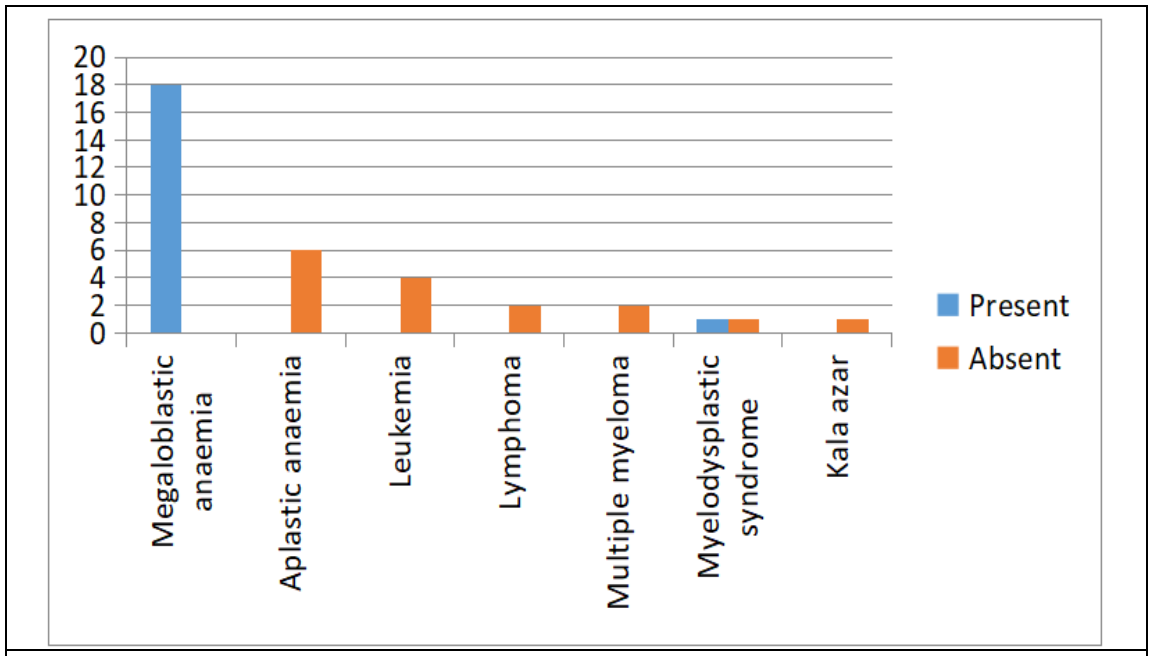

Figure 9: Comparison of presence of Erythroid hyperplasia between megaloblastic anaemia and with rest of the disease groups of pancytopenia

Present study showed erythroid hyperplasia in 94% of megaloblastic anaemia and 5.88% in rest of disease groups. ’p’ value: 0.0005 (significant).

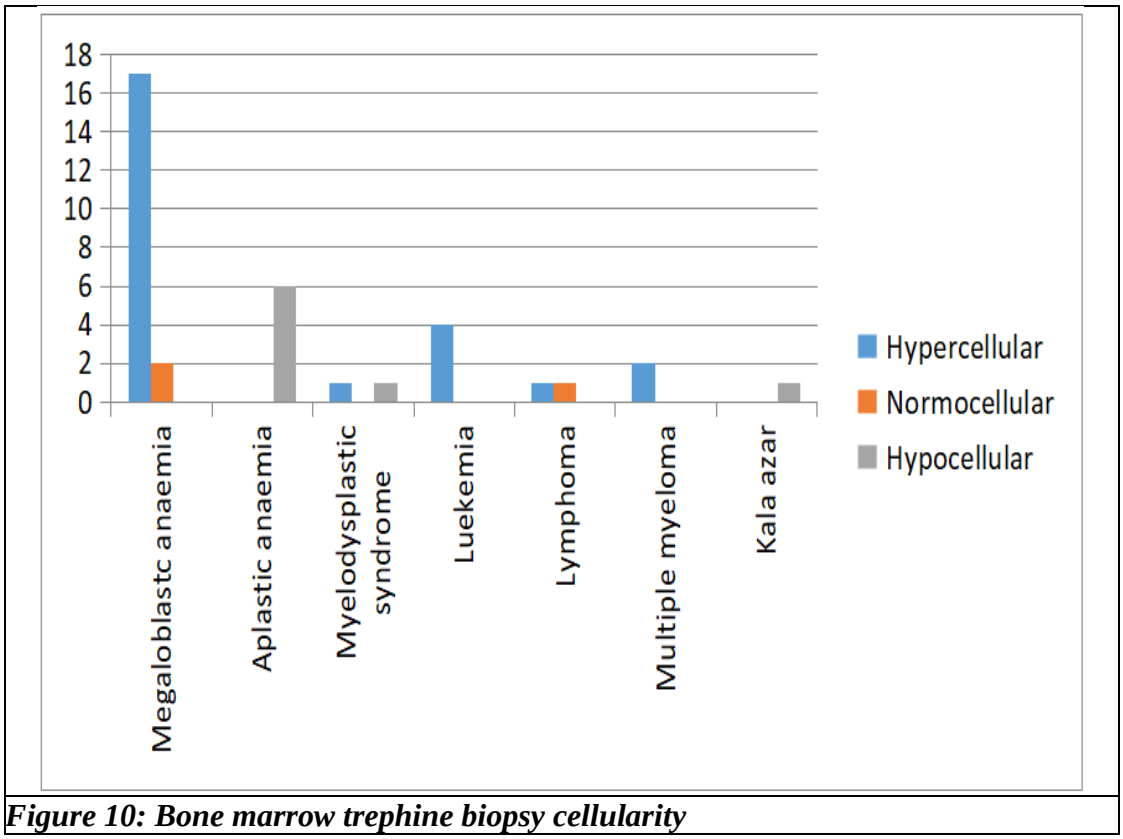

Figure 10: Bone marrow trephine biopsy cellularity

Present study showed bone marrow cellularity in 89.47% of megaloblastic anaemia and rest of disease groups showed 47.08%. 'p' value: 0.0008 (significant).

DISCUSSION

A total of 36 cases of pancytopenia were studied in two year study in which Megaloblastic anemia was the commonest cause which constitute about 19 cases. Age, gender, presenting complaints, peripheral blood picture, bone marrow aspirate smears, and bone marrow trephine biopsy were studied in all cases of pancytopenia with special reference to megaloblastic anaemia and observations were compared with those in studies published in the literature.

Megaloblastic anemia (52.8%) was the commonest cause of Pancytopenia (Fig. 1) followed by Aplastic anemia (16.67%) This was comparable to the study done by Kishore Khodke et al,[6] Gayathri et al[7] and Osama Ishtiaq et al[8] (table 1) who also got Megaloblastic anemia as the commonest cause of pancytopenia with the percentage of 44%, 39%, and 74.04% respectively. Tariq et al[9] got megaloblastic anaemia as the second commonest cause of pancytopenia. This seems to reflect the higher prevalence of nutritional anemia in Indian subjects as well as in developing countries.

The present study of megaloblastic anaemia were more common among 61 to 70 years (Table 2) similar to the study done in the Caucasian and Chinese populations where Megaloblastic anemia is reported to occur more in older age groups.[10]

In the our study Megaloblastic anemia was more common among female with 66.7% (table 2) similar to a study done by Uma Khanduri and Archana Sharma[11] in 175 cases of Megaloblastic anemia in which 71% were female patients.

The most common presenting symptom in this study was weakness (86%) followed by breathlessness (16.67%) (Table 3).This is similar to the study by Kishore Khodke et al in which 40% of cases showed weakness, another study by Mussarat et al[12] in which 68.2% of cases and Gayathri et al in which 100% of cases showed weakness. The present study showed pallor as the most common sign (100%) followed by hepatomegaly and splenomegaly(Table 3) A similar finding was also seen in the study by Kishore Khodke et al, Mussarat et al, Gayathri et al and Santra et al.[13]

The present study showed megaloblastic anaemia cases with Haemoglobin value less than 5gms% ranged were more common.This was similar to the study done by Santra et al in which cases with Haemoglobin value less than 5 gm% was more common. (Table 4). 61.1% of cases showed leukocyte count between the range of 1000/cumm to 3000/cumm (Table 4). This was comparable to the study by Santra et al which showed 54.94% of cases with the total leukocyte count between 1000/cumm to 3000/cumm. Platelet count in present study showed 33.8% of megaloblastic anaemia cases in the range of 20000 to 50000 (Table 4)

In this study of Peripheral smear examination (Table 5) Megaloblastic anemia shows anisocytosis (100%) (Fig. 2) and poikilocytosis (84 %) with a 'p' value of 0.008 (significant) (fig. 3) and 0.007(significant %) which was comparable to the study by Osama Ishtiaq et al[9] in which all cases of megaloblastic anemia shows poikilocytosis. Macrocytes were seen in all cases of Megaloblastic anemia in the present study with the 'p' value 0.006% (significant) (fig. 4). Macro ovalocytes (fig. 5) were noted in 15 cases of pancytopenia (41.7%) and among them, 14 patients (73.68%) had Megaloblastic anemia with 'p' value 0.000285(significant) (fig. 6), these observations were comparable to the findings of Osama Ishtiaq et al.

Hypersegmented neutrophils in 10 cases which constitute 27.8% of the total number of 36 cases of pancytopenia and among which 22.22% of cases were contributed by Megaloblastic anemia with a 'p' value of 0.046 (significant) (fig. 7). This finding was comparable to the study by Gayathri et al and Tilak et al.[14] Normoblast was noted in 5 cases of megaloblastic anaemia, 3 cases of leukemia and 1 case of MDS. Kishore et al[4] noted 8% (4/50) of circulating erythroblast(fig. 8) in their study

in which 2 cases were of megaloblastic anemia, 1 case was of leukemia and 1 case from Myelodysplastic syndrome.
Erythroid hyperplasia was noted in 19/36 cases in this study, Megaloblastic anemia shared 18 cases (94%)(fig. 9) and one case was MDS. Bone marrow aspiration showed megaloblastic erythroid maturation with the characteristic feature of sieved nuclear chromatin, asynchronous nuclear and cytoplasmic maturation, giant metamyelocytes and giant band forms. This finding was similar to the study of Gayathri et al in which 74.04% of cases showed megaloblastic maturation. Erythroid hyperplasia was a significant finding in megaloblastic anemia with a 'p' value 0.0005. One case showed erythroid hyperplasia with megaloblastic maturation, dysmyelopoeisis with hypogranular neutrophils and large hypolobated megakaryocytes this was diagnosed as Myelodysplastic syndrome.

The present study showed bone marrow trephine hypercellularity (fig. 10) in 69.44% cases (fig. 13) of which 17 cases showed megaloblastic maturation with giant metamyelocyte and giant band forms Finding was comparable to findings noted by Santra et al. Normal bone marrow cellularity was noted in 3 cases, among which 2 were megaloblastic anaemia and 1 was lymphoma. Hypocellularity in bone marrow trephine was noted in 8 cases in which bone marrow of 6 cases showed replacement of normal hematopoietic cells by fat cells in all patients There was a relative increase in plasma cells and lymphocytes and was diagnosed to be Aplastic anaemia.

CONCLUSION

Megaloblastic anaemia was the commonest cause of pancytopenia in this two years study, this seems to reflect the higher prevalence of nutritional anaemia in our population. Females were found to be more common than males. Commonest age group in this study were between 60 to 70 years. The haematological parameters in patients with megaloblastic anaemia were comparable to the findings of other authors. Uncommon etiologies like leukemia, multiple myeloma, lymphoma, myelodysplastic syndrome and kala azar were also identified in this study. Though the results of this study was in concordance with the other studies in India, the main limitation that we encountered was that of the small sample size and which may probably be due to the unavailablity of bone marrow aspirates and trephine biopsies in many of the pancytopenic patients. Paediatric cases and patients on chemotherapy were excluded from this study.

ACKNOWLEDGEMENT: I would like to extend my sincere gratitude to everyone who have supported and contributed for this study.

REFERENCES

1. Green R, Datta Mitra A. Megaloblastic anemias: nutritional and other causes. Med Clin North Am 2017;101(2):297–317.
2. Hunt A, Harrington D, Robinson S. Vitamin B12 deficiency. Br Med J 2014;349:g5226.
3. Suthar NR, Sharma R, Madhulata. Pancytopenia: etiology and it's variables. J Assoc Physicians India 2022;70 (4):11-2.
4. Niazi M, Fazl-i-Raziq. The incidence of underlying pathology in pancytopenia – an experience of 89 cases. 2004;18(1):76-9.
5. Khunger JM, Arculselvi S, Sharma U, Ranga S, Talib VH. Pancytopenia-A Clinico-haematological study of 200 cases. Indian J Pathol Microbiol 2002;45(3):375-9.
6. Khodke K, Marwah S, Buxi G, Yadav RB, Chaturvedi NK. Bone marrow examination in cases of pancytopenia. JIACM 2001;2(1-2):55-9.
7. Gayathri B N,Kadam Satyanarayanan Rao.Pancytopenia: a clinico Hematological study, Journal of Laboratory Physicaians 2011;3(1):15-20.

8. Ishtiaq O, Baqai HZ, Anwer F, Hussain N. Patterns of pancytopenia patients in a general medical ward and a proposed diagnostic approach. Journal of Ayub Medical College Abbottabad 2004;16(1).
9. Tariq M, Khan N, Basri R, Amin S. Aetiology of pancytopenia. Professional Med J 2010;17(2):252-6.
10. Chan JCW, Lui HSY, Kho BCS, Chu RW, Ma Esk, Ma KM, et al ,Megaloblastic anaemia in Chinese patients: a review of 52 cases. Hong Kong Med J 1998;4:269-74.
11. Khanduri U, Sharma A. Megaloblastic anaemia: prevalence and causative factors. The National Medical Journal of India 2007;20(4):172-75.
12. Niazi M, Raziq F. The incidence of underlying pathology in pancytopenia-an experience of 89 cases. JPMI 2000;18(1):76-9.
13. Santra G, Das BK. A cross-sectional study of the clinical profile and aetiological spectrum of pancytopenia in a tertiary care centre. Singapore Med J 2010;51(10):806.
14. Tilak V. Jam R. Pancytopenia - a clinic-hematologic analysis of 77 cases. Indian J Pathol Microbiol 1999;42:399-404.