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Frequency of Pancytopenia Among Patients with Megaloblastic Anemia

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ABSTRACT

Background: Megaloblastic anemia is common nutritional disorder caused by deficiency of vitamin B12 and folate which lead to defective synthesis of deoxyribonucleic acid and ineffective hematopoiesis. Pancytopenia may occur as complication and can cause serious infections and bleeding. **Objective:** To determine the frequency of pancytopenia among patients with megaloblastic anemia. **Study Design:** Cross sectional study. **Duration and Place of Study:** This study was conducted from 01-12-2023 to 01-06-2024 in General Medicine Department, Saidu Group of Teaching Hospital, Swat. **Methodology:** A total of 165 patients aged 18–65 years diagnosed with megaloblastic anemia were included. Pancytopenia was defined as red blood cell count $<4\times10^{12}$/L, white blood cell count $<4\times10^{9}$/L and platelet count $<150\times10^{9}$/L. Data were analyzed using Statistical Package for Social Sciences version 21. Post stratification chi-square test and Fisher exact test were applied. **Results:** Mean age was 41.77 ± 13.96 years. Pancytopenia was found in 123 (74.50%) patients while 42 (25.50%) did not have pancytopenia. No significant association was observed between pancytopenia and age (p = 0.395), gender (p = 0.891), education (p = 0.755), employment (p = 0.197), economic status (p = 0.966), diabetes mellitus (p = 0.866) and hypertension (p = 0.185). **Conclusion:** Pancytopenia is highly frequent among patients with megaloblastic anemia and majority of cases were affected. Demographic factors were not showing significant association.

INTRODUCTION

Megaloblastic anemia is a type of macrocytic anemia caused by impaired synthesis of DNA, which leads to the formation of large, immature cells called megaloblasts.[1] Impaired synthesis of DNA is caused by a deficiency of Vitamin B12 and folate, as these two play an important role as cofactors in nuclear maturation. Impaired nuclear maturation, compared with cytoplasmic maturation, leads to nuclear-cytoplasmic asynchrony, which causes these cells to be large with open nuclei.[2] Clinical manifestations include pallor, weakness, fatigue, and sometimes shortness of breath. Laboratory tests reveal low hemoglobin concentration, with a mean corpuscular volume above 100 fL. Macro-ovalocytes and hypersegmented neutrophils may be seen in peripheral blood smear. Hypercellularity with megaloblastic changes may be seen in bone marrow. [3]

Pancytopenia, as a manifestation of megaloblastic anemia, is a consequence of ineffective hematopoiesis, which affects not only erythropoiesis but also leukopoiesis and thrombopoiesis.[4] Defective DNA synthesis leads to the destruction of blood cells within the bone marrow, resulting in pancytopenia. Patients may present with a variety of signs and symptoms, which include recurrent infections due to low white blood cell counts, a tendency for bleeding, such as bleeding gums, and signs of anemia.[5] Laboratory tests will show low hemoglobin levels, a total leukocyte count below the normal range, and a low platelet count. Pancytopenia is a manifestation of a disease but is not a disease by itself. Among the common causes of pancytopenia in developing countries is megaloblastic anemia. [6]

The treatment of megaloblastic anemia accompanied by pancytopenia is dependent upon the treatment of the underlying cause and the appropriate supportive care.[7] In cases where vitamin B12 deficiency is diagnosed, the recommended treatment is the initial administration of vitamin B12, especially in severe cases and neurological manifestations.[8] Folic acid supplementation is recommended in cases of folate deficiency. In folate deficiency, it is important to note that vitamin B12 deficiency should first be ruled out to avoid exacerbating neurological damage by administering folate.[9] In cases where patients present with severe anemia and pancytopenia, blood and platelet transfusions may be indicated.[10]

There is a scarcity of data on the incidence of pancytopenia among patients with megaloblastic anemia in Swat. Nutritional deficiencies, especially vitamin B12 and folate deficiencies, are common in this area due to

socioeconomic factors and dietary habits, but the actual incidence of pancytopenia is not well documented. It is important to diagnose pancytopenia to avoid complications, such as severe infections and bleeding, and this study will be useful in establishing the actual incidence of pancytopenia, which will be helpful for developing strategies for its early detection and management in tertiary care hospitals of Swat.

METHODOLOGY

This cross sectional study was carried out in the General Medicine department of Saidu Group of Teaching Hospital, Swat, from 01-12-2023 to 01-06-2024. Ethical permission was obtained from hospital ethical board before starting the study (57/ERB/2023 dated 07-06-2023) and all procedures were conducted according to institutional research regulations. Sample size was calculated by using WHO sample size calculator by taking expected frequency of pancytopenia 70%,[11] confidence level 95%, and margin of error 7%. The calculated sample size was 165 patients. Patients of age 18–65 years, both Male and Female, diagnosed with megaloblastic anemia were included. Patients taking methotrexate or cytotoxic drugs, patients having acute leukemia and patients with hyperthyroidism were excluded from study. Megaloblastic anemia was considered when complete blood count showed hemoglobin level ≤9 mg/dl together with macrocytosis defined as mean corpuscular volume >100 fL. Demographic information including age, gender, marital status, economic status, educations status and employment status were recorded. A detailed medical history was taken, and a general physical examination was conducted. The skin was cleaned with an antiseptic solution, and a tourniquet was placed on the upper arm. A sterile needle was inserted into a vein to extract the blood needed for the test in labeled vials. After this, the tourniquet was removed and pressure was placed on the site where the blood was extracted and covered with a bandage. The blood samples were then sent to the hospital lab for the assessment of hemoglobin, mean corpuscular volume, red blood cell count, white blood cell count, and platelet count. After the blood test results were obtained, the patients were checked for pancytopenia. Pancytopenia was defined as the reduction of all peripheral blood cell types, which includes red blood cell count < 4 * 10^12/L, white blood cell count < 4 * 10^9/L, and platelet count < 150 * 10^9/L. The percentage of pancytopenia was determined for the entire population of patients with megaloblastic anemia. All collected data were entered and analyzed using IBM SPSS version 21. Mean ± standard deviation were calculated for quantitative variables including age, height, weight, BMI, HB level, red blood cell level, platelet count and white blood cell count. Frequencies and percentages were computed for categorical variables such as gender, pancytopenia, hypertension, diabetes mellitus, marital status, economic status, educations status and employment status. Pancytopenia was stratified with respect to age, gender, hypertension, diabetes mellitus, marital status, economic status, educations status and employment status. Post stratification chi-square test was applied and p value <0.05 was considered statistically significant.

RESULTS

The mean age of participants were 41.77±13.96 years and weight was found to be 74.70±11.07 kg while height were recorded as 1.63±0.08 m. Body mass index was calculated which show mean value of 28.12±3.34 kg/m^2. Laboratory parameters demonstrate that hemoglobin level was 7.28±1.03 g/dL, red blood cell count were 3.65±0.76 ×10^{12}/L, white blood cell count was 3.97±1.93 ×10^9/L, and platelet count were found 137.35±88.27 ×10^9/L. Gender distribution was showing 81 (49.1%) male and 84 (50.9%) female patients. Majority of participant were married 158 (95.8%) compare to single 7 (4.2%). Educational status reveal that 83 (50.3%) were educated while 82 (49.7%) were uneducated. Employment status showing 77 (46.7%) employed and 88 (53.3%) unemployed individuals. Economic distribution were consisted of 17 (10.3%) upper class, 74 (44.8%) middle class, and 74 (44.8%) lower class. Comorbidities analysis show diabetes mellitus present in 30 (18.2%) and absent in 135 (81.8%) patients, while hypertension was present in 40 (24.2%) and absent in 125 (75.8%) patients (Table-I).

Table I

Patient Demographics

Demographics	Mean ± SD
Age (years)	41.77±13.96
Weight (kg)	74.70±11.07
Height (m)	1.63±0.08
BMI (kg/m^2)	28.12±3.34
HB (g/dL)	7.28±1.03
Red Blood Cells (×10^{12}/L)	3.65±0.76
White Blood Cells (×10^9/L)	3.97±1.93
Platelet Count (×10^9/L)	137.35±88.27
Gender	
Male n (%)	81 (49.1%)
Female n (%)	84 (50.9%)
Marital Status	
Married n (%)	158 (95.8%)
Single n (%)	7 (4.2%)
Education Status	
Educated n (%)	83 (50.3%)
Uneducated n (%)	82 (49.7%)
Employment Status	
Employed n (%)	77 (46.7%)
Unemployed n (%)	88 (53.3%)
Economic Status	
Upper n (%)	17 (10.3%)
Middle n (%)	74 (44.8%)
Lower n (%)	74 (44.8%)
Diabetes Mellitus	
Yes n (%)	30 (18.2%)
No n (%)	135 (81.8%)
Hypertension	
Yes n (%)	40 (24.2%)
No n (%)	125 (75.8%)

Frequency analysis of pancytopenia among patient with megaloblastic anemia was demonstrated that 123 (74.50%) patient was having pancytopenia while 42 (25.50%) was not having pancytopenia from total 165 patient (Table-II).

Table II

Frequency of Pancytopenia Among Patients with Megaloblastic Anemia

Pancytopenia	Frequency	% age
Yes	123	74.50%
No	42	25.50%
Total	165	100%

Age stratification show that among patient ≤40 years, 58 (71.6%) have pancytopenia and 23 (28.4%) does not have, while in patient >40 years, 65 (77.4%) have pancytopenia and 19 (22.6%) not having it, with p-value of 0.395 indicating no significant association. Gender wise distribution showing in male 60 (74.1%) have and 21 (25.9%) not have pancytopenia, whereas female showing 63 (75.0%) have and 21 (25.0%) not have pancytopenia, p-value was 0.891 which show no significant difference. Marital status reveal married patient 118 (74.7%) having and 40 (25.3%) not having pancytopenia compare to single patient 5 (71.4%) having and 2 (28.6%) not having, p-value 1.000 demonstrate no significant association. Education status indicate educated patient 61 (73.5%) having and 22 (26.5%) not having pancytopenia while uneducated 62 (75.6%) having and 20 (24.4%) not having, p-value 0.755 showing no significance. Employment status data reveal employed patient 61 (79.2%) having and 16 (20.8%) not having pancytopenia whereas unemployed 62 (70.5%) having and 26 (29.5%) not having, p-value 0.197 which was not significant. Economic status stratification showing upper class 13 (76.5%) having and 4 (23.5%) not having, middle class 54 (73.0%) having and 20 (27.0%) not having, and lower class 56 (75.7%) having and 18 (24.3%) not having pancytopenia, p-value 0.966 indicate no significance. Diabetes mellitus analysis reveal patient with diabetes 22 (73.3%) having and 8 (26.7%) not having pancytopenia compare to without diabetes 101 (74.8%) having and 34 (25.2%) not having, p-value 0.866 showing no significant association. Hypertension data showing patient with hypertension 33 (82.5%) having and 7 (17.5%) not having pancytopenia while patient without hypertension 90 (72.0%) having and 35 (28.0%) not having, p-value 0.185 which was not showing statistical significance (Table-III).

Table III

Association of Pancytopenia with Demographic Factors

Demographic Factors		Pancytopenia		p-value
		Yes n(%)	No n(%)	
Age (years)	≤40	58 (71.6%)	23 (28.4%)	0.395**
	>40	65 (77.4%)	19 (22.6%)	
Gender	Male	60 (74.1%)	21 (25.9%)	0.891**
	Female	63 (75.0%)	21 (25.0%)	
Marital Status	Married	118 (74.7%)	40 (25.3%)	1.000*
	Single	5 (71.4%)	2 (28.6%)	
Education Status	Educated	61 (73.5%)	22 (26.5%)	0.755**
	Uneducated	62 (75.6%)	20 (24.4%)	
Employment Status	Employed	61 (79.2%)	16 (20.8%)	0.197**
	Unemployed	62 (70.5%)	26 (29.5%)	
Economic Status	Upper	13 (76.5%)	4 (23.5%)	0.966*
	Middle	54 (73.0%)	20 (27.0%)	
	Lower	56 (75.7%)	18 (24.3%)	
Diabetes Mellitus	Yes	22 (73.3%)	8 (26.7%)	0.866**
	No	101 (74.8%)	34 (25.2%)	
Hypertension	Yes	33 (82.5%)	7 (17.5%)	0.185**
	No	90 (72.0%)	35 (28.0%)	

***Fischer Exact Test **Chi-square Test**

DISCUSSION

The present study observed that out of 165 patients with megaloblastic anemia, 123 patients (74.50%) presented with pancytopenia. This suggests that megaloblastic anemia has a profound effect on all three cell lines present in the bone marrow. The pathophysiology of megaloblastic anemia is due to a deficiency of vitamin B12 and folate, which affects DNA synthesis and has a profound impact on rapidly proliferating cell populations present in the bone marrow. The ineffective hematopoiesis observed is due to a deficiency of vitamin B12 and folate, where abnormal cell populations are produced by the bone marrow, which eventually undergo apoptosis. There is a concurrent reduction observed in red cell, white cell, and platelet counts. In patients with megaloblastic anemia, characteristic megaloblastic cell changes are observed in all cell lines present in the bone marrow, which is responsible for the concurrent reduction observed. The lab parameters observed in patients with megaloblastic anemia were a mean hemoglobin level of 7.28 ± 1.03 g/dL, white cell counts of 3.97 ± 1.93 × 10^9/L, and a platelet count of 137.35 ± 88.27 × 10^9/L, which is concurrently decreased, thereby confirming pancytopenia.

Frequency of pancytopenia among patients with megaloblastic anemia and findings reveal that 123 (74.50%) patients have pancytopenia out of 165 patients. This frequency is considerably higher than reported by Khattak MB *et al.* [12] who found pancytopenia in 63 (70%) patients with megaloblastic anemia which show similar trend but slightly lower prevalence. The difference of 4.5% between two studies may be attributed to variations in diagnostic criteria or patient selection methods, however both studies was confirming that majority of megaloblastic anemia patients developing pancytopenia due to ineffective hematopoiesis affecting all cell lineages. Similarly, Niazi HT *et al.* [13] reported much lower frequency of pancytopenia at 13 (5%) among patients with vitamin B12 deficiency which is significantly different from current findings. This marked difference is explained by fact that Niazi *et al.* study population was vitamin B12 deficient patients not specifically megaloblastic anemia patients, and not all vitamin B12 deficiency cases progress to megaloblastic anemia or severe enough to cause pancytopenia. The pathophysiological progression from vitamin deficiency to megaloblastic changes and then to pancytopenia require time and severity of deficiency which vary among individuals.

Current study show mean age of 41.77±13.96 years which is comparable to Khan Y *et al.* [14] who reported mean age 42±15.84 years and Niazi HT *et al.* [13] with mean age 42±15.84 years, suggesting that megaloblastic anemia predominantly affecting middle-aged population. However, Razaq S *et al.* [15] found lower mean age of 35.85±6.92 years while Khattak MB *et al.* [12] reported mean age 28±15.84 years and Qamar U *et al.* [16] found mean age 26.04±11.97 years which indicate younger population being affected in these studies. This variation in age distribution could be due to differences in dietary habits, socioeconomic status, and regional variations in nutrition availability across different areas of Pakistan. Younger age in some studies possibly reflecting earlier onset of nutritional deficiencies in populations with poor socioeconomic conditions. The gender distribution in present study showing 81 (49.1%) male and 84 (50.9%) female which demonstrate almost equal distribution. This finding contrast with Khattak MB *et al.* [12] who found 54 (60%) males and 36 (40%) females, and Razaq S *et al.* [15]

showing 66.1% males and 33.9% females indicating male predominance. However, Niazi HT *et al.* [13] reported 96 (38%) males and 156 (62%) females showing female predominance. These gender variations across different studies suggesting that megaloblastic anemia not having consistent gender predilection and distribution may vary based on regional factors, dietary patterns, and gender-specific risk factors like pregnancy and menstruation in females increasing demands for vitamins.

In context of megaloblastic anemia as cause of pancytopenia, Memon S *et al.* [17] found megaloblastic anemia in 30 (13.04%) of pediatric pancytopenia cases while Qamar U *et al.* [16] reported higher frequency at 55 (36.6%) among adult pancytopenia patients. Shinwari N *et al.* [18] found 27 (27%) cases of megaloblastic anemia causing pancytopenia and Shafaat SS *et al.* [19] reported megaloblastic anemia as commonest etiology in 68 (25.85%) of pancytopenia cases. These studies was demonstrating that megaloblastic anemia is significant cause of pancytopenia across different age groups and settings. Khan S *et al.* [20] reported frequency of megaloblastic anemia in pediatric cytopenia as 23 (13.14%) which is lower compared to adult studies, this difference possibly reflecting that nutritional deficiencies taking longer time to manifest and cause severe hematological abnormalities in children compared to adults. The economic status distribution in current study showing 74 (44.8%) middle class and 74 (44.8%) lower class with only 17 (10.3%) upper class which is similar pattern to Khan S *et al.* [20] who found 94 (53.71%) from poor families, 40 (22.85%) middle class, and 41 (23.42%) rich families. This distribution supporting the notion that lower socioeconomic status is risk factor for megaloblastic anemia due to poor nutrition and limited access to vitamin-rich foods.

The limitations of the current study are recognized. The study is a single-center study, meaning that it was carried out in one hospital. This may limit the results' generalization to other populations or geographic locations. The study also has a small sample size of 165 patients, which may limit the power of the study. The nature of the study is also a limitation. The study is a cross-sectional study, and thus temporal relationships cannot be established.

CONCLUSION

It was concluded from this study that pancytopenia is highly prevalent in patients with megaloblastic anemia. The demographic factors like age, gender, education, employment, and economic status do not have any significant association with pancytopenia in patients with megaloblastic anemia.

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