Research Article

Etiological profile of pancytopenia in a tertiary care hospital

Nikhil Goli[1]*, Srikanth Koguru[1], Rustom S. Wadia[2], Sanjay Agarwal[3], Pradeep Patel[4], Pradeep Reddy[4], Karthik Nallam[4], Datta Kharwade[4]

[1]Senior Resident, Department of General Medicine, Malla Reddy Institute of Medical Sciences, Suraram, Hyderabad, Telangana, India
[2]Consultant Neurologist, [3]HOD of Internal Medicine, [4]Resident of Internal Medicine, Ruby Hall Clinic, Pune, Maharashtra, India

*Correspondence:
Dr. Nikhil Goli,
E-mail: drnikhilgoli@gmail.com

ABSTRACT

Background: As the severity of pancytopenia and the underlying pathology determines the management and prognosis of these patients, identifying the correct etiopathology in a given case is crucial and helps in implementing timely and appropriate treatment.The objective of the study was to determine the etiological profile of pancytopenia.
Methods: The present study included 44 patients with pancytopenia comprising of all ages and both sex. All cases were analyzed with respect to age, sex, clinical features at presentation, hemogram, peripheral smear, serum B12, plasma Folate, serum ferritin and bone marrow aspiration and etiological profile of pancytopenia was ascertained. Whether critical analysis of peripheral smear provides clue to the underlying pathology and how frequently bone marrow aspiration yields the diagnosis were also studied.
Results: Megaloblastic anemia was the most common cause of pancytopenia in this study (54.5%). Malignant and premalignant conditions (20.5%) were the second commonest cause of pancytopenia. Among 24 patients of megaloblastic anemia, 13 patients (54.16%) had macrocytic picture on peripheral smear suggestive of uncomplicated megaloblastic anemia. Of the 24 patients of megaloblastic anemia, 22 patients (91.66%) had low serum B12 levels. The distribution of cellularity, megakaryopoiesis, erythropoiesis, granulopoiesis on bone marrow aspiration differed significantly across various causes of pancytopenia (p-value <0.001).
Conclusions: Megaloblastic anemia should be considered first when managing a patient of pancytopenia and peripheral smear should be carefully examined for features of megaloblastic anemia.

Keywords: Etiological profile, Pancytopenia, Hospital, Clinical features

INTRODUCTION

Pancytopenia refers to a reduction in all the three formed elements of blood- erythrocytes, leukocytes and platelets. It is not a disease entity, but rather a triad of findings that may result from a number of disease processes.[1] The frequency with which each condition is associated with pancytopenia differs considerably. For example, it is essentially always present at some stage in the course of aplastic anemia, very common in subleukemic leukemia, relatively uncommon in lymphomas and rare in metastatic carcinoma involving bone marrow. Also the prognosis depends on both, the severity of pancytopenia and on the nature of underlying condition. Thus, the causes of pancytopenia vary from treatable causes like vitamin B_{12}. Folate deficiency and hypersplenism to virtually an untreatable cause like aplastic anemia (except by bone marrow transplantation which is out of reach of

most of our patients). Sometimes the clinical picture and examination of peripheral smear readily indicate the nature of causative disorder. The major diagnostic problems occur in those patients who have pancytopenia on peripheral smear but do not have any specific features either clinically or in the smear to provide a clue, either to the diagnosis or to an associated feature, such as splenomegaly or lymphadenopathy. Pancytopenia is generally equated to aplastic anemia and the prognosis considered grim even in textbooks of hematology.[1,2] Pancytopenia has been discussed along with aplastic anemia, though it was not considered its most common cause.[2] A review of available literature, revealed only few international studies 3 and few Indian studies on common etiological causes of pancytopenia.[1]

As the severity of pancytopenia and the underlying pathology determines the management and prognosis of these patients, identifying the correct etiopathology in a given case is crucial and helps in implementing timely and appropriate treatment. This study was therefore taken up to find out the common etiological causes of pancytopenia, to determine if a critical analysis of peripheral smear would provide vital clues and to conclude how frequently bone marrow aspiration yields the diagnosis.

METHODS

The study was conducted in Ruby Hall Clinic, a tertiary care hospital, Pune, India. Patients of all age groups, both males and females, consulting medical OPD and admitted in a medical unit with pancytopenia and meeting the inclusion criteria were studied. Prospective observational study was done. Sample size calculation was based on the results (effect sizes) from the previously published studies.[4-6]

Thus a sample of size 44 cases (minimum) satisfying the inclusion criteria would produce 80.0% statistical power (type II error=0.20) and 5% type I error probability (p=0.05) to be able to detect the statistical significant association between various risk factors and the prevalence of pancytopenia. The study was done from January 2013 to October 2014.

Inclusion criteria

All patients consulting medical OPD and admitted into the medical unit of Ruby Hall Clinic with the following indices on hemogram were included, based on criteria defined by De Gruchy.[1]

- Hemogram% <13.5 gm% (males).
  <11.5 gm% (females)
- WBC count <4000 cells/mm^3
- Platelets <1,50,000/mm^3

Patients with the above indices either in the initial hemogram or subsequent hemogram, during the workup prior to the diagnosis were included. Those patients, who were in remission due to previous anticancer drug therapy and on maintenance dose with follow up, if developed pancytopenia freshly, were also included in the study.

Exclusion criteria

All those patients who developed pancytopenia immediately after starting anti-malignant therapy in the hospital were excluded.

Methodology

The patients admitted in medical ward and OPD patients at Ruby Hall Clinic, from January 2013 to October 2014, meeting the inclusion criteria, were subjected to a detailed history- including age, sex, various symptoms, dietary habits, smoking status, treatment history, intake or exposure to potential toxic chemicals, agents or drugs, radiation exposure, symptoms such as bone pains, fever, night sweats, malaise, weight loss and pruritus.

Detailed Physical examination for pallor, jaundice, hepatosplenomegaly, lymphadenopathy, sternal tenderness, gum hypertrophy and for other signs was done.

The following investigations were done in all patients; hemogram, basic biochemical parameters, peripheral smear. For hemogram, 3 ml of K_2EDTA (ethylene diamine tetra-acetic acid) anticoagulated blood was collected and processed through coulter LH750 fully automated hematology analyzer; hemoglobin, total leukocyte count, differential counts, absolute neutrophil count, absolute lymphocyte count were obtained.

Peripheral smear was stained by leishman stain and was examined in detail. Bone marrow aspiration was done whenever indicated avoiding in cases where the cause of pancytopenia was obvious.

Bone marrow aspiration was done under aseptic precautions after obtaining written consent from the patient or guardian. Additional investigations were done as required and wherever feasible, depending upon the history and physical examination, like ESR (by westergren's method), urine and stool examination, liver and renal function tests, ELISA for HIV, hepatitis B and C viruses, chest and bone radiographs, abdominal ultrasonography, urine bence Jones proteins and serum electrophoresis.

The noted features peripheral smear

Morphology of red cells, presence of macroovalocytes, the degree of anisocytosis and poikilocytosis, presence of atypical cells, blast cells.

The evaluated features of bone marrow

Cellularity, myeloid:erythroid ratio, megaloblasts, dysplastic cells, proportion of cells and fat spaces, abnormal fibrosis and blast cells.

If the bone marrow resulted in a dry tap, it was repeated at a different site, after one week. A bone marrow was considered if the second attempt also resulted in a dry tap or an inadequate sample. All the patients were investigated systematically. Clinicopathological correlation was done in all cases before reaching a definite diagnosis.

Statistical methods

The entire data was entered and cleaned in MS Excel before it was statistically analyzed in SPSS. All the results are shown in tabular as well as graphical format to visualize the statistically significant difference more

clearly. The p-values less than 0.05 are considered to be statistically significant.

RESULTS

Megaloblastic anemia constituted major percentage of cases (54.5%). Malignant conditions included were all L1, all L2, acute promyelocytic leukemia, hodgkin's lymphoma. Premalignant condition was myelodysplastic syndrome.

Table 1: Distribution of causes of pancytopenia.

Causes of pancytopenia	Number	Percentage
Megaloblastic anemia	24	54.5
Aplastic anemia	5	11.4
Malignant and pre-malignant conditions	9	20.5
Others	6	13.6
Total	44	100.0

Table 2: The distribution of hepatomegaly and splenomegaly according to bone marrow impression.

Parameters		Bone marrow impression					P-value
		Megaloblastic anemia	Aplastic anemia	all	APL	Others	
Hepatomegaly	Yes	14 (53.8%)	02 (7.7%)	04 (15.4%)	03 (11.5%)	03 (11.5%)	0.128 (NS)
	No	10 (55.6%)	03 (16.7%)	00	00	05 (27.8%)	
Splenomegaly	Yes	15 (55.6%)	01 (11.1%)	03 (11.1%)	03 (11.1%)	05 (18.5%)	0.212 (NS)
	No	09 (52.9%)	04 (23.5%)	01 (5.9%)	00	03 (17.6%)	

Values are N (% of cases), p-values by chi-square test. P-value <0.05 is considered to be statistically significant. S: statistically significant, NS: Statistically non-significant.

The distribution of hepatomegaly did not differ significantly across various bone marrow impression groups (p-value >0.05). The distribution of splenomegaly did not differ significantly across various bone marrow impression groups (p-value >0.05).

This table describes the association of bleeding risk with level of platelet count. The association was found to be statistically significant (p=0.010).

Values are n (% of cases). P-values by chi-square test. P-value <0.05 is considered to be statistically significant. S: statistically significant, NS: statistically non-significant.

Table 3: Distribution of thrombocytopenia according to bleeding status.

Thrombocytopenia		Bleeding		
		Yes	No	Total
<25000	Grade 4	4 (25.0)	1 (3.6)	5 (11.4)
25000-50000	Grade 3	7 (43.8)	6 (21.4)	13 (29.5)
50000-75000	Grade 2	3 (18.8)	4 (14.3)	7 (15.9)
75000-150000	Grade 1	2 (12.5)	17 (12.5)	19 (43.2)
Total		16 (100.0)	28 (100.0)	44 (100.0)

Values are n (% of cases). P-value = 0.010 (Significant)

Table 4: The distribution of hypersegmented neutrophils and the cause of pancytopenia.

Hypersegmented neutrophils	Cause of pancytopenia					P value
	Megaloblastic anemia	Aplastic anemia	Malignant and pre malignanat	Others	Total	
Yes	20 (100%)	00	00	00	20 (100%)	0.001 (S)
No	04 (16.7%)	05 (20.8%)	09 (37.5%)	06 (25%)	24 (100%)	

The distribution of cause of pancytopenia differs significantly according to presence or absence of hyper segmented neutrophils (p-value <0.001).

Values are n (% of cases). P-values by chi-square test. P-value <0.05 is considered to be statistically significant. S: Statistically Significant, NS: Statistically Non-Significant.

The distribution of cellularity differs significantly across various bone marrow impression groups (p-value<0.001). The distribution of megakaryopoiesis differs significantly across various bone marrow impression groups (p-value <0.001). The distribution of Erythropoiesis differs significantly across various bone marrow impression groups (p-value <0.001). The distribution of granulopoiesis differs significantly across various bone marrow impression groups (p-value <0.001).

Table 5: The association between peripheral smear morphology and the cause of pancytopenia.

Peripheral smear	Causes of pancytopenia					P value
	Megaloblastic anemia	Aplastic anemia	Malignant and pre-malignant	Others	Total	
Normocytic	03 (15%)	05 (25%)	06 (30%)	06 (30%)	20 (100%)	0.001 (S)
Macrocytic	13 (100%)	00	00	00	13 (100%)	
Microcytic	00	00	03 (100%)	00	03 (100%)	
Dimorphic	08 (100%)	00	00	00	08 (100%)	

DISCUSSION

At 54.5%, megaloblastic anemia as the most common cause of pancytopenia in this study was in contrast to the literature. The most common cause of pancytopenia reported in various studies throughout the world has been aplastic anemia.[3,5,6] However other studies conducted in India reported megaloblastic anemia as the most common cause of pancytopenia.[7,8] Even few Indian studies reported hypoplastic/aplastic anemia as the commonest cause of pancytopenia.[4,9]

The radically different dietary habits, predominantly low socioeconomic group patients in this study and a proportion of strict vegetarians explain the discrepancy with worldwide studies. This also explains why hematological malignancies (including lymphoid malignancies) were their commonest cause but second most common cause in this study (20.5%).[3]

At 20.5%, the incidence of malignant and premalignant conditions as a cause of pancytopenia in this study was lower than that described in the literature. Study by Devi et al reported 32% cases; Tariq M et al reported 26% cases.[4,6] This could be due to geographic variation, variation in the genetics of the population studied, and environmental factors. The diagnosis of leukemia was established by bone marrow aspiration. In this study malignant and premalignant conditions included were; 2 cases of all L1 type, 2 cases of all L2 type, 3 cases of acute promyelocytic leukemia, 1 case of hodgkin's lymphoma, 1 case of myelodysplastic syndrome.

At 2.27%, the incidence of MDS was lower than that described in the literature. MDS as a cause of pancytopenia, reported in other studies varied from 0-18%.[4,6,10] This is due to variation in genetics, geography and environmental factors.

At 11.4%, aplastic anemia as a cause of pancytopenia in this study was lower than that described in the literature. Other studies from India reported aplastic anemia as a cause of pancytopenia between 20-30%.[7,8] It was lower even when compared to international studies.[5,6] This could be due to the differences in the population studied, genetics, environmental factors and number of cases. However this percentage of aplastic anemia in this study was comparable to few studies.[3,10] The diagnosis of aplastic anemia was established on bone marrow aspiration.

At 4.54%, hypersplenism as a cause of pancytopenia was comparable to that described in the literature. Various studies have reported hypersplenism between 3 to 68%.[7,8,10] This wide range of reports were due to enormous geographic variation.

Myelofibrosis constituted 31% in the study by Imbert et al but this study could not record any such case.[3] 80% of the patients in the present study were below 40 years during which myelofibrosis is uncommon, its peak incidence being between 40-70 years of age.[5]

The variation in causes of pancytopenia observed among various studies could be due to differences in methodology and the stringency of diagnostic criteria, period of observation, epidemiology of infections, environmental factors, socioeconomic discrepancies, geography, age distribution, dietary patterns, and cultural differences.

At 83.33%, the presence of hyper segmented neutrophils in megaloblastic anemia on peripheral smear is consistent with that described in the literature. Other Indian studies reported as follows; Tilak V et al reported 84.9%.[12] But recent study by Gayatri et al reported 51.35%.[13] Khunger JM et al demonstrated no neutrophils in megaloblastic

anemia.[8] This variation could be due to subjective variation in analysis of peripheral smear.

Bone marrow aspiration was sufficient to make a diagnosis in all cases and bone marrow biopsy was not required. In the study by Imbert et al bone marrow aspiration yielded diagnostic material in 55% cases, trephine biopsy in 30% of cases.[3] The rest 15% needed bone marrow biopsy. A high incidence of myelofibrosis (31%) in their study, where marrow fibrosis commonly results in a dry tap, may be responsible for higher percentage of trephine biopsy and bone marrow biopsy. In contrast in this study, 54.5% of cases were megaloblastic by marrow which is usually hypercellular readily yielding diagnostic material on aspiration. The diagnosis of leukemia and aplastic anemia was proved on bone marrow aspiration.

Nutritional megaloblastic anemia is a commoner cause of anemia, than generally considered and its typical features on peripheral smear are masked by concurrent iron deficiency state. This is suggested by the presence of microcytes with macrocytes (dimorphic); normocytes with macrocytes and hypochromic macrocytes. This is in concurrence with various studies.[15,16]

CONCLUSIONS

Megaloblastic anemia was the most common cause of pancytopenia in this study, critical analysis of peripheral smear provided clue to megaloblastic anemia, bone marrow aspiration was sufficient to diagnose the cause of pancytopenia. Megaloblastic anemia should be considered first when managing a patient of pancytopenia and peripheral smear should be carefully examined for features of megaloblastic anemia.

REFERENCES

1. Jain A, Naniwadekar M. An etiological reappraisal of pancytopenia- largest series reported to date from a single tertiary care teaching hospital. British Med Hemato. 2013;13(1):10.
2. De GGC. Pancytopenia, aplastic anemia. In Edited by Firkin F, Chesterman C, Penington D, Rush B. Berlin . De Gruchy's clinical hematology in medical practice. 5th edition. Germany. Blackwell Science;1989:119-36.
3. Imbert M, Scoazec JY, Mary JY. Adult patients presenting with pancytopenia: a reappraisal of underlying pathology and diagnostic procedures in 213 cases. Hematol Pathol. 1989;3(4);159-67.
4. Devi PM, Laishram RS, Sharma PS, Singh AM, Singh MK, Singh YM, et al. Clinic haematological profile of pancytopenia in Manipur, India. J Kuwait Med Assoc. 2008;40:221-4.
5. Jalbani A, Ansari IA, Shah AH. Pancytopenia: study of 40 patients at CMC hospital, Larkana. Professional Med J. 2010;17(1):105-10.
6. Tariq M, Khan NU, Basri R, Said A. Aetiology of pancytopenia. Prof Med J. 2010;17(2):252-6.
7. Kumar R, Kalra SP, Kumar H. Pancytopenia- a six year study. J Assoc Physicians India. 2001;49:1078-81.
8. Khunger JM, Arulselvi S, Sharma U. Pancytopenia-a clinic hematological study of 200 cases. Indian J Pathol Microbiol. 2002;45(3):375-9.
9. Tilak V, Jain R. Pancytopenia- a clinic hematological analysis of 77 cases. Indian J Pathol Microbiol. 1999;42(4):399-404.
10. Ishtiaq O, Baqai HZ, Anwer F, Hussain N. Patterns of pancytopenia patients in a general medical ward and a proposed diagnostic approach. J Ayub Med Coll Abbottabad. 2004;16(1):8-13.
11. Frank F. Megaloblastic anemia, aplastic anemia and other cytopenias. In Edited by Firkin F, Chesterman C, Penington D, Rush B. De Gruchy's clinical hematology in medical practice. 5th edition. Berlin, Germany: Blackwell Science;1989:158-72.
12. Tilak V, Jain R. Pancytopenia-a clinic hematological analysis of 77 cases. Indian J Pathol Microbiol. 1999;42(4):399-404.
13. Gayatri BN, Rao KS. Pancytopenia: a clinico hematological study. J Lab Physicians. 2011;3(1):15-20.
14. Spivak JL. Masked megaloblastic anemia. Arch Intern Med. 1982;142(12):2111-4.
15. Saraya AK, Kumar R, Kailash S, Sehgal AK. Vit B_{12} and folic acid deficiency in beta heterozygous thalassemia. Indian J Med Research. 1984;79:783-8.