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Clinicopathological Spectrum of Pancytopenia in Adults Without Known Malignancy at a Tertiary Care Hospital in Central India

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Abstract

Background

Pancytopenia has diverse nutritional, infectious, systemic, and marrow-related causes. Its etiological distribution depends on the population evaluated and the diagnostic setting. The primary objective was to describe diagnostic categories among adults with pancytopenia without known malignancy. Secondary objectives were to characterize clinical, hematological, and marrow findings and explore differences in selected characteristics across diagnostic groups.

Materials and methods

This prospective observational study included 162 consecutively enrolled adults over 18 months. Pancytopenia was defined as hemoglobin <13 g/dL in men or <12 g/dL in women, leukocyte count <4,000/µL, and platelet count <150,000/µL. Patients with known malignancy or previous or current chemotherapy or radiotherapy were excluded. Clinical assessment, complete blood counts, peripheral smears, and clinically indicated investigations were evaluated. Bone marrow aspiration was performed when indicated. Categorical associations were assessed using chi-square or Fisher-Freeman-Halton exact tests.

Results

The mean age was 39.71 ± 13.40 years; 93 patients (57.4%) were men. Generalized weakness occurred in 147 patients (90.7%) and pallor in 160 (98.8%). Mean hemoglobin was 6.87 ± 1.62 g/dL. Nutritional deficiency accounted for 77 cases (47.5%), hypersplenism for 51 (31.5%), and infectious or immune-mediated systemic disorders for 34 (21.0%). Megaloblastic anemia was the most frequent individual diagnosis (62; 38.3%). Bone marrow aspiration was performed in 147 patients (90.7%); 101 aspirates (68.7%) were hypercellular, and 58 (39.5%) showed megaloblastic erythropoiesis. Age category was associated with etiological group ($p = 0.017$), whereas sex was not ($p = 0.592$).

Conclusions

Nutritional deficiency, particularly megaloblastic anemia, and hypersplenism were the most frequent diagnostic categories in this selected adult cohort. Hypercellularity was the predominant marrow cellularity pattern, and megaloblastic erythropoiesis was the most frequent morphological finding among aspirated patients.

Categories: Hematology, Pathology
Keywords: bone marrow aspiration, hypersplenism, megaloblastic anaemia, nutritional deficiency, pancytopenia, peripheral blood smear

Introduction

Pancytopenia is the simultaneous reduction of red cells, leukocytes, and platelets in peripheral blood. It may result from ineffective hematopoiesis, marrow failure or infiltration, peripheral destruction, or splenic sequestration. Its evaluation requires integration of clinical findings, blood counts, peripheral smear morphology, and targeted investigations [1].

The etiological spectrum varies across clinical settings. Nutritional deficiency, infections, and hypersplenism account for substantial proportions of cases in Indian hospital series, whereas specialist hematology cohorts also include many patients with leukemia and marrow failure [2,3]. Other Indian studies similarly demonstrate a heterogeneous distribution of causes [4,5]. Differences in patient selection, referral patterns, and diagnostic thresholds influence these distributions and make local clinical data useful when interpreting pancytopenia at presentation.

The primary objective was to describe the distribution of diagnostic categories among eligible adults with pancytopenia without known malignancy. Secondary objectives were to describe clinical, hematological, and marrow findings, with exploratory comparisons of age, sex, fever, bleeding manifestations, organomegaly, and marrow aspiration status across diagnostic groups. This study adds contemporary regional estimates for a defined adult population, integrating clinical presentation and laboratory findings with marrow morphology in most participants, and complements existing evidence from Central India [4].

Materials And Methods

Study design and participants

This prospective observational study was conducted over 18 months, from March 15, 2024, to September 14, 2025, in the Department of Pathology, Amaltas Institute of Medical Sciences and Research Centre, Dewas, Madhya Pradesh, India. The study included 162 consecutively enrolled adults aged ⩾18 years with pancytopenia on complete blood count, confirmed by peripheral smear examination, who provided written informed consent.

Pancytopenia was operationally defined as the simultaneous presence of hemoglobin <13 g/dL in men or <12 g/dL in women, total leukocyte count <4,000/µL, and platelet count <150,000/µL. Patients with known malignancy, previous or current chemotherapy or radiotherapy, or exposure to cytotoxic drugs used for cancer chemotherapy were excluded. Patients with isolated cytopenia or bicytopenia and those who declined consent were not enrolled.

Sample size was estimated using the single-proportion formula, $n = \frac{Z^2 p(1-p)}{d^2}$ [6], with a 95% confidence level (Z = 1.96), an assumed pancytopenia prevalence of 12% (p = 0.12), and an absolute precision of five percentage points (d = 0.05). The calculation yielded 162 consecutively enrolled patients.

Clinical and laboratory evaluation

A structured proforma recorded demographic characteristics, presenting symptoms, relevant medical and drug histories, previous transfusions, and examination findings. Clinical evaluation included pallor, bleeding manifestations, lymphadenopathy, hepatomegaly, and splenomegaly. Symptoms and physical findings were recorded independently and could coexist.

Venous blood samples were collected under aseptic precautions for hematological and clinically indicated biochemical, serological, and coagulation investigations. Complete blood counts and red cell indices were measured using an automated five-part hematology analyzer (ELite 580; Transasia Bio-Medicals Ltd., Mumbai, India). Peripheral blood smears were stained with Leishman stain and examined for red cell morphology, leukocyte abnormalities, platelet adequacy, abnormal cells, and parasites.

Additional investigations were selected according to the clinical presentation and initial laboratory findings. These included serum vitamin B12 and folate, iron studies, liver and renal function tests, lactate dehydrogenase, uric acid, and coagulation tests. Infection screening included malaria, dengue, human immunodeficiency virus (HIV), hepatitis B, and hepatitis C testing. Antinuclear antibodies, anti-double-stranded DNA antibodies, and the direct antiglobulin test were used when autoimmune disease was suspected. Urine specimens were collected for routine examination when clinically indicated. Imaging investigations included abdominal ultrasonography, chest radiography, and bone radiography, selected according to clinical findings.

Bone marrow examination and etiological classification

Bone marrow aspiration was undertaken for unexplained pancytopenia, inconclusive peripheral smear findings, or suspected marrow failure, neoplasia, or infiltration. Aspiration was performed under aseptic precautions using a Salah needle after written informed consent. Aspirate smears were stained with Leishman stain for morphological evaluation and Perls Prussian blue stain for iron stores. Clinical findings, blood counts, smear morphology, ancillary investigations, and marrow findings, when available, were considered together in assigning the final diagnosis.

Final diagnoses were grouped as nutritional deficiency, hypersplenism, or infectious or immune-mediated systemic disorders. Classification integrated the clinical assessment, peripheral blood findings, ancillary investigations, and marrow morphology when performed. Nutritional deficiency comprised megaloblastic anemia and other nutritional deficiencies. Megaloblastic anemia was supported by vitamin B12 and/or folate deficiency together with compatible blood or marrow morphology, including macro-ovalocytes, hypersegmented neutrophils, and megaloblastic erythropoiesis. Other nutritional deficiency was characterized by microcytic hypochromic morphology and biochemical evidence of iron depletion, with marrow iron findings providing additional support when examined. Hypersplenism was assigned in patients with pancytopenia and splenomegaly in the clinical context of portal hypertension or chronic liver disease, integrating ultrasonographic findings, liver function tests, and marrow findings when performed. The

systemic group included malaria, dengue, HIV-associated disease, miliary tuberculosis, sepsis, and systemic lupus erythematosus (SLE). These diagnoses integrated the clinical presentation with the relevant infection-screening results, autoimmune investigations, imaging, and marrow findings obtained during evaluation.

When a relevant ancillary investigation or marrow examination was not performed, the assigned diagnosis reflected the clinical assessment and the investigations completed during evaluation. The reported distribution therefore represents integrated clinical diagnostic categories, with the degree of diagnostic confirmation dependent on the investigations performed. Marrow findings were summarized separately among patients who underwent aspiration.

Statistical analysis

Data were entered into Microsoft Excel (Microsoft Corporation, Redmond, WA) and analyzed using IBM SPSS Statistics, version 25.0 (IBM Corp., Armonk, NY). Continuous variables were summarized as mean ± standard deviation and range, where available; categorical variables were presented as counts and percentages. Percentages were calculated using the full cohort or the explicitly stated subgroup denominator. Wilson 95% confidence intervals were calculated for the principal proportions. For exploratory comparisons, age was categorized as 18-40 years and >40 years. Pearson's chi-square test was used for age category and sex, and the Fisher-Freeman-Halton exact test was used for marrow aspiration status. A p-value <0.05 was considered statistically significant for these exploratory comparisons. Fever, bleeding manifestations, hepatomegaly, splenomegaly, and hepatosplenomegaly were summarized descriptively across diagnostic groups because these findings also contributed to diagnostic classification, introducing potential incorporation bias.

Ethical approval

The Institutional Ethics Committee approved the study on February 15, 2024. Written informed consent was obtained from all participants before enrollment and separately before bone marrow aspiration. Patient confidentiality was maintained.

Results

Participant characteristics and clinical presentation

The cohort comprised 162 adults, including 93 men (57.4%) and 69 women (42.6%). Mean age was 39.71 ± 13.40 years. The age distribution was 18-30 years in 43 patients (26.5%), 31-40 years in 47 (29.0%), 41-50 years in 38 (23.5%), 51-60 years in 22 (13.6%), and >60 years in 12 (7.4%).

Generalized weakness was the most common symptom (147; 90.7%), followed by palpitations (84; 51.9%), dyspnea (79; 48.8%), loss of appetite (63; 38.9%), and headache (54; 33.3%). Fever occurred in 31 patients (19.1%) and bleeding manifestations in seven (4.3%). Pallor was present in 160 patients (98.8%). Splenomegaly was recorded in 73 (45.1%), hepatomegaly in 59 (36.4%), and concurrent hepatosplenomegaly in 51 (31.5%).

Hematological and peripheral smear findings

The hematological profile is summarized in Table *1*. Hemoglobin was <7 g/dL in 87 patients (53.7%), including 20 (12.3%) with values <5 g/dL. Platelet counts were <50,000/µL in 49 patients (30.2%), including three (1.9%) with counts <20,000/µL.

Parameter	Mean ± SD	Observed range (minimum-maximum)	Adult reference interval
Hemoglobin (g/dL)	6.87 ± 1.62	3.70-10.70	Men: 13.00-17.00; women: 12.00-15.00
Total leukocyte count (×10³/μL)	2.98 ± 0.52	1.68-3.94	4.00-10.00
Platelet count (×10³/μL)	63.54 ± 24.14	12.69-126.30	150.00-410.00
Packed cell volume (%)	20.45 ± 4.97	11.20-32.70	Men: 40.00-50.00; women: 36.00-46.00
Mean corpuscular volume (fL)	95.26 ± 16.00	55.50-123.90	83.00-101.00
Mean corpuscular hemoglobin (pg)	30.75 ± 4.49	17.40-38.70	27.00-32.00
Mean corpuscular hemoglobin concentration (g/dL)	32.70 ± 1.28	27.80-34.60	31.50-34.50
Red cell distribution width (%)	18.58 ± 3.73	13.00-28.00	11.60-14.00

TABLE 1: Hematological parameters at presentation (n = 162)

Observed ranges represent the minimum and maximum values recorded in the study cohort. Adult reference intervals are external comparators provided for clinical context.

SD: standard deviation

Peripheral smears showed a normocytic normochromic pattern in 79 patients (48.8%), a macrocytic or megaloblastic pattern in 68 (42.0%), and a microcytic hypochromic pattern in 15 (9.3%). Hypersegmented neutrophils were recorded in 62 patients (38.3%). Isolated vitamin B12 deficiency was documented in 42 patients (25.9%), isolated folate deficiency in six (3.7%), and combined deficiency in 14 (8.6%).

Diagnostic distribution

Nutritional deficiency was the most frequent broad etiological group (47.5%; 95% CI: 40.0-55.2%), followed by hypersplenism (31.5%; 95% CI: 24.8-39.0%) and infectious or immune-mediated systemic disorders (21.0%; 95% CI: 15.4-27.9%). Megaloblastic anemia was the most frequent individual diagnosis (38.3%; 95% CI: 31.1-45.9%). The complete distribution of final diagnoses is shown in Figure *1*. Among the 51 patients classified as having hypersplenism, ultrasonographic features of portal hypertension were present in 42 (82.4%), and abnormal liver function tests were present in 39 (76.5%). These findings were not mutually exclusive.

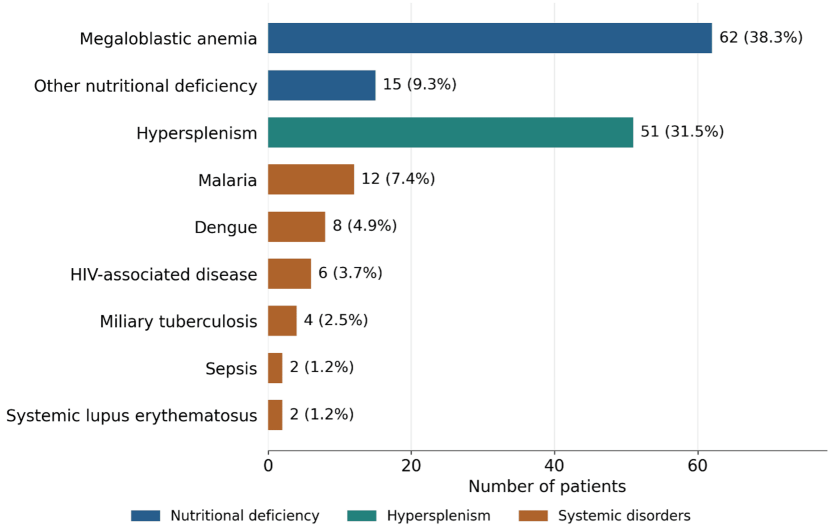


FIGURE 1: Recorded diagnostic categories among adults with pancytopenia

Values are n (%) of 162 patients. Each patient is represented once. Other nutritional deficiency denotes deficiency not attributed to vitamin B12 or folate.

HIV: human immunodeficiency virus

Bone marrow findings

Bone marrow aspiration was performed in 147 patients (90.7%). Among these aspirates, 101 (68.7%) were hypercellular, 43 (29.3%) were normocellular, and three (2.0%) were hypocellular. Iron stores were adequate or increased in 119 (81.0%), reduced in 18 (12.2%), and absent in 10 (6.8%). Morphological findings are summarized in Table 2. Of the three hypocellular aspirates, two occurred in patients with other nutritional deficiencies and one in a patient with dengue. Among the 15 patients with other nutritional deficiency, 14 underwent aspiration; iron stores were absent in 10 and reduced in four. Reduced iron stores were also present in 14 patients assigned to other diagnostic categories.

Morphological finding	n (%)
Megaloblastic erythropoiesis	58 (39.5)
Erythroid hyperplasia or hypersplenism-related reactive marrow	48 (32.7)
Micronormoblastic erythropoiesis or nutritional marrow	14 (9.5)
Infective marrow with hemozoin pigment	9 (6.1)
Reactive plasmacytosis or HIV-associated marrow changes	8 (5.4)
Granulomatous marrow	4 (2.7)
Megakaryocytic prominence or changes	3 (2.0)
Reactive myeloid changes or hemophagocytosis	2 (1.4)
Transient viral marrow suppression	1 (0.7)

TABLE 2: Bone marrow morphology in aspirated patients (n = 147)

Percentages are based on 147 aspirates. Combined morphological categories do not denote a single underlying disease.

HIV: human immunodeficiency virus

Characteristics across diagnostic groups

Age composition differed across diagnostic groups (p = 0.017; Table 3). Fever and bleeding were recorded only in the systemic disorder group, whereas organomegaly was most frequent in the hypersplenism group. Differences in sex distribution and marrow aspiration status were not statistically significant.

Characteristic	Nutritional deficiency (n = 77)	Hypersplenism (n = 51)	Systemic disorders (n = 34)	Test statistic	p-value
Age ≤40 years	48 (62.3)	20 (39.2)	22 (64.7)	8.102	0.017
Male sex	41 (53.2)	31 (60.8)	21 (61.8)	1.047	0.592
Fever	0 (0.0)	0 (0.0)	31 (91.2)	-	-
Bleeding manifestations	0 (0.0)	0 (0.0)	7 (20.6)	-	-
Hepatomegaly	5 (6.5)	40 (78.4)	14 (41.2)	-	-
Splenomegaly	7 (9.1)	51 (100.0)	15 (44.1)	-	-
Hepatosplenomegaly	2 (2.6)	40 (78.4)	9 (26.5)	82.294	<0.001
Marrow aspiration performed	72 (93.5)	48 (94.1)	27 (79.4)	N.A.	0.061

TABLE 3: Clinical characteristics and marrow aspiration by diagnostic group

Values are n (%), with percentages calculated within each diagnostic group. - indicates that the clinical finding is presented descriptively because it contributed to diagnostic classification. These distributions should not be interpreted as independent diagnostic associations.

N.A.: test statistic not reported for the Fisher-Freeman-Halton exact test

Discussion

Nutritional deficiency and hypersplenism were the most frequent diagnostic categories in this selected adult cohort. The distribution should be interpreted within the eligibility criteria and diagnostic setting. The absence of marrow failure, neoplastic, and other less common diagnoses from the reported cohort does not establish their absence in the source population.

The proportion of megaloblastic anemia (38.3%) was similar to the 41.4% reported by Negotia et al. [4] and the 37% reported by Jain et al. [7]. Khunger et al. reported a higher proportion of 72% in an older series [8]. By contrast, acute leukemia was the leading diagnosis in the specialist hematology cohort of Patel and Prajapati, accounting for 17.9% of cases, while megaloblastic anemia accounted for 15.4% [3]. Differences in age eligibility, referral setting, diagnostic thresholds, and investigation protocols limit direct comparison of these proportions. Weakness and pallor predominated, consistent with the low mean hemoglobin concentration and frequent values below 7 g/dL. Similar presentations have been reported in other hospital cohorts [5,7].

Macrocytic or megaloblastic blood-film appearances and hypersegmented neutrophils were consistent with the prominence of megaloblastic anemia. Isolated vitamin B12 deficiency was more frequent than isolated folate or combined deficiency. Kaur et al. also documented all three deficiency patterns among patients with megaloblastic anemia [9]. Nevertheless, almost half of the present cohort had a normocytic normochromic smear, illustrating the morphological diversity of pancytopenia. Mean corpuscular volume should therefore be interpreted alongside blood-film morphology and targeted biochemical testing [1].

Hypersplenism accounted for 31.5% of cases, compared with 29.2% in the series of Jain and Naniwadekar, which included children and drug-related cytopenia [2]. Hypersplenism represents a mechanism of cytopenia rather than a single underlying disease [1]. In the current study, its clinical context included portal hypertension and liver dysfunction; the corresponding ultrasonographic and biochemical findings are reported separately. In a separate population with cirrhosis and otherwise unexplained cytopenia, Koschade et al. reported normal, nonspecific, or reactive marrow findings in 117 of 118 patients [10]. Their findings highlight the importance of the systemic disease context when interpreting marrow morphology.

Patients aged >40 years constituted 60.8% of the hypersplenism group, compared with 37.7% of the nutritional deficiency group and 35.3% of the systemic disorder group. This exploratory association reflects differences in group composition and does not establish an independent effect of age.

Bone marrow aspiration was performed in most participants. Hypercellularity predominated, and megaloblastic erythropoiesis was the most frequent morphological impression. Ullah et al. similarly identified megaloblastic anemia as the leading diagnosis in a marrow-based pancytopenia cohort [11]. Although aspiration supplied morphological information in the present study, its incremental diagnostic yield was not measured. These findings therefore describe marrow abnormalities among patients selected for aspiration and do not define criteria for safely omitting the procedure.

The three hypocellular aspirates occurred within the nutritional deficiency and dengue groups. Hypocellularity was a morphological finding and did not itself establish a diagnosis of aplastic anemia [1]. Similarly, the 28 aspirates with reduced or absent iron stores represented marrow iron status across diagnostic groups, whereas the 15 other nutritional deficiency cases represented a primary diagnostic category. These totals therefore describe different aspects of the cohort.

Comparisons with other hospital cohorts further illustrate the variation in etiological distribution. In a Pakistani series of 117 patients, Saleem et al. reported aplastic anemia in 17.1% and megaloblastic anemia in 12.8% [12]. In a Turkish series of 112 patients, Çordan and Tukek identified vitamin B12 deficiency and hypersplenism in 20.5% each [13]. Both series included adolescents and a broader range of diagnoses, limiting direct comparison with the present selected adult cohort.

Nutritional causes also require consideration beyond megaloblastic anemia. Meena et al. described six adults with severe iron deficiency and pancytopenia whose blood counts improved following iron replacement [14]. In a mixed-age Indian cohort of 154 patients, Das et al. found differences in immature reticulocyte fraction and mean reticulocyte volume between megaloblastic and dual-deficiency anemia [15]. These findings support integrating morphological, biochemical, and hematological information when evaluating nutritional causes. Treatment response and reticulocyte indices were not evaluated in the present analysis.

Limitations

The single-center setting and eligibility restrictions limit generalizability. Clinically selected ancillary investigations and bone marrow aspiration may have introduced selection and diagnostic ascertainment bias. Variation in diagnostic confirmation across patients may also have resulted in etiological misclassification. The analysis did not include trephine biopsy correlation, limiting assessment of marrow architecture and some hypocellular or infiltrative disorders. Clinical findings contributed to diagnostic classification, and the diagnostic groups encompassed heterogeneous disorders. Exploratory comparisons were not adjusted for confounding or multiple testing. Diagnostic accuracy and treatment response were not evaluated. The prevalence-based sample-size calculation did not determine precision for individual diagnostic proportions.

Conclusions

Nutritional deficiency, particularly megaloblastic anemia, and hypersplenism were the most frequent diagnostic categories in this selected adult cohort. Hypercellularity was the predominant marrow cellularity pattern, and megaloblastic erythropoiesis was the most frequent morphological finding among aspirated patients.

Additional Information

Author Contributions

All authors have reviewed the final version to be published and agreed to be accountable for all aspects of the work.

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Disclosures

Human subjects: Informed consent for treatment and open access publication was obtained or waived by all participants in this study. Amaltas Institute of Medical Science Institutional Ethical Committee issued approval N/A. **Animal subjects:** All authors have confirmed that this study did not involve animal subjects or tissue. **Conflicts of interest:** In compliance with the ICMJE uniform disclosure form, all authors declare the following: **Payment/services info:** All authors have declared that no financial support was received from any organization for the submitted work. **Financial relationships:** All authors have declared that they have no financial relationships at present or within the previous three years with any organizations that might have an interest in the submitted work. **Other relationships:** All authors have declared that there are no other relationships or activities that could appear to have influenced the submitted work.

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