

**Bone Marrow Studies in Geriatric Anaemic Patients**

Ayushi Bansal<sup>1</sup>, Narayanan Subramanian Mani<sup>2</sup>, Amit Nisal<sup>3</sup>, Anjali Kelkar<sup>4</sup>

<sup>1</sup>Resident, Department of Pathology, <sup>2</sup>Professor, Department of Pathology

<sup>3</sup>Assistant Professor, Department of Pathology, <sup>4</sup>Associate Professor, Department of Pathology

<sup>1,2,3,4</sup>Bharati Vidyapeeth Deemed University Medical College and Hospital, Pune-411043, India.

**Correspondence Author:** Amit Nisal, Assistant Professor, Department of Pathology, Bharati Vidyapeeth Deemed University Medical College and Hospital, Pune-411043, India.

**Type of Publication:** Original Research Paper

**Conflicts of Interest:** Nil

**Abstract**

**Purpose:** To evaluate Bone marrow studies in anaemic geriatric patients.

**Methods:** Bone marrow in 75 cases of anaemia in elderly >65yrs as defined by the WHO were studied in 24 months. Clinical History, Biochemical, Radiological investigations, Haematological parameters using LH750 and Peripheral blood smears were recorded and correlated. These anaemias were classified etiologically and morphologically in correlation with their bone marrow findings

**Results:** Bone marrow studies in 75 cases of anaemic elderly were studied. There were 14 cases of Multiple Myeloma, 9 cases of haemophagocytosis, 8 cases of reactive marrow, 7 cases of megaloblastic anaemia and 6 cases of aplastic anaemia, 5 cases of Immune thrombocytopenia purpura (ITP), 5 cases of Lymphoproliferative disorder (LPD), 3 cases each of Acute myeloid leukemia (AML), Iron deficiency anaemia (IDA), hypocellular marrow, Myelodysplastic syndromes (MDS) and infectious etiology respectively. There were 2 cases of hypersplenism and 1 case each of myelofibrosis, metastasis from prostatic adenocarcinoma, eosinophilia and Chronic myeloid leukemia (CML) respectively. These findings were correlated with the

etiologically and morphologically classification of these anaemias.

**Conclusions:** The two most common entities on bone marrow examination in our study were Multiple Myeloma followed by haemophagocytosis. These were more often anaemias due to chronic diseases (etiologically classification) and normocytic normochromic anaemias (morphologically classification). Bone marrow was found to be an essential requirement of the workup of elderly patients with anaemias in view of the variety of findings and unexplained cytopenias seen in geriatric age group.

**Introduction**

Anaemia is defined according to the World Health Organization (WHO) as Hemoglobin (Hb) <13g/dl in males and Hb<12g/dl in females [1]. The WHO has also defined range for anaemia to be either mild (11-12.9g/dL), moderate (8-10.9g/dL) or severe (<8g/dL) [2]. According to the WHO, most developed countries of the world have accepted chronological age of 65 as definition of elderly [1]. Guralnik et al. categorized elderly above 85 years of age as very elderly [3]. Prevalence of anaemia in various studies conducted by Denny et al. and Patel et al. ranged from 9.2% to 23.9% in elderly men and 8.1% to 24.7% in elderly women. In the analysis of the 1991-98 National Health and Nutrition Examination Survey (NHANES), the

prevalance of anaemia (by WHO standards) began to rise after 50 years to a rate greater than 20% at age of 85 years and was found to be associated with poor physical performance and lower muscle strength [3].

Anaemia is of common concern in older people because it can have significant morbidity and mortality. Because anaemia is a sign and not a diagnosis, an evaluation is almost always warranted to identify the underlying cause and to establish whether anaemia is a marker of disease burden or a mediator in the causal pathway leading to adverse events. Studying the morbidities associated with the geriatric age group becomes a major health issue for the country, anaemia being one of the most important, being a treatable condition. It is a multifactorial condition with varied pathophysiology and increased comorbidities. Thus in this population, it can have significantly more severe complications than in younger adults and can greatly hamper the quality of life [4]. Bone marrow examination becomes an essential part of natural course of patient management and establishment of diagnosis in elderly due to various reasons like, pyrexia of unknown origin, cytopenias, lymphomas/leukemias and MDS and involvement of marrow by secondary malignancies and to differentiate myelodysplasias from megaloblastic anaemias. Rectification of any of these abnormalities contributes significantly to overall improved outcome with respect to physiological parameters as well as quality of life [5].

### Materials and Methods

- **Study Area:** Bharati Vidyapeeth Deemed to be University Medical College Hospital and Research Centre, Pune
- **Study Design:** Prospective study
- **Inclusion Criteria** - Age >65 years presenting in Bharati hospital with anaemia (hemoglobin (Hb)

<13g/dl in males, Hb<12g/dl in females) undergone bone marrow examination

- **Study Duration:** August 2015 to August 2017

A cross sectional observational study was carried out on 75 patients aged 65 years and above, presenting to our hospital and undergoing bone marrow examination, fulfilling the WHO criteria of anaemia (hemoglobin (Hb) <13g/dl in males, Hb<12g/dl in females). These patients were further investigated as follows, first to classify these anaemias morphologically, and then to assign them to the closest possible etiology on the basis of laboratory results. The study was carried out over a period of 24 months. The patient selection was based on inclusion criteria which included all the patients presenting to Bharati hospital of age 65 years and above with anaemia and undergoing bone marrow examination.

- The demographic profile of the patient with detailed history was recorded with special focus on possible etiology and details of general and systemic examination was recorded. Results from other investigations prescribed to the patient were included in the data collection.
- The first line of investigation included Hb level, Total leucocyte count (TLC), platelet count, blood indices, reticulocyte count, Peripheral blood smear examination carried out on Automated hematology analyser and other standard techniques.
- Bone marrow aspiration and trephine biopsy was done as a part of natural course of patient management and analysed. Bone marrow aspirate was stained using Romanowsky stains. Bone marrow trephine biopsy was stained using Hematoxylin and Eosin stain. Wherever required special stains like Periodic Acid Schiff (PAS), Myeloperoxidase (MPO) Prussian blue and reticulin stain were done.

- Based on the above parameters , the anaemias were classified into one of the following subtype.

**A) Normocytic normochromic anaemia**

**B) Microcytic hypochromic anaemia**

**C) Macrocytic anaemia**

- Based on the morphological classification, further investigations were carried out to determine etiology of anaemia as follows:
- Microcytic Hypochromic and Macrocytic Anaemia**
  - Serum Ferritin and Iron, Total Iron Binding Capacity, % Iron Saturation
  - Serum cobalamin, Serum folate
- Normocytic Normochromic Anaemia**
  - Serum Creatinine
  - Creatinine Clearance
  - Erythropoietin levels (where indicated and feasible)
  - Serum Iron, Total Iron Binding Capacity, %Iron Saturation (where indicated)
  - Lactate Dehydrogenase (LDH),Indirect Bilirubin ( in cases where hemolysis was suspected).
  - Special investigations (as per clinical profile of patients such as Functional Tests, protein electrophoresis, Autoimmune disease markers, Tumor markers, Immunohistochemistry and flowcytometry)
- These anaemias were then subclassified into the four groups as under:
  - Nutritional anaemia
  - Anaemia due to Chronic Kidney Disease
  - Anaemia due to Chronic illness/ Malignancy
  - Unexplained Anaemia
- The morphological and etiological classifications were correlated with the bone marrow findings in order to elucidate causative factors and related to the clinical presentation.

**Results**

**Table 1: Distribution of Bone Marrow cases in 75 cases out of 400 who underwent B.M examination.**

S.no	Bone marrow diagnosis	No. of cases
1	Multiple Myeloma	14
2	Haemophagocytosis	9
3	Reactive marrow	8
4	Megaloblastic Anaemia	7
5	Aplastic Anaemia	6
6	ITP	5
7	LPD	5
8	AML	3
9	Infectious etiology	3
10	Hypocellular	3
11	MDS	3
12	IDA	3
13	Hypersplenism	2
14	CML	1
15	Metastasis to bone	1
16	Myelofibrosis	1
17	Eosinophilia	1
	Total	75

Out of total 75 cases who underwent BM examination 55 cases had NCNC anaemia forming the largest morphological anaemia category. 11 cases had MCHC anaemia and 9 cases had macrocytic anaemia. As shown in **Table 1**, largest group was of 14 cases of MM. The percentage of plasma cells ranged from 20-60% in these cases. (**Figure1** ). 12 of these cases had NCNC anaemia and 2 had MCHC anaemia. Of these 11 had severe and 3 had moderate anaemia. 6 cases presented with skeletal complaints like backpain and other further workup showed lytic lesions in bones. 2 cases had Bence Jones protein in urine. 2 cases had acute tubular necrosis (ATN), 1 case had pancytopenia and 3 cases had acute kidney

injury (AKI). Out of these 14 cases, 1 had severe leucopenia. 1 case was diagnosed as Waldenstrom's macroglobulinemia. 3 cases underwent protein electrophoresis. Of these 2 showed monoclonal band on gamma region and 1 showed biclonal gammopathy, band 1 in beta and band 2 in gamma region.

There were 9 cases who had evidence of haemophagocytosis (**Figure 3**), out of which 4 had pancytopenia and rest had bicytopenia. 7 of these cases had NCNC anaemia, 1 had macrocytosis and 1 had MCHC anaemia. 8 cases gave a history of chronic infection with low grade fever for long duration. 1 of these cases had peritoneal TB. 2 had hepatomegaly and rest had no specific infection. 1 case had pyrexia of unknown origin with pancytopenia and severe anaemia. Out of 9 cases 4 had moderate anaemia and 5 had severe anaemia.

The third largest group comprised of 10.6 % (8) cases of reactive marrow. In 7 of these 8 cases marrow were done to rule out involvement in K/c/o NHL where none showed involvement and in 1 case with moderate anaemia the patient had pyrexia of unknown origin. In this case bone marrow finding were also normal.

There were 7 (9.3%) cases comprising of megaloblastic anaemia. B12 and folate levels were low in all these cases. 4 cases had pancytopenia and rest three had bicytopenia. 6 cases had severe anaemia while only one case had moderate anaemia. 2 cases even had history of chronic alcoholism.

There were 6 cases of Aplastic anaemia. All these patients presented with generalised weakness and pancytopenia. 5 cases had severe anaemia and 1 case had moderate anaemia. All cases had NCNC blood picture. Due to missing records of previous admissions of the

patient in some other hospital, proper drug or causative history leading pancytopenia could not be elucidated.

There were 5 cases of ITP, 3 had mucosal and skin bleed, while the other 2 had persistent thrombocytopenia. 4 cases of ITP had NCNC blood picture while 1 case showed microcytic hypochromic blood picture. 4 cases had moderate anaemia and 1 case had mild anaemia.

There were 5 cases of lymphoproliferative disorder (LPD). All the cases had splenomegaly while 3 cases had both splenomegaly and hepatomegaly. 3 cases were of chronic lymphocytic leukemia and rest 2 were highly suggestive of LPD. 1 case had severe, 1 had mild and rest 3 had moderate anaemia. 3 cases had NCNC blood picture while 2 had MCHC blood picture. 2 cases had leucocytosis, 2 had leucopenia and only 1 case had normal TLC count.

There were 3 cases of Acute Myeloid Leukemia (AML). All 3 cases presented to hospital with fever and weakness. 2 cases had macrocytic and 1 case had NCNC blood picture. 2 cases had severe anaemia and 1 had moderate anaemia. All the cases had thrombocytopenia. 1 case had leucocytosis and other 2 had leucopenia. 1 case of AML was seen to arise from a dysplastic marrow.

1 case of myelofibrosis occurred in a 66 year old male with persistent seizure disorder who was on phenytoin for past 25 years and showed a NCNC blood picture with a progressive fall in platelet count. When patient came to hospital the platelet count was  $<10,000/\text{cumm}$ . BM revealed marked hypocellular marrow and increased fibrosis. Reticulin stain showed grade 4 myelofibrosis. Probably this was a case of secondary myelofibrosis due to long term intake of phenytoin. (**Figure 4**).

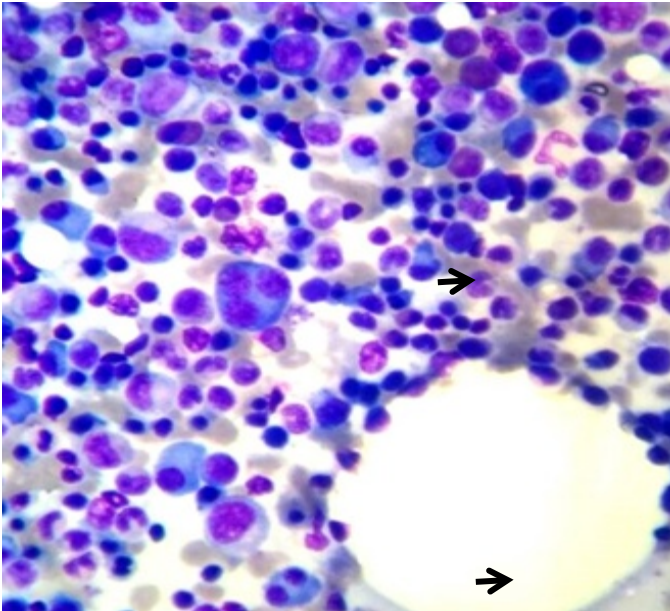
1 case of prostatic adenocarcinoma with Gleason score of  $4+3 = 7$ , group grade III presented with NCNC anaemia with leucoerythroblastic blood picture on peripheral blood smear (PBS) and bony pain. BM revealed clusters of



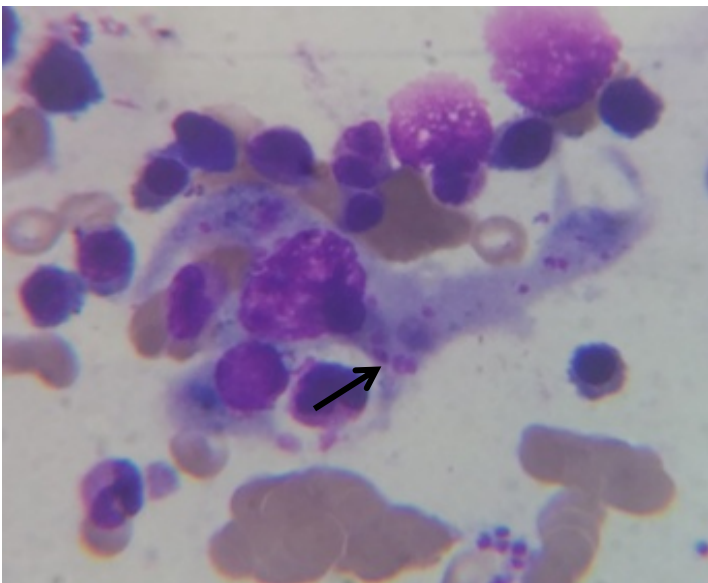
atypical non haematopoietic cells suggesting metastasis from carcinoma prostate. (Figure 5).

#### Case Images

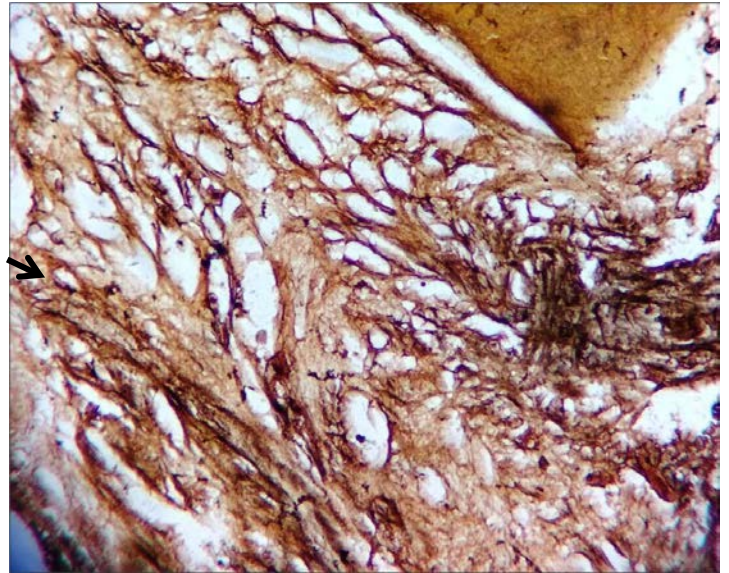
**Figure 1:** Bone marrow aspirate in case of Plasma cell dyscrasia showing abnormal binucleate and trinucleate plasma cells



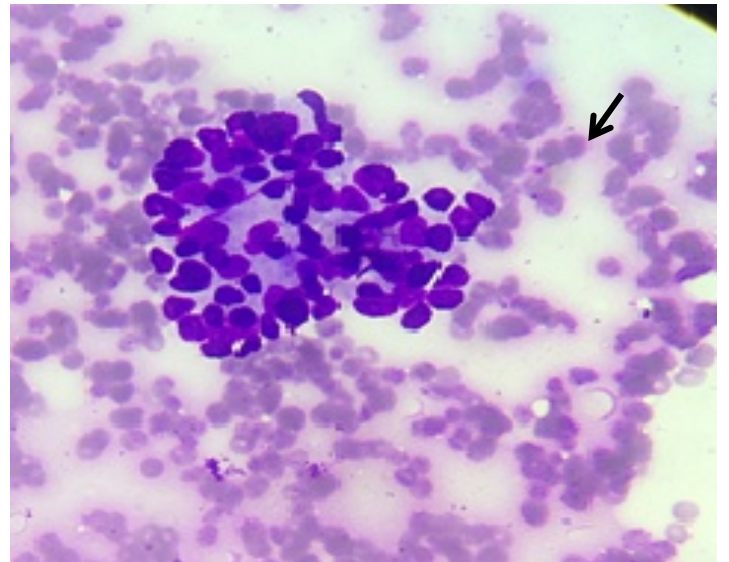
**Figure 2:** Bone marrow aspirate showing a reticulum cell which has engulfed lymphocyte, neutrophil, platelets; showing evidence of haemophagocytosis



**Figure3 :** Reticulin stain on bone marrow biopsy in a case of myelofibrosis showing Grade 4 myelofibrosis



**Figure 4:** Bone marrow aspirate in a case of carcinoma prostate showing clusters of metastatic tumor cells.



#### Discussion

In the present study 75 elderly patients with anaemia underwent bone marrow examination for various indications like, pyrexia of unknown origin, cytopenias ,lymphomas/leukemias and MDS, involvement of marrow by secondary malignancies and to differentiate myelodysplasias from megaloblastic anaemias. In the present study the largest group comprised of 18.6% (14) cases of MM. The likely cause of anaemia in these cases

is infiltration of marrow spaces by increased number of abnormal plasma cell thus suppressing the normal hematopoiesis. Bone marrow thus helped in making a confirmed diagnosis in these cases. The second largest group comprised of 12%(9) cases of marrow with evidence of haemophagocytosis. All these cases had a history of either pancytopenia, bicytopenia, organomegaly, pyrexia of unknown origin or chronic illness like tuberculosis with most having moderate to severe anaemia. These cases could not be labelled as Hemophagocytic Lymphohistiocytosis (HLH) due to non-availability of adequate diagnostic criteria as defined by Janka GE et al. According to this definition following should be present to label as HLH.

- 1) Fever
- 2) Splenomegaly
- 3) Cytopenia including 2 or more cell lines
- 4) Hypertriglyceridemia/hypofibrinogenemia
- 5) Hepatitis
- 6) Lower absent Natural killer cell activity
- 7) Serum ferritin >500 Hg/L
- 8) Soluble CD 25 >2400 U/mL
- 9) Hemophagocytosis as demonstrated in lymph node, spleen or bone marrow.

HLH has been associated with mutations of genes coding for proteins like perforin 1, UNC13D and syntaxin 11 and is triggered by stimuli such as infection or severe uncontrolled T cell and histiocytic reaction leading to pancytopenia also causing anaemia. HLH can be fatal and high diagnostic suspicion should be entertained. BM studies must be performed as it provides a direct evidence of haemophagocytosis.

The third largest group comprised of 10.6 % (8) cases of reactive marrow. In 7 of these 8 cases marrow were done to rule out involvement in K/c/o NHL. None of these cases showed involvement and in 1 case with moderate

anaemia the patient had pyrexia of unknown origin. In this case bone marrow findings were also normal. Thus the cause could not be attributed to any etiology here. In this group all cases had mild to moderate anaemia. BM studies help us in staging the lymphomas and plan additional therapy.

There were 7 (9.3%) cases comprising of megaloblastic anaemia. B12 and folate deficiency are classic causes of macrocytic anaemia and commonly present with anaemia and thrombocytopenia. In elderly it can be associated with chronic alcoholism and decreased absorption. Also long term megaloblastic anaemia can lead to an unrecognized Myelodysplasia occurring in marrow. BM studies thus help in early identification of underlying MDS on a background of megaloblastic anaemia. In the present study there was one case of AML which was seen to arise from a dysplastic marrow.

The present study comprised of 8 % (6) cases of aplastic anaemia (AA) (Figure 12). 5 of the cases presented with severe anaemia and pancytopenia. AA can be caused by multiple etiologies including drugs, chemicals, radiation, viruses etc. Although the exact cause of AA is unknown majority of AA are idiopathic and are thought to be a result from attack of effector T cell leading to BM failure and peripheral pancytopenia [6]. The cause of aplastic anaemia could not be established in these cases due to non-availability of the patient history records and previous hospital admissions elsewhere.

In the study by Tilak et al, MM comprised of 21.5%, megaloblastic anaemia comprised 12.2 % and AA comprised of 4.3% of all bone marrow cases and the figures in the present study were favourably comparable with his study [7].

### Conclusion

Anaemia among elderly is commonly a multifactorial condition associated with a variety of adverse outcomes

including mortality. Bone marrow forms an important part of the workup of the unexplained and persistent cytopenias in the elderly due to higher frequencies of primary malignancies, metastatic malignancies, myelodysplasias and myeloproliferative neoplasms in them. This study is thus a small contribution in the Indian context to the various epidemiologic and hematologic aspects of anaemia in the geriatric population and aims to address geriatric care in an attempt to reduce the disease burden in this population.

## References

1. World Health Organization. Definition of an older or elderly person: Proposed Working Definition of an Older Person in Africa for the MDS Project. URL: <http://www.who.int/healthinfo/survey/ageingdefnolder/en/index.html> [accessed 2012-11-03][WebCite Cache]. 2013 Oct.
2. WHO. Haemoglobin concentrations for the diagnosis of anaemia and assessment of severity. Vitamin and Mineral Nutrition Information System. Geneva, World Health Organization, 2011 (WHO/NMH/NHD/MNM/11.1)
3. Guralnik JM, Eisenstaedt RS, Ferrucci L, Klein HG, Woodman RC: Prevalence of anaemia in persons 65 years and older in the United States: evidence for a high rate of unexplained anaemia. *Blood* 2004 oct 104:2263-2268.
4. Ania Lafuente BJ, Fernandez-Burriel Tercero M, Suarez Almenara JL, Betancort Mastrangelo CC, Guerra Hernandez L (2001) Anaemia and functional capacity at admission in a geriatric home. *An Med Interna* 18(1):9-12.
5. Penninx BW, Guralnik JM, Onder G, Ferrucci L, Wallace RB, Pahor M (2003) Anaemia and decline in physical performance among older persons. *Am J Med* 115:104-110
6. Weinzierl EP, Arber DA. The differential diagnosis and bone marrow evaluation of new-onset pancytopenia. *American journal of clinical pathology*. 2013 Jan 1;139(1):9-29.
7. Tilak V, Rani D, Ambhir IS. Characteristics Of geriatric anaemia in and around Varanasi: A hospital based study. *Indian J. Prev Soc Med*. 2013 Jan;44:1-2