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Original Research Article

A Study of the Peripheral Smears in a Tertiary Care Teaching Hospital of Andaman & Nicobar Islands

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ABSTRACT

Background: Peripheral blood film is one of the basic and highly informative haematological tool for screening, diagnosis and monitoring of disease progression and to look for therapeutic response. The understanding of peripheral blood interpretation is very important for a successful clinical practice as it exposes the various morphology of peripheral blood cells, which ensures its place in diagnosis of various blood and its related disorders. Therefore this study was done to know the prevalence of haematological conditions in a relatively untouched population of Andaman & Nicobar Island.
Methods: This study was conducted over a period of 1 year from January 2016 to January 2017 in tertiary care teaching hospital of Port Blair. Peripheral smear along with CBC were done of all the patients. Smears were prepared by Leishman stain and Microscopy was done. The data was compiled as per Age, gender and peripheral smear findings and was presented in percentage and proportions.
Observations: A total of 1629 patients were enrolled in this study, out of which 940 were females and 689 were males with female: male ratio of 1.49:1. Out of 1629, Normal PBF picture was seen in 266(16.3%) patients, Anaemia of different types was seen in 1015(62.3%) patients. Severe Cases of Anaemia were 184(18.1%) females and 96(9.5%) males. Incidence of neutrophilic leucocytosis was observed in 131(8.04%) and eosinophilia in 60(3.6%) patients. Prevalence of anaemia, thrombocytopenia and leukaemia was 1015(62.3%), 106(6.5%), 07(0.42%) respectively.
Conclusion: In a resource limited area such as Andaman group of Islands, Peripheral blood smear examination is a simple and inexpensive tool, yet its value is very high in diagnosing various haematological disorders which serves as a guide for further management & patient welfare.

Keywords: Anaemia, peripheral blood smear, Leishman stain, neutrophilic leucocytosis, thrombocytopenia.

INTRODUCTION

Peripheral blood film is one of the basic and highly informative haematological tools for screening, diagnosis and monitoring of disease progression and to look for therapeutic response. The understanding of peripheral blood interpretation is very important for a successful clinical practice. It exposes the various morphology of peripheral blood cells, which ensures its place in diagnosis of various blood and its related disease. [1] Routinely analysing of blood films helps to facilitate interpretation of various haematological disorders as well as diagnostic tool for Etio-pathological work up of various anaemia. [2] Peripheral blood film analysis is done for common clinical conditions include unexplained cytopenias: anaemia, leucopenia or thrombocytopenia; unexplained leucocytosis, lymphocytosis; unexplained jaundice or hemolysis; suspected chronic or acute myelo-proliferative neoplasm; severe bacterial

sepsis and parasitic infections; malignancies with possible bone marrow involvement; suspected cases of nutritional anaemia; suspected chronic lympho-proliferative diseases such as chronic lymphocytic leukaemia; lymphoma with leukaemic spills; evaluation of therapeutic response in haemoglobinopathies. [1] Microscopic examination of the peripheral blood is used to supplement the information which is provided by automated cell counter and sometimes it provides information that cannot be obtained from automated cell counter i.e hemoparasites. [3]

Anaemia is functionally defined as an insufficient RBC mass to adequately deliver oxygen to peripheral tissue. [4] As per WHO criteria it is defined as haemoglobin concentration lower than 12g/dl in women and 13g/dl in men. Along with most grading classification systems, mild grade anaemia was defined as a haemoglobin concentration between 10.0 and 11.9 g/dl in women and between 10.0 and 12.9 g/dl in men, moderate anaemia was defined as haemoglobin level of 7-9.9 g/dl and severe anaemia was defined as haemoglobin levels less than 7 g/dl for both males and females respectively. For children 6 months to 5 years of age, anaemia is defined as a Hb level < 11g/dl, children 5–11 years of age Hb< 11.5 g/dl, adults males Hb< 13 g/dl; non-pregnant females Hb< 12g/dl, and pregnant females Hb< 11g/dl. [5]

NEUTROPHILIC LEUCOCYTOSIS: Leukocytes are usually elevated in acute infections and inflammatory conditions. Leukocytes count more than 11000/ µl were considered as leucocytosis. Neutrophilia is defined as an increase in the absolute blood neutrophil count to a level greater than two standard deviations above the mean value for normal individual. This level is approximately 7.5 x10/l. Extreme neutrophilia is referred as leukemoid reaction. [6] Neutrophils with toxic granules were defined by the presence of dark blue to purple colored granules in the cytoplasm. The most common tissue response in acute inflammation is dominated by neutrophils. [7]

EOSINOPHILIA: Eosinophils can be numerated in the blood either by wet counts in modified Neubauer chambers or dried blood films. It should be enumerated in absolute number rather than as a percentage of white cells, as the latter will depend on the total cell count. The normal eosinophil count is generally less than 0.4x10/µl, mild eosinophilia as less than 1.0x10/µl, a moderate as 1.0to5.0x10µ/l and high as greater than 5 .0x10µ /l. [8]

THROMBOCYTOPENIA: Thrombocytopenia is defined as platelet count below the normal range for the population150000 to 450000.For the cancer patient receiving treatment NCI has developed the severity criteria of thrombocytopenia. Platelet count of 75000 to 150000 as grade 1,50000 to 75000 grade 2,25000 to 50000 as grade 3,below 25000 as grade 4 thrombocytopenia. [9] Prolonged thrombocytopenia and absence of relative increase in the platelet count was also associated with a greater risk of mortality and thus require proper diagnosis.

PANCYTOPENIA: Pancytopenia is a disorder in which all three formed elements of blood (RBC, WBC, and Platelets) are decreased in number. Inclusion criteria require presence of all 3 of the following: haemoglobin, <9 g/dl; total leukocyte count (TLC), <4000/ul; platelet count, <100,000/ul. The severity of pancytopenia and the underlying pathology determine the management and prognosis of the patient. [10]

LEUKAEMIA: It is a haematological malignancy that arise when there is dysregulation of the division or life span of a blood cell or its precursor. It is characterised by rapid and disorderly proliferation of leukocytes and its precursor in the blood. Leukaemia is one of the most frequently occurring cancers in all races or ethnicities with relative proportion varying between 25-40%. Diagnosis of leukaemia was done by 20% blast criteria and

cytochemical stain was used to distinguish between AML & ALL. [11]

MATERIALS AND METHODS

This was a cross-sectional study of 1 year duration conducted in Andaman & Nicobar Islands Institute of medical sciences which is a tertiary care teaching hospital in Port Blair, India after obtaining approval from Institutional Ethics Committee. Patients fulfilling the inclusion criteria were enrolled in the study. Data thus obtained was filled in Case Record Forms and was presented as percentage & Proportions.

RESULTS

In the present Study, total numbers of patients with anaemia were 1015 with female preponderance as the numbers of female patients with anaemia were 637(62.7%). (TABLE -1)

Table-1 Distribution of patients based on gender and severity of anaemia

GENDER	MILD ANAEMIA	MODERATE ANAEMIA	SEVERE ANAEMIA	TOTAL
Females	16.7%	27.8%	18.1%	62.7%
Males	10.5%	8.8%	9.5%	28.8%

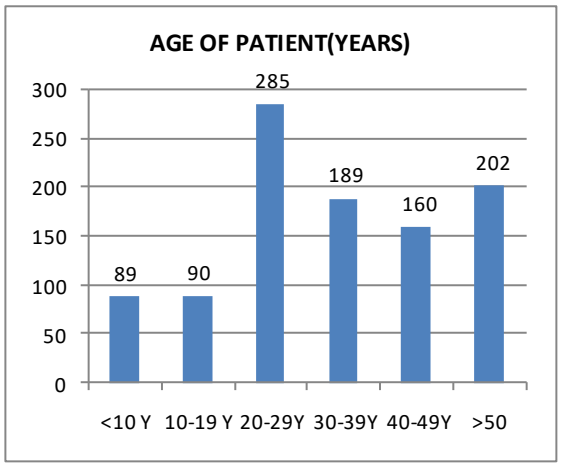

Figure-1 Distribution of patients based on their age

In this study, most common age group of anaemia is 20-29 years as 285 (28%) patients were in this age group, followed by >50 years age group with 202(20%) patients, followed by 30-39 years age group with 189(18.6%) patients, then 40-49 years age group with 160(15.8%) patients, least common age group with anaemia was <10 years age group with 89 (8.8%) patients. (FIGURE-1)

In present study, most common type of anaemia is microcytic hypochromic in 655(64.5%)no of patient, followed by dimorphic anaemia observed in 178(17.5%)no of patient, then normocytic normochromic anaemia in 140 patients (13.8%), followed by macrocytic anaemia in 30 patients(03%). Only 12(1.2%) patients were having haemolytic type of anaemia. (TABLE-2)

Table- 2 Distribution of patients based on type of anaemia

TYPE OF ANEMIA	NO OF PATIENTS	PERCENTAGE (%)
MICROCYTIC HYPOCHROMIC	655	64.5%
DIMORPHIC ANEAMIA	178	17.5%
NORMOCYTIC NORMOCHROMIC	140	13.8%
MACROCYTIC ANAEMIA	30	3%
HEMOLYTIC ANAEMIA	12	1.2%

The prevalence of Neutrophilic Leucocytosis (NL) with toxic granules was observed in 8.04% of cases. It was more commonly seen in females than males being 55.7 % and 44.2% respectively. The relationship between toxic granules in mature neutrophils in infectious disease and inflammation has been described. These neutrophils show toxic granules with shift to left. The prevalence of thrombocytopenia in our study was 6.5%. Major causes of thrombocytopenia were severe infections, drug induced, gestational thrombocytopenia, sepsis and other haematological malignancies. Also, patients had pancytopenia on their peripheral blood smear that made a frequency of 2.7% out of the total admissions during the study period.

Out of these cases, there were 56.8% males and 43.1% were female patients with male to female ratio were 1.3:1.

Leukaemia / myeloproliferative disorders were seen in 0.42% of the total study group. Only 6 patients were diagnosed with acute leukaemia, one patient was diagnosed with CML. Total 7 patients were having haemoparasite in peripheral smear. (FIGURE-2)

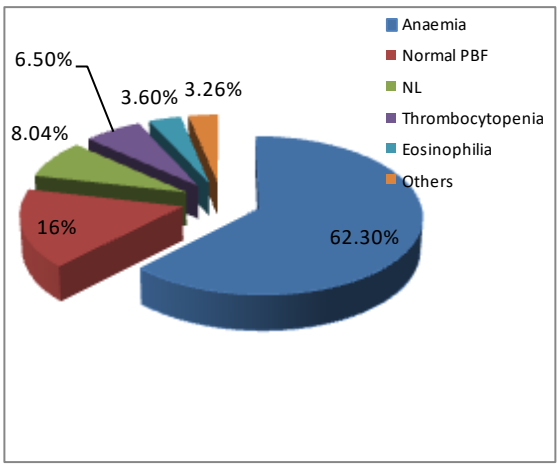

Figure-2 Distribution of patients based on findings in peripheral blood smear.

DISCUSSION

Anaemia and iron deficiency anaemia are often used interchangeably. There are however mild to moderate forms of iron deficiency in which the host is not yet anaemic, but tissues are functionally iron deficient. In addition, iron deficiency accounts for most of the anaemia that occurs in underprivileged environments, multiple other causes exist independently or co-existent with this micro-nutrient deficiency. Iron deficiency is the result of long-term negative iron balance.

Iron stores in the form of haemosiderin and ferritin are progressively diminished and no longer meet the needs of normal iron turnover. India is facing a grave public health problem, with the prevalence of anaemia in India being > 40%. [5] Anaemia is an indicator of poor nutrition and poor health with major consequences for the human health, as well as for the social and economic development.

TYPES OF ANAEMIA

Anemia is one of the most common health related problems faced by the society. Although nutritional deficiency is the most common cause, other causes like hemolysis are also common. [(12)] Iron deficiency anaemia was considered if the patient had low serum iron (lower than 50 µg/dl in women and 60 µg/dl in men), low ferritin (lower than 15 ng/ml), low transferrin saturation rate (lower than 16%) or increased total iron binding capacity (higher than 450 µg/dl). Anaemia of chronic disease was defined as low circulating iron in the presence of increased iron stores (normal or increased ferritin higher than 100 ng/ml, transferrin saturation higher than 25% and lower than 50%) and decreased total iron binding capacity (lower than 250 µg/dl). Hemolytic anaemias are characterised by increased red cell destruction. Amongst the inherited causes of haemolytic anaemias, thalassemias are the most common. Thalassemia is a quantitative hemoglobinopathy due to reduced synthesis of one or more globin chains. [12] Thalassemia trait was considered when the following conditions were present: low or very low mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH), increased red blood cell (RBC) count, normal or increased circulating iron in the presence of normal or increased iron stores. Anaemia associated with folate or vitamin B12 deficiency was defined as concentrations of folate lower than 3.0 ng/ml or vitamin B12 lower than 200 pg/ml and MCV higher than 95 fl. Anaemias that could not be classified into any of the previous categories were considered to be of unexplained origin. The hematologic characteristics consistent with the possible presence of myelodysplastic syndromes were defined consistent with the possible presence of myelodysplastic syndromes were defined as macrocytosis in the absence of folate or B 12 deficiency, and leukopenia. Present study showed the most common type of anaemia in portblair is microcytic

hypochromic being 64.5% followed by dimorphic anaemia,17.5% .

In present study, the prevalence of anaemia among males (10-88 years) was 28.8%, which was more as compared to NFHS 3 data according to which it is 24%. [13] In a study conducted by Vitull et al. in 2011 and show the prevalence of anemia among males was 89.9%. [14] The present study highlights a higher prevalence of anemia in females and males as compared to previous data of National Family Health Survey-3data showed that 55% of the women and 24% of the men were anemic and that Punjab, along with Manipur, Mizoram, Goa and Kerala had the lowest levels of anemia throughout the country. [13] Even in these states, however, more than 30 % of the women were found to be anaemic. (TABLE-3)

Table- 3 Showing comparison of percentage of anaemia among females and males with other studies.

STUDY	FEMALE	MALE
NFH3	55%	24%
Gupta VK et al(2011) [14]	89.5%	89.9%
Present study	62.7%	28.8%

Anaemia is graded as mild, moderate and severe. In Comparison with other studies done by Vitull k gupta & Bentley, [14,15] present study highlights more cases of moderate and severe anaemic as compared to mild anaemia. (TABLE-4)

Table- 4 Showing the comparison of grading of anaemia among females with other studies

Study	Mild (%age)	Moderate (%age)	Severe (%age)
Gupta VK [14]	49.8	38.2	1.5
Bentley ME [15]	32.4	14.19	2.2
PRESENT STUDY(2016)	16.7	27.8	18.1

In this study, Neutrophilic Leucocytosis (NL) with toxic granules was observed in 8.04% of cases. In 1932 Kugeland Rosenthal described toxic granules as large, dark, irregular, basophilic granules in the cytoplasm of neutrophilic granulocytes. They are seen in patients with severe infection in comparison to the typical fine granular staining pattern and are helpful in predicting acute infection. In Liu et al. described the usefulness of these morphological changes, that the presence of TG was more predictive of bacteraemia than the total leukocyte count. [16] In our study, the incidence of thrombocytopenia was 6.5% which was slightly higher than reported by in 2016 observed the prevalence of thrombocytopenia as 2.5%. [18] (TABLE-5)

Table- 5 Showing comparison of percentage of thrombocytopenia with other studies.

Study	No of Cases	Thrombocytopenia(%)
Nadir et al [17]	18000	2.3
Khatib Y et al [18]	15000	2.5
Present study(2016)	106	6.5

The total incidence of pancytopenia reported in our study was 25 patients in male group and 19 patients in female group with female to male ratioof 1.3:1 which is in accordance with the other studies done by gayathri et al, [10] and Mallick M. [20] (TABLE-6)

Table- 6 Comparison of incidence of pancytopenia among males and females with other studies.

STUDY	MALES	FEMALES	RATIO
Lakhey et al [19]	39/54	15/54	2.6:1
Mallik M [20]	512/817	305/817	1.67:1
Gayathri et al [10]	57/104	47/104	1.2:1
Present study(2016)	25/1629	19/1629	1.3:1

Megaloblastic anemia was considered the most common cause of pancytopenia in their study. The high prevalence of nutritional anemia in India has been cited for the increased frequency of Megaloblastic anemia. Total incidence of leukaemia was 0.42%. Acute leukaemia was more prevalent as compared to chronic leukaemia. Out of total 7 patient 6 were diagnosed as acute leukaemia and one was diagnosed with chronic leukaemia.

CONCLUSION

The present study shows the incidence of pancytopenia and other haematological diseases in the patients of Andaman & Nicobar group of islands. Furthermore studies with a larger population/sample size should be done in order to have a better understanding of the factors responsible for various diseases like

pancytopenia, so that further management can be planned accordingly.

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