

Original Research Article

# To Study Automated Histogram Patterns with morphological features noticed on peripheral smear at CMCH Bhopal.

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## Abstract:

**Background & Method:** The aim of this study is to Study Automated Histogram Patterns with morphological features noticed on peripheral smear at CMCH Bhopal.

**Result:** Mean hemoglobin was significantly least among Microcytic hypochromic anemia ( $7.57\pm 1.55$ ) whereas it was  $8.50\pm 1.44$  among subjects with Pancytopenia,  $9.68\pm 0.23$  among subjects with Dimorphic Anemia and  $9.80\pm 0.23$  among subjects with Normocytic Anemia. Mean MPV was significantly lesser among subjects with Pancytopenia compared to other groups.

**Conclusion:** Mean MPV was significantly lesser among subjects with Pancytopenia compared to other groups. Mean ANC was significantly lesser among subjects with Pancytopenia compared to other groups. Mean ALC was significantly lesser among subjects with Pancytopenia compared to other groups. Mean AMC was significantly lesser among subjects with Pancytopenia compared to other groups.

**Keywords:** automated, histogram, morphological, peripheral smear & blood.

**Study Designed:** Observational Study.

## 1. INTRODUCTION

Anemia is major public health problem over worldwide, especially among females in developing countries. Anemia is defined as a reduction of total circulating red cell mass below normal limits. It is estimated that approximately 33% of the world's population has anemia with iron deficiency considered to be the leading cause of anemia accounts for almost 9% of the world's population living years with a disability burden. It has also been estimated that worldwide 273 million preschool age children having anemia (43% of all children), 32 million pregnant women having anemia (38% of all pregnant women); and 496 million non-pregnant women having anemia (29% of all non-pregnant women). Anemia is most prevalent in central and west Africa and south Asia.[1]

Thirty to Forty years ago, Laboratory hematology was labor intensive and time consuming. Procedures were manual. Reagents were prepared in the laboratory from raw chemicals. Hemoglobin measurement was based on the cyanmethemoglobin method, which involve tedious procedure. The automated hematology analyzer has replaced the traditional manual

methods for hematological parameters as the initial screening and detection system for hematological abnormalities in modern clinical setups. While enhancing the speed, accuracy and precision of test results, this has also added a new dimension to hematology reporting.[2] From the earlier instruments that used electrical impedance as the sole counting principle for blood cells, modern day analyzers, in addition, use conductivity differences, cytochemical staining, light scatter, and flow cytometric principles.[3]

Blood cell histograms are produced by the modern automated haematology analysers which are routinely used to count blood cells. A good interpretation of this histogram provides a wealth of information on many haematological conditions than mere cell counts, helping to narrow down the differential diagnosis at a very early stage even before higher level investigations are ordered. Histogram interpretation needs careful analysis of RBC, WBC and platelet distribution curves. The overall pattern of histogram by itself is meaningless unless it is compared with a reference normal curve and confirmed microscopically.[4]

## **2. MATERIAL & METHOD**

The Study will be undertaken in the Central Clinical Laboratory in Department of pathology CMCH(Chirayu Medical College And Hospital), Bhopal(M.P.)

### **Method of Collection-**

3 ml of EDTA venous blood sample will be collected. Leishman staining will be done on it. Histogram will be obtained from Automated haematology analyzer and comparison and interpretation of both will be done

### **Inclusion Criteria**

1. All Blood Sample from patients with age of 15yr and above will be included.

### **Exclusion Criteria**

2. All cases that have undergone blood transfusion will be excluded from the study  
3. Inadequate quantity of blood sample for automated analyzer(less than 3ml)will be excluded.

The hematological parameters & histogram will be recorded from the automated hematology analyzer. Peripheral Smear will be prepared and stained with Leishman stain. Obtained Histogram will be evaluated in relation to the RBCs, WBCs and platelets with their peripheral blood smear picture and with the Clinical diagnosis of cases.

Pour Leishman stain drop wise on the slide and wait for 2 minutes.This allows fixation of blood film in methyl alcohol.Add double the quantity of buffered water over the slide. Dry in air and examine under oil immersion lens of the microscope.

### 3. RESULTS

**Table 1: Distribution of study population according to Age**

|                               | Age(years) |       |        |       | cases |
|-------------------------------|------------|-------|--------|-------|-------|
|                               | Mean       | SD    | 95% CI |       |       |
| ALL                           | 16.80      | 13.66 | 12.69  | 20.91 | 45    |
| AML                           | 46.24      | 9.30  | 43.60  | 48.88 | 50    |
| CLL                           | 73.17      | 2.76  | 76.00  | 76.00 | 23    |
| CML                           | 33.13      | 2.15  | 32.22  | 34.03 | 24    |
| Dimorphic Anemia              | 52.84      | 13.29 | 50.27  | 55.41 | 104   |
| Macrocytic Anemia             | 47.05      | 1.68  | 43.69  | 50.41 | 102   |
| Hemolytic Anemia              | 37.33      | 1.93  | 33.46  | 41.18 | 98    |
| Microcytic hypochromic anemia | 43.18      | 16.44 | 41.80  | 44.56 | 498   |
| Normocytic Anemia             | 43.45      | 16.05 | 42.31  | 44.58 | 318   |
| Pancytopenia                  | 48.50      | 1.54  | 47.73  | 49.27 | 75    |
| Reactive Lymphocytosis        | 6.50       | 1.54  | 5.73   | 7.27  | 252   |

The mean age was significantly lesser among subjects with Reactive Lymphocytosis ( $6.50 \pm 1.54$  years) followed by ALL ( $16.80 \pm 13.66$ ), CML ( $33.13 \pm 2.15$ ), Pancytopenia ( $48.50 \pm 1.54$ ), AML ( $46.24 \pm 9.30$ ) Microcytic hypochromic anemia ( $43.18 \pm 16.44$ ), Normocytic Anemia ( $43.45 \pm 16.05$ ), Dimorphic Anemia ( $52.84 \pm 13.29$ ) and CLL ( $73.17 \pm 2.76$ ).

**Table 2: Distribution of study population according to Hemoglobin**

|                               | Hemoglobin gm% |      |        |       |
|-------------------------------|----------------|------|--------|-------|
|                               | Mean           | SD   | 95% CI |       |
| ALL                           | 8.20           | 0.99 | 5.90   | 6.50  |
| AML                           | 8.32           | 1.13 | 6.00   | 6.64  |
| CLL                           | 14.50          | 0.00 | 14.50  | 14.50 |
| CML                           | 9.52           | 0.09 | 9.48   | 9.56  |
| Dimorphic Anemia              | 9.68           | 0.23 | 9.63   | 9.72  |
| Microcytic hypochromic anemia | 7.57           | 1.55 | 7.44   | 7.70  |
| Normocytic Anemia             | 9.80           | 1.96 | 9.66   | 9.93  |
| Macrocytic Anemia             | 9.04           | 2.18 | 4.68   | 13.4  |
| Hemolytic Anemia              | 9.8            | 2.49 | 4.82   | 14.78 |
| Pancytopenia                  | 8.50           | 1.44 | 7.78   | 9.22  |
| Reactive Lymphocytosis        | 11.95          | 0.26 | 11.82  | 12.08 |

Mean hemoglobin was significantly least among Microcytic hypochromic anemia ( $7.57 \pm 1.55$ ) whereas it was  $8.50 \pm 1.44$  among subjects with Pancytopenia,  $9.68 \pm 0.23$  among subjects with Dimorphic Anemia and  $9.80 \pm 0.23$  among subjects with Normocytic Anemia.

**Table 3: Distribution of study population according to Platelet count**

|                               | Platelet count |           |           |            |
|-------------------------------|----------------|-----------|-----------|------------|
|                               | Mean           | SD        | 95% CI    |            |
| ALL                           | 81200.00       | 56449.74  | 64240.62  | 98159.38   |
| AML                           | 48209.00       | 36887.60  | 37725.66  | 58692.34   |
| CLL                           | 338000.00      | 0.00      | 338000.00 | 338000.00  |
| CML                           | 163627.08      | 1081.36   | 163170.46 | 164083.70  |
| Dimorphic Anemia              | 746096.67      | 146478.25 | 717749.51 | 774443.82  |
| Microcytic hypochromic anemia | 219038.32      | 202593.42 | 202038.47 | 236038.17  |
| Normocytic Anemia             | 271751.89      | 137278.10 | 262034.01 | 281469.76  |
| Macrocytic Anemia             | 225372.5       | 98660.2   | 283456.90 | 4224692.78 |
| Hemolytic Anemia              | 248121.5       | 140897.5  | 256989.20 | 306607.80  |
| Pancytopenia                  | 27500.00       | 18007.35  | 18545.15  | 36454.85   |
| Reactive Lymphocytosis        | 396500.00      | 89007.77  | 352237.48 | 440762.52  |

Mean platelet count was significantly lesser among subjects with Pancytopenia compared to other groups.

**Table 4: Distribution of study population according to MPV**

|                               | MPV   |      |        |       |
|-------------------------------|-------|------|--------|-------|
|                               | Mean  | SD   | 95% CI |       |
| ALL                           | 12.48 | 2.41 | 11.76  | 13.20 |
| AML                           | 10.66 | 0.33 | 10.56  | 10.75 |
| CLL                           | 8.80  | 0.00 | 8.80   | 8.80  |
| CML                           | 9.45  | 0.21 | 9.36   | 9.54  |
| Dimorphic Anemia              | 9.42  | 0.34 | 7.35   | 7.48  |
| Microcytic hypochromic anemia | 9.38  | 1.46 | 9.26   | 9.50  |
| Normocytic Anemia             | 9.96  | 1.59 | 9.84   | 10.07 |
| Macrocytic Anemia             | 9.68  | 1.24 | 7.2    | 12.16 |
| Hemolytic Anemia              | 9.92  | 1.59 | 6.74   | 13.1  |
| Pancytopenia                  | 8.00  | 1.44 | 8.28   | 9.72  |
| Reactive Lymphocytosis        | 11.00 | 1.23 | 10.39  | 11.61 |

Mean MPV was significantly lesser among subjects with Pancytopenia compared to other groups.

#### 4. DISCUSSION

Morphological typing of anemia is done by PBS, helps clinician and hematologist in making early diagnosis and appropriate therapeutic intervention although automated hematology analyzer has improved accuracy, precision and reduced the subjective error and time consumption.

Even in era of automation and development of sophisticated auto analyzers, peripheral smear is still used as a basic and important diagnostic tool for anemia, leukemia and other hematological disorders.[5] The RBC histogram is an integral part of automated haematology analysis and is available routinely on all automated cell counters. The RBC histogram follows well-known coulter principle of counting and sizing red cells providing the basis for generating the histogram. The histogram in association with other CBC parameters such as RDW, MCV has been found abnormal in various haematological conditions.

In our study the mean age was significantly lesser among subjects with Reactive Lymphocytosis ( $6.50\pm 1.54$  years) followed by ALL ( $16.80\pm 13.66$ ), CML ( $33.13\pm 2.15$ ), Pancytopenia ( $48.50\pm 1.54$ ), AML ( $46.24\pm 9.30$ ), Microcytic hypochromic anemia ( $43.18\pm 16.44$ ), Normocytic Anemia ( $43.45\pm 16.05$ ), Dimorphic Anemia ( $52.84\pm 13.29$ ) and CLL ( $73.17\pm 2.76$ ). Aravind et al.[3] study showed a predominance of age group between 21-49 years, the mean being 36.08 years. This can be comparable to the age group in other studies. Bhadran et al.[6] found that maximum (77.04%) cases were present among age group of more than 15 years and the mean age was 42 years.

The most common morphological type of anemia in the study was Microcytic hypochromic anemia (72.2%) followed by Normocytic normochromic anemia (14%). This is in concordance with various other studies. Iron deficiency anemia is the most common cause of microcytic hypochromic blood picture. WHO has estimated that prevalence of anemia in pregnant women is 14.0% in developed and 51.0% in developing countries and 65-75 percent in India. About one third of the global population (over 2 billion) are anaemic.[7]Prevalence of anemia in all the groups is higher in India as compared to other developing countries.[8]

#### 5. CONCLUSION

There were 559 (35.6%) males and 1,030 (64.4%) females among study population with male to female ratio of 0.55. The mean age was significantly lesser among subjects with Reactive Lymphocytosis ( $6.50\pm 1.54$  years) followed by ALL ( $16.80\pm 13.66$ ), CML ( $33.13\pm 2.15$ ), Pancytopenia ( $48.50\pm 1.54$ ), AML ( $46.24\pm 9.30$ ), Microcytic hypochromic anemia ( $43.18\pm 16.44$ ), Normocytic Anemia ( $43.45\pm 16.05$ ), Dimorphic Anemia ( $52.84\pm 13.29$ ) and CLL ( $73.17\pm 2.76$ ). Mean hemoglobin was significantly least among Microcytic hypochromic anemia ( $7.57\pm 1.55$ ) whereas it was  $8.50\pm 1.44$  among subjects with Pancytopenia,  $9.68\pm 0.23$  among subjects with Dimorphic Anemia and  $9.80\pm 0.23$  among subjects with Normocytic Anemia.

Mean MPV was significantly lesser among subjects with Pancytopenia compared to other groups. Mean ANC was significantly lesser among subjects with Pancytopenia compared to other groups. Mean ALC was significantly lesser among subjects with Pancytopenia compared to other groups. Mean AMC was significantly lesser among subjects with Pancytopenia compared to other groups.

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