Establishment of Haematological Reference Values for Healthy Individuals Attending Ruhengeri Referral Hospital in Rwanda

Ally Dusabimana and Amie de Dieu Musabyimana

Department of Biomedical Laboratory Sciences (BLS), Faculty of Applied sciences

ABSTRACT

Background: The Laboratory investigations are very important for reaching to definitive diagnosis of diseases orientation and thus enabling optimal patient management based on informed diagnoses. Decision, and these are very difficult to obtain in the absence of reference values. In many cases, laboratory diagnosis such as haematological analyses are dependent on pre-defined locally established reference values.

Objective of the study: The objective of this study was to describe ranges of haematological reference values for healthy individuals attending Ruhengeri referral Hospital in Rwanda

Methodology: The cross sectional study was carried out in Northern Province at Ruhengeri referral hospital from July 2018 to September 2018. Participants were 252 healthy individuals aged less than one year to 68 years. From each participant, 4mL of blood samples were collected using K3 EDTA containers, and then analysed by Sysmex XS-500i automated haematology analyser.

Results: Haemoglobin levels varied with age and sex. The level decreased with the increasing age, and males had high haemoglobin level than females (15.69g/dL versus 14.46g/dL). Minimum mean values of haematological parameters for study participants were slightly on the high side with narrow confidence intervals compared to the Manufactured Sysmex values.

Conclusion: The findings may be used to define normal haematological values for Rwandan population and help physicians to better define haematological abnormalities in patients

INTRODUCTION

The concept of reference values was conceived by a group of Scandinavian scientists in the 1970s, and then developed by many works of French and Spanish Societies as well as the International Federation of Clinical Chemistry and Laboratory Medicine (IFCC-LM) and the National Committee for Clinical Laboratory Standards (NCCLS) in the United States during the 1980s.

According to the study done in Iran appropriate local reference values for haematological parameters are essential for screening, follow up, interpreting laboratory data and detecting haematological abnormalities. Haematological parameters may be affected by individual factors such as age, sex and lifestyle, and ecological factors such as ethnic background, climate, exposure to pathogens and altitude. They vary not only between individuals but also between populations.

Study conducted at Ethiopia have shown that the normal ranges of red blood cell counts (RBCs), haemoglobin (Hb) concentration, haematocrit, mean cell volume (MCV), total white cell count (WBC), platelet counts are known to vary with age, sex, dietary patterns, ethnic origin, genetic and environmental factors.

Moreover, the reference values, which were established by the studies conducted in different geographic locations, may not reflect the normalcy of the population in question and therefore, it is always desirable to identify the region specific reference intervals. In some of African countries, laboratories uses reference values obtained from the literature or inserts accompanying the reagent kits or instrument manuals. The few small studies conducted in African population syndicate differences in normal values compared with those for populations in industrialized countries. Lower values for haemoglobin, red blood cells, haematocrit, mean corpuscular volumes, platelets and neutrophils and higher monocytes and eosinophils levels were reported in African population compared to similar population in Western parts.

Many studies have revealed that reference values vary with several parameters such as ethnic origin, genetics, gender, altitude and environmental factors. It is important to recognize that reference values may differ between different laboratories. It is therefore important to be careful when interpreting patients’ results. Thus, unsuitable reference values of haematological profile might elevate the risk of
either unessential further investigations or failure to determine underlying disease.\textsuperscript{10} Laboratory reference intervals for healthy populations have not been established in most African countries. Currently many countries in sub-Saharan Africa including Rwanda uses the reference intervals derived from populations in Europe and North America.\textsuperscript{3} This type of study is essential to establish haematological reference values for Rwanda population. Using local laboratory haematological reference values may help to improve diagnosis and treatment of individuals with haematological disorders.

**METHODS**

**Ruhengeri Referral Hospital Demographic information of participants**

Ruhengeri is served as a public District Referral Hospital for many years, and is located in Musanze District of North Province in Rwanda. Since 2013, the facility serves as the National Referral Hospital and receive referred patients from its neighbouring hospitals and health facilities in northwest Rwanda. The hospital provides services to 406,557 people who live in Musanze District, and to 386,078 people from surrounding districts.

**Study Design**

Across sectional study was conducted at Ruhengeri referral hospital with the aims of establishing haematological reference parameters for healthy individuals.

**Population and Sample Size Estimation**

A total of 350 individuals were screened from July to September 2018 at Ruhengeri referral hospital. Out of 350 screened individuals, 252 health individuals selected and 98 excluded because of their disease conditions that would alter haematological values.

**Inclusion Criteria**

All healthy individuals aged 1 to 68 years who visited Ruhengeri Referral Hospital during data collection period (July to September 2018) were eligible for the study.

**Exclusion Criteria**

Individuals using anticoagulants, those with history of significant blood loss, blood donation, surgical operation within three months, those with disease conditions that would alter haematological values, those with pre malaria symptoms were excluded.

**Sample Collection**

Study participants’ particulars were recorded and registered in haematology register books. Blood sample was collected after cleaning the venepuncture site with pad soaked in 70% isopropyl alcohol. About 4ml of venous blood sample were collected in K3 EDTA containers, then Sysmex XS-500i automated haematology analyser was used for analysing haematological parameters after running daily control according to the manufacturer’s instructions.

**Data Analysis**

Data was analysed using Microsoft Excel sheet (2013) and Statistical Package for Social Sciences (SPSS) computer program (version 22). The percentile range (2.5%-97.5 %) was used to determine the higher and lower values of normal ranges.

**Ethical Consideration**

Ethical clearance was obtained from Ruhengeri referral hospital ethics committee with reference number: 1089/ HDR/HRR/2018. Informed consent was used from participants before collection of samples. The specimens and information were collected from the individuals under privacy and confidentiality and were not used for any purposes rather than this study. Each individual included in this study was given his/her tests results.

**RESULTS AND DISCUSSION**

**Social Demographic Characteristics of Study Population**

Out of 252 study participants, 23.6% were children under the age of 18 years, of which 27.2% were females. For adults, significantly high proportion of participants were males (Table 1).

<table>
<thead>
<tr>
<th></th>
<th>Under 18 years (%)</th>
<th>18+ years (%)</th>
<th>All (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Males</td>
<td>25 (19.7)</td>
<td>102 (80.3)</td>
<td>127 (50.4)</td>
</tr>
<tr>
<td>Female</td>
<td>34 (27.2)</td>
<td>91 (72.8)</td>
<td>125 (49.6)</td>
</tr>
<tr>
<td>Total</td>
<td>59 (23.6)</td>
<td>193 (76.6)</td>
<td>252 (100.0)</td>
</tr>
</tbody>
</table>

**Haematological Reference Ranges**

Table 2 summarized mean reference values for haematological parameters age group and sex. Independent student’s t-test was used to assess the differences between age groups and sex.

Haematological reference parameters for children below 18 years and adults aged 18 to 68 years are depicted in Table 2. As expected, haemoglobin levels varied with age and sex. The level decreased with the increasing age, and males had high haemoglobin level than females (15.69g/dL versus 14.46g/dL).

The sex difference in haemoglobin level is a well-established fact that has been reported in other studies\textsuperscript{11,12}.

**Comparison of Study Participants’ Ranges of Normal Haematological References with System Values**

Study participants’ normal ranges had narrow intervals compared to the manufactured Sysmex values (Table 3). haematological values among healthy individuals attending Ruhengeri referral hospital are lower on some parameters than sysmex reference values, obtained values of monocytes, white blood cells, platelets, neutrophil, eosinophils and lymphocytes on both genders are lower compared to sysmex values, the means of basophils are same as sysmex reference values while haemoglobin and red blood cells are higher (Figure 1).

Studies from other African countries have reported high mean Hb, PLT, RBC and WBC compared to the findings of the current study.\textsuperscript{13,14} The reason for the lower values in this study might be due to variation of the standardization.
TABLE 3: Comparison of Study Participants’ Ranges of Normal Haematological References with Sysmex Values

| Study participants’ normal haematological parameter ranges | Manufactured Sysmex Values |
|-----------------|-----------------|-----------------|-----------------|-----------------|
|                  | Males   | Females   | Females | Males   | Females   | Males   |
| RBC              | 4.83-5.85 | 4.52-5.31 | 3.94-5.22 | 4.63-6.08 | 4.63-6.08 |
| HB               | 14.50-16.88 | 13.541-15.38 | 11.2-15.7 | 13.7-17.5 | 13.7-17.5 |
| PLT              | 213.01-323.59 | 211.05-322.00 | 182-369 | 163-337 | 163-337 |
| NEUT             | 43.00-59.96 | 43.67-59.29 | 34-71.1 | 34-67.9 | 34-67.9 |
| LYM              | 30.19-46.05 | 31.56-45.66 | 19.3-51.7 | 21.8-53.1 | 21.8-53.1 |
| MON              | 6.36-9.85 | 6.21-9.63 | 4.7-12.5 | 5.3-12.2 | 5.3-12.2 |
| EOSI             | 0.49-3.07 | 0.50-3.01 | 0.7-5.8 | 0.8-7 | 0.8-7 |
| BASO             | 0.17-0.53 | 0.16-0.48 | 0.1-1.2 | 0.2-1.2 | 0.2-1.2 |

FIGURE 1: Comparison of Haematological Parameter Values

- Study participants’ mean haematological parameter values
- Manufactured Sysmex mean haematological parameter values

The findings of this study were similar to those obtained in Gondar Norwest of Ethiopia. With exception of Hb level, males’ haematological parameter values reported in the current study were slightly similar to values reported in a study conducted in Sudan. On the contrary, PLT and WBC values were higher in women, the difference was significant as compared to men just as reported in other studies, and men have high values of haemoglobin and red blood cells compared to females. The reason for these differences may be due to the variations in hormone types and concentrations in the different sexes and the effect of erythropoietin release in response to regular menstruation cross-stimulating megakaryopoiesis. However, the platelet counts are lower when compared to the US derived values and other African studies. The reason for these lower values is still unclear and may require additional studies but may be due to the diet, genetic factors or other environmental or genetic factors.
| Table 2: Haematological Reference Ranges for Individuals Attending Ruhengeri Hospital |
|---------------------------------|------------------|------------------|------------------|------------------|------------------|
|                                  | Children aged 0 to 17 years (N=193) | Adults aged 18 to 68 years (N=193) |
|                                  | 0 day to 7 day | 8 day to 5 months | 6 months to 2 yrs | 3 yrs to 6 yrs | 7 yrs to 17 yrs |
|                                  | (n=6) | (n=9) | (n=12) | (n=12) | (n=52) |
| WBC (n=6) | 2.38±0.95 | 2.35±0.94 | 2.37±0.93 | 2.34±0.92 | 2.36±0.94 |
| RBC (n=6) | 4.30±0.89 | 4.31±0.88 | 4.32±0.87 | 4.33±0.86 | 4.34±0.85 |
| HB (n=6)  | 13.2±2.4 | 13.3±2.5 | 13.4±2.6 | 13.5±2.7 | 13.6±2.8 |
| PLT (n=6) | 2.9±0.7 | 2.9±0.8 | 2.9±0.9 | 2.9±1.0 | 2.9±1.1 |
| NEUT (n=6) | 5.8±1.2 | 5.9±1.3 | 6.0±1.4 | 6.1±1.5 | 6.2±1.6 |
| LYMPH (n=6) | 3.9±1.0 | 4.0±1.1 | 4.1±1.2 | 4.2±1.3 | 4.3±1.4 |
| MON (n=6)  | 1.1±0.3 | 1.2±0.4 | 1.3±0.5 | 1.4±0.6 | 1.5±0.7 |
| EOSI (n=6) | 0.5±0.2 | 0.6±0.3 | 0.7±0.4 | 0.8±0.5 | 0.9±0.6 |
| BASO (n=6) | 0.2±0.1 | 0.3±0.2 | 0.4±0.3 | 0.5±0.4 | 0.6±0.5 |

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CONCLUSION
The findings may be used to define normal haematological values for Rwandan population and help physicians to better define haematological abnormalities in patients. The reference values obtained in this study are recommended to be used in the medical practice. Another study with representative sample may be conducted to determine effect of geographical location on haematological reference parameters in the population of Rwanda.

REFERENCES

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