ORIGINAL ARTICLE

HEMATOIMMUNOLOGICAL PROFILE AT GILGEL GIBE FIELD RESEARCH CENTER, SOUTHWEST ETHIOPIA

Abraham Haileamlak², Ayalew T. Muluneh¹, Fessahaye Alemseged¹, Fasil Tessema¹, Kifle Woldemichael¹, Makonnen Asefa¹, Yoseph Mamo³, Solomon Tamiru³, Gemeda Abebe⁴

ABSTRACT

BACKGROUND: Health and disease can only be distinguished by accurate and reliable reference values of a particular laboratory test. In interpreting laboratory test results, usually the reported values are compared with established reference values from developed countries. Now it is a fact that there is considerable variation in hematology reference intervals by several variables. However, such data at a population level are scanty in the Ethiopian situation. Therefore, this study was conducted to determine the hematological and immunological values in a community setting.

METHODS: A population-based cross-sectional study was conducted in Gilgel Gibe Field Research Center (GGFRC) from late September 2008 to end of January 2009. A sample of 1,965 individuals was included in the study. Blood sample was collected by vacutainer tube and transported to Jimma University Specialized Hospital laboratory. Data were entered into EpiData and analyzed using SPSS for Windows version 16.0 and STATA 11.

RESULTS: A total of 1965 (955 men and 1010 women) individuals were studied. The mean red blood cell count for men and women was 4.55 x 10¹²/L and 4.34 x 10¹²/L (95 percentile range between 2.9 and 5.7 x 10¹²/L) and 4.34 x 10¹²/L (95 percentile range between 2.8 and 5.2 x 10¹²/L), respectively. On the other hand, the red blood cell count of 95% of the men and women lied between 2.9-5.7 x 10¹² cells/L and 2.8-5.2 x 10¹² cells/L, respectively. The mean hemoglobin value for men was 13.6 gm/dl and for women 12.7 gm/dl. The mean corpuscular volume for men and women was 90.2 fl and 90.8 fl, respectively. The mean platelet value for men was 229.1 x 10⁹ cells/L and for women 241.3 x 10⁹ cells/L. The mean white blood cells count for men and women was 6.08 x 10⁹ cells/L and 6.12 x 10⁹ cells/L, respectively. The mean CD4 value was 809 cell/µl for men and 868 cell/µl for women. Forty two percent of the study participants had O blood group.

CONCLUSION: the hematologic and immunologic profile of the studied population in Southwest Ethiopia is different from the reports from other countries and the standards described in western literature. We recommend conducting similar nationwide study to determine the immunological and hematological reference values of the Ethiopian population as a whole.

KEYWORDS: Hematoimmunologic parameters, Southwest Ethiopia

INTRODUCTION

Lack of appropriate local reference values for hematoimmunological parameters are challenges in interpreting results for management of patients and other decision making. Health professionals usually use textbook reference values to compare the reported values. In addition to lack of evidence based practice, such values have substantial variability between subjects by age, sex, geographic, environment and genetic variation (1, 2).

¹Department of Epidemiology, College of Public Health and Medical Sciences, Jimma University
²Department of Pediatrics and Child Health, College of Public Health and Medical Sciences, Jimma University
³Department of Internal Medicine, College of Public Health and Medical Sciences, Jimma University
⁴Department of Medical Laboratory Sciences and Pathology, College of Public Health and Medical Sciences, Jimma University
Moreover, the laboratory method used to establish the reference values can lead to different results. Inappropriate reference values may increase the risk of either unnecessary additional investigations, failure to detect underlying disease or mismanagement of patients. Normality of values for a given individual are usually defined in terms of the range of results typically encountered from similar subjects who are known to be in good health.

Textbook reference values have been derived from a variety of samples (3-6). The reference values of hematological and immunological values currently used in Africa and Asia are borrowed from data collected from populations living in developed countries (7). Some studies conducted in Asian and African countries showed lower values compared to the established Western references as presented elsewhere (2, 8, 9-13). Some Asian studies also showed that there were considerable differences in the reference values by sex and among population groups (7, 8).

Even though very limited studies are available from developing countries, a multicenter study in Africa showed lower hematocrit (Hct) and hemoglobin (Hgb) levels, lower white blood cell and neutrophil counts (14). Studies from some parts of Africa showed differences in biomarkers profile (10, 12).

Lower CD₄ T-cell numbers have been reported in Asians and Ethiopians compared to Caucasians, although absolute CD₄ T-cell counts in Africans from the Central African Republic have been reported to be similar to Europeans (11). Studies from Uganda and Kenya showed CD₄ counts that differed with age and gender (9, 10).

The few studies conducted on African populations indicated differences in normal values compared with those from populations in developed countries (9, 10, 15, 16). These studies were conducted on certain segment of healthy workers and might not be representative of the normal population and thus lack generalizability.

As described above the ethnic origin, genetics, gender, geography, and environmental factors, may influence some values of hematological and immunological profiles suggesting that the development of reference values for the Ethiopian population is imperative to improve quality of health care. Such valuable data at a local population level are scanty in the Ethiopian situation.

Therefore, this study was conducted to determine the hematological and immunological values for general population in community settings. If validated with other studies, this can be used as reference values in the future evidence-based practices. Moreover, this study would serve as baseline information for further studies at national level.

SUBJECTS MATERIALS AND METHODS

Population-based cross-sectional survey was conducted from late September 2008 to end of January 2009 at Gilgel Gibe Field Research Center (GGFRC) of Jimma University (JU). GGFRC is located around Gilgel Gibe Hydroelectric Dam, 55 kilometers Northeast of Jimma Town on the way to Addis Ababa.

This study was part of the survey for determination of magnitude of chronic non-communicable diseases (CNCDs), risk factors of CNCDs and biochemical value determination for the community at GGFRC. Residents in the 10 kebeles of GGFRC with age range from 15 to 64 years of both sexes were included as per the WHO recommendation for CNCDs survey. It was planned to collect blood samples for hematological values measurement from 60% (3,300) of the total sample for CNCDs survey as per the recommendation of WHO (17). Individuals selected from the total sample by simple random sampling were asked to give blood sample. The study included all apparently healthy individuals; however, those who were disabled and acutely ill during the data collection were excluded. The sampling procedure is showed schematically in figure 1.

New formats were developed for recoding of hematological and immunological values. Two laboratory technicians and two nurses were identified and trained on blood sample collection, completing the recording format, labelling blood sample, storage and transportation of the sample to Jimma University Hospital (JUSH) laboratory. They were provided with manuals that covers the standard operating procedures of blood sample collections.

After completion of the interview, all respondents who were selected for hematoimmunological tests were given early
morning appointment. Whole blood sample was collected in the morning (8:00am to 12:00 noon) after cleaning the cubital area by 70% alcohol. Venous blood was drawn from each subject using vacutainer system and stored in vacutainer tube containing ethylenediamine tetraacetic acid (EDTA). Subsequently the blood was mixed with anticoagulant. The test tubes then were placed in ice-box and transported to the JUSH laboratory in the afternoon.

Six laboratory technicians trained on the purpose of the study, laboratory procedures and analysis, format completion and repository storage did the laboratory work within 12 hours of blood sample collection at Jimma University Hospital Laboratory.

Complete blood cell (CBC) count was performed using an automated hematology analyzer KX-21, (Sysmex Coporation, Germany). The analyzer was calibrated each day using commercial standards recommended by the manufacturer. The machine automatically dilutes a whole-blood sample, lyses, counts and gives a printout result of hematological values. The analyzed hematological values were Hgb, Hct, red blood cell (RBC) count, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), white blood cell (WBC) count (including neutrophils, mixed white cells and lymphocytes) and platelet count. Fluorescence-activated cell sorter (FACS) system (Becton Dickinson San Jose California, USA) was used to enumerate absolute values for CD4, CD8 and CD3 cells. The ratio of CD4 to CD8 count was computed.

ABO blood grouping was carried out using HEMA-CLONE monoclonal ABO blood grouping reagents while RH blood group was determined using Anti-D (RHO) IgM+IgG monoclonal blood grouping reagents. Blood groupings were done using slide methods where blood grouping reagents were placed on a test slide followed by a drop of blood from EDTA tube. The reagents and the blood were mixed using clean stick and spread by moving gently the test slide back and forth and checked for agglutination within 2 minutes.

Supervisors checked each completed format and rechecked during data entry. Data entry personnel coded and entered the data using a double entry method.

Data were entered using Epidata and analyzed using SPSS for Windows version 16.0 and STATA 11. Background of study participants and hematoimmunological summary values with measures of dispersion were determined when appropriate.

Ethical clearance was obtained from Jimma University’s Research and Publication Office. Signed informed consent was obtained from study participants before blood sample collection. Detailed information on methods is described in article 1 of this special issue.

RESULTS

This study presented distribution of hematological and immunological profiles of 1965 (955 men and 1010 women) randomly selected individuals from a community setting.

The mean RBC count (95% CI) for men and women was 4.55 X 10^{12} cells/L (4.51-4.59) and 4.34 X 10^{12} cells/L (4.22-4.29), respectively. The lowest RBC mean value was observed in age group 55 years and above in both men (4.42 X 10^{12} cells/L) and women (4.21 X 10^{12} cells/L). The median value of RBC count for different age strata and sex is similar with the mean but with wider range. The RBC count of 95% of the men and women lies between 2.9-5.7 X 10^{12} cells/L and 2.8-5.2 X 10^{12} cells/L, respectively. The corresponding values for women were 12.7 gm/dl (12.6-12.8) and 40.9% (40.6-41.3). The mean Hgb and Hct value (95% CI) for men were 13.6 gm/dl (12.6-12.8) and 40.9% (40.6-41.3). The mean (95% CI) MCV for men and women was 90.2 fl (89.7-90.7) and 90.8 fl (90.0-91.7), respectively with the highest values in those age group 55 years and above for both sexes. The mean (95% CI) MCHC for men and women was 33.1 Pg/cell (32.9-33.4) and 32.9 Pg/cell (32.7-33.2), respectively while the MCH was the same 30.1 gm/dl (29.8-30.4) for both sexes. The median value for MCV, MCH and MCHC was similar with mean value with slight differences. However, the 95th percentile range is wide (Table 2).
| Age group | Sex          | RBC ($10^{12}$ cells/L) | Hemoglobin (gm/dl) | Hematocrit (%) |
|-----------|--------------|--------------------------|-------------------|----------------|
|           |              | Mean [95% CI]            | Median [95% CI]   | Mean [95% CI] |
|           |              | Median [95% percentile range] | Median [95% percentile range] | Median [95% percentile range] |
| 15-24     | Men (n=152)  | 4.58 (4.44-4.73)         | 4.8 (2.5-5.7)     | 13.8 (13.3-14.2) | 41.0 (39.8-42.3) |
|           | Women (n=155)| 4.35 (4.23-4.46)         | 4.4 (2.6-5.4)     | 13.0 (12.5-13.5) | 38.5 (37.5-39.5) |
| 25-34     | Men (n=172)  | 4.61 (4.50-4.73)         | 4.8 (3.0-5.7)     | 13.8 (13.4-14.1) | 41.2 (40.2-42.1) |
|           | Women (n=231)| 4.24 (4.16-4.32)         | 4.3 (3.1-5.2)     | 12.7 (12.4-12.9) | 38.4 (37.7-39.2) |
| 35-44     | Men (n=199)  | 4.59 (4.48-4.69)         | 4.7 (2.8-5.7)     | 14.1 (13.4-14.8) | 41.1 (40.2-42.1) |
|           | Women (n=211)| 4.24 (4.16-4.33)         | 4.2 (2.7-5.3)     | 12.8 (12.3-12.8) | 38.1 (37.3-38.9) |
| 45-54     | Men (n=201)  | 4.57 (4.47-4.67)         | 4.6 (2.4-5.7)     | 13.7 (13.5-14.0) | 41.2 (40.4-42.0) |
|           | Women (n=202)| 4.28 (4.19-4.37)         | 4.4 (2.8-5.3)     | 12.9 (12.6-13.1) | 38.9 (38.1-39.7) |
| 55+       | Men (n=331)  | 4.42 (4.33-4.50)         | 4.4 (3.0-5.4)     | 13.3 (13.0-13.5) | 40.3 (39.3-41.3) |
|           | Women (n=211)| 4.21 (4.12-4.30)         | 4.3 (2.5-5.1)     | 12.7 (12.4-12.9) | 38.6 (37.8-39.4) |
| Total     | Men (n=955)  | 4.55 (4.51-4.59)         | 4.6 (2.9-5.7)     | 13.6 (13.5-13.8) | 40.9 (40.6-41.3) |
|           | Women (n=1010)| 4.34 (4.22-4.29)        | 4.3 (2.8-5.2)     | 12.7 (12.6-12.8) | 38.5 (38.2-38.8) |
| Both sexes|              | 4.40 (4.37-4.43)         | 4.44 (2.8-5.5)    | 13.1 (13.0-13.2) | 39.7 (39.4-39.9) |

RBC= red blood cell
Table 2: Distribution mean value with 95% CI and median with 95 percentile range of RBC indices, GGFRC, Sept 2008-Jan 2009.

| Age group | Sex          | MCV (fl)         | MCH (gm/dl)        | MCHC (Pg/cell)     |
|-----------|--------------|------------------|--------------------|--------------------|
|           |              | Mean [95% CI]    | Median [95 percentile range] | Mean [95% CI]    | Median [95 percentile range] | Mean [95% CI]    | Median [95 percentile range] |
| 15-24     | Men (n=152)  | 89.4 (87.7-91.0) | 89.8 (70.1-102.3)  | 30.2 (29.4-31.0)   | 29.9 (20.9-6.6)   | 33.6 (32.9-34.2)   | 32.9 (28.5-40.4)   |
|           | Women (n=155)| 89.4 (88.1-90.7) | 89.8 (72.4-102.1)  | 30.2 (28.9-31.5)   | 30.0 (20.0-45.8)  | 33.2 (32.4-34.1)   | 32.8 (27.1-39.1)  |
| 25-34     | Men (n=172)  | 89.8 (88.5-91.2) | 90.9 (68.3-103.6)  | 30.0 (29.4-30.7)   | 30.2 (20.1-34.8)  | 33.5 (32.9-34.0)   | 32.8 (26.0-39.5)  |
|           | Women (n=231)| 90.5 (89.2-91.8) | 91.2 (75.0-100.6)  | 30.1 (29.4-30.6)   | 30.0 (22.5-39.4)  | 33.0 (32.7-33.4)   | 32.5 (28.1-40.4)  |
| 35-44     | Men (n=199)  | 90.6 (89.4-91.8) | 91.4 (71.5-105.6)  | 30.6 (29.6-31.6)   | 30.4 (20.8-37.5)  | 33.5 (32.8-34.1)   | 32.8 (27.8-40.1)  |
|           | Women (n=211)| 90.0 (88.7-91.3) | 91.7 (71.2-102.7)  | 30.1 (29.3-30.9)   | 30.0 (20.4-40.6)  | 33.0 (32.5-33.4)   | 32.4 (28.1-39.4)  |
| 45-54     | Men (n=201)  | 90.0 (88.6-91.3) | 92.0 (65.5-111.9)  | 30.3 (29.7-31.0)   | 30.0 (18.8-35.5)  | 33.5 (32.9-34.0)   | 32.8 (27.5-39.6)  |
|           | Women (n=202)| 89.9 (88.4-91.4) | 91.3 (68.6-104.0)  | 30.4 (29.6-31.2)   | 30.0 (23.6-40.9)  | 33.4 (32.8-34.0)   | 32.4 (29.4-40.1)  |
| 55+       | Men (n=331)  | 91.1 (90.1-92.0) | 91.9 (68.5-102.0)  | 30.3 (29.8-30.7)   | 30.5 (20.4-36.6)  | 32.9 (32.5-33.3)   | 32.7 (27.2-39.1)  |
|           | Women (n=211)| 94.0 (90.0-98.0) | 92.5 (77.6-103.6)  | 30.6 (29.8-31.1)   | 30.0 (23.7-40.1)  | 33.0 (32.2-33.7)   | 32.3 (28.5-39.6)  |
| Total     | Men (n=955)  | 90.2 (89.7-90.7) | 91.2 (68.9-102.1)  | 30.1 (29.8-30.4)   | 30.2 (20.5-36.1)  | 33.1 (32.9-33.4)   | 32.8 (27.6-39.6)  |
|           | Women (n=1010)| 90.8 (90.0-91.7) | 91.3 (73.0-102.3)  | 30.1 (29.8-30.4)   | 30.0 (21.9-38.9)  | 32.9 (32.7-33.2)   | 32.4 (28.4-39.8)  |
| Both sexes|              | 90.5 (90.0-91.0) | 91.2 (71.6 - 102.0)| 30.1 (29.9-30.3)   | 30.0 (20.8 - 39.6)| 33.0 (32.9-33.2)   | 32.6 (28.1 - 39.6)|

MCV = mean corpuscular volume, MCHC = mean corpuscular hemoglobin concentration, MCH = mean corpuscular Hemoglobin. Pg=Picograms
The mean platelet value for the different age strata ranged from 221-235 x10^9/L for men and 220-260 x10^9/L for women with mean (95% CI) of 229.1 x10^9 cells/L (223.6-234.7) and 241.3 x10^9 cells/L (236.0-246.5), respectively. The median platelet value for all men and women were 219 x10^9 cells/L and 233 x10^9 cells/L where the values for 95% of the subjects lied between 91-429 x10^9 cells/L and 101-437 x10^9 cells/L, respectively. The mean (95% CI) WBC count for men and women was 6.08 x10^9 cells/L (5.95-6.21) and 6.12 x10^9 cells/L (5.97-6.27), respectively. The mean WBC value for the different age strata for men ranged from 5.95-6.51 x10^9 cells/L and for women from 5.88-6.31 x10^9 cells/L. WBC count of 95% of all men was between 3.1-10.5 x10^9 cells/L and that of women 3.1-10.8 x10^9 cells/L. The mean (95% CI) neutrophil, lymphocyte and mixed white cells proportion for all age was 48.9% (48.1-49.7), 35.7% (35.1-36.4) and 15.8% (15.2-16.4) and for women 49.1% (48.1-49.8), 35.4% (34.8-36.0) and 16.0% (15.4-16.6), respectively (Table 3).

The mean (95% CI) value for CD3, CD4 and CD8 T cells were 1743 (1689-1797) cell/µl, 809 cell/µl (787-832) and 812 (775-848) cell/µl for men and 1774 (1719-1830) cell/µl, 868 cell/µl (840-895) and 818 (783-853) cell/µl for women, respectively. The lowest mean CD4 value for men, 757 (711-803) cell/µl was observed in those age 55 years and above while for women, 810 (749-872) cell/µl was in those age 45-54 years. Except for the age stratum 45-54 years, the mean CD4 count is higher for women. The median values of CD3, CD4 and CD8 cells count were lower than the mean. The CD4 count of 95% of the men lied between 361-1,404 cell/µl and women 400-1,553 cell/µl (Table 4).

ABO and Rhesus (RhD) blood group was determined for 1965 study subjects. Accordingly; 42%, 31%, 21% and 6% had O, A, B and AB blood group, respectively. Fifty seven (2.9%) subjects of them were RhD negative (Fig 1). Two percent of those with blood group A, 6.5% of AB, 3.3% of B and 3% of O were RhD negative (Fig 2).

**DISCUSSION**

Hematologic and immunologic reference values are essential for evidence based practice. Many studies all over the world showed variability of reference values. Though few studies tried to show hematologic and immunologic profiles on segments of Ethiopian population, to the best of the authors’ knowledge, the present study is more comprehensive in terms of the quantity and type of hematoimmunologic parameters included and the size of the population studied.

The mean and median RBC, Hgb and Hct values at all age strata were higher for men than women. Such difference by sex is in line with the findings of other previous studies from different parts of the world (2, 8, 10, 13, 14, 18-21). However, one previous study from north-western Ethiopia reported high Hct in women (22). Slight differences of mean RBC, Hgb and Hct values by age groups within same sex are observed with no specific pattern. This study revealed that the mean values of MCV, MCHC and MCH by sex and age...
### Table 3: Distribution mean value with 95% CI and median with 95 percentile range of WBC and platelet count and mean value with 95% CI differential count, GGFRC, Sept 2008-Jan 2009.

| Age group | Sex   | Platelet ($x10^9$ cells/L) | WBC count ($x10^9$ cells/L) | Neutrophil (%) | Mixed (%) | Lymphocyte (%) |
|-----------|-------|-----------------------------|-----------------------------|----------------|-----------|----------------|
|           |       | Mean [95% CI]                | Median [95 percentile range] | Mean [95% CI] | Median [95 percentile range] | Mean [95% CI] | Mean [95% CI] | Mean [95% CI] |
| 15-24     | Men   | 223.85 (208.36-239.35)       | 211.0 (78.0-425.2)          | 6.03 (5.67-6.40) | 5.7 (3.1-11.2) | 46.9 (44.8-49.0) | 16.5 (15.1-18.0) | 36.6 (34.7-38.5) |
|           | Women | 220.44 (207.71-233.17)       | 219.0 (102.7-437.4)         | 6.21 (5.86-6.57) | 5.9 (3.1-11.0) | 50.4 (48.4-52.4) | 15.1 (14.2-16.9) | 34.3 (32.8-35.8) |
| 25-34     | Men   | 235.76 (218.78-252.75)       | 216.0 (92.3-428.8)          | 6.10 (5.75-6.44) | 5.6 (3.2-11.0) | 48.5 (46.5-50.4) | 16.2 (14.9-17.4) | 35.5 (33.7-37.3) |
|           | Women | 233.17 (219.90-246.44)       | 229.0 (103.8-450.3)         | 6.31 (6.02-6.60) | 6.0 (3.2-10.9) | 49.2 (47.5-50.9) | 16.2 (15.0-17.3) | 34.8 (33.4-36.2) |
| 35-44     | Men   | 221.01 (207.78-234.23)       | 216.0 (81.0-412.0)          | 5.95 (5.61-6.29) | 5.6 (3.1-10.5) | 49.2 (47.3-51.0) | 15.0 (13.9-16.2) | 35.6 (34.1-37.2) |
|           | Women | 239.03 (225.16-252.91)       | 230.0 (83.8-435.9)          | 6.05 (5.74-6.36) | 5.7 (3.1-11.0) | 48.3 (46.3-50.2) | 16.7 (15.4-18.1) | 34.9 (33.4-36.5) |
| 45-54     | Men   | 233.14 (220.40-245.88)       | 227.0 (106.0-406.9)         | 6.11 (5.83-6.39) | 5.7 (3.1-9.8) | 47.9 (45.9-49.9) | 15.8 (14.6-17.1) | 36.3 (34.8-37.8) |
|           | Women | 260.52 (247.14-273.90)       | 250.0 (127.3-458.0)         | 5.88 (5.60-6.16) | 5.8 (3.1-10.2) | 49.1 (47.2-51.0) | 15.5 (14.1-16.9) | 35.3 (33.8-36.9) |
| 55+       | Men   | 235.56 (222.69-248.44)       | 224.0 (92.4-483.4)          | 6.51 (6.17-6.84) | 6.2 (2.8-10.9) | 51.0 (49.3-52.8) | 15.6 (14.4-16.8) | 33.6 (32.3-35.0) |
|           | Women | 243.93 (231.06-256.81)       | 233.0 (119.5-439.4)         | 5.98 (5.67-6.29) | 5.7 (3.0-11.2) | 47.9 (46.3-49.5) | 16.2 (15.1-17.3) | 36.0 (34.6-37.4) |
| Total     | Men   | 229.14 (223.57-234.71)       | 219.0 (91.0-429.0)          | 6.08 (5.95-6.21) | 5.7 (3.1-10.5) | 48.9 (48.1-49.8) | 15.8 (15.2-16.4) | 35.7 (35.1-36.4) |
|           | Women | 241.27 (236.01-246.54)       | 233.0 (101.3-437.4)         | 6.12 (5.97-6.27) | 5.8 (3.1-10.8) | 49.0 (48.1-49.8) | 16.0 (15.4-16.6) | 35.4 (34.8-36.0) |
| Both sexes|       | 235.44 (231.67-239.22)       | 227.0 (93.0-435.0)          | 6.09 (6.00-6.19) | 5.8 (3.1-10.5) | 48.9 (48.3-49.5) | 16.0 (15.6-16.4) | 35.5 (35.1-36.0) |

WBC = white blood cell
### Table 4: Distribution mean value with 95% CI and median with 95 percentile range of CD3, CD4 and CD8, GGFRC, Sept 2008-Jan 2009.

| Age group | Sex       | CD3 (cells/µl) | CD4 (cells/µl) | CD8 (cells/µl) |
|-----------|-----------|----------------|----------------|---------------|
|           |           | Mean [95% CI]  | Median (95 percentile range) | Mean [95% CI]  | Median (95 percentile range) | Mean [95% CI]  | Median (95 percentile range) |
| 15-24     | Men       | 1891 (1754-2028) | 1,782 (969-3,500) | 848 (788 907) | 797 (387-1,485) | 876 (786-967) | 736 (273-2,000) |
|           | Women     | 1789 (1658-1920) | 1,741 (777-3,423) | 904 (817-990) | 805 (363-2,023) | 779 (706-853) | 760 (271-1,961) |
| 25-34     | Men       | 1679 (1577-1780) | 1,624 (724-2,976) | 780 (735-826) | 783 (307-1,306) | 754 (687-820) | 696 (242-1,510) |
|           | Women     | 1816 (1698-1934) | 1,729 (781-3,396) | 921 (871-970) | 896 (475-1,564) | 805 (732-879) | 713 (259-2,000) |
| 35-44     | Men       | 1751 (1625-1877) | 1,705 (573-3,500) | 810 (761-858) | 800 (302-1,418) | 818 (735-901) | 706 (237-2,000) |
|           | Women     | 1753 (1627-1890) | 1,633 (781-3,500) | 845 (791-899) | 821 (380-1,649) | 814 (731-897) | 697 (231-2,000) |
| 45-54     | Men       | 1841 (1699-1983) | 1,712 (817-3,500) | 841 (781-899) | 853 (372-1,509) | 894 (790-998) | 741 (280-2,000) |
|           | Women     | 1714 (1583-1845) | 1,629 (644-3,445) | 810 (749-872) | 775 (320-1,566) | 819 (735-903) | 728 (256-2,000) |
| 55+       | Men       | 1575 (1471-1680) | 1,542 (765-3,032) | 757 (711-803) | 741 (387-1,367) | 734 (665-803) | 653 (228-1,784) |
|           | Women     | 1762 (1626-1898) | 1,723 (754-3,446) | 843 (773-913) | 774 (394-1,551) | 860 (770-949) | 790 (230-2,000) |
| Total     | Men       | 1743 (1689-1797) | 1,643 (760-3,465) | 809 (787-832) | 792 (361-1,404) | 812 (775-848) | 706 (258-2,000) |
|           | Women     | 1774 (1719-1830) | 1,685 (762-3,420) | 868 (840-895) | 823 (400-1,553) | 818 (783-853) | 728 (250-2,000) |
| Both sexes|           | 1754 (1715-1791) | 1,662 (763-3,421) | 836 (819-854) | 800 (380-1468) | 813 (788-838) | 716 (255-2000) |
strata had no big difference which is in agreement with the American study (21), however, the Kenyan (10), Malaysian (8) and Turkish (18) studies showed higher values in men than women. The mean RBC, hemoglobin and Hct values in the present study are higher than the findings from Kenya (10). However, the values of the current study (done at an average altitude of 1700 m) are lower than the findings of Tsegaye et al in Ethiopia which was done at an altitude of about 2200 meters above sea level (15) and the standards described in another literature (21). The lower values in the current study as compared to the previous Ethiopian study (15) could be explained by the fact that the altitude of current study setting is low by 500 meter. Despite the fact that malaria is prevalent around Gilgel Gibe (23, 24), the lower hemoglobin level in our study population might not be attributable to malaria as the MCV is high which could in turn be affected by other factors like parasitic infestations and micronutrient deficiencies. The mean value for MCV of this study is similar with the Australian finding (19) but higher compared to other reports (8, 18, 20). The MCH and MCHC mean values of this study are similar to the American and Turkish findings (18, 20), however, higher than the Malaysian report (8).

Slight differences of the mean WBC, differential and platelet count by age and sex are observed in this study without specific trend. Such minor differences by sex are seen on previous reports from different parts of the world (8, 13, 14, 19). However, the mean WBC and platelet count in men was lower than mean value of women (2, 10, 18). The WBC count in our study is comparable with findings of another study done in Ethiopia (25) but lower than the standard described in the textbook (4). However, the WBC count in the present study is higher than the findings of other studies in other parts of Africa (9, 10, 26). The proportion of neutrophil is similar with report on Eastern and Southern African population (14) but, is low compared to the Malaysian report (8).

Lower CD4 T-cell numbers have been reported in Ethiopians and Botswana (15, 25, 26) compared to Caucasians, although absolute CD4 T-cell counts in Africans from the Central African Republic have been reported to be similar to Europeans (11). A study from Uganda showed CD4 counts that differed with age and gender (9). The mean values of this study for CD3, CD4 and CD8 T cell has no remarkable differences by age strata, however, the mean CD4 value is higher for women. The mean values of CD3, CD4 and CD8 T cells in the present study are higher compared to other studies in Kenya (10), Uganda (21), Ethiopia (25) and Tanzania (27). However, the reference range of CD4 and CD8 lymphocyte in this study are far below the finding of Klose et al in Burkina Faso (28) and the western standards (21). The lower reference range of CD4 and CD8 lymphocyte counts in our setting as compared to the international standards suggests researchers and policy makers to use locally available reference value to monitor disease progress such as HIV/AIDS and to assess the health status of the population during vaccine and drug trial. In this study, we observed that females have a higher CD4, CD8 lymphocyte counts and CD4/CD8 ratio. Similar findings have been reported in different settings (9, 10, 25). We observed an important change is the progressive decline of the mean values of CD4 and CD8 lymphocyte counts as the age increases. These findings are in line with observations from similar investigations carried out in other parts of the world (29-31).

Comparison of hematol immunological values of the current study findings with other studies from different parts of the world is presented in table 5.

The ABO blood group distribution varies in different geographical and ethnic groups. Nevertheless, in almost all, O positive blood group is the predominant followed by A blood group while AB is the rarest which is in line with the finding of this study. In Caucasians in the United States, the distribution is type O, 47%; type A, 41%; type B, 9%; and type AB, 3%. Among African Americans, the distribution is type O, 46%; type A, 27%; type B, 20%; and type AB; 7%. Among Western Europeans, 42% have group A, 9% group B, 3% group AB and the remaining 46% group O (32-37). RhD distribution also varies worldwide. RhD negative blood group is documented as 5.5% in south India, 5% in Nairobi, 4.8% in Nigeria, 7.3% in Lahore, 7.7% in Rawalpindi. About 95% of African – Americans are Rh-positive whereas indigenous Africans are virtually 100% Rh-positive (32-37). In this study 2.9% of the subjects were rhesus D negative.
In conclusion, this study has established baseline values for haematological and immunological parameters in “apparently healthy” Ethiopians. The hematologic and immunologic parameters of the studied population in Southwest Ethiopia are different from the reports from other countries and the standards described in western literature. Monitoring of the health status of the population in the study areas should be based on the local hematologic and immunologic values. We recommend conducting similar nationwide study to determine the hematological and immunological reference values of the Ethiopian population as a whole.

Table 5: Immune-hematological values comparative table for showing the currents study findings, textbook references and some other countries, Sept 2008-Jan 2009.

| Profile         | Current study | Text book References | Uganda (9)† | Malaysia | Turkey (20) |
|-----------------|---------------|-----------------------|-------------|----------|-------------|
| **Men**         |               |                       |             |          |             |
| RBC (10^12/L)   | 4.51-4.59     | 4.5-5.9               | 5.1 (4.1–6.0) | 5.12 (5.04–5.21) | 5.12 ± 0.4 |
| Hct %           | 40.6-41.3     | 41-53                 | 43.62 (43.05–44.19) | 45.0 ± 3.9 |
| MCV (fl)        | 89.7-90.7     | 78-100                | 254.90 (246.73-263.08) | 235 ± 52 |
| Platelet (10^9/L) | 223.57-234.71 | 150-350               | 6.74 (6.46–7.01) | 7.8 ± 2 |
| WBC (10^9/L)    | 5.95-6.21     | 4.5-11                | 6.02 (54.73–57.31) | 33.09 (31.72–34.46) |
| Neutrophil %    | 48.1-49.8     | 40-70                 | 56.02 (54.73–57.31) | 33.09 (31.72–34.46) |
| Lymphocyte %    | 35.1-36.4     | 22-44                 | 33.09 (31.72–34.46) | 33.09 (31.72–34.46) |
| CD4 (cells/µl)  | 787-832       | -                     | -            | -        |             |
| **Women**       |               |                       |             |          |             |
| RBC (10^12/L)   | 4.22-4.29x10^12/L | 4.0-5.2x10^12/L        | 4.34 (4.25–4.42) | 4.64 ± 0.4 |
| Hct %           | 38.2-38.8%    | 36-46                 | 37.08 (36.51–37.65) | 40.0 ± 3.8 |
| MCV             | 90.0-90.7fl   | 78-102fl              | 85.99 (85.19–86.78) | 86.7 ± 4.6 |
| Platelet (10^9/L) | 236.01-246.54 | 150-350               | 275.24 (261.64-288.85) | 253 ± 56 |
| WBC (10^9/L)    | 5.97-6.27x10^9/L | 4.5-11x10^9/L         | 6.73 (6.36–7.09) | 7.46 ± 2 |
| Neutrophil %    | 48.1-49.8     | 40-70                 | 56.88 (55.34–58.41) | 33.28 (31.75–34.82) |
| Lymphocyte %    | 34.8-36       | 22-44                 | 33.28 (31.75–34.82) | 33.28 (31.75–34.82) |
| CD4 (cells/ml)  | 480-895       | -                     | -            | -        |             |

† mean and 90% reference interval

REFERENCES

1. Soldberg HE. Approved recommendation (1986) on the theory of reference values, part 1. The concept of reference values. *Clin Chim Acta*, 1987;165:111-118.
2. Khalid Usman, Zeeshan Ali Syed, Arif Ashfaq Rao. Reference Range Values of Haematological Parameters in Healthy Pakistani Adults. *Pak J Physiol*, 2007;3(1):19-22.
3. Bain B. Normal ranges. In: Blood cells: a practical guide, 2nd ed. London: Blackwell Science Ltd., 1995:148–52.
4. Lipschitz D, Mitchell C, Thompson C. The anemia of senescence. *Am J Hematol*, 1981; 11:51-54.
5. Fielding J, Karabus C, Brunstrom G. Storage iron depletion in male blood donors: its significance for iron status in women. *J Clin Pathol*, 1968;21(3):402-405.
6. Kalchthaler T, Tan ME. Anemia in institutionalized elderly patients. *J Am Geriatr Soc*, 1980;28:108–13.
7. Shaper AG, P. Lewis. Genetic neutropenia in people of African origin. *Lancet*, 1971;2(7732):1021-1023.
8. Roshan TM, Rosline H, Ahmed SA, Rapiaah M, Wan Zaidah A, Khattak MN. Hematological reference values of healthy Malaysian population. *Int J Lab Hematol*, 2009; 31: 505-512.
9. Lugada ES, Mermin J, Kaharuza F, *et al.* Population-Based Hematologic and Immunologic Reference Values for a Healthy
Ugandan Population. Clin Diagn Lab Immunol, 2004;11(1):29-34.
10. Kibaya RS, Bautista CT, Sawe FK, et al. Reference Ranges for the Clinical Laboratory Derived from a Rural Population in Kericho, Kenya. PLoS One, 2008; 3 (e 10): e3327.
11. Gill GV, England A, Marshal C. Low platelet counts in Zambians. Trans R Soc Trop Med Hyg. 1979;73:111–112.
12. Saathoff E, Schneider P, Kleinfeldt V, Geis S, Haule D, et al. Laboratory reference values for healthy adults from southern Tanzania. Trop Med Int Health, 2008;13:612-625.
13. Koram KA, Addae MM, Ocran JC, Adumankwah S, Rogers WO, Nkrumah FK. Population Based Reference Intervals for Common Blood Hematological and Biochemical Parameters in the Akuapem North District. Ghana Medical Journal, 2007;41(4):160-166.
14. Karita E, Ketter N, Price MA, et al. CLSI-Derived Hematology and Biochemistry Reference Intervals for Healthy Adults in Eastern and Southern Africa. PloS one, 2009;4(2): e4401.
15. Tsegaye A, Messele T, Tilahun T, et al. Immunohematological reference ranges for adult Ethiopians. Clin Diagn Lab Immunol, 1999;6:410–414.
16. Tugume SB, Piwowar EM, Lutalo T, et al. Hematological reference ranges among healthy Ugandans. Clin Diagn Lab Immunol, 1995;2:233–235.
17. World Health Organization. Chronic diseases and health promotion. STEPwise approach to surveillance (STEPS). STEPS Manual. Available at: http://www.who.int/chp/steps.
18. Kaya H, Kyky Y, Akarsu E, Gundoddu M, BaPol Tekyen SB, Ýnandi T. Hematological Values of Healthy Adult Population Living at Moderate Altitude (1869 m, Erzurum, Turkey). Turk J Haematol, 2000;17(3):123-128.
19. Tsang CW, Lazarus R, Smith W, Mitchell P, Koutts J, Burnett L. Hematological indices in an older population sample: derivation of healthy reference values. Clinical Chemistry, 1998;44:196–101.
20. Viteriv FE, De Tunaan V, Guzman MA. Normal Haematological Values in Population the Central American. Br J of Hematol, 1972; 23: 189-203.
21. Wintrobe MM, Lee GR. Wintrobe’s clinical hematology, 10th ed. The Williams & Wilkins Co, Baltimore, Md. 1999
22. Zein AZ, Makonnen A. The prevalence of Anemia among Populations Living at Different Altitudes in North-Western Ethiopia. Ethiop Med J, 1987;25:105-111.
23. Yewhalaw D, Legesse W, Van Bortel W, et al: Malaria and water resource development: the case of Gilgel-Gibe hydroelectric dam in Ethiopia. Malar J, 2009; 8:21.
24. Deribew A, Alemsged F, Zewudie B, et al. Effect of training on the use of long-lasting insecticide-treated bed nets on the burden of malaria among vulnerable groups, south-west Ethiopia: baseline results of a cluster randomized trial. Malaria Journal, 2010; 9:121.
25. Kassu A, Tsegaye A, Petros B, et al. Distribution of Lymphocyte Subsets in Healthy Human Immunodeficiency Virus-Negative Adult Ethiopians from Two Geographic Locales. Clinical and Diagnostic Laboratory Immunology, 2001; 8(1), 1171-6.
26. Bussmann H, Wester CW, Kereng V, et al. Low CD4 T-Lymphocyte Values in Human Immunodeficiency Virus-Negative Adults in Botswana. Clinical and Diagnostic Laboratory Immunology, 2004; 11(5) : 930–935.
27. Ngowi BI, Mfinanga SG, Bruun JN, Morkve O. Immunohematological reference values in human immunodeficiency virus-negative adolescent and adults in rural northern Tanzania. BMC Infectious Diseases, 2009, 9:1 doi:10.1186/1471-2334-9-1.
28. Klose N, Coulibaly B, Tebit DM, et al. Immunohematological Reference Values for Healthy Adults in Burkina Faso. Clinical and Vaccine Immunology, 2007;14(6):782-4.
29. Swaminathan S, Elizabeth L, Raja A, Sankaran K, Arunkumar N. Age related change in lymphocyte subsets in South Indian Children. Natl Med J India, 2003;16:249–52.
30. Robinson M, O’Donohoe J, Dadian G, Wankowicz A, Barltop D, Hobbs JR. An analysis of the normal ranges of lymphocyte subpopulations in children aged 5–13 years. Eur J Pediatr 1996;155:535–9.
31. Comans-Bitter WM, de Groot R, van den Beemd R, et al. Immunophenotyping of blood lymphocytes in childhood. Reference values for lymphocyte subpopulations. *J Pediatr* 1997;130:388–93.

32. Odokuma EI, Okolo AC, Aloamaka PC. Distribution of ABO and Rhesus Blood Groups in Abraka, Delta State. *Nigerian Journal of Physiological Sciences*, 2007; 22(1-2): 89-91.

33. Chavhan A, Pawar S, Baig MM. Allelic Frequency of ABO And Rh D Blood Group Among the Banjara Backward Caste of Yavatmal District, Maharashtra, India. *Nature Precedings*: hdl:10101/npre.2010.5482.1: Posted 31 Dec 2010.

34. Pramanik T, Pramanik S. Distribution of ABO and Rh blood groups in Nepalese students: a report. *Eastern Mediterranean Health J*. 2000;6(1):156-158.

35. Mawuagi J. Blood group distribution in an urban population of patient targeted blood donors. *East Afr Med J*, 1999;76(11):615-618.

36. Bhalti FA, Amin. Spectrum of ABO and D blood groups of donors at Rawalpindi/Islamabad. *Pakistan J Pathol*, 1996;7(2):26-28.

37. Omotade OO, Adeyemo AA, Kayode CM, Falade SL, Ikpeme S. Gene frequencies of ABO and Rh (D) blood group alleles in a healthy infant population in Ibadan, Nigeria. *West Afr J Med*, 1999;18:294-297.

38. Kasper DL, Fauci AS, Longo DL, (eds). In: Harrisons Principles of Internal Medicine. 16th edition, McGraw-Hill Companies, Inc. 2005.