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ESTABLISHMENT OF HEMATOLOGICAL REFERENCE VALUES FOR YOUNG ADULTS IN BANGANGTE, CAMEROON: PRELIMINARY FINDINGS FROM A PILOT STUDY TOWARDS HEALTHCARE QUALITY IMPROVEMENT

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ABSTRACT

Introduction: Blood count is an essential routine examination in biological diagnosis. The interpretation of its results involves the comparison of values obtained with values or ranges of references usually specific to the Caucasian population. The aim of this study was to establish reference values specific to young adults in the city of Bangangté (Western Region, Cameroon). **Materials and methods:** This study was a cross sectional descriptive study carried out at the Cliniques Universitaires des Montagnes between December 2017 and May 2018. We recruited 312 young, healthy adults made up of 131 men and 181 women between the ages of 15 and 29 living in the city of Bangangté. After signing a consent form, all participants were voluntarily allowed to carry out clinical, radiological and biomedical examinations analysis to ensure that they are healthy. After selection, we performed the blood count with hematology automat (BC-2800). **Results and Discussion**: The results from this analysis allowed us to establish the reference values of the blood count parameters, which include: white blood cells between $3.5x10^9$-$9.7x10^9$ / L (women) and $3.3x10^9$-$8.5x10^9$ / L (men), red blood cells between $3.5x10^{12}$-$6x10^{12}$ / L (women) and $4.3x10^{12}$-$6.9x10^{12}$ / L (men), and platelets between $130x10^9$ - $464x10^9$ / L (women) and $165x10^9$-$412x10^9$ / L (men). The Comparison of these values with the values described elsewhere has revealed the existence of several dissimilarities. **Conclusion**: This preliminary study permitted us to establish the references values of the blood count for young adults in the city of Bangangté and to identify potential differences between results obtained in this study and result obtained in other setting. Hence the need to establish our own reference intervals through wider studies and in different regions of Cameroon.

KEYWORDS: Reference values, young adults, Bangangté.

INTRODUCTION

Hematology which is a branch of medicine concerned with the study of the cause, prognosis, treatment, and prevention of diseases related to blood is based on comparisons of the patient values with reference values. Reference values play an important role in patient diagnosis, management, disease prognosis, monitoring of response to therapy and in monitoring possible adverse reaction to therapy.[1-3]

Nowadays, the reference values of hematological indices currently used in many Africa countries have been derived from studies on Caucasian populations and may not be applicable in most local settings in non-Caucasian populations.[4] This is because several factors affect the measurements determined in different populations such as age, gender, ethnicity and environment including altitude and climate.[4] Moreover, many constitutional hemoglobin abnormalities (Thalassemia, sickle cell disease and other hemoglobinopathies) or pathologic conditions (malaria, HIV, HBV, HCV and other viral infections), influence hematological values.[5] Recently, several authors in Africa tried to establish reference values in hematology for African countries, however there are some discrepancies from one study to another which may be related to different factors (age, gender, ethnicity and environment including altitude and climate).

Inappropriate reference values can cause unnecessary follow up investigations, treatment and mismanagement of patient.[4] The Clinical and Laboratory standards

institute (CLSI) of the USA and World Health Organization (WHO) recommended that reference values should be established for each region.[4] Thereby, for a better interpretation of hematologic results in our country (Cameroon), we decided to perform this preliminary study in aim to establish the hematological reference values of the city of Bangangté (west Cameroon).

MATERIALS AND METHODS

Ethical consideration

The go-ahead was obtained from the Head of the Cliniques Universitaires des Montagnes under reference number: Ref: 0213/CUM.

Setting and populations

This study was a cross sectional descriptive study carried out between December 2017 and May 2018 in town of Bangangté, western region of Cameroon. The town is located between Latitudes 5° 08′ 49″ N and Longitudes 10° 24′ 54″ E., altitude of 1350 m above sea level and temperature is generally ranges between 14^{o} and 27^{o}. Typically semi-urban, Bangangté hosts a mixed population of 28 000 inhabitants.

Selection of reference population

During study, 512 young adults living in Bangangté for at least 3 months who gave their informed consent were allowed to participate in the study. All participants underwent clinical, radiological and biological analysis to confirm their health status.

Clinical evaluation of the participants of the study was performed by a medical doctor. During this evaluation, medical parameters which can influence hematological values as high temperature, symptoms of anemia, pregnancy, abdominal or body pains, history of drug, alcohol, tobacco, asthenia, infectious diseases, cancer, diabetes, blood donation during the last 3 months, blood transfusion, hemophilia. Radiological analysis has been performed by a radiologist technician, and this analysis had consisted to do a chest x-ray in order to detect a pulmonary infection. Biological analysis was performed by laboratory technician, this analysis consisted to do serological tests and to detect hemoglobinopathy as sickle cells and hemoparasite after perform blood smear (May-Grünwald-Giemsa). All participants who had a positif result after clinical, radiological and biological analysis had been excluded from study.

Blood collection, serological testing, hematological analysis and conservation

Blood collection

Five milliliters of venous blood was collected in Dipotassium Ethylene Diaminetetraacetic acid (EDTA) tubes and five milliliters venous blood was collected in tube without anticoagulant. The collection of blood was performed by vacutainer system. The samples were collected between 8:00 a.m. and 11:30 a.m. After mixing the blood (EDTA tube) with the anticoagulant; the test tubes were appropriately labeled. The samples were then placed in ice-box, transported to laboratory of Cliniques Universitaires des Montagnes and processed within 2h. All bad samples were excluded.

Serological testing

Serological test for hepatitis B virus (HBV) and hepatitis C virus (HCV) were undertaken using Determine Ag HBs (FASTEP Lot No. IHBSG-401; 09/2019), Ac HCV (CJ SMART Lot No. HCV17120016; 09/2019) and Ac HBC (DIAGNOS Lot No. 20160528; 05/27/2019)).

Hematological Analysis

Hematological analysis was performed using MINDRAY AUTO HEMATOLOGY ANALYZER (BC-2800) for whole-blood analysis of hematological parameters. This instrument automatically counts and gives a printout result of absolute numbers of white blood cells (WBC) (G/L), red blood cells (RBC) (T/L), hemoglobin (g/dL), hematocrit (%), mean cell volume (MCV) (fl), mean cell hemoglobin (MCH) (pg), mean cell hemoglobin concentration (MCHC) (g/dL), platelets (G/L). Neutrophils (G/L), lymphocytes (G/L), eosinophils (G/L), basophils (G/L), and monocytes cells (G/L) were estimated after performing leucocytes count on blood smear stained with May grundwald-Giemsa (MGG).

Conservation

After analysis, blood collected in EDTA tubes were conserved at - 4^{o}C and serum were conserved in cryoconsever after were appropriately labeled and kept at - 20^{o}C.

Statistical analysis

Data were recorded in the Excel software and analyzed with R software. The mean and 2.5th and 97.5th percentiles were calculated for each parameter. Differences were statistically significant when $p < 0.05$.

RESULTS

During this study, 512 young adults living in Bangangté for at least 3 months who gave their informed consent were allowed to participate in the study. Of the 512, 200 had been excluded and 312 (131 men and 181 women) with age between 15 and 29 years old had been retained after clinical, radiological and biological analysis. After screening healthy population, hematological analysis were performed to establish reference values of hematological parameters. These parameters were summarized as showed in **Table** 1 and **Table** 2.

Table 1: References values of parameters of erythrocyte lineage and platelet lineage.

Parameters	SEX	Mean	RV (2.5th–97.5th‰)	p-value (gender)
Red blood cells (RBC) (T/L)	Combined	5.19 (±0.76)	3.8-6.9	**<0.001**
	Male	5.67 (±0.70)	4.3-6.9	
	Female	4.84 (±0.60)	3.5-6	
Hemoglobin (Hgb) (g/dL)	Combined	13.3 (±2.14)	8-17	**<0.001**
	Male	14.6 (±1.66)	11-17	
	Female	12.4 (±1.97)	7.8-16	
Hematocrit (Hct) (%)	Combined	44.1 (±6.98)	29-55	**<0.001**
	Male	47.7 (±5.97)	33-57	
	Female	41.4 (±6.49)	28-52	
Mean cell volume (MCV) (fL)	Combined	85.6 (±6.54)	71-95	**0.35**
	Male	85.6 (±5.06)	75-94	
	Female	85.6 (± 7.46)	69-95	
Mean cell hemoglobin (MCH) (pg)	Combined	25.8 (± 2.89)	20-30	**0.49**
	Male	26.0 (±2.64)	21-30	
	Female	25.7 (±3.06)	19-30	
Mean cell hemoglobin concentration (MCHC) (g/dL)	Combined	30.1 (±2.05)	27-32	**< 0.001**
	Male	30.6 (±2.54)	28-33	
	Female	29.7 (±1.51)	27-32	
Platelet (PLT) (G/L)	Combined	290 (±76.2)	152-438	**< 0.001**
	Male	268 (±58.9)	165-412	
	Female	305 (±83.4)	130-464	

RV: Reference value.

Table 1 shown on the one hand that there was a significant difference in RBC count (p<0.001); hemoglobin (p<0.001), Hct (p<0.001), MCHC (p<0.001) and platelets (p< 0.001) between the two sexes. And on the other hand that there was no significant difference between male and female in MCV (p=0.35) and MCH (p= 0.49).

Table 2: References values of parameters of leucocyte lineage.

Parameters	SEX	Mean	RV (2.5th–97.5th‰)	p-value (gender)
White blood cells (WBC) (G/L)	Combined	5.75 (±1.54)	3.5-9	**0.065**
	Male	5.55 (± 1.51)	3.3-8.5	
	Female	5.89 (±1.55)	3.5-9.7	
Lymhocytes (G/L)	Combined	2.80 (±1.07)	1.3-4.8	**0.16**
	Male	2.72 (±096)	1.4-4.5	
	Female	2.87 (±1.17)	1.3-5.2	
Monocytes (G/L)	Combined	0.35 (±0.25)	0.08-0.81	**0.25**
	Male	0.34 (±0.26)	0.07-0.76	
	Female	0.35 (±0.25)	0.11-0.81	
Neutrophils (G/L)	Combined	2.31 (±0.91)	1-4.4	**< 0.001**
	Male	2.12 (±0.75)	0.96-3.7	
	Female	2.49 (±1.01)	1.2-4.7	
Eosinophils (G/L)	Combined	0.15 (±0.18)	0.04-0.63	**0.62**
	Male	0.17(±0.22)	0.04-0.98	
	Female	O.12 (±0.13)	0.044-0.49	
Basophils (G/L)	Combined	0.01(±0.04)	0.036-0.22	**0.093**
	Male	0.01(±0.03)	0.04-0.16	
	Female	0.01(±0.004)	0.039-0.26	

RV: Reference value.

Table 3: Comparison of the references values of erythrocyte lineage and platelet obtained in this study with values obtained in other countries.

Parameters	SEX	Our study	C. Africa 2003[8]	Ghana 2012[7]	Togo 2011[5]	Ethiopie 2016[6]	Eritrea 2018[1]	Morocco 2016[11]	France 2014[10]	USA 2004[9]
RBC (T/L)	Combined	3.8-6.9	NA	3.39-5.83	3.1-6.4	NA	4.07-6.02	NA	NA	NA
	Male	4.3-6.9	4.5-6.1	3.79-5.96	3.3-6.4	3.53-6.93	4.2-6.07	4.83-7.03	4.53-5.79	4.50-5.90
	Female	3.5-6	3.42-5.44	3.09-5.30	3.1-6.0	3.45-6.25	4-5.7	3.9-5.96	4.01-5.19	4.00-5.20
Hemoglobin (Hgb) (g/dL)	Combined	8-17	NA	9.8-16.0	10-18.4	NA	12.6-17.7	NA	NA	NA
	Male	11-17	12.3-17.3	11.3-16.4	10-18.4	11.5-18	12.6-17.8	13.8-20.21	13.4-16.7	13.5-17.5
	Female	7.8-16	9.1-14.9	8.8-14.4	10.3-17.1	11.0-16.7	12.5-17.6	10.10-16.97	34.4-43.9	12.0-16.0
Hematocrit (Hct) (%)	Combined	29-55	NA	28.9-48.7	28-54	NA	38.3-54.4	NA	NA	NA
	Male	33-57	39-52	33.2-50.5	28-54	36.2–58.6	40.5-55	43.38-61.60	39.2-48.6	41.0-53.0
	Female	28-52	28-44	26.4-45	28-47	32.1-56.6	37.9-52	31.39-51.64	34.4-43.9	36.0-46.0
Mean cell volume (MCV) (fL)	Combined	71-95	NA	72-97	80-99	85-100	85.8-100	NA	NA	80-100
	Male	75-94	NA	70-98	80-99	NA	85.7-100	77.97-100.6	78.0-91.9	NA
	Female	69-95	NA	73-93	80-95	NA	85.5-100	74-100.08	75.7-92.7	NA
Mean cell hemoglobin (MCH) (pg)	Combined	20-30	NA	22.6-33.5	25-37	NA	27.4-32.8	NA	NA	26-34
	Male	21-30	NA	22.7-33.5	26-36	26.6-33.3	28-33	24.80-32.30	26.3-32.1	NA
	Female	19-30	NA	22.3-33.6	25-37	25.8-32.8	26.5-32.6	19.91-31.97	75.7-92.7	NA
MCHC (g/dL)	Combined	27-32	NA	30.5-36.2	29-41	NA	30.2-33.8	NA	NA	31-37
	Male	28-33	NA	30.6-36.0	29-39	29.5-34.4	30.4-33.7	29.68-34.20	32.4-36.3	NA
	Female	27-32	NA	30.4-36.5	30-41	28.5-34.4	30-33.7	27.50-33.90	31.9-35.8	NA
Platelet (G/L)	Combined	152-438	NA	89-380	120-443	128-432	134-344.2	NA	NA	150-350
	Male	165-412	124-378	88-352	120-443	NA	128-318.4	108-327.25	172-398	NA
	Female	130-464	117-382	89-403	150-436	NA	145-351.6	121.13-341	185-445	NA

From **Table 2**, it appears on one hand that there was significant difference in neutrophil count ($p< 0.001$) between two sexes. And on other hand that no significant difference between WBC ($p=0.065$), lymphocyte ($p=0.16$), monocyte ($p=0.25$), eosinophil ($p=0.62$) and basophil ($p=0.093$). The hematological reference interval established in this study were compared to reference interval established in similar studies in other counties. This comparison were summarized as shown in **Table 3** and **Table 4**.

Table 4: Comparison of the references values of leucocyte lineage obtained in this study with values obtained in other countries.

Parameters	SEX	Our study	C. Africa 200[8]	Ghana 2012[7]	Togo 2011[5]	Ethiopie 2016[6]	Eritrea 2018[1]	Morocco 2016[11]	France 2014[10]	USA 2004[9]
WBC (G/L)	Combined	3.5-9	NA	3.4-9.2	1.9-10.1	3.2-8.8	3.4-9.0	NA	NA	4.5-11
	Male	3.3-8.5	2.9-8.3	3.5-9.2	1.9-10.1	NA	3.7-9.3	4.60-13.81	4.09-11.0	NA
	Female	3.5-9.7	2.7-8.0	3.4-9.3	2.2-7.8	NA	3.3-8.9	5.10-11.58	4.02-11.4	NA
Lymphocytes (G/L)	Combined	1.3-4.8	NA	1.2-4.4	1.1-4.3	1-3.5	NA	NA	NA	NA
	Male	1.4-4.5	1.5-4.22	1.2-5.2	1.1-4.3	NA	NA	0.59-4.41	1.34-3.92	NA
	Female	1.3-5.2	1.4-3.68	1.2-4.4	1.2-4.3	NA	NA	0.42-4.18	1.37-3.96	NA
Monocytes (G/L)	Combined	0.08-0.81	NA	0.2-1.0	0.05-0.8	NA	NA	NA	NA	NA
	Male	0.07-0.76	NA	0.2-1.4	0.05-0.8	NA	NA	0-1.44	0.23-0.77	NA
	Female	0.11-0.81	NA	0.2-0.9	0.05-0.8	NA	NA	0-1.18	0.20-0.71	NA
Neutrophils (G/L)	Combined	1-4.4	NA	NA	0.5-5.4	1.6-5.1	NA	NA	NA	NA
	Male	0.96-3.7	NA	NA	0.5-5.4	NA	NA	1.60-9.80	1.78-6.95	NA
	Female	1.2-4.7	NA	NA	0.5-4.4	NA	NA	2.05-9.46	1.75-7.50	NA
Eosinophils (G/L)	Combined	0.04-0.63	NA	NA	0-0.5	NA	NA	NA	NA	NA
	Male	0.04-0.98	NA	NA	0-0.5	NA	NA	0-0.80	0.05-0.63	NA
	Female	0.044-0.49	NA	NA	0-0.5	NA	NA	0-0.80	0.04-0.58	NA
Basophils (G/L)	Combined	0.036-0.22	NA	NA	NA	NA	NA	NA	NA	NA
	Male	0.04-0.16	NA	NA	NA	NA	NA	0-0.50	0.00-0.10	NA
	Female	0.039-0.26	NA	NA	NA	NA	NA	0-0.20	0.00-0.08	NA

C. Africa: Central Africa; WBC: white blood cells; USA: United State of America; NA: not available.

Results of the comparisons of the reference values obtained in this study with similar studies undertaken in Central Africa, Ghana, Togo, Ethiopia, Morocco, France and United States of America (USA) showed that globally references values obtained in this study were similar to reference values obtained in Ghana and different to reference values obtained in other countries.

DISCUSSION

This study showed that there was a difference between gender in the reference values for specific RBC parameters including RBC count, Hgb, Hct and RBC indices like MCHC. Historically, the observed gender differences have been attributed to a range of factor including: menstruation, hormonal influences of androgen, estrogens. testosterone on erythropoiesis and low oxygen consumption in women compare to men.[1,8] However, MCV did not exhibit a significant difference. These finding is similar to other results from some studies conducted elsewhere in Africa.[1,6,12] There was statistically significant difference in platelet count between the two sexes; this result is different to results from studies undertaken in Eritrea, Ghana and Kenya.[1,8]

In addition, the references values of total WBC count for male and females did not show any significant variation. This is similar to finding from Eritrea, Ethiopia and northern Nigeria.[6,12] However, a study conducted in Turkey showed statistically significant sex-related difference for WBC count.[13]

Comparison of the reference interval obtained with reference intervals finding in other countries show higher dissimilarity between reference values obtained in this study and reference values obtained in others countries. We don't know the real cause of these dissimilarity but these results could be explained by many factors influences the hematological values such as sex, age, ethnic origin, geographic location, season, and genetic disease.[1,5] Globally, RBC count obtained in this study (male: 4.3-6.9 T/L; female 3.5-6 T/L) was similar to values obtained in Central Africa (male: 4.5-6.1 T/L; female: 3.42-5.44 T/L), France (male: 4.53-5.79 T/L; female: 4.01-5.19 T/L), United States of America (male: 4.50-5.90 T/L; female: 4.00-5.20 T/L) and Togo (male: 3.3-6.4 T/L; female: 3.1-6.0 T/L).[1,5,10] Concerning hemoglobin, value obtained among men (11-17 g/dL) was higher to values of female (7.8-16 g/dL), this result is similar to result obtained in other countries Central Africa (male: 12.3-17.3 g/dL; female: 9.1-14.9 g/dL), Ghana (male:11.3-16.4 g/dL; female: 8.8-14.4 g/dL), Togo (male: 10-18.4 g/dL; female: 10.3-17.1 g/dL).[1,5,6] Globally hematocrit (male 33-57%; female 28-52%) was similar to results showed in Togo ((male 28-54%; female 28-47%) but low to values obtained in Morocco (male:43.38-61.60%; female: 31.39-51.64%), Eritrea (male: 40.5-55%; female:37.9-52%), France (male:39.2-48.6%; female: 34.4-43.9%), USA (male:41.0-53.0%; female: 36.0-46.0%), and Ethiopia (male: 36.2-58.6%; female: 32.1-56.6%). MCV (71-95 fl) was similar to values obtained in Ghana (72-97 fl) but low to values obtained in Ethiopia (85-100 fl), Eritrea (85.8-100 fl), USA (80-100 fl) and Togo (80-99 fl).[1,5,6] MCH (20-30 pg) were practically equal to values obtained in Ghana (22.6-33.5 pg) but low to value found in Togo (25-37 pg), Eritrea (27.4-32.8 pg), and USA (26-34 pg). In general, MCHC were low to values found in other countries.

Platelets (152-438 G/L) were similar to results found in Togo (120-443 G/L), Ethiopia (128-432 G/L) but higher to values obtained in Ghana (89-380 G/L), and Eritrea (134-344.2 G/L).[5,6,11]

The current study revealed that reference values of WBC count were 3.5-9 G/L. these values were similar to reference values obtained in Ghana (3.4-9.2 G/L), Ethiopia (3.2-8.8 G/L), and Eritrea (3.4-9.0 G/L) but higher to values obtained in Central Africa (male: 2.9-8.3 G/L; female: 2.7-8.0 G/L) and low to values obtained in Morocco (male:4.60-13.81 G/L; female: 5.10-11.58 G/L), in France (male: 4.09-11.0 G/L; female: 4.02-11.4 G/L) and USA (4.5-11 G/L).[5,6] Concerning lymphocyte (male 1.4-4.5 G/L; female 1.3-5.2), the values were practically similar to values obtained in other setting Ghana (male 1.2-5.2 G/L; female 1.2-4.4 G/L), Togo (male 1.1-4.3 G/L; female 1.2-4.3 G/L) but dissimilar to values found in Morocco (male: 0.59-4.41 G/L; female 0.42-4.18 G/L).

Monocytes (0.08-0.81 G/L) were almost equal to the value found in Togo (0.05-0.8 G/L), but low compared to those obtained in Morocco (male: 0- 1.44 G/L, female: 0-1.18 G/L). Neutrophilia were similar to values obtained in many countries of black Africa, this value can be explained by a genetic factor in black population but mechanisms by which black population have lower values of neutrophils is still not clear.[5,8] Eosinophils and Basophils values in this current study was similar to values obtained in other countries, these results can be explained by the fact that the populations of this study had no infections and were healthy.[5,6]

CONCLUSION

This study aimed to establish the hematological reference values of young adults in Bangangté, Cameroon. This study showed firstly that hematological reference values were very different between one country and others; secondary that, these differences can be explained by factors influencing the hematological values such as sex, age, ethnic origin, geographic location, season, and genetic disease; in end that, the references values obtained in one setting can not be apply in other country. For this reason, each country must establish its own reference values.

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