Gender-Specific Distribution of Hematological Parameters in Adults Living in Nouna, Burkina Faso

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Abstract: We examined the distribution of hematological parameters in 186 adults (89 female, 97 male) living in Nouna, Burkina Faso. Median (5^{th} – 95^{th} percentile) hemoglobin concentration was 12.6 [10.3-15.3] g/dl, which was lower than reported from other populations in Africa. Iron deficiency may be the most important reason for this observation.

Keywords: Anemia, hemoglobin, red blood cell indices, malaria, iron deficiency.

There is ample evidence that clinicians and medical researchers should use method-specific reference ranges in their laboratories which account for gender differences and variances in the ethnic composition of the local society [1]. In particular, reference ranges of hematological indices based on results from western individuals are not in agreement with those calculated from African populations [2].

From July 2004 to September 2005 a study was performed by the Nouna Health Research Center (Centre de la Recherche en Santé de Nouna, CRSN) in order to generate gender-specific reference values for immunological parameters which could be used in immunological monitoring of individuals living with HIV/AIDS [3]. We now analysed data on hematological parameters in order to assess the distribution of the measurements in our study population.

STUDY DESIGN

The study was part of an ongoing longitudinal trial on the prevention of mother-to-child transmission of the human immune deficiency virus in Nouna [4] which was approved by the Institutional Ethics Committee of the University of Heidelberg, Germany and the Institutional Ethics Committee at the CRSN. Donors were recruited after informed consent from female clients of the mother and infant clinic at the Nouna Health Center and during three blood donation campaigns in the community of Nouna town. Details of the study protocol are described elsewhere [3].

Any positive serological test for the presence of antibodies against HIV or syphilis as well as for HBsAg led to exclusion from the study. In addition, based on history and physical examination, eligible donors were classified as "clinically healthy", i.e. they did not show or report signs and symptoms of illness (>5 kg weight loss in the preceding month, fever during the last 2 weeks, asthma, diabetes, cardiovascular and renal diseases). Clinically inapparent chronic conditions (e.g., hepatitis) were not specifically screened for by laboratory testing.

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An automated device (Sysmex KX21N, Sysmex Corporation, Kobe, Japan) was used to measure erythrocyte count (RBC), hemoglobin concentration (Hb), hematocrit (Hct), RBC mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC), platelet count (PLT), total leukocyte count (WBC), and the automated white blood differential (percentages of lymphocytes (LYM), neutrophils (NEUT), and the mixed population of monocytes, basophiles and eosinophiles [MXD]). The measurements were performed on 50µl of whole blood collected *via* venipuncture into EDTA. Quality control was done at regular intervals using EIGHTCHECK 3 WP high, low and normal manufactured by Sysmex.

Descriptive statistics were calculated using SAS for Windows; for comparison between male and female subjects the TTEST procedure and for comparison between different age groups linear regression analysis (GLM procedure) was used.

RESULTS AND DISCUSSION

Data from 186 adults (89 female, 97 male) aged 18 to 78 years were available for final analysis (Table **1**). Age distribution was as follows: <20 years, n = 28 (19 female, 9 female); 20 to 29 years, n = 99 (59 female, 40 male); 30 to 39 years, n = 31 (16 female, 15 male); 40 years and more, n = 28 (14 female, 14 male). Male blood donors had significantly higher RBC counts than females (mean difference: $0.5*10^{12}$/l, 95% CI: 0.3 to $0.6*10^{12}$/l; $p<0.0001$), higher Hb values (mean difference: 1.7 g/dl, 95% CI: 1.4 to 2.1 g/dl; $p<0.0001$), and a higher Hct (mean difference: 0.05 l/l, 95% CI: 0.04 to 0.06 l/l; $p<0.0001$). No statistically significant influence of age on hematological parameters could be detected in our cohort.

RBC, Hb, and Hct values in Nouna were lower than those reported from other African populations (Table **2**). Gender-specific differences (female<male) were similar to those published in the literature [5-7]. Beutler and Waalen recently suggested specific lower limits of normal Hb concentrations for black US-American males (12.9 g/dl) and females (11.5 g/dl) [8]. When we applied those limits to our study population, approximately 30% of all clinically healthy adults in Nouna were classified as anemic.

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Table 1. Distribution of Values for Hematological Parameters Obtained from Healthy Adults in Nouna, Burkina Faso. The Marker "§" Indicates a Statistically Significant Difference Between Males and Females (p<0.01)

Total (n=186)	RBC§ (10^{12}/l)	Hb§ (g/dl)	Hct§ (l/l)	MCV (fl)	MCH (pg)	MCHC (g/dl)	PLT (10^9/l)	WBC (10^9/l)	LYM (%)	MXD (%)	NEUT (%)
95th centile	5.4	15.3	0.44	92	32	36.2	356	8.5	59	20	63
75th centile	4.8	13.8	0.40	88	30	35.2	281	5.9	50	14	51
50th centile (median)	4.4	12.6	0.37	85	29	34.4	236	5.2	44	11	45
25th centile	4.1	11.9	0.35	81	27	33.2	202	4.3	38	9	37
5th centile	3.6	10.3	0.31	75	24	31.7	146	3.4	27	6	28
Male (n=97)											
95th centile	5.6	15.6	0.46	94	33	36.3	365	9.2	60	22	64
75th centile	5.0	14.3	0.41	88	31	35.5	271	5.9	50	15	52
50th centile (median)	4.7	13.5	0.39	85	29	34.6	217	5.1	44	12	43
25th centile	4.3	12.5	0.37	81	28	33.5	190	4.3	39	9	36
5th centile	3.8	11.3	0.34	73	25	31.7	127	3.2	26	4	27
Female (n=89)											
95th centile	4.9	13.5	0.39	92	32	36.1	356	7.4	57	18	62
75th centile	4.5	12.5	0.37	87	30	35.0	291	5.9	49	14	51
50th centile (median)	4.2	12.0	0.35	84	28	34.2	252	5.2	43	11	46
25th centile	3.9	11.0	0.33	80	27	32.9	214	4.5	37	9	39
5th centile	3.5	9.8	0.30	75	24	31.4	159	3.4	27	7	30

Abbreviations: RBC = erythrocyte count, Hb = hemoglobin concentration, Hct = hematocrit, MCV = mean corpuscular volume, MCH = mean cellular hemoglobin, MCHC = mean cellular hemoglobin concentration, PLT = thrombocyte count, WBC = leukocyte count, LYM = lymphocytes, MXD = monocytes, eosinophiles and basophiles, NEUT = neutrophils.

Table 2. Comparison of the Distribution of Values for Hematological Parameters Retrieved from Distinct Populations in Burkina Faso (Present Study), Ethiopia [5], Uganda [7], the Central African Republic (CAR) [6], and the USA (Black Americans) [9]. Data are Given as Median and 5th and 95th Centiles Unless Otherwise Indicated

Region (range of age)	sex	RBC (10^{12}/l)	Hb (g/dl)	Hct (l/l)	MCV (fl)	MCH (pg)	MCHC (g/l)	PLT (10^9/l)	WBC (10^9/l)	LY (10^9/l)	LY (%)	NEUT (%)
Burkina Faso (18-78 years) n = 186	M	4.7 (3.8-5.6)	13.5 (11.3-15.6)	0.39 (0.34-0.46)	85 (73-94)	29 (25-33)	34.6 (31.7-36.3)	217 (127-365)	5.1 (3.2-9.2)	2.1 (1.3-4.0)	44 (26-60)	43 (27-64)
	F	4.2 (3.5-4.9)	12.0 (9.8-13.5)	0.35 (0.30-0.39)	84 (75-92)	28 (24-32)	34.2 (31.4-36.1)	252 (159-356)	5.2 (3.4-7.4)	2.2 (1.4-3.2)	43 (27-57)	46 (30-62)
Uganda (25-92 years) n = 845	M	4.9 (3.8-6.0)	14.1 (11.1-16.8)	0.41 (0.32-0.48)	84 (70-95)	NA	NA	171 (80-288)	5.3 (3.4-8.7)	2.4 (1.4-4.2)	NA	NA
	F	4.5 (3.7-5.3)	12.5 (10.1-14.3)	0.36 (0.30-0.41)	81 (68-93)	NA	NA	198 (100-297)				

(Table 2). Contd…..

Region (range of age)	sex	RBC (10^{12}/l)	Hb (g/dl)	Hct (l/l)	MCV (fl)	MCH (pg)	MCHC (g/l)	PLT (10^9/l)	WBC (10^9/l)	LY (10^9/l)	LY (%)	NEUT (%)
Ethiopia* (15-45 years) n = 485	M	5.0 (4.3-5.9)	16.1 (13.9-18.3)	0.48 (0.42-0.55)	NA	NA	NA	203 (97-324)	5.9 (3.0-9.8)	1.8 (0.9-3.5)	36 (16-55)	56 (32-79)
	F	4.5 (3.7-5.2)	14.4 (12.2-16.6)	0.42 (0.35-0.49)	NA	NA	NA	193 (98-352)	5.9 (3.0-12.2)	1.7 (1.1-3.5)	34 (20-64)	58 (23-73)
CAR* (16-58 years) n = 150	M	5.1 (4.5-6.1)	14.9 (12.3-17.3)	0.45 (0.39-0.52)	NA	NA	NA	225 (124-378)	5.1 (2.9-8.3)	2.5 (1.5-4.2)	NA	NA
	F	4.5 (3.4-5.4)	12.5 (9.1-14.9)	0.38 (0.28-0.44)	NA	NA	NA	228 (117-382)	4.9 (2.7-8.0)	2.4 (1.5-3.7)	NA	NA
USA (31-40 years) n = ca. 940	M	4.9 (4.1-5.8)	15.0 (12.8-16.8)	0.45 (0.39-0.51)	92 (79-104)	31 (26-35)	33.4 (30.5-37.2)	NA	6.5 (4.1-10.4)	NA	NA	NA
	F	4.3 (3.7-5.1)	13.3 (11.5-15.1)	0.40 (0.33-0.46)	91 (79-104)	31 (26-35)	33.3 (30.5-37.6)		6.7 (4.4-11.0)			
SYSMEX reference range ("adults")	M	4.6-6.2	14.0-18.0	0.43-0.49	85-95	27-33	32.0-36.0	140-400	4.0-10.0	1.0-4.0	25-40	50-70
	F	4.2-5.4	12.0-16.0	0.36-0.46								

*median ± 2 standard deviations or 2.5th and 97.5th centiles; Abbreviations: M = male, F = female, RBC = erythrocyte count, Hb = hemoglobin concentration, Hct = hematocrit, MCV = mean corpuscular volume, MCH = mean cellular hemoglobin, MCHC = mean cellular hemoglobin concentration, PLT = thrombocyte/platelet count, WBC = leukocyte count, LY = lymphocytes, NEUT = neutrophils, NA = not available.

Comparison our data on mean corpuscular volume with MCV percentiles for black Americans living in the metropolitan area of Washington, D.C. revealed a tendency towards microcytosis in our study population: median MCV (50^{th} percentile) was in the range of the 25^{th} MCV percentile of black Americans [9]. The ratio of MCV (fl) over RBC (10^6 cells μl^{-1}) [10] was >14 in all study participants, indicating that iron deficiency may be the most important reason for microcytic anemia in healthy adults in Nouna. No data are yet available, however, on the prevalence of iron deficiency, or on the frequency of diseases that may cause iron deficiency, e.g. infestation with hookworms. We do not know the prevalence of hookworms in Nouna, but infestation with helminths was not associated with anemia in pregnant women in Uganda [11].

Malaria is a frequent cause of clinically apparent anemia in Nouna and infection with Plasmodium species is highly prevalent after the rainy season (approximately 80% as detected by microscopic examination of thick blood films and more than 90% by PCR techniques) [12]. The persistence of submicroscopic Plasmodium gametocyte carriage which has been shown to be common in an area of low and seasonal transmission [13] may lead to increased hemolysis and clearance of uninfected erythrocytes and have also have a negative impact on hematopoesis [14].

Previous studies had revealed a high prevalence (29.8%) of abnormal hemoglobins in the study area, but the frequency of thalassaemia was very low and only the carriers of hemoglobin SC (present in 1.2% of the female population) had a high risk of being anemic [15]. Other potential genetic causes of hereditary anemia such as glucose-6-phosphate-dehydrogenase deficiency or glutathion reductase deficiency were diagnosed in 6-16% of the study population but these enzymopathies do not cause microcytic anemia [16].

We do not know whether the distribution of hematological parameters in our study population shows seasonal changes; this question requires further study. Another potential limitation of data interpretation relates to the fact that we did not specifically screen for clinically inapparent chronic diseases and relied on history and assessment of symptoms when recruiting our healthy study population. We do not expect, however, that failure to exclude rare cases of chronic diseases does significantly change the results of our study. Gender-specific distribution of RBC, Hb, and Hct values in adults in Nouna was lower than the reference ranges reported for black US-American adults and also below those of other populations in Africa. Iron deficiency may be the most important explanation for this observation.

The clinical consequences of low RBC, Hb and Hct values in African adults should be studied in more detail. Recent epidemiological data from the US showed that anemia was significantly associated with increased risk of death and mobility disability only in older whites while older blacks with anemia were not at risk for adverse events [17]. Thus,

the question "what is the lower limit of normal of the blood haemoglobin concentration" [8], eventually has to be answered differently for people living in areas where anemia, malaria, iron deficiency and high mortality rates prevail.

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