

## Hematological Parameters in Moderately Trained Students

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### Abstract

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#### **Purpose:**

To create first reference values of blood volume and hematological parameters in young and moderately trained people.

#### **Introduction:**

The hemoglobin mass and the total blood volume determine the oxygen transport capacity. The hemoglobin mass and the maximum cardiac output are the most relevant dependent variables of maximum aerobic capacity. Maximum aerobic capacity is often calculated in relation to the whole body mass and the body composition. The aim of this study is to determine the total hemoglobin mass and other hematological parameters in relation to body composition parameters as measured by using a bio-impedance method.

#### **Methods:**

Measurements were taken in 100 students (54 men, 46 women, mean age 24 or 21 years) of the University of Leipzig during their clinical courses. The data acquisition included the following parameters: body weight and height, water (TBW), fat mass (FM), body cell mass (BCM), and lean body mass (LBM); hematocrit (Hct), hemoglobin fraction (Hb) and total hemoglobin mass (tHb). The total hemoglobin mass was measured by using a modified closed circle rebreathing CO-method. The total blood volume (BV) and the total erythrocyte volume (RCV) were calculated from tHb and hematocrit values. The data were assessed separately for men and women to also calculate gender differences. A correlation analysis was performed for the relation between body composition and hematological parameters.

#### **Results:**

##### Mixed group:

Mean values for tHb, BV, RCV: 842 g, 5975 ml, 2408 ml. Body weight and LBM: 68 kg, 55 kg. Relation between body composition and hematological parameters: BV vs weight:  $r = 0.84$ ; tHb vs weight:  $r = 0.8$ ; BV vs LBM:  $r = 0.92$ ; tHb vs LBM:  $r = 0.91$ . BV may be estimated from LBM as follows:  $BV (l) = 0.122 \text{ LBM (kg)} - 0.818$ ;  $r = 0.9$  or approximately 0.106 l per kg LBM.

##### Gender-specific results:

Women: Mean values for tHb, BV, RCV: 637 g, 4952 ml, 1861 ml. Body weight and LBM: 62 kg, 47 kg. Relation between body composition and hematological parameters: BV vs weight:  $r = 0.69$ ; tHb vs weight:  $r = 0.63$ ; BV vs LBM:  $r = 0.76$ ; tHb vs LBM:  $r = 0.64$ . BV may be estimated from LBM as follows:  $BV (l) = 0.153 \text{ LBM (kg)} - 2.233$ ;  $r = 0.76$  or approximately 0.1 l per kg LBM.

Men: Mean values for tHb, BV, RCV: 1045 g, 6846 ml, 2873 ml. Body weight and LBM: 73 kg, 63 kg. Relation between body composition and hematological parameters: BV vs weight:  $r = 0.74$ ; tHb vs weight:  $r = 0.65$ ; BV vs LBM:  $r = 0.79$ ; tHb vs LBM:  $r = 0.67$ . BV may be estimated from LBM as follows:  $BV (l) = 0.125 \text{ LBM (kg)} - 1.014$ ;  $r = 0.79$  or approximately 0.1 l per kg LBM.

#### **Conclusions:**

The major result of this study is that the relation between the blood volume and the lean body mass is almost equal in men and women. So on the one hand, the results prove that for general purposes the fat-free mass should be used to make estimations about the hemoglobin mass or the blood volume. On the other hand, a relatively large variation between the subjects indicates that the total hemoglobin should be measured directly whenever higher precision is desired. Almost all parameters require different regression equations for men and women if the body weight is used. The total blood volume is about 0.1 l per kg lean body mass in men and women. Consequently, the difference in the relative maximum O<sub>2</sub>-uptake (ml/kg body weight) can be explained by a minor portion of body fat in men. Other factors such as the cardiac output may play a major role.

#### **Key words:**

Hemoglobin mass, blood volume, body composition, gender

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## Introduction:

The knowledge about the individual total hemoglobin mass and the derived blood volume (BV), plasma volume (PV) and erythrocyte volume (RCV) as basic parameters of the circulation, is in the field of clinics, sports science and sports medicine still of great interest. Changes of the circulation under various pathological conditions as well as during diagnostic and therapeutic measures are not to be disregarded, too [1]. The total hemoglobin mass and the blood volume represent the oxygen transport capacity in the body. So, in addition to the cardiac output per minute, they are the important variables of the maximum aerobic capacity. In the inter-individual comparison, the maximum oxygen uptake, as key parameter of the aerobic capacity, is often standardized by using the body mass.

In the past, hematological parameters such as the blood volume, plasma volume and the erythrocyte volume have been relativized by the weight and the height [2, 3]. Therefore, obese people show a pathological lower relative BV, RCV and PV because adipose tissue is relatively avascular. So, the hematological measurements are partially seen in relation to the body surface whereby the weight but also the height of people

## Methods

### Study Group:

All 100 study participants were moderately trained students (mainly sports students) of the University of Leipzig. They give were written informed consent after they were informed about the study and the risks.

**Table 1 basic characteristic of the study participants separated by gender.**

	Male (n = 54)	Female (n = 46)
Age (years)*	23.5 ± 3.5	21.0 ± 2.0
Height (cm)*	180 ± 7	169 ± 8
Weight (kg)*	73.2 ± 7.5	62.0 ± 6.1
BMI (kg/m <sup>2</sup> )*	22.7 ± 1.7	21.8 ± 2.2
Fat-free mass (kg)*	62.7 ± 5.7	47.0 ± 2.8
Sports activity per week in (h)*	9.8 ± 5.7	8.6 ± 4.4

\* Mean value and standard deviation in the study group

### Examination procedure:

The total hemoglobin mass and the derived blood parameters of all participants were determined with the CO-rebreathing method [10]. Before determining the blood volume, the body composition was measured with the help of a bio-impedance measurement (Akern impedance analyser STA/BIA, Italy).

The bioelectrical impedance analysis (BIA) has been used for about 20 years as a fast and non-invasive method for determining the body composition. After placing two electrodes on each, the hand and the foot of the participant, the resistance (R, ohmic impedance) and reactance (Xc, capacitive impedance) are measured in

influences the relative blood volumes. Already in 1995, the International Council for Standardisation in Haematology has demanded an individual anthropometric reference to reflect the body composition [4]. The fat-free body mass was suggested as the ideal parameter [4] and was confirmed as such by different examinations. [5, 6, 7, 8].

The nowadays simple determination of the body composition and, among others, of the fat-free body mass, permits a relativization of the hematological parameters, independent from the fat mass.

In this study, the total hemoglobin mass and the derived parameters were determined via the CO-rebreathing method. Additionally, connections with measurements of the body composition were investigated in general but also separately for men and women. The aim of this study was to investigate relations between the hemoglobin mass and other hematological measurements by using parameters of the body composition that were determined with the help of a bio-impedance analysis.

the high frequency alternating current field (mainly 50 kHz) at constant current.

Basically, the used CO-rebreathing method is characterized by the combination of three components:

1. Defined individual CO-bolus application [cf. 9]
2. Rebreathing in the closed breathing system with oxygen substitution for inducing a HbCO-steady state
3. Quantification of CO that is unbound to hemoglobin and located in the respiratory system (remaining CO)

The individual **CO-Dose**, which was determined during pre-examinations, amounted to 1 ml CO per kilogram fat-free mass (FFM) in female participants and to 1.2 ml CO per kilogram fat-free mass in male participants.

In the first minute after starting the ventilation, an arterialized blood sample was taken from a capillary in the ear lobe. It was directly analyzed in a hemoximeter (OSM3 radiometer, Denmark). Further blood samples were taken in minute 3, 5, 7, 9, 11, 13 and 15 and were also analyzed [cf. 10]. The tHb mass was calculated as dilution of the CO bolus, where  $\Delta\text{COHb}$  was the difference between the steady state COHb concentration minus the COHb concentration at rest [9, 11].

### Statistics:

All statistical evaluations were made with the programs SPSS 11.0 (SPSS Inc., Illinois, USA) and GraphPad Prism 4.0 (GraphPad Software Inc., California, USA). The arithmetic mean and the standard deviation were calculated. The correlation and regression analysis was made for the determination of relations and the t-test was administered for the determination of differences in the study group.

## Results

### The entire study group

Table 2 shows the determined anthropometric and hematological measurement parameters in the study group

**Table 2 anthropometric and hematological parameters of the entire study group**

Height (m)	1.75 ± 0.08
Weight (kg)	68.0 ± 8.9
LBM (kg)	55.5 ± 9.1
FM (kg)	12.5 ± 4.4
BCM (kg)	29.5 ± 6.0
TBW (l)	40.8 ± 7.0
BSA (m <sup>2</sup> )	1.81 ± 0.15
tHb (mmol/l)	9.14 ± 0.9
Hct (%)	44 ± 4
tHb-mass (g)	842 ± 226
RCV (ml)	2408 ± 608
PV (ml)	3568 ± 653
BV (ml)	5976 ± 1216

### The relation between hematological measurements (tHb-mass, RCV, PV and BV) and body composition parameters (KG, FFM, BCM, TBW, ECW, BSA) of the participants.

The correlation coefficient for tHb-mass, RCV, PV and BV in respect of KG, FFM, BCM, TBW and BSA always indicates a close connection and is of high significance ( $p < 0.0001$ ).

The closest relation shows the blood volume with the fat-free mass with  $r = 0.92$ . The tHb-mass and the RCV also correlate the most with the fat-free mass (in each case  $r = 0.91$ ). The PV shows the closest connection with the TBW ( $r = 0.88$ ). Of all hematological measurements, the body weight indicates the lowest connection with the body composition parameters.

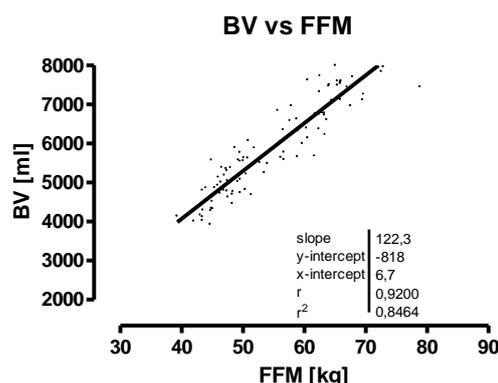
**Table 3 Correlation between hematological and body composition parameters in the study group (n = 100)**

$r$ ( $r^2$ )	tHb-mass (g)	RCV (l)	PV (l)	BV (l)
<b>KG (kg)</b>	0.798 (0.64)	0.796 (0.63)	0.830 (0.69)	0.844 (0.71)
<b>FFM (kg)</b>	<b>0.908 (0.82)</b>	<b>0.908 (0.82)</b>	0.868 (0.75)	<b>0.920 (0.85)</b>
<b>BCM (kg)</b>	0.885 (0.78)	0.884 (0.78)	0.823 (0.68)	0.884 (0.78)
<b>TBW (l)</b>	0.895 (0.80)	0.897 (0.80)	<b>0.876 (0.77)</b>	0.919 (0.84)
<b>BSA (m<sup>2</sup>)</b>	0.829 (0.67)	0.828 (0.69)	0.865 (0.75)	0.878 (0.77)

Figure 1 shows the regression between the blood volume and the fat-free mass. The regression line was calculated as follows:

$$BV (l) = 0.122 LBM (kg) - 0.818$$

As a result, the blood volume in the study group amounts to approx. 0.106 l per kg LBM.



**Figure 1 Correlation and regression of the blood volume with the fat-free mass in the study group (n = 100)**

### Gender-specific results

Table 4 shows the absolute measurements of the hematological and anthropometric parameters separately for men and women. All measurement parameters exceed the significance threshold in the gender comparison. Male participants had an average hematocrit of 46.4 % with a hemoglobin concentration of 9.8 mmol/l. Female participants presented a hematocrit of 41.9 % with a respective hemoglobin concentration of 8.6 mmol/l. The total hemoglobin mass amounted to 1015 g in male participants and to an average of 637 g in female participants. Resulting from this, the total blood volume in women amounted to 4952 ml and to 6846 ml in men.

Compared to the parameters body weight, fat-free mass, body cell mass, total body water, extra cellular water and body surface, the fat mass is the only measured parameter that was not significantly higher in men than in women, but significantly lower.

**Table 4 Hematological and anthropometric measurements of the study group separated by gender**

	Male n = 54	Female n = 46	P<
tHb (mmol/l)	9.8 ± 0.8	8.6 ± 0.7	0.0001
Hct (%)	46.4 ± 3	41.9 ± 3	0.0001
tHb-mass (g)	1015 ± 152.5	637 ± 78.0	0.0001
RCV (ml)	2873 ± 408	1861 ± 227	0.0001
PV (ml)	3973 ± 551	3091 ± 378	0.0001
BV (ml)	6846 ± 899	4952 ± 568	0.0001
Weight (kg)	73.20 ± 7.5	62.01 ± 6.09	0.0001
LBM (kg)	62.77 ± 5.75	47.01 ± 2.83	0.0001
FM (kg)	10.27 ± 3.24	15.00 ± 4.25	0.0001
BCM (kg)	34.20 ± 3.61	24.01 ± 2.25	0.0001
TBW (l)	46.12 ± 4.98	34.68 ± 2.53	0.0001
BSA (m <sup>2</sup> )	1.91 ± 0.12	1.70 ± 0.09	0.0001

\* Mean value and standard deviation in the study group

### A gender comparison of standardized blood parameters.

Table 5-8 show the mean values for the total hemoglobin mass (tHb-mass), the erythrocyte volume (RCV), the blood volume (BV) and the plasma volume (PV), calculated from the difference of BV and RCV). All of them have been standardized through the body weight (KG), the fat-free mass (FFM), the body cell mass (BCM), the body water (TBW) and the body surface (BSA). The values are separated by gender.

The standardized hemoglobin mass and the relative erythrocyte volumes show a significant difference between genders for all investigated parameters (Table 5 and 6). Male participants have significantly higher hemoglobin masses and higher erythrocyte volumes per kg body weight, kg fat-free mass and kg body cell mass than female participants. With regard to the body surface and the hydration parameters the same result is to be seen.

Based on the body weight of the male participants, the plasma volume (Table 7) is significantly higher than the one of the female participants. In relation to the fat-free mass and the body cell mass, female participants present a significantly higher plasma volume than men. However the plasma volume in relation to the body water does not differ between genders. The plasma volume in relation to the body surface is significantly higher in male participants than in female participants.

**Table 5 Mean value of the total hemoglobin mass standardized according to parameters of the body composition and separated by gender.**

	Male (n = 54)	Female (n = 46)	p<
tHb-mass (g) / KG (kg)	13.9 ± 1.6	10.3 ± 1.0	0.0001
tHb-mass (g) / FFM (kg)	16.1 ± 1.8	13.5 ± 1.3	0.0001
tHb-mass (g) / BCM (kg)	29.7 ± 3.6	26.6 ± 3.0	0.0001
tHb-mass (g) / TBW (l)	22.0 ± 2.5	18.4 ± 1.7	0.0001
tHb-mass (g) / BSA (m <sup>2</sup> )	530.8 ± 65.4	373.9 ± 34.6	0.0001

\* Mean value and standard deviation in the study group

**Table 6 Mean values of the erythrocyte volume standardized according to parameters of the body composition and separated by gender.**

	Male (n = 54)	Female (n = 46)	P<
RCV (ml) / KG (kg)	39.3 ± 4.4	30.1 ± 2.9	0.0001
RCV (ml) / FFM (kg)	45.6 ± 4.8	39.5 ± 3.7	0.0001
RCV (ml) / BCM (kg)	83.9 ± 9.5	77.8 ± 8.5	0.001
RCV (ml) / TBW (l)	62.2 ± 6.4	53.6 ± 5.0	0.0001
RCV (ml) / BSA (m <sup>2</sup> )	1501.7 ± 173	1091.5 ± 103	0.0001

\* Mean value and standard deviation in the study group

**Table 7 Mean value of the plasma volume standardized according to parameters of the body composition and separated by gender.**

	Male (n = 54)	Female (n = 46)	P<
PV (ml) / KG (kg)	54.4 ± 5.3	49.9 ± 4.9	0.0001
PV (ml) / FFM (kg)	63.1 ± 5.6	65.6 ± 6.1	0.036
PV (ml) / BCM (kg)	116.1 ± 11.5	129.2 ± 15.0	0.0001
PV (ml) / TBW (l)	86.0 ± 7.5	89.0 ± 7.8	0.057ns
PV (ml) / BSA (m <sup>2</sup> )	2076 ± 210	1812 ± 172	0.0001

\* Mean value and standard deviation in the study group

Male participants present (93.7 ml/kg) significantly higher blood volumes per kg body weight than female participants (80.0 ml/kg). If the blood volume is seen in relation to the fat-free mass and the body cell mass, there are no significant differences between genders. By dividing the blood volumes by the hydration parameters and the body surface, there are significantly higher values in male participants than in women.

**Table 8 Mean value of the blood volume standardized according to the body composition parameters and separated by gender.**

	Male (n = 54)	Female (n = 46)	P<
BV (ml) / KG (kg)	93.7 ± 8.5	80.0 ± 6.8	0.0001
BV (ml) / FFM (kg)	108.8 ± 9.0	105.2 ± 8.4	0.0433
BV (ml) / BCM (kg)	200.1 ± 18.5	206.7 ± 21.5	0.09 ns
BV (ml) / TBW (l)	148.3 ± 11.8	142.7 ± 10.9	0.017
BV (ml) / BSA (m <sup>2</sup> )	3578 ± 340	2903 ± 239	0.0001

\* Mean value and standard deviation in the study group

### Relationships between hematological and anthropometric parameters in the study group; separated by gender.

**Table 9 Correlation between hematological and body composition parameters separated by gender.**

Male/ Female	tHb-mass (g)	RCV (l)	PV (l)	BV (l)
KG (kg)	0.65 / 0.63	0.64 / 0.62	0.73 / 0.65	0.74 / 0.69
FFM (kg)	<b>0.68</b> / 0.64	0.68 / <b>0.66</b>	0.71 / 0.48	<b>0.79 / 0.76</b>
BCM (kg)	0.64 / 0.45	0.63 / 0.48	<b>0.79</b> / 0.73	0.72 / 0.52
TBW (l)	<b>0.68</b> / 0.63	<b>0.69</b> / 0.63	0.78 / <b>0.74</b>	<b>0.79 / 0.76</b>
BSA (m <sup>2</sup> )	0.65 / <b>0.67</b>	0.65 / 0.65	0.77 / 0.71	0.77 / 0.74

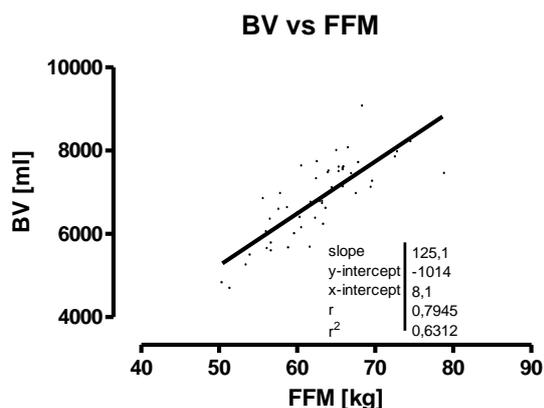


Figure 2 Correlation and regression of the blood volume with the fat-free mass of male participants (n = 54)

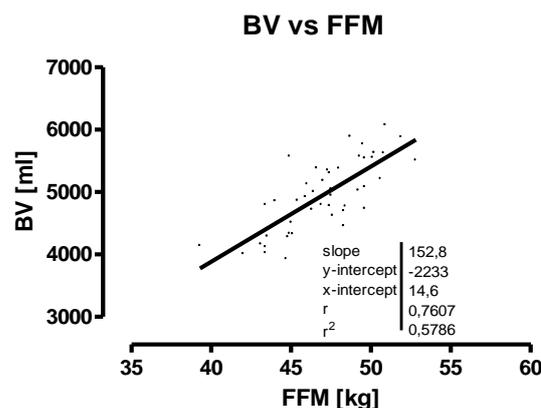


Figure 3 Correlation and regression of the blood volume with the fat-free mass of female participants (n = 46)

## Discussion

In this study, connections between hematological measurements and parameters of the body composition were investigated in general but also separately for men and women.

The comparison of the blood and erythrocyte volumes and the total hemoglobin mass, which were determined by us, with statements made in literature, shows great coincidences with recent studies [11, 3, 12, 13, 14, 15]. Previous studies [2, 6] that partially used radioactive determination methods reported about lower hematological measurement values.

The absolute values of the blood parameters and the body composition are strongly gender-dependent. Without exception, male subjects have most significantly higher hematological measurement values (BV, RCV, PV, tHb-mass) than female subjects. The values of the body composition (KG, FFM, BCM, TBW, BSA) are also most significantly lower in women than in men. In contrast to the other body composition parameters, only the fat mass (FM) is significantly higher in females than in males.

In the total study group, the correlation analyses carried out between the body composition and blood parameters show the highest connections between the fat free mass (FFM) and the blood volume (BV) or more specifically the hemoglobin mass (tHb-mass). Separated by gender, the highest correlations could be detected between BC and FFM, too. These results seem to be reasonable because blood carries oxygen and the muscle mass, which mainly determines the FFM, is the main consumer of oxygen during physical strain. Participants with a high amount of muscle mass probably have the highest BV per kg body weight (KG) and the highest amount of tHb-mass per kg KG. Ahlgrim 2010 [17] could determine that the blood parameters, which were standardized with the KG, significantly correlate with the body fat content. That means that a high FM is associated with a small BV per kg KG. The KG, and in some cases the body surface (BSA), is still used for the comparison of blood parameters [1, 4, 11, 13, 18, 19, 20]. However, KG and BSA reflect the body composition only insufficiently [17], especially because the amount of avascular adipose

tissue in KG and BSA does not become clear. In subjects with the same KG or same BSA, FM and FFM can be completely different. The standardization of the blood parameters with the KG and BSA does not correspond with the blood amount/hemoglobin amount per muscle mass.

Similar high connections, as those of the FFM with the hematological parameters, could be determined between the total body water (TBW) and the hematological parameters. In the examined parameters of the body composition with the blood parameters, the lowest correlation coefficients were, with only few exceptions, detected in the KG.

These results clarify the use of the FFM as anthropometric reference for the blood composition parameters [5, 6, 7, 8]. At the same time, the large scatter range of the measurement values shows that a direct measurement of the blood parameters needs to be preferred over the indirect determination with body composition parameters. Studies on sportsmen describe a high correlation between BV, RCV or tHb-mass and parameters of the aerobic performance [11, 21]. Martino et al. 2002 [14] observed that the BV correlated with the endurance performance in untrained participants, too. This indicates a substantial genetic predisposition of the BV that is similar to the assumption of Schmidt und Prommer 2008 [22]. In conclusion, the investigation of Martino, Gledhill and Jamnik 2002 [14] recommended the BV as parameter for the recognition of talents of endurance sports. At the same time, endurance performance is strongly gender-dependent. Practically, this fact shows itself in the higher oxygen uptake of males as compared to females [23]. On the average, maximum oxygen uptake in women is 10 – 20 % lower than the one of men [23, 24]. Regarding the running performance at distances of 3 000, 5 000 and 10 000 m (high amount of aerobic energy supply) in gender comparison, the world record time of women is between 10 % and 12 % higher than the one of men. The results of the standardized BV per FFM only show slightly higher values for males than for females. The values of men and women do not differ by relating the BV to the body

cell mass (BCM). At least for the BCM, female and male participants have similar hematological conditions. The blood composition of female participants negatively affects the oxygen transport capacity. The significantly lower values for the tHb-mass (absolute and standardized) and the RCV (absolute and standardized) in women show this very descriptively. The deriving lower hemoglobin concentration in women expresses this, too. So, women can transport less oxygen per heartbeat although the cardiac output per minute is theoretically the same. A further point additionally limits the performance

of female subjects in relation to male subjects. Female participants have significantly higher fat values than males, both absolutely and relatively. In all kind of sports in which KG and consequently FM directly affect the performance, men benefit from the low relation between body weight and muscle.

Among others, the gender-dependent, different relative oxygen uptake may be explained by the low fat content in men and the low hemoglobin concentration in women. Other factors such as the cardiac output may play a major role here, too.

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