Hemoglobin, Red Blood Cell Count, Hematocrit and Derived Parameters for Diagnosing Anemia in Elderly Males

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Abstract: Anemia is one of the most common micronutrient deficiency in our community. Nutritional anaemias are caused when there is an inadequate body store of a specific nutrient needed for hemoglobin synthesis. The most common nutrient deficiency is of iron. Therefore, a cross-sectional survey was conducted on the healthy elderly male, aged ≥ 40 and ≤77 years (n=60) volunteers in order to assess their blood parameters, such as hemoglobin concentration (Hb), hematocrit (HCT), red blood cell count (RBC), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) for the diagnosis of anemia. The demographic results showed mean values  (50.10±8.79) years for age, 66-68 ± 1.95 inches for height , 71.43 ± 6.43 kg body weight, 98.34±0.48 °F body temperature, 124 ± 8.67 systolic blood pressure, 82.17 ± 4.15 diastolic pressure while, The pulse rate was found to be  74.63 ± 7.02/minute. Similarly, mean values for lean body weight (LBW) found to be 49.9±2.89, ideal body weight (IBW) 60.9 ± 4.49, body surface area (BSA) was 1.8 ± 0.1 m² whereas, body mass index (BMI) showed mean value 24.9 ± 2.6 kg/m². More so, overall mean Hb found to be 13.60 g/dl, RBC 4.6 mill/mm³, HCT/PCV 43%, MCV 92.95fl, MCH 29.42 pg and MCHC was found to be 31.73 g/dl. The normal range of Hb for men was 13-17 g/dl and 31.67% of the subjects participated in the study was considered to be anemic showing less Hb than normal range. The volunteers were suggested to improve the dietary habits and to take iron supplements in order to overcome the iron deficiency anemia.

Keywords: Hemoglobin, red blood cells, anemia, dietary habits, elder age

1. INTRODUCTION

The human body can survive three weeks without food, three days without drinking, but can’t survive even for three minutes without oxygen. Sufficient oxygen to each cell in the body is the basis of life itself. Iron is essential for life, as it is the agent that carries oxygen. Oxygen provides the energy the body needs for all of its normal activities. Only red blood cells are capable of carrying oxygen to cells. Each red blood cell contains between 200 and 300 hemoglobin molecules which bind with oxygen. The normal hemoglobin values range from 14-18 g per deciliter of blood for men and 12-16 g for women [1]. The normal hematocrit is 42-54% for men and 36-48% for women [1]. Anemia occurs when the number of red blood cells (or the Hb in them) fall below normal and the body gets less oxygen and therefore has less energy than it needs to function properly [2]. Anemia is a great problem globally and worse in developing countries, like ours, but by no means is absent in industrialized nations of the world. Anemia can occur from a malfunction at any point in the production, recycling or regulating of red blood cells in the body.

Anemia is not a single disease but is a condition, like fever, with many possible causes and many forms. The normal red blood cell count ranges from 4.2-5.4 million/mm³ for males and for women about...
Red blood cell indices include Mean Corpuscular Volume (MCV), Mean Corpuscular Hemoglobin (MCH) and Mean Corpuscular Hemoglobin Concentration (MCHC). MCV is the mean volume of all the erythrocytes counted in the sample. The value is expressed in volume units, femtoliters (fL). The normal range is 80-100 fL. When MCV is low, the blood is said to be microcytic and when it is high blood is said to be macrocytic. Normocytic refers to blood with a normal MCV [3]. Mean Corpuscular Hemoglobin (MCH) means the average weight of hemoglobin per cell. Normal values are 27-33 picograms. When MCH is low blood is said to be hypochromic and when it is high the blood is said to be hyperchromic [3]. Mean Corpuscular Hemoglobin Concentration (MCHC) the average weight of hemoglobin per cell per its volume. Its normal values are 32-36 g/dL [3].

2. MATERIALS AND METHODS

The present study was planned to measure hemoglobin, hematocrit and red blood cells count in elder males. The males were selected at random for collection of blood samples determinant of anemia and its relation with the dietary habits in this group of subjects.

2.1 Collection of Blood Samples

Sixty blood samples were collected from male volunteers aged above 40 years. The blood was collected from the left arm vein using disposable syringes for each subject. About 3ml of blood was drawn from each subject. The blood was taken into centrifuge tubes heparin containing as an anticoagulant.

2.2 Anthropometric/Demographic Measurements

The data of height and weight of each subject was measured to calculate the parameters including lean body weight (LBW), ideal body weight (IBW), body mass index (BMI) and body surface area (BSA) by using formulae given by [4]. The following parameters were then studied:

Physical parameters which included age (years), body temperature (F), height, weight and blood pressure. Similarly, dietary intake of the volunteers under study was made through food frequency questionnaire. The volunteers were presented with a list of foods and were interviewed to assess how often each of common foods eaten well made on the basis of data thus obtained.

2.3 Hemoglobin Determination

N/10 HC1 was taken into an ordinary pipette and was poured in the graduated dilution tube up to 20% mark. The heparinized blood was filled into the hemoglobin pipette up to 0.02 ml and transferred it into the dilution tube. The blood and HCl were stirred in the dilution tube with the stirrer. Distilled water was added until the colour of the dilution and standard tubes matched with each other. The reading was noted which gave hemoglobin as g/dl of blood [5].

2.4 Red Blood Cell Count

For RBC counting blood with an anticoagulant was used. Blood was drawn into the RBC diluting pipette exactly to the 0.5 mark, using gentle suction on the mouth piece. The lip of the pipette was wiped free of blood before inserting it in to the diluting fluid (Toission Solution). The diluting fluid was drawn up to the mark 101 above the bulb. The tube was rotated in a horizontal position to ensure uniform dispersion of the blood cells in the pipette [5].

RBCs were calculated by using the following formula:

\[
\text{RBC (million/mm)} = \frac{\text{Cells counted} \times 10 \times 200}{5}
\]

2.5 Packed Cell Volume (PCV)

Packed cell volume was measured using the heparinized blood in the plain capillary tubes (75 mm x 1 mm). Tubes were filled approximately 1 cm from the end. Holding it in the flame sealed the vacant ends of the tubes. Care was taken not to heat the blood. Capillary tubes were fixed in the hematocrit centrifuge machine. Then centrifugation was done at 13000 rpm for 5 minutes [5].

2.6 Red Blood Cell Indices

From the values of PCV, Hb and RBC count following useful erythrocyte indices were
empirically calculated.

2.7 Mean Corpuscular Volume (MCV)

MCV expresses the average volume of the individual RBC and is calculated from the formula as given by Wintrobe [5] and Diem and Clenter [6].

\[ MCV = \frac{\text{Hematocrit} \times 10}{\text{R.B.C.}} \]

MCV is expressed in femtoliter.

2.8 Mean Corpuscular Hemoglobin (MCH)

MCH is the amount of hemoglobin by weight in average Red blood cell count and is calculated by the formula as given by Wintrobe [5] and Diem and Clenter [6].

\[ MCH = \frac{\text{Hemoglobin} \times 10}{\text{R.B.C.}} \]

It is expressed in picogram.

2.9 Mean Corpuscular Hemoglobin Concentration (MCHC)

MCHC is the concentration of hemoglobin in the average red blood cells or the ratio of weight of hemoglobin to the volume in which it is contained and is calculated from the formula as given by Wintrobe [5] and Diem and Clenter [6].

\[ MCHC = \frac{\text{Hemoglobin} \times 100}{\text{Hematocrit}} \]

2.10 Data Analysis

The data regarding demographic parameters, hemoglobin, hematocrit, red blood cell count, MCV, MCH, and MCHC, were statistically analyzed by regression/correlation analysis by using Microsoft excel program in Computer.

3. RESULTS

The study was designed to measure hemoglobin concentration, hematocrit and red blood cell count for the diagnosis of anemia in elderly males above 40 years of age to assess any correlation between these parameters. The results of the demographic and blood parameters are given below.

3.1 Demographic and Anthropometric Parameters

The mean values for age (50.10), height (66.68), actual and predicted body weight, the blood pressure, pulse rate (74), body temperature, BSA (1.8), LBW (49.9), IBW (60.9) and BMI of the elderly male volunteers who participated in the study on the hemoglobin (13.60), hematocrit and red blood cell count (4.65) for diagnosis of anemia in elderly males above 40 years of age are presented in Table 1 and Table 2.

3.2 Complete Blood Count

The blood samples were collected from 60 healthy elderly male volunteers for the study of hemoglobin, hematocrit, and red blood cells count, MCH (29.42), MCV (92.95), and MCHC for the diagnosis of presence or absence of anemia and

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Age (Year)</th>
<th>Weight (kg)</th>
<th>Height (inch)</th>
<th>Sys. (mmHg)</th>
<th>Dis. (mmHg)</th>
<th>Temp. (°F)</th>
<th>Pulse / min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>50.10</td>
<td>71.43</td>
<td>66.68</td>
<td>124</td>
<td>82</td>
<td>98.34</td>
<td>74</td>
</tr>
<tr>
<td>± SD</td>
<td>8.79</td>
<td>6.43</td>
<td>1.95</td>
<td>8.67</td>
<td>4.2</td>
<td>0.48</td>
<td>7.0</td>
</tr>
<tr>
<td>Minimum</td>
<td>40.00</td>
<td>57</td>
<td>63</td>
<td>100</td>
<td>80</td>
<td>97</td>
<td>68</td>
</tr>
<tr>
<td>Maximum</td>
<td>77.00</td>
<td>87</td>
<td>69</td>
<td>140</td>
<td>90</td>
<td>99</td>
<td>120</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Parameter</th>
<th>BSA (m²)</th>
<th>BMI (kg/m²)</th>
<th>IBW (kg)</th>
<th>LBW (kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>1.8</td>
<td>25</td>
<td>60.9</td>
<td>49.9</td>
</tr>
<tr>
<td>± SD</td>
<td>0.1</td>
<td>2.6</td>
<td>4.5</td>
<td>2.9</td>
</tr>
<tr>
<td>Minimum</td>
<td>1.61</td>
<td>20.64</td>
<td>50.10</td>
<td>43</td>
</tr>
<tr>
<td>Maximum</td>
<td>1.96</td>
<td>32.66</td>
<td>66.20</td>
<td>55</td>
</tr>
</tbody>
</table>
results are presented in Table 3.

### 3.3 Regression Correlation

The correlation between Hb and RBC has been presented in Fig. 1 which shows statistically significant relationship between these two parameters. The value of regression coefficient was \( R^2 = 27 \) which was found to be statistically significant \((P < 0.05)\). Similarly, correlation between Hb and PCV was also carried out as presented in Fig. 2. Both the parameters show highly significant positive relationship with the regression coefficient value of \( R^2 = 0.80 \). Moreover, correlation between Hb, MCV and MCH was also checked and the Fig. 3 showed that regression coefficient \( R^2 = 0.14 \) was non-significant between Hb and MCV \((P < 0.05)\). However, relationship between Hb and MCH was significant with the value of regression coefficient \( R^2 = 0.32 \) \((P < 0.05)\). Furthermore, correlation between Hb and MCHC (Fig. 4) was evaluated. The value of regression coefficient \( R^2 = 0.10 \) depicted non-significant relationship between Hb and MCHC \((P < 0.05)\).

As for as correlation between RBC and PCV is concerned (Fig. 5), it was found to be statistically non-significant as the value of regression coefficient was \( R^2 = 0.20 \). A similar correlation between RBC, MCV and MCH (Fig. 6) were observed which were found to be non-significant. The values of regression coefficient were \( R^2 = 0.24 \) and \( R^2 = 0.16 \), respectively. In Fig. 7, correlation between RBC and MCHC is presented that also showed non-significant relationship as the regression coefficient value is \( R^2 = 0.04 \) so \((P < 0.05)\). On contrast to the above, correlation between PCV, MCV and MCH (Fig. 8) showed significant relationship between PCV and MCV as the value of regression coefficient was \( R^2 = 0.31 \) and \( R^2 = 0.27 \) respectively. Finally, the correlation between PCV and MCHC were carried out which were presented in Fig. 9. The regression coefficient value \( R^2 = 0.02 \) showed non-significant relationship between these two parameters.

### 4. DISCUSSION

The present study was conducted on the elderly male (above 40 years of age) volunteers in order to assess their Hb, HCT, RBC, MCV, MCH and MCHC for the diagnosis of anemia in elderly males. A total of 60 volunteers participated in the study. The demographic parameters of the volunteers were recorded and blood samples were drawn from each volunteer. The results of the demographic and blood parameters are discussed below.

#### 4.1 Demographic / Anthropometrics Parameters

The volunteers had the mean \pm SD values for age 50.10 \pm 8.79 years, ranged from 40-77 years. Their height was 66-68 \pm 1.95 inches ranging between 62-69 inches and the body weight was 71.43 \pm 6.43 kg with a range of 57-87 kg. The mean \pm SD value for the body temperature was 98.34 \pm 0.48 F ranged from 97-99 F. Systolic blood pressure was 124 \pm 8.67 mmHg and varied between 100-140 mmHg, while diastolic was 82.17 \pm 4.15 mmHg with a range of 80-90 mmHg. The pulse rate was 74.63 \pm 7.02 / minute and varied from 68-120/ minute. The lean body weight (LBW) showed mean \pm SD value 49.9 \pm 2.89, ranged from 42.79 – 54.76 kg. The mean \pm SD value for ideal body weight (IBW) was 60.9 \pm 4.49, ranged between 50.10-66.20 kg. The mean \pm SD value for the body surface area (BSA) was 1.8 \pm 0.1 m\(^2\) and ranged between 1.61- 1.96 m\(^2\). The body mass index (BMI) showed mean \pm SD value of 24.9 \pm 2.6 Kg/m\(^2\) varying between 20.64- 32.66 Kg/m\(^2\). All the volunteers participated in the study.

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### Table 3

Mean hemoglobin (Hb), hematocrit or packed cell volume (PCV), red blood cell count (RBC), mean corpuscular hemoglobin (MCH), mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC) of male volunteers.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Hb (g/dl)</th>
<th>PCV (%)</th>
<th>RBC (mil/mm(^3))</th>
<th>MCH (Pg)*</th>
<th>MCV (Fl)**</th>
<th>MCH (g/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>13.60</td>
<td>42.90</td>
<td>4.65</td>
<td>29.42</td>
<td>92.95</td>
<td>31.73</td>
</tr>
<tr>
<td>± SD</td>
<td>1.71</td>
<td>5.04</td>
<td>0.50</td>
<td>3.48</td>
<td>11.49</td>
<td>1.79</td>
</tr>
<tr>
<td>Minimum</td>
<td>10.10</td>
<td>31</td>
<td>3.50</td>
<td>21.96</td>
<td>67.39</td>
<td>23.40</td>
</tr>
<tr>
<td>Maximum</td>
<td>17.70</td>
<td>53</td>
<td>5.80</td>
<td>41.14</td>
<td>128.57</td>
<td>36.45</td>
</tr>
</tbody>
</table>

*Pg = Picogram/cell  
**fl = Fematoliter.
Fig. 1. Relation between Hb and RBC of male volunteers.

Fig. 2. Relationship between Hb and PCV of male volunteers.
Fig. 3. Relationship between Hb, MCV and MCH of male volunteers.

Fig. 4. Relationship between Hb and MCHC of male volunteers.
Fig. 5. Relationship between RBC and PCV of male volunteers.

Fig. 6. Relationship between RBC, MCV and MCH of male volunteers.
Fig. 7. Relationship between RBC and MCHC of male volunteers.

\[ y = 0.7278x + 28.346 \\
R^2 = 0.042 \]

Fig. 8 Relationship between PCV, MCV and MCH of male volunteers.

\[ y = 1.2737x + 38.312 \\
R^2 = 0.3123 \]
\[ y = 0.3608x + 13.951 \\
R^2 = 0.2729 \]

Fig. 9 Relationship between PCV and MCHC of male volunteers.

\[ y = -0.0498x + 33.863 \\
R^2 = 0.0198 \]
were healthy and having no sign of any disease or disorder.

4.2 Complete Blood Count Parameters

Anemia, one of the more common blood disorders, occurs when the number of healthy red blood cells decreases in the body. The nutritional or hypochromic anemia is common and in chronic blood loss where iron or protein intakes are in adequate [7]. The overall mean concentration for Hb was 13.60 g/dl which showed that in Pakistan, elder males above 40 years of age have Hb levels between the normal values i.e. 13-17 g/dl [1]. As [8] evaluated prevalence of anemia and degree of correlation of aging with changes in Hb level in a healthy, elderly population (age range, 60 to 96 years; mean age, 70 years). They reported that twelve percent (12%) of participants were anemic overall, although there was a gender difference; more males (17.7%) were anemic than females (8.4%). Mean Hb level did not change significantly with age group except in males over 85 years of age. Our results, also found same as reported by [8]. Aging is one the important factor that affects RBCs, Hb and HCT [9]. Our findings were also proved to be more or less same. Two groups, less than 75 years and more than 75 years of age showed Hb 11.0 g/dl in group 1 and Hb 10.0 g/dl in group 2. So they were considered as anemic. Our results also show some tendency of decrease in Hb levels with aging and 31.6667% of the subjects participated were showing low Hb levels.

The overall mean RBC value in elderly male volunteers above 40 years of age was found to be 4.6 mill/mm³ which is between the normal limits i.e. 4.2-5.4 mill/mm³. However, 20% of the subjects participated in the study showed RBC count below the normal limits. As [10] reported that irrespective of subject age, Hb, Hct, RBC, MCV, MCH, MCHC were higher in males than in females, but sex-related differences tended to decrease with aging. In males, there was a trend toward lower Hb, and Hct with aging, due to an increase in RBC, MCV, and MCH. The overall HCT or PCV in elderly male volunteers above 40 years of age was estimated to be 43% and ranged between 31-53% which is between the normal limits i.e. 40-54% [1]. Nozaki et al [11] defined the range of normal hematocrit and hemoglobin levels in residents of high altitude is required to diagnose chronic mountain sickness (CMS) and other conditions defined, in part, by hematocrit or hemoglobin values. Male hematocrit averaged 52.7% and hemoglobin averaged 17.3 m/dl whole blood. The corresponding female values were 48.3% and 15.8 g/dl whole blood, respectively. The range of normal values was 45% to 61% for hematocrit and 13 to 21 g/dl for hemoglobin in the men and 41% to 56% for hematocrit and 12 to 19 g/dl for hemoglobin in the women. These data indicate that hematocrit values above 61% in men or 56% in women and hemoglobin values above 21 g/dl whole blood in men or 19 g/dl whole blood in women are outside the normal range.

The overall MCV mean value in elderly male volunteers above 40 years of age was 92.95 fl with 10% showing lower MCV levels and 20% showing high levels than normal limits i.e. 80-100 fl. So in our study, 10% of the volunteers were considered as microcytic anemic and 20% as macrocytic anemic which is in collaboration with [3].

Alian et al. (1997) studied the range of full blood count (FBBC), vitamin B12 and folate levels and determined the prevalence of occult hematological abnormalities in older Zimbabweans. Healthy Zimbabweans aged > 65 years were randomly selected. The median Hb was males 14.0 (range 8 to 18.3), females 13.1 g/dl (7.9 to 18.1). 23% were anemic (Hb < 13 g/dl in males, < 12 g/dl in females), 3% with microcytic and 20% with macrocytic indices. Overall 13% had low vitamin B12 and 30% had low folate levels. Folate levels were significantly lower in urban subjects and B12 levels were significant lower in rural subjects. Fifty-four subjects (21%) had an MCV > 100 fl. In this group, low folate levels were found in 22, low B12 levels in nine, excessive alcohol in eight and two subjects had elevated TSH. The MCV was higher in urban subjects. This study has revealed a large amount of occult hematological abnormally and interesting differences between rural and urban subjects. It focuses attention on low levels of folate, which should be preventable by simple nutritional education, as an extensive problem in the community.

The overall mean value for MCH in elderly male volunteers (above 40 years of age) was 29.42
and 27% of the subjects participated in the study showed lower levels while 21% showed high levels then the normal limits i.e. 27-33pg. In the present study, 27% of the subjects were considered as hypochromic anemic and 21% were considered as hyperchromic anemic. Similar results were also reported by [3]. The overall MCHC mean value in elderly male volunteers was 31.73 g/dl, ranged between 23.40-36.45 g/dl and 50% were showing lower MCHC values then the normal limits i.e. 32-36 g/dl. As [12] established the normal ranges in RBC parameters for healthy centenarians is necessary for diagnostic criteria of anemia and they found MCHC=32.1 +/- 1.1 g/dl.

5. REFERENCES