Is it necessary to consider obesity when constructing norms for hemoglobin or when screening for anemia using hemoglobin levels?

Fahaid H. Al-Hashem, MBBS, PhD (UK).

ABSTRACT

Objectives: To assess the relationship of total adiposity and abdominal adiposity on hemoglobin levels in Saudi nonpregnant women.

Methods: We carried out this cross-sectional study during winter of 2002 (from January to March) in and around Abha city, Kingdom of Saudi Arabia. Included in the study were 530 non-pregnant women between the age of 18-65 years. Body weight was measured using an Avery Beam weighing scale, while height was measured using a stadiometer and waist circumference using a fiberglass. Hemoglobin levels were estimated using cyanmethemoglobin method. Total obesity was defined as body mass index (BMI) ≥30 and abdominal obesity as WC >88 cm.

Results: The mean and median hemoglobin levels were significantly higher in abdominally obese women compared with totally obese ($p<0.04$ versus <0.02) and non obese ones ($p<0.04$ versus <0.03). No significant differences in the mean and median hemoglobin levels were observed when abdominally obese women were compared with both abdominally and totally obese ones ($p<0.7$ for both). The mean and the median hemoglobin levels were virtually identical in non-obese and totally obese women. Statistical analysis showed that the mean hemoglobin level was positively and significantly associated with WC ($p<0.005$) and negatively and insignificantly associated with BMI ($p<0.8$).

Conclusion: In view of the positive and significant association between abdominal obesity and the mean hemoglobin level in this population, abdominal obesity should be considered when constructing norms for hemoglobin or when screening for anemia using hemoglobin levels.

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Address correspondence and reprint request to: Dr. Fahaid H. Al-Hashem, Chairman, Department of Physiology, College of Medicine, King Khalid University, PO Box 641, Abha 61421, Kingdom of Saudi Arabia. Fax. +966 (7) 2264438. E-mail: fahaid999@yahoo.com

The association of hemoglobin levels with obesity has been the subjects of many studies.[1-7] Thus, while Scheer and Guthrie1 found no significant differences in hemoglobin levels between lean and obese children as defined by skinfold thickness, Garn and Ryan[2] using the same data and the same indicator for obesity found significant differences in hemoglobin levels between lean and obese infants, children, adolescents and adults. The difference between the 2 conclusions may be attributed to the criteria by which the triceps skinfold thickness was used to define obesity. Studies were further extended into the pregnancy period. Hemoglobin levels were found to be significantly higher in obese pregnant women compared to lean pregnant ones.[3] Thus, despite the hemodilution which occurs during pregnancy the fatness effect was manifest. Subsequent studies in different population confirmed the original findings of the earlier ones in the field. Significantly higher levels of hemoglobin were found in overweight than in control adults[4] and in other studies hemoglobin levels were shown to increase with increase in body mass index (BMI) in children[5] and adults[6] and with body weight in adults.[7] It appears therefore, that there is a general agreement that obesity is associated with increase in the hemoglobin level. However, the magnitude and the significance of this association remained a point of contradiction between different investigators. Thus, while some investigators,[8] recommended that higher adjusted cut-off values of hemoglobin levels be used for obese individuals in screening for anemia, others reported that exclusion of obese individuals has virtually no effect on ranges or mid points of hemoglobin levels in different races.[1,9] However, most of the previous investigators, if not all, used either skinfold thickness or BMI as an indicators for total obesity and to my knowledge no previous study had looked specifically into the effect of fat distribution on hemoglobin level at least

in the Kingdom of Saudi Arabia. The present study was therefore undertaken to determine the relationship of abdominal adiposity and total adiposity on hemoglobin levels in non-pregnant women ranging in age from 18-65 years. The objective was to see whether it is necessary to consider fat distribution when constructing norms for hemoglobin in this population or when screening for anemia using hemoglobin levels.

Methods. This study was carried out during the winter of 2002 (from January to March) in and around Abha City. Abha is the capital of Aseer region, which lies in the Southwestern part of the Kingdom of Saudi Arabia. Five primary health care centers were selected for this study, one in central Abha and 4 around it. The primary health care centers belong to the Ministry of Health and are run by qualified physicians who use a central referral hospital within easy reach by built roads to manage their difficult cases.

A total of 530 non-pregnant women with age ranging from 18-65 years were involved in this study, which was a part of a survey on the effect of parity on blood pressure and hypertension. The local people working in the primary health care centers were instructed to recruit all non-pregnant women registered in the 5 health centers. This was carried out by inquiring regarding menstrual history and urine pregnancy test. The inclusion criteria for this study were healthy nonpregnant Saudi women with age ranging from 18-65 years, born and were in permanent residence in the area of study and who did not have diabetes, hypertension, anemia, cardiac and respiratory diseases. Using the World Health Organization manual for sample size determination in health studies at 95% confidence interval with a conservative estimate of the anticipated population proportion of 15% and with an absolute precision of 3%, the minimal sample size required for the study were calculated to be 545 cases.[10] A total of 662 women were recruited (85% of the total number of non-pregnant women registered in the 5 health centers) of whom 132 were excluded as they did not fulfill the criteria for inclusion in this study. Those chosen for the study enjoy modern amenities such as adequate diet (comprising mainly of meat, chicken and rice), potable drinking water and electricity.

For each woman selected, body weight was measured and recorded using an Avery Beam weighing scale to the nearest of 0.1 kg. Subjects were weighed partly dressed and a correction of 0.5 kg was made for clothing. Standing height was measured (without shoes) and recorded to the nearest of 0.1 cm with stadiometer. Body mass index was calculated from the weight and the height (BMI = weight in kg/height in meters square). Waist circumference (WC) was measured midway between the inferior margin of the last rib and the crest of the ilium and recorded to the nearest of 0.1 cm using a fiber-glass tape. Total obesity was defined as BMI ≥30 and abdominal obesity as WC>88 cm.[11] Venous blood samples were collected into heparinized tubes for determination of hemoglobin levels using cyanmethemoglobin method.[12] The basis of this method is to dilute blood in a Darbkin's reagent, which consists of potassium cyanide, potassium ferricyanide and sodium bicarbonate. Hemoglobin, methemoglobin and carboxyhemoglobin are all converted into cyanmethemoglobin. A medium standard was provided and was labeled with the concentration of hemoglobin. The absorbance of the cyanmethemoglobin solution and the medium standard are then measured in a photoelectric colorimeter at a wave length of 540 nm (to give maximum absorbance). The hemoglobin concentration was then computed as follow: Hemoglobin (gm/dl) = (absorbance of the test/absorbance of the standard) × concentration of the standard.

To determine the effect of age on hemoglobin levels, women were divided into 3 age groups (18-29, 30-44 and 45-65 years).

To determine the effect of different types of obesity on hemoglobin levels, subjects were divided into 4 groups: 1. Non-obese (BMI < 30 and WC ≤ 88 cm) 2. Totally obese (BMI ≥ 30 and WC < 88 cm) 3. Abdominally obese (BMI < 30 and WC > 88 cm) 4. Totally and abdominally obese (BMI ≥ 30 and WC > 88 cm). At different stages of the study, the collected data were compiled and fed into an IBM computer. The Statistical Package for Social Sciences package version 10 was used for standard statistical analysis including multiple linear regressions. Unpaired Student's t-test and Man-Whitney U test were used where appropriate to determine statistical significance. A $p<0.05$ was considered statistically significant.

Results. Table 1 shows some of the characteristic of Saudi women by age. The mean weight and calculated BMI increased significantly between the youngest and the middle age groups ($p<0.001$ for both) and insignificantly in the oldest age group ($p<0.3$ and <0.1). The WC increased significantly between the 3 age groups ($p<0.001$ for all) while the mean height increased significantly between the youngest and the middle age groups ($p<0.04$) but declined insignificantly in the oldest age group ($p<0.1$). Displayed in the same table are the mean hemoglobin levels in each age group. No significant differences in the mean hemoglobin levels were observed between the 3 age groups although the mean levels increased by 0.1 gm/dl between the youngest and the middle age groups ($p<0.3$).

Table 2 shows the mean, median and 95% normal range of hemoglobin levels in non-obese and the 3 obesity types. The mean and median hemoglobin levels were significantly higher in abdominally obese compared to totally obese ($p<0.04$ versus <0.02) and non-obese women ($p<0.04$ versus <0.03). When abdominally obese women were compared with both abdominally and totally obese ones, no significant differences in the mean and median hemoglobin levels were observed ($p<0.7$ for both). A striking similarity of the mean and median hemoglobin levels were observed between the totally obese and non-obese women. The lower limit of normal ranges for hemoglobin calculated as 2.5 percentile[8] in abdominally obese was 0.8 gm/dl higher than the totally obese and non-obese and 0.6 gm/dl higher than the both abdominal and totally obese women. However, the upper limit of normal ranges were nearly similar in non-obese and in the other obesity types.

Multiple linear regression analysis was performed to assess the impact of BMI and WC on hemoglobin levels in Saudi non-pregnant women. Age was not entered as there were no significant differences in hemoglobin levels between the 3 age groups. The multiple linear regression model (hemoglobin as function of BMI and WC) was statistically valid and shows positive and significant correlation between hemoglobin levels and WC ($p<0.005$) and negative and insignificant correlations between hemoglobin levels and BMI ($p<0.8$).

Discussion. The results presented in this paper showed that the mean and median hemoglobin levels were significantly higher in abdominally obese women compared with totally obese and non obese ones and no significant differences in the mean and median hemoglobin levels were observed when the abdominally obese women were compared with both abdominally and totally obese ones. However, the mean and the median hemoglobin levels were virtually identical in non-obese

Table 1 - The mean ± standard deviation of weight, height, BMI, WC and hemoglobin levels of Saudi women by age.

Age group (year)	Weight (Kg)	Height (cm)	BMI (Kg/m²)	WC (cm)	Hemoglobin (gm/dl)
18-29 N=283	62.4 ± 13.4	152.4 ± 8.1	27.0 ± 6.5	71.8 ± 15.8	13.5 ± 1.2
30-44 N=193	73.3 ± 14.4	153.8 ± 6.3	31.0 ± 6	85.6 ± 16.1	13.6 ± 1.2
45-65 N=54	75.6 ± 14.1	152.1 ± 5.5	32.7 ± 6.1	92.1 ± 13.7	13.6 ± 1.1
Total N=530	67.7 ± 15	152.9 ± 7.3	29.1 ± 6.6	78.9 ± 17.5	13. 5 ± 1.2

BMI – body mass index, WC – waist circumference.

Table 2 - The mean ± standard deviation (SD), median and 95% normal range of hemoglobin levels in Saudi women by obesity type.

Obesity type	Mean ± SD	Median	95% normal range	
			Lower limit (2.5 percentile)	Upper limit (97.5 percentile)
Non-obese N=275	13.4 ± 1.2	13.4	11.2	16.1
Abdominally obese N =33	13.8 ± 0.9	13.9	12	16
Totally obese N =97	13.4 ± 1.1	13.4	11.2	16
Abdominally and total obese N=125	13.7 ± 1.2	13.7	11.4	16.1

All hemoglobin values are expressed as gm/dl.

and totally obese women. In addition hemoglobin levels in this study sample were significantly and positively correlated with WC and negatively and insignificantly with BMI.

Among various indices, the BMI was selected as an indicator for total obesity. This is due to the fact that the index is easy to derive and apply to all populations without the need of reference population. Furthermore, in this particular population the BMI is significantly and highly correlated with other indices used for estimation of total body fatness.[13] Accurate assessment of abdominal adiposity requires the use of computed tomography and magnetic resonance.[14,15] However, these techniques are expensive and there is a possible risk of radiation and therefore, they are not suitable for large scale epidemiological surveys. Instead of the WC, the waist hip ratio (WHR) was selected as an indicator for abdominal obesity. The WC is easy to measure and the measurement error is low due to the large circumference. In addition, the WC has been endorsed as the best anthropometric surrogate of abdominal adiposity,[15,16] which is highly correlated with visceral adipose tissue accumulation, especially in women.[17,18] The WHR was not selected due to the inherent weakness as a ratio index[19,20] and as it is influenced by pelvic structure.[21]

Previous investigators who found significantly higher hemoglobin levels among obese compared to non-obese used either skinfold thickness[2] or BM[15,7] to quantify total adiposity and it is most likely therefore, that their samples included abdominally obese individuals. However, our findings are at variance with previous studies which showed positive and significant correlation between BMI and hemoglobin levels.[4,5,7] The discrepancy may be due to selection of target study groups in relation to age, gender and race.

The mechanism underlying the relationship between obesity and hemoglobin level is not well established. It has been suggested that obese individuals have higher food consumption and therefore, ingest more iron.[22] However, obesity is known to be more common among individuals of poor socioeconomic status of whom their diet is usually characterized by higher carbohydrate intake and relatively low intake of dietary items rich in iron such as meat.[23] Recently, it has been suggested that Leptin-The OB gene secreted by adipocytes may be responsible for the increase in hemoglobin level seen in obese individuals. In an in vitro study, it has been shown that leptin plus erythropoietin acted synergistically to increase erythroid development.[24] Subsequent epidemiological studies showed statistically significantly higher levels of serum leptin and hemoglobin level in the overweight than in control adults,[4] although other investigators failed to show

significant correlation between leptin and red blood cell count.[25] However, erythropoiesis is known to be regulated by erythropoietin, which in adults secreted by the kidney is response to hypoxia.[26] Leptin is produced mainly by adipocytes but there is no evidence that adipocytes have receptors for hypoxia.

The present study showed no significant relationship between hemoglobin level and total obesity as defined by BMI, but there was a significant relationship between hemoglobin level and abdominal obesity as defined by WC. There are well documented instances where abdominal obesity is associated with certain biochemical changes in some population groups such as hyperinsulinemia[27,28] and hyperlipidemia[27-29] independently of total adiposity and there is no reason not to believe that abdominal adiposity is associated with certain hematological changes. Abdominal adiposity is highly correlated with visceral adipose tissue accumulation[17,18] and the connection of visceral adiposity with the biochemical changes was thought to be due to enlarged visceral fat depots discharging free fatty acids into the portal and systemic circulations.[27,28] It is possible therefore, that there is a direct effect of fatty acids on erythropoiesis like a direct effect of fatty acids on serum protein and serum lipids. In this context Bug et al[30] found that valproic acid increases both proliferation and self renewal of normal hematopoietic stem cells. In a more recent study by Cao et al[31] showed that the hydroxamic acid derivatives of some short-chain fatty acids stimulate in vivo the erythropoiesis and fetal globin production in experimental animals.

Another possible mechanism of the association of abdominal obesity and hemoglobin level is that the extra weight of the abdomen of abdominally obese individuals may interfere with breathing and result in hypoventilation and relative hypoxia with the resultant increase in red blood cells production and hemoglobin levels.

In conclusion, although the effect of abdominal adiposity on hemoglobin level shown in this study may be modest but it is significant and therefore, should be considered when constructing norms for hemoglobin in this population or when screening for anemia using hemoglobin level. Whether this relationship is the same for adult men or children remained to be investigated.

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