A method to estimate prevalence of iron deficiency and iron deficiency anemia in adolescent Jamaican girls1-3

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ABSTRACT  A method is presented to estimate a cutoff for hemoglobin concentration appropriate for estimating the prevalence of iron deficiency anemia in poor Jamaican girls 13-14 y of age. Iron deficiency was determined from a three-variable model of iron status (serum ferritin, erythrocyte protoporphyrin, and mean corpuscular volume). The most appropriate hemoglobin cutoff was considered the one that minimized misclassification of iron deficiency: that yielding the maximum kappa coefficient for correctly classifying iron deficiency between 100 and 120 g/L, at 1-g/L intervals. By using this method, a hemoglobin cutoff of 107 g/L was considered most appropriate. This cutoff and the other indicators were used to estimate prevalences of iron deficiency and iron deficiency anemia in the Jamaican girls: 7.6% and 4.3%, respectively. This approach should be appropriate for determining hemoglobin cutoffs for iron deficiency anemia in other populations. Am J Clin Nutr 1997:65:831-6.

KEY WORDS  Iron deficiency, iron deficiency anemia, hemoglobin, prevalence estimation

INTRODUCTION

In black individuals of African heritage, specification of meaningful cutoffs for anemia or iron deficiency anemia based on hemoglobin concentration has been difficult because blacks may have systematically lower hemoglobin values than whites with comparable iron status. Studies have repeatedly reported differences in mean hemoglobin from 2 to 10 g/L between samples from blacks and whites after iron status and other possible confounding factors were controlled for (1-3). Accordingly, a need for population-specific criteria for hemoglobin and anemia has been expressed (4, 5), but no firm recommendations have been made.

Based on a 10-g/L difference observed between mean hemoglobin values for black and white non-iron-deficient women, Johnson-Spear and Yip (5) found that a hemoglobin cutoff of 110 g/L had a sensitivity and specificity to identify iron deficiency in black women that was similar to that of the conventional 120-g/L cutoff used for white women. These results suggest that the relation between hemoglobin concentration and iron deficiency status can be maintained for blacks with a lower hemoglobin cutoff, and that an appropriate hemoglobin cutoff for iron deficiency anemia may be ~110 g/L in adult black women. Nevertheless, a method to determine the most appropriate hemoglobin cutoff for iron deficiency anemia in blacks has not been elucidated previously.

Typically, cutoffs for anemia based on low hemoglobin concentration have been determined from distributions of healthy populations, excluding those individuals known to have iron deficiency, thalassemias, and other conditions that may alter hemoglobin values (6-8). Then, a fixed proportion of the healthy sample (usually 2.5% or 5%) is assumed to be below the value corresponding to the appropriate cutoff. Those with iron deficiency anemia are the subset with hemoglobin concentrations below the cutoff for anemia who also are considered iron-deficient based on other hematologic and biochemical indicators (9, 10). For populations, the prevalences of anemia and iron deficiency are best estimated by adjusting the observed frequencies for the expected within-subject variation in the indicators (11, 12).

Supplementation trials in which anemia is defined by hemoglobin response to iron therapy show that distributions of hemoglobin values of truly iron-deficient individuals (those who respond) and nondeficient individuals overlap (13, 14). James (15) provided a method of estimating a hemoglobin cutoff point that minimizes the variance due to misclassification between a mixture of two Gaussian distributions. Means and SDs of the mixing distributions (eg, iron-deficient and non-iron-deficient), and the approximate prevalence of the abnormal condition are required. Cook et al (16) estimated population prevalences of anemia in Latin America assuming a mixture of Gaussian distributions for hemoglobin. Distributions of anemic individuals, however, are not always Gaussian (13). Deviations from the Gaussian distributions of individuals with normal transferrin saturation or hemoglobin concentration have been used to estimate population prevalences of iron...

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deficiency and anemia (17), but this approach has not been expanded to include simultaneously the multiple indicators required to estimate prevalences of iron deficiency anemia.

Dallman et al (18) estimated the prevalence of iron deficiency anemia using the “median shift” method in the hemoglobin distribution, after excluding individuals with one or more laboratory values indicative of iron deficiency. This approach assumes that the central tendency of the hemoglobin distribution reflects changes at the extreme tail of the distribution, which may not be true, especially when the prevalence of iron deficiency is low.

An epidemiologic method of selecting an optimum cutoff for hemoglobin concentration to identify those who respond to iron supplementation was used by Leibel et al (19). This approach, derived from screening methodology, used the maximum sum of sensitivity and specificity of selected hemoglobin cutoffs as the criterion for choosing the optimum cutoff value. This yielded the hemoglobin value that maximized the proportions of iron responders and nonresponders that were correctly identified, while giving equal weight to both outcomes (20).

The screening method has the advantages that it makes no assumptions regarding the shapes of the constituent distributions and it requires no prior approximation of prevalence. The maximum sum of sensitivity and specificity, however, does not necessarily minimize the misclassification of the criterion populations (in this case, iron responders from nonresponders). Maximum efficiency, or the proportion of individuals correctly classified by the test or screen, calculated on serial hemoglobin cutoffs would minimize the misclassification. Because observed efficiencies are biased, they should be adjusted for chance occurrence (20).

Adolescence is a period of increased requirements for dietary iron because of the adolescent spurt in body mass, and is additionally so in girls because of the onset of menstrual losses (21). Iron status during adolescence may be complicated further by low dietary intakes (8). In Jamaica, available data indicate that anemia has probably been relatively prevalent among preadolescent and adolescent girls. In a case-control study of health exposures and school failure of poor Kingston children in grade 5 (9.5–12.5 y of age), 31% of succeeding children and 43% of failing children had hemoglobin concentrations < 110 g/L (22). Corresponding proportions with hemoglobin concentrations < 100 g/L were 14% and 27%, respectively. Girls represented 65% of succeeding children and 35% of failing children. A national anemia survey conducted in 1987 indicated that 17% of girls aged 10–14 y, and 32% of girls 15–19 y were anemic according to hemoglobin cutoffs of 114 and 120 g/L, respectively (M Campbel, PJ Bennett, K Fox, S Rawlins, MA Green, WN Gibbs, unpublished observations, 1990).

The present study evaluated the iron status of a sample of black adolescent girls living in the poorest areas of Kingston, Jamaica. Iron deficiency was determined from a model including three indicators of iron status: serum ferritin, erythrocyte protoporphyrin, and mean corpuscular volume (MCV). A new approach to determine an appropriate cutoff for hemoglobin concentration relative to iron deficiency was taken that used the screening efficiencies of potential hemoglobin cutoffs and a statistic to correct for chance agreement and misclassification bias. The prevalences of iron deficiency and iron deficiency anemia in this population were then estimated, taking into account the expected within-subject variation in measurement of the laboratory values.

SUBJECTS AND METHODS

The sample was selected from a pool of all 13- and 14-y-old girls on the school registers in grade 8 in all Kingston inner-city All-Age schools (n = 4) and New Secondary schools (n = 5) that had > 40 girls enrolled. These schools include children who did not qualify to attend the more academic high schools and they include 77% of children from the poorest quintile of the population (23). Girls were sampled from the pool randomly and contribute to the total sample in proportion to the size of the school. Approximately 87% of Kingston girls of this age attend school. Data were collected on 452 girls, 95% of the original target sample. The sample should be representative of poor girls who attend grade 8 in Kingston schools. Further details of the sampling frame and process are available elsewhere (23). Complete data for blood variables were available for 435 girls.

Blood samples were obtained by antecubital venipuncture. Serum ferritin was assayed by an enzyme-linked immunosorbent assay (ELISA) method (Diagnostic Products Corporation, Los Angeles); standards were included in each assay and the CV of replicates was 4%. Erythrocyte protoporphyrin was determined by using an extraction method (24), and two quality control samples were included in each assay. The CVs for erythrocyte protoporphyrin in 28 replicates were 6.9% and 14.2% at 102 and 238 g/L red blood cells (RBCs), respectively. Hemoglobin concentration, MCV, and hematocrit were determined by automated methods with standards on a Cell Dyn 700 cell counter (Sequoia, Turner, CA), and the analytic variation of replicates was within the published range of values (25).

Electrophoresis studies were carried out for all blood samples with hemoglobin < 100 g/L by using a standard hemoglobin electrophoresis method with a cellulose acetate membrane (26). The electrophoresis identified three girls with sickle cell disease and two girls with sickle cell trait; these five girls were excluded from the analyses. Also excluded from the analyses were eight girls with hemoglobin > 100 g/L who reported they had sickle cell disease, and three iron-deficient girls who were considered to probably have thalassemia minor according to a Metzner index ≤ 13.0 MCV/RBC (27). There was no evidence of macrocytosis that would indicate anemias due to vitamin deficiencies. Evidence from a nutritional survey in Jamaica confirms the unlikely existence of vitamin B-12 and folate deficiencies (Campbell et al, unpublished observations, 1990). After exclusions, the final sample included 419 girls of African descent who lived at approximately sea level. Five girls (1.2%) reported smoking more than five times in the past month. Elevated blood lead concentrations are rare in this population (22).

Cutoff values used for indicators of iron deficiency were as follows: hematocrit < 36%, serum ferritin < 10 µg/L, erythrocyte protoporphyrin > 700 µg/L RBCs, and MCV < 78 fl (21, 28). Iron deficiency was considered present if individuals had two or more abnormal values from among these three indicators (10, 28).

Height, weight, and triceps skinfold thickness were measured by using recommended protocols (29), with errors of
measurement well within recommended ranges (30). Body mass index (BMI) was calculated as weight (kg)/height (m)^2. Menarcheal status was obtained by direct questioning. All procedures were approved by the Ethics Committee at the University of the West Indies.

An appropriate cutoff for hemoglobin concentration was obtained by adapting the approach of Kraemer (20) for screening tests. The optimal hemoglobin cutoff relative to iron deficiency was considered the concentration of hemoglobin with the maximum kappa coefficient (κ) (31) for efficiency, from among 2 × 2 tables of iron-deficiency status versus membership in groups defined by hemoglobin cutoffs and indexed by 1-g/L intervals, as follows:

\[ \kappa = \frac{P_o - P_c}{1 - P_c} \]

where \( P_o \) is the observed proportion correctly identified as iron deficient (efficiency) by the hemoglobin cutoff, and \( P_c \) is the proportion expected by chance.

\[ P_o = \frac{(TP + TN)}{n} \]

where TP is the number of true-positive results, TN is the number of true-negative results, and \( n \) is the total number in the sample.

\[ P_c = \frac{[(TP + FN)/n] [(TP + FP)/n]}{[(FP + TN)/n] [(FN + TN)/n]} \]

where FP is the number of false-positive results and FN is the number of false-negative results.

Adjustment of the observed prevalences for within-subject variation was carried out by following the method of Looker et al (11). Because the distributions of MCV, erythrocyte protoporphyrin, and serum ferritin were distinctly non-Gaussian, these variables were transformed to approximate normality before the variance adjustments. The transformations used for each measure were MCV^3, −1/3 erythrocyte protoporphyrin, and log serum ferritin, respectively. Variance ratios used for hemoglobin, MCV, and erythrocyte protoporphyrin were from US women included in the Hispanic Health and Nutrition Examination Survey (HHANES) (11). The variance ratio for serum ferritin was based on a separate sample of US women (32).

RESULTS

Summary statistics for age and anthropometric variables are presented in Table 1. Heights, weights, and BMIs for the Jamaican girls approximate age-specific medians for National Center for Health Statistics (NCHS) reference data based on US youth (33), although the median triceps skinfold thickness indicates considerably less subcutaneous fat than the US median (34). More detailed analysis of their anthropometric status is reported elsewhere (30). The median age at menarche (estimated from probit analysis) was 13.1 y.

Medians and quartile values of hematologic and biochemical indicators of iron status are presented in Table 2. Because some of the distributions deviated markedly from Gaussian expectations, medians are presented rather than means. The median values of hemoglobin, hematocrit, and MCV are lower than those reported for US girls of comparable age (6, 7), and samples with appreciable numbers of black youth (35, 36). The frequencies beyond iron-deficiency cutoffs are consistent with elevated rates of iron deficiency compared with US youth. The proportions observed with hemoglobin concentrations less than two conventional cutoffs are presented for information purposes only. The pattern of elevated frequencies of abnormal values for hemoglobin and hematocrit relative to those for serum ferritin and erythrocyte protoporphyrin are consistent with the view that the conventional hemoglobin and hematocrit cutoffs are probably inappropriate relative to iron deficiency in this population.

Mean hemoglobin concentrations are presented in Figure 1 according to the number of abnormal values in the three-variable model of iron deficiency. Analysis of variance resulted in significant differences in mean hemoglobin concentration among groups (\( F = 60.3, \text{df} = 3, P < 0.001 \)), and the trend of decreasing mean hemoglobin concentration with increasing number of abnormal values was highly significant (\( F = 172.0, \text{df} = 1, P < 0.001 \)). This hemoglobin pattern supports the validity of the model of iron deficiency. Because at least two abnormal values are required to designate iron deficiency, all those considered iron-deficient must have had either low serum ferritin or MCV values. Consequently, it is unlikely that iron deficiency is overestimated by the model because of elevations in erythrocyte protoporphyrin that may be associated with inflammatory disease or lead exposure. The observed proportion of girls with iron deficiency using this model (those having two or more abnormal values) was 7.6%. The mean (± SD) hemoglobin concentrations of those considered iron-deficient

<table>
<thead>
<tr>
<th>Indicator</th>
<th>Median</th>
<th>25th, 75th percentiles</th>
<th>Frequency beyond cutoff %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemoglobin (g/L)</td>
<td>125</td>
<td>118, 131</td>
<td>28.2</td>
</tr>
<tr>
<td>&lt;120 g/L</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>&lt;115 g/L</td>
<td></td>
<td></td>
<td>14.1</td>
</tr>
<tr>
<td>Hematocrit (I)</td>
<td>0.371</td>
<td>0.354, 0.391</td>
<td>32.5</td>
</tr>
<tr>
<td>Serum ferritin (μg/L)</td>
<td>25.7</td>
<td>14.1, 42.1</td>
<td>15.8</td>
</tr>
<tr>
<td>Erythrocyte protoporphyrin (μg/L RBCs)</td>
<td>386</td>
<td>321, 476</td>
<td>7.6</td>
</tr>
<tr>
<td>Mean corpuscular volume (fL)</td>
<td>85.5</td>
<td>81.4, 89.2</td>
<td>12.9</td>
</tr>
</tbody>
</table>

^1 RBCs, red blood cells.
and not iron-deficient were 106.4 ± 13.6 and 126.1 ± 9.2 g/L, respectively.

Values for efficiency and the κ for hemoglobin relative to iron deficiency are presented for cutoffs indexed by 1 g/L from 100 to 120 g/L (Figure 2). A single hemoglobin cutoff was maximally able to correctly classify 95.5% of those considered iron deficient, so little misclassification occurred. By using the criterion of maximal value for κ, 107 g/L is the most appropriate hemoglobin cutoff for iron deficiency. In this population, maximum efficiency and maximum κ identified the same cutoff, although this may not always be so (20).

Population prevalences for iron status, adjusted for expected within-subject variation, are presented in Table 3. The observed frequencies (Table 2) are reduced by considering within-subject variation and the multiple indicators in estimating the prevalences (Table 3). The relative pattern of prevalences based on the new hemoglobin cutoff conforms to theoretical expectations of the iron-deficiency model (9).

**FIGURE 1.** Mean (± SD) hemoglobin concentration in groups classified by the number of abnormal indicators of iron deficiency. Sample sizes are presented at the base of each column.

**FIGURE 2.** Kappa coefficients (κ) and efficiency of hemoglobin cutoffs in identifying individuals with iron deficiency (at least two abnormal indicators). Hemoglobin cutoff (107 g/L) with the maximum κ is identified.

<table>
<thead>
<tr>
<th>Iron status</th>
<th>Indicator</th>
<th>Population prevalence</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Depleted iron stores</td>
<td>SF &lt; 10 μg/L</td>
<td>15.0</td>
<td>11.6, 18.5</td>
</tr>
<tr>
<td>Impaired erythropoiesis</td>
<td>EP &gt; 700 μg/L/RBCs,</td>
<td>7.4</td>
<td>4.9, 9.9</td>
</tr>
<tr>
<td>Iron deficiency</td>
<td>SF &lt; 10 μg/L,</td>
<td>7.4</td>
<td>4.9, 9.9</td>
</tr>
<tr>
<td></td>
<td>MCV &lt; 78 FL</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>(≥ 2 abnormal)</td>
<td>7.6</td>
<td>5.1, 10.1</td>
</tr>
<tr>
<td>Iron deficiency and</td>
<td>Iron deficiency and</td>
<td></td>
<td></td>
</tr>
<tr>
<td>anemia</td>
<td>hemoglobin &lt; 107 g/L</td>
<td>4.3</td>
<td>2.4, 6.2</td>
</tr>
</tbody>
</table>

1 SF, serum ferritin; EP, erythrocyte protoporphyrin; MCV, mean corpuscular volume.
2 Adjusted for within-subject variation by the method of Looker et al (11).

**DISCUSSION**

We assumed that the most appropriate hemoglobin cutoff to define iron deficiency anemia in this population is the one that correctly classifies the largest proportion of individuals relative to iron deficiency, when corrected for bias due to chance agreement. The maximum κ statistic for overall efficiency from possible hemoglobin cutoffs identified the appropriate cutoff as 107 g/L. This concentration is considerably lower than the more conventional hemoglobin cutoffs for anemia, which range from 115 to 120 g/L (6, 7, 28, 37).

Because of the method used, the 107 g/L cutoff is specific to iron deficiency anemia. When all individuals with one or more abnormal values for serum ferritin, erythrocyte protoporphyrin, or MCV were excluded from the sample (n = 102), the 2.5th and 5th percentile concentrations of hemoglobin were 110 and 112 g/L, respectively. Consequently, the 107 g/L cutoff for iron deficiency anemia is more restrictive than those that would define anemia if the traditional methodology for determining anemia cutoff were applied to this sample. It is unclear whether the hemoglobin cutoff considered appropriate for iron-deficiency anemia in this group is applicable more generally to other black women. The method, however, can be applied to other populations to determine its generalizability.

By using the hemoglobin shift model to estimate anemia (18), the relative prevalence was 10% for the Jamaican girls when the presence of any measure of iron deficiency (serum ferritin, erythrocyte protoporphyrin, or MCV) was used to adjust the hemoglobin distribution. When only abnormal erythrocyte protoporphyrin or MCV values were used, the relative prevalence of anemia in Jamaican girls due to hemoglobin distribution shift was 7.2%. These rates are within the 95% CI for prevalences of impaired erythropoiesis and iron deficiency (Table 3), but they are greater than that for iron deficiency anemia estimated by the new method.

Direct comparison of the prevalences of impaired iron status in Jamaican girls with other studies is hampered because of differences in available measures and methods; nevertheless, some comparisons can be made. Iron deficiency was assessed for the US population based on data from the second National
Health and Nutrition Examination Survey (NHANES II: 12). Prevalences of iron deficiency in young women 11–14 y of age were estimated at 6.1% and 3.4%, by using a ferritin model (serum ferritin, erythrocyte protoporphyrin, and transferrin saturation) and an MCV model (MCV, erythrocyte protoporphyrin, and transferrin saturation), respectively (28). The estimate of 7.6% iron deficiency in the Jamaican adolescents used serum ferritin, erythrocyte protoporphyrin, and MCV and would be expected to roughly correspond to the ferritin model in sensitivity to moderately impaired iron status.

With use of the MCV model and a hemoglobin concentration cutoff of < 118 g/L, prevalences reported for iron deficiency anemia in US girls aged 11–14 y were 0.2% (non-Hispanic whites), 0.6% (Mexican Americans), and 2.7% (Puerto Ricans) (12). Corresponding estimates were not provided for African Americans. The estimate of prevalence of iron deficiency anemia for Jamaican girls was 4.3% when a the three-variable model and a hemoglobin concentration < 107 g/L. Even when the lower hemoglobin cutoff was used, the Jamaican girls had from 1.6 to 21 times the prevalence of iron deficiency anemia as their same-aged US peers.

The appropriate hemoglobin cutoff for iron deficiency anemia in the Jamaican girls (< 107 g/L) has high specificity (0.987) but rather low sensitivity (0.588). Even so, the sensitivity and specificity both exceed the those seen for conventional hemoglobin cutoffs relative to iron deficiency in women (5). The proposed cutoff is appropriate for estimates of prevalence and should be viewed as an average value in the population. The new method selects for the hemoglobin cutoff that correctly classifies the largest proportion of individuals, whether iron-deficient or not iron-deficient. The pattern of high specificity and low sensitivity is typical of the indicators of iron status, and it occurs because an appreciable proportion of iron-deficient individuals may have hemoglobin values in the normal range. If more liberal definitions of iron deficiencies were used, eg, serum ferritin < 15 μg/L, the sensitivity of a fixed hemoglobin cutoff would rise, as would the most discriminating hemoglobin cutoff.

Because there may be truly iron-deficient individuals who are not anemic (9), the hemoglobin cutoff selected by our method may be slightly biased toward a higher value. If this is true, our prevalence of iron-deficient anemia is probably slightly overestimated. A hemoglobin cutoff of 107 g/L would probably not be appropriate for screening individuals if the purpose was to identify a large proportion of the individuals at risk of iron-deficiency for possible intervention. For such a purpose, a higher cutoff with greater sensitivity may be desirable. For interventions, the choice of the most appropriate cutoff is influenced by prevalence and by available resources (1).

A concern in establishing a hemoglobin cutoff for anemia in blacks that is lower than that for others has been the possibility that the lower hemoglobin distributions observed in blacks reflect a subset of individuals with mild hereditary hematologic abnormalities, such as hemoglobinopathies or thalassemia traits, that impair RBC or hemoglobin production (38). If this position is correct, an inappropriately low hemoglobin cutoff would tend to miss many iron-deficient individuals and to misclassify some individuals as iron-deficient who are not. The traditional approach of assuming that a fixed proportion of the non-iron-deficient hemoglobin distribution is anemic would be susceptible to such “hidden” individuals with low hemoglobin but who are not truly iron deficient. Because our method did not assume a fixed proportion of anemic individuals and the appropriate hemoglobin cutoff was determined relative to iron deficiency per se, it should be less susceptible to such bias. We attempted to exclude known or probable cases of sickle cell disease or thalassemias that may be related to impaired RBC or hemoglobin production. Nevertheless, because some hematologic abnormalities may be difficult to detect, we cannot completely rule out the possibility of misclassification of such individuals if they exist in this population. In our population, all but one (17 of 18) of the individuals with iron deficiency anemia had a serum ferritin value ≤ 10 μg/L, so it is unlikely they were not truly iron deficient.

We have provided a method to determine an appropriate hemoglobin cutoff designating iron deficiency anemia for poor Jamaican girls 13–14 y of age. The results suggest that a hemoglobin cutoff for blacks that effectively discriminates between iron-deficient and non-iron-deficient individuals (107 g/L) is lower than conventional cutoffs. The method proposed can be applied to determine whether 107 g hemoglobin/L is appropriate for black women at other ages or in other populations.

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REFERENCES


