

Diagnostic accuracy of hemoglobin for iron deficiency in pregnancy: disclosing results of a cited clinical trial

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ABSTRACT

Objective. To analyze the accuracy of hemoglobin (Hb) concentrations as a diagnostic indicator of iron deficiency in pregnant women and to measure the efficacy of oral iron therapy using Hb z-scores rather than Hb absolute values.

Methods. The sensitivity and specificity of Hb < 11.0 g/dL, and its receiver operating characteristic (ROC) curve, in the diagnosis of iron deficiency (serum ferritin (SF) < 12.0 ng/mL) were determined in 318 women in their second trimester of pregnancy who had been screened for a clinical trial conducted in 2001 in Northeast Brazil. A secondary analysis of iron therapy efficacy was carried out using data from the trial's three different treatments (60 mg of oral iron once per week (n = 46), twice per week (n = 50), and once per day (n = 44)). The mean differences between post- and pre-treatment Hb absolute values (g/dL) and z-scores (standard deviation (SD)) were calculated for the three treatment groups for study participants with and without iron deficiency.

Results. Hb sensitivity, specificity, and area under the ROC curve were 60.7%, 44.3%, and 0.54 respectively. Women without iron deficiency showed improvements in Hb absolute values (as in the clinical trial's overall results) but did not have improved Hb z-scores (with scores of -0.6 SD (95% confidence interval (CI): -0.99, -0.28); -0.2 SD (95% CI: -0.47, 0.08); and -0.1 SD (95% CI: -0.33, 0.18) for weekly, twice-per-week, and daily iron treatment schemes respectively). In contrast, iron-deficient women treated with the intermittent schemes had reductions in both Hb absolute values and Hb z-scores, respectively: weekly = -0.42 g/dL (95% CI: -0.72, -0.12) and -1.4 SD (95% CI: -1.74, -0.99); twice per week = -0.14 g/dL (95% CI: -0.46, 0.17) and -1.1 SD (95% CI: -1.44, -0.75).

Conclusions. These analyses revealed that Hb concentrations were not an accurate indicator of either iron needs or iron-therapy response in pregnant women.

Key words

Anemia, iron-deficiency; pregnancy; erythrocyte indices; sensitivity and specificity; clinical trial; Brazil.

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Maternal anemia still has a high prevalence, affecting 50% of pregnant women worldwide and 30% in Latin America (1, 2). Anemia during pregnancy has been associated with several adverse perinatal outcomes, including prematurity, low birth weight, and maternal and perinatal mortalities (1–9). Since the 1980s, iron

supplementation during pregnancy has been widely recommended as a health policy to control anemia (10, 11), based on the assumption that low hemoglobin (Hb) concentrations would be a proxy of iron-deficiency anemia (11). However, several studies conducted in pregnant women have shown not only that ane-

mia has multiple causes but also that Hb levels have low correlation with maternal body iron deficiency (12–18). Although serum ferritin (SF) is the most accurate biomarker of body iron stores in pregnant women (18–20), it is underutilized in clinical practice and trials (21, 22). Diagnostic misclassification resulting from the use of the Hb criterion (concentration levels < 11.0 g/dL) in the selection of patients for iron therapy could lead to low therapeutic efficacy in clinical sets (19, 21, 22), as well as ineffective control programs (1, 11, 23).

Moreover, physiological fluctuations in Hb values could bias the measurement of therapeutic efficacy during pregnancy. Homeostatic hemodilution throughout pregnancy induces a variation in Hb and hematocrit levels (12–18), leading to a U-shaped curve with a nadir between the 24th and 28th gestational weeks (9, 24–27). Considering this phenomenon, Beaton & McCabe (22) employed the z-scores to adjust the Hb values according to the gestational age in several trials using oral iron in pregnant women and showed strikingly different results when compared to the therapeutic effects previously reported. The z-score quantifies (in standard deviation (SD) units) the difference between an observed Hb value and the reference mean for a specific gestational week derived from the Hb distribution curve found in iron-supplemented healthy women from Europe and North America (28).

Even though Hb z-scores can measure the erythrocyte mass more precisely than absolute Hb values throughout the gestational period, by taking into account the hemodilution distortion on Hb values, the former method has received little attention from those comparing iron treatments in clinical trials (21, 22). The dissenting results produced by the two different methods can be seen in two studies described in a recent meta-analysis on maternal iron-deficiency anemia treatments (21) that compared daily and intermittent schemes of oral iron treatment (29, 30). The first study, by Mumtaz et al. (29), measured the treatment outcomes using Hb z-scores, while the second study by Souza et al. (30) used Hb absolute values and found different results.

This study analyzed the Souza et al. (30) clinical trial databank with two objectives: 1) to estimate the accuracy of Hb

for iron-deficiency diagnosis in the study population screened for the trial and 2) to reanalyze and measure the efficacy of the three iron therapy schemes used in the trial based on Hb z-scores rather than Hb absolute values.

MATERIALS AND METHODS

Study protocol

A diagnostic validation analysis was performed using data for 318 pregnant women screened for a clinical trial conducted in 2001 by Souza et al. (30) at a prenatal care center in an urban city in Northeast Brazil. In addition, among 150 anemic women enrolled for the oral iron therapies, a treatment efficacy reanalysis was undertaken introducing two new variables: 1) SF pretreatment values and 2) pre- and post-treatment Hb values adjusted by the z-score according to the gestational week. The full protocol of the primary clinical trial is available elsewhere (30), as well as the data on therapeutic compliance and adverse effects (31).

Of the 347 screened women with low-risk pregnancy, 180 (55%) had anemia according to the World Health Organization (WHO) criterion (Hb < 11.0 g/dL) (11). The mean age was 22 years and the mean gestational age was 17 weeks at diagnosis. A total of 150 anemic women agreed to participate and were randomly allocated into three treatment groups with different schemes for treatment with ferrous sulphate pills (60 mg of elementary iron) (group 1, one pill weekly ($n = 48$); group 2, one pill twice per week ($n = 53$); group 3, one pill daily ($n = 49$)) and without supplementation with any other micronutrient. The women were treated for 16 (± 1) weeks and submitted to Hb measurements at enrollment and at the 8th and 16th weeks of follow-up (30).

The erythrogram was taken using the cyanmethemoglobin method and a Coulter T890[®] automated cell counter (Beckman Coulter, Brea, California, USA). The SF was measured using the enzyme-immunoassay (EIA) method and a COBAS Core Ferritin EIA device (Roche SA, Basel, Switzerland) and a measurement zone between zero and 1 200 ng/mL. This method is calibrated according to WHO international standards (IS 80/602) by the National Institute for Biological Standards and Control (Hertfordshire, England).

Analysis of the accuracy of Hb versus SF

Among the 347 screened pregnant women, 318 had SF pretreatment results available for this analysis, which was a sufficient number to estimate a sensitivity of 90% ($\pm 9\%$) and a specificity of 80% ($\pm 8\%$), with a relative error of 10%, assuming a 50% frequency for iron-deficiency anemia in the local population (2, 30). The agreement (kappa test), sensitivity, specificity, and predictive values of Hb pretreatment according to WHO's cutoff point (Hb < 11.0 g/dL) were estimated against SF pretreatment as a gold standard for iron-deficiency diagnosis (11). SF values under 12.0 ng/mL were classified as iron deficiency according to WHO and U.S. Centers for Disease Control and Prevention (CDC) recommendations (11, 28, 32, 33).

The discriminatory power of Hb was verified by the receiver operating characteristic (ROC) curve and represented by the area under the curve (AUC). The ROC curve plots the correlation between sensitivity and 1-specificity of all test values observed in a population distribution and allows for evaluation of the discriminatory power of the test to enable selection of the most accurate cutoff point (34). In general, distinctions can be made between non-informative (AUC = 0.5), less accurate ($0.5 > \text{AUC} \leq 0.7$), moderately accurate ($0.7 > \text{AUC} \leq 0.9$), highly accurate ($0.9 > \text{AUC} < 1.0$), and perfect tests (AUC = 1.0) (34). A perfect test has a cutoff point with both sensitivity and specificity at 100% (34).

Analysis of therapeutic efficacy using Hb z-scores and SF stratification

Therapeutic efficacy reanalysis in each treatment group employed z-scores to measure the variation of Hb, overall and within the SF pretreatment strata (< 12.0 ng/mL or ≥ 12.0 ng/mL). The stratified analysis was performed on 140 participants tested for body iron deposits at baseline among the 150 pregnant women enrolled in the three treatment groups (weekly ($n = 46$), twice per week ($n = 50$), and daily ($n = 44$)), as 10 women did not comply with the SF measurement. At the 16th week of follow-up, the mean difference between Hb post- and pretreatment was estimated by Hb absolute values (in g/dL) and z-scores (SD) for each treatment group and SF stratum

pretreatment. The Hb z-scores were obtained from each woman according to the curve proposed by Beaton & McCabe (22). Along this curve, the Hb SD had to be 0.9 g/dL at any gestational age. The z-score was calculated using the following equation: $z\text{-score} = (\text{observed Hb} - \text{expected Hb mean}) / \text{SD}$ (reference population). Therefore, each z-score unit corresponds to one SD from the Hb reference mean (22).

A two-tailed Student's *t*-test was used to assess the difference in therapeutic response between the strata for each treatment group, with the statistical significance level set at 5%. The results were robust to outliers, according to a test using a quantile regression model (35, 36) in which the estimation is based on the median rather than the mean. Data were analyzed using Epi Info™ 6.04b (Centers for Disease Control and Prevention, Atlanta, Georgia, USA); Minitab 14.2 (State College, Pennsylvania, USA); and Stata 9.2 (StataCorp LP, College Station, Texas, USA). The study followed ethical principles for medical research involving human subjects according to the Declaration of Helsinki and received approval from the ethics committee of the Instituto de Medicina Integral Prof. Fernando Figueira in Recife (#650/2005). Written informed consent was obtained from all patients before their admission into the study.

RESULTS

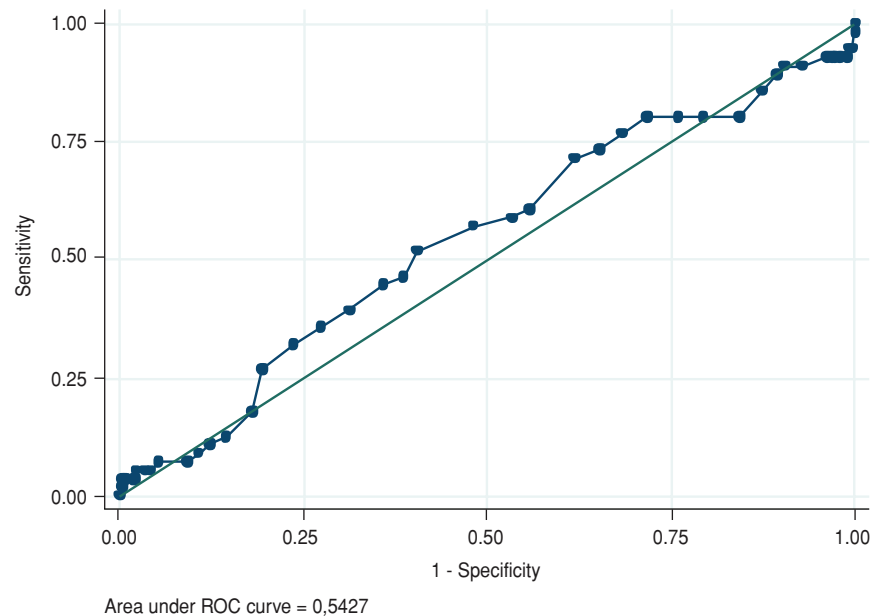
Accuracy of Hb versus SF

Table 1 shows the results distribution of the Hb criterion (< 11.0 g/dL) versus the gold standard for iron-deficiency diagnosis (SF < 12.0 ng/mL). The frequency of iron deficiency estimated by the Hb criterion was 57% while the frequency estimated by the SF criterion was 18%. Among pregnant women classified as anemic according to the WHO Hb criterion, 81% (146 out of 180) had body iron stores at baseline (SF ≥ 12.0 ng/mL). Diagnostic agreement between both tests occurred in 10.7% of cases (Cohen's kappa = 0.027; 95% confidence interval (CI): -0.050, 0.103). Based on the results in Table 1, the accuracy parameters were estimated. The Hb criterion showed a sensitivity of 60.7% (95% CI: 46.8, 73.5); a specificity of 44.3% (95% CI: 38.2, 50.5); accuracy of 47.2% (95% CI: 41.7, 52.7); positive predictive value of 18.9% (95% CI: 13.5, 25.4); and negative predictive

TABLE 1. Distribution of hemoglobin and serum ferritin (SF): results among 318 women in second trimester of pregnancy, Recife, Pernambuco, Brazil, 2000–2001

Hemoglobin level	Iron deficiency (SF < 12.0 ng/mL)		Total (%)
	Present (%)	Absent (%)	
Low (< 11.0 g/dL)	34 (10.7)	146 (45.9)	180 (56.6)
Normal (≥ 11.0 g/dL)	22 (6.9)	116 (36.5)	138 (43.4)
Total	56 (17.6)	262 (82.4)	318 (100)

FIGURE 1. Receiver operating characteristic (ROC) curve of hemoglobin concentration for diagnosis of iron deficiency among 318 women in their second trimester of pregnancy, Recife, Pernambuco, Brazil, 2000–2001



value of 84.1% (95% CI: 76.9, 89.7). The Hb ROC curve was less accurate for diagnosis of iron deficiency (AUC = 0.54 (95% CI: 0.46, 0.63)) (Figure 1).

Therapeutic efficacy using Hb z-scores and SF stratification

Table 2 shows overall and stratified therapeutic responses assessed by Hb absolute values and z-scores in each treatment group. For overall therapeutic response, the mean differences in absolute values of Hb indicated positive effects with twice-per-week and daily treatments, but the effects on z-scores were negative after the intermittent treatments (twice per week or weekly). Among those with the higher level of stored iron (SF ≥ 12.0 ng/mL), the stratified therapeutic analyses showed that the mean differences in Hb absolute val-

ues were positive, similar to the overall therapeutic responses obtained in the primary analysis. However, among the stratum with lower SF (< 12.0 ng/mL) the results showed an inversion of that effect among the groups who underwent intermittent treatments (twice per week or weekly). As shown in Table 2, only the women with some stored iron at baseline (SF ≥ 12.0 ng/mL) treated at least twice per week and those in both strata treated with daily doses achieved positive effects in their Hb values and maintained their Hb z-scores.

DISCUSSION

The population studied in the current research was screened for anemia in a Brazilian study conducted before the implementation of the National Iron Supplementation Program (*Programa Nacio-*

TABLE 2. Therapeutic responses in second-trimester pregnant women with and without iron deficiency (serum ferritin < 12 ng/mL) for three oral iron therapy schemes, based on mean differences between post- and pre-treatment hemoglobin (Hb) concentrations (absolute values or z-scores), Recife, Pernambuco, Brazil, 2000–2001

Oral iron therapy scheme	Overall therapeutic response						Stratified therapeutic response ^e							
	n	Hb g/dL (CI) ^b	P ^b	Hb z-score	SD ^c (CI)	P ^d	Iron deficiency		No iron deficiency		Iron deficiency		No iron deficiency	
							No.	Hb g/dL (CI)	No.	Hb g/dL (CI)	P ^f	Hb z-score	SD (CI)	Hb z-score
Weekly	48	0.1 (-0.14, 0.42)	0.314	-0.71 (-1.02, -0.40)	< 0.001		9	-0.42 (-0.72, -0.12)	37	0.20 (-0.12, 0.53)	0.066	-1.4 (-1.74, -0.99)	-0.6 (-0.99, -0.28)	0.051
Twice per week	53	0.5 (0.30, 0.74)	< 0.001	-0.33 (-0.57, -0.09)	0.008		7	-0.14 (-0.46, 0.17)	43 ^g	0.64 (0.39, 0.90)	0.017	-1.1 (-1.44, -0.75)	-0.2 (-0.47, 0.08)	0.012
Daily	49	0.9 (0.64, 1.08)	< 0.001	0.057 (-0.19, 0.30)	0.643		7 ^h	1.5 (0.49, 2.51)	37 ^g	0.75 (0.51, 0.98)	0.023	0.8 (-0.35, 1.94)	-0.1 (-0.33, 0.18)	0.016

^a CI: 95% confidence interval.

^b Primary results (30).

^c SD: standard deviation.

^d Student's two-tailed t-test for statistical significance of mean difference for each therapy scheme.

^e Results of reanalysis after excluding 10 observations without serum ferritin (SF) data.

^f Student's two-tailed t-test for statistical significance of mean difference comparison between results for women with and without iron deficiency for each therapy scheme.

^g Women with iron stores (SF ≥ 12 ng/mL) at baseline treated twice per week or daily that achieved positive effects in their Hb absolute values and maintained their Hb z-scores.

^h Women with iron deficiency treated daily who achieved positive effects in their Hb absolute values and maintained their Hb z-scores.

nal de Suplementação de Ferro, PNSF) and the fortification of wheat flour and corn (37, 38). For that study, the frequency of hypoferritinemia was lower than would be expected in a context of severe magnitude of epidemiological anemia (11, 33) and lower than estimated iron deficiency reported by developed countries such as the United States (39) and the United Kingdom (40), which may reflect the need to establish diagnostic criteria for each population.

The accuracy analyses confirm the low reliability of Hb for iron-deficiency diagnosis versus SF and the lack of comparability between both tests as shown by the low diagnostic agreement (41). Moreover, the ROC curve indicated Hb had a low discriminatory power for iron-deficiency diagnosis; according to its AUC it would not be possible to achieve both sensitivity and specificity above 50% at any Hb cutoff point (34). The diagnostic accuracy of other erythrocyte indices (red blood cell count, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, and mean corpuscular hemoglobin concentration) was also tested in this study population (results not shown) and their AUCs were found to be between 0.45 (red blood cell count) and 0.65 (mean corpuscular volume) (42).

These results show that even the use of a cutoff point of 10.5 g/dL for the Hb in the 2nd trimester, as recommended by the CDC (28, 39) and the British Committee for Standards in Haematology (40), would not be informative, in accordance with previous validation stud-

ies of erythrocyte indices against SF in pregnant women. Tam & Lao (12) and Casanova et al. (17) found low sensitivity of Hb during pregnancy (64% (Hb ≤ 11.0 g/dL) and 43% (Hb ≤ 9.6 g/dL) respectively), as well as moderately accurate ROC curves with AUCs of about 0.7 for all erythrocyte indices. Recently, Mei et al. reported 16% sensitivity of Hb according to the ratio transferrin receptor to SF, based on a representative sample of pregnant women from the U.S. population (39).

Low levels of Hb have been considered a proxy of iron-deficiency anemia in population surveys and have been proposed as a screening test in pregnant women based on the assumption that such test would have high sensitivity (11). However, evidence points to the inadequacy of using Hb absolute values as a unique and invariable diagnosis criterion during pregnancy (12, 13, 17, 18). In the current study, almost 40% of the women with deficient body iron stores (SF < 12 ng/mL) (22 out of 56; Table 1) were not selected to receive iron therapies because they had normal Hb, and only 16% of the women who underwent treatments (23 out of 140; Table 2) had low SF. These diagnostic misclassifications are a potential source of selection bias and might have contributed to the low cure rates (approximately 50%) found in the primary analyses by Souza et al. (30). They might also help explain similar findings from other clinical trials with oral iron therapy in pregnancy (21, 22, 24, 27).

Furthermore, measurements of the increase in Hb levels after iron treatments are prone to distortions during pregnancy due to the hemodilution phenomenon (22, 24, 27). A recent trial by Roberfroid et al. (24) concluded that, despite the iron doses (30 mg versus 60 mg), micronutrient intake did not prevent anemia (which was 51%) in the 3rd trimester of pregnancy, partly because of physiologic hemodilution. Therefore, when Hb values are adjusted for the gestational physiologic curve by z-scores, the therapeutic effects can be worse than those indicated by Hb absolute values, as observed in Beaton & McCabe's meta-analysis (22). In a recent Cochrane meta-analysis (21), only two clinical trials compared daily and intermittent iron therapies in pregnant women (29, 30). A study by Mumtaz et al. (29) was the only one to apply Hb z-scores among 23 reviewed clinical trials and found results supporting the daily posology. On the other hand, the trial conducted by Souza et al. (30) that used absolute Hb values as the criterion for indicating iron treatments and for evaluating their therapeutic efficacy found favorable results for intermittent therapies.

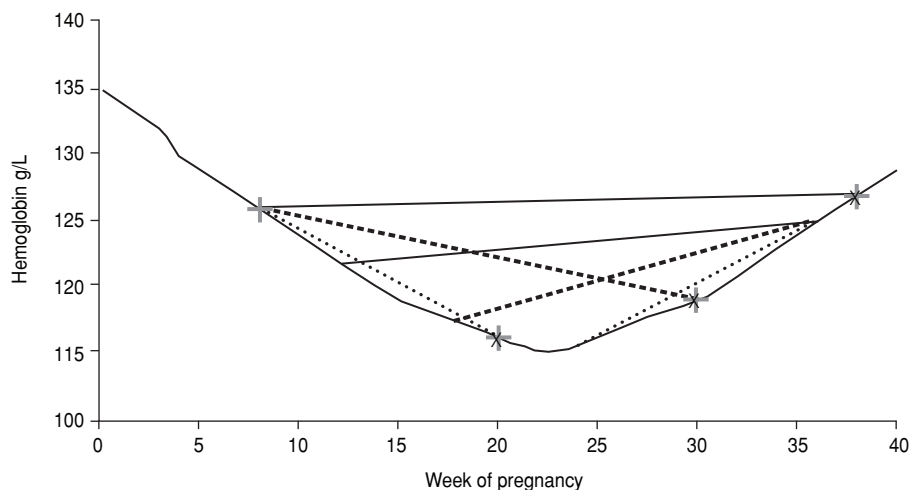
Therefore, the current study analyzed, retrospectively, the data from the therapeutic trial conducted by Souza et al. (30), considering pretreatment body iron stores and the adjustment of the Hb values for the gestational week. The similarity between the positive effects of the twice-per-week and daily posology observed by the primary analyses (30) was

not observed in these secondary analyses, suggesting a detrimental effect in pregnant women who underwent the intermittent treatment schemes, meaningful in the iron-deficient stratum, which had an opposite effect direction. The current findings match those of Beaton & McCabe (22) and Mumtaz et al. (29), which demonstrated greater therapeutic efficacy with the daily treatment versus the intermittent oral iron doses when using the Hb z-score methodology to evaluate the outcomes.

The demonstration of the poor accuracy of Hb levels as the criterion for patient selection for this trial was also coherent with the worst therapeutic response disclosed in the current study's efficacy reanalysis, probably because the selection bias resulted in a study population with low iron needs. Moreover, while Beaton & McCabe (22) demonstrated that the hemodilution phenomenon can interfere with inter-study comparability, the current findings indicate that it may also affect the comparison between different treatments within the same study (intra-study comparability). The most striking finding, however, was the negative or neutral effect of all therapeutic schemes after adjusting the Hb values according to the gestational week by the z-score method.

This seemingly dissenting result may reflect distortions from the hemodilution on the longitudinal observation of Hb values during the follow-up period between the indication and the assessment of the outcomes. The mean of the participants' gestational age at the time of enrollment in the trial was 17 weeks and the women were followed for 16 weeks (30), a period that could have coincided with the ascendant loop of the Hb physiological curve (24–28), as can be seen in Figure 2 from Beaton & McCabe (22) that simulates how iron therapies could behave during different periods of pregnancy. Hence, the positive primary findings for Hb absolute values could be largely attributable to a physiological effect, as the sample was composed mainly of women with iron stores at baseline who showed a non-increase of Hb z-scores when undergoing iron therapy. In addition, the non-improvement in these women's Hb z-score could indicate the presence of other causes of anemia (beyond "physiological anemia"), such as infections and other nutritional deficiencies (27, 28, 40).

FIGURE 2. Illustrative effect of varying the gestational age of start and duration of iron therapy trials^{a,b}



^a Source: (22).

^b Reference curve from supplemented well-nourished women reported by U.S. Centers for Disease Control and Prevention (28).

Limitations

Limitations of this study might be related to a counterpoint to the above interpretations, which is the increasing magnitude of the effects with the increasing posology of iron therapy in both the absolute values and z-scores of Hb. There is insufficient evidence on cutoff points of SF to differentiate iron-deficient and iron-sufficient pregnant women (13, 27, 33). Therefore, there could be some level of functional iron deficiency in the women that were studied that could make them somehow iron-responsive regardless of the SF stratification (11, 33). After WHO's standardization for human ferritins for immunoassays in 1985, a study by van den Broek et al. (in which half of the pregnant women had human immunodeficiency virus (HIV)) showed SF < 30 ng/mL had 90% sensitivity and 85% specificity against the iron in the bone marrow but 34% sensitivity and 94% specificity at a cutoff point of 12 ng/mL (13). To address this possibility, the current analyses were performed again using the cutoff point of 30 ng/mL, a criterion recommended by the British Committee for Standards in Haematology (40), and the ROC curves of erythrocyte indices and therapeutic effects were not modified (results not shown).

Other limitations include the fact that the Hb z-scores were calculated using

a reference curve for North American and European pregnant women (22, 28), which may not be relevant to the Brazilian population due to its ethnic heterogeneity. The use of a control in the trial could have helped limit the effect of this local factor, but this was not considered at the time of the study (30). In addition, the number of pregnant women with SF < 12 ng/mL was low, which may have affected the precision of the results, along with the long period of time that has elapsed since the completion of the trial. Nevertheless, the main aim of these analyses was to investigate intra-study distortions when using Hb absolute values as a diagnostic or curative criterion in pregnancy. Hence, the methodological rationale used in this report guided the protocol of the authors' most recent diagnostic trial, which was published elsewhere (43) and may enable studies with wider samples from other populations to improve the external validity and applicability of the results on the usefulness of Hb and SF to detect iron needs in pregnant women, as stressed in the most recent WHO guidelines (32, 33).

Conclusions

Regardless of its limitations related to temporality, the current study showed the diagnostic inaccuracy of Hb for maternal iron deficiency, which can result in

the nontreatment of iron-deficient pregnant women or the treatment of iron-sufficient pregnant women, and thus limit and distort reported results on iron-therapy efficacy. Therefore, the physiological curve of Hb and new biomarkers of iron metabolism must be addressed in

future surveys and therapeutic and diagnostic trials on maternal anemia.

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Conflicts of interest. None.

REFERENCES

- World Health Organization. Worldwide prevalence of anaemia 1993–2005: WHO Global Database on Anaemia. Geneva: WHO; 2008. Available from: http://whqlibdoc.who.int/publications/2008/9789241596657_eng.pdf
- Stoltzfus RJ. Iron deficiency: global prevalence and consequences. *Food Nutr Bull.* 2003;24(4 Suppl):S99–103.
- Rasmussen K. Is there a causal relationship between iron deficiency or iron-deficiency anemia and weight at birth, length of gestation and perinatal mortality? *J Nutr.* 2001; 131(2S-2):S590–601.
- Steer P, Alam MA, Wadsworth J, Welch A. Relation between maternal haemoglobin concentration and birth weight in different ethnic groups. *Br Med J.* 1995;310(6978):489–91.
- Allen LH. Anemia and iron deficiency: effects on pregnancy outcome. *Am J Clin Nutr.* 2000;71(5 Suppl):S1280–4.
- Murphy JF, O’Riordan J, Newcombe RG, Coles EC, Pearson JF. Relation of haemoglobin levels in first and second trimesters to outcome of pregnancy. *Lancet.* 1986;1(8488):992–5.
- Cogswell ME, Parvanta I, Ickes L, Yip R, Brittenham GM. Iron supplementation during pregnancy, anemia, and birth weight: a randomized controlled trial. *Am J Clin Nutr.* 2003;78(4):773–81.
- Scholl TO, Reilly T. Anemia, iron and pregnancy outcome. *J Nutr.* 2000;130(2S Suppl): S443–7.
- Yip R. Significance of an abnormally low or high hemoglobin concentration during pregnancy: special consideration of iron nutrition. *Am J Clin Nutr.* 2000;72(1 Suppl):S272–9.
- World Health Organization. Control of nutritional anaemia with special reference to iron deficiency. WHO Technical Report Series No. 580. Geneva: WHO; 1975. Available from: http://whqlibdoc.who.int/trs/WHO_TRS_580.pdf
- World Health Organization. Iron deficiency anaemia: assessment, prevention and control. A guide for programme managers. Geneva: WHO; 2001. Available from: http://www.who.int/nutrition/publications/micronutrients/anaemia_iron_deficiency/WHO_NHD_01.3/en/index.html
- Tam KF, Lao TT. Hemoglobin and red cell indices correlated with serum ferritin concentration in late pregnancy. *Obstet Gynecol.* 1999;93(3):427–31.
- van den Broek NR, Letsky EA, White SA, Shenkin A. Iron status in pregnant women: which measurements are valid? *Br J Haematol.* 1998;103(3):817–24.
- Puolakka J, Jänne O, Pakarinen A, Vihko R. Serum ferritin in the diagnosis of anemia during pregnancy. *Acta Obstet Gynecol Scand.* 1980;95 Suppl:S57–63.
- Thompson WG. Comparison of tests for diagnosis of iron depletion in pregnancy. *Am J Obstet Gynecol.* 1988;159(5):1132–4.
- Volpi E, De Grandis T, Alba E, Mangione M, Dall’Amico D, Bollati C. [Variations in ferritin levels in blood during physiological pregnancy]. *Minerva Ginecol.* 1991;43(9):387–91.
- Casanova BF, Sammel MD, Macones GA. Development of a clinical prediction rule for iron deficiency anemia in pregnancy. *Am J Obstet Gynecol.* 2005;193(2):460–6.
- Milman N, Byg KE, Agger AO. Hemoglobin and erythrocyte indices during normal pregnancy and postpartum in 206 women with and without iron supplementation. *Acta Obstet Gynecol Scand.* 2000;79(2):89–98.
- Mei Z, Cogswell ME, Parvanta I, Lynch S, Beard JL, Stoltzfus RJ, et al. Hemoglobin and ferritin are currently the most efficient indicators of population response to iron interventions: an analysis of nine randomized controlled trials. *J Nutr.* 2005;135(8):1974–80.
- Milman N. Iron and pregnancy—a delicate balance. *Ann Hematol.* 2006;85(9):559–65.
- Revez L, Gyte GM, Cuervo LG, Casasbuenas A. Treatments for iron-deficiency anaemia in pregnancy. *Cochrane Database Syst Rev.* 2011;(10):CD003094.doi:10.1002/14651858.CD003094.pub3.
- Beaton GH, McCabe GP. Efficacy of intermittent iron supplementation in the control of iron deficiency anaemia in developing countries. An analysis of experience: final report to the Micronutrient Initiative. Toronto: Canadian International Development Agency; 1999. Available from: <http://www.idpas.org/pdf/125EfficacyOfIntermittent.pdf>
- World Health Organization Regional Office for Europe; United Nation’s Children’s Fund Regional Office for Central and Eastern Europe the Commonwealth of Independence States and the Baltic States. Prevention and control of iron deficiency anaemia in women and children. Report of the UNICEF/WHO Regional Consultation. Geneva, Switzerland, 3–5 February 1999. Geneva: UNICEF; 2001. Available from: http://www.who.int/nutrition/publications/micronutrients/anaemia_iron_deficiency/e73102/en/
- Roberfroid D, Huybregts L, Habicht JP, Lanou H, Henry MC, Meda N, et al. Randomized controlled trial of 2 prenatal iron supplements: is there a dose-response relation with maternal hemoglobin? *Am J Clin Nutr.* 2011;93(5):1012–8.
- Bendell J, Benz Jr EJ. Hematologic changes of pregnancy. In: Hoffman R, Benz Jr EJ, Shattil SJ, Furie B, Cohen HJ, editors. *Hematology: basic principles and practice.* Orlando, FL: Churchill Livingstone; 2005. Pp. 2629–32.
- van Buul EJ, Steegers EA, Jongsma HW, Eskes TK, Thomas CM, Hein PR. Haematological and biochemical profile of uncomplicated pregnancy in nulliparous women: a longitudinal study. *Neth J Med.* 1995;46(2):73–85.
- Cao C, O’Brien KO. Pregnancy and iron homeostasis: an update. *Nutr Rev.* 2013;71(1): 35–51.
- Centers for Disease Control and Prevention (US). Recommendations to prevent and control iron deficiency in the United States. *MMWR Recomm Rep.* 1998;47(RR-3):1–29.
- Mumtaz Z, Shahab S, Butt N, Rab MA, DeMuynck A. Daily iron supplementation is more effective than twice weekly iron supplementation in pregnant women in Pakistan in a randomized double-blind clinical trial. *J Nutr.* 2000;130(11):2697–702.
- Souza AI, Batista Filho M, Ferreira LO, Figueirôa JN. Efetividade de três esquemas com sulfato ferroso para tratamento de anemia em gestantes. *Rev Panam Salud Publica.* 2004;15(5):313–9.
- Souza AI, Batista Filho M, Bresani CC, Ferreira LO, Figueirôa JN. Adherence and side effects of three ferrous sulfate treatment regimens on anemic pregnant women in clinical trials. *Cad Saude Publica.* 2009;25(6):1225–33.
- World Health Organization. Haemoglobin concentrations for the diagnosis of anaemia and assessment of severity. Vitamin and Mineral Nutrition Information System. Geneva: WHO; 2011. (WHO/NMH/NHD/MNM/11.1). Available from: <http://www.who.int/vmnis/indicators/haemoglobin.pdf>
- World Health Organization. Serum ferritin concentrations for the assessment of iron status and iron deficiency in populations. Vitamin and Mineral Nutrition Information System. Geneva: WHO; 2011. (WHO/NMH/NHD/MNM/11.2). Available from: http://www.who.int/vmnis/indicators/serum_ferritin.pdf
- Greiner M, Pfeiffer D, Smith RD. Principles and practical application of the receiver-operating characteristic analysis for diagnostic tests. *Prev Vet Med.* 2000;45(1-2):23–41.
- Koenker R, Bassett Jr G. Regression quantiles. *Econometrica.* 1978;46(1):33–50.
- Hallock K, Koenker R. Quantile regression. *J Econ Perspect.* 2001;15(4):143–56.
- Szarfarc SC. Public policies to control iron deficiency in Brazil. *Rev Bras Hematol Hemoter.* 2010;32 Suppl 2:S2–7.
- Côrtes MH, Vasconcelos IAL, Coitinho DC. Prevalence of iron-deficiency anemia in Bra-

- zilian pregnant women: a review of the last 40 years. *Rev Nutr.* 2009;22(3):409–18.
39. Mei Z, Cogswell ME, Looker AC, Pfeiffer CM, Cusick SE, Lacher DA, et al. Assessment of iron status in US pregnant women from the National Health and Nutrition Examination Survey (NHANES), 1999–2006. *Am J Clin Nutr.* 2011;93(6):1312–20.
40. Pavord S, Myers B, Robinson S, Allard S, Oppenheimer C; British Committee for Standards in Haematology. UK guidelines on the management of iron deficiency in pregnancy. *Br J Haematol.* 2012;156(5):588–600.
41. Walker P, Subasinghe R, editors. DNA-based molecular diagnostic techniques: research needs for standardization and validation of the detection of aquatic animal pathogens and diseases. FAO Fisheries Technical Paper 395. Rome: Food and Agriculture Organization of the United Nations; 2000. Available from: <http://www.fao.org/docrep/005/X4946E/x4946e0b.htm>
42. Bresani CC. Utilidade diagnóstica dos parâmetros eritrocitários, da classificação morfológica das hemácias e da ferritina sérica em gestantes de baixo risco: um estudo transversal [dissertation]. Recife, PE: Instituto de Medicina Integral Prof. Fernando Figueira; 2007. Available from: http://www.imip.org.br/site/ARQUIVOS_ANEXO/mestrado%20cristiane%20campello;20071130.pdf
43. Bresani CC, Braga MC, Felisberto DF, Tavares-de-Melo CE, Salvi DB, Batista-Filho M. Accuracy of erythrogram and serum ferritin for the maternal anemia diagnosis (AMA): a phase 3 diagnostic study on prediction of the therapeutic responsiveness to oral iron in pregnancy. *BMC Pregnancy Childbirth.* 2013;13:13.

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RESUMEN

Precisión diagnóstica de los valores de hemoglobina para determinar la carencia de hierro durante el embarazo: revelación de los resultados de un estudio clínico citado

Objetivo. Analizar la precisión de las concentraciones de hemoglobina (Hb) como indicador diagnóstico de la carencia de hierro en mujeres embarazadas, y medir la eficacia del tratamiento con hierro por vía oral empleando las puntuaciones z de Hb en lugar de sus valores absolutos.

Métodos. Se determinaron la sensibilidad y la especificidad de un valor de Hb < 11,0 g/dl, y su curva de eficacia diagnóstica (ROC, por sus siglas en inglés), en el diagnóstico de la carencia de hierro (ferritina sérica [FS] < 12,0 ng/ml) en 318 mujeres que estaban en su segundo trimestre de embarazo y habían sido sometidas a tamizaje para un estudio clínico llevado a cabo en el noreste del Brasil el año 2001. Se realizó un análisis secundario de la eficacia del tratamiento con hierro mediante el empleo de los datos de tres pautas de tratamiento diferentes incluidas en el estudio (60 mg de hierro por vía oral una vez por semana [$n = 46$], dos veces por semana [$n = 50$], y una vez al día [$n = 44$]). Se calcularon las diferencias medias entre los valores absolutos (en g/dl) de Hb y las puntuaciones z (desviaciones estándar [DE]), posteriores y previos al tratamiento, correspondientes a los tres grupos de tratamiento de participantes en el estudio con y sin carencia de hierro.

Resultados. La sensibilidad, la especificidad y el área bajo la curva ROC de la Hb fueron de 60,7, 44,3 y 0,54%, respectivamente. Las mujeres que no tenían carencia de hierro mostraron incrementos de los valores absolutos de Hb (de manera similar a los resultados globales del estudio clínico) pero no presentaron mejores puntuaciones z de Hb (puntuaciones de -0,6 DE [IC de 95%: -0,99, -0,28]; -0,2 DE [IC de 95%: -0,47, 0,08]; y -0,1 DE [IC de 95%: -0,33, 0,18], correspondientes a las pautas de feroterapia de una vez por semana, dos veces por semana y una vez al día, respectivamente). Por el contrario, las mujeres que tenían carencia de hierro y fueron tratadas mediante las pautas intermitentes presentaron reducciones tanto de los valores absolutos como de las puntuaciones z de Hb, respectivamente: una vez por semana = -0,42 g/dl (IC de 95%: -0,72, -0,12) y -1,4 DE (IC de 95%: -1,74, -0,99); dos veces por semana = -0,14 g/dl (IC de 95%: -0,46, 0,17) y -1,1 DE (IC de 95%: -1,44, -0,75).

Conclusiones. Estos análisis revelaron que las concentraciones de Hb no eran un indicador preciso de las necesidades de hierro ni de la respuesta al tratamiento con hierro en las mujeres embarazadas.

Palabras clave

Anemia ferropénica; embarazo; índices de eritrocitos; sensibilidad y especificidad; ensayo clínico; Brasil.