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Anemia, Iron Deficiency, Meat Consumption, and Hookworm Infection in Women of Reproductive Age in Northwest Vietnam

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Abstract. Iron deficiency anemia poses an important public health problem for women of reproductive age living in developing countries. We assessed the prevalence of iron deficiency and anemia and associated risk factors in a community-based sample of women living in a rural province of northwest Vietnam. A cross-sectional survey, comprised of written questionnaires and laboratory analysis of hemoglobin (Hb), ferritin, transferrin receptor, and stool hookworm egg count, was undertaken, and the soluble transferrin receptor/log ferritin index was calculated. Of 349 non-pregnant women, 37.53% were anemic (Hb < 12 g/dL), and 23.10% were iron deficient (ferritin < 15 ng/L). Hookworm infection was present in 78.15% of women, although heavy infection was uncommon (6.29%). Iron deficiency was more prevalent in anemic than non-anemic women (38.21% versus 14.08%, $P < 0.001$). Consumption of meat at least three times a week was more common in non-anemic women (51.15% versus 66.67%, $P = 0.042$). Mean ferritin was lower in anemic women (18.99 versus 35.66 ng/mL, $P < 0.001$). There was no evidence of a difference in prevalence (15.20% versus 17.23%, $P = 0.629$) or intensity (171.07 versus 129.93 eggs/g, $P = 0.412$) of hookworm infection between anemic and non-anemic women. Although intensity of hookworm infection and meat consumption were associated with indices of iron deficiency in a multiple regression model, their relationship with hemoglobin was not significant. Anemia, iron deficiency, and hookworm infection were prevalent in this population. Intake of meat was more clearly associated with hemoglobin and iron indices than hookworm. An approach to addressing iron deficiency in this population should emphasize both iron supplementation and deworming.

INTRODUCTION

It has been estimated that more than one third of the world's women are anemic; the vast majority of this burden occurs in developing countries.[1] This produces an enormous public health impact, because anemia poses a significant mortality and morbidity risk, particularly to women of reproductive age and their children.[2] Iron deficiency is recognized as perhaps the most common cause of anemia worldwide. Even in the absence of anemia, mild to moderate iron deficiency has been associated with impaired physical and cognitive growth of children,[3] reduced work capacity of adolescents and adults,[4] and increased morbidity and mortality in pregnant women and their infants.[5] Iron deficiency exists in a continuum, from a reduction in body storage iron, to iron deficient erythropoiesis, and ultimately to iron-deficient anemia.[6] Women of reproductive age who are iron deficient but not anemic may become anemic during pregnancy as a consequence of increased iron requirements and expanded plasma volume. Hookworm (*Ancylostoma duodenale* and *Necator americanus*) infection causes chronic gastrointestinal blood loss and is the most common worldwide cause of iron deficiency.[7] The impact of hookworm infection on iron stores depends on the intensity of infection and the amount of iron consumed in the diet.

The World Health Organization has suggested that anemia is of "moderate" public health importance where its prevalence is between 20% and 39.9% and "severe" if it occurs in 40% or more of the population. It is recommended that an approach to iron deficiency anemia be especially targeted toward women of reproductive age who risk the attendant consequences of anemia during pregnancy.[8] Integrating this strategy into a community-based program has been found to be effective.[9] A recent meta-analysis has confirmed that antenatal iron supplementation improves both antenatal and postnatal hemoglobin concentration and that side effects of iron supplementation appear more common in women who receive daily iron.[10] For countries in the Western Pacific Region, where anemia prevalence constitutes a public health problem, a strategy of weekly iron supplementation has been recommended, with hookworm control where prevalent.[11]

In Southeast Asia, almost 215 million women are estimated to be anemic. In Vietnam, the prevalence of anemia was 42.8% in 1995,[12] and a recent survey found that 40% of non-pregnant women were anemic.[13] The prevalence of hookworm infection in northern Vietnam has previously been reported as 52%.[14] However, in many districts of Vietnam, the prevalence of anemia, iron deficiency and hookworm infection remains largely unknown. Thus, before the development and implementation of strategies in Vietnam to control iron deficiency anemia, further analysis of these parameters is necessary.

We conducted a cross-sectional survey in Yen Bai province, in northwest Vietnam, to assess the prevalence of and factors associated with iron deficiency and anemia in non-pregnant women of reproductive age. This study represents the baseline assessment before implementing a model program in this area to address anemia and hookworm infection in Vietnamese women of reproductive age.[15]

MATERIALS AND METHODS

Study site and subjects. Yen Bai province was selected for the project because previous surveys conducted by the Na-

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tional Institute of Malariology, Parasitology, and Entomology (NIMPE) had determined that the prevalence of hookworm was high. The province is in a mountainous area of northwest Vietnam, with an area of 6,882.9 km^2. It is relatively poor and contains sizable ethnic minority populations. Malaria has been largely eliminated from the province, with only two imported cases reported in 2005 (Phu L, Director Malaria Control Programme, Yen Bai, Personal Communication).

The province is divided into nine administrative towns and districts. Two districts, Yen Binh and Tran Yen, were chosen for the study. An estimated 51,623 women of reproductive age (16–45 years) live within the study area. Twenty-eight percent of the communes in this area are classified as having very poor economic status.[16]

Villages and women were selected using the population proportional to size cluster sampling method. Thirty-three villages were selected, and in each, 13 women were invited for a cross-sectional baseline evaluation. Sample size was calculated to produce a 90% power ($\alpha = 0.05$) to detect a change in hemoglobin of 10% at follow-up analyses incorporating the cluster-based nature of the survey. A total of 354 women completed the survey and laboratory evaluation.

Laboratory methods. Blood and stool samples were collected from each participant. The intensity of hookworm, *Ascaris lumbricoides*, and *Trichuris triciura* infection was evaluated from the fecal specimen using standard Kato Katz methodology[17] and expressed as eggs per gram of feces (epg). Intensity of hookworm infection was classified according to WHO guidelines: 0 epg classified as nil, up to 2,000 epg classified as light, 2,000–4,000 epg as moderate, and > 4,000 epg classified as severe.[18] The count of eggs per gram of stool has been previously shown to rise with overall gastrointestinal hookworm burden.[19] Hemoglobin was evaluated in the field from finger prick blood, using a hemoglobinometer (HemoCue AB, Angelholm, Sweden). A 3-mL sample of venous blood was collected using a closed collection system into tubes containing fast clotting agent. The tubes were spun at 4,000 rpm for 20 minutes at room temperature, after which serum was collected and stored at −4°C. The samples were transported on dry ice to Sydney, Australia, for analysis by the South Eastern Sydney Illawarra Area Health Service (SEALS). Serum ferritin was measured using a sandwich immunoenzymatic assay (IEA; Beckman Coulter Access Reagents, Fullerton, CA). Soluble transferrin receptor was evaluated using enzyme-linked immunoassay (ELISA; IT; Orion Diagnostica, Espoo, Finland). The ratio of transferrin receptor to log (to base 10) serum ferritin (TfR-F index) was calculated from these results.[20] Serum C-reactive protein (CRP) levels were assessed by an ELISA method. We chose a level of > 10 mg/L to represent a state of inflammation, because this has been previously reported as an appropriate clinical cut-off.[21]

Anemia was defined as a hemoglobin concentration of < 12 g/dL and iron deficiency as a serum ferritin of < 15 ng/mL, except where otherwise stated, in concordance with WHO recommendations for women of reproductive age.[8] Transferrin receptor levels of 2.3 μg/mL or above were considered abnormal, based on the manufacturer's reference interval (0.8–2.3 mg/L). The TfR-F index has been previously shown to provide an excellent indicator of iron reserves, with depletion of iron stores implied once the ratio exceeds 1.8.[22] We adopted this cut-off as an alternative indicator of iron deficiency.

Survey. Participating women were asked to complete a questionnaire covering potential demographic risk factors for iron deficiency and hookworm infection, including dietary meat consumption, ethnic group, education, number of children, domestic sanitary facilities, and frequency of wearing shoes. Current pregnancy status was also assessed. Local village health workers assisted the survey team in administering the survey.

Statistical analysis. The data were entered in the field into spreadsheets using Microsoft Excel (Microsoft Corp., Redmond, WA). The spreadsheets were imported into Stata (Intercooled Stata 9.2 for Windows; StataCorp, College Station, TX) for further analysis. The distribution of laboratory variables was assessed.

Hemoglobin values were approximately normally distributed, enabling use of the arithmetic mean. Ferritin, transferrin receptor, and the TfR-F index were right-skewed; thus, these were log-transformed for subsequent analysis. Hookworm eggs per gram data were right skewed, with a considerable number of zero values. Thus, egg count data were analyzed using the Poisson distribution, incorporating the clustered nature of the survey (by village), with comparisons between groups made by calculating the incidence rate ratio. Egg count data were logarithmically transformed after addition of 1 to facilitate regression calculations.[18] Median meat consumption was determined.

The prevalence of anemia, iron deficiency, hookworm infection, and iron deficiency anemia were calculated using the cut-offs defined above. Average iron indices, hookworm burden, and meat consumption were calculated and compared between those women who were anemic and those who were non-anemic; the analysis was based on a linear regression model that incorporated the effect of clustering by village. Prevalences of iron deficiency, hookworm infestation, and meat consumption were also compared between anemic and non-anemic groups. Comparison of significance of differences between meat consumption in the anemic and non-anemic groups was performed using a two-sample Wilcoxon rank-sum (Mann-Whitney) test.

Multiple regression analysis incorporating the cluster design of the survey was performed to evaluate the effect of hookworm eggs per gram, meat consumption, and the demographic variables of age, number of children, and level of education on hemoglobin and log transformation of iron indices (ferritin, transferrin receptor, and TfR-F index). The regression equation used the log(eggs per gram) value as described above, and as such, the results for association with hookworm are reported after transformation by taking the inverse logarithm.

Ethical considerations. Before the implementation of the project, extensive consultation was undertaken between the project team and the community, as well as careful liaison with village, district, and provincial health staff. The local village health workers provided participants with information regarding the reasons for the survey, and verbal informed consent was obtained at the time of enrollment. The survey team assisted the village health workers where participants experienced concerns or uncertainty relating to any aspect of their participation.

The project was approved by the Human Research Ethics

Committee of the National Institute of Malariology, Parasitology, and Entomology (Hanoi, Vietnam), the Walter and Eliza Hall Institute of Medical Research (Melbourne, Australia), and Melbourne Health (Melbourne, Australia).

RESULTS

The demographic and socio-economic data for the 354 women of reproductive age who participated in the study are presented in Table 1. Given the small sample (5/354), pregnant women were excluded from further analysis. There were nine women for whom pregnancy status was not recorded. The interviewers believed these women were unlikely to be pregnant, based on the degree of familiarity between the village health workers who assisted with the interviews and the participants, and they were included in the analysis. Two women had CRP levels > 10 mg/L, but because neither had hemoglobin < 12 g/dL and thus did not have evidence of anemia of chronic disease, they were included in the analysis. There were no significant differences in any of the demographic or laboratory analysis parameters between the two districts.

The most frequent type of latrine used by women in the district was a simple hole dug in the ground (151/340, 44.4%). Owning a single stool collection box was the second most common (127/140, 37.4%), whereas using a sanitary toilet or pan was infrequent (36/340, 10.6%).

The district and total means for hemoglobin, iron studies, meat consumption, and stool parasite counts, together with prevalences of anemia, iron deficiency, and hookworm infection, are presented in Table 2. The geometric mean eggs per gram was 145.04 (95% CI 105.63–199.01). To indicate the mean worm burden among women who were actually infected, the value was recalculated after the exclusion of the 21.85% who had a count of 0. After this adjustment, the mean was 586.19 (492.90–697.15). Using the Poisson distribution, the mean hookworm egg count was 1,184.48 (952.96–1,472.24).

The prevalence of anemia, defined as a hemoglobin < 12 g/dL, was 37.53% (131/349), and the prevalence of iron deficiency (defined as a serum ferritin concentration < 15 ng/mL) was 23.10% (76/329). Borderline iron depletion (serum ferritin between 15 and 30 ng/mL) was observed in 22.18% (73/329) of participants. The prevalence of soluble transferrin receptor concentration above the cut-off of 2.3 mg/mL was 22.02% (74/336), similar to iron deficiency. The proportion of participants with both low ferritin and high transferrin receptor concentrations was 13.57% (46/339; data not shown). One or both of these markers were outside the cut-off level in 32.33% (107/331), and the TfR-F index was < 1.8 for 19.94% (65/326) of the participants.

The overall prevalence of hookworm infection was 78.15% (261/334); however, most participants (61.68%, 206/334) had a light worm burden.

A comparison of indices of iron stores, hookworm infection, and meat consumption between anemic and non-anemic women is shown in Table 3. The mean indices of iron stores among the anemic group were significantly different to those of the non-anemic group. Median meat consumption was also lower among the anemic population ($P = 0.004$). However, there was no evidence of significant difference in hookworm eggs per gram [incidence rate ratio, 1.31; 95% confidence interval (CI), 0.81–2.11; $P = 0.270$].

A comparison between the prevalence of iron deficiency, meat consumption, and hookworm infection in anemic and non-anemic women is shown in Table 3. Of 123 anemic participants in this analysis, 47 (38.21%) had a ferritin level < 15 ng/mL, compared with 29/206 (14.1%) who were not anemic ($P < 0.001$). There was no evidence of difference in prevalence of borderline iron deficiency (serum ferritin, 15–30 ng/mL) between anemic and non-anemic women. Similar differences between the two groups were observed for the other markers of iron deficiency. The proportion of participants consuming meat more than three times per week was greater among the non-anemic population ($P = 0.042$). Logistic regression analysis showed that meat consumption above the

TABLE 1
Demographic and socioeconomic data for women of reproductive age in two districts of Yen Bai Province, Vietnam

Variable		Yen Binh*	Tran Yen*	Combined*
n		163	191	354
Mean age (95% CI)		32.2 (30.97–33.45)	31.34 (30.31–32.39)	31.73 (30.93–32.53)
Marital status	Married	134 (82%)	166 (87%)	300 (85%)
	Never married	27 (17%)	23 (12%)	50 (14%)
	Divorced	0 (0%)	2 (1%)	2 (1%)
	Widow	1 (1%)	0	1 (< 1%)
Number of children	Median (range)	2 (0–5)	2 (0–5)	2 (0–5)
Ethnicity	Kinh	97 (60%)	136 (71%)	233 (66%)
	Tay	12 (7%)	30 (16%)	42 (12%)
	Cao Lan	16 (10%)	12 (6%)	28 (8%)
	Dao	33 (20%)	12 (6%)	45 (13%)
Education level	Illiterate	18 (11%)	5 (3%)	23 (7%)
	To grade 5	27 (17%)	28 (15%)	55 (16%)
	Grades 6–9	88 (54%)	110 (58%)	198 (56%)
	Grade 10–12	28 (17%)	39 (20%)	67 (19%)
	Higher	2 (1%)	9 (4.7%)	11 (3%)
Current pregnancy status	Pregnant	2 (1%)	3 (2%)	5 (1%)
	Not pregnant	155 (95%)	185 (96%)	340 (96%)†
Frequency of wearing shoes	Never	24 (15%)	17 (9%)	41 (12%)
	Occasionally	91 (56%)	71 (38%)	162 (47%)
	Always	46 (29%)	97 (52%)	143 (41%)

* Failure of percentages to add to 100 implies missing data.
† Nine patients were included for whom pregnancy data were missing but the research team concluded that pregnancy was unlikely.

TABLE 2
Hemoglobin, iron indices, meat intake, and hookworm results for women surveyed in two districts of Yen Bai Province

	Tran Yen	Yen Binh	Combined
Hemoglobin (g/dL)			
N	188	161	349
Mean* (95% CI)	12.11 (11.87–12.35)	12.38 (12.11–12.64)	12.23 (12.06–12.41)
Proportion anemic (%)			
< 12 g/dL	38.30 (31.31,45.29)	36.65 (29.15,44.14)	37.53 (32.43,42.64)
< 10 g/dL	9.57 (5.34,13.81)	6.83 (2.91,10.76)	8.31 (5.40,11.21)
< 7 g/dL	1.60 (−0.20, 3.39)	0.62 (−0.59, 1.83)	1.15 (0.02, 2.27)
Ferritin (ng/mL)			
N	175	154	329
Mean† (95% CI)	27.66 (23.89–32.02)	28.77 (24.38–33.96)	28.17 (25.25–31.43)
Proportion			
Ferritin < 15 ng/mL	22.86 (16.64,29.09)	23.38 (16.69,30.06)	23.10 (18.52,27.68)
Ferritin < 30 ng/mL	48.00 (40.55,55.45)	42.21 (34.35,50.06)	45.29 (39.88,50.70)
Ferritin: 15 to < 30 ng/mL	25.14 (18.72,31.57)	18.83 (12.66,25.01)	22.19 (17.68,26.70)
Transferrin receptor (mg/mL)			
N	178	158	336
Mean† (95% CI)	1.64 (1.53–1.76)	1.74 (1.63–1.86)	1.69 (1.61–1.77)
Proportion			
Transferrin receptor > 2.3	20.22 (14.29–26.16)	24.05 (17.34–30.76)	22.02 (17.57–26.48)
TfR-F index			
N	174	152	326
Mean† (95% CI)	1.20 (1.08–1.33)	1.27 (1.14–1.44)	1.23 (1.15–1.34)
TfR-F index > 1.8	19.54 (13.61–25.47)	20.39 (13.94–26.85)	19.94 (15.58–24.30)
Proportion			
Ferritin < 15 and/or TfR-F index > 1.8	25.14 (18.72–31.57)	26.62 (19.64–33.60)	25.84 (21.08–30.59)
Meat intake			
N	184	160	344
Median‡ (range)	3 (0–14)	3 (0–15)	3 (0–15)
Hookworm (epg)			
N	179	155	334
Mean§ (95% CI)	144.52 (95.61–218.43)	145.64 (88.93–238.12)	145.04 (105.63–199.01)
Proportion with infection			
None	19.55 (13.71–25.40)	24.52 (17.70–31.33)	21.85 (17.40–26.31)
Light	66.48 (59.52–73.44)	56.12 (48.26–63.99)	61.68 (56.43–66.91)
Moderate	9.49 (5.17–13.82)	10.97 (6.01–15.92)	10.18 (6.92–13.44)
Heavy	4.44 (1.42–7.51)	8.39 (3.99–12.78)	6.29 (3.67–8.90)

* Arithmetic mean.
† Geometric mean.
‡ Median serves per week.
§ Geometric mean using log (eggs per gram + 1) and subsequent subtraction of 1. If uninfected women are excluded, mean = 586.19 (492.90–697.15).

TABLE 3
Mean indices of hemoglobin, iron, meat intake, and hookworm and proportions of anemia, iron deficiency, low meat intake, and hookworm infection by anemia classification

Variable	Anemic (Hb < 12 g/dL) *N*	Anemic (Hb < 12 g/dL) Mean (95% CI)	Non-anemic (Hb > 12 g/dL) *N*	Non-anemic (Hb > 12 g/dL) Mean (95% CI)	*P* coefficient (95% CI)
Ferritin*	123	18.99 (15.47, 23.30)	206	35.66 (31.80, 39.98)	< 0.001 0.53 (0.41, 0.68)
Percent ferritin < 15 ng/mL	47/123	38.21 (29.62, 46.80)	29/206	14.08 (9.33, 18.83)	< 0.001
Percent ferritin < 30ng/mL	76/123	61.79 (53.20, 70.38)	73/206	35.44 (28.91, 41.97)	< 0.001
Ferritin 15 to < 30 ng/mL	29/123	23.58 (16.08, 31.09)	44/206	21.36 (15.76, 26.96)	0.640
Transferrin receptor*	126	2.04 (1.86, 2.22)	210	1.51 (1.44, 1.59)	< 0.001 1.35 (1.22, 1.49)
Percent transferrin receptor > 2.3	48/126	38.10 (29.62, 46.57)	26/210	12.38 (7.93, 16.84)	< 0.001
TfR-F index*	122	2.74 (2.11, 3.37)	204	1.16 (1.06, 1.26)	< 0.001 1.71 (1.44, 2.03)
Percent TfR-F index > 1.8	45/122	36.89 (28.32, 45.45)	20/204	16.50 (11.44, 21.57)	< 0.001
Meat consumption†	131	3 (2, 5)	213	3.5 (2, 6)	0.004
Percent meat consumption > 3 servings/wk	67/131	51.15 (42.59, 59.70)	142/213	66.67 (60.34, 73.00)	0.042
Hookworm eggs per gram‡	125	173.07 (103.79, 288.59)	209	131.93 (88.31, 197.11)	0.270 1.31 (0.81, 2.11)
Percent hookworm > 2,000 epg (moderate/severe infection)	19/125	1.20 (8.91, 21.49)	36/209	17.23 (12.11, 22.34)	0.629

* Geometric mean: *P* value calculated by linear regression accounting for clustering.
† Median, interquartile range: *P* value calculated by Wilcoxon rank-sum (Mann-Whitney) test.
‡ Geometric mean using log (eggs per gram + 1), *P*, and incidence rate ratio calculated by Poisson regression accounting for clustering and dispersion.

median (≥ 3 servings/wk) was protective against anemia [odds ratio (OR), 0.52; 95% CI, 0.32, 0.85; $P = 0.009$] and iron deficiency (OR, 0.46; 95% CI, 0.28, 0.76; $P = 0.002$). Prevalence of moderate or severe hookworm infection was not significantly different between the anemic and non-anemic groups. In particular, severe hookworm infection (> 4,000 epg) was not significantly more common among anemic women (7.20%) than non-anemic women (5.74%; $P = 0.595$).

The logarithm of hookworm eggs per gram was not associated with age (coefficient, –0.03; 95% CI, –0.10, 0.04; $P = 0.392$), level of education (coefficient, 0.06; 95% CI, –0.43, 0.56; $P = 0.794$), or number of children (coefficient, 0.32; 95% CI, –0.20, 0.84; $P = 0.224$), but there was weak evidence of inverse association with meat consumption (coefficient, –0.17; 95% CI, –0.35, 0.00; $P = 0.048$). By logistic regression, ethnicity was not associated with moderate/heavy hookworm infection (OR, 1.12; 95% CI, 0.87, 1.44; $P = 0.387$), anemia (OR, 1.15; 95% CI, 0.97, 1.35; $P = 0.109$), or ferritin level < 15 ng/mL (OR, 1.11; 95% CI, 0.96, 1.28; $P = 0.168$). Hookworm, hemoglobin, and iron indices were not associated with frequency wearing of shoes or type of latrine used. Consumption of meat was not associated with age or number of children but was positively associated with increased level of education on regression analysis (coefficient, 1.19; 95% CI, 0.84, 1.55; $P < 0.001$).

Table 4 shows the results of a multiple regression analysis performed to determine the relationship between dependent variables (hemoglobin, iron indices) and amount of meat consumption, degree of hookworm infection, and demographic factors. There was a positive association between meat consumption and both hemoglobin and iron indices. Hemoglobin was not associated with hookworm eggs per gram, but iron indices, particularly ferritin, were. Hemoglobin and iron indices were unrelated to age, number of years of education, and number of children of women of reproductive age. After education, age, and number of children were withdrawn from the model (data not shown), hemoglobin was unrelated to hookworm eggs per gram (coefficient, 0.98; 95% CI, 0.92, 1.04; $P = 0.506$) but was associated with meat consumption (coefficient, 0.07; 95% CI, 0.00, 0.14; $P = 0.042$). Hookworm (coefficient, 0.93; 95% CI, 0.89, 0.98; $P = 0.007$) and meat intake (coefficient, 0.07; 95% CI, 0.04, 0.11; $P < 0.001$) were both associated with logarithmic transformation of ferritin. Transferrin receptor was not associated with hookworm (coefficient, 1.00; 95% CI, 0.98, 1.02; $P = 0.795$), but there was a weak association with meat intake (–0.02; 95% CI, –0.04, 0.00; $P = 0.074$). The results were similar for transferrin receptor/log ferritin index and associations with hookworm (coefficient, 1.02; 95% CI, 0.99, 1.06; $P = 0.200$) and meat intake (coefficient, –0.04; 95% CI, –0.07, –0.01; $P = 0.006$).

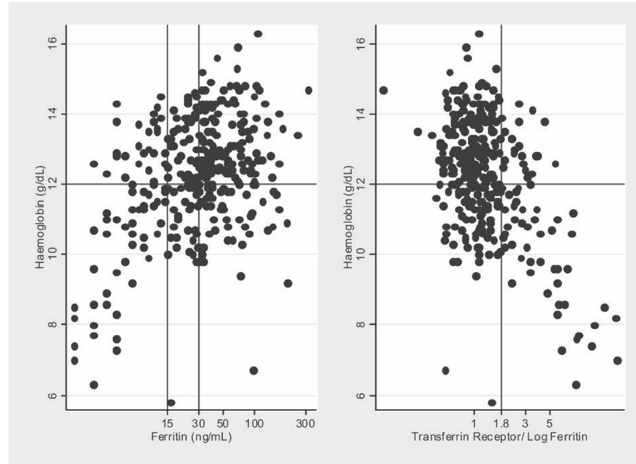

Figure 1. Relationship between hemoglobin and (A) ferritin and (B) transferrin receptor/log ferritin in women from Yen Bai Province, Vietnam.

Figure 1 shows the log linear relationship between iron indices and hemoglobin. The graphs also show subsets of women who were anemic but not iron deficient, and iron deficient but not anemic. The coefficient of the slope between hemoglobin and (log)ferritin was 0.70 ($P < 0.001$; 95% CI, 0.53, 0.86), between hemoglobin and (log)soluble transferrin receptor was –1.60 ($P < 0.001$; 95% CI, –1.98, –1.23), and between hemoglobin and (log)TfR-F index was –1.18 ($P < 0.001$; –1.40, –0.95).

DISCUSSION

We found that the prevalence of anemia and iron deficiency was 37.5% and 23.1%, respectively, in women of reproductive age in two districts of a rural province of northwest Vietnam. Iron deficiency was more common among anemic women, although less than one half of the cases of anemia could be attributed to iron deficiency. Both iron deficiency and anemia were associated with a lower weekly consumption of meat. Hookworm infection was common and, although not associated with anemia in this population, was correlated with iron deficiency. Demographic indicators such as age, number of children, and education were not associated with iron deficiency or anemia.

The main limitation of the study was that, in < 10% of

TABLE 4

Multiple regression analyses of risk factors (meat consumption, hookworm infection, demographic) as predictors of hemoglobin and iron indices

	Hemoglobin		Log (ferritin)		Log (TfR)		Log (TfR/F index)	
	Coefficient (95% CI)	*P*	Coefficient (95% CI)	*P*	Coefficient (95% CI)	*P*	Coefficient (95% CI)	*P*
Meat consumption (servings/wk)	0.09 (0.02, 0.16)	0.020	0.08 (0.03, 0.13)	0.002	−0.02 (−0.04, 0.01)	0.185	−0.04 (−0.08, −0.01)	0.020
Hookworm* (epg)	0.99 (0.92, 1.06)	0.770	0.91 (0.86, 0.97)	0.003	1.01 (0.98, 1.03)	0.567	1.03 (1.00, 1.07)	0.087
Age (years)	0.00 (0.04, 0.03)	0.918	0.00 (-0.03, 0.02)	0.601	0.00 (−0.01, 0.01)	0.718	0.01 (−0.01, 0.02)	0.548
Education (years)	−0.18 (−0.46, 0.10)	0.195	−0.08 (−0.26, 0.10)	0.365	0.00 (−0.07, 0.07)	0.984	0.03 (−0.07, 0.14)	0.508
Number of children (*N*)	−0.03 (−0.34, 0.28)	0.848	−0.01 (−0.23, 0.25)	0.939	0.01 (−0.07, 0.01)	0.984	0.02 (−0.16, 0.20)	0.835

* Values for coefficient and confidence intervals (CIs) are inverse log to base e of calculated figure because of log(eggs per gram) being used in regression equation. Note that inverse log(0) = 1.

cases, demographic data and test results were missing. This was because of occasional unanswered questions at the time of interview, and in some cases, low volumes of venous blood taken, such that not all laboratory analyses could be performed for all participants. Although we have no reason to believe that this biased the analysis, it remains a possibility. A proportional cluster sampling method was used to ensure that the findings were representative for the 51,623 women living in Tran Yen and Yen Binh districts. However, these districts were relatively easily accessible, and the relationship between iron deficiency, anemia, diet, and hookworm infection may differ in more remote districts in Vietnam.

Associations between hemoglobin and iron with meat consumption were not part of the *a priori* hypothesis. However, the association is robust ($P < 0.05$) and physiologically feasible, although the coefficient for association was not especially high (0.07 for both hemoglobin and ferritin). It is unlikely that our study was underpowered to detect an association between hookworm and hemoglobin because the inverse logarithm of the confidence interval for the association was narrow (0.92, 1.04) and centered around 1.

In this study, iron deficiency was more prevalent among the anemic population, but only approximately one third of women with anemia had a low serum ferritin. The true prevalence of iron deficiency anemia may be higher than this because ferritin is an acute phase protein that may be elevated in inflammation. To address this, we used alternative indices of iron deficiency—soluble transferrin receptor and the TfR-F index[20]—and confirmed that the prevalence of iron deficiency was similar using the TfR (> 2.3 μg/mL), the TfR-F index (> 1.8), and serum ferritin (< 15 ng/mL), both in the overall and in the anemic groups. This suggests that the newer indices are comparable to ferritin for detecting iron deficiency in this population. Combining the results of both serum ferritin and the TfR-F index provides a higher prevalence of iron deficiency, both overall and among the anemic group, implying that the cases detected by each method do not completely overlap. Iron depletion (ferritin = 15–30 ng/mL) was present in 23.58% of anemic women (and a similar proportion of the non-anemic sample), raising the possibility that iron supplementation may have a greater than expected impact in the long term because both anemic and non-anemic women would be at risk of developing or exacerbating iron deficiency anemia should their iron requirements increase, for example, during pregnancy.

Inadequate iron intake, particularly caused by reduced access to heme iron, which is chiefly found in meat and is highly bioavailable, can contribute to iron deficiency.[23] Our analysis showed that meat intake was associated with iron status, and there was a strong trend toward a positive correlation with hemoglobin. This suggests that failure of adequate iron intake is an important contributor to iron deficiency in this region. Although demographic factors were not associated with outcomes of iron deficiency or anemia, there was an association between education and increasing meat intake. This suggests better educated women can afford, or make it a priority to include, meat in their diet. These findings provide a rationale to include regular iron fortification or supplementation in any strategy aimed at alleviating iron deficiency. A large survey in Vietnam has recently shown that low meat intake is associated with iron deficiency anemia,[13] but unlike our findings, it found that hookworm infection was the stronger risk factor. Similar findings were found among pregnant women. Hookworm infection has been previously found to affect appetite,[24] and we found a subtle inverse association between hookworm and meat intake.

This study showed that hookworm infestation is prevalent in northwest Vietnam, with 78.14% of women in the study infected. However, heavy infection (> 4,000 epg) was relatively uncommon. The mean hookworm burden remained "light," even if the uninfected population was removed from the analysis, suggesting that hookworm burden was low among those who were infected. This may be explained by recent school-based deworming activities conducted in the area. Increasing intensity of hookworm infestation was related to iron deficiency, suggesting that hookworm is a contributing factor to gastrointestinal blood loss in women in this area of Vietnam.

The lack of an association with anemia may be caused by the low prevalence of moderate and heavy hookworm infection and the large number of anemic women who were not iron deficient, suggesting other causes of anemia may be prevalent in this population. Other nutritional deficiencies, such as of folate, vitamin B_{12},[25] and vitamin A,[26] have been shown to contribute to anemia. Inflammation can produce an anemia of chronic disease, characteristically associated with an elevated CRP and ferritin.[21] However, the finding of a normal CRP in > 99% of the study group makes it unlikely that this had an impact on the results. Genetic hemoglobinopathies, such as α thalassemia and hemoglobin E may also cause anemia. These have been found to be common among certain populations in Southeast Asia[27] and may have contributed to anemia in some of the study participants.

Our study confirmed that iron deficiency anemia is a major public health problem in women of reproductive age in northwest Vietnam. Lack of dietary iron and hookworm infection both contribute to iron deficiency, and therefore, a program of iron supplementation and deworming would be beneficial in this area. Further study to define the other causes of anemia will be important for planning national anemia prevention strategies.

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