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Iron status of adolescent girls from two boarding schools in southern Benin

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Abstract

Iron deficiency (ID) is the most prevalent micronutrient deficiency in the world, particularly in developing countries. Blood samples and a qualitative FFQ on Fe- and vitamin C-rich foods were obtained in 180 adolescent girls aged 12 to 17 years living in two boarding schools from south Benin. ID, defined as serum ferritin either <20 μg/l or 20–50 μg/l, plus two of the following parameters: serum Fe < 11 μmol/l, total iron-binding capacity > 73 μmol/l or transferrin saturation < 20 %, was found in 32 % of subjects. Anaemia (Hb < 120 g/l) was found in 51 % of adolescents, while 24 % suffered from iron-deficiency anaemia (IDA) (ID and Hb < 20 g/l). After adjusting for confounding factors (age, mother's and father's occupation, household size) in a logistic regression equation, subjects having a low meat consumption (beef, mutton, pork) (<4 times/week) were more than twice as likely to suffer from ID (OR = 2·43; 95 % CI 1·72, 3·35; $P = 0·04$). Adolescents consuming less fruits (<4 times/week) also had a higher likelihood of suffering from ID (OR = 1·53; 95 % CI 1·31, 2·80; $P = 0·03$). Finally, subjects whose meat consumption was low were twice as likely to suffer from IDA (OR = 2·24; 95 % CI 1·01, 4·96; $P = 0·04$). The prevalence of ID represents an important health problem in these Beninese adolescent girls. A higher consumption of Fe-rich foods and of promoters of Fe absorption (meat factor and vitamin C) is recommended to prevent ID deficiency in these subjects.

Keywords
Iron-deficiency anaemia
Food consumption
Adolescents
Boarding schools
Benin

Iron deficiency (ID) continues to be the most prevalent micronutrient deficiency in the world, particularly in developing countries[1,2], with the WHO estimating that ID occurs in about 66–80 % of the world's population[3]. The detrimental public health effects of ID include anaemia, decreased intellectual and work performance, and functional alterations of the small bowel[4,5]. Besides other vulnerable age groups such as infancy and early childhood, adolescence is also considered a high-risk period for developing ID owing to the combination of rapid physical growth and losses of Fe through menstruation[4].

Recent work has focused on the inadequacy of methods for obtaining global estimates of ID, which are calculated indirectly from indices of anaemia such as Hb or haematocrit (Ht) concentration that are neither sensitive nor specific to ID[6,7]. These measures fail to detect mild to moderate forms of ID, which even in the absence of anaemia may be associated with functional impairments. The accuracy of ID assessment is improved by combining Hb or Ht with independent measures of Fe status, such as serum ferritin (SF). This method is particularly useful in a tropical context, where confounding factors (liver disease, infection, inflammatory processes, protein malnutrition, etc.) may interfere with the significance of each test of Fe status[6,8,9]. The usual approach of separating normal from Fe-deficient subjects on the basis of only one criterion inevitably involves errors in the diagnosis of both normal and Fe-deficient subjects. Unfortunately, surveys performed in Africa were based only on a few laboratory parameters which did not reflect the true prevalence of ID[6,8,9]. In Benin, only two studies carried out in the 1980s have defined ID using a multi-parameter index composed of two or more abnormal values in the four independent indicators of Fe status used (SF, transferrin saturation (TFS), erythrocyte protoporphyrin and mean corpuscular volume (MCV))[8,10]. Although a recent study performed in a group of adolescent girls from Benin also defined ID using a multi-parameter index, SF, a measure of the first stage of ID, was not included[11].

In Benin, the results of a limited number of studies indicate that ID is a significant public health problem[8,10,11]. However, most of these studies were performed around 20 years ago and included adolescent girls

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as a part of a larger sample of adult women, pregnant women or children[8,10]. Overall, the prevalence of ID has been estimated to range from 34 to 41 % among menstruating women[8,10]. To our knowledge, only one study published in 2007 was conducted in adolescent girls[11]. In that study, the prevalence of anaemia was 43 %.

Moreover, the impact of promoters and inhibitors of Fe absorption has not been studied in Beninese adolescents. Yet studying Fe status and food consumption of adolescents provides an opportunity to understand and intervene at a point in the life cycle before potential problems become serious in later life. The aims of the present study were therefore: (i) to assess the Fe status of adolescent girls; and (ii) to examine the association between food consumption evaluated by means of a qualitative FFQ and ID or iron-deficiency anaemia (IDA).

Subjects and methods

Study population

The present cross-sectional study was carried out from October to November 2005. The study population consisted of adolescent girls aged 12 to 17 years living in two boarding schools of south Benin, i.e. Lycée Toffa 1er of Porto-Novo (*n* 80) and CEG1 Bertrand Dagnon de Ouidah (*n* 100). The selected schools were located in two departments, Ouémé and Littoral, and are 70 km apart. These schools were selected on the basis of similarity of cafeterias' menus and supply of local foods. Baseline data including blood samples and a qualitative FFQ on Fe- and vitamin C-rich foods were collected in both schools. This step was followed by the implementation of a nutrition intervention programme to reduce IDA in adolescent girls boarding at Lycée Toffa 1er of Porto-Novo (intervention school); CEG1 Bertrand Dagnon de Ouidah served as control school. Results obtained at baseline are presented herein, while data on the nutrition intervention will be published elsewhere[12].

Recruitment

Meetings with personnel from the two boarding schools, as well as with adolescent girls and their parents, were carried out in order to explain the study protocol. Participants were selected from lists of students at each of the selected boarding schools, which included names and dates of birth. Only girls aged 12–17 years were included in the present study. At the beginning of the study, Lycée Toffa 1er of Porto-Novo included 1680 girls living at home (92 %) and 140 girls who were boarding at the school (8 %), while CEG1 Bertrand Dagnon de Ouidah counted respectively 721 girls (83 %) and 148 girls (17 %) in these categories. Among girls boarding at school, eighty girls (57 % of 140 girls) from Lycée Toffa 1er and 100 girls (68 % of 148 girls) from CEG1 Bertrand Dagnon de Ouidah were selected in the sixth to the ninth grade to participate in the study. Thus, 108 girls (38 % of 288 girls) who were boarding in the two schools were not included in the study. Twelve girls (6 %) aged 12 to 17 years did not participate in the study: seven refused to participate, three suffered from malaria and two were not present at the time of the study. Also, the girls who were not included in the sample were under 12 years of age (*n* 9) or older than 17 years of age (*n* 87). Written consent was required from both parents in order for their child to participate in the study. Data were collected using an FFQ and laboratory analysis of blood. The subjects received oral and written notification of test results. Approval was obtained from the Ethical Committee of Laval University and the Departmental Direction of the Primary and Secondary Education of the concerned departments in Benin.

General questionnaire and FFQ

Each study participant was interviewed by the first author using a standardized and pre-tested questionnaire which included information on socio-economic characteristics (age, religion, parents' occupation and their level of education, number of rooms and running water in the house) and health history (recent febrile and non-febrile illnesses, menstrual history, history of recent drug use, main source of information on health issues, home hygiene). Also, a qualitative FFQ that assessed the frequency with which specific food items were consumed during a given period (≥4 times/week, 2–3 times/week, 2–3 times/month, <1 time/month) was administered[13]. The selected food items were meat (beef, mutton and pork), poultry, fish, legumes, vegetables, fruits and coffee (tea consumption is not a food habit in Beninese adolescent girls and it was not served in the boarding school cafeterias). These food items were selected as indicators of vitamin C, haem Fe, non-haem Fe, phytate and polyphenol intake.

Anthropometric data

Participants' weight was determined to the nearest 0·1 kg on an electronic load cell scale (Precision Health Scale, UC-300; A&D Company Limited, Japan). Height was measured to the nearest 1 cm with a stature meter (2 m), with a movable bar and a steel tape, mounted on a wall. All girls wore light clothing and removed their shoes. BMI was calculated as weight (kg)/square of height (m^2). BMI was plotted on individual percentile charts developed by the National Center for Health Statistics[14]. These charts are specific for age and gender: underweight was defined as BMI <5th percentile, normal weight as BMI between 5th and 85th percentile, overweight as BMI between 85th and 95th percentile, obesity as BMI >95th percentile.

Blood sampling

A venous blood sample ($\approx$5 ml) was drawn from each non-fasting adolescent into an EDTA-containing Vacutainer tube (Becton Dickinson, Plymouth, UK) and sent to TOXI-LABO laboratory in Cotonou (Benin) for automated blood analysis. A haematology analyser (Sysmex KX-21; Sysmex Corporation, Kobe, Japan) was used for full blood count and differential white blood cell count. The full blood count analysis included: Hb concentration, Ht, MCV, mean corpuscular Hb concentration (MCHC) and mean corpuscular Hb (MCH).

To obtain serum for determination of parameters of Fe status, a blood sample ($\approx$5 ml) was drawn from each adolescent into a trace mineral-free Vacutainer tube (Becton Dickinson) and allowed to coagulate at room temperature. The coagulated blood was centrifuged and the serum was divided into aliquots for the measurement of SF, serum Fe, total iron-binding capacity (TIBC), TFS and C-reactive protein (CRP). An enzyme-linked fluorescent assay (Vidas Ferritin; Biomérieux, Marcy-l'Etoile, France) was used to determine SF. Serum Fe was measured without deproteinization using a commercial photometric test (Ferrimat-Kit; Biomérieux). After saturation of transferrin by the addition of Fe, the same assay system was used for determination of TIBC. TFS was calculated by expressing serum Fe as a percentage of TIBC. CRP was detected using a latex agglutination test (Slidex CRP kit; Biomérieux).

Laboratory measurements were considered to be abnormal at the following levels: SF $<$ 20 μg/l, serum Fe $<$ 11 μmol/l, TIBC $>$ 73 μmol/l, TFS $<$ 20 %, Hb $<$ 120 g/l, Ht $<$ 37 %, MCV $<$ 80 fl, MCHC $<$ 300 g/l, MCH $<$ 30 pg, CRP $>$ 6 mg/l[15–17].

Prevalence of iron deficiency and iron-deficiency anaemia

Perturbations in Fe nutriture can be classified into three stages: Fe depletion, ID and IDA. During the depletion phase, Fe stores are exhausted; however, decrease of serum Fe or anaemia is not present. ID occurs as Fe stores decline and a decrease in transport Fe is present. Anaemia and hypochromia are still not detectable. IDA occurs as the synthesis of Fe-containing proteins, such as Hb, becomes compromised to the point at which values fall below a specified cut-off value[13,18,19].

Malaria, inflammation and infection disorders are common in West African populations and can influence Fe status indices. Thus they complicate the accurate detection of ID and may decrease the effect of additional dietary Fe by reducing the body's ability to absorb and utilize it[20]. SF value $<$20 μg/l is often used to indicate reduced Fe stores. SF, however, is an acute-phase protein that increases with malaria, inflammation or infection disorders[13]. In an attempt to adjust for the observed high prevalence of malaria and other infection disorders, we increased the SF cut-off value to 50 μg/l[21–23].

In the present study, participants were categorized as Fe-deficient (ID) if they presented SF value of either $<$20 μg/l or 20–50 μg/l plus two abnormal values among the three following biochemical parameters: serum Fe $<$ 11 μmol/l, TIBC $>$73 μmol/l or TFS $<$ 20 %[16]. The adolescents were classified as having IDA if they were Fe-deficient and had Hb $<$ 120 g/l.

Statistical analysis

Statistical analyses were performed using the commercially available statistical software NCSS 2004 (NCSS, Kaysville, UT, USA). Descriptive statistics are presented as either means with their standard deviation in the case of continuous variables or frequency for categorical variables. The χ^2 test was used to test for differences in characteristics between girls from the two boarding schools, Fe status indices and CRP values. Although both schools were chosen on the basis of similarity of cafeterias' menus and supply of local foods, each adolescent girl was free to choose the amount of foods she desired in accordance with her taste and appetite. For example, in the case of white rice with tomato sauce and fried fish, some adolescents chose only white rice and tomato sauce, while others took only tomato sauce and fried fish. This would have an impact on their Fe status. Also, as mentioned above, it was necessary to obtain baseline data in each school before implementation of the nutrition intervention programme whose results will be published elsewhere[12].

The independent samples *t* test (two-tailed) procedure was used to compare group means for Fe status indicators. SF had a skewed distribution; therefore, this parameter was log-transformed for all statistical calculations and converted back to the original units as means and SD.

The association between food consumption and ID or IDA (0 = absence; 1 = presence) was examined by multiple logistic regression analyses which adjusted for potential confounders. The following variables were included in the multivariate model because they had been previously identified as potential confounders in the published literature: age (1 = 12–14 years, 2 = 15–17 years); parents' occupation (1 = high-level non-manual employees, 2 = medium-level non-manual employees, 3 = manual workers) and household size (1 = ≤5 persons, 2 = >5 persons). Consumption of meat (beef, mutton and pork), poultry, legumes, vegetables, fruits and coffee was classified into two groups (1 = <4 times/week, 2 = ≥4 times/week) for the logistic regression analysis presented in Tables 4 and 5. However, consumption of fish was classified as follows: 1 = <5 times/week, 2 = ≥5 times/week. Statistically significant differences were indicated by $P < 0{\cdot}05$.

Results

Subjects' characteristics

The characteristics of the subjects are shown in Table 1. The mean age of adolescents was 14·5 (SD 1·8) years and 51 %

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Table 1 Characteristics of subjects: adolescent girls living in two boarding schools of south Benin, 2005

Variable	Overall (*n* 180)	School: Lycée Toffa 1er (*n* 80)	School: CEG1 Ouidah (*n* 100)	*P**
Age (years)				
12–14	51·1	58·7	45·0	0·036
15–17	48·9	41·3	55·0	
BMI (kg/m^2)				
Underweight (<5th percentile)	8·3	3·7	12·0	NS
Normal (5th to 85th percentile)	80·6	82·5	79·0	
Overweight (85th to 95th percentile)	7·8	8·8	7·0	
Obese (>95th percentile)	3·3	5·0	2·0	
Use of vitamin/mineral supplements during last two months before interview†	19·1	22·5	16·0	NS
Religion				
Catholic	73·8	72·5	75·0	NS
Muslim	5·6	7·5	4·0	
Protestant	7·8	8·7	7·0	
Others‡	12·8	11·3	14·0	
Mother's education				
Illiterate	15·0	10·0	19·0	0·001
Literate	85·0	90·0	81·0	
Primary	14·5	15·0	14·0	
Secondary	23·9	35·0	15·0	
University	5·5	10·0	2·0	
Not available	41·1	30·0	50·0	
Father's education				
Illiterate	4·1	1·3	6·0	0·001
Literate	95·9	98·7	94·0	
Primary	8·2	3·8	12·0	
Secondary	17·6	20·7	16·0	
University	20·6	34·5	9·0	
Not available	49·5	39·7	57·0	
Mother's occupation				
High-level non-manual employee§	5·0	8·7	2·0	0·018
Medium-level non-manual employee‖	15·0	16·3	14·0	
Manual worker¶	77·8	70·0	84·0	
Deceased	2·2	5·0	0	
Father's occupation				
High-level non-manual employee§	19·4	15·0	23·0	NS
Medium-level non-manual employee‖	29·4	35·0	25·0	
Manual worker¶	45·6	47·5	44·0	
Deceased	5·6	2·5	8·0	
Household size				
≤5 persons	17·8	18·7	17·0	NS
>5 persons	82·2	81·3	83·0	

*Comparison between boarding schools using the χ^2 test; NS, $P > 0·05$.
†Almost 10 % of adolescents had taken Fe (200 mg iron fumarate) and folic acid (0.25 mg) supplements, whereas 6 % had taken one tablet of effervescent UPSA vitamin C (500 mg) while 4 % used both vitamin and mineral supplements.
‡Others: Eckankar, celestial Christianity and Vaudou.
§High-level non-manual employees: business executives, doctors, engineers and university teachers.
‖Medium-level non-manual employees: nurses, accountants and high-school teachers.
¶Manual workers: vehicle mechanics, metal workers, construction workers and retailers.

were less than 15 years old. The average BMI of the girls was 20·1 (SD 3·8) kg/m^2. Of the total surveyed population, 3 % were obese, 8 % were overweight and 8 % were found to be underweight. The majority of the study population (81 %) was within the normal weight range. Approximately 20 % used vitamin and mineral supplements during the last two months before the study: about 10 % of the adolescents took Fe and folic acid supplements daily (one tablet containing 200 mg iron fumarate and one tablet containing 0·25 mg folic acid), whereas 6% took vitamin C (one tablet of effervescent UPSA vitamin C containing 500 mg vitamin C). Finally, 4% used both vitamin and mineral supplements. More than 70 % of the girls were Catholic. Muslims represented about 6 % of the sample, while other religions (Eckankar, celestial Christianity and Vaudou) comprised 13 %. Religious beliefs are important because certain foods may be considered taboo by the rules promulgated by a religion concerning what is and what is not allowed to be eaten. For example, pork is forbidden in the Islamic faith, while Catholics are required to perform some specific acts of penance, which includes fasting and abstaining at times each year, especially during Lent.

In the present study, about 86 % of mothers were literate and only 5 % went to university, while 96 % of fathers were literate and 21 % went to university. Also, more than two-thirds of the adolescents' mothers were

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Table 2 Iron status indices and C-reactive protein concentration among subjects: adolescent girls living in two boarding schools of south Benin, 2005

		School		
Indicator	Overall (*n* 180)	Lycée Toffa 1er (*n* 80)	CEG1 Ouidah (*n* 100)	*P**
SF (μg/l)				
Mean	40·4	41·7	39·2	NS
SD	29·9	36·1	24·1	
% with value <20 μg/l	25·0	30·0	21·0	NS
Serum Fe (μmol/l)				
Mean	17·8	16·9	18·7	NS
SD	6·3	6·3	6·3	
% with value <11 μmol/l	4·5	6·2	3·0	NS
TIBC (μmol/l)				
Mean	72·7	73·8	71·7	NS
SD	9·8	10·0	10·3	
% with value >73 μmol/l	45·6	46·2	45·0	NS
TFS (%)				
Mean	25·1	23·3	26·5	0·02
SD	9·5	8·8	9·9	
% with value <20 %	26·7	36·2	19·0	0·04
Hb (g/l)				
Mean	120·0	119·0	120·9	NS
SD	10·6	10·3	10·9	
% with value <120 g/l	50·5	52·5	49·0	NS
Ht (%)				
Mean	37·4	35·9	38·6	0·0001
SD	3·4	2·6	3·5	
% with value <37 %	35·0	51·2	22·2	0·0001
MCV (fl)				
Mean	82·8	81·4	83·8	0·02
SD	6·6	5·8	6·9	
% with value <80 fl	28·9	32·5	26·0	NS
MCHC (g/l)				
Mean	321·4	330·9	313·8	0·0001
SD	16·9	14·6	14·8	
% with value <300 g/l	10·0	1·2	17·0	0·0001
MCH (pg)				
Mean	26·6	26·9	26·3	NS
SD	2·5	2·3	2·6	
% with value <30 pg	92·8	95·0	91·0	NS
CRP (mg/l)				
Mean	15·8	10·5	18·0	NS
SD	15·7	9·0	7·0	
% with value >6 mg/l	7·2	5·0	9·0	NS

SF, serum ferritin; TIBC, total iron-binding capacity; TFS, transferrin saturation; Ht, haematocrit; MCV, mean corpuscular volume; MCHC, mean corpuscular Hb concentration; MCH, mean corpuscular Hb; CRP, C-reactive protein.
*Comparison between boarding schools using Student's *t* test (means) or the χ^2 test (%); NS, $P > 0·05$.

manual workers (78 %). Almost half of the fathers were manual workers (46 %); a lower proportion of them, about 20 %, were high-level non-manual employees (business executives, doctors, engineers and university teachers). The majority of families consisted of six or more individuals. No significant differences were observed between subjects from the two boarding schools for the following variables: religion of the subjects, father's occupation, household size, use of supplements during the last two months and BMI. However, age of the adolescents, mother's occupation and parents' education differed significantly between the two boarding schools.

Haematological and biochemical parameters

Means (SD) and percentage of abnormal values for Fe status indices and CRP concentration are presented in Table 2. Mean values were significantly lower in Lycée Toffa 1er compared with CEG1 Ouidah for TFS ($P = 0·02$), Ht ($P = 0·0001$) and MCV ($P = 0·02$), and significantly higher for MCHC ($P = 0·0001$).

Of the 180 subjects included in the survey, 25 % had low SF (<20 μg/l) while 5 % had low serum Fe (<11 μmol/l). Also, 46 % had elevated TIBC (>73 μmol/l) indicative of ID, while more than a quarter of subjects had TFS below 20 %. A low Hb level according to WHO reference values was observed in half of the adolescents, whereas about a third of them had low Ht and MCV values. Finally, a small proportion of adolescents (10 %) had low MCHC, while the majority of adolescents (93 %) had insufficient MCH. Low CRP was detected in 7 % of subjects. The percentage of abnormal values was significantly higher in Lycée Toffa 1er compared with

CEG1 Ouidah for TFS ($P=0{\cdot}04$) and Ht ($P=0{\cdot}0001$) and significantly lower for MCHC ($P=0{\cdot}0001$).

Prevalence of anaemia, iron deficiency and iron-deficiency anaemia

The prevalence of anaemia, ID and IDA is shown in Table 3. As indicated in this table, the prevalence of anaemia was 51 %. Thirty-two per cent of adolescents were classified as Fe-deficient and had SF value of either <20 μg/l or 20–50 μg/l, plus two abnormal values in the following three biochemical parameters: serum Fe < 11 μmol/l, TIBC > 73 μmol/l or TFS < 20 %. Twenty-four per cent suffered from IDA. There was no difference in the prevalence of anaemia, ID or IDA in adolescents from the two boarding schools. Also, using the χ^2 test, the prevalence of ID ($P=0{\cdot}24$) and IDA ($P=0{\cdot}15$) was not lower in adolescent girls who took vitamin and mineral supplements during the two months preceding the interview (results not shown).

Food consumption and iron status

The qualitative FFQ permitted us to gather information on food consumption patterns of these adolescents. In Lycée Toffa 1[er], breakfast always included a maize-based porridge (*gbagba*) or milk chicory made with powdered skimmed milk or concentrated milk, while in CEG1 Bertrand Dagnon de Ouidah it included porridge made from corn flour and hot water (*akassa*) or rice porridge with milk. For lunch and dinner, girls in both schools received, for example: *pâte rouge* (spicy corn paste served with tomato sauce) and fried fish (mostly mackerel); white rice with tomato sauce and fried fish; white rice with fried sardines; chicken in peanut/tomato sauce; *riz au gras* (beef stew with onions, tomatoes, oil and rice); legumes (red or white kidney beans) with *gari* (the dried and ground form of cassava) or *atassi* (a stiff corn flour porridge) and served with fried fish or fried chicken; *pâte rouge* accompanied by fresh or fried fish and *crain-crain* (a sludgy green sauce made with tossa jute) or okra (results not presented). Thus, food consumption patterns were very similar in the two boarding schools.

Results of the multiple logistic regression analyses that investigated the relationship between ID and IDA and food consumption are presented in Tables 4 and 5, respectively. The majority of girls suffering from ID and IDA had a low consumption frequency of meat, poultry, legumes, vegetables and fruit (<4 times/week). In contrast, three-quarters of the subjects consumed fish ≥5 times/week. Also, the majority of adolescents had coffee less than 4 times weekly.

Girls with a frequency of meat consumption of <4 times/week were twice as likely as those who consumed meat ≥4 times/week to suffer from ID (OR = 2·43; 95 % CI 1·72, 3·35; $P=0{\cdot}04$) and IDA (OR = 2·24; 95 % CI 1·01, 4·96; $P=0{\cdot}04$). Likewise, adolescents with a low frequency of fruit consumption (<4 times/week) also had a higher risk of suffering from ID (OR=1·53; 95 % CI 1·31, 2·80; $P=0{\cdot}03$).

Table 4 Results of multiple logistic regression analyses of the association of food consumption with iron deficiency* among adolescent girls living in south Benin, 2005

Promoters and inhibitors of Fe absorption	Percentage†	Adjusted OR	95 % CI	*P*‡
Meat§				
<4 times/week	77·2	2·43	1·72, 3·35	0·04
≥4 times/week	22·8	1·00	–	
Poultry				
<4 times/week	89·5	2·33	0·81, 6·72	NS
≥4 times/week	10·5	1·00	–	
Fish				
<5 times/week	13·3	1·03	0·44, 2·41	NS
≥5 times/week	86·7	1·00	–	
Legumes				
<4 times/week	82·4	1·56	0·66, 3·70	NS
≥4 times/week	17·6	1·00	–	
Vegetables				
<4 times/week	70·2	0·70	0·11, 4·48	NS
≥4 times/week	29·8	1·00	–	
Fruits				
<4 times/week	77·2	1·53	1·31, 2·80	0·03
≥4 times/week	22·8	1·00	–	
Coffee				
<4 times/week	57·9	0·54	0·26, 1·09	NS
≥4 times/week	42·1	1·00	–	

*Adjusted for age, parent's occupation and household size.
†Food consumption of girls suffering from iron deficiency (*n* 57).
‡NS, $P>0{\cdot}05$.
§Meat: beef, mutton, pork.

Table 3 Prevalence (%) of iron deficiency and iron-deficiency anaemia among subjects using multiple criteria: adolescent girls living in two boarding schools of south Benin, 2005

		School		
	Overall (*n* 180)	Lycée Toffa 1[er] (*n* 80)	CEG1 Ouidah (*n* 100)	*P**
Anaemia†	50·6	52·5	49·0	NS
Iron deficiency‡	31·7	37·5	27·0	NS
Iron-deficiency anaemia§	23·9	26·2	22·0	NS

*Comparison between boarding schools using the χ^2 test; NS, $P>0{\cdot}05$.
†Defined as Hb < 120 g/l.
‡Defined as serum ferritin <20 μg/l or 20–50 μg/l, plus two abnormal values in the following three biochemical parameters: serum Fe <11 μmol/l, total iron-binding capacity >73 μmol/l or transferrin saturation <20 %.
§Defined as Fe-deficient and Hb < 120 g/l.

Table 5 Results of multiple logistic regression analyses of the association of food consumption with iron-deficiency anaemia* among adolescent girls living in south Benin, 2005

Promoters and inhibitors of iron absorption	Percentage†	Adjusted OR	95 % CI	P‡
Meat§				
<4 times/week	83·0	2·24	1·01, 4·96	0·04
≥4 times/week	17·0	1·00	–	
Poultry				
<4 times/week	92·5	2·44	0·72, 8·31	NS
≥4 times/week	7·5	1·00	–	
Fish				
<5 times/week	24·5	1·05	0·41, 2·67	NS
≥5 times/week	75·5	1·00	–	
Legumes				
<4 times/week	83·0	1·66	0·26, 10·53	NS
≥4 times/week	17·0	1·00	–	
Vegetables				
<4 times/week	77·4	0·99	0·41, 2·39	NS
≥4 times/week	22·6	1·00	–	
Fruits				
<4 times/week	75·5	1·31	0·62, 2·77	NS
≥4 times/week	24·5	1·00	–	
Coffee				
<4 times/week	69·8	0·48	0·35, 1·63	NS
≥4 times/week	30·2	1·00	–	

*Adjusted for age, parent's occupation and household size.
†Food consumption of girls suffering from iron-deficiency anaemia (*n* 43).
‡NS, $P > 0·05$.
§Meat: beef, mutton, pork.

Discussion

The majority of adolescents in the present study were within the healthy weight range. The average BMI of these adolescents (20·1 (sd 3·8) kg/m^2) is slightly higher than the BMI of 19·3 (sd 0·1) kg/m^2 and 19·5 (sd 0·2) kg/m^2 reported respectively in 214 migrant Senegalese adolescent girls aged 14·5–16·6 years and 405 girls aged 8–16 years living in an urban area of Ashanti in Ghana[24,25]. However, it is slightly lower than BMI of 21·5 (sd 3·8) kg/m^2 observed in a previous study conducted in 2003 in southern Benin among a group of 100 adolescent girls aged 14–16 years[11]. Also, in the latter study, the prevalence of underweight was lower (5·0 % *v.* 8·3 %) while the rate of overweight was higher (11·5 % *v.* 7·8 %), which explains the higher BMI. In comparison with African data, the prevalence of overweight is higher for US students, with 26·5 % of 1089 girls aged from 12 to 19 years found to be overweight[26].

Adolescence is a critical period for the development of IDA[27]. Its prevalence shows great variability in different countries, and for different races and socio-economic levels. The worldwide prevalence of anaemia during adolescence is 15 % (27 % in developing countries and 6 % in developed countries)[2]. In the present study, the prevalence of anaemia was high and reached 51 %. Similar results were reported by Alaofè *et al.*[11] in 100 Beninese adolescent girls from Lycée Toffa 1er, fifty boarding at the school and fifty living outside the boarding environment. In these subjects, the prevalence of anaemia was 36 % and 50 %, respectively, although the difference was not significant[11]. About 20 years ago, Hercberg *et al.*[8,10] reported similar results in 102 menstruating women aged 22–40 years (46 %) and in 517 menstruating women (mean age 30·0 (sd 0·3) years) of south Benin (42·4 %). To our knowledge, no other study has been conducted in Benin to evaluate the prevalence of anaemia in adolescents or menstruating women.

The high prevalence of anaemia found in these studies is consistent with data from surveys performed in other West African countries, including Ghana and Mali[28,29]. However, these figures are lower than what was reported previously from community- and school-based cross-sectional surveys of adolescent populations from Zanzibar (62·3 %) and Tanzania (62·6 %)[30,31]. While one may be tempted to attribute these differences to nutritional and socio-economic discrepancies between Benin and other areas in sub-Saharan Africa, they may also be due, at least partly, to the difficulties in comparing boarding school and school- or population-based studies.

Serum Fe, TIBC and, more recently, free erythrocyte protoporphyrin measurements provide information on the adequacy of the Fe supply to the erythroid marrow. SF is directly proportional to the body level of Fe stores[5,6]. However, the use of each of these indicators is problematic in a tropical context because of the frequency of confounding factors which may interfere with their significance and may be responsible for false positives or false negatives. In such contexts, no single measurement when used alone may be considered specific enough to identify ID[5,6,32]. As suggested in other studies[33,34], in the current research ID was defined by at least two abnormal values among the three parameters of Fe status used. According to this multiple criteria model, we found that almost 32 % of the adolescent girls were Fe-deficient which is similar to the prevalence seen among menstruating women from Côte d'Ivoire (35 %)[20]. However, a lower prevalence of ID was observed in adolescent girls from Kenya (19·8 %), Kazakhstan (13 %), Turkey (20·8 %) and Malaysia (26·4 %)[4,35–37]. These studies were based on a definition of ID that relied solely on measures of SF levels below 12 μg/l and TFS below 16 %[35] or on a single variable (SF < 15 μg/l[4], SF < 12 μg/l[36]). This partly explains the lower prevalence of ID.

Adolescents enrolled in the present study came from an urban area and were living in boarding schools. The prevalence of ID observed in these subjects is most likely lower than in adolescent girls from the general population[8,10]. Hence, the sampling method may have skewed the sample towards subjects from middle- to upper-class socio-economic strata, a factor that would undoubtedly influence many determinants of Fe status including dietary intake. Although socio-economic status was not measured

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specifically, the geographic location of the participating boarding schools was in a middle-income department of south Benin. Moreover, about 86 % of mothers were literate. Recently, researchers in Brazil and Benin have revealed that the lowest mean Hb concentrations were associated with fewer years of maternal schooling[11,38,39].

Moreover, in the present study, the majority of girls had a low consumption frequency of meat, poultry, legumes, vegetables and fruits. Several researchers have documented low Fe intakes in Beninese populations[10,11]. Hercberg *et al.*[10] indicated that the typical diet in this area is usually monotonous, containing cereals (maize) and/or roots (cassava) with negligible quantities of meat, fish and ascorbic acid. This type of diet is poor in Fe and contains a preponderance of foods rich in phytate and tannins, which inhibit Fe absorption. These results were confirmed by Alaofè *et al.*[11], who found that 73 % of 100 adolescents (fifty boarding at school and fifty living at home) had estimated absorbable Fe intake below the average requirement for absorbed Fe. Comparative data describing differences in dietary intake and lifestyle determinants of Fe status between adolescents in Benin and other countries are lacking.

Girls who consumed meat <4 times/week were twice as likely as those who consumed meat more often (≥4 times/week) to suffer from ID. Similar results were observed with fruit consumption. Higher meat consumption was also related to a lower risk of IDA. Meat is as an excellent source of bioavailable Fe while the vitamin C contained in fruits and vegetables enhances Fe absorption, thus reducing the possibility of ID[40,41]. Tympa-Psirropoulou *et al.*[42] also reported that infrequent consumption of meat, fish, eggs, vegetables and fruit were risk factors for IDA. Inadequate consumption of vegetables and a previous history of pica were reported as risk factors for anaemia by Kara *et al.*[2] in a group of 400 students aged 14 to 16 years. In Benin, cost, availability and food habits affect fruit and meat consumption. In spite of the fact that adolescent girls in the present study took most of their meals in the school cafeteria, they were free to buy street foods with their own money. Some fruits rich in vitamin C, such oranges, mango, papaya and bananas, are seasonal and less abundant in the dry season, the period during which the study was conducted. Also, the consumption of fruit at meal times, which can increase non-haem Fe absorption, is not part of Beninese food habits. Finally, the low meat intake in the present population might be due to the higher cost of some animal products such as beef and/or insufficient cooking skills concerning less expensive but highly nutritious meat such as 'organ' meats. Alaofè *et al.*[11] reported that the mean intake of meat in a group of adolescent girls from Lycée Toffa 1^er^ was low, 71·1 (SD 17·9) g/d for boarders and 45·7 (SD 19·2) g/d for adolescents living at home. In the present study, fish was not significantly related to ID or IDA. That it was consumed by the majority of subjects most likely explains why it was not a contributing factor.

Limitations are inherent to all FFQ and must be considered when reviewing the data presented here. Specifically, the reliability of FFQ data is influenced by each subject's ability to accurately recall their food choices and consumption patterns over extended periods of time[43]. Also, qualitative FFQ do not include portion size estimates, which does not allow the derivation of energy and selected nutrient intakes[13]. Over- and underestimation of intakes are also a notable limitation of all self-reported dietary assessment data. However, in spite of all of these limitations, significant associations were found between low meat and fruit consumption and ID.

Considering the fact that adolescents enrolled in the present study do not constitute a representative sample of all Beninese adolescents, we do not know whether Beninese adolescents in general are more at risk for ID than other subgroups of the population. As mentioned earlier, these young girls possibly have better Fe status which would indicate that the situation concerning anaemia and ID in Benin is alarming. A survey of 126 pregnant women in Cotonou found anaemia in 55 % and ID in 73 %[44]. Also, in 586 subjects living in a rural district of Benin, IDA was present in 68 % of children from 6 months to 2 years of age and in 54 % of children aged 2–14 years[8]. Surveys simultaneously including at-risk groups such as young children, adolescents and adult women are needed to estimate adolescents' vulnerability to both ID and anaemia relative to other population groups.

Conclusion and recommendations

In conclusion, the present study shows that ID is a serious public health problem in these Beninese adolescent girls. Also, the study confirms the observation by Gibson[13] that 'the use of several different indices of iron status simultaneously provides a more valid assessment of iron status than any single measurements because the misclassification that can occur due to overlapping normal and abnormal values for a single measure is minimized and differentiate the severity of iron deficiency more rapidly'.

Increasing access to animal foods and fruits is recommended in order to improve the Fe status of these adolescent girls. Animal foods are high in haem iron and are enhancers of non-haem Fe absorption. Vitamin C in vegetables and fruits also enhances non-haem Fe absorption. In fact, these are household dietary strategies that can be used to alter the content of Fe absorption modifiers in plant-based diets and thus successfully alleviate ID in developing countries[45]. Also, strategies to reduce ID must include fortification of staple foods with Fe and, when necessary, the use of Fe supplements. Finally, to enhance effectiveness and sustainability, all strategies should be integrated with ongoing national food, nutrition and health education programmes, and implemented using education and social-marketing techniques.

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