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Original article

Study on etiology of iron deficiency anemia and effect of iron supplementation in women of childbearing age

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Summary. *Objective:* Iron deficiency is the most common nutritional deficiency and is more likely occur at certain times in life such as adolescence, pregnancy and breastfeeding. The main objective of this study is to assess the dietary iron intake, analyze the levels of some iron deficiency biomarkers and to assess the effect of iron supplement (ferrous sulphate) in women of child bearing age living in Al Riyadh. *Methods:* A structured performa was designed and filled with patients consent to get demographic and health information. Twenty four hour dietary recall method was used for dietary assessment and biochemical tests were performed to analyze the indicators of iron level in serum and to assess the effect of iron supplement (ferrous sulphate) in women of child bearing age. *Results:* Results shows that 50% subjects were of normal weight and majority of the subjects had enough family income. In this study the dietary intake of iron and vitamin C was far below then the DRI and it was revealed that 27.08% of the subjects were iron deficient as 12.5% of the women's showed abnormal (lower value) mean cell volume, serum ferritin and transferrin saturation and 14.58% had abnormal serum ferritin and MCV. It was noted that iron supplementation had significant ($p \geq 0.05$) effect on total iron binding capacity, transferrin, ferritin, red blood corpuscles and hemoglobin. *Conclusion:* This study infers that majority of the subjects didn't have any iron supports. Iron supplementation resulted in improvement in iron status. Nutritional education should be provided to women of childbearing age for increasing nutritional awareness.

Key words: ferritin, hemoglobin, iron, ferrous sulphate, iron supplementation

Introduction

Iron deficiency is the most common form of nutritional deficiency and continues to be the leading single nutrient deficiency in the world (1), affecting millions of people; in spite of significant efforts to decrease its occurrence. Iron deficiency anemia (IDA) is characterized by deficiency of hemoglobin resulting in abnormal and insufficient red blood cells containing little hemoglobin. The main risk factor of IDA includes pure vegetarian diet, less iron intake, poor absorption of iron from diets high in phytates or phenol compounds (2), repeated pregnancies, menorrhagia, less awareness about importance of iron supplements, increased plasma volume during pregnancy (3). There are various functional consequences of anemia (4). Anemia affects the quality of life by decreasing fetal growth (5), affecting mental development and learning capacity, work performance, cognitive functioning, immunity and thermoregulation (6).

In Saudi Arabia, children and women are particularly affected by such a disease (1). In previous studies the daily average intake of iron for childbearing women was 11 mg/day (7) and 10.2 mg/day (1). Randomized controlled trials specified that universal supplementation with iron to meet pregnancy requisite reduced the risk of low birth weight and preterm births (8). Iron supports can be considered as an effective treatment in maintaining iron level (9). Supplementation of folic acid before pregnancy reduces the occurrence of birth de-

fects, especially neural tube defects (10). Till date hardly any studies are available on the effect of iron supports on iron deficiency indexes in blood in women of child bearing age living in Al Riyadh. The main objective of this study was to assess the dietary iron intake, to analyze the prevalence of iron deficiency anemia and to assess the effect of iron supplement on iron deficiency indexes in blood in women of child bearing age.

Methods

Research design

A descriptive cross sectional survey approach was used in present study to assess the prevalence and knowledge regarding anemia among women.

Sample and sampling technique

Total of 48 women of reproductive age group (15-49 years) was recruited through random sampling method. Initially a house to house survey was done to identify the women and then through lottery method desired sample of women was chosen out by some another person who was not aware about the study (11).

Ethical considerations

The study was approved by the Deanship of Graduate Studies, King Saud University and study was in accordance with the Policy of Research Centre, King Saud University. The aim of this study was explained to all participants. Written consent was obtained from the respondents involved in this research and the study abided by the principle of voluntary participation. Blood was withdrawn by a qualified nurse and each subject was assured that the information given was solely for scientific purposes and would be kept confidential.

Inclusion criteria

All women of child bearing age who are not taking any iron supplements were included in this study.

Exclusion criteria

All the women's lesser than 18 and higher than forty nine years, patients on non-prophylactic antibiotics, pregnant and lactating women, smokers, women's receiving erythropoietin, i.e. iron therapy or blood transfusion in the previous 3 months or women with any chronic disease were excluded from the study.

Demographic characteristics

A structured performa was designed and filled with subjects consent while interviewing them to get their demographic information. Their names were not required on the questionnaire and each subject was assured that the information given was solely for scientific purposes and would be kept confidential. A set of multiple choice questions related to causes, preventions, symptoms and control of anemia, approved by panel of experts (in Arabic) were used to find the response of women on the importance of having iron supplementation and factors affecting iron absorption.

Anthropometric measurements

Anthropometric measurements in this study included the body weight and height. A self reported height and weight were used to calculate BMI i.e BMI = weight (kg)/height^2 (m)^2. Women's were classified as underweight if their BMI was <18.5, normal if it was between 18.5 and 24.9, overweight if it was 25-29.9 and obese if their BMI was greater than 30 (12-14).

Dietary assessment

Twenty four hour dietary recall method (15) was used for dietary assessment which was then analyzed by using food processor software (16). Subjects were interviewed by the nutritionist to report all food and beverages they had consumed during a 24 hour period (for 3 days). Then each item was described as much as possible. The subjects were instructed to report everything eaten or drunk, including meals, snacks juices and tea etc. for the past 24 hrs. The questionnaire included details regarding the exact portion of the food consumed (serving size), type of food (low fat, full fat,

skimmed, sugar free, fortified), method of preparation (boiled, deep fried, baked, grilled), brand name (commercial, ready to eat), parts consumed (whole, half, quarter). Participants were given a validated two-dimensional (such as food photographs or computer graphics) or 3-dimensional (e.g. food models, household measures (eg- a glass, a slice, a teaspoon, a plateful) or real food samples (17-19).

Biochemical tests

Women fasted for eight hours before venipuncture. They consumed fefol (500 mg folic acid, 150 mg ferrous sulphate) and after completing three months they underwent blood analysis again. Hematology Analyzer Abbott Cell Dyn 3700 was used to estimate some basic blood components (red blood cells (RBC), hemoglobin and hematocrit) according to method of Riedinger and Rodak (20). Serum ferritin level was measured as reported by Mortal (21). Serum iron and total iron binding capacity (TIBC) was measured calorimetrically with kits purchased from Technicon (Tarrytown, New York).MCV was calculated by the following formula: MCV= hematocrit (%)*10/ RBC (million mm^3). According to CDC (22); iron deficiency status is based on 3 laboratory tests: mean cell volume (MCV), serum ferritin (SF), and transferrin saturation (TS). To be diagnosed as iron deficient, the individual had to have an abnormal value of 2 or more of these indicators. The cutoff value for low hemoglobin was less than 120 g/l (according to WHO criteria for diagnosis of anemia). Mean cell value was considered low if the value was less than 80 fl. Accepted lower limits for SF were values less than 12 μg/l for adults.
Transferrin saturation was calculated by dividing serum iron by total iron binding capacity, and cut-off point was less than 16% (23).

Statistical analysis

Data were analyzed using SPSS statistical software package (version 22, NY) and expressed as mean ±standard deviation. The differences among the dietary treatment groups were analyzed by ANOVA at a significance level of $p \leq 0.05$; if significant differences were found, a Post-hoc analysis using Duncan's multiple range tests was performed.

Results

Table 1 explores that maximum number of participants were from the age group of 18-24. By classifying subjects according to body mass index (BMI) it was found that only 50% subjects were of normal weight. Almost 63% subjects were high school passed

Table 1. Demographic information of women of childbearing age

Variables	Numbers	Percentage
Age		
18-24	30	62.5
25-31	8	16.7
32-40	7	14.6
41 and above	3	6.3
BMI		
Less than 18	4	8.3%
18.5-24.5	24	50%
25-29.9	10	20.8%
30 and above	10	20.8%
Education level		
High	30	62.5%
Secondary	12	25%
Intermediate	3	6.3%
Postgraduate	3	6.3%
Monthly income		
1000-3000	17	35.4
3000-6000	2	4.2
6000-9000	9	18.8
More than 9000	20	41.7
Occupation		
Housewife	11	22.9
Employee	5	10.4
Student	32	66.7
Marital status		
Unmarried	31	64.6
Married	17	35.4
Health Condition		
Iron deficiency	8	16.7
No iron deficiency	40	83.3

and 42% subjects had monthly family income more than 9000 riyal. Maximum (66.7%) subjects were students and majority (83.3%) of the women reported absence of any iron deficiency.

Table 2 shows the dietary intake of the subjects and it was found that the average of calories, protein, carbohydrate, fats, iron, vitamin C, vitamin B_{12} and folic acid was 916.46±305.74 cal, 36.35± 14.71 gm, 38.34±16.82 gm, 126.04±36.56 gm, 9.7±4.40 mg, 10.84±10.75 mg, 2.66±1.01 mcg and 379.04±189.96 mcg respectively. Seventy five percent of subject's consumed less than 50% of the recommended DRI of calories. The consumption of protein, fats, carbohydrate, folic acid and vitamin B_{12} was almost within the normal range but range was far below than the normal for iron and vitamin C.

Table 3 shows some indicators of iron levels such as TIBC, transferrin, ferritin, RBC, hemoglobin, and hematocrit and severity of anemia was classified on the basis of hemoglobin concentrations (Fig. 1). In this study 54% women had mild, 4% had moderate anemia and none of them were severe anemic. The average of serum iron and TIBC was 13.94±4.39 and 56.34±9.58 µmol/l respectively. The average of transferrin and ferritin was 283.59±46.09 mg/dl and 22.75±18.18 µg/l respectively. Almost 35.41% subjects had less than 12 µg/l of ferritin. The average of RBC and hemoglobin was 4.49±0.32 million/mm^3 and11.82±1.12g/dl respectively and 58.33% of women had less than 12 g/dl of hemoglobin. The average of hematocrit was 34.70±2.6% and it was found out that 75% of women had less than 36% of hematocrit. The average of MCV and Transferrin saturation was 77.27± 5.02f/l and 25.58 ±9.60% and 64.58% of women had less than 80 f/l of MCV while only 12.5% subjects had saturation % less than 16. From the study it was revealed that 27.08% of the subjects were iron deficient as 12.5% were deficient in mean cell volume, serum ferritin and transferrin saturation and 14.58% were deficient in serum ferritin and MCV.

Despite the importance of having iron supports

Table 2. Dietary intake of women of childbearing age

Nutrients	Intake Amount	DRI	Adequate intake of nutrient (DRI)	Greater than or equal to 50% of DRI	Lesser than or equal to 50% of DRI
Calorie (cal)	916.46± 305.74	2200	41.65%	25%	75%
Protein (gm)	36.35± 14.71	46	79.02%	81.25%	18.75%
Fats (gm)	38.34± 16.82	20-35	139.41%	92.9%	27.1%
CHO (gm)	126.04± 36.56	130	96.95%	95.8%	4.2%
Iron (mg)	9.70± 4.40	18	53.88%	58.3%	41.7%
Vitamin C (mg)	10.84± 9.75	75.01	14.45%	4.2%	95.8%
Folic acid (µg)	379.04± 189.96	400	94.76%	83.4%	16.6%
Vitamin B12 (µg)	2.66± 1.01	2.4	110.83%	95.8%	4.2%

CHO-Carbohydrates, DRI- Dietary Recommended Intake

Table 3. Indicators of iron level in serum of women of childbearing age

Parameters	Average	Standard deviation	Less than minimum	More than maximum	Reference values
Iron (µmol/l)	13.94	4.39	0%	0%	4.6-30.4
TIBC (µmol/l)	56.34	9.58	8.3%	16.66%	45-66
Transferrin (mg/dl)	283.59	46.09	4.16%	0%	210-360
Ferritin (µg/l)	22.75	18.18	35.41%	0%	12-150
RBC(million mm^3)	4.49	0.32	18.75%	0%	4.2-5.9
MCV (fm/l)	77.278	5.02	64.58	0	80-100
Haemoglobin (g/dl)	11.82	1.12	58.33%	0%	12-16
Hematocrit (%)	34.70	2.60	75%	0%	36-48
Transferrin saturation (%)	25.58	9.60	12.5%	0%	15-50

TIBC-Total iron binding capacity, RBC-Red blood Corpuscles

for childbearing women indicated by numerous studies only few Saudi childbearing women (8.3%) had iron supports although 64.6% women believed that iron tablet is important to compensate the loss of iron during menstruation and to prevent anemia during pregnancy. Fifty eight percent of women were not aware that iron tablets after meal reduces the feeling of vomiting. Maximum (68.8%) subjects were aware that iron tablets with citrus increases its absorption but 75% didn't know that iron tablets with anti-acids obstruct iron absorption (Table 4).

Figure 2 (A-D) depicts that an improvement was observed in most of the indicators of iron levels in blood after consuming iron tablets. Eighty percent of the subjects showed an increase in iron, TIBC and transferrin level, 90% of the subjects showed an increase in ferritin level, 70% of the subjects showed an increase in RBC and 100% of the subjects showed an

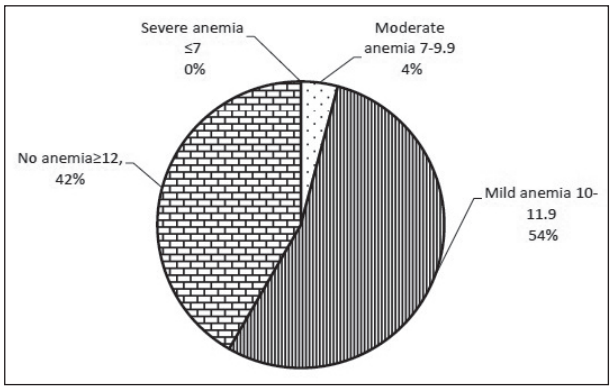

Figure 1. Distribution of anemia on the basis of hemoglobin content

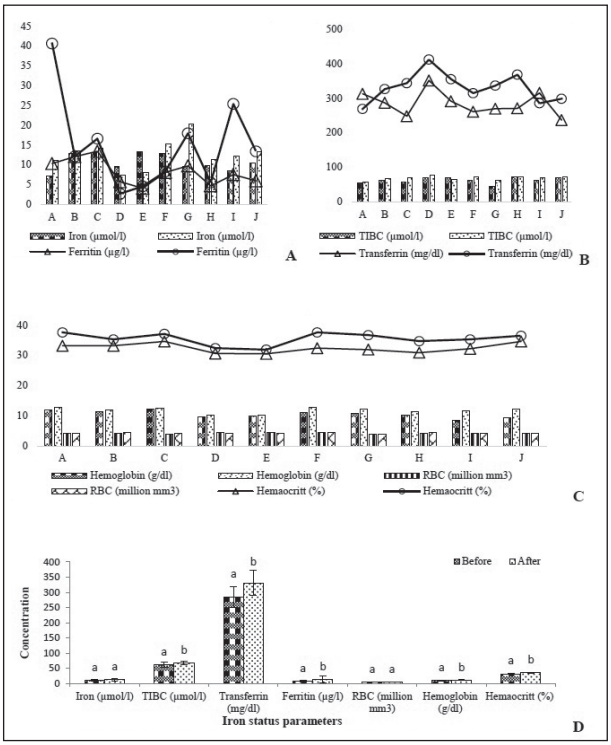

Figure 2. Effect of iron supplementation on biomarkers of iron status. (A-C) Effect of iron supplents on each subject (D) Average of the iron status parameters for the subjects involved in the study. A-J are the subjects. Different letters for each parameter indicates significant effect of supplementation as indicated by ANOVA followed by Duncan's multiple range tests

Table 4. Response of subjects towards the importance of iron supplements and factors affecting iron absorption

S.No	Variables	Percentage
1	Do you have iron supports?	
	Yes	8.3%
	No	91.7%
	I don't know	0%
2	Is it possible to have iron tablets daily during childbearing period?	
	Yes	60.4%
	No	39.6%
	I don't know	0%
3	Is it important for pregnant woman to have iron tablets during childbearing age?	
	Yes	64.6%
	No	0%
	I don't know	35.4%
4	Is it important to have iron tablet during childhood period to prevent anemia?	
	Yes	64.6%
	No	0%
	I don't know	35.4%
5	Is it important to have iron tablets during childbearing age to compensate the loss of iron during menstruation?	
	Yes	64.6%
	No	0%
	I don't know	35.4%
6	Does having iron tablets during childbearing age has other importance?	
	Yes	2.1%
	No	4.2%
	I don't know	93.7%
7	Do iron tablets after meals reduce the feeling of vomiting?	
	Yes	25%
	No	16.7%
	I don't know	58.3%
8	Do Tea, coffee and milk is among materials that reduces iron absorption?	
	Yes	83.3%
	No	4.2%
	I don't know	12.5%
9	Does having iron tablets with orange juice or citrus fruits and juices increases iron absorption?	
	Yes	68.8%
	No	8.3%
	I don't know	22.9%
10	Does having iron tablets with anti-acids obstruct iron absorption?	
	Yes	16.7%
	No	8.3%
	I don't know	75%
11	Does having iron tablets before meal causes the feeling of vomiting and pyrosis?	
	Yes	35.4%
	No	8.3%
	I don't know	56.3%

increase in hemoglobin and hematocrit level. Statistical analysis shows that the increase in iron and hematocrit level was insignificant ($p \leq 0.05$) but the increase in TIBC, transferrin, ferritin, RBC and hemoglobin was significant ($p \geq 0.05$).

Discussion

The rate of iron deficiency anemia remains high in many parts of the world in spite of remarkable effort to reduce this problem. The current nutritional status will decide the wellbeing of the present as well as the future generations (24) and the women's nutritional status is closely tied to their reproductive study (25). Sociodemographic characteristic of the enrolled women showed that 41.6% of subjects were either overweight or obese. This percentage is lesser than previous local study (26). Various factors such as levels of income, education and the size of family members affect the iron level (16). Nutritional status and knowledge regarding anemia was performed in 48 females of child bearing age. Dietary habits within a family should be appropriately examined to identify the inhibitors and enhancers of iron intake (27). Most of the local and global studies focus on the phenomenon of the prevalence of decreased dietary intake of iron. But the dietary intake of iron found in this study was even lesser than the previous reported studies (1,28). The positive effect on iron status will be temporary if the diets do not contain adequate bioavailable iron (29). Vitamin C enhances iron absorption and plays significant role in prevention of anemia. The folic acid plays an important role in maintaining good health, providing a protection against chronic diseases such as anemia, prevent neural tube defects in embryos (30).

The average of serum iron in this study is quiet similar to a previous study in United States (7). In a study on 162 adolescents participants with anemia in India, 104 (64.2%) had mild, 59 (36.2%) had moderate, and only 1 participant (0.6%) had severe anemia which is much higher than the present study (31). In another study in Bangladesh the prevalence of anemia among non-pregnant, ever-married women was 41.3 %. Among anemic women, 35.5 % had mild anemia, 5.6 % had moderate anemia, and 0.2 % had severe anemia (32). In this study none of the women showed serum iron level much below or above the reference range. Adequacy of iron may be due to the haem iron and the absence of its blocking factors such as phytic-containing grains (33) The average of TIBC in this study was lesser than previous study (34). Serum ferritin concentration is an early indicator of the status of iron stores and is the most specific especially when used in conjunction with other tests to assess iron status (35). The level of serum ferritin was quiet less than the value reported by Lahti-Koski *et al*. (28), in Finland but similar to the study of Al- Buhairan and Oluboyede (34) in Saudi Arabia. Ferritin is the common storing source of iron in the human body. Although Ferritin is commonly used for detecting iron deficiency, it is reported that there may be normal serum ferritin values despite a true iron deficiency. Low hemoglobin reflects a later stage of iron deficiency (27). The value of hemoglobin in this study is lesser than the previous studies (1,28). Hemoglobin concentration is the most reliable indicator of anemia. Due to low cost and quick assessment hemoglobin and hematocrit are most commonly used to screen for iron deficiency, and these measures reflect the amount of functional iron in the body but the limitation is that changes in hemoglobin concentration and hematocrit occur only at later stage of deficiency, so both these tests are late indicators of iron deficiency. The average of hematocrit reported in a local study (1) was higher than the value reported in this study. Patterson *et al.* (36), mentioned that if the serum ferritin is low (less than 15 μg/liter) or if they have low iron stores and at least two abnormal values among the three following biochemical parameters: serum iron <10 μmol/l, TIBC≥68 μmol/l and TSAT < 15% than, this is an indication of iron-deficiency anemia. Previous study suggests that iron deficiency anemia is common in Saudi Arabia ranging from 30% to 56% (37).

Supplementation is mostly targeted to the high risk groups of population such as pregnant women, infants and women of reproductive age. A study showed that iron supplements improve the iron status of the mother during pregnancy and during postpartum period even in women who enter pregnancy with reasonable iron stores (38). The main reason of anemia was women's ignorance and irregularity with taking daily

iron supplements. Most of the women were ignorant of the factors affecting iron absorption which indicates the unconsciousness and unawareness of a large section of female regarding such important information.

The dietary style helped in absorption as all indexes have been raised. This result is in line with the result obtained by Verdon's *et al.* (39). Iron supplementation has shown its effectiveness in raising blood iron level of sample and combat iron deficiency anemia in childbearing women (39). Nutritional programs at large scale is still in need to improve performance.

Conclusions

The findings presented here conclude that the diet was found to be deficient in iron and ascorbic acid. Iron supplementation has significant effect on TIBC, transferrin, ferritin, RBC and hemoglobin. Majority of the subjects involved in the study didn't have any iron supports although they were aware of the importance of iron supplements. Consumption of iron supplements are strongly recommended in order to prevent iron deficiency anemia in child bearing age.

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