

Iron Intake and Risk of Ovulatory Infertility

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OBJECTIVE: To evaluate whether iron supplement use or greater intake of total, heme and nonheme iron is associated with lower risk of ovulatory infertility.

METHODS: We conducted a prospective cohort study among 18,555 married, premenopausal women without a history of infertility who attempted a pregnancy or became pregnant between 1991 and 1999 (mean baseline age \pm standard deviation 32.6 \pm 3.6). Diet was assessed twice during follow-up and prospectively related to the incidence of infertility due to ovulatory disorder.

RESULTS: During the 8 years of follow-up, 438 women reported infertility due to ovulatory disorder. Women who consumed iron supplements had a significantly lower risk of ovulatory infertility than women who did not use iron supplements (relative risk 0.60, 95% confidence interval 0.39–0.92), after adjusting for potential confounders. Total nonheme iron intake, primarily consumed as multivitamins and iron supplements, was inversely associated with the risk of infertility (relative risk Quintile 1 compared with 5, 95% confidence interval 0.39–0.92; *P*, trend .005.) Heme iron intake was unrelated to ovulatory infertility in multivariable adjusted analyses.

CONCLUSION: Consumption of iron supplements and nonheme iron from other sources may decrease the risk of ovulatory infertility.

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The inability to become pregnant results in important emotional and economic tolls among those affected. Anxiety and depression are more common in infertile couples than in fertile couples.¹ Assisted reproductive technologies are available to treat infertility, but these are costly.^{2,3} The identification of modifiable risk factors for infertility could lead to inexpensive and effective interventions aimed at prevention.

Although the relation of dietary factors to infertility has not been extensively investigated, they could be important. A small clinical trial testing the efficacy of an iron-containing supplement among women who had unsuccessfully tried to become pregnant documented a higher pregnancy rate in the treatment group.⁴ In addition, women with celiac disease, a condition usually accompanied by deficiencies of iron and other micronutrients, have altered reproductive function, including delayed menarche, early menopause, and infertility of unknown cause,^{5–8} suggesting that some component of this disease may affect ovulatory function. To our knowledge, whether iron intake could influence risk of ovulatory infertility among apparently healthy women without a history of previous infertility is not known. Therefore, we evaluated the association between iron supplement use, total iron intake, and the incidence of ovulatory infertility in a group of women participating in the Nurses' Health Study II.

MATERIALS AND METHODS

The Nurses' Health Study II is a prospective cohort study of 116,671 female registered nurses ages 24 to 42 at the study inception in 1989. The present study is an analysis of incident ovulatory infertility among married women who provided dietary information as part of their participation in the Nurses' Health Study II. The study was approved by the Institutional Review Board of Brigham and Women's Hospital.

Follow-up for the current analysis started in 1991, when diet was first measured. Every two years participants were asked if they had tried to become preg-



nant for more than 1 year without success and to indicate whether their inability to conceive was caused by tubal blockage, ovulatory disorder, endometriosis, cervical mucous factor, spousal factor, was not found, was not investigated, or was caused by other reason. In a validation study among a group of 90 randomly selected women who reported ovulatory infertility in 1989, this diagnosis was confirmed by review of the medical record in 95% of the cases.⁹ Participants were also asked whether they became pregnant during the preceding 2-year period, including pregnancies resulting in miscarriages or induced abortions. Using this information we simulated a cohort of women trying to become pregnant. Only married women, with available dietary information and without a history of infertility, were eligible to enter the analysis. These women contributed information to the analysis during each 2-year period in which they reported a pregnancy or a failed pregnancy attempt and were followed up until they reported infertility from any cause, reached menopause, or underwent a sterilization procedure (themselves or their partner), whichever came first. Ten diabetic women met these criteria. Diabetes has been associated with iron intake^{10,11} and may compromise ovulatory function.¹² However, the small number of diabetics would preclude meaningful statistical adjustment or exploration of modification of associations by diabetes; thus diabetic women were excluded. After exclusions, we identified 18,555 women without a history of infertility who tried to become pregnant or became pregnant between 1991 and 1999.

In each 2-year period, women who met the selection criteria for the study and reported infertility due to ovulatory disorder were considered cases and the remaining women were considered noncases. If a pregnancy and infertility were reported in the same 2-year period it was assumed that infertility preceded the pregnancy.

Dietary information was collected in 1991 and 1995 using a semiquantitative food-frequency questionnaire with more than 130 food items. Participants were asked to report how often, on average, they consumed each of the foods and beverages included in the food-frequency questionnaire during the previous year. The questionnaire had nine options for frequency of intake, ranging from never or less than once per month to six or more times per day. Participants were also asked whether they used multivitamin supplements and other nutrient supplements. Multivitamin users were asked to specify the brand of the multivitamin and its frequency of use. Iron supplement users were asked to specify the daily

dose of their supplement. Nutrient intakes were estimated by summing the nutrient contribution of all food items in the questionnaire, taking into consideration the brand, type, and dose of dietary supplements used. The nutrient content of each food and specified portion size was obtained from a nutrient database derived from the U.S. Department of Agriculture¹³ and additional information obtained from food manufacturers. To reduce extraneous variation in nutrient intakes, these were adjusted for total energy intake using the nutrient residual method.¹⁴

The food-frequency questionnaire has been previously found to be a reasonably valid instrument for measuring iron intake; the correlation coefficient between an estimate of iron intake using the food-frequency questionnaire and the estimated intake from the average of four 1-week dietary records was 0.55 for total iron intake.¹⁵ In a later study, the deattenuated correlation coefficient between food-frequency questionnaire estimates and the average of two 1-week diet records was 0.60 for iron intake excluding supplements.¹⁶ Furthermore, the estimated long-term iron intake using this questionnaire has been found to be associated with biomarkers of body iron stores.¹⁷

We defined dietary intakes in two ways. First, we used the most recent intakes, whereby the 1991 food-frequency questionnaire was used for the 1991–1995 follow-up period and the 1995 food-frequency questionnaire was assigned to the 1995–1999 follow-up. To represent long-term diet we calculated cumulative averaged intakes. Specifically, the 1991 intakes were used to represent diet during the 1991–1995 follow-up period and the average of the 1991 and 1995 intakes was used for the 1995–1999 period. The results obtained using the two methods were similar. Since cumulative averaged intakes may reduce measurement error due to within-person random variation over time,¹⁸ and only results using this method are presented.

The relative risk (RR; calculated as an odds ratio) of ovulatory infertility according to categories of iron intake and iron supplement use was estimated using logistic regression. The generalized estimating equation approach¹⁹ with an exchangeable working correlation structure, was used to account for the within-person correlation in outcomes at different time periods. Women were initially divided into users and nonusers of iron supplements, according to their most recent dietary assessment. Users were further divided into those using a supplement with low iron content (because the categories for the amount of iron differed in the 1991 and 1995 questionnaires, low iron content



was defined as less than 51 mg/d for the 1991 questionnaire and less than 41mg/d for the 1995 questionnaire), those consuming a supplement with high iron content (51 mg/d or more for the 1991 questionnaire and 41mg/d or more for the 1995 questionnaire) and those who did not know the iron content of their supplement. In these models, the RR was computed as the risk of ovulatory infertility among women in a specific level of iron supplement use divided by the risk among nonusers. Then, we divided women into five groups according to quintiles of their intake of total iron, heme iron (found in meats), and nonheme iron (found in supplements and some plant foods). In these models, the RR was computed as the risk of infertility in a specific quintile of intake compared with the risk in the lowest quintile. Tests for linear trend²⁰ were conducted by using the median values of intake in each category as a continuous variable.

To control for confounding by age and calendar time, all models were adjusted for age in years at the beginning of each mailing cycle and calendar time of the current questionnaire cycle. All models of iron intake were adjusted for total energy intake. In multivariable models, we considered body mass index (BMI), parity, smoking history, physical activity, history of contraceptive use, and dietary factors found to be related to infertility in preliminary analyses (intakes of alcohol, coffee, multivitamins, retinol, and α -carotene) or known to affect iron metabolism (vitamin C) as potential confounders of the association. Due to the small number of exposed cases, multivariable models for iron supplements included only terms for variables associated with supplement use and ovulatory infertility that changed the age-adjusted estimate by more than 1% after including them in the model (parity, history of oral contraceptive use, use of multivitamins, and intakes of retinol, vitamin C, coffee, and alcohol) or were thought to be biologically relevant regardless of how much their inclusion changed the original estimates (BMI). Of the variables included in this multivariate model, only parity, use of multivitamins, and retinol intake changed the original estimate by more than 5%. Adjustment for use of other contraceptive methods (injectable or depot progestins, barrier methods, and spermicides) did not affect the original associations, thus adjustment for these variables was not included in the final models. Multivariable models for iron intake included all other potential confounders and additional terms for major types of fatty acids and protein intake to account for any association that might be observed as a result of common food sources. The values of

covariates were updated as new data became available from follow-up questionnaires. Last, terms for multivitamin supplement use and iron supplement use were added to the multivariate models for nonheme iron intake to determine whether these supplements were responsible for any observed association.

We explored whether the association between iron intake and ovulatory infertility was modified by age, BMI, factors affecting iron metabolism (vitamin C intake, history of oral contraceptive use, presence of menstrual cycles longer than 40 days) or was different for primary and secondary infertility by introducing cross-product terms between iron intake, as a linear term, and the variables of interest. The same procedure was used to evaluate whether the association between nonheme iron intake and ovulatory infertility was modified by heme iron intake. All *P* values were 2-tailed. Analyses were performed in SAS 9.1 (SAS Institute Inc., Cary, NC).

RESULTS

During the 8 years of follow-up, 26,971 eligible pregnancies or pregnancy attempts were accrued. Thirteen percent of these events were incident infertility reports, of which 2,165 were women who underwent medical investigation of infertility and 438 were cases of ovulatory infertility (208 among nulliparous and 230 among parous women). At baseline, women differed in some of their characteristics according to their intake of heme and nonheme iron (Table 1). Women with a higher heme iron intake tended to be heavier, smoke, and consume coffee more frequently, to be less physically active, to have a lower vitamin C intake, and to use oral contraceptives more often at the beginning of their first 2-year period in the study. On the other hand, higher nonheme iron intake was associated with less smoking, consumption of coffee, and use of oral contraceptives, higher levels of physical activity, and greater intake of vitamin C. In addition, women with the highest nonheme iron intake were more likely to be parous and to consume less alcohol than women with lower nonheme iron intakes.

In age-adjusted models, use of iron supplements was associated with approximately one half the risk of developing ovulatory infertility compared with nonuse of these supplements ($P<.001$) (Table 2). After subdividing the supplement users by the daily dose of iron, we observed that women consuming iron supplements with low iron content had a similar risk of ovulatory infertility as nonusers, whereas consumption of supplements with high iron content was associated with a 70% lower risk of ovulatory infertility



Table 1. Baseline* Characteristics of the Study Population by Quintiles of Heme and Nonheme Iron Intake (N=18,555)

	Quintiles of Intake					
	Heme Iron			Nonheme Iron		
	1	3	5	1	3	5
Age (y)	32.9	32.5	32.4	32.4	32.9	32.1
Alcohol intake (g/d)	2.8	2.9	2.7	3.5	3.0	1.9
Vitamin C intake (mg/d)	286	236	216	170	248	312
Coffee intake 2 cups/d or more. (%)	21	24	25	27	25	14
Current smoker (%)	5	7	9	12	6	4
Body mass index (kg/m ²)	23.1	24.0	24.8	24.0	23.6	24.4
Physical activity (METs/wk)	24.6	20.7	19.7	19.1	22.0	21.0
Cycles 40 days or more (%)	3	3	3	3	3	3
Hyperandrogenism (%)	0.3	0.4	0.2	0.3	0.2	0.4
Nulliparous (%)	26	22	24	24	26	18
Oral contraceptive use at the beginning of the mailing cycle (%)	14	16	19	21	19	6

MET, metabolic equivalent.

Values are presented as age-standardized means and proportions with the exception of values for age.

* Baseline refers to the year of entry into the study for each individual.

Table 2. Relative Risk and 95% Confidence Intervals for Ovulatory Infertility by Categories of Iron Supplement Use

	Cases/ Noncases*	Age Adjusted [†] RR (95% CI)	Multivariable Adjusted [†] RR (95% CI)
Iron supplement use			
Non users	413/23,563	1.00 (referent)	1.00 (referent)
Users	25/2,970	0.49 (0.33–0.74)	0.60 (0.39–0.92)
Iron supplement dose [§]			
Nonusers	413/23,563	1.00 (referent)	1.00 (referent)
Low iron content	10/610	0.96 (0.51–1.80)	1.13 (0.58–2.18)
High iron content	10/1908	0.30 (0.16–0.57)	0.38 (0.20–0.72)
<i>P</i> , trend		<.001	.003

RR, relative risk; CI, confidence interval.

* A total of 457 supplement users (5 cases and 452 noncases) did not provide information about the iron dose in their supplement.

[†] Adjusted for age (continuous) and calendar time (4 2-year intervals).

[‡] Age-adjusted model further adjusted for body mass index (less than 20, 20–24.9, 25–29.9, 30 or more, and missing), parity (0, 1, 2 or more, and missing), oral contraceptive use (current user, never user, past user 0–23 months ago, past user 24–47 months ago, past user 48–71 months ago, past user 72–95 months ago, past user 96–119 months ago, past user 120 months ago or longer, and missing), multivitamin use (nonusers, 2/wk or less, 3–5/wk, 6/wk or more) and intakes of vitamin C, retinol, and total energy.

[§] The cutoff value for high iron content supplements is 51 mg/day or more for the 1991–1995 follow-up period and 41 mg/day or more for the 1995–1999 follow-up period. The change in cutoff value followed a change in the iron supplement dose question in 1995.

^{||} Calculated with the median iron supplement dose in each category as a continuous variable.

(95% confidence interval [CI] –43% to –84%). Adjustment for BMI and variables changing the age-adjusted estimate by more than 1% attenuated the association, but the same pattern was still present. Further adjustment for smoking, physical activity, and intakes of α -carotene, major types of fatty acids, and protein did not change the results.

Next, we examined the associations between intakes of total iron and heme and nonheme iron intake and occurrence of ovulatory infertility (Table 3). In age and energy-adjusted models, heme iron intake was associated with an increased risk of ovula-

tory infertility, whereas total and nonheme iron intakes were inversely related to this condition. When compared with women in the lowest quintile of heme iron intake, women in the highest category of intake had a 31% greater risk of ovulatory infertility (95% CI 0.97–1.76). Although the estimate for this group did not reach conventional levels of statistical significance, there was a statistically significant trend toward increasing risk of ovulatory infertility with increasing heme iron intake. Further adjustment for potential confounders attenuated this association and the trend lost statistical significance. In contrast, women in the



Table 3. Relative Risks and 95% Confidence Intervals for Ovulatory Infertility by Quintiles of Cumulative Averaged Iron Intake

Type of Iron	Quintile of Intake					P, Trend*
	1	2	3	4	5	
Total iron						
Median intake (mg/d)	11	14	19	37	77	
Cases/noncases	107/5,333	105/5,255	105/5,283	71/5,311	50/5,351	
Age and energy adjusted RR [†]	1.00 (referent)	1.01 (0.77–1.33)	1.01 (0.76–1.33)	0.68 (0.50–0.93)	0.48 (0.34–0.67)	<.001
Multivariable adjusted RR [‡]	1.00 (referent)	0.91 (0.68–1.21)	0.86 (0.63–1.17)	0.66 (0.45–0.97)	0.53 (0.35–0.82)	.003
Heme iron						
Median intake (mg/d)	0.6	0.9	1.0	1.2	1.5	
Cases/noncases	75/4,883	80/5,634	85/5,969	84/4,519	114/5,528	
Age and energy-adjusted RR [†]	1.00 (referent)	0.95 (0.69–1.30)	0.93 (0.68–1.27)	1.19 (0.87–1.63)	1.31 (0.97–1.76)	.02
Multivariable adjusted RR [‡]	1.00 (referent)	1.02 (0.72–1.43)	0.97 (0.69–1.38)	1.20 (0.82–1.75)	1.26 (0.83–1.91)	.17
Nonheme iron						
Median intake (mg/d)	9.7	12.4	18.3	36.3	76.0	
Cases/noncases	98/5,267	109/5,303	109/5,296	72/5,325	50/5,342	
Age and energy adjusted RR [†]	1.00 (referent)	1.13 (0.85–1.48)	1.13 (0.85–1.50)	0.75 (0.55–1.03)	0.52 (0.37–0.73)	<.001
Multivariable adjusted RR [‡]	1.00 (referent)	1.02 (0.76–1.38)	1.00 (0.72–1.37)	0.75 (0.51–1.11)	0.60 (0.39–0.92)	.005

RR, relative risk.

Data are n or relative risk (95% confidence interval).

* Calculated with median iron intake in each quintile as a continuous variable.

[†] Adjusted for age (continuous), calendar time (4 2-year intervals), and total energy intake (continuous).

[‡] Age and energy adjusted model further adjusted for body mass index (less than 20, 20–24.9, 25–29.9, 30 or more, and missing), parity (0, 1, 2 or more, and missing), smoking history (never, past 1–4 cigarettes per day, past 5–14 cigarettes per day, past 15–24 cigarettes per day, past 25 cigarettes per day or more, or unknown amount, current 1–4 cigarettes per day, current 5–14 cigarettes per day, current 15–24 cigarettes per day, and current 25 cigarettes per day or more, or unknown amount), physical activity (less than 3 MET-h/wk, 3–8.9 MET-h/wk, 9–17.9 MET-h/wk, 18–26.9 MET-h/wk, 27–41.9 MET-h/wk, 42 MET-h/wk or more, and missing), oral contraceptive use (current user, never user, past user 0–23 months ago, past user 24–47 months ago, past user 48–71 months ago, past user 72–95 months ago, past user 96–119 months ago, past user 120 months ago or longer, and missing), intake of alcohol (no intake, less than 2 g/d, 2–4.9 g/d, 5 g/day or more), coffee (less than 1 serving per month, 1 serving per month, 2–6 servings per week, 1 serving per day, 2–3 servings per day, 4 servings per day or more), and intake of vitamin C, retinol, and α -carotene, major types of fatty acids and protein.

highest category of nonheme iron intake had about one half the risk of ovulatory infertility when compared with women in the lowest category of intake. After adjusting for potential confounders, women in the highest quintile of nonheme iron intake had a 40% lower risk of ovulatory infertility (95% CI –8 to –61%) when compared with women in the lowest quintile, and a statistically significant linear trend toward decreasing risk with increasing intake was present. The results for total iron intake closely resembled those for nonheme iron intake.

To assess whether multivitamins and iron supplements were the iron sources explaining the association between nonheme iron intake and ovulatory infertility, we introduced terms for these in the multivariable adjusted model for nonheme iron. As expected, inclusion of multivitamin and iron supplements attenuated the association of nonheme iron with risk of ovulatory infertility. The RRs (95% CI) of ovulatory infertility for women in successively higher quintiles of nonheme iron intake were 1.03 (0.76–1.39), 1.03 (0.75–1.43), 0.82 (0.55–1.22), and 0.73 (0.45–1.19), compared with women in the lowest

quintile of intake (*P*, trend .13). In the same model, the RR (95% CI) of ovulatory infertility for women using iron supplements with high iron content was 0.46 (0.24–0.90, *P*=.02), compared with women who did not use iron supplements.

We assessed the possibility that age, parity, and factors that affect iron homeostasis, namely vitamin C intake, menstrual cycle length, and past use of oral contraceptives, could modify the association between iron intake and ovulatory infertility. We found no evidence of interaction of vitamin C intake with iron supplement use (*P*=.53), heme iron intake (*P*=.79), or nonheme iron intake (*P*=.84). Similarly there were no interactions between intake of these kinds of iron and menstrual cycle length (*P*=.32 for iron supplements, *P*=.55 for heme iron, and *P*=.09 for nonheme iron) or history of oral contraceptive use (*P*=.93, *P*=.21, and *P*=.58, respectively). In addition, we found no evidence that these associations were modified by age, BMI, or parity (*P*>.05, in all cases). Last, we examined whether the association between nonheme iron intake and ovulatory infertility was modified by heme iron intake. There was a strong inverse association



between nonheme iron intake and ovulatory infertility among women in the lowest one half of heme iron intake (P , trend $<.001$) but not among the women with higher heme iron intake (P , trend $.22$) (Table 4). The test for interaction, however, was not statistically significant ($P=.78$).

DISCUSSION

We evaluated the association between intakes of different forms of iron and ovulatory infertility and found that use of iron supplements and nonheme iron intake were inversely related to the risk of this disease. Moreover, our data suggested that the consumption of iron from dietary supplements, the most important iron sources in this population, explained some of the association between nonheme iron and ovulatory infertility.

We are unaware of other studies exploring the association between iron intake and infertility resulting from ovulatory disorder or other causes (PubMed, from January 1950 to June 2006, no language restrictions, limited to humans, using the search terms “iron, diet, micronutrients” combined with “fertility, infertility, ovulation”). However, other sources indirectly suggest that iron may have an important role in ovulatory function and fertility. In a small randomized trial of the efficacy of an iron-containing dietary supplement on fertility, 30 women who had not been able to become pregnant after at least 6 months of attempts were randomly assigned to receive the supplement or placebo for 3 months.⁴ At the end of this period, four women in the supplement group and none in the control group had achieved a pregnancy. Resumption of fertility has also been incidentally

observed among women with low body iron stores receiving iron supplements for unrelated conditions.²¹ Additional evidence comes from studies that have documented a greater risk of infertility among women with subclinical celiac disease. In two studies conducted in Europe, undiagnosed celiac disease was found more frequently among women with unexplained infertility than among fertile controls.^{6,7} Moreover, some of these infertile women had signs of iron deficiency, including iron deficiency anemia⁷ and low ferritin levels, without evidence of other nutrient deficiencies.⁶ It is important to keep in mind that the main infertility diagnosis in celiac disease is not ovulatory disorder but rather idiopathic infertility, and not all women in the trial had an initial diagnosis of ovulatory infertility. However, it is possible that iron may be important for other forms of infertility and that subclinical ovulatory disorders may be the underlying cause of some infertility cases where no diagnosis can be made.

Iron deficiency is the most prevalent nutritional deficiency worldwide. In the United States, women of child bearing age are at increased risk of this condition.²² A high prevalence of depleted iron stores (serum ferritin less than 12 ng/mL) has been documented in national surveys among young women (21%).²³ In addition, iron supplements have been associated with a lower prevalence of iron deficiency among women of reproductive age.²⁴ The role iron status may have on reproduction is further highlighted by studies regarding iron-transporting proteins in key ovarian cells. The presence of transferrin and its receptor in granulosa cells and oocytes has been documented in several studies.^{25–27} More recently, it

Table 4. Multivariable-Adjusted* Relative Risks and 95% Confidence Intervals by Tertiles of Nonheme Iron Intake Stratified by Heme Iron Intake

	Tertiles of Nonheme Iron Intake (mg/d)			P , Trend
	1 (≤ 13)	2 (13.01–32.59)	3 (≥ 32.6)	
Halves of heme-iron intake				
1 (1 mg/d or less)	1.00 (ref)	0.92 (0.65–1.29)	0.42 (0.26–0.66)	$<.001$
2 (more than 1 mg/d)	0.92 (0.65–1.30)	0.93 (0.65–1.33)	0.74 (0.48–1.14)	.22
P , interaction			0.78	

Data are relative risk (95% confidence interval).

* Adjusted for age (continuous), calendar time (4 2-year intervals), total energy intake (continuous), body mass index (less than 20, 20–24.9, 25–29.9, 30 or more, and missing), parity (0, 1, 2 or more, and missing), smoking history (never, past 1–4 cigarettes per day, past 5–14 cigarettes per day, past 15–24 cigarettes per day, past 25 cigarettes per day or more or unknown amount, current 1–4 cigarettes per day, current 5–14 cigarettes per day, current 15–24 cigarettes per day and current 25 cigarettes per day or more, or unknown amount), physical activity (less than 3 MET-h/wk, 3–8.9 MET-h/wk, 9–17.9 MET-h/wk, 18–26.9 MET-h/wk, 27–41.9 MET-h/wk, 42 MET-h/wk or more, and missing), oral contraceptive use (current user, never user, past user 0–23 months ago, past user 24–47 months ago, past user 48–71 months ago, past user 72–95 months ago, past user 96–119 months ago, past user 120 months ago or more, and missing), intake of alcohol (no intake, less than 2 g/d, 2–4.9 g/d, 5 g/d or more), coffee (less than 1 serving per month, 1 serving per month, 2–6 servings per week, 1 serving per day, 2–3 servings per day, 4 servings per day or more), and intakes of vitamin C, retinol, and α -carotene, major types of fatty acids and protein.



has been reported that granulosa cells can synthesize transferrin, which may be translocated to the oocytes.²⁷ Although it is possible that transferrin and transferrin receptor are redundant in the ovary or do not play an important role in local iron metabolism, it has been suggested that these proteins are essential for ovum development and are required to support the increased iron demand of the developing follicle.²⁶ Our results are in agreement with the latter hypothesis.

Limitations of our study include that it was not a cohort of women known to be planning a pregnancy. Cases, who were clearly attempting to conceive, may have been more health-conscious than the pregnancy noncases, who may have conceived accidentally. However, this would lead to results wrongly suggesting a positive association of iron intake with risk of infertility, rather than the inverse associations we observed for iron supplements, total iron intake, and nonheme iron intake. Moreover, we simulated a cohort of pregnancy planners in our study by including only married women (whose pregnancies are more likely to be intentional than those of unmarried women²⁸) and by including in the noncase group women who were diagnosed with infertility from other causes. This makes it less likely that pregnancy intention would affect our results.

In summary, we found that use of iron supplements with high iron content was associated with a reduced risk of ovulatory infertility. We also observed an inverse association between nonheme iron intake and risk of ovulatory infertility, due primarily to the use of supplements containing iron. In addition, we found intake of heme iron to be unrelated to ovulatory infertility at intake levels equivalent to consuming one serving of meat per day. Because these results have not been reported previously it is important that they are reproduced, preferably in prospective studies using diverse measures of iron intake and in randomized trials among healthy and infertile women. Nevertheless, given that iron deficiency is commonly found among women of reproductive age and may be associated with adverse pregnancy and perinatal outcomes,^{29,30} women planning to become pregnant should consider using iron supplements because they may help them prevent iron deficiency and also improve fertility.

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