

Predictors of iron levels in 14,737 Danish blood donors: results from the Danish Blood Donor Study

Andreas Stribolt Rigas,¹ Cecilie Juul Sørensen,² Ole Birger Pedersen,³ Mikkel Steen Petersen,² Lise Wegner Thørner,¹ Sebastian Kotzé,² Erik Sørensen,¹ Karin Magnussen,¹ Klaus Rostgaard,⁴ Christian Erikstrup,² and Henrik Ullum¹

BACKGROUND: Dietary studies show a relationship between the intake of iron enhancers and inhibitors and iron stores in the general population. However, the impact of dietary factors on the iron stores of blood donors, whose iron status is affected by blood donations, is incompletely understood.

STUDY DESIGN AND METHODS: In the Danish Blood Donor Study, we assessed the effect of blood donation frequency, physiologic factors, lifestyle and supplemental factors, and dietary factors on ferritin levels. We used multiple linear and logistic regression analyses stratified by sex and menopausal status.

RESULTS: Among high-frequency donors (more than nine donations in the past 3 years), we found iron deficiency (ferritin below 15 ng/mL) in 9, 39, and 22% of men, premenopausal women, and postmenopausal women, respectively. The strongest predictors of iron deficiency were sex, menopausal status, the number of blood donations in a 3-year period, and the time since last donation. Other significant factors included weight, age, intensity of menstruation, iron tablets, vitamin pills, and consumption of meat and wine.

CONCLUSION: The study confirms iron deficiency as an important problem, especially among menstruating women donating frequently. The risk of iron depletion was largely explained by sex, menopausal status, and donation frequency. Other factors, including dietary and supplemental iron intake, had a much weaker effect on the risk of iron depletion.

Lack of iron could affect as many as 5 billion people worldwide.¹ Blood donors are at increased risk of developing iron deficiency because of losses through repeated blood donations.^{2,3} Indeed, the strongest predictor of having iron deficiency in blood donors is the number of blood donations.³ Iron is needed for several physiologic functions, such as cellular respiration, electron transport, and gene regulation.⁴ In addition to anemia, iron deficiency may cause neuropsychologic changes,⁵ restless leg syndrome,⁶ hair loss,⁷ and impaired innate and specific immunity.^{8,9}

Different periods in life demand higher iron absorption because of increased requirements; for example, the

ABBREVIATIONS: AIS = absent iron stores; IDE = iron-deficient erythropoiesis; IUD(s) = intrauterine device(s).

From the ¹Department of Clinical Immunology, Copenhagen University Hospital, Copenhagen, Denmark; the ²Department of Clinical Immunology, Aarhus University Hospital, Aarhus, Denmark; the ³Department of Clinical Immunology, Naestved Hospital, Naestved, Denmark; and the ⁴Department of Epidemiology, State Serum Institute, Copenhagen, Denmark.

Address reprint requests to: Henrik Ullum, Department of Clinical Immunology 2034, Copenhagen University Hospital, Blegdamsvej 9, 2100 Copenhagen, Denmark; e-mail: Henrik.ullum@regionh.dk.

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adolescent growth spurt, pregnancy, lactation periods,^{10,11} or physiologic blood loss due to menstruation.^{11,12} The intensity of menstruation is influenced by contraception. Hormonal contraception suppresses menstruation bleeding, whereas intrauterine devices (IUDs) often increase uterine bleeding. Consequently, the use of oral contraception is positively associated with ferritin levels, whereas the use of IUDs is negatively associated with ferritin levels.¹²

Many factors affect the absorption of heme and nonheme iron.¹³ Nevertheless, the extent to which these factors influence iron stores (estimated by ferritin levels) in blood donors is unknown. We chose to study these effects among blood donors, whose iron homeostasis is especially challenged.

Heme and nonheme iron is absorbed through different physiologic pathways. Nonheme iron is transported through the apical mucosal barrier in the proximal small intestine by a divalent metal transporter (DMT1), whereas the transport of heme iron is facilitated by heme carrier protein 1.¹⁴ Heme iron is found in meat, fish, and poultry. Nonheme iron occurs in both vegetables and animal products. The iron in meat, fish, and poultry consists of approximately 40% heme iron and 60% nonheme iron. Consequently, nonheme iron contributes most to the total intake of dietary iron, but heme iron is usually better absorbed.¹³

Several factors inhibit or enhance iron absorption. Among enhancing factors are ascorbic acid and a "meat factor."^{13,15} The meat factor, which enhances both nonheme iron and heme iron absorption,^{13,15,16} has not been identified. Possibly, cysteine-containing peptides are responsible for the enhancing effect in meat.^{17,18} Calcium is, besides inhibiting nonheme iron absorption, the only known factor to inhibit absorption of heme iron.¹⁶ Nonheme iron absorption inhibitors are eggs, milk (dairy products), tea, and coffee.^{13,19} Furthermore, alcohol intake is associated with higher iron stores and a reduced risk of iron deficiency.²⁰

Ferritin levels are also influenced by genetic mutations²¹ in iron metabolism or absorption (such as hereditary hemochromatosis) and general iron status (replete vs. deficient).¹⁹ In addition, iron supplements significantly increase iron stores and improve iron status when compared with control or placebo.²² The present epidemiologic blood donor study examines the extent to which dietary factors and blood donation history determine the current iron status in blood donors.

MATERIALS AND METHODS

The Danish Blood Donor Study²³ is an ongoing epidemiologic cohort study comprising more than 80,000 healthy blood donors aged 18 to 67 years recruited prospectively from the Danish blood banks since 2010. All blood donors attending the blood banks for their second or later blood

donation were invited to participate in the study if the staff had time to inform them about the study. At inclusion, all blood donors complete a four-page questionnaire. Plasma ferritin and blood hemoglobin (Hb) levels were determined in 18,070 participants enrolled from March 1, 2010, to December 31, 2010. All blood donors older than 17 years were eligible for inclusion. Less than 5% of invited donors choose not to participate.²³

The questionnaire addressed the average weekly dietary intake of meat, fish, and egg stated as number of meals. Questions regarding milk, coffee, tea, wine, beer, liquor, and intake of vitamins were assessed on a four- or five-item ordinal scale. Vitamin intake was dichotomized to "rarely" or "frequently" with "frequently" defined as at least weekly consumption of vitamin pills. Participants were also asked whether they received iron tablets at their last blood donation. All participants reported anthropometric measures and smoking status. Female participants provided information about age of menarche, menopause ("have your menstruations stopped"), duration of menstruation, contraceptive medication, and pregnancies (nullipara, primipara/multipara).

The Scandinavian Donation and Transfusion Database (SCANDAT)²⁴ is a binational (Denmark and Sweden) hemovigilance system for monitoring blood donors and transfusion recipients. Historical data on blood donation were extracted from the database and combined with the questionnaire data.

Statistical analysis

Statistical analysis was performed using computer software (Stata/IC 11.2 for Macintosh, StataCorp, College Station, TX). Characteristics were described by means (\pm standard deviation [SD]) for normally distributed data, medians (25th, 75th percentiles) for nonnormally distributed data, and frequency statistics. Ferritin levels were log-transformed to approximate normal distribution. Premenopausal women, postmenopausal women, and men were compared by two-sample t test for normally distributed data, Mann-Whitney U test for nonnormally distributed data, and chi-square test for binary data.

All analyses were stratified for sex and menopausal status. Predictors of ferritin levels were assessed by multivariable linear regression analysis and results presented as regression coefficients and beta-coefficients. Beta-coefficients depict the change in the outcome variable in SDs with a SD change in the explanatory variables. Multivariable logistic regression analysis was used to identify risk factors for iron deficiency. Iron deficiency was defined as ferritin below 15 ng/mL.²⁵ The annual donation frequency was calculated from the average number of donations during the 3 years preceding inclusion. The variable menstruation (duration of menstruation) was dichotomized with the cutoff at more than 4 days on average per period.

TABLE 1. Characteristics of the study population

Variables	Men (1)	Premenopausal women (2)	Postmenopausal women (3)	p values		
				(1) vs. (2)	(1) vs. (3)	(2) vs. (3)
Blood donation factors						
Number of donations (3 years)*	6.4 (2.8)	5.2 (2.5)	6.9 (2.7)	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻⁴
Days since last donation†	141 (105, 223)	158 (112, 266)	145 (105, 231)	<10 ⁻⁴	0.19	<10 ⁻⁴
Physiologic factors						
Age (years)*	40 (12)	34 (9)	54 (8)	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻⁴
BMI*	25.7 (3.7)	24.4 (4.2)	25.1 (4.1)	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻⁴
Ferritin level (ng/mL)†	50 (32, 77)	23 (15, 34)	31 (22, 46)	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻⁴
Hb level (mmol/L)*	9.4 (0.6)	8.3 (0.6)	8.6 (0.6)	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻⁴
Giving birth (%)		46%	84%			<10 ⁻³
Days of menstruation†		5 (4, 6)				
Lifestyle or supplemental factors						
Smoking (yes)	15.6%	16.1%	20.2%	0.53	<10 ⁻³	<10 ⁻³
Vitamin pills (yes)	30.9%	33.1%	39.9%	<10 ⁻²	<10 ⁻³	<10 ⁻³
Iron tablets (yes)	3.5%	36.2%	19.6%	<10 ⁻³	<10 ⁻³	<10 ⁻³
Dietary factors						
Meat per week†	6 (5, 7)	6 (4, 7)	5 (4, 6)	<10 ⁻³	<10 ⁻³	<10 ⁻³
Fish per week†	1 (1, 2)	1 (1, 2)	1 (1, 2)	<10 ⁻³	<10 ⁻⁴	<10 ⁻⁴
Eggs per week†	2 (1, 3)	2 (1, 3)	2 (1, 3)	<10 ⁻⁴	0.9	<10 ⁻⁴
Milk (1, 2, 3, 4)‡	15, 8, 14, 63	18, 8, 14, 59	32, 6, 12, 50	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻⁴
Tea (1, 2, 3, 4)‡	45, 21, 16, 17	23, 24, 25, 29	27, 15, 17, 42	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻³
Coffee (1, 2, 3, 4)‡	18, 3, 8, 71	30, 5, 10, 54	11, 2, 5, 82	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻⁴
Beer (1, 2, 3, 4)‡	11, 40, 43, 6	39, 46, 14, 0.4	43, 36, 18, 2	<10 ⁻⁴	<10 ⁻⁴	0.17
Wine (1, 2, 3, 4)‡	20, 44, 31, 5	23, 52, 23, 2	12, 34, 41, 13	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻⁴
Liquor (1, 2, 3, 4)‡	51, 41, 7, 0.4	56, 41, 3, 0.1	72, 23, 5, 0.5	<10 ⁻⁴	<10 ⁻⁴	<10 ⁻⁴

* Mean (SD). [Correction added on 5 March 2014. Hb level units have been changed from g/dL]

† Median (25th, 75th percentile).

‡ Shown as percentage of the frequency distribution of the four categories: 1 (“never, almost never”), 2 (“a few times per month”), 3 (“a few times per week”), 4 (“every day, almost every day”).

RESULTS

A total of 25,877 blood donors were included in the study between March 1, 2010, and December 31, 2010. Plasma ferritin and Hb were measured in 18,070 blood donors. We excluded 3054 participants from analyses because of missing values in at least one of the 19 investigated factors, and an additional 279 participants were excluded from analyses because of extreme physiologic outliers. Complete, relevant questionnaire data were thus available from 14,737 donors, comprising 7922 men, 5403 premenopausal women, and 1412 postmenopausal women. Basic characteristics of the included donors are presented in Table 1.

Compared with women, men had higher ferritin and Hb levels, consumed more meat, and drank more milk and beer. Postmenopausal women had the highest proportion of tobacco users. Also, postmenopausal women were the most frequent consumers of tea, coffee, and wine. Compared with men and premenopausal women, postmenopausal women were significantly more likely to use supplemental vitamin pills. The proportion of participants who received iron tablets was significantly higher among premenopausal women compared with the two other groups. Indeed, approximately one in three premenopausal women in our study received iron tablets.

A linear regression analysis (Table 2) was carried out to identify factors associated with iron storage (as measured by plasma ferritin) and stratified by sex and menopausal status. A logistic regression analysis (Table 3) on factors predicting iron deficiency was performed and likewise stratified by sex and menopausal status. Both models were, for the sake of clarity, divided into blood donation factors, physiologic factors, lifestyle or supplemental factors, and dietary factors.

Blood donation factors

For all groups, the time since last donation and the number of blood donations within the past 3 years correlated positively and negatively, respectively, with iron store levels for all groups (Table 2). With each additional blood donation per year (in a 3-year period), the iron stores were, on average, reduced by 27.9% (95% confidence interval [CI], 29.0%-26.8%), 12.0% (95% CI, 13.8%-10.1%), and 18.4% (95% CI, 21.2%-15.4%) for men, premenopausal women, and postmenopausal women, respectively. The blood donation factors were also the most important predictors of iron levels assessed by the standardized beta-coefficients. Increasing number of blood donations increased the risk of iron deficiency, whereas prolonging the time since last donation protected against development of iron deficiency (Table 3). Thus,

TABLE 2. Multiple linear regression analyses*

Outcome: ln(ferritin)	Men (n = 7922)			Premenopausal women (n = 5403)			Postmenopausal women (n = 1412)		
	Coefficient	p value	Beta	Coefficient	p value	Beta	Coefficient	p value	Beta
Blood donation factors									
Time since last donation (years)	0.09	<10 ⁻³	0.10	0.10	<10 ⁻³	0.14	0.14	<10 ⁻³	0.15
Blood donations (3 years)	-0.11	<10 ⁻³	-0.46	-0.04	<10 ⁻³	-0.17	-0.07	<10 ⁻³	-0.31
Physiologic factors									
Weight (5 kg)	0.06	<10 ⁻³	0.22	0.02	<10 ⁻³	0.09	0.01	0.028	0.06
Age (10 years)	0.05	<10 ⁻³	0.09	0.07	<10 ⁻³	0.11	0.07	0.001	0.09
Height (cm)	-0.01	<10 ⁻³	-0.07	0.00	0.647	-0.01	0.00	0.301	0.03
Giving birth				-0.08	<10 ⁻³	-0.07	-0.06	0.113	-0.04
Menstruation				-0.01	0.001	-0.04			
Lifestyle or supplemental factors									
Smoker	0.00	0.910	0.00	0.03	0.156	0.02	0.06	0.081	0.04
Iron tablets	-0.16	<10 ⁻³	-0.05	-0.05	0.001	-0.04	-0.14	<10 ⁻³	-0.09
Vitamin pills	0.06	<10 ⁻³	0.04	0.13	<10 ⁻³	0.10	0.13	<10 ⁻³	0.11
Dietary factors									
Meat	0.01	<10 ⁻³	0.04	0.01	<10 ⁻³	0.05	0.01	0.015	0.06
Fish	-0.01	0.004	-0.03	0.00	0.766	0.00	0.00	0.459	0.02
Egg	0.00	0.266	0.01	0.00	0.373	0.01	0.00	0.670	0.01
Milk	0.00	0.506	-0.01	-0.01	0.077	-0.02	-0.01	0.264	-0.03
Tea	-0.03	<10 ⁻³	-0.05	-0.01	0.080	-0.02	-0.03	0.017	-0.06
Coffee	-0.01	0.277	-0.01	0.00	0.623	0.01	-0.05	0.003	-0.07
Beer	0.04	<10 ⁻³	0.04	0.02	0.142	0.02	0.07	<10 ⁻³	0.10
Wine	0.08	<10 ⁻³	0.09	0.06	<10 ⁻³	0.07	0.10	<10 ⁻³	0.14
Liquor	0.01	0.466	0.01	0.02	0.300	0.02	0.00	0.992	0.00

* The factors time since last donation (in years), blood donations (per 3 years), weight (divided by 5), age (divided by 10), height, menstruation (days), meat, fish, and egg were all entered as continuous data. The factors giving birth, smoker, iron tablets, and vitamin pills were all entered as dichotomous data. The factors milk, tea, coffee, beer, wine, and liquor were all entered as ordinal data.

TABLE 3. Logistic regression analyses*

Outcome: iron deficiency (ferritin < 15 ng/mL)	Men (n = 7922)			Premenopausal women (n = 5403)			Postmenopausal women (n = 1412)		
	OR	p value	95% CI	OR	p value	95% CI	OR	p value	95% CI
Blood donation factors									
Time since last donation (per year)	0.02	<10 ⁻³	0.01-0.07	0.62	<10 ⁻³	0.53-0.72	0.24	0.004	0.09-0.63
Blood donations per year	2.25	<10 ⁻³	1.89-2.68	1.45	<10 ⁻³	1.33-1.59	1.88	<10 ⁻³	1.45-2.43
Physiologic factors									
Hb	0.41	<10 ⁻³	0.32-0.52	0.41	<10 ⁻³	0.36-0.47	0.49	<10 ⁻³	0.34-0.70
Age (10 years)	0.93	0.264	0.81-1.06	0.84	0.004	0.75-0.95	0.83	0.152	0.65-1.07
Weight (5 kg)	0.83	<10 ⁻³	0.78-0.89	0.94	<10 ⁻³	0.91-0.97	0.98	0.552	0.90-1.06
Height	1.01	0.394	0.99-1.03	1.00	0.886	0.99-1.01	0.98	0.337	0.95-1.02
Giving birth				1.37	0.001	1.13-1.66	1.05	0.850	0.63-1.74
Menstruation (≥5 days)				1.56	<10 ⁻³	1.36-1.80			
Lifestyle or supplemental factors									
Smoker	0.93	0.73	0.63-1.38	0.99	0.955	0.83-1.20	0.56	0.044	0.32-0.98
Iron tablets	0.97	0.907	0.58-1.63	1.17	0.02	1.03-1.34	1.66	0.012	1.12-2.45
Vitamin pills	0.47	<10 ⁻³	0.34-0.65	0.70	<10 ⁻³	0.60-0.80	0.48	<10 ⁻³	0.32-0.72
Dietary factors									
Meat	0.92	0.003	0.88-0.97	0.96	0.004	0.94-0.99	0.93	0.154	0.85-1.03
Fish	1.09	0.066	0.99-1.19	0.99	0.559	0.94-1.04	0.98	0.687	0.88-1.09
Egg	1.02	0.235	0.99-1.06	1.00	0.836	0.97-1.03	1.05	0.376	0.94-1.17
Milk	1.01	0.863	0.90-1.14	1.02	0.43	0.97-1.08	1.07	0.307	0.94-1.23
Tea	1.23	<10 ⁻³	1.09-1.37	1.04	0.175	0.98-1.10	1.14	0.109	0.97-1.33
Coffee	0.94	0.314	0.84-1.06	0.98	0.419	0.93-1.03	1.27	0.025	1.03-1.57
Beer	0.90	0.251	0.75-1.08	0.88	0.022	0.79-0.98	0.90	0.406	0.70-1.16
Wine	0.74	0.002	0.62-0.90	0.85	0.002	0.77-0.94	0.62	<10 ⁻³	0.50-0.79
Liquor	0.98	0.829	0.78-1.22	0.97	0.62	0.85-1.10	0.78	0.206	0.53-1.15

* The factors time since last donation (in years), blood donations (per year over a 3-year period), weight (divided by 5), age (divided by 10), height, meat, fish, and egg were all entered as continuous data. The factors giving birth, smoker, iron tablets, vitamin pills, and menstruation (more than 4 days) were all entered as dichotomous data. The factors milk, tea, coffee, beer, wine, and liquor were all entered as ordinal data.

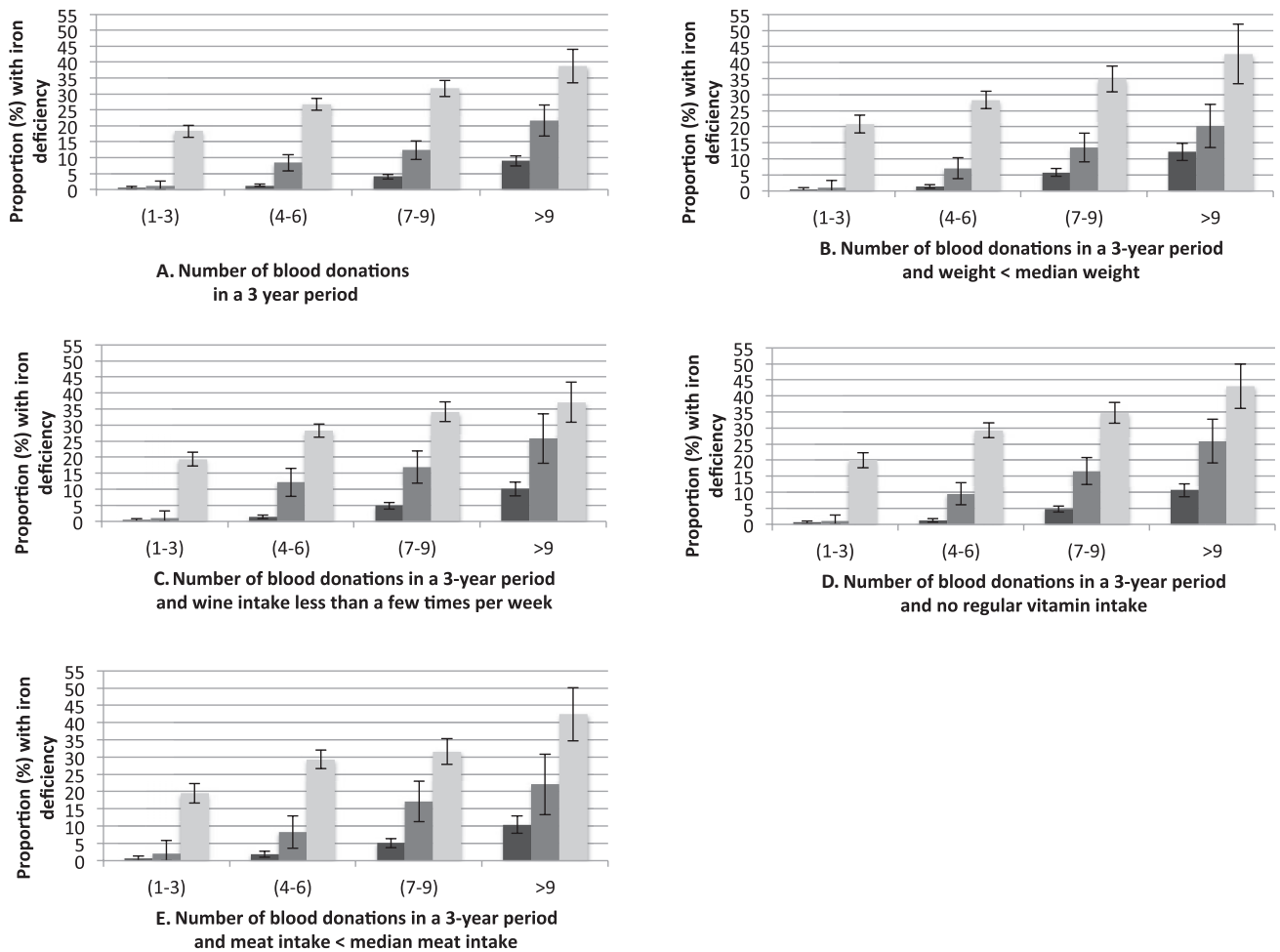


Fig. 1. Proportion (%) of men (■), postmenopausal women (▒), and premenopausal women (░) with iron deficiency (ferritin < 15 ng/mL) when (A) stratified by donation frequency and further selected by (B) weight below median weight, (C) wine intake less than a few times per week, (D) no regular vitamin intake, or (E) a meat intake below median meat intake. All analyses were performed separately for men, premenopausal women, and postmenopausal women. [Correction made after online publication on 1 February 2014. Colors for pre- and post-menopausal women have been switched]

the odds ratio (OR) of having iron deficiency was 2.25 (95% CI, 1.89-2.68), 1.45 (95% CI, 1.33-1.59), and 1.88 (95% CI, 1.45-2.43) per additional blood donation per year in a 3-year period for men, premenopausal women, and postmenopausal women, respectively (Table 3). If bled thrice or less in a 3-year period roughly 0.5, 18.5, and 1.1% of men, premenopausal women, and postmenopausal women were iron deficient, respectively (Fig. 1A). In comparison, if bled nine times or more in a 3-year period 9.0, 38.7, and 21.7% of men, premenopausal women, and postmenopausal women had iron deficiency, respectively (Fig. 1A).

Physiologic factors

Weight and age were the only significant predictors of iron store levels for all three groups (Table 2). Weight and age

correlated positively with iron stores (Table 2). However, only for premenopausal women were both factors associated with a slightly reduced risk of iron deficiency (Table 3). Although positively correlated with ferritin levels in univariate analysis, increased height predicted lower ferritin levels when adjusted for weight among men (Table 2). Giving birth and the duration of menstruation measured as number of days correlated negatively with iron stores (Table 2) and were associated with an increased risk of iron deficiency (Table 3) in premenopausal women. As a predictor of low iron stores, the Hb level was added to the logistic regression model. Unsurprisingly, a high Hb level predicted a low risk of iron deficiency for all groups (Table 3). When considering standardized beta coefficients physiologic factors influenced iron stores to a lesser degree compared with blood donation factors.

Lifestyle or supplemental factors

Supplemental vitamin intake correlated positively with iron stores and was associated with a reduced risk of iron deficiency for all groups (Table 2 and Table 3). Supplemental iron tablets were negatively associated with iron stores for all groups (Table 2). Tabulating iron deficiency and iron tablets revealed that iron-deficient participants were more likely to receive iron tablets.

In a subanalysis, IUD and combined oral contraception were added to the model for premenopausal women, comprising 3690 participants. This model revealed that using IUD correlated negatively with iron stores and was associated with an increased risk of iron deficiency whereas intake of combined oral contraception had no effect. Furthermore, smoking status did not affect iron levels (Table 2) but was marginally significant when predicting iron deficiency for postmenopausal women (Table 3).

Dietary factors

Meat intake was positively associated with iron stores for all three groups, whereas consumption of fish, egg, and milk had no effect on iron stores, except for fish intake in men (Table 2). Meat consumption decreased the risk of iron deficiency to a minor extent for men and premenopausal women (Table 3). A frequent wine intake correlated with high iron stores in all groups, whereas liquor had no effect and beer only an effect in men and postmenopausal women (Table 2). However, beer intake was associated with a slightly reduced risk of iron deficiency in premenopausal women (Table 3). Wine intake also reduced the risk of iron deficiency in all groups (Table 3).

Finally, there were very few donors ($n = 130$) who did not eat meat at all. The vegetarian group had a median level of ferritin of 18.25 ng/mL (25th, 75th percentile, 13.50, 27.50 ng/mL) and 33.1% (95% CI, 24.9%-41.3%) had iron deficiency.

When depicting ferritin levels stratified according to donation frequency, a steady decrease in median ferritin levels for all groups was observed (Fig. 2). In accordance with the observed low ferritin levels for high-frequency donors, a substantial proportion of donors were iron deficient. Roughly one in 10, one in five, and one in three of men, postmenopausal women, and premenopausal women, respectively, had iron deficiency if bled more than nine times in a 3-year period (Fig. 1).

Interestingly, there were almost no iron-deficient men unless they had been bled at least seven times in a 3-year period. Likewise, we found very few iron-deficient postmenopausal women unless bled more than thrice in a 3-year period (Fig. 1). Premenopausal women were more prone to develop iron deficiency, and a substantial proportion of low-frequency premenopausal donors were iron deficient (Fig. 1). In fact, 9.6% (95% CI, 4.1%-15.0%)

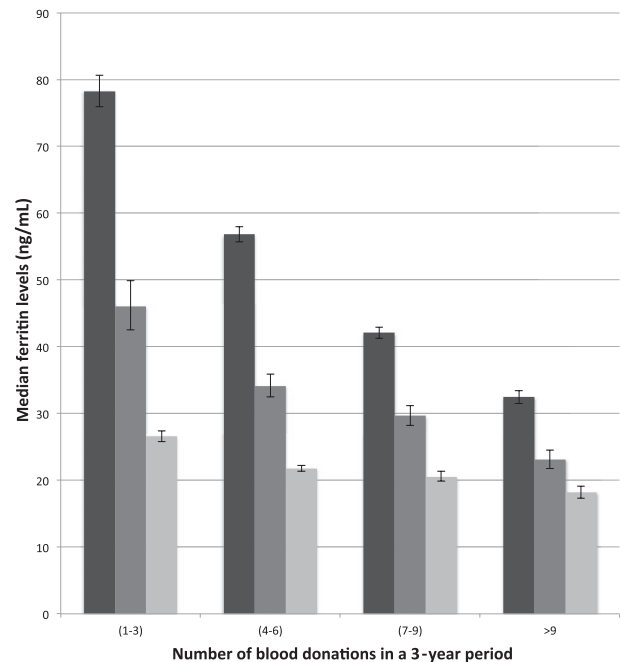


Fig. 2. Median ferritin levels (ng/mL) for men (■), postmenopausal women (■), and premenopausal women (■) when stratified by donation frequency 3 years before inclusion. [Correction made after online publication on 1 February 2014. Colors for pre- and post-menopausal women have been switched]

re-evoked premenopausal female donors ($n = 115$) with a 3-year pause from donation had iron deficiency.

To compare the effect of other factors, we depicted some of them as evaluated by regression analysis. The impact of the donation frequency remained substantial, even when participants were further selected by a weight below the median, no regular vitamin pill consumption, meat intake below median meat intake, or consumption of wine less than a few times per week (Fig. 1). Similar results were obtained by selecting for other physiologic, supplemental, or dietary factors (data not shown).

Despite a modest effect of many of the single physiologic, supplemental, or dietary factors, their combined effect could be important. For example, a large risk of iron deficiency was found for menstruating women younger than 35 years with a weight below 67 kg who donated more than three times during a 3-year period, had an average meat consumption of less than six times per week, and who did not take vitamin pills regularly. The prevalence of iron deficiency in this group was 36.2% (95% CI, 31.2%-41.1%) and the group constituted approximately 7% of premenopausal women in the study.

DISCUSSION

Blood donors have an increased risk of developing iron deficiency.^{2,3} Even among blood donors who meet the Hb

criteria for donating blood, iron deficiency is prevalent.²⁶ The present, large epidemiologic study on the influence of blood donation factors, physiologic factors, lifestyle and supplemental factors, and dietary factors on iron store levels confirmed that premenopausal women were more prone to develop iron deficiency compared with postmenopausal women and men. In fact, among high frequency donors with more than nine donations in a 3-year period, a substantial proportion of donors and especially female donors were iron deficient.

Fairweather-Tait¹⁹ describes a number of dietary enhancers (meat, fish, poultry, ascorbic acid, alcohol) and dietary inhibitors (milk, egg, calcium, tea, coffee) of iron absorption. However, only a few large-scale population studies investigate whether these iron absorption enhancers and inhibitors affect iron store levels.^{27,28}

These studies, alongside others on factors influencing iron stores, report that heme iron, supplemental iron, total iron intake, dietary vitamin C, alcohol, age, meat, and fish have a positive impact on iron stores. These studies also find that the intake of coffee, tea, and dairy products correlate negatively with iron levels and that the duration of menstruation and giving birth for premenopausal women result in additional loss of iron.^{11,20,27-29} A recent study by Cable and colleagues³ investigated factors that predict absent iron stores (AIS) and iron-deficient erythropoiesis (IDE) in blood donors. They defined AIS as a ferritin level below 12 ng/mL and IDE as present if the log of the ratio of soluble transferrin receptor to ferritin was greater than or equal to 2.07. Cable and colleagues³ find that donation frequency is the strongest predictor for iron stores. Furthermore, they report that sex, weight, iron supplements, dairy products, and age for women are significantly associated with AIS. They also report that age in men, smoking, menstruation status, giving birth, and beef are associated with IDE. Furthermore, smoking and time since last donation are associated with a lower risk of AIS.³⁰

In accordance with these studies, we found that intake of wine and meat correlated positively with iron stores for all groups. Only a few donors did not eat meat at all and among the few vegetarians one-third had iron deficiency. This suggests that vegetarians have problems maintaining the required Hb levels for repeated blood donations. The current study also showed that weight, age, and supplemental vitamin pills were positively associated with iron stores for all groups. Conversely, we found no consistent effect of coffee or tea on iron stores, and intake of milk and eggs were not associated with iron levels. The association between consumption of fish and iron stores was only significant for men and correlated negatively with iron stores. In addition, the intensity of menstruation in premenopausal women also correlated negatively with iron stores. As previously shown, giving birth affected iron stores in premenopausal women, but a history of previous

birth(s) was not associated with iron levels in postmenopausal female donors.

Only the number of blood donations and the intake of iron tablets were negatively associated with iron stores in all groups. However, the study was not designed to measure the effect of iron supplementation, since iron routinely was given to donors considered at risk for iron deficiency. Iron tablets were therefore probably negatively associated with iron stores because of confounding by indication. We found no effect of smoking on iron stores, but smoking was marginally significant for prediction of iron deficiency in postmenopausal women.

In general, the blood donation frequency, the time since last donation, sex, and menopausal status were the most important predictors of current iron status, whereas physiologic, lifestyle or supplemental factors, and dietary factors were associated with iron stores but were less influential.

Several factors can predict the development of iron deficiency. The number of blood donations significantly increased the risk of iron deficiency. Among high-frequency donors with more than nine blood donations in the preceding 3 years, a substantial proportion of donors were iron deficient, even when the "protective" effect of certain physiologic, supplemental, and dietary factors were accounted for.

In general, blood donors became more prone to iron deficiency and experienced a steady decrease in ferritin levels with increasing donation frequency regardless of dietary intake. The eligibility criteria for blood donors in Denmark ensure that the study population in general was a healthy group with few cases of chronic disease or sickness at time of donation.

The internal validity of this study was strengthened by the large number of participants and the minimal nonresponder rate.²³ Furthermore, the questionnaire included the most relevant factors influencing iron stores. Because blood donors differ from the general population as their iron stores are stressed from repeated blood donations, the findings do not necessarily apply to other groups.

Our study confirms iron deficiency to be highly prevalent among blood donors. High-frequency donors are particularly exposed to iron deficiency and decreasing ferritin levels regardless of dietary intake. Iron stores in blood donors are affected by many factors previously found to either enhance or inhibit iron absorption, by physiologic factors, and by supplements such as iron tablets and vitamin pills.

Although the strongest predictor of iron stores in blood donors was the number of donations in the past 3 years, both sex and menopausal status were also strong predictors. The physiologic factors (weight, age, and duration of menstruation), the lifestyle or supplemental factors (iron tablets, vitamin pills, and IUD), and the dietary factors (meat and wine) were all significant but

less useful to predict current iron status in blood donors. Therefore, the composition of dietary iron inhibitors and enhancers in the diet influenced iron stores to a lesser degree when compared with blood donation history.

The study does not answer whether sufficient iron stores can be maintained by guided iron supplementation or whether reduction of donation frequency may be necessary for donors at high risk of iron depletion.

CONFLICT OF INTEREST

The authors report no conflicts of interest or funding sources.

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