

Effect of Blood Donation on Iron Stores As Evaluated by Serum Ferritin

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Serum ferritin was measured in 2982 blood donors. First-time male donors had a geometric mean of 127 $\mu\text{g/liter}$ and female donors 46 $\mu\text{g/liter}$. While values were essentially constant in the women between the ages of 18 and 45, there was a rapid increase in the men between 18 and 30 years of age consistent with the establishment of iron stores during that time. Blood donation was associated with a decrease in serum ferritin. One unit per year, equivalent to an increased requirement of 0.65 mg/day, halved the serum ferritin level in the male. More frequent

donations were associated with further decreases. From the data obtained it would appear that male donors, while depleting their iron stores, were able to donate 2–3 U/yr without an appreciable incidence of iron deficiency. Women could donate only about half that amount, and more frequent donations were associated with a high incidence of iron deficiency and donor dropout. These data have provided information on the effect of graded amounts of iron loss through bleeding on iron balance.

BLEEDING results in mobilization of iron from body stores.¹ As stores decrease iron absorption increases.² With continued bleeding an individual either reaches equilibrium at a lower level of iron stores or becomes anemic.³ These considerations apply directly to people who serve as blood donors. The frequency of blood donation has been so adjusted as to prevent anemia in most donors, but quantitative information concerning the iron status of donors is limited. With the development of the ferritin assay, an evaluation of iron stores became possible;^{4,5} such an evaluation is the purpose of this study.

MATERIALS AND METHODS

Donors presenting themselves at two bleeding depots of the Puget Sound Blood Center gave permission for the use of their blood for ferritin analyses, and potential donors rejected because of anemia gave permission for more extensive hematologic measurements. Except for the presence of anemia in the rejected donor group, all individuals were healthy by history and fulfilled criteria for suitability as a blood donor. The cyanmethemoglobin method was used to screen the hemoglobin concentration of all blood donors and only those women and men whose hemoglobin concentrations were ≥ 12.5 and ≥ 13.5 g/100 ml respectively, were bled. The average blood donation exclusive of the volume of anticoagulant was 472 ± 11 (1 SD) ml of blood, and it was assumed in the calculations of iron loss that the average hemoglobin level in the male was 15 g/100 ml and in the female 13.5 g/100 ml of blood. Individuals were identified by age, sex, and by the number of blood donations over the past 4½ yr. There were 803 first-time male donors and 812 first-time female donors. In addition, there were 737 male and 630 female donors who had given 4–18 units over the 4½-yr period. The 2982 sera collected for ferritin determinations were fro-

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zen and analyzed within 12 mo by the method of Miles et al.⁶ Results of serum ferritin analyses were expressed as geometric means because of the skewed normal distribution.²

The blood of 51 rejected donors was also analyzed within a few days of their visit to the Blood Bank. Hemoglobin was measured by the cyanmethemoglobin method, plasma iron as outlined by the International Standardization Committee,⁷ unsaturated iron binding capacity using $MgCO_3$ as described by Cook,⁸ red cell protoporphyrin by the method of Labbe,⁹ and ferritin by the method of Miles et al.⁶ In evaluating the iron status of these rejected donors, a transferrin saturation below 16% was considered to represent iron deficiency, 16%–20% borderline, and >20% normal. Protoporphyrin:heme ratios below 20 were normal, between 20 and 30 borderline, and above 30 iron deficient. Ferritin values below 12 $\mu g/liter$ were considered iron deficient, 12–20 borderline, and above 20 normal. Hemoglobin values below 12.5 g/100 ml in the female and below 13.5 g/100 ml in the male were considered to represent anemia.¹⁰

RESULTS

Results of ferritin assays on new donors are shown in Fig. 1 and Table 1. The geometric mean value for serum ferritin determined on 812 first-time female donors was 46 $\mu g/liter$, and this value showed little change over an age range of 18–45. Values below 12 $\mu g/liter$ were found in 48 women, of whom 30 were younger than 30 yr old. By contrast, the mean ferritin of 803 male subjects showed an increase, starting at 66 $\mu g/liter$ at age 18, rising to 170 μg in the 30–35 age range, and climbing still higher to 196 in the 42–47 age range. No first-time male donor had a value below 12 $\mu g/liter$.

Ferritin values of 1367 repeat donors (737 men, 630 women) who had given from 4 to over 13 donations during the 4½-yr period were also determined (Table 1). These donors were divided into seven groups of about 100 subjects each based on the number of blood donations given over the previous 4½ yr. The estimated daily loss of iron due to phlebotomy in each group is shown. Mean ferritin values decreased with an increased frequency of blood donation, while the number of individuals with a ferritin value of <12 $\mu g/liter$ increased. The age difference between first-time blood donors and multiple donors averaged 6 yr in males and 8 yr in females, which was to be expected since the multiple donors had been giving blood a minimum of 5 yr. At higher donation

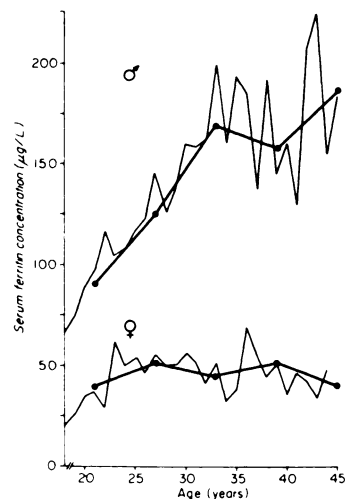


Fig. 1. Serum ferritin levels as a function of age in 803 male and 812 female first-time blood donors. The light line indicates mean values for subjects at each year and the heavy line represents a 6-yr average.

Table 1. Results of Ferritin Assays on Blood Donors

Number of Subjects	Blood Donations* (no/yr)	Estimated Iron Requirement† (mg/day)	Mean Donor Age (yr)	Mean Ferritin‡ (µg/liter)	Ferritin < 12 µg/liter (percent of subjects)
Males					
803	0	0.90	28 ± 7.0	127.3 (66.4, 244.1)	0
105	0.89	1.48	32 ± 7.7	65.9 (29.4, 147.5)	3.8
116	1.11	1.62	34 ± 9.1	70.0 (36.3, 134.9)	0.9
111	1.33	1.76	34 ± 8.0	66.8 (34.3, 129.8)	0.9
99	1.56	1.91	35 ± 9.5	56.3 (27.9, 113.4)	2.0
106	1.89	2.13	35 ± 9.2	54.9 (29.4, 102.4)	2.8
98	2.44	2.49	35 ± 9.5	42.9 (18.8, 97.8)	5.1
102	3.10	2.91	36 ± 9.2	31.3 (15.0, 65.5)	12.7
Females					
812	0	1.30	27 ± 7.0	46.0 (19.8, 107.1)	5.9
108	0.89	1.82	32 ± 8.5	33.2 (14.9, 74.0)	8.3
106	1.11	1.94	33 ± 9.9	26.2 (10.8, 63.6)	14.2
110	1.33	2.04	39 ± 10.9	40.1 (17.7, 90.6)	5.4
104	1.56	2.20	36 ± 11.5	26.9 (11.1, 65.2)	15.4
95	1.89	2.40	35 ± 10.2	24.3 (10.0, 59.2)	18.9
66	2.44	2.72	39 ± 13.5	25.0 (10.8, 58.2)	19.8
41	3.10	3.10	40 ± 12.2	21.6 (7.6, 61.2)	26.8

*Average number over 4.5 yr.

†Calculated from basal requirements of 0.9 mg/day and 1.3 mg/day plus allowance of 0.65 and 0.58 mg/day for each yearly blood donation for male and female subjects, respectively.

‡Geometric mean.

rates, there were fewer female donors, so that in the last two groups it was not possible within the time limits of the study to bring the number of female donors to 100.

Among the 51 donors rejected on the basis of a low hemoglobin, 44 were female and 7 were male. A repeat hemoglobin determination was normal in 2 of these, and in an additional 7 subjects iron deficiency was excluded on the basis of normal serum ferritin, transferrin saturation, and red cell protoporphyrin: heme values. In the remaining 44 subjects, unequivocal iron deficiency was found in 35, based on two (10 subjects) or all three (25 subjects) of the iron parameters being in the iron deficient range. The remaining 9 prospective donors had questionable iron deficiency on the basis of one abnormal iron parameter, one borderline value (6 subjects), or two borderline values (3 subjects).

DISCUSSION

Most of the blood in the Puget Sound Blood Center is donated through organized donor groups, and the lack of any blood replacement penalty eliminates financial coercion. Thus, the donor population in this study may be presumed to represent public-spirited citizens who give blood on a voluntary basis. The geometric mean ferritin level of these individuals who had not given blood before was higher than previously reported norms. For example, in a nutritional survey of another Northwest population, Cook et al.¹¹ found a mean level of 25 µg/liter in 370 women (to be compared with 46 µg/liter in the 812 women of this study) and a mean of 94 µg/liter in 240 men (as compared with 127 µg/liter

in the 803 men of this study). Perhaps the higher values we found reflected better iron nutrition due to better general economic status among this community-involved donor group.

The change in serum ferritin levels in women between 18 and 45 yr of age (Fig. 1) was minimal, consistent with the very limited iron stores in women of childbearing age. In men there was an increase in serum ferritin levels with age. This increase was particularly marked between the ages of 18 and 30, when it may be assumed that the larger iron stores in men have been established. These effects of sex and age on serum ferritin levels were similar to those reported elsewhere.^{11,12}

Significant additional iron requirements are imposed by blood donation. In our calculations it was assumed that basal losses are 0.9 mg/day in the adult male and 1.3 mg/day in the adult female.^{13,14} The average phlebotomy of 472 ml represented the removal of about 236 mg of iron from the male and 213 from the female. In individuals donating 1 unit of blood yearly, this increased the daily iron requirement by 0.65 mg/day in the male and 0.58 mg/day in the female. The relationship between iron requirements and ferritin concentration in people who donated multiple units over a period of 4½ yr is shown in Fig. 2. In men, when the basal requirement of 0.9 mg/day was increased to 1.5 mg/day by one blood donation per year, serum ferritin levels were halved. They were further reduced to about one-fourth basal when the daily requirements were increased to 2.9 mg/day by about three blood donations per year. None of the first-time male donors had ferritin levels below 12 µg/liter, but the frequency increased to 12.7% in men giving over 3 units yearly.

In female donors with a basal iron requirement of 1.3 mg/day, iron stores were approximately 30% lower than in men giving 1 unit of blood yearly. Iron requirements of the two groups were similar, and the difference probably reflected the higher iron intake in male subjects due to their higher caloric intake. The serum ferritin levels in women did not fall as much with multiple donations as did those in men. This difference may be misleading, however, because far fewer women than men donated blood more than twice a year. The greater sus-

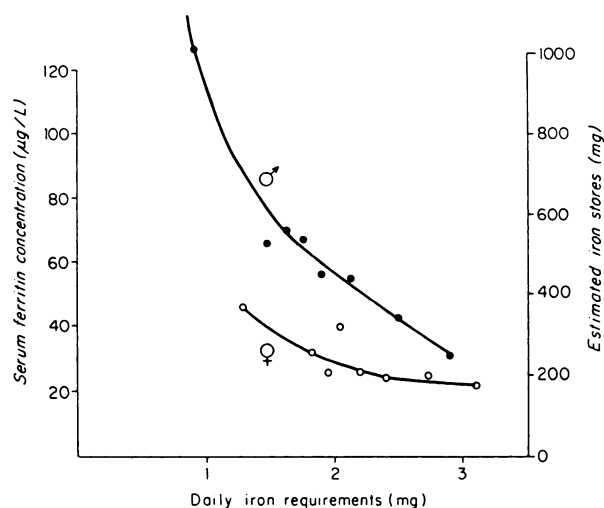


Fig. 2. Mean serum ferritin values of groups of subjects in Table 1 are plotted to indicate the relationship between estimated iron requirement and serum ferritin concentration. On the right hand vertical axis is indicated the estimated change in body iron stores corresponding to the change in serum ferritin.

ceptibility of females to depletion of iron stores with repeated blood donation was supported by the fact that 26.8% of women giving 3 or more units annually had ferritin levels below 12 $\mu\text{g}/\text{liter}$ as compared with only 6% of the first-time female donors. It seems likely that blood donation-induced iron deficiency among women may be an important reason for the predominance of male donors among those individuals donating blood frequently and that the relationship between multiple blood donations and serum ferritin levels of women may be altered at the high frequency of blood donations by the dropouts. More direct evidence of the selection of females was the fact that 44 of the 51 donors rejected on the basis of low hemoglobin were women. The ability to donate blood would also be influenced by the self-initiated ingestion of medicinal iron by the blood donor, about which we have no information.

The literature dealing with the effects of blood donation on iron balance includes observations on changes in hemoglobin concentration, plasma iron concentration, total iron-binding capacity, and iron stores. Initially, hemoglobin concentration was the chief concern. Most reports agree that anemia occurs much more frequently among female donors¹⁵⁻¹⁷ and that this sex difference diminishes after the menopause.¹⁸ Hervey et al.¹⁵ found that 12.6% of 6911 women who had an adequate hemoglobin level for blood donation the first time failed to meet the accepted hemoglobin concentration of 12.3 g/100 ml on the next donation, the average interval being 5.3 mo. On the other hand, Denicourt and Goudemand¹⁶ measured the hemoglobins of 394 women who had given three donations yearly for 2-10 yr and found no significant differences compared to new donors. Other studies suggest that no anemia is seen among donors of both sexes giving 1700-3100 ml of blood annually.¹⁹⁻²¹ One of the difficulties of interpreting these reports is the lack of precise information about menstruation and iron ingestion and the degree of donor selection that may have occurred.

Decreases in plasma iron and transferrin saturation might be expected to provide a more sensitive index of iron status in blood donors. Schaffrin et al.²² monitored the effect 6 mo after a single donation in forty 18-yr-old girls and found the transferrin saturation to be <15% in 33% of these girls as compared to 8% in a control group. Gerland²³ found a marked decrease in plasma iron in both male and female donors giving over 1200 ml of blood per year. Hagberg et al.¹⁹ found an average decrease in plasma iron of 32 $\mu\text{g}/100\text{ ml}$ in a group of 26 donors who had been phlebotomized 1750 ml or more per year as compared to unbled subjects and donors giving smaller amounts of blood. Hagberg et al. also found an increase in total iron-binding capacity after the donation of 1000-1400 ml/year, whereas Schaffrin et al. reported a significant increase in this parameter 6 mo after a single donation in women.

Depletion of iron stores in blood donors has also been demonstrated by more direct measurements. For example, Olsson²⁰ measured iron stores by phlebotomy and found the average stored iron to be 110 mg in 14 male donors giving over 2 liters of blood per year. Weinfeld²⁴ demonstrated a decreased marrow hemosiderin as well as an increase in total iron-binding capacity in blood donors. Other measurements such as urinary iron excretion following desferrioxamine^{20,24} and plasma iron tolerance test measurements²⁵ were consistent with

a decrease in iron stores in regular donors. Assuming that there is a quantitative relationship between ferritin level and iron stores of approximately 8 mg of storage iron for 1 μg /liter of serum ferritin,²⁶ the effect of phlebotomy on iron stores in the present study can be estimated and is shown in Fig. 2.

These observations illustrate the varied potential of people to give blood. A significant portion of the female population cannot give blood without developing a significant degree of iron deficiency, whereas other individuals, largely males, can replace losses up to 2000 ml of blood per year without developing iron-deficiency anemia. The chief variables are available iron in the diet and blood loss, and perhaps ingestion of medicinal iron. Some 10% of menstruating women have iron requirements of 2 or more mg/day due to large menstrual losses.^{14,27} The difference in the prevalence of iron deficiency observed in the new donor populations of menstruating women studied here, estimated at 5.9%, and that of a previous nutritional study in the same area of 20%¹¹ could be related to dietary composition. While there is evidence that male subjects or non-menstruating females can absorb 3–4 mg of dietary iron per day, equivalent to a yearly blood loss of 2000 ml,^{1,28} this figure can be greatly influenced by the amount of meat and ascorbic acid in the diet.^{29,30} No general rules of blood donation are likely to fit each individual, especially when important variables such as available iron in the diet and the amount of menstrual blood loss in the female are not known.

The hematologic criterion of suitability of a donor for blood donation has been to establish that the individual's hemoglobin is above some cutoff point. This criterion is known to be highly inaccurate in identifying a pathologic hemoglobin concentration, i.e., one below the individual's physiologic norm.^{11,31} Inasmuch as the effect of phlebotomy is more specifically the reduction of body iron, it would seem desirable to define the donor's iron balance. Individuals already iron deficient by such measurements as transferrin saturation, red cell protoporphyrin, and serum ferritin should not give blood. Ferritin determination would be even more informative in identifying those individuals who may not be iron deficient by laboratory criteria, but who have reduced iron stores and would therefore be likely to develop iron deficiency if bled. It seems clear that some individuals who are bled are already iron deficient although not overtly anemic; some will be made iron deficient by a single blood donation, and the iron stores of all will be reduced.

This investigation and other studies suggest that insufficient attention has been given to the iron status of blood donors. Further information is needed about the background of donors in respect to economic status, racial derivation, and intake of supplemental iron. Factors responsible for the dropout of donors, particularly women in the older age groups, should be explored. Additional studies should be directed toward determining the extent to which iron stores may be replaced by supplemental iron after phlebotomy, what iron dose is optimal, and what duration of treatment is indicated.

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