Effect of iron supplementation on iron stores and total body iron after whole blood donation

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BACKGROUND: Understanding the effect of blood donation and iron supplementation on iron balance will inform strategies to manage donor iron status.

STUDY DESIGN AND METHODS: A total of 215 donors were randomized to receive ferrous gluconate daily (37.5 mg iron) or no iron for 24 weeks after blood donation. Iron stores were assessed using ferritin and soluble transferrin receptor. Hemoglobin (Hb) iron was calculated from total body Hb. Total body iron (TBI) was estimated by summing iron stores and Hb iron.

RESULTS: At 24 weeks, TBI in donors taking iron increased by 281.0 mg (95% confidence interval [CI], 223.4-338.6 mg) compared to before donation, while TBI in donors not on iron decreased by 74.1 mg (95% CI, −112.3 to −35.9; $p < 0.0001$, iron vs. no iron). TBI increased rapidly after blood donation with iron supplementation, especially in iron-depleted donors. Supplementation increased TBI compared to controls during the first 8 weeks after donation: 367.8 mg (95% CI, 293.5-442.1) versus −24.1 mg (95% CI, −82.5 to 34.3) for donors with a baseline ferritin level of not more than 26 ng/mL and 167.8 mg (95% CI, 116.5-219.2) versus −68.1 mg (95% CI, −136.7 to 0.5) for donors with a baseline ferritin level of more than 26 ng/mL. A total of 88% of the benefit of iron supplementation occurred during the first 8 weeks after blood donation.

CONCLUSION: Donors on iron supplementation replaced donated iron while donors not on iron did not. Eight weeks of iron supplementation provided nearly all of the measured improvement in TBI. Daily iron supplementation after blood donation allows blood donors to recover the iron loss from blood donation and prevents sustained iron deficiency.

Blood donation is known to reduce body iron stores, and frequent blood donors are often iron depleted.[1,2] A number of studies have demonstrated that iron supplementation can reverse the iron depletion and reduce hemoglobin (Hb) deferrals associated with blood donation.[3,4] Our group has recently documented that use of daily iron supplements is

ABBREVIATIONS: EBV = estimated blood volume; HEIRS = Hemoglobin and Iron Recovery Study; sTfR = soluble transferrin receptor; TBI = total body iron.

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Protocol available at: https://reds-iii.rti.org/ref/REDS_III_HEIRS_Protocol.pdf.

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necessary for efficient recovery of donor Hb and iron to support blood donation.[5] To date studies have separately assessed the effect of blood donation on iron stores, as assessed by serum or plasma ferritin, and Hb, as assessed by venous Hb concentration. To more fully understand the dynamics of iron balance into and out of the body, however, it is necessary to assess these two major iron compartments together. We propose here to use the concept of total body iron (TBI) as a novel and comprehensive model to quantify iron after blood donation. We applied this model to data collected in the Hemoglobin and Iron Recovery Study[5] (HEIRS); this allowed us to carefully describe iron balance after blood donation and the effects of iron supplementation on iron recovery.

MATERIALS AND METHODS

The HEIRS was conducted, as previously reported, at four US blood centers.[5] An eligible participant was a volunteer, repeat blood donor at least 18 years of age who successfully donated a whole blood unit on the day of enrollment, but not in the prior 4 months. Participants agreed not to use iron supplements outside of the study. Participants returned 3 to 8 days later for their first postdonation visit and classification of index donation ferritin results. Participants were classified for predonation iron status as iron-depleted ("low ferritin," $\leq$26 ng/mL) and iron-replete ("higher ferritin," >26 ng/mL) based on the ferritin measurement at enrollment. Randomization was conducted within eight strata, based on ferritin classification, sex, and age. If a participant met stratification requirements, based on their stratum remaining open for enrollment, they were randomly assigned (in equal numbers within each stratum) to receive either daily oral ferrous gluconate 325 mg containing 37.5 mg of elemental iron for 24 weeks or no treatment. During a 24-week follow-up period, they provided periodic blood samples.

Participants studied

A total of 215 donors were randomly assigned and followed further; 111 (51 low ferritin and 60 higher ferritin) were assigned to ferrous gluconate, and 104 (50 low ferritin and 54 higher ferritin) were assigned to no supplemental iron. Twelve participants (nine on iron; three not on iron) withdrew from HEIRS before completion of all the visits, and 10 additional participants (six on iron; four not on iron) were missing Hb values on two or more follow-up visits, leaving 193 participants analyzed for all parameters (96 on iron, 97 not on iron).

Available samples and laboratory methods

Plasma ferritin (Advia Centaur) and complete blood count were measured on EDTA blood samples collected before index donation and once between Days 3 and 8 and 2, 4, 8, 12, 16, and 24 weeks after donation. Soluble transferrin receptor (sTfR; Roche Tina-quant) was tested at the predonation and 24-week visits only.[1]

Iron stores

Iron stores (mg/kg body weight) were calculated at enrollment and at the 24-week visit, where both ferritin and sTfR were available, from the reported relationship between iron stores and the log ratio of serum transferrin receptor/ferritin:

$$\text{Iron stores (mg/kg)} = -[\log(\text{sTfR/ferritin}) - 2.8229]/0.1207.^{6}$$

Plasma ferritin values were increased by 5% to better reflect serum ferritin values required for use in the formula (Assay Manual, ADVIA Centaur, Siemens Healthcare Diagnostics, 2012). Values for the Roche sTfR were converted to the original ("Flowers")[7] sTfR values used in Cook from a previous comparison of the two assays according to the formula:

$$\text{Flowers sTfR} = 1.5\ \text{Roche sTfR} + 0.35\ \text{mg/L}.^{8}$$

To evaluate TBI at intermediate points during follow-up, we generated estimates of iron stores at all seven follow-up visits. These were obtained by fitting a regression of iron stores (calculated as above at enrollment) on log ferritin using enrollment data from the 193 participants included in the analysis (Fig. 1). A similar regression formula derived from the same subjects' 24-week samples gave nearly identical results (data not shown). Iron stores at all seven follow-up visits were then estimated from this regression and TBI was calculated as described below.

TBI calculations

We calculated TBI as

$$\text{TBI} = \text{red blood cell (RBC) iron (mg)} + \text{iron stores (mg/kg)} \times \text{weight (kg)}.$$

To determine RBC iron (i.e., iron in Hb), we estimated that Hb contained 3.4 mg iron/g.[9] We calculated estimated blood volume (EBV) from height, weight, and sex using the Nadler formula[10] and then calculated RBC iron from EBV and venous Hb (per L) as EBV $\times$ Hb $\times$ 0.91.[11]

Changes in TBI

Changes in TBI were calculated over the entire 24-week period of observation using predonation TBI values and comparing them to 24-week TBI values, as well as over three consecutive adjacent intervals: Interval 1, 0- to 8-week sample; Interval 2, 8-week sample to 16-week sample; and Interval 3, 16-week sample to 24-week sample. These intervals were chosen in part to reflect the 8-week interdonation interval currently allowed in the United

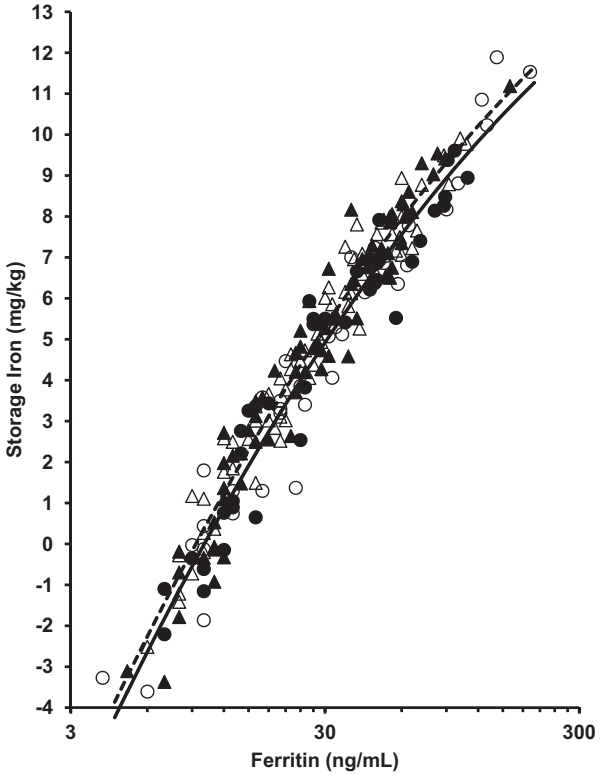

Fig. 1. Quadratic regression of predonation storage iron (mg/kg) against predonation ferritin (ng/mL) for 193 subjects enrolled in the HEIRS study. Storage iron was determined from ferritin and sTfR as described under Materials and Methods. (—) Male regression; (– – –) female regression. HEIRS randomization assignment of the individual donors (see Materials and Methods): (▲) female, iron; (△) female, no iron; (●) male, iron; (○) male, no iron.

States. Using shorter intervals for analysis also led to less precise estimates as these were based on fewer visits, which resulted in an increased proportion of missing data in given intervals. However, because of interest in defining the optimal duration of iron supplementation, a secondary analysis was performed on 4-week intervals during Weeks 0 to 16.

The TBI at the beginning of Interval 1 (Time 0, i.e., the TBI immediately after donation) was calculated by taking the predonation TBI and subtracting the amount of iron in the donated whole blood. The iron in the donated unit was calculated as predonation venous Hb multiplied by donated volume of 525 mL multiplied by 3.4 mg iron/g Hb.

Statistical methods and models used

Linear models were employed for all statistical comparisons. To examine the effects of treatment (iron or not), baseline ferritin, and other predictors on changes in TBI over the three 8-week intervals, we employed a repeated-measures model. Reduced forms of the model were employed to analyze subsets of the data, such as the effect of treatment on changes in a single interval. A t test was employed for comparing two means, such as the mean 24-week change in storage iron in those taking and not taking iron.

We used multiple imputation to account for the manner in which storage iron was calculated from ferritin alone. A separate normally distributed random variate with mean of 0 and standard deviation of 0.80 was added to each estimate of storage iron to account for the residual uncertainty in the estimates. This involved creating 10 data sets with separate imputed values in each, analyzing each of the data sets and then combining the results to obtain final estimates and p values. Data analysis took place using a computer (SAS/STAT software Version 9.3 of the SAS System for Windows, SAS Institute).

TABLE 1. Iron content of whole blood donated by the 193 participants (mg iron/donation)

	Number	Mean (95% CI)	Median
Men	75	260.0 (255.1-264.9)	260.6
Women	118	238.4 (235.5-241.3)	239.2
Total	193	246.8 (243.8-249.8)	246.3

RESULTS

Iron in donated whole blood

The iron content of the units of whole blood donated by the 193 participants is shown in Table 1. As expected there is a 9% difference between men and women, reflecting the higher male Hb (men 260 mg iron, women 238.4 mg iron). The median and mean are nearly the same, indicating a symmetric distribution, with a small confidence interval (CI) around the mean. Individual donor's iron losses, however, varied considerably with a range from low to high of nearly 100 mg.

TBI: changes over 24 weeks

TBI at enrollment (before donation) and at 24 weeks is shown in Table 2. The two treatment groups had nearly identical iron stores and RBC iron before donation. As expected, only 15% of TBI was found bound to ferritin as iron stores, with 85% found in the RBC mass. At 24 weeks, TBI in the iron-supplemented group increased on average by 281.0 mg (95% CI, 223.4 to 338.6) compared to before donation. Most of this increase occurred in the iron stores, with a smaller increase in RBC iron. Since on average 247 mg of iron was lost through a blood donation (Table 1), these donors not only replenished the Hb iron lost during their donation but added another RBC equivalent to their TBI. In contrast, the group not on iron decreased TBI by 74.1 mg (mean, −74.1 mg; 95% CI, −112.3 to −35.9; $p < 0.0001$, iron vs. no iron), the decrease equally divided between iron stores and RBC iron. These

TABLE 2. TBI (mg) at enrollment and change at final visit*

	Iron (n = 96)		No iron (n = 97)	
	Before donation	24-week change (Δ)	Before donation	24-week change (Δ)
Iron stores	344.8 (287.1-402.5)	219.4 (179.4-259.4)	347.6 (287.1-408.0)	−35.1 (−66.1 to −4.1)
RBC iron	2038.4 (1943.1-2133.6)	61.6 (35.7-87.5)	2034.1 (1936.8-2131.4)	−39.0 (−65.7 to −12.3)
TBI	2383.1 (2252.8-2513.5)	281.0 (223.4-338.6)	2381.7 (2251.2-2512.1)	−74.1 (−112.3 to −35.9)

* Data are reported as mean (95% CI). All Δ values, $p < 0.0001$, iron vs. no iron.

donors thus failed to replace the RBC iron lost during their donation, even after 24 weeks.

Regression for iron stores at interim visits, based solely on ferritin

To track changes in TBI, we developed a regression to estimate iron stores based solely on ferritin, since sTfR was not available at interim bleeds. Figure 1 plots a quadratic regression of iron stores against log ferritin where stored iron was calculated from ferritin and sTfR, as described above.

The formula for the regression is:

$S = -13.8588 + 0.3929G + 15.5999\log(F) - 2.0519(\log(F))^2$, where S is storage iron in mg/kg, F is ferritin level in ng/mL, and G is 0 for men and 1 for women. The model explained most of the variance in stored iron ($r^2 = 0.95$). Analysis of residuals showed an excellent fit to the data throughout the range.

Figure 2 shows the mean change in TBI over time in the four subgroups (low or high ferritin, iron or no iron treatment) within HEIRS. All groups showed an initial decrease in the Day 3 to 8 sample, reflecting the initial blood donation, with recovery shown by later follow-up samples. Within 4 to 13 weeks, depending on baseline ferritin levels, donors receiving iron supplements had replaced the donated iron. For donors on iron who were iron depleted before donation, replacement occurred in 4 weeks (to predonation iron-deficient levels), but then iron absorption continued so that by 24 weeks these donors had absorbed an additional 400 mg of iron (Fig. 2). For donors with higher ferritin levels at baseline, recovery to their predonation iron-replete status was slower but still complete by 13 weeks. Donors not on iron supplementation did not completely recover TBI by 24 weeks—although the low ferritin group nearly did so; however, their "recovery" was to an iron-deficient level.

Kinetics of recovery of TBI after whole blood donation

Table 3 shows the change in TBI over three consecutive adjacent 8-week intervals after blood donation. Subjects receiving iron increased TBI significantly compared to

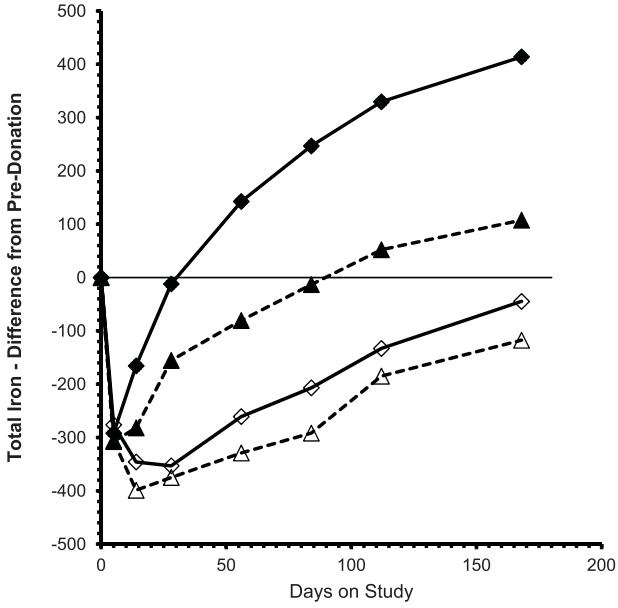

Fig. 2. Change in TBI (mg) from predonation TBI at seven postdonation visits from Day 3 to 8 to 24 weeks after donation (see Materials and Methods). (◆, ▲) Donors randomized to oral iron supplements; (◇, △) donors randomized to no iron. (_____, ◇, ◆) Donors whose predonation ferritin level was not more than 26 ng/mL; (----------, △, ▲) donors whose predonation ferritin level was more than 26 ng/mL.

subjects not on iron during the first 8 weeks after blood donation: a 367.8-mg increase (95% CI, 293.5 to 442.1 mg) versus a decrease of 24.1 mg (mean change, −24.1 mg; 95% CI, −82.5 to 34.3; $p < 0.0001$, iron vs. no iron) for those with a baseline ferritin level of not more than 26 ng/mL and a 167.8-mg increase (95% CI, 116.4 to 219.2) versus a decrease of 68.1 mg (mean change, −68.1; 95% CI, −136.7 to 0.5; $p < 0.0001$, iron vs. no iron) for donors with a baseline ferritin level of more than 26 ng/mL. However, there was no significant effect of iron supplementation compared to no iron in either ferritin group during Intervals 2 and 3.

For those donors on iron, the increase in TBI was highest in Interval 1. For those donors on iron with a

TABLE 3. Change in TBI (mg/interval) at three consecutive adjacent intervals after blood donation*

		Ferritin level (ng/mL)			
		≤26		>26	
Weeks after donation	Interval	Iron	No iron	Iron	No iron
0-8	1	367.8 (293.5-442.1)	−24.1 (−82.5 to 34.3)	167.8 (116.4-219.2)	−68.1 (−136.7 to 0.5)
8-16	2	183.9 (132.2-235.6)	138.3 (87.7-188.9)	129.8 (81.8-177.8)	140.9 (75.6-206.2)
16-24	3	90.0 (44.1-135.9)	81.6 (27.9-135.3)	66.0 (13.9-118.1)	65.2 (4.6-125.8)
p value†	Overall	<0.0001	<0.0001	<0.0001	<0.0001
	1 vs. 2	0.0001	0.0003	0.38	0.0006
	2 vs. 3	0.0309	0.19	0.13	0.17
	1 vs. 3	<0.0001	0.0124	0.0064	0.0060
p values for comparison of changes in total iron (mg/interval) in those taking and not taking iron‡					
0-8	1	<0.0001		<0.0001	
8-16	2	0.19		0.79	
16-24	3	0.81		0.98	

* Data are reported as mean (95% CI) change for donors with at least 6 follow-up Hb values.
† p value for comparing changes among intervals.
‡ p values are based on t tests.

baseline ferritin level of not more than 26 ng/L, TBI in Intervals 2 and 3 had smaller increases than in Interval 1. For those donors on iron with a baseline ferritin level of more than 26 ng/mL, TBI in successive intervals was not statistically different, but the increase in TBI in Interval 1 was greater than that in Interval 3.

Donors not on iron experienced a decrease in TBI during Interval 1–beyond that already accounted for from the blood donation. This observation is reflected in the TBI recovery curves for these donors shown in Fig. 2. TBI continued to decrease during the first 2 weeks after blood donation and then flattened out at 4 weeks, partially recovering only between the 4- and 8-week samples. As shown in Table 3 the decrease in Interval 1 was reversed by a substantial increase in TBI in Interval 2, which was not statistically different from that observed in Interval 3.

To better define the optimal duration of iron supplementation, the above analysis was repeated for successive 4-week intervals over the first 16 weeks: 0 to 4, 4 to 8, 8 to 12, and 12 to 16 weeks. The benefit of iron decreased in each successive 4-week interval and no benefit was shown during weeks 12-16. (Table 4) A statistically significant improvement in TBI due to iron supplementation could only be demonstrated in the 0-4 week interval. In initially iron-depleted donors 88% of the benefit of iron occurred in the first 8 weeks after blood donation, and 87% in the iron-replete donors.

DISCUSSION

Our approach to estimating body iron in blood donors is to focus only on the two major compartments, Hb and storage iron. For convenience we have termed the total of these two compartments TBI. While this measurement ignores minor iron compartments such as transferrin, myoglobin, and iron cofactors in enzymes and cytochromes, it is ideally suited to assess changes in body iron over time, since any change in minor compartments is likely to be small in early iron depletion. By combining changes in TBI with a precise estimator of iron lost in a blood donation, it is possible to assess the effect of iron supplements expressed as milligrams per time interval. This approach allows a dynamic look at iron balance in blood donors.

We have shown that TBI increases from predonation levels in iron supplemented donors by a mean of 281 mg/dL over 24 weeks. A unit of RBCs was found to represent approximately 247 mg of iron. Since this index blood donation needs replacement to maintain TBI at preenrollment levels, this means that approximately 500 mg of iron was absorbed from the gastrointestinal tract (from iron supplements and dietary iron) over and above that necessary to replace insensible iron losses occurring through loss of skin, hair, and gastrointestinal mucosa. Put another way, the donor absorbed the iron to produce 2 units of RBCs, the one donated and another one. In the absence of iron, however, donors lost 74 mg of iron from preenrollment levels; put another way, over 24 weeks the nonsupplemented donors could only replace two-thirds of the index donation's iron. Since diet and insensible iron losses would not be expected to differ between the iron-supplemented donors and the no-iron donors, the differences in TBI between the two groups are explained solely by the iron supplements. By combining the gain of

TABLE 4. Benefit of iron supplementation on TBI over 4-week intervals after blood donation*

	Ferritin level (ng/mL)			
	≤26		>26	
Interval (weeks)	Benefit of iron (mg)	% of total iron benefit†	Benefit of iron (mg)	% of total iron benefit†
0-4	323.1‡	73	215.7‡	78
4-8	65.6	15	24.4	9
8-12	53.8	12	37.1	13
12-16	(11.8)§	NA	(46.1)§	NA

* Benefit from iron is shown as differences in change in TBI (mg/interval) between iron supplemented and no iron donors at four consecutive 4-week intervals after blood donation.
† Total iron benefit is the total of Weeks 1 to 12 and ignores Weeks 12 to 16.
‡ $p < 0.0001$, TBI in iron group compared to no iron group. All others are NS.
§ TBI increased in the no iron group more than in the iron group.

281 mg in the supplement group with the loss of 74 mg in the no-iron group, the difference of 355 mg of iron is shown to be attributable to and a benefit of the iron supplements. This represents 5.6% of the total iron content in the 24 weeks of provided iron supplements.

By plotting TBI over sequential time periods after blood donation, the benefits of supplemental iron in recovery from donation are clear. The recovery curves in Fig. 2 resemble the Hb recovery curves that were observed for the same groups in HEIRS,[5] which is expected given that the majority of TBI is represented by Hb (Table 2). The more rapid recovery in the initially iron-depleted donors is likely due to increased absorption of supplemental iron seen in iron-depleted individuals as well as higher erythropoietic drive due to their relative anemia before donation. In comparison, recovery of TBI in the no iron groups was delayed and incomplete, even by 24 weeks.

Importantly, by estimating the change in TBI over consecutive adjacent 8-week intervals, we have demonstrated that the benefit of iron supplementation is most clearly shown during the first 8 weeks after donation (Tables 3 and 4). During this period, and especially in the first 4 weeks, the rate of increase in TBI is quite high (e.g., 3-6 mg/day), with higher rates in supplemented donors with preenrollment iron deficiency (ferritin level ≤ 26 ng/mL). There was some suggestion from the data in Table 3 that continued iron supplements beyond 8 weeks might benefit some donors. There is a higher rate of TBI increase in Interval 2 (8-16 weeks) versus Interval 3 (16-24 weeks) particularly in the iron-depleted group ($p = 0.03$). To analyze this further, the increase in TBI during successive 4-week intervals was analyzed over the first 16 weeks (Table 4). Perhaps because the statistical power of this shorter interval analysis was reduced compared to the 8-week interval analysis, a significant benefit of iron could only be demonstrated during the first 4 weeks after donation. An analysis of the contribution of each successive interval to the overall iron benefit showed that 87% to 88% of the benefit of iron supplementation occurred during the first 8 weeks after donation. The rapid increase in TBI in iron-supplemented donors contrasts with small decreases in TBI in nonsupplemented donors over the first 8 weeks after donation. The reasons for the first 8-week decrease in TBI in this group are not clear. They may relate to changes in iron compartments that are not included in TBI. In this model, TBI is the sum of iron stores and RBC (Hb) iron but does not include minor compartments such as transport (i.e., transferrin), marrow, and cytochrome iron. Changes in iron in these minor compartments would not be detected in the observed TBI compartments, thus distorting the apparent changes in TBI shown in Table 3. For example, it is possible that the expected expansion of the RBC precursors in the marrow after donation could account for postdonation undercounting of body iron in the TBI calculation. Although this would be expected in all groups, it is possible that this effect is overwhelmed by the positive effect on iron supplements, so that it is observed only in the nonsupplemented donors. Finally, the observations may be due to other small measurement inaccuracies in the model, which are apparent only in the nonsupplemented donors, being hidden by the effect of the iron supplementation in the iron groups. Further validation of this model would require the use of radiolabeled iron studies, which would not be feasible in the context of a large study such as HEIRS. However, the model presented here illustrates the dynamics of iron balance in blood donors and the substantial impact of blood donation on body iron.

Given the delay in recovery of the Hb iron compartment in donors who were not taking a supplement, it is no surprise that studies have reported that donors who are deferred for low Hb (most of whom are iron-depleted)[12] improve their chances of successfully donating very little by waiting longer between donations.[13] For example, a deferred donor with Hb level of 12.0 gm/dL with a 60% chance of passing the fingerstick Hb screen after waiting 1 to 2 weeks until his or her next donation attempt only improved to 70% passing by waiting 21 to 24 weeks between deferral and return. From the data

presented here and earlier-reported results,[5] it is now clear that oral iron supplementation at small, well-tolerated doses is necessary to recover iron and Hb levels promptly after whole blood donation and to maintain iron homeostasis in frequent blood donors. Further, the model provides important data indicating that 8 weeks of iron supplementation after blood donation is an effective and efficient way to replenish iron lost from donation. Since it is increasingly apparent that routine iron supplementation of many or most blood donors is needed, it is very important to understand how long after a whole blood donation oral iron supplementation should be offered or recommended. Our data clearly indicate that no more than 8 weeks of oral iron supplementation is necessary. This information should be useful to US blood centers that have implemented or are currently evaluating how best to implement oral iron supplementation among their donors. However, further blood center operational trials to optimize oral iron supplementation methods and to determine the acceptability of and compliance with various methods for donors are needed.

There are study limitations that may affect the generalizability of our findings. First, whereas HEIRS studied an iron dose of 37.5 mg/day, many center-proposed operational protocols anticipate a daily iron dose of 19 mg, either as a supplement or included in a multivitamin. Second, we found excellent compliance from the paid participants in this study. It is possible that reduced compliance in more routine settings may affect the ingested iron dose over the recommended supplementation period. Consequently it is possible that 19 mg/day should be recommended for longer than 8 weeks in routine donor management. However, the very brisk iron response shown in our study in the first 4 weeks indicates that 8 weeks would likely be an adequate recommended supplementation period, even for the reduced dose of 19 mg/day or a somewhat reduced compliance. Findings from STRIDE[14] reinforce the likelihood that an 8-week supplementation period using 19 mg would probably be effective since no difference was noted in response of donor iron and Hb variables between doses of 19 and 38 mg/day, when given to frequent whole blood donors for 8 weeks after a whole blood donation. Finally, we only evaluated responses to iron supplementation after a whole blood donation and did not address iron management after double-RBC donation or after multiple plateletpheresis donations. The data presented here do show, however, that the response to oral supplemental iron is immediate and brisk, particularly in iron-depleted donors, and our findings should be considered in planning and studying iron supplementation interventions in these other populations.

The assessment of TBI in HEIRS represents a novel approach to studying iron balance, which is also applicable to a variety of other clinical and community settings. Most of the literature has focused on the storage iron component of TBI.[6,15] While the storage iron approach works well in populations with relatively stable Hb levels,[16] it is not a comprehensive assessment of iron balance in a blood donation setting, where lowering of the Hb is expected, with or without subsequent recovery and where Hb represents the major reservoir of iron in the body. The current TBI approach can be used in other cross-sectional and longitudinal studies of blood donor iron depletion and in trials of iron supplementation. Applying this model to other studies should reveal additional important facets of donor iron management, leading to improved donor well-being in addition to enhancing the blood supply.

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CONFLICT OF INTEREST

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