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Time-Dependent Associations between Iron and Mortality in Hemodialysis Patients

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The independent association between the indices of iron stores or administered intravenous iron, both of which vary over time, and survival in patients who are on maintenance hemodialysis (MHD) is not clear. It was hypothesized that the observed associations between moderately high levels of three iron markers (serum ferritin, iron, and iron saturation ratio) or administered intravenous iron and all-cause and cardiovascular death is due to the time-varying confounding effect of malnutrition-inflammation-cachexia syndrome (MICS). Time-dependent Cox regression models were examined using prospectively collected data of the 2-yr (July 2001 to June 2003) historical cohort of 58,058 MHD patients from virtually all DaVita dialysis clinics in the United States. After time-dependent and multivariate adjustment for case mix, administered intravenous iron and erythropoietin doses, and available surrogates of MICS, serum ferritin levels between 200 and 1200 ng/ml (reference 100 to 199 ng/ml), serum iron levels between 60 and 120 μg/ml (reference 50 to 59 μg/ml), and iron saturation ratio between 30 and 50% (reference 45 to 50%) were associated with the lowest all-cause and cardiovascular death risks. Compared with those who did not receive intravenous iron, administered intravenous iron up to 400 mg/mo was associated with improved survival, whereas doses >400 mg/mo tended to be associated with higher death rates. The association between serum ferritin levels >800 ng/ml and mortality in MHD patients seems to be due mostly to the confounding effects of MICS. For ascertaining whether the observed associations between moderate doses of administered intravenous iron and improved survival are causal or due to selection bias by indication, clinical trials are warranted.

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In patients who are on maintenance hemodialysis (MHD), markers of anemia, including low blood hemoglobin concentrations, are associated with poor clinical outcomes (1,2). Consequently, management of anemia by recombinant human erythropoietin (EPO) by increasing serum hemoglobin toward the target of 11.0 to 12.0 g/dl is reported consistently to improve outcome measures in MHD patients (3–6). However, much controversy exists with regard to the association between measures of iron stores or changes in these values by iron administration and clinical outcome in these individuals (7). Although in the pre-EPO era iron overload was a major cause of morbidity in MHD patients, its significance in the post-EPO era remains unclear. A recent observational study in 1283 MHD patients indicated that a low, rather than a high, baseline serum iron quartile was associated with increased mortality and hospitalization over 12 mo of observation (8). Even though quartile analysis enables us to uncover nonlinear relationships, large sample sizes with repeated measures are needed to compare the mortality predictability of smaller increments of iron markers. Moreover, the association between other iron markers and survival remains to be determined.

Many observational studies have examined the association between a baseline laboratory value at the start of the cohort and the survival and ignored variations in clinical and laboratory measures over time. It is not clear whether longitudinal changes in laboratory markers of iron stores or injected intravenous iron have meaningful associations with survival after adjustment for time-varying confounders and, if so, in which direction. Time-dependent multivariate adjustments are especially important for serum ferritin and total iron binding capacity (TIBC), which may also be markers of inflammation and/or nutritional status (9,10), conditions that may fluctuate over time in MHD patients.

Changes in serum concentrations of iron markers over time may also be a result of the time-varying administered EPO and intravenous iron. Because the medical treatment of MHD patients is usually based on the periodically repeated measure-

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ment of blood tests, rather than mere use of a set of old baseline data, examining such associations using traditional Cox regression models that use baseline data and ignore subsequent changes over time may be flawed. Hence, we examined models that are based on time-varying values of repeated measures of not only the iron markers but also a large number of potential confounding covariates. We hypothesized that the observed and apparently positive associations between moderately high levels of serum ferritin, iron, and iron saturation ratio (ISAT), also known as transferrin saturation ratio, or administered intravenous iron and the increased all-cause and cardiovascular death risk is essentially due to the confounding by the time-varying surrogates of malnutrition and inflammation.

Materials and Methods

Patients

We examined prospectively collected data of a 2-yr (July 1, 2001, through June 30, 2003) historical cohort of all incident and prevalent MHD patients in the national database of DaVita, Inc., the then second largest dialysis care provider in the United States. Database creation has been described elsewhere (11–13). In summary, this database included information on approximately 40,000 maintenance dialysis patients at any given time. All repeated measures for each patient within a given quarter (13-wk interval) were averaged to obtain one quarterly mean value and to mitigate the effect of short-term variations. Hence, up to eight repeated and quarterly varying values were available for each measure in each MHD patient over a 2-yr observation period. The study was approved by the Institutional Review Committees of Harbor-UCLA and DaVita, Inc.

Age was estimated subtracting the date of birth from the first day of the entry quarter. Six race/ethnic groups were defined: Caucasians (including non-Hispanic whites and Middle Easterners), self-described blacks (including African Americans and sub-Saharan Africans), Asians (including Pacific Islanders), American Indians, Hispanic, and others. Patients' postdialysis weight from each hemodialysis treatment was averaged over each 13-wk quarter, and the body mass index (BMI; weight in kg divided by height in m^2) was calculated.

Cohort time included the number of days that a patient participated in the cohort and was a number between 1 and 731 d. Dialysis vintage was defined as the duration of time between the first day of dialysis treatment and the first day that the patient entered the cohort. Four categories of vintage were formed: (*1*) First 6 mo, (*2*) between 6 and 24 mo, (*3*) between 2 and 5 yr, and (*4*) >5 yr. The entry quarter was defined as the first quarter in which the patient's dialysis vintage was >3 mo for at least half the duration of the quarter. By implementing this criterion, any patient who did not remain in the cohort beyond the first 3 mo of MHD was excluded. The computerized causes of death, reflecting the reported information in the Cause of Death form (Form 2746), were obtained and summarized into five main categories: Cardiovascular (CV), infectious, gastrointestinal, cancer related, and others/unspecified/unknown. CV death included death as a result of myocardial infarction, cardiac arrest, heart failure, cerebrovascular accident, sudden death, and other cardiac events.

Laboratory Methods

All laboratory measurements were performed by DaVita Laboratories in Deland, FL, using standardized and automated methods. For each laboratory measure, the average of all available values obtained within any given calendar quarter were used in all analyses. Most blood samples were collected predialysis, with the exception of the postdialysis serum urea nitrogen to calculate urea kinetics. The normalized protein nitrogen appearance (nPNA), also known as protein catabolic rate, and the Kt/V (single pool) were calculated using urea kinetic modeling formulas (14,15). Blood samples were drawn using uniform techniques in all DaVita dialysis clinics across the nation and were transported to the DaVita Laboratory in Deland, FL, usually within 24 h. Most laboratory values, including complete blood cell counts and serum levels of urea nitrogen, albumin, creatinine, calcium, phosphorus, bicarbonate, iron, and TIBC, were measured monthly. Ferritin was usually measured at least once during each calendar quarter. Both nPNA and Kt/V were calculated monthly. Hemoglobin was measured weekly to biweekly in most patients.

Twelve categories of ferritin and 11 categories of ISAT and serum iron were created to cover the entire range of each of the foregoing iron markers. The choice for cutoff levels of increments was based on both the clinical meaningfulness and applicability and the availability of adequate population, including death cases in each incremental group. Special efforts were made to accommodate some more often used cutoff levels of such known practice guidelines such as Kidney Disease Outcome Quality Initiative (K/DOQI) of the National Kidney Foundation (16) and the Best Practice Guidelines of the European Renal Association (17), including cutoff levels of 100, 500, and 800 ng/ml for serum ferritin and 20 and 50% for ISAT.

In addition to the BMI, nine time-varying (quarterly changing) laboratory variables with known association with mortality in MHD patients were selected as the surrogates of the nutritional status and/or inflammation, together also known as malnutrition-inflammation-cachexia (or complex) syndrome (MICS) (18): (*1*) nPNA, a urea kinetic indicator of protein intake (19); (*2*) serum albumin, a known outcome indicator with strong correlations with both C-reactive protein (CRP) and dietary protein intake (20,21); (*3*) serum TIBC, which has a strong association with subjective global assessment of nutritional status (9,22); (*4*) serum creatinine, an indicator of muscle mass and possibly dietary protein intake (23); (*5*) serum phosphorus, a correlate of food intake, clinical outcome, and compliance with recommended phosphorus binders (24); (*6*) serum calcium, as described for phosphorus (24); (*7*) serum bicarbonate, an indicator of interdialytic protein intake (25); (*8*) peripheral white blood cell count, which correlates with serum CRP and survival in MHD patients (26,27); and (*9*) lymphocyte percentage, a known nutritional marker that can decrease with protein-energy malnutrition (28) and correlates independently with mortality in MHD patients (27).

Administered In-Center Medications

The dose of the in-center (dialysis facility) administered medications that were related to the management of iron and anemia were also included in all case-mix adjusted models as additional covariates. The dose of EPO is expressed in units/wk, whereas the dose of intravenous iron is in mg/mo. Three different types of intravenous iron was administered: (*1*) Iron gluconate (Ferrlecit; Watson Pharmaceuticals, Corona, CA) (29,30), (*2*) Iron sucrose (Venofer; American Regent, Inc., Shirley, NY) (31,32), and (*3*) Iron dextran (Infed, Watson, or Dexferrum, American regent) (33,34). During each of the eight quarters, on average 95% of MHD patients received EPO and 65% received intravenous iron at least once. During the first 3 quarters, >90% of MHD patients who were administered intravenous iron received iron gluconate, whereas during the last three quarters, this proportion was 5 to 10%, because iron sucrose was the dominating form of the administered intravenous iron. Intravenous iron dextran was given to <5% of the patients throughout all eight quarters. For examining the effect of administered intravenous iron on survival, all three forms of intravenous iron were merged into one single variable regardless of the type of the adminis-

Table 1. Baseline data of the nonconcurrent (left truncated) cohort of 58,058 MHD patients, including 37,049 from the first quarter (q1) and 21,009 from subsequent quarters (q2 to q8)[a]

Variable	Received IV Iron at Baseline[b] (n = 38,569)	No IV Iron at Baseline[b] (n = 19,489)	P Value
Age (yr)	61 ± 15	61 ± 15	0.7
Gender (% women)	46	46	0.06
Diabetes (%)	46	42	<0.001
Race and ethnicity (%)			
white	37	37	0.10
black	32	31	0.004
Hispanic	17	18	0.04
3 to 6 mo on dialysis (%)	48	26	<0.001
Medicare (%)	59	61	<0.001
Known causes of death (%)			
cardiovascular	52	51	0.09
infectious	11	11	0.6
Standardized mortality ratio	0.80 ± 0.23	0.79 ± 0.23	<0.001
Cohort time (d)	447 ± 255	465 ± 263	<0.001
BMI (kg/m^2)	26.3 ± 6.3	25.8 ± 5.6	<0.001
Kt/V (single pool)	1.5 ± 0.3	1.6 ± 0.3	<0.001
nPNA (g/kg per d)	1.0 ± 0.2	1.0 ± 0.2	<0.001
Serum albumin (g/dl)	3.7 ± 0.4	3.8 ± 0.4	<0.001
creatinine (mg/dl)	8.8 ± 3.3	9.4 ± 3.3	<0.001
TIBC (mg/dl)	203 ± 42	200 ± 43	<0.001
ferritin (ng/ml)	556 ± 448	718 ± 596	<0.001
iron (ng/ml)	59 ± 25	65 ± 29	<0.001
iron saturation ratio (%)	29 ± 11	33 ± 13	<0.001
bicarbonate (mEq/L)	21.7 ± 2.8	22.0 ± 2.9	<0.001
phosphorus (mg/dl)	5.7 ± 1.5	5.7 ± 1.6	<0.001
calcium (mg/dl)	9.2 ± 0.7	9.3 ± 0.8	<0.001
Blood hemoglobin (g/dl)	12.1 ± 1.3	11.8 ± 1.3	<0.001
WBC (per fl)	7.3 ± 2.3	7.3 ± 2.4	<0.001
lymphocyte (% of total WBC)	21 ± 8	21 ± 8	<0.001
Administered EPO			
proportion received EPO (%)	99	82	<0.001
EPO dose (units/wk)	21,560 ± 17,660	16,580 ± 30,440	<0.001

[a]MHD, maintenance hemodialysis; IV, intravenous; BMI, body mass index; nPNA, normalized protein nitrogen appearance; TIBC, total iron binding capacity; WBC, white blood cell; EPO, recombinant human erythropoietin.
[b]Baseline period of the cohort pertains to the first 3 mo (first calendar quarter of the cohort).

tered intravenous iron, and four groups of MHD patients were created: (*1*) Those who did not receive any intravenous iron during the entire 13 wk of a given quarter; (*2*) those who received intravenous iron of any type between 1 and 199.9 mg/mo; (*3*) those who received intravenous iron between 200 and 399.9 mg/mo; and (*4*) those who received intravenous iron of 400 mg/mo or greater.

Statistical Analyses

Eight quarterly data sets were linked using unique patient identifiers. A nonconcurrent cohort with quarterly units and quarterly values for each time-varying variable was also formed to include all existing MHD patients of the first quarter (q1) and all new MHD patients of the subsequent quarters (q2 through q8). In addition to eight quarterly values for every variable, a baseline value was also created for each measure by left-truncating the first available value of the entry quarter for each patient. Standard descriptive statistics were performed. Both conventional and time-dependent Cox proportional hazard regression analyses for truncated and censored data (PROC PHREG) (35) were examined. These statistical analyses were conducted to determine whether the 2-yr survival was associated with baseline or time-varying categories of serum indices of iron or administered intravenous iron. A hazard ratio >1 indicates increased death risk and <1 suggests improved survival.

For each analysis, three types of models were examined on the basis of the level of multivariate adjustment: (*1*) Unadjusted models included one of the three iron markers (serum ferritin, ISAT, or iron) as the predicting variable, entry quarter as the covariate, and mortality as the outcome variable; (*2*) case-mix adjusted models included additional covariates age (continuous), gender (two groups), race and ethnicity (six groups), diabetes (yes/no), vintage categories (four groups), pri-

mary insurance (Medicare, Medicaid, private, and other), marriage status (married, single, divorced, widowed, and other), and standardized mortality ratio of the dialysis clinic during entry quarter (continuous), as well as continuous values of Kt/V (single pool), blood hemoglobin, and administered doses of each of the three intravenous iron medications and EPO; and (*3*) case-mix and MICS adjusted models included all of the above-mentioned covariates as well as 10 indicators of nutritional status and inflammation, including BMI and all nine above-mentioned laboratory values. In time-dependent Cox models, in addition to time-varying quarterly iron markers and intravenous iron categories, 10 indicators of MICS, Kt/V, and administered doses of medication were included as time-varying covariates with up to eight separate quarterly values per variable per patient. When the intravenous iron composite was modeled as the predicting variable, all three serum iron markers were included as case-mix covariates. Missing covariate data (<5%) were imputed by the mean or the median of the existing values, whichever was most appropriate. All descriptive and multivariate statistics were carried out with SAS (version 8.02; SAS Institute, Inc., Cary, NC). Because of the large sample size, *P* values are small.

Results

The original 2-yr national database of all MHD patients included 69,819 cumulative subjects. After deleting MHD patients who did not maintain beyond 3 mo of hemodialysis treatment (5600 patients from the first seven quarters and 5870 patients from the last quarter), 58,349 MHD patients remained for analysis, 291 of whom had missing data. Hence, the resulting cohort included 58,058 MHD patients, 37,049 (64%) of whom originated from the first-quarter data set (q1) and the rest from the subsequent quarters (q2 through q8). Table 1 shows baseline demographic, clinical, and laboratory characteristics of the cohort according to the administration of intravenous iron. Diabetes was more prevalent among patients who received intravenous iron. Almost half of patients who received intravenous iron were among new patients (*i.e.*, dialysis vintage of <6 mo), whereas only one quarter of those who did not receive intravenous iron were new. The BMI was 0.5 kg/m^2 higher whereas serum albumin and creatinine levels were slightly lower in the intravenous iron group. Among indices of iron and anemia, hemoglobin was 0.3 g/dl higher whereas serum ferritin was 162 ng/ml lower among those who received iron. Patients who received intravenous iron also received approximately 5000 units/wk higher dose of EPO.

Table 2 shows correlation coefficients between the three serum markers of iron stores (continuous values) and some clinically relevant variables at the baseline of the cohort. Serum ferritin had only weak to moderate correlations with serum iron and ISAT. Serum iron was positively associated with serum albumin level and TIBC. Blood hemoglobin had negative (inverse) association with serum ferritin but positive (direct) association with serum iron. Table 3 shows the 12 ferritin and 11 ISAT categories. Both all-cause and CV mortality exhibited increasing rates across increasing ferritin categories, whereas the opposite (inverse) association was observed for ISAT increments.

Table 4 and Figure 1 show the hazard ratios of death for time-varying ferritin categories. In unadjusted model, a serum ferritin >800 ng/ml during each quarter was associated with increased death rate, whereas in case-mix adjusted model, a tendency toward increased death rate first was observed when serum ferritin was >1000 ng/ml. However, after additional multivariate adjustment for the confounding effect of surrogates of inflammation and malnutrition, there was no increased death rate for ferritin levels as high as 1200 ng/ml. Figure 2 shows similar death risk analyses for time-varying ISAT categories. In all three models, a serum ISAT between 30 and 50% was associated with the lowest risk for all-cause mortality. Similarly, the risk for CV death was the lowest in the ISAT range of 35 to 50%. Figure 3 shows hazard ratios of death for time-varying serum iron categories. A moderately high serum iron between 60 and 100 μg/ml was associated with significantly better survival, whereas a serum iron <50 μg/ml displayed increased all-cause and CV death risk. Higher serum iron levels in the range of 100 to 120 μg/ml also tended to be associated with better survival, and levels even higher than 120 μg/ml still did not show any meaningful increase in risk for death when compared with the low serum iron levels <60 μg/ml. Figure 4 shows the associations between administered doses of intravenous iron and estimated relative risk for death. Compared with patients who did not receive any intravenous iron, administered intravenous iron up to 400 mg/mo was associated with lower relative risk for death, whereas doses >400 mg/mo correlated with increased death risk. To explore further the association between administering any intravenous iron (yes/no) and 2-yr survival and to explore possible interactions, we performed subgroup analysis using time-varying Cox model (Figure 5). The survival advantages of administer-

Table 2. Bivariate (unadjusted) correlation coefficients between iron markers and relevant variables at baseline in 58,058 MHD patients[a]

Variable	Serum Ferritin	Serum ISAT	Serum Iron
Serum ISAT	0.24		
iron	0.13	0.82	
albumin	−0.01	0.09	0.27
TIBC	−0.28	−0.18	0.30
creatinine	0.03	0.10	0.12
bicarbonate	0.05	−0.01	−0.07
phosphorous	−0.05	−0.02	0.02
calcium	0.05	0.00[b]	0.06
Blood hemoglobin	−0.15	0.11	0.18
WBC	0.09	−0.16	−0.17
lymphocyte (%)	−0.06	0.20	0.23
Age	0.06	−0.07	−0.10
Dialysis vintage	0.06	−0.07	−0.10
Kt/V	0.11	0.09	0.08
BMI	−0.07	−0.07	−0.01[b]
IV iron dose (composite)	−0.06	−0.09	−0.07
EPO dose	−0.03	−0.13	−0.17

[a]ISAT, iron saturation ratio.
[b]$P > 0.05$; all other $P < 0.001$.

Table 3. Selected serum ferritin and ISAT groups and 2-yr mortality among 58,058 MHD patients[a]

	Sample Size (%)	All-Cause Death (%)	CV Death (%)
Ferritin categories (ng/ml)			
<50	1909 (4)	299 (16)	138 (7)
50 to 99	2706 (5)	533 (20)	225 (9)
100 to 199 (reference)	5880 (11)	1149 (20)	513 (9)
200 to 299	5556 (11)	1162 (21)	473 (9)
300 to 499	9827 (19)	2261 (23)	935 (10)
500 to 649	6214 (12)	1548 (25)	700 (12)
650 to 799	5313 (10)	1401 (26)	630 (12)
800 to 999	5580 (11)	1540 (28)	686 (13)
1000 to 1199	3532 (7)	1053 (30)	458 (14)
1200 to 1499	3355 (6)	1060 (32)	475 (15)
1500 to 1999	1634 (3)	583 (36)	258 (17)
≥2000	601 (1)	249 (41)	92 (17)
ISAT categories (%)			
<15	353 (1)	122 (35)	59 (18)
15 to 19.9	2061 (4)	671 (33)	261 (14)
20 to 24.9	6019 (11)	1832 (30)	797 (14)
25 to 29.9	10,719 (20)	2950 (28)	1270 (12)
30 to 34.9	11,078 (21)	2686 (24)	1224 (12)
35 to 39.9	8737 (16)	1997 (23)	884 (11)
40 to 44.9	5907 (11)	1272 (22)	556 (10)
45 to 49.9 (reference)	3925 (7)	878 (22)	380 (10)
50 to 54.9	2268 (4)	430 (19)	171 (8)
55 to 59.9	1395 (3)	288 (21)	104 (8)
≥60	824 (2)	176 (21)	63 (8)

[a]Note that the denominator for all-cause mortality is the total sample of MHD patients with at least one baseline TSAT value, whereas the denominator of cardiovascular (CV) death is slightly smaller as a result of missing causes of death.

ing any intravenous iron seemed relatively consistent in four subgroups of serum ferritin and ISAT on the basis of cutoff values of 500 ng/ml and 50%, respectively,

Discussion

Examining prospectively collected historical data of a large national database of 58,058 MHD patients in the United States with comprehensive time-varying quarterly laboratory values and medication doses for up to 2 yr, we found that after extensive time-dependent and multivariate adjustment for case mix, administered intravenous iron and EPO doses, and surrogates of nutritional status and inflammation, serum ferritin levels between 200 and 1200 ng/ml, serum iron levels between 60 and 120 μg/ml, and ISAT between 30 and 50% were associated with the lowest all-cause and CV death risks. We also found that compared with those who did not receive intravenous iron, administered intravenous iron up to 400 mg/mo was associated with improved survival. Although patients who received intravenous iron had significantly different demographic, clinical, and laboratory features at baseline, the survival benefits of intravenous iron were relatively consistent in different subgroups of MHD patients, including those who had high ferritin but low ISAT values. Hence, the previously observed associations between moderately high levels of serum ferritin and mortality in unadjusted and case-mix adjusted models (36) seem to be mostly due to the confounding effects of malnutrition and inflammation, because additional adjustments for surrogates of MICS mitigates or even reverses some of these associations.

High serum ferritin levels have been found to be associated with increased hospitalization, and a recent rise in serum ferritin is reported to be an imminent death risk in MHD patients (36). An association between dialysis morbidity, including risk for infection, and iron overload reflected by a high serum ferritin have also been reported (37). However, the foregoing studies had small sample size and did not control extensively for confounders. Hyperferritinemia-associated morbidity may be due to non–iron-related factors. Because serum ferritin is a positive inflammatory marker (10), hyperferritinemia-associated increased risk for infection and death may be a mere epiphenomenon. Thus, considering high ferritin levels as the primary cause of increased mortality in the setting of inflammation or infection and preventing optimal anemia management with intravenous iron for serum ferritin levels >500 ng/ml (17) or 800 ng/ml (16) may be flawed. Indeed, the recommended 800-ng/ml threshold as the upper level for serum ferritin in K/DOQI guidelines was mostly opinion based without adequate evidence (16).

Table 4. Hazard ratio of death for ferritin categories based on time-dependent Cox regression models[a]

	Unadjusted		Case-Mix Adjusted		Case-Mix and MICS Adjusted	
	HR (95% CI)	*P* Value	HR (95% CI)	*P* Value	HR (95% CI)	*P* Value
All to cause death ferritin range						
<50	0.81 (0.70 to 095)	0.009	0.74 (0.63 to 0.87)	<0.001	0.84 (0.72 to 0.99)	0.04
50 to 99	1.02 (0.90 to 1.15)	0.8	0.90 (0.8 to 1.02)	0.09	0.93 (0.82 to 1.05)	0.3
100 to 199 (reference)	1.00	NA	1.00	NA	1.00	NA
200 to 299	0.94 (0.86 to 1.03)	0.20	0.97 (0.89 to 1.07)	0.5	0.94 (0.86 to 1.04)	0.22
300 to 499	0.89 (0.82 to 0.93)	0.005	0.93 (0.86 to 1.00)	0.06	0.87 (0.80 to 0.94)	<0.001
500 to 649	0.93 (0.86 to 1.01)	0.09	1.006 (0.926 to 1.094)	0.9	0.95 (0.87 to 1.03)	0.21
650 to 799	0.93 (0.85 to 1.01)	0.08	0.95 (0.872 to 1.035)	0.24	0.87 (0.80 to 0.95)	0.001
800 to 999	1.13 (1.04 to 1.23)	0.006	0.99 (0.91 to 1.07)	0.7	0.89 (0.82 to 0.97)	0.01
1000 to 1199	1.32 (1.20 to 1.44)	<0.001	1.07 (0.97 to 1.17)	0.18	0.90 (0.82 to 0.99)	0.02
1200 to 1499	1.67 (1.53 to 1.83)	<0.001	1.32 (1.20 to 1.44)	<0.001	1.05 (0.96 to 1.15)	0.3
1500 to 1999	2.31 (2.09 to 2.56)	<0.001	1.59 (1.43 to 1.76)	<0.001	1.13 (1.02 to 1.25)	0.02
≥2000	3.00 (2.65 to 3.39)	<0.001	1.78 (1.57 to 2.01)	<0.001	1.16 (1.02 to 1.32)	0.02
CV death ferritin range						
<50	0.80 (0.63 to 1.01)	0.06	0.73 (0.58 to 0.93)	0.01	0.81 (0.64 to 1.03)	0.09
50 to 99	1.08 (0.90 to 1.30)	0.4	0.97 (0.81 to 1.16)	0.7	1.0 (0.83 to 1.20)	1.0
100 to 199 (reference)	1.00	NA	1.00	NA	1.00	NA
200 to 299	0.96 (0.84 to 1.1)	0.6	0.99 (0.86 to 1.14)	0.9	0.97 (0.84 to 1.11)	0.6
300 to 499	0.89 (0.79 to 1.00)	0.06	0.92 (0.82 to 1.04)	0.1835	0.88 (0.78 to 0.99)	0.04
500 to 649	0.97 (0.86 to 1.10)	0.6	1.05 (0.92 to 1.19)	0.5	1.00 (0.89 to 1.14)	1.0
650 to 799	0.95 (0.84 to 1.08)	0.4	0.99 (0.87 to 1.12)	0.8	0.91 (0.80 to 1.04)	0.15
800 to 999	1.11 (0.98 to 1.27)	0.09	0.98 (0.87 to 1.12)	0.8	0.91 (0.80 to 1.04)	0.16
1000 to 1199	1.26 (1.09 to 1.45)	0.002	1.04 (0.9 to 1.19)	0.6	0.90 (0.78 to 1.03)	0.13
1200 to 1499	1.64 (1.43 to 1.88)	<0.001	1.33 (1.16 to 1.53)	<0.001	1.10 (0.96 to 1.27)	0.18
1500 to 1999	2.09 (1.78 to 2.45)	<0.001	1.50 (1.28 to 1.77)	<0.001	1.13 (0.96 to 1.33)	0.16
≥2000	2.55 (2.09 to 3.12)	<0.001	1.67 (1.36 to 2.04)	<0.001	1.15 (0.93 to 1.41)	0.189

[a]HR, hazards ratio; CI, confidence interval; MICS, malnutrition-inflammation complex syndrome; NA, not applicable.

We found that ISAT in range of 30 to 50% was associated with the best survival. This range is essentially within the recommended guidelines (16,17). However, it is important to note that ISAT is not a measured entity but a mathematical derivation of two other measures (serum iron divided by TIBC). Serum TIBC is a negative acute-phase reactant and a marker of MICS and poor outcome in MHD patients (9,18,36). In iron deficiency, the TIBC level tends to be elevated in individuals without kidney disease, whereas serum iron and TIBC levels change parallel to each other in MHD patients as shown in our study ($r = 0.30$). In addition to uremia, ISAT can be significantly confounded by MICS, because the denominator of ISAT (TIBC) is a nutritional and/or inflammatory marker (38,39). This feature of TIBC as a component of ISAT may explain why in our study a more prominent J-shaped association was observed with ISAT than with serum iron (Figures 2 and 3). Indeed, our found association between ISAT levels >50% and increased death risk may be an artificial finding and a mere reflection of the known association between low TIBC levels (which erroneously increases the calculated ISAT value) and death risk in MHD patients.

Among the three indicators of iron stores, the serum iron concentration, which is usually measured monthly in US dialysis facilities, is used less frequently by clinicians. Similarly, K/DOQI guidelines include inadequate practical recommendations that pertain to the interpretation of serum iron for clinical application (16). Serum iron levels may be decreased in conditions besides iron deficiency; the most frequent is probably the anemia associated with chronic conditions such as uremia and inflammation (40–42). Hypoferremia (*i.e.*, low serum iron; not to be confused with hypoferritinemia) is observed during inflammation, neoplasia, trauma, myocardial infarction, surgery, and stressful conditions (41). Hence, our found association between low serum iron (<50 μg/ml) and increased death risk may be due to inflammation. Although, we showed the same associations between low iron and poor outcome even after adjustment for available surrogates of MICS, such conventional inflammatory markers as CRP were not available in our study. Nevertheless, in a recent study of a prospective 12-mo cohort of 1283 MHD patients, low baseline values of serum iron and ISAT were also associated with higher rates of mortality and hospitalization, and in the subcohort of the same study, these associations were also independent of inflammation as measured by serum CRP (8).

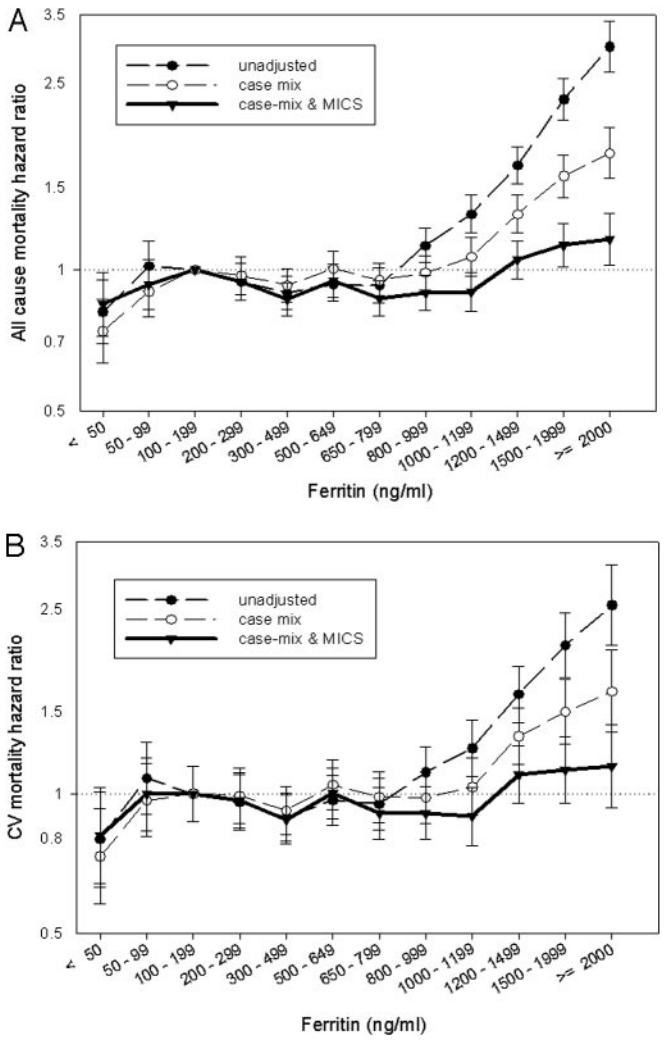

Figure 1. Association between serum ferritin and all-cause (top) and cardiovascular (CV; bottom) mortality.

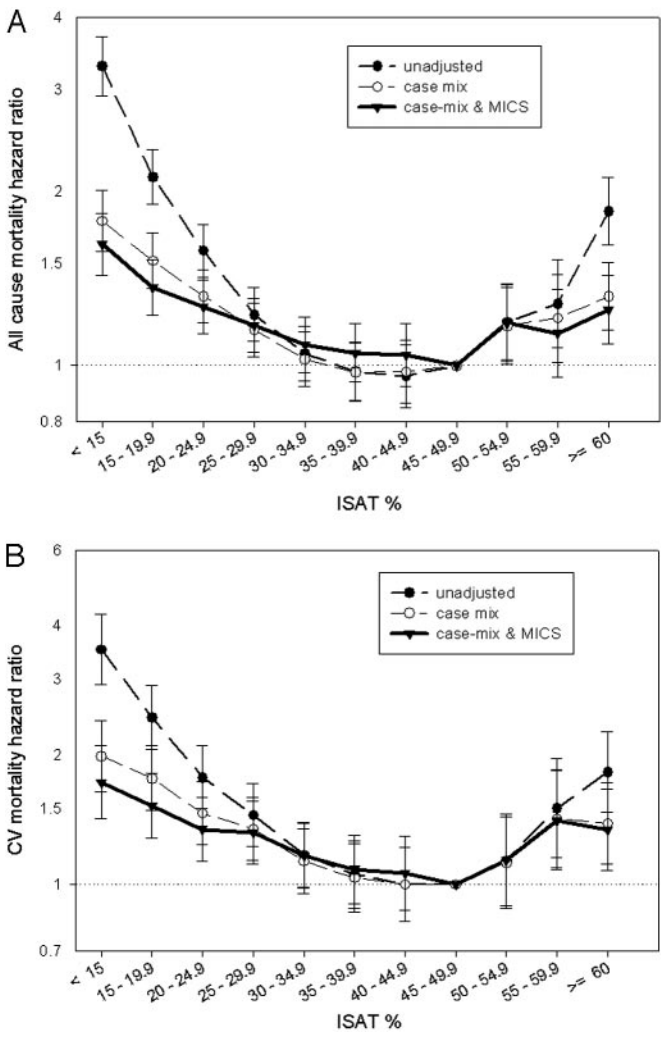

Figure 2. Association between serum iron saturation ratio (ISAT) and all-cause (top) and CV (bottom) mortality.

In our study, we found an unexpected association between moderate doses of intravenous iron (up to 400 mg/mo) and improved survival. This finding seems to be inconsistent with the notion that excessive intravenous iron is deleterious by leading to oxidative stress and predisposition to infection. Such concerns have led judiciously to relatively conservative policies including in K/DOQI guidelines for iron administration to dialysis patients (16). However, most reports concerning adverse effects of iron in dialysis patients are based on *in vitro* studies (43). Very few *in vivo* studies have shown an association between iron administration or higher iron indices and poor outcome in MHD patients, and most of these studies have a small sample sizes and high likelihood of selection bias (44). In another study, by Feldman *et al.* (45), although there was a tendency to increased mortality in MHD patients who had received higher doses of intravenous iron, additional analyses by the same investigators using time-varying marginal structural models did not confirm previous findings (46). A recent study showed that intravenous iron administration to a group of dialysis patients reduced levels of circulating TNF (47). Hence, it is possible that the observed survival advantage of intravenous iron is due to its mitigating effect on inflammation. It is of utmost importance to appreciate that examining associations between a prescribed medication and outcome is amenable to bias by indication (48); such treatments are usually the result of sophisticated processes of active decision making by well-trained and specialized physicians. Many nephrologists may practice according to the K/DOQI or other guidelines. Indeed, we found that MHD patients who did not receive intravenous iron had substantially higher serum ferritin levels, which may indicate that intravenous iron was not prescribed to such high-ferritin patients on the basis of the current guidelines. Although use of such novel epidemiologic tools as marginal structural modeling may mitigate the degree of bias by indication (49), such models were not examined in our study. Hence, the found associations should not be interpreted as causal relationships between the administered intravenous iron and improved survival.

Our study should be qualified because it is observational,

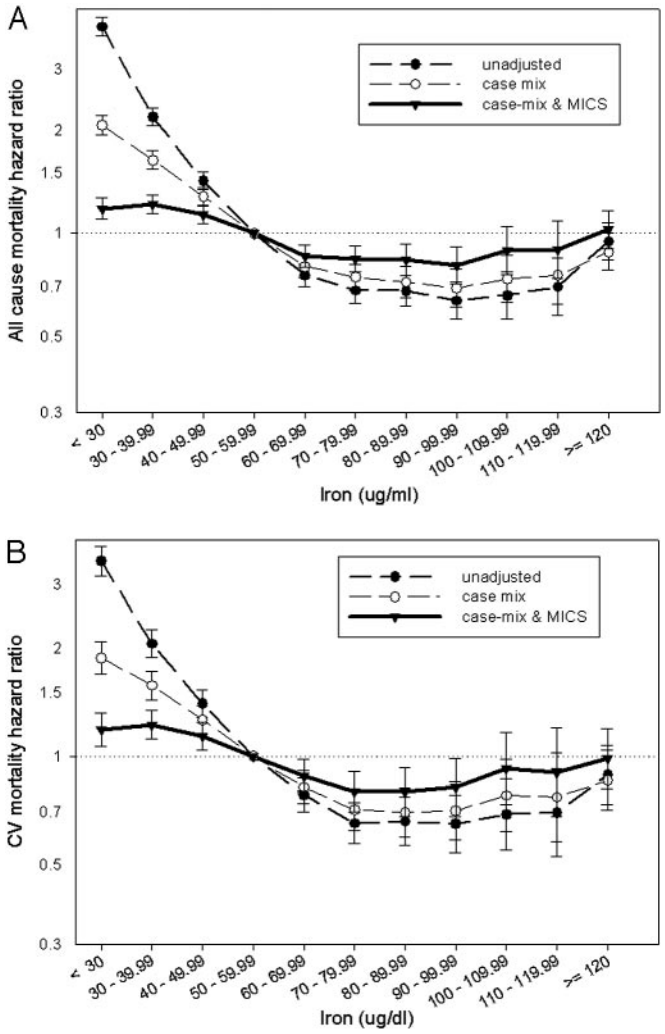

Figure 3. Association between serum iron and all-cause (top) and CV (bottom) mortality.

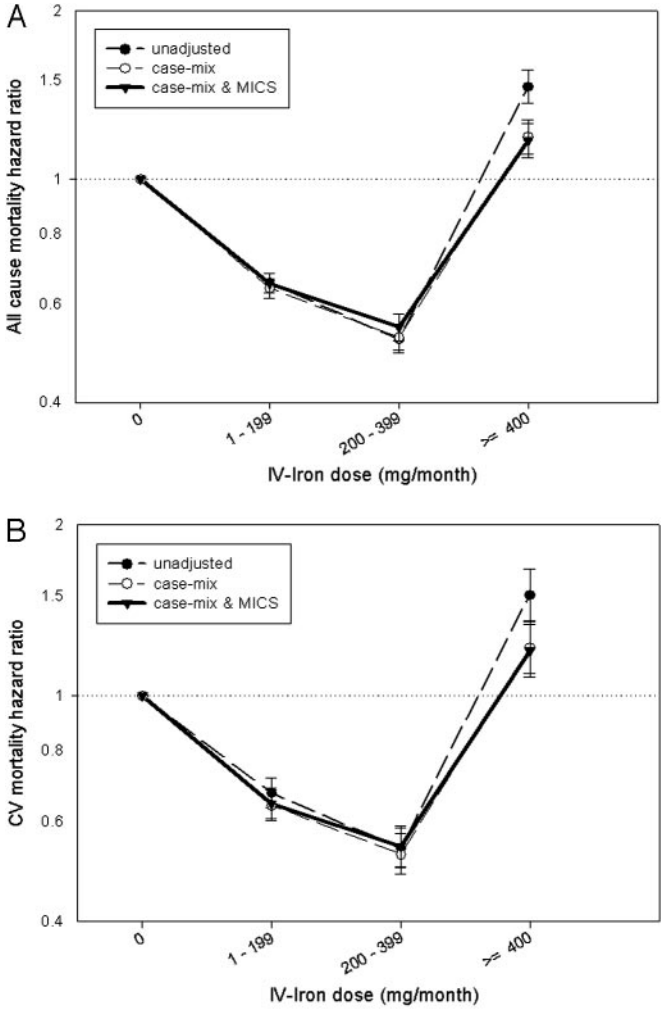

Figure 4. Association between administered intravenous iron and all-cause (top) and CV (bottom) mortality.

rather than interventional, and because a mixed incident/prevalent MHD population was examined. Nevertheless, because essentially all MHD patients of the DaVita dialysis facilities were included in our analyses, the likelihood of selection bias is minimal. Moreover, all dialysis facilities were under uniform administrative care, and all laboratory tests were performed in one single laboratory with optimal quality assurance monitoring. Furthermore, we used 3-mo averaged measures rather than one single measure at baseline, and we adjusted for dialysis vintage in all multivariate models. However, a switch from iron gluconate to iron sucrose in the middle of the cohort might have confounded some associations. Moreover, the actually administered dose may be different from the billed dose, and the discrepancy may differ according to the type of intravenous iron medication used. Hence, caution should be practiced in interpreting our medication dose data. Another limitation of our study is the possible inclusion of cases with gastrointestinal bleeding, other sources of blood loss, or malignancies, which may lead to low serum iron and poor outcome. Moreover, MHD patients with intercurrent infection or systemic inflammatory diseases, in whom inflammation-induced hypoferremia can be observed, were not excluded. However, these cases are not too frequent to cause major confounding, especially because the entire national database was examined and because the laboratory values were 13-wk averaged values.

Although our study is based exclusively on mere observational data and, hence, no cause-effect association can be inferred with certainty, it is consistent with the thesis that moderately high levels of serum ferritin are not *per se* indicators of iron overload and should not be regarded as means to restrict medications for optimal anemia management. A low iron status may be as harmful as, if not more harmful than, the so-called iron overload. Indeed, in the previously reported cases of hemochromatosis and/or hemosiderosis among dialysis patients, the observed serum ferritin levels were well above the currently observed ranges in MHD patients, usually in 3000- to 10,000-ng/ml range (50). With widespread EPO administration to dialysis patients since the early 1990s, there has been much fewer reported, if any, cases of hemochromatosis or hemosiderosis in MHD patients despite rigorous use of intravenous iron (50). Although our results may have major clinical implications,

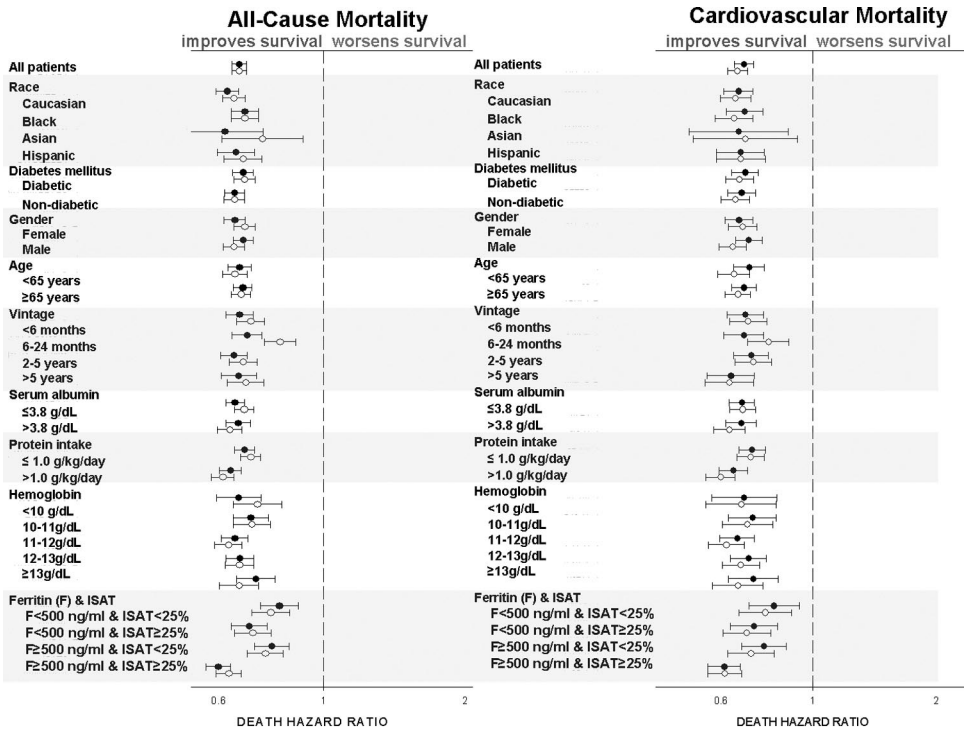

Figure 5. Association between administered intravenous iron and survival in different subgroups of patients on maintenance hemodialysis. Filled circles are based on unadjusted Cox models, whereas empty circles result from multivariate adjusted models. F, ferritin.

the observational nature of our study prompts caution in interpreting and generalizing our findings. Interventional studies including randomized clinical trials are required to ascertain whether (*1*) in MHD patients with a low serum iron and/or moderately high serum ferritin, intravenous iron administration can effectively increase serum iron and (*2*) whether such an interventional increase in serum iron improves clinical outcome in these individuals.

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See related editorial, "Intravenous Iron: How Much Is Too Much?," on pages 2833–2835.