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Criteria for Iron Deficiency in Patients With Heart Failure

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ABSTRACT

BACKGROUND Guidelines on heart failure (HF) define iron deficiency (ID) as a serum ferritin <100 ng/mL or, when 100-299 ng/mL, a transferrin saturation (TSAT) <20%. Inflammation (common in HF) may hinder interpretation of serum ferritin.

OBJECTIVES This study sought to investigate how different definitions of ID affect its prevalence and relationship to prognosis in ambulatory patients with chronic HF.

METHODS Prevalence, relationship with patients' characteristics, and outcomes of various ID definitions were evaluated among patients with HF referred to a regional clinic (Hull LifeLab) from 2001 to 2019.

RESULTS Of 4,422 patients with HF (median age 75 years [range: 68-82 years], 60% men, 32% with reduced left ventricular ejection fraction), 46% had TSAT <20%, 48% had serum iron ≤13 μmol/L, 57% had serum ferritin <100 ng/mL, and 68% fulfilled current guideline criteria for ID, of whom 35% had a TSAT >20%. Irrespective of definition, ID was more common in women and those with more severe symptoms, anemia, or preserved ejection fraction. TSAT <20% and serum iron ≤13 μmol/L, but not guideline criteria, were associated with higher 5-year mortality (HR: 1.27; 95% CI: 1.14-1.43; $P < 0.001$; and HR: 1.37; 95% CI: 1.22-1.54; $P < 0.001$, respectively). Serum ferritin <100 ng/mL tended to be associated with lower mortality (HR: 0.91; 95% CI: 0.81-1.01; $P = 0.09$).

CONCLUSIONS Different definitions of ID provide discordant results for prevalence and prognosis. Definitions lacking specificity may attenuate the benefits of intravenous iron observed in trials while definitions lacking sensitivity may exclude patients who should receive intravenous iron. Prespecified subgroup analyses of ongoing randomized trials should address this issue. (J Am Coll Cardiol 2022;79:341-351) © 2022 The Authors. Published by Elsevier on behalf of the American College of Cardiology Foundation. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/).

Iron deficiency (ID), inferred from the results of blood tests, is common in chronic heart failure (HF) and, in the presence or absence of anemia, is associated with poorer quality of life, exercise capacity, and prognosis.[1-4] Many definitions of ID have been proposed, but consensus is lacking on which should be used in clinical practice for patients with HF.

The World Health Organization defines ID as a serum ferritin <15 ng/mL, and most clinical laboratories define ID as <30 ng/mL. However, international guidelines on HF define ID as a serum ferritin <100 ng/mL or, when ferritin is 100-299 ng/mL, a transferrin saturation (TSAT) <20%.[5,6] These criteria were based on a consensus of opinion mainly among nephrologists[7] and on the selection criteria for successful clinical trials of intravenous (IV) iron in HF, such as the FAIR-HF (Ferric Carboxymaltose in Patients with Heart Failure and Iron Deficiency) trial.[8] However, a definition based primarily on

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ABBREVIATIONS AND ACRONYMS

CV = cardiovascular

HF = heart failure

HFpEF = heart failure with preserved ejection fraction

HFrEF = heart failure with reduced ejection fraction

ID = iron deficiency

IV = intravenous

Ln = natural logarithm

LVEF = left ventricular ejection fraction

LVSD = left ventricular systolic dysfunction

NT-proBNP = N-terminal pro-B-type natriuretic peptide

SqR = square root

TSAT = transferrin saturation

ferritin has several limitations. Most ferritin resides in cells where it binds to iron to prevent free radical production. Any cell damage, including activation of inflammatory pathways, may cause ferritin to be released; an increase in serum ferritin may occur even in the presence of ID.[9] Bone marrow biopsy, the gold-standard for diagnosing ID, may demonstrate ID even when ferritin is high.[10] Observational studies suggest that serum iron concentration and TSAT might both be more strongly associated with prognosis than serum ferritin and might be a better guide to which patients benefit from IV iron.[10-14]

Accordingly, we investigated the prevalence, associations, and prognostic significance of ID using diverse criteria in a large cohort of patients attending an HF clinic.

METHODS

STUDY POPULATION. Patients referred between December 2001 and June 2019 with suspected or confirmed HF to a regional HF clinic (the Hull LifeLab), serving a population of around 550,000 inhabitants, were included. All patients gave written informed consent for their data to be electronically stored and used for research. Demographic data, medical history, symptoms and signs, and an electrocardiogram and echocardiogram were recorded. Blood samples were obtained for hematological and biochemical tests, including N-terminal pro-B-type natriuretic peptide (NT-proBNP), serum iron, TSAT, and ferritin. The study was approved by the Hull and East Yorkshire Local Research Ethics Committee.

SEE PAGE 352

DEFINITIONS. HF was defined by the presence of symptoms and signs of HF and either 1) a left ventricular ejection fraction (LVEF) ≤40% (HF with reduced EF [HFrEF] or moderate or severe LV systolic dysfunction [LVSD]) or 2) a plasma NT-proBNP ≥125 ng/L. Those with an NT-proBNP ≥125 ng/L and an LVEF >40% were further classified as those with HF with an LVEF in the mid-range (HF with mid-range EF: LVEF 40%-49% or mild or mild-moderate LVSD) or preserved (HF with preserved EF [HFpEF]: LVEF ≥50% or trivial or no LVSD). If NT-proBNP was high but information on LV function was not available, patients were grouped as HF-↑NT-proBNP. Patients with an LVEF >40% who had no available measurement of NT-proBNP or an NT-proBNP<125 ng/L or lacked echocardiographic data were excluded from this analysis.

Anemia was defined according to World Health Organization criteria as hemoglobin <12.0 g/dL in women and <13.0 g/dL in men.[15] Serum ferritin, TSAT, and serum iron were used as biomarkers of ID. Serum concentrations of ferritin and iron were measured and TSAT (%) calculated using the formula: [iron (μmol/L) / (transferrin [g/L] × 25.2) × 100].[16] Only patients with all these iron biomarkers and hemoglobin values available were included in this analysis. Patients were followed up clinically and by electronic records until June 3, 2019. The cause of death was adjudicated based on available clinical and electronic records, following a protocol described elsewhere.[17]

STATISTICAL ANALYSIS. Continuous variables are presented as median with 25th and 75th percentile and compared using 1-way analysis of variance or Kruskal-Wallis test. Categorical variables are presented as numbers and percentages and compared using chi-square tests. Pearson's or Spearman's ρ correlations were used, for parametric and nonparametric variables, respectively. Natural logarithmic (Ln) or square root (SqR) transformation were used for non-normally distributed variables. No imputation was performed for missing data. Variables with a high percentage of missing values were not included in the multivariable analysis to reduce uncertainty.

Cox proportional hazards models were used to identify variables associated with 5-year all-cause and cardiovascular (CV) mortality. For guideline ID definition, the Fine-Gray subdistribution hazard model for CV mortality was used, considering non-CV death as a competing event. Restricted cubic splines were constructed for each continuous biomarker. Univariable interaction analyses for HFrEF vs HF with mid-range EF, HFpEF, and HF-↑NT-proBNP were done. Multivariable models were built including only those variables associated with outcome ($P \leq 0.1$) in univariable analysis. Definitions of ID associated with outcome with a P value >0.10 in univariable analysis were not tested in the multivariable model. HRs with 95% CIs are reported. Kaplan-Meier cumulative mortality curves for all-cause death within 5 years were used to compare mortality among patients grouped by different ID definitions. Differences between groups were evaluated using the log-rank test.

Iron biomarkers were tested both as continuous and as categorical variables. As categorical variables, different thresholds were used. We used: 1) international guideline criteria (ferritin <100 ng/mL or TSAT <20% if ferritin 100-299 ng/mL); 2) ferritin <100 ng/mL; 3) TSAT <20%; and 4) serum iron ≤13 μmol/L. Serum iron ≤13 μmol/L was examined based on the results of a study using bone marrow iron staining as a gold-standard in patients

TABLE 1 Characteristics of Patients According to HF Phenotypes

	Missing	HFrEF (n = 1,429 [32%])	HFmrEF (n = 820 [19%])	HFpEF (n = 1,832 [41%])	HF ↑NT-proBNP (n = 341 [8%])	*P* Value
Demographics and comorbidities						
Age, y	0 (0)	72 (64-79)	76 (68-82)	77 (71-83)	75 (68-83)	<0.001
Female	0 (0)	376 (26)	262 (32)	978 (53)	147 (43)	<0.001
BMI, kg/m^2	85 (2)	27 (24-31)	28 (25-33)	29 (25-33)	28 (24-33)	<0.001
IHD	0 (0)	812 (57)	465 (57)	494 (27)	105 (31)	<0.001
Hypertension	0 (0)	602 (42)	460 (56)	1,222 (67)	171 (50)	<0.001
Diabetes	0 (0)	351 (25)	222 (27)	506 (28)	73 (21)	0.04
COPD	11 (<1)	134 (9)	61 (8)	176 (10)	40 (12)	0.10
eGFR, mL/min/1.73 m^2	74 (2)	59 (44-74)	61 (46-74)	60 (45-76)	59 (44-77)	0.45
Atrial fibrillation/flutter	151 (3)	366 (27)	303 (38)	714 (39)	124 (44)	<0.001
Signs and symptoms						
NYHA functional class III/IV	57 (1)	533 (37)	255 (31)	446 (25)	86 (27)	<0.001
Edema (≥ankle)	355 (8)	322 (25)	227 (30)	576 (34)	93 (34)	<0.001
Laboratory						
Serum iron, μmol/L	0 (0)	14 (10-19)	14 (10-18)	13 (10-17)	13 (10-18)	<0.001
Serum iron ≤13 μmol/L	0 (0)	635 (44)	377 (46)	918 (50)	171 (50)	<0.01
TSAT, %	0 (0)	22 (16-30)	22 (16-29)	21 (15-26)	21 (15-28)	<0.001
TSAT <20%	0 (0)	621 (44)	361 (44)	890 (49)	159 (47)	0.02
Ferritin, ng/mL	0 (0)	102 (54-184)	94 (46-171)	71 (38-135)	78 (38-158)	<0.001
Ferritin <100 ng/mL	0 (0)	697 (49)	432 (53)	1,175 (64)	202 (59)	<0.001
Guideline ID criteria	0 (0)	872 (61)	534 (65)	1,373 (75)	232 (68)	<0.001
Hemoglobin, g/dl	0 (0)	13.4 (12.2-14.6)	13.3 (12.0-14.5)	12.9 (11.8-14.1)	12.7 (11.8-13.8)	<0.001
Anemia	0 (0)	473 (33)	267 (33)	670 (37)	136 (40)	0.02
NT-proBNP, ng/L	152 (3)	1,935 (841-4,333)	1,164 (497-2,587)	865 (332-1,825)	1,329 (490-2,766)	<0.001
hs-CRP, mg/L	238 (5)	4.2 (1.7-8.8)	4.0 (1.7-8.5)	3.9 (1.6-8.4)	4.2 (1.8-11.0)	0.22
Medications						
Loop diuretic	73 (2)	1,104 (77)	532 (65)	977 (54)	212 (74)	<0.001
ACE inhibitor or ARB	73 (2)	1,161 (81)	608 (75)	1,063 (58)	196 (68)	<0.001
MRA	73 (2)	547 (38)	176 (22)	172 (9)	89 (31)	<0.001
BB	73 (2)	990 (69)	567 (70)	957 (53)	214 (74)	<0.001
Anticoagulant	0 (0)	429 (30)	261 (32)	528 (29)	120 (35)	0.08
Antiplatelet	0 (0)	707 (50)	399 (49)	722 (39)	100 (29)	<0.001

Values are n (%) or median (IQR). Guideline ID criteria were ferritin <100 ng/mL or TSAT <20% if ferritin was 100-299 ng/mL.

ACE = angiotensin-converting enzyme; ARB = angiotensin receptor blocker; BB = beta-blocker; BMI = body mass index; COPD = chronic obstructive pulmonary disease; eGFR = estimated glomerular filtration rate; HFmrEF = heart failure with mid-range ejection fraction; HFpEF = heart failure with preserved ejection fraction; HFrEF = heart failure with reduced ejection fraction; hs-CRP = high-sensitivity C-reactive protein; ID = iron deficiency; IHD = ischemic heart disease; MRA = mineralocorticoid receptor antagonist; NT-proBNP = N-terminal pro-B-type natriuretic peptide; NYHA = New York Heart Association; TSAT = transferrin saturation.

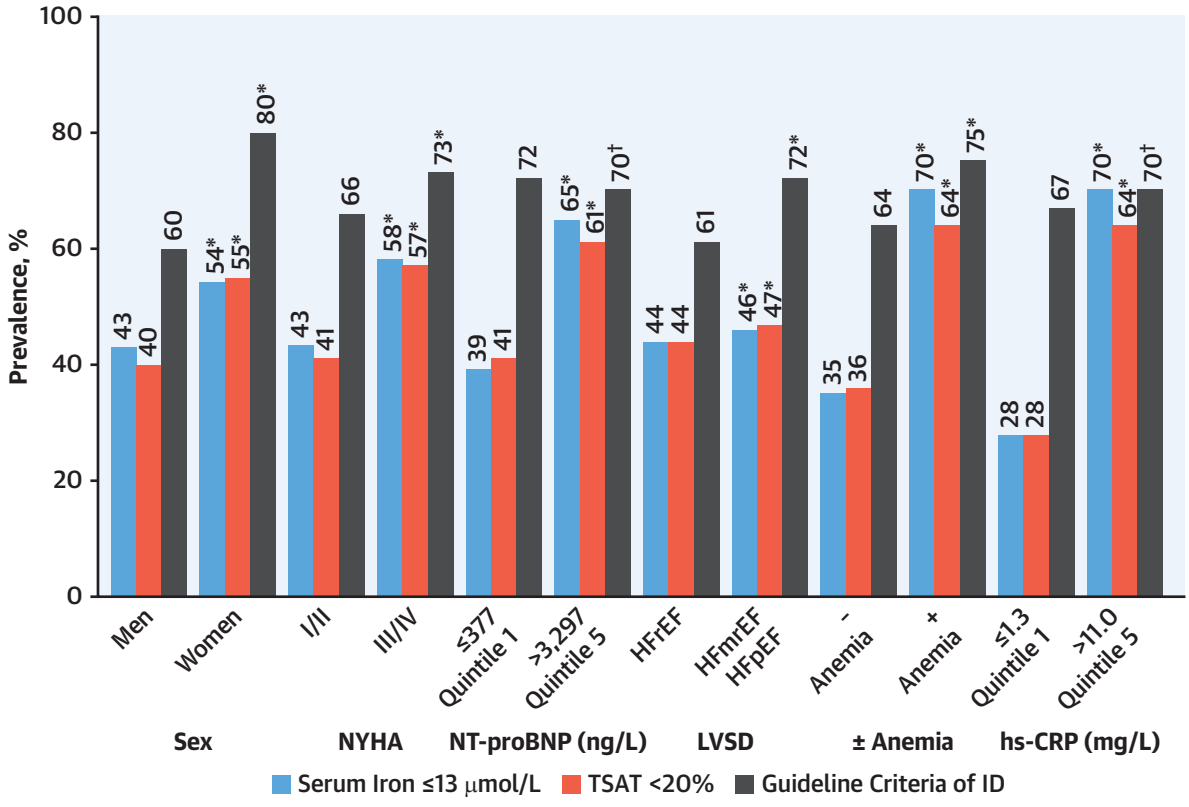
with HF.[10] We also explored prevalence and outcomes of additional thresholds (ferritin <30 ng/mL, ferritin <100 ng/mL, ferritin <300 ng/mL, serum iron ≤10 μmol/L, and TSAT <30%).

The HR for all-cause mortality for each decile of serum ferritin, iron, and TSAT was determined by univariable Cox proportional model. Receiver-operating characteristic analysis was also performed for each iron biomarker to determine the best cutoff values to predict 1-year all-cause mortality, excluding patients with ≤12 months follow-up. The optimal threshold for prediction was defined as the value on the receiver-operating characteristic curve closest to the upper left corner: $d^2 = (1 - \text{sensitivity})^2 + (1 - \text{specificity})^2$. *P* values and 95% CIs presented in this report have not been adjusted for multiplicity; therefore, inferences drawn from these statistics may not be reproducible. All analyses were performed with SPSS statistical software, version 27 (IBM) and STATA statistical software, version 17 (StataCorp). The 2-tailed level of statistical significance was set at $P < 0.05$.

RESULTS

Of the 9,321 patients evaluated in the clinic, 7,160 were considered to have HF. Of these, 4,422 (62%) had all the required iron indices (Supplemental Figure 1). Patients with HF enrolled after January 1, 2009 (n = 4,182), subsequent to a revision of the clinical pathway, were more likely to have all the required iron indices (n = 2,959 [71%]). Compared with those with HFrEF, patients with HFpEF were

FIGURE 1 Prevalence of ID by Definition and Subgroup



Prevalence (%) of serum iron ≤13 μmol/L, transferrin saturation (TSAT) <20%, and guideline criteria for iron deficiency (ID) according to patients' subgroups. *Significant difference ($P < 0.05$) between patients subgroups; †P = NS. HFmrEF = heart failure with mid-range ejection fraction; HFpEF = heart failure with preserved ejection fraction; HFrEF = heart failure with reduced ejection fraction; hs-CRP = high-sensitivity C-reactive protein; LVSD = left ventricular systolic dysfunction; NT-proBNP = N-terminal pro-B-type natriuretic peptide; NYHA = New York Heart Association functional class.

older, were more likely to be women, and were more likely to have hypertension and atrial fibrillation, but despite these differences, they had a lower median plasma NT-proBNP and similar renal function.

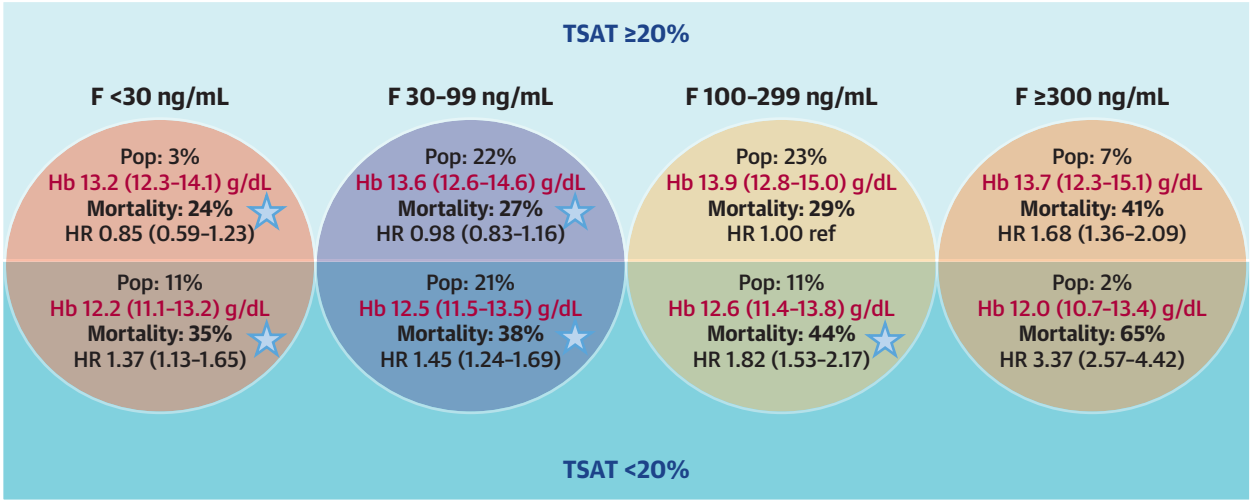
Serum iron and TSAT were highly correlated (r = 0.92, $P < 0.001$); correlations between ferritin and serum iron (r = 0.27, $P < 0.001$) or TSAT (r = 0.41, $P < 0.001$) were weaker.

PREVALENCE OF ID AND ANEMIA AND ASSOCIATION WITH PATIENT CHARACTERISTICS. The prevalence of ID ranged from 44% to 68% depending on the definition (Table 1). Of 3,011 patients who met guideline criteria for ID, 2,506 (83%) had a ferritin <100 ng/mL. Many patients fulfilled 1 definition of ID but not others (Supplemental Figure 2). Of 3,011 patients with ID according to guideline criteria, 32% had a TSAT ≥20% and a serum iron >13 μmol/L, while of those who did not have ID according to guideline criteria, 20% (n = 282) had a TSAT <20% or a serum iron ≤13 μmol/L.

Figure 1 shows the prevalence of ID defined by TSAT, serum iron, and guideline criteria in various patient subgroups. Using any of these definitions, ID was more common in women, in those with more severe symptoms, and in those who did not have HFrEF. Compared with those in the lowest quintile, those in the highest quintiles of NT-proBNP and high-sensitivity C-reactive protein were more likely to have a low serum iron or TSAT but not ID using guideline criteria. Patients with HFpEF were more likely to have a low serum iron, TSAT, and ferritin compared with those with other HF phenotypes (Table 1, Supplemental Figure 3).

Anemia was present in 1,543 (35%) patients. Compared with those without anemia, those with anemia had a higher prevalence of ID, irrespective of

FIGURE 2 Hb and Mortality by Serum Ferritin and TSAT



Percentage of patients (Pop), median and first to third quartile of hemoglobin (Hb), mortality at 5 years (Mortality), and adjusted HR for 5-year mortality (95% CI) according to serum ferritin (F) (<30 ng/mL, 30-99 ng/mL, 100-299 ng/mL, and ≥300 ng/mL) and transferrin saturation (TSAT) (<20% and ≥20%). Model adjusted for age (/5), sex, and heart failure phenotype. **Stars** represent those patients who fulfill current guideline criteria for iron deficiency. Ref = reference group.

the ID criteria used (Figure 1). Figure 2 shows the hemoglobin concentration (median [25th and 75th percentile]) and 5-year mortality according to TSAT above or below 20% and by different concentrations of ferritin (<30 ng/mL, 30-99 ng/mL, 100-299 ng/mL, and ≥300 ng/mL).

Compared with those with a higher TSAT, those with a TSAT <20%, had a lower hemoglobin concentration, which was similar across the range of serum ferritin. Patients with a TSAT <20% and serum ferritin ≥300 ng/mL had the highest prevalence of anemia. These patients were more likely to be men, had lower body mass index, had higher median NT-proBNP and high-sensitivity C-reactive protein, and had worse renal function compared with those with ferritin <300 ng/mL (Supplemental Table 1). The guideline criteria for ID were met by 3,011 (68%) patients, but 1,079 (36%) of them had a TSAT >20% (Figure 2).

OUTCOMES BY DIFFERENT ID CRITERIA. The median duration of follow-up was 49 (25th and 75th percentile: 18-89) months. In total, 2,321 (52.5%) patients died. The 5-year mortality was 34.5%. Mortality was lowest for those with a serum ferritin <100 ng/mL and a TSAT >20% and highest for those with a serum ferritin >100 ng/mL with a TSAT <20% (Figure 2). In univariable analysis, lower TSAT and serum iron, but higher serum ferritin, were associated with a higher all-cause and CV mortality (Table 2, Figure 3). In a multivariable analysis that included demographic factors, comorbidities, findings on physical examination, electrocardiography, echocardiography, and biochemical testing (Supplemental Tables 2 and 3), a higher SqR[serum iron] and Ln[TSAT] were associated with a lower all-cause mortality (HR: 0.84; 95% CI: 0.78-0.91; $P < 0.001$; and HR: 0.83; 95% CI: 0.74-0.92; $P < 0.01$, respectively), while a higher Ln[ferritin] was associated with a higher all-cause and CV mortality (HR: 1.09; 95% CI: 1.02-1.16; $P < 0.01$; and HR: 1.11; 95% CI: 1.02-1.20; $P = 0.02$, respectively) (Table 2). After including hemoglobin in the models, SqR[serum iron] and Ln[TSAT] were no longer associated with all-cause mortality, but the associations with Ln[ferritin] did not change (Supplemental Table 4).

As categorical variables, TSAT <20% (vs ≥20%) and serum iron ≤13 μmol/L (vs >13 μmol/L) were associated with a higher all-cause and CV mortality in the univariable model (Table 2), while a ferritin <100 ng/mL (vs ≥100 ng/mL) was associated with a better survival ($P < 0.01$ for all-cause and CV mortality). ID defined by guideline criteria was not associated with either all-cause or CV-mortality ($P = 0.16$ and $P = 0.98$, respectively) (Table 2, Figure 4).

In a multivariable analysis, TSAT <20% and serum iron ≤13 μmol/L were independently associated with greater all-cause mortality (HR: 1.27; 95% CI: 1.14-1.43; $P < 0.001$; and HR: 1.37 95% CI: 1.22-1.54; $P < 0.001$, respectively) but not with CV mortality. Ferritin <100 ng/mL was associated with lower CV

TABLE 2 Cox Regression Model for 5-Year Mortality

	All-Cause Mortality					Cardiovascular Mortality				
	Univariable Model			Multivariable Model[a]		Univariable Model			Multivariable Model[a]	
	HR (95% CI)	*P* Value	*P* for Interaction HFrEF vs Non-HFrEF	HR (95% CI)	*P* Value	HR (95% CI)	*P* Value	*P* for Interaction HFrEF vs Non-HFrEF	HR (95% CI)	*P* Value
Serum iron biomarkers										
SqR[serum iron], μmol/L	0.66 (0.62-0.71)	<0.001	0.09	0.84 (0.78-0.91)	<0.001	0.73 (0.67-0.80)	<0.001	0.08	0.97 (0.87-1.07)	0.50
Ln[TSAT], %	0.63 (0.58-0.70)	<0.001	0.45	0.83 (0.74-0.92)	<0.01	0.68 (0.60-0.78)	<0.001	0.61	0.93 (0.80-1.09)	0.36
Ln[ferritin], ng/mL	1.11 (1.05-1.17)	<0.001	0.23	1.09 (1.02-1.16)	<0.01	1.17 (1.09-1.26)	<0.001	0.06	1.11 (1.02-1.20)	0.02
ID definitions										
Iron ≤10 μmol/L	1.81 (1.63-2.01)	<0.001	0.01	1.32 (1.18-1.49)	<0.001	1.71 (1.48-1.97)	<0.001	0.02	1.17 (0.99-1.38)	0.054
Iron ≤13 μmol/L	1.81 (1.63-2.00)	<0.001	0.08	1.37 (1.22-1.54)	<0.001	1.55 (1.35-1.78)	<0.001	0.18	1.11 (0.95-1.30)	0.18
TSAT <20%	1.56 (1.41-1.72)	<0.001	0.09	1.27 (1.14-1.43)	<0.001	1.41 (1.22-1.61)	<0.001	0.61	1.06 (0.93-1.27)	0.29
TSAT <30%	1.39 (1.22-1.59)	<0.001	0.49	1.10 (0.95-1.28)	0.20	1.20 (1.01-1.43)	0.04	0.22	0.95 (0.78-1.16)	0.60
Ferritin <30 ng/mL	0.97 (0.84-1.12)	0.66	0.54	0.96 (0.85-1.17)	0.96	0.88 (0.72-1.09)	0.24	0.64	0.98 (0.78-1.23)	0.88
Ferritin <100 ng/mL	0.89 (0.81-0.99)	0.03	0.02	0.91 (0.81-1.01)	0.09	0.78 (0.68-0.89)	<0.001	<0.01	0.83 (0.71-0.96)	0.02
Ferritin <300 ng/mL	0.65 (0.56-0.76)	<0.001	0.20	0.69 (0.58-0.81)	<0.001	0.67 (0.54-0.83)	<0.001	0.26	0.78 (0.61-0.99)	0.048
Guideline ID criteria	1.08 (0.97-1.20)	0.16	0.07	1.02 (0.90-1.15)	0.75	1.00 (0.87-1.16)	0.98	0.02	0.96 (0.81-1.14)	0.66

Guideline ID criteria were ferritin <100 ng/mL or TSAT <20% if ferritin was 100-299 ng/mL. [a]Model adjusted for age (/5), sex, body mass index (/5), systolic blood pressure (/5), ischemic heart disease, diabetes, New York Heart Association functional class III or IV, heart rate (/5), atrial fibrillation or atrial flutter, HF phenotype (HFrEF vs HFmrEF, HFrFF vs HFpEF, HFrEF vs HF ↑NT-proBNP), Ln[NT-proBNP], and eGFR (/5). SqR[serum iron], Ln[TSAT], and Ln[ferritin] were included one at a time. No corrections for multiple testing were applied.

Ln = natural logarithmic; SqR = square root; other abbreviations as in Table 1.

mortality and tended to be associated with lower all-cause mortality. Ferritin <300 ng/mL was associated with both lower all-cause and CV mortality (**Table 2**).

Figure 5 and Supplemental Figures 4 and 5 show the Kaplan-Meier curves for all-cause mortality according to prespecified thresholds of each biomarker with their relative HRs while Supplemental Figure 6 shows the unadjusted HRs for all-cause death for each decile of TSAT, serum iron, and ferritin. Compared with the reference range (38-50 ng/mL), the highest decile of ferritin (>274 ng/mL) was associated with a higher mortality (HR: 1.67; 95% CI: 1.34-2.08; $P < 0.001$). Mortality was higher for patients in deciles of TSAT <19.2% and serum iron <13 μmol/L.

No significant interaction was found in the adjusted models between definitions of ID and HF phenotypes for all-cause or cardiovascular mortality.

Patients with a serum ferritin ≥300 ng/mL and a TSAT <20%, who do not fulfill guideline criteria for ID, had the highest risk of death, while those with a ferritin <100 ng/mL with a TSAT ≥20%, who do fulfill HF guideline criteria for ID, had a similar prognosis to those with ferritin 100-299 ng/mL and TSAT ≥20% (**Figure 2**, Supplemental Figure 7).

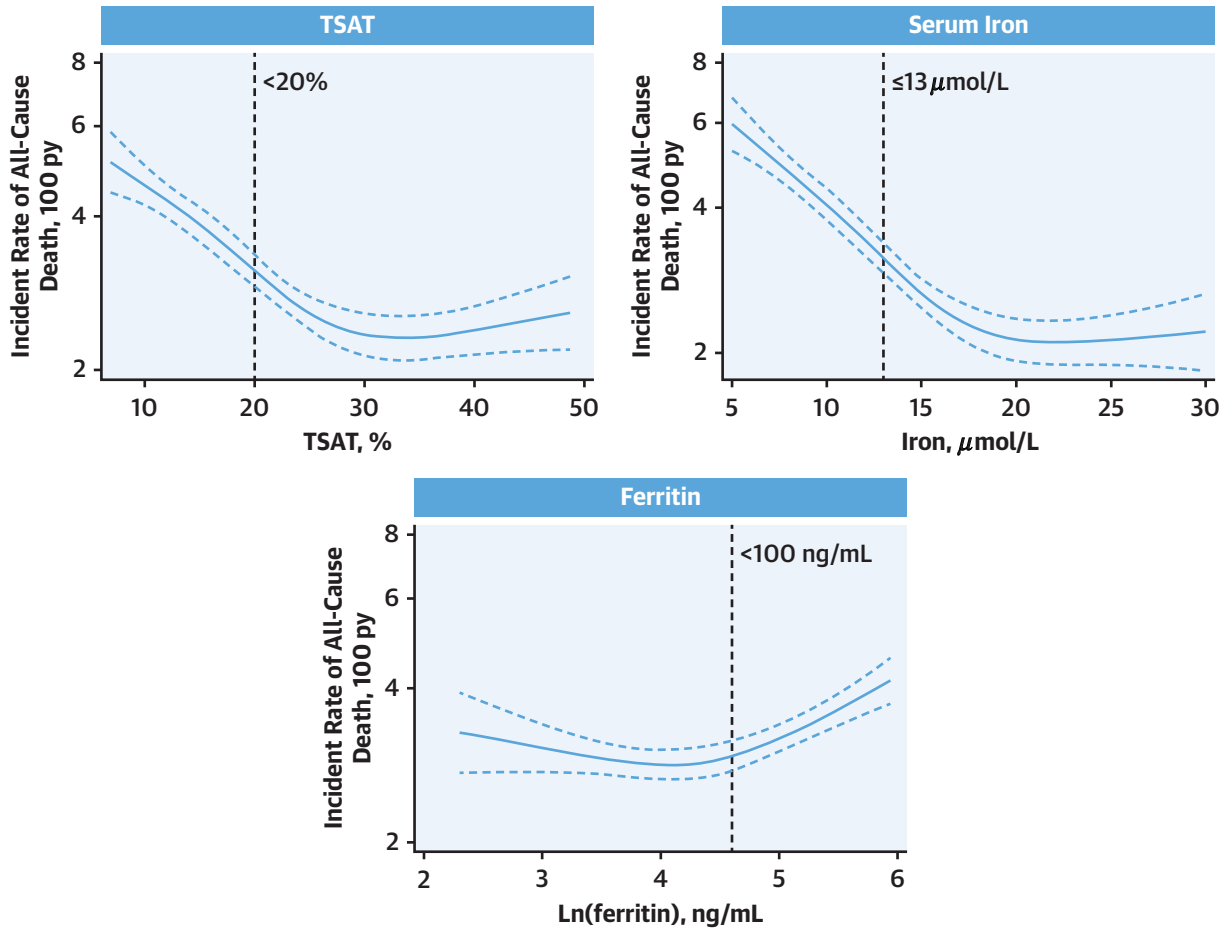
The area under the curve for all-cause mortality at 1 year for serum iron and TSAT were 0.64 (95% CI: 0.61-0.67) and 0.61 (95% CI: 0.58-0.64) with optimal predictive values of <12.5 μmol/L and <19.0%, respectively. For ferritin, area under the curve was 0.56 (95% CI: 0.53-0.59) and the optimal cutoff was >143.5 ng/mL (Supplemental Figure 8).

DISCUSSION

Getting the definition of ID right is important for clinical trials and clinical practice. We found that ID is common among ambulatory patients with HF, but the prognostic implications differ according to definition. We did not find any association between the current guideline definition of ID and mortality; indeed, lower serum ferritin concentrations were associated with a better survival (**Central Illustration**). Among other definitions of iron deficiency, TSAT <20% and serum iron ≤13 μmol/L were independently associated with death, with no interaction between HF phenotypes. About two-thirds of patients fulfilled the guideline definition of ID but of these, about one-third had a TSAT >20%. If this latter group of patients does not truly have ID but is being included in trials of IV iron, this could attenuate any observed benefit and might even lead to a neutral result.

In the human body, iron is central to hematopoietic (erythropoiesis, oxygen transport, and storage) and nonhematopoietic processes (substrate utilization and mitochondrial energy production).[18] As a result, iron deficiency can lead to reduced oxygen delivery and impaired oxygen utilization,[19,20] both of which can contribute to breathlessness and reduced exercise capacity. Suggested mechanisms for the development of ID in patients with HF are many and include reduced intake, reduced absorption of dietary iron secondary to increased secretion of hepcidin, sequestration of iron within the reticuloendothelial

FIGURE 3 Association Between ID Biomarkers and All-Cause Mortality



Restricted cubic splines showing the association between serum iron, TSAT, and ferritin and risk of all-cause mortality. Ln = natural logarithm; py = person-years; other abbreviations as in Figure 1

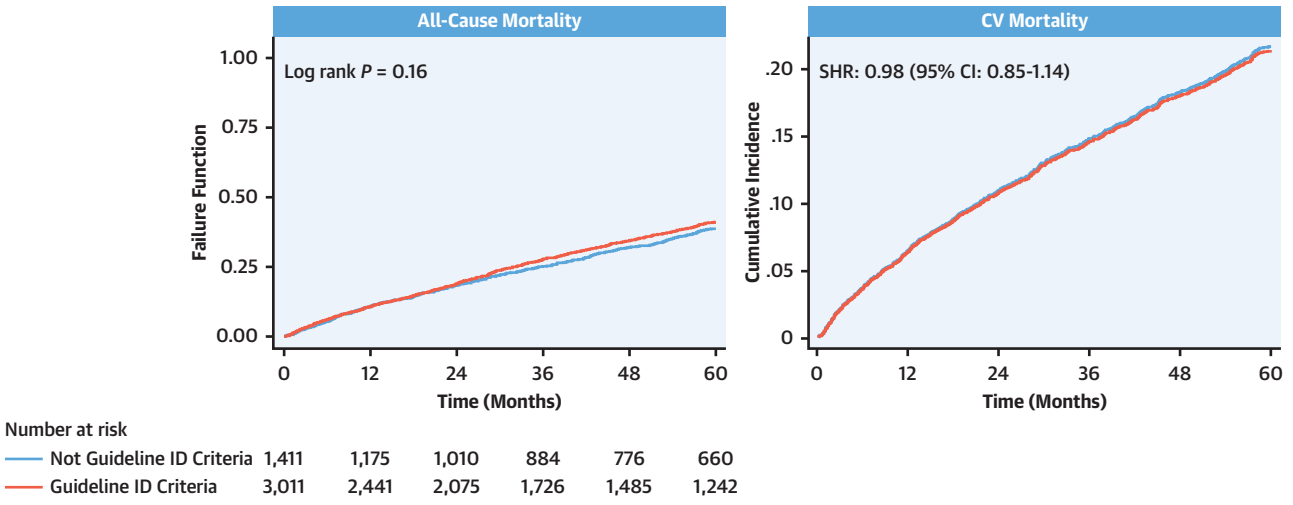
system as a result of inflammation, and subclinical gastrointestinal blood loss, which might be common in those taking antithrombotic agents.[21]

To our knowledge, this is the first study to assess systematically the prevalence and prognostic implications of different definitions of ID across all HF phenotypes in a large population of patients with chronic HF. Our findings confirm that ID is common in patients with HF, in line with previous reports.[2-4,11] However, many previous studies have included only patients with HFrEF and assessed ID using only the guideline definition. We extend these findings and suggest that many patients with HFpEF also have ID, which might reflect their advanced age, the high proportion of women, and the high comorbidity burden.

The relationship between serum iron biomarkers and adverse outcomes in those with ID remains uncertain. ID defined by guideline criteria was an independent predictor of mortality in a study of 546 patients with HFrEF and in another study of 1,506 patients with HF, of whom 87% had HFrEF (LVEF ≤45%).[4,22] However, others have questioned both the diagnostic and prognostic utility of the guideline criteria in patients with HF.[10-14]

In a European multicenter study of 1,821 patients with chronic HF, a TSAT <20% but not a ferritin <100 ng/mL independently predicted mortality.[12] In another study, in which ID was defined by bone marrow iron staining,[10] only a serum iron ≤13 μmol/L or TSAT ≤19.8%, but not guideline criteria, predicted bone marrow ID in patients with HFrEF (defined by an EF ≤45%; n = 42) undergoing coronary artery bypass. In a previous analysis of the Hull LifeLab cohort, the highest quintiles of ferritin had the worst all-cause and CV mortality.[13] The current analysis includes many more patients, has longer

FIGURE 4 All-Cause and CV Mortality by Guideline-Defined ID

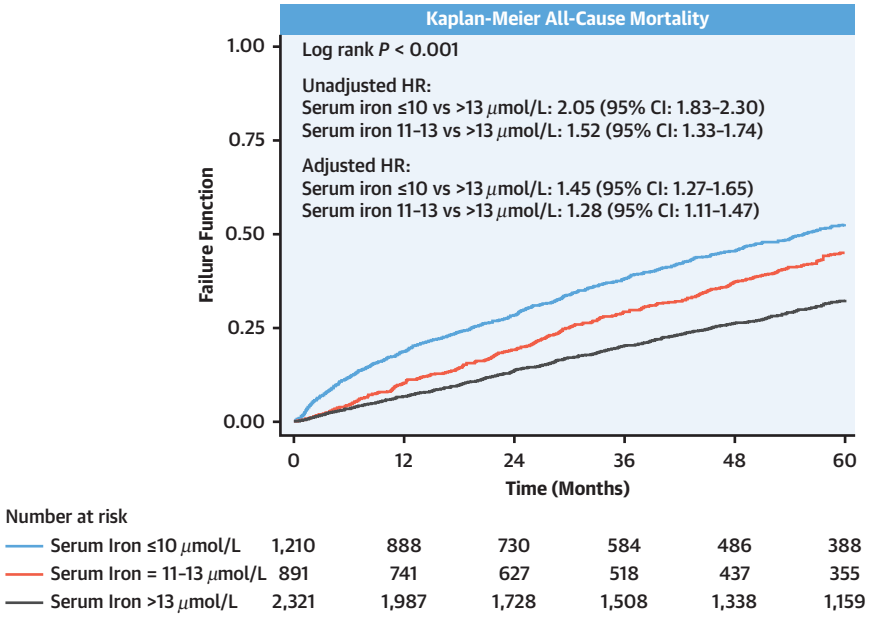


Kaplan-Meier cumulative curves for all-cause mortality and cumulative incidence curves using Fine and Gray method for cardiovascular (CV) mortality according to guideline iron deficiency (ID) criteria. SHR = subdistribution HR.

follow-up and classifies patient phenotype in line with international guidelines on HF.

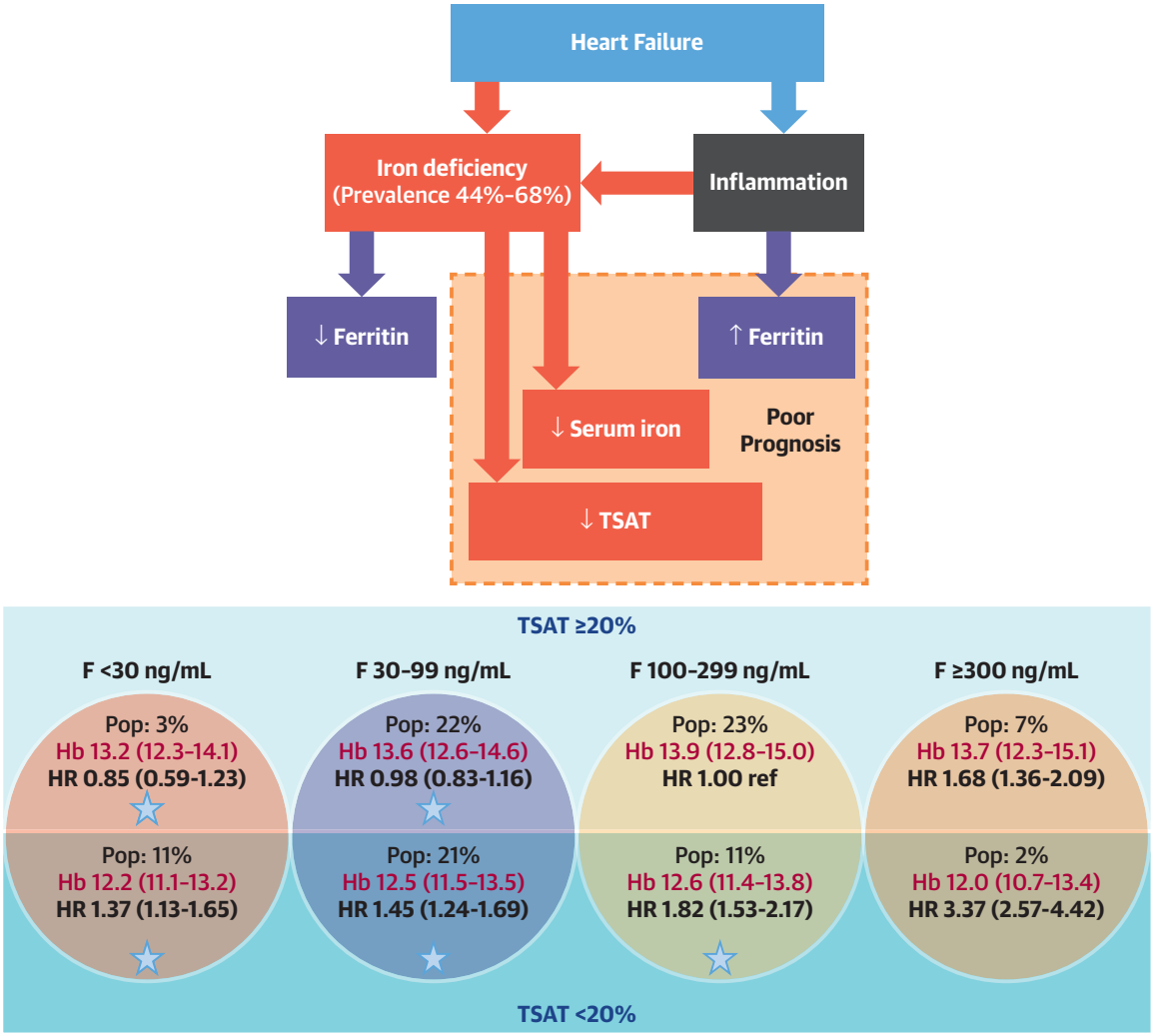
An individual patient data meta-analysis of 4 randomized controlled trials comparing IV ferric carboxymaltose with placebo in patients with HFrEF suggested that the prognostic benefit of ferric carboxymaltose might be limited to those with a TSAT ≤19.8%, irrespective of ferritin concentration. These findings support the use of TSAT rather than ferritin to select those more likely to benefit from IV

FIGURE 5 All-Cause Mortality and HRs by Iron Strata



Kaplan-Meier cumulative curves for all-cause mortality with unadjusted and adjusted HRs for 5-year mortality according to serum iron <10 μmol/L, 10-13 μmol/L, and >13 μmol/L (reference group). Model adjusted for age (/5), sex, body mass index (/5), ischemic heart disease, diabetes, systolic blood pressure (/5), New York Heart Association functional class III or IV, heart rate (/5), atrial fibrillation or atrial flutter, heart failure phenotype, Ln[NT-proBNP], and estimated glomerular filtration rate (/5). Ln = natural logarithm.

CENTRAL ILLUSTRATION **Prevalence and Prognosis of ID Using Different Criteria**



Masini, G. et al. J Am Coll Cardiol. 2022;79(4):341-351.

In 4,422 patients with chronic heart failure, prevalence of iron deficiency (ID) is high, regardless of the definition used **(top)**. Current guideline definition of ID excludes patients who may have ID (transferrin saturation [TSAT] <20%) and a high mortality and includes many who may not be iron deficient (transferrin saturation [TSAT] ≥20%) who are at lower risk. **Stars** represent those patients who fulfill current guideline ID criteria. Data shown are the percentage of overall population (Pop), median (1st-3rd quartile) of hemoglobin (Hb) (g/dL), and the adjusted HR for 5-year mortality (95% CI). Model adjusted for age, sex, and heart failure phenotype. F = ferritin; ref = reference group.

iron. Identifying those most likely to respond to IV iron might be considered the best method for diagnosing clinically relevant ID.[10] These considerations should be taken into account when interpreting results from randomized trials testing the benefit of IV iron in patients with HF. Results from the ongoing IRONMAN (Intravenous Iron Treatment in Patients With Heart Failure and Iron Deficiency: IRONMAN; NCT02642562) trial in patients with TSAT <20% or ferritin <100 ng/L, and future, prespecified subgroup analyses of other ongoing trials of IV iron[23] assessing alternative diagnostic criteria such as those highlighted in our study will determine whether the current definition of ID should be revised.

Serum iron is almost entirely transferrin bound, and therefore a close association between serum iron and TSAT is not surprising.[13] Serum ferritin increases in response to cellular damage and inflammation, highlighted by reports of extremely high serum ferritin concentrations associated with severe COVID-19 (coronavirus disease-2019).[24] While

a serum ferritin <15 ng/mL may be specific for ID,[25] higher concentrations may reflect a complex interplay between ID, inflammation, and other causes of cell damage and do not rule out ID, rendering ferritin of little diagnostic use in patients with coexisting diseases with an inflammatory component, including HF.[9,17,26,27]

Grote Beverborg et al[10] detected bone marrow ID in 2 patients with a serum ferritin ≥300 ng/mL and a TSAT <20%. In our study, 26% of those with ferritin ≥300 ng/mL had a TSAT <20%, and a similar proportion of patients had a serum iron ≤13 μmol/L. Such patients had a lower hemoglobin and higher mortality than any other group. This may reflect more severe renal and cardiac dysfunction and congestion,[28] which may be a key stimulus to inflammation and hepcidin secretion, which leads, in turn, to reduced iron absorption and sequestration but a higher serum ferritin and a worse mortality.[29] However, very few patients, perhaps none,[30] with a serum ferritin ≥300 ng/mL have been included in trials of IV iron therapy because of concerns about possible iron overload.

These concerns may not be valid if ferritin is such a poor guide to ID. The ongoing IRONMAN trial includes patients with either a TSAT <20% or ferritin <100 ng/mL and excludes patients only when serum ferritin is >400 ng/mL. Even higher thresholds have been proposed as an indicator of iron overload in patients with a variety of diseases.[31,32] In the PIVOTAL (Intravenous Iron in Patients Undergoing Maintenance Hemodialysis) trial of patients on renal dialysis, redosing with IV iron was encouraged provided serum ferritin was <700 ng/mL in the high-dose intervention arm.[33] Better validation of serum biomarkers of ID together with data on the efficacy and safety of IV iron in those with higher serum ferritin concentrations are required. New formulations of IV iron currently in use do not release large amounts of labile iron into the circulation, which may reduce the risks of iron loading, although excessive intracellular accumulation is still a concern.[34]

STUDY LIMITATIONS. Other biomarkers of interest, such as soluble transferrin receptor or hepcidin were not measured. Although soluble transferrin receptor might be a better test for ID than TSAT or serum iron,[35,36] it is not widely used clinically. We did not collect information on the rate of IV iron administration or blood transfusions that some of our patients might have received during follow-up. However, the first guideline on HF to recommend IV iron was published in May 2016,[37] and IV iron is not yet recommended by guidelines for HF in the United Kingdom.[38] We expect that few patients had received IV iron prior to June 2019. We did not investigate for the presence of genetic mutations associated with ID; however, they are rare and international guidelines do not recommend specific genetic testing for patients with ID. We enrolled patients during a period of 20 years, and evidence-based therapies for HF have evolved. However, applying sensitivity analysis, the period of enrolment did not affect our main results. This is a single-center study including a predominantly white British population, and therefore data should be extrapolated with caution to a population with characteristics different from ours.

CONCLUSIONS

Irrespective of how it is defined, ID is common in patients with HF. When defined by current guideline criteria, ID was not associated with poor outcome; indeed, lower serum ferritin concentrations were associated with a better survival. TSAT <20% and serum iron ≤13 μmol/L were associated with a higher mortality and this was independent of HF phenotype.

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PERSPECTIVES

COMPETENCY IN PATIENT CARE AND PROCEDURAL SKILLS: Irrespective of HF phenotype, low serum iron or TSAT is associated with higher mortality, while ID, as defined in the current HF guidelines, is not. Serum ferritin may better reflect inflammation than ID in patients with HF.

TRANSLATIONAL OUTLOOK: Clinical trial data should be analyzed to determine the criteria for ID that best identify patients with HF likely to benefit from iron replacement.

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KEY WORDS definition, heart failure, iron deficiency

APPENDIX For supplemental tables and figures, please see the online version of this paper.