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Iron Deficiency in Heart Failure Patients with Reduced Ejection Fraction

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List of Abbreviations

6MWT: 6-Minute Walk Test

ACCF: American College of Cardiology Foundation

AHA: American Heart Association;

ACD: anemia of chronic disease

ACEIs: angiotensin-converting-enzyme inhibitors

ADH: anti diuretic hormone

AHF :acute heart failure

ANP: atrial natriuretic peptide

ANS: adrenergic nervous system

ARBs: Angiotensin receptor blockers

ARNI: Angiotensin receptor neprilysin inhibitor

ARVD: Arrhythmogenic right ventricular dysplasia

ASD: Atrial septal defect

BDI: beck depression inventory

BNP: brain natriuretic peptide

CAD: Coronary artery disease

CKD: chronic kidney diseases

CRF: chronic renal failure

CRP :c reactive protein

DCM: dilated cardiomyopathy

ECG: Electrocardiogram

EPO: erythropoietin

ESAs: erythropoiesis stimulating agents

ESC: European Society of Cardiology

FCM: ferric carboxymaltose

FCM: ferric carboxymaltose

FGF: fibroblast growth factor

Fpn: ferroportin

GDMT: Guideline-Directed Medical Treatment

HAMP: Hepcidin antimicrobial peptide

Hb: Hemoglobin

HCM: hypertrophic cardiomyopathy

HF: Heart Failure

HFpEF: Heart failure with preserved ejection fraction

HFrEF: Heart failure with reduced ejection fraction

HMWD: High-Molecular Weight Dextran

HT: Hypertension

ICD: Implantable cardioverter-defibrillator

ID : Iron deficiency

IDA: iron deficiency anemia

IHD: Ischemic heart disease

ISC: iron sucrose complex

IV: intravenous

JCS/JHFS: Japanese Circulation Society/ Japanese Heart Failure Society

LVEDP: LV end-diastolic pressure

LVEF: Left ventricle ejection fraction

LVH: Left Ventricular Hypertrophy

LVNC: Left ventricular noncompaction

MCH: mean corpuscular Hb

MCHC: mean corpuscular Hb concentration

MCV: Mean corpuscular volume

MI: myocardial infarction

MRA: Mineralocorticoid/aldosterone receptor antagonists

MRI: Magnetic resonance imaging

MR-proANP: mid-reginal atrial natriuretic peptide prohormone

ND-CKD : nondialysis-dependent CKD

NE : norepinephrine

NSAID: Non-steroidal anti-inflammatory drugs

NTBI: non–transferrin- bound iron

NT-pro-BNP: N-terminal pro- brain natriuretic peptide

NYHA: New York Heart Association

PGA: patient global assessment

PIC: polysaccharide iron complex

PND: Paroxysmal Nocturnal Dyspnea

QALY: quality-adjusted life year

Qol : quality of life

RAAS: renin-angiotensin-aldosterone system

RDA: Recommended dietary allowances

rHuEPO: recombinant human erythropoietin

ROS: reactive oxygen species

Tf: Transferrin

TfR: transferrin receptor

TIBC: total iron binding capacity

TSAT: Transferrin saturation

VSD: ventricular septal defect

WHO: world health organization

1. INTRODUCTION

Heart failure is a heterogeneous clinical syndrome that results in a decline of the heart's pumping capacity and subsequent inability to meet the body's circulatory demands. Remarkable advances in our understanding of the pathogenesis of heart failure that have led to therapies with considerable improvement in patient outcomes. Despite this, however, the prognosis of HF remains poor (1).

Iron deficiency and anemia are common comorbidities in HF that can complicate treatment and affect clinical outcomes. Iron deficiency was known as a worldwide nutritional disorder, affecting an estimated 2 billion people, and now aside from being traditionally linked to anemia, iron deficiency was noted as a separate condition with a high prevalence (30% to 50%) in the HF community (2). Therefore, it emerged as a new therapeutic target of chronic heart failure. Recent European guidelines (3) recommend the monitoring of iron parameters (serum ferritin, transferrin saturation) for patients with heart failure.

The treatment of ID in heart failure with the use of oral iron supplementation versus intravenous (IV) iron remains a topic of controversy, at present, IV iron is the preferred route. The European guidelines (3) recommends treatment with IV ferric carboxymaltose in symptomatic heart failure patients with iron deficiency to improve heart failure symptoms and quality of life, but additional clinical trials are needed to more fully characterize the therapeutic potential and its safety.

In this thesis, the focus was on chronic heart failure with reduced ejection fraction (< 40%), iron physiology and the pathophysiology of iron homeostasis in HF. We discussed contemporary and possible diagnostic tools of iron deficiency in HF. We also highlighted the global perspective of iron deficiency in HF with regard to prevalence, clinical implications, and provide an overview of recent, ongoing and emerging future therapeutic approaches to treat iron deficiency in HF. In study, our primary aim was to assess the prevalence of iron deficiency and anemia in chronic heart failure with reduced ejection fraction.

Section I:

Theory

2. Chapter One: Chronic Heart Failure with Reduced Ejection Fraction

2.1 Definition of Heart Failure:

Heart Failure (HF) is a chronic, progressive condition commonly referred to as a "condition in which the heart cannot pump enough blood to meet the body's needs" (4). HF is a clinical syndrome with different etiologies and pathophysiology rather than a specific disease which makes defining HF a difficult task. Various definitions of HF currently exist in the medical literature, in contemporary guidelines and in medical practice (**Table 1**). Differing definitions have been developed for different purposes. Overall, the existing definitions of HF comprise 3 elements: evidence of structural heart disease, a history of symptoms that are commonly reported in HF, and objective signs commonly seen in HF (5).

In this thesis, we will be following the European Society of Cardiology's 2016 heart failure guidelines (3). It defines heart failure as "a clinical syndrome characterized by typical symptoms (eg, breathlessness, ankle swelling and fatigue) that may be accompanied by signs (eg, elevated jugular venous pressure, pulmonary crackles and peripheral oedema) caused by a structural and/or functional cardiac abnormality, resulting in a reduced cardiac output and/or elevated intracardiac pressures at rest or during stress."

Table 1: HF Definitions in Contemporary Clinical Practice Guidelines.

ACCF/AHA (2013) (6)	HF is a complex clinical syndrome that results from any structural or functional impairment of ventricular filling or ejection of blood. The cardinal manifestations of HF are dyspnea and fatigue, which may limit exercise tolerance, and fluid retention, which may lead to pulmonary and/or splanchnic congestion and/or peripheral oedema. Some patients have exercise intolerance but little evidence of fluid retention, whereas others complain primarily of oedema, dyspnea, or fatigue.
JCS/JHFS (2017) (7)	HF is a clinical syndrome consisting of dyspnea, malaise, swelling and/or decreased exercise capacity due to the loss of compensation for cardiac pumping function due to structural and/or functional abnormalities of the heart.

2.2 Classification of HF:

2.2.1 Stages of the HF Continuum:

A recent report published in the journal of cardiac failure (5) proposed revised stages of the HF Continuum as the following:

- **At Risk for HF (stage A):**

Patients at risk for HF, but without current or prior symptoms or signs of HF and without structural cardiac changes or elevated biomarkers of heart disease. Patients with hypertension, atherosclerotic cardiovascular disease, diabetes, obesity, known exposure to cardiotoxins, a positive family history of cardiomyopathy, or genetic cardiomyopathy would be in this category. Not all of these patients will develop HF, but risk factor intervention may be warranted.

- **Pre-HF (Stage B):**

Patients without current or prior symptoms or signs of HF with evidence of one of the following:

- Structural Heart Disease: for example, left ventricular hypertrophy, cardiac chamber enlargement, ventricular wall motion abnormality, myocardial tissue abnormality, valvular heart disease.
- Abnormal cardiac function: for example, reduced left or right ventricular systolic function, evidence of increased filling pressures (by invasive or noninvasive measures), abnormal diastolic dysfunction.
- Elevated natriuretic peptide levels or elevated cardiac troponin levels, especially in the setting of exposure to cardiotoxins.

➢ **HF (Stage C):**

Patients with current or prior symptoms and/or signs of HF caused by a structural and/or functional cardiac abnormality.

➢ **Advanced HF (Stage D):**

Severe symptoms and/or signs of HF at rest, recurrent hospitalizations despite guideline-directed medical treatment (GDMT), refractory or intolerant to GDMT, requiring advanced therapies such as consideration for transplantation, mechanical circulatory support, or palliative care.

2.2.2 New York Heart Association (NYHA) functional classification (6) :

Class I: No limitation of physical activity. Ordinary physical activity does not cause undue fatigue, palpitation, dyspnea (shortness of breath).

Class II: Slight limitation of physical activity. Comfortable at rest. Ordinary physical activity results in fatigue, palpitation, dyspnea (shortness of breath).

Class III: Marked limitation of physical activity. Comfortable at rest. Less than ordinary activity causes fatigue, palpitation, or dyspnea.

Class IV: Unable to carry on any physical activity without discomfort. Symptoms of heart failure at rest. If any physical activity is undertaken, discomfort increases.

The NYHA functional classification is important to characterize symptoms and functional capacity of patients with symptomatic HF (Stage C) or advanced HF (Stage D). It is important to specify a baseline NYHA functional class after the initial diagnosis, and after treatment through the period of care of a patient with HF (5). For example, patients with symptomatic HF (Stage C) may become asymptomatic with treatment. These patients will still be categorized as having HF Stage C but NYHA functional class I can further specify the absence of current symptoms. Worsening NYHA functional class is associated with a worse prognosis. Further optimization of GDMT is advised for any symptomatic patient with HF (NYHA functional class II to IV).

2.2.3 Classification According to Left Ventricle Ejection fraction (LVEF):

Left ventricle ejection fraction (LVEF) is a measurement, expressed as a percentage, of how much blood in the left ventricle gets pumped out with each systole (contraction) (**Equation 1)**. Echocardiography is widely used to non-invasively measure EF.

Equation 1: Ejection Fraction Formula.

$$Ejection\ fraction = \frac{Stroke\ volume}{End - diastole\ volume} \times 100$$

For clinical purposes, HF due to left ventricular (LV) dysfunction is categorized according to LV ejection fraction (LVEF). However, this categorization is not based upon etiology or pathophysiology,

but rather by clinical convention given the prognostic value of LVEF, the inclusion of LVEF thresholds as criteria in clinical HF trials, and the widespread availability of methods to measure LVEF (8). Consequently, patients with the same LVEF may not have necessarily the same underlying pathophysiology and prognosis. In addition, LVEF is not a robust measure of contractility (9), commonly changes over time, and is subject to substantial variability among and within modalities. Despite these reservations, classification by LVEF has proven to be clinically and epidemiologically useful (5). It also provides prognostic information as depressed LVEF is an adverse prognostic indicator in HF patients, with increasing morbidity and mortality as LVEF drops below 40 to 50 % (8).

All current clinical practice guidelines propose a classification of HF according to LVEF (**Table 2**). While they all use the terminology of Heart failure with reduced ejection fraction (HFrEF) and Heart failure with preserved ejection fraction (HFpEF), they differ in the terminology used for patients with EFs between 40 and 49%. In this thesis, we followed the ESC's 2016 classification presented in **Table 2.**

Table 2: Current HF Classifications According to LVEF in Contemporary Clinical Practice Guidelines.

	HF Classification According to LVEF	LVEF	Additional requirements
ACCF/AHA (2013) (6)	Heart failure with reduced ejection fraction (HFrEF)	≤40%	Symptoms and signs
	Heart failure with preserved ejection fraction (HFpEF)	≥50%	Symptoms and signs
	HFpEF, borderline	41%-49%	Symptoms and signs
	HFpEF improved	>40%	Symptoms and signs
ESC (2016) (3)	Heart failure with reduced ejection fraction (HFrEF)	<40 %	Symptoms and signs
	Heart failure with mid-range ejection fraction (HFmrEF)	40 to 49%	Symptoms and signs, elevated levels of natriuretic peptides and ≥ 1 additional criterion of relevant structural heart disease (LVH or LAE) or diastolic dysfunction
	Heart failure with preserved ejection fraction (HFpEF)	≥50%	Symptoms and signs, elevated levels of natriuretic peptides and ≥ 1 additional criterion of relevant structural heart disease (LVH or LAE) or diastolic dysfunction
JCS/JHFS (2017)(7)	Heart failure with reduced ejection fraction (HFrEF)	<40%	
	Heart failure with mid-range ejection fraction (HFmrEF)	40% to <50%	
	Heart failure with preserved ejection fraction (HFpEF)	≥50%	
	Heart failure with preserved ejection fraction, improved (HFpEF improved) or heart failure with recovered EF (HFrecEF)	≥40%	

2.3 Epidemiology of Heart Failure:

Ageing of the population and the prolongation of cardiac patients' lives by modern medicine has led to an increase in the prevalence of HF (10). Despite therapeutic improvements, HF's mortality rate remains high (11) highlighting the importance of early detection and preventive measures.

2.3.1 Prevalence:

The American heart association (AHA) estimated the presence of 6.2 million people with HF in the united states between 2013 and 2016 (12). Globally, there are an estimated 23 million people with HF (13). A steep increase in the prevalence of HF and LV dysfunction was observed with age no matter the definition used (11,14–19). The Framingham Heart Study, for example, described an increase in the prevalence of HF in men from 8 per 1000 at ages 50 to 59 years to 66 per 1000 at ages 80 to 89 years; similar findings (8 and 79 per 1000) were also found in women (11).

An increase in the prevalence of HF in the population over time has been observed. From 1989 to 1999, the average increase, in one study, was 1/1000 and 0.9/1000 for women and men, respectively (20). This has been associated with a three- to fourfold rise in HF's hospitalization rate from 1971 to 1999 (20,21). This rise has several contributing elements from an ageing population to improved treatment of hypertension and valvular and coronary disease allowing patients to survive their diseases only to later develop HF. In the united states, a rise in the prevalence of HF is projected over the next forty years, with an estimated 772, 000 new HF cases projected in the year 2040 (22) and a total of 8 million prevalent cases by 2030 (23).

2.3.2 Incidence:

The incidence of HF, similarly to its prevalence, increases with age (19,24). In the Framingham Study, the incidence approximately doubled over each successive decade of life. It rose more steeply with age in women than in men. The annual incidence in men rose from 2 per 1000 at ages 35 to 64 years to 12 per 1000 at ages 65 to 94 years. The lifetime likelihood of developing HF is approximately 20 % at all ages above 40 because the increase in risk with age is balanced by the decreased life expectancy with older age (24).

2.3.3 The Lifetime risk of developing HF:

The Framingham Heart Study found, at age 40, the lifetime risk of developing HF for both men and women was one in five (24). At age 40 and without antecedent myocardial infarction (MI), the lifetime risk of HF occurring was one in nine for men and one in six for women. The Physicians' Health Study observed a lower lifetime risk, one in seven at age 40, which may be attributed to healthy lifestyle factors (25).

2.4 Pathophysiology of Heart Failure with Reduced Ejection Fraction:

2.4.1 Physiology of the heart:

The heart is a specialized muscular organ that rhythmically contracts and pumps blood from the low-pressure venous side to the high-pressure arterial side of the circulation. Efficient pumping occurs thanks to orderly contraction sequence of the four heart chambers (left and right atria and left and right ventricles) and the valves that exist within the heart that ensure a unidirectional flow of blood (26). The heart can be viewed functionally as two pumps with the pulmonary and systemic circulations situated between the two pumps (**Figure 1**).

The right atrium receives deoxygenated blood from the systemic circuit via the superior vena cava and the inferior vena cava and pumps it into the right ventricle through the right atrioventricular valve (tricuspid valve). The right ventricle receives the deoxygenated blood from the right atrium and ejects

it into the pulmonary circuit through the pulmonary semilunar valve (pulmonary valve) to the pulmonary artery. Gas exchanges occur in the pulmonary capillaries (oxygen into the blood, carbon dioxide out).

The left atrium receives the newly oxygenated blood from the pulmonary circuit via the pulmonary veins and pumps it into the left ventricle through the left atrioventricular valve (mitral or bicuspid valve). The left ventricle receives that oxygenated blood from the left atrium and ejects it into the systemic circuit through aortic semilunar valve (the aortic valve) to the aorta. Following exchanges in the systemic capillaries (oxygen and nutrients out of the capillaries and carbon dioxide and wastes in), blood returns to the right atrium and the cycle is repeated.

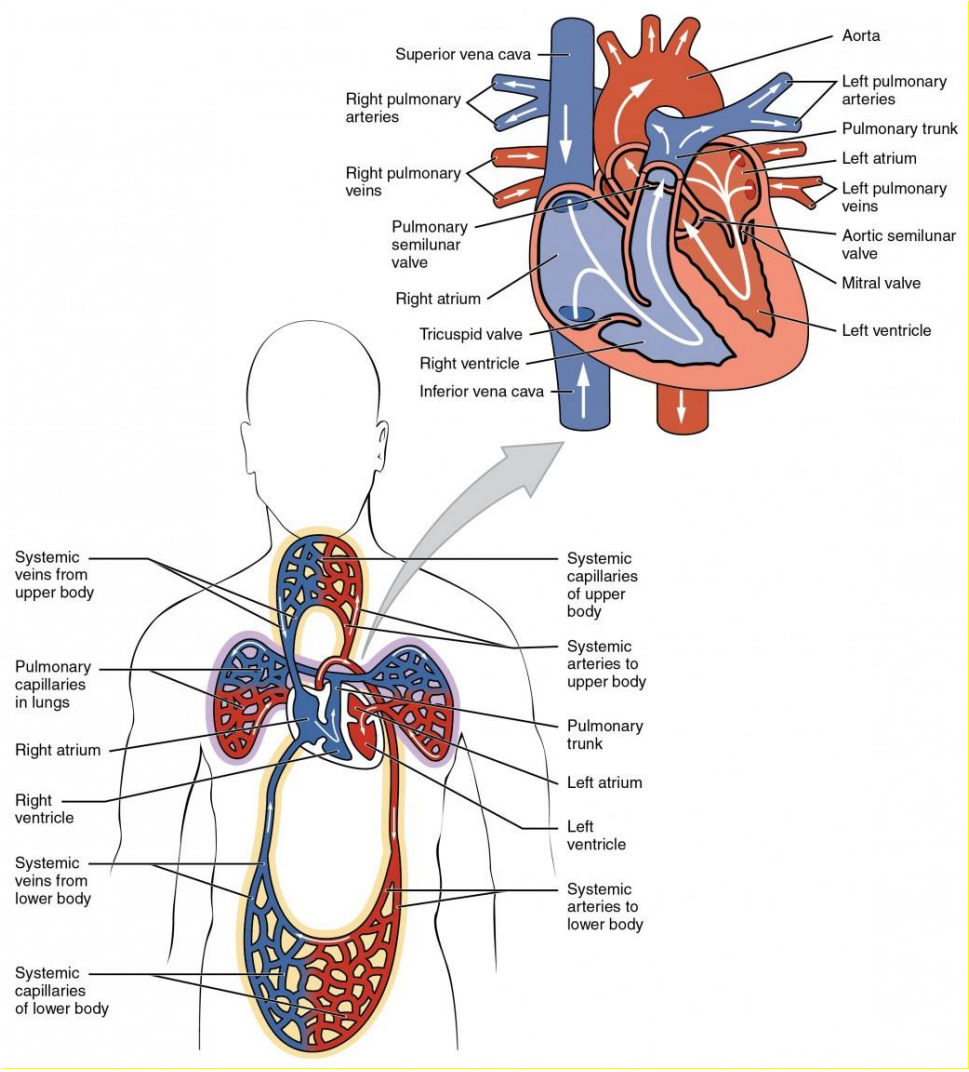

Figure 1: Chambers and Circulation through the Heart (27)

Cardiac cycle refers to the sequence of events that take place when the heart beats. The cardiac cycle is divided into two general categories: systole and diastole. Systole refers to events associated with ventricular contraction and ejection. Diastole refers to the rest of the cardiac cycle, including ventricular relaxation and filling. The cardiac cycle is further divided into phases: atrial diastole, atrial systole, isovolumic contraction, isovolumic relaxation, ventricular filling stage (**Figure 2**).

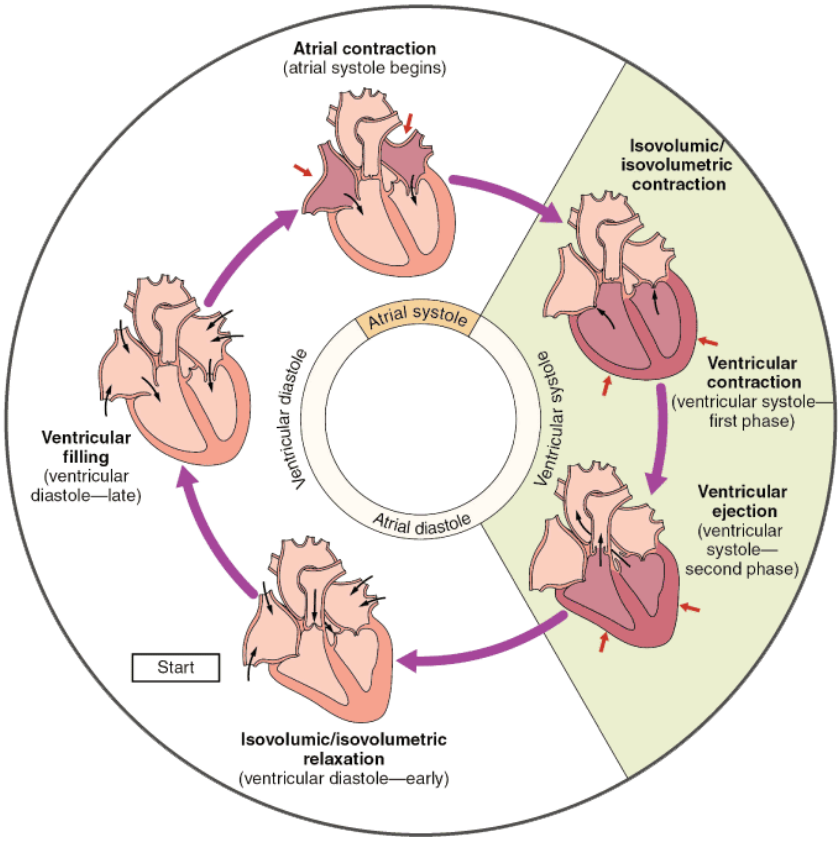

Figure 2: The phases of cardiac cycle (28).

2.4.2 Pathophysiology of HFrEF:

HF is a progressive disorder that is initiated after an index event either damages the heart muscle, resulting in the loss of functioning cardiac myocytes, or disrupts the ability of the myocardium to generate force, consequently preventing the heart from contracting normally (29). This index event may have an abrupt onset (myocardial infarction) or a gradual onset (hemodynamic pressure or volume overloading) or it may be hereditary (genetic cardiomyopathies). Regardless of the nature of the inciting event, they all in some manner produce a decline in the pumping capacity of the heart.

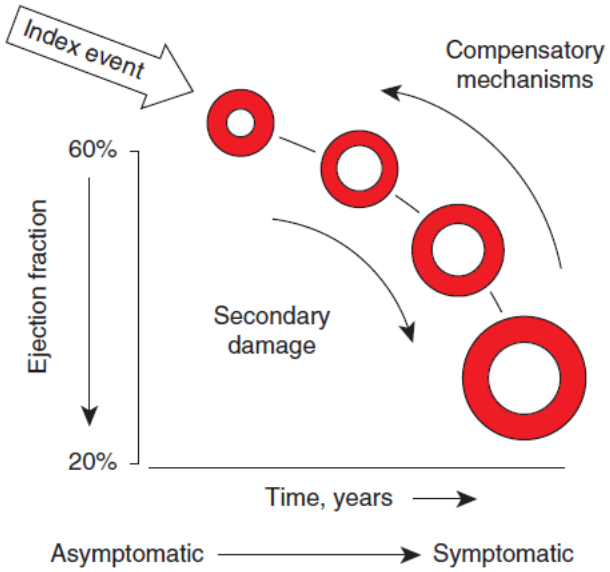

Figure 3: Pathogenesis of heart failure with a depressed ejection fraction (29).

Global LV systolic dysfunction predominates in HFrEF (also called systolic HF). LV systolic dysfunction refers to a decrease in myocardial contractility defined as an alteration in the relationship between preload (frequently defined by LV filling pressure) and stroke volume (8). This change is associated with a drop in stroke volume. The left ventricular ejection fraction (LVEF) is thus reduced which results in a decrease in cardiac output and an increase in filling pressures (LV end-diastolic pressure (LVEDP) and consequently pulmonary capillary pressure). It is this increase that is responsible for the congestive signs of HF (30).

The failing heart is progressively more afterload-dependent, and small changes in afterload can produce large changes in stroke volume (**Figure 4**). Reducing afterload in patients with HF via pharmacological treatments has the dual advantage of increasing cardiac output and, over the long term, slowing the rate of loss of myocardial function (8).

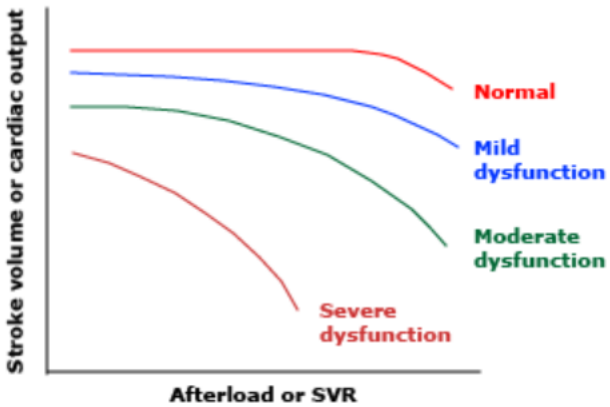

Figure 4: Curves relating stroke volume or cardiac to afterload or systemic vascular resistance (SVR) in normal subjects and those with heart failure and increasing degrees of ventricular dysfunction (8).

2.4.3 Compensatory mechanisms:

After the initial decline in pumping capacity, a variety of compensatory mechanisms are activated, including the adrenergic nervous system, the renin-angiotensin-aldosterone system, and the cytokine system. In the short term, these systems are able to restore cardiovascular function to a normal homeostatic range which allows the patient to remain asymptomatic (29). However, sustained activation of these systems leads to secondary end-organ damage within the ventricle, with worsening left ventricular remodeling and subsequent cardiac decompensation (31).

2.4.3.1 Left Ventricular remodeling:

Ventricular remodeling refers to the changes in LV mass, volume, and shape and the composition of the heart that occur after cardiac injury and/or abnormal hemodynamic loading conditions, in combination with neurohormonal activation and other factors (8,29). The cardiac myocytes and the interstitial collagen matrix undergo structural, functional, cellular, and molecular changes during the process of remodeling (**Table 3**). It may transition from an apparently compensatory (adaptive) process to a maladaptive one (32).

Table 3: Overview of Left Ventricular Remodeling (33).

Alterations in Myocyte Biology	Myocardial Changes	Alterations in Left Ventricular Chamber Geometry
- Excitation-contraction coupling. - Myosin heavy chain (fetal) gene expression. - β-Adrenergic desensitization. - Hypertrophy. - Myocytolysis. - Cytoskeletal proteins.	- Myocyte loss: - Necrosis. - Apoptosis. - Autophagy. - Alterations in extracellular matrix: - Matrix degradation. - Myocardial fibrosis.	- Left ventricular (LV) dilation. - Increased LV sphericity. - LV wall thinning. - Mitral valve incompetence.

2.4.3.2 Neurohumoral adaptations:

2.4.3.2.1 Activation of the sympathetic (adrenergic) nervous system:

The role of sympathetic nervous system (SNS) activation in HF is complex, with both beneficial and adverse effects. SNS activation is one of the first responses to a decrease in cardiac output which is sensed as a fall in blood pressure (34,35). Initially, activation of adrenergic nervous system (ANS) results in (30,35) :

- Increased heart rate and inotropism to restore cardiac output.
- Peripheral vasoconstriction to maintain adequate blood pressure.
- Redistribution of blood flow in favour of vital organs such as the brain and coronary arteries.
- Stimulation of the renin-angiotensin-aldosterone system (RAAS).

However, its sustained activation leads to (30):

- The exhaustion of the effect of catecholamines on the myocardium due to a reduction in the number of adrenergic receptors (down-regulation). In addition, catecholamines have a direct

toxic effect on the myocardium. These notions are the basis for the use of beta-blockers in HF. This also explains the decreased sensitivity to positive inotropes (like dobutamine) in these patients.

- By increasing peripheral resistance, peripheral vasoconstriction increases afterload which decreases stroke volume and worsens HF.

2.4.3.2.2 Activation of the renin-angiotensin-aldosterone system (RAAS):

The activation of RAAS is secondary, on the one hand, to the action of catecholamines on renin secretion and, on the other hand, to the drop in pressure in the afferent arterioles of the renal glomerulus (due to low output), which leads to the secretion of renin (**Figure 5**) (30,34). Renin stimulates the secretion of angiotensin II. The latter has a powerful peripheral vasoconstrictive action (to maintain blood pressure) and is responsible for increased aldosterone secretion. Aldosterone induces fluid retention which, by increasing blood volume, maintains blood pressure and increases venous return (thus preload and thus stroke volume).

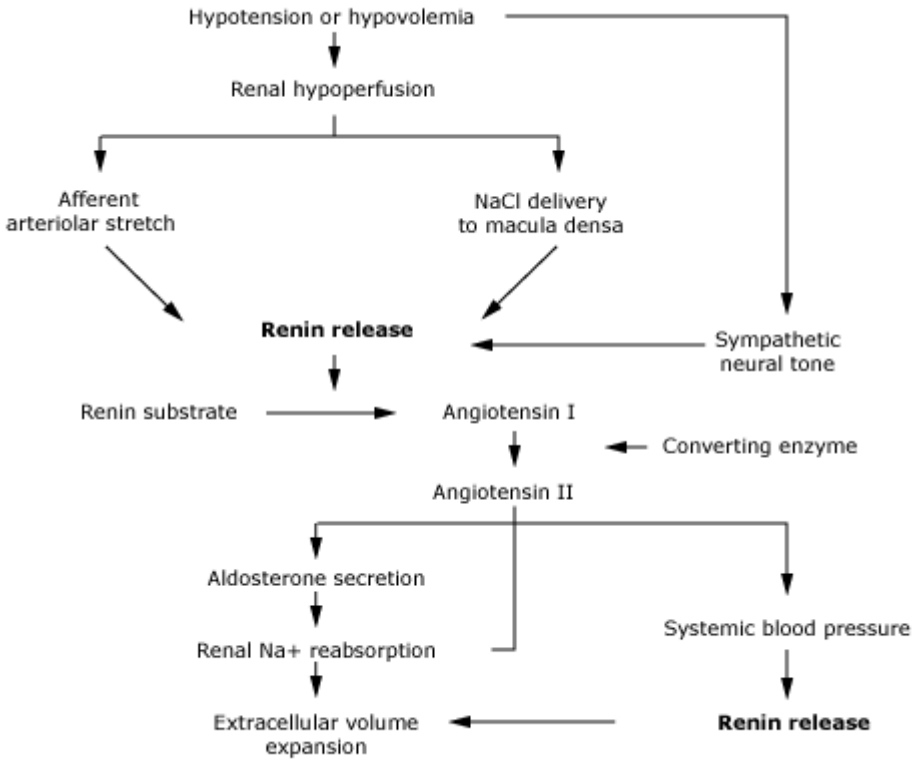

Figure 5: Regulation of renin release (35).

However, there are a number of maladaptive consequences of its persistent activation. The increase in blood volume induced by RAAS activation may lead to an increase in filling pressures, which increases congestive signs. The increase in blood volume does not always translate into an increase in effective blood volume because of the low cardiac output and the presence of eodema. Therefore, renin secretion remains high. It is a vicious circle that must be opposed by diuretic treatment. In general, water retention is greater than sodium retention, which leads to hyponatremia in severe heart failure. Finally, aldosterone has a pro-fibrosing action on the myocardium. This fibrosis may be responsible for sudden death due to ventricular rhythm disorders which is an additional reason for targeting RAAS in the treatment of HF.

2.4.3.2.3 Increased secretion of natriuretic peptides:

Volume expansion and/or increased intra-cardiac pressures leads to increased atrial and ventricular strain which triggers the release of atrial natriuretic peptide (ANP), primarily from the atria, and brain natriuretic peptide (BNP), primarily from the ventricles. Thus, plasma levels of these natriuretic peptides are elevated in patients with HF (35).

Expression of the BNP gene leads to the secretion of a precursor pro-BNP, which is then converted (proteolysis and glycolysis) into BNP with hormonal action (diuretic action) and N-terminal pro-BNP (NT-pro-BNP) without hormonal action (no diuretic action of its own) (30).

While plasma natriuretic peptide levels are useful biomarkers of disease severity, there is also evidence that natriuretic peptides, and in particular BNP, play an important beneficial role in the pathophysiology of HF. The natriuretic peptides exert a wide array of cellular and systemic effects which oppose those of the adverse effects of the sympathetic nervous system and renin-angiotensin-aldosterone system (35). For example, they oppose vasoconstriction, promote salt and water excretion, and protect target organs from the adverse effects of norepinephrine (NE) and angiotensin.

2.4.3.2.4 Increased secretion of antidiuretic hormone (ADH):

Activation of the carotid sinus and aortic arch baroreceptors by the low cardiac output in HF leads to enhanced release of ADH (also called vasopressin) and stimulation of thirst. Elevated levels of ADH may contribute to the increase in systemic vascular resistance in HF, and also promote water retention by enhancing water reabsorption in the collecting tubules. The combination of decreased water excretion and increased water intake via thirst often leads to a fall in the plasma sodium concentration. The severity of these defects tends to parallel the severity of the HF. As a result, the degree of hyponatremia is an important predictor of survival in these patients (35).

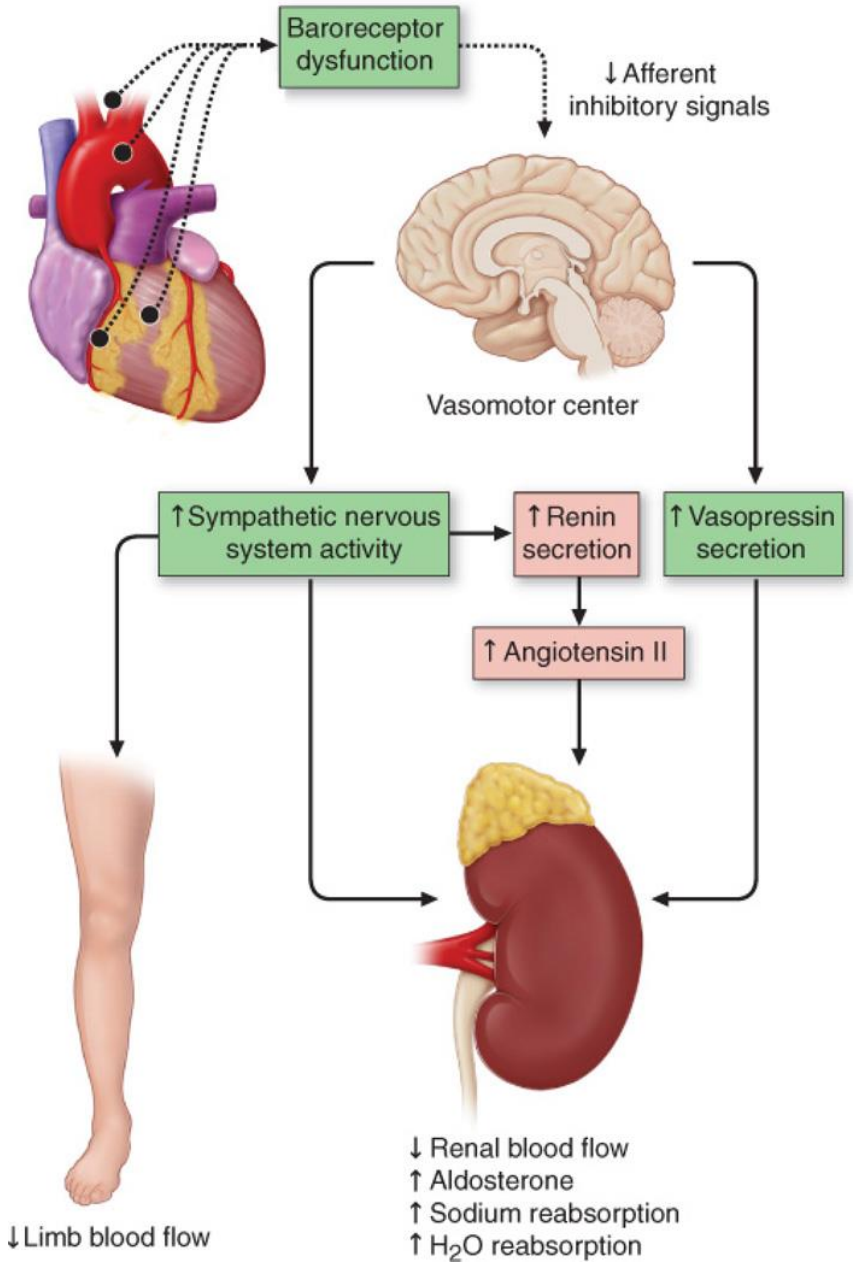

Figure 6: Overview of the activation of neurohormonal systems in heart failure (34).

2.5 Etiologies of HFrEF:

The search for an underlying cardiac abnormality is essential for the diagnosis of HF. Any disorder that results in an abnormality in LV structure or function can predispose a patient to develop HF (

Table 4). It is most often direct damage of the myocardial muscle, leading to systolic dysfunction (with reduced LVEF). However, HF can also be secondary to valvular, pericardial, rhythmic disease, etc. Although the etiology of HFrEF is distinct from that of HFpEF, there is significant overlap between the two disorders' etiologies.

Table 4: Etiologies of Heart Failure (29).

[a]Indicates conditions that can also lead to heart failure with a preserved ejection fraction.

HF with Reduced Ejection Fraction (<40%)	
Coronary artery disease: Myocardial infarction[a] Myocardial ischemia[a]	**Nonischemic dilated cardiomyopathy:** Familial/genetic disorders Infiltrative disorders[a]
Chronic pressure overload: Hypertension[a] Obstructive valvular disease[a]	**Toxic/drug-induced damage:** Metabolic disorder[a] Viral
Chronic volume overload: Regurgitant valvular disease[a] Intracardiac (left-to-right) shunting Extracardiac shunting	Chagas' disease
Chronic lung disease: Cor pulmonale Pulmonary vascular disorders	**Disorders of rate and rhythm:** Chronic bradyarrhythmia Chronic tachyarrhythmia

Coronary artery disease (CAD) has become the leading cause of heart failure (HF) in men and women in developed countries, accounting for 60–75% of cases. In 75% of patients, hypertension plays a role in the development of HF, including the majority of CAD patients (29). Both CAD and hypertension, as well as diabetes mellitus, interact to increase the risk of heart failure. Although the exact role of diabetes mellitus in HF is unclear, diabetes speeds up atherosclerosis and is frequently linked with hypertension. In 20–30% of the cases of HF with a reduced EF, the exact etiologic premise is not known (29). These patients are said to have non-ischemic, dilated, or idiopathic cardiomyopathy. Prior viral infection, toxin exposure (e.g., alcoholic or chemotherapeutic) or genetic defects also may lead to a dilated cardiomyopathy.

Rheumatic heart disease is still a prominent cause of heart failure in Africa and Asia, particularly among the young (29). In South America, Chagas disease remains a prevalent cause of HF. The epidemiology of HF in developing countries is growing more similar to that of Western Europe and North America as they progress socioeconomically, with CAD becoming the most common cause of HF. **Table 5** presents the principal etiologies of heart failure with reduced ejection fraction and their effects on cardiac function or structure.

Table 5: Principal etiologies of heart failure with reduced ejection fraction (36)

Etiology	Conditions	Description	Effect on Cardiac Function/Structure
Myocardial injury	Coronary artery disease	Blockages in coronary arteries that limit blood flow to the heart muscle.	It weakens or damages the myocardium and impairs its ability to eject blood.
	Cardiomyopathy	A progressive myocardial disorder	Weakens the myocardium impairing contractility and decreasing stroke volume.
	Viral myocarditis	Viral infection of the myocardium	Causes inflammation in the myocardium affecting its ability to contract and eject blood.
	Toxins	Alcohol, chemotherapy agents and radiation	Continued exposure may affect the myocardium and impairs its ability to eject blood.

Abnormal loading conditions	Arterial hypertension	Elevates arterial pressure	Increases cardiac workload to eject blood against increased pressure, which weakens the myocardium
	Aortic stenosis	Narrows the opening of aortic valve and impairs blood flow.	Increases cardiac workload to eject blood through the narrowed valve, weakening the myocardium.
	Mitral regurgitation	Improper closure of the mitral valve, leading to leakage on left side of the heart.	Increases blood volume leading to dilatation and weakened myocardium.
Arrhythmias	Tachyarrhythmias, bradyarrhythmias	Causes irregular heart rhythm.	Irregular rhythm decreases cardiac pumping effectiveness

2.6 Diagnosis of HFrEF:

The diagnosis of HF is relatively straightforward when the patient presents with classic signs and symptoms of HF (**Table 6**). However, the signs and symptoms of HF are neither specific nor sensitive. Accordingly, the key to making the diagnosis is to have a high index of suspicion, particularly for high-risk patients. When these patients present with signs or symptoms of HF, additional testing should be performed.

Table 6: Symptoms and Signs of HF (5)**.**

Symptoms of HF	**Signs of HF**
Typical	**More specific**
- Breathlessness - Orthopnea* - Paroxysmal nocturnal dyspnea* - Reduced exercise tolerance* - Fatigue, tiredness ± - Ankle swelling* - Inability to exercise* - Swelling of parts of the body other than ankles - Bendopnea	- Elevated jugular venous pressure* - Third heart sound* - Summation gallop with third and fourth heart sounds - Cardiomegaly, laterally displaced apical impulse - Hepatojugular reflux - Cheyne Stokes respiration in advanced HF ±
Less typical	**Less specific**
- Nocturnal cough - Wheezing - Bloated feeling ≠ - Postprandial satiety ≠ - Loss of appetite - Decline in cognitive function, confusion (especially in the elderly) ± - Depression - Dizziness, syncope ±	- Peripheral oedema (ankle, sacral, scrotal) - Pulmonary rales* - Unintentional weight gain (>2 kg/week) - Weight loss (in advanced HF) with muscle wasting and cachexia - Cardiac murmur - Reduced air entry and dullness to percussion at lung bases suggestive - of pleural effusion - Tachycardia, irregular pulse - Tachypnea - Hepatomegaly/ascites - Cold extremities ± - Oliguria Narrow pulse pressure

*Commonly used in clinical trials, registries, risk scoring, and have been tested for sensitivity and specificity.
±Common in low perfusion, low cardiac output states.
≠Can be typical in the setting of right HF or biventricular failure.

2.6.1 Symptoms:

In most instances, patients remain asymptomatic or minimally symptomatic after the initial decline in pumping capacity of the heart or develop symptoms only after the dysfunction has been present for some time. Although the precise reasons why patients with LV dysfunction may remain asymptomatic is not certain, one potential explanation is that a number of compensatory mechanisms become activated in the presence of cardiac injury and/or LV dysfunction allowing patients to sustain and modulate LV function for a period of months to years. Thus, patients may remain asymptomatic or minimally symptomatic for a period of years; however, at some point patients become overtly symptomatic, with a resultant striking increase in morbidity and mortality rates (29).

- **Dyspnea:**

It is the main symptom of left ventricular failure. Dyspnea is classified according to NYHA functional classification. In the early stages of HF, dyspnea is observed only during exertion. However, as the disease progresses, dyspnea occurs with less strenuous activity, and it ultimately may occur even at rest. The origin of dyspnea in HF is probably multifactorial. The most important mechanism is pulmonary congestion with accumulation of interstitial or intra-alveolar fluid, which activates juxtacapillary J receptors, which in turn stimulate the rapid, shallow breathing characteristic of cardiac dyspnea. Other factors that contribute to dyspnea on exertion include reductions in pulmonary compliance, increased airway resistance, respiratory muscle and/or diaphragm fatigue, and anemia (29).

- **Orthopnea:**

It is defined as dyspnea occurring in the recumbent position (laying down). It is usually a later manifestation of HF. It results from redistribution of fluid from the splanchnic circulation (blood supply to the gastrointestinal tract, liver, spleen, and pancreas) and lower extremities into the central circulation during recumbency, with a resultant increase in pulmonary capillary pressure. Nocturnal cough is a common manifestation of this process. Orthopnea generally is relieved by sitting upright or sleeping with additional pillows. Although orthopnea is a relatively specific symptom of HF, it may occur in patients with abdominal obesity or ascites and patients with pulmonary disease whose lung mechanics favor an upright posture (29).

- **Paroxysmal Nocturnal Dyspnea (PND):**

This term refers to acute episodes of severe shortness of breath and coughing that generally occur at night and awaken the patient from sleep. PND may manifest as coughing or wheezing. Patients with PND often have persistent coughing and wheezing even after they have assumed the upright position (29,30).

- **Cheyne-Stokes Respiration:**

Also referred to as periodic respiration or cyclic respiration, Cheyne-Stokes respiration is present in 40% of patients with advanced HF and usually is associated with low cardiac output. It is caused by an increased sensitivity of the respiratory center to arterial partial pressure of carbon dioxide (PCO_2) and a lengthy circulatory time. There is an apneic phase, during which arterial partial pressure of oxygen (PO_2) falls and arterial PCO_2 rises. These changes in the arterial blood gas content stimulate the respiratory center, resulting in hyperventilation and hypocapnia (low PCO_2), followed by recurrence of apnea (29).

- **Fatigue:**

Although fatigue traditionally has been ascribed to the low cardiac output in HF, it is likely that skeletal-muscle abnormalities and other noncardiac comorbidities (e.g., anemia) also contribute to this symptom (29,30).

- **Acute Pulmonary Edema**
- **Other Symptoms:**

Gastrointestinal symptoms. Cerebral symptoms such as confusion, disorientation, and sleep and mood disturbances may be observed in patients with severe HF, particularly elderly patients with cerebral arteriosclerosis and reduced cerebral perfusion. Nocturia is common in HF and may contribute to insomnia (29).

2.6.2 Tests and cardiac imaging:

2.6.2.1 Electrocardiogram (ECG):

A routine 12-lead ECG is recommended. ECG is helpful when looking for the etiology of HF. It can show signs related to the initial heart disease. The major importance of the ECG is to assess cardiac rhythm and determine the presence of LV hypertrophy or a prior MI (presence or absence of Q-waves). A normal ECG virtually excludes LV systolic dysfunction.

2.6.2.2 Chest X-Ray:

A chest x-ray provides useful information about cardiac size and shape, as well as the state of the pulmonary vasculature, and may identify noncardiac causes of the patient's symptoms. The majority of patients with chronic HF do not have evidence of pulmonary hypertension, interstitial edema, and/or pulmonary edema, contrary to patients with acute HF.

2.6.2.3 Assessment of LV Function:

Noninvasive cardiac imaging is essential for the diagnosis, evaluation, and management of HF.

- **Echocardiogram/Doppler:**

The most useful test is the two-dimensional (2-D) echocardiogram/Doppler, which can provide a semi-quantitative assessment of LV size and function as well as the presence or absence of valvular and/or regional wall motion abnormalities (indicative of a prior MI). The presence of left atrial dilation and LV hypertrophy. In addition to measuring EF, it can be used to search for the etiology and potential complications of HF, and to guide and follow up on treatments.

- **Magnetic Resonance Imaging (MRI):**

MRI also provides a comprehensive analysis of cardiac anatomy and function and is now the gold standard for assessing LV mass and volumes. MRI also is emerging as a useful and accurate imaging modality for evaluating patients with HF, both in terms of assessing LV structure and for determining the cause of HF.

2.6.3 Biomarkers:

Circulating levels of natriuretic peptides are useful and important adjunctive tools in the diagnosis of patients with HF. Both BNP and NT-proBNP, which are released from the failing heart, are relatively sensitive markers for the presence of HF with depressed EF.

Table 7: BNP thresholds to rule out HF in a patient with dyspnea (30).

BNP thresholds to rule out HF in a patient with dyspnea	
Chronic dyspnea	Acute dyspnea:
BNP < 35 pg/mL.	BNP < 100 pg/mL.

NT-pro-BNP < 125 pg/mL.	NT-pro-BNP < 300 pg/mL.

2.6.4 Routine Laboratory Testing:

Patients with new-onset HF and those with chronic HF and acute decompensation should have a complete blood count, a panel of electrolytes, blood urea nitrogen, serum creatinine, hepatic enzymes, and a urinalysis. Selected patients should have assessment for diabetes mellitus (fasting serum glucose or oral glucose tolerance test), dyslipidemia (fasting lipid panel), and thyroid abnormalities (thyroid-stimulating hormone level).

2.6.5 Exercise Testing:

Treadmill or bicycle exercise testing is not routinely advocated for patients with HF, but either is useful for assessing the need for cardiac transplantation in patients with advanced HF.

2.6.6 Differential Diagnosis:

HF should be distinguished from conditions in which there is circulatory congestion secondary to abnormal salt and water retention but without disturbance of cardiac structure or function (e.g., renal failure), and noncardiac causes of pulmonary edema. It is difficult differentiating the dyspnea that arises from cardiac and pulmonary causes. In this regard, noninvasive cardiac imaging, biomarkers, pulmonary function testing, and chest x-ray may be useful. A very low BNP or NT-proBNP may be helpful in excluding a cardiac cause of dyspnea in this setting. Ankle edema may arise secondary to varicose veins, obesity, renal disease, or gravitational effects.

2.7 Comorbidities:

Patients with CHF often have multiple comorbidities, both cardiovascular and non-cardiovascular, which accelerate disease progression, to a greater or lesser extent, and worsen the response to treatment (37,38). However, while most deaths are due to cardiovascular causes, non-cardiovascular causes (chronic renal failure, anemia, diabetes) are responsible for most hospitalizations (37–42). Likewise, it is known that patients with non-cardiovascular comorbidities present a higher risk of mortality and increased length of hospitalization compared to patients with CHF without comorbidities or those with only cardiovascular comorbidities (37,39,40,43,44).

2.8 Evolution:

The condition is rarely reversible after treatment of the etiology. In general, the functional signs stabilise (or even improve) with treatment, but ventricular function gradually deteriorates. This progressive worsening is usually interspersed with flare ups. The final stage is global HF, combining signs of left and right HF. The causes of death in CHF are mainly cardiovascular: sudden death and worsening of HF(30).

2.9 Complications:

Ventricular rhythm disorders (tachycardia and ventricular fibrillation) become common as left ventricular function deteriorates. 50% of deaths are due to sudden death secondary to a ventricular rhythm disorder. Thromboembolic events are also common. Low output may be responsible for cerebral hypo-perfusion (asthenia, memory problems, even confusion in very advanced HF). In the terminal stage, we can observe a very low output with severe hypotension, functional renal failure with oliguria and shock liver. Signs of pulmonary overload may be present or absent. These patients are often dependent on positive inotropic drugs such dobutamine.

2.10 Prognosis:

Despite recent advances in the management of HF, the development of symptomatic HF still carries a poor prognosis. Community-based studies indicate that 30–40% of patients die within 1 year of diagnosis and 60–70% die within 5 years, mainly from worsening HF or as a sudden event (probably

because of a ventricular arrhythmia). Although it is difficult to predict prognosis in an individual, patients with symptoms at rest (NYHA class IV) have a 30–70% annual mortality rate, whereas patients with symptoms with moderate activity (NYHA class II) have an annual mortality rate of 5–10%. Thus, functional status is an important predictor of patient outcome (29).

2.11 Management of HFrEF*:

2.11.1 Goals:

- Reduction of symptoms and improvement of quality of life.
- Reduction in the number and duration of hospital admissions.
- Reduction in mortality.
- Slowing down of disease progression.

2.11.2 Therapeutic Strategy:

The treatment of heart failure (HF) has several components:

2.11.2.1 Treatment of the underlying heart disease (etiological treatment):

It must systematically be sought because it is the only treatment likely to prevent the progression of heart failure. For example, revascularization techniques in ischemic heart disease (angioplasty, bypass surgery) and valve replacement in valvular heart disease…

2.11.2.2 Education and lifestyle changes:

- **Patient education:**

It is important to explain the disease to patients, its risks, its complications and potential side effects of treatments. In addition to emphasizing the need for compliance with diet, treatment and regular monitoring.

- **Rest and exercise:**

- Rest required in case of acute decompensation or NYHA stage IV dyspnea.
- Regular physical activity is recommended (walking 3 times, 1 hour/week) in patients in NYHA stage II.
- Cardiovascular rehabilitation (exercise rehabilitation).

- **Dietary consultation**:

Salt-free diet to adapted to the severity of the heart disease (generally < 6g/day), water restriction in case of hyponatremia or very severe LV dysfunction, fight against obesity and management of malnutrition (albumin< 30 g/L).

- Control of cardiovascular risk factors (hypertension, cessation of smoking, management of diabetes etc.).
- Avoidance of self-medication (NSAIDs, corticosteroids, etc.).
- Influenza and pneumococcal vaccination.

2.11.2.3 Other measures:

- Treatment of concomitant hypertension.
- Management of comorbidities.
- Oxygen therapy: necessary during flare-ups (pulmonary oedema), but not indicated for long-term use.
- Prevention of thromboembolic events.

* According to the 2016 European ESC guidelines on the management of chronic heart failure (3).

2.11.3 Pharmacological treatment:

Staged medication strategy, adapted according to NYHA stage and persistence of symptoms and/or severe LV dysfunction (**Figure 7**).

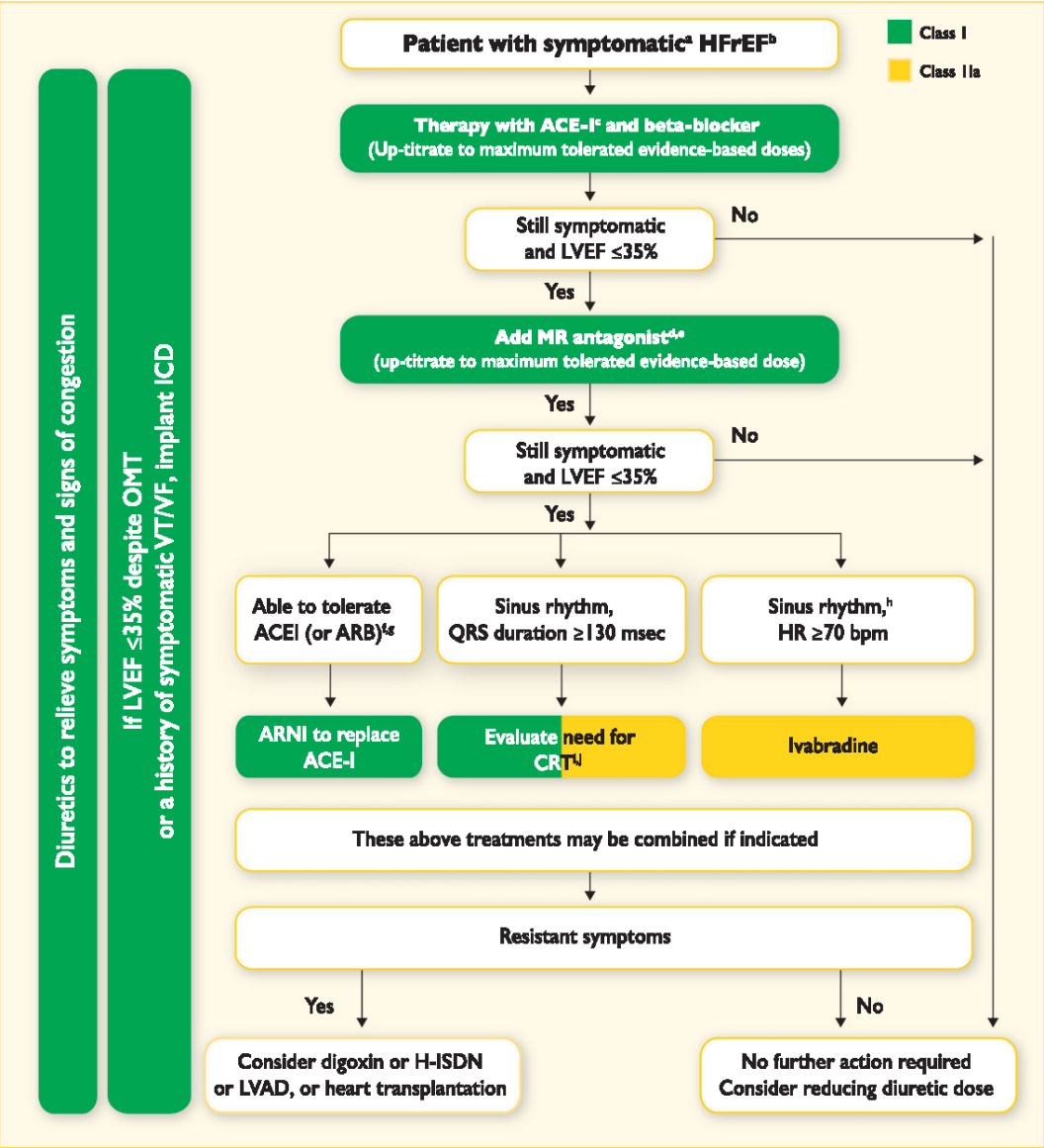

Figure 7: Therapeutic algorithm for a patient with symptomatic heart failure with reduced ejection fraction (3).

2.11.3.1 Angiotensin-converting-enzyme inhibitors (ACEIs) (ramipril, lisinopril, and perindopril):

ACE inhibitors are currently the most commonly used. They are arterial vasodilators. They decrease systemic arterial resistance and thus afterload. This promotes LV ejection and increases cardiac output. In addition, they limit ventricular dilation (ventricular remodelling) because of their action on the tissue RAAS (30).

ACEIs have been shown to reduce mortality and morbidity in patients with HFrEF (45–49) and are recommended unless contraindicated or untolerated in all symptomatic patients. They are also recommended in patients with asymptomatic LV systolic dysfunction to reduce the risk of HF

development, HF hospitalization and death (3). Therefore, they represent the basis of treatment for HFrEF, regardless of NYHA class (symptomatic or asymptomatic).

ACE inhibitors should be up-titrated to the maximum tolerated dose in order to achieve adequate inhibition of the RAAS (3). Urea, creatinine, kaliemia (K+), and blood pressure should be monitored after their introduction and then regularly during treatment (30).

2.11.3.2 Angiotensin II type I receptor blockers (ARBs) (candesartan, valsartan and losartan):

ARBs are recommended only as an alternative in patients intolerant of an ACEI, in symptomatic patients with LVEF < 40%, in combination with beta blockers and aldosterone receptor antagonists. They showed to have the same efficacy as ACEIs in terms of morbidity and mortality in CHF (30).

2.11.3.3 Beta-blockers (carvedilol, bisoprolol, metoprolol, and nevibolol):

Beta-blockers reduce mortality and morbidity in symptomatic patients with HFrEF, despite treatment with an ACEI and, in most cases, a diuretic. But they have not been tested in congested or decompensated patients. There is consensus that beta-blockers and ACEIs are complementary, and can be started together as soon as the diagnosis of HFrEF is made (3). There is no evidence favouring the initiation of treatment with a beta-blocker before an ACEI has been started (50).

In chronic patients, beta-blockers decrease heart rate. In addition, they improve myocardial supply/need ratio and energy utilization, contractility, and ventricular remodeling by reducing ventricular stress and volumes. They also increase stroke volume and LVEF. The later may increase by 25 to 30 %, but the effect only appears after the second month of treatment. Finally, they decrease ventricular rhythm disturbances and thus the risk of sudden death (30).

Beta blockers are recommended in patients with a history of myocardial infarction and asymptomatic LV systolic dysfunction to reduce the risk of death. Beta blockers should be initiated in clinically stable patients at a low dose and gradually up-titrated to the maximum tolerated dose (3). Each time the dosage is started or increased the following should be monitored: supine (lying on the back or with the face upward) and standing blood pressure, heart rate (bradycardia), functional tolerance (dyspnoea , congestive signs, weight), and ECG (30) .

2.11.3.4 Mineralocorticoid/aldosterone receptor antagonists (MRAs) (spironolactone and eplerenone):

Spironolactone or eplerenone are recommended in all symptomatic patients (despite treatment with an ACEI and a beta-blocker) with HFrEF and LVEF ≤35%, to reduce mortality and HF hospitalization (51,52). MRAs block receptors that bind aldosterone and, with different degrees of affinity, other steroid hormone (e.g. corticosteroids, androgens) receptors (3).

Their main side effects are renal failure and hyperkalaemia. Their use is contraindicted if blood potassium > 5.0 mmol/L at initiation of therapy and/or severe renal impairment (glomerular filtration rate (GFR) < 30 mL/minute/1.73 m^2). Regular checks of serum potassium levels and renal function should be performed according to clinical status (30).

2.11.3.5 Loop diuretics (furosemide, bumetamide..)

Diuretics are recommended to reduce the signs and symptoms of congestion in patients with HFrEF (3). Loop diuretics have natriuretic and kaliuretic effects. They can be used in case of renal failure with adjusted doses. In the case of resistant oedema, thiazide diuretics or spironolactone in small doses can be combined with loop diuretics. The aim is to achieve euvolaemia (no congestive signs) with the lowest dose of diuretics. In advanced HF, the patient can be educated to increase the dose himself in case of the worsening or the appearance of congestive signs (e.g. peripheral edema and/or weight gain…). The

main side effects are hypokalaemia (systematic substitution, except in the case of renal failure) and dehydration (in elderly subjects). In very high doses, furosemide can cause deafness if the injection is too rapid (30).

2.11.3.6 Angiotensin receptor neprilysin inhibitor (ARNI) (LCZ696):

A new therapeutic class of agents acting on the RAAS and the neutral endopeptidase system has been developed. The first in class is LCZ696, which is a molecule that combines the moieties of valsartan (ARB) and sacubitril (neprilysin inhibitor) in a single substance (3). LCZ696 (sacubitril/valsartan) results in the inhibition of the RAAS (valsartan) and increased concentrations of natriuretic peptides and bradykinin and other peptides (sacubitril). The Paradigm trial published in the New England Journal of Medicine in 2014 showed that this molecule had a superior effect to ACE inhibitors in the management of patients with reduced LVEF in terms of reduced mortality and hospital admissions (53). It is recommended in patients with LVEF ≤ 35%, still symptomatic, and on the optimal maximal treatment of ACE (or ARB) and beta-blockers and aldosterone receptor antagonists. ACE inhibitors and ARB should be stopped 36 hours and 24 hours, respectively, before the first dose of sacubitril/valsartan (30).

- **Contraindications** (30)**:** kaliemia > 5.4 mmol/L, blood pressure < 100 mmhg, end-stage renal disease, severe hepatic failure, biliary cirrhosis or cholestasis, concomitant use of ACEIs, history of angioedema related to previous treatment with ACEIs or ARB IIs, hereditary or idiopathic angioedema, and pregnancy (2nd and 3rd trimester).
- **Side effects** (30)**:** symptomatic hypotension, hyperkaliemia, impaired renal function, and angioedema with a concomitant prescription of an ACE inhibitor. If side effects do occur, the dose should temporarily be reduced or discontinued.

Sacubitril/valsartan inhibits the degradation of BNP but not NT-pro-BNP. Consequently, BNP levels are no longer interpretable. In a patient treated with sacubitril/valsartan, it is, therefore, necessary to measure NT-pro-BNP for monitoring.

2.11.3.7 Other medications that may also be useful in HFrEF:

2.11.3.7.1 If-channel inhibitor (Ivabradine):

Ivabradine slows the heart rate through inhibition of the If channel in the sinus node and therefore should only be used for patients in sinus rhythm. It is recommended for symptomatic patients with LVEF ≤35%, in sinus rhythm and with a heart rate ≥70 beats per minute (bpm) (3). It can be used instead of beta-blockers in case of intolerance or formal contraindications and is always combined with ACE inhibitors and aldosterone receptor antagonists (30).

2.11.3.7.2 Digoxin and other digitalis glycosides:

Digitalis have long been a staple treatment for CHF. They have no effect on mortality, but they reduce symptoms and re-hospitalization. Their positive inotropic and parasympathomimetic bradycardia effects improve LV function and symptoms. They can be combined with beta-blockers. They are second line treatment for patients in sinus rhythm, remaining symptomatic on a association of ACE inhibitors and beta-blockers and aldosterone receptor antagonists. Digoxin has a renal elimination. In case of renal failure, the dosage is reduced and the serum levels are monitored (30).

2.11.3.7.3 Combination of hydralazine and isosorbide dinitrate:

Hydralazine is an arterial and arteriolar vasodilator and isosorbide dinitrate is derivative of nitrate (results in relaxation of vascular smooth muscle and vasodilation). There is no clear evidence to suggest the use of this fixed-dose combination therapy in all patients with HFrEF (3). It is recommended as second line treatment in symptomatic black patients with LVEF ≤ 35% or LVEF < 45%, associated

with a cardiomyopathy, under treatment of a combination of ACE inhibitors and beta-blockers and aldosterone receptor antagonists (3,30).

2.11.3.7.4 Antiarrhythmics:

Beta-blockers and amiodarone. They are not systematic. Antiarrhythmics are recommended in case of atrial fibrillation and/or ventricular tachycardia. Class I antiarrhythmics (Flecainide in particular) are contraindicated (30).

2.11.3.7.5 Oral anticoagulants:

They are never systematically administered. Direct factor Xa inhibitors (nouvel oral anticoagulant) or vitamin k antagonists (Acenocoumarol) are recommended in case of atrial fibrillation. Vitamin k antagonists are also recommended in case of left ventricular thrombus (30). Successful therapy with vitamin K antagonists requires maintaining a patient in the therapeutic international normalized ratio (INR) range for the particular indication under treatment (54).

2.11.3.8 Medication to be avoided in HFrEF :

- The combination of ACEI and ARB II and aldosterone receptor antagonists is prohibited because of the risk of renal failure, hyperkaliemia and ventricular rhythm disorders (sudden death).
- Negative inotropic calcium channel blockers (diltiazem, verapamil) increase the risk of worsening HF.
- Dronedarone (non-iodinated class III anti-arrhythmic) increase risk of worsening HF and sudden death.
- Non-steroidal anti-inflammatory drugs (NSAIDs)
- Glitazones (oral hypoglycemic) (30).

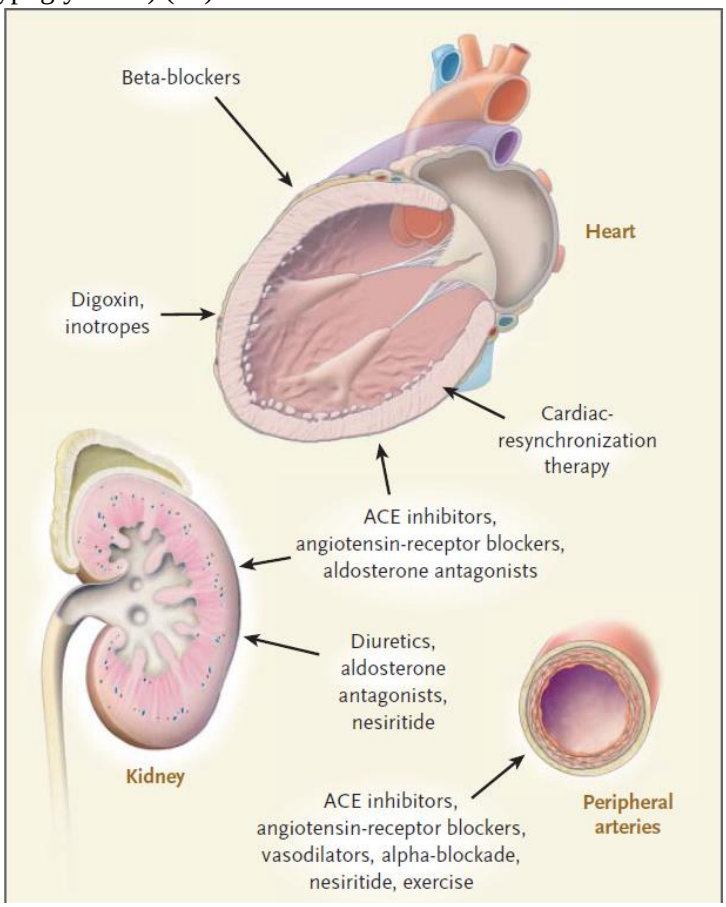

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Figure 8: Primary Targets of Treatment in Heart Failure (55).

2.11.4 Non pharmacological treatments:

2.11.4.1 Implantable cardioverter-defibrillator (ICD) :

An implantable cardioverter-defibrillator or automated implantable cardioverter defibrillator is a device implantable inside the body, able to perform cardioversion, defibrillation, and pacing of the heart. The device is therefore capable of correcting most life-threatening cardiac arrhythmias. Its main goal is to prevent sudden death, a major cause of death in HFrEF.

- **Secondary prevention** (30)**:**

It is recommended for patients with heart failure, regardless of LVEF, who have experienced resuscitated cardiac arrest on ventricular fibrillation or ventricular tachycardia with hemodynamic instability, provided that their life expectancy is greater than 1 year and there is no cause for the arrhythmia (infarction, hyperkaliemia, etc.).

- **Primary prevention** (30)**:**

This is before any episode of ventricular tachycardia and ventricular fibrillation. It is recommended for patients with reduced LVEF (≤ 35 %) remaining symptomatic in NYHA class II to III, despite 3 months of optimal medical treatment, provided that life expectancy is greater than 1 year.

It is important to note that LVEF is reassessed 40 days following an infraction.

2.11.4.2 Cardiac resynchronization therapy (pace maker) (30):

It is recommended for patients in sinus rhythm, remaining symptomatic despite optimal medical treatment, with an LVEF ≤ 35% and a wide QRS, provided that life expectancy provided that life expectancy is greater than 1 year.

2.11.4.3 Heart transplantation (30):

It is recommended for patient with irreversible severe chronic heart failure (NYHA stages III-IV), under optimal treatment, in the absence of contraindication. It is preceded by temporary circulatory support. There is also permanent circulatory support (artificial heart).

3. Chapter Two: Physiology of Iron

Iron is a necessary micronutrient that acts as a catalytic center for a broad spectrum of metabolic functions(56). As present in hemoglobin, iron is required for the transport of oxygen, and carbon dioxide critical for cell respiration. It is also a component of various tissue enzymes such as cytochromes that are critical for energy production and enzymes necessary for immune system functioning. The importance of iron as an element necessary for life derives from its redox reactivity as it exists in two stable, interchangeable forms, ferrous (Fe^{2+}) and ferric (Fe^{3+}) (57). Although other transition metals can similarly exist in different oxidation states, iron appears unique in its capacity to form complexes having redox potentials that are highly coordination-dependent.(58). Unfortunately, the redox reactivity of iron is a two-edged sword; the single-electron transfer reactions of iron can lead to the production of harmful free radicals and their derivatives such as peroxide, Superoxide, and hydroxyl radical (56,57). Clearly, a potentially harmful situation will exist in the presence of iron that is freely redox-active, for this reason, under normal in vivo situations, the availability of iron for participation in redox cycling is rigorously limited during transport and storage by an elaborate system of iron-binding proteins including transferrin, ferritin, and hemosiderin. (56,59,60).

Transferrin is an iron transport protein that mediates the exchange of iron among body tissues, while ferritin and hemosiderin serve to store iron within the body. These iron-binding proteins perform an important, although perhaps secondary, function by withholding iron necessary for bacterial growth and thereby preventing infectious growth of microorganisms(56,58).

The bodily iron content of the 70-kg adult man is approximately 4 to 5 g, with up to 80% of it located in erythrocytes' hemoglobin, 10%–15% in muscle myoglobin and other Fe-containing proteins and enzymes and only a very minor quantity of iron (n <0.1 %) circulate in the plasma bound to transferrin (Tf). Within cells, iron storage is managed by specialized iron storage proteins, the most important of which is ferritin (61).

3.1 Nutritional sources of Iron:

The body must find the necessary amount of iron in its diet. This later is present in variable quantities in many foods, but only a fraction of the iron consumed is actually absorbed, so the "real" iron intake depends on the content of the diet (dietary composition), physiological iron status and the bioavailability of this iron (as in its capacity to be absorbed and used)(62,63). It should be noted that more than the quantity of iron in the diet, it is the quality that determines the coverage of the needs. In fact, various studies carried out using foods marked with radioactive iron) have shown that the average absorption of iron in healthy subjects is very variable from one food to another (64). These differences can be explained by the form of the iron contained in the food: heme iron or non-heme iron.

Heme iron is present only in foods of animal origins where it represents about 40% of the total iron. Non-heme iron is present in both plant and animal derived food (65).

In terms of both, quantity and bioavailability, beef, red meat, and liver are excellent dietary sources of iron. Other good sources of iron include oysters, lentils, shrimps, spinach, lima beans, and whole-grain cereals.

3.2 Iron metabolism:

3.2.1 Absorption and distribution in the body

Iron absorption occurs primarily in the proximal small intestine and involves the uptake and transfer of iron across the enterocytes into the systemic circulation.

Dietary iron is absorbed in the intestine by a highly regulated active mechanism, mainly in the duodenum and the proximal part of the jejunum. Two situations can be distinguished according to the origin of the iron (66).

Heme iron has greater bioavailability and may be absorbed more easily without the need for absorption enhancers. Non-heme iron, the most significant dietary source in vegetarians, has a reduced bioavailability; its absorption is dependent on the balance of dietary enhancers and inhibitors, as well as body iron reserves. Approximately 20–30% of heme iron is absorbed via endocytosis of the entire heme molecule. Iron is then released into the enterocyte by a heme oxidase.

Non-heme iron is reduced from the ferric **Fe3+** to the ferrous form **Fe2+(**the form used for the synthesis of hemoglobin) in the intestinal lumen, this reduction is carried out by a specialized enzyme: duodenal cytochrome b (DcyB) (67), which is expressed on the brush border membrane of duodenal enterocytes, the major site for the absorption of dietary iron. Then Once in the form of Fe 2+, iron can then be taken up by a specific membrane transporter: The DMT1 (divalent metal transporter) which is present at the apical pole of the enterocytes. Once inside the enterocyte, iron from heme and non-heme sources is similarly transported through the cell and across the basolateral membrane by the ferroportin transporter in conjunction with the ferroxidase hephaestin after that it can be taken up by transferrin into the circulation. The regulation of iron across the basolateral membrane of the enterocyte is considered the most important aspect of iron absorption.

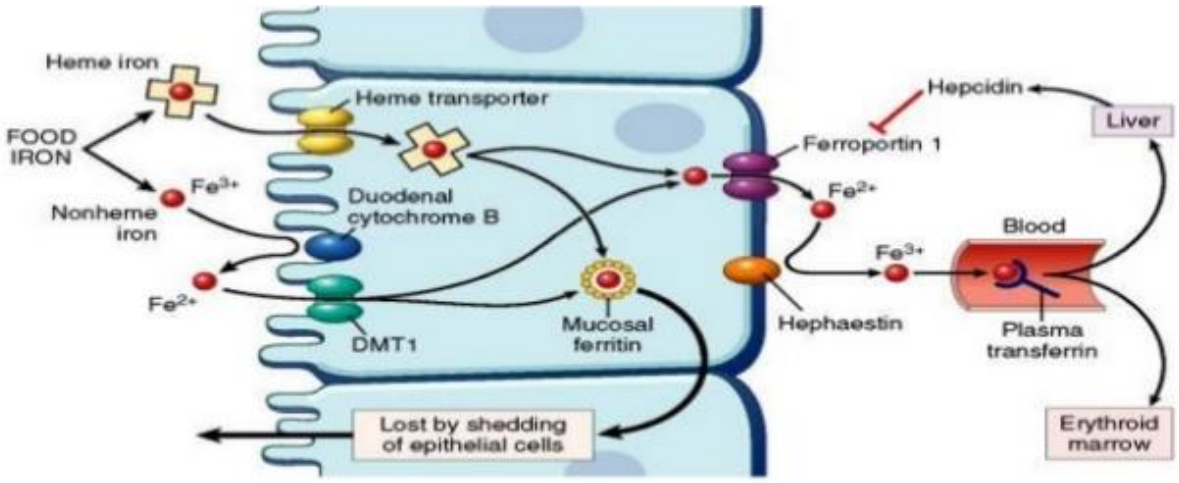


Figure 9: Iron absorbtion and regulation (68).

3.2.1.1 Bioavailability of dietary iron:

The terms bioavailability and absorption are often used interchangeably in the literature. Absorption refers to the passage of a nutrient, or other dietary constituents, from the intestinal lumen into the body. It usually comprises the uptake of a nutrient into the enterocytes of the gut mucosa and its subsequent transfer across the cell and into the body. Bioavailability describes the systemic utilization of a dietary nutrient and refers to the proportion of a nutrient that is taken up and transferred by the intestinal mucosa and subsequently used systemically. Heme and non-heme iron are transported into the intestinal

mucosal cells by independent mechanisms. The intestinal uptake and transfer of iron are dependent on the body's need for iron. The availability of dietary iron for uptake by the gut mucosa is affected by the chemical form of iron (heme iron or inorganic non-heme iron) and the character of the complexes of inorganic iron with other dietary constituents.

3.2.1.2 Dietary factors influencing iron absorption and bioavailability:

Dietary components that affect iron absorption have been identified and partly characterized from single meal studies using isotope labels (69). Single meal studies have shown that heme iron is more efficiently absorbed from the diet (20–30%) than non-heme iron (5–15%). Non-heme iron absorption in single meal studies is very variable and is influenced by the balance of dietary factors enhancing (e.g., meat, ascorbic acid) and inhibiting (e.g: soy protein, polyphenols, calcium…) absorption. Except for calcium, which has an inhibitory effect on both heme and non-heme iron (70), absorption of heme iron is not affected by other components of the diet.

3.2.2 Transport of Iron:

Iron is distributed systemically in the circulation as transferrin. Transferrin comprises a core carrier glycoprotein, apo transferrin, which can bind one or two atoms of ferric iron to form holo-transferrin, which is usually called transferrin. Fe3+ is the form of iron that binds to it, so the Fe2+ transported through ferroportin must be oxidized to Fe3+ first. 2 copper-containing proteins catalyze this oxidation of Fe2+: hephaestin and ceruloplasmin.

Hephaestin is found in the membrane of enterocytes, while ceruloplasmin is the major copper transport protein in the blood. Hephaestin facilitates basolateral iron export from the intestinal epithelial cells by oxidizing the ferrous iron back to its ferric form and so it performs this function in a coupled manner (need to occur together) with transport through ferroportin. Ceruloplasmin, which is found in plasma, is also a ferroxidase and may be involved in the oxidation of ferrous iron to ferric iron during binding to transferrin. Once Fe2+ is oxidized, Fe3+ binds to transferrin and is transported to a tissue cell that contains a transferrin receptor. Transferrin (Tf) binds to the transferrin receptor(TfR) and is endocytosed [37] as shown in (**Figure 10**).

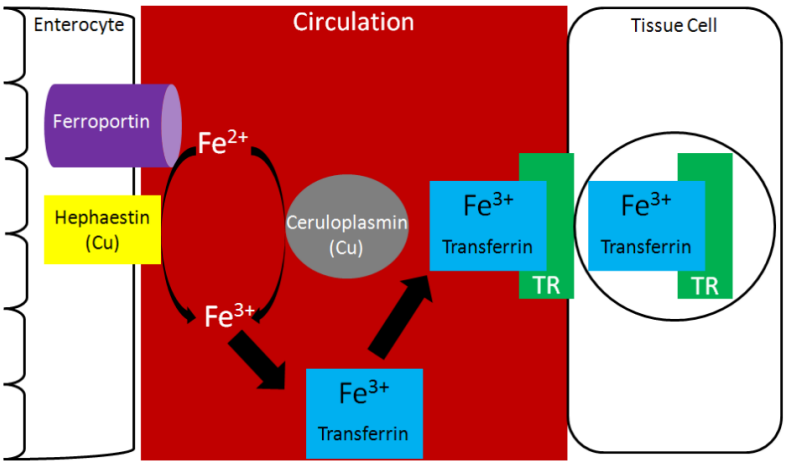


Figure 10: scheme that expresses the iron transport in the enterocyte (71).

3.2.3 The storage of iron:

The liver is the primary storage site in the body, with the spleen and bone marrow being the other major storage sites: 65% of iron is present in red blood cells, 20% is stored in the liver and it is stored too in smaller quantities in macrophages (10%), bone marrow (2%), muscles (10%), and enterocytes (0.05%).

Once inside cells, the iron can be used for cellular purposes (cofactor for enzyme, etc.) or it can be stored in the iron storage proteins ferritin or hemosiderin (**Figure 11**). Ferritin is the primary iron storage protein and is critical to iron homeostasis (72). Ferritin stores iron in its unreactive Fe3+ form and at higher concentrations, iron is also stored in hemosiderin.

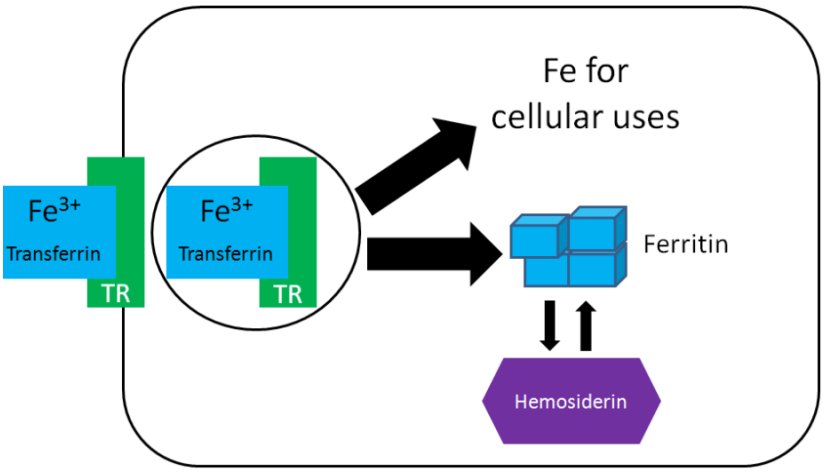


Figure 11: scheme showing how iron is being stored as ferritin or hemosiderin (71).

Haemosiderin is produced by lysosomal denaturation of ferritin(73,74), in which the protein shells degrade and the iron cores aggregate. Haemosiderin iron is found in lysosomes and cytosol and, as it is less soluble than ferritin iron, it is less easily mobilized.

3.2.4 Iron recycling and elimination:

- **Recycling :**

As previously stated, red blood cells carry the majority of the iron in the human body. When these cells reach the end of their lives, the iron they release is a key source of systemically accessible iron that may be utilized to make new erythrocytes in the bone marrow(75), so the needs are covered from senescent erythrocytes phagocyted by macrophages. Once phagocytosed, heme is degraded by a heme oxygenase, the iron is then released into the cytosol of the macrophages where it will be either stored and bound to ferritin, or exported to the outside of the cell via ferroportin and released into the plasma to be redistributed.

- **Excretion and losses :**

In healthy individuals, obligatory iron losses from the skin and gastrointestinal mucosa are thought to be approximately 1 mg/day in males (76), and slightly more in women of child-bearing age because of additional losses due to menstruation, pregnancy, and lactation.

Iron is not actively excreted from the body in urine or in the intestines. The majority of absorbed iron is lost in the feces. Daily iron losses from urine, gastrointestinal tract, and skin are approximately 0.08, 0.6, and 0.2 to 0.3 mg/day, respectively. These basal losses may drop to 0.5 mg/day in iron deficiency and may be as high as 2 mg/day in iron overload. The total amount lost is estimated at 14 µg/kg body weight/day.

3.3 Iron regulation

3.3.1 Systemic regulation of iron:

Iron levels in the blood must be kept at ideal levels for cells and tissues to operate properly. Too little iron, for example, can cause iron-restricted erythropoiesis and anemia, whereas too much iron can cause tissue iron overload and accompanying disorders, elaborate mechanisms have evolved to sense iron levels and modify iron absorption and recycling accordingly to maintain iron homeostasis, allowing the body to satisfy its requirements. Inflammatory/infectious stimuli, as well as hypoxia (reduced amount of oxygen in the tissues) and erythropoietic signals, cause these processes to decrease or increase iron availability. The mechanisms regulating systemic iron homeostasis are largely centered on the liver and involve two molecules, « hepcidin and ferroportin », that work together to regulate the flow of iron from cells into the systemic circulation.

The principal regulator of iron absorption is **hepcidin** (77) a small peptide of 20–25 amino acids encoded by the hamp (hepcidin antimicrobial peptide.) Gene, in vitro studies in macrophage cells have shown that hepcidin exerts its effects by directly binding to and degrading the iron exporter molecule“ferroportin”, on the cellular membrane; as a consequence, iron is prevented from leaving the cell (78). In fact, the mechanism of hepcidin action may be twofold and cell-type specific. Studies using in vitro and in vivo models have indicated that hepcidin inhibits iron export from macrophages by causing degradation of ferroportin; however, in enterocytes, it may also down regulate iron uptake by inhibiting dmt1 transcription (79).

When iron stores are excessive, hepatic hepcidin synthesis increases as well as during inflammation, because in this case, there is a synthesis of pro-inflammatory cytokines like IL-6, and these cytokines are responsible for an increase in its synthesis. The secreted hepcidin affects ferroportin and prevents iron transport from the enterocyte to plasma transferrin. the iron that is not transferred is sequestered within the enterocytes and is eventually lost in the gut lumen when the enterocytes are shed and lost in the feces. But when systemic iron needs are raised, iron reserves are low, or both, Hepcidin synthesis is reduced, permitting intestinal iron transfer and the release of iron from macrophage depots. In addition, hepcidin production is reduced by systemic hypoxia, which also stimulates the production of erythropoietin which, in turn, stimulates the production of red blood cells (erythropoiesis). The coincident depression of hepcidin, therefore, ensures a supply of iron needed for the synthesis of hemoglobin as part of the erythropoietic response.

Ferroportin is a cellular iron export protein. It is expressed most highly in macrophages, duodenal enterocytes, and hepatocytes. The gene was previously known under a variety of names, including SLC11A3, IREG1 (iron regulated-transporter-1), and MTP1 (metal transporter protein-1), The expression of ferroportin can be controlled at the transcriptional, translational, and post-translational levels. The ferroportin mRNA contains a functional IRE in its 5′UTR (Untranslated Transcribed Region) and, similar to H and L ferritin, its translation is repressed under iron-deficient conditions, with resultant reduction in cellular iron export.

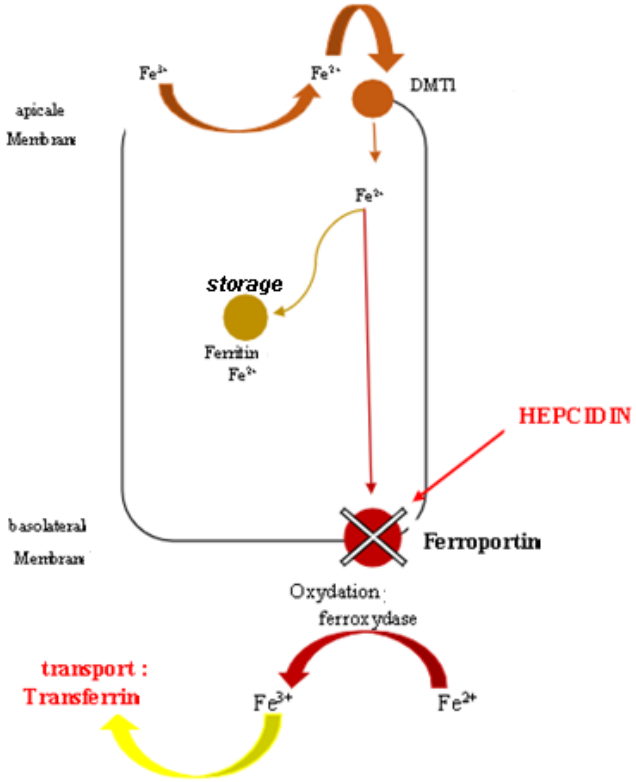


Figure 12: Hepcidin's action (80).

3.3.2 Cellular Iron Regulation :

On a cellular level, iron metabolism is primarily regulated by the iron-responsive element (IRE)/iron regulatory protein (IRP) system. Many proteins involved in iron metabolism have motifs called iron-responsive elements (IREs), which are stem-loop structures in their 3′ or 5′ untranslated regions.IRPs (IRP1 and IRP2, also named ACO1 and IREB2, respectively), cytoplasmic proteins belonging to the aconitase superfamily, regulate its metabolism by binding with high affinity and specificity to IRE, and thereby control the expression of mRNAs that encode for the proteins involved in iron uptake, storage, usage, and export, including TfR1(transferrin receptor1), DMT1, ferritin, hypoxia-inducible factor 2 (HIF2), and ferroportin (81).

Under conditions of iron deficiency, IRPs are active and bind to IRE and stabilize the mRNA for TfR1 while also decreasing translation of mRNA for ferritin eventually increasing iron uptake and availability within the cell. (Binding to five IREs in the 3′ untranslated region of TfR1 mRNA inhibits mRNA degradation, thereby increasing TfR1 expression and iron uptake. Binding to one IRE in the 5′ untranslated region of ferritin mRNA inhibits ferritin translation, thereby reducing cellular iron storage). Conversely, high iron levels decrease IRE-binding activity, leading to efficient translation of ferritin mRNA and decreased stability of TfR1 mRNA, IRP1 inserts a 4Fe-4S cluster, which converts it into a cytosolic aconitase, while IRP2 is targeted for proteasomal degradation,this ultimately enhancing iron sequestration over uptake (**Figure 13**).

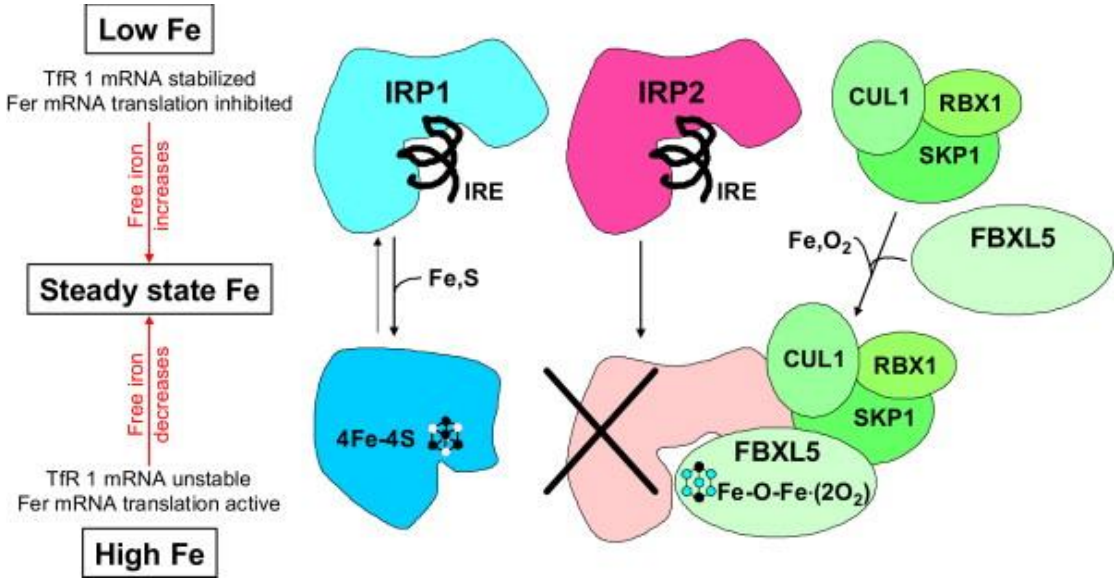


Figure 13: Mechanisms that control cellular iron homeostasis (82).

The scheme depicts the two iron regulatory proteins, IRP1 and IRP2, which are active RNA-binding proteins at low-free-iron conditions. They control post-transcriptionally the fate of mRNAs encoding the most essential proteins in iron metabolism, TfR1, and ferritin (Fer), which function in cellular iron import and storage, respectively, thus adjusting the free iron pool. Free iron contributes to the assembly of the 4Fe-4S cluster that inactivates IRP1 (light blue), converting it to a cytoplasmic aconitase (dark blue) and to the di-iron-oxygen center in the hemerythrin domain of FBXL5, which then binds IRP2 (fuchsia pink) and induces its degradation by the proteasomal pathway. Thus, free iron acts on its own level through these elaborate feedback loops. Its steady-state will equilibrate at the concentration required for the iron center synthesis.

3.3.2.1 Cellular Iron Regulation in The Heart:

Iron is essential to the cell. Both iron deficiency and overload impinge negatively on cardiac health. Thus, effective iron homeostasis is important for cardiac function.

Cardiomyocytes, like other cell types, have only one pathway for iron export and that is through Fpn1. However, there are several ways that iron can enter cardiomyocytes. This makes cardiomyocytes particularly vulnerable to iron overload. Hepcidin is also produced in cardiomyocytes. Cardiac hepcidin has important autocrine effects and participates in the autonomous regulation of iron in cardiomyocytes that are distinct from systemic iron regulation (83). In contrast to systemic hepcidin, the level of cardiac hepcidin increases in iron deficiency to preserve the cellular iron.

Iron uptake by cardiomyocytes primarily occurs through TfR1. Only Tf-bound iron can enter cardiomyocytes via this pathway. Mice lacking TfR1 in the heart die early from a cardiomyopathy that is associated with cardiac ID (84). Another important pathway for the influx of iron into the cardiomyocytes is through the divalent metal transporter 1 protein (DMT1), which mediates the import of non–transferrin- bound iron (NTBI). Inside the cell, iron can either be stored as ferritin, where it is redox inert; or go through biosynthetic pathways to generate heme or Fe/S clusters; or remain as labile iron. Under normal conditions, the level of labile iron is kept very low to prevent Reactive oxygen species (ROS) formation. However, pathological states of iron overload can dramatically increase the labile iron pool.

Cellular iron homeostasis is maintained by iron regulatory proteins (IRPs). When the cellular iron concentration is low, IRPs stabilize the messenger RNA of TfR1 and DMT1 to promote iron influx; at the same time, they inhibit messenger RNA translation of Fpn1 and ferritin to inhibit iron efflux and iron storage, respectively (85,86). Cardiac TfR1 and DMT1 are not regulated by systemic iron (85). In addition to the TfR1- and DMT1-mediated pathways, cardiomyocytes carry L-type and T-type calcium channels as well as zinc transporters, all of which are capable of transporting NTBI into the cardiac cells (85,87,88). Although the cellular regulatory mechanisms of iron in the heart can modify the import of Tf-bound iron via TfR1 and control the entry of NTBI via DMT1, the influx of NTBI into the cardiomyocytes through calcium channels and zinc transporters are not regulated by these cellular regulatory mechanisms (89).

4. Chapter Three: Iron Deficiency in Chronic Heart Failure

4.1 Prevalence:

Chronic heart failure is still a common and serious condition with a bad prognosis. Comorbidities (such as diabetes, COPD, chronic renal failure, and anemia) are frequently associated with it and can complicate the therapy and have a severe impact on outcomes. Even though iron deficiency is now recognized as an important comorbidity, it is still frequently overlooked or disregarded, if not associated with anemia. Although the two can coexist, iron deficiency is not always the cause of anemia in HF, and iron deficit can exist without producing anemia (90,91). Therefore, iron deficiency emerges as a new therapeutic target of chronic heart failure. The prevalence of ID varies depending on the definition used. A lot of studies have reported a high prevalence of iron deficiency—up to 30%–50%—in chronic heart failure, even in the absence of anemia (90). In a large study of 955 heart failure patients due to left ventricular systolic dysfunction were investigated for the prevalence of anemia and its cause and followed for a median of 531 days, they found that 43% of anemic patients and 15% of non-anemic patients had iron deficiency (92). In another study including 37 patients with advanced heart failure, iron deficiency anemia diagnosed by bone marrow aspiration was present in up to 73% of patients. (93). A more recent study reported an iron deficiency prevalence of 37% in 546 heart failure patients; importantly, the prevalence was 32% in patients without anemia (94), in another cohort of 1506 chronic heart failure patients, the prevalence of iron deficiency was 50%, including 45.6% among patients without anemia (90).

4.2 Diagnosis:

4.2.1 Biological examinations to assess iron status:

4.2.1.1 Bone marrow biopsy or aspiration:

The gold standard for evaluating iron stores in target tissues is a bone marrow biopsy or aspiration after a specific iron staining: the Prussian blue staining with potassium ferrocyanide (ferric ion (+3) in the tissue combines with the ferrocyanide form a bright blue pigment called 'Prussian blue" or ferric ferrocyanide) (95). It is used to assess the presence of nonheme-bound iron in the erythroblasts and the extracellular space by 2 independent analysts. The percentage of erythroblasts containing iron, that is, sideroblasts, reflects the amount of iron incorporated in the erythrocyte precursor cells and thus the functional availability of iron for erythropoiesis (96).

In normal conditions, 20% to 50% of the erythroblasts contain iron, 10% to 20% is considered low normal, and patients with sideroblasts <10% are considered functionally iron deficient (97). The iron stores are assessed as the amount of iron present in the extracellular space and graded using Gale histological grading method (98). It is an invasive and traumatic procedure. That gives semi-quantitative results. Phiri et al(99) proposed a histological grading by iron smear assessment with separate detection of iron in macrophages (stored iron) and erythroblasts (utilized iron), differentiating between a normal status, absolute ID, functional ID, and combined functional and absolute ID.

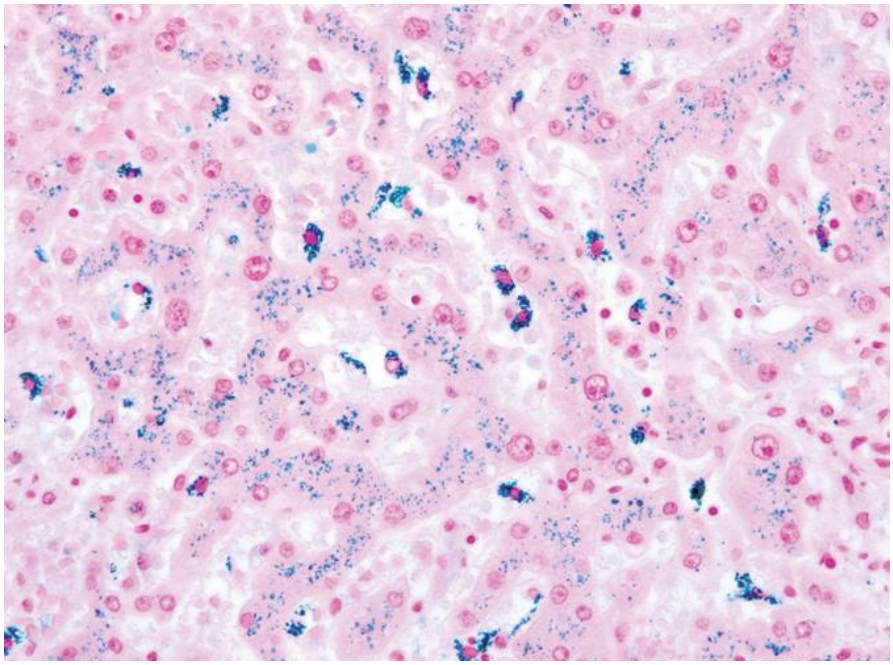


Figure 14: Bone marrow smear with iron accumulation (100).

4.2.1.2 Magnetic resonance imaging (MRI):

There is even, a more recently developed, non-invasive method for examining the iron stores in multiple tissues with magnetic resonance imaging (MRI), although the sensitivity remains an important issue. MRI does not image the iron directly but instead images water protons as they diffuse near iron deposits in the tissue of interest. The iron acts as little magnets, destroying the homogeneity of the magnetic field in iron-laden tissues. The moving water protons each experience significantly different magnetic profiles and become desynchronized from one another. This causes the image to darken at a rate proportional to the iron concentration(101).

However, the most used method today for the evaluation of iron's statue is the measurement of several blood biomarkers since each of the recent methods from before has its limitations.

4.2.1.3 Blood biomarkers:

The following tests can be performed on serum or plasma (heparin tube).

4.2.1.3.1 Serum Iron:

It measures the concentration of iron in the serum or plasma, nearly all of which is bound to transferrin. Serum iron shows significant intra-individual nycthemeral variations: several old observational studies carried out in anemic and non-anemic subjects, report that the amplitude during the day is 30% to 40% on average, according to a circadian cycle(102,103), it can increase after a meal rich of iron (red meat) or increase in iron overload, hepatitis and cirrhosis, chronic alcoholism, hemolysis, that's why serum iron alone should not be used as a marker of iron status (104).

Usual values: Males: 65–177 µg/dL (11.6–31.7 µmol/L).

Females: 50–170 µg/dL (9.0–30.4 µmol/L).

4.2.1.3.2 Serum Transferrin:

It measures the concentration of transferrin in the serum or plasma. There is a lesser physiological variation in transferrin values. Factors that can decrease transferrin levels are iron overload, hepatocellular insufficiency, nephrotic syndrome, and inflammatory syndrome by increased catabolism.

The concentration of transferrin is increased by iron deficiency but also by oral contraceptives (105), and during pregnancy too.

Usual values: Transferin (adults): 1.6 to 3,2 g/l.

TIBC: 250–370 μg/dL (45-66 μmol/L)

4.2.1.3.3 Transferrin saturation (TSAT):

It is calculated by dividing the serum iron concentration by the total iron-binding capacity (TIBC). It reflects an estimate of how many transferrin iron-binding sites are occupied (a marker of the availability of circulating iron to supply metabolizing cells).

TSAT (%) = [Serum iron (μg⁄dL) / TIBC (μg⁄dL)]× 100

TIBC = $[Tf]\ X\ 25$

Usual values: Males: 20–50%.

Female: 15–50%.

4.2.1.3.4 Serum Ferritin:

The ferritin levels measured usually have a direct correlation with the total amount of iron stored in the body. Usually, 1 μg/l of serum ferritin corresponds to approximately 10 mg of iron in reserve. However, ferritin is an acute-phase reactant and elevates in response to processes that do not correlate with iron status, including inflammation, chronic disease, malignancy, and infection.

Usual values: They vary in literature depending on age and sex:

Males: 20-250 μg/mL.

Females: 15-150 μg/mL.

Serum ferritin is the primary diagnostic parameter (106). As previously mentioned in the first chapter, the 2016 ESC HF guidelines recommend treating iron deficiency based on a serum ferritin level < 100 μg/L, or 100–299 μg/L when TSAT <20% here Two different cut-offs are used because ferritin levels may become elevated in the presence of inflammation and may, therefore, appear to be within the normal range (100–300 μg/L), As ferritin, the TSAT levels <20 % is what indicate the functional iron deficiency(107). If the first assessment of iron status is normal, the interval between the next iron tests should be 6 months for NYHA functional classes III-IV and 1 year for NYHA functional classes I—II.

A low ferritin level (< 15 μg/L) is a pathognomonic sign of iron deficiency.

Following a normal ferritin level, a CRP test should be performed. Normal ferritin and CRP concentrations exclude an iron deficiency.

For intermediate ferritin values coexisting with an increased CRP, the soluble transferrin receptor dosage can be proposed.

4.2.1.3.5 Soluble transferrin receptors sTfRs :

The concentration of sTfR is an indicator of iron status. STfRs are proteins found in the blood that are cleaved from the membrane-bound transferrin receptors found on nearly all cells. The number of transferrin receptors found on the surface of cells correlates with the level of iron within cells. Iron repletion results in decreased sTfR levels while iron deficiency causes overexpression of transferrin receptor and sTfR levels. As more receptors are produced, more are cleaved from cell surfaces and enter the blood, increasing the level of soluble transferrin receptors. Thus, measuring the level of sTfR is one

way of evaluating the amount of iron available in the body. The human soluble transferrin receptor (sTfR)/log ferritin index is intended as an aid in the diagnosis of iron deficiency anemia (IDA) and for the differential diagnosis of IDA and anemia of chronic disease (ACD).

In contrast to ferritin, sTfR is not an acute-phase reactant and the interpretation of iron status using sTfR measurement is not affected by inflammation and other conditions. sTfR is elevated in patients with thalassemia and sickle cell disease. Caution should be exercised in managing anemia in these individuals based on the sTfR test results (108).

Usual values: 1.8-4.6 mg/L.

It needs to be acknowledged that there are attempts to develop more precise definitions of ID based on experimental biomarkers such as the combined assessment of serum hepcidin (correlates with iron stores more precisely than ferritin) and soluble transferrin receptor (sTfR) (109,110).

Note: Mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and MCH concentration (MCHC) have been found to be unreliable markers of iron deficiency status. Measuring their levels is not recommended for the assessment of iron deficiency in patients with HF. In addition(111). Measuring their levels is not recommended for the assessment of iron deficiency in patients with HF.

4.3 Diagnosis of ID in heart failure patients:

The 2016 ESC HF guidelines (3) recommend that the evaluation of iron status should be considered in the diagnostic work-up of all newly diagnosed HF patients. Iron status should also be evaluated in patients with existing CHF, particularly if they are symptomatic despite receiving optimal background HF medications. In addition, as part of routine follow-up, consideration should be given to the re-evaluation of iron status one to two times per year, as well as after hospitalization for HF. It is also recommended to assess hemoglobin levels in HF patients. The diagnosis of iron deficiencies is more complex when associated with underlying chronic conditions like HF. Two types of ID need to be distinguished: absolute, and functional ID.

4.3.1.1 Absolute Iron Deficiency:

Absolute iron deficiency is defined as depleted systemic iron stores and is characterized by low or absence of stainable iron in the bone marrow (112,113). According to 2016 ESC guidelines, absolute ID refers to ferritin levels under 100 ng/ml and TSAT levels under 20%. It is also associated with low circulating hepcidin levels as well as elevated serum levels of sTfR (114). Iron-depleted stores result in a major tissue avidity for iron and require a greater expression of sTfR to maximize the internalization of iron in peripheral cells (109). If low serum hepcidin is representative of depleted iron stores, a high level of sTfr is a parameter of metabolically unmet cellular needs and depleted utilized iron. The most profound iron deficiency can be identified with both low serum hepcidin and high plasma sTfR levels, whereas low serum hepcidin but normal sTfR plasma level reflects a situation of bone marrow depleted iron stores with still satisfied metabolic cellular iron needs. However, this status stands for a greater future probability of developing severe iron deficiency (112).

4.3.1.2 Functional iron deficiency:

It occurs when peripheral iron availability is inadequate despite an adequate iron pool. The cut-off values for functional ID, according to 2016 ESC guidelines (3), are ferritin levels between 100-299 ng/ml and TSAT under 20%. It is associated with high sTfR levels (reflecting low tissue utilized iron) (115).

4.4 Etiologies:

Iron deficiency in chronic heart failure has a multifactorial genesis. However, the exact etiologies of negative systemic iron balance in the course of HF are still not fully understood (116).

4.4.1 Absolute iron deficiency:

The following mechanisms are presumed to be involved in the development of absolute ID in HF: insufficient dietary iron intake (117,118), intestinal malabsorption and chronic gastrointestinal bleeding (iron loss), very frequent in cardiologic patients, not only for gastrointestinal pathological condition but also drug-induced, such as in oral anticoagulant.

- **Insufficient iron dietary intake:**

HF is accompanied by many nutritional deficiencies, including dietary iron (119). Based on a 4-day food diary, Hughes et al. (117) showed that 46% of patients with stable HF consumed less iron than the dietary reference value, and average daily iron intake was markedly reduced in patients in NYHA class III–IV when compared with NYHA class II. Lourenço et al. (118) assessed the nutritional status using an interview by nutritionists in 125 outpatients with stable HF, and in 12–35% found an inadequate dietary iron intake (113).

- **Iron intestinal malabsorption:**

Proton pump inhibitors can also be responsible for further reduction in iron absorption because of the decreased gastric acidity level. Duodenal wall edema, frequently associated with acute and chronic heart failure, can play a role in the reduced absorption of iron by duodenal enterocytes (120,121). Theoretically, reduced absorption may result from increased circulating hepcidin levels, analogous to a reported experimental model of chronic kidney disease (113).

4.4.2 Functional iron deficiency:

In several chronic diseases (not only HF but also chronic renal failure, chronic obstructive pulmonary disease, and diabetes mellitus…) deranged iron status is attributed to the magnitude of systemic inflammation and circulating proinflammatory mediators, such as interleukin 6 (122–124). However, in patients with HF, the complex pathophysiological interplay between chronic low-grade inflammation and systemic iron status is not fully understood. Moreover, some iron biomarkers play a role in the immune system and response, for example, ferritin, which is an acute-phase reactant. From this point of view, interesting data have been demonstrated for hepcidin, which is not only the key iron regulator but also an element of an innate immune response (125). However, in stable patients with heart failure with reduced ejection fraction (HFrEF), hepcidin correlates neither with low hemoglobin levels nor inflammation and is increased only in the early stages of the disease (126).

Although ID is considered an “energetic insult” for tissues utilizing high amounts of energy, the complex interplay between systemic, intercellular, and mitochondrial ID and iron overload is still not fully understood, in particular in cardiomyocytes and the heart as a whole. Additionally, it remains to be established how inflammation, a hallmark of cardiovascular disease and a known regulator of hepcidin, impinges on the cardiac hepcidin/ferroportin axis, and on cardiomyocyte iron levels (127).

Table 8 shows the relevant Cut-offs for the Diagnosis of ID in Different Fields of Internal Medicine.

Table 8: Relevant cut-offs for the Diagnosis of ID in Different Fields of internal medicine.							
Study name (Ref. #)	FERRIC-HF (Okonko et al 2008) (128)	Toblli et al(129) 2007	FAIR-HF (130) (Anker et al 2009)	CONFIRM-HF (131)(ponikowski et al 2015)	EFFECT-HF (41) (van veldhuisen et al 2017)	IRON5(132,133)	IRONOUT(134) (Lewis et al2017)
Diagnosis	HFrEF	HFrEF	HFrEF	HFrEF	HFrEF	HF	HFrEF
Number of patients	35	40	459	304	172	54	225
Randomization	2:1 (Iron:placebo)	1:1 (Iron:placebo)	2:1 (FCM:placebo)	1:1 (FCM:placebo)	1:1 (FCM:standard of care) 1	1:1(iron « Ferrous Sulfate oral capsule » :Placebo)	1:1 (FCM:placebo)
Blinding	Double-blind	Double-blind	Double-blind	Double-blind	Open-label	Double-blind	Double-blind
Symptoms	NYHA functional class II-III	NYHA functional class II-IV	NYHA functional class II-III	NYHA functional class II-III	NYHA functional class II-III	NYHA functional class II-III	NYHA functional class II-IV
LVEF	<or=45%	<or =35%	<or =40% in NYHA functional class I, <or =45% in NYHA functional class III	<or =45%	<or =45%	<50%	< or =40

Definition of iron deficiency	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%	TSAT <20% ferritin<100ng/ml	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%	Serum ferritin <100ng/ml, or100–300 ng/ml with TSAT <20%	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%
Hemoglobin	<12.5 g/dl [anemic group] vs. 12.5 to 14.5 g/dl [nonanemic group]	<12.5 g/dl	<or=9,5 to <or =13,5 g/dl	< 15 g/dl	<or=15 g/dl	>8 and[< 13 gr/dL (if male), < 12 gr/dL (female)]	>9 to <or =13.5 g/dl
Natriuretic peptides	Not included	NT-pro-brain natriuretic peptide (NT-proBNP	Not included	BNP >100 pg/ml, NT-proBNP >400 pg/ml	BNP >100 pg/ml, NT-proBNP >400 pg/ml	NT-proBNP >4,000 pg/ml	NT-pro BNP
Duration)	16 week	25 weeks	24 weeks	52 weeks	24 weeks	90 days	16 week
Dosage	Iron sucrose 200 mg weekly until ferritin >500 ng/ml	Iron sucrose 200 mg weekly for 5 weeks	200 mg ferric carboxymaltose	an initial single dose of 1,000 mg iron as FCM (or 500 mg for those with Hb > 14 g/dL).	500 mg ferric carboxymaltose 3 times	Ferrous Sulfate 200 mg 3 times daily for 90 days	Oral Iron 150 mg Polysaccharide iron complex (Feramax) twice daily

Primary end point	Change in absolute pVO2 (ml/min) from baseline to week 18	Change in the NT-proBNP level and inflammatory status by CRP	Change in self reported PGA score and NHYA class from baseline to week 24	Change in 6-min walk distance from baseline to week 24	Change in peak VO2 from baseline to week 24	Change in 6MWT from baseline to 90 days	change in peak oxygen uptake (V̇o2) from baseline to 16 weeks
conclusion	Intravenous iron loading improved exercise capacity and symptoms in patients with CHF and evidence of abnormal iron metabolism. Benefits were more evident in anemic patients	Intravenous iron therapy without rhEPO substantially reduced NT-proBNP and inflammatory status in anemic patients with CHF and moderate CRF. This situation was associated with an improvement in LVEF, NYHA functional class, exercise capacity,	Treatment with intravenous ferric carboxymaltose in patients with chronic heart failure and iron deficiency, with or without anemia, improves symptoms, functional capacity, and quality of life; the side-effect	Treatment of symptomatic, iron-deficient HF patients with FCM over a 1-year period resulted in sustainable improvement in functional capacity, symptoms, and QoL and may be associated with risk reduction of hospitalization for worsening H	Treatment with intravenous FCM in patients with HF and iron deficiency improves iron stores. Although a favorable effect on peak VO2 was observed on FCM, compared with standard of care in the primary analysis, this effect was	Trial was terminated	among participants with HFrEF with iron deficiency, high-dose oral iron did not improve exercise capacity over 16 weeks. These results do not support use of oral iron supplementation in patients with HFrEF

		renal function, and better Qol	profile is acceptable		highly sensitive to the imputation strategy for peak VO2 among patients who died. Whether FCM is associated with an improved outcome in these high-risk patients needs further study.		

Table is continued bellow : The second part is about some of the anticipated studies :

Study name (Ref. #)	FAIR-HFpEF (135,136)	FAIR-HF 2(137)	AFFIRM-AHF(138)	HEART-FID(139)	Differential Gene Expression in Patients With Heart Failure and Iron Deficiency - Effects of Ferric Carboxymalto(140)	IRONMAN(141)	Ferric Carboxymaltose to Improve Skeletal Muscle Metabolism in Heart Failure Patients With Functional Iron Deficiency(142)
Diagnosis	HF**p**EF	HFrEF	AHF after restabilization	HFrEF	HFrEF	HFrEF	HFrEF
Number of patients	200	1200	1100	3014	20	1300	32
Randomization	1:1 (FCM:placebo	1:1 (FCM:placebo	1:1 (FCM:placebo	1:1 (FCM:placebo	1:1 (FCM:placebo	1:1 (iron:placebo)	1:1 (FCM:placebo
Blinding	Double-blind	Double-blind	Double-blind	Double-blind	Double-blind	Open label	Double-blind
Symptoms	NYHA functional class II-III	NYHA functional class II-III	NYHA functional class II-III	NYHA functional class III-IV	NYHA functional class II-III	NYHA functional class II-IV	NYHA functional class II-III
LVEF	>or=45% together with evidence of diastolic dysfunction	<or=45%	<50%	<or=35%	<or=40%	<45%	<40% or >or=40% with left atrial volum index >28 ml/m2) and/or

							left ventricular mass index >95 g/m2 (women) or >115 g/m2(men)
Definition of iron deficiency	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%	Serum ferritin <100ng/ml, or100–300 ng/ml with TSAT <20%	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%	Serum ferritin <100ng/ml, with TSAT <20%	Serum ferritin <100ng/ml, or100–299 ng/ml with TSAT <20%
Hemoglobin	>9 to <or=14 g/dl	(>or=)9.5 to < or=14 g/d	>8to<or=15 g/dl	>9.0 and<13.5 g/dl (women) or<15.0 g/dl (men)	9.5-13.5 g/dl	>9.0 and<13.5 g/dl (women) or<15.0 g/dl (men)	< 13 gr/dL (if male), < 12 gr/dL (female)]
Natriuretic peptides	BNP >100 pg/ml, NT-proBNP >400 pg/ml, MR-proANP >125pmol/l	Not included	Not included	NT-proBNP >600 pg/ml or BNP >200 pg/ml for patients with normal sinus rhythm or NT-proBNP >1,000 pg/ml (or BNP	Not included	NT-proBNP >250 ng/l in sinus rhythm or >1,000 ng/l in atrial fibrillation (or BNP of >75 pg/ml or 300 pg/ml, respectively	Not included

				>400 pg/ml) for patients with atrial fibrillation			
Duration	52 weeks	Event driven	52 weeks	Event driven	12 weeks	120 weeks	4 weeks
Dosage	500–2,000 mg ferric carboxymaltose according to Hb and weight value	500–2,000 mg ferric carboxymaltose according to Hb and weight value	500–1500 mg ferric carboxymaltose according to Hb and weight value	500–2,000 mg ferric carboxymaltose according to Hb and weight value	500–2,000 mg ferric carboxymaltose according to Hb and weight value	iron (III) isomaltoside 1,000	750 mg ferric carboxymaltose
Primary end point	Change in 6-min walk distance from baseline to week 24	Combined rate of recurrent hospitalizations for HF and of cardiovascular death from baseline to at least 12 month of follow-up	HF hospitalizations and cardiovascular death up to 52 weeks after randomization	Incidence of death and incidence of hospitalization for heart failure at least 12 month of follow-up and change in 6MWT distance after 6 months	Evaluate the effect of ferric carboxymaltose on mitochondrial gene activation pattern after12 weeks of treatment	Cardiovascular mortality or hospitalization for worsening heart failure	Change in skeletal muscle mitochondrial oxidative capacity
conclusion	Estimated Study Completion Date :July 2021	Estimated Study Completion :December 2021	Actual study completion date : July 21, 2020	Estimated completion : March 2023	Still in the phase of recruitment /no updates till now	Estimated completion : March 2022(still recruiting)	Estimated completion August 2021

4.5 Clinical and prognostic consequences of iron deficiency in heart failure patients:

Iron deficiency was labeled clinically as relevant, in relation to anemia traditionally. However, the current and prevailing point of view is to consider the anemia, as the end of a process that began much earlier, with the gradual depletion of iron deposits shown in the physical capacity and tolerance to daily activities.

4.5.1 Iron deficiency and exercise intolerance in heart failure:

Iron is a strategic micronutrient, which is directly involved in oxygen transport (as a hemoglobin component of 'heme structure'), oxygen storage (as a myoglobin component), muscular energetic metabolism, and mitochondrial function, as a cofactor and catalyst for oxidative phosphorylation enzymes and respiratory chain proteins (in cytochromes) (112).

Animal models have convincingly demonstrated a causal connection between iron deficiency and physical labor capability. The effects of iron deficiency (with and without anemia) on aerobic capacity, endurance capacity, physical performance, and work efficiency have been established in humans in numerous studies, in patients with HFrEF, ID is associated with impaired aerobic performance, as reflected by lower peak oxygen consumption (VO2), higher ventilatory response to exercise (VE-VCO2 slope)(143,144) and lower 6-min walk test (6MWT) distance. Importantly, in patients with HFrEF, the impact of ID on both peak VO2 and VE-VCO2 slope is independent of and much stronger than the effect of anemia alone (145).

4.5.2 Iron deficiency and depression symptoms in heart failure:

Iron deficiency carries also a risk of depression in men with systolic HF. Moderate depression by beck depression inventory (BDI) (≥16 points) was more prevalent (48 vs. 25%), and the lack of depression symptoms (BDI,10 points) was less common (13 vs. 51%) in men with ID than in those without ID. Iron deficiency was associated with more severe depression symptoms, irrespective of HF severity, neurohormonal activation, hemoglobin, and inflammation (113).

Iron deficiency also had consequences on cognitive performance(concentration, intelligence, memory, psychomotor skills, and scholastic achievement), emotions, fatigue, and behavior; moreover, iron supplementation improved cognition and exercise performance in iron-deficient patients (146,147).

4.5.3 Iron deficiency and prognosis in heart failure:

The prognostic impact of ID in HF patients was investigated in two observational prospective studies(94,148). Varma et al. (148) investigated 120 consecutive patients with systolic dysfunction (LVEF ≤45%) undergoing percutaneous coronary intervention with a median follow-up of 30 months. They demonstrated that anemia accompanied by ID strongly predicted cardiac mortality (33 vs. 1% in non-anemics), malignancy-associated anemia was related to high-non-cardiac mortality (57 vs. 4% in non-anemics), whereas anemia of chronic disease predicted neither cardiac nor non-cardiac death (148). Among 546 patients with systolic HF we found that ID was a strong independent predictor of death and heart transplantation during a 3-year follow-up. The presence of ID increased the risk of a poor outcome by 60% during the 3-year follow-up (94). A prospective study in 157 chronic heart failure patients showed that non-anemic, iron-deficient patients had a twofold greater risk of death than anemic, iron-replete patients(144).

4.5.4 Iron deficiency and dysfunction of myocardium and skeletal muscle

Mechanisms underlying links between ID and poor clinical status, exercise intolerance, and an unfavorable outcome in HF remain unclear. Dysfunction of both the myocardium and skeletal muscles

are at the center of the pathophysiology of HF (149–151). These organs have high energy demands, and their function is independent on intact iron metabolism.

4.6 Anemia in heart Failure

4.6.1 Prevalence:

The prevalence of anemia in heart failure varies from one study to another: from 13.5 to 57%, depending on the hematological definition criteria used and the characteristics of the patients included (152–157). However, it can be estimated at approximately 35% in patients hospitalized for systolic heart failure (158). It increases with the severity of the disease and with age. It is therefore more frequent in patients with NYHA stage III or IV and in hospitalised patients. In contrast, it is less frequent in ambulatory patients, found in 15 to 35% of them. It may also be less frequent in dilated cardiomyopathy than in ischemic heart disease and in Africans (159).

4.6.2 Causes:

The etiopathogenesis of anemia in heart failure is multifactorial (93,160,161). Iron deficiency is involved in approximately 50% of anemias found in this disease (159). The other causes of deficiency are dominated by a lack of vitamin B12 or B9, which may be the result of malnutrition or alcoholism. Renal failure, which results in a decrease in renal erythropoietin (EPO) production, also plays an important role (162,163). Renal failure is particularly common in heart failure, where the plasma hemoglobin concentration is significantly related to glomerular filtration rate (164). Inflammatory processes are also involved, with pro-inflammatory cytokines causing resistance to the bone marrow action of EPO (165). In addition, malnutrition promotes bone marrow dysfunction. Thus, a deficiency in the bone marrow action of EPO is involved in approximately 70% of anemias in heart failure**.**

4.6.3 Diagnosis:

Anemia is defined by the world health organization (WHO) as a hemoglobin level of less than 13 g/dl in men or 12 g/dl in women. In theory, hemoglobin concentration should be determined at a distance from hospitalization for cardiac decompensation, after signs of fluid retention have disappeared, to rule out false anemia due to hemodilution. In case of doubt, a second hemoglobin test should be carried out at a distance. A distinction must be made between severe anemia, defined by the hemoglobin of less than 9 g/l, which requires specific diagnostic and therapeutic measures independent of heart failure, and moderate anemia, which is part of the chronic disease that is heart failure. The etiological tests include, in addition to iron assessment and the estimation of the glomerular filtration rate, the calculation of creatinine clearance, reticulocytes, thyroid-stimulating hormone and vitamins B9 and B12.

It may be guided by the mean corpuscular volume (MCV) value:

- **Normocytic anemias** (MCV: 80-100 fl): are most often associated with renal failure. If they are regenerative (reticulocytes > 120,000/mm3), they may be the consequence of acute hemorrhage or hemolysis with increased lactate dehydrogenases, decreased haptoglobin and the presence of schizocytes (fragments of red blood cell).
- **Microcytic anemias** (MCV < 80 fl): are mostly iron deficiency anemia, resulting in either from absolute iron deficiency or functional iron deficiency. Much more rarely, serum iron levels may be elevated, and hemoglobin electrophoresis may show thalassemia.
- **Macrocytic anemia** (MCV > 100 fl): are the rarest and may be the consequence of a B9 or B12 vitamin deficiency, hypothyroidism, liver disease or alcoholism.

This etiological workup frequently finds a cause justifying specific treatment. In other cases, the anemia of heart failure is multifactorial, linked to renal failure and the inflammatory syndrome which almost systematically accompany this disease in its advanced stages. We speak then anemia of chronic disease.

4.6.4 Consequences:

Anemia worsens the symptomatology of heart failure patients by reducing the supply of O_2 to the tissues, thus promoting asthenia. As the heart of heart failure patients has limited capacity to adapt (increased circulatory output and arteriovenous O_2 difference), the deleterious effects of anemia are manifested at higher hemoglobin levels than in healthy subjects. There is a correlation between peak VO_2 and distance covered in the 6-minute walk test and hemoglobin concentration. Furthermore, anemia is a factor of decompensation in heart failure patients. To compensate for the peripheral vasodilatation resulting from tissue hypoxia, it causes a volumetric overload (**Figure 15**) increasing myocardial parietal stress which may worsen ventricular dysfunction. In anemic patients without heart failure, there is an inverse correlation between hemoglobin and natriuretic peptide concentrations Furthermore, if coronary artery disease is the cause of the heart disease, it aggravates the underlying myocardial ischemia. Anemia could thus be involved in 13% of hospitalizations of patients with heart failure. Finally, anemia could have a negative prognostic impact. Heart failure patients with anemia have a higher risk of death or hospitalization. However, not all studies agree that anemia is an independent prognostic effect, as most did not adjust for associated comorbidities and their severity. In addition, there is no direct relationship between hemoglobin concentration and mortality in heart failure, but a U-shaped relationship. In the ELITE II study, a hemoglobin value between 14.5 and 15.4 g/dl is associated with the best prognosis, whereas the risk of death is higher in patients with polycythemia. Thus, it is still unclear whether anemia is a worsening factor in heart failure or simply a risk marker (159).

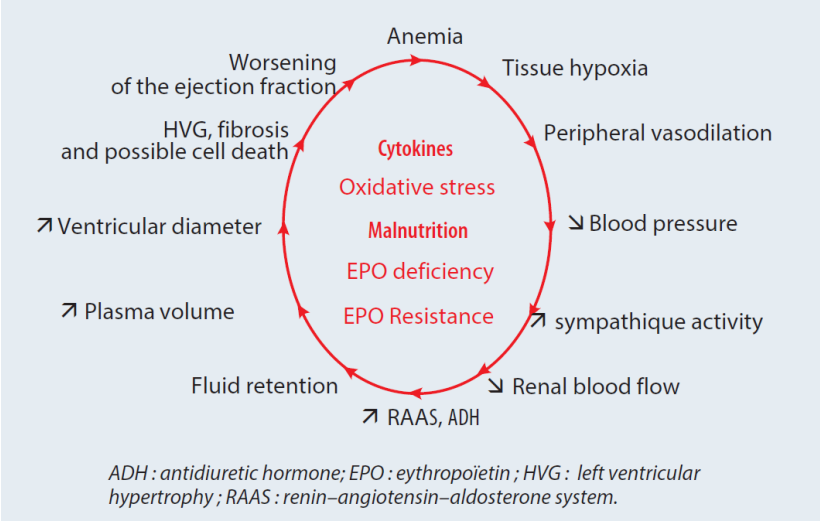


Figure 15: Reciprocal worsening of anemia and heart failure (159)**.**

4.7 The History of Research studies of ID in HF:

Patients with HF are affected by severely reduced exercise capacity and quality of life. On the lookout for new ways of improving exercise capacity, researchers from Tel Aviv started in the late 1990s to treat anemic patients with HF with a combination of subcutaneous erythropoietin and intravenous (IV) iron sucrose, they conclude in the end that The correction of the anemia is associated with an impressive improvement in cardiac function that is reflected in a marked improvement in the NYHA functional class, an improvement in renal function and a striking reduction in hospitalizations (166) these findings sparked an avalanche of research into the treatment of anemia in HF culminating in a program of large trials of the novel long-acting erythropoietin derivative darbepoetin-alf (Aranesp®) which is a hyperglycosylated EPO (erythropoietin) analogue designed for prolonged survival in the circulation and

with consequent greater bioavailability than epoetins that was was developed by Amgen. Darbepoetin-alfa was approved by the FDA and the EMA in 2001(167).

The anemia treatment pathway was ultimately left only after a large trial of darbepoetin-alfa ended in disappointment. In retrospect, it can be said that the early study design, consisting of a combination therapy of erythropoietin and IV iron, lead to the discovery that ID treatment may be worthwhile even in the absence of anemia.

4.8 Treatment of iron deficiency in patients with heart failure:

4.8.1 Historical considerations in Iron therapy:

Iron therapy dates back to the seventeenth century, when Thomas Sydenham (1624–1689) first proposed the use of oral iron salts for the treatment of "chlorosis", although the disorder was initially believed as a hysterical problem rather than due to IDA(168).

The first iron compound to be used for intravenous (IV) route (iron saccharide) entered the clinical scenario near to the second half of the past century(1947), followed by High-Molecular Weight Dextran (HMWD) iron, Despite documented success in correcting IDA, rare cases of severe hypersensitivity reactions were reported, some of them being fatal. This led to extreme caution in prescribing IV iron, which was deemed to be reserved only for conditions where oral iron could not be used(169).

The medical community experienced a long-lasting generalized prejudice against IV iron, whatever the preparation used. Only relatively recently, it was realized that severe and potentially lethal reactions were almost exclusively due to HMWD-iron (170)which, in the meantime, was no longer produced since 1992 and replaced by other preparations (**Figure 16**).

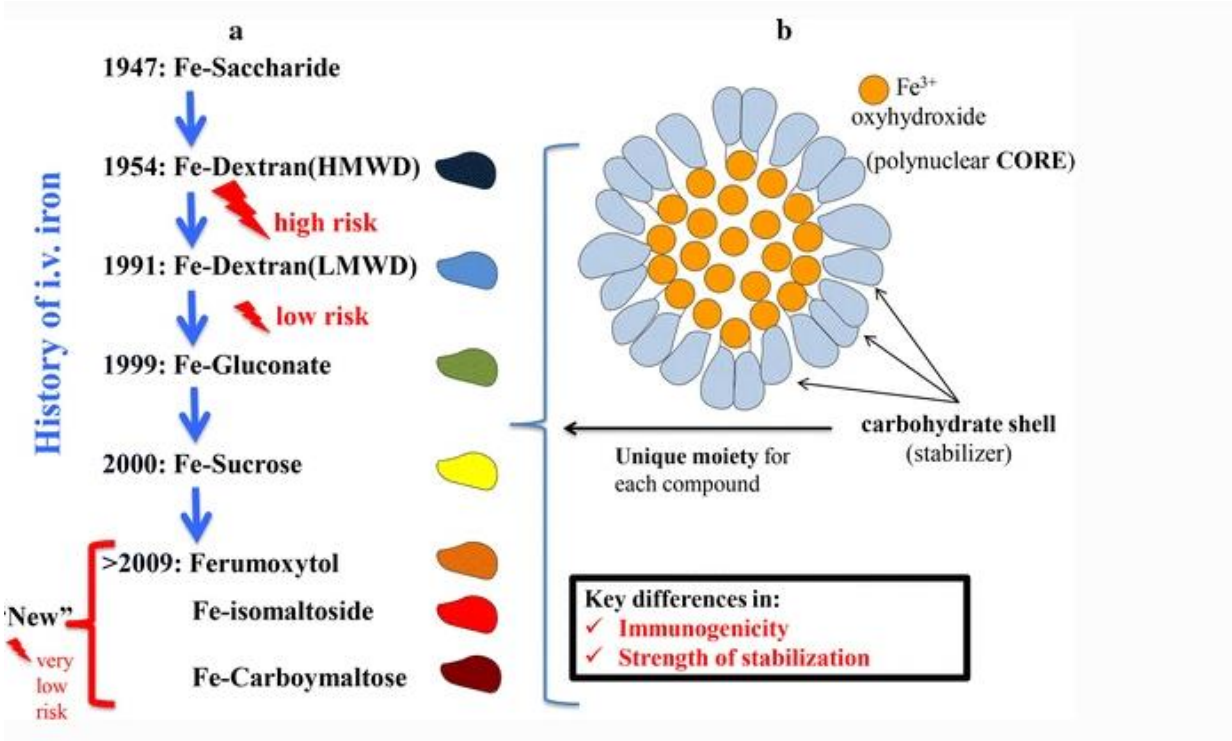


Figure 16: IV iron preparations: historical and chemical perspective (171).

a: A widespread use of IV iron has been historically hampered by unacceptable risks with early preparations, particularly anaphylactic reactions with high molecular weight dextran (HMWD) iron.

b : All IV iron preparations consist of an iron polynuclear core surrounded by a carbohydrate shell that acts as a stabilizer, preventing uncontrolled release of toxic free iron

Both oral and IV traditional iron formulations are known to be far from ideal, mainly because of tolerability and safety issues, respectively. From a pharmacological point of view, iron replacement therapy has progressed relatively slowly until recently. At the beginning of this century, concomitantly with the pathophysiological advances mentioned above, improvements in pharmaceutical technologies have allowed the production of newer iron formulations, particularly for IV administration, aimed at minimizing the problems inherent with traditional compounds. Noteworthy, the pharmacokinetics of oral iron is completely different from that of IV iron. Oral iron is incorporated into plasma transferrin after release from the basolateral membrane of intestinal cells, providing that there is no condition leading to malabsorption (i.e. celiac disease, autoimmune ..). By contrast, IV iron compounds are first taken up by macrophages and then released into the bloodstream. As the two treatments cannot be considered merely interchangeable and have different indications (**Figure 17**).

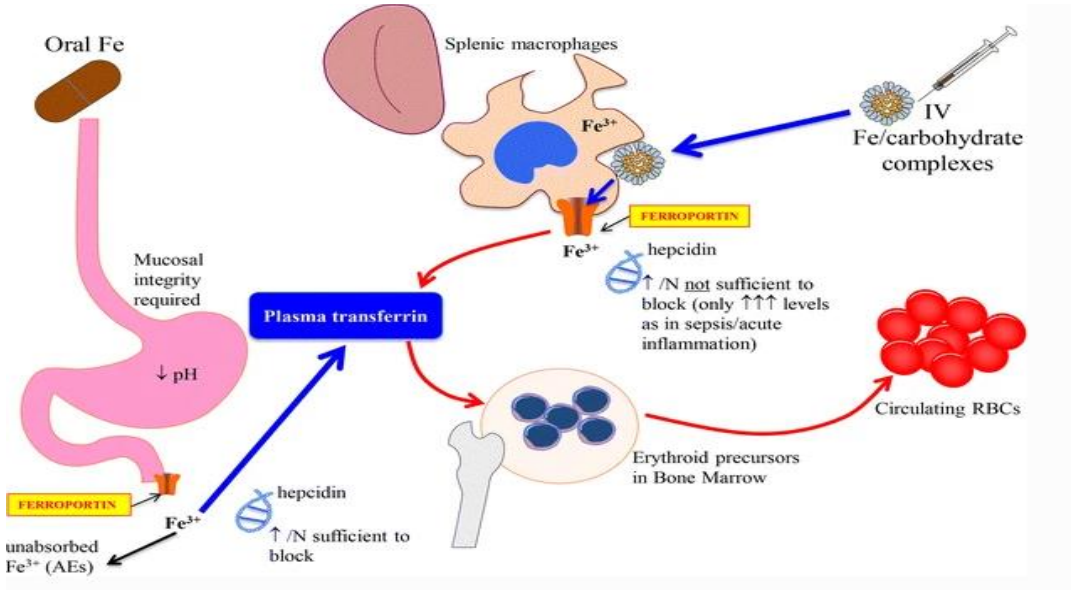


Figure 17: The pharmacokinetics of IV and Oral iron (171).

Pharmacokinetic of oral iron requires the integrity of the mucosa of the stomach (acidity is needed to solubilize iron) and duodenum/proximal jejunum (where most of the iron is absorbed). This integrity can be compromised by several conditions leading to malabsorption and the unabsorbed iron is mainly responsible for gastrointestinal adverse effects.

IV iron has a completely different one, The iron-carbohydrate complexes(read Parenteral Iron Supplementation bellow for more informations**)** are rapidly taken up by macrophages, then iron atoms of the core are slowly released in the circulation through ferroportin. Both oral and IV iron requires ferroportin to be released in the plasma. Hepcidin production is typically suppressed in uncomplicated IDA, allowing maximal iron absorption. However, slightly elevated (or even inappropriately normal) hepcidin levels appear sufficient to inhibit intestinal ferroportin. This can be due to a genetic disorder (IRIDA, Iron-refractory iron deficiency anemia) to concomitant low-grade inflammation (eg: in chronic heart failure), or even to transient stimulation after a first dose of oral iron. This constitutes the basis for the current recommendation of using oral iron on an alternate day schedule instead of the classical daily schedule (see the text for details). On the other hand, macrophage ferroportin, whose expression is much higher than at the intestinal level, requires much more elevated hepcidin levels (i.e. like during acute inflammation) to be substantially suppressed.

4.8.2 Oral Iron Supplementation :

In healthy persons, an overall iron absorption depends on several factors as mentioned before. In the general population, oral iron supplementation is usually **''the first-choice therapy''** for iron deficiency **anemia**, but frequently the response to the treatment is suboptimal. It is a reasonable option for healthy subjects without absorption disorders, in whom ID is usually mild and rapid replenishment is not necessary. The tolerance of iron differs depending on the used oral iron formulation, there are two forms of absorbable iron: ferrous (Fe^{2+}) and ferric (Fe^{3+}). Due to lower solubility, ferric iron is less bioavailable than ferrous iron (ferrous products are effective, but they are associated with more gastrointestinal side effects than ferric products and ferric products tend to have lower absorption comparison). **Ferrous fumarate, ferrous sulfate, and ferrous gluconate** are the major types of ferrous iron supplements, with comparable bioavailability (172,173), the gold standard of oral iron treatment is ferrous sulfate. The recommended therapeutic dosage ranges from 150 to 180 mg/day of elemental iron delivered in divided doses two to three times a day. Importantly, the therapy with oral iron supplements can be complicated by gastrointestinal side effects, such as abdominal discomfort, nausea, vomiting, and constipation due to the oxidative properties of iron on the gastrointestinal mucosa, which occur frequently, especially when the iron is taken while fasting. For this reason, few patients fully adhere to the prescribed dose for the entire duration of treatment that should last 3-6 months in order to replenish iron stores.

Iron administration with meals or dose reduction may increase tolerance. Interference by diet and drugs of common use (eg, antacids, H2 blockers, and proton pump inhibitors) may also reduce pharmacologic iron absorption. The newest formulation of iron is a delayed-release polysaccharide iron complex (PIC)(174). This is a combination of ferric iron and a low-molecular-weight polysaccharide, which has been designed to minimize gastrointestinal upset by delaying iron release in the intestines but On the other hand, in Taiwanese populations, delayed release of iron from PIC results in slower treatment of IDA (studying the improvement in ferritin and hemoglobin) as compared with an equivalent daily dose of ferrous fumarate (175).

Oral iron is effective in correcting anemia; however, results of the same treatment on the reversal of iron deficiency in other tissues are controversial. Few studies suggest that fatigue and quality-of-life improve in iron-deficient non-anemic females treated with oral iron. However, the studies are small, heterogeneous in design, and not always unbiased (176).

The majority of studies regarding oral iron supplementation and its effectiveness recruited subjects with IDA or patients with CKD (chronic kidney diseases). In patients with IDA and non-dialysis CKD in seven randomized studies, the superiority of intravenous (IV) iron over oral iron as a faster and more efficient support for erythropoiesis has been demonstrated (**Table 9**).

Table 9: Randomized studies of ORAL vs IV iron in patients with iron deficiency anemia and chronic kidney disease.

The study name	Conclusions
A randomized study of oral vs intravenous iron supplementation in patients with progressive renal insufficiency treated with erythropoietin(177)	In pre-dialysis patients, the efficacy of monthly 300 mg iron sucrose given intravenously is not superior with regard to hemoglobin response and rHuEpo dose as compared with a daily oral dose of 600 mg of ferrous sulfate or equivalent. Where intravenous iron is preferred, lower doses may help to reduce the incidence of allergic or "free iron" reactions, especially in patients with low body mass.

FIND-CKD: a randomized trial of intravenous ferric carboxymaltose versus oral iron in patients with chronic kidney disease and iron deficiency anemia(178)	compared with oral iron, IV FCM targeting ferritin of 400-600 µg/L quickly reached and maintained Hb level, and delayed and/or reduced the need for other anemia management including ESAs(erythropoiesis-stimulating agents). Within the limitations of this trial, no renal toxicity was observed, with no difference in cardiovascular or infectious events.
A randomized, controlled trial comparing IV iron sucrose to oral iron in anemic patients with nondialysis-dependent CKD(179)	IV iron administration using 1000 mg iron sucrose in divided doses is superior to oral iron therapy in the management of ND-CKD patients with anemia and low iron indices.
Comparison of intravenous iron sucrose to oral iron in the treatment of anemic patients with chronic kidney disease not on dialysis(180)	These CKD patients had increases in both hemoglobin and ferritin following IV iron therapy, whereas those treated with oral iron had increases in hemoglobin without increases in iron stores. Iron sucrose, given weekly as 200 mg IV push over 5 min is an effective and safe anemia treatment in this population
A randomized controlled trial of oral versus intravenous iron in chronic kidney disease(181)	Oral and intravenous iron similarly increase Hgb in anemic iron-depleted ND-CKD patients not receiving ESAs. Although in comparison to oral iron, intravenous iron may result in a more rapid repletion of iron stores and greater improvement in quality of life, it exposes the patients to a greater risk of adverse effects and increases inconvenience and cost.
Comparison of oral versus intravenous iron therapy in predialysis patients of chronic renal failure receiving recombinant human erythropoietin(182)	Concomitant use of intravenous iron is better than oral iron in CRF patients(chronic renal failure) treated with rHuEPO(recombinant human erythropoietin). The intravenous route of iron administration may be a preferred route along with rHuEPO therapy, more so in the Indian context where prevalence of iron deficiency anaemia is fairly high

- **In patients with HF :**

It needs to be acknowledged that available clinical evidence on the effectiveness of oral iron therapy in patients with HF and ID is very limited.

Two clinical studies evaluated the use of oral iron preparations in patients with HF :

The first study, published in 2013, was named **IRON 5- HF** (Short Term Oral Iron Supplementation in Systolic Heart Failure Patients Suffering From Iron Deficiency Anemia) and enrolled patients with an LVEF of 20%, and a ferritin concentration of <500 µg /l. Patients were randomized into 3 groups: to receive IV iron (iron sucrose), oral iron (ferrous sulfate), or placebo. the primary endpoint of change in peak VO2 numerically increased by 3.5 ml/kg/min in the IV group without a detectable increase with the use of oral iron. However, there was a significant increase in the serum ferritin and TSAT (42).

The second trial to use oral iron therapy in HF was the **IRONOUT HF** (Oral Iron Repletion Effects On Oxygen Uptake in Heart Failure) (134) a National Institutes of Health-sponsored, double-blind,

placebo-controlled trial to test the hypothesis that oral iron compared with placebo can improve exercise capacity in HF patients with ID.

In this study, 225 HF patients with LVEF ≤40% and ID (ferritin <100 or TSAT <20%) were randomized to receive oral iron polysaccharide at 150 mg twice daily or placebo. The primary endpoint was a change in peak VO2 after 16 weeks of therapy. At week 16, peak VO2 virtually remained unchanged compared with baseline in both groups. Similarly, there was no signification between-group differences in the amount of change in the 6 MWT distance, N-terminal pro-B-type natriuretic peptide level, or HF-related quality of life score from baseline to week 16. However, compared with placebo, oral iron increased TSAT by 3.3% (p = 0.003) and the ferritin level by 11.3 ng/ml (p = 0.06). The following points should be considered in interpreting the findings of this trial:

1. In this trial, there was no direct comparison between IV and oral iron
2. the peak VO_2 remained virtually unchanged after 16 weeks of therapy with oral iron. However, therapy with IV iron either has not been shown to increase peak VO_2 in nonanemic HF patients.
3. The best method to diagnose ID in the HF population is controversial. In the treatment group of this trial, the median ferritin, TSAT, and iron levels at baseline were 75 ng/ml, 19%, and 12.6 g/dl. One could argue that not of all the participants were iron deficient and therefore were unlikely to show any benefit from iron supplementation. Considering the size (N = 225) and duration (16 weeks) of the study, this study might have been underpowered to detect a meaningful benefit in the subgroup of participants with true ID
4. The pharmacokinetics of oral iron is drastically different from that of IV iron and despite the lack of direct comparison, it is conceivable that IV iron therapy would improve iron indexes faster than oral formulations. Thus, a duration of therapy longer than 16 weeks might have been needed in the IRONOUT-HF trial to detect any beneficial effects of oral iron therapy.

These 2 studies collectively suggest that the use of oral iron can improve iron indexes. The question remains whether the improvement of iron indexes with oral iron would translate into better clinical status.

In a retrospective study, regarding iron-deficient patients with HFrEF, where oral iron supplementation over 180 days resulted in an increase in ferritin, TSAT, serum iron, and hemoglobin concentration. It was suggested that oral iron supplementation could be an alternative for IV iron, but it is worth noting that after 5 months of the therapy, the level of ferritin was still far below the threshold for an absolute ID in HF (ferritin <100 lg/L). Moreover, this study did not reveal any clinical benefits beyond improvement in serum iron indices and hemoglobin; particularly, there was no difference regarding re-hospitalization rates(183).

We note that in Australia and New Zealand 2018 guidelines practice advice, it was mentioned that :

- Oral iron supplementation is ineffective at normalizing iron status or improving QOL in patients with CHF.
- IV iron should be considered in patients with CHF associated with iron deficiency, with or without anemia.

4.8.2.1 Dosing schedule of oral iron :

Emerging evidence suggests that the conventional schedule of 2 or 3 times daily of oral iron therapy could lead to a rapid and transient response in hepcidin production and result in limited absorption of the subsequent dose of oral iron(184), some controlled trials suggest that oral iron supplementation on alternate days and in single doses can optimize iron absorption and might be the preferred dosing regimen(185).

4.8.2.2 New formulation of oral iron

Sucrosomial iron is One of the most innovative oral iron preparation consisting of ferric pyrophosphate protected by a phospholipid bilayer membrane made up of primarily a sunflower lecithin(186) developed by Alesco srl (Pisa, Italy), Preclinical data have shown that sucrosomial iron retains the iron in the sucrosome when in stomach acid, which allows intact sucrosomes to reach the small intestine where they are absorbed. A randomized open-label trial evaluated oral sucrosomial iron in non dialysis dependent patient with iron deficiency anemia. Patients were randomized 2:1 to receive oral sucrosomial iron 30 mg/d for 3 months or IV ferrous gluconate 125 mg/wk to a total dose of 1000 mg, with follow-up of 4 months. The study indicated that short-term oral sucrosomial iron was **as effective as IV ferrous gluconate** at correcting anemia (187).

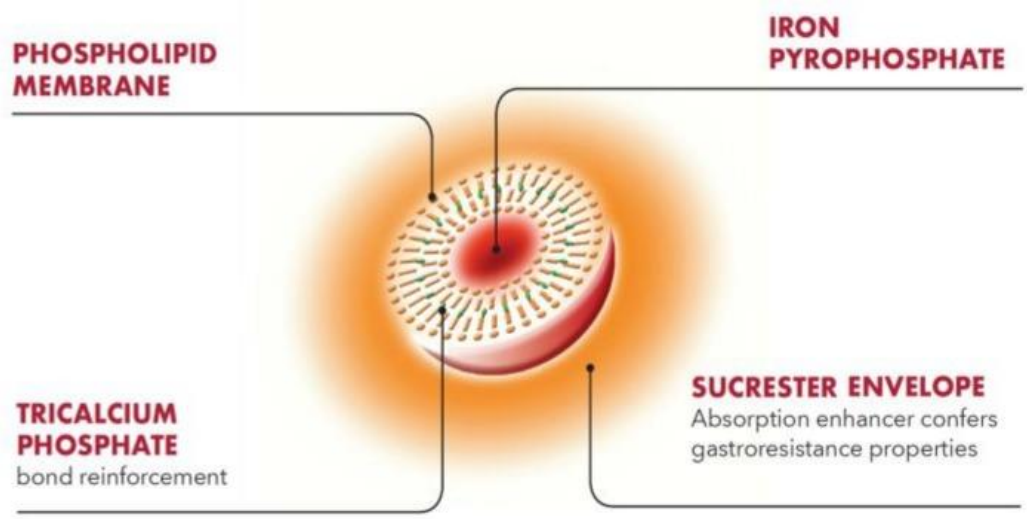


Figure 18: schematic structure of sucrosomial iron (186).

4.8.3 Parenteral Iron Supplementation:

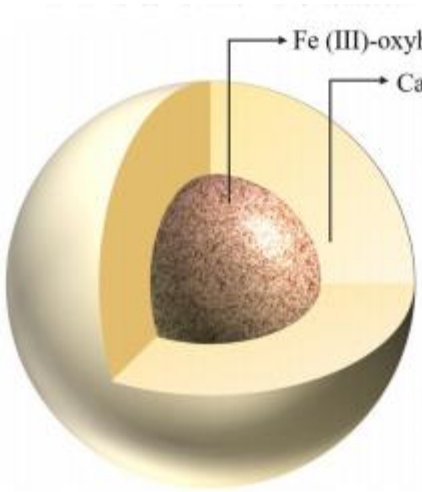
The only alternative to the oral iron route is intravenous treatment. The intramuscular route has been abandoned because of the inconvenience of painful injection, dark discoloration of the skin, and the development of sarcoma at the site of injection in treated animals. Intravenous iron is indicated when intestinal absorption is presumably poor when rapid Hb increase is needed, as in severe anemia in the second to third trimester of pregnancy, or in chronic bleeding due to inherited defects, as in hereditary hemorrhagic telangiectasia.

With the intravenous route of administration, the exact dose of iron needed to normalize Hb levels and to replenish the stores can be calculated. Thus, it is not unexpected that hemoglobin response is better to intravenous than to oral iron, as documented in several studies (188).

Historically, in the general population with anemia, the first parenteral iron preparations were administered as an iron oxyhydroxide complex (189,190) which resulted in large amounts of non-transferrin-bound iron and therefore increased oxidative stress and so several side effects, such as hypotension, nausea, vomiting, and peripheral oedema….

This problem has been solved with the introduction of compounds containing iron in a core surrounded by a carbohydrate shell (known as iron-carbohydrate complexes), which influences the molecular size, pharmacokinetics, and adverse reaction profiles, The carbohydrate shell consists of molecules such as dextran, sucrose, dextrin or gluconate (191). Structurally, all intravenous iron-carbohydrate complexes are NPs(nano particles) consisting of spheroidal polynuclear Fe (III)-oxyhydroxide/oxide cores shielded by carbohydrate shells with an overall diameter between 8 and 30 nm. This carbohydrate shell

surrounding the iron plays crucial role in stabilizing the iron core, slowing down the release of bioactive iron, protecting the particles from further aggregation, as well as sustaining the particles in a colloidal suspension (192).



Trade name	Carbohydrate shell
InFeD	Low molecular weight dextran
Ferrlecit	Gluconate
Venofer	Sucrose
Ferinject	Carboxymaltose
FeraHeme	Polyglucose sorbitol carboxymethyl ether
Monofer	Isomalto-oligosaccharides
Maltofer	Polymaltose

Figure 19: schematic representation of iron-carbohydrate complexes (192).

Since the beginning of ID treatment in HF, oral iron preparations were not the most promising treatment approach due to pathophysiological considerations, such as the established overactivity of inflammatory mediators in HF(193) ,In addition to that , the gut wall itself has been shown to display increased thickness in cases of HF(194) , Therefore, early intervention trials were carried out with IV iron treatment using one of the several different formulations available on the market. Presently, the usual route of administration is the IV route.

There are at least ten parenteral iron formulations approved for therapeutic use: **ferric sorbitol, iron dextrans** (high- and low-molecular weight dextran), **iron polymaltose, iron sucrose** complex (ISC), **ferric gluconate, ferric carboxymaltose** (FCM: hydroxide polymaltose complex), **iron isomaltoside 1000, and ferumoxytol** (**Table 10:** All Intavenous Iron formulations.).

FCM, iron isomaltoside 1000, and ferumoxytol are considered more stable iron compounds, and are characterized by slower degradation. These formulations make it possible to administer high single doses of iron. Iron dextran can also be administered in a large single dose, but its safety profile in comparison to FCM or iron isomaltoside 1000, and ferumoxytol is worse as it can more often cause severe immunological reactions, including life threatening anaphylaxis, and delayed hypersensitivity-like reactions. Most studies in patients with HF and ID have used either iron sucrose (maximum dose in 1 sitting: 200 mg) or ferric carboxymaltose (1,000 mg). Iron dextran does not play a major role anymore in clinical routine.Also a combination of the products in one sitting is generally not recommended.

Table 10: All Intavenous Iron formulations.

	High-molecular	Low-molecular	Ferric carboxy-Maltose	Iron sucrose	Ferric gluconate	Iron isomaltoside	Ferumoxytol	Iron polymaltose

	weight iron dextran	weight iron dextran	(FCM)					
Trade name	DexFerrum	INFeD	Ferinject	Venofer	Ferrlecit	Monofer	Feraheme	Ferrosig
Carbohydrate shell	Complex branched glucan	Complex branched glucan	Carboxy-maltose (branched polysaccharide	Sucrose (di-saccharide)	Gluconate (mono-saccharide)	Linear chemical structure of average 5.2 glucose units	Polyglucose sorbitol carboxymethyl ether	Dextrin
Molecular weight by manufacturer (Dalton x 1000)	265	<165	150	34–60	289–444	150	750	462
Dosage used for the pharmaco-dynamic characteristics, (mg Fe)	Not stated	500–2000	100-1000	100	125	100-200	316	100
Terminal half-life(h)	9.4–87.4, average 58.9	5.20	7.4-9.4	5.3	1.42	20.8–23.5	14.7	22.4
Test dose required	yes	yes	No	No	No	No	No	No

4.8.3.1 Iron formulations administered in patients with HF :

4.8.3.1.1 Ferric Carboxymaltose : FCM [Ferinject or Injectafer, Vifor (International) :

It is useful for rapid and high-dose replenishment of depleted iron stores (195,196) ,It has been observed that serum iron concentration increases rapidly after administration of a single dose of IV FCM equivalent to 100–1000 mg of iron, FCM is rapidly distributed from plasma not only to bone marrow but also liver and spleen,its Rapid uptake by the bone marrow occurs in the first 10 min following the administration, with subsequent uptake occurring at a slower but steady rate.

In patients receiving a single dose of FCM equivalent to 100–1000 mg of iron, the half-life of elimination of FCM from the plasma is 7–12 h, the renal elimination of iron is negligible (197). Weekly administration of FCM (up to two infusions of 1000 mg of iron and four infusions of 500 mg of iron) does not result in accumulation of iron in the serum and Being dextran-free, FCM does not react with anti-dextran antibodies so a test dose is not required.

In the FAIR-HF study(130,198), the Ganzoni formula was applied to calculate the required total FCM dose. In the CONFIRM-HF study, FCM was administered according to a fixed scheme based on the subject's weight and hemoglobin concentration at screening, additional doses of FCM can be administered at weeks 12, 24, and 36 if the patient remains iron deficient and it is worth noting that more than 75% of treated patients required a maximum of two injections of FCM.

4.8.3.1.2 Iron Sucrose :

ISC (Venofer, Vifor Pharma Ltd.) contains iron(III)-hydroxide sucrose complex. In healthy volunteers, a single dose of ISC equivalent to 100 mg of elemental iron is quickly cleared from serum, with a half-life of 5 to 6 hours, Renal elimination is negligible (on average <5%). Serum ferritin level increases significantly after 8–10 h and doubles after 24 h. In anaemic patients, a single-dose administration of radiolabelled ISC equivalent to 100 mg of elemental iron is followed by rapid uptake of this microelement by the liver, spleen, and bone marrow, reaching maximum rates at 10, 20 and 100 min after an administration, respectively. Up to 97% of administered iron is utilized for erythropoiesis, and both ferritin and TSAT return to baseline levels within 3–4 weeks. The extensive safety and tolerability record of ISC (including a low prevalence of hypersensitivity reactions) supports the recommendations of the European Medicines Agency (2013) that a test dose need no longer be applied prior to ISC administration. Special caution is recommended with every dose of IV iron instead, even in patients who responded well previously. The total dose of ISC should be determined individually, based on the calculated total iron deficit according to Ganzoni's formula (depending on the target level of haemoglobin; the frequently applied concentration is 15 g/dL).

4.8.3.1.3 Iron Isomaltoside 1000 :

Iron isomaltoside 1000 (Monofer, Pharmacosmos, Copenhagen, Denmark) consists of iron tightly bound in a carbohydrate matrix structure that guarantees a slow release of iron. The plasma half-life is 5 h for unbound circulating iron and 20 h for total iron. Due to the low anaphylactic potential, a test dose is not required, and this formulation can be administered in high doses (up to 20 mg/kg within 30–60 min). To date, only one small study with iron isomaltoside 1000 has been performed in HF patients(199) .

4.8.3.1.4 Iron Dextran :

There are two formulations of iron dextran: low-molecularweight iron dextran (INFeD, Watson Pharmaceuticals) and (CosmoFer, Pharmacosmos), and highmolecular weight iron dextan (DexFerrum, Watson Pharmaceuticals),the low molecular weight reduces the risk of anaphylaxis.. These products still require the test dose, and their parenteral use is associated with increased risk of anaphylactic reactions as compared with ISC and ferric gluconate, CosmoFer can be administered in a maximal

single dose of 20 mg/kg of body weight, but in a very slow IV infusion (over 4–6 h). The maximum daily dose of INFeD and DexFerrum should not exceed 2.0 mL (100 mg of iron).

4.8.3.1.5 Ferric Gluconate :

Ferric gluconate (Ferrlecit) is a labile weak complex of iron and requires multiple IV injections with a maximal single dose of 125–250 mg(200), An intensive IV dosing scheme (250 mg in 2 h infusions twice daily) improves haematological parameters and is well tolerated in hospitalized patients with advanced HF.

4.8.3.2 Iron formulations still not used in HF :

4.8.3.2.1 Ferumoxytol :

A novel therapeutic option for HF patients is ferumoxytol (Feraheme/Rienso, AMAG Pharmaceuticals, Inc., Lexington, MA, USA/Takeda Pharmaceutical Company Limited, Tokyo, Japan). It is a non-stoichiometric magnetite (superparamagnetic iron oxide) coated with polyglucose sorbitol carboxymethyl ether. The efficacy of this formulation in patients with CKD(chronic kidney disease)and IDA (iron deficiency anemia) appears to be non-inferior to iron sucrose (201). In addition, the safety profile of ferumoxytol is similar to placebo in anaemic patients with CKD .Since only small amounts of free iron are present in the preparation, doses of 510 mg have been administered safely during an infusion of less than 30 s. In patients receiving a single dose of ferumoxytol equivalent to 316 mg of elemental iron, the mean serum terminal elimination half-life is 15 h .

In short-term studies (201,202), IV ferumoxytol was safe and more effective than oral iron therapy in the correction of anaemia in anaemic patients with CKD .

4.8.3.2.2 Iron Polymaltose (Iron Dextrin) :

This active substance is available in an oral, intramuscular, and IV form, and contains an iron(III)-hydroxide polymaltose complex with non-ionic iron. It has not been tested in HF patients. After an IV administration of 100 mg of iron polymaltose, the mean terminal half-life is 22 h. The maximal intramuscular dose for adults is 200 mg (this dose may be repeated every second day), and for IV, the maximal dose is up to 2500 mg in a slow infusion lasting over 5h (203).

Initial test doses are not obligatory. Although a parenteral formulation has been registered for more than 50 years, the evidence on the safety of this formulation is limited. Self-limiting side effects that occur up to 2 days after an IV infusion affect 26% of patients and include headache, fever, and arthralgia (204).

4.8.3.2.3 Iron Sorbitol

Iron sorbitol is administered intramuscularly only (maximal daily dose of 100 mg), and has not been tested in HF patients.

4.8.3.3 Studies assessing efficacy of iron replacement in patients with heart failure and iron deficiency:

See Table 8 on the relevant cut-offs for the diagnosis of ID in different fields of internal medicine for details .

4.8.3.3.1 Evidence from Randomized Uncontrolled studies

4.8.3.3.1.1 Bolger et al (205) 2006:

One of the first studies on the effects of intravenous iron substitution in CHF patients published in 2006 (205). It provided data on 16 cases, that IV iron sucrose given for 5–17 days in anemic ID HF patients was well tolerated, increased hemoglobin, and improved symptoms and exercise capacity over a 3-month follow-up period.

4.8.3.3.1.2 **Usmanov et al (206) 2008 :**

Demonstrated that IV iron given for 26 weeks to patients with advanced HF, anemia, and chronic renal insufficiency exerted favorable anti-remodeling effects on the myocardium assessed by echocardiography, and improved the functional class (only in NYHA class III patients).

4.8.3.3.2 Evidence from Randomized Controlled Trials :

4.8.3.3.2.1 **Toblli et al (129) 2007 :**

It is the first randomized controlled trial involving 40 patients with chronic HF and moderate chronic kidney disease with ID anemia who were treated with a weekly infusion of IV iron sucrose for 5 weeks or placebo (isotonic saline). At 6-month follow-up the iron-treated patients when compared with the placebo group had significant improvements in HB and TSAT levels, reduction in NTpro-BNP, and diuretic requirement, improved NYHA class, LVEF, and creatinine clearance.

4.8.3.3.2.2 **Okonko et all «The FERRIC-HF: ferric iron sucrose in heart failure) » study (128) 2008 :**

16 weeks of iv. iron therapy was well tolerated, and improved exercise tolerance and symptoms. Interestingly, benefits were also observed in non-anemic ID patients although to a lesser extent, and an increase in the peak oxygen consumption was not related to changes in hemoglobin, but to an increment in the TSAT.

4.8.3.3.2.3 **Anker et al «FAIR-HF : Fer inject Assessment in patients with IRon deficiency and chronic Heart Failure study » (130) 2009 :**

This study was the one with the greatest magnitude of benefit observed with IV iron in HF patients, a randomized double-blind placebo-controlled multi-center trial, which so far recruited the greatest number of patients with chronic systolic HF and ID (both anemics and non-anemics) (n = 459) who subsequently received a 24-week therapy of iv iron or placebo (2:1).

Beneficial effects of iv iron therapy on the NYHA class and the patient's global assessment were seen across the whole clinical spectrum of HF (regardless of the baseline NYHA class, hemoglobin, LVEF, HF etiology, the presence of co-morbidities).

There was no increased risk of side-effects in the treated vs. the non-treated group, but the observation was limited to 6 months, although the FAIR-HF trial was not designed to test the effects of iron therapy on the outcome, the authors reported a trend towards a reduced rate for the first cardiovascular hospitalization in the treated vs. the non-treated group, which is similar to other reports. Undoubtedly, there is a need for more and longer running, randomized, double-blind, placebo-controlled trials that could validate the findings of FAIR-HF and also investigate the impact of this novel treatment modality on the morbidity and mortality in HF patients with ID.

4.8.3.3.2.4 **Ponikowski et al CONFIRM-HF « Ferric Carboxymaltose Evaluation On Performance In Patients With Iron Deficiency In Combination With Chronic Heart Failure » Trial (131) 2015:**

The CONFIRM-HF trial, published in 2015, was a placebo-controlled multicenter trial that enrolled 304 ambulatory symptomatic patients with HF ,it is The second large-scale trial of ferric carboxymaltose in HF and has the longest published follow-up period in HF patients treated with iron(52 weeks),it showed similair results as the ones in the FAIR study. The treatment with FCM significantly improved the 6MWD test at week 24, in addition, improvements in NYHA class and other functional/QoL measures were reported with statistical significance noted from week 24 and onwards. Furthermore, treatment with FCM was associated with a significant reduction in the risk of hospitalizations for worsening HF .

4.8.3.3.2.5 Vandelhuisen et al EFFECT-HF « Effect of Ferric Carboxymaltose on Exercise Capacity in Patients With Iron Deficiency and Chronic Heart Failure) (207) 2017 :

EFFECT-HF (2017) was an open-label RCT that enrolled 174 HF patients with ID in NYHA functional class II and III with LVEF ≤45%. FCM was injected once at baseline and another time at week 6 for a total dose of 500 to 2,000 mg elemental iron. At week 12, another dose of FCM was given at a fixed dose of 500 mg if ID was still present.

4.8.3.3.3 Evidence from Meta-Analyses :

Two meta-analyses were published in follow-up of the previously mentioned studies.

- **The first study** included subjects from FAIR-HF and CONFIRM-HF as well as the 2 previously described small RCTs (Toblli et al. and FERRIC-HF) (208) It also included subjects from the IRON-HF (Iron Supplementation in Heart Failure Patients With Anemia) study that enrolled only 23 anemic HF patients with a ferritin cutoff of 500 ng/ml and was a study to compare IV versus oral iron (209) .

  This meta-analysis showed that therapy with IV iron could reduce HF hospitalization in HF patients with anemia, albeit with significant limitations associated with the analysis.

- **The second meta-analysis** included subjects from FAIR-HF and CONFIRM-HF as well as 2 small studies, PER-CARS-01 (30 patients received FCM and 15 placebo) and EFFICACY-HF (20 patients received FCM and 14 placebo) (210).

  This meta-analysis also showed a reduction in HF hospitalization with the use of IV iron.

The previously mentioned studies on the use of IV iron in HF collectively suggest a symptomatic benefit from the use of IV iron in HF patients with ID, as defined by the serum iron indexes. In chronic diseases such as HF, improvements in symptoms and patient-reported outcomes are important, however unlike therapies that improve mortality and morbidity, interventions that purely improve symptoms require more convincing evidence before widely being incorporated into clinical practice. This is particularly relevant if there is potential for harm with those interventions.

Table 11:clinical trials with IV IRON in pateints with HF.

Authors/study	Number of patients	Iron and anaemia status	intravenous iron formulations	Administration method
Bolger et al 2006	16	Anaemia	ISC	Injection of undiluted ISC over 10 min
Usmanov et al 2008	32	Anaemia with iron deficiency	ISC	Injection of ISC in a 150 mL normal saline over 30 min
Toblli et al 2007	40	Iron deficiency and anaemia	ISC	Injection of ISC in 200 mL normal saline over 60 min(Iron sucrose

				200 mg (iv) weekly for 5 weeks)
Okonko et al 2008(FERRIC – HF)	35	Iron deficiency with and without anaemia	ISC	Injection of ISC in 50 mL normal saline over 30 min; patients observation for up to 1 h after injection; a test infusion (10 mL over 10 min) before the first treatment(ron sucrose 200 mg (iv) weekly for 16 weeks or until ferritin >500 ng/mL)
Anker et al (FAIR-HF) 2009	459	Iron deficiency with or without anaemia	FCM	Bolus injection(Ferric carboxymaltose 200 mg weekly until iron replaced then 200 mg 4 weekly)
ponikowski et al (CONFIRM-HF) 2015	304	Iron deficiency with or without anaemia	FCM	Bolus injection over at least 1 min(Ferric carboxymaltose 500–2000 mg at week 1 and 6 (+500 mg at weeks 12, 24, 36 if still ID)

Van veldhuisen et al (EFFECT-HF) 2017	174	Iron deficiency with or without anaemia	FCM	(Administered in ≥1 minute) or an infusion. Infusions of 10 or 20 mL (which is the amount of FCM that is equivalent to 500 or 1000 mg of iron, respectively) were administered diluted in ≈100 mL of sterile 0.9% sodium chloride solution and given in ≥6 minutes for 10 mL (infusion of 20 mL diluted in ≈200 mL and administered in ≥15 minutes)

4.8.3.4 Limits of the clinical trials on iron administration in chronic heart failure :

Before the FAIR HF study, the relationship between iron deficiency and chronic heart failure remained largely ignored. Nevertheless, this research area is recent, with only a few clinical trials that included limited numbers of patients (except for the FAIR-HF study). Therefore, many questions remain unanswered. As an example, although pathophysiological and clinical arguments are in favour of the intravenous route in heart failure, no trial has formally compared the oral versus intravenous routes. In addition, optimal values of iron parameters and the effects of long-term management of iron deficiency in chronic heart failure should be more precisely defined in future studies. It has to be underscored that blinding is difficult in these studies since intravenous iron is dark and it is not possible to obtain a dark placebo; therefore, particular attention to blinding is required.

4.8.3.5 Side Effects of IV Iron :

The adverse cardiovascular effects of iron are mainly related to the redox properties of elemental iron that promote the generation of ROS (reactive oxygen species) . Excess iron can overwhelm the iron-carrying capacity of transferrin, causing accumulation of NTBI (non-transferrin-bound **iron**) as well as a highly reactive labile iron pool inside the cells. This nonbound iron can react with hydrogen peroxide and generate highly toxic hydroxyl radicals (**Figure 20**), Moreover, ferroptosis an iron-dependent and reactive oxygen species (ROS)-reliant cell death with characteristics mainly of cytological changes, including decreased or vanished mitochondria cristae, a ruptured outer mitochondrial membrane, and a condensed mitochondrial membrane,has been shown to be involved in several cardiovascular processes

such as ischemia/reperfusion (I/R) injury and doxorubicin-induced cardiomyopathy(211) Interestingly, the administration of an iron chelator in vivo was protective against I/R-induced cardiomyopathy(212).

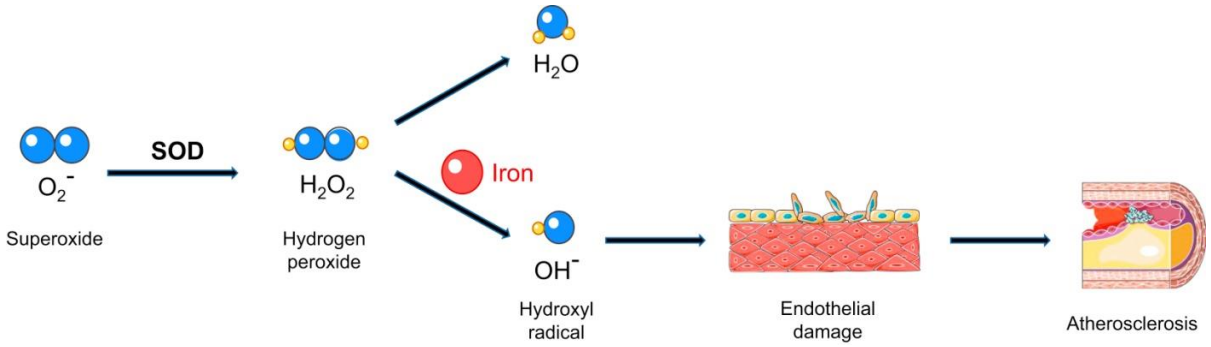


Figure 20: Free iron promotes the formtaion of reactive oxygen species and atherosclerosis: A graphic depicting the stepwise production of hydroxyl radicals via the fenton reaction. Hydroxyl radical is a strong reactive oxygen species that can damage cells and tissues by oxidizing lipid and proteins. Oxidative damage to endothelial cells lining the blood vessels can promote the formation of atherosclerotic lesions (89).

The potential toxic effects of increased iron on endothelial cells was demonstrated in a study using an apolipoprotein E–deficient mouse model in which it was shown that the iron load was associated with the progression of atherosclerosis via inducing a pro-inflammatory state (213). In another study, the restriction of dietary iron in a similar mouse model resulted in significant inhibition of atherosclerosis (214) .

IV iron has also been shown to be associated with markers of endothelial dysfunction(215,216). In a human study, the administration of IV iron at therapeutic doses to healthy volunteers resulted in transient endothelial dysfunction and was associated with a significant rise in NTBI and a biomarker of oxidative stress (217). Conversely, treatment with iron chelators can reduce endothelial dysfunction in patients with coronary artery disease (218).

Accumulation of NTBI in the serum and labile iron pool (LIP) inside the cells can lead to endothelial cell damage and the progression of atherosclerosis. In contrast, absorption of oral iron is tightly controlled by the function of hepcidin on ferroportin-1 (Fpn1) to minimize the reactive unbound iron pool while ensuring a sufficient supply of iron to the body**.**

Recent studies have shown an association between the use of IV iron and elevated levels of the biologically active form of fibroblast growth factor (FGF)-23. This hormone is primarily produced by the bone cells and is involved in phosphate homeostasis. Abnormally elevated levels of FGF-23 reduce phosphate absorption in the kidney and result in hypophosphatemia, bone resorption, and ultimately osteomalacia a disease that weakens bones and can cause them to break more easily (219). Both hypophosphatemia and osteomalacia have been reported in connection with the use of IV iron (220–222). In the CKD population, enhanced levels of FGF-23 have been associated with left ventricular hypertrophy, myocardial fibrosis, all-cause mortality, and adverse cardiovascular events, independent of traditional cardiovascular risk factors (223–225).

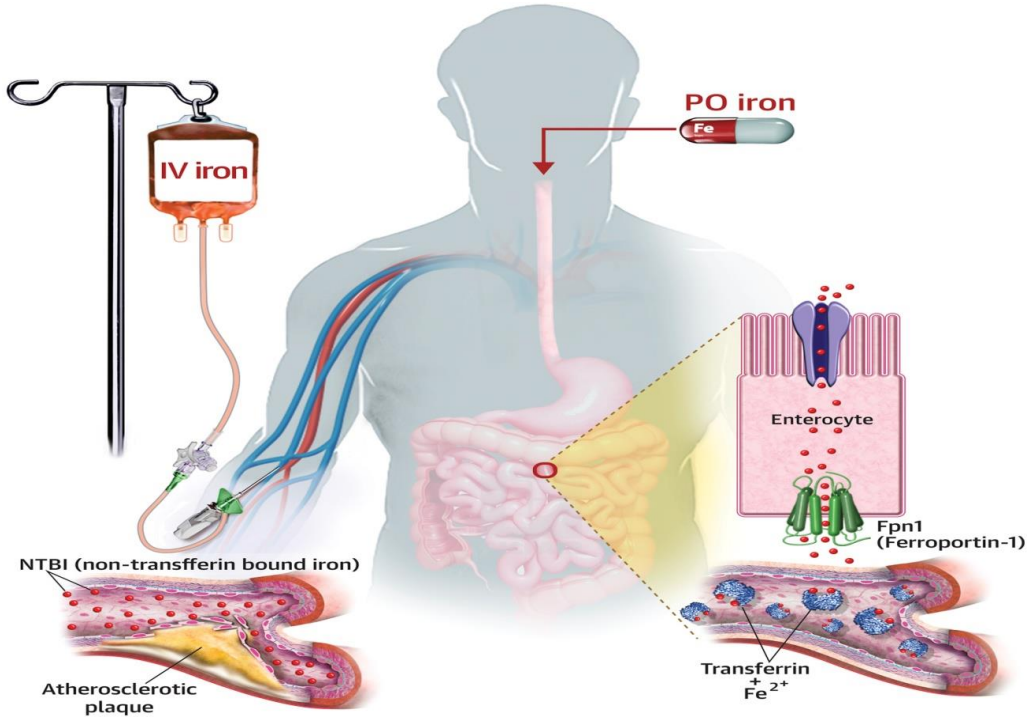


Figure 21: Absorption and distribution of iron : Iron infusion introduces large amount of non-transferrin bound iron (NTBI) into the vasculature and bypasses homeostatic mechanisms of the body that meticulously regulate influx of iron into the circulation (89).

4.8.3.5.1 Risks of ferric carboxymaltose FCM use:

Interestingly, it appears that FCM, which is the only recommended form of IV iron in HF by the European guidelines, has the unique capability of increasing the FGF-23 level as opposed to other forms of IV iron.

In a double-blind RCT comparing 2 different forms of IV iron in the treatment of adults with ID anemia (FCM 750 mg vs. ferumoxytol 510 mg), the incidence of severe hypophosphatemia at week 2 was significantly higher in the FCM group, this was correlated with the doubling of FGF-23 levels after each infusion of FCM, whereas the level of FGF-23 remained unchanged in the other group. At week 5, about 30% of patients in the FCM group continued to have severe hypophosphatemia (226).

These findings collectively raise concerns about the safety of the prolonged use of IV iron in the HF population. In particular, hypophosphatemia and osteomalacia were not monitored as side effects in the trials of IV iron in HF. The long-term safety of the use of IV iron in HF patients remains to be determined.

4.8.4 Treatment of ID in HF Guidelines:

Since 2012, the ESC guidelines have recommended that all patients with HF should undergo testing for ID by using serum assessment of ferritin and TSAT. Anemia should be ruled out by using full blood count assessment, both are Class I, Level of Evidence: C recommendations. This point has not been changed in the 2016 version of the guidelines, in which the treatment recommendation had been updated after the publication of the CONFIRM-HF trial to Class IIa, Level of Evidence: A. The statement reads, « IV ferric carboxymaltose should be considered in symptomatic patients (serum ferritin <100 or

between 100-299 µg/l and TSAT <20%) in order to alleviate HF symptoms and improve exercise capacity and quality of life » (227) .

Figure 22 below shows the diagnostic algorithm for the treatment of iron deficiency in patients with HF as recommended by current ESC guidelines:

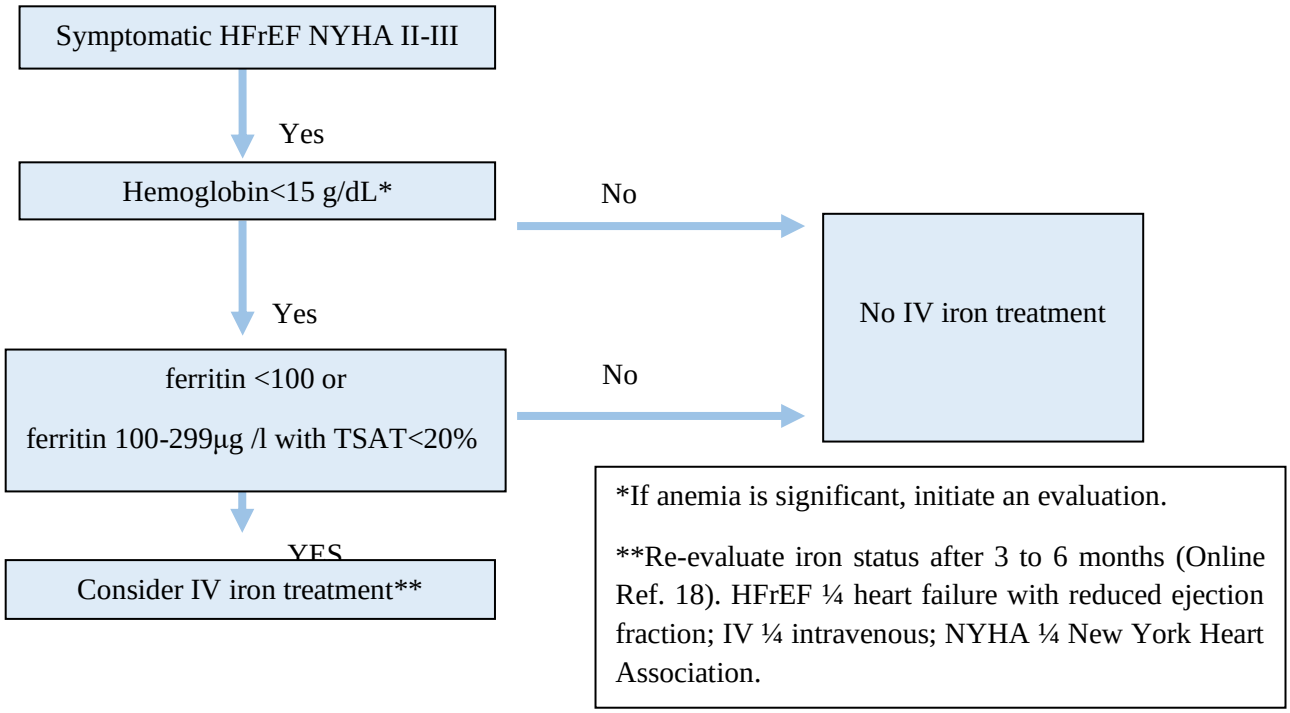


Figure 22: Diagnostic Algorithm for Treatment of Iron Deficiency in Patients with HF According to ESC Guidelines and Expert Consensus Recommendations.

The 2017 update of the joint guidelines of the American Heart Association (AHA) and American College of Cardiology (ACC) states that "in patients with NYHA functional class II–III and iron deficiency (serum ferritin <100 µg/L, or ferritin between 100–299 µg/L if TSAT <20%) IV iron might be reasonable to improve functional status and quality of life." The level of evidence is IIb. No specific type of iron formulation was recommended .

The European guidelines currently advocate only the use of ferric carboxymaltose, because large-scale trials have been undertaken, and the drug's safety has been demonstrated in the HF population only with this drug. The US guidelines do not mention a specific type of iron but rather mention IV iron administration. Previous smaller trials have also used other iron preparations, particularly iron sucrose, but largescale trials that could have been considered by the guideline committees are still missing. Therefore, it remains unclear if these are able to achieve similar results.

4.8.5 Economic analyses of iron deficiency and anemia:

A recent pharmacoeconomic study has been performed by Gutzwiller et al(228) based on the data of the FAIR-HF trial. Results were expressed as quality-adjusted life-year (QALY), an index that includes both the quality and the quantity of life lived. From the perspective of the UK National Health Service, managing iron deficiency in chronic heart failure patients using iron carboxymaltose is cost-effective. The cost per QALY-gained ratio of iron carboxymaltose compared with placebo was €4414 for the

FAIR-HF dosing regimen. This result was clearly below the threshold of €22200–€33 300/QALY gained typically used by the UK National Institute for Health and Clinical Excellence for prioritizing health expenses. These results were mainly related to improved symptoms.

Another economic model based on the Spanish National Health System, with a time horizon of 24 weeks were taken from the Ferinject® assessment in patients with iron deficiency and chronic Heart Failure trial, showed that this treatment with ferric carboxymaltose in patients with chronic heart failure and iron deficiency, with or without anemia, is cost-effective in Spain(229) .

An evaluation of the cost-effectiveness of iron repletion with ferric carboxymaltorse (FCM), in iron-deficient heart failure (HF) patients in Greece, also showed the same results(228).

4.8.6 A list of some iron products commercialized in Algeria:

- **Ferinject (**Ferric Carboxymaltose) by Hikma Pharmaceuticals.

- Ferro Sanol Gyn (Complexe f Iron Ii - Glycine Sulfate / Folic Acid) by Merinal.
- Feromax (complexe of ferric hydroxide-saccharose expressed in ferric iron) by Julphar.
- Venofer (complexe of ferric hydroxide-saccharose expressed in ferric iron) by Vifor International Inc.
- Encifer (complexe of ferric hydroxide-saccharose expressed in ferric iron) by Emcure Pharmaceuticals Ltd.
- Trifer Fol (hydroxide ferric complex polymaltose expressed in ferric iron).
- Novo Fer Plus (hydroxide ferric complex polymaltose expressed in ferric iron / folic acid).
- Razifer Ferric (hydroxide complex-saccharose expressed in ferric iron).
- Selofer (hydroxide ferric complex polymaltose expressed in ferric iron / folic acid).
- Fer 3+ (hydroxide ferric complex polymaltose expressed in ferric iron).
- Ferrum Hausmann (hydroxide ferric complex polymaltose expressed in ferric iron).

5. Conclusion:

Iron is an indispensable element for the human body, both within and beyond the process of erythropoiesis. Patients with HF are prone to become iron deficient and constitutes a globally frequent comorbidity in HF, not only as a factor leading to anemia, but also as a separate condition with adverse clinical and prognostic consequences. Conventional markers of iron status such ferritin and TSAT have certain limitations in HF and other chronic diseases. Therefore, more studies are needed to identify potentially new and/or additional serum markers reflecting iron status with comparison to the gold standard of bone marrow iron staining in patients with HF.

Although the pathophysiology of iron deficiency in HF is still partially unclear, a number of clinical trials have demonstrated that treatment with IV iron has favourable effects on NYHA functional class, exercise capacity, renal function, echocardiographic parameters, and quality of life. That is why ESC recommends it, but it is still not known whether treatment affects outcomes, since FCM, the most studied iv iron and the one recommended by ECS presents a certain risk. In addition, till now the use of oral supplementation and its effectiveness is still up for debate because a definitive head-to-head comparison between oral and IV iron supplementation in HF is still pending and so no definitive conclusion is made.

Section II:

Study

6. Objectifs :

6.1 Primary objective:

To evaluate the prevalence of iron deficiency in a chronic heart failure population with reduced left ventricular ejection fraction (LVEF < 40%).

6.2 Secondary objectives:

- To evaluate the prevalence of anemia, with or without iron deficiency, in this population.
- To search for factors associated with iron deficiency or anemia in this population.

7. Material and methods:

7.1 Study design:

A prospective study conducted between March 1st, 2021 and May 31th, 2021 at the cardiology and internal medicine service in the university hospital centre of Blida, Algeria. The investigation methods included the use of a structured questionnaire, clinical evaluation and laboratory investigations.

7.2 Population:

Recruitment was performed among patients with CHF visiting the cardiology and internal medicine service between March 1st, 2021 and May 31st, 2021.

7.2.1 Inclusion criteria:

Adult male or female patients (18 years old and above) clinically diagnosed with chronic Heart failure with reduced left ventricular function (HFrEF) defined as an ejection fraction (EF) < 40%.

7.2.2 Exclusion criteria:

- Acute heart failure.
- Patients for whom iron assessment was not available.

7.3 Biological materials:

7.3.1 Blood samples:

Peripheral venous blood samples were collected from our patients in EDTA and lithium heparin test tubes for an evaluation of various blood assays along with the measurement of haemoglobin concentration. The various tests were performed at the central hospital laboratory and at the laboratory of the anti-cancer center (CAC).

Figure 23: EDTA (left) and lithium heparin (right) test tubes.

Our patients were assessed for their hemoglobin level in whole blood using EDTA test tubes. Serum creatinine and iron status measurements (serum iron, serum ferritin, total iron-binding capacity, and transferrin saturation) were performed using lithium heparin test tubes after centrifugation.

The determination of TSAT values was only possible after the availability of TIBC reagents (Cromatest, Linear chemicals) at the central laboratory. After centrifugation and separation, patient plasma samples were frozen for the future assessment of TIBC and consequently, transferrin saturation.

7.4 **Equipment:**

Hemoglobin testing was performed using Symex hematology analyzer at the central labotary (**Figure 24**).

Figure 24: Hematology analyzer.

Figure 25: ROTOFIX 32 A centrifuge machine.

The biochemical analysis was performed in the automatic COBAS INTEGRA 400 plus analyzer (**Figure 26**).

Figure 26: COBAS INTEGRA 400 plus.

7.5 Methods:

7.5.1 Assessment of TIBC and transferrin saturation:

We used the Lineare Cromatest TIBC kit (REF 1137005). The method used involves a manual manipulation of samples by realizing a pretreatment step, where the iron is added in excess to fully saturate all the serum iron-binding sites in the plasma collected of the patients and then any remaining excess iron is removed by a solid-phase absorbent (magnesium carbonate adsorption and centrifugation).

➢ **Principle for TIBC measurement:**

Serum iron is bound to transferrin, but only about one third of the iron binding sites are saturated with iron. The unsaturated iron-binding capacity of transferrin (UIBC) denotes the available iron-binding sites of serum. The amount of iron that serum transferrin can bind when completely saturated with an excess of Fe+3 is the total iron-binding capacity (TIBC). The method1,2 measures the TIBC by first saturating the transferrin with excess of Fe+3. The remaining iron is adsorbed with magnesium carbonate, and once the binding process is complete the quelator is removed by centrifugation, and an assay for iron content performed in the supernatant. From this measurement the TIBC value is obtained. When the serum iron (SI) determination is performed concurrently with the TIBC and the result substracted from the TIBC value, the difference yields the unsaturated iron-binding capacity (UIBC), or seric transferrin not bound to iron.

➢ **Reagent composition:**

- R1: Iron solution. 500 µg/dL Fe+3 (89.5 µmol/L)
- R2: Magnesium carbonate. Powder.

➢ **Procedure**

- Pipette into disposable centrifuge tubes:
  - **Sample** : 0.5 mL
  - **R1** :1ml.
  - **Ratio** = sample/R1=1/2
  - **Dilution factor** = 3
- Mix and allow to stand for 5-20 minutes at room temperature.
- Add to each tube one scoop (aprox. 100 mg) of R2 and allow to stand for 30 minutes, mixing vigorously at 5-minute intervals.

- Centrifuge for 10 minutes at 3000 r.p.m. Separate off the clear supernatant.

➢ **Calculations:**

- Once the serum iron (SI) test is performed as described in the technical insert of the kit, TIBC is calculated.

Total iron-binding capacity (TIBC) = µg/dL supernat x 3 (Dilution Factor).

The normal range of TIBC is between 250 – 450 µg/dL which corresponds to 44.8 – 80.5 µmol/L.

- We then calculated the transferrin saturation:

Transferrin saturation (%) =Serum iron x 100/ TIBC.

Normal values are 15% to 50%.

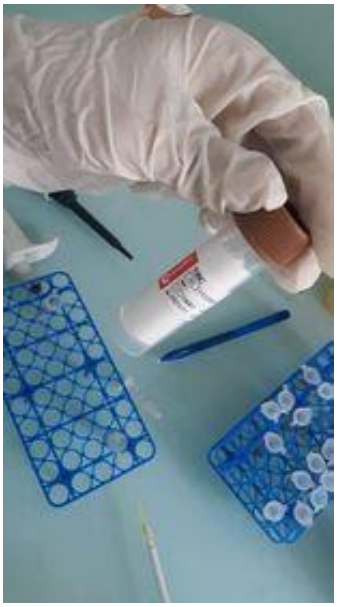


Figure 27: Measurement of TIBC.

Figure 28: Reagent (2): magnesium carbonate.

7.5.2 Data collection:

A carefully prepared patient recruitment form (**Annex I**) was used to collect data. Out of patient data collected, the following characteristics were included in the study:

➢ **Patient characteristics:**
- Date of birth (age).
- Gender.
- Weight, height and body mass index (BMI).
- Left ventricular ejection fraction (LVEF).
- Etiology of heart failure: Ischemic or non-ischemic.
- New York Heart Association (NYHA) Functional Classification (I- IV).

➢ **Risk factors for cardiovascular disease**:
- Hypertension
- Diabetes
- Smoking
- Dyslipidimia

➢ **Biological parameters:**

- Hemoglobin.
- Serum ferritin.
- Serum iron.
- Transferrin saturation coefficient.
- Creatinine clearance.

➢ **Medications taken by the patient:**

- Beta blockers
- ACE (Angiotensin-converting enzyme) inhibitors.
- ARB (angiotensin receptor blockers).
- VKA (Vitamin K antagonist).
- Digoxin.
- Furosemide.
- Antiplatelet Agent (aspirine and/or clopidogrel).
- Spironolactone.
- Statins.

Patient information was obtained from our direct patient questionnaire, and medical records, collected by the doctors of the cardiology and internal medicine service.

7.5.3 Definitions :

- Anemia was defined as hemoglobin level <12 g/dl in women and <13g/dl in men.
- Absolute iron deficiency was defined as serum ferritin <100ng/ml.
- Functional iron deficiency was defined as serum ferritin between 100-299 ng/ml and TSAT <20%.
- BMI categories were defined according to WHO.

8. Statistical analysis:

Statistical analysis was performed using IBM SPSS statistics, version 26 and Microsoft Excel 2016. Qualitative variables were expressed as relative frequencies (N) and percentages (%) while continuous quantitative variables were expressed as mean ± standard deviation. Comparison of means (quantitative variables) between 2 groups was performed with Student's *t* test for independent groups. Comparison between groups for qualitative variables was performed with Pearson's chi-squared test (χ^2) or Fisher's exact test . A *p*-value < 0.05 was considered to be statistically significant.

9. Results:

9.1 Descriptive analysis:

9.1.1 Population:

In this study, we analyzed patients' data procured through a prospective study conducted between March 1st, 2021 and May 31st, 2021 in the cardiology and internal medicine service at the university hospital center of Blida, Algeria.

The study population consisted of 125 initial patients with diagnosed chronic heart failure (CHF) based on the presence of current or previous symptoms or characteristic clinical signs and evidence of left ventricular dysfunction (defined as < 40%). However, 28 out of those patients had to be excluded for the lack of iron assessment tests which left in the end, a population of 97 patients was included for analysis.

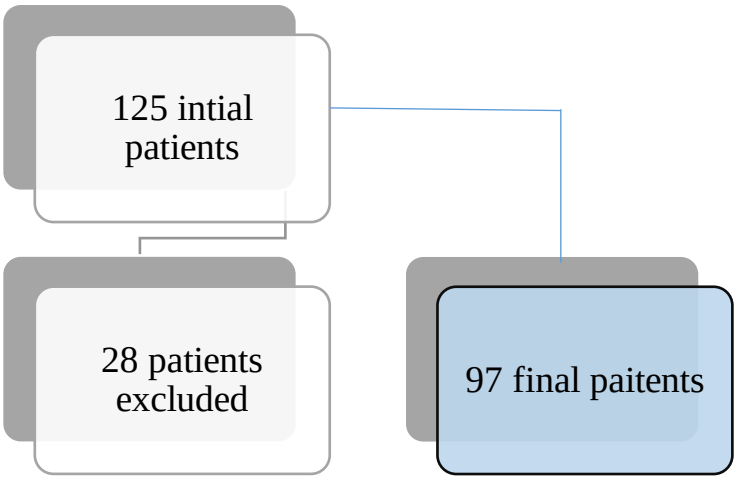


Figure 29: Composition of our study population.

9.1.2 Demographic data:

9.1.2.1 Sex Distribution:

The sex distribution of the population (**Table 12**) shows that more than twice as many men were in this study compared to women, with a sex ratio of 2,12 (M/F).

Table 12: Distribution of patients by sex.

Sex	Men	Women	Total
N	66	31	97
%	68,04	31,96	100

9.1.2.2 Age distribution:

Table 13 shows the distribution of our population by age group. The age of patients included in this study ranged from 23 to 83 years old with a mean age of 55,89 years (**Table 14**). We note that more than half of patients (55,67%) were between 50 and 69 years of age with 28,87% reflecting patients aged between 50 and 59 years and 26,8% for those aged between 60 and 69 years.

Table 13: Distribution of patients by age group (years).

Age group (years)	20 - 29	30 - 39	40 - 49	50 - 59	60 - 69	70 - 79	80+	Total
N	3	10	16	28	26	13	1	97
%	3,09	10,31	16,49	28,87	26,80	13,40	1,03	100

Table 14: Descriptive statistics of patients' ages.

Age (years)	N	Minimum	Maximum	Mean	Std. Deviation
	97	23	82	55,89	13,184

9.1.2.3 Distribution according to the body mass index (BMI) categories:

Table 15 shows the distribution of our population by BMI categories. The patients' BMIs ranged from 16,33 to 37,11 (**Figure 30:** Descriptive statistics of patients' body mass index.

Table 16). The majority of patients (89,69%) were either normal or overweight. Overweight patients had the highest percentages (47,42%) which is little more than that of patients with normal weight (42,27%).

Table 15: Distribution of patients by body mass index categories.

BMI	Underweight	Normal	Overweight	Obese	Extremely obese	Total
	<18,5	18,5–24,9	25–29,9	30–34,9	> 35	
N	2	41	46	5	3	97
%	2,06	42,27	47,42	5,15	3,09	100

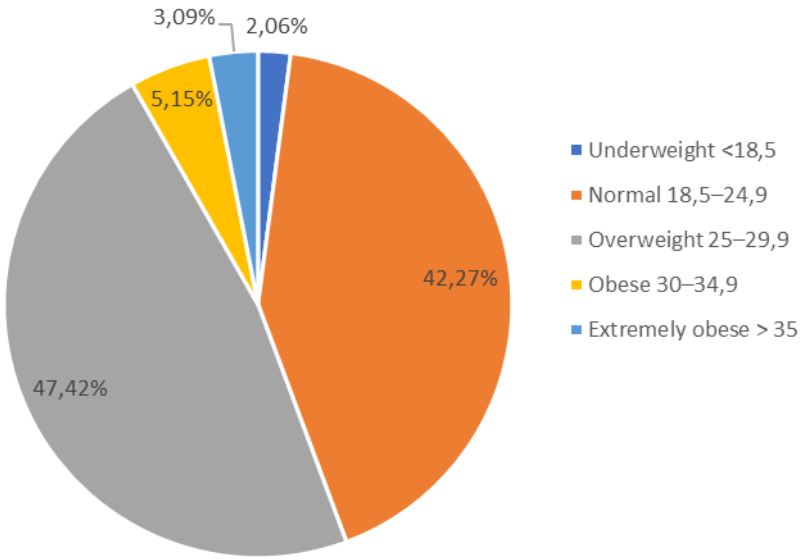


Figure 30: Descriptive statistics of patients' body mass index.

Table 16: Descriptive statistics of patients' body mass index.

BMI	N	Minimum	Maximum	Mean	Std. Deviation
	97	16,33	37,11	25,24	3,818

9.1.3 Clinical data:

9.1.3.1 Distribution by Etiology of Heart Failure:

Table 17 shows the distribution of patients by the etiology of heart failure. We observe that more than half (53,61%) of patients had an ischemic etiology compared to 46,39% with non-ischemic etiology.

Table 17: Distribution of patients by the etiology of heart failure.

Etiology	Ischemic	Non-ischemic	Total
N	52	45	97
%	53,61	46,39	100

9.1.3.2 Distribution by the New York Heart Association (NYHA) Functional Classes:

Table 18 shows the distribution of patients by the New York Heart Association (NYHA) functional classes. We observe that more than half of patients (63.92%) were symptomatic (class II to IV) patients. Class II have the highest percentage with 39%.

Table 18: Distribution of patients by the New York Heart Association (NYHA) functional classes.

NYHA	Class I	Class II	Class III	Class IV	Total
N	35	39	21	2	97

%	36,08	40,21	21,65	2,06	100

9.1.3.3 Distribution by Left Ventricular Ejection Fraction (LVEF):

Table 19 shows the distribution of patients by left ventricular ejection fraction. The latter ranged from 10 to 40% (**Table 20**). The majority of patients (88,66%) had a LVEF between 21 to 40%. Patients with a LVEF between 21 to 30% had the highest percentage of 47,42%, closely followed by patients with a LVEF between 31 to 40% with 40%.

Table 19: Distribution of patients by left ventricular ejection fraction.

LVEF	≤20%	21 – 30%	31 – 40%	Total
N	11	46	40	97
%	11,34	47,42	41,24	100,00

Table 20: Descriptive statistics of patients' left ventricular ejection fractions.

LVEF	N	Minimum	Maximum	Mean	Std. Deviation
	97	10	40	29,72	6,347

9.1.3.4 Distribution by risk factors for cardiovascular disease/comorbidities:

The studies factors are hypertension, diabetes, smoking and dyslipidemia.

Table **21** shows the distribution of patients by these factors. We observe that out of 97 patients, 46,39% had hypertension, 44,32% had diabetes and almost 33% were smokers. 10 patients included in this study did not have a lipid profile (total cholesterol, HDL cholesterol, LDL cholesterol and triglycerides). Out of the remaining 87 patients, 32,18 % had dyslipidemia.

Table 21: Distribution of patients with hypertension, diabetes, smoking habits and dyslipidemia.

CRF	Hypertension	Diabetes	Smokers	Dyslipidemia
N	45	43	32	28
%	46,39	44,32	32,98	32,18
Total	97	97	97	87

9.1.3.5 Distribution by Pharmacological treatments of heart failure:

Table 22 shows the distribution of patients by pharmacological treatments of heart failure. We observe the following association of medications:

- 30,92% were taking an angiotensin-converting-enzyme inhibitor (ACEI) or an angiotensin receptor blocker (ARB) plus a beta blocker.
- 54,63% were taking an angiotensin-converting-enzyme inhibitor (ACEI) or an angiotensin receptor blocker (ARB) plus a beta blocker plus a spironolactone.

Table 22: Distribution of patients by pharmacological treatments of heart failure (n=97).

ACEI or ARB (+)	N	87
	%	89,69
Beta-blockers (+)	N	88
	%	90,72
Spironolactone (+)	N	59
	%	60,82
ACEI/ARB + Beta blockers	N	30
	%	30,92
ACEI/ARB + Beta blockers +	N	53

Spironolatone	%	54,63
Vitamin K antagonists (+)	N	27
	%	27,84
Statins (+)	N	60
	%	61,86
Digoxin (+)	N	5
	%	5,15
Antiplatelet Agents (+)	N	56
	%	57,73
Furosemide (+)	N	84
	%	86,60

9.1.4 Biological data:

9.1.4.1 Distribution by creatinine clearance (CrCl):

Creatinine clearance was calculated using the MDRD formula. Table 23 shows the distribution of patients by creatinine clearance. The latter ranged from 9.09 to 139 mL/min/ 1.73 m^2 (**Table 24**). CrCl had the highest percentage of 39,18%.

Table 23: Distribution of patients by creatinine clearance (MDRD).

CrCl (mL/min/1,73m^2)	**<15**	**15 - 29**	**30 - 44**	**45 - 59**	**60 - 89**	**≥90**	**Total**
N	3	6	12	18	38	20	97
%	3,09	6,19	12,37	18,56	39,18	20,62	100

Table 24: Descriptive statistics of patients' Creatinine clearance (CrCl)

CrCl	**N**	**Minimum**	**Maximum**	**Mean**	**Std. Deviation**
	97	9,09	132,00	64,86	26,029

9.1.5 Prevalence of Iron deficiency (ID) in heart failure patients:

Table 25 and Figure 31 show the distribution of iron deficient and non-iron deficient heart failure patients. We observe that majority of our population (82,47%) had some type of iron deficiency. Out of the 80 iron-deficient patients, more than half (53,75%) had an absolute iron deficiency while the remaining 46,25% had a functional iron deficiency (**Figure 32**).

Table 25: Distribution of patients by the presence or absence of iron deficiency.

	Iron Deficiency (-)	**Iron Deficiency (+)**		**Total**
		Absolute ID	**Functional ID**	
N	17	80		97
		43	37	
%	17,53	82,47		100
		44,33	38,14	

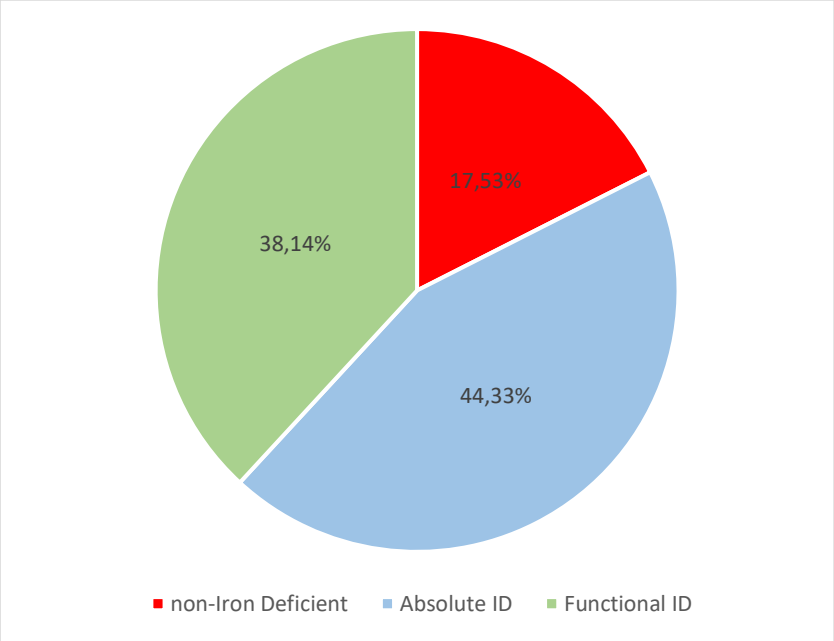


Figure 31: Distribution of patients by the presence or absence of iron deficiency (n=97).

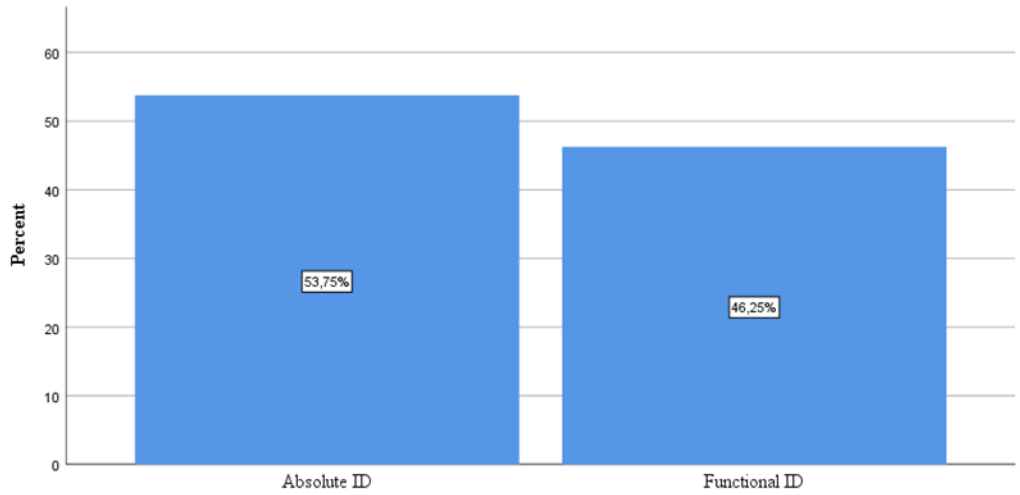


Figure 32: Distribution of iron-deficient patients by the type of iron deficiency (ID) (n=80).

Figure 33 shows the distribution of our population by the presence or absence of iron deficiency and by sex. We observe a male predominance among iron-deficient patients with men accounting for 53,61% of those patients.

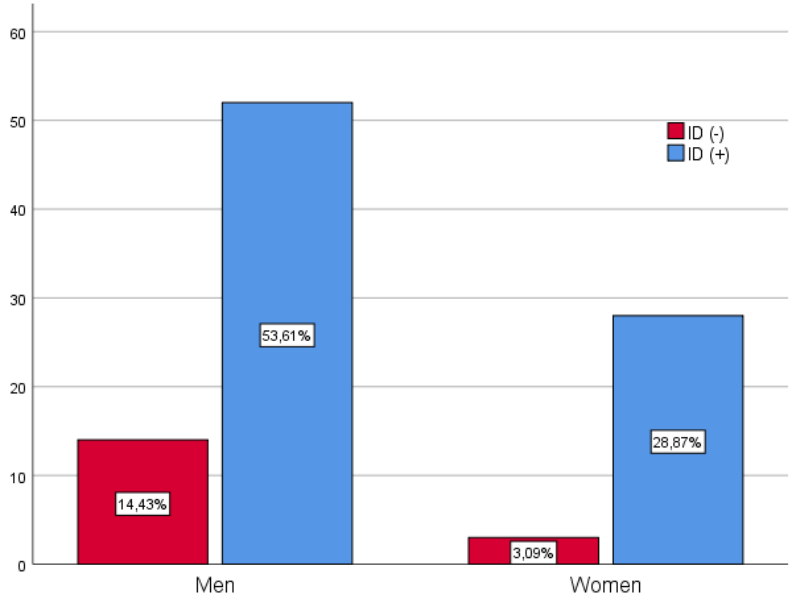


Figure 33: Distribution of our population by the presence (+) or absence (-) of iron deficiency (ID) and by sex.

Figure 34 shows the distribution of our population by the presence or the absence of iron deficiency and by age group. We observe that iron deficient patients aged between 50-59 years and between 60-69 years each account for 22,68% of the whole population (45,36% in total).

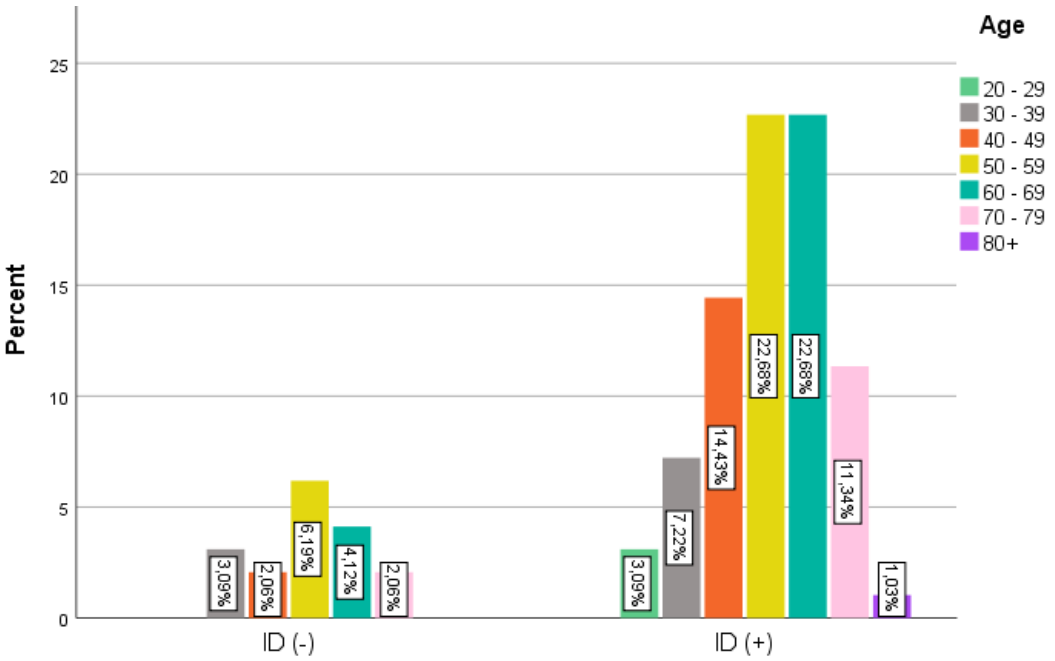


Figure 34: Distribution of our population by the presence (+) and absence (-) of iron deficiency (ID) and by age group (n=97).

9.1.6 Prevalence of Anemia in heart failure patients:

Table 26 and Figure 35 show the distribution of anemic and non-anemic heart failure patients. We observe that more than half of our population (59,79%) had anemia.

Table 26: Distribution of patients by the presence (+) or absence (-) of anemia.

	Anemia (-)	Anemia (+)	Total
N	39	58	97
%	40,21	59,79	100

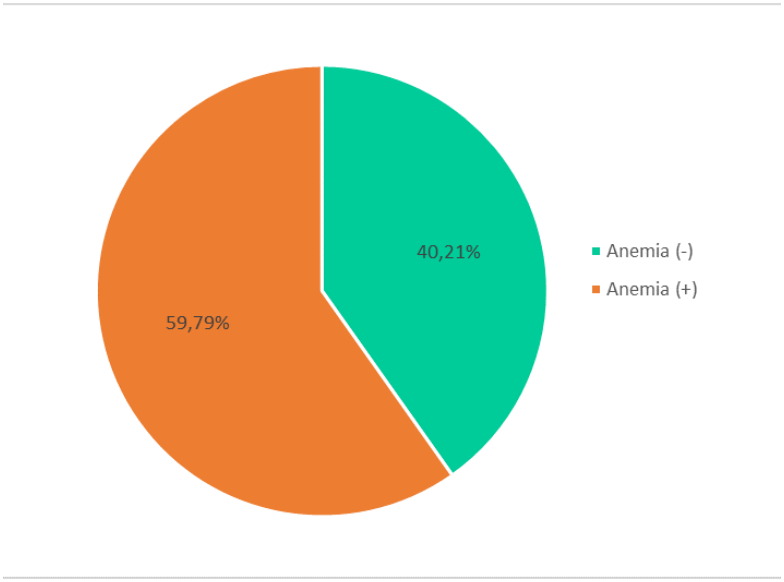


Figure 35: Distribution of patients by the presence (+) or absence of anemia (-) (n=97).

From Table 27 and Figure 36, we observe that, more than half (63,75%) of iron deficient patients had anemia and less than half (41,18%) of non-iron deficient patients had anemia.

Table 27: Distribution of iron and non-iron deficient patients by the presence (+) or absence (-) of anemia.

		Anemia +	Anemia -	Total
ID +	N	51	29	80
	%	63,75	36,25	100
ID -	N	7	10	17
	%	41,18	58,82	100

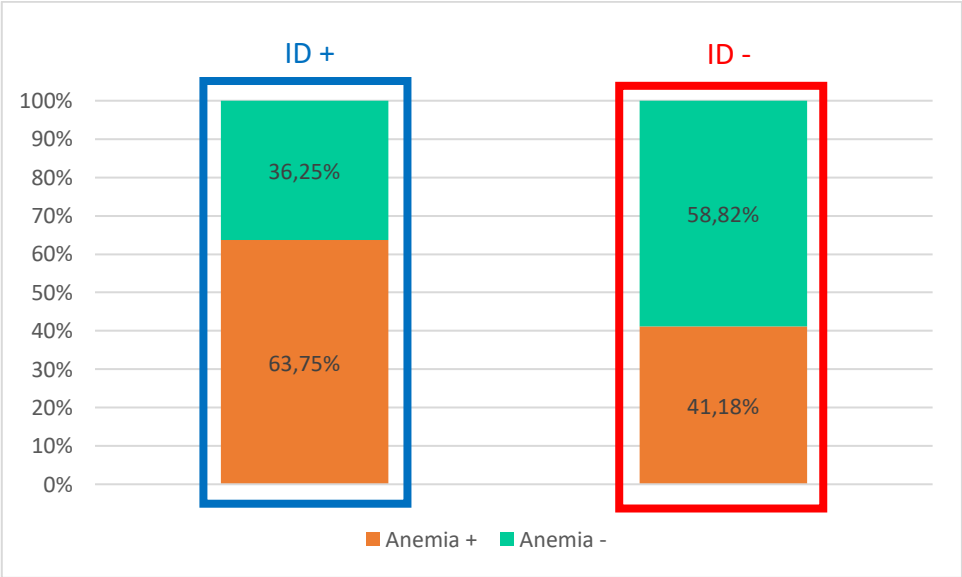


Figure 36: Distribution of iron and non-iron deficient patients by the presence (+) or absence (-) of anemia.

9.1.6.1 Prevalence of anemia in women and men:

Figure 37 shows the distribution of men and women by the presence or absence of anemia. We observe that more than half of men (66,67%) and of women (54,84%) are anemic.

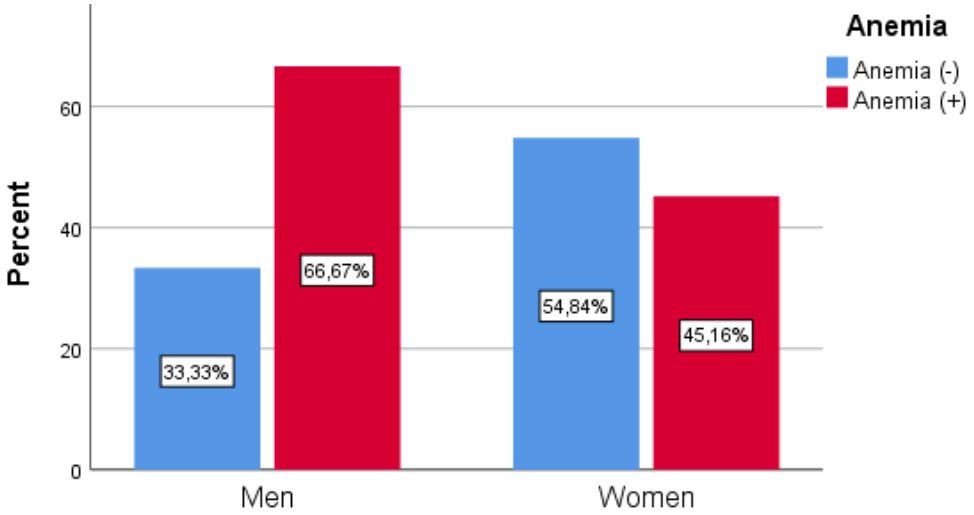


Figure 37: Distribution of our population by the presence (+) or absence (-) of anemia and by sex.

9.2 Analytical study:

9.2.1 Comparison of characteristics of iron deficient and non-iron deficient patients:

A bi-variate analysis, presented in Tables **Table 28** and **Table 29**, was performed to compare the characteristics of iron deficient and non-iron deficient patients.

Table 28: General characteristics of iron deficient and non-iron deficient patients.

Variables		Iron deficiency (-) (N)	Iron Deficiency (+) (N)	*P*-value
Sex	Men	14	52	0,163
	Women	3	28	
Etiology	Non ischemic	7	38	0,635
	Ischemic	10	42	
NYHA Class	Class I	6	29	0,559*
	Class II	6	33	
	Class III	4	17	
	Class IV	1	1	
ACEI/ARB	No	4	6	0,070*
	Yes	13	74	
Beta Blockers	No	3	6	0,191*
	Yes	14	74	
Vitamin K antagonists	No	13	57	0,773*
	Yes	4	23	
Digoxin	No	16	76	1*
	Yes	1	4	
Statins	No	7	30	0,777
	Yes	10	50	
Furosemide	No	3	10	0,694*
	Yes	14	70	
Antiplatelet Agent	No	5	36	0,237
	Yes	12	44	
Spironolactone	No	11	27	0,018

	Yes	6	53	
Hypertension	No	9	43	0,952
	Yes	8	37	
Diabetes	No	9	45	0,803
	Yes	8	35	
Dyslipidemia	No	9	50	0,762*
	Yes	5	23	
Smoking	No	9	56	0,174
	Yes	8	4	
Anemia	No	10	29	0,085
	Yes	7	51	

+: presence. –: absence. NYHA: New York Heart associations. ACEI: angiotensin-converting-enzyme inhibitor. ARB: angiotensin receptor blocker. P-value: X^2 test. P-value*: (2-sided) Fischer's exact test.

Table 29: Bioclinical characteristics of iron deficient and non-iron deficient patients.

Variables	Iron deficiency (-)		Iron deficiency (+)		*P*-value
	mean	Std.dev	mean	Std.dev	
Age	55,76	13.165	55.91	13.271	0,967
Body mass index	26,13	3,56	25,06	3,87	0,296
Left ventricular ejection fraction	32,65	5,488	29,10	6,374	0,036
Hemoglobin	12,71	1,428	12,01	1,519	0,088
Creatinine clearance	67,28	32,022	64,28	24,773	0,636

+: presence. –: absence. P-value: Student's t-test.

From **Table 28** and **Table 29**, we note that the *P*-values for spironolactone (*P*=,018), and left ventricular ejection fraction (*P*=,036), are each below 0,05 and thus considered statistically significant. This allows us to reject the null hypothesis, and to say that the association between each of variables and the variable of iron deficiency is significant.

9.2.2 Comparison of general characteristics of anemic and non-anemic patients:

A bi-variate analysis, presented in Tables **Table 30** and **Table 31**, was performed to compare the characteristics of anemic and non-anemic patients.

Table 30: General characteristics of anemic and non-anemic patients.

Variables		Anemia (-) N	Anemia (+) N	*P*-value
Sex	Men	22	44	0,044
	Women	17	14	
Etiology	Non ischemic	14	31	0,089
	Ischemic	25	27	
NYHA Class	Class I	16	19	0,337*
	Class II	17	22	
	Class III	5	16	
	Class IV	1	1	
ACEI/ARB	No	4	6	1*
	Yes	35	52	
Beta Blockers	No	1	8	0,080*

	Yes	38	50	
Vitamin K antagonists	No	28	42	0,947
	Yes	11	16	
Digoxin	No	38	54	0,645*
	Yes	1	4	
Statins	No	20	17	0,029
	Yes	19	41	
Furosemide	No	7	6	0,281
	Yes	32	52	
Antiplatelet Agent	No	18	23	0,525
	Yes	21	35	
Spironolactone	No	18	20	0,248
	Yes	21	38	
Hypertension	No	23	29	0,385
	Yes	16	29	
Diabetes	No	23	31	0,591
	Yes	16	27	
Dyslipidemia	No	25	34	0,722
	Yes	13	15	
Smoking	No	27	38	0,703
	Yes	12	20	
Iron deficiency	No	10	7	0,085
	Yes	29	51	

+: presence. –: absence. NYHA: New York Heart associations. ACEI: angiotensin-converting-enzyme inhibitor. ARB: angiotensin receptor blocker. P-value: Chi squared (X^2) test. P-value*: (2-sided) Fischer's exact test.

Table 31: Bioclinical characteristics of anemic and non-anemic patients.

Variables	Anemia (-)		Anemia (+)		*P*-value
	mean	Std.dev	mean	Std.dev	
Age	54.87	13.647	56.57	12.943	0,537
Body mass index	25.58	3.557	25.02	3.998	0,476
Left ventricular ejection fraction	28.62	6.285	30.47	6.333	0,160
Creatinine clearance	71.29	24.031	60.54	26.625	0,046

+: presence. –: absence. P-value: Student's t-test.

From tables **Table 30** and **Table 31** we note that the *P*-values for sex (*P*=,044), statins (*P*=,029), and creatinine clearance (*P*=0,046) are each below 0,05 and thus considered statistically significant. This allows us to reject the null hypothesis, and to say that the association between each of variables and the variable of anemia is significant.

10. Discussion:

In recent years, iron deficiency and anemia have become a point of interest in heart failure patients with reduced ejection fractions. In this study, we aimed to evaluate the prevalence of iron deficiency and anemia in that population and to pursue an analysis of diverse variables which could possibly relate to iron deficiency or anemia. Thus clinical, echocardiographic, pharmacological and laboratory variables were included.

The prevalence of ID in our heart failure patients with reduced ejection fraction was 82,47% with 44,33% having an absolute ID and 38,14% having a functional ID. Iron-deficient men accounted for 53,61% and iron-deficient women accounted for 28,87% of the whole population. The prevalence of ID in our study is high compared to other studies. A study by Jankowska et *al*. (94) documenting the prevalence of ID regardless of presence of a concomitant anemia exhibited a 37% ID prevalence using the same definitions as us for absolute and functional ID. An important difference between our study and theirs is the inclusion cut-off values for LVEF, ours was ≤40% (mean LVEF 29,10 ± 6,3 %) while theirs was ≤45% (mean LVEF 26 ± 7%). We found a significant decrease (P=0,036) of mean LVEF in iron deficient patients compared to non-iron deficient patients (29,10 ± 6,374 vs 32,65 ± 5,488). Our Patients were highly symptomatic CHF with a severely reduced left ejection fraction (21 – 30%) representing the biggest part of the population (47,42%: n=46) which may lead to the idea linking the severity of systolic heart failure to the prevalence of ID.

Anemia in regards to ID was found to be marginally significant (P=0.085).

Our analysis showed a significant association (P=,029) between the variable of spironolactone and the variable of iron deficiency in heart failure. There is no similar data available on this topic. But it maybe associated with our hospitalized patients' aggravated status which led to the administration of spironolactone.

59,79% of our population had anemia. The prevalence of anemia varied in academic writing. Its prevalence in patients with HF (defined as hemoglobin <13 g/dL in men and <12 g/dL in women) was found to be ≈30% in stable and ≈50% in hospitalized patients, regardless of whether patients have HFrEF or HF with preserved ejection fraction (1).

We found a significant association between the variable of sexes and the variable of anemia (P=,044).

A significant association between the use of statins and anemia in heart failure was noted in our study (P=,029). There was no available data on this matter but it is worth mentioning that a study by Anna Masajtis (230) on the effect of 6-month administration of atorvastatin within chronic kidney disease patients, found there was a small but significant increase of serum hemoglobin in the course of atorvastatin administration (from 11.6 ± 1.6 g/dl to 11.9 ± 1.5 g/dl; P = ,002).

In our population, anemic patients had a significant decrease in mean creatinine clearance (P=,045) compared to non-anemic patients (60.54 ± 26.625 vs 71.29 ± 24.031). This decrease invokes possible presence of CKD among the anemic population. This in turn suggests that anemia in HF can be associated with chronic kidney disease. This correlates with existing information that compared with non-anemic patients with HF, anemic patients are more likely to have CKD (1) .

Our study showed a prevalence of anemia of 59,79%. Focusing on the anemic population, we calculated an ID prevalence of 87,93% (n=58) which is high compared to the ones documented. Iron deficiency was evaluated in many subgroups of patients who presented with anaemia in different studies. The prevalence of ID in the anemic patients varied from 10 to 60% (Witte et al. 2004 (231), De Silva et al. 2006 (92), Annand 2008 (91). Nanas et al. 2006 (93) were the first to show that ID assessed by bone

marrow aspiration was present in 73% of patients with anemia despite seemingly adequate iron stores assessed by serum iron and ferritin, this study caused a change in the definition of ID in heart failure by Okonko et al. (144), including now the functional form of ID. In our non-anemic population, the ID prevalence was estimated at 74,36% (n=39). The study by Jankowska et al (94), found the prevalence of ID in the anemic patients was 57 ± 10% vs. 32 ± 4% in non-anemic patients. Thus the prevalence of iron deficiency in the anemic and non-anemic subgroup in our present study was also significantly higher than in the study of Jankowska et al (94).

Our population had a mean age of 55,89 years and more than half of patients were aged between 50 to 69. This agrees with academic writing. According to a review article published in Trends in research (36), individuals aged 50 years or below are unlikely to have HFrEF, but the prevalence and incidence increase progressively in those aged above 50 years. In an age controlled population study in Denmark, Raymond *et al.*(232), found that impaired left ventricular systolic function and heart failure increased substantially with age. However, we also had young HFrEF patients (<40 years) in our study.

We observe a male predominance among our population of heart failure patients with reduced ejection fraction with a sex ratio of 2,12. This predominance goes in the same direction with other data available. In a prospective population-based study in Spain, Gomez-Soto *et al.* (233) found higher rates of HFrEF in men than women. In a Scottish study, McDonagh et *al* (16), found that the prevalence of symptomatic and asymptomatic LV systolic dysfunction (LVEF) ⩽ 30%) were both greater in men than women. Raymond *et al.*(232) found that LV systolic impairment and HF was more than twice as frequent among men as among women.

The highest percentage of our population was overweight with a percentage of 47,42%. Hubert HB, et al. (234) found that weight gain after the young adult years conveyed an increased risk of cardiovascular disease in both sexes.

53,61% of our population had an ischemic etiology. This goes in the same direction as in available data but the prevalence of ischemic heart disease (IHD) in heart failure varies by source. According to Harrison's principles of internal medicine, coronary artery disease has become the predominant cause in men and women and is responsible for 60–75% of cases of HF in industrialized countries (29). In clinical trials, HF has been ascribed to IHD in about 70% of patients. Epidemiological studies also suggest that IHD is the most common cause of heart failure (235). McDonagh et *al* (16), suggested that 83% of an unselected group of patients with heart failure with left-ventricular systolic dysfunction had evidence of ischemic heart disease, although only about half had had an overt myocardial infarction.

More than half of our population (63,92%) were symptomatic (Class II-IV). Our results were high compared to other studies. The Carla study showed that for the examined population aged 45 years and over (n=78), symptomatic HFrEF could be shown in 48% of patients (236). In the Rotterdam study, 40% of persons with left ventricular systolic dysfunction had shortness of breath, ankle oedema or pulmonary crepitations (237). A study by Flu et al (238), where echocardiography was performed preoperatively in 1,005 consecutive vascular surgery patients with a systolic LV dysfunction (defined as less than 50%) found that 80% of 506 diagnosed patients with LV dysfunction were asymptomatic. The difference in the prevalence of symptomatic patients can be attributed to differing definitions of LV systolic dysfunction, the prevalence of cardiovascular risk factors in the studied populations and the subjectivity of reporting symptoms.

Creatinine clearance is an estimate of glomerular filtration rate (GFR) which is a widely accepted indicator of renal function. Chronic kidney disease (CKD) is usually defined as an estimated glomerular filtration rate (eGFR) < 60 mL/min/ 1.73 m^2. We note that 40,21% of our patients presented a creatinine

clearance below 60ml/min/1,73m2 suggesting the presence of chronic kidney disease. This can also be seen in scientific literature. A large meta-analysis of patients with HFrEF found that around 55% had CKD stage G3 or higher (eGFR < 60 ml/min per 1.73 m2) (239). In a Swedish study by Ida Löfman et *al* (240), 47716 patients were divided based on estimated glomerular filtration rate (eGFR). The ending result was that 51% of the patients had eGFR <60 mL/min/1.73m^2 and 11% had eGFR <30, in addition to an increase in mortality with decreasing kidney function regardless of age, lead to the conclusion that kidney dysfunction is common and strongly associated with short-term and long-term outcomes in patients with heart failure.

46,39% of our patients had hypertension. The latter is an important coexisting disorder but also contributes pathogenetically to the development of systolic and diastolic heart failure (241). Braunstein et *al.* (39) found essential hypertension to be the most common among chronic heart failure patients with 55%. A study by Levy et *al.* (242) correlates with the same results, finding that during up to 20.1 years of subjects follow-up (mean, 14.1 years), there were 392 new cases of heart failure; in 91% hypertension antedated the development of heart failure and this what makes it the most common risk factor for CHF.

44,32% of our patients had diabetes which is a common comorbidity in chronic heart failure. Braunstein et al. (39) found diabetes mellitus to be the second most common comorbidity in chronic heart failure patients with 31%. In a report from Framingham study (243), in which 5209 middle-aged people in the community were followed up prospectively for 10 years, diabetes was associated with a two-fold increase in risk of chronic heart failure in men and a fivefold increase in risk in women. Moreover, the increased risk persisted after adjustment for other potential contributors such as known coronary-artery disease, age, blood pressure, and cholesterol.

Among our heart failure patients, 30,92% were taking first intention treatment for HFrEF: an angiotensin-converting-enzyme inhibitor (ACEI) or an angiotensin receptor blocker (ARB) plus a beta blocker. 54,63% were taking second intention treatment: ACEI or ARB plus a beta blocker plus a mineralocorticoid/aldosterone receptor antagonist (spironolactone). This is per the 2016 ESC guidelines (3) for management of heart failure.

11. Study limitations:

In this work, we analyzed a limited number of variables. Biological evaluations of recruited patients were not followed up upon, so this study can not characterize the impact of changes in iron status and hemoglobin levels over time. The unavailability of the necessary reagents for iron statue assessment in central laboratory, and the high analysis cost in private laboratories, made it hard to collect enough data which lead to a delay in the study and a difficulty in recruitment.

12. Conclusion and Outlook:

Iron deficiency, with or without anemia, has already been established as common comorbidity of HFrEF. The results of this work showed high prevalence of ID (82.47 %) and anemia (59,79%) in a chronic heart failure population with a reduced left ventricular ejection fraction < 40%. Spironolactone has been inexplicably associated with ID, LVEF was also a significant parameter in the prevalence of iron deficiency within our population. Anemia was found to be marginally significant in ID.

Both conditions, together or independently, were found in scientific literature to be associated with poor clinical status and worse outcomes. Therefore, it is important to test all symptomatic patients with HF with reduced ejection fraction for the presence of ID. The diagnosis of ID in the heart failure population has and still remains a point of controversy. ESC currently defines absolute ID as ferritin levels <100 ng/ml and function ID as ferritin levels between 100 to 299 ng/ml and transferrin saturation of <20%. Anemia is defined as HB<13g/dl in men and HB<12 g/dl in women.

This study remains superficial and limited. It would be interesting to have further profound studies on the impact of changes in iron status and hemoglobin levels over time in HFrEF patients, or the impact of possible treatment of ID in the same population, with more specific and detailed parameters.

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ANNEXES:

Annex I: Patient recrutement form.

Etude de la carence martiale au cours de l'insuffisance cardiaque à FEVG altérée

Les Internes en pharmacie : **SELATNI Anfal et ABDICHE Amel** (**EMAIL :** Selatni.abdiche@gmail.com)

L'encadreur : **Dr BOUAFIA Mohamed**

Fiche de renseignements : N°..... (...../...../2021)

1/Identification du patient :

- Nom et Prénom : ..... (Lit:.....)
- Date et lieu de naissance...../...../.....à.....
- Age du patient : .....
- Numéro de téléphone : 1/..... /2.....
- Sexe : Homme ☐ Femme ☐ ☐ / Poids : ..... Kg / Taille : ..... C m
- Groupe sanguin : ..... IMC : ..... Kg/m²
- Durée d'hospitalisation : .....

2/Insuffisance cardiaque :

- FEVG :..... % (.....)
- Origine d'IC: Ischémique ☐ Valvulaire ☐ Congénitale ☐ Hypertensive ☐ Autres :.....

  .....
- Classe fonctionnelle (NYHA): Classe I ☐ Classe II ☐ Class III ☐ Classe IV ☐

3/Bilan Biologique : DATE DE PRELEVEMENT (...../...../2021)

Paramètres	Résultats	Unités	Valeurs de référence
Formule numération sanguine :			
Leucocytes			
Hémoglobine			
Hématocrite			
VGM			
CCMH			
PLAQUETTES			
GLYCEMIE A JEUN			
UREE SANGUINE			
CREATININE SANGUINE			
Clairance de la créatinine (MDRD)			
T.Q m (T.Q t = ...)			
T.C.Km (T.C.K t = ...)			
TP			
INR			
CHOLESTEROL TOTAL			
CHOLESTEROL HDL			
CHOLESTEROL LDL			
TRIGLYCERIDES			
ACIDE URIQUE			
IONOGRAMME SANGUIN :			
NATREMIE			

KALIEMIE			
Ferritinémie			
SATURATION DE TRANSFERRINE %(CST)			
ASAT			
ALAT			
CRP			
TSH			

Comorbidités et Antécédents

……

……

……

……

Facteurs de risques

HTA ☐ Diabète type ..... ☐ Tabagisme ☐ Obésité ☐ Hyper Cholestérolémie ☐ Alcoolisme ☐

Autres : ……

Traitements pharmacologiques indiqués

IEC ☐
- Captopril ☐
- Ramipril ☐

AUTRE :……
……

Bêtabloquants ☐
- Bisoprolol ☐
- Carvédilol ☐
- Succinate de metoprolol (Lp) ☐

AUTRE :……
……

ARAII ☐
- Valsartan ☐
- Losartan ☐
- Candésartan ☐

AUTRE:……
……

Diurétiques de l'anse ☐
- Furosémide ☐

AUTRE :……

Diurétiques épargneurs du potassium ☐
- Spironolactone/éplérénone ☐

Anti-Acides : ☐
- Pantoprazole ☐
- Oméprazole ☐
- Ranitidine ☐

AUTRE:……

- **Aspirine** ☐
- **Clopidogrel** ☐

Statines ☐
- Atorvastatine ☐
- Rosuvastatine ☐
- Fluvastatine ☐
- Simvastatine ☐
- Pravastatine ☐

AUTRE: ……
……

Anticoagulants ☐
- **AVK** ☐
  - Acenocoumarol ☐
  - Warfarine ☐
  - Fluindione ☐
- **AOD** ☐
  - Apixaban ☐
  - Rivaroxaban ☐
  - Dabigatran ☐
  - Edoxaban ☐

Autres médicaments : ……

……

Abstact:

The diagnosis of iron deficiency in chronic heart failure with a reduced ejection fraction is very important because it can be a key determinant of health-related quality of life in patients with chronic heart failure regardless of anemia status.

We analyzed data of 97 patients obtained from a prospective study of 3 months with the aim to evaluate the prevalence of iron deficiency within a population of chronic heart failure patients presenting a reduced ejection fraction defined as left ventricular ejection fraction LVEF < 40%. All while respecting the cut-off values for serum ferritin and TSAT (transferrin saturation) recommended in ESC's 2016 guidelines for iron deficiency diagnosis in chronic heart failure. In addition to an assessment of the prevalence of anemia in the same population. Moreover, we looked for significant associations with iron deficiency or anemia and other parameters.

Our results showed a high prevalence of ID, with it being present in 82,47% of our patients. We also found a significant association between ID and each of LVEF and aldosterone antagonist (spironolactone). In addition, 59,79% of our population were anemic, out of these 87,93% had a coexisting iron deficiency. A significant association between anemia and each of sex and statins was also noted.

Keywords: Iron Deficiency, LVEF, chronic heart failure, transferrin saturation, ferritin, Anemia.

Résumé :

Le diagnostic de la carence en fer dans l'insuffisance cardiaque chronique avec fraction d'éjection réduite est très important car elle peut être un déterminant clé de la qualité de vie liée à la santé chez les patients atteints d'insuffisance cardiaque chronique, indépendamment du statut anémique.

Nous avons analysé les données de 97 patients obtenues à partir d'une étude prospective de 3 mois dans le but d'évaluer la prévalence de la carence en fer au sein d'une population de patients insuffisants cardiaques chroniques présentant une fraction d'éjection réduite définie comme une fraction d'éjection ventriculaire gauche LVEF < 40%. Le tout en respectant les valeurs seuils de la ferritine sérique et de la saturation de transférine recommandées par l'ESC (2016) pour le diagnostic de la carence en fer dans l'insuffisance cardiaque chronique. En plus d'une évaluation de la prévalence de l'anémie dans la même population. De plus, nous avons recherché des associations significatives avec la carence en fer ou l'anémie et d'autres paramètres.

Nos résultats ont montré une prévalence élevée de la carence en fer, puisqu'elle était présente chez 82,47% de nos patients. Nous avons également trouvé une association significative entre la carence en fer et chacun des paramètres suivants : FEVG et antagoniste de l'aldostérone (spironolactone). Par ailleurs, 59,79% de notre population était anémique, dont 87,93% présentaient une carence en fer coexistante. Une association significative entre l'anémie et chacun des paramètres suivants : sex et statines, a également été notée.

Mots clés : Carence en fer, FEVG, insuffisance cardiaque chronique, saturation de la transferrine, ferritine, anémie.

ملخص:

تشخيص نقص الحديد في قصور القلب المزمن مع انخفاض في الكسر القذفي مهم جدا لأنه يمكن أن يكون عاملا رئيسيا في تحديد جودة الحياة الصحية للمرضى الذين يعانون من قصور مزمن في القلب بغض النظر عن حالة فقر الدم لديهم.

قمنا بتحليل بيانات 97 مريض تم الحصول عليهم بدراسة مستقبلية لمدة 3 أشهر بهدف تقييم مدى انتشار نقص الحديد بين هؤلاء المرضى الذين يعانون قصورا مزمنا في القلب بكسر قذفي منخفض تم تعريفه بالكسر القذفي للبطين الأيسر الأقل من 40%. هذه الدراسة أجريت وبنيت اعتمادا على القيم والتعاريف الموضوعة لمصل الفيريتين وتشبع الترانسفيرين و الموصى بها عام 2016 لتشخيص نقص الحديد عند مرضى قصور القلب المزمن في المبادئ التوجيهية لمنظمة القلب الأوروبية .ESC إضافة إلى تقييم نسبة انتشار فقر الدم في نفس مجموعة المرضى المدروس عليها, قد قمنا ايضا بالبحث عن اي رابط مهم موجود بين نقص الحديد أو فقر الدم وبين معايير أخرى مختلفة.

أظهرت نتائجنا انتشارا كبيرا لنقص الحديد بين الفئة المدروسة، حيث ان 82.47% من مرضانا كانوا يعانون من هذا النقص. ولقد وجدنا أيضا ارتباطا هاما بين نقص الحديد وكل من الكسر القذفي اليساري للقلب وكذا دواء السبيرونولاكتون. فضلاً عن ذلك فإنه ثبت على 59.79% من مرضانا إصابتهم بفقر الدم، ومن بين المصابين وجد أن 87.93% منهم يعانون من نقص في الحديد. ولوحظ أيضا وجود ارتباط هام بين فقر الدم وكل من الجنس وادوية الستاتين.

الكلمات المفتاحية: نقص الحديد، الكسر القذفي اليساري، قصور القلب المزمن، تشبع الترانسفيرين، الفيريتين ، فقر الدم.