Laboratory and metabolomic fingerprint in heart failure with preserved ejection fraction: from clinical classification to biomarker signature

Alberto Palazzuoli MD PhD FESC*, Francesco Tramonte MD*, Matteo Beltrami MD PhD^

*Cardiovascular Diseases Unit Cardio thoracic and vascular Department Le Scotte Hospital University of Siena Italy

^ San Giovanni di Dio Hospital Florence Italy

Alberto Palazzuoli MD, PhD, FESC, FEACVI

Cardiovascular Diseases Unit, Cardio thoracic and vascular Department,

Le Scotte Hospital, Viale Bracci 53100 Siena Italy

Fax: +39577233480

Phone: +39577585363- +39577585461

Email:palazzuoli2@unisi.it

Short Title: biomarkers signature in HFpEF

Word count: 4432

Key Words: Biomarkers; metabolomic, microRNA; HFpEF

 doi:10.20944/preprints202211.0551.v1

SUMMARY

Heart failure with preserved ejection fraction (HFpEF) remains a poorly characterized syndrome with many dark aspects related to different patients profile, various associated risk factors and wide aetiologies. It comprises several pathophysiological pathways related to endothelial dysfunction, myocardial fibrosis, extracellular matrix deposition and high inflammatory response. Up to now, it has been described only for clinical appearance and most common associated risk factors without an effective characterization of biological processes responsible for cardiovascular deteriorations. Recent advances in laboratory and metabolomic researches showed that HFpEF appears strictly related to specific cells and molecular mechanisms dysregulation. Some biomarkers are capable to early identify these processes adding new insights into diagnosis and risk stratification. Additionally recent advances on intermediate metabolites reflecting provide relevant information on intrinsic cellular and energetic substrate alterations. The systematic combination of clinical imaging and laboratory data may lead to a precision medicine approach providing prognostic and therapeutic advantages. Current review reports traditional and emerging biomarkers recently investigated in HFpEF setting, and it purpose a new diagnostic approach based on integrative information achieved from risk factors burden, hemodynamic dysfunction and biomarkers signature partnership.

INTRODUCTION

Heart failure with preserved ejection fraction (HFpEF) is an heterogenous syndrome with specific molecular, genetic, metabolomic features reflecting vascular and myocardial cell adaptations. [1] HFpEF encompasses different pathophysiological and cardiac structural profile compared to HF with reduced ejection fraction (HFrEF).[2] It accounts half of whole patients affected by heart failure (HF) but it shows a peculiar clinical profile, cardiac structural and functional alterations. Therefore, the selection criteria are often elusive and mainly based on ejection fraction cut off rather than distinct clinical and laboratory phenotype.[3] Most of inclusion criteria comprise the contemporary presence of left ventricular hypertrophy (LVH), altered diastolic dysfunction and elevation of natriuretic peptide (NP) associated with exertional dyspnea or reduced exercise tolerance. Indeed, recent clinical trials adopted wide inclusion criteria and characteristics of patients enrolled resulted inhomogeneous with various morphological and comorbidities patterns.[4,5] In this framework, an advanced analytic research investigating specific biomarkers in a well phenotyped population, could lead to a better understanding of molecular pathways and biological mechanisms responsible for HFpEF syndrome. The interaction among clinical variables, imaging features and biomarkers could became the model for future researches, and a combined network analysis may change current approach based on traditional knockdown/knockout study. [6]

 doi:10.20944/preprints202211.0551.v1

Conversely from HF with reduced systolic function in which myocyte loss, cellular death and consequent cardiac chamber enlargement, are the main features responsible for disease progression, HFpEF is characterised by collagen overexpression, myocardial fibrosis, extracellular matrix deposition and high inflammatory response.[7] All these mechanisms differently occur according to specific risk factors, comorbidities association, vascular and cardiac remodelling. [8] Thus an analysis starting from detailed phenotyping, cardiac structural alteration and distinctive laboratory investigation may challenge the current scenario going towards a precision medicine model with better targeted therapeutic profiling. [9] This personalized setting begin from machine learning analysis of big data in order to resolve disease heterogeneity by identifying patients within particular subtypes and predicting response to the therapy.

DIFFERENT HFpEF PHENOGROUPS

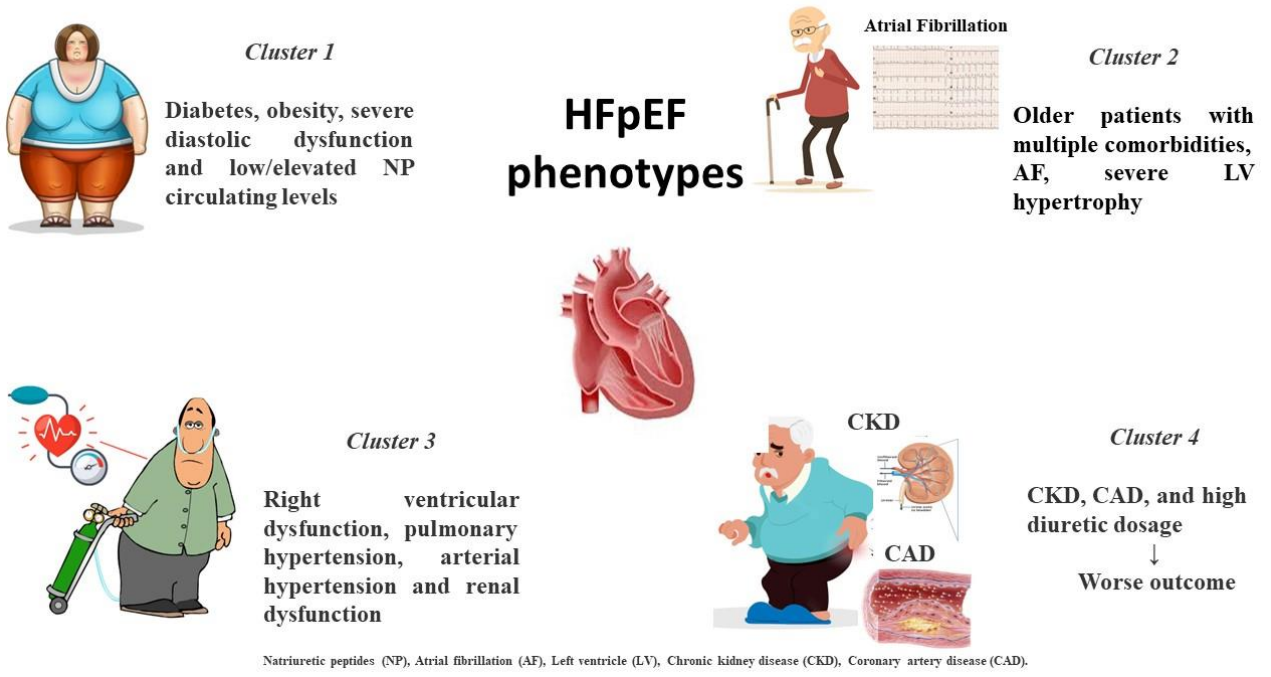
Despite recent advances in treatment diagnosis and recognition, HFpEF remains a poorly characterized syndrome with many dark aspects related to different patients profile, associated risk factors and pathophysiological pathways.[10] Large trials showed a wide prevalence of LVH left atrial dilatation diastolic dysfunction and pulmonary hypertension. Moreover patients presented various extracardiac comorbidities such as diabetes CKD anemia chronic lung diseases obesity metabolic dysfunction. [5,11-13] Because the interventional Trials did not distinguish among different risk factors and underlying diseases, the one-size-fits-all approach might explain the lack of efficacy and benefit for the tested treatments. Based on the different pathophysiological drivers, some Authors suggested different HFpEF subtypes linked to cardiometabolic alterations, body size conformation or peripheral maladaptation. These appraisals may be related respectively to the presence of systemic disorders leading to skeletal muscle metabolism alterations and vascular rarefaction.[14] Since all these features are widely expressed in HFpEF , the diagnosis based only on cardiac morphology and dysfunction remains difficult to interpret and often misleading. Current pictures may configure a wide HFpEF phenotype that differently interfere on cardiac structure and cardiovascular remodelling related to the underlying biological process and pathophysiological contributor, despite a similar EF.

Notably, recent machine learning analysis attempt to cluster specific phenotype by latent class study. In a post hoc analysis of TOPCAT patients were classified into three categories according to vascular and cardiac remodelling: patients with mild LV hypertrophy and chronic pulmonary disease with normal vascular stiffness characterized for increased expression of metalloproteinase; older patients with multiple comorbidities LV hypertrophy and reduced vascular compliance, characterized for elevated tissue calcification biomarker; obese subgroup with several metabolic

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

alterations increased levels of renin angiotensin system activity, lipidic profile derangement and increased inflammatory pattern. [15] Similarly, another study identified a group including youngest with increased body mass index, typical abnormalities in cardiac structure and function and low natriuretic peptide levels; a pattern with high prevalence of diabetes and obesity characterized for severe diastolic dysfunction and elevated NP circulating levels; a cluster characterized by RV dysfunction, pulmonary hypertension, and renal dysfunction, experiencing the worst outcomes.[16]

Finally a combined analysis of SwedeHF and CHECK HF registries distinguished among 5 different phenogroups accordingly to a combined approach including risk factors and associated comorbidities confirming that cluster with CKD, coronary artery disease (CAD) and high diuretic amount revealed the worst outcome. [17] FIGURE 1



Natriuretic peptides (NP), Atrial fibrillation (AF), Left ventricle (LV), Chronic kidney disease (CKD), Coronary artery disease (CAD).

Taken together these data reveal some common features but also some discrepancies underlying the needing of a more homogenous classification. Of note a detailed laboratory screening capable to identify specific cluster by the combination of structural cardiac abnormalities with underlying molecular mechanisms responsible for disease progression may open the way for a better HFpEF understanding opening targeted therapeutic opportunities. [18]

CURRENT BIOMARKERS IN HFpEF

Several risk prediction scores have been produced including biomarkers,(mainly natriuretic peptides NP) ,but important gaps exists regarding the knowledge of underlying pathophysiological

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

mechanisms, biological process And disease evolution. Circulating biomarkers should reflect cardiac and extra cardiac disorders responsible for HFpEF development and related pathological pathways. [19,20] Since HFpEF is characterized by LVH with increased parietal stress, systemic vascular damage and stiffness, increased inflammation, and enhanced fibrosis, we recognize four main biomarkers targets: Myocardial injury, extracellular fibrosis, inflammation and endothelial dysfunction. TABLE 1

	Name of Biomarker	Mechanism of action
Markers of myocardial injury	↑↑ High sensivity troponin	the final results of microvascular dysfunction, and subendocardial layer damage due to systemic oxygen reduction .
	↑ Natriuretic peptides	related to diuresis and natriureis that fovour the congestion reduction and euvolemia
	↑↑ Adrenomedullin	regulatory peptide produced by endothelial and smooth muscle cells with antiproliferative vasodilatatory and antiapoptotic effects.
Markers of extracellular fibrosis	↑↑ Galectin -3	inflammatory and pro fibrotic processes and it is synthetized by macrophage
	↑↑ Soluble ST2	roduced by myocardial cells, but also smooth muscle cells and endothelium are capable to synthetize the peptide in relation to congestion
	↑↑ Matrix Metalloproteinases	involved in collagen synthesis and collagen degradation
	↑↑ Pro collagen type I (PIP) and procollagen type III N-terminal peptide (PIINP),	reflect collagen increases deposition and turnover.
Markers of inflammation	↑↑ CRP and pentraxin	inducing complement and cytokine stimulation causing myocyte loss and endothelial dysfunction by NO production decrease.
	↑↑ Grow differentiation factor 15	expressed in inflammatory chronuic diseases, lung, kidney, and cardiovascular diseases and providing additional informations on LV

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

		remodeling and function.
	↑↑ Intereleukin-6	contributes by direct myocites damage and indirect inflammatory burden elevation.
	↑ Tumor necrosis factor α	correlates with atrial dimension and diastolic dysfunction degree
Markers of endothelial dysfunction	↑↑ vascular cel adhesion molecules (VCAM) and E selectin	activates von villerbrand and other pro thrombotic factor
	↑ endothelin 1	secreted by the endothelial cells in response to renin angiotensin system activation.
	↑↑ Plasminogen activator inhibitor	in association with D-dimer levels suggesting an association with pro thrombotic and procoagulant state
	↑↑ Insulin grow factor binding	left atrial dysfunction and dilatation reflecting diastolic dysfunction in HFpEF.

Myocardial Injury-High sensivity troponin (HsTn) is universally considered a markers of myocardial damage in acute coronary syndrome, however it has a prognostic significance in HF and it implies myocardial damage apoptosis and progressive fibers loss independently on coronary vessel diseases. It could be the final results of microvascular dysfunction, and subendocardial layer damage due to systemic oxygen reduction configuring an altered supply-demand mismatch. Other features such as increased wall tension, high LV filling pressure and right ventricular dysfunction are related with increased HsTn levels.[21] In patients with HFpEF increased HsTn level correspond to more severe diastolic degree and higher pulmonary pressure. Elevated levels are also associated with increased wall stress and much more pronounced LV hypertrophy subtending and increased workload. [22] Many reports showed that HsTn predict poor outcome in HFpEF, with better prediction in men than in women. In hospitalized patients with acute HFpEF the persistence of increased HsTn levels at both admission and discharge is related with increased events rate in terms of rehospitalization and death.[23,24] Similarly in TOPCAT trial, elevation of HsTn was independently associated with increased risk of hospitalization and cardiovascular events. [25]Current findings were confirmed in the PARAGON study in which even a mild HsTn elevation was associated with worse outcome across 3 years follow-up, and patients taking salcubitril valsartan showed a significative reduction compared to placebo. [26]

 doi:10.20944/preprints202211.0551.v1

Natriuretic peptides (NPs) are the hallmark biomarkers in HF and its measurement is accounted in HF guidelines across the spectrum of the whole EF.[27] The biologically active NP form and its precursor amino-termianl portion pro B type natriuretic peptide is cleaved into NT-pro BNP and BNP are released in response to enhanced cardiac wall tension and increased filling pressure, their levels increase proportionally to the degree of systolic dysfunction. The two peptides are released in response to sympatethic activity systemic vasoconstriction and and fluid retention as opposite response to the increased neuro hormonal overdrive.[28] NP activity counteract sympathetic activity promoting cardiac afterload reduction and myocardial relaxation by direct vasodilatation and myocardial relaxation effects. The main mechanism of action is related to diuresis and natriureis that fovour the congestion reduction and euvolemia. [29] The Levels of NPs are directly related to intracardiac pressure values including LV end diastolic pressure, wedge pressure, and pulmonary systolic pressure. Both peptides are largely analyzed in patients with reduced systolic function as valuable diagnostic and prognostic features, in HFpEF they are generally less increased but they keep their diagnostic relevance.[30] Some authors believe that this appaeranace is due to the reduced wall stress in this setting together with extracardiac conditions such as metabolic syndrome chronic lung disease and obesity in which adipocyte cells favor a reduced receptor expression. [31] These comorbidities are often associated in HFpEF causing a relevant NPs levels variations. Despite some studies reveals that some HFpEF cluster experienced low NPs below 100 pg/ml, recent metanalysis showed an optimal diagnostic accuracy in this setting (AUC 0.80 CI 0.73-0.87). [32] Moreover, a combined analysis of NPs and Tn showed that patients with higher plasma level have the increased risk for death and hospitalization. Finally in acute patients, NP assay reveals similar prognostic informations in HFpEF as In HFrEF, and the related changes during hospitalization. confer equal risk assessment adjusted for potential confounding factors.[34]

Adrenomedullin is a regulatory peptide produced by endothelial and smooth muscle cells with antiproliferative vasodilatatory and antiapoptotic effects. It is synthetized mainly by adrenal medulla but its receptors are expressed in many tissues such as lung heart and kidney. {35] It is an important biomarker of pulmonary and systemic congestion and its is produced in relation to sympathetic activity.[36] It counteracts systemic vasoconstriction induced by renin angiotensin system activation facilitating vascular permeability and elastance. Due to its instability with plasma protein and short half life, a reliable quantification of ADM is difficult to achieve and its precursor mid regional pro- hormone (MRpro-ADM) is usually measured. [37] A large study conformaed the close relationship between ADM and congestion in patients with worseining heart failure, therefore high plasma level appears related to increased risk and recurrent hospitalization for HF. [38]MRpro-

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

ADM measured at admission is also related to all cause of cardiovascular mortality, sudden death and cardiac arrest. In patients with acute coronary syndrome (ACS) elevated MRpro-ADM levels predict the risk for HF occurrence . Finally in the PROTECT trial the MRproADM was related to longer hospitalization increased congestion signs and elevated NP levels, the assessment before discharge confers relevant prognostic information related to incomplete decongestion statuis and early rehospitalization risk.[40]

Extracellular Fibrosis-Collagen deposition and increased myocardial fibrosis are two relevant features in HFpEF. The more extensively analyzed biomarkers of this process are Galectin 3 and soluble ST2- Galectin -3 is a glycoprotein involved in many inflammatory and pro fibrotic processes as galactosidases family member, and it is synthetized by macrophage . [41] It directly increases fibroblast proliferation and fibrogenesis in animal models inducing myocardial and vascular stiffness. Its is also associated with renal dysfunction and LV remodelling.[42] Galectin-3 Inhibition attenuated myocardial fibrosis and induce a reverse remodelling by reduction of systemic overaload. [44] High Galectin 3 levels are associated with poor outcome in both patients with HFrEF and HFpEF. In patients with elevated levels, galectin is associated with other comorbidities such as hypertension and CKD and it is an useful markers for target therapy and risk stratification. [45] Therefore, changes in galectin 3 over time provide prognostic insights in patients with HFpEF.[46]

Soluble ST2 is another markers reflection myocardial fibrosis and it is overexpressed in HFrEF and HFpEF patients. It is primarily produced by myocardial cells, but also smooth muscle cells and endothelium are capable to synthetize the peptide in relation to congestion or pro fibrotic stimuli.[47] In HFpEF patients the addition of ST2 to NPs provide additive prognostic information, therefore higher ST2 phenotype indicated a more compromised diastolic dysfunction. [48,49] Notably a mataanalysis demonstrated that ST2 is capable to predict outcome indepentently on EF values.[50]

Matrix Metalloproteinases (MMP) and tissue inhibitor of metalloproteinases (TIMP) are two endopeptidases inducing extracellular collagen deposition reasonable accounted as biomarkers of fibrosis in HFpEF. [51] Collagenases is an enzyme family with different characteristics and may be considered in the context between collagen synthesis and collagen degradation. Elevated levels of MMP2 and MMP9 are related to increased risk in HFpEF but are increased also in HFrEF after myocardial infraction. [52]In the PARAGON trial high level of TIMP, a markers of impaired collagen degradation, is associated with increased event rate. [53]

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

Additional collagen biomarkers such as Pro collagen type I (PIP) and procollagen type III N-terminal peptide (PIINP), demonstrated a predictive role in high risk patients for HFpEF development. Both biomarkers reflect collagen increases deposition and turnover. They appear to be associated with extent of collagen deposition in myocardial biopsies. [54] However in cross sectional analysis showed contrasting results: in Framingham sub-study PIIINP was not associated with echocardiographic abnormalities, whereas in Cardiovascular Health Study it was associated with increased risk of incident HF. [55,56]

Inflammation-Systemic inflammation is a typical features of HFpEF, it reflects the immune response to cardiac remodelling, systemic vascular injury and underling triggers often associated with diseases such as metabolic syndrome diabetes chronic lung disease and anemia. [57] Inflammation can occur differently in every HFpEF phenotype and it can be analyzed by several biomarkers showed a positive association with outcome. C-Reactive protein is the wider analyzed markers and it is assocatide with increased risk in ACS and HF. A comparison study differentiating CRP between HFrEF and HFpEF demonstrated that in the latter it has a better prognostication, adding new information respect to NP. [58] CRP and pentraxin are significantly higher in acute HFpEF patienst compared to stable and they correlated with diastolic dysfunction degree. [59]CRP has a direct role in inducing complement and cytokine stimulation causing myocyte loss and endothelial dysfunction by NO production decrease. CRP increase is also related to immune response mediated by lymphocyte T and monocyte cells. Current immune- inflammatory status may trigger microvascular dysfunction by inducing endothelial permeability and adhesion molecules, and the increase of reactive oxygen species bioavailability. [60]

Grow differentiation factor 15 (GDF-15) is a member of cytokines and it belongs to transforming grow factor beta family. It is highly expressed in inflammatory chronuic diseases, lung, kidney, and cardiovascular diseases. [61] Since it integrates information from cardiac and systemic diseases it reflects the interplay among different apparatus, but it is not specific of CV diseases or HF. [62] Recent metanalysis demonstrated in patients with high risk burden it is related to increased incidence of HF providing additional informations on LV remodeling and function. [63] In HFpEF it is similarly elevated than HFrEF but it has additional prognostic power compared to NTproBNP. Indeed, subjects with low NTpro BNP levels and high GDF-15 values, the risk of cardiovascular death is comparable to those with high NP. [62] This finding confirm the role of GDF15 as intermediate markers of inflammatory and multi organ injury.

Intereleukin-6 (IL6) and interleukin 1ß (ILß) are the most famous members of citokyne family. They are produced by activated macrophages and they are involved in several inflammatory

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

and immunitary processes. [64] They contributes by direct myocites damage and indirect inflammatory burden elevation, to the cardiac damage and remodelling. Therefore cytokine mediated systemic inflammation impairs skeletal muscle metabolism and circulation. [65] Notably, the IL-1 inhibitor Anakinra, is capable to reduce hospitalization improving exercise tolerance in HFpEF. therefore the drug improves treadmill parameters and quality of life.[66]

Tumor necrosis factor α(TNF) is another interleukin highly expressed in HFpEF, it correlates with adverse otcome in a cross sectional analysis.[67] However several confounding factors related to systemic immune system, may influence its levels. In stable HFpEF patients it correlates with atrial dimension and diastolic dysfunction degree, moreover it is additive compared with NP.In Health ABC study it correlates with HFpEF but it did not provide additional prognostic informations., finally anticitokyne treatment with specific antibody etanercept did not improve quality of life neither outcome. [68,69]

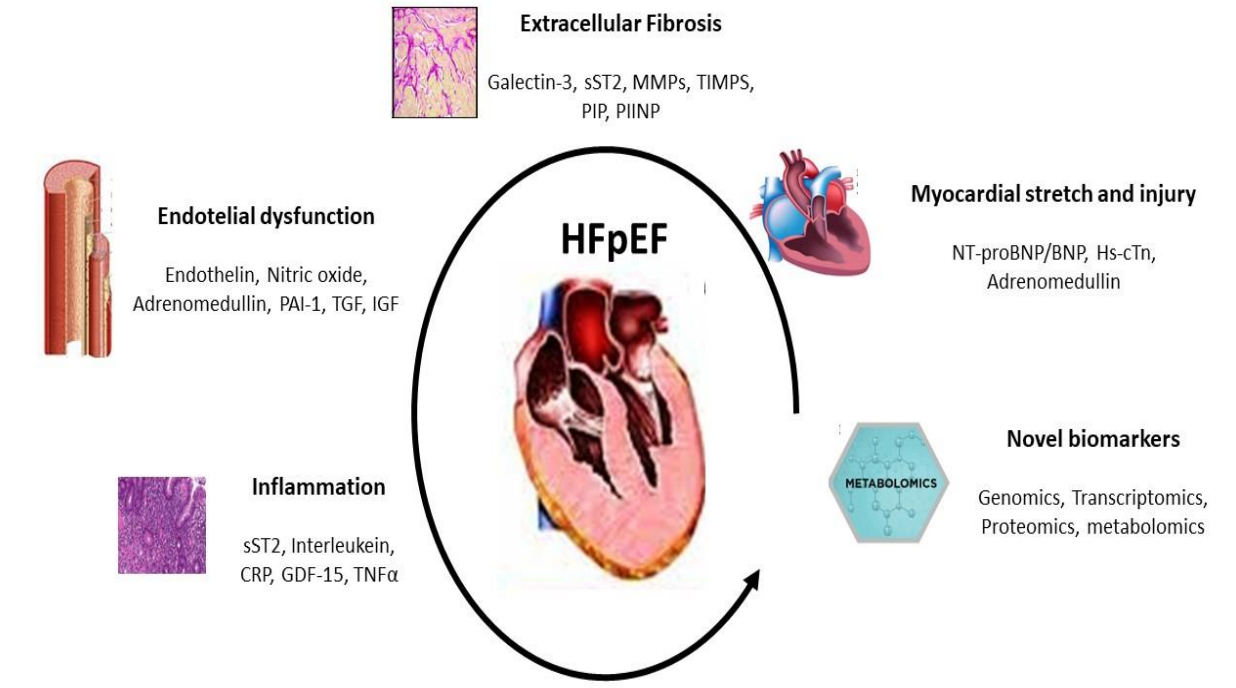
Endothelial dysfunction- microcirculation and endothelial cells are two important features for HFpEF occurrence, and microvascular dysfunction is one of the most therapeutic target. Dysfunctiona endothelium increases the expersion of adhesion molecules such as vascular cel adhesion molecules (VCAM) induced cell adhesion molecules (ICAM) and E selectin that activates von villerbrand and other pro thrombotic factors. Therefore Tissue grow factor (TGF) and Insulin grow factors (IGF) are other two items of vascular alteration and increased proliferation . [70] The pro thrombotic cascade is also emphasized by several coagulation alteration involving Factor V and VII, Tissue plasminogen activator (TPA), inducing endothelial damage and loss of vascular integrity. Current vascular, coagulative, and thrombotic alterations may lead to a progressive microvascular obstruction capillary obliteration and loss of capillary integrity. [71] These processes induce increased resistance and enhanced cardiac workload at systemic and pulmonary districts. Therefore vascular damage is characterized by intima media hyperplasia, disarray of smooth muscle cells, intimal fibrosis leading to progressive capillary reduction and narrowing. Current alterations reduces nitric oxide (NO) production and its mediator guanosine monophosphate cyclase (GMPc) causing vasoconstriction, reduction of viscoelastic properties, altered oxygen consumption and utilization with increased oxidative stress.[72,73] Unfortunately no reliable blood biomarker exist to measure these processes,and only in vitro studies can document these endothelial alterations. Nevertheless, a direct GMPc activator Vericiguat is capable to improve vascular tone and to reduce cardiac stiffness. A reliable marker of vascocntrictction and vascular tone is endothelin 1 (ET1) directly secreted by the endothelial cells in response to renin angiotensin system activation, hyperglicemia, hypertension and systemic inflammation. It is the most powerful vasoconstrictors

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

and is highly exporessed in pulmonary hypertension,severe hypertensive status and HF irrespective of EF.[74] Et-1 levels were predictive of all cause mortality and it was associated with increased hospitalization rate in a longitudinal study of HFpEF. [75]Therefore in a RELAX analysis ET1 correlates with reduced exercise oxygen consumption and significantly associate with hifghre NTproBNP and Galectin 3 levels.[76]

Plasminogen activator inhibitor (PAI-1) is the main inhibitor of tissue plasmimogen activator and intrinsic fibrinolytic system. It is increased in patients with HFpEF in association with D-dimer levels suggesting an association with pro thrombotic and procoagulant state in this setting. [77]In the LURIC study it is a prognostic index of mortality and CV events although a longitudinal study confirmed only an association with markers of renal damage and NP.[78]

Insulin grow factor binding (IGFBP) is associated with inflammation, cell adhesion and senescence. It is increased according to left atrial dysfunction and dilatation reflecting diastolic dysfunction in HFpEF. [79]In a machine learning study subjects with high inflammatory phenotype elevated comorbidity burden and renal dysfunction it is elevated and associated with increased hospitalization risk. [80]In I-PRESERVE trial IGFBP was associated with increased risk of CV events and HF severity.[81] Finally in asymptomatic patients with LV hypertrophy IGFBP identifies subjets with altered diastolic function suggesting a role in early identification and screening of HFpEF.[82] FIGURE 2 (graphical abstract)



Soluble ST2 (sST2), Matrix metalloproteinases (MMPs), Tissue inhibitors of metalloproteinases (TIMPs), Plasminogen activator inhibitor 1 (PAI-1), C-reactive protein (CRP), N-terminal pro–B-type natriuretic peptide (NT-proBNP), high-sensitivity cardiac troponin (hs-cTn), Pro collagen type I (PIP), procollagen type III N-terminal peptide (PIINP), Grow differentiation factor 15 (GDF-15), Tumor necrosis factor α (TNFα), Tissue grow factor (TGF), Insulin grow factors (IGF)

 doi:10.20944/preprints202211.0551.v1

METABOLOMIC SIGNATURE

HFpEF is associated with indices of increased inflammation and oxidative stress, impaired lipid metabolism, increased collagen synthesis, and downregulated nitric oxide signaling. [83] . The most studied metabolites involved in the metabolic profile of HFpEF are serine, Lysophosphatidilcolyne (LPC), kynurenine and cystine, hydroxyproline, lactate, cGMP, symmetric dimethylarginine (SDMA), arginine, cAMP and acylcarnitine. [83]

In HFpEF mouse models, serine deficiency has been associated with inflammatory response and oxidative stress [84] Serine is a non-essential amino acid and has different physiological functions, including the immunoregulatory actions. Through one-carbon metabolism in macrophages, serine is critical for the generation of phospholipid, biosynthesis of purine and thymidine, and production of methyl donor of S-adenosyl-methionine (SAM) and cellular glutathione. Serine is essential for production of proinflammatory cytokines in M1 macrophages [85]. In the inflammatory process the activation of serine proteases induces a serine deficiency. Cathepsin G is another serine protease of PMN azurophile granules that hydrolyses several types of proteins. Cathepsin G exerting strong pro-inflammatory effects with vascular and systemic impact. [86] Elastase is the most involved serine protease contained in the azurophile granules of polymorphonuclear cells (PMN, or neutrophils). When discharged upon PMN activation elastase, it has a direct effects on the degradation of collagen, elastin and fibronectin. These processes could represent the potential basis of HFpEF development and strictly related to the reduced myocardial compliance with diastolic dysfunction and consequent LA dilatation. Similarly, few studies found a significant increase of hydroxyproline. It is produced by hydroxylation of proline by prolyl hydroxylase and have the role of stability of collagen and this dysregulation contribute to the myocardial fibrosis in HFpEF. [87,88]

In endothelial cells, NO is generated by endothelial nitric oxide synthase (eNOS) through the conversion of its substrate, L-arginine to L-citrulline. Arginine is known to act as a substrate for NO production by endothelial cells [89]. The lower levels of NO substrate arginine reflects higher concentrations of SDMA associated with worsening renal function and microvascular dysfunction reduced vasodilatory properties [90,91].

Activation of cGMP precursors via natriuretic peptide increase the cGMP levels and a subsequent protein-kinase G (PKG), [83]. Furthermore the cGMP/PKG signaling cascade phosphorylates many sarcomeric and cytosolic proteins. Downregulation of myocardial cGMP-PKG signalling in HFpEF is related to reduced myocardial brain-type NP (BNP) expression and

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

increased microvascular inflammation and oxidative stress, which impair both the NP-cGMP and NO-cGMP axes. Decreased levels of cGMP in HFpEF, and subsequently of PKG, were associated with increased resting tension and myocyte stiffness. This activation leads to Titin phosphorilation reduction that modulates passive stiffness of cardiac muscle that negatively affects passive diastolic distention. [92,1]

Another finding is high levels of cystine that is an indirect index of inflammation [93] . Cystine enters the cell and then is reduced to cysteine, which is involved in the synthesis of glutathione (GSH). Glutathione peroxidase 4 uses GSH as a substrate to scavenge lipid peroxidation and reduce oxidative stress. The important role of cysteine in maintaining and transducing redox signals in the mitochondria. [94] Redox-dependent cysteine modification has been studied most extensively in cardiac tissue following ischemia/reperfusion injury, which deprives cardiac tissue of oxygen in the ischemic state and generates a ROS burst.[95]

Kynurenine (Kyn) is a regulator of immune response, metabolized from tryptophan (Trp) during inflammatory conditions. [96]. The Kyn pathway of Trp is the most active process of Trp metabolism and produces metabolites including kynurenic acid and nicotinamide adenine dinucleotide (NAD+). Notable is the involvement of NAD+ in oxidative phosphorylation. The Kyn pathway is initiated by the enzymes tryptophan 2,3-dioxygenase (TDO) and indoleamine 2,3-dioxygenase (IDO and IDO2). Kyn is incresed in HFpEF and have a role in the regulation of inflammation response mediated through the function as a ligand of the aryl hydrocarbon receptor (AhR) and as a transcription factor that controls local and systemic immune responses.

cAMP is produced via β-AR signalling and inhibited via AMP-hydrolysing enzyme phosphodiesterases , at least 5 families expressed in the heart (PDE1, PDE2, PDE3, PDE4 and PDE8) Low cAMP levels in HFpEF suggesting impaired cell signaling. Disruptions in β-Adrenergic R microdomains in HFpEF, in the Obese individuals with type 2 diabetes heart exhibiting altered β-AR expression levels, blunted β-AR responsiveness and evidence of altered β2-AR-coupled PDE activity [97] . Current signal cell dysregulation may alter both intracellular calcium (Ca) membrane signal, and myocyte energetic process linked to glycogenolysis and lipolysis.

Additionally, HFpEF patients displayed an elevated concentrations of medium and long-chain acylcarnitines [98] . The acyl derivatives play a key role in fatty acid uptake and mitochondrial metabolism, an increase in acylcarnitines may imply inefficient β-oxidation in HFpEF patients[99] Several reports have demonstrated the detrimental effects of long-chain

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

acylcarnitines, in this HF model because of pro-inflammatory and pro-arrhythmogenic activity [99, 100].

Low levels of lysophosphatidylcholine suggests a dysregulated phospholipid metabolism.[83] Lysophosphatidylcholine (LPC), also called lysolecithins, is a class of lipid biomolecule derived by the cleaving of phosphatidylcholine (PC) via the action of phospholipase A2 (PLA2). Phosphatidylcholine is required for assembly of VLDLs and chylomicrons. Moreover, small alterations in phospholipid levels appear to have large implications related to the metabolic syndrome and fatty acid oxidation signaling. [101]. Dysregulated lipid metabolism could drive to adipose accumulation around pericardium and muscle compartment. Also, circulating fatty acids impair insulin sensitivity through binding to the plasma membrane receptor Toll-like receptor 4 in tissues of obese animals. This process results in the activation of signaling proteins, such as inhibitor of nuclear factor-κB kinase, c-Jun N-terminal kinase , and mitogen-activated protein kinase, that negatively dysregulate the metabolic axis of macrophage and favoring chronic inflammation. [102] TABLE 2

	Biomarkers	**Altered Cell Mechanism**
Increased inflammation	↓ Serine	immunoregulatory actions: essential for production of proinflammatory cytokines in M1 macrophages
	↑ Cathepsin G	stimulating the production of cytokines and chemokines.
	↑ Cystine	key player in conditions of oxidative stress
	↑ Kynurenine	controls local and systemic immune responses
Increased collagen syntesis and reduced myocardial compliance	↑ Hydroxyproline	role of stability of collagen and this dysregulation contribute to the myocardial fibrosis
	↑ Elastase	degradation of extracellular matrix components, including collagen, elastin and fibronectin
	↓ cGMP/PKG signaling	phosphorilation reduction associated to passive stiffness of cardiac muscle

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 29 November 2022 doi:10.20944/preprints202211.0551.v1

Endothelial dysfunction	↓ Arginine ↑ SDMA	substrate for NO production by endothelial cells with reduced vasodilatory effects alternative methylation product of L-arginine associated with worsening renal function and microvascular dysfunction
Energetic impairment	↓ cAMP ↑ Acylcarnitine ↑ Tryptophan	is produced via β-AR signalling imply inefficient β-oxidation produces metabolites including kynurenic acid and nicotinamide adenine dinucleotide
Metabolic lipides impairment	↓ Lysophosphatidylcholine ↓ cAMP	is required for assembly of VLDLs and chylomicrons involved in lipolysis

CIRCULATING MicroRNA EVIDENCES

The non coding genome which indicates small and long non coding RNA is involved in gene regulation. Multi-microRNA are 21-22 nucleotide single stranded RNAs that bind complementary messenger leading the degradation. They have been implicated in phathophisiologic processes that conduct to HFpEf. miRNA correlated with NT-proBNP are highly discriminatory and improved specificity and accuracy in identifying nonacute HF. [103] The subtype stratification, mir-24-3p has been reported to regulate apoptosis and vascularity in ischemic heart disease; mir-503-5p has been implicated in driving cardiomyocytes specification; miR-30a-5p has been shown to regulate autophagy during myocardial injury induced by Angiotensin II; and miR-106a-5p promotes hypertrophy through targeting mitofusin 2, a mitochondrial protein in regulating cardiac function . [104] . There are also pro-hypertrophic miRNAs, such as miR-208, miR-22, miR-21, miR-25, miR-34, miR-199a, miR-212/132, and miR-23. [105] miR-3135b and miR-3908 were significantly upregulated in HFpEF and are involved in metabolic regulators for serum lipids and blood glucose[106]. Certain circulating miRNA may serve as markers of response of therapy: Sucharov et al. identified a set of miRNAs (miRNA 208a-3p and miRNA-591) differentially expressed in HF patients who respond to beta-blockers therapy.[107] Despite increasing literature in this setting, there is no current consensus on the choice of a specific circulating miRNA serve as HFpEF biomarker. This is due to the lack of standardized methods, different population analyzed

and deficienty in multi center trials combining laboratory data in systematic methods with homogeneous analysis.

CONCLUSIONS

Since HFpEF is an heterogeneous syndrome characterized by multiple risk factors and several associated conditions, by the simple phenotypical classification is hard to distinguish the main pathophysiological driver. In this framework a detailed laboratory screening may better elucidate underlying mechanisms responsible for HFpEF appearance and evolution. Behind new and traditional biomarkers of inflammation, cardiovascular dysfunction and fibrosis, some emerging metabolites responsible for altered cell signals , energetic substrate, and excessive immune response revealed additional diagnostic properties. The challenge of future researches may systematically address the real value of clinical laboratory and metabolomic combination, to effectively initiate a precision medicine methodology.

FIGURES AND TABLES LEGEND

FIGURE 1: distinct HFpEF clinical phenotypes based on clinical presentation associated metabolic disorders and comorbidities

FIGURE 2: potential pathophysiological mechanisms occurring in HFpEF: each disorders can be recognized by specific biomarker increase and overexpression. The partnership between clinical and laboratory information may better target HFpEF profile

TABLE 1: Circulating biomarkers responsible for cardiac remodeling reflecting myocardial injury, collagen overexpression, inflammation, and vascular damage

TABLE 2: most common metabolomic pathways analyzed in HFpEF: different mechanisms suggest metabolic and energetic substrate alterations involving several cells including myocytes, macrophages, fibroblasts, and endothelium.

REFERENCES

1. Lewis GA, Schelbert EB, Williams SG, Cunnington C, Ahmed F, McDonagh TA, Miller CA. Biological Phenotypes of Heart Failure With Preserved Ejection Fraction. J Am Coll Cardiol. 2017 Oct 24;70(17):2186-2200. doi: 10.1016/j.jacc.2017.09.006. PMID: 29050567.
2. Borlaug BA, Paulus WJ. Heart failure with preserved ejection fraction: pathophysiology,

diagnosis, and treatment. Eur Heart J. 2011 ;32(6):670-9. doi: 10.1093/eurheartj/ehq426

3. Luo H, Xu Y, Yue F, Zhang C, Chen C. Quality of inclusion criteria in the registered clinical trials of heart failure with preserved ejection fraction: Is it time for a change? Int J Cardiol. 2018 ;254:210-214. doi: 10.1016/j.ijcard.2017
4. Shah AM, Shah SJ, Anand IS, Sweitzer NK, O'Meara E, Heitner JF, Sopko G, Li G, Assmann SF, McKinlay SM, Pitt B, Pfeffer MA, Solomon SD; TOPCAT Investigators. Cardiac structure and function in heart failure with preserved ejection fraction: baseline findings from the echocardiographic study of the Treatment of Preserved Cardiac Function Heart Failure with an Aldosterone Antagonist trial. Circ Heart Fail. 2014 Jan;7(1):104-15. doi: 10.1161/CIRCHEARTFAILURE.113.000887. Epub 2013 Nov 18. PMID: 24249049; PMCID: PMC4467731.
5. Shah AM, Cikes M, Prasad N, Li G, Getchevski S, Claggett B, Rizkala A, Lukashevich I, O'Meara E, Ryan JJ, Shah SJ, Mullens W, Zile MR, Lam CSP, McMurray JJV, Solomon SD; PARAGON-HF Investigators. Echocardiographic Features of Patients With Heart Failure and Preserved Left Ventricular Ejection Fraction. J Am Coll Cardiol. 2019 Dec 10;74(23):2858-2873. doi: 10.1016/j.jacc.2019.09.063. PMID: 31806129.
6. Shah SJ, Katz DH, Deo RC. Phenotypic spectrum of heart failure with preserved ejection fraction. Heart Fail Clin. 2014 Jul;10(3):407-18. doi: 10.1016/j.hfc.2014.04.008. Epub 2014 May 22. PMID: 24975905; PMCID: PMC4076705.
7. Kelly JP, Mentz RJ, Mebazaa A, Voors AA, Butler J, Roessig L, Fiuzat M, Zannad F, Pitt B, O'Connor CM, Lam CSP. Patient selection in heart failure with preserved ejection fraction clinical trials. J Am Coll Cardiol. 2015 Apr 28;65(16):1668-1682. doi: 10.1016/j.jacc.2015.03.043. PMID: 25908073; PMCID: PMC4713836.
8. Vaduganathan M, Michel A, Hall K, Mulligan C, Nodari S, Shah SJ, Senni M, Triggiani M, Butler J, Gheorghiade M. Spectrum of epidemiological and clinical findings in patients with heart failure with preserved ejection fraction stratified by study design: a systematic review. Eur J Heart Fail. 2016 Jan;18(1):54-65. doi: 10.1002/ejhf.442. Epub 2015 Dec 3. PMID: 26634799.
9. Shah SJ. Innovative Clinical Trial Designs for Precision Medicine in Heart Failure with Preserved Ejection Fraction. J Cardiovasc Transl Res. 2017 Jun;10(3):322-336. doi: 10.1007/s12265-017-9759-8. Epub 2017 Jul 5. PMID: 28681133; PMCID: PMC5571647.
10. Palazzuoli A, Caravita S, Paolillo S, Ghio S, Tocchetti CG, Ruocco G, Correale M, Ambrosio G, Perrone Filardi P, Senni M; Italian Society of Cardiology Heart Failure Study Group. Current gaps in HFpEF trials: Time to reconsider patients' selection and to target phenotypes. Prog Cardiovasc Dis. 2021 Jul-Aug;67:89-97. doi: 10.1016/j.pcad.2021.03.007. Epub 2021 Apr 9.

PMID: 33839166.

11. Shah AM, Claggett B, Sweitzer NK, Shah SJ, Anand IS, O'Meara E, Desai AS, Heitner JF, Li G, Fang J, Rouleau J, Zile MR, Markov V, Ryabov V, Reis G, Assmann SF, McKinlay SM, Pitt B, Pfeffer MA, Solomon SD. Cardiac structure and function and prognosis in heart failure with preserved ejection fraction: findings from the echocardiographic study of the Treatment of Preserved Cardiac Function Heart Failure with an Aldosterone Antagonist (TOPCAT) Trial. Circ Heart Fail. 2014 Sep;7(5):740-51. doi: 10.1161/CIRCHEARTFAILURE.114.001583
12. Zile MR, Gottdiener JS, Hetzel SJ, McMurray JJ, Komajda M, McKelvie R, Baicu CF, Massie BM, Carson PE; I-PRESERVE Investigator Prevalence and significance of alterations in cardiac structure and function in patients with heart failure and a preserved ejection fraction. Circulation. 2011 ;124(23):2491-501. doi: 10.1161/CIRCULATIONAHA.110.011031
13. Campbell RT, Jhund PS, Castagno D, Hawkins NM, Petrie MC, McMurray JJ. What have we learned about patients with heart failure and preserved ejection fraction from DIG-PEF, CHARM-preserved, and I-PRESERVE? J Am Coll Cardiol. 2012 Dec 11;60(23):2349-56. doi: 10.1016/j.jacc.2012.04.064
14. Iorio A, Senni M, Barbati G, Greene SJ, Poli S, Zambon E, Di Nora C, Cioffi G, Tarantini L, Gavazzi A, Sinagra G, Di Lenarda A. Prevalence and prognostic impact of non-cardiac co-morbidities in heart failure outpatients with preserved and reduced ejection fraction: a community-based study. Eur J Heart Fail. 2018 Sep;20(9):1257-1266. doi: 10.1002/ejhf.1202. Epub 2018 Jun 19. PMID: 29917301.
15. Cohen JB, Schrauben SJ, Zhao L, Basso MD, Cvijic ME, Li Z, Yarde M, Wang Z, Bhattacharya PT, Chirinos DA, Prenner S, Zamani P, Seiffert DA, Car BD, Gordon DA, Margulies K, Cappola T, Chirinos JA. Clinical Phenogroups in Heart Failure With Preserved Ejection Fraction: Detailed Phenotypes, Prognosis, and Response to Spironolactone. JACC Heart Fail. 2020 Mar;8(3):172-184. doi: 10.1016/j.jchf.2019.09.009. Epub 2020 Jan 8. PMID: 31926856; PMCID: PMC7058514.
16. Shah SJ, Katz DH, Selvaraj S, Burke MA, Yancy CW, Gheorghiade M, Bonow RO, Huang CC, Deo RC. Phenomapping for novel classification of heart failure with preserved ejection fraction. Circulation. 2015 Jan 20;131(3):269-79. doi: 10.1161/CIRCULATIONAHA.114.010637. Epub 2014 Nov 14. PMID: 25398313; PMCID: PMC4302027.
17. Uijl A, Savarese G, Vaartjes I, Dahlström U, Brugts JJ, Linssen GCM, van Empel V, Brunner-La Rocca HP, Asselbergs FW, Lund LH, Hoes AW, Koudstaal S. Identification of distinct phenotypic clusters in heart failure with preserved ejection fraction. Eur J Heart Fail. 2021 Jun;23(6):973-982. doi: 10.1002/ejhf.2169. Epub 2021 May 1. PMID: 33779119; PMCID: PMC8359985.

18. Tromp J, Westenbrink BD, Ouwerkerk W, van Veldhuisen DJ, Samani NJ, Ponikowski P, Metra M, Anker SD, Cleland JG, Dickstein K, Filippatos G, van der Harst P, Lang CC, Ng LL, Zannad F, Zwinderman AH, Hillege HL, van der Meer P, Voors AA Identifying Pathophysiological Mechanisms in Heart Failure With Reduced versus Preserved Ejection Fraction. J Am Coll Cardiol. 2018 Sep 4;72(10):1081-1090. doi: 10.1016/j.jacc.2018.06.050
19. Meijers WC, Bayes-Genis A, Mebazaa A, Bauersachs J, Cleland JGF, Coats AJS, et al. Circulating heart failure biomarkers beyond natriuretic peptides: updated review from the biomarker study Group of the Heart Failure Association (HFA), European Society of Cardiology (ESC). *Eur J Heart Fail*. 2021; **23**: 1610– 32
20. Chow SL, Maisel AS, Anand I, Bozkurt B, de Boer RA, Felker GM,et al. . Role of Biomarkers for the Prevention, Assessment, and Management of Heart Failure: A Scientific Statement From the American Heart Association. Circulation. 2017 May 30;135(22):e1054-e1091. doi: 10.1161/CIR.0000000000000490. Epub 2017 Apr 26. Erratum in: Circulation. 2017 Nov 7;136(19):e345. PMID: 28446515.
21. Kociol RD, Pang PS, Gheorghiade M, Fonarow GC, O'Connor CM, Felker GM. Troponin elevation in heart failure prevalence, mechanisms, and clinical implications. J Am Coll Cardiol. 2010 Sep 28;56(14):1071-8. doi: 10.1016/j.jacc.2010.06.016. PMID: 20863950.
22. Greenberg B. Heart failure preserved ejection fraction with coronary artery disease: time for a new classification? J Am Coll Cardiol. 2014 Jul 1;63(25 Pt A):2828-30. doi: 10.1016/j.jacc.2014.03.033. Epub 2014 Apr 23. PMID: 24768875.
23. Obokata M, Reddy YNV, Melenovsky V, Kane GC, Olson TP, Jarolim P, Borlaug BA. Myocardial Injury and Cardiac Reserve in Patients With Heart Failure and Preserved Ejection Fraction. J Am Coll Cardiol. 2018 Jul 3;72(1):29-40. doi: 10.1016/j.jacc.2018.04.039. PMID: 29957229; PMCID: PMC6034112.
24. Gohar A, Chong JPC, Liew OW, den Ruijter H, de Kleijn DPV, Sim D, Yeo DPS, Ong HY, Jaufeerally F, Leong GKT, Ling LH, Lam CSP, Richards AM. The prognostic value of highly sensitive cardiac troponin assays for adverse events in men and women with stable heart failure and a preserved vs. reduced ejection fraction. Eur J Heart Fail. 2017 Dec;19(12):1638-1647. doi: 10.1002/ejhf.911. Epub 2017 Aug 28. PMID: 28849609.
25. Myhre PL, O'Meara E, Claggett BL, de Denus S, Jarolim P, Anand IS, Beldhuis IE, Fleg JL, Lewis E, Pitt B, Rouleau JL, Solomon SD, Pfeffer MA, Desai AS. Cardiac Troponin I and Risk of Cardiac Events in Patients With Heart Failure and Preserved Ejection Fraction. Circ Heart Fail. 2018 Nov;11(11):e005312. doi: 10.1161/CIRCHEARTFAILURE.118.005312. PMID: 30571192
26. Gori M, Senni M, Claggett B, Liu J, Maggioni AP, Zile M, Prescott MF, Van Veldhuisen DJ,

Zannad F, Pieske B, Lam CSP, Rouleau J, Jhund P, Packer M, Pfeffer MA, Lefkowitz M, Shi V, McMurray JJV, Solomon SD. Integrating High-Sensitivity Troponin T and Sacubitril/Valsartan Treatment in HFpEF: The PARAGON-HF Trial. JACC Heart Fail. 2021 Sep;9(9):627-635. doi: 10.1016/j.jchf.2021.04.009. Epub 2021 Jul 7. PMID: 34246603.

27. Meijers WC, Bayes-Genis A, Mebazaa A, Bauersachs J, Cleland JGF, Coats AJS, Januzzi JL, Maisel AS, McDonald K, Mueller T, Richards AM, Seferovic P, Mueller C, de Boer RA. Circulating heart failure biomarkers beyond natriuretic peptides: review from the Biomarker Study Group of the Heart Failure Association (HFA), European Society of Cardiology (ESC). Eur J Heart Fail. 2021 Oct;23(10):1610-1632. doi: 10.1002/ejhf.2346. Epub 2021 Oct 10. PMID: 34498368; PMCID: PMC9292239.
28. Maisel A, Mueller C, Nowak R, Peacock WF, Landsberg JW, Ponikowski P, Mockel M, Hogan C, Wu AH, Richards M, Clopton P, Filippatos GS, Di Somma S, Anand I, Ng L, Daniels LB, Neath SX, Christenson R, Potocki M, McCord J, Terracciano G, Kremastinos D, Hartmann O, von Haehling S, Bergmann A, Morgenthaler NG, Anker SD. Mid-region pro-hormone markers for diagnosis and prognosis in acute dyspnea: results from the BACH (Biomarkers in Acute Heart Failure) trial. J Am Coll Cardiol. 2010 May 11;55(19):2062-76. doi: 10.1016/j.jacc.2010.02.025. PMID: 20447528.
29. Goetze JP, Bruneau BG, Ramos HR, Ogawa T, de Bold MK, de Bold AJ. Cardiac natriuretic peptides. Nat Rev Cardiol. 2020 Nov;17(11):698-717. doi: 10.1038/s41569-020-0381-0. Epub 2020 May 22. PMID: 32444692.
30. Tromp J, Khan MA, Klip IT, Meyer S, de Boer RA, Jaarsma T, Hillege H, van Veldhuisen DJ, van der Meer P, Voors AA. Biomarker Profiles in Heart Failure Patients With Preserved and Reduced Ejection Fraction. J Am Heart Assoc. 2017 Mar 30;6(4):e003989. doi: 10.1161/JAHA.116.003989. PMID: 28360225; PMCID: PMC5532986.
31. Sakane K, Kanzaki Y, Tsuda K, Maeda D, Sohmiya K, Hoshiga M. Disproportionately low BNP levels in patients of acute heart failure with preserved vs. reduced ejection fraction. Int J Cardiol. 2021 Mar 15;327:105-110. doi: 10.1016/j.ijcard.2020.11.066. Epub 2020 Dec 3. PMID: 33279592.
32. Remmelzwaal S, van Ballegooijen AJ, Schoonmade LJ, Dal Canto E, Handoko ML, Henkens MTHM, van Empel V, Heymans SRB, Beulens JWJ. Natriuretic peptides for the detection of diastolic dysfunction and heart failure with preserved ejection fraction-a systematic review and meta-analysis. BMC Med. 2020 Oct 30;18(1):290. doi: 10.1186/s12916-020-01764-x. PMID: 33121502; PMCID: PMC7599104.
33. Lopuszynski JB, Downing AJ, Finley CM, Zahid M. Prognosticators of All-Cause Mortality in

Patients With Heart Failure With Preserved Ejection Fraction. Am J Cardiol. 2021 Nov 1;158:66-73. doi: 10.1016/j.amjcard.2021.07.044. Epub 2021 Aug 29. PMID: 34465456; PMCID: PMC8497433.

34. Kociol RD, Horton JR, Fonarow GC, Reyes EM, Shaw LK, O'Connor CM, Felker GM, Hernandez AF. Admission, discharge, or change in B-type natriuretic peptide and long-term outcomes: data from Organized Program to Initiate Lifesaving Treatment in Hospitalized Patients with Heart Failure (OPTIMIZE-HF) linked to Medicare claims. Circ Heart Fail. 2011 Sep;4(5):628-36. doi: 10.1161/CIRCHEARTFAILURE.111.962290. Epub 2011 Jul 8. PMID: 21743005; PMCID: PMC3465672.
35. Nishikimi T, Nakagawa Y. Adrenomedullin as a Biomarker of Heart Failure. Heart Fail Clin. 2018 Jan;14(1):49-55. doi: 10.1016/j.hfc.2017.08.006. Epub 2017 Oct 7. PMID: 29153200.
36. Voors AA, Kremer D, Geven C, Ter Maaten JM, Struck J, Bergmann A, Pickkers P, Metra M, Mebazaa A, Düngen HD, Butler J. Adrenomedullin in heart failure: pathophysiology and therapeutic application. Eur J Heart Fail. 2019 Feb;21(2):163-171. doi: 10.1002/ejhf.1366. Epub 2018 Dec 28. PMID: 30592365; PMCID: PMC6607488.
37. Kremer D, Ter Maaten JM, Voors AA. Bio-adrenomedullin as a potential quick, reliable, and objective marker of congestion in heart failure. Eur J Heart Fail. 2018 Sep;20(9):1363-1365. doi: 10.1002/ejhf.1245. Epub 2018 Jun 22. PMID: 29932477.
38. Ter Maaten JM, Kremer D, Demissei BG, Struck J, Bergmann A, Anker SD, Ng LL, Dickstein K, Metra M, Samani NJ, Romaine SPR, Cleland J, Girerd N, Lang CC, van Veldhuisen DJ, Voors AA. Bio-adrenomedullin as a marker of congestion in patients with new-onset and worsening heart failure. Eur J Heart Fail. 2019 Jun;21(6):732-743. doi: 10.1002/ejhf.1437. Epub 2019 Mar 6. PMID: 30843353.
39. Ter Maaten JM, Kremer D, Demissei BG, Struck J, Bergmann A, Anker SD, Ng LL, Dickstein K, Metra M, Samani NJ, Romaine SPR, Cleland J, Girerd N, Lang CC, van Veldhuisen DJ, Voors AA. Bio-adrenomedullin as a marker of congestion in patients with new-onset and worsening heart failure. Eur J Heart Fail. 2019 Jun;21(6):732-743. doi: 10.1002/ejhf.1437. Epub 2019 Mar 6. PMID: 30843353.
40. Pandhi P, Ter Maaten JM, Emmens JE, Struck J, Bergmann A, Cleland JG, Givertz MM, Metra M, O'Connor CM, Teerlink JR, Ponikowski P, Cotter G, Davison B, van Veldhuisen DJ, Voors AA. Clinical value of pre-discharge bio-adrenomedullin as a marker of residual congestion and high risk of heart failure hospital readmission. Eur J Heart Fail. 2020 Apr;22(4):683-691. doi: 10.1002/ejhf.1693. Epub 2019 Dec 3. PMID: 31797505.
41. Sharma UC, Pokharel S, van Brakel TJ, van Berlo JH, Cleutjens JP, Schroen B, André S, Crijns

HJ, Gabius HJ, Maessen J, Pinto YM. Galectin-3 marks activated macrophages in failure-prone hypertrophied hearts and contributes to cardiac dysfunction. Circulation. 2004 Nov 9;110(19):3121-8. doi: 10.1161/01.CIR.0000147181.65298.4D. Epub 2004 Nov 1. PMID: 15520318.

42. de Boer RA, Voors AA, Muntendam P, van Gilst WH, van Veldhuisen DJ. Galectin-3: a novel mediator of heart failure development and progression. Eur J Heart Fail. 2009 Sep;11(9):811-7. doi: 10.1093/eurjhf/hfp097. Epub 2009 Jul 31. PMID: 19648160.
43. Sharma UC, Pokharel S, van Brakel TJ, van Berlo JH, Cleutjens JP, Schroen B, André S, Crijns HJ, Gabius HJ, Maessen J, Pinto YM. Galectin-3 marks activated macrophages in failure-prone hypertrophied hearts and contributes to cardiac dysfunction. Circulation. 2004 Nov 9;110(19):3121-8. doi: 10.1161/01.CIR.0000147181.65298.4D. Epub 2004 Nov 1. PMID: 15520318.
44. de Boer RA, Lok DJ, Jaarsma T, van der Meer P, Voors AA, Hillege HL, van Veldhuisen DJ. Predictive value of plasma galectin-3 levels in heart failure with reduced and preserved ejection fraction. Ann Med. 2011 Feb;43(1):60-8. doi: 10.3109/07853890.2010.538080. Epub 2010 Dec 28. PMID: 21189092; PMCID: PMC3028573.
45. Edelmann F, Holzendorf V, Wachter R, Nolte K, Schmidt AG, Kraigher-Krainer E, Duvinage A, Unkelbach I, Düngen HD, Tschöpe C, Herrmann-Lingen C, Halle M, Hasenfuss G, Gelbrich G, Stough WG, Pieske BM. Galectin-3 in patients with heart failure with preserved ejection fraction: results from the Aldo-DHF trial. Eur J Heart Fail. 2015 Feb;17(2):214-23. doi: 10.1002/ejhf.203. Epub 2014 Nov 24. PMID: 25418979.
46. Ghorbani A, Bhambhani V, Christenson RH, Meijers WC, de Boer RA, Levy D, Larson MG, Ho JE. Longitudinal Change in Galectin-3 and Incident Cardiovascular Outcomes. J Am Coll Cardiol. 2018 Dec 25;72(25):3246-3254. doi: 10.1016/j.jacc.2018.09.076. PMID: 30573026; PMCID: PMC6516745.
47. Weinberg EO, Shimpo M, De Keulenaer GW, MacGillivray C, Tominaga S, Solomon SD, Rouleau JL, Lee RT. Expression and regulation of ST2, an interleukin-1 receptor family member, in cardiomyocytes and myocardial infarction. Circulation. 2002 Dec 3;106(23):2961-6. doi: 10.1161/01.cir.0000038705.69871.d9. PMID: 12460879; PMCID: PMC1460012.
48. Ky B, French B, McCloskey K, Rame JE, McIntosh E, Shahi P, Dries DL, Tang WH, Wu AH, Fang JC, Boxer R, Sweitzer NK, Levy WC, Goldberg LR, Jessup M, Cappola TP. High-sensitivity ST2 for prediction of adverse outcomes in chronic heart failure. Circ Heart Fail. 2011 Mar;4(2):180-7. doi: 10.1161/CIRCHEARTFAILURE.110.958223. Epub 2010 Dec 22. PMID: 21178018; PMCID: PMC3163169.

49. Wang YC, Yu CC, Chiu FC, Tsai CT, Lai LP, Hwang JJ, Lin JL. Soluble ST2 as a biomarker for detecting stable heart failure with a normal ejection fraction in hypertensive patients. J Card Fail. 2013 Mar;19(3):163-8. doi: 10.1016/j.cardfail.2013.01.010. PMID: 23482076.
50. Aimo A, Vergaro G, Ripoli A, Bayes-Genis A, Pascual Figal DA, de Boer RA, Lassus J, Mebazaa A, Gayat E, Breidthardt T, Sabti Z, Mueller C, Brunner-La Rocca HP, Tang WH, Grodin JL, Zhang Y, Bettencourt P, Maisel AS, Passino C, Januzzi JL, Emdin M. Meta-Analysis of Soluble Suppression of Tumorigenicity-2 and Prognosis in Acute Heart Failure. JACC Heart Fail. 2017 Apr;5(4):287-296. doi: 10.1016/j.jchf.2016.12.016. Epub 2017 Feb 8. PMID: 28189578.
51. Krebber MM, van Dijk CGM, Vernooij RWM, Brandt MM, Emter CA, Rau CD, Fledderus JO, Duncker DJ, Verhaar MC, Cheng C, Joles JA. Matrix Metalloproteinases and Tissue Inhibitors of Metalloproteinases in Extracellular Matrix Remodeling during Left Ventricular Diastolic Dysfunction and Heart Failure with Preserved Ejection Fraction: A Systematic Review and Meta-Analysis. Int J Mol Sci. 2020 Sep 14;21(18):6742. doi: 10.3390/ijms21186742. PMID: 32937927; PMCID: PMC7555240.
52. Ferreira JM, Ferreira SM, Ferreira MJ, Falcão-Pires I. Circulating Biomarkers of Collagen Metabolism and Prognosis of Heart Failure with Reduced or Mid-Range Ejection Fraction. Curr Pharm Des. 2017;23(22):3217-3223. doi: 10.2174/1381612823666170317124125. PMID: 28317477.
53. Cunningham JW, Claggett BL, O'Meara E, Prescott MF, Pfeffer MA, Shah SJ, Redfield MM, Zannad F, Chiang LM, Rizkala AR, Shi VC, Lefkowitz MP, Rouleau J, McMurray JJV, Solomon SD, Zile MR. Effect of Sacubitril/Valsartan on Biomarkers of Extracellular Matrix Regulation in Patients With HFpEF. J Am Coll Cardiol. 2020 Aug 4;76(5):503-514. doi: 10.1016/j.jacc.2020.05.072. PMID: 32731928.
54. Duprez DA, Gross MD, Kizer JR, Ix JH, Hundley WG, Jacobs DR Jr. Predictive Value of Collagen Biomarkers for Heart Failure With and Without Preserved Ejection Fraction: MESA (Multi-Ethnic Study of Atherosclerosis). J Am Heart Assoc. 2018 Feb 23;7(5):e007885. doi: 10.1161/JAHA.117.007885. PMID: 29475876; PMCID: PMC5866330.
55. Wang TJ, Larson MG, Benjamin EJ, Siwik DA, Safa R, Guo CY, Corey D, Sundstrom J, Sawyer DB, Colucci WS, Vasan RS. Clinical and echocardiographic correlates of plasma procollagen type III amino-terminal peptide levels in the community. Am Heart J. 2007 Aug;154(2):291-7. doi: 10.1016/j.ahj.2007.04.006. PMID: 17643579; PMCID: PMC3170820.
56. Agarwal I, Glazer NL, Barasch E, Djousse L, Gottdiener JS, Ix JH, Kizer JR, Rimm EB, Siscovick DS, King GL, Mukamal KJ. Associations between metabolic dysregulation and circulating biomarkers of fibrosis: the Cardiovascular Health Study. Metabolism. 2015 Oct;64(10):1316-23.

doi: 10.1016/j.metabol.2015.07.013. Epub 2015 Jul 21. PMID: 26282733; PMCID: PMC4939831.

57. Pasceri V, Willerson JT, Yeh ET. Direct proinflammatory effect of C-reactive protein on human endothelial cells. Circulation. 2000 Oct 31;102(18):2165-8. doi: 10.1161/01.cir.102.18.2165. PMID: 11056086.
58. Koller L, Kleber M, Goliasch G, Sulzgruber P, Scharnagl H, Silbernagel G, Grammer T, Delgado G, Tomaschitz A, Pilz S, März W, Niessner A. C-reactive protein predicts mortality in patients referred for coronary angiography and symptoms of heart failure with preserved ejection fraction. Eur J Heart Fail. 2014 Jul;16(7):758-66. doi: 10.1002/ejhf.104. Epub 2014 May 7. PMID: 24806206.
59. Abernethy A, Raza S, Sun JL, Anstrom KJ, Tracy R, Steiner J, VanBuren P, LeWinter MM. Pro-Inflammatory Biomarkers in Stable Versus Acutely Decompensated Heart Failure With Preserved Ejection Fraction. J Am Heart Assoc. 2018 Apr 12;7(8):e007385. doi: 10.1161/JAHA.117.007385. PMID: 29650706; PMCID: PMC6015440.
60. DuBrock HM, AbouEzzeddine OF, Redfield MM. High-sensitivity C-reactive protein in heart failure with preserved ejection fraction. PLoS One. 2018 Aug 16;13(8):e0201836. doi: 10.1371/journal.pone.0201836. PMID: 30114262; PMCID: PMC6095520.
61. Kempf T, von Haehling S, Peter T, Allhoff T, Cicoira M, Doehner W, Ponikowski P, Filippatos GS, Rozentryt P, Drexler H, Anker SD, Wollert KC. Prognostic utility of growth differentiation factor-15 in patients with chronic heart failure. J Am Coll Cardiol. 2007 Sep 11;50(11):1054-60. doi: 10.1016/j.jacc.2007.04.091. Epub 2007 Aug 24. PMID: 17825714
62. Chan MM, Santhanakrishnan R, Chong JP et al. Growth differentiation factor 15 in heart failure with preserved vs. reduced ejection fraction. *Eur J Heart Fail.* 2016;18:81–8. doi: 10.1002/ejhf.431
63. Kato ET, Morrow DA, Guo J, Berg DD, Blazing MA, Bohula EA, Bonaca MP, Cannon CP, de Lemos JA, Giugliano RP, Jarolim P, Kempf T, Kristin Newby L, O'Donoghue ML, Pfeffer MA, Rifai N, Wiviott SD, Wollert KC, Braunwald E, Sabatine MS. Growth differentiation factor 15 and cardiovascular risk: individual patient meta-analysis. Eur Heart J. 2022 Oct 28:ehac577. doi: 10.1093/eurheartj/ehac577. Epub ahead of print. PMID: 36303404
64. Abbate A, Toldo S, Marchetti C et al. Interleukin-1 and the inflammasome as therapeutic targets in cardiovascular disease. *Circ Res.* 2020;126:1260–80. doi: 10.1161/CIRCRESAHA.120.315937
65. Chia YC. Interleukin 6 and development of heart failure with preserved ejection fraction in the general population. J Am Heart Assoc. 2021;10(11):e018549

66. Everett BM, Cornel JH, Lainscak M et al. Anti-inflammatory therapy with canakinumab for the prevention of hospitalization for heart failure. *Circulation.* 2019;139:1289–99. doi: 10.1161/CIRCULATIONAHA.118.038010.
67. Hage C, Michaëlsson E, Linde C, Donal E, Daubert JC, Gan LM, Lund LH. Inflammatory Biomarkers Predict Heart Failure Severity and Prognosis in Patients With Heart Failure With Preserved Ejection Fraction: A Holistic Proteomic Approach. Circ Cardiovasc Genet. 2017 Feb;10(1):e001633. doi: 10.1161/CIRCGENETICS.116.001633. PMID: 28100627.
68. Marti CN, Khan H, Mann DL, Georgiopoulou VV, Bibbins-Domingo K, Harris T, Koster A, Newman A, Kritchevsky SB, Kalogeropoulos AP, Butler J; Health ABC Study. Soluble tumor necrosis factor receptors and heart failure risk in older adults: Health, Aging, and Body Composition (Health ABC) Study. Circ Heart Fail. 2014 Jan;7(1):5-11. doi: 10.1161/CIRCHEARTFAILURE.113.000344. Epub 2013 Dec 9. PMID: 24323631; PMCID: PMC3990649.
69. Mann DL, McMurray JJ, Packer M, Swedberg K, Borer JS, Colucci WS, Djian J, Drexler H, Feldman A, Kober L, Krum H, Liu P, Nieminen M, Tavazzi L, van Veldhuisen DJ, Waldenstrom A, Warren M, Westheim A, Zannad F, Fleming T. Targeted anticytokine therapy in patients with chronic heart failure: results of the Randomized Etanercept Worldwide Evaluation (RENEWAL). Circulation. 2004 Apr 6;109(13):1594-602. doi: 10.1161/01.CIR.0000124490.27666.B2. Epub 2004 Mar 15. PMID: 15023878.
70. Shantsila E, Wrigley BJ, Blann AD, Gill PS, Lip GY. A contemporary view on endothelial function in heart failure. Eur J Heart Fail. 2012 Aug;14(8):873-81. doi: 10.1093/eurjhf/hfs066. Epub 2012 Jun 7. PMID: 22677484.
71. Shah SJ, Lam CSP, Svedlund S, Saraste A, Hage C, Tan RS, Beussink-Nelson L, Ljung Faxén U, Fermer ML, Broberg MA, Gan LM, Lund LH. Prevalence and correlates of coronary microvascular dysfunction in heart failure with preserved ejection fraction: PROMIS-HFpEF. Eur Heart J. 2018 Oct 1;39(37):3439-3450. doi: 10.1093/eurheartj/ehy531. Erratum in: Eur Heart J. 2019 Feb 7;40(6):541. PMID: 30165580; PMCID: PMC6927847.
72. Rush CJ, Berry C, Oldroyd KG, Rocchiccioli JP, Lindsay MM, Touyz RM, Murphy CL, Ford TJ, Sidik N, McEntegart MB, Lang NN, Jhund PS, Campbell RT, McMurray JJV, Petrie MC. Prevalence of Coronary Artery Disease and Coronary Microvascular Dysfunction in Patients With Heart Failure With Preserved Ejection Fraction. JAMA Cardiol. 2021 Oct 1;6(10):1130-1143. doi: 10.1001/jamacardio.2021.1825. PMID: 34160566; PMCID: PMC8223134.
73. Emdin M, Aimo A, Castiglione V, Vergaro G, Georgiopoulos G, Saccaro LF, Lombardi CM, Passino C, Cerbai E, Metra M, Senni M. Targeting Cyclic Guanosine Monophosphate to Treat

Heart Failure: JACC Review Topic of the Week. J Am Coll Cardiol. 2020 Oct 13;76(15):1795-1807. doi: 10.1016/j.jacc.2020.08.031. PMID: 33032741.

74. Schiffrin EL. Role of endothelin-1 in hypertension and vascular disease. Am J Hypertens. 2001 Jun;14(6 Pt 2):83S-89S. doi: 10.1016/s0895-7061(01)02074-x. PMID: 11411770.
75. Chowdhury MA, Moukarbel GV, Gupta R, Frank SM, Anderson AM, Liu LC, Khouri SJ. Endothelin 1 Is Associated with Heart Failure Hospitalization and Long-Term Mortality in Patients with Heart Failure with Preserved Ejection Fraction and Pulmonary Hypertension. Cardiology. 2019;143(3-4):124-133. doi: 10.1159/000501100. Epub 2019 Sep 12. PMID: 31514181.
76. Bevan GH, Jenkins T, Josephson R, Rajagopalan S, Al-Kindi SG. Endothelin-1 and peak oxygen consumption in patients with heart failure with preserved ejection fraction. Heart Lung. 2021 May-Jun;50(3):442-446. doi: 10.1016/j.hrtlng.2021.02.004. Epub 2021 Feb 23. PMID: 33636416.
77. Winter MP, Kleber ME, Koller L, Sulzgruber P, Scharnagl H, Delgado G, Goliasch G, März W, Niessner A. Prognostic significance of tPA/PAI-1 complex in patients with heart failure and preserved ejection fraction. Thromb Haemost. 2017 Feb 28;117(3):471-478. doi: 10.1160/TH16-08-0600. Epub 2016 Dec 15. PMID: 27975104.
78. Jug B, Vene N, Salobir BG, Sebestjen M, Sabovic M, Keber I. Procoagulant state in heart failure with preserved left ventricular ejection fraction. Int Heart J. 2009 Sep;50(5):591-600. doi: 10.1536/ihj.50.591. PMID: 19809208.
79. Hage C, Bjerre M, Frystyk J, Gu HF, Brismar K, Donal E, Daubert JC, Linde C, Lund LH. Comparison of Prognostic Usefulness of Serum Insulin-Like Growth Factor-Binding Protein 7 in Patients With Heart Failure and Preserved Versus Reduced Left Ventricular Ejection Fraction. Am J Cardiol. 2018 Jun 15;121(12):1558-1566. doi: 10.1016/j.amjcard.2018.02.041. Epub 2018 Mar 14. PMID: 29622288.
80. Sabbah MS, Fayyaz AU, de Denus S, Felker GM, Borlaug BA, Dasari S, Carter RE, Redfield MM. Obese-Inflammatory Phenotypes in Heart Failure With Preserved Ejection Fraction. Circ Heart Fail. 2020 Aug;13(8):e006414. doi: 10.1161/CIRCHEARTFAILURE.119.006414. Epub 2020 Jul 29. PMID: 32809874; PMCID: PMC7439286.
81. Gandhi PU, Chow SL, Rector TS, Krum H, Gaggin HK, McMurray JJ, Zile MR, Komajda M, McKelvie RS, Carson PE, Januzzi JL Jr, Anand IS. Prognostic Value of Insulin-Like Growth Factor-Binding Protein 7 in Patients with Heart Failure and Preserved Ejection Fraction. J Card Fail. 2017 Jan;23(1):20-28. doi: 10.1016/j.cardfail.2016.06.006. Epub 2016 Jun 16. PMID: 27317843.

82. Gandhi PU, Gaggin HK, Redfield MM, Chen HH, Stevens SR, Anstrom KJ, Semigran MJ, Liu P, Januzzi JL Jr. Insulin-Like Growth Factor-Binding Protein-7 as a Biomarker of Diastolic Dysfunction and Functional Capacity in Heart Failure With Preserved Ejection Fraction: Results From the RELAX Trial. JACC Heart Fail. 2016 Nov;4(11):860-869. doi: 10.1016/j.jchf.2016.08.002. Epub 2016 Oct 12. PMID: 27744089; PMCID: PMC5500914.
83. Bayes-Genis A, Cediel G, Domingo M, Codina P, Santiago E, Lupón J. Biomarkers in Heart Failure with Preserved Ejection Fraction. Card Fail Rev. 2022 Jun 23;8:e20. doi: 10.15420/cfr.2021.37. PMID: 35815256; PMCID: PMC9253965
84. Zhou X, He L, Zuo S, Zhang Y, Wan D, Long C, Huang P, Wu X, Wu C, Liu G, Yin Y. Serine prevented high-fat diet-induced oxidative stress by activating AMPK and epigenetically modulating the expression of glutathione synthesis-related genes. Biochim Biophys Acta Mol Basis Dis. 2018 Feb;1864(2):488-498. doi: 10.1016/j.bbadis.2017.11.009. Epub 2017 Nov 17. PMID: 29158183.
85. Chen Siyuan, Xia Yaoyao, He Fang, et al. Serine Supports IL-1β Production in Macrophages Through mTOR Signaling. Front. Immunol., 27 August 2020. Sec. Nutritional Immunology https://doi.org/10.3389/fimmu.2020.01866
86. Fu Z, Akula S, Thorpe M, Hellman L. Potent and Broad but not Unselective Cleavage of Cytokines and Chemokines by Human Neutrophil Elastase and Proteinase 3. International Journal of Molecular Sciences. 2020; 21(2):651. https://doi.org/10.3390/ijms21020651
87. Sibille Lejeune, Nassiba Menghoum, Julie Thompson, Isabelle Robillard, Julie Hussin, Luc Bertrand, Sandrine Horman, Christine Des Rosiers, Christophe Beauloye, Anne-Catherine Pouleur. Plasma metabolomics identify hydroxyproline as a potential player in the pathophysiogy of HFpEF, Archives of Cardiovascular Diseases Supplements, Volume 14, Issue 2, 2022, Page 193, ISSN 1878-6480, https://doi.org/10.1016/j.acvdsp.2022.04.083.
88. Chen F, Lucas R, Fulton D. The subcellular compartmentalization of arginine metabolizing enzymes and their role in endothelial dysfunction. Front Immunol. 2013 Jul 9;4:184. doi: 10.3389/fimmu.2013.00184. PMID: 23847624; PMCID: PMC3705211.
89. Gambardella J, Khondkar W, Morelli MB, Wang X, Santulli G, Trimarco V. Arginine and Endothelial Function. Biomedicines. 2020 Aug 6;8(8):277. doi: 10.3390/biomedicines8080277. PMID: 32781796; PMCID: PMC7460461.
90. Tang WH, Tong W, Shrestha K, Wang Z, Levison BS, Delfraino B, Hu B, Troughton RW, Klein AL, Hazen SL. Differential effects of arginine methylation on diastolic dysfunction and disease progression in patients with chronic systolic heart failure. Eur Heart J. 2008 Oct;29(20):2506-13. doi: 10.1093/eurheartj/ehn360. Epub 2008 Aug 6. PMID: 18687662; PMCID: PMC2567021.

91. Lugrin, Jérôme, Rosenblatt-Velin, Nathalie, Parapanov, Roumen and Liaudet, Lucas. "The role of oxidative stress during inflammatory processes" Biological Chemistry, vol. 395, no. 2, 2014, pp. 203-230. https://doi.org/10.1515/hsz-2013-0241
92. Zhulan Cai , Cencen Wu , Yuan Xu , Jiageng Cai , Menglin Zhao , Lingyun Zu. The NO-cGMP-PKG Axis in HFpEF: From Pathological Mechanisms to Potential Therapies. Aging and disease. 2022 https://doi.org/10.14336/AD.2022.0523
93. Phillips CM, Chen LW, Heude B, Bernard JY, Harvey NC, Duijts L, Mensink-Bout SM, Polanska K, Mancano G, Suderman M, Shivappa N, Hébert JR. Dietary Inflammatory Index and Non-Communicable Disease Risk: A Narrative Review. Nutrients. 2019 Aug 12;11(8):1873. doi: 10.3390/nu11081873. PMID: 31408965; PMCID: PMC6722630.
94. Bak DW, Weerapana E. Cysteine-mediated redox signalling in the mitochondria. Mol Biosyst. 2015 Mar;11(3):678-97. doi: 10.1039/c4mb00571f. Epub 2014 Dec 18. PMID: 25519845.
95. Jay L. Zweier, M.A. Hassan Talukder, The role of oxidants and free radicals in reperfusion injury, Cardiovascular Research, Volume 70, Issue 2, May 2006, Pages 181–190, https://doi.org/10.1016/j.cardiores.2006.02.025
96. Bekfani T, Bekhite M, Neugebauer S, Derlien S, Hamadanchi A, Nisser J, Hilse MS, Haase D, Kretzschmar T, Wu MF, Lichtenauer M, Kiehntopf M, von Haehling S, Schlattmann P, Lehmann G, Franz M, Möbius-Winkler S, Schulze C. Metabolomic Profiling in Patients with Heart Failure and Exercise Intolerance: Kynurenine as a Potential Biomarker. Cells. 2022 May 18;11(10):1674. doi: 10.3390/cells11101674. PMID: 35626711; PMCID: PMC9139290.
97. De Jong KA, Nikolaev VO. Multifaceted remodelling of cAMP microdomains driven by different aetiologies of heart failure. FEBS J. 2021 Dec;288(23):6603-6622. doi: 10.1111/febs.15706. Epub 2021 Jan 23. PMID: 33415835.
98. Zordoky BN, Sung MM, Ezekowitz J, Mandal R, Han B, Bjorndahl TC, Bouatra S, Anderson T, Oudit GY, Wishart DS, Dyck JR; Alberta HEART. Metabolomic fingerprint of heart failure with preserved ejection fraction. PLoS One. 2015 May 26;10(5):e0124844. doi: 10.1371/journal.pone.0124844. PMID: 26010610; PMCID: PMC4444296.
99. Adams SH, Hoppel CL, Lok KH, Zhao L, Wong SW, Minkler PE, Hwang DH, Newman JW, Garvey WT. Plasma acylcarnitine profiles suggest incomplete long-chain fatty acid beta-oxidation and altered tricarboxylic acid cycle activity in type 2 diabetic African-American women. J Nutr. 2009 Jun;139(6):1073-81. doi: 10.3945/jn.108.103754. Epub 2009 Apr 15. PMID: 19369366; PMCID: PMC2714383.
100. Ferro F, Ouillé A, Tran TA, Fontanaud P, Bois P, Babuty D, Labarthe F, Le Guennec JY. Long-chain acylcarnitines regulate the hERG channel. PLoS One. 2012;7(7):e41686. doi:

10.1371/journal.pone.0041686. Epub 2012 Jul 25. PMID: 22848566; PMCID: PMC3404973.

101. Francesca M Trovato, Rabiya Zia, Salvatore Napoli et al. Dysregulation of the Lysophosphatidylcholine/Autotaxin/Lysophosphatidic Acid Axis in Acute-on-Chronic Liver Failure Is Associated With Mortality and Systemic Inflammation by Lysophosphatidic Acid-Dependent Monocyte Activation. Hepatology Aug 2021 ;74(2):907-925. doi: 10.1002/hep.31738. Epub 2021 Jun 15.

102. Longo M, Zatterale F, Naderi J, Parrillo L, Formisano P, Raciti GA, Beguinot F, Miele C. Adipose Tissue Dysfunction as Determinant of Obesity-Associated Metabolic Complications. Int J Mol Sci. 2019 May 13;20(9):2358. doi: 10.3390/ijms20092358. PMID: 31085992; PMCID: PMC6539070.

103. Vegter EL, van der Meer P, de Windt LJ, Pinto YM, Voors AA. MicroRNAs in heart failure: from biomarker to target for therapy. Eur J Heart Fail. 2016 May;18(5):457-68. doi: 10.1002/ejhf.495. Epub 2016 Feb 11. PMID: 26869172.

104. He, X., Du, T., Long, T. et al. Signaling cascades in the failing heart and emerging therapeutic strategies. Sig Transduct Target Ther 7, 134 (2022). https://doi.org/10.1038/s41392-022-00972-6

105. Wong LL, Zou R, Zhou L et al. Combining Circulating MicroRNA and NT-proBNP to Detect and Categorize Heart Failure Subtypes. J Am Coll Cardiol. 2019 Mar 26;73(11):1300-1313. doi: 10.1016/j.jacc.2018.11.060. PMID: 30898206.

106. Feng Chen, Jiefu Yang, Yingying Li, et al. Circulating microRNAs as novel biomarkers for heart failure. Hellenic Journal of Cardiology, Volume 59, Issue 4, July–August 2018, Pages 209-214 https://doi.org/10.1016/j.hjc.2017.10.002.

107. Sucharov CC, Kao DP, Port JD, Karimpour-Fard A, Quaife RA, Minobe W, Nunley K, Lowes BD, Gilbert EM, Bristow MR. Myocardial microRNAs associated with reverse remodeling in human heart failure. JCI Insight. 2017 Jan 26;2(2):e89169. doi: 10.1172/jci.insight.89169. PMID: 28138556; PMCID: PMC5256135.