# Heart Failure with Preserved Ejection Fraction: Microvascular Dysfunction

Subjects: Biology
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Heart failure with preserved ejection fraction (HFpEF) is a condition with increasing incidence, leading to a health care problem of epidemic proportions for which no curative treatments exist. Consequently, an urge exists to better understand the pathophysiology of HFpEF. Accumulating evidence suggests a key pathophysiological role for coronary microvascular dysfunction (MVD), with an underlying mechanism of low-grade pro-inflammatory state caused by systemic comorbidities.

heart failure with preserved ejection fraction | microcirculation | microvascular dysfunction

## 1. Introduction

Heart failure with preserved ejection fraction (HFpEF) is a health care problem of epidemic proportions, currently accounting for roughly 3 million patients in the United States alone [1][2]. Over 50% of all patients with heart failure (HF) suffer from HFpEF, and its incidence is increasing by 1% each year [1]. This rising incidence parallels increasing rates of comorbidities and age. However, HFpEF is more than just a correlate of comorbidities [3]. Rather, HFpEF seems to be driven by them [4]. Moreover, growing evidence suggests HFpEF is a heterogeneous syndrome comprising of different phenotypes [5].

The paucity of effective treatments for HFpEF creates an urge to better understand the pathophysiology of this condition. Important comorbidities associated with HFpEF such as obesity, diabetes mellitus, renal dysfunction and hypertension have been linked with a systemic-low grade pro-inflammatory state [6]. This pro-inflammatory state is associated with increased oxidative stress and reduced nitric oxide (NO) bioavailability in endothelial cells (ECs), marking an EC phenotype shift towards an activated, pro-inflammatory state [7][8]. It has been postulated that, as a consequence, abnormal remodelling occurs (cardiomyocytes hypertrophy and stiffen, and local fibroblasts are activated to produce collagen), ultimately leading to diastolic dysfunction and HF [7].

Both coronary MVD and peripheral MVD, here defined as MVD in vascular beds other than the heart, have previously been reported in HFpEF [9][10][11][12][13][14][15][16]. These findings raise the question of whether either isolated coronary MVD or a more generalized, systemic MVD, consisting of both coronary and peripheral MVD, contribute to developing HFpEF. In spite of that, MVD is not limited to HFpEF. Comorbidities highly prevalent in HFpEF, such as hypertension, diabetes mellitus, and obesity, are also associated with MVD [17]. The reported associations between both HFpEF and its comorbidities with MVD complicate the discussion of whether MVD is a cause of HFpEF or a bystander.

## 2. Defining Microvascular Function and Dysfunction

Adequate interpretation of HFpEF studies that focus on MVD requires knowledge of the microcirculation and its functions from the vessel network and vessel type to a molecular level. Interpretation is complicated by the absence of a gold standard to define and diagnose microvascular dysfunction. Rather, different techniques evaluate different functional and structural aspects of the microcirculation and in different tissues, and a variety of these have been used in clinical research in HFpEF. The techniques have been reviewed elsewhere [18].

Microcirculatory beds are highly dynamic networks that constantly adapt to a variety of systemic and local signals from surrounding tissue and the vascular lumen, acting both acutely and chronically [19]. These signals include humoral, physical, neurogenic, cellular, and metabolic factors [19]. The primary function of the microcirculation is to meet demands for delivery of nutrients including oxygen to local tissue through flow regulation (mainly through regulation of vascular tone), structural adaptation (such as angiogenesis or rarefaction), permeability, haemostasis, immunity, and inflammation [20]. The way a microvessel functions differs per vessel type (arteriole, capillary, venule), organ, and position in the highly heterogeneous vascular tree [21]. These factors all contribute to different responses to local haemodynamics, rheology, and signalling factors/metabolites. **Figure 1** displays a simplified example of the intricate interplay between different cell types in an arteriole and alterations that have been reported in HFpEF patients. The following sections will elaborate on the microvascular alterations found in prior research.

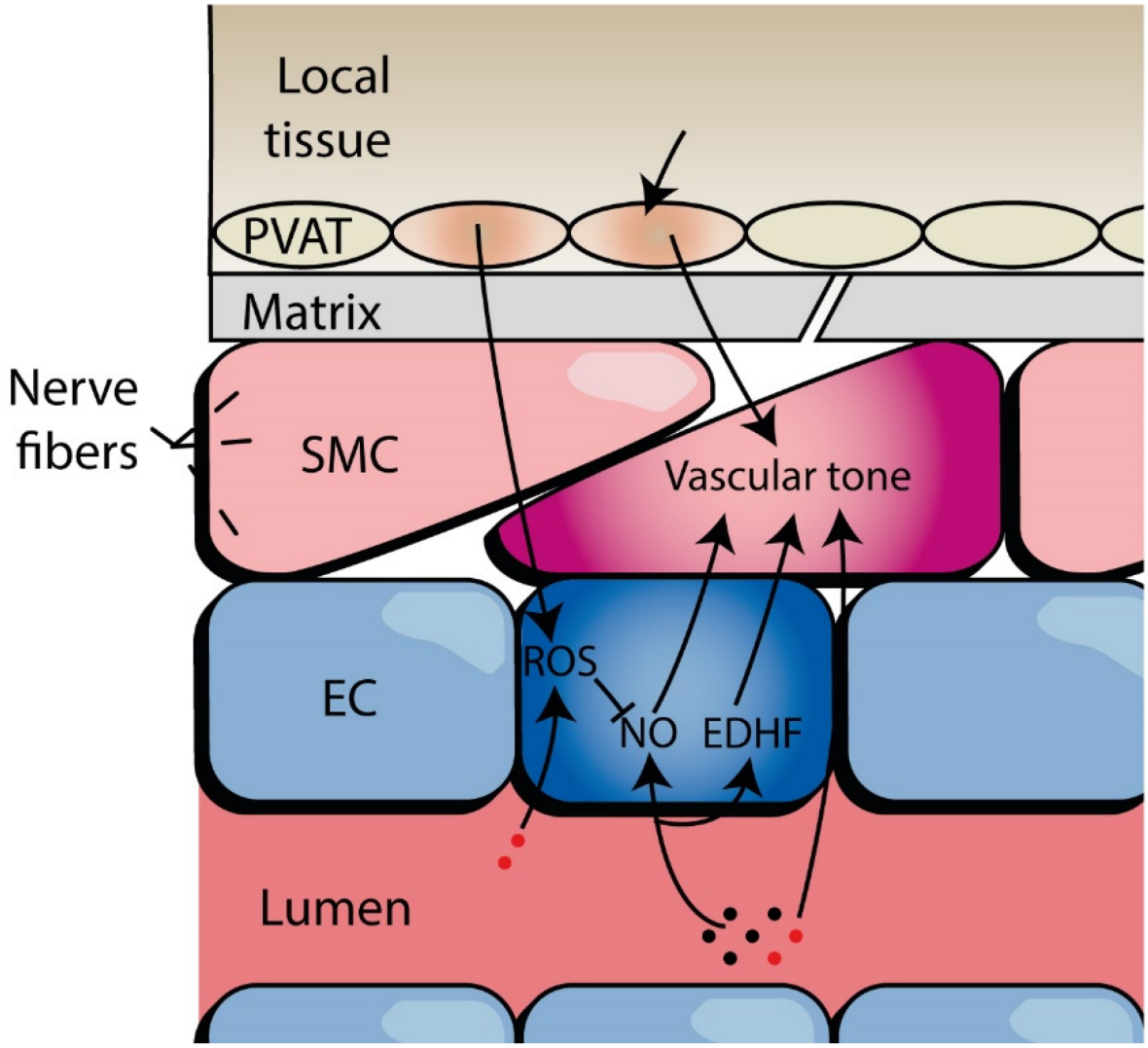


**Figure 1.** Example of intercellular signalling in an arteriole. A simplified example of the complex intercellular signalling of the microcirculation is displayed. This signalling is different per vessel type. It comprises a variety of systemic and local signals from surrounding tissue and the blood, acting in both the short- and long-term, including humoral, physical, neurogenic, cellular, and metabolic factors. Alterations in these signalling pathways and cellular abnormalities have been reported in HFpEF patients, including changes in matrix cell types and stiffness; adipose tissue cell phenotype and adipokine secretion; muscle cell hypertrophy and oxidative stress, and vasodilator response; endothelium-dependent vasodilation; microvascular rarefaction, and microvessel morphology. EC, endothelial cells; EDHF, endothelium-derived hyperpolarizing factors; NO, nitric oxide; PVAT, perivascular adipose tissue; ROS, reactive oxygen species; SMC, smooth muscle cells.

MVD is often used as a broadly defined term encompassing all aspects of abnormal functioning of the microcirculation. MVD is not a synonym for endothelial dysfunction since it is not limited to functional or structural alterations of ECs but can include any (cellular) component of the microcirculation such as smooth muscle cells (SMCs), matrix, or pericytes. Here, researchers define microvascular function as a continuum between normal function and dysfunction, instead of a binary phenomenon, and microvascular dysfunction as a state where the primary microvascular functions are suboptimal and affect the surrounding tissue.

# 3. Evidence of Microvascular Dysfunction in HFpEF

Since Paulus and Tschöpe postulated a central role for MVD in the aetiology of HFpEF, a substantial number of studies have investigated MVD in HFpEF [7]. At first, the focus was on coronary MVD, also referred to as coronary microvascular dysfunction (CMD), but with the understanding that HFpEF is a condition related to systemic comorbidities such as hypertension and diabetes mellitus, the involvement of systemic MVD was proposed [15][16]. These include vasoreactivity, the response of vascular tone to an external stimulus, and capillary rarefaction, a reduction in the capillary density within tissues. Evidence is further grouped according to the location of the microvascular bed. Data are mainly available on different microvascular abnormalities throughout the body as correlates of HFpEF. An overview of the studies on HFpEF performed to date, to the best of their knowledge, is provided in **Table 1** for peripheral MVD studies and in **Table 2** for coronary MVD studies.

**Table 1.** Studies on peripheral microvascular function in HFpEF.

| Study Design | HFpEF Population | Control Population | Method (Measurement) | Stimulus | Microvascular Function Assessed | Outcome (SD/IQR) |
|---|---|---|---|---|---|---|
| | | | Skin-finger | | | |
| Prospective [22] | *n* = 321 | Controls without HF, matched for age, sex, HT, and DM (*n* = 173) | Peripheral arterial tonometry (endoPAT): (RHI) | Ischemia | Hyperaemia | Log RHI: 0.53 ± 0.20 vs. 0.64 ± 0.20, *p* < 0.001 |
| Prospective [10] | *n* = 202 | No controls | endoPAT (RHI) | Ischemia | Hyperaemia | Log RHI: no absolute values reported. Correlation with CFR of R 0.21, *p* = 0.004 |
| Retrospective [23] | *n* = 159 | No controls | endoPAT (RHI) | Ischemia | Hyperaemia | Log RHI: 0.50 ± 0.09. Event free 0.52 ± 0.09 vs. Events 0.46 ± 0.08, *p* < 0.001 |
| Prospective (cross-sectional) [24] | *n* = 62 | Controls matched for age, sex, HT, DM, dyslipidaemia | endoPAT (RHI) | Ischemia | Hyperaemia | RHI: 2.01 [1.64–2.42] vs. 1.70 [1.55–1.88], *p* < 0.001 |

| Study Design | HFpEF Population | Control Population | Method (Measurement) | Stimulus | Microvascular Function Assessed | Outcome (SD/IQR) |
|---|---|---|---|---|---|---|
| | | and CAD (*n* = 64) | | | | |
| Prospective [25] | *n* = 42 | HFrEF (*n* = 46) | endoPAT (RHI) | Ischemia | Hyperaemia | RHI: 1.77 [1.67–2.16] vs. 1.53 [1.42–1.94], *p* = 0.014. |
| Prospective [26] | *n* = 26 | Healthy controls, matched for age and sex (*n* = 26) | endoPAT (RHI) | Ischemia | Hyperaemia | RHI interpretation from boxplots: 1.9 [1.6–2.9] vs. 1.8 [2.0–3.3], *p* = 0.036. No effect of exercise |
| Prospective [27] | *n* = 21 | HT controls without HF (*n* = 19) Healthy controls (*n* = 10) | endoPAT (RHI) | Ischemia | Hyperaemia | Log RHI: 0.85 ± 0.42 vs. 0.92 ± 0.38 vs. 1.33 ± 0.34, *p* = n.s. between HFpEF and HT controls |
| | | | Skin-arm | | | |
| Prospective [28] | *n* = 45 | HT controls, matched for age, sex and diabetic status (*n* = 45) | Laser Doppler flowmetry (LDF), power spectral density (PSD) of the LDF signal | None, ischemia | Vasomotion, hyperaemia | LDF PSD: lower in HFpEF, no absolute numbers reported, *p* < 0.05. Peak blood flow (PU): 135 [104–206] vs. 177 [139–216], *p* = 0.03 |
| Prospective [11] | HFpEF with CAD *n* = 12 | HFrEF with CAD (*n* = 12) CAD without HF (*n* = 12) | Laser Doppler imaging (LDI) coupled with transcutaneous iontophoresis of vasodilators | acetylcholine, sodium nitroprusside | Hyperaemia | Vasodilation due to Acth: No absolute values reported. *p* = 0.00099 (HF |

| Study Design | HFpEF Population | Control Population | Method (Measurement) | Stimulus | Microvascular Function Assessed | Outcome (SD/IQR) |
|---|---|---|---|---|---|---|
| | | | | | | vs. controls). Vasodilation due to nitroprusside: $p = 0.006$ (HF vs. controls) |
| | | | Muscle-leg | | | |
| Prospective [16] | $n = 22$ | Healthy controls, age-matched ($n = 43$). | Histology (skeletal muscle biopsy of thigh) | | Capillary density | Capillary-to-fibre ratio: 1.35 ± 0.32 vs. 2.53 ± 1.37, $p = 0.006$ |
| Prospective [29] | $n = 7$ | No controls. | Near-infrared spectroscopy: | | Diffusion | Muscle deoxygenation |

t failure; ion; HT,

| Study Design | Study Population | Method (Measurement) | Stimulus | Microvascular Function Assessed | Outcome (SD/IQR) | Outcome Adjusted for Confounders |
|---|---|---|---|---|---|---|
| | | | Heart-autopsy | | | |
| Retrospective [12] | Deceased: HFpEF ($n = 124$); Controls (no HF) ($n = 104$) | Histology: microvessels/mm$^2$ (microvascular density) | | Rarefaction | Microvascular density: 961 (800–1370) vs. 1316 (1148–1467), $p < 0.0001$ | Not performed, unmatched population |
| | | | Invasive coronary function assessment | | | |
| Retrospective [14] | CAG after positive stress test: HFpEF > 65 ($n = 32$); HFpEF < 65 ($n = 24$); Controls ($n = 31$) | Invasive CFR and IMR | Adenosine | Hyperaemia | CFR: 1.94 ± 0.28 vs. 1.83 ± 0.32 vs. 3.24 ± 1.11, $p \leq 0.04$ IMR: 39.2 ± 6.8 vs. 27.2 ± 6.4 vs. 18.3 ± 4.4, $p \leq 0.03$ | Age, sex, HT, DM, CKD, AF, BMI, LVMI. Unmatched controls |
| Retrospective [9] | HFpEF ($n = 162$) | Invasive CFR and coronary blood flow (CBF) | Adenosine, acetylcholine | Hyperaemia | No absolute values reported. Mortality is increased in coronary | Age, sex, BMI, DM, HT, hyperlipidaemia, smoking, Hb, creatinine, uric acid |

| Study Design | Study Population | Method (Measurement) | Stimulus | Microvascular Function Assessed | Outcome (SD/IQR) | Outcome Adjusted for Confounders |
|---|---|---|---|---|---|---|
| | | | | | MVD (HR 2.8–3.5). | |
| Retrospective [30] | HFpEF (*n* = 22); no HFpEF (*n* = 29) | Invasive CFR and CBF | Adenosine, acetylcholine | Hyperaemia | CFR: 2.5 ± 0.6 vs. 3.2 ± 0.7, *p* = 0.0003 Median CBF % increase: 1 (−35;34) vs. 64 (−4;133), *p* = 0.002 | Age, sex |
| Prospective [31] | HFpEF with obstructive epicardial CAD (*n* = 38); HFpEF without epicardial CAD (*n* = 37) | CAG (CFR, coronary reactivity, IMR) and MRI | Adenosine, acetylcholine | Hyperaemia | CFR: 2.0(1.2–2.4) vs. 2.4(1.5–3.1), *p* = 0.06. IMR: 18(12–26) vs. 27(19–43), *p* = 0.02. 24% microvascular spasm due to Acth. | Clinical characteristics are compared between groups based on coronary results. |
| Prospective (cross-sectional) [13] | Clinical indication for CAG: HFpEF (*n* = 30); Controls (*n* = 14) | Invasive CFR and IMR | Adenosine | Hyperaemia | CFR: 2.55 ± 1.60 vs. 3.84 ± 1.89, *p* = 0.024 IMR: 26.7 ± 10.3 vs. 19.7 ± 9.7, *p* = 0.037 | Exploratory analysis on age, BMI, GFR, BNP, echocardiographic data, hemodynamic data. Unmatched controls |
| Retrospective [32] | Patients with angina presented to the ER: HFpEF (*n* = 155); Controls (*n* = 135) | Total myocardial blush grade score (TMBGS) | None, nitroglycerin | Blood flow | TMBGS: 5.6 ± 1.22 vs. 6.1 ± 1.26, *p* = 0.02 | Not performed, unmatched population |
| | | | Non-invasive coronary assessment | | | |
| Prospective [33] | HFpEF (*n* = 19); Matched | PET (C-acetate-11): myocardial blood flow (MBF) | Dobutamine | Blood flow, hyperaemia, diffusion | MBF increase: 78% vs. | LVH, Hb. Healthy controls were |

| Study Design | Study Population | Method (Measurement) | Stimulus | Microvascular Function Assessed | Outcome (SD/IQR) | Outcome Adjusted for Confounders |
|---|---|---|---|---|---|---|
| | healthy controls (*n* = 19) | and myocardial oxygen consumption ($MVO_2$) | | | 151%, *p* = 0.0480 MVO2 increase: 59% vs. 86%, *p* = 0.0079 Absolute values during stress test not significantly different. | matched for age and sex. |
| Retrospective [34] | Indication for cardiac PET: HFpEF (*n* = 78); HT without HF (*n* = 112); No HF no HT (*n* = 186) | PET (Rb-82): global myocardial flow reserve (MFR) | Dipyridamole | Hyperaemia | MFR: 2.16 ± 0.69 vs. 2.54 ± 0.80 vs. 2.89 ± 0.70, *p* ≤ 0.001 | Age, sex, BMI, smoking, DM, HT, hyperlipidaemia, HT, AF, statin use. Controls matched for HT. |
| Retrospective [35] | Suspected CAD: Cohort without HF (*n* = 201) | PET (Rb-82): (CFR) | Regadenoson or dipyridamole | Hyperaemia | 18% of the patients had a HFpEF event during follow-up. Independent HR with CFR <2.0 of 2.47 (1.09–5.62) | In entire cohort: AF, CKD, troponin, LVEF, CFR, E/e' septal |
| Prospective [36] | HFpEF (*n* = 25); LVH (*n* = 13); Controls (*n* = 18) | MRI (CFR) | Adenosine | Hyperaemia | CFR: 2.21 ± 0.55 vs. 3.05 ± 0.74 vs. 3.83 ± 0.73, *p* ≤ 0.002 | BNP, LVEF, E/e', LA dimension |
| Retrospective [37] | HFpEF without events (*n* = 137), with events (*n* = 26) | MRI (CFR) | Adenosine | Hyperaemia | CFR: 2.67 ± 0.64 vs. 1.93 ± 0.38 | Not performed |

Abbreviations: AF, atrial fibrillation; BMI, body mass index; CAD, coronary artery disease; CAG, coronary angiography; CFR; coronary flow reserve; CKD, chronic kidney disease; CMD, coronary microvascular dysfunction; DM, diabetes mellitus; ER, emergency room; GFR, glomerular filtration rate; HF, heart failure; HFpEF, heart failure with preserved ejection fraction; HR, hazard ratio; HT, hypertension; IMR, index of microcirculatory resistance; LAVI; left atrial volume index; LV, left ventricle/ventricular; LVEDI, left ventricular end-diastolic volume indexLVEF,

| Study Design | Study Population | Method (Measurement) | Stimulus | Microvascular Function Assessed | Outcome (SD/IQR) | Outcome Adjusted for Confounders |
|---|---|---|---|---|---|---|
| Prospective [38] | HFpEF (*n* = 6); Post MI (*n* = 6); Healthy controls (*n* = 20) | MRI: intravascular volume of basal septum (IVV) | Gadofosveset | Permeability | IVV: 0.155 ± 0.033 vs. 0.146 ± 0.038 vs. 0.135 ± 0.018, *p* = 0.413 | Not performed, unmatched controls |
| Prospective [10] | HFpEF (*n* = 202) [40] | Echocardiography [5] (CFR) | Adenosine | Hyperaemia | CFR: 2.13 ± 0.51 | Age, sex, BMI, AF, DM, CAD, smoking, LV mass, 6MWT, KCCQ, urinary albumin-creatinine ratio. No controls. |
| Prospective [39] | HFpEF (*n* = 77); Healthy controls (*n* = 30) | Echocardiography (CFR) | Adenosine | Hyperaemia | CFR: 1.7 ± 0.2 (with MVD) vs. 3.1 ± 0.4 (no MVD) vs. 3.4 ± 0.3 (control) | Age, LAVI, LVMI, LVEF, E/e', 6MWT distance |

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mutually exclusive, as most of the current techniques do not show cell type specific vasodilation due to the dynamic nature of the microcirculation in vivo. For instance, adenosine is an endothelium-independent vasodilator [41], but its responses in vivo are also modulated by flow-dependent NO production of endothelial cells [42][43]. Moreover, supra-physiological adenosine concentrations, as used in coronary tests, are used to assess maximal hyperaemia; increased blood flow as a consequence of maximal vasodilatation [44]. Maximal hyperaemia is influenced by structural microvascular properties such as rarefaction as well as endothelium-independent vasodilation. In addition, stimuli that induce tachycardia also influence blood flow through active hyperaemia [45][46]. Findings of normal maximal hyperaemia do not exclude impairment of active hyperaemia under physiological conditions due to the involvement of more processes that could compensate for the processes occurring under physiological conditions [47]. Nevertheless, the classification of endothelium-dependent and independent vasoreactivity, as used in this review, can help to identify different underlying mechanisms for MVD or disturbed downstream pathways.

### 3.1.1. Endothelium-Dependent Vasodilation

Impaired endothelium-dependent vasodilation and hyperaemia of peripheral microvascular beds have been studied scarcely in HFpEF. In a subgroup of coronary artery disease patients with HFpEF, HF with reduced ejection fraction (HFrEF), and no HF, one small study featured laser Doppler imaging of the forearm skin blood flow coupled with transcutaneous iontophoresis (delivery of a substance through the skin using a small electric current) using acetylcholine (endothelium-dependent) and sodium nitroprusside (endothelium-independent) [11]. Impaired endothelium-dependent and endothelium-independent hyperaemia were observed in both HFpEF and HFrEF patients compared to controls [11]. Another study reported impaired endothelium-dependent vasomotion measured as a rhythmic variation of blood flow using laser Doppler flowmetry in HFpEF patients compared to hypertensive controls [28]. However, the small sample size of the studies and the inadequate matching of comorbidities in the

control groups limit the generalization of these results. Since all patients in the first mentioned study had a form of coronary artery disease [11], the findings of this study could mainly be driven by triggers involved in atherosclerosis [48] rather than HF(pEF) [49], although similar pathways have been identified [48][49].

In the heart, impaired coronary endothelium-dependent vasoreactivity has been demonstrated in HFpEF [9][30][31]. Yang et al. showed impaired coronary microvascular function in 72% of HFpEF patients who underwent invasive coronary microvascular assessment. This entailed 29% of the patients with isolated impaired endothelium-dependent coronary vasoreactivity measured by coronary blood flow (CBF, cut-off ≤ 0% increase) using intracoronary acetylcholine infusion; isolated impaired coronary maximal hyperaemia in 33% by coronary flow reserve (CFR, cut-off < 2.5) using adenosine infusion; and combined impaired coronary vasoreactivity in only 10% (both CFR < 2.5 and CBF ≤ 0%) [9]. Similar findings were confirmed in hospitalized HFpEF patients using invasive measurements [31]. Those results suggest that impaired coronary vasoreactivity is underreported when only endothelium-dependent vasodilatation is assessed [9][31]. In contrast, another recent study similarly used CFR and CBF and reported more endothelium-dependent MVD (86%) than impaired maximal hyperaemia (46%) in HFpEF patients, which were both more prevalent compared to controls without HFpEF (35% and 21%, respectively) [30]. Moreover, at rest, endothelium-dependent MVD, but not impaired maximal hyperaemia, was associated with higher cardiac filling pressures, a sign of cardiac congestion and a hallmark of HFpEF [1]. During exercise, both forms of MVD showed this association [30]. These results could, on the one hand, reflect that some HFpEF patients show different coronary microvascular alterations as compared to other HFpEF patients within the same population and compared to other populations, which could represent unequal disease progression or different underlying MVD mechanisms within the broad HFpEF spectrum. On the other hand, interpretation of differences between study results is limited by the low sample size and inadequate correction for confounders, absence of a control group, or a retrospective design that take in a selection bias. This bias encompasses that included patients have a higher likelihood of having coronary abnormalities than a general HFpEF population because of clinical suspicion of coronary abnormalities being the indication for the diagnostic test.

A promising vascular bed to provide knowledge on endothelium-dependent and independent MVD in HFpEF is the retina, mainly because the retina allows the evaluation of both structural and functional microvascular features by direct imaging of microvessels [50][51]. Studies have suggested a link between alterations in the retinal microcirculation and HFpEF; an association was shown between retinal microvascular changes, i.e., decreased arteriolar calibres (vessel widths) and increased venular calibres, and increased LV concentric remodelling [52][53], one of the characteristics for HFpEF [3]. Moreover, it was shown that retinopathy and widening of retinal venular calibres, but not narrowing of arteriole calibres, independently predicted HF incidence in large datasets [53][54]. Finally, an association was found between current and incident HFpEF and (self-)reported retinopathy as a complication of diabetes mellitus [55][56], as well as neuropathy and nephropathy [55]. This reported association was stronger than the association with HFrEF [55]. However, retinal alterations are associated with many other factors in similar datasets [57]. Thus, retinal microvascular alterations are presumably present in HFpEF, but which specific alterations are present in HFpEF remain to be identified.

Several other techniques have been used more frequently to assess endothelium-dependent and endothelium-independent flow responses in HFpEF, including flow-mediated dilation (FMD) of larger arteries [58]. As those studies have focused on the macrocirculation and its associated endothelial phenotype [21][59], this evidence falls outside of the scope of this review but has been reviewed elsewhere [60].

Furthermore, reactive hyperaemia by peripheral arterial tonometry of the finger is often referred to as microvascular endothelial dysfunction in studies but is mostly investigated by vasoactive stimuli that are not endothelium specific. Data from this technique are presented in the next section, while researchers provide more context on the technique here. This assessed reactive hyperaemia resembles post-ischemic hyperaemia after 5 min arterial occlusion involving many microvascular metabolites, including adenosine and NO, rather than only endothelium-dependent hyperaemia, which would occur after 1–3 min occlusion [61]. This response being not fully endothelium-specific is further supported by studies showing poor comparability of results of digital post-occlusive hyperaemia with other microvascular assessments such as laser Doppler flowmetry with iontophoresis of an endothelium-dependent stimulus [62][63]. Moreover, the reactive hyperaemia is minimally affected after smoking cessation [64][65], while smoking is a well-known trigger for oxidative stress and its effects on NO-dependent endothelial vasodilatation [66][67]).

### 3.1.2. Endothelium-Independent Vasodilation

Peripheral endothelium-independent vasodilation has been studied in HFpEF predominantly in the digital microcirculation. Following the reported association of post-occlusive reactive hyperaemia in the finger using peripheral arterial tonometry with multiple cardiovascular risk factors in the Framingham Heart study [68], most HFpEF studies have used this technique. Several studies reported a lower hyperaemic response after 5 min occlusion of the brachial vasculature in HFpEF patients compared to matched control subjects [22][24][26][27], which was associated with worse outcome [23]. However, HFpEF patients showed a better digital hyperaemic response than HFrEF patients [25]. Impaired endothelium-independent vasodilation was also shown in the forearm skin in coronary artery disease patients with HFpEF compared with patients without HF [11]. Nonetheless, multiple comorbidities have been associated with MVD regardless of HF (**Table 3**) [18][69][70][71]. Therefore, it is important to take comorbidities into account when interpreting these results. Some studies were small, and the control individuals were often inadequately matched for comorbidities (**Table 1**) [11][26][27]. Controls matched for hypertension showed worse digital hyperaemia compared to healthy controls and showed no difference compared to HFpEF in one study [27], but in two other larger studies, HFpEF subjects had worse digital hyperaemia compared to controls matched for at least hypertension, age, sex, and diabetes mellitus [22][24]. These data underline that MVD is influenced by multiple clinical factors, but particularly show that HFpEF seems to have exaggerated MVD compared to its comorbidities.

**Table 3.** Clinical factors associated with microvascular dysfunction.

| Clinical Factor | Measurement Method | Microvascular Bed Assessed | Effect on Microvascular Function |
|---|---|---|---|
| Age [34][69][72][73][74] | | Skin, eye, skeletal muscle, heart | Function decreases by increasing age |
| Hormonal status [75][76][77][78] | Oestrogen levels, together with oestrogen receptor activity, are most accurate. Menopausal status and oral contraceptive therapy use are alternative surrogate markers. | Skin, skeletal muscle, heart | Function decreases with lower oestrogen activity |
| Hypercholesterolemia [70][79][80] | Serum cholesterol panel | Skin, eye, heart | Function decreases with higher serum low-density lipoprotein cholesterol levels |
| Hyperglycaemia [81][82] | Glucose tolerance test, fasting glucose, HbA1c | Skin, eye, heart | Function decreases with higher plasma glucose levels |
| Hypertension [34][36][69][83][84] | 24-h systolic blood pressure shows the highest correlation | Skin, eye, skeletal muscle, heart | Function decreases with higher systolic blood pressure and by duration of hypertension |
| Dietary intake [85] | Caffeine | Skin | Function is temporarily increased |
| Dietary intake [86][87] | High-fat diet | Skin, heart | Function is temporarily decreased |
| Physical inactivity [29][88][89][90] | 24-h accelerometer, physical activity questionnaire | Skin, eye, skeletal muscle | Function decreases with more physical inactivity. |
| Obesity [8][69][91][92] | Waist circumference is more correlated than BMI or BSA. | Skin, eye, skeletal muscle, heart | Function decreases with increasing level of obesity |
| Sex [93][94] | | Skin, eye, skeletal muscle, heart | Effect on function depends on other confounders. |
| Smoking [74][95] | Self-reported use | Skin, eye, heart | Function decreases with smoking and more pack years. |

Abbreviations: BMI, body mass index; BSA, body surface area; HbA1c, glycated haemoglobin.

Clinical studies focusing on endothelium-independent vasodilation of the coronary microcirculation have assessed hyperaemia using different stimuli. Invasive studies have revealed impaired maximal hyperaemia in HFpEF patients compared to controls without HF, both free of epicardial stenosis. In particular, these studies reported impaired coronary flow reserve (CFR) and an increased index of microcirculatory resistance (IMR) after intracoronary adenosine infusion [13][14][30][32]. In parallel, non-invasive imaging studies assessing coronary vasoreactivity using positron emission tomography (PET), magnetic resonance imaging (MRI), or echocardiography revealed similar results, mainly using adenosine for maximum hyperaemia and dipyridamole or dobutamine as a stimulus for active hyperaemia [33][34][35][36][39] (**Table 2**). Moreover, impaired maximal hyperaemia was associated with more adverse events in HFpEF [37]. All but two of these coronary MVD studies were retrospective and, thus, also take in a selection bias with a higher likelihood of having coronary abnormalities than a general HFpEF population. In addition, the controls in these studies had fewer comorbidities than HFpEF patients, and limited confounder correction was performed. Nevertheless, the results all pointed in the same direction; coronary MVD is present in HFpEF.

To date, only one study assessed both coronary and peripheral (micro)vascular function in HFpEF. In their assessment, Shah et al. used novel echocardiographic tools (Doppler imaging to assess CFR of the left anterior descending artery after adenosine infusion) and observed a high prevalence of impaired maximal coronary hyperaemia in HFpEF (75% of 202 patients, no controls). This was significantly but weakly correlated with impaired peripheral vasoreactivity as measured by endoPAT [10]. This direct comparison between the peripheral skin microvasculature and coronary microvasculature is particularly limited by the use of different triggers to assess vasoreactivity (adenosine vs. ischemia), which would already result in different hyperaemic responses in the same vascular bed [61].

Taking all published results regarding vasoreactivity of different (micro)vascular beds together, HFpEF patients consistently show impaired (micro)vascular vasoreactivity throughout the body, suggestive of systemic MVD. However, not all patients show identical MVD phenotypes. Nonetheless, a causative conclusion on the gradual development of MVD and HFpEF requires more robust evidence.

### 3.2. Other alterations of the microcirculation in HFpEF

Elaboration on these topics are reviewed elsewhere [96]. Briefly, capillary rarefaction of vascular beds in the upper legs and the heart, as well as biomarkers of microvascular function such as vascular cell adhesion molecule 1 (VCAM-1), have been reported in HFpEF compared to controls. Moreover, tissues such as adipocytes and cardiomyocytes have alterations in HFpEF compared to controls.

## 4. Systemic Microvascular Dysfunction in HFpEF

Systemic MVD is present in HFpEF, based on interpretation of abundant data from many correlational studies that show impairments in microvascular function, both endothelium-dependent and endothelium-independent, in different vascular beds. MVD should be seen as a continuum between function and dysfunction, which can

influence HFpEF and comorbidity progression, and vice versa. Hitherto, due to a lack of clear causative evidence, it remains unknown how systemic MVD could drive HFpEF.

Furthermore, HFpEF patients unequally show different elements of MVD, which might reflect different underlying mechanisms and therapeutic targets. Future research on MVD and HFpEF is, therefore, needed to uncover the true diagnostic and therapeutic value of microvascular assessments. This will require more uniformity and confounder considerations in study design, analyses, and reporting. However, the incorporation of peripheral microvascular assessments is feasible and should be considered in clinical HFpEF trials.

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