CORONARY FLOW RESERVE

TO ASSESS MICROCIRCULATION WITH ECHOCARDIOGRAPHY:

BASIC CONCEPTS, NEW INSIGHT AND FUTURE PRESPECTIVES.

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Abstract

Coronary flow reserve is the capacity of the coronary circulation to augment the blood flow in response an increase in myocardial metabolic demands and has a powerful prognostic significance in different clinical situations. It might assess with invasive and non-invasive technique. Transthoracic echocardiography Doppler is an emerging diagnostic technique, noninvasive, highly feasible, safe for patient and physician, without radiation, able to detect macrovascular and microvascular anomalies in the coronary circulation. This review aims to describe the benefit and limits of noninvasive assessment of coronary flow reserve, in particular his evaluation with echocardiography.

Keywords: coronary flow reserve; Doppler echocardiography; coronary microvascular dysfunction; microcirculation; coronary physiology.

INTRODUCTION

Coronary flow reserve (CFR), assessed by transthoracic echocardiography Doppler (TTE), is a non-invasive diagnostic technique able to reflect presence of macrovascular as well as microvascular disease in the coronary circulation, with the advantages of being highly feasible, safe for patient and physician, and not associated with any radiation.

In particular, it could serve as a diagnostic tool for coronary microvascular dysfunction, while in the setting of ischemic cardiac disease is useful in identification and assessment of functional significance of coronary lesions as well as for the follow-up of patients after coronary interventions. In addition, CFR has also showed a powerful prognostic significance in different clinical situations.

CFR and microcirculation: general concepts and physiology.

Coronary circulation is characterized by complex morphology and physiology. Coronary arteries bifurcate into smaller vessels [1], divided into three categories in terms of functional and anatomical characteristics:

- large arteries (diameter > 500 μm) with capacitance function and little resistance to blood flow;

- small arteries or pre-arterioles (diameter between 100 and 500 μm), that represent the intermediate compartment with a measurable pressure drop along their length

- arterioles (diameter <100 μm) that represent the distal compartment with a very large pressure drop along their length [2].

The major part of the coronary resistance vessel is assigned to arterioles and small arteries, which are referred to as resistance vessels, but capillaries might play an important role in the regulation of CFR [3,4]. Unlike other organ blood flow, myocardial perfusion is predominantly diastolic with unique profile of coronary blood flow velocity (CBFV), characterized by a diastolic predominant pattern, and by a significant systolic retrograde flow which is called a “coronary slosh phenomenon” [5]. The coronary slosh is enhanced by

coronary artery stenosis and further increases following the administration of vasodilatory substances [5].

There are various regulation mechanisms of coronary flow mediated by metabolic, neurohumoral and myogenic factors: adenosine, nitric oxide, oxygen, norepinephrine, endothelin, acetylcholine, angiotensin II, endothelium-derived hyperpolarizing factor [6], maintaining the luminal pressure of capillaries within a physiological range [7-9]. The myogenic response is endothelium-independent, but the flow-induced vasodilation is endothelium-dependent [10,11].

The coronary microcirculation plays a key role in the myocardial perfusion. Therefore, the presence of functional and/or structural abnormalities of this circulatory pathway may impair the myocardial perfusion, a condition referred as *coronary microvascular dysfunction* (CMD). The coronary microvasculature cannot directly imaged *in vivo* (vessels <300 μm), but the CMD may be assessed by many invasive and noninvasive technique to evaluate the coronary microvascular function, such as the CFR, which represents an integrated measure of coronary blood flow (CBF) in both the macro- and microcirculation and consists in the ratio of hyperaemic to baseline blood flow [2].

The CFR is the capacity of the coronary circulation to increase the blood flow in response an increase in myocardial metabolic demands, reducing vascular resistance through the dilatation of the arteriolar bed [4].

In the absence of obstructive stenosis of the epicardial arteries, reduced CFR is a marker of CMD, but because obstructive disease of the epicardial arteries and CMD often coexist, discrimination between the effects of these two conditions on myocardial perfusion can be difficult. CMD is related to several pathological conditions, and is influenced by several factors: metabolic demand, diastolic time, systolic arterial blood pressure and heart rate. Some authors suggest to correct CFR for the rate–pressure product and to take into

account the differences of myocardial blood flow (MBF) related to sex and age [12,13]. Coronary flow reveals marked spatial heterogeneity of CFR across the myocardial wall. The highest CFR is that measurable in the subepicardial layer of myocardium, whilst is significantly lower in the subendocardial layer, also due to elevated left ventricular (LV) diastolic pressure increasing extravascular compressive forces [14].

Pharmacological stressors.

To produce maximum vasodilatation for CFR calculation, pharmacological stressors are usually used. The agents most frequently chosen to induce coronary vasodilatation are papaverine [15-17], regadenoson, [18], adenosine [19] and dipyridamole [20], but in clinical setting the most used are dipyridamole and adenosine. Dipyridamole is an adenosine transport blocker that produces elevation of tissue adenosine levels. Moreover, it inhibits phosphodiesterases and causes an increase in cellular content of cyclic Adenosine Monophosphate (cAMP) and cyclic guanosine monophosphate (cGMP), with the result of coronary dilation [21]. Adenosine and dipyridamole have an advantage over exercise and dobutamine, which represent submaximal stimuli for CFR [22]. Coronary vasodilators are usually administered intravenously in noninvasive techniques, but adenosine is generally given intracoronary when measuring CFR in the catheterization laboratory. Cold pressure testing can also be used in combination to assess endothelium-dependent coronary vasodilator function, to unmask abnormalities of endothelial function, when pharmacological vasodilatation is still preserved [2].

CMD: pathogenesis and mechanisms.

Multiple mechanisms are potentially responsible for CMD and can be classified in structural, functional and extravascular changes.

Structural anomalies include vascular wall infiltration, vascular remodeling, perivascular fibrosis and luminal obstruction. Smooth cell hypertrophy and the increased deposition of collagen, such as in hypertension or in hypertrophic cardiomyopathy (HCM), determine thickening of the media and intima tunica [23].

Functional alterations are related to impaired dilatation or increased constriction of the coronary arterioles and pre-arterioles [2]. Impaired vasodilatation can be due to a pathological endothelium-dependent pathway (such as acetylcholine, and serotonin and flow-mediated) or to endothelium-independent pathways (eg, involving adenosine). On the other hand, increased vasoconstriction can be caused by increased levels of endothelin-1, catecholamines, and acetylcholine [23].

Extravascular anomalies consist in increased heart rate, with reduction in diastolic perfusion time, reduced driving blood pressure, extramural systolic compression, more severe in the endocardial layers [24].

These different mechanisms frequently are simultaneously and can be observed in the absence of myocardial disease and obstructive CAD as well as in conditions with myocardial disease without obstructive CAD and is usually diffuse and affects the entire ventricle. Camici and Crea originally proposed a clinical and pathogenic classification of CMD [24]. (**Table 1**).

CFR ASSESSMENT IN ECHO-LAB

CFR reserve could be estimated with various non-invasive techniques, each of which has got limitations. We will focus CFR assessment in echo-lab: actually, echocardiography represents the most reliable and easy method to evaluate CFR and coronary microcirculation.

The left anterior descending (LAD) coronary CBVF profile can be recorded with pulsed-wave Doppler, either with TEE (sampling the proximal tract) or transthoracic echocardiography (TTE) (exploring the mid-distal tract). Recently, the success rates to measure CBFV of right coronary artery (RCA) and LCA have been increasing owing to the advancement in ultrasonic technology [25,26]. CBVF by Doppler is represented by a biphasic wave, with a lower peak during systole and a higher peak during diastole, for the effect of myocardial contraction [27]. Several parameters might be measured from Doppler tracings of LAD artery flow, including systolic flows, time–velocity integrals, and mean flows, but the best parameter is peak diastolic flow: it is easy to measure, reproducible and it has a closest correlation with CRF measured with PET [28]. Second harmonic imaging and high frequency transducers (up to 8 MHz) provide better definition of coronary artery and improved resolution. Contrast agents also improved the signal-to-noise ratio, increasing the feasibility of transthoracic imaging of the LAD artery [29].

Methodology

Detection of coronary arteries

The principal coronary artery investigated is the LAD branch. It can be divided in three tracts: proximal, intermediate and distal. The key reference points to detect the proximal tract are the left atrial appendage and the pulmonary artery. The intermediate tract key reference points are the septal perforans branches. The distal LAD tract is more suitable to investigate coronary microvascular function because it is between large epicardial arteries and microvasculature. This tract can be investigated in B-mode and under Color Doppler guidance and by using growing delivery frequencies (5-7 MHz) in the second harmonic [30,31]. For a better visualization, the setting depth should be reduced approximately to 6-10 cm. The acoustic window is, in the left decubitus position, around the midclavicular line in the fourth

or fifth intercostal space (**Figure 1**). The distal LAD can be assessed from the low left parasternal position to a modified apical five-chamber position at varying levels using different short- and long-axis views in the anterior interventricular groove before, at or after the apex of the LV [32]. The CBF in the distal LAD is searched for under the color Doppler flow. Angle correction is necessary in each examination. CBF is characterized by a biphasic flow pattern with a larger diastolic component and a smaller systolic one (**Figure 1**).

PDA, usually the distal part of the RCA can be assessed from fourth- and fifth intercostal spaces in the apical long-axis position in a modified two- or three-chamber view with caudal tip of the transducer in the posterior interventricular groove [32] (Figure 1). Unlike the LAD, the scanning depth for the RCA and LCA should be set at 12-15 cm. It's possible to assess the RCA and LCA proximal and mid tract in a modified parasternal short axis focused on great vessels, but pulse-wave Doppler examination is rarely correct because the angle between the direction of CBF and Doppler beam exceeds 60 degrees. The first or second obtuse marginal branches (OMB) presenting distal parts of the LCA can be assessed from fourth- and fifth intercostal spaces in the apical long-axis position in a modified four- or five chamber view at either lateral or inferior wall of the LV [32].

Feasibility of TTE in assessing coronary arteries with the addition of harmonics and contrast agents has been reported to be as high as 100% for distal LAD and 33-97% for the PDA [30,31,33].

Pitfalls in detecting coronary arteries

There are many pitfalls regarding the CFR execution by TTE. First, regarding the assessment of coronary arteries: some branches of coronary arteries can occasionally be confused with the LAD. To avoid this mistake, the LAD should be visualized in the anterior interventricular groove in its entire length with the evidence of left main coronary artery

(LMCA) and distal LAD. Furthermore, LAD can be confused with extracardiac arteries on color Doppler, such as the left internal thoracic artery (LITA), but, in this case, the pulse wave Doppler shows a typical peripheral arterial flow with high systolic flow velocity. Larger cardiac veins can be distinguished for their three-phasic, predominantly systolic, flow with respiratory variations and for their position, closer to the right ventricle [34].

CBFV and CFR assessment with pulse wave color Doppler.

To obtain a correct CBVF and CFR assessment, it's necessary that the correct Doppler angle and the sample volume are maintained in the correct position throughout the infusion of the vasodilator agent [32]. The optimal incidence angle between the Doppler beam and flow direction is of 30 degrees. The sample volume should be sized (1.5-3.0 mm) and placed within the coronary artery [35]. Time velocity integral, peak velocity and mean velocity in systole and diastole should assessed at rest and during the pharmacological stimuli. The diastolic blood flow velocity is the most used parameter. Furthermore, it's possible to assess the diastolic and systolic flow length of time [36,37] (**Figure 2**). The normal value of the peak diastolic velocity in distal LAD was 21.2±7.9 cm/s and the duration of diastolic coronary artery flow was 58.5±6.4% of the R-R interval at rest and with normal heart rates (60-100 b/ m), also coronary flow velocities showed a non-significant decrease from proximal segments to distal segments [38,39].

Like invasive CFR assessment, adenosine is the first choice to assess CFR. Adenosine protocol usually consists in intravenously infusion at the rate of 140 mcg/kg/min for 2 minutes, measuring CBFV before and after of adenosine infusion. Adenosine has a short half-life, compared with dipyridamole (up to 30 minutes) and CFVR achieved with adenosine, can be obtained with dipyridamole infusion rate of 0.84 mg/kg for 6 minutes [40].

After measuring the baseline and the hyperemic coronary flow velocities, CFR should be calculated as the ratio of CBFV under maximal vasodilatation to CBVF at rest [41]. Many authors assume that the peak velocity (VpD) can be used for CFVR evaluation and the cut-off <2 for predicts significant LAD stenosis in distal LAD and in PDA [42,43], whereas CFVR <1 denotes LAD or PDA occlusion [44,45].

In patient with chronic artery disease (CAD), CFR is related to the severity of coronary disease, while in case of angiographically normal arteries it is a marker of microvascular dysfunction [46] (Figure 3).

Recently, the ABCDE protocol, validated by the Stress Echo 2020 study group of the Italian Society of Echocardiography and Cardiovascular Imaging, has been proposed during stress echocardiography to investigate dyspnea of cardiac origin, analyzing 5 parameters: regional wall motion (step A); pulmonary congestion with B-lines (step B); LV contractile reserve (step C); coronary microvascular dysfunction with CFR (step D); and EKG-based heart rate reserve during exercise (step E) [47,48]. Reduced CRF is often accompanied by regional wall motion anomalies, abnormal LV contractile reserve, and pulmonary congestion during stress, and shows independent value over segmental motion anomalies in predicting an adverse outcome [49].

CRF assessed by TEE.

CBFV can be assessed by TEE with Doppler examination of the proximal LAD coronary artery. This is a simple, reproducible, safe, and reliable method for coronary flow reserve assessment, and is correlated with CBFV derived from intracoronary Doppler guide wire studies [50]. Coronary artery imaging was performed after routine cardiac examination. The LMCA was visualized, with the probe approximately 28 cm from the mouth, at a level above the aortic leaflets, [51]. The LMCA, arising from the corresponding sinus, is seen as an

echo-free structure and small adjustments in transducer orientation are necessary to visualize the artery, its bifurcation and LAD. CBFV was evaluated by pulsed wave Doppler of the ostial part of the LAD [51].

Myocardial contrast echocardiography (MCE).

Quantitative contrast echocardiography has been shown to be useful for assessing global and regional MBF reserve using different pharmacologic stimuli. This technique seems feasible for the assessment of mechanistic insights at coronary microcirculation [52]. MCE utilizes gas-filled microbubbles to produce myocardial opacification on ultrasound examination. After complete destruction of microbubbles using a beam of high-intensity ultrasounds, the replenishment of myocardial microcirculation may be assessed as a time-intensity curve, with a mathematical function [53]. In patients without significant stenosis of coronary arteries, contrast infusion determines homogeneous opacification of the myocardial wall, because microbubbles continuously replaced those that are destroyed by ultrasounds. In case of microvascular defects, myocardial flow increase during pharmacological/physical stress and determines regional differences of opacification. An excellent correlation was found with PET [53]. MCE did not receive regulatory authorities approval for clinical use, and the technique remains an option for research [53].

CORONARY MICROVASCULAR EVALUATION IN DIFFERENT CLINICAL SETTINGS

Aging and gender physiologic variations

Recent evidence have shown that age is correlated with a decline in endothelial function in rodent models and humans, also in the absence of cardiovascular risk factors. With advancing age the endothelium is characterized by dilatory dysfunction, occurring a decade earlier in men compared to women [54]. This dysfunction is manifested as a reduced flow reserve while maintaining baseline flow capacity similar to young levels [54]. It seems to be a limited vasodilation response to various stimuli in advanced age. In their study, Cortigiani et al analyzed data of 5,577 patients who underwent dipyridamole stress echocardiography with CFR assessment: they have shown sex-independent value of CFVR <2.0 indicates the "prognosis" cut-off for all age groups, except for those ≥85 years in whom a cut-off ≤1.90 is needed [55].

CFR differs according to gender and menstrual cycle in women. CFR increase was associated with the level of 17-beta-estradiol serum concentration, during the menstrual cycle, and acute estrogen replacement in postmenopausal women [56,57]. Estrogen are associated with augmentation in CFR and have a protective effect on coronary micro-vessels, by increasing activity of NO synthase [56].

Impact of cardiovascular risk factors

Coronary microvascular function is compromised in patients with hypertension and/or diabetes. Hypertension impairs endothelial function and increases microvascular resistance [58]. CFR was impaired in hypertensive patients compared with healthy subjects, also in patients with prehypertension [59,60]. The impairment of CFR occurs very early in hypertension, before organ damage is evident [60]. Resistant hypertension was associated with abnormal CFR as compared to patients with non-resistant hypertension [61]. Histological studies have showed increased arteriolar media area and interstitial fibrosis in patients with arterial hypertension and angina pectoris in the absence of significant coronary artery disease [62].

Coronary microcirculation dysfunction has been reported in patients with diabetes mellitus (DM) and normal coronary arteries [63]. Hyperglycemia and oxidative stress determine accumulation of advanced glycation end products, with consequent increased interstitial fibrosis [64]. Also, vasomotor function is impaired in patients with type 2 DM for decreased level of nitric oxide and increased secretion of endothelin and angiotensin II [65]. Consequently, diabetic patients have a reduced CRF, as demonstrated with different techniques such as intracoronary Doppler [66], TEE [67], and PET [68] and CFVR <2 is an independent predictor of death and nonfatal myocardial infarction [65]. In diabetic subjects, a tighter glycemic control is associated with normal CFR and better event-free survival in unselected patients and in subjects with normal coronary arteries [69,70].

CFR evaluated by TEE and PET is also impaired in patients with increased cholesterol level, without a history of coronary artery disease who did not receive lipid-lowering therapy [71,72].

Finally, tobacco smoking is associated with epicardial atherosclerosis and impaired endothelial function. It was reported that CRF was lower in smokers than in controls. CFR decreases, with no change in hemodynamic parameters, after acute smoking, determined invasively [72] and using PET [73].

Suspected CAD

CMD, evaluated by TTE, is independent predictor of cardiovascular events and adds incremental prognostic value in patients with suspected CAD. In a study by Li-Ming Gan, 371 patients underwent to myocardial perfusion scintigraphy for suspected myocardial ischemia. CFR was a significant independent predictor of major cardiovascular events. The major cardiovascular events rate was 7.5% in patients without myocardial ischemia and normal CFR, whereas event rate was 24.2% in patients without ischemia but with reduced CFR, and 46.5% in patients with both myocardial perfusion scintigraphy–detected

myocardial ischemia and a reduced CFR (P<0.001) [74].

Cardiac syndrome X

The term "syndrome X" was created to stress the uncertain pathophysiology of chest pain. This term should not be used in patients with risk factors for CAD or cardiomyopathies, in whom myocardial ischemia may be due to CMD. The syndrome X almost certainly encompasses several pathophysiological disease entities, perhaps including a group of diseases associated with coronary microvascular dysfunction, previously unidentified cardiomyopathy processes, disorders of visceral nociception and insulin resistance [75-77]. In a subgroup of these patients, angina and ST wave may be caused by pre-arteriolar dysfunction. CFR, evaluated by TTE and MCE, is impaired in patients with syndrome X [78]. Several studies have shown an abnormal CBF response to vasodilator stimuli, indicative of CMD, using thermodilution, a xenon washout method [79], PET [80], cardiac magnetic resonance (CMR) [81], and intracoronary Doppler wire recording [82].

Furthermore, patients with non-obstructive CAD can also have microvascular dysfunction, with an abnormal response to endothelial vasodilators. These coronary microvascular abnormalities might mediate ischemia and cause angina in patients with obstructed or unobstructed coronary arteries [24].

Valve heart diseases

Aortic stenosis (AS). Patients with moderate and severe AS have shown an impairment of CFR [83]. Also asymptomatic subjects have abnormal CFR, with the major impairment in severe AS [84]. CFR has a prognostic value in asymptomatic AS with preserved EF. Marko et al have shown that CFR < 1.85 has high sensitivity and specificity in predicting adverse outcome during long term follow-up [84]. The resolution of stenosis with transcatheter

aortic valve implantation (TAVI) determines a decrease of microvascular resistance and an improvement in coronary vasodilatory reserve [85].

Aortic insufficiency (AI). CFR assessed by dipyridamole-induced coronary vasodilation, is greatly impaired in patients with aortic regurgitation, LV hypertrophy, exertional chest pain and normal coronary arteries [86]. A reduced CFR in this population could be due to 1) a low coronary driving pressure (mean aortic pressure – left ventricular diastolic pressure); the diastolic aortic pressure is reduced and LV diastolic pressure may be elevated; 2) impaired vasodilator capacity for deposition of interstitial fibrous tissue; 3) failure of growth of coronary microcirculation in parallel with myocardial hypertrophy [86].

Ardehali et al created a model of acute AI using a percutaneous approach in closed-chest dogs. They showed that CFR decreased with the induction of acute aortic insufficiency. A direct relation is showed between the severity of aortic regurgitation and the impairment of CFR: with the worsening of acute aortic regurgitation, the diastolic to systolic blood flow ratio decreases, CBF occurs mainly in systole, and there is retrograde flow during diastole [87].

Mitral regurgitation (MI). CFR is impaired in patients with MI and improves after successful mitral surgery for the reduction of LV preload, LV volume and LV hypertrophy [88].

Cardiomyopathies

HCM.

Reduction in CFR is a recognized feature in HCM and is a strong predictor for future cardiovascular events [89,90]. Patients with HCM show important wall thickening of coronary arterioles, due to intimal hyperplasia and interstitial fibrosis reduces capillary density, with angina, progressive deterioration of LV function [91] (**Figure 4**). Elevated LV end-diastolic and end-systolic pressure might contribute to perfusion defects, mainly in sub-endocardial layers of myocardium. Severe CMD is situated in the hypertrophied septum, but also in the non-

hypertrophied LV free wall using PET [92] and CMR [93]. Severe impairment of microvascular function is more frequent in HCM patient with sarcomere mutations [94]. CFVR was lower in patients with obstructive HCM compared with non-obstructive HCM [95] and alcohol septal ablation results in improvement of CFR [96].

Athlete's heart

Endurance athletes showed a supra-normal CFR, probably for diminished resting coronary flow and increased hyperaemic coronary flow, despite the presence of cardiac hypertrophy [90,97]. In athletes, resting bradycardia is likely to account for some of the observed increase in CFR, since CFR diminishes linearly with increasing resting heart rate [90,97,98].

Idiopathic dilated cardiomyopathy (DCM)

CFR is decreased in patients with non-ischemic DCM [99]. An impaired CFR identifies patients at higher risk of, such as death and worsening of clinical conditions [100]. In addition to LV hypertrophy and LV pressure overload, other causes for reduced CFR in DCM patients may also be the impaired endothelium dependent vascular relaxation and the vascular wall structural abnormalities, such as interstitial and perivascular fibrosis [100].

Finally, the left bundle branch block (LBBB), in this population, was associated with reduced CFVR and diminished myocardial contractile reserve during dipyridamole stress echocardiography [101], probably for the delayed mechanical activation of the left ventricle [101].

Heart transplant

Cardiac allograft vasculopathy (CAV) is characterized by diffuse concentric myo-intimal thickening of the distal epicardial artery and the small endocardial vessels [102]. Annual coronary angiography is the most common approach to evaluate the progression of CAV. Recently, the assessment of CFR on TTE has been proposed as an alternative method to

diagnosis CAV. A recent study has shown that CFR is sensitive to assess microvascular and macrovascular dysfunction and when CFR is combined with dobutamine stress echocardiography, CAV can be accurately diagnosed with high specificity [103].

Chronic systemic inflammation and microvascular dysfunction.

Patients with autoimmune diseases, such as rheumatoid arthritis and systemic lupus erythematosus, have increased cardiovascular morbidity and mortality for premature coronary artery disease [104]. Systemic inflammation damages the coronary microcirculation. Leucocytes and soluble factors play an important role in accelerating vessel atherosclerosis. These patients have a impaired CFR without significant coronary disease, and the impairment is correlated with the duration of the disease [105].

Limitations

CFVR measured with Doppler has some limitations. The coronary blood flow is not calculated directly, because it's not possible to measure accurately the diameter of the vessel.; CFR is calculated without considering the change of coronary artery diameter during drug infusion. The changes in coronary diameter during vasodilator infusion introduce a source of error [106].

Conclusions

TTE, a noninvasive and widely used method, can be used for the diagnosis of microvascular coronary dysfunction.

After an adequate period of training, detection and measurement of distal LAD and RCA flow and CFR by TTE is feasible in more than 90% of patients. Noninvasive serial measurements of coronary flow velocity at rest and after stress is useful for understanding the

physiology and pathophysiology of microcirculation, for diagnosis and the follow-up of different clinical conditions.

The protocols of studies cited in this review, reporting the results of human experimentation, were approved by local institutional review board (IRB) and informed consents for the studies were obtained from all human subjects in accordance with the WORLD Medical Association Declaration of Helsinki: Ethical principles for medical research involving human subjects, 2013.

The figures in our review are original, not published previously and that subjects granted permission for the use of their data/images in published articles.

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FIGURE LEGENDES

Figure 1. Color Doppler images of the distal left anterior descending coronary artery (LAD) and interventricular posterior coronary artery (IVP). The modified apical 4-chamber (A) and 2-chamber (B) positions, with cranial angulation of the transducer and a Nyquist limit of 20-67 cm/s, allow optimal identification of the diastolic coronary flow.
LV=left ventricle; RV=right ventricle; LA= left atrium.

Figure 2: an example of normal coronary flow reserve in a healthy subject. Velocity patterns are registered by pulse-wave Doppler. Resting laminar peak diastolic velocity in normal coronary artery is from 0.21 ± 0.08 m/s to 0.28 ± 0.09 m/s, and the velocity will not exceed 1 m/s even in case of its 3-4-fold increase in the stenosis site. CFR is expressed as the ratio of coronary flow velocity under maximal vasodilatation to coronary flow velocity at rest. In this case the basal peak diastolic flow velocity is normal, and it increases more than 2 times after dipyridamole, and therefore CFR is normal (2.5).
LV= left ventricle; Vp(d)= velocity peak (diastolic); CFVR= coronary flow velocity ratio.

Figure 3: overview of CFR responses in different clinical settings. In the healthy subject (left), there is a normal CFR response [ratio of hyperemic (light red) to basal (dark red) peak diastolic coronary flow velocity >2. A mildly abnormal CFR response can be noted in the presence of microvascular disease or mild-to-moderate epicardial coronary artery stenosis (middle). With more significant epicardial coronary artery stenosis (right), further reduction of CFR can be found.

Figure 4: impaired CFR in an HCM patient with normal epicardial coronary arteries. Basal increased coronary flow with high peak diastolic flow velocity in distal LAD at rest; after dipyridamole, the increase of peak diastolic flow velocity is slight, CFR is equal to 1.2.
Vp(d)= velocity peak (diastolic)

CLASSIFICATION OF CMD
1. **Type 1: CMD in absence of myocardial disease and obstuctive CAD.** Syndrome X; Hypertension; Obesity; Smoking; Diabetes; hyperlipidemia and insulin resistance, syndrome; chronic inflammation (chronic inflammatory rheumatoid disease such as systemic lupus erythematosus and rheumatoid arthritis). 2. **Type 2: CMD in myocardial diseases without obstructive CAD** Primary inherited (HCM and idiopathic DCM) and secondary cardiomyopathies (eg, hypertensive heart disease, aortic stenosis, and valvular heart disease); Myocarditis; Tako-Tsubo syndrome; infiltrative cardiac diseases (such as amyloidosis and Anderson-Fabry's disease). 3. **Type 3: CMD in obstructive CAD** Stable coronary artery disease; acute coronary syndromes. 4. **Type 4: Iatrogenic CMD** Coronary vasoconstriction occurs after PCI; embolization of atheromatous debris

during PCI; Chemotherapy and radiation therapy.

CMD= coronary microvascular disease; CAD= coronary artery disease; HCM= hypertrophic cardiomyopathy; DCM= dilated cardiomyopathy; PCI=percutaneous coronary intervention.

Table 1: Classification of coronary microvascular disease.