Transmural heterogeneity of myocardial contraction and ischemia. Diagnosis and clinical implications

Paolo Colonna, Christian Cadeddu, Roberta Montisci, Lijun Chen, Luigi Meloni, Sabino Iliceto

Department of Cardiovascular and Neurological Sciences, University of Cagliari, Italy

Key words:
Coronary artery disease; Echocardiography; Myocardial ischemia; Ventricular function.

Myocardial contraction behaves heterogeneously, being greater in subendocardial than in subepicardial layers. Similarly, during acute myocardial ischemia or infarction, the subendocardium is the first myocardial layer to suffer. Conventional two-dimensional echocardiography cannot distinguish the transmural extension of myocardial ischemia or infarction, showing akinesia also when only the subendocardium is affected. Novel ultrasonographic techniques (like tissue characterization with integrated backscatter or Doppler tissue imaging) and nuclear magnetic resonance tagging can investigate myocardial contraction in different transmural layers and distinguish subendocardial from transmural ischemia or infarction.

With the advent of thrombolysis and primary angioplasty in the acute phase of myocardial infarction a correct diagnosis of the extension of myocardial necrosis cannot ignore its transmural wavefront development. The salvage of the subepicardial layer does not give direct information on overall myocardial thickening but is one of the major determinants of overall left ventricular dysfunction and size.

Although it is still necessary to investigate this phenomenon, new ultrasonographic techniques give us important information and more opportunities to appropriate diagnosis and future treatment of cardiac patients.

(Ital Heart J 2000; 1 (3): 174-183)

Received December 14, 1999; revision received March 3, 2000; accepted March 8, 2000.

Address:

Dr. Paolo Colonna

Dipartimento di Scienze Cardiovascolari e Neurologiche
Universit degli Studi
Ospedale S. Giovanni di Dio
Via Ospedale, 46
09124 Cagliari
E-mail:
pcolcard@pacs.unica.it

Introduction

In the everyday evaluation and treatment of patients with coronary artery disease information about myocardial perfusion and contraction regarding both regional and global function is usually required. Experimental studies have shown that microvascular flow[1], metabolic consumption and the strength of contraction[2,3] are heterogeneous not only between adjacent regions, but also in the different transmural layers of the myocardium.

Methods to study transmural heterogeneity of myocardial blood flow and contraction

Radiolabeled microspheres. Several methods nowadays permit coronary blood flow measurement, and between them the most common are the ones using the electromagnetic catheter (electromagnetic flowmeters implanted around the coronary arteries, Doppler flow wires or coronary sinus catheters) to determine blood flow. Conversely, it is difficult to separate the flow as it is distributed to the different layers of the cardiac muscle. The most used technique to measure blood flow in the different layers of the myocardium, is an invasive and time consuming one: the assessment of blood flow using radiolabeled microspheres[1,4]. This method consists in injecting microspheres of policarbonate with a diameter of 7-15 μ into the left atrium (to favor the mixing with blood), radiolabeled with 125I, 51Cr, 141Ce, 85Sr, 46Sc[1]. For each step of the protocol requiring a blood flow measurement a different marker of the microspheres (e.g. 125I, 51Cr, 141Ce) is used, depending on how many steps are required during the protocol (for example in different steps of a progressive coronary occlusion). At this point, a blood sample is taken from the aorta to use in the counting of the radiolabeled microspheres as a reference point. At the end, after the animal sacrifice, the different slices of the ventricle are analyzed in

the subendocardium and subepicardium. Blood flow is calculated in each myocardial layer, counting the different radiomarkers and obtaining a result with a radiomarker for each protocol step (e.g. the different levels of coronary occlusion).

According to these techniques, available only in animals or *post-mortem* human studies, the subendocardial/subepicardial myocardial flow ratio calculated at rest is approximately 1.25, thanks to a lower microvascular tone in the subendocardium[5]. This lower subendocardial microvascular tone makes this layer more susceptible to ischemia[5].

Myocardial contrast echocardiography. Recently myocardial contrast echocardiography has also been used to quantify myocardial perfusion. Several myocardial contrast agents, suspensions of microbubbles of variable diameter, have been injected initially intracoronary and nowadays also intravenously.

It must be noted, however, that the increase in contrast videointensity after myocardial contrast injection is not a direct sign of myocardial blood flow or myocardial perfusion because, different from the radiolabeled perfusion agents such as thallium or technetium, the microbubbles do not reach the myocytes out of the microvascular vessels. Despite these considerations, echocardiographic videointensity after myocardial contrast correlates with microvascular blood volume; in fact, a close correlation between the mean myocardial red blood cell transit rate and microbubble myocardial transit rate has been demonstrated ($r = 0.89$), indicating that the transit rates of microbubbles during myocardial contrast echocardiography in experimental studies can be used to assess regional myocardial blood flow in the *in vivo* beating heart[6].

Intracoronary injection of contrast media in conjunction with echocardiography has the potential of providing information about transmural differences in vascular volume[7]. It shows a good correlation when compared with myocardial flow by radioactive microspheres, and it is able to detect a reduction in coronary flow reserve (as the dipyridamole/baseline flow ratio) in the subendocardium of dogs during coronary ligation[8]. Thanks to contrast echocardiography, Rovai et al.[9] demonstrated that coronary blood flow is primarily subendocardial in distribution during diastole and subepicardial during systole.

Using myocardial contrast echocardiography and atrial pacing, Lim et al.[10] showed that, in patients with coronary artery stenosis, pacing stress test induced a decrease in the subendocardial/subepicardial gray-level ratio only in segments supplied by a stenotic coronary artery, thus suggesting the occurrence of subendocardial myocardial ischemia.

Another study demonstrated the ability of myocardial contrast echocardiography in detecting transmural differences in videointensity and their changes induced by papaverine after coronary angioplasty[11]: the subendocardial/subepicardial gray-level ratio decreased after papaverine infusion in the segments supplied by a stenotic coronary artery, but increased in the same segments after coronary angioplasty.

Some other studies, using intracoronary injected contrast agents during conventional echocardiography, have shown controversial results for the demonstration of the ability of contrast echo to assess the transmural (subendocardial/subepicardial) distribution of blood flow[12,13]: myocardial opacification in subendocardial, mid and subepicardial layers weakly correlated with the corresponding regional blood flow measured by microspheres in normal conditions and during myocardial ischemia.

The advent of new resonating echocontrast agents and of the second harmonic ultrasound technology will probably ameliorate the resolution of the images and will allow the separate measurement of contrast opacification in the different myocardial layers.

Myocardial thickening with intramyocardial references. When investigating myocardial contractility of different transmural layers the anatomic arrangement of myocardial fibers has to be taken into account. Subendocardial and subepicardial fibers are mostly parallel to the long axis of the left ventricle and the midwall fibers are oriented circumferentially; during systole all these fibers contract and both the longitudinal axis and transversal axis of the left ventricle shorten; therefore ventricular wall thickness increases[2,3].

The first technique employed to measure transmural heterogeneity of contractility and ischemia utilized pairs of sonomicrometers implanted in the different layers of the ventricular wall, with the ultrasonic crystals aligned to the anatomic direction of myocardial fibers. The fiber shortening between each pair of sonomicrometers was then calculated. The superficial pair, aligned along the longitudinal fiber direction in the subepicardium, measured the ventricular long-axis shortening[14,15]. The inner-intermediate pair, aligned along the circumferential direction in the subendocardium-mid-myocardium, investigated the circumferential shortening. Thanks to this first method it was possible to demonstrate that the subendocardial fibers contract much more than the subepicardial ones[14,16,17].

A similar technique, single epicardial pulsed Doppler transducer, measured myocardial contraction velocity in different myocardial layers evaluating the movements of a single point of myocardial tissue. This quicker and less invasive technique showed a good correlation with the paired sonomicrometer technique and confirmed the previous findings of greater subendocardial contribution to overall myocardial thickening[18]. Similar results were obtained with intramyocardial pressure measurements in different transmural layers.

Also traditional M-mode echocardiography with an epicardial transducer has been used in conjunction with sutures placed at various distances within myocardial

thickness to measure the distance between myocardial layers along the cardiac cycle[19,20].

Integrated backscatter. The integrated backscatter tissue characterization studies the structural and functional state of the myocardium, providing quantitative indexes of its physical properties and variations along the cardiac cycle. Tissue characterization is also capable of evaluating different layers of myocardial transmural thickness, while conventional echocardiography cannot differentiate the relative role of the different layers of the myocardium.

With tissue characterization technique the wavelength of the incident beam is much greater than the boundaries between the different components of the tissue, so ultrasounds become scattered in a multidirectional phenomenon[21].

The built-in software, currently available for tissue characterization, measures the waves that are scattered and redirected back to the same transducer. This integrated backscatter is calculated by processing the signal directly in the time domain, producing real-time integrated backscatter two-dimensional images. It is then possible to select a region of interest to calculate the integrated backscatter values in a particular segment or layer of the myocardium.

The integrated backscatter value changes during contraction and relaxation of the normal myocardium. These cyclic variations of integrated backscatter (IBScv) are consistent and reproducible along the cardiac cycle with higher values near end-diastole and lower values at end-systole[22] (Fig. 1). These IBScv are the expressions of regional intramural myocardial contractile performance[23,24] and are related to contractility[25], but are not directly dependent on inotropic state changes[23,24]. In fact, during stress-induced myocardial ischemia the IBScv are blunted and, after stress interruption, they recover earlier than myocardial thickening[26]. A similar dissociation between blunting of conventional video image gray-level cyclic variations and segmental dysfunction, has been observed in patients studied immediately after coronary artery occlusion during angioplasty or during intraoperative ischemia[27,28]. It is also possible to place the region of interest in different transmural layers, obtaining a selective behavior of IBScv correlated with myocardial contraction in that particular myocardial layer[29,30].

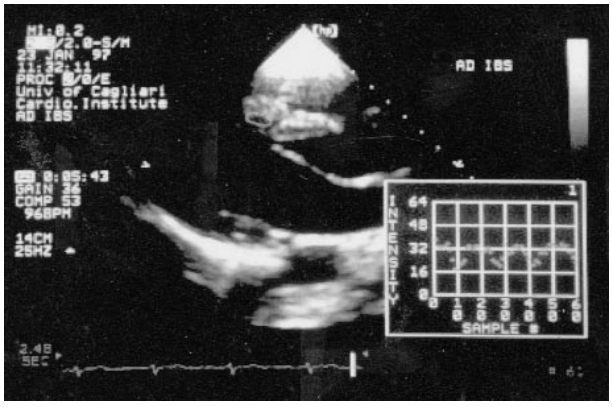


Figure 1. *Example of integrated backscatter cyclic variations calculated by means of transthoracic echocardiography in the subendocardium of a normal patient. The region of interest is the oval placed at the level of anterior interventricular septum. The time/intensity graph is drawn in the* lower right *corner.*

However, at the moment the tissue characterization technology shows some limitations: the method is time consuming and it gives results in an unattractive format (a videodensitometric systo-diastolic cyclic variation graph). Moreover, due to anisotropy, it is necessary to have an insonification angle (of the ultrasonic beam) perpendicular to the myocardial segment to obtain precise IBScv curves. This problem can be solved thanks to the transesophageal approach[31].

Tissue Doppler imaging. Another ultrasound technique which can be used to detect transmural heterogeneity is tissue Doppler imaging. This technique derives from Doppler ultrasound, conventionally implemented in most echocardiographic equipment. Conventional Doppler echocardiography detects any signal in the ultrasound beam with high velocity despite a low intensity; this is due to filters that remove signal with low velocity and high amplitude. The tissue Doppler imaging system enhances the wall motion signal suppressing blood flow: in fact it removes all the low amplitude and high velocity signal (from blood flow)[32].

Myocardial wall movement can be displayed in colors similar to those represented in conventional color Doppler: red for the wall moving toward the transducer, blue for the wall moving away from the transducer. Several studies showed that the velocity measured by color tissue Doppler imaging correlated well with the real velocity of rotational moving phantoms[33,34]. Also in the clinical setting, there was a good correlation of Doppler tissue velocity with conventional M-mode echocardiography[35].

This technique is largely limited due to the dependence of myocardial Doppler velocity on heart translocation in the thoracic cage[32,36]. However, a great potential of this technique is the opportunity to obtain information on the velocity in the different transmural layers. It is possible to detect along the myocardial wall represented with M-mode a range of brightness of the color from the subendocardium to the subepicardium, indicating a myocardial velocity gradient .

This myocardial velocity gradient can be calculated by tissue Doppler imaging in normal subjects and in different pathologic myocardial states, involving left ventricular myocardial function.

Nuclear magnetic resonance. Recently it has been possible to determine the contribution of different transmural layers to myocardial contraction in normal subjects and in patients with acute myocardial infarction also with the use of cardiac magnetic resonance tagging[37-40].

Tagged images are obtained using a breath-hold multiple phase-encoded gradient-echo method. The tag pattern is produced creating tags every 7 mm in a diagonal orientation. Image planes 7 mm thick are created spanning the entire left ventricular cavity from base to apex.

The advent of fast magnetic resonance imaging permits the acquisition of magnetic resonance tomograms during a breath hold. This creates the possibility of studying the influence of paramagnetic contrast agents on myocardial signal intensity with much greater temporal resolution than achieved before by spin-echo magnetic resonance imaging. This major resolution can be useful when studying myocardial contraction in the ischemic myocardium and provides spatial information on myocardial perfusion and myocardial tissue damage additional to that supplied by thallium scintigraphy. Given the time course of regional enhancement patterns after contrast bolus administration, the temporal and topographic features of hypoenhanced zones within the injured territory are best characterized by fast imaging techniques. The demonstration of myocardial contractile heterogeneity and the possibility of its non-invasive evaluation in human beings have potential clinical implications to evaluate the transmural extent a) of myocardial infarction, b) of myocardial viability, and c) of acute myocardial ischemia during stress test. Breath-hold tagged magnetic resonance imaging is safe and effective in detecting regional and transmural changes in function, also during incremental infusion of dobutamine to evaluate transmural contractility in patients without myocardial wall abnormalities. This technique has shown heterogeneous enhancement patterns within the infarcted territory in patients with acute myocardial infarction; moreover, it was able to demonstrate a reduced remote non-infarcted region dysfunction in patients 1 week after anterior myocardial infarction. With this study magnetic resonance tagging demonstrated the feasibility of characterizing regional intramyocardial function in a quantitative and topographic manner[41].

Magnetic resonance tagging could differentiate subendocardial from transmural myocardial infarctions; this finding permits the identification of clinical situations at different risk of infarct expansion (the major cause of left ventricular remodeling[42]), and the functional relevance of recovery of viable subepicardium for the improvement in ejection fraction. In fact there was a 3-month ejection fraction improvement in patients showing subendocardial and subepicardial layer viability at positron emission tomography studies, but there was no improvement in patients with absence of myocardial viability. A good correlation between recovery in subepicardial layers (viable segments) and recovery in global ejection fraction was also shown.

During transient stress-induced myocardial ischemia, transmural heterogeneity has not been studied with magnetic resonance tagging, mainly because of the complexity of this technique. Therefore, the data available so far during acute myocardial ischemia are those obtained with direct invasive experimental measurements in dogs[20] or with transesophageal integrated backscatter in humans.

Clinical implications of transmural heterogeneity

The contribution of different transmural layers to myocardial contraction is difficult to study, but can provide very important information for the diagnostic and prognostic evaluation of ischemic and non-ischemic cardiac pathologies. In fact myocardial ischemia and tissue necrosis progress along the different transmural layers, from the subendocardium to the subepicardium, creating the necrosis wavefront phenomenon . This phenomenon has important clinical implications, but is very difficult to recognize because invasive techniques cannot be used to calculate the transmural differences of myocardial contraction and ischemia.

At present it is possible to use cardiac ultrasounds and magnetic resonance imaging to obtain information about the heterogeneity of myocardial contraction in the human heart. This information can be applied especially to the evaluation of cardiac ischemia and myocardial infarction, but also to several other pathologies of the myocardium in its different transmural layers.

Transmural heterogeneity in normal subjects. The heterogeneity in myocardial contraction has been shown in animal studies by using epicardial echocardiography and intramyocardial references (crystals, sutures, etc.). These techniques demonstrated that, also during the increase in contractility due to physical exercise or inotropic dobutamine stimulus, the subendocardium contributes to most of wall thickening, accounting for 83% of total systolic thickening.

In normal human beings a transmural heterogeneity of myocardial contraction at rest has recently been determined with the use of cardiac magnetic resonance tagging and ultrasound techniques.

Using tissue characterization by IBScv, several experimental studies evaluated transmural heterogeneity of contraction, but few of them pointed out how the IBScv behave in the subendocardium and subepicardium. To measure the behavior of integrated backscatter in the different layers, the region of interest has to be placed at different depths in the myocardial wall: at first in the subepicardium and then in the subendocardium. At rest there were no differences in absolute values of backscatter, but IBScv were higher in the subendocardium than in the subepicardium. In two studies the endo/epicardial gradient was approximately 2 dB, and the values measured in different layers in the two studies were highly different.

Wickline et al.[23] demonstrated that this transmural gradient does not change with inotropic state modifications produced by infusion of isoproterenol (β stim-

ulus) or β-blockers. Similar data have been obtained in humans by our[30] and other groups[43].

Several limitations can affect the calculation of IBScv in different layers of the myocardium: a) the spatial definition of echocardiography that limits the sensitivity of the sample volume, mostly during transthoracic studies, b) the intrinsic cardiac movements that have different orientation in the different myocardial layers (twisting, longitudinal or circumferential shortening).

The heterogeneity of myocardial contraction has also been demonstrated by the color analysis of tissue Doppler imaging both in M-mode and in two-dimensional imaging[44]. During systole and diastole there is a transmural heterogeneity in colors, reflecting the differences in velocity of contraction: the subendocardium looks brighter (a greater contraction velocity) while the subepicardium darker (lower contraction). This analysis has also been confirmed by computer measurement of the color gradient, comparing the velocity with the rate of wall thickening from conventional M-mode. This myocardial velocity gradient changes during the different phases of the cardiac cycle, but it is present in both systolic and diastolic phases in normal subjects.

Acute ischemia and subendocardial contraction. The basal greater subendocardial blood flow (normally about 20% greater than that of subepicardial muscle) appears to be secondary to the lower subendocardial vascular tone (because of a greater oxygen consumption). Consequently, the coronary reserve (the ability to increase local blood flow thanks to a reduction in microvascular tone) is physiologically reduced in the subendocardium. In the presence of an intermediate (> 50 and 70%) coronary artery stenosis the subendocardial resistance vessels become fully dilated and their perfusion becomes pressure dependent. Redistribution of flow from the subendocardium to the subepicardium develops as the transstenotic pressure gradient increases (> 70%) and pressure distal to the stenosis falls and the subendocardium is the first layer to be vulnerable to ischemia. During physical activity, coronary blood flow rises to meet the increase in myocardial oxygen demand, leading to an increase in trans-stenotic pressure gradient and a fall in the distal perfusion pressure, resulting in redistribution of blood flow from the subendocardium toward the subepicardium. The fall in intraluminal pressure may lead to a collapse of the vessel at the level of the obstruction, thereby increasing the degree of stenosis.

Due to this low subendocardial flow reserve, the subendocardium is the first layer to be vulnerable to ischemia when an acute reduction in coronary blood flow occurs. In dogs the effects of a coronary artery progressive occlusion[45-48] and/or physical exercise performed in the presence of a significantly narrowed coronary artery[49] have been studied. The subendocardium suffered a severe reduction in perfusion and kinesis (detected with intramyocardial electronic crystals), but only a trivial reduction of both factors can be detected in the subepicardium. Therefore, during partial coronary occlusion the reduction in flow is limited to the inner layers and contractility is mainly depressed in the subendocardium. These results were also confirmed by pulsed Doppler[50] and the intramyocardial suture technique described above[51].

Also the ultrasonic backscatter technique can detect myocardial ischemia. IBScv are severely blunted during acute myocardial ischemia, obtained with coronary artery ligation[52]. Such a behavior has also been found in coronary artery disease patients, during stress-induced myocardial ischemia[53].

IBScv blunting during myocardial ischemia is related to the duration of coronary artery occlusion. If reperfusion is achieved very early (after 5 min occlusion), IBScv recovery is immediate and complete; if reperfusion is delayed (after 30 min), the recovery period lasts several hours[54]. Moreover, IBScv recovery during the reperfusion phase is much faster and complete than other conventional regional left ventricular functional parameters (myocardial thickening, systolic shortening, wall motion score index)[55].

The only data on transmural gradient in human beings have been obtained from a transesophageal study of 37 patients with suspected coronary artery disease during a stress test with atrial pacing[30]. We divided the 37 patients into two groups: one with normal coronary arteries and the other with significant coronary stenosis. In both groups at rest there was a 2 dB transmural gradient of integrated backscatter, equal to the one found in dogs.

Moreover, IBScv are capable of detecting ischemia in different transmural layers (Fig. 2). Acute occlusion of the left anterior descending coronary artery caused a reduction of IBScv in the subendocardial layer, abolishing the basal transmural gradient. During the same experiment there was also a depression of subendocardial blood flow. A similar behavior of IBScv in the subendocardium and subepicardium has been found during myocardial ischemia induced by atrial pacing stress test in patients with suspected coronary artery disease compared to a control group without significant coronary disease. At rest IBScv were greater in the subendocardium than in the subepicardium (transmural gradient of

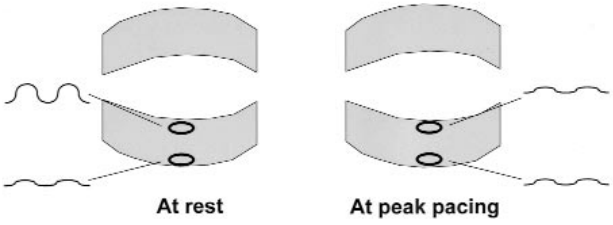
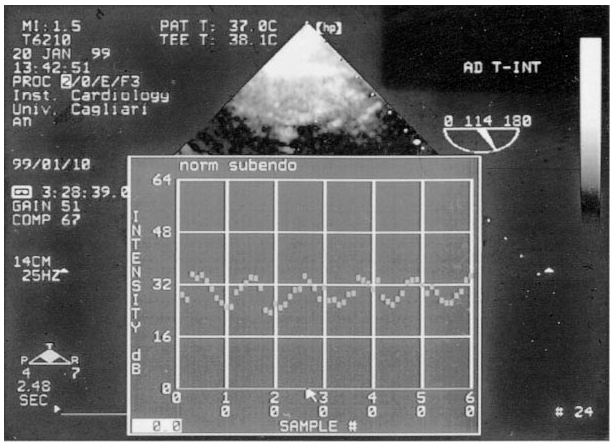


Figure 2. *Scheme of the behavior of integrated backscatter cyclic variations in different layers at rest and during atrial pacing stress test. In the* left panel *there is a transmural heterogeneity gradient of integrated backscatter cyclic variations favoring the subendocardium. In the* right panel *(at peak pacing stress stimulus) there is a blunting in subendocardial integrated backscatter cyclic variations and disappearance of the subendocardial/subepicardial transmural gradient.*

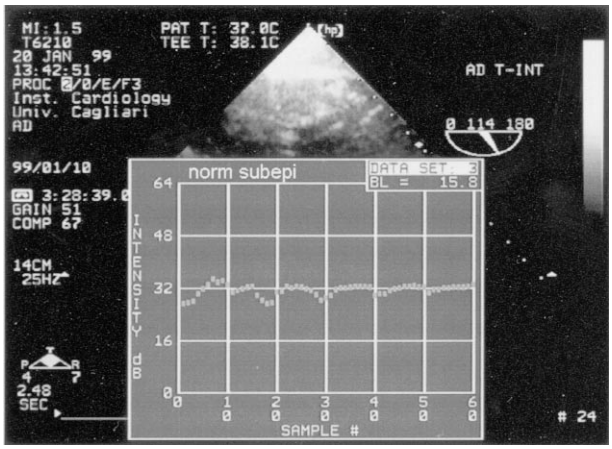
IBScv) both in myocardial segments supplied by a stenotic or a non-stenotic coronary artery. At peak transesophageal atrial pacing the IBScv myocardial segments supplied by a stenotic coronary artery showed a reduction in subendocardial IBScv (Fig. 3) and no reduction in subepicardial IBScv (Fig. 3), abolishing the transmural myocardial gradient (Fig. 4). In normal subjects there were no changes in IBScv in both the subendocardium and subepicardium; this was the first demonstration of subendocardial stress-induced ischemia in human beings[30].

Myocardial infarction transmural heterogeneity. Thanks to an early and more complete reperfusion in the acute phase of myocardial infarction (due to thrombolysis or primary angioplasty), myocardial salvage is frequently achieved.

With a progressive increase in ischemia duration the necrosis progresses from the endocardium to the epicardium[56-58] and this wavefront phenomenon is not dependent on the collateral circulation. This sequence of



A



B

Figure 3. A: *example of integrated backscatter cyclic variations in the subendocardium of a normal segment at peak pacing stress test (150 b/min). There is a value of peak-to-nadir integrated backscatter cyclic variations of 11.4 dB.* B: *example of integrated backscatter cyclic variations in the corresponding subepicardium at peak pacing stress test. There is a much lower value of peak-to-nadir integrated backscatter cyclic variations (4.3 dB).*

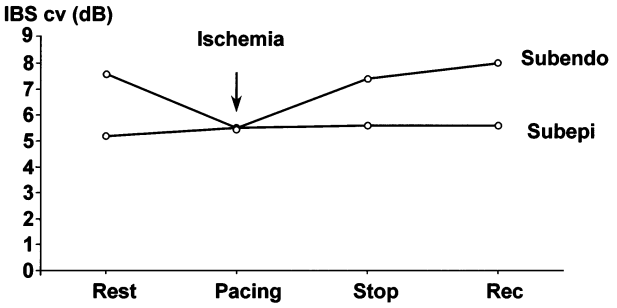


Figure 4. *Graph showing the behavior of integrated backscatter cyclic variations (IBScv) in different transmural layers of the group of ischemic myocardial segments of patients with coronary artery disease. Adapted from Colonna et al.[30].*

events cannot be demonstrated with conventional echocardiography, which evaluates myocardial wall thickening (by means of segmental myocardial wall motion analysis). This is due to the necrosis transmural threshold : in fact, when the histological necrosis affects less than 20% of myocardial wall thickness, a proportional reduction in myocardial thickening occurs; if the necrosis exceeds the threshold of 20%, a total absence of myocardial thickening is observed[59]. Therefore a subendocardial necrosis involving the inner 30% of the myocardial layers produces a wall motion akinesia similar to that of a transmural myocardial infarction.

The clinical importance of the above data is underlined by some interesting recent findings. In two groups of dogs with comparable infarct size (measured as grams of necrotic tissue), the group with a greater infarct transmurality showed larger diastolic and systolic volumes and a greater regional and global left ventricular dysfunction after 6 weeks (left ventricular remodeling). In this group of dogs there was also a greater number of complications like left ventricular thrombus, arrhythmias and deaths[42].

Moreover, myocardial infarction causes also a disappearance in myocardial velocity gradient at tissue Doppler imaging. In fact myocardial velocity gradient was significantly lower in infarcted regions both in the interventricular septum and in posterior wall when compared to non-infarcted regions or to normal myocardial wall in control subjects.

At rest a poor correlation between histological infarct size and myocardial thickening has been observed; this correlation remarkably improved during dobutamine infusion[60]. Dobutamine did not improve the correlation when the segments were supplied by a coronary artery with residual stenosis. This correlation improvement was explained by the fact that dobutamine infusion increased contractility in the viable segments with a basal akinesia, but without transmural necrosis (viability in the mid-superficial myocardium). In the presence of residual stenosis of the infarct-related artery, dobutamine infusion did not ameliorate myocardial contractility probably because of a biphasic response to dobutamine of the stunned myocardium, thus correlation remained

poor[60,61]. This finding marks the importance of myocardial salvage (even isolated subepicardial salvage) to limit the post-infarction cavity expansion.

Transmural heterogeneity in post-ischemic stunning recovery. Similarly to the ischemia wavefront also myocardial post-ischemic stunning is heterogeneous, being more intense in subendocardial layers. Myocardial contractility has been invasively studied in three different transmural layers after a 15 min coronary artery occlusion followed by reperfusion. Post-ischemic myocardial contractility recovery was delayed in the subendocardial layer compared to the other two.

This transmural heterogeneity in post-ischemic stunning recovery could be explained by a more intense subendocardial ischemia, with a delayed subendocardial microvascular reperfusion (microcirculatory subendocardial stunning)[62], or with a greater subendocardial overload of reperfusion toxic metabolites (free radicals, granulocytes).

Tissue characterization has potential in detecting stunned myocardium. After acute myocardial infarction, only in patients with an open infarct-related artery (with a high probability of post-infarction myocardial stunning) IBScv recovered from early (within 24 hours of chest pain onset) to late (pre-discharge) studies, while wall motion score index did not[63].

Moreover, in a dog model of ischemia-reperfusion IBScv recovered earlier than myocardial thickening; similarly, in a group of patients with coronary artery disease undergoing stress-induced myocardial ischemia, after atrial pacing interruption there was an immediate (3 s) IBScv recovery, while myocardial thickening recovered after 2 min[30]. This delayed IBScv recovery can be due to post-ischemic stunning and can be studied along the different transmural layers.

In fact, a possible explanation of this temporal dissociation between IBScv and myocardial thickening is related to the subendocardial stunning. Since the subendocardial layer is responsible for a great part of the overall transmural contraction, the ischemic loss of its contribution can provoke a reduction in overall transmural thickening with two-dimensional echocardiography. Therefore, immediately after coronary ligation release or stress interruption the mid-outer myocardium (not important for the overall myocardial thickening) has already recovered and there are normal IBScv, because they are calculated in all the different myocardial layers[64].

Transmural heterogeneity in other clinical conditions. Important findings about transmural heterogeneity have been demonstrated in clinical conditions different from ischemic heart disease. Mostly, new echocardiographic techniques have been used (the integrated backscatter with absolute values for increases in myocardial fibrosis, or magnitude of the cyclic variations and tissue Doppler imaging) to evaluate transmural heterogeneity in clinical conditions as hypertrophic[43,65] and dilated[66] cardiomyopathy, initial acute cardiac allograft rejection[67], cardiac dysfunction from iron overload[68] or cardiac muscle changes in diabetic patients without overt heart disease[69].

Diagnosis of transmural heterogeneity in hypertrophic cardiomyopathy has been accomplished by the analysis of the transmural trend of the absolute myocardial integrated backscatter and tissue Doppler imaging. The transmural gradient in integrated backscatter did not correlate with ventricular wall thickness in patients with hypertrophic cardiomyopathy either in the septum or posterior wall. Conversely most patients in the hypertrophic cardiomyopathy group showed an altered integrated backscatter gradient in the septum, and about half of them in the posterior wall.

By means of tissue Doppler imaging the intramural function of patients with left ventricular hypertrophy of different etiologies can be assessed. The analysis and quantitation of myocardial velocity gradient, in fact, showed a significant reduction in transmural myocardial velocity gradient in patients with hypertrophic cardiomyopathy, while patients with left ventricular hypertrophy secondary to hypertension had values comparable to normal subjects. A similar transmural reduction was also observed in diastolic velocity behavior. Thus, myocardial velocity gradient measurement has a potential in the identification of patients with hypertrophic cardiomyopathy within the group of hypertrophies of different origins.

Also in dilated cardiomyopathy with impairment of left ventricular contraction, the endo-epicardial transmural velocity gradient at tissue Doppler was reduced. In both antero-septal and posterior wall, the myocardial velocity gradient demonstrated to be significantly reduced in patients with dilated cardiomyopathy compared to normal subjects. This reduction in velocity gradient is evident in all systolic cardiac phases, and during diastole only in the atrial contraction phase.

Important changes in the subendocardium occur also in acute cardiac rejection. Several studies showed that moderate and severe but not mild acute cardiac rejection is associated with significant acoustic changes. The use of ultrasonic tissue analysis with two-dimensional integrated backscatter allows for a reliable identification of mild acute cardiac rejection, with an end-diastolic backscatter increase in both the posterior wall and interventricular septum. This significant myocardial acoustic change occurs only in the subendocardium and is independent of changes in contractile performance.

Moreover, early muscular dysfunction can be detected in secondary cardiomyopathy such as cardiac dysfunction in thalassemic patients without clinical signs of heart failure and diabetic patients without overt heart disease abnormalities. The data obtained by myocardial tissue characterization with integrated backscatter in these studies, even without specific information on different transmural layers, demonstrated that myocardial re-

flectivity is abnormally increased probably due to iron overload, fibrosis and histopathological alterations. These quantitative abnormalities can be detected within the posterior wall and the septum when conventional echocardiography cannot clearly differentiate them from normal segments.

Conclusions

Nowadays, the most important goal of myocardial infarction treatment is early myocardial salvage. With the advent of effective reperfusion therapeutic strategies (thrombolysis, percutaneous transluminal coronary angioplasty) in the acute phase of myocardial infarction, myocardial salvage is frequently achieved. Thus, a correct diagnosis of the extension of the myocardial necrosis cannot ignore its transmural wavefront development.

Due to this necrosis wavefront progression, the myocardial salvage is more easily obtained in the subepicardial layer. Thus, after myocardial ischemia followed by reperfusion, myocardial infarction can manifest different anatomic and/or functional characteristics (subendocardial infarction). The salvage of the subepicardial layer does not give direct information on overall myocardial thickening but is the major determinant of overall left ventricular dysfunction and size, this latter being the most powerful predictor of prognosis after myocardial infarction.

Wall motion akinesia can be caused by a variable extension of non-contracting, scarred myocardium (in the presence of successful and timely reperfusion scar is usually confined to subendocardial layers); an important role is played by the extent of salvaged myocardium beyond the scar. Such myocardium can show different functional status (alive and normo-contracting; alive but stunned and temporarily dysfunctioning; alive but chronically dysfunctioning). These different anatomic and functional intramyocardial patterns represent the basis for different functional outcome of regional and hence global left ventricular function.

New non-invasive technologies, based on ultrasounds and magnetic resonance, can detect subendocardial contractility and the transmural extension of myocardial damage. Although it is still necessary to investigate this phenomenon, these new techniques give fundamental information in an unexplored field of cardiac disease diagnosis. The impact of a better understanding of cardiac diseases and their pathophysiology will give us more opportunities to appropriate diagnosis and treatment of cardiac patients.

Acknowledgments

The authors would like to thank Barbara Hildenbrand for her assistance in the preparation of the manuscript.

References

1. Domenech RJ, Hoffman JIE, Noble MI, Saunders KB, Henson JR, Subijanto S. Total and regional coronary blood flow measured by radioactive microspheres in conscious and anesthetized dogs. Circ Res 1969; 25: 581-96.
2. LeWinter MM, Kent RS, Kroener JM, Carew TE, Cowell JW. Regional differences in myocardial performances in the left ventricle of the dog. Circ Res 1975; 37: 191-9.
3. Sabbah HN, Marzilli M, Stein PD. The relative role of subendocardium and subepicardium in left ventricular mechanics. Am J Physiol 1981; 240: H920-H926.
4. Brazier J, Cooper N, Buckberg G. The adequacy of subendocardial oxygen delivery: the interaction of determinants of flow, arterial oxygen content and myocardial oxygen need. Circulation 1974; 49: 968-77.
5. Hoffman JIE. Determinants of prediction of transmural myocardial perfusion. Circulation 1978; 58: 381-91.
6. Jayaweera AR, Edwards N, Glasheen WP, Villanueva FS, Abbott RD, Kaul S. In vivo myocardial kinetics of air-filled albumin microbubbles during myocardial contrast echocardiography. Comparison with radiolabeled red blood cells. Circ Res 1994; 74: 1157-65.
7. Chandwaney RH, Zajac E, Saldivar J, et al. Contrast echocardiography displays increased subendocardial perfusion after nitroglycerin administration. J Am Soc Echocardiogr 1997; 10: 210-4.
8. Cheirif J, Zoghbi WA, Bolli R, O Neill PG, Hoyt BD, Quinones MA. Assessment of regional myocardial perfusion by contrast echocardiography. II: Detection of changes in transmural and subendocardial perfusion during dipyridamole-induced hyperemia in a model of critical stenosis. J Am Coll Cardiol 1989; 14: 1555-65.
9. Rovai D, L Abbate A, Lombardi M, et al. Nonuniformity of the transmural distribution of coronary blood flow during the cardiac cycle. In vivo documentation by contrast echocardiography. Circulation 1989; 79: 179-87.
10. Lim YJ, Nanto S, Masuyama T, et al. Visualization of subendocardial myocardial ischemia with myocardial contrast echocardiography in humans. Circulation 1989; 79: 233-44.
11. Perchet H, Dupouy P, Duval-Moulin AM, et al. Improvement of subendocardial myocardial perfusion after percutaneous transluminal coronary angioplasty: a myocardial contrast echocardiography study with correlation between myocardial contrast reserve and Doppler coronary reserve. Circulation 1995; 91: 1419-26.
12. Rovai D, Ghelardini G, Lombardi M, et al. Myocardial washout of sonicated iopamidol does not reflect the transmural distribution of coronary blood flow. Eur Heart J 1993; 14: 1072-8.
13. Kaul S, Jayaweera AR, Glasheen WP, Villanueva FS, Gutgesell HP, Spotnitz WD. Myocardial contrast echocardiography and the transmural distribution of flow: a critical appraisal during myocardial ischemia not associated with infarction. J Am Coll Cardiol 1992; 20: 1005-16.
14. Gallagher KP, Osakada G, Hess OM, Koziol JA, Kemper WS, Ross J Jr. Subepicardial segmental function during coronary stenosis and the role of myocardial fiber orientation. Circ Res 1982; 50: 352-9.
15. Weintraub WS, Hattori S, Agrawal JR, Bodenheimer MM, Banka VS, Helfant RH. The relationship between myocardial blood flow and contraction by myocardial layer in the canine left ventricle during ischemia. Circ Res 1981; 48: 430-8.
16. Gallagher KP, Stirling MC, Choy M, et al. Dissociation between epicardial and transmural function during acute myocardial ischemia. Circulation 1985; 71: 1279-91.
17. Gallagher KP, Osakada G, Matsuzaki M, Miller M, Kemper WS, Ross J Jr. Nonuniformity of inner and outer systolic wall thickening in conscious dogs. Am J Physiol 1985; 249: H241-H248.

18. Hartley CJ, Latson LJ, Michael LH, Seidel CL, Lewis RM, Entman ML. Doppler measurement of myocardial thickening with a single epicardial transducer. Am J Physiol 1983; 245: H1066-H1072.
19. Zhu WX, Myers ML, Hartkeh CJ, Roberts R, Bolli R. Validation of a single crystal for measurement of transmural and epicardial thickening. Am J Physiol 1986; 251: H1045-H1055.
20. Myers JH, Stirling MC, Choy M, Buda AJ, Gallagher KP. Direct measurement of inner and outer wall thickening dynamics with epicardial echocardiography. Circulation 1986; 74: 164-72.
21. PØrez JE, Miller JG, Wickline SA, et al. Quantitative ultrasonic imaging: tissue characterization and instantaneous quantification of cardiac function. Am J Cardiol 1992; 69: 104H-111H.
22. Madaras EI, Barzilai B, PØrez JE, Sobel BE, Miller JG. Changes in myocardial backscatter throughout the cardiac cycle. Ultrason Imaging 1983; 5: 229-39.
23. Wickline SA, Thomas LJ III, Miller JG, Sobel BE, PØrez JE. The dependence of myocardial ultrasonic backscatter on contractile performance. Circulation 1985; 72: 183-92.
24. Wickline SA, Thomas LJ III, Miller JG, Sobel BE, PØrez JE. A relationship between ultrasonic integrated backscatter and myocardial contractile function. J Clin Invest 1985; 76: 2151-60.
25. Sagar KB, Pelc L, Rhyne TL, Wann LS, Warltier DC. Influence of heart rate, preload, afterload, and inotropic state on myocardial ultrasonic backscatter. Circulation 1988; 77: 478-83.
26. Iliceto S, Galiuto L, Colonna P, Napoli VF, Rizzon P. Effects of atrial pacing stress test on ultrasonic integrated backscatter cyclic variations in normals and in patients with coronary artery disease. Eur Heart J 1997; 18: 1590-8.
27. Picano E, Faletra F, Marini C, et al. Increased echodensity of transiently asynergic myocardium in humans: a novel echocardiographic sign of myocardial ischemia. J Am Coll Cardiol 1993; 21: 199-207.
28. Pingitore A, Kozakova M, Picano E, Paterni M, Landini L, Distante A. Acute myocardial gray intensity changes detected by transesophageal echocardiography during intraoperative ischemia. Am J Cardiol 1993; 72: 465-6.
29. Sagar KB, Rhyne TL, Warltier DC, Pelc L, Wann S. Intramyocardial variability in integrated backscatter: effect of coronary occlusion and reperfusion. Circulation 1987; 75: 436-42.
30. Colonna P, Montisci R, Galiuto L, Meloni L, Iliceto S. Effects of acute myocardial ischemia on intramyocardial contraction heterogeneity: a study performed with ultrasound integrated backscatter during transesophageal atrial pacing. Circulation 1999; 100: 1770-6.
31. Eaton MH, Lappas D, Waggoner AD, PØrez JE, Miller JG, Barzilai B. Ultrasonic myocardial tissue characterization in the operating room: initial results using transesophageal echocardiography. J Am Soc Echocardiogr 1991; 4: 541-6.
32. Sutherland GR, Steward MJ, Grounstroem KWE, et al. Color Doppler myocardial imaging: a new technique for the assessment of myocardial function. J Am Soc Echocardiogr 1994; 7: 441-58.
33. Fleming AD, Xia X, McDicken WN, Sutherland GR, Fenn L. Myocardial velocity gradient by Doppler imaging. Br J Radiol 1994; 67: 679-88.
34. Uematsu M, Miyatake K, Tanaka N, et al. Myocardial velocity gradient as a new indicator of regional left ventricular contraction: detection by a two-dimensional tissue Doppler imaging technique. J Am Coll Cardiol 1995; 26: 217-23.
35. Miyatake K, Yamagishi M, Tanaka N, et al. New method for evaluating LV wall motion by color-coded tissue Doppler imaging (TDI): in vitro and in vivo studies. J Am Coll Cardiol 1995; 25: 717-24.
36. Gorcsan J III, Gulati VK, Mandarino WA, Katz WE. Color coded measures of myocardial velocity throughout the cardiac cycle by tissue Doppler imaging to quantify regional left ventricular function. Am Heart J 1996; 131: 1203-13.
37. Bogaert J, Maes A, Van de Werf F, et al. Functional recovery of subepicardial myocardial tissue in transmural myocardial infarction after successful reperfusion. Circulation 1999; 99: 36-43.
38. Lima JA, Judd RM, Bazille A, Schulman SP, Atalar E, Zerhouni EA. Regional heterogeneity of human myocardial infarcts demonstrated by contrast-enhanced MRI: potential mechanisms. Circulation 1995; 92: 1117-25.
39. Power TP, Kramer CM, Shaffer AL, et al. Breath-hold dobutamine magnetic resonance myocardial tagging: normal left ventricular response. Am J Cardiol 1997; 80: 1203-7.
40. Lima JAC, Jeremy R, Guier W, et al. Accurate systolic wall thickening by nuclear magnetic resonance imaging with tissue tagging: correlation with sonomicrometers in normal and ischemic myocardium. J Am Coll Cardiol 1993; 21: 1741-51.
41. Kramer CM, Rogers WJ, Theobald TM, Power TP, Petruolo S, Reichek N. Remote noninfarcted region dysfunction soon after first anterior myocardial infarction, a magnetic resonance tagging study. Circulation 1996; 94: 660-6.
42. Jugdutt BI, Khan MI. Impact of increased infarct transmurality on remodeling and function during healing after anterior myocardial infarction in the dog. Can J Physiol Pharmacol 1992; 70: 949-58.
43. Naito J, Masujama T, Tanouchi J, et al. Analysis of transmural trend of myocardial integrated ultrasound backscatter for differentiation of hypertrophic cardiomyopathy and ventricular hypertrophy due to hypertension. J Am Coll Cardiol 1994; 24: 517-24.
44. Palka P, Lange A, Fleming AD, et al. Age-related transmural peak mean velocities and peak velocity gradients by Doppler myocardial imaging in normal subjects. Eur Heart J 1996; 17: 940-50.
45. Bache RJ, Schwartz JS. Effect of perfusion pressure distal to a coronary stenosis on transmural myocardial blood flow. Circulation 1982; 65: 928-35.
46. Edwards NC, Sinusas AJ, Bergin JD, Watson DD, Ruiz M, Beller GA. Influence of subendocardial ischemia on transmural myocardial function. Am J Physiol 1992; 262: H568-H576.
47. Canty JM, Klocke FJ. Reduced myocardial perfusion in the presence of pharmacologic vasodilator reserve. Circulation 1985; 71: 370-7.
48. Gallagher KP, Matsuzaki M, Koziol JA, Kemper WS, Ross J Jr. Regional myocardial perfusion and wall thickening during ischemia in conscious dogs. Am J Physiol 1984; 247: H727-H738.
49. Homans DC, Sublett E, Lindstrom P, Nesbitt T, Bache RJ. Subendocardial and subepicardial wall thickening during ischemia in exercising dogs. Circulation 1988; 78: 1267-76.
50. Bolli R, Patel BS, Hartley CJ, Thornby JI, Jeroudi MO, Roberts R. Nonuniform transmural recovery of contractile function in stunned myocardium. Am J Physiol 1989; 257: H375-H385.
51. Torry RJ, Myers JH, Adler AL, Liut CL, Gallagher KP. Effects of nontransmural ischemia on inner and outer wall thickening in the canine left ventricle. Am Heart J 1991; 122: 1292-9.
52. Glueck RM, Mottley JG, Miller JG, Sobel BE, PØrez JE. Effect of coronary artery occlusion and reperfusion on cardiac cycle-dependent variation of myocardial ultrasonic backscatter. Circ Res 1985; 56: 683-9.
53. Vitale DF, Bonow RO, Gerundo G, et al. Alterations in ultrasonic backscatter during exercise-induced myocardial ischemia in humans. Circulation 1995; 92: 1452-7.

54. Wickline SA, Thomas LJ III, Miller JG, Sobel BE, PØrez JE. Sensitive detection of the effects of reperfusion on myocardium by ultrasonic tissue characterization with integrated backscatter. Circulation 1986; 74: 389-400.
55. Milunski MR, Mohr GA, Wear KA, Sobel BE, Miller JG, Wickline SA. Early identification with ultrasonic integrated backscatter of viable but stunned myocardium in dogs. J Am Coll Cardiol 1989; 14: 462-71.
56. Homans DC, Pavek T, Laxson DD, Bache RJ. Recovery of transmural and subepicardial wall thickening after subendocardial infarction. J Am Coll Cardiol 1994; 24: 1109-16.
57. Reimer KA, Lowe JE, Rasmussen MM, Jennings RB. The wavefront phenomenon of ischemic cell death. I: Myocardial infarct size vs duration of coronary occlusion in dogs. Circulation 1977; 56: 786-94.
58. Lowe JE, Cummings RG, Adams DH, Hullryde EA. Evidence that ischemic cell death begins in subendocardium independent of variations in collateral flow or wall tension. Circulation 1988; 68: 190-202.
59. Lieberman A, Weiss JL, Judgutt BI, et al. Two-dimensional echocardiography and infarct size relationship of regional wall motion and thickening to the extent of myocardial infarction in the dog. Circulation 1981; 63: 739-46.
60. Sklenar J, Ismail S, Villanueva FS, Goodman NC, Glasheen WP, Kaul S. Dobutamine echocardiography for determining the extent of myocardial salvage after reperfusion. An experimental evaluation. Circulation 1994; 90: 1502-12.
61. Kaul S. There may be more to myocardial viability than meets the eye! Circulation 1995; 92: 2790-3.
62. Bolli R, Triana JF, Jeroudi MO. Prolonged impairment of coronary vasodilation after reversible ischemia: evidence for microvascular stunning . Circ Res 1990; 67: 332-43.
63. Milunski MR, Mohr GA, Vered Z, et al. Ultrasonic tissue characterization with integrated backscatter. Acute myocardial ischemia, reperfusion, and stunned myocardium in patients. Circulation 1989; 80: 491-503.
64. Colonna P, Galiuto L, Marangelli V, Iliceto S, Rizzon P. Transmural gradient time-course of integrated backscatter cyclic variations during atrial pacing in patients with coronary artery disease. (abstr) Eur Heart J 1996; 17 (Suppl): 373.
65. Palka P, Lange A, Fleming AD, et al. Assessment of transmural velocity gradients in hypertrophic hearts - a new diagnostic index for hypertrophic cardiomyopathy. (abstr) J Am Coll Cardiol 1996; Special Issue: 21A.
66. Palka P, Lange A, Bouki KP, et al. A new ultrasonic index of left ventricular function-myocardial velocity gradient in dilated cardiomyopathy. (abstr) Circulation 1995; 92: I-277.
67. Angermann CE, Nassau K, Stempfle HU, et al. Recognition of acute cardiac allograft rejection from serial integrated backscatter analyses in human orthotopic heart transplant recipients. Comparison with conventional echocardiography. Circulation 1997; 95: 140-50.
68. Lattanzi F, Bellotti P, Picano E, et al. Quantitative ultrasonic analysis of myocardium in patients with thalassemia major and iron overload. Circulation 1993; 87: 748-54.
69. Perez JE, McGill JB, Santiago JV, et al. Abnormal myocardial acoustic properties in diabetic patients and their correlation with the severity of disease. J Am Coll Cardiol 1992; 19: 1154-62.