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Cyclic variation in myocardial gray level as a marker of viability in man

A videodensitometric study

C. Marini, E. Picano, A. Varga, P. Marzullo, A. Pingitore and M. Paterni

CNR, Institute of Clinical Physiology, Pisa, Italy

Experimental and human studies have shown that a preserved cyclic (diastolic-to-systolic) echoreflectivity variation, assessed by radiofrequency sampling of backscatter signal with non-commercially available prototypes, identifies viability in a myocardial segment with a resting dyssynergy.

The objective of this study was to assess whether a videodensitometric analysis of myocardial gray level variation during cardiac cycle might identify viable but dyssynergic myocardium in a clinical setting. Thirty-four patients with a resting dyssynergy (akinesis in 26, marked hypokinesis in eight) in the septum and/or infero-posterior wall were evaluated by videodensitometry. All echo studies were performed with commercially available instruments in the long axis parasternal view, with quantitative analysis of gray levels performed off-line on digitized images. Segmental wall motion was assessed with a 16 segment model of the left ventricle, each scored from 1, normal, to 4, dyskinetic. A follow-up echo study was obtained in all patients >4 weeks after successful revascularization (in 22 by angioplasty, in 12 by bypass surgery). Two groups of segments were identified: 18 viable segments (contractile improvement of 1 grade or more in resting function after revascularization); 16 necrotic segments (no contractile improvement in resting function after revascularization). The % cyclic variation was higher in viable vs necrotic segments (26 ± 16 vs $1 \pm 13\%$, $P<0.01$), in spite of similar % systolic thickening (5 ± 5 vs $4 \pm 6\%$, P=ns) and end-diastolic thickness (10 ± 2 vs 10 ± 2 mm, P=ns). When individual patient analysis was performed, % cyclic variation was below the 95% confidence limits obtained from normal control regions (n=34; % cyclic variation=38 ± 14) in two out of 18 viable and in 14 out of 16 necrotic segments. A cut-off of $\geq 9.4\%$ cyclic variations in a dyssynergic segment yielded 89% sensitivity and 88% specificity for predicting functional recovery following successful revascularization.

In conclusion, viable dyssynergic myocardial segments show a cyclic gray level variation at rest, which can be detected by simple videodensitometric analysis, much less technologically demanding than radiofrequency backscatter evaluation.
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Key Words: Echocardiography, viability, necrosis, videodensitometry, tissue characterization.

Introduction

In patients with coronary artery disease and impaired left ventricular function, the distinction between ventricular dysfunction due to myocardial fibrosis and post-ischaemic, viable, although dyssynergic, myocardium has important clinical implications. The accepted clinical standard for measuring myocardial viability is nuclear medicine, with either positron emission tomography imaging of fluorodeoxyglucose uptake or thallium-201 scintigraphy[1]. Recently, the possibility of obtaining reliable information on viability by means of echocardiography has emerged. Such a possibility is very attractive in principle, owing to the lower cost, widespread availability, non-ionizing and environment-friendly nature of ultrasonic imaging. Different methods have been proposed in combination with ultrasound, focusing of different physiological variables. Low dose pharmacological stress echocardiography with either dobutamine or dipyridamole identifies viable myocardium through the presence of contractile reserve, which can be recruited, in a basally dyssynergic region, by a cathecolamine or flow-mediated inotropic stimulus[2–3]. Myocardial contrast echocardiography also recognizes viable myocardium, through the physiological variable of preserved perfusion in dyssynergic regions[4].

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Correspondence. Eugenio Picano, MD, PhD, FESC, CNR, Institute of Clinical Physiology, Via Paolo Savi, 8, 56100 Pisa, Italy.

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An additional method for ultrasound detection of myocardial viability is based upon tissue characterization techniques. The hypothesis underlying the tissue characterization approach is that pathological changes in myocardial structure and function result in alterations of the fundamental physical properties of tissue that can be quantified with indexes dependent of ultrasonic backscatter[5]. In particular, it has been shown that physiological contraction and relaxation of the myocardium are paralleled by a cardiac cycle-dependent variation of integrated backscatter that reflects regional, intramural myocardial contractile performance[6–11] and sarcomere length[12]. With reperfusion of a previously occluded vessel, cyclic variation recovers before wall thickening and provides a reliable index with which to assess viability both in the experimental[13] and in the clinical setting[14]. The impact of these findings has been limited, mainly because the employed method was based upon quantitative backscatter imaging which requires modified ultrasound equipment and is technically demanding[5]. Indeed, a myocardial cyclic gray level variation has been found in normal myocardium also by simple videodensitometric analysis of two-dimensional echocardiographic images conventionally obtained by commercially available ultrasound systems[15]. This index is simple to obtain, poses a low technological burden, and shares the same physiological background and the potential domain of application with the more complex cyclic backscatter.

The aim of this study was to establish whether a cyclic echodensity variation — detected by videodensitometry in conventional images acquired by a commercially available ultrasound system could be helpful in recognizing viable, although dysfunctioning, myocardium in a clinical model traditionally ascribed to hibernation: i.e. resting dysfunction in patients with chronic (>3 month) myocardial infarction[16,17]. Accordingly, a quantitative texture analysis on videodata from a standard two-dimensional echocardiogram was applied to 34 patients with a resting dyssynergy and scheduled for revascularization. Since cyclic gray level variation — due to the anisotropic properties of the myocardium — can be consistently and best detected in myocardial regions with fibres oriented perpendicularly to the ultrasonic beam[18,19], we prospectively enrolled only patients with a resting dyssynergy of the septum and infero-posterior was adequately imaged in the parasternal long axis view. The gold standard of viability was the subsequent recovery of myocardial function, documented with a follow-up echo examination obtained after successful revascularization.

Methods

Study patients

We initially considered 45 patients scheduled for coronary revascularization, with angiographically documented coronary artery disease and abnormal resting left ventricular function (wall motion abnormalities in at least 2 adjacent segments by ventriculography and/or 2-dimensional echocardiography). Patients with recent (<3 month) myocardial infarction were not considered. Of these 45, 11 were excluded for: (1) echo examination of insufficient quality for quantitative gray level assessment (n=2); (2) presence of a resting dyssynergy in segments other than the septal and infero-posterior walls, and not visualized in the parasternal long axis view (n=6); (3) inability to complete the study for unsuccessful revascularization (n=3). A subset of 34 patients (30 men and 4 women; age 54 ± 9 years) completed the study. All of them had: (a) an obvious regional dyssynergy (severe hypokinesis or akinesis) adequately imaged in the parasternal long axis view (the projection in which myocardial cyclic gray level changes are most evident and consistent); (b) successful coronary revascularization achieved with either coronary artery bypass surgery (n=12) or percutaneous transluminal coronary angioplasty (n=22). None of these patients manifested clinical evidence of a peri-operative myocardial infarction, and all were considered to have had successful revascularization. The revascularized artery was the left anterior descending in 22 patients, the left circumflex in 16, the right coronary artery in 17, the diagonal branch in two patients. The successful revascularization involved one vessel in 19, two vessels in nine and three vessels in six patients; (c) an echocardiographic follow-up examination obtained at least 4 weeks (42 ± 14 days) after successful coronary revascularization.

Mean left ventricular ejection fraction (at contrast ventriculography) was 46 ± 11%. Seven patients had single, 17 double, and 10 triple vessel disease (>50% diameter reduction of a major coronary vessel).

Baseline echocardiographic examination

Two dimensional echocardiograms were obtained by using commercially available imaging systems (Hewlett-Packard 77020, 77025 or Toshiba Sonolayer FFA270A, 2·5 and 3·5 MHz transducers). The gray scale transfer function was adjusted to be linear for the entire video signal range[15]. Echocardiographic images were recorded on VHS videotape for subsequent playback and analysis. Regional wall motion was assessed according to the recommendations of the American Society of Echocardiography with a 16-segment model[20]. In all studies, segmental wall motion was semiquantitatively graded as follows: normal=1; hypokinetic, marked reduction of endocardial motion and thickening=2; akinetic, virtual absence of inward motion and thickening=3; and dyskinetic, paradoxic wall motion away from the centre of the left ventricle in systole=4. Baseline echocardiography was obtained before coronary angioplasty for cardiac surgery and regional wall motion analysis was performed in the parasternal long-axis view in all patients.

Videodensitometric analysis of the baseline echocardiographic examination

Images of the baseline echocardiographic examination corresponding to the end-diastolic and end-systolic phases were selected in three consecutive cardiac cycles and their mean was analysed. End-diastole was defined as the point in cardiac cycle at the onset of the electrocardiographic (ECG) Q wave. End-systole was defined as the time of apparent minimal left ventricular chamber size. The selected frames were digitized off-line by an array processor-based computer[19,21]. Images were converted into frames of 256 × 256 pixels of 256 gray levels each (0=black, 255=white). The asynergic zone and a remote control region were analysed. The region of interest was always the same size and was placed in the same location of the wall at the different study points (end-diastole and end-systole). A region of interest of 200 × 300 pixels was selected with use of the digitizing pad and the mouse. Regions of interest were individually selected during diastole and systole so as to include the myocardium, exclude the endocardial and epicardial specular reflections and avoid areas of echo dropout and obvious artifacts. By selection, the 'control' segments had end-diastolic thickness and % systolic thickening within the normal range in the baseline (pre-revascularization) two-dimensional echocardiographic study.

A region of interest was identified at similar locations at the two points in the cardiac cycle with the use of anatomic landmarks by an experienced observer who was blind to the hypothesis of the study. The gray level 'cyclic variation index' was calculated by the following formula: (end-diastolic echo amplitude — end-systolic echo amplitude/end-diastolic echo amplitude) × 100.

The intra- and interobserver reproducibility of the measurements was previously assessed and was shown to be acceptable, with an intra- and interobserver variation of 9 and 10% respectively[21]. In each segment evaluated for videodensitometric assessment, the end-diastolic and end-systolic wall thicknesses were also measured from 2-D targeted M-mode tracings.

Echocardiographic follow-up

Follow-up echocardiograms were obtained in all patients at least 4 weeks (42 ± 14 days) after successful coronary revascularization. The revascularized vessels corresponded to the territory investigated for viability: left anterior descending artery for anterior septum; right coronary or circumflex artery for infero-posterior wall.

Postoperative resting wall motion score was determined as previously described by an experienced echocardiographer who was blinded as to patient identification, clinical history, and videodensitometric data obtained on pre-revascularization echocardiogram. Digital acquisition of images was obtained with a side by side display of baseline (pre-revascularization) and follow-up (post-revascularization) echocardiograms. Improved segmental wall motion at follow-up was defined as endocardial excursion and wall thickening (score 1 or 2) in areas of akinesis or dyskinesis (score 3 or 4) at baseline, or normalization (score 1) of reduced endocardial excursion and wall thickening (score 2) at baseline. In presence of paradoxical septal motion following cardiac surgery, we emphasized systolic wall thickening.

Statistical analysis

Continuous variables are expressed as mean value ± standard deviation. Multiple group comparisons were performed by analysis of variance followed by the Scheffe' test. Intragroup differences were tested for significance by the Student's t-test for paired values. Upper and lower 95% confidence limits for each variable were calculated from the 2 tails of the Student's t-test distribution by using the following formulas: mean+(2·042 × SD), and mean − (2·042 × SD), respectively[22]. Linear regression analysis was used to correlate the percentual systolic thickening and percentual cyclic variation of gray level amplitude in the asynergic segments. The accepted level of significance was $P<0.05$.

Results

Echocardiographic findings

The resting echocardiogram identified 34 asynergic regions (septum in 15, infero-posterior wall in 19), showing akinesis in 26 and severe hypokinesis in eight. Follow-up echocardiograms showed improvement in 18 segments (viable: group I), whereas fixed abnormalities were observed in 16 segments (necrotic: group II).

Videodensitometric findings

The data of the main conventional echocardiographic and videodensitometric parameters of the two groups, as well as of the normal control segments, are reported in Table 1. Control segments showed a consistent cyclic gray level variation (38 ± 14%), with gray level values higher at end-diastole (48 ± 22) than at end-systole (30 ± 18, $P<0.01$). No differences were observed between the two groups regarding segmental wall motion score index, end-diastolic thickness or percentual systolic thickening. In segments of group I, mean gray level amplitude in the basally dyssynergic zones decreased from 52 ± 20 (end-diastole) to 34 ± 16 (end-systole), $P<0.01$, with a cyclic variation of 26 ± 16% ($P<0.05$ vs normal regions). A typical videodensitometric pattern of a viable segment is shown in Fig. 1. In viable segments, no correlation was observed between percentual systolic thickening and percentual systo-diastolic variation of gray level amplitude

Table 1 Conventional echocardiographic and videodensitometric data

	Control segments	Asynergic, viable segments (group I)	Asynergic, necrotic segments (group II)
Baseline echo			
Segmental WMSI (baseline)	1	2·7 ± 0·5	2·9 ± 0·3
End-diastolic thickness (mm)	11 ± 1	10 ± 2	10 ± 2
% Systolic thickening	38 ± 11	5 ± 5	4 ± 6
Cyclic variation index	38 ± 14	26 ± 16	1 ± 13
Follow-up echo			
Segmental WMSI (baseline)	1	1·5 ± 0·6	2·9 ± 0·3
End-diastolic thickness (mm)	11 ± 1	10 ± 2	11 ± 2
% Systolic thickening	40 ± 9	31 ± 17	5 ± 7

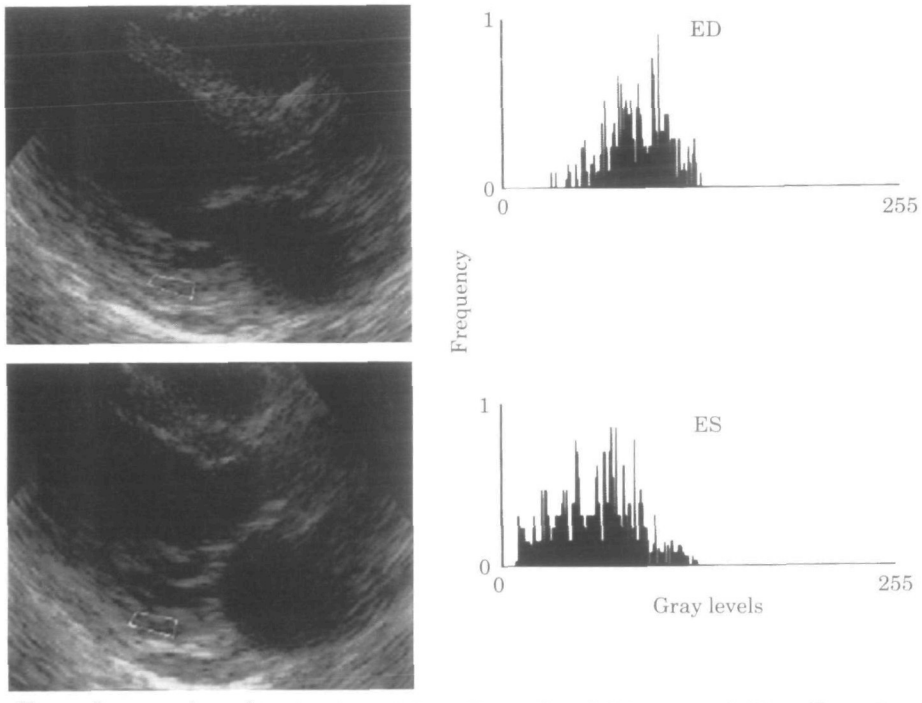

Figure 1 **A typical videodensitometric pattern of a viable segment. Two-dimensional end-diastolic (ED, upper panel) and end-systolic (ES) frames are shown on the left. The region of interest is located in the inferior wall, which appears akinetic. On the right, the gray level frequency histogram corresponding to the region of interest exhibits a cyclic gray level variation, with an end-diastolic peak shifted rightward (towards brighter gray levels) in comparison with the end-systolic peak.**

(r=0·15, P=ns). By contrast, in group II, mean gray level amplitude in the basally dyssynergic regions did not change during cardiac contraction (end-diastole=60 ± 23 vs end-systole 60 ± 24, P=ns). A typical videodensitometric pattern of a necrotic segment with a flat gray level response is shown in Fig. 2. The cyclic variation was significantly higher in viable than in necrotic segments (26 ± 16 vs 1 ± 13%, P<0·01). When individual segment analysis was performed, the cyclic variation index was below the 95% confidence limits obtained for the normal control segments in two (11%) of the group I (viable) segments and in 14 (88%) of the group II (necrotic) segments (Fig. 3). Assuming 9·4% as a cut-off for viability, the cyclic variation index yielded a 89% sensitivity and 88% specificity for predicting post-revascularization functional recovery.

Discussion

Ultrasonic tissue characterization by videodensitometric assessment of regional cyclic gray level variation in conventional images acquired with standard ultrasound

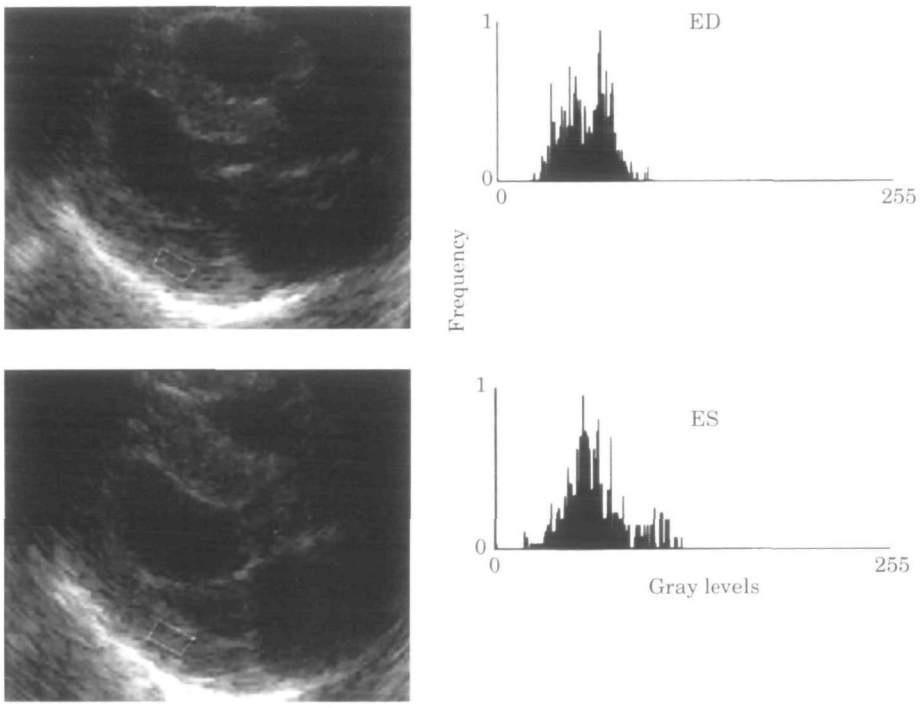

Figure 2 **A typical videodensitometric pattern of a necrotic segment. Two-dimensional end-diastolic (ED, upper panel) and end-systolic (ES) frames are shown on the left. The region of interest is located in the inferior wall, which appears akinetic. On the right, the gray level frequency histogram corresponding to the region of interest does not exhibit a cyclic gray level variation, with an end-diastolic peak virtually identical to the end systolic peak.**

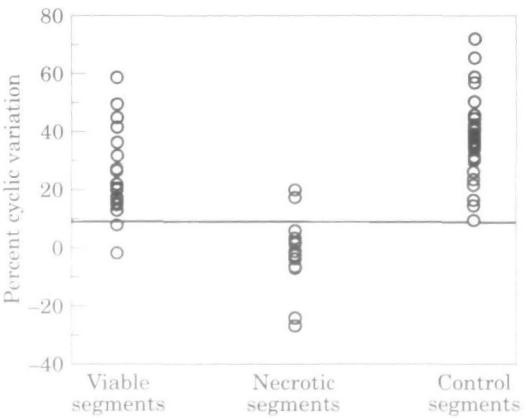

Figure 3 **Scatterplots showing individual values for cyclic gray level variation in the asynergic viable, the asynergic necrotic and the normofunctioning control segments. Horizontal line represents the 95% normal confidence limits for the control group.**

equipment allows the reliable identification of viable, but dyssynergic, myocardial regions, in a simple way, require low-tech apparatus of image digitization and analysis, with no need administering intravenous pharmacological stress, and circumventing the observer-dependence of any echocardiographic method based on wall motion analysis.

The physiological basis of the cyclic variation as a marker of viability

A large body of experimental and clinical evidence suggests that cyclic variation reflects intrinsic contractile function, although the physiological basis of this index has not yet been completely understood and is almost certainly multifactorial[23,24]. As with most phenomena involving the heart, ultrasonic backscatter varies throughout the cardiac cycle, with peak levels of ultrasound reflected at end-diastole and a nadir occurring near end-systole[6–10]. This cyclic variation was first described in the normal myocardium in the pioneering work of scientists in the laboratory at Washington University, and confirmed by several other groups and techniques[6–10,15,19,20–25]. Since the contractile function of sarcomeres so closely parallels ventricular systole, it must be an important contributor to the cyclic variation[26,27]. However, contractile function does not totally explain the phenomenon of cyclic variation, because it has been convincingly shown that, after release of a coronary occlusion in an animal model of ischaemia, cyclic variation of backscatter is restored substantially before regional left ventricular thickening[13,14]. Regional systolic thickening reflects, in an integrated way, the transmural wall function. Mid-myocardial and sub-epicardial contractile function may persist despite diminished wall thickening during stunning in dogs[28]. These observations are consistent with the data from

Gallagher *et al.*, showing that subepicardial contraction can be relatively well preserved despite substantially reduced wall thickening[29]. Cyclic echo amplitude variation represents a physiological measurement that is non-linearly related to, but also distinct from wall thickening. An educated guess is that the preserved intramural contractile function — mainly subepicardial, and therefore not translated into percent thickening — is mirrored by the preserved cyclic variation.

Study limitations

Conventional two-dimensional echocardiographic images were analysed by videodensitometry. It is well known that electronic processing heavily manipulates the radiofrequency native signal to optimize the display of specular reflectors such as endocardial borders[5]. Therefore, videodensitometry is influenced by native ultrasound signal modifications due to manifold electronic processing, and mainly by the type of preprocessing map[5]. Conversely, radiofrequency analysis being independent of native ultrasound signal modifications, allows a more accurate characterization of myocardial acoustic properties[5]. However, this type of analysis is more complex, and data acquisition requires prototypes that are not commercially available. Clearly, this limits its clinical implementation. On the other hand, the advantages of videodensitometric analysis include the use of standard echocardiographic instrumentation with substantially smaller data storage and processing requirements for video data[5]. In addition, the use of a parameter such as cyclic variation — focusing on relative changes of gray level amplitude in the cardiac cycle, rather than on its absolute values — allows averaging out the many potential sources of error in assessing absolute echocardiographic values by analysis of conventionally acquired images. In addition, in all our studies, by inclusion criteria, the gray level scale transfer function was adjusted to be linear for the entire video signal range. Therefore making the assessment of cyclic changes more reliable[15].

We focused our analysis only on myocardial regions imaged by transthoracic echocardiography in the parasternal long axis view. This decision was related to the anisotropic acoustic properties of myocardial tissue, determining that cyclic echodensity variations, assessed by either backscatter or videodensitometry, can be consistently detected only in certain regions, utilizing given projections in which the ultrasonic beams impact perpendicularly to the fibre orientation. A consistent cyclic variation can probably be found in all myocardial regions, given a proper perpendicular angle of insonation. For instance, with epicardial echocardiography in a short axis mid-papillary view, all myocardial regions exhibited a cyclic gray level variation[19], and with transoesophageal echocardiography[30], both the anterior and the inferior wall showed a cyclic backscatter excursion of comparable entity.

The echocardiographically documented improvement of wall motion at follow-up was used as the gold standard for judging the predictive accuracy of cyclic gray level variation. We did not use an independent standard such as fluorodeoxyglucose uptake with positron emission tomography or Thallium uptake with rest-redistribution protocols[1]. On the other hand, our approach had also obvious advantages, from both a methodological and a conceptual viewpoint. We matched the same segments imaged with the same 2D-echo technique in the same projections without the inaccuracies and approximations inherent to any attempt to match segments visualized by different techniques, such as echo and planar or tomographic thallium or positron emission tomography. It is also true that there is little dispute that a segment recovering after revascularization was viable at the time of the index echo evaluation; furthermore — from a clinical viewpoint — our aim was to predict recovery following revascularization, not metabolic activity which may not always correlate with reversible dysfunction[31]. Therefore, we felt justified in measuring the accuracy of the videodensitometric approach against the post-revascularization outcome in systolic segmental wall motion as a yardstick of myocardial viability.

Our population was highly selected: all patients had a resting dyssynergy well imaged in the parasternal long axis view; the number of patients and of segments was relatively limited; and other standard approaches for determining viability were not assessed. In fact, we believe that, according to Diamond[32], we should not mistake the seed of feasibility and potential usefulness with the fruit of effectiveness. This actually should be true for all techniques, which are all too often implemented in clinical practice after inadequate validations and head to head comparisons with time-honoured approaches.

Comparison with previous studies

Our results are conceptually germane to the findings obtained in animal experiments and in man by Perez, Miller and coworkers at the Washington University of St. Louis. They were the first to show that: (1) normal myocardium exhibits a cyclic variation in echo intensity[6,7]; (2) the overall relationship between wall thickening and cyclic variation is non-linear, indicating that cyclic variation represents a physiological measurement distinct from wall thickening and probably related to intramural function[11,12]; (3) stunned myocardium exhibits a normal cyclic variation in the presence of severe wall motion abnormalities[13,14].

Their data were obtained with quantitative backscatter imaging method, based on the quantitation of the ultrasonic power scattered by the myocardium. Returning echo signals are digitized in the radiofrequency range, after prospective acquisition with non-commercially available dedicated prototypes. Recently, the potential of a much simpler videodensitometric analysis of gray level variation in standard images acquired with conventional ultrasound equipment, was

shown to mirror the information of the much more technologically demanding backscatter analysis in many conditions, ranging from normal contraction[15] to acute myocardial ischaemia[21]. It was therefore conceivable to apply this parameter to the study of viable myocardium, a field in which cyclic integrated backscatter yields attractive information. The step-up in clinical appeal going from backscatter analysis to videodensitometry approach might be comparable to that achieved by moving from positron emission tomography to gamma-camera imaging[1]: there is an abatement of costs, an increase in applicability and a simplification of procedures. The videodensitometric approach appears to be a suitable low-tech alternative to backscatter imaging for viability detection, with potential to integrate and expand the possibility of echocardiography — by pharmacological stress and myocardial contrast echocardiography — in the difficult task of reliably identifying viable myocardium[33].

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