UNIVERSITA' DEGLI STUDI DI MESSINA

Dipartimento di Medicina Clinica e Sperimentale

AOU Policlinico "G.Martino" Di Messina

Dottorato di Ricerca XXX° Ciclo in

Scienze Biomediche Cliniche e Sperimentali

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ASSESSMENT OF LEFT VENTRICULAR MECHANICS IN ACUTE AND CHRONIC PHASES OF MYOCARDITIS: A STUDY WITH CARDIAC MAGNETIC RESONANCE IMAGING AND 2D STRAIN ECHOCARDIOGRAPHY

Tesi di Dottorato della Dott.ssa

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SSD: MED 11

***Anno Accademico* 2016-2017**

SUMMARY

INTRODUCTION

Acute myocarditis (AM) is an inflammatory disease of myocardium usually due to viral infection. A definite etiological diagnosis of AM is generally achieved by myocardial biopsy meeting established histological, immunological, immunohistochemical criteria (1,2).

Typically, myocardial damage is located in the epicardium and/or mid-layer of the left ventricular (LV) wall and results in a variable effect on LV function, ranging from preserved wall kinesis to severe systolic impairment (3). Accordingly, the clinical presentation is heterogeneous (recent-onset heart failure, bradi- and tachi-arrhythmias, infarct-like symptoms, but also without a symptomatic pictures) as well as long-term outcomes (4,5). A complete recovering of systolic function has been reported in some patients, but adverse remodelling (up to irreversible dilated cardiomyopathy) has been often described (3,6,7).

Usually, patients with infarct-like presentation AM (*hereinafter infarct-like AM*) show preserved LV ejection fraction (EF). In these subsets of patients with AM, biopsy is not strictly indicated in order to make diagnosis, and specific pharmacological therapy against adverse remodelling is not mandatory, also considering their quite good prognosis (1,4).

Over the last 15 years, Cardiac Magnetic Resonance (CMR) has been demonstrated to discriminate AM from myocardial infarction (8,9). It allows to assess LV volumes and function, as well as to identify the acute myocardial damage (oedema, hyperemia and/or fibrosis). Particularly, high accuracy and diagnostic impact of Late Gadolinium Enhancement (LGE) in detection of AM has been confirmed (8). Recently, two-dimensional (2D) strain echocardiography was also validated in order to provide important information on myocardial dysfunction in patients with AM, even if no wall motion abnormalities are detected. In these patients, a significant longitudinal and (less frequent) circumferential dysfunction have been recognized (10,11,12).

However, despite such studies, only few data on long-term myocardial functional outcomes following the acute inflammation are available.

CHAPTER 1

MIOCARDIAL STRUCTURE AND LEFT VENTRICULAR FUNCTION

1.1 Myocardial Fibers Architecture and Two-Dimensional Speckle-Tracking Echocardiography (2D STE) Role

Recent microscopic studies have shown that the orientation of the fibers in the myocardial wall is not constant but varies in a distinct manner: the more internal, subendocardial fibers have a predominantly longitudinal direction, while as it moves towards the epicardium, orientation of the fibers change, taking a transverse direction; moving further towards the epicardium, the orientation of the fibers continues to change, until the more superficial ones reengage longitudinally. Overall, it is as if from the endocardium to the epicardium the fibrocells rotated by 180°, describing a spiral: the subendocardial fibers describe a direct propeller, and are first activated by the wave of depolarization; the subepicardial fibers, however, have a left-handed helical pattern, and are reached from the wavefront immediately afterwards. (13)

The subendocardial fiber shortening, which generates the isovolumetric contraction, causes the stretching of the subepicardial fibers and creates the presupposition for their contraction, which is responsible for the ejection. During the ejection, although the contraction affects all segments, the Longitudinal Strain (LS) is greater at the apex than at the base, and this explains why at this stage the mitral ring approaches the apex.

Left ventricular torsion studies have shown that in normal hearts during the isovolumetic contraction phase, subendocardial fibrous shortening causes a short and modest apical rotation, and an initial anti-clockwise rotation at baseline. With the contraction of the subepicardial fibers, during the ejection phase, a clear apical anti-clock rotation is realized, with rotation in the opposite direction, that is, hourly and baseline. This series of phenomena is probably due to the fact that the activation begins in the subendocardium and then advances to the subepicardium: therefore the contraction of the subendocardial layers precedes that of subepicardics. (14)

Finally, during the diastole, the opposite events occur: clockwise apical rotation, and anti-clockwise basal rotation. Changes in the shape of such a complex structure, which occur during systole, are circumferential, radial, and twist shortening. These variations are determined by the different positions and angles that take the fibers in the various phases of the heart cycle and are in turn determinant of the systolic and diastolic heart function. The longitudinal shortening of these fibers results in a reduction in the LV length of about 10-12%, while the radial one causes a decrease in the transverse diameter of approximately 25%, resulting in a transformation of the ventricular cavity into the systole, which assumes a less spherical shape than diastolic. During the systole there is a myocardial shortening on a longitudinal plane (longitudinal strain) which consists in moving the base to the apex, resulting in a reduction in the long axis of the left ventricle; this longitudinal deformation is accompanied by a circumferential shortening (circumferential strain) with a reduction in the circumference of the LV cavity and, finally, radial thickening (radial strain) with increased thickness of the myocardial walls and hence the endocardial-epicardial distance (Figure 1) The twist of the left ventricle around its long axis is caused by the contraction of the fibers with helical orientation, with a rotation of the apex which is opposite to that of the base. These movements, recalling those of a vortex, induce suction and ejection forces and ensure efficient distribution of stress and regional strain. (14) The difference between the peak rotation at the apex and the peak rotation at the base, in the systole, determines the degree of twisting of the left ventricle. In the diastolic phase there is a return of twisting or relaxation ("recoil", "untwist") with the base of the ventricle rotating anti-clockwise and the apex rotating clockwise. (15) Alterations in motion and amplitude of LV apex rotation were found in pathological conditions such as hypertrophic and dilated cardiomyopathy, myocardial ischemia, acute myocardial infarction, and aortic stenosis. Magnetic Resonance Imaging (MRI) methods have shown that in patients with severe aortic stenosis pressure overload is associated with an increase of LV systolic torsion which is a likely compensation mechanism. (16,17) Until recently, MRI was the only non-invasive technique for measuring LV twist; this technique is able to evaluate three-dimensional (3D)

movement of the heart in a non-invasive way, but is limited by the low spread and the high costs that make it difficult to apply in routine clinical practice.

The recent development of Speckle Tracking Echocardiography (STE) technique, which provides the evaluation of regional myocardial deformation (radial, circumferential and longitudinal) directly from two-dimensional images (2D), has allowed to get a non invasive estimation of accurate rotation and of LV strain, providing high reproducibility comparable to those of the MRI. (18)

This technique allows to overcome the strains derived from Tissue Doppler Imaging (TDI) such as the angle of dependence and the ability to obtain deformation measures only on the longitudinal plane. Speckle-Tracking is based on the recognition of pixel groups within the myocardial wall with specific acoustic characteristics that precisely because of their reflectivity characteristics are followed frames per frame during the heart cycle. "Speckles" are natural acoustic markers that appear as light and small elements in conventional, grayscale images. Such speckles are the result of constructive and destructive interference of the ultrasonic beam, resulting from the back-scattering of smaller structures of an ultrasonic beam wavelength. These markers are equally distributed in the myocardium and can be identified and followed in consecutive frames during cardiac cycles. Measurements can be made simultaneously by multiple regions of interest (ROI) with a previously acquired, grayscale image. The distance between the selected speckles is measured within a predetermined myocardial area as a time function and the myocardial deformation parameters can thus be derived. This is in contrast to strain analysis from the TDI, where the sample volume is a fixed area in the space and all measurements are made with reference to an external point (transducer). Therefore, the strain obtained by STE method is a direct measure of myocardial deformation while the derived TDI strain is a strain rate integration. (19)

1.2 Cardiac Magnetic Resonance

Cardiac magnetic resonance (CMR) imaging is a non invasive diagnostic tool for detection of acute and chronic myocarditis. (20,21) In 2009, the International Consensus Group on CMR in

Myocarditis proposed ‘Lake Louise Criteria’ for myocarditis. At least two of the following CMR criteria must be present: increased of regional or global myocardial signal intensity in T2-weighted images (edema), increased global myocardial early (3 min) gadolinium enhancement ratio between myocardium and skeletal muscle in gadolinium- enhanced T1-weighted images (hyperemia), nonischemic regional distribution in late gadolinium enhancement (LGE) most frequently involving the subepicardial portion of the lateral left ventricular (LV) wall and less frequently the mid-wall of the interventricular septum. (22)

The concomitant existence of skeletal muscle inflammation with edema, observed in acute myocarditis, can lead to false negative results for myocardial edema. (23)

Hyperemia identification, as proposed by Lake Louise criteria, is very challenging and poorly used in the clinical setting. Recently, postcontrast cine steady state free precession (SSFP) sequences were proposed as a new method for hyperemia identification *(Figure 2).* (24, 25)

The sensitivity of CMR for the diagnosis of myocarditis is high for infarct-like presentation, low for myocarditis with heart failure and with arrhythmic clinical presentation.

CMR findings should be used cautiously in these two latter conditions. The persistence of inflammation in chronic myocarditis is associated with LV dilatation and dysfunction and to tissue abnormalities on CMR, but the role of CMR in assessing chronic myocarditis patients is uncertain. Recent data suggest that chronic myocardial inflammation may be associated with generalized myocardial edema and to an increase of global myocardial enhancement. (26,27).

Recent reports demonstrated that hyperenhancement areas can decrease or even disappear over time. (28,29)

A study of comparison with endomyocardial biopsy (EMB) demonstrated that CMR with LGE is highly accurate to detect acute myocardial inflammation, in which LGE is positive in 95% cases, but it is not sensitive to detect chronic myocarditis (LGE positive in only 40%). (30)

CHAPTER 2

ORIGINAL CONTRIBUTION

Aim of the present study was to assess changes in LV myocardial deformation over time, and the relationship between myocardial deformation measures and LGE in patients with infarct-like AM.

2.1 Study Population

From February 2007 to October 2013, all patients admitted to our Cardiac department (University Hospital of Messina, Messina, Italy) for suspected infarct-like AM were prospectively enrolled.

Diagnosis of infarct-like AM was based on all the following criteria: (a) history of flu-like symptoms within 8 weeks prior admission; (b) new onset of symptoms like fatigue/breathlessness, chest pain, mild dyspnea and/or palpitation; (c) ischemic ECG pattern (ST-segment elevation and/or T wave anomalies); (d) increase of inflammation markers (non-high-sensitivity CRP > 8 mg/l and/or white blood cell count > 11.000/mm^3) and cardiac enzymes; and (e) preserved global systolic function (EF > 50%).

We excluded patients with New York Heart Association (NYHA) from II to IV, patients with LVEF<50% and those with electrocardiographic evidence of bradyarrhythmias (≥ second-degree atrioventricular block) or tachyarrhythmias (ventricular or supraventricular arrhythmias).

Every patient underwent standard transthoracic echocardiogram with strain evaluation, and CMR on the same day, in random order. Either, coronary angiography or coronary multislice computed tomography scan was performed to rule out active coronary artery disease on the basis of LGE ischemic pattern or in patients aged >35 years, more likely to have a coronary disease.

On admission, clinical and instrumental findings from the enrolled patients were compared to those from a control group of 25 apparently healthy male subjects (mean aged 25±7 years) with no history or symptoms of heart disease, absence of hypertension, obesity, glucose intolerance, smoking, metabolic syndrome, and normal ECG and transthoracic echocardiogram.

Clinical outcomes were evaluated over time, as follows: 1) new occurrence of AM; 2) hospitalization for any cardiac reasons; 3) new signs of / worsening heart failure; 4) arrhythmias; 5) cardiac death.

The study protocol was approved by the local ethics review committee and the investigation conformed to the principles outlined in the Declaration of Helsinki. Written informed consent was obtained from all participants.

2.2 Cardiac Magnetic Resonance

CMR was performed using a 1.5-T system (Gyroscan NT; Philips Medical Systems, Best, The Netherlands) with cardiac phased-array coil and vectorcardiogram synchronization. A breath-hold balanced fast field echo sequence was used to evaluate LV volumes and global LV function. In each patient, depending on LV volume, a total of 9 to 14 short-axis views and 2 long-axis views (four-chamber view and two-chamber view, respectively) were acquired.

Left ventricular volumes, mass, and EF were measured using a previously validated software (EasyVision, version 4.0; Philips Medical Systems, Best, The Netherlands) and categorized according with reference values (32).

On T2-weighted images, areas of myocardial edema were visually evaluated. The presence of edema was confirmed if the signal intensity of the myocardium was >5 SD of the mean signal intensity of the skeletal muscle (33).

LGE images by a gradient echo inversion recovery sequence were obtained within 10 – 20 min after bolus injection of 0.2 mmol/Kg of gadobutrol (Gadovist®, Schering, Germany). Inversion time (200-320 msec) was optimized to a null signal from normal myocardium. As reported in previous studies (34,35), the areas of LGE (LGE positive) were assessed by visual approach with a scheme based on the extent and localization of LGE (epicardial or mid-layer localization). According with Lake Louise criteria, patients had a definite diagnosis of AM if was observed both edema and LGE (36).

LGE images were obtained in each patient with suspected myocarditis. LGE images don't were acquired in control group.

2.3 Strain-by 2D Speckle Tracking Echocardiography

Ultrasound studies were performed using a commercial ultrasound machine (MyLabAlpha Esaote, Florence, Italy) equipped with an1 to 4 MHz phased-array adult cardiology transducer (SP2430 probe, Esaote, Florence , Italy). Parasternal short-axis views at the basal, mid and apical levels and three standard apical views (four-chamber, two-chamber and LV outflow long-axis) were acquired. LV volumes and LVEF were assessed by the apical biplane Simpson method (37). LV mass was calculated using the ASE recommended criteria from validated studies.

The ratio between the E-wave velocity of the pulsed-wave Doppler mitral flow image and the average of early diastolic velocity from the septum and lateral side of the mitral annulus (E' wave) on tissue Doppler imaging (TDI) was used as a marker of filling pressures (38).

A dedicated software package for two-dimensional speckle tracking (2D STE) strain analysis (XStrain™, Esaote, Florence, Italy), validated both on synthetic ultrasound data (39) and in vivo (40) as part of the Initiative to standardize deformation imaging promoted by ASE and EACVI, was used to quantify both endocardial (ENDO) and epicardial (EPI) strains.

To explain in more detail, it relies on a "feature tracking" algorithm and, in order to improve the border tracking results, it combines speckle tracking with other information such as tissue-to-blood border detection, the periodicity of the cardiac cycle and the fact that the cardiac borders maintain their own "overall spatial coherence" over time. Digitalized 2-D video clips were further analyzed using XStrain software: the ENDO border to be tracked was drawn by the operator and was identified as a sequence of points (feature tracking); the EPI border was automatically tracked and manually adjusted when required; frame by- frame displacement of these points (both ENDO and EPI) was automatically evaluated, generating strain curves for each segment. The tracking quality was verified for each segment and subsequent manual adjustments were performed, when required.

All data were analyzed with the aid of Fourier techniques, which ensure greater accuracy using the periodicity of the heart motion.

Global strain values were obtained as the average of each valuable segment measurement, from apical 4-, 2- and 3-chamber (global longitudinal strain and short-axis views (radial and circumferential), at basal, mid and apical level, respectively, as previously described (41,42).

Two skilled cardiologists performed all examinations and measurements blinded respect to clinical presentation and CMR findings.

2.4 Statistical analysis

Continuous variables were expressed as mean ± SD and categorical variables as percentages. One-way analysis of variance (ANOVA) was used to compare data among the three groups (Controls, AM and follow-up). Paired two-sample t-tests were used to assess the differences between acute and follow-up myocarditis. Inter-observer and intra-observer variability were assessed using the Bland–Altman method. For any statistical comparison, a p value <0.05 was considered significant. Analyses were performed using SPSS version 12 (SPSS Inc., Chicago, Illinois).

CHAPTER 3

RESULTS

Each of the 39 enrolled patients completed the CMR examination without major complications and with sufficient image quality. Four out of them were excluded according to MRI findings because an ischemic LGE pattern was found in 2 cases and the absence of oedema in 2 more patients. Therefore, the remaining 35 male patients (aged 25±8 years) with a definite CMR diagnosis of acute myocarditis consisted in the AM group. As showed on table 1, each patient was male, had history of flu-like symptoms and elevated CRP, chest pain, ST segment elevation and elevated troponin.

Both patients and controls were free from any other relevant systemic disorder, including hypertension or diabetes.

The median follow-up length was 22 months (range: 3.5-61). During such period, 6/35 patients with no cardiac events were lost at follow-up and 3/35 patients showed a relapse of AM. The 3 patients suffering of relapse of AM has similar LV segments extent regarding edema (mean 3 segments), LGE and LV deformation.

The remaining 26 patients (FU-group) experienced uneventful follow-up.

Both patients and controls underwent clinical evaluation and strain echocardiography study, but cardiac MRI was performed only in AM patients. As shown in table 2, although in a normal range, LV septum and mass were significantly higher in AM and FU groups than in controls. There was no difference about mean value of EDV among groups; however, comparing each patient during its acute phase respect to FU, during FU was found a significant mild increase of EDV. All AM patients had normal LV volumes, mass and EF, with the presence of oedema and LGE pattern at cardiac MRI suggestive for AM (Table 3).

A total of 147 out of 595 (25%) LV segments were LGE-positive: 138 with sub-EPI distribution in the anterior-lateral and/or inferior-lateral wall, and 9 with mid-wall septal distribution.

Endocardial function

Global ENDO longitudinal strain was similar between AM and FU groups, and lower in magnitude in both AM and FU groups than in controls (Table 4). During the acute disease, ENDO longitudinal strain was poorer in magnitude in LGE-positive than LGE-negative myocardial segments (p=0.04) (figure 4, panel A).

Compared to baseline, an improvement in ENDO longitudinal deformation, both in LGE-positive (p=0.06) and LGE-negative (p=0.02) segments, was observed in the FU group (figure 4, panel A).

The mean value of global ENDO circumferential strain were similar among controls, AM group and FU group. Comparing each patient during its acute phase respect to FU, an increase in magnitude of global ENDO circumferential strain was observed (p=0.001) during FU.

ENDO circumferential strain was found to be tendentially more lower in magnitude in LGE-positive segments, both in AM (p=0.07) and FU patients (p=0.08), with a significant improvement in magnitude at follow-up in both LGE-positive (p=0.002) and LGE-negative (p<0.0001) segments (figure 5, panel A).

Global radial function was not different among groups (table 4) and in segments with LGE positive and negative LGE both in AM and FU (figure 5, panel C).

Epicardial function

The mean value of global EPI longitudinal and circumferential strains were similar among groups (AM group, FU group and controls). On the contrary, comparing each patient during its acute phase respect to FU, a global EPI longitudinal and circumferential deformation significantly improved (supranormal) in magnitude in FU compared to AM patients (Table 4).

The mean value of EPI circumferential strain was significant higher in magnitude (supranormal) in FU respect AM group ($P < 0.05$) and Control group ($P < 0.05$). This difference was significant increased ($P < 0.0001$) comparing each patient during its acute phase respect to FU.

In AM and FU groups respectively, EPI longitudinal and EPI circumferential strains of segments with LGE-negative and LGE-positive were similar. Considering the change in LV deformation from acute phase to FU phase, EPI longitudinal strain was improved in magnitude in LGE-positive segments only ($p=0.01$), whereas circumferential strain was improved in magnitude in both LGE positive ($p=0.001$) and LGE negative ($p<0.0001$) segments (Fig. 4B and Fig 5B).

Reproducibility

There was good interobserver agreement concerning ENDO longitudinal (mean –1%, SD 3), EPI longitudinal (mean 2%, SD 3), ENDO circumferential (mean 2%, SD 3), EPI circumferential (mean 2%, SD 4) and radial strain (mean −1%, SD 4). Intra-observer agreement of ENDO longitudinal (mean 1%, SD 3), EPI longitudinal (mean 2%, SD 3), ENDO circumferential (mean 1.5%, SD 3), circumferential (mean 2%, SD 3) and radial strain (mean 1 %, SD 3) was also good.

CHAPTER 4

DISCUSSION AND LIMITS

In this study we have analysed the effect of acute myocarditis damage on LV myocardial deformation and remodelling both in acute myocarditis phase and during follow-up (about 2 years later) and have compared these data with a matched controls group.

The main findings of our study show that: a) an impairment of ENDO longitudinal strain and an increase of LV septum dimension and mass were observed in myocarditis (both during acute and follow-up period) respect to controls; b) an increase in magnitude (supra-normal) of EPI circumferential deformation was observed in myocarditis during FU respect to both AM and controls; c) comparing each patient from his acute phase to FU was observed a mild increase of EDV, an increase in magnitude of EPI longitudinal deformation and an increase in magnitude of ENDO circumferential deformation and a further increase in magnitude (supra-normal) of EPI circumferential deformation; d) no impairment in radial deformation; e) epicardial LGE influences both ENDO and EPI longitudinal and circumferential deformation (segments without LGE showed a greater increase in magnitude of LV deformation during FU respect to segment with LGE).

Interesting all these data were obtained in patients with preserved global systolic function both during acute phase and FU.

As previously showed of many Authors, longitudinal LV function is impaired in AM with epicardial LGE and preserved EF. In addition to these data, we have demonstrated that global longitudinal dysfunction observed in AM persists about 2 years during FU.

Our results are similar to those of Caspar et al (43) showing that longitudinal dysfunction persists 2 years later AM. Differently from Caspar's study, we analyzed circumferential and radial deformation at endocardial and epicardial layers both in acute and FU phases. Additionally, we observed that longitudinal deformation is due to functional abnormalities of endocardium although there is epicardial damage detected by LGE also showing that ENDO

The complexity of orientation of myocardial fibers, physiological and pathophysiological data on LV deformation can contributes to explain the greater impairment of endocardial respect to epicardial longitudinal deformation (44-48).

From a physiological point of view, longitudinal deformation is higher in endocardium than in epicardium (46) because myocardial fibers have proportionally greater changes in dimension with decreasing radius. Chen et al have showed that subendocardial necrosis has a longitudinal strain reduction similar to transmural infartion (47) suggesting that subepicardial layer don't plays a role in longitudinal function. Our data suggest that global longitudinal is the result of the synchronized systolic deformation of both subendocardial (right-handed helix) and subepicardial (left-handed helix) layers; when endocardial or epicardial longitudinal fibers are damaged, longitudinal function, that is prevalent expressed from endocardium, is impaired.

Khoo et al (48) had found a mild improvement 1-year after AM, remaining global strain values lower than in controls. In our data, we found a similar trend on ENDO longitudinal function because it was mild increase (not significant) in magnitude in FU respect AM but it persisting significant impaired in FU than controls.

In the present study, ENDO and EPI circumferential function was found to increase in magnitude (supra-normal value) in the FU group. This can be explained by the fact that circumferential function chiefly locates in the mid-wall myofibers, while LGE is prevalent in epicardial layers.

Previous Authors have showed that circumferential function can increase as a compensatory mechanism in early stages of many cardiac diseases such as cardiomyopathies and diabetic heart.

On the contrary, radial function was not impaired in our AM patients because it is prevalent due to endocardial layer (49-50).

The mild dilatation of EDV during FU can be the effect of a mild adverse remodelling due to myocarditis damage but also a compensative adaptive phenomenon interrelated with the improvement of circumferential function and EPI longitudinal function.

Another interesting data is the increased of LV septal thickness and LV mass in both AM and FU respect to controls; these findings could be due to LV tumefaction (pseudo inflammatory "hypertrophy") (51) but also representing the substrate of the functional compensative phenomena observed in epicardial longitudinal and circumferential deformation.

A further evidence that epicardial LGE indirectly influences endocardial function can be observed through the greater recovery of ENDO longitudinal deformation in segments without respect to segments with LGE; similarly, the increase in magnitude of EPI longitudinal, ENDO and EPI circumferential deformation was found only in segments without LGE.

Accordingly, Løgstrup et al. recently provided a demonstration that longitudinal dysfunction is significantly related to myocardial edema in myocarditis (52); other Authors have showed that longitudinal function tends to ameliorate in patients without persistent inflammation at biopsy (53).

STUDY LIMITATIONS

Despite the present results are quite intriguing, some limitations should be acknowledged. First of all, our study population was entirely composed by male Caucasian patients and we cannot extend our findings to women or patients of different ethnicity. Also, the number of patients enrolled is quite limited and the range of follow up times is broad.

An additional shortcoming is the absence of histopathological findings. Endomyocardial biopsy is the gold standard for a definite diagnosis of AM particularly in patients having HF or severe arrhythmias presentation, but in patients without wall motion abnormalities and preserved EF, this approach is unnecessary or even contraindicated (54). We have made an accurate non-invasive diagnosis because CMR criteria (2 of 3 among edema, hyperemia, LGE) have a high specificity and positive predictive value (both 91%) in acute phase of AM (36, 55). However, many patients with positive endomyocardial biopsy for AM don't show 2 of 3 CMR criteria. Therefore, another limitation of our study is that the selection based only on CMR excludes many patients having AM on biopsy with a false negative CMR. Also for these reasons, the absence of new CMR techniques

as T1 and/or T2 mapping that have showed high accuracy in detecting AM is a further limitation of our study.

Moreover, patients in the FU group were belonging to the AM group. Thus, such functional differences we reported between AM and FU groups over-time changes within the same group.

Cardiac MRI examination at follow-up was missed due to budget restrictions from the original proposal. Thus, changes in LGE signals from the acute to the chronic phase of the myocarditis cannot be provided. Whether these findings can be related to long-term functional recovery or not remains an interesting issue to be investigated in future studies.

Finally, the LGE technique was not performed in normal participants. However, in this group of young participants who had no history or symptoms of heart disease, and normal ECG and echocardiogram, it was very unlikely to find a scar on LGE-CMR.

CHAPTER 5

CONCLUSIONS

The main findings from the present study demonstrate a steady ENDO longitudinal dysfunction in patients with infarct-like AM over a two-year follow-up period. Overall, myocardial deformation chiefly improves in LGE-negative segments, as detected by cardiac MRI in the acute phases of inflammation. EPI longitudinal and circumferential strain deformation can improve as potential adaptive mechanisms, so that the majority of patients present with good clinical condition.

These preliminary results indicate the need for further powered study aimed at establishing long-term outcomes in these patients, potential correlation with microbic agents, as well as cardioprotective therapeutic approach to the early phase of infarct-like AM.

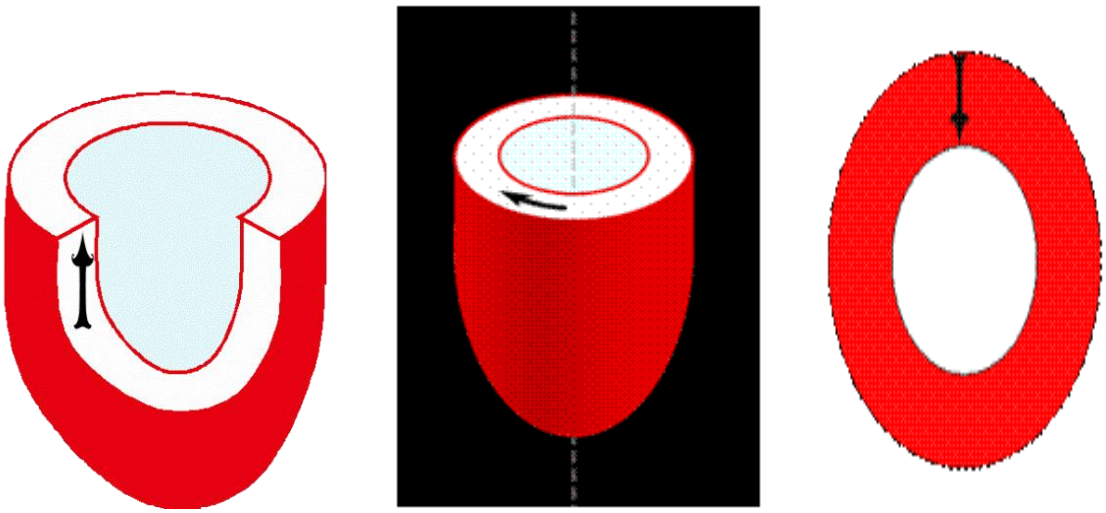

Figure 1
From left to right: myocardial shortening on a longitudinal plane (longitudinal strain) which consists in moving the base to the apex, resulting in a reduction in the long axis of the left ventricle; circumferential shortening (circumferential strain) with a reduction in the circumference of the LV cavity; finally, radial thickening (radial strain).

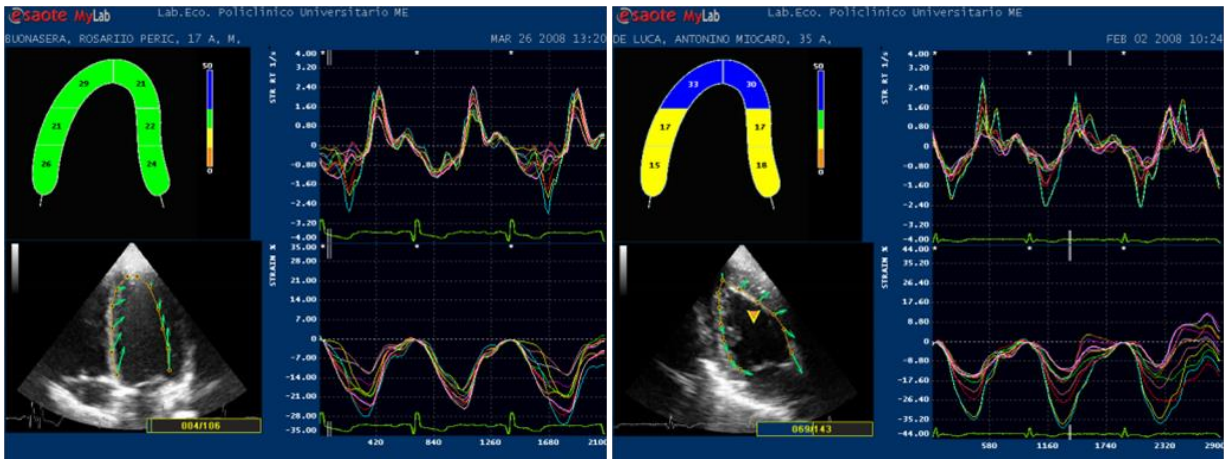

Figure 2
Example of 2-Dimensional Speckle-Tracking analysis: on the left in a normal subject, on the right in a subject with acute myocarditis.

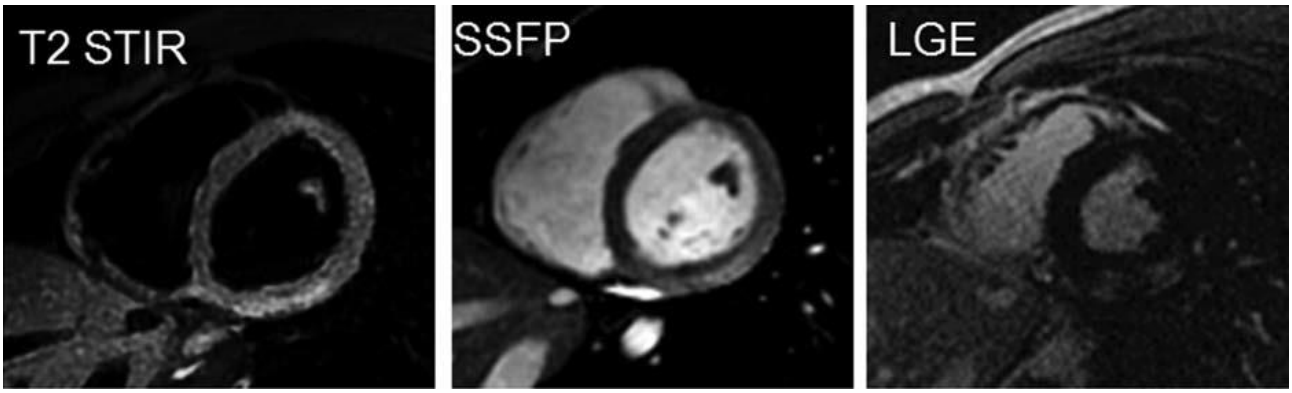

FIGURE 3
Cardiac MRIs of a case of 'infarct like' presentation of acute myocarditis. In T2-STIR image, myocardial hyperintensity of edema is found in inferior and inferolateral wall. In steady-state free precession images (SSFP), acquired early after gadolinium injection, hyperemia is detectable as well as positive LGE in the same myocardial regions.

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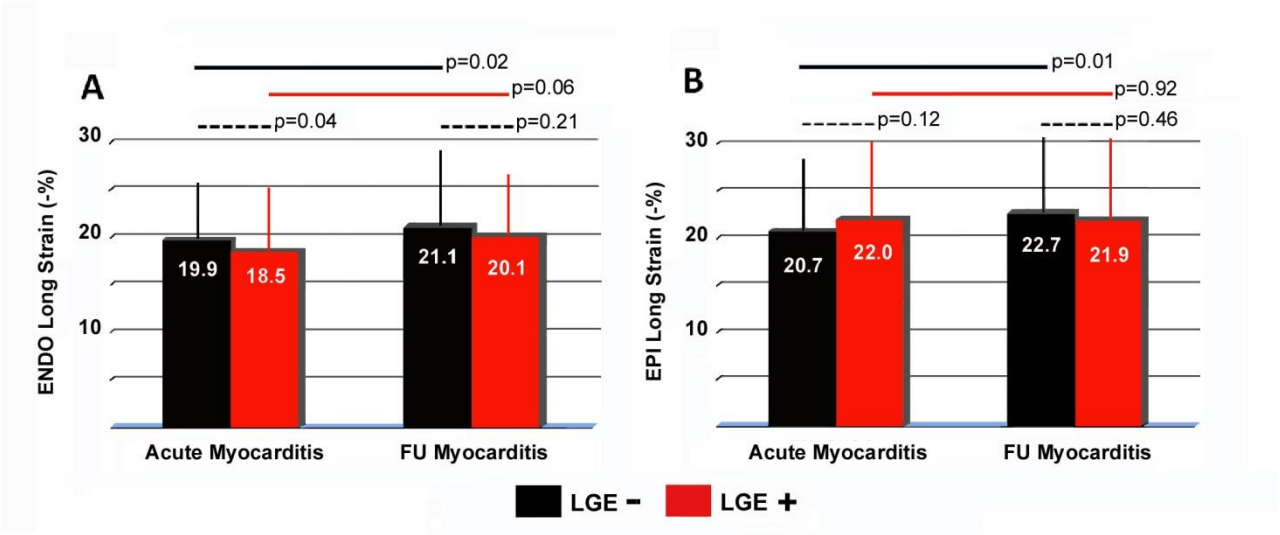

Figure 4
ENDO (panel A) and EPI (panel B) longitudinal strain in LGE-positive (LGE+) and LGE-negative (LGE-) segments in the acute and chronic stages of infarct-like AM.

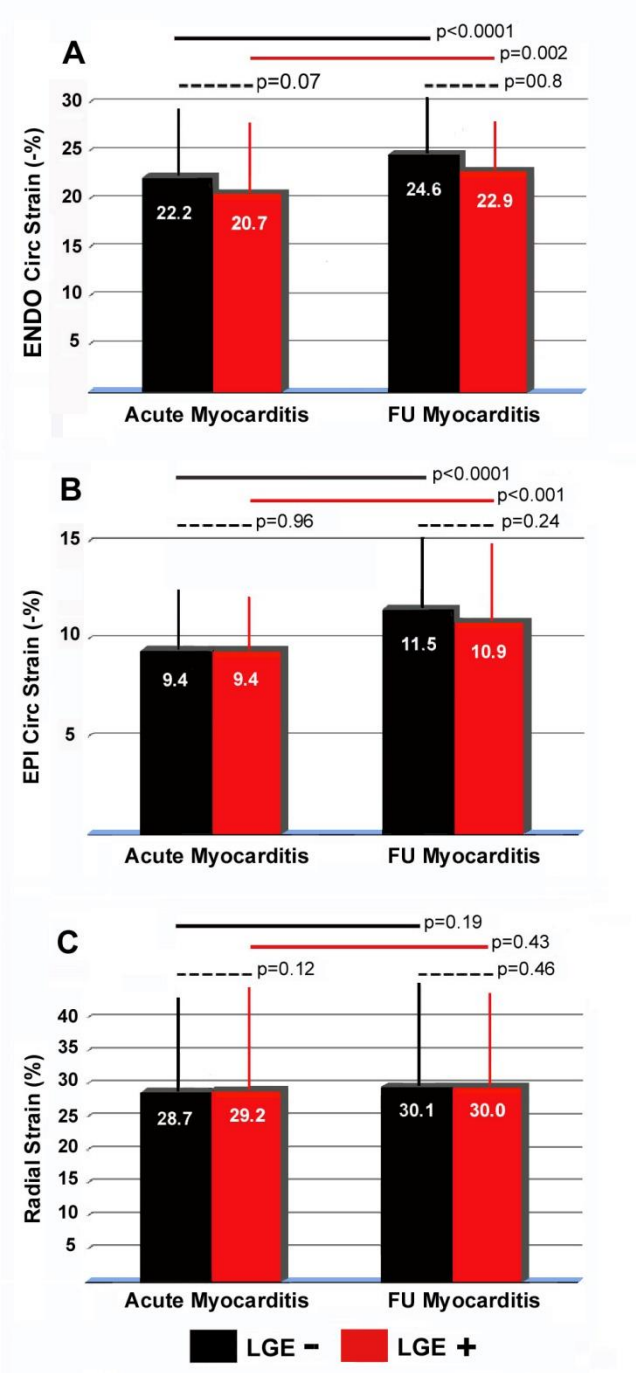

Figure 5

ENDO (panel A) and EPI (panel B) circumferential strain and radial strain (panel C) in LGE-positive (LGE+) and LGE-negative (LGE-) segments in the acute and chronic stages of infarct-like AM.

Table 1

Characteristics of patients with infarct-like acute myocarditis

	Control Group (n=25)	*Infarct-like AM* (n=35)	*P value*
Age	**25±7**	**25±8**	**1**
Gender (male %)	**100**	**100**	**1**
Smokers (%)	**32**	**29**	**0.9**
BMI > 30 (%)	**0**	**3**	**0.9**
History of flu-like symptoms within 8 weeks (%)	**0**	**100**	**<0.0001**
Chest pain (%)	**0**	**100**	**<0.0001**
Fatigue/malaise (%)	**0**	**26**	**0.01**
Dyspnea (%)	**0**	**14**	**0.1**
Palpitation (%)	**0**	**17**	**0.08**
ECG: ST-segment elevation (%)	**0**	**100**	**<0.0001**
ECG: T wave abnormalities (%)	**0**	**34**	**0.003**
Abnormal C-Reactive Protein (%)	//	**100**	//
abnormal white blood cell count (%)	//	**94**	//
Abnormal Troponine I (%)	//	**100**	//

BMI: body surface area; //: not performed.

Table 2
Left ventricular echocardiographic findings in controls and patients with acute and follow-up myocarditis.

	Control Group (n = 25)	AM group (n = 35)	FU group (n = 26)	ANOVA Global P	Paired two-sample P value AM vs FU groups
Left ventricle					
LV Septum (mm)	8.9 ± 0.7	9.8 ± 1.1*	9.8 ± 1*	0.003	0.153
LV IL wall (mm)	8.7 ± 0.9	8.8 ± 1.1	9 ± 1.5	0.795	0.254
LV EDV (ml/BSA)	59 ± 13	57 ± 14	62 ± 14	0.412	0.03
LV ESV (ml/BSA)	27 ± 6	25 ± 8	24 ± 7	0.265	0.143
LV EF (%)	62 ± 3	61 ± 4	62 ± 4	0.704	0.779
LV Mass (gr/BSA)	77 ± 14	97 ± 23*	98 ± 18*	0.001	0.592
E/E'	6.3 ± 1.9	5.5 ± 1.4	5.2 ± 1.9	0.351	0.201

Global P = ANOVA among 3 groups
* P <0.05 vs Control group

** P <0.001 vs Control group

§ P <0.05 vs AM group

§§ P <0.001 vs AM group

EDV = end-diastolic volume, velocity, ESV = end systolic volume, IL = infero-lateral, LV = left ventricular, E/E' = early diastolic velocity at mitral valve inflow/tissue early diastolic, BSA = body surface area.

Table 3
CMR Parameters in infarct-like acute myocarditis

CMR Parameters	*Infarct-like AM* (n=35)
LV-EDV (ml/m^2)	89 ± 14
LV-ESV(ml/m^2)	44 ± 21
LV-EF (%)	58 ± 6
LV Mass (gr/m^2)	62 ± 12
Positive T2-weighted images – edema (%)	100
Segments with LGE (%)	25
Segments mid-wall LGE (%)	6
Segments with epicardial LGE (%)	94
in inferior wall (%)	22
in anterior wall (%)	7
in anterolateral wall (%)	27
in inferolateral wall (%)	38
in anterior and inferior septum (%)	6

LV = left ventricular; EDV= end-diastolic volume; ESV= end-systolic volume; EF= ejection fraction; LGE= late gadolinium enhancement

Table 4

Analysis of ENDO and EPI Longitudinal, Circumferential and Radial function in controls and acute and follow-up myocarditis.

	Control Group (n = 25)	**AM group (n = 35)**	**FU group (n = 26)**	**ANOVA Global P**	**Paired two-sample P value AM vs FU groups**
Longitudinal Function					
Global ENDO S	- 24 ± 1.1	- 19.2 ± 3.1*	- 20.8 ± 5.4*	0.001	0.107
Global EPI S	- 19.7 ± 6	- 20.6 ± 3.4	- 22.6 ± 4.6	0.11	**0.02**
Circumferential Function					
Global ENDO S	- 21.7 ± 2.3	- 21.4 ±	- 23.3 ± 4.9	0.25	**0.001**
Global EPI S	- 9.1 ± 2.2	- 8.9 ± 2.2	- 10.7 ± 2.3*§	0.009	**<0.0001**
Radial Function					
Global Radial S	28.3 ± 3.9	28.1 ± 9.4	29 ± 9.1	0.76	0.088

ENDO S = endocardial strain; EPI S = epicardial strain.

* P <0.05 vs Control group

** P <0.001 vs Control group

§ P <0.05 vs AM group

§§ P <0.001 vs AM group

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