

Assessment of cardiac abnormalities in Duchenne's muscular dystrophy by ^{99m}Tc -MIBI gated myocardial perfusion imaging

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Abstract

Cardiac abnormalities in Duchenne's muscular dystrophy (DMD) are often detected in adult patients and their early detection is warranted. Studies have suggested that myocardial damage may be detected by cardiovascular magnetic resonance imaging or by echocardiography at the early stage of DMD. We aimed to identify early changes of cardiac abnormalities in children with DMD by technetium 99m-methoxyisobutylisonitrile (^{99m}Tc -MIBI) gated myocardial perfusion imaging (G-MPI). Forty-three boys aged 3 to 14 years (mean age 8.2 ± 3.6 years) with DMD and 12 age-matched normal boys as control were studied by G-MPI. These patients were at early stage according to previous studies on DMD. Uptake of ^{99m}Tc -MIBI in 7 regional walls and 17 segments of the left ventricle were visually analyzed. Quantitative gated single photon emission tomography (QGS-SPET) analysis of myocardium was performed to evaluate left ventricular function (LVEF). Gated myocardial perfusion imaging revealed cardiac abnormalities in 81.4% of all patients. Regional perfusion decrease involving multiple walls of LV was present. Four of the patients demonstrated mild abnormalities (11.4%), 7 moderate (20.0%) and 24 severe abnormalities (68.6%). Evident LV ejection fraction (EF) decrease ($42.1\pm 6.4\%$) and dilation with globally poor perfusion were found in three patients, aged 10 to 14, which had significant difference compared with the control group ($EF=58.4\pm 4.7\%$, $P=0.001$). The rest cases, aged 3 to 9 years, had normal LVEF. In conclusion, from the 35 cases of DMD patients (aged 3 to 14 years), regional myocardial perfusion decrease was detected in multiple walls by ^{99m}Tc -MIBI G-MPI at an early stage, while left ventricular function decrease (3/35, 8.6%) appeared late at about 10 years of age or older as compared with the control group in this study.

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Introduction

Duchenne's muscular dystrophy (DMD) is an X-linked recessive disorder that affects approximately 1 of 3,500 male births and is caused by the absence of dystrophin, a sarcolemmal protein of the skeletal and cardiac muscle cells [1]. Although the most characteristic symptom in DMD patients is severe skeletal muscle weakness from early childhood, the most important and fatal symptom is cardiac involvement. Cardiac involvement is characterized by degeneration and severe fibrosis, found by endomyocardial biopsy and postmortem examination [2-3]. Dilated cardiomyopathy (DCM) and heart failure also frequently occur. Early pre-symptomatic manifestations of myocardial abnormalities although important often go unrecognized due to lack of classical signs and symptoms. In this study we used technetium 99m-methoxyisobutylisonitrile (^{99m}Tc -MIBI) gated myocardial perfusion imaging (G-MPI) optimizing the schedule of treatment.

Nuclear medicine has played for years a major role in research of cardiac involvement in patients with heart diseases. It has been reported that myocardial fibrosis can be evaluated by ^{201}Tl myocardial imaging in patients with DMD [4-5]. An extensive search of methods in new technology development that are capable of detecting myocardial abnormalities has been observed. Gated myocardial perfusion imaging by ^{99m}Tc -MIBI is suggested as a precise and reliable method for evaluating myocardial abnormalities [6]. The combined information of perfusion and function can improve accuracy for detecting myocardial abnormalities [7]. This method may potentially be helpful in dealing with DMD although this has not been reported up to now [8]. Myocardial perfusion imaging has been often used to evaluate myocardial ischemia and cardiomyopathy [9]. Studies on myocardial perfusion abnormalities with DMD, which were commonly seen by thallium-201 chloride (^{201}Tl)-SPET perfusion imaging, have not been reported by ^{99m}Tc -MIBI G-MPI [4, 10].

Thus, the aim of this study was to determine whether ^{99m}Tc -MIBI G-MPI enables us to detect any changes in myocardial properties in patients with DMD at the early stage of the disease.

Cardiac involvement is an important component of DMD, usually in the form of DCM [11]. However, most DMD patients remain asymptomatic for years in spite of the progression of cardiac dysfunction because of their limited daily activities. Early detection of latent myocardial involvement and early use of drugs would be beneficial for delaying progression of heart failure in DMD patients. Therefore, it is important to identify patients at high risk of development of DCM and heart failure by using a noninvasive method. This study attempts to evaluate myocardial abnormalities in the early stage of DMD by using the method of ^{99m}Tc -MIBI G-MPI.

Subjects and methods

Patients and study design

We enrolled 43 DMD children aged 3 to 14 years (mean age 8.2 ± 3.6 y) and 12 age-matched controls (aged 3 to 13y, mean age 7.3 ± 2.2 y, $P = \text{NS}$) between September 2008 and March 2011. The control and the study groups were taken from the neuromuscular disorders department of the 3rd Hospital of Hebei Medical University (Hebei province, China). Written informed consent was obtained from the parents in all cases. In all patients, the diagnosis of DMD was confirmed by biceps brachial biopsy of the left arm, which showed dystrophin deficiency on the sarcolemma of skeletal muscle, by immunohistochemical stains using anti-dystrophin monoclonal antibodies. The inclusion criterion for patients with DMD was a firm diagnosis of muscular dystrophy with DMD, based on clinical characteristics, serum creatine kinase levels, electromyograms and muscle biopsy. The exclusion criteria included the presence of valvular heart disease, left ventricular hypertrophy, or other systemic diseases in addition to the muscular dystrophy.

All patients underwent full clinical and biochemical characterization, including skeletal muscle biopsy for immunohistochemistry. A cardiac history and examination were

undertaken, height and weight were measured, from which body surface area (BSA) was calculated, and G-MPI and left ventricular function recordings were taken.

The range of the myocardial abnormalities was analyzed by 7 regional walls (apex, anterior wall, anteroseptal wall, inferoseptal wall, inferior wall, inferolateral wall, and anterolateral wall); and the degree of the myocardium abnormalities was analyzed by 17 segments based on the polar maps [12]: using basal, mid-cavity, and apical as part of the actual definition of the location of the abnormality along the long axis of the ventricle from the apex to base. The basal and mid-cavity slices were divided into 6 segments of 60° each. The circumferential locations in the basal and mid-cavity were: anterior, anteroseptal, inferoseptal, inferior, inferolateral, and anterolateral. The remained segments were: apical anterior, apical septal, apical inferior and apical lateral. The 17 segments are presented in Figure 1.

Data collection

Ninety minutes after ^{99m}Tc -MIBI (^{99m}Tc , HTA Co., Ltd; MIBI, Beijing Shihong Pharma Centre, China) was injected intravenously, resting data were acquired over a 180° arc (20s 36 steps, 140KeV peak, window width: $\pm 20\%$) from a 45° right anterior oblique position to a 45° left posterior oblique position by a 2-head SPET (Infinia Vc Hawkeye, GE, USA) with low-energy-high-resolution collimators (64X64 matrix, 32 projections over 180° , 8 frames per cardiac cycle, 30s per projection). A zoom factor of 2.3 was used. The pixel size was 5.4mm. Short axial, horizontal longitudinal and sagittal longitudinal tomograms were reconstructed with a Butterworth filter (cutoff frequency, 0.45), and SPET images and polar maps were generated from the short axial images. No attenuation or scatter correction was applied.

To determine the range and the degree of myocardial abnormalities, the SPET and polar maps were visually inspected by two experienced nuclear medicine physicians who were blinded to patients' clinical data.

Left ventricular end-diastolic volume (LVEDV), end-systolic volume (LVESV) and EF, which were used to estimate LVF, were calculated by means of the commercially available software packages: Quantitative gated SPET (QGS). The EF was compared with the echocardiography EF values. End-diastolic volume index (LVEDVI) and end-systolic volume index (LVESVI), which considered BSA were also used in our research.

The perfusion, motion, and thickening scores were expressed as summed rest score (SRS), summed motion score (SMS), and summed thickening score (STS), respectively. A 17-segment model of the left ventricle was used for SRS, with a 5-point scoring system for perfusion defect severity (0=normal uptake of ^{99m}Tc -MIBI; 1=mildly decreased uptake; 2=moderately decreased uptake; 3=severely decreased uptake; 4=absent uptake) [13]. A likewise scoring system was used for SMS (0: normal, 1: mild hypokinesis, 2: moderate hypokinesis, 3: severe hypokinesis, 4: akinesis, 5: dyskinesis) and STS (0: normal, 1: mildly impaired, 2: moderately impaired, 3: severely impaired, 4: absent thickening) as well [14]. If there was a disagreement regarding the score, a final consensus was reached after discussion.

The injection dose of ^{99m}Tc -MIBI (range, 148-740MBq) was determined by the weight of each one [15]. The zoom factor in the image acquisition and the cutoff frequency of the Butterworth filter in the process protocol were re-



- | | | |
|------------------------|-----------------------|---------------------|
| 1. basal anterior | 7. mid anterior | 13. apical anterior |
| 2. basal anteroseptal | 8. mid anteroseptal | 14. apical septal |
| 3. basal inferoseptal | 9. mid inferoseptal | 15. apical inferior |
| 4. basal inferior | 10. mid inferior | 16. apical lateral |
| 5. basal inferolateral | 11. mid inferolateral | 17. apex |
| 6. basal anterolateral | 12. mid anterolateral | |

Figure 1. Display of the 17 myocardial segments of the left ventricle on a circumferential polar plot

vised according to others [16]. The indices, EDVI and ESVI, which were the standardization of EDV and ESV by using BSA were calculated as follows: $EDVI=EDV(mL)/BSA(m^2)$, $ESVI=ESV(mL)/BSA(m^2)$

Statistical analysis

Clinical data, including age, are expressed as mean (\pm SD). Student's *t* test was used for comparison between the DMD groups and the control group. Man-Whitney U test and Kruskal-Wallis test were used to compare scores among different segments. A P value of <0.05 was considered statistically significant. Software SPSS 15.0 was used for statistical analysis.

Results

Comparability of groups

The median age was 6 (range 8.2 ± 3.6) for the DMD group and 7 (range 7.3 ± 2.2) for the control group. This difference in age was not statistically significant. Details of our study data are given in Table 1. The skeletal muscle biopsy was characteristic in the DMD group (Fig. 2).

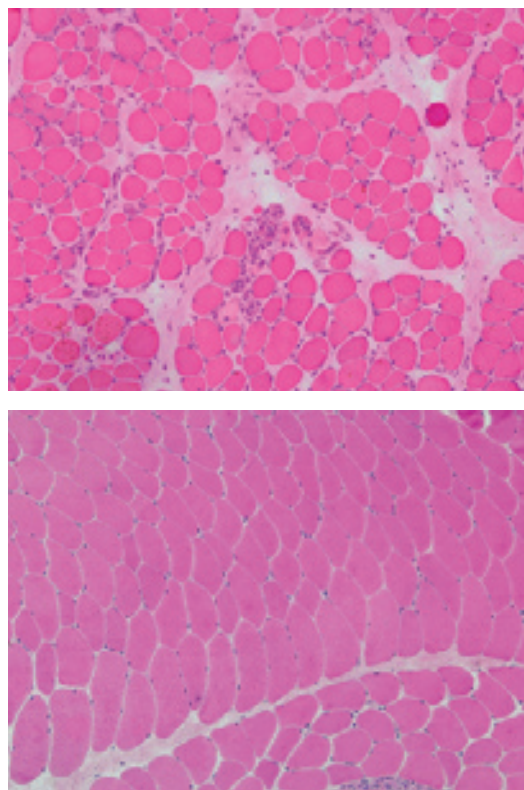


Figure 2. The typical image (left) of biceps brachial biopsy of the left arm in DMD, which was obviously different from the normal image (right). HE staining, X100

Table 1. Clinical data and the EF calculated by QGS and echocardiography in DMD and the control group

	DMD	Control
Number	43	12
Age (year)	8.2 ± 3.6	7.3 ± 2.2
Height (cm)	118 ± 13	104 ± 17
Weight (kg)	24 ± 8	21 ± 4
BSA (m^2)	0.87 ± 0.11	0.81 ± 0.16
Injection dose (MBq)	355 ± 81	316 ± 93
QGS EF (%)	56.2 ± 9.1	58.4 ± 4.7
ECHO EF (%)	62.4 ± 11.2	64.1 ± 8.6

BSA: Body surface area; ECHO: Echocardiography; EF: Ejection fraction; DMD: Duchenne's muscular dystrophy; QGS: Quantitative gated SPET.

The seven regional walls of G-MPI

The ^{99m}Tc -MIBI G-MPI results were abnormal in 35 of the 43 DMD patients and normal in 7. The rate of abnormalities was 81.4% (35/43). The range of myocardial abnormalities was as follows: 1 wall abnormality was described in 4 cases (mild 11.4%), 2 walls abnormalities in 7 patients (moderate 20.0%), 3 and more in 24 patients (severe 68.6%).

Seventeen segments of G-MPI

We used a half quantitative method of 17 segments and 5 points. In a total of 595 segments of 35 positive patients, 235 segments with abnormal distribution of the tracer within the myocardium were detected: 67 were characterized as mild decrease (1 point 28.5%), 93 segments were characterized as moderate decrease (2 points 39.6%) and 59 segments as serious decrease (3 points 25.1%). Sixteen segments had no distribution of the tracer (4 points 6.8%). The distribution and the score of 235 abnormal segments in 14 patients are given in Table 2.

The results of the distribution in the 235 abnormal segments as for the left ventricular walls were found in Table 3, which did not show significant difference. A typical image of DMD with perfusion defects is presented in Figure 3.

The most prominent abnormality of the myocardium was the wall thickening, followed by motion abnormalities and perfusion defects. The results were calculated for STS, SMS, and SRS as 6.3 ± 2.9 , 5.1 ± 3.4 , and 4.2 ± 2.3 , respectively. The QGS parameters between DMD and control groups are given in Table 4.

Left ventricular function

The results of the left ventricular function parameters are shown in Table 5. There was no significant difference between

Table 2. Two hundred and thirty five abnormal segments in 35 Duchenne's muscular dystrophy patients, 17-segment model by 5-point scoring system

Sum	%	Basal			Mid-cavity							Apical							
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	
1	67	28.5	3	3	5	3	3	5	4	2	6	8	3	4	5	4	2	3	4
2	93	39.6	4	6	5	7	6	4	7	6	6	5	7	5	4	3	5	7	6
3	59	25.1	4	4	4	3	2	3	3	4	3	2	5	3	2	5	4	3	5
4	16	6.8	1	0	1	1	1	0	0	1	1	1	1	1	1	2	1	1	2

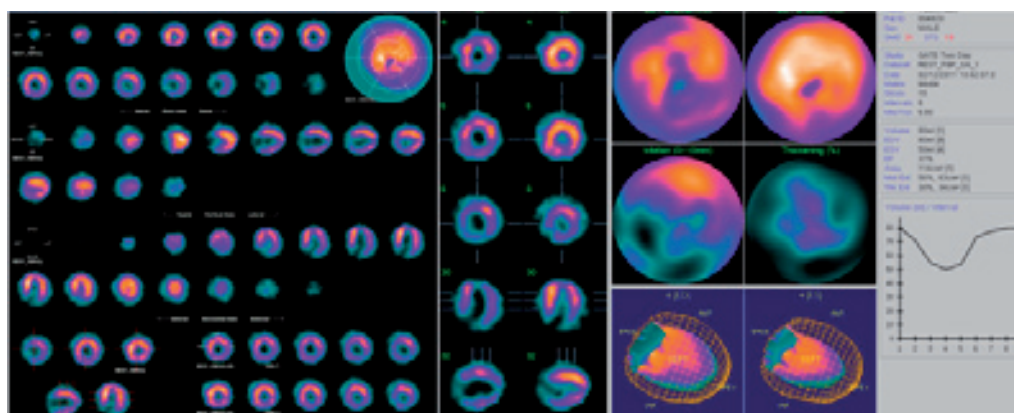


Figure 3. The typical images with respective perfusion defects of a 13 years old male patient in DMD. The ventricular cavity is larger than expected and perfusion defects were observed on the apex, inferior, and inferolateral walls. The semi-quantitative analysis of the patient's G- MPI revealed SMS and STS calculated as 31 and 19, respectively. The calculated ejection fraction value was 37%.

Table 3. Two hundred and thirty five abnormal segments in 35 Duchenne's muscular dystrophy patients, 17-segment model distributed in the seven regional walls

Wall	Anterior	Anteroseptal	Inferoseptal	Inferior	Inferolateral	Anterolateral	Apex
Abnormal segments	30	36	41	36	38	35	17

The distribution in the 235 abnormal segments as for the left ventricular walls did not show significant difference. Since the total segments number was significantly less than that of the other 6 walls, only 17 abnormal segments were observed in the apex.

Table 4. The QGS parameters in DMD and control groups

	DMD(%)	Control(%)	P value
EDVI	60.8±11.2	64.7±9.6	0.78
ESVI	21.3±7.5	25.2±9.4	0.67
SRS	4.2±2.3	0.4±0.3	0.001*
SMS	5.1±3.4	0.5±0.4	0.001*
STS	6.3±2.9	0.3±0.5	0.001*

EDVI: End-diastolic volume index; ESVI: End-systolic volume index; QGS: Quantitative gated SPET; SMS: Summed motion score; SRS: Summed rest score; STS: Summed thickening score

Table 5. Left ventricular function parameters in DMD, control groups and 3 patients (aged 10 to 14)

	DMD	Control	Three patients: 10-14y
EDV (mL)	57.6±13.8	61.2±9.3	69.2±11.4*
EDVI (mL/m ²)	60.8±11.2	64.7±9.6	71.1±10.2*
ESV (mL)	21.3±7.5	24.1±8.7	42.9±5.7*
ESVI (mL/m ²)	21.8±7.1	25.2±9.4	43.2±7.1*
EF (%)	56.2±9.1	58.4±4.7	42.1±6.4*

Three patients (aged 10 to 14) showed statistically significant difference compared with the control group (P<0.05).

the DMD and the control group for EDV, EDVI, ESV, ESV and EF. The calculated EF of G-MPI and corresponding echocardiography EF (%) were 56.2±9.1 and 62.4±11.2, respectively. Although G-MPI EF was lower than the ECHO EF, there was no

statistically significant difference between them.

Three patients (aged 10 to 14), with mild DCM, had EF decreased to 42.1±6.4%, EDV was 69.2±11.4mL, EDVI was 71.1±10.2mL/m², ESV was 42.9±5.7mL, ESVI was 43.2±7.1mL/m². They showed statistically significant difference compared with the control group (P<0.05).

The main findings in our study were that early changes in myocardial properties in DMD can be detected by ^{99m}Tc-MIBI G-MPI despite normal EF; and unlike the findings of previous studies, no definite regularity in distribution and no significant difference in extent were found in myocardial abnormalities.

Discussion

Perfusion abnormalities which were observed in most patients characterized by distributed diffusely and involved multiple ventricle walls. That was different from segmental spread in ischemic cardiomyopathy.

It has been reported that ²⁰¹Tl-SPET perfusion defects are frequently seen in the posterolateral wall of the left ventricle in patients with DMD [4-5]. Both the range and the degree of myocardial fibrosis were high in these studies but they were studied in a small group of patients, and the majority of cases were in the final stage of the disease. In the present study, more patients at the early stage of the disease were studied. Only three cases (aged 10 to 14 years) had obvious regional perfusion defects. This could indicate that perfusion abnormalities are related to the patients' age and the stage of the disease.

The mechanism for development of perfusion abnormalities in DMD is not well understood. Chronic progressive cellular death and fibrous tissue replacement have

been suggested for both skeletal and cardiac muscles loss. Studies have shown that dystrophin is integral to the structure and the stability of the cell membrane [17] and its associated proteins form a scaffold underneath the cardiomyocyte membrane connecting the intracellular cytoskeleton to the extracellular matrix. Dystrophin mutations may result in DMD and in X-linked dilated cardiomyopathy since localizes at the X chromosome. Dystrophin may also participate in the organization of ion channels and neurotransmitter receptors, which are necessary for cellular integrity [18]. These characteristics assure the role of dystrophin in maintaining structural integrity and force transmitting of myocytes.

Myocardial uptake and retention of ^{99m}Tc -MIBI involve passive diffusion across the plasma and the mitochondrial membranes [19]. Several studies have indicated that the cellular mechanism of ^{99m}Tc -MIBI metabolic exchange involves non-mediated diffusion across both plasma and mitochondrial membrane in response to negative trans-membrane potentials [20]. Mitochondrial membrane integrity is required for the intracellular binding of ^{99m}Tc -MIBI, and mitochondrial injury results in extracellular leakage of the tracer. Thus, ^{99m}Tc -MIBI could monitor the state of mitochondrial energetics as a fundamental aspect of tissue injury in addition to ischemia [19].

Perfusion abnormalities may exist early in the course of cardiomyopathy despite normal EF and would be progressive during the course of the disease, until cardiac dysfunction becomes more evident and can be detected. Comparison of QGS parameters (SRS, SMS, STS) showed statistically significant differences between DMD and the control groups. This finding showed that ^{99m}Tc -MIBI G-MPI might be valuable for the diagnosis and evaluation of myocardial damage or dysfunction, which is compatible with the results of a previous study [21].

These present results suggest that ^{99m}Tc -MIBI G-MPI may be useful for identifying early myocardial abnormalities in DMD. There have been few reports demonstrating early changes in myocardial features in asymptomatic patients with DMD studied by noninvasive methods like us [22-23]. This is the first study showing cardiac abnormalities in DMD by ^{99m}Tc -MIBI G-MPI.

These preliminary results must be confirmed in further studies involving a larger DMD population, and studying the influence of cardiomyopathy in mortality of the DMD patients.

In conclusion, this method is an important diagnostic tool in patients with cardiac abnormalities and for quantitative assessment of regional wall motion of the left ventricle in various heart diseases. The high-count density achieved by ^{99m}Tc -MIBI can lead to a higher quality of the myocardial perfusion images as compared to ^{201}Tl . An important advantage of G-MPI over other methods detecting myocardial viability is the possibility of obtaining simultaneously myocardial perfusion and function during one acquisition.

The authors declare that there are no conflicts of interest.

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Rembrandt von Rijn, 1632: Anatomy lessons by Professor Tulp.