Original Research

Differentiation of patients with two-dimensional echocardiography false positive non-compaction of ventricular myocardium by contrast echocardiography

Mingjun Tian, Minjuan Zheng*, Hui Ma, Yicui Niu, Ying Zhang and Pan Xu

Department of Ultrasound, Xijing Hospital, Air Force Medical University, Xi'an, Shananxi 710032, P.R China.

*Correspondence: zhengmj@fmmu.edu.cn (Minjuan Zheng)

Although echocardiography can be used to detect patients with non-compaction of the ventricular myocardium, it is often difficult to diagnose. In this study, the endocardium may be clearly visualized by contrast echocardiography to improve the diagnostic accuracy of patients with non-compaction of the ventricular myocardium. Twenty-four patients (n = 24) suspected with non-compaction of the ventricular myocardium Underwent transthoracic echocardiography including an intracardiac contrast echocardiography. The clinical data, Left ventricular opacification, and contrast echocardiography results were analyzed retrospectively. Twenty-four patients (n = 24) suspected with non-compaction of the ventricular myocardium were classified with transthoracic echocardiography and contrast echocardiography results into two groups: false positive and true positive. There were no significant differences in age, predisposing segments, Left Ventricular End-Diastolic Diameter, Left Ventricular End-Diastolic Volume, Left Ventricular End-Systolic Diameter, Left Ventricular End-Systolic Volume and ejection fraction between the two groups ($P > 0.05$). The thickness ratio of noncompacted to compacted myocardium (N/C) in the true positive group was significantly higher than that in the false positive group (3.47 ± 1.31 vs. 4.96 ± 1.28; $P < 0.05$). The range of non-compact myocardium in non-compaction of the ventricular myocardium patients can be observed clearly by Left ventricular opacification. Contrast medium in trabecular space and crypt is plentiful and ultrasonic contrast is more objective in measuring the thickness of dense myocardium. Two-dimensional echocardiography plays a characteristic role in the diagnosis of non-compaction of the ventricular myocardium; however, some suspected patients were observed to be false positive. Left ventricular opacification can greatly improve the clarity and accuracy of the endocardial margin by enhancing left ventricular imaging, displaying the true dense and non-dense layers, and improve the accuracy of ultrasonic diagnosis of non-compaction of the ventricular myocardium. The purpose of this paper was to explore the applied value of contrast echocardiography for heart diagnosis.

1. Introduction

Non-compaction of ventricular myocardium (NVM) is a congenital heart disease caused by abnormal development and maturation of the ventricular myocardium during embryonic period. The main reason is that the process of compaction of ventricular myocardium is disturbed and stagnated, which leads to abnormal structure and function of myocardium. Pathological sections show that the inner layer of the diseased myocardium is composed of a large number of loose muscle trabeculae. The abnormal trabeculae and deep recesses between trabeculae form a loose grid or spongy structure, which are also known as the myocardial sinusoidal status, spongy myocardial degeneration, or embryonic myocardium (Menendez-Montes et al., 2016). The diseased region of non-compaction of ventricular myocardium usually involves the left ventricle; however, it will seldom involve the right ventricle, both ventricles, interventricular septum, or inferior wall. Isolated non-compaction of ventricular myocardium is due to malformation of the heart, which involves a single ventricle and associated without other types. The myocardial trabecula and trabecular space, which contain a loose spongy structure, are formed during the first four weeks of normal embryonic heart development. At the time of the fifth week, the ventricles, apex, epicardium, endocardium, and the formation of the coronary arteries takes place. If compaction of the ventricular myocardium is obstructed, then the development and maturation of the myocardial trabecula will be blocked. This development will lead to a persistent recess between the myocardial trabeculae that will eventually form the non-compaction of ventricular myocardium.

Non-compaction of ventricular myocardium was first reported by Chin et al. (1990). where most NVM cases reported were sporadic. As more studies observed NVM, more family clustering NVMs have been reported. Genomic analysis revealed that some NVMs were genetically related to the family. The American Heart Association (AHA) classified them as hereditary cardiomyopathy (Maron et al., 2006). In 2008, ESC (the European Society of Cardiology) considered that although there were NVMs related

to family clustering, most cases were sporadic and related to some gene mutations in autosomal chromosomes. The NVM is still classified as unspecified cardiomyopathy. The patients with NVM are clinically diverse and lack specificity. Most patients, including the middle-aged and elderly, have no symptoms during the early period. Varying severities of NVM range from asymptomatic to progressive cardiac function deterioration, congestive heart failure, arrhythmia, embolic events, and sudden death. The onset of NVM is insidious with prognosis of patients with NVM being poor and the mortality rate high. NVM clinical symptoms are easily missed and often times misdiagnosed. With the continuous progress of imaging technology and deepening of understanding, the diagnostic rate of NVM has been greatly improved. Although it is congenital dysplasia, the average time from the onset of clinical symptoms to diagnosis is about 3.5 years (Towbin et al., 2018). Presently, it has not been widely accepted by clinicians as a diagnostic method or gold standard for NVM.

Iconography examination can provide valuable left ventricular morphological information. Ultrasonic examination can show an enlarged left ventricular cavity and decreased motion. The innumerable thick muscle trabeculae are intricately arranged in the ventricle from the middle of the interventricular septum to the apex of the heart. Recesses of varying sizes can be found between them, and muscle trabeculae and recesses can form a reticulated structure. Compact myocardium is thinner than normal myocardium. Color doppler imaging can detect blood flow with low velocity in crypt, which communicates with the heart cavity (Carminati et al., 2018). Although MRI and 201TI myocardial imaging can provide evidence for the diagnosis of NVM, ultrasound is a simple and effective imaging method that has been widely used in clinical diagnosis of NVM because it is composed of many muscle trabeculae and deep trabecular recesses (Maron et al., 2006; Alkar et al., 2018). Due to the limitation of resolution, conventional echocardiography is sometimes difficult to distinguish the anatomical structure of myocardial trabecula and recess. It is often difficult to distinguish the anatomical structure of the myocardial trabecula and recess andobtain satisfactory ultrasonic images when patients are obese. In this population, it is easy to to misjudge the non-dense myocardium layer, which leads to a decrease in diagnostic accuracy of TTE in patients with NVM. After LVO, the left ventricular endocardial structure images of the undiagnosed NVM are improved significantly, which also greatly improved the imaging quality of the patients' echocardiographic images. By enhancing the display of endocardial edges, the myocardial trabeculae could be accurately displayed and the endocardial boundary could be observed (Hussein et al., 2015; Towbin et al., 2018).

Although general echocardiography has high sensitivity and specificity in the diagnosis of NVM. Reports that the missed diagnosis rate of two-dimensional echocardiography in children with NVM is more than 50% (Jacquier et al., 2010; Ichida et al., 1999). Therefore, LVO improves the image quality of NVM, which is more advantageous than two-dimensional ultrasound in NVM identification (Nagueh et al., 2016; Song, 2008).

2. Materials and methods

Twenty-four patients (n = 24) suspected with NVM were selected from Xijing Hospital, First Affiliated Hospital of Air Force Military Medical University from 2016 to 2018. Transthoracic echocardiography showed several large muscle trabeculae, which were highly suspected of NVM. LVO suggested that contrast medium filled the crypt formed by left ventricular trabeculae and met the diagnostic criteria of NVM (Hook et al., 1996; Hussein et al., 2015). The outer layer of the diseased ventricular wall is compact myocardium, while the inner layer is noncompact myocardium. At the end of systole, the thickness of subendocardial noncompact myocardium was twice as thick as that of subepicardial noncompact myocardium. Different size recesses can be seen between the intricate muscle trabeculae. Color Doppler flow imaging can detect blood flow with low velocity in the recess or compare contrast medium with the heart cavity.

Twenty-four eligible NVM patients were included after routine ultrasonography and LVO (n = 19 males and n = 5 females; ages 18-40 years). Patients with basic heart diseases (confirmed by angiography), congenital heart disease, and cardiomyopathy)were excluded. Patients over 18 years old with clinical symptoms of chest tightness and chest pain, and who were willing to make a definite diagnosis by 2D echocardiography for further treatment were included. Contrast echocardiography was performed with patient consent.

In order to reduce measurement error, the blind method was used to measure the ultrasonic images. According to the guidelines outlined by the American Society of Echocardiography, 17 myocardial segments utilizing the wall images of conventional ultrasound and LVO echocardiography were (segmentation method) evaluated (Li et al., 2018). According to the 17-segment method of left ventricle, the number of noncompact segments of 2D echocar-diography was determined independently by an attending physi-cian and a deputy chief physician in the Department of Echocar-diography. Patients with NVM diagnosed by routine ultrasonogra-phy and those with negative contrast echocardiography were clas-sified in the false-positive group. There were 12 males and 4 fe-males aged 43.08 ± 14.66 years. Patients with NVM diagnosed by routine ultrasonography and those with negative contrast echocar-diography were classified as positive group. There were 8 cases (7 males and 1 female) confirmed in the group. There was no sig-nificant difference in gender and age between the two groups ($P > 0.05$).

Contrast echocardiography of left ventricle was done utilizing Philips EPIQ 7C color Doppler ultrasonic diagnostic instrument. It has S5-1 probe and frequency is 1-5 MHz. In routine 2D echocardiography, S5-1 probe and 1-5 MHz frequency was used. Changes in heart rate and blood pressure were closely observed before and after LVO in both groups ($P > 0.05$).

2.1. Measurement of echocardiography

Before LVO, real-time three-dimensional echocardiography was used to determine the end-diastolic and end-systolic phases. ECG was used to determine the position of the mitral annulus and left ventricular apex, respectively. The software automatically outlines the endocardium and manually adjuststo fit the actual boundary. LVEDV (Left Ventricular End-Diastolic Volume), LVESV (Left Ventricular End-Diastolic Volume), LVEF (Left Ventricular Ejection Fraction), LVSV (Left Ventricular stroke volume) and CO (Cardiac Output) were measured. The filling of contrast medium in the left ventricular cavity was observed in apical four-chamber,
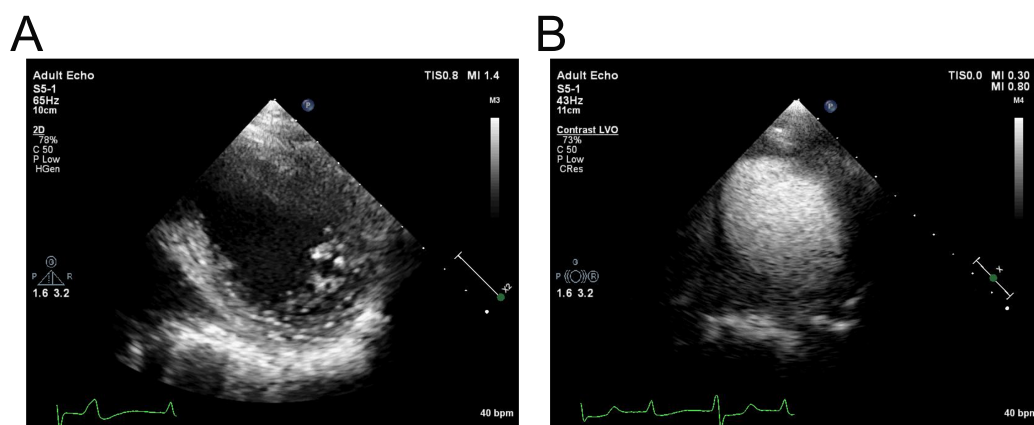
Figure 1. False-positive groups (A) Transthoracic echocardiography (B) Acoustic imaging
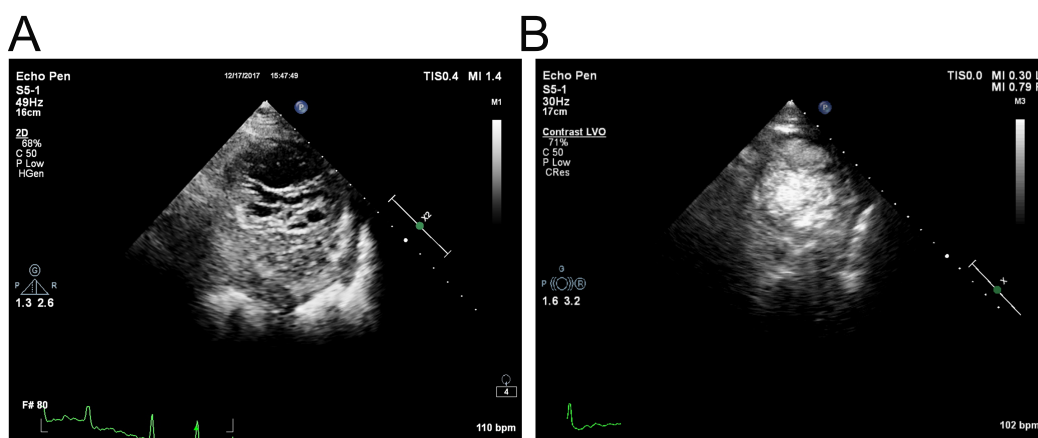

Figure 2. NVM-positive groups (A) Transthoracic echocardiography (B) Acoustic imaging

two-chamber, three-chamber, and left ventricular short-axis sections. The ratio of noncompact layer to compact layer (N/C) was measured in diastolic pseudo-positive group and confirmed group, respectively.

2.2. Statistical methods

SPSS 19.0 software is used and the measurements were expressed by mean and standard deviation. Parameters of echocardiography (LVEDV, LVESV, LVEF, LVSV, CO) were measured by paired t-test in false positive group and confirmed group. The detection rate of non-compaction of ventricular myocardium in different imaging modes was detected by chi-square test, $P < 0.05$. There was statistical significance between the false positive group and confirmed group ($P < 0.05$).

3. Result and Discussion

Patients treated with chest tightness and pain with a doubtful diagnosis NVM in cardiology department of our hospital. If patients have already performed cardiac ultrasonic contrast or cardiac magnetic resonance, which are considered as NVM patients, and want to come to our department to confirm their diagnosis, they have to talk and sign the informed consent before cardiac ultrasonic contrast.

Materials: There are 16 cases in false positive group (12 males and 4 females). There are 8 cases (7 males and 1 female) confirmed in the group. Both groups were mostly male (71% and 83%, $P > 0.05$). The average age of onset was younger in both groups, but there was no statistical significance ($P > 0.05$).

Contrast echocardiography of left ventricular function changes in false-positive and confirmed NVM groups (Table 1). There were no significant differences in left ventricular diastolic, end systolic (diameter and volume), and EF between the two groups ($P > 0.05$).

Left ventricular structural changes in two groups: After LVO examination, contrast medium filled the left ventricular cavity in both NVM-positive and false-positive groups. In false-positive group, smooth and continuous wall intima was observed and could be clearly distinguished from the left ventricular cavity (Fig. 1). In the NVM confirmed group, the left ventricular multiple enlarged trabeculae were filled with contrast echocardiography, which is showed deep lacunae (Fig. 2). No allergic symptoms were found in twenty-four patients by contrast echocardiography. After LVO, the clearness of the endocardial boundary was improved, and the identification of ventricular septum and left ventricular lateral wall was enhanced. The contrast medium was injected into the recess between trabeculae. The muscle trabeculae and recess were easily observed. It was observed that the endocardium was incomplete and honeycombed. The muscle trabeculae showed a small filling

Table 1. Index comparison table

	Age	ESV (ml)	EDV (ml)	EF (%)	EDD (mm)	ESD (mm)
fase-positive groups	29.50 ± 8.735	76.33 ± 16.133	129.67 ± 19.99	39.67% ± 7.84	56.75 ± 4.77	45.75 ± 6.20
NVM-positive groups	43.08 ± 14.66	63.75 ± 35.04	124.92 ± 40.66	51.54% ± 10.80%	53.23 ± 9.58	42.42 ± 8.96

defect near the apex of the left ventricular cavity.

Predilection site: According to the left ventricular ASE17 segment method, the partial compact myocardium in the false positive group and the confirmed group occurred in the lateral and posterior walls below the level of the low papillary muscle in patients with NVM (i.e. 10, 12 and 16 segments (100% *vs.* 100%), 10 and 15 segments were more common. There was no significant difference between the two groups in the predisposing sites (57% *vs.* 50%, $P > 0.05$).

Results of contrast echocardiography: The N/C ratio in the confirmed group was significantly higher compared to the false positive group (3.47 ± 1.31 *vs.* 4.96 ± 1.28). The N/C ratio under contrast echocardiography had statistical significance ($P < 0.05$).

3.1. Pathogenesis and clinical characteristics of NVM

Non-compaction of the ventricular myocardium, also known as cavernous myocardium or myocardial sinusoidal space persistence, is characterized by large reticular trabeculae and deep intertrabecular recesses connected with the left ventricular cavity. The main clinical manifestations are heart failure, arrhythmia and embolism events. At present, the clinical manifestations and initial age of patients are different making the diagnosis difficult. It is often difficult to diagnose NVM when heart failure occurs. Recent studies have found that mutations in genes encoding dysrtobrevin and Cypher/ZASP may lead to the failure of left ventricular densification in the embryonic stage, which may lead to NVM. Similar genes are also present in dilated cardiomyopathy. Therefore, we can explain why these two types of myocardial lesions are more common in both the left and right ventricles. All 8 lesions in this group contained these characteristics. The earliest reports of NVM are mostly in children; however, in recent years, reports of adult morbidity are increasing. Although NVM is a congenital disease, its initial symptoms vary greatly in age, ranging from birth to greater than 70 years. Some patients remain asymptomatic for many years, butsome patients die of severe heart failure at the early stage of symptoms that are mainly related to the extent of non-dense myocardium and the degree of chronic ischemia. Three major clinical manifestations of NVM are progressive cardiac dysfunction, systemic thrombosis, and arrhythmia. Other clinical manifestations and complications reported in the references include sudden death, special face, cyanosis, and growth restriction. Cardiac enlargement can be found during physical examination and cardiac murmur can often be found during cardiac auscultation. It is very important to diagnose by imaging, family history, laboratory examination, electrocardiogram, echocardiography, and cardiac magnetic resonance.

The number of suspected patients with NVM is increasing. The onset age is younger, mostly male, but there are no significant differences between the false positive group and the confirmed NVM patients in regards to left ventricular function changes, left ventricular diastolic, end systolic (diameter and volume) increase, and EF

decrease($P > 0.05$). This may be related to the insufficient sample size; however, this study found that the diagnostic rate of suspected NVM patients with clinical symptoms may be improved by combining contrast echocardiography. Therefore, for suspected patients with NVM, priority should be given to myocardial contrast to assist in diagnosis, in order to improve the clinical prognosis and achieve the purpose of early treatment.

3.2. Advances in the application of cardiac ultrasonic contrast in the diagnosis of NVM

Echocardiography examination is simple and economical compared to other imaging examinations. It is of great value to patients with non-compaction of ventricular myocardium. Relatively accurate identification of myocardial structural abnormalities in NVM is currently the preferred and best imaging method for diagnosis of NVM (Sun et al., 2019; Arenas et al., 2018; Xu et al., 2018). However, when some patients have poor sound transmission conditions and cannot obtain satisfactory echocardiography images, the accuracy of ultrasonic evaluation of NVM is significantly reduced. Transthoracic echocardiography is difficult to diagnose especially in suspected patients with NVM. Combined with contrast echocardiography, the endocardium can be clearly displayed and the diagnostic rate of NVM can be improved (Lufundo et al., 2019; Arenas et al., 2018; Friedberg, 2018). In this study, LVO was performed in suspected patients with NVM to be diagnosed by TTE. The characteristics of echocardiography and contrast echocardiography in two-dimensional suspected patients with NVM were analyzed. The purpose was to evaluate the diagnostic value of contrast echocardiography in two-dimensional false-positive patients with NVM and to provide evidence for improving the accuracy of NVM. Compared with two-dimensional ultrasound, contrast echocardiography has better correlation and less difference between observers.

This paper utilizes Jenni's standard: (1) Do not merge other congenital or secondary cardiomyopathy; (2) The ventricular wall is divided into two layers. The thinner densification layer and the thicker non-densification layer with reticular trabeculae and crypts, and the thickness ratio of noncompacted to compacted myocardium at the end of systole is greater than 2; (3) The lesions are mainly located at the apex, inferior wall, and lateral wall, rarely involving the basal part (Roberts et al., 2014). However, in recent years, it has been found that end-diastolic measurements are more accurate, so the thickness ratio of N/C is measured during diastolic period. In this study, the thickness ratio of N/C is measured at the end of diastolic phase under the parasternal left ventricular short axis and four-chamber, two-chamber and three-chamber views under cardiac ultrasonic contrast and two-dimensional echocardiography.

In these cases, numerous deep trabecular recesses with sizes and with blood flow connected with the ventricular cavity can be seen by color doppler imaging. It is very helpful for 2D TTE to

differentiate other non-dense incomplete cases with color doppler technique; however, color Doppler echocardiography cannot be used in cardiac ultrasonic contrast. Color doppler echocardiography cannot be used to diagnose patients with NVM. Suspected patients with NVM can also show dark blood in the recess between the trabeculae of the myocardium connected with the cardiac cavity. Energy doppler could show low-velocity blood flow, but it is not directional. Its speed is not measurable. It has no advantage in the diagnosis of NVM patients. Cardiac ultrasonic contrast mode can display myocardium more accurately. Cardiac contrast echocardiography can increase the contrast of myocardial structure, improve the display rate of endocardium, and make the coarse trabecular structure and the recess between trabeculae clearer.

3.3. Prevalent segment of NVM patients

NVM is formed by the persistence of deep recesses between the trabeculae of ventricular wall protrusion and incomplete densification of cavernous myocardium. LVO examination can clearly show that the left ventricular function of NVM patients is significantly different from that of normal people. According to ASE17 segmental method, the incomplete densification segments of myocardium in the false positive group and the confirmed group all appeared in the lateral and posterior walls below the level of low papillary muscle, i.e. 10, 12 and 16 segments (100% *vs.* 100%), 10 and 15 segments were more common, and the possibly incomplete densification segments generally had a lower incidence. There was no significant difference in the predisposing sites between the two groups (57% *vs.* 50%, *p* > 0.05). This study showed that the ratio of diastolic noncompact layer to compact layer (N/C) in NVM patients after LVO was significantly higher than that in the false positive group (3.47 ± 1.31 *vs.* 4.96 ± 1.28). The results of this study confirm that LVO enhanced imaging of intracardiac structure is helpful to differentiate and diagnose suspected patients with NVM. Normal left ventricular apex may also have noncompact structure (Uribe et al., 2012). Depending on the ratio of noncompact myocardial thickness to subepicardial compact myocardial thickness, abnormal myocardial trabeculae may also occur, but the shape of normal myocardial trabeculae is different from that of NVM patients. In NVM patients undergoing intracardiac contrast echocardiography, contrast medium echoes can be seen filling the recess between the muscle trabeculae, which helps to enhance the display of the apex and recess. Intracardiac contrast echocardiography not only enhanced the myocardial trabeculae and recesses in apical region, but was also more useful in displaying the recesses. During intracardiac contrast echocardiography, contrast medium could be seen to be filled in the heart cavity, and the muscle trabeculae could not be shown to be filled. The recess was distinguished from the myocardial trabeculae and the heart cavity due to blood storage, and the block echoes of contrast agents were formed at the whole interface, so that the diagnosis of NVM could be clearer to make a diagnosis.

Previously, researches focused on normal healthy people and patients with NVM, compared with their 2D echocardiography and cardiac ultrasonic contrast results. It was found that cardiac ultrasonic contrast could improve the diagnostic rate of patients. In this study, patients have already performed 2D echocardiography and also considered as NVM; however, some patients find diagnostic errors after further cardiac ultrasonic contrast. They are false positive results. In some patients only muscle trabeculae increased and not NVM. These false positive patients are screened to improve the diagnostic accuracy of this disease.

4. Conclusion

LVO was used to improve endocardial imaging, which could clearly display the myocardial trabeculae and recesses in NVM patients with the help of contrast medium. In particular, the imaging features of the parallel arrangement of contrast medium filling areas with uneven echoes in the apex of left ventricle are helpful for the diagnosis and differential diagnosis of NVM, especially for the screening of first-degree relatives of NVM patients and the follow-up after treatment. Data published by the NVM Registry of the Italian Echocardiographic Association show that symptomatic patients have worse prognosis, while asymptomatic patients have better prognosis (Berti et al., 2018). Early ultrasonic screening and diagnosis of NVM is conducive to early intervention, symptomatic treatment, and prolonged life span of patients. With the rapid development of contrast echocardiography technology in recent years, the display of cardiac cavity structure in ultrasonic imaging becomes clearer, which greatly improves the diagnostic rate of NVM and provides important help for clinical diagnosis and treatment of NVM.

Conflict of Interest

The authors declare that there is no conflict of interest.

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