Gene 642 (2018) 159–162

Contents lists available at ScienceDirect

Gene

journal homepage: www.elsevier.com/locate/gene

Research paper

The pathogenic gene screening in a Chinese familial dilated cardiomyopathy pedigree from Hubei

Yongnan Lyu, Jingjing Chen, Hongxin Xu*

Department of Cardiology, Renmin Hospital of Wuhan University, Wuhan 430060, China

ARTICLE INFO

Keywords:
Familial dilated cardiomyopathy
Gene mutation
TTN

ABSTRACT

Dilated cardiomyopathy arises from mutations in many genes. TTN, the gene encoding the sarcomere protein titin, has been insufficiently analyzed for cardiomyopathy mutations because of its enormous size. In this study, we report a Chinese family with two members affected by TTN. Blood samples were collected from all family members. Genomic DNA was isolated from blood, and all coding exons and adjacent intronic sequences of the TTN gene were examined for mutation analysis using polymerase chain reaction (PCR)-based sequencing. The proband (III3) and his sister (III2) carry a TTN c.100126A > G (p.Thr33376Ala) missense mutation. The proband currently exhibits decreased cardiac function accompanied by malignant arrhythmia, and his sister has no obvious clinical symptoms and no abnormal ultrasound findings. The study found that there is a missense mutation in the TTN gene, c.100126A > G (p.Thr33376Ala), in a family whose members suffer from familial dilated cardiomyopathy in Hubei province. TTN is closely related to dilated cardiomyopathy and is an important causative gene of familial dilated cardiomyopathy.

Dilated cardiomyopathy (DCM) is a cardiomyopathy characterized by left ventricular or biventricular dilatation with systolic dysfunction. The main clinical symptoms are heart failure, arrhythmia and embolic stroke. Severe cases can even lead to sudden cardiac death (Franz et al., 2001). The 5-year mortality rate of patients with DCM ranges from 15% to 50% (Komajda et al., 1990). One study found that approximately 30% to 50% of patients with DCM have familial inheritance, in which case it is known as familial dilated cardiomyopathy (FDCM) (Mcnally et al., 2013; Dellefave and McNally, 2010). Patients with FDCM typically have an age of onset between 20 and 30 years, and they show rapid disease progression, poor quality of life, poor prognosis and high mortality (Gerull et al., 2006). At present, with the exception of heart transplantation, there is no effective treatment method available. Early diagnosis and intervention is an effective means of delaying the progression. FDCM shows significant heritability, and it is mainly caused by genetic mutations, but the clinical genetic features exhibit great heterogeneity. There are 65 causative genes that have been identified for FDCM, and they mainly have an autosomal dominant inheritance pattern (Hershberger et al., 2010). In the present study, a genetic screen was carried out for family members in a family exhibiting FDCM to identify potential causative genes for early screening of and intervention for DCM.

1. Methods

1.1. Ethics statement

Human blood samples were collected upon approval of the Ethical committee of Renmin Hospital of Wuhan University, China. Written informed consent was obtained from the participants, their parent, or legal guardian in accordance with the institutional requirements and the Declaration of Helsinki. All methods were performed in accordance with the relevant guidelines and regulations. This study was approved by the Ethics Committee of Renmin Hospital of Wuhan University, China.

1.2. Patients

The proband was diagnosed with DCM in April 2017 at Renmin Hospital of Wuhan University. The proband and his family members are from Daye City, Hubei Province.

1.3. General clinical data collection

The medical history and family history of the proband and his

Abbreviations: DCM, dilated cardiomyopathy; FDCM, familial dilated cardiomyopathy; ECG, electrocardiogram; BP, blood pressure; R, rhythm; P, pulse; T, temperature; HR, heart rate; LAD, left atrium diameter; LVDD, left ventricle diastolic diameter; RAD, right atrium diameter; RVD, right ventricle diameter; LVEF, left ventricle ejection fraction

* Corresponding author.
E-mail address: 303876340@qq.com (H. Xu).

https://doi.org/10.1016/j.gene.2017.11.001
Received 2 September 2017; Received in revised form 29 October 2017; Accepted 2 November 2017
Available online 03 November 2017

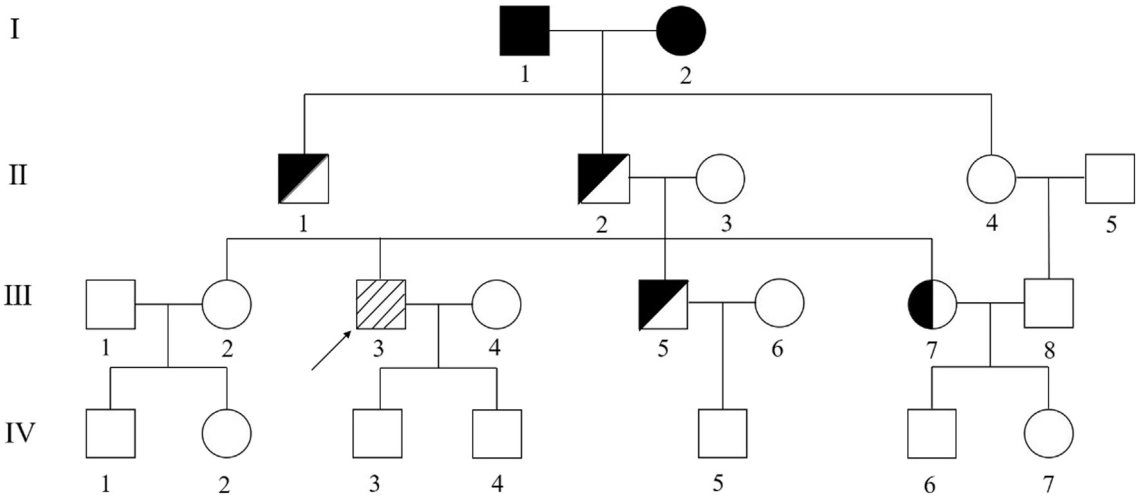

Fig. 1. The pedigree chart of the proband with dilated cardiomyopathy.

family members were recorded in detail. Based on the information provided by the proband, his family pedigree chart was plotted (Fig. 1). A physical examination, blood test, cardiac ultrasound and ECG examination were performed for the proband. Cardiac ultrasounds were performed for his family members.

1.4. Gene screening

The proband and his family members provided their written consent. The clinical molecular diagnostic center of People's Hospital of Wuhan University carried out high-throughput sequencing for the exons of all potential causative genes in the proband to identify suspicious mutations. Sanger sequencing was then performed to verify whether his family members carried the same suspicious mutations.

2. Results

2.1. General clinical information

The patient is male and 46 years old. In the past week, the patient experienced chest tightness, excessive panting and fatigue when he climbed stairs and performed intense activities. The discomfort was reduced after resting. The patient did not have other discomfort, such as coughing, hemoptysis, chest pain and dizziness, and did not receive any treatment. In the last two days, the patient experienced aggravated discomfort, with decreased mobility, and he could not sleep through the night in a supine position. Physical examination: BP 130/80 mm Hg, R 26 breaths/min, P 88 bpm, T 36.3 °C. The patient was alert with normal appearance. The skin and mucous membranes showed no bleeding nor yellow tinge. The superficial lymph nodes were not enlarged. Jugular vein distention and stiff neck were not observed. Breath sounds were normal, but a fine crackle could be heard on the lower left lung lobe. Left atrial enlargement, HR 88 bpm. Normal heart sound was observed with no obvious heart murmur. The abdomen was soft and showed no tenderness nor rebound tenderness. The liver and spleen were under the ribs, the kidney area was without percussion pain, and lower extremities were without edema. Auxiliary examination: cardiac ultrasound - LAD = 50 mm, LVDD = 62 mm, RAD = 41 mm, RVD = 21 mm, and LVEF% = 35%. Long-term electrocardiogram - normal sinus rhythm; average heart rate of 72 bpm; ventricular premature beats 2357, and non-sustained ventricular tachycardia, 32 arrays. The patients NT-proBNP level was 8734 ng/L.

2.2. General information of family members of the proband

I1 and I2 died of unknown causes. II1 and II2 were diagnosed with DCM and had passed away. III5 is a younger brother of the proband who died at the age of 26. III7 is an older sister of the proband who died at age 33. III2 is a younger sister of the proband, 43 years old, and does not currently have the disease; heart ultrasound: LAD = 26 mm, LVDD = 41 mm, RAD = 31 mm, RVD = 18 mm, and LVEF% = 60%. IV1–IV7 do not currently have the disease, and the cardiac ultrasound found no obvious abnormalities (Fig. 1).

2.3. Potential causative gene screening results

Genomic DNA was extracted from the peripheral blood of the subjects. An Ion PGM high-throughput sequencer (Life Technologies) was used to sequence 65 DCM related genes, such as ACTC1 and ACTN2. The sequencing results were analyzed using Ion Reporter software, and 583 mutations were identified. By referencing the ClinVar database, Common SNPs in the UCSC database, the dbSNP database and common mutations with ≥ 1% MAF in the 1000-genome-project, 5 deleterious or potentially deleterious mutations were identified after excluding synonymous mutations and intron mutations (Table 1). SIFT and PROVEAN were used to predict the harmfulness of those 5 mutations. Among them, the c.100126A > G (p.Thr33376Ala) variant is Deleterious/Damaging; the c.49762G > A (p.Val16588Met) variant is Neutral/Damaging; the c.17456G > A (p.Arg5819Gln) variant is Neutral/Tolerated; and the c.15758C > T (p.Thr5253Ile) variant is Deleterious/

Table 1
Gene mutation results of the proband.

Gene name	Transcriptions	Codon	Amino acids	Type of mutation	Heterozygous/homozygous	Mutation rate	Coverage
TTN	NM_001256850.1	c.100126A > G	p.Thr33376Ala	Missense mutation	Heterozygous	48.52%	1964
TTN	NM_001256850.1	c.49762G > A	p.Val16588Met	Missense mutation	Heterozygous	51.23%	851
TTN	NM_001256850.1	c.17456G > A	p.Arg5819Gln	Missense mutation	Heterozygous	49.56%	1715
TTN	NM_001256850.1	c.15758C > T	p.Thr5253Ile	Missense mutation	Heterozygous	46.98%	464
KCNQ2	NM_172107.2	c.1154_1155delT CinsCT	p.Ile385Thr	Missense mutation	Heterozygous	19.53%	129