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Case Report

Whole Exome Sequencing Identified the Causative Mutation in a 4-Year-Old Female with Mulibrey Nanism: A Case Report

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Abstract

Mulibrey Nanism is a rare multisystem disorder inherited in an autosomal recessive manner caused by mutations in the *TRIM37* gene. Most of the reported cases are from Finland, but this condition has rarely occurred in other countries. Although the clinical diagnosis of Mulibrey nanism is a challenge during the first months of life, the disease can be suspected clinically due to the distinctive features of the patients. A 4-year-old female with pneumonia, cardiomyopathy, growth retardation, peripheral edema, and characteristic craniofacial features was referred to Tehran Hope Generation Foundation Genetic diagnosis Center, in October 2021. Genomic DNA was isolated from peripheral blood samples of the patient and her parents and Whole exome sequencing was performed for the patient. Whole exome sequencing revealed a homozygous G>A splice site variant (*TRIM37*; c.370-1G>A). Sanger sequencing confirmed the segregation of the variant with phenotype in this family. Whole exome sequencing can be helpful in the diagnosis of the patients suspecting to Mulibrey nanism and lacking sufficient clinical presentation according to the diagnostic algorithm.

Keywords: Mulibrey nanism; Whole exome sequencing; *TRIM37* protein; Pericardial constriction

Introduction

Mulibrey (MUscle–LIver–BRain–Eye) nanism (OMIM #253250) is one of the extremely rare multisystem disorders in the world with prenatal onset, inherited by autosomal recessive mode of inheritance (1). Mulibrey nanism is characterized by occasional progressive cardiomyopathy, characteristic facial features, failure of sexual maturation, insulin resistance and an increased risk for Wilms tumor (2). Constrictive pericarditis, low birth weight, short stature, sever progressive growth delays, hypotonia, and hepatomegaly are the most signs and symptoms of the disease (3). It is caused by mutations in the human *TRIM37* gene located on chromosome 17q22-q23, which encodes the peroxisomal TRIM37 protein (4). The TRIM family proteins comprised more than 100 members and have several important roles in cellular functions. The TRIM family proteins are associated with various diseases including autoimmune and inflammatory disorders (5). The most

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reported cases with Mulibrey nanism are from Finland, however, there are some reports of sporadic cases from all over the world (6, 7). About 50% of the patients develop congestive heart failure and constrictive pericarditis, early diagnosis for this syndrome is important. Treatment of the disease includes surgery for constrictive pericarditis, medications for progressive heart failure, and hormone replacement therapy (8, 9).

Case presentation

A 4-year-old female born to healthy Iranian consanguineous parents originating from Kurdish ethnic group was referred to Tehran Hope Generation Foundation Genetic diagnosis Center, in October 2021. There was a history of pneumonia, cardiomyopathy, growth retardation, peripheral edema, and characteristic craniofacial features but without mental retardation in the patient. Cardiovascular evaluation using Magnetic Resonance study had been suggested constrictive pericarditis. CT-SCAN of the thorax also had revealed cardiomegaly, pericardial thickness, hepatomegaly, and mosaic lung attenuation. Molecular evaluation for Familial Mediterranean fever had been previously performed for the patient and no mutation was found in the related gene.

Informed consent and the consent of publishing the study results was obtained from the parents. The Medical Ethics Committee of the Hope Generation Fouandation permitted this study (Code No: IR.SBMU. MSP.REC.1396.792).

Differential diagnosis

After genetic counseling and evaluating the pedigree (Fig.1), an autosomal recessive disorder was suspected to the patient. Based on the patient past medical history, one of the probable diagnosis was Mulibrey Nanism. Therefore, due to the high number of the exons (24 exons) in the disease related gene (TRIM37), and to exclude other probable disorders, Whole Exome Sequencing (WES) was offered for the patient.

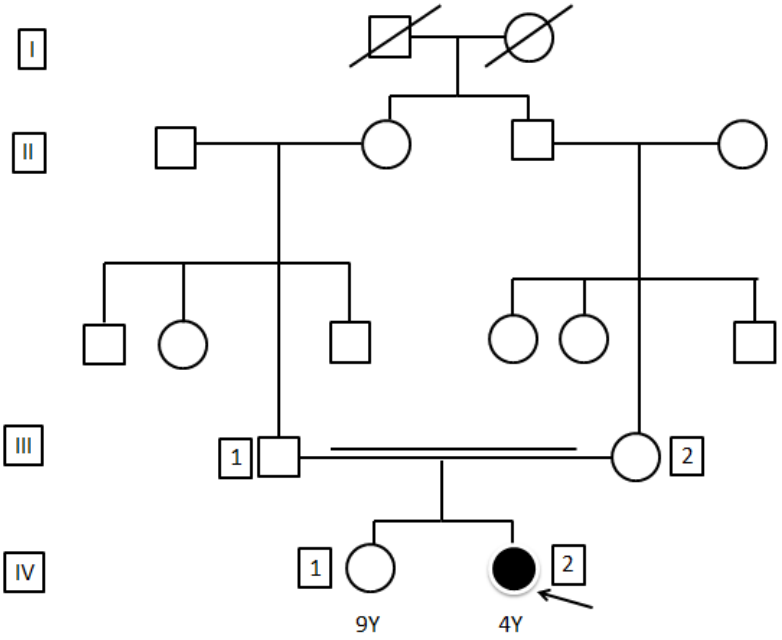

Fig. 1: The family pedigree showing an autosomal recessive pattern of inheritance. The proband has shown by the arrow

DNA Extraction and Whole-Exome Sequencing

Genomic DNA was isolated from peripheral blood samples of the proband and her parents using the salting out method. The proband genomic DNA was fragmented, and enriched for exome sequencing using Agilent SureSelect V6 kit. The libraries were sequenced to mean >90x coverage on an Illumina Novaseq 6000 platform. The initial sequencing component of this test was performed by the Illumina genome sequencing service in Macrogen (Seoul, Korea).

Bioinformatics Analysis of Whole-Exome Data

After aligning the obtained sequences to the human reference genome (GRCh37/hg19) using the Burrows-Wheeler- Aligner (BWA) program, variant calls are made using the Genomic analyzer tool kit (GATK) to identify variants relevant to the clinical indication. Gene annotation of the variants is performed using Variant Effect Predictor (VEP) program. Common variants are filtered based on the available information from databases (Including HGMD, ClinVar, 1000 Genome, ExAC, LSDBs, dbSNP and locally database, Iranome), published literature, clinical correlation and its predicted function.

Sanger Sequencing Validation and Segregation Analysis

Using bioinformatics filtering strategies and in silico analysis, a homozygous G>A splice site variant (*TRIM37*; c.370-1G>A) was detected in the patient. According to the ACMG guidelines for the interpretation of sequence variants, this G to A transition classified as likely pathogenic. This variant destroys the canonical splice acceptor site in intron 5 of *TRIM37* gene, in which loss of function is a known mechanism of the disease. In addition, most bioinformatics prediction tools are in favor of its pathogenicity. Segregation analysis in family members confirmed the cosegregation of the variant with the phenotype (Fig. 2).

Finally, for variant validation and segregation analysis in the family, polymerase chain reaction (PCR) was performed to amplify the targeted region using specific primers (F-5'CAGATGATTT-GGGCTGTGG3' and R-5'**TCTCTGACTCCAACTGCTTCC3'**). The PCR products were then subjected to Sanger sequencing on an automated ABI PRISM 3130XL (Applied Biosystems, USA).

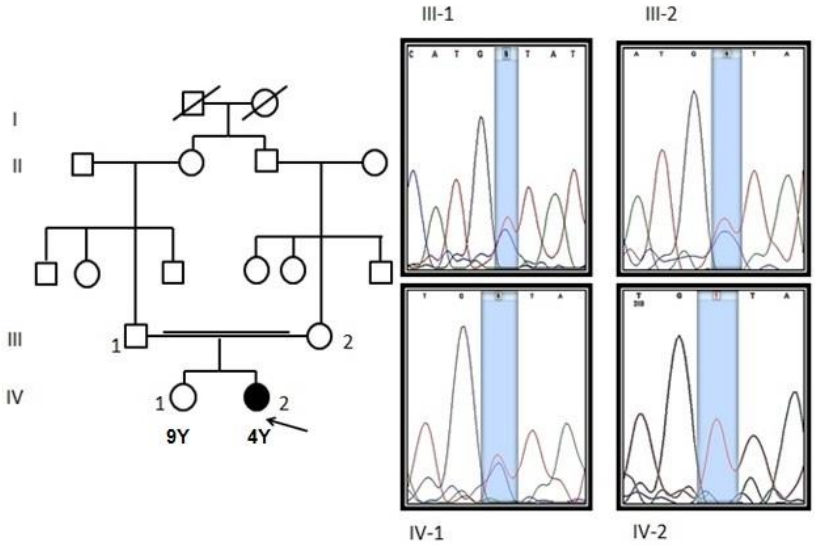

Fig. 2: The variant was segregated with the phenotype. The patient (IV-2) was homozygous for c.370-1G>A, and her parents (III-3 & III-4) as well as her healthy sister (IV-1) were in the heterozygous state

Discussion

Mulibrey nanism is an extremely rare genetic manifestation with unique clinical features. According to Karlberg and colleague's revised algorithm, diagnosis of Mulibrey nanism is based on the presence of major and minor clinical criteria (1, 10). Our case was a 4-year-old female with a history of pneumonia, cardiomyopathy, growth retardation, peripheral edema, and characteristic craniofacial features but without mental retardation. The patient was suspected to Mulibrey nanism and had a negative result for Familial Mediterranean Fever (FMF) at the time of referral, although, genetic analysis confirmed Mulibrey nanism in the patient. Based on our knowledge only 2 cases of Mulibrey nanism have been reported in Iran to date, but none of them were genetically confirmed (11, 12). For some referrals, limited phenotypic information makes it difficult to have an accurate diagnosis. Genomic technologies not only provide diagnoses, but they can cause essential alteration to the patient management. WES is now extremely strong and affordable. It is becoming more suitable and cost-efficient to prefer WES to other methods of screening, particularly in rare diseases with a recessive pattern (13). Using WES in a Mulibrey nanism suspected patient, we found a new splice site variant (*TRIM37*; c.370-1G>A) and give a definite molecular diagnosis for the patient. Parents were heterozygote for the variant, so, this family has a 25% chance for having an affected child in every future pregnancy without prenatal testing.

Conclusion

This study highlighted the potential of clinical WES in the diagnosis of rare genetic diseases including Mulibrey nanism. Its early use in the diagnostic strategy reduces the time to diagnosis of the disease.

Journalism Ethics considerations

Ethical issues (Including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

Acknowledgements

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Conflict of interest

The authors declare that there is no conflict of interest.

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