

Pangenia Genomics' tailor-made genetic testing service finds a potential genetic cause of mitochondrial myopathy bewildering a family for eight years

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Pangenia Genomics offers tailor-made genetic testing service to meet customers' needs across different diseases, genetic causes and family structures. In this series, we will share stories of families that we helped with by employing different testing methods and customized counselling service.

Problem

Eight years ago, at the age of 39, Keith was admitted to hospital for generalized weakness and swallowing difficulty that lasted for a few weeks. His condition deteriorated rapidly, complicated with pneumonia and respiratory failure that required intubation and mechanical ventilation. Three days later, he was transferred to the Intensive Care Unit

(ICU). Examinations further revealed ptosis (drooping eyelid), and severe muscle wasting. Neuromuscular disease was suspected. Subsequent muscle biopsy showed accumulation of abnormal mitochondria in muscle cells, indicating **mitochondrial myopathy**.

Mitochondrial myopathy (1) is a group of muscular diseases caused by genetic mutations in either nuclear DNA or mitochondrial DNA, which result in defective mitochondria. Mitochondria are small organelles inside almost all the cells, responsible for generating energy. When mitochondria have defects, the cells with high energy needs, such as muscle and nerve cells, are most severely affected. The underlying genetic causes are various. In Keith's case, doctors initially had two suspects: the *TYMP* gene, the cause of MNGIE (mitochondrial

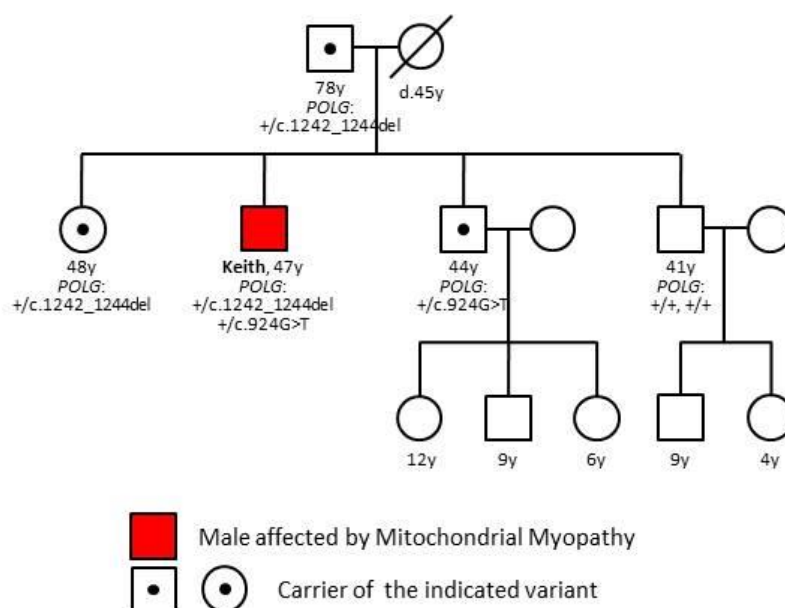


Figure 1 Pedigree and genotypes of Keith's family.

neurogastrointestinal encephalopathy syndrome); and four hotspot mutations of mitochondrial DNA, the cause of 90% of MELAS (mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes). But the investigation was unfruitful: DNA sequencing in both scenarios only detected wildtype sequence.

Treatment of mitochondrial myopathy was commenced and rehabilitation was supported. Keith had progressive recovery and was able to restart walking without intense support. But his life was never the same as before. A tube was placed into his stomach through the abdominal wall to facilitate long-term non-oral feeding. Another tube was kept in his windpipe to facilitate sputum suction. He can no longer work and needs support from family for everyday life. Furthermore, a question mark still remains in the family: why did this happen to Keith while his three siblings all appear to be healthy?

Solution

Upon reviewing Keith's case, scientists at Pangenia Genomics decided to use whole genome sequencing to search for the answer. Whole genome sequencing has the technical advantage of relatively uniform sequencing depth of across the genome, the coverage of exonic and intronic regions, minimal PCR bias, and identification of copy number variation (CNV) and structural variants (SV) (2). After pre-test counselling and informed consent, whole blood samples were collected from Keith, his father and two siblings. DNA was extracted from the blood samples and sent for whole genome sequencing. Variants were evaluated by PhD-trained scientists, facilitated by the QIAGEN Clinical Insight (QCI) Interpret software.

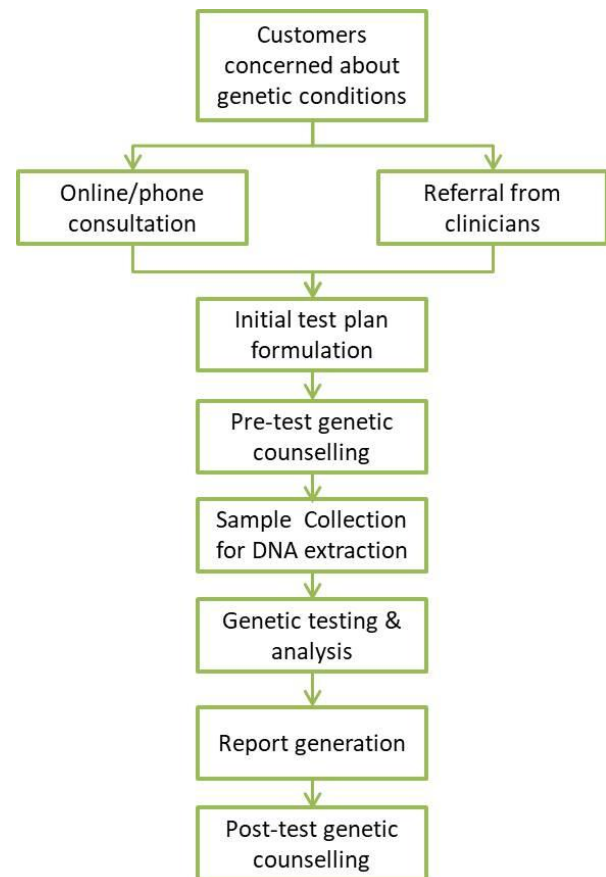


Figure 2 Pangenia Genomics' individualized genetic testing service flowchart.

Two compound heterozygous variants in the *POLG* gene were identified in Keith's DNA while only one of the two variants was found in his unaffected father and two siblings' samples. Sanger sequencing of the two variants in the last unaffected brother whose genome was not sequenced detected only wildtype sequence. This indicates Keith inherited one of the variants from his father and the other one from his mother, consistent with an autosomal recessive pattern of inheritance. The *POLG* gene encodes the catalytic subunit of DNA polymerase γ , which is responsible for the replication of the mitochondrial genome. A number of mutations have been mapped to the *POLG* gene, and found to be associated with mitochondrial diseases, most of which are inherited in an autosomal recessive manner (3). Although by the published ACMG

guideline (4), these two variants are classified as variants of unknown significance (VUS), they are likely to be disease-related in Keith's case, taking into account the compatibility with the observed phenotype and the expected mode of inheritance.

Keith and his family were relieved to get an answer to their eight-year-old mystery. This information also allows carrier screening for other family members.

Table 1: Variants of unknown significance related to Keith's condition were detected.

Gene	Variant	Zygosity	Frequency in population	Computational prediction	Classification	Criteria ^a
<i>POLG</i>	NM_002693.2: exon6:c.1242_1244del (p.Phe414del)	heterozygous	N/A	N/A	Variant of Unknown Significance	PM2, PM4
<i>POLG</i>	NM_002693.2: exon4:c.924G>T (p.Gln308His)	heterozygous	0.00005445 ^c	deleterious ^d	Variant of Unknown Significance	PM2, PP3

^a According to ACMG 2015 guideline (4)

^b Frequency in East Asian in the gnomAD database

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