Cordis DX

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Scientific Director: George Nasioulas PhD

SAMPLE INFORMATION

Name :	-	**Date Received :**	-
Medical ID :	-	**Date of Report :**	-
Date of Birth :	-	**Req. Physician :**	-
Location :	-	**Barcode :**	-
Material :	-	**Reason of referal:**	-

Cordis DX by Next Generation Sequencing

Results associated with the reason of referral

PATHOGENIC VARIANT IDENTIFIED

Gene	Variant	Clinical Significance	Zygosity
MYBPC3	NM_000256.3:c.1505G>A, p.(Arg502Gln)	Pathogenic variant	Heterozygous
FLNC	NM_001458.5:c.5668+6G>A	Variant of Uncertain Significance (VUS)	Heterozygous

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Additional results (secondary findings) in clinically important genes (according to ACMG_SF_V3.0 guidelines)

Findings in the 84 clinically important genes that international guidelines suggest that they should be reported (https://www.ncbi.nlm.nih.gov/clinvar/docs/acmg/) even not associated with the reason of referral and with the patient's consent.

NO PATHOGENIC/LIKELY PATHOGENIC VARIANTS IDENTIFIED

Table 1: Genes analyzed for clinically important secondary findings.

ABCD1	*ACVRL1*	*ACTA2*	*ACTC1*	*APC*	*APOB*	*ATP7B**	*BAG3*	*BMPR1A*
BRCA1	*BRCA2*	*BTD**	*CACNA1S*	*CALM1*	*CALM2*	*CALM3*	*CASQ2*	*COL3A1*
*CYP27A1**	*DES*	*DSC2*	*DSG2*	*DSP*	*ENG*	*FBN1*	*FLNC*	*GAA**
GLA	*HFE***	*HNF1A*	*KCNH2*	*KCNQ1*	*LDLR*	*LMNA*	*MAX*	*MEN1*
MLH1	*MSH2*	*MSH6*	*MUTYH**	*MYBPC3*	*MYH11*	*MYH7*	*MYL2*	*MYL3*
NF2	*OTC*	*PALB2*	*PCSK9*	*PMS2*	*PKP2*	*PLN*	*PRKAG2*	*PTEN*
RB1	*RBM20*	*RET*	*RPE65**	*RYR1*	*RYR2*	*SCN5A*	*SDHAF2*	*SDHB*
SDHC	*SDHD*	*SMAD3*	*SMAD4*	*STK11*	*TGFBR1*	*TGFBR2*	*TMEM127*	*TMEM43*
TNNC1	*TNNI3*	*TNNT2*	*TP53*	*TPM1*	*TRDN*	*TSC1*	*TSC2*	*TTN*
TTR	*VHL*	*WT1*						

* Will be reported only if two likely pathogenic and/or pathogenic variants are identified (homozygous or compound heterozygous state).
**HFE p.Cys282Try homozygous only.

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Variants Details

MYBPC3, Exon 16, NM_000256.3:c.1505G>A, p.(Arg502Gln)

ClinGen | HPO | ClinVar

This sequence change replaces arginine, which is basic and polar, with glutamine, which is neutral and polar, at codon 502 of the MYBPC3 protein (p.Arg502Gln). This variant is not present in population databases (gnomAD no frequency). This missense change has been observed in individuals with hypertrophic cardiomyopathy (PMID: 9562578, 16566405, 18403758, 18533079, 20433692, 22386539). ClinVar contains an entry for this variant (Variation ID: 42541). Algorithms developed to predict the effect of missense changes on protein structure and function (SIFT, PolyPhen-2, Align-GVGD) all suggest that this variant is likely to be disruptive. This variant disrupts the p.Arg502 amino acid residue in MYBPC3. Other variant(s) that disrupt this residue have been determined to be pathogenic (PMID: 12707239, 20378854, 22267749, 23396983). This suggests that this residue is clinically significant, and that variants that disrupt this residue are likely to be disease-causing. For these reasons, this variant has been classified as Pathogenic.

MYBPC3 encodes the cardiac isoform of myosin-binding protein C. Myosin-binding protein C is a myosin-associated protein found in the cross-bridge-bearing zone (C region) of A bands in striated muscle. MYBPC3 is expressed exclusively in heart muscle and is a key regulator of cardiac contraction. Pathogenic/likely pathogenic variants in MYBPC3 gene are the cause of familial hypertrophic cardiomyopathy in 15-40% of the cases (PMID: 30674652, 20031618).

FLNC ,Intron 34, NM_001458.5:c.5668+6G>A

ClinGen | HPO | ClinVar

Gene	***FLNC***	OMIM
Disease associated with pathogenic variants in this gene	[Arrhythmogenic right ventricular dysplasia, familial;Autosomal dominant],[Cardiomyopathy, familial hypertrophic, 26;Autosomal dominant],[Cardiomyopathy, familial restrictive 5;Autosomal dominant],[Myopathy, distal, 4;Autosomal dominant],[Myopathy, myofibrillar, 5;Autosomal dominant]	
Variant	NM_001458.5:c.5668+6G>A	
Zygosity	Heterozygous	
Type of variant	Splicing/Intronic	
Allele frequency (dbSNP)	0.0004%	rs773119692
Grantham score	-	

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Protein position information	-
ClinVar	Variation ID: 539341
In silico analysis	Probably not affecting splicing
Clinical Significance	Variant of Uncertain Significance (VUS)

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Methodology

Genomic DNA was extracted from the sample under investigation. The DNA was analyzed using a target enrichment method (exome capture) covering all coding exons and flanking intronic regions of the human genome (21,285 genes), utilizing the Twist Human Core Exome EF Multiplex Complete kit (Twist Bioscience). Sequencing of the enriched targets was performed on the MGI DNBSEQ-T7 platform. Bioinformatic analysis and variant interpretation were carried out using the Breakthrough Genomics bioinformatics platform VG PLUS ver3.0.8, aligned to the reference genome GRCh37/hg19.

The mean coverage depth was …, with …% of target regions sequenced at a depth ≥20x. Large genomic rearrangements (CNVs) were evaluated in silico using validated algorithms of the Breakthrough Genomics bioinformatics platform VG PLUS ver3.0.8.

Based on the available clinical data and the reported phenotype, a phenotype-driven analysis approach was followed.

-Genes were prioritized according to the OMIM and Human Phenotype Ontology (HPO) databases, selecting those associated with the patient's phenotype.

- Variant classification was performed according to the ACMG/AMP guidelines (PMID: 25741868).

-Variants predicted to have a deleterious impact (frameshift, nonsense, missense, or splice-site changes), as well as potential de novo variants, were evaluated. Only variants classified as pathogenic, likely pathogenic, or variants of uncertain significance (VUS) relevant to the phenotype were reported.

- All clinically significant variants were confirmed by Sanger sequencing, when technically feasible.

RECOMMENDATIONS

1)Genetic counseling is recommended to discuss the implications of this test and to interpret the results in the context of the patient's overall clinical evaluation and family history.

2) Reinterpretation of genome sequencing data is recommended on an annual basis and may be requested by the referring clinician and one should be cautious about that variant classification and/or interpretation may change over time if more information becomes available and identification of new variants associated with disease phenotype during the re-assessment.

3) Targeted testing of the identified pathogenic variant in the VCP gene is recommended in the extended family members if deemed necessary, for identifying those at risk for the clinical condition or reproductive planning. Consult with the referring physician to discuss about risk assessment and disease management measures.

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*Note:

- Every molecular test has an internal 0,5-1% chance of failure. This is due to rare molecular events and factors related to the preparation and analysis of the samples.

-Unless otherwise stated, the target regions enriched in this assay include all coding exons and 15 base pairs of the flanking intronic sequences on either side. Therefore, this method does not detect variants located deep within introns, in regulatory regions (enhancers or promoters), or in non-coding RNAs.

-The applied methodology achieves >99% sensitivity and specificity for the detection of single-nucleotide variants (SNVs) and small insertions/deletions (INDELs) as well as >90% sensitivity for the detection of large genomic rearrangements (CNVs) using validated computational algorithms.

- CNV calls generated from sequencing coverage data should be interpreted with caution and confirmed by an independent method. In addition, due to limitations in technology, certain regions may either not be covered or may be poorly covered, where variants cannot be confidently detected.

- This methodology does not detect structural alterations such as translocations, balanced rearrangements, or nucleotide repeat expansions in genes associated with these disorders. In addition, it cannot detect low-level mosaicism (coverage <25%).

-Next generation sequencing technologies and our bioinformatics analysis significantly reduce the contribution of pseudogene sequences or other highly-homologous sequences, these may still occasionally interfere with the technical ability of the assay to identify pathogenic variant alleles in both sequencing and deletion/duplication analyses.

GLOSSARY OF USED ABBREVIATIONS:

AD = autosomal dominant

AR = autosomal recessive

HEM = hemizygous

HET = heterozygous

HOM = homozygous

gnomAD = genome Aggregation Database (reference population database; >138,600 individuals)

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Details about non-pathogenic variants

Each individual carries a large number of genetic variants, most of which are not associated with an increased risk of disease. Variants that, according to bioinformatic analysis and the ACMG/AMP classification criteria, are considered benign or likely benign are not reported, as they are not known to confer an increased risk of disease and do not alter medical management beyond what is indicated by the patient's family and personal history. Only variants that are relevant to the reported clinical phenotype and that have been classified, according to the ACMG/AMP guidelines (2015) and ClinGen specifications (2021), as pathogenic or likely pathogenic are included in this report. Variants of uncertain clinical significance (VUS) are reported only when found in genes potentially related to the patient's phenotype and predicted by most computational algorithms (e.g., REVEL, MetaLR) to have a damaging effect on protein function. VUS identified in autosomal recessive genes are not reported unless another variant (pathogenic, likely pathogenic, or VUS) is detected in the same gene. Furthermore, variants that are not related to the indication for testing are not reported. Secondary findings are not included unless the patient has opted to receive such information, in accordance with ACMG SF v3.1 (2022) (PMID: 35802134).

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Family tree

Note: **The information shown on the family tree has been provided by the patient and not by medical records.**

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