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Patient identification

Surname XXXX
First name XXXX
Gender X
EADnr 00000000
Birth date 00/00/0000

Sampling time 00/00/0000 00:00
Sample type blood
Sample number XXXX
GC code GC000000 (A_GC000000)
Project code MD1-MT260-Genpanel-14

Subpanel HCM ⊆ gene panel HCM_LQT (v1.0)

Annex 1 General specifications of the test and analysis

The general characteristics of the full *HCM_LQT* panel are described below. The subpanel *HCM* consists of a subset of the captured genes. **Please refer to Annex 2 for the specific statisitics of the application of the subpanel *HCM* on sample *GC000000*.**

List of the 75 genes available in gene panel *HCM_LQT*. In total the gene panel *HCM_LQT* covers a maximum of **100%** of the combined coding regions ± 2bp intronic sequence of the following genes:

ABCC9
ACTC1
ACTN2
AKAP9
ANK2
ANKRD1
CACNA1C
CACNA1D
CACNA2D1
CACNB2
CALR3
CASQ2
CAV3
CSRP3
DES
DMD
DSC2
DSG2
DSP
DTNA
GJA5
GLA
GPD1L
HCN4
JPH2
JUP
KCNA5
KCNE1
KCNE1L
KCNE2
KCNE3
KCNH2
KCNJ2
KCNJ5
KCNJ8
KCNQ1
LAMP2
LDB3
LMNA
MYBPC3
MYH6
MYH7
MYL2
MYL3
MYLK2
MYOZ2
NKX2-5
NOS1AP
NPPA
NUP155
PKP2
PLN
PRKAG2
RYR2
SCN1B
SCN2B
SCN3B
SCN4B
SCN5A
SGCD
SNTA1
TAZ
TBX3
TBX5
TCAP
TGFB3
TMEM43
TMPO
TNNC1
TNNI3
TNNT2
TPM1
TRPM4
VCL
WWTR1

Method For each sample 2 μg of genomic DNA was sonicated in fragments of 300bp on average. The TruSeq DNA Sample Preparation kit of Illumina was used to make the library. After library preparation 6 samples were pooled for *in-solution* capturing with the Nimblegen SeqCap EZ kit (Lot No. xxxxxx). The enriched fragments were then amplified and sequenced on the HiSeq2500 rapid mode, 2x 100bp paired-end sequencing. This was performed in the Genomics Core facility (UZ Leuven).

Analysis (v1.6) After demultiplexing, the reads were aligned to the human reference genome hg19 (human_g1k_v37.fasta) using BWA (0.7.8). Duplicate reads were removed with Picard MarkDuplicates (1.118), local realignment around indels was performed with GATK RealignerTargetCreator (3.2.2), base scores were recalibrated with GATK BaseRecalibrator (3.2.2), and then variants were called with GATK HaplotypeCaller (3.2.2). Variants were annotated using RefSeq (release 65) and Cartagenia (Cartagenia Bench Lab NGS 3.1.2).

Sanger sequencing Mutations retained as pathogenic were confirmed by Sanger sequencing.

General limitations of the technique The analysis was optimised to identify base pair substitutions with a high sensitivity. The sensitivity for small insertions and deletions was lower. Deep-intronic mutations, mutations in the promoter region, repeats, large exonic deletions and duplications, and other structural variants were not detected by this test.

Annex 2 Quality parameters

Gene panel *HCM_LQT* applied to sample *GC000000*

- The data satisfy the proposed quality criteria.
- **99.06%** ▬ of the gene panel *HCM_LQT* is genotyped
- Total number of variants *HCM_LQT*: **1595**

Subpanel *HCM* applied to sample *GC000000*

- **99.99%** ▬ of subpanel *HCM* is genotyped; the 26 genes transcripten of subpanel *HCM* are completely genotyped unless it is indicated otherwise (e.g. ◧ = 75%):

ACTC1 NM_005159.4
ACTN2 NM_001103.3
ANKRD1 NM_014391.2
CALR3 NM_145046.4
CASQ2 NM_001232.3
CAV3 NM_033337.2
CSRP3 NM_003476.4
GLA NM_000169.2
JPH2 NM_020433.4
LAMP2 NM_002294.2
MYBPC3 NM_000256.3
MYH6 NM_002471.3 ▬
MYH7 NM_000257.2
MYL2 NM_000432.3
MYL3 NM_000258.2
MYLK2 NM_033118.3
MYOZ2 NM_016599.4
PLN NM_002667.3
PRKAG2 NM_016203.3
RYR2 NM_001035.2
TCAP NM_003673.3
TNNC1 NM_003280.2
TNNI3 NM_000363.4
TNNT2 NM_001001430.2
TPM1 NM_001018005.1
VCL NM_014000.2

- The table below shows the incomplete exons for some subpanel *HCM* genes that are not entirely genotyped:

Gene	**Transcript**	**List of incomplete exons**	**% gene genotyped**
MYH6	NM_002471.3	ex29	99.9 ▬

Annex 3 List of variants

The variants are assigned to one of the following classes: (1) benign, (2) likely benign, (3) variants of unknown significance, (4) likely pathogenic, or (5) pathogenic. Variants of class (1) and (2) are not included below.

There is no (likely) pathogenic mutation associated with the phenotype.

There is no variant of unkown significance that could be associated with the phenotype.