ORIGINAL ARTICLE

Targeted exome analysis of Russian patients with hypertrophic cardiomyopathy

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Abstract

Background: Hypertrophic cardiomyopathy (HCM), described as the presence of hypertrophy of left ventricular, is the most prevalent heritable cardiovascular disease with predominantly an autosomal dominant type of inheritance. However, pathogenic alleles are not identified in at least 25% of patients with HCM, and the spectrum of pathogenic variants that contribute to the development of HCM in Russia has not been fully described. Therefore, the goal of our study was to identify genetic variants associated with the etiopathogenesis of HCM in Russian patients.

Methods: The study cohort included 98 unrelated adult patients with HCM. We performed targeted exome sequencing, an analysis using various algorithms for prediction of the impact of variants on protein structure and the prediction of pathogenicity using ACMG Guidelines.

Results: The frequency of pathogenic and likely pathogenic variants in all HCM-related genes was 8% in our patients. We also identified 20 variants of uncertain significance in all HCM-related genes.

Conclusions: The prevalence of individual pathogenic variants in HCM-related genes in Russian population appears to be lower than in general European population, which could be explained by ethnic features of Russian population, age characteristics of our sample, or unidentified pathogenic variants in genes previously not linked with HCM.

KEYWORDS
exome, genetics, hypertrophic cardiomyopathy, next generation sequencing, pathogenic variants

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1 | INTRODUCTION

Hypertrophic cardiomyopathy (HCM), described as the presence of idiopathic left ventricular hypertrophy, is the most prevalent heritable cardiovascular disease and affects more than 1 in 500 individuals (Semsarian et al., 2015). This condition in some cases leads to heart failure, sudden cardiac death (SCD), and atrial fibrillation followed by embolic stroke (Cecconi et al., 2016; Maron & Maron, 2013).

Hypertrophic cardiomyopathy is predominantly a genetically caused disorder with an autosomal dominant type of inheritance (Greaves et al., 1987; Ho et al., 2015; Maron & Maron, 2013; Maron et al., 2012). Rare autosomal recessive and X-linked types of inheritance have also been described (Branzi et al., 1985; Hagen et al., 2015; Hartmannova et al., 2013; Santorelli et al., 1999) and may be a phenocopy condition (Marian, 2016). Moreover, numerous sporadic cases associated with de novo pathogenic variants have also been reported (Maron et al., 2012; Watkins et al., 1995).

Currently, HCM is associated with over two dozens of different mutant genes, which predominantly encode thick and thin myofilament (TTm) proteins of the sarcomere. Disease-causing pathogenic variants in myosin heavy chain 7 (MYH7) and myosin binding protein C, cardiac (MYBPC3) genes, which encode myofilament proteins, account for ~70% of more than 1,350 alleles with proven pathogenetic significance that have been identified in patients with HCM (van Velzen et al., 2018). Pathogenic variants not encoding for TTm proteins have also been detected in several patients with this condition (Arimura et al., 2009; Chiu et al., 2007; Hayashi et al., 2004; Osio et al., 2007; Siegert et al., 2011; Vasile et al., 2006).

Nevertheless, pathogenic alleles are not present in 28–40% of patients with HCM and a family history of the disease and in up to 70% of sporadic HCM cases (Alfares et al., 2015; Walsh, Buchan, et al., 2017). Conversely, up to 5% of patients with HCM carry several pathogenic variants affecting one or more gene(s) (Burns et al., 2017). Moreover, a large genetic diversity may also be associated with a large number of clinical manifestations from asymptomatic to SCD (Maron et al., 2012), even in representatives of the same family with the same pathogenic variant (Roberts et al., 2013). Some pathogenic variants demonstrate incomplete penetrance, which may depend on environmental and/or other genetic factors. Many rare pathogenic variants with low-to-moderate penetrance are detected in patients with sporadic form of HCM and in small families with this disorder. Thus, it is very difficult to establish the role of such variants in the etiopathogenesis of HCM (Marian & Braunwald, 2017).

In this context, it is very important to study the genetic landscape of HCM in diverse populations. Moreover, apart from several reports on single pathogenic variants in small samples of Russian patients (Glotov et al., 2015; Kostareva et al., 2006; Savostyanov et al., 2017; Seleznev et al., 2005), the spectrum of genes and pathogenic variants in them that contribute to the development of this disease in our population has not been described on a large scale. Conducting such studies will complement the knowledge on the range of pathogenic variants and genes involved in the development of HCM and allow the expansion of our understanding of the mechanisms of pathogenesis of HCM and the functioning of the cardiovascular system as a whole. Therefore, the goal of our study was to identify genetic variants associated with the etiopathogenesis of HCM in patients from Russia – both known pathogenic variants in genes responsible for the development of the disease and new likely pathogenic variants with the possible identification of new genes associated with the development of HCM.

2 | METHODS

2.1 | Ethical compliance

The study was approved by the Ethics Committees of RNRMU and Institute of Molecular Genetics.

2.2 | Patients

Studied cohort included 98 unrelated adult patients with HCM. The subjects were all Russians from the Moscow region. Patients were selected and investigated according to the European diagnostic criteria for familial HCM (interventricular septal thickness (IVS) ≥15 mm in the absence of other known causes of hypertrophy) (Elliott et al., 2014) by Krylova N.S. at Cardiological Department of City Clinical Hospital No. 52 of the Moscow City Health Department). The average age of the patients at the time of enrollment in the study was 58.59 ± 14.66 years, and the sex ratio was 39/59 (M/F). 35 patients had an older relative with SCD and/or HCM (24 patients had only an older relative with SCD, 6 patients had only an older relative with HCM, 5 patients had both). Clinical characteristics are presented in Table 1. Written informed consent was obtained from all participating patients and families according to the Declaration of Helsinki.

2.3 | DNA preparation and sequencing

Genomic DNA was obtained from leukocytes using a Quick-DNA Miniprep Kit (Zymo Research), according
to the manufacturer’s instructions. The concentration of isolated nucleic acids was measured using a Qubit fluorometer (Invitrogen) and a Quant-iT DNA BR Assay Kit (Invitrogen), according to the manufacturer’s instructions. Exome sequencing was performed using a SureSelect Focused Exome Enrichment kit (Agilent Technologies, Inc.) on an Illumina HiSeq 2500 sequencer. The sequence consisted of ∼4,800 disease-associated genes and regions with an average coverage of 30 reads. Pathogenic and likely pathogenic variants were validated by Sanger sequencing. The sequences of the primers used are available upon request.

### 2.4 Annotation and functional assessment

Bioinformatic analysis of obtained sequences was carried out using various bioinformatics resources. Bioinformatic sequence analysis was performed in the “R” software environment using Ensembl Variation and Ensembl Gene data (Zerbino et al., 2018). Ensembl data was accessed using the “R” BioMart package (Smedley et al., 2015). We used the GRCh38 reference assembly (Schneider et al., 2017). Genome alignment was performed with Burrows-Wheeler Alignment (BWA) tool (Li & Durbin, 2009). The genome mapping and variant calling was conducted using GATK HaplotypeCaller, as described in GATK best practices (Poplin et al., 2017). The replacement effect was classified using the Variant Effect Predictor (McLaren et al., 2016). The impact on protein structure of the identified variants was further assessed using Polyphen-2 (Polymorphism Phenotyping v2) (Adzhubei et al., 2010), SIFT (Sorting Intolerant from Tolerant) (Sim et al., 2012), REVEL (Rare Exome Variant Ensemble Learner) (Ioannidis et al., 2016), and CADD (Combined Annotation Dependent Depletion) (Rentzsch et al., 2019). The missense variants were considered “probably damaging” (or “potentially damaging”) if they had a Polyphen score > 0.5 or SIFT score < 0.05 (deleterious), and CADD PHRED >20 and REVEL Score > 0.5. Only CADD algorithm was able to evaluate the deleteriousness of variants leading to frame-shift or formation of stop-codon. These variants were considered “probably damaging” (or “potentially damaging”) if they had a CADD PHRED score > 20. The pathogenicity of known variants was also assessed using ClinVar database (Landrum et al., 2018). All possibly pathogenic variants were classified in accordance with ACMG Standards and Guidelines for the Interpretation of Sequence Variants (Kelly et al., 2018; Richards et al., 2015).

### 3 RESULTS

It is currently believed that HCM is predominantly an autosomal dominant disease. Therefore, the first stage of our analysis included only missense, nonsense, and small indel heterozygous variants in the coding regions of genes of interest. We selected only heterozygous variants (autosomal dominant type of inheritance was assumed) with a genotyping quality more than 99 and a coverage of at least 20 reads. The analysis of the selected variants revealed that there were strong differences in the ratio between two allelic variants that were read during the sequencing. As a result, an approach for the elimination of false-positive heterozygotes was implemented at the initial stage of the primary data analysis (Shulskaya et al., 2018). The result of the formula (AD1-AD2)/DP, where DP is the approximated read depth, and AD1 and AD2 are the approximated read depth for the first and the second allele, respectively, was used as a criterion for the selection of reliable heterozygous positions. For further analysis, we selected only variants for which the result of the formula met a specific condition, i.e., it had to be in the range of −0.3 and 0.3. The approach was implemented as a Python...
As the goal of our study was to identify genetic variants associated with the development of HCM in patients from Russia, both known pathogenic variants and new likely pathogenic variants with the possible identification of new genes associated with the development of HCM, for further analysis we selected only variants with MAF <0.0001 according to gnomAD, or ALFA or ExAc, if there was no data on MAF in gnomAD.

To reduce the number of candidate variants, we analyzed only 174 genes with known associations to 17 different inherited cardiac conditions according to the TruSight Cardio Sequencing Kit (Illumina) (https://emea.support.illumina.com/downloads/trusight-cardio-product-files.html, last access: 10:08.2020). Next, we used bioinformatic resources to predict the impact of variants on protein structure. The use of the Polyphen-2, SIFT, REVEL, and CADD allowed us to select 54 possibly pathogenic variants that: (a) met the CADD criteria for nonsense variants and at least two of the criteria used for missense variants and (b) were located in the coding regions of 28 genes, including the definitive genes responsible for the development of most of the cases of familial HCM (Marian & Braunwald, 2017). Further, we analyzed only variants in the genes that are associated with the development of HCM according to the OMIM database and ClinGen resource (Rehm et al., 2015) (Table S1). The data on these variants with the scores of all algorithms used are presented in Table S2.

At the second stage, an analysis of definitive HCM genes (ACTC1, MYBPC3, MYH7, MYL2, MYL3, TNNI3, TNNT2, TPM1, and PRKAG2), according to the ClinGen resource, was carried out, which led to the identification of several possibly pathogenic variants. Four of these variants are now considered pathogenic variants: p.Q1233X (two probands), R495Q (one proband), and p.Y847X (one proband) in the MYBPC3 gene and p.G741R (one proband) in the MYH7 gene (Table 2). All of them were validated by Sanger sequencing.

We also identified several likely pathogenic variants and variants of uncertain significance in the MYH7, MYBPC3, MYL2, MYL3, and TPM1 genes (Table 3), which were classified as probably damaging all algorithms used in the study; however, they are very rare and there are few data on their clinical significance in ClinVar database and ClinGen resource. It should be noted that three variants in the MYH7 and MYBPC3 genes were discovered for the first time here. The change of adenine to thymine at position 23415221 in NC_000014.9 (GRCh38.p12) (also validated by Sanger sequencing) leads to the H1778L amino-acid change, which could alter the structure of the MYH7 protein significantly. A similar situation was detected for variant K1173X (NC_000011.10:g.47332676T > A (GRCh38.p12)) (Table 3 and Table S1), which may lead to the mRNA degradation by nonsense-mediated decay or synthesis of truncated MYBPC3 protein. Unfortunately, we were unable to analyze the parents and/or children of this patient. However, the novel variant K1173X, resulting in synthesis of truncated protein, could be the cause of HCM in this case.

We also identified 13 variants of uncertain significance in the moderate and limited, according to ClinGen resource, HCM genes encoding actinin alpha 2 (ACTN2), LIM domain binding 3 protein (LDB3), titin (TTN), myosin heavy chain 6 (MYH6), nexilin F-actin binding protein (NEXN), and vinculin (VCL), which are linked to the development of HCM (Table 4). All of these variants were classified as possibly damaging by the algorithms used here. However, they are extremely rare (with the exception of some variants in the TTN gene) and there are no or few data on their actual pathogenicity in the ClinVar database.

4 DISCUSSION

In this study, we aimed to identify genetic variants associated with the ethiopathogenesis of HCM in patients from Russia by screening for both known pathogenic variants and new likely pathogenic variants in genes currently associated with the development of HCM.

It should be noted that we were unable to identify major pathogenic variants that may be characteristic of our patients. The frequency of pathogenic variants in the definitive HCM genes in our patients was only 5%. However, when considering all pathogenic variants (i.e., pathogenic as well as likely pathogenic variants) in definitive, moderate and limited HCM-related genes, the frequency

| Gene     | Existing variation | Position in genome | Protein position | Number of probands |
|----------|--------------------|--------------------|------------------|--------------------|
| MYBPC3   | rs397516037        | NC_000011.10:g.47332189G>A | Q1233X           | 2                  |
| MYBPC3   | rs397515974        | NC_000011.10:g.47337452G>C | Y847X            | 1                  |
| MYBPC3   | rs200411226        | NC_000011.10:g.47342718C>T | R495Q            | 1                  |
| MYH7     | rs121913632        | NC_000014.9:g.23425760C>T | G741R            | 1                  |
of pathogenic variants was 8% in our patients, whereas the mutation rate in these genes worldwide has been estimated to be 30–60% on average (Burke et al., 2016). In Europe alone, the frequency of HCM causing pathogenic variants is estimated as 17–63% (average 33.5%) (Andersen et al., 2009; Berge & Leren, 2014; Brito et al., 2012; Cecconi et al., 2016; Erdmann et al., 2003; Fokstuen et al., 2008, 2011; Garcia-Castro et al., 2009; Kaski et al., 2009; Lopes et al., 2013; Millat et al., 2010; Morner et al., 2003; Richard et al., 2003; Waldmuller et al., 2011; Zeller et al., 2006). This discrepancy of our results with the data obtained on other populations could be explained by the ethnic characteristics of our sample. Moreover, the fact that our sample consists mainly of middle-aged patients without family history of the disease may be associated with unidentified pathogenic variants in genes previously not linked with HCM, which alone or in combination could result in manifestation of mild form of HCM.

It is widely recognized that HCM is caused by rare pathogenic variants. These variants are usually found in domains of genes that encode sarcomere proteins and proteins associated with this cell structure. Accordingly, the majority of pathogenic variants in our sample were identified in the sarcomere-related genes MYH7 and MYBPC3, the mutation of which is causative in majority of cases of HCM with a proven genetic cause (Marian & Braunwald, 2017). Overall the frequency of these pathogenic variants (Table 2) in our sample was 5%, which was higher than that reported in various European samples (average 0.86%) (Berge & Leren, 2014; Brito et al., 2012; Cecconi et al., 2016; Christiaans et al., 2010; Ehlermann et al., 2008; Erdmann et al., 2001; Fokstuen et al., 2008,

**Table 3** Likely pathogenic variants and variants of uncertain significance in definitive HCM genes

| Gene    | Existing variation | Position in genome | Protein position | Number of probands |
|---------|--------------------|--------------------|------------------|--------------------|
| MYBPC3  | rs397515905        | NC_000011.10:g.47342719G>A | R495W            | 1                  |
| MYBPC3  | rs730880711        | NC_000011.10:g.47342928_47342929insG | V453X           | 1                  |
| MYBPC3  |                  | NC_000011.10:g.47332676T>A | K1173X          | 1                  |

**Table 4** Variants of uncertain significance in other HCM genes

| Gene    | Existing variation | Position in genome | Protein position | Number of probands |
|---------|--------------------|--------------------|------------------|--------------------|
| ACTN2   | rs397516574        | NC_000001.11:g.236761033C>T | R796C            | 1                  |
| NEXN    | rs397516574        | NC_000001.11:g.236761033C>T | R796C            | 1                  |
| VCL     | rs749628307        | NC_000010.11:g.74074809G>A | R230H            | 1                  |
| LDB3    | rs774815578        | NC_000010.11:g.86732915C>T | P598L            | 1                  |
| MYH6    | rs201989347        | NC_000014.9:g.23387854G>A | R1477C           | 1                  |
| TTN     | rs192360370        | NC_000002.12:g.178538825G>C | P31361A          | 1                  |
| TTN     | rs192360370        | NC_000002.12:g.178538825G>C | P31361A          | 1                  |
| TTN     | rs551496477        | NC_000002.12:g.178563052G>A | R26053C          | 1                  |
| TTN     | rs1214607347       | NC_000002.12:g.178568461G>T | P24250T          | 1                  |
| TTN     | rs756003188        | NC_000002.12:g.178713277G>A | P8636S           | 1                  |
2011; Garcia-Giustinianini et al., 2015; Helms et al., 2014; Ingles et al., 2013; Kapplinger et al., 2014; Kaski et al., 2009; Lopes et al., 2013; Millat et al., 2010; Ng et al., 2013; Niimura et al., 1998; Ferrot et al., 2005; Toth et al., 2011; Weissler-Snir et al., 2017; Zeller et al., 2006) (Table S3).

However, the prevalence of individual pathogenic variants in an HCM population appears to be very low (Marian & Braunwald, 2017). The majority of other pathogenic variants occur at a frequency of <0.01 in the HCM population and nearly half are detected in a single proband or family (Alfares et al., 2015). Our data support these findings in our population, as most of the pathogenic variants identified here were detected in single probands. We found only one pathogenic variant, rs397516037 in MYBPC3, in two probands, which represented 2% of all non-related cases of HCM in our sample. This variant is rare in general population. However, Tóth T. et al. reported results that were similar to ours (Toth et al., 2011). The high occurrence of this pathogenic variant in our sample and various other cohorts (Ehlermann et al., 2008; Erdmann et al., 2001, 2003; Fokstuen et al., 2008; Ingles et al., 2005; Kapplinger et al., 2014; Toth et al., 2011; Zeller et al., 2006) might reflect “hot spots” for pathogenic variants or probably a founder effect (Page et al., 2012), which was confirmed by Erdmann et al. (Erdmann et al., 2001).

The other pathogenic variant in MYBPC3 which leads to formation of the stop codon p.Y847X is also very rare even in individuals with HCM. This variant was also identified in patients from diverse populations (Berge & Leren, 2014; Chan et al., 2014; Kapplinger et al., 2014; Marsiglia et al., 2013; Zhao et al., 2017). Thus, this pathogenic variant in MYBPC3 is responsible for the development of less than 1% of all cases of this condition in general population. The higher occurrence of this pathogenic variant in our population can be explained either by the peculiarities of the Russian population or by the small size of our sample.

The same is true for rs200411226 which leads to p.R495Q substitution in the MYBPC3 protein. Its frequency is also very low even in individuals with HCM. This pathogenic variant occurs on average in one patient out of 100, ranging from 0.5% to 8% (Brito et al., 2012; Christiaans et al., 2010; Ehlermann et al., 2008; Fokstuen et al., 2008, 2011; Helms et al., 2014; Kapplinger et al., 2014; Lopes et al., 2013; Maron et al., 2001; Marsiglia et al., 2013; Millat et al., 2010; Ng et al., 2013; Niimura et al., 1998; Van Driest et al., 2004; Zeller et al., 2006). Our data indicate that our population does not differ much from the European population in the case of this variant.

The pathogenic variant in the MYH7 gene, which leads to the p.G741R substitution, was first associated with the development of HCM in Chinese patients by Song et al (Song et al., 2005). This pathogenic variant was also discovered in less than 0.01 in other samples worldwide (Berge & Leren, 2014; Garcia-Giustinianini et al., 2015; Kapplinger et al., 2014; Kaski et al., 2009; Marsiglia et al., 2013; Miller et al., 2013; Murphy et al., 2016; Otsuka et al., 2012; Ferrot et al., 2005), whereas it was 1 out of 98 in patients with HCM in our sample.

As it was mentioned earlier also, we identified several probably damaging variants in MYH7, MYBPC3, MYL3, and TPM1 genes, which are considered to be definitive HCM genes, as pathogenic variants in these genes are responsible for the development of HCM in the majority of cases of this condition (Table 3). These variants are also very rare, as they occur exclusively in single cases worldwide. Therefore, it is very difficult to prove their pathogenic significance by co-segregation analysis. However, some variants in the MYBPC3 gene were classified as likely pathogenic.

As mentioned above, we also detected 13 probably damaging (that could affect a protein structure) variants in the moderate (ACTN2) and limited (LDB3, TTN, MYH6, NEXN, and VCL) genes of HCM, which may be causative pathogenic variants of HCM in 24 patients from our sample (Table 4). Computational prediction tools and a conservation analysis suggested that these variants have an impact on the structures of the proteins, although this information is not sufficiently predictive to determine pathogenicity, i.e., the clinical significance of these variants is uncertain (Ng et al., 2013). Some of these genes encode proteins that are structural components of sarcomere (ACTN2, TTN, MYH6), whereas others are linked to the sarcomere structurally (NEXN) or are involved in the maintenance of sarcomere function (LDB3, VCL). Several of the variants were detected in the same patients (Table S2). This fact may complicate the definition of pathogenicity of these variants. In general, there are few data on these extremely rare variants, which we identified, and much less on their involvement in the pathogenesis of HCM (Chen et al., 2012; Ploski et al., 2014; Walsh, Thomson, et al., 2017). Moreover, the frequencies of some of these variants were significantly higher in our sample than they were in the general population (Table 4). This fact may indicate both the potential pathogenicity of these variants and their ethno-specificity for our population. It is possible that these variants do not lead to the development of HCM by themselves; rather, their combination with another similar variant may participate in the pathogenic process.

In summary, further investigation of these potentially pathogenic variants is needed to prove their causal role in the pathogenesis of HCM.

5 | CONCLUSIONS

In this study, we described partially the spectrum of pathogenic variants that cause HCM in the Russian population.
Most of the pathogenic variants in our patients were detected in the MYBPC3 and MYH7 genes. The total frequency of pathogenic and likely pathogenic variants in our sample was 8%. However, the overall prevalence of individual pathogenic variants identified in our population appeared to be very low. Nevertheless, we were able to detect some novel likely pathogenic variants in HCM genes in our sample. We also identified 20 variants of uncertain significance in all HCM-related genes: 7 variants in the definitive genes and 13 variants in the moderate and limited genes.

It should be mentioned that, despite the fact that the exact pathogenic variants identified are not unique to our sample and are found in patients worldwide, the frequencies of these variants in our population differ slightly from those reported in Europe. This could be explained by ethno-specific traits of our population. Moreover, some rare probably damaging variants, especially those that were detected for the first time, could also be specific to our population. In addition, our findings may be explained by the features of our sample (mainly middle-aged patients without family history of the disease) and unidentified pathogenic variants in genes previously not linked with HCM, which alone or in combination could result in manifestation of mild form of HCM.

Since the initial discovery of the hereditary nature of HCM in the late 60s, many HCM causative genes have been discovered. However, it should be emphasized that, for a fairly large number of rare pathogenic variants, and despite the demonstrated connection with the development of HCM, there is still no reliable confirmation of a direct causal relationship between the variant and the disease. Furthermore, despite many years of research, genes that may be associated with the development of this disease remain undiscovered; the genetic basis of HCM remains undetected in at least one quarter of all cases of this condition\(^8\). Therefore, it is necessary to continue the search for new genes associated with the development of HCM using modern methods of genome-wide analysis, as well as molecular genetics and cellular technologies.

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CONFLICT OF INTERESTS
The authors declare that there is no conflict of interest.

AUTHORS’ CONTRIBUTION
SPA and SMI developed the concept of the study. FEV, KNS, PNG, SPA, and SMI organized and coordinated the study. FEV, KNS, VIN, MMS, SPA, and SMI conducted the study and analyzed the data. FEV wrote the manuscript. SPA and SMI revised and reviewed the manuscript.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE
The study was approved by the Ethics Committees of RNRMU and Institute of Molecular Genetics. Written informed consent was obtained from all participating patients and families according to the Declaration of Helsinki.

PATIENT CONSENT FOR PUBLICATION
Written informed consent for publication was obtained from all participating patients and families.

DATA AVAILABILITY STATEMENT
All data generated or analyzed during this study are included in this published article (and its Supplementary Information files).

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REFERENCES
Adzhubei, I. A., Schmidt, S., Peshkin, L., Ramensky, V. E., Gerassimova, A., Bork, P., Kondrashov, A. S., & Sunyaev, S. R. (2010). A method and server for predicting damaging missense mutations. Nature Methods, 7(4), 248–249. https://doi.org/10.1038/nmeth0410-248
Alfares, A. A., Kelly, M. A., McDermott, G., Funke, B. H., Lebo, M. S., Baxter, S. B., Shen, J., McLaughlin, H. M., Clark, E. H., Babb, L. J., Cox, S. W., DePalma, S. R., Ho, C. Y., Seidman, J. G., Seidman, C. E., & Rehm, H. L. (2015). Results of clinical genetic testing of 2,912 probands with hypertrophic cardiomyopathy: expanded panels offer limited additional sensitivity. Genetics in Medicine, 17(11), 880–888. https://doi.org/10.1038/gim.2014.205
Andersen, P. S., Havndrup, O., Houg, L., Sorensen, K. M., Jensen, M., Larsen, L. A., Hedley, P., Bie Thomsen, A. R., Moolman-Smook, J., Christiansen, M., & Bundgaard, H. (2009). Diagnostic yield, interpretation, and clinical utility of mutation screening of sarcomere encoding genes in Danish hypertrophic cardiomyopathy patients and relatives. Human Mutation, 30(3), 363–370. https://doi.org/10.1002/humu.20862
Arimura, T., Bos, J. M., Sato, A., Kubo, T., Okamoto, H., Nishi, H., Harada, H., Koga, Y., Moulik, M., Doi, Y. L., Towbin, J. A., Ackerman, M. J., & Kimura, A. (2009). Cardiac ankyrin repeat protein gene (ANKRD1) mutations in hypertrophic cardiomyopathy. Journal of the American College of Cardiology, 54(4), 334–342. https://doi.org/10.1016/j.jacc.2008.12.082
Berge, K. E., & Leren, T. P. (2014). Genetics of hypertrophic cardiomyopathy in Norway. Clinical Genetics, 86(4), 355–360. https://doi.org/10.1111/cge.12286
Branzi, A., Romeo, G., Specchia, S., Lolli, C., Binetti, G., Devoto, M., Bacchi, M., & Magnani, B. (1985). Genetic heterogeneity of
hypertrophic cardiomyopathy. *International Journal of Cardiology*, 7(2), 129–138. https://doi.org/10.1016/0167-5273(85)90352-3

Brito, D., Miltenberger-Milenyi, G., Vale Pereira, S., Silva, D., Diogo, A. N., & Madeira, H. (2012). Sarcomeric hypertrophic cardiomyopathy: genetic profile in a Portuguese population. *Revista Portuguesa De Cardiologia*, 31(9), 577–587. https://doi.org/10.1016/j.rcpi.2011.12.020

Burke, M. A., Cook, S. A., Seidman, J. G., & Seidman, C. E. (2016). Clinical and mechanistic insights into the genetics of cardiomyopathy. *Journal of the American College of Cardiology*, 68(25), 2871–2886. https://doi.org/10.1016/j.jacc.2016.07.079

Burns, C., Bagnall, R. D., Lam, I., Semsarian, C., & Ingles, J. (2017). Multiple gene variants in hypertrophic cardiomyopathy in the era of next-generation sequencing. *Circulation: Cardiovascular Genetics*, 10(4), e001666. https://doi.org/10.1161/CIRCGENETICS.116.001666

Cecconi, M., Parodi, M. I., Formisano, F., Spirito, P., Autore, C., Musumeci, M. B., Favale, S., Forleo, C., Biagini, E., Davi, S., Canepa, E., Pennese, L., Castagnetta, M., Degiorgio, D., & Coviello, D. A. (2016). Targeted next-generation sequencing helps to decipher the genetic and phenotypic heterogeneity of hypertrophic cardiomyopathy. *International Journal of Molecular Medicine*, 38(4), 1111–1124. https://doi.org/10.3892/ijmm.2016.2732

Chan, R. H., Maron, B. J., Olivotto, I., Pencina, M. J., Assenza, G. E., Haas, T., Lesser, J. R., Groner, C., Crean, A. M., Rakowski, H., Udelson, J. E., Rowin, E., Lombardi, M., Cecchi, F., Tomberli, B., Spirito, P., Formisano, F., Biagini, E., R apezzi, C., ... Maron, M. S. (2014). Prognostic value of quantitative contrast-enhanced cardiovascular magnetic resonance for the evaluation of sudden death risk in patients with hypertrophic cardiomyopathy. *Circulation*, 130(6), 484–495. https://doi.org/10.1161/CIRCULATIONAHA.113.007094

Chen, S. N., Czernuszeewicz, G., Tan, Y., Lombardi, R., Jin, J., Willerson, J. T., & Marian, A. J. (2012). Human molecular genetic and functional studies identify TRIM63, encoding Muscle RING Finger Protein 1, as a novel gene for human hypertrophic cardiomyopathy. *Circulation Research*, 111(7), 907–919. https://doi.org/10.1161/CIRCRESAHA.112.270207

Chiu, C., Tebo, M., Ingle, J., Yeates, L., Arthur, J. W., Lind, J. M., & Semsarian, C. (2007). Genetic screening of calcium regulation genes in familial hypertrophic cardiomyopathy. *Journal of Molecular and Cellular Cardiology*, 43(3), 337–343. https://doi.org/10.1016/j.ymecoc.2007.06.009

Christiaans, I., Birnie, E., van Langen, I. M., van Spaandonck-Zwarts, K. Y., van Tintelen, J. P., van den Berg, M. P., Atsma, D. E., Helderman-van den Enden, A. T. J. M., Pinto, Y. M., Hermans-van Ast, J. P., Bonsel, G. J., & Wilde, A. A. M. (2010). The yield of risk stratification for sudden cardiac death in hypertrophic cardiomyopathy myosin-binding protein C gene mutation carriers: Focus on predictive screening. *European Heart Journal*, 31(7), 842–848. https://doi.org/10.1093/eurheartj/ehp539

Ehlermann, P., Weichenhan, D., Zehelien, J., Steen, H., Prib, R., Zeller, R., Lehrke, S., Zugck, C., Ivanidc, B. T., & Katus, H. A. (2008). Adverse events in families with hypertrophic or dilated cardiomyopathy and mutations in the MYBPC3 gene. *BMC Medical Genetics*, 9, 95. https://doi.org/10.1186/1471-2350-9-95

Elliott, P. M., Anastasakis, A., Borger, M. A., Borggrefe, M., Cecchi, F., Charron, P., & Watkins, H. (2014). 2014 ESC Guidelines on diagnosis and management of hypertrophic cardiomyopathy: the Task Force for the Diagnosis and Management of Hypertrophic Cardiomyopathy of the European Society of Cardiology (ESC). *European Heart Journal*, 35(39), 2733–2779. https://doi.org/10.1093/eurheartj/ehu284

Erdmann, J., Daehmlow, S., Wischke, S., Senyuva, M., Werner, U., Raible, J., Tanis, N., Dyachenko, S., Hummel, M., Hetzer, R., & Regitz-Zagrosek, V. (2003). Mutation spectrum in a large cohort of unrelated consecutive patients with hypertrophic cardiomyopathy. *Clinical Genetics*, 64(4), 339–349. https://doi.org/10.1034/j.1399-0004.2003.00151.x

Erdmann, J., Raible, J., Maki-Abadi, J., Hammann, J., Wollnik, B., Frantz, E., Fleck, E., Regitz-Zagrosek, V., Hummel, M., & Hetzer, R. (2001). Spectrum of clinical phenotypes and gene variants in cardiac myosin-binding protein C mutation carriers with hypertrophic cardiomyopathy. *Journal of the American College of Cardiology*, 38(2), 322–330. https://doi.org/10.1016/s0735-1097(01)01387-0

Fokstuen, S., Lyle, R., Munoz, A., Gehrig, C., Lerch, R., Perrot, A., Osterziel, K. J., Geier, C., Beghetti, M., Mach, F., Szatzjel, J., Sigwart, U., Antonarakis, S. E., & Blouin, J.-L. (2008). A DNA resequencing array for pathogenic mutation detection in hypertrophic cardiomyopathy. *Human Mutation*, 29(6), 879–885. https://doi.org/10.1002/humu.20749

Fokstuen, S., Munoz, A., Melacini, P., Iliicto, S., Perrot, A., Ozcelik, C., Jeanrenaud, X., Rieubland, C., Farr, M., Faber, L., Sigwart, U., Mach, F., Lerch, R., Antonarakis, S. E., & Blouin, J.-L. (2011). Rapid detection of genetic variants in hypertrophic cardiomyopathy by custom DNA resequencing array in clinical practice. *Journal of Medical Genetics*, 48(8), 572–576. https://doi.org/10.1136/jmg.2010.083345

Garcia-Castro, M., Coto, E., Reguero, J. R., Berazaleta, J. R., Alvarez, V., Alonso, B., & Moris, C. (2009). Mutations in sarcomeric genes MYH7, MYBPC3, TNN2, TNN1, and TPM1 in patients with hypertrophic cardiomyopathy. *Revista Española De Cardiología*, 62(1), 48–56. https://doi.org/10.1016/s0300-8932(09)70020-x

Garcia-Giustianini, D., Arad, M., Ortiz-Genga, M., Barrialles-Villa, R., Fernandez, X., Rodriguez-Garcia, I., & Monserrat, L. (2015). Phenotype and prognostic correlations of the converter region mutations affecting the beta myosin heavy chain. *Heart*, 101(13), 1047–1053. https://doi.org/10.1136/heartjnl-2014-307205

Glotov, A. S., Kazakov, S. V., Zhukova, E. A., Alexandrov, A. V., Glotov, O. S., Pakin, V. S., Danilova, M. M., Poliakova, I. V., Niyazova, S. S., Chakova, N. N., Komissarova, S. M., Kurnikova, E. A., Sarana, A. M., Sherbak, S. G., Sergushichev, A. A., Shaltyo, A. A., & Baranov, V. S. (2015). Targeted next-generation sequencing (NGS) of nine candidate genes with custom AmpliSeq in patients and a cardiomyopathy risk group. *Clínica Chimica Acta*, 446, 132–140. https://doi.org/10.1016/j.cca.2015.04.014

Greaves, S. C., Roche, A. H., Neutze, J. M., Whitlock, R. M., & Veale, A. M. (1987). Inheritance of hypertrophic cardiomyopathy: A cross sectional and M mode echocardiographic study of 50 families. *British Heart Journal*, 58(3), 259–266. https://doi.org/10.1136/hrt.58.3.259

Hagen, C. M., Afdt, F. H., Havndrup, O., Hedley, P. L., Jensen, M. K., Kanters, J. K., Pham, T. T., Bundgaard, H., & Christiansen, M. (2015). Private mitochondrial DNA variants in danish patients with hypertrophic cardiomyopathy. *PLoS One*, 10(4), e0124540. https://doi.org/10.1371/journal.pone.0124540

Hartmannova, H., Kubanek, M., Sramko, M., Piberova, L., Noskova, L., Hodanova, K., Stranecky, V., Pristoupilova, A., Sovova, J., Marek, T., Maluskova, J., Ridzon, P., Kautzner, J., Hulka, H., & Knoch, S. (2013). Isolated X-linked hypertrophic cardiomyopathy caused by a novel mutation of the four-and-a-half LIM
domain 1 gene. Circulation: Cardiovascular Genetics, 6(6), 543–551. https://doi.org/10.1161/CIRCGENETICS.113.000245
Hayashi, T., Arimura, T., Ueda, K., Shibata, H., Hirota, R., Takahashi, M., Hori, H., Koga, Y., Oka, N., Imazumi, T., Yasumami, M., & Kimura, A. (2004). Identification and functional analysis of a caveolin-3 mutation associated with familial hypertrophic cardiomyopathy. Biochemical and Biophysical Research Communications, 313(1), 178–184. https://doi.org/10.1016/j.bbrc.2003.11.101
Helms, A. S., Davis, F. M., Coleman, D., Bartolone, S. N., Glazier, A. A., Pagani, F., Yob, M. J., Sadayappan, S., Pedersen, E., Lyons, R., Westfall, M. V., Jones, R., Russell, M. W., & Day, S. M. (2014). Sarcomere mutation-specific expression patterns in human hypertrophic cardiomyopathy. Circulation: Cardiovascular Genetics, 7(4), 434–443. https://doi.org/10.1161/CIRCGENETICS.113.000448
Ho, C. Y., Charron, P., Richard, P., Girolami, F., Van Spall, M., Ho, C. Y., Charron, P., Richard, P., Girolami, F., Van Spaendonck-Zwarts, K. Y., & Pinto, Y. (2015). Genetic advances in sarcomeric cardiomyopathies: State of the art. Cardiovascular Research, 105(4), 397–408. https://doi.org/10.1093/cvr/cv025
Ingles, J., Doolan, A., Chiu, C., Seidman, J., Seidman, C., & Semsarian, C. (2005). Compound and double mutations in patients with hypertrophic cardiomyopathy: implications for genetic testing and counselling. Journal of Medical Genetics, 42(10), e59. https://doi.org/10.1136/jmg.2005.033886
Ingles, J., Sarina, T., Yeates, L., Hunt, L., Macciocca, I., McCormack, L., Winship, I., McGaughan, J., Atherton, J., & Semsarian, C. (2013). Clinical predictors of genetic testing outcomes in hypertrophic cardiomyopathy. Genetics in Medicine, 15(12), 972–977. https://doi.org/10.1038/gim.2013.44
Ioannidis, N. M., Rothstein, J. H., Pejaver, V., Middha, S., McDonnell, S. K., Baheti, S., Musolf, A. L., Qi, J., Holzinger, E., Karyadi, D., Cannon-Albright, L. A., Teerlink, C. C., Stanford, J. L., Isaacs, W. B., Xu, J., Cooney, K. A., Lange, E. M., Schleutker, J., Carpten, J. D., ... Sieh, W. (2016). REVEL: An ensemble method for predicting the pathogenicity of rare missense variants. American Journal of Human Genetics, 99(4), 877–885. https://doi.org/10.1016/j.ajhg.2016.08.016
Kapplinger, J. D., Landstrom, A. P., Bos, J. M., Salisbury, B. A., Callis, T. E., & Ackerman, M. J. (2014). Distinguishing hypertrophic cardiomyopathy-associated mutations from background genetic noise. Journal of Cardiovascular Translational Research, 7(3), 347–361. https://doi.org/10.1007/s12265-014-9542-2
Kaski, J. P., Syrris, P., Esteban, M. T. T., Jenkins, S., Pantazis, A., Deanfield, J. E., McKenna, W. J., & Elliott, P. M. (2009). Prevalence of sarcomere protein gene mutations in preadolescent children with hypertrophic cardiomyopathy. Circulation: Cardiovascular Genetics, 2(5), 436–441. https://doi.org/10.1161/CIRCGENETICS.108.821314
Kelly, M. A., Caleshu, C., Morales, A., Buchan, J., Wolf, Z., Harrison, S. M., Cook, S., Dillon, M. W., Garcia, J., Haverfield, E., Jongbloed, J. D. H., Macaya, D., Manrai, A., Orland, K., Richard, G., Spoonmore, K., Thomas, M., Thomson, K., Vincent, L. M., ... Funke, B. (2018). Adaptation and validation of the ACMG/AMP variant classification framework for MYH7-associated inherited cardiomyopathies: Recommendations by ClinGen’s Inherited Cardiomyopathy Expert Panel. Genetics in Medicine, 20(3), 351–359. https://doi.org/10.1038/gim.2017.218
Kostareva, A., Gudkova, A., Sjoberg, G., Kiselev, I., Moiseeva, O., Karelkina, E., & Sejersen, T. (2006). Desmin mutations in a St. Petersburg cohort of cardiomyopathies. Acta Myologica, 25(3), 109–115.

Landrum, M. J., Lee, J. M., Benson, M., Brown, G. R., Chao, C., Chitipiralla, S., Gu, B., Hart, J., Hoffman, D., Jang, W., Karapetyan, K., Katz, K., Liu, C., Maddipatla, Z., Malheiro, A., McDaniel, K., Ovetsky, M., Riley, G., Zhou, G., ... Maglott, D. R. (2018). ClinVar: improving access to variant interpretation and supporting evidence. Nucleic Acids Research, 46(D1), D1062–D1067. https://doi.org/10.1093/nar/gkx1153
Li, H., & Durbin, R. (2009). Fast and accurate read alignment with Burrows-Wheeler transform. Bioinformatics, 25(14), 1754–1760. https://doi.org/10.1093/bioinformatics/btp324
Lopes, L. R., Zekavati, A., Syrris, P., Hubank, M., Giambartolomei, C., Dalageorgou, C., Jenkins, S., McKenna, W., Plagnol, V., & Elliott, P. M. (2013). Genetic complexity in hypertrophic cardiomyopathy revealed by high-throughput sequencing. Journal of Medical Genetics, 50(4), 228–239. https://doi.org/10.1136/jmedgenet-2012-101270
Marian, A. J. (2016). Challenges in the diagnosis of anderson-fabry disease: A deceptively simple and yet complicated genetic disease. Journal of the American College of Cardiology, 68(10), 1051–1053. https://doi.org/10.1016/j.jacc.2016.06.026
Marian, A. J., & Braunwald, E. (2017). Hypertrophic cardiomyopathy: genetics, pathogenesis, clinical manifestations, diagnosis, and therapy. Circulation Research, 121(7), 749–770. https://doi.org/10.1161/CIRCRESAHA.117.311059
Maron, B. J., & Maron, M. S. (2013). Hypertrophic cardiomyopathy. Lancet, 381(9862), 242–255. https://doi.org/10.1016/S0140-6736(12)60397-3
Maron, B. J., Maron, M. S., & Semsarian, C. (2012). Genetics of hypertrophic cardiomyopathy after 20 years: Clinical perspectives. Journal of the American College of Cardiology, 60(8), 705–715. https://doi.org/10.1016/j.jacc.2012.02.068
Maron, B. J., Niimura, H., Casey, S. A., Soper, M. K., Wright, G. B., Seidman, J. G., & Seidman, C. E. (2001). Development of left ventricular hypertrophy in adults in hypertrophic cardiomyopathy caused by cardiac myosin-binding protein C gene mutations. Journal of the American College of Cardiology, 38(2), 315–321. https://doi.org/10.1016/s0735-1097(01)01386-9
Marsiglia, J. D. C., Credidio, F. L., de Oliveira, T. G. M., Reis, R. F., Antunes, M. D. O., de Araujo, A. Q., Pedrosa, R. P., Barbosa-Ferreira, J. M. B., Mady, C., Krieger, J. E., Arteaga-Fernandez, E., & Pereira, A. D. C. (2013). Screening of MYH7, MYBPC3, and TNNT2 genes in Brazilian patients with hypertrophic cardiomyopathy. American Heart Journal, 166(4), 775–782. https://doi.org/10.1016/j.ahj.2013.07.029
McLaren, W., Gil, L., Hunt, S. E., Riat, H. S., Ritchie, G. R. S., Thormann, A., Flicke, P., & Cunningham, F. (2016). The Ensembl variant effect predictor. Genome Biology, 17(1), 122.
Millat, G., Bouvagnet, P., Chevalier, P., Dauphin, C., Simon Jouk, P., Da Costa, A., Prieur, F., Bresson, J.-L., Faivre, L., Eicher, J.-C., Chassaing, N., Crehalet, H., Porcher, R., Rodriguez-Lafraise, C., & Roussin, R. (2010). Prevalence and spectrum of mutations in a cohort of 192 unrelated patients with hypertrophic cardiomyopathy. European Journal of Medical Genetics, 53(5), 261–267. https://doi.org/10.1016/j.ejmg.2010.07.007
Miller, E. M., Wang, Y., & Ware, S. M. (2013). Uptake of cardiac screening and genetic testing among hypertrophic and dilated cardiomyopathy families. J Genet Couns, 22(2), 258–267. https://doi.org/10.1007/s10897-012-9544-4
Morner, S., Richard, P., Kazzam, E., Hellman, U., Hainque, B., Schwartz, K., & Waldenstrom, A. (2003). Identification of the
genotypes causing hypertrophic cardiomyopathy in northern Sweden. Journal of Molecular and Cellular Cardiology, 35(7), 841–849. https://doi.org/10.1016/s0022-2828(03)00146-9

Murphy, S. L., Anderson, J. H., Kapplinger, J. D., Kruiselbrink, T. M., Gersh, B. J., Ommen, S. R., Ackerman, M. J., & Bos, J. M. (2016). Evaluation of the myo phenotype-based genotype predictor score in patients with clinically diagnosed hypertrophic cardiomyopathy. Journal of Cardiovascular Translational Research, 9(2), 153–161. https://doi.org/10.1007/s12265-016-9681-5

Ng, D., Johnston, J. J., Teer, J. K., Singh, L. N., Peller, L. C., Wynter, J. S., Lewis, K. L., Cooper, D. N., Stenson, P. D., Mullikin, J. C., & Biesecker, L. G. (2013). Interpreting secondary cardiac disease variants in an exome cohort. Circulation: Cardiovascular Genetics, 6(4), 337–346. https://doi.org/10.1161/CIRCGENETICS.113.000039

Niimura, H., Bachinski, L. L., Sangwatanaroj, S., Watkins, H., Chudley, A. E., McKenna, W., Kristinsson, A., Roberts, R., Sole, M., Maron, B. J., Seidman, J. G., Seidman, C. E., Thierfelder, L., Jarcho, J. A., Anastasakis, A., Toutouzas, P., Elstein, E., Liew, C.-C., Liew, J., ... Bjornsdottir, H. (1998). Mutations in the gene for cardiac myosin-binding protein C and late-onset familial hypertrophic cardiomyopathy. New England Journal of Medicine, 338(18), 1248–1257. https://doi.org/10.1056/NEJM199804303381802

Osio, A., Tan, L., Chen, S. N., Lombardi, R., Nagueh, S. F., Shete, S., Roberts, R., Willerson, J. T., & Marian, A. J. (2007). Myozenin 2 is a novel gene for human hypertrophic cardiomyopathy. Circulation Research, 100(6), 766–768. https://doi.org/10.1161/01.RES.0000263008.66799.aa

Otsuka, H., Arimura, T., Abe, T., Kawai, H., Aizawa, Y., Kubo, T., Kitaoka, H., Nakamura, H., Nakamura, K., Okamoto, H., Ichida, F., Ayusawa, M., Nunoda, S., Isobe, M., Matsuzaki, M., Doi, Y. L., Fukuda, K., Sasaki, T., Izumi, T., ... Kimura, A. (2012). Prevalence and distribution of sarcomeric gene mutations in Japanese patients with familial hypertrophic cardiomyopathy. Journal of Circulation, 76(2), 453–461. https://doi.org/10.1253/circj.cj-11-0876

Page, S. P., Kounas, S., Syrris, P., Christiansen, M., Frank-Hansen, R., Andersen, P. S., Elliott, P. M., & McKenna, W. J. (2012). Cardiac myosin binding protein-C mutations in families with hypertrophic cardiomyopathy: Disease expression in relation to age, gender, and long term outcome. Circulation: Cardiovascular Genetics, 5(2), 156–166. https://doi.org/10.1161/CIRCGENETICS.111.960831

Perrot, A., Schmidt-Traub, H., Hoffmann, B., Prager, M., Bit-Avragim, N., Rudenko, R. I., Usupbaeva, D. A., Kabaeva, Z., Imanov, B., Mirrakhimov, M. M., Dietz, R., Wycisk, A., Tendera, M., Geßner, R., & Osterziel, K. J. (2005). Prevalence of cardiac beta-myosin heavy chain gene mutations in patients with hypertrophic cardiomyopathy. Journal of Molecular Medicine (Berlin), 83(6), 468–477. https://doi.org/10.1007/s00109-005-0635-7

Ploski, R., Pollak, A., Muller, S., Franaszczyk, M., Michalak, E., Kosinska, J., & Bilinska, Z. T. (2014). Does p. Q247X in TRIM63 cause human hypertrophic cardiomyopathy? Circulation Research, 114(2), e2–5. https://doi.org/10.1161/CIRCRESAHA.114.302662

Poplin, R., Ruano-Rubio, V., DePristo, M. A., Fennell, T. J., Carneiro, M. O., Van der Auwera, G. A., & Banks, E. (2017). Scaling accurate variant discovery to tens of thousands of samples. bioRxiv, 201178. https://doi.org/10.1101/201178

Rehm, H. L., Berg, J. S., Brooks, L. D., Bustamante, C. D., Evans, J. P., Landrum, M. J., Ledbetter, D. H., Maglott, D. R., Martin, C. L., Nussbaum, R. L., Plon, S. E., Ramos, E. M., Sherry, S. T., & Watson, M. S. (2015). ClinGen—the clinical genome resource. New England Journal of Medicine, 372(23), 2235–2242. https://doi.org/10.1056/NEJMsr1406261

Rentzsch, P., Witten, D., Cooper, G. M., Shendure, J., & Kircher, M. (2019). CADD: predicting the deleteriousness of variants throughout the human genome. Nucleic Acids Research, 47(D1), D886–D894. https://doi.org/10.1093/nar/gky1016

Richard, P., Charron, P., Carrier, L., Ledeuil, Céline, Chev, T., Pichereau, C., Benaiche, A., Isnard, R., Dubourg, O., Burban, M., Gueffet, J.-P., Millaire, A., Desnos, M., Schwartz, K., Hainque, B., & Komajda, M. (2003). Hypertrophic cardiomyopathy: Distribution of disease genes, spectrum of mutations, and implications for a molecular diagnosis strategy. Circulation, 107(17), 2227–2232. https://doi.org/10.1161/01.CIR.0000066323.15244.54

Richards, S., Aziz, N., Bale, S., D., Das, S., Gastier-Foster, J., Grody, W. W., Helg, M., Lyon, E., Spector, E., Voelkerding, K., & Rehm, H. L. (2015). Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. Genetics in Medicine, 17(5), 405–424. https://doi.org/10.1038/gim.2015.30

Roberts, W. C., Roberts, C. C., Ko, J. M., Grayburn, P. A., Tandon, A., Kuiper, J. J., Capehart, J. E., & Hall, S. A. (2013). Dramatically different phenotypic expressions of hypertrophic cardiomyopathy in male cousins undergoing cardiac transplantation with identical disease-causing mutation. American Journal of Cardiology, 111(12), 1818–1822. https://doi.org/10.1016/j.amjcard.2013.02.042

Santorelli, F. M., Tanji, K., Manta, P., Casali, C., Krishna, S., Hays, A. P., Mancini, D. M., DiMauro, S., & Hirano, M. (1999). Maternally inherited cardiomyopathy: An atypical presentation of the mtDNA 12S rRNA gene A1555G mutation. American Journal of Human Genetics, 64(1), 295–300. https://doi.org/10.1086/302188

Savostyanov, K. V., Namazova-Baranova, L. S., Basargina, E. N., Vakhramadze, N. D., Zhurkova, N. V., Pushkov, A. A., Zhanin, I. S., Sdvigova, N. A., Lukanina, V. Y., & Nikitin, A. (2017). The new genome variants in Russian children with genetically determined cardiomyopathies revealed with massive parallel sequencing. Annals of the Russian Academy of Medical Sciences, 72(4), 242–253. https://doi.org/10.15690/vramn872

Schneider, V. A., Graves-Lindsay, T., Howe, K., Bouk, N., Chen, H.-C., Kitts, P. A., Murphy, T. D., Pruitt, K. D., Thibaud-Nissen, F., Albracht, D., Fulton, R. S., Kremitzki, M., Magrini, V., Markovic, C., McGrath, S., Steinberg, K. M., Auger, K., Chow, W., Collins, J., ... Church, D. M. (2017). Evaluation of GRCh38 and de novo haploid genome assemblies demonstrates the enduring quality of the reference assembly. Genome Research, 27(5), 849–864. https://doi.org/10.1101/gr.213611.116

Seleznev, D. M., Gabrusenko, S. A., Parfenova, E. V., Naumov, V. G., Stambol'skii, D. V., & Tkachuk, V. A. (2005). The role of mutation in cardiac beta-myosin heavy chain gene in population of patients. Kardiologiia, 45(4), 15–20.

Semsarian, C., Ingles, J., Maron, M. S., & Maron, B. J. (2015). New perspectives on the prevalence of hypertrophic cardiomyopathy.
Novel correlations between the genotype and the phenotype of hypertrophic and dilated cardiomyopathy: Results from the German Competence Network for Cardiomyopathy. **European Journal of Heart Failure**, 13(11), 1185–1192. https://doi.org/10.1093/eurjhf/hfr074

Walsh, R., Buchan, R., Wilk, A., John, S., Felkin, L. E., Thomson, K. L., Chiaw, T. H., Loong, C. C. W., Pua, C. J., Raphael, C., Prasad, S., Barton, P. J., Funke, B., Watkins, H., Ware, J. S., & Cook, S. A. (2017). Defining the genetic architecture of hypertrophic cardiomyopathy: Re-evaluating the role of non-sarcomeric genes. **European Heart Journal**, 38(46), 3461–3468. https://doi.org/10.1093/eurheartj/ehw603

Walsh, R., Thomson, K. L., Ware, J. S., Funke, B. H., Woodley, J., McGuire, K. J., Mazzarotto, F., Blair, E., Seller, A., Taylor, J. C., Minikel, E. V., MacArthur, D. G., Farrall, M., Cook, S. A., & Watkins, H. (2017). Reassessment of Mendelian gene pathogenicity using 7,855 cardiomyopathy cases and 60,706 reference samples. **Genetics in Medicine**, 19(2), 192–203. https://doi.org/10.1038/gim.2016.90

Watkins, H., Anan, R., Coviello, D. A., Spirito, P., Seidman, J. G., & Seidman, C. E. (1995). A de novo mutation in alpha-tropomyosin that causes hypertrophic cardiomyopathy. **Circulation**, 91(9), 2302–2305. https://doi.org/10.1161/01.cir.91.9.2302

Weissler-Snir, A., Hindieh, W., Gruner, C., Fourey, D., Appelbaum, E., Rowin, E., & Chan, R. H. (2017). Lack of phenotypic differences by cardiovascular magnetic resonance imaging in MYH7 (beta-Musin Heavy Chain)- vs MYBPC3 (Myosin-Binding Protein C)-related hypertrophic cardiomyopathy. **Circulation: Cardiovascular Imaging**, 10(2), e005311. https://doi.org/10.1161/CIRCIMAGING.116.005311

Zeller, R., Ivanidc, B. T., Ehlermann, P., Mücke, O., Zugck, C., Remppis, A., Giannitsis, E., Katus, H. A., & Weichenhan, D. (2006). Large-scale mutation screening in patients with dilated or hypertrophic cardiomyopathy: a pilot study using DGGE. **Journal of Molecular Medicine (Berlin)**, 84(8), 682–691. https://doi.org/10.1007/s00109-006-0056-2

Zerbino, D. R., Achuthan, P., Akanni, W., Amode, M. R., Barrell, D., Bhai, J., Bills, K., Cummins, C., Gall, A., Girón, C. G., Gil, L., Gordon, L., Haggerty, L., Haskell, E., Hourlier, T., Izugu, O. G., Janacek, S. H., Juettemann, T., To, J. K., ... Flicek, P. (2018). Ensembl 2018. **Nucleic Acids Research**, 46(D1), D754–D761. https://doi.org/10.1093/nar/gkx1098

Zhao, B., Wang, S., Chen, J., Ji, Y., Wang, J., Tian, X., & Zhi, G. (2017). Echocardiographic characterization of hypertrophic cardiomyopathy in Chinese patients with myosin-binding protein C3 mutations. **Experimental and Therapeutic Medicine**, 13(3), 995–1002. https://doi.org/10.3892/etm.2017.4089

**SUPPORTING INFORMATION**

Additional Supporting Information may be found online in the Supporting Information section.

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