Supplementary material

Osadnik T, Frycz-Kurek A, Lejawa M, et al. Genotype — phenotype correlations in Polish patients with hypertrophic cardiomyopathy. Preliminary report. Kardiol Pol. 2022.

Genetic analyses. Genomic DNA was extracted from blood samples using MagCore HF 16 automated nucleic acid extractor (RBC Bioscience, Taiwan) and MagCore® Genomic DNA Whole Blood Kit according to the manufacturer's instructions. Patients' DNA samples were screened by targeted sequencing of 174 genes using TruSight™ Cardio Sequencing Kit (Illumina, USA). The gene panel (TruSight™ Cardio Sequencing Kit) was designed to screen coding sequences of the genes (exons) associated with 17 inherited cardiac conditions including HCM. Hybridization capture was carried out using the Illumina reagents. The library was prepared according to the manufacturer's instructions using a Veriti Dx PCR instrument (Thermo Fisher Scientific, USA). The concentration of the final library was verified using QuantiFluor ONE dsDNA System reagents (Promega Corporation, USA) on a Quantus Fluorometer (Promega Corporation, USA). The quality of the final library was assessed by capillary electrophoresis using the bio-fragment analyzer Qsep100 (BiOptic Inc, Taiwan). Enriched fragment libraries were sequenced by 2×150bp paired end sequencing protocol on the MiSeq System (Illumina, USA) using MiSeq Reagent Kit v2 chemistry (Illumina, USA). All the reads were aligned against the human reference genome hg19.

Bioinformatics analysis. Genomic variants were filtered and annotated by Variant Studio (Illumina, USA). We excluded variants with a frequency greater than 1% in the available databases. Synonymous, intronic, and variants outside the flanking regions were excluded. Missense variants were analysed with functional annotation algorithms – SIFT https://sift.bii.a-star.edu.sg) and PolyPhen-2 (http://genetics.bwh.harvard.edu/pph2/). Missense variants were considered to be pathogenic/likely pathogenic mainly by comparisons with data in ClinVar (https://www.ncbi.nlm.nih.gov/clinvar/), Varsome (https://varsome.com) and others. Databases were accessed from 1st December 2021 to 6th January 2022. Additionally, we run *in silico* prediction with SIFT and PolyPhen-2 algorithms. In this preliminary paper we decided to report

only variants that were reported as pathogenic or likely pathogenic by multiple submitters in clinical databases or variants that were reported as pathogenic by some submitters and variant of uncertain significance (VUS) by others but with the evidence of pathogenicity as defined in American College of Medical Genetics criteria (ACMG) [1]. Coding DNA reference transcripts are given in accordance with the HGVS nomenclature [2].

REFERENCES

1. Richards S, Aziz N, Bale S, et al. 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. Genet Med. 2015; 17: 405–424.
2. den Dunnen JT, Dalgleish R, Maglott DR, et al. HGVS Recommendations for the Description of Sequence Variants: 2016 Update. Hum Mutat. 2016; 37: 564–569.