Title: Next-generation sequencing of 68 genes in sudden unexplained death of young individuals in forensics

Author: L. Buscemi F. Alessandrini G. Perna A. Tagliabracci

PII: S1875-1768(15)30086-X
DOI: http://dx.doi.org/doi:10.1016/j.fsigss.2015.09.056
Reference: FSIGSS 1053

To appear in:

Received date: 24-8-2015
Accepted date: 14-9-2015

Please cite this article as: L.Buscemi, F.Alessandrini, G.Perna, A.Tagliabracci, Next-generation sequencing of 68 genes in sudden unexplained death of young individuals in forensics, Forensic Science International: Genetics Supplement Series http://dx.doi.org/10.1016/j.fsigss.2015.09.056

Next-generation sequencing of 68 genes in sudden unexplained death of young individuals in forensics

L. Buscemi[a], F. Alessandrini, G. Perna[b] and A. Tagliabracci
[a]Institute of Legal Medicine, Università Politecnica delle Marche, Ancona, Italy
[b] Department of Cardiology, Ospedali Riuniti di Ancona, Ancona, Italy

Corresponding author: buscemi@univpm.it, Tel: +39 071 5964721, Fax: +39 071 5964723.

Abstract
Sudden cardiac death in the young is a very dramatic event that occurs in apparently healthy individuals without an explainable cause of death. The advancement of knowledge in the field of molecular biology has suggested that genetic abnormalities are the leading cause of sudden unexplained death in young people. Three forensic SUD cases were studied. 68 genes most frequently associated with SUD were analyzed by NGS.

Keywords: Molecular autopsy, sudden cardiac death, young, next generation sequencing

1. Introduction

Sudden cardiac death in the young is a very dramatic event that occurs in apparently healthy individuals, under the age of 35 years, without an explainable cause of death. The advancement of knowledge in the field of molecular biology has suggested that genetic abnormalities are the leading cause of sudden unexplained death in young people. The molecular autopsy may identify a cause of death in 30% of cases of SUD.
Three forensic SUD cases were studied. 68 genes most frequently associated with SUD were analyzed by NGS on an Ion Torrent PGM instrument.

2. Materials and Methods

Three forensic SUD cases were studied: Case 1: ♀, 13, was found dead in her room. In her history was a syncopal episode at age 10. Autopsy was negative. Case 2: ♂, 18, who died during a football game. In his history there have been two syncopal episodes at 14 years. The autopsy revealed a circumferential subepicardial fibrosis of the left ventricle, with extension to the septum. Case 3: ♂, 29, died during a diving. The autopsy gave evidence of an asphyxiation by drowning. A paternal cousin had a SUD at 21 with a sibling suffering from arrhythmogenic cardiomyopathy, bearer of a defibrillator.
DNA was extracted from blood samples with the QIAamp DNA Micro Kit (Qiagen). 68 genes present in the Ion AmpliSeq IDP, associated with inherited channelopathies were investigated: Sequencing was performed on an Ion 318 chip using the Sequencing kit 200; alignment of sequences to the reference human genome (GRCh37/hg19) and base calling were performed using the Torrent Suite software v4.0.2 (Life Technologies). Variants passing the quality criteria were subjected to a filtering process in order to prioritize putative pathogenic mutations [1,2,3] and by taking into account the published estimates for the cardiac inherited disease prevalence [4]. The putatively pathogenic variants were confirmed by Sanger sequencing.

3. Results

Case 1: Missense mutation in KCNH2 gene (L955V), pathogenic for LQT2 syndrome (Fig. 1). Cause of death: sudden cardiac death in LQT2 syndrome. A clinical and genetic screening of family members was made and a maternal aunt with long QT was found.
Case 2: Nonsense mutation in MYH7 gene (K1587X) (Fig. 2). Mutations in MYH7 gene (myosin, heavy chain 7) have been associated with sudden cardiac death. Cause of death: sudden cardiac death in arrhythmogenic cardiomyopathy with prevalent left ventricular involvement.
Case 3: Mutation in ABCC9 gene (V1137I) (Fig. 3) associated with early repolarization syndrome, linked to increased risk of arrhythmic death. Cause of death: asphyxiation by drowning in the early repolarization syndrome.

4. Discussion

The introduction of NGS technology, especially the Ion Torrent PGM sequencing platform, consents to rapidly identify at a low cost a higher number of genetic variants potentially implicated in cardiac diseases and they might be used in routine practice in forensic genetics. A multidisciplinary approach including the forensic pathologist, the molecular biologist, the cardiologist and the genetic counselor is crucial to prevent further deaths in existing family members.

Conflict of interest: none

References

[1] M. Brion, A. Blanco-Verea, B. Sobrino, et al., Next generation sequencing challenges in the analysis of cardiac sudden death due to arrhythmogenic disorders, Electrophoresis 35 (2014) 3111–3116.
[2] C.L. Hertz, S.L. Christiansen, L. Ferrero-Miliani, et al., Next-generation sequencing of 34 genes in sudden unexplained death victims in forensics and in patients with channelopathic cardiac diseases, Int. J. Legal Med. (2014).