Reviewer's report

Title: Functional Effects of the TMEM43 Ser358Leu Mutation in the Pathogenesis of Arrhythmogenic Right Ventricular Cardiomyopathy

Version: 1 Date: 23 November 2011

Reviewer: Cristina Basso

Reviewer's report:

Rajkumar and colleagues have performed a multitask study comprising:
- The investigation of a small cohort of ARVC patients for mutations in the 6 ARVC-related genes (DSP, PKP2, DSG2, DSC2 and TMEM43);
- The effect of mutant S358L TMEM43 mutation on the desmosomal and nuclear protein localization. To this regard, they have transfected cells with TMEM43-containing plasmids and they examined the localization of desmosomal proteins by western blotting and immunofluorescence;
- The expression analysis of genes associated with laminopathies.

Major Compulsory Revisions

The present study is original in aiming to investigate the function of a TMEM43 mutation (S358L) and mechanism by which this mutation causes ARVC; however the value added to the present study by reporting genetic screening results of desmosomal genes in a small cohort of patients remains unclear. The authors should stress further the link between these mutations and the experimental investigation described on the present study.

Further, I would have few suggestions as follow:

Background section:

“Furthermore, mutant TMEM43 did not alter the expression of genes that are suggested to be associated with cardiomyopathy.” (page5, 13) The above mentioned statement of the authors implies that genes for a range of cardiomyopathies has been investigated. It should be more appropriate to refer only to “lamin-cardiomyopathies or laminopathies”.

Minor Essential Revisions

Methods section:

Please follow the MIQE guidelines (Bustin SA et al., Clinical Chemistry, 2009; 55:611-622) to report your qPCR experiments. Experimental procedures of gene expression analysis typically include sampling-processing steps, quality and quantity assessment, as well as reverse transcription steps. Consequently, sequence information for primers should be provided, how many biological and technical replicants were included and so on.

Moreover, refer to the description of genes (location and reason why investigated) targeted by qPCR experiments in the method section and not in the
Results section:
- Please refer to the HGVS guidelines regarding mutation nomenclature (i.e. page 11, Thr412AsnfsX2)
- Figure 1 should be correctly resized.
- Please note, that the plasmid ratio and protein extraction method should be described in methods section and not in results section (page 1, lanes 2,3) as well as the method to obviated solubility effects of TMEM43 in Triton-X100 (page 13 lanes 10,11)
- Figure 2A, the blot for DSP is not proper. Please provide a better picture of this blot.
- Figure 2A and 2B. Pictures from the immunoblot of a reference protein are lacking.
- Please move the description of genes (location and reason why investigated) targeted by qPCR experiments in the method section and not in the result section. (page 14 lanes 1,2)

Discussion section:
Compound/digenic heterozygosity in ARVC patients that leads to a more severe disease phenotype has been extensively described by Xu et al., JACC, 2010 and Bauce et al., Heart Rhythm, 2010.

Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests:
I declare that I have no competing interests