Re-classification of variants: Implications from the cardiac perspective

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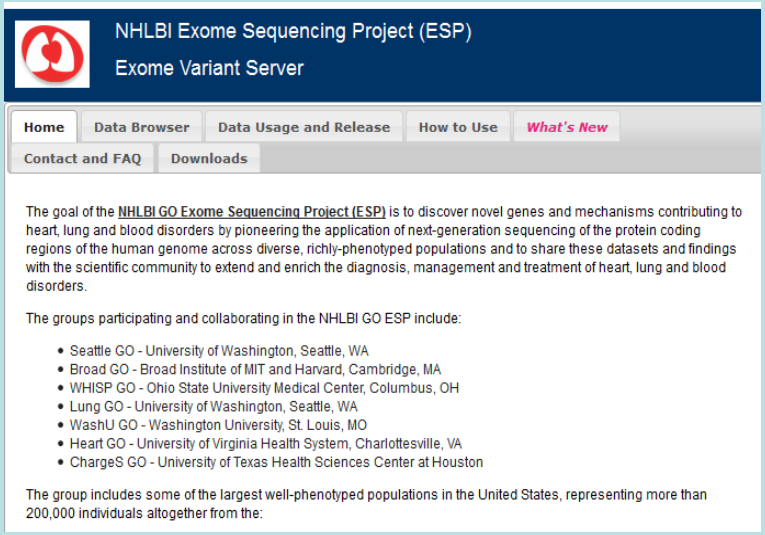
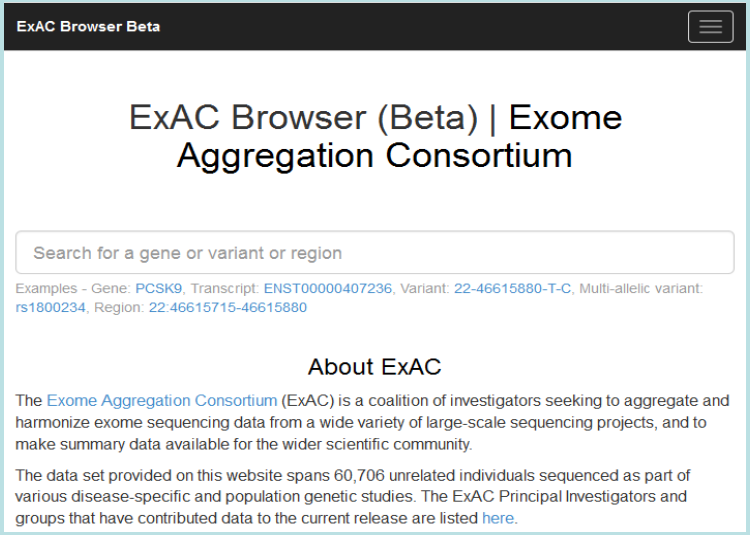
Re-classification

New evidence that changes the original interpretation and classification of the variant

Enhance Pathogenicity	Question Pathogenicity
De novo occurrence	Detection in multiple phenotypes
Segregation with disease	Detected alongside another putative pathogenic variant
Additional functional studies e.g. RNA	Non segregation
	Frequency in population based cohorts
UPGRADE	**DOWNGRADE**

Population-based cohorts





- Population ascertainment of samples
- Not pre-selected for having cardiomyopathy
- General prevalence of 1/500
- Allows investigation into general frequency of genetic variants in cardiac genes

Population-based cohorts

REPORT

Burden of Rare Sarcomere Gene Variants in the Framingham and Jackson Heart Study Cohorts

Alexander G. Bick,[1,2] Jason Flannick,[2,3] Kaoru Ito,[1] Susan Cheng,[4] Ramachandran S. Vasan,[5,6] Michael G. Parfenov,[1] Daniel S. Herman,[1] Steven R. DePalma,[1] Namrata Gupta,[2] Stacey B. Ga Birgit H. Funke,[7,15] Heidi L. Rehm,[7,15] Emelia J. Benjamin,[5,6,8] Jayashri Aragam,[4] Herman A. Taylor, Jr.,[9,10,16] Ervin R. Fox,[9] Christopher Newton-Cheh,[3,5,11] Sekar Kathiresan,[3,4] Christopher J. O'Donnell,[5,12] James G. Wilson,[13] David M. Altshuler,[2,3] Joel N. Hirschhorn,[1,2] J.G. Seidman,[1] and Christine Seidman[1,4,17,*]

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Population-Based Variation in Cardiomyopathy GenesClinical Perspective

Jessica R. Golbus, Megan J. Puckelwartz, John P. Fahrenbach, Lisa M. Dellefave-Castillo, Don Wolfgeher and Elizabeth M. McNally

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Evaluating Pathogenicity of Rare Variants From Dilated Cardiomyo Era

Nadine Norton, Peggy D. Robertson, Mark J. Rieder, Stephan Züchner, E Eden Martin, Duanxiang Li, Deborah A. Nickerson and Ray E. H

Circ Cardiovasc Genet. 2012;5:167-174; originally published online F
doi: 10.1161/CIRCGENETICS.111.961805
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ARTICLE

New population-based exome data are questioning the pathogenicity of previously cardiomyopathy-associated genetic variants

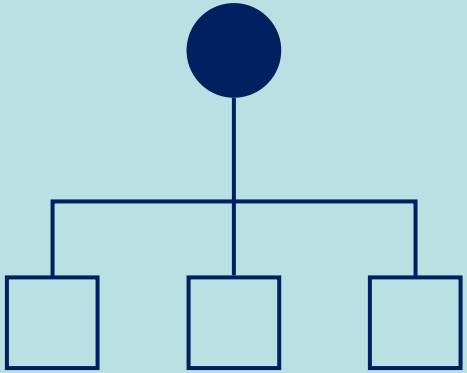
Charlotte Andreasen[1,2,5], Jonas B Nielsen[1,2,5], Lena Refsgaard[1,2], Anders G Holst[1,2], Alex H Christensen[1,2], [illegible][1,2], Ahmad Sajadieh[3], Stig Haunsø[1,2,4], Jesper H Svendsen[1,2,4] and Morten S Olesen[*,1,2]

Reassessment of Mendelian gene pathogenicity using 7,855 cardiomyopathy cases and 60,706 reference samples

Roddy Walsh, Kate Thomson, James S Ware, Birgit H Funke, Jessica Woodley, Karen J McGuire, Francesco Mazzarotto, Edward Blair, Anneke Seller, Jenny C Taylor, Eric V Minikel, Exome Aggregation Consortium, Daniel G MacArthur, Martin Farrall, Stuart A Cook, Hugh C Watkins

Genetics In Medicine, in press.

TNNT2 p.Arg288Cys



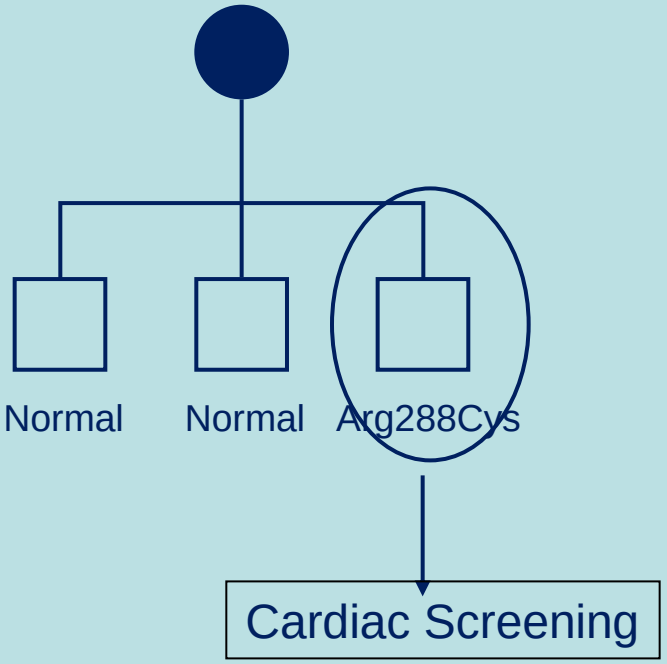
- Diagnosis of HCM aged 80
- No family history of HCM
- Targetted 4 gene screen
- *TNNT2* p.Arg288Cys detected

- Conserved across species
- Region of the protein where other pathogenic mutations were reported
- Detected in several individuals with HCM from multiple groups including UK[1], Spanish[2], Italian[3], Australian[4] and American[5].
- Not detected in normal controls

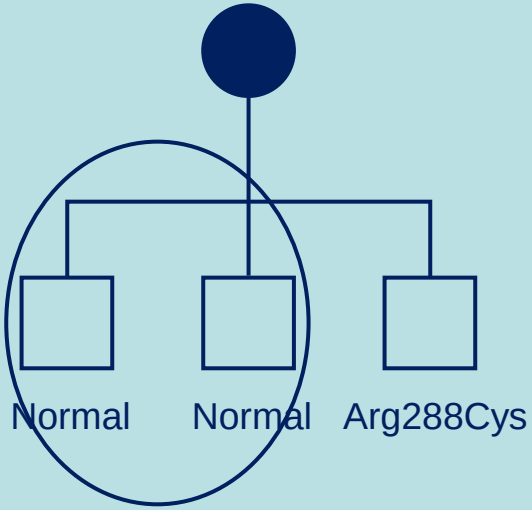
- **Highly likely to be pathogenic (2007)**

References: **[1]** Watkins *et al* (1995) *NEJM* **332** 1058 **[2]** Garcia-Castro et al. (2003) Clin Chem 49(8):1279-1285 **[3]** Torricelli et al. (2003) Am J Cardiol 92(11):1358-1362 **[4]** Ingles et al. (2005) J Med Genet 42(10)e59 **[5]** Van Driest et al (2003) Circ. 108(4):445-451

TNNT2 p.Arg288Cys

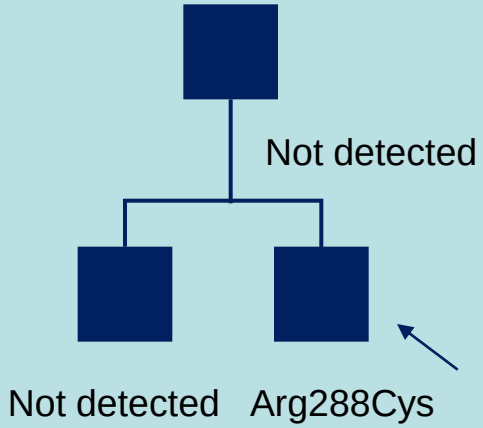


TNNT2 p.Arg288Cys



No clinical screening

TNNT2 p.Arg288Cys



Is this variant not a pathogenic mutation?

or

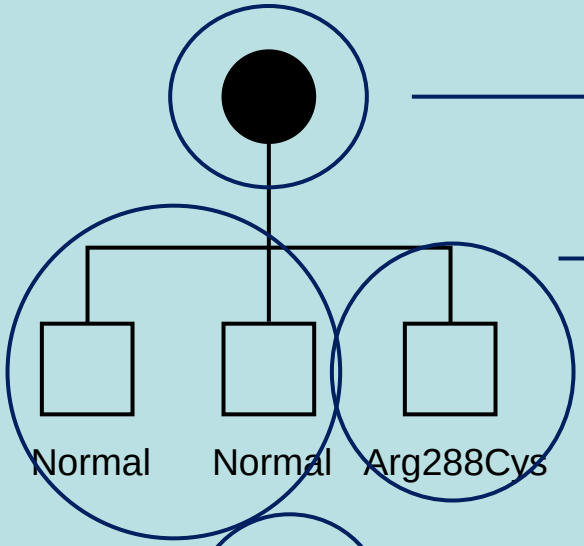
Is there a second pathogenic variant in this family?

TNNT2 p.Arg288Cys

Detected in 5 of >4,000 individuals (0.1%) of European ancestry and 1 of >2,000 individuals (0.04%) of African ancestry in NHLBI-EVS cohort.	**Is this a rare polymorphism or population specific polymorphism?**
Detected in 12 of 3,000 individuals in our own cohort (0.4%); however, 4 of these individuals also had a second putative pathogenic variant. This information is reflected in cases reported in the literature.	**Does this variant work in concert with other mutations to cause cardiomyopathy?** **Is it a mild variant?** **Is it sufficient to cause disease alone?**
Functional studies *in vitro* indicated that the variant does not have a significant effect on protein function.	**Is this variant sufficient to cause disease as a sole variant?**

Re-Classified as Unknown Pathogenicity

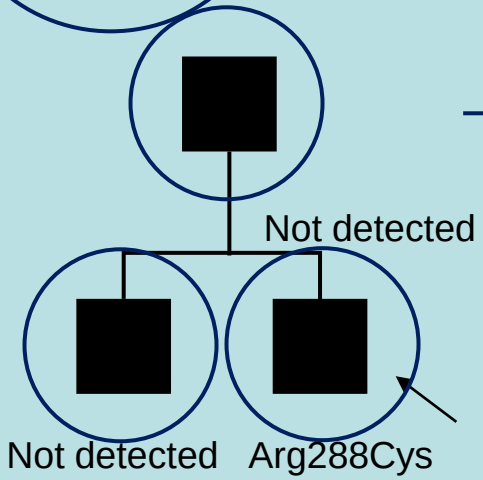
Clinical impact of re-classification



No further family testing offered

May not be “at risk” of developing HCM

May be ‘at-risk’
Offer clinical cardiac screening



Offered screening for known genes
Family testing is no longer informative

Offer extended screening / new panels to identify additional variants

Re-classifications 2013 - present

Mutation details	Class change	Families	Individuals
***TNNT2* c.862C>T p.(Arg288Cys)**	**Highly likely to UV**	**15**	**31**
***MYBPC3* c.2429G>A p.(Arg810His)**	**Highly likely to UV**	**7**	**13**
***MYBPC3* c.13G>A p.(Gly5Arg)**	**Likely to unlikely**	**13**	**45**
***MYBPC3* c.2003G>A p.(Arg668His)**	**Likely to unlikely**	**4**	**6**
***MYH7* c.2945T>C p.(Met982Thr)**	**Likely to unlikely**	**10**	**21**
MYBPC3 c.2441_2443delAGA p.(Lys814del)	Likely to UV	4	14
MYBPC3 c.3392T>C p.(Ile1131Thr)	Likely to UV	1	1
MYH7 c.5135G>A p.(Arg1712Gln)	Likely to UV	2	2
MYBPC3 c.530G>A p.(Arg177His)	UV to unlikely	2	2
MYBPC3 c.649A>G p.(Ser217Gly)	UV to unlikely	3	7
MYBPC3 c.961G>A p.(Val321Met)	UV to unlikely	3	6
MYBPC3 c.1519G>A p.(Gly507Arg)	UV to unlikely	3	5
MYBPC3 c.2870C>G p.(Thr957Ser)	UV to unlikely	2	2
MYBPC3 c.2914C>T p.(Arg972Trp)	UV to unlikely	1	1
PRKAG2 c.250C>T p.(Arg84Trp)	UV to unlikely	1	1
	Total	**70**	**156**

Variant Review

This data highlights a need for all clinical laboratories to review variants in light of new information.

- Review active variants with a frequency of 12 months
  - Variant detected in a new proband
  - Family test requested
  - Requested by the referring Clinician
  - Large new cohort of data is released
- Always report on the current classification
  - Sometimes discrepancy between different laboratories classifications
  - Contact with clinical team - OK to proceed?
  - Additional explanatory information on the report
- Review variants prior to clinic date for family testing

Future improvements

- **Guidelines on:**
  - Who is responsible for flagging variants for review?
  - How often should variants be reviewed?
  - What is the mechanism for review?
  - Which re-classifications require communication?

- **Bioinformatic solutions:**
  - Automatically extrapolate new information for review
  - Automatic variant reviews
  - Administration of re-classification letters

- Consistent approach to variant review and re-classification amongst ACGS laboratories.

Acknowledgements

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