A 49-year-old man with bicuspid aortic valve (BAV) and mild-to-moderate aortic valvular stenosis was referred for investigation of a dilated cardiomyopathy. At the time of presentation, he was physically active, New York Heart Association class I. Physical examination revealed a grade III/VI harsh crescendo-decrescendo murmur at right upper sternal border radiating to both carotids. His 12-lead ECG revealed a normal sinus rhythm and left ventricular hypertrophy with strain. He was treated with an angiotensin-converting enzyme inhibitor, β-blocker and was routinely followed up for 2 years. Further work up, which included a transthoracic echocardiogram (Figure 1Ai–1Av) and cardiac magnetic resonance imaging (Figure 1Av and 1Avi), established the diagnosis of left ventricular noncompaction (LVNC). Diagnosis of LVNC is primarily based on transthoracic echocardiogram and cardiac magnetic resonance imaging of the 2 layered structure of the myocardium with epicardial compacted and endocardial noncompacted zones, with estimation of the ratio of these 2 layers (noncompacted/compacted ≥2 by echo and ≥2.3 by cardiac magnetic resonance) and visualization of the perfused recesses. Evaluation with transthoracic echocardiogram and contrast (Figure 1Aii) and color Doppler (Figure 1Aiii) revealed prominent trabeculations, left ventricular apical recesses (sinusoids) in continuation with the ventricular cavity, reduced LV systolic function (ejection fraction [EF], 30%–35%) with concomitant LV dilatation. His BAV showed severe aortic valvular stenosis (aortic valve area 0.55 cm²/m², mean gradient of 70 mm Hg). Because of progressive dyspnea and reduced exercise tolerance coupled with proximal aortic dilatation and reduced cardiac output (EF <30%), he underwent successful aortic valve replacement with a 29-mm freestyle aortic valve and proximal ascending aorta replacement. He is currently asymptomatic with excellent exercise tolerance at New York Heart Association class I and EF of 40% to 45%.

Discussion

LVNC is an important cause of dilated cardiomyopathy caused by the cessation of intratubular compaction of myocardial fibers during endomyocardial morphogenesis in the first trimester. LVNC is characterized by multiple trabeculations with profound intratubecular recesses perfused by blood in the ventricular cavity. LVNC is regarded as a genetic disease, which can either affect as an isolated and distinct cardiomyopathy or be in combination with other forms of congenital heart diseases including BAV. In our proband, genetic studies revealed a novel MYH7:c.1316T>G mutation in exon 14 of the MYH7 gene resulting in a nonconserved single amino acid substitution of arginine for methionine in position 439 (M439R) of the cardiac β-myosin heavy chain head region and expressed in an autosomal dominant manner. Subsequent analysis of segregation studies followed up to 3 successive generations of the proband established an autosomal dominant pattern for disease inheritance with the mutation preserved in the affected relatives (Figure 1B).

The structural stability of the head region in facilitating actin–myosin interaction as an integral component of cardiomyocyte contraction is well known. Importantly, a comparative sequence analysis of β-myosin heavy chain (MHC) from different species confirmed a well-conserved hydrophobic amino acid in position 439 (Figure 2). To investigate the role of the point mutation at position 439, we performed comparative analysis of the homology model of the mutant (Figure 3Ai and 3Aii) with the crystal structure of the wild-type protein. A 3-dimensional energy-minimized model of the β-MHC head domain in the methionine-439 wild-type and arginine-439 mutant proteins illustrates a destabilized modeled mutant protein (Figure 3B). A rotamer analysis clearly suggests that the incorporation of a long flexible charged amino acid (Arg) in place of hydrophobic amino acid Met is associated with potential multiple radical alterations in the structural environment of the β-MHC head region (Figure 3C), which generally favors an evolutionary conserved hydrophobic amino acids (Met/Leu/Val) as typically found in normal β-MHC proteins in different species (Figure 2).

Mutations in the MYH7 gene are associated with a diverse phenotype of cardiomyopathies, including hypertrophic,
dilated, and restrictive cardiomyopathies. Indeed, MYH7 is the most common sarcomeric gene mutated in patients with LVNC and includes mutations in exons 8, 9, 30, and 37 in both familial and sporadic LVNC in the absence of other congenital heart defects. However, LVNC is a heterogeneous genetic disorder harboring mutations in other sarcomere protein genes with variable modes of inheritance. Apart from a solitary case report, which linked 22q11.2 distal deletion encompassing the BCR gene, little is known on the genetic association of LVNC concomitant with BAV. The association between LVNC and BAV may reflect other genetic alterations in these patients and the impact of early embryonic LVNC on the aortic valve morphogenesis. Our results highlight a novel MYH7 mutation in exon 14 associated with LVNC concomitant with BAV, and its potential impact on the function of the head region of the β-MHC protein, a critical determinant of the actin–myosin cross-bridge interaction in the heart.

Acknowledgments
We thank Gendia-Genetic Diagnostic Network, Antwerp, Belgium for conducting genetic testing using next-generation sequencing technology. We also thank the patients and their family members for their support.

Sources of Funding
This work was supported by operating grants from the Canadian Institutes of Health Research, Heart and Stroke Foundation of Canada, and Alberta Innovates–Health Solutions.

Disclosures
None.

References

Key Words: bicuspid aortic valve • cardiomyopathies • mutation
The diagnosis of left ventricular noncompaction (LVNC) and bicuspid aortic valve (BAV) was based on 2-dimensional (transesophageal and transthoracic) echocardiography with color flow Doppler and further confirmed by cardiac magnetic resonance (CMR). The imaging diagnostic criteria for LVNC and BAV were used as described previously. A, Transthoracic apical 4-chambered view with contrast enhancement showing prominent recesses at the apical and mid-lateral wall depicted with an arrow (i), color Doppler image revealing flow of blood into the intratrabecular recesses (shown with arrow) in continuation with the LV (ii), prominent trabeculations demonstrating noncompacted to compacted region (shown in higher magnification) in the LV apex and mid-lateral wall as they are seen without any contrast (iii) and short axis view demonstrating noncompacted to compacted ratio >2 (iv). Real-time CMR cine imaging (steady state free precession) with 4-chamber view and (v) short axis view in systole (vi) demonstrating endomyocardial trabeculation (shown with arrows) in the noncompacted apical and mid-lateral regions of the LV and LVNC concomitant to BAV. B, Based on our genetic study, we found parent to child transmission affecting both sexes in an autosomal mode of inheritance associated with a high penetrance and expressivity as demonstrated by a pedigree chart for ≥2 successive generations. Genetic test was performed by next-generation sequencing using an Illumina Solexa platform for the coding regions including the splice sites of 27 genes known to be associated with familial dilated cardiomyopathy. C indicates compacted; LA, left atrium; LV, left ventricle; NC, noncompacted; RA, right atrium; and RV, right ventricle.
Figure 3. Crystal structure of wild-type human myosin head-like domain (PDBID: 4DB1) was used as a template for homology modeling of the M439R mutant protein. The molecular visualization programs, COOT (Crystallographic Object-Oriented Toolkit) and SWISSPDB (Swiss-PdbViewer), were used to manipulate the position of amino acid residues. In silico analysis of the cardiac human beta-myosin heavy chain (MHC) containing the methionine 439 arginine mutation. The predicted model was improved by steepest descent energy minimization method using the GROMACS (GROningen MAchine for Chemical Simulations) software package and the OPLS-AA (Optimized Potentials for Liquid Simulations-All Atom) force field modeled structure. Comparison of a 3-dimensional energy-minimized model of the head-like domain of beta-MHC between normal (Met439wt) and mutated (Arg439mut) proteins, and rotamer analysis of the M439R mutant protein revealing the presence of 5 different possible conformers.

Figure 2. Sequence comparison showing presence of a conserved hydrophobic amino acid (Met/Leu/Val) at position 439 (site of mutation) in the beta-myosin heavy chain protein across different species. Hs-myo1c refers to a member of the unconventional cytoplasmic human myosin protein family. Dd indicates Dictyoselium discoideum; Dm, Drosophila melanogaster; Gallas, Gallas gallas; Hs, Homo sapiens; Pig, Sus scrofa; Rn, Rattus norvegicus; Scallop, Argopecten irradians; and Squid, Doryteuthis pealeii.
Novel Mutation in Exon 14 of the Sarcomere Gene MYH7 in Familial Left Ventricular Noncompaction With Bicuspid Aortic Valve
Ratnadeep Basu, Saugata Hazra, Miriam Shanks, David I. Paterson and Gavin Y. Oudit

Circ Heart Fail. 2014;7:1059-1062
doi: 10.1161/CIRCHEARTFAILURE.114.001666

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circheartfailure.ahajournals.org/content/7/6/1059