Circulation: Genomic and Precision Medicine

EDITORIAL

Additional Genetic Variants in Inherited Dilated Cardiomyopathy

Just Another Brick in the Wall?

See Article by Cowan et al

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Clinical diagnostic genetic testing for different types of cardiomyopathies has increasingly become a part of clinical practice during the last decade or so.[1] In dilated cardiomyopathy (DCM), a large number of genes has been identified underlying genetic, mostly autosomal dominantly inherited forms of the disease. The key role for genetic testing is cascade genetic screening in family members after the identification of a class 4 (likely pathogenic) or a class 5 (pathogenic) variant in an index patient. Interpreting such a probabilistic outcome and translating it into clinical practice requires a team effort and may lead to reassurance of family members who do not have the pathogenic variant and subsequent dismissal from regular cardiological follow-up, whereas those not having the class 4 (likely pathogenic) variant are followed with larger intervals or are dismissed after being informed about the risks and potential symptoms that belong to the respective disorder.[2] Family members that do have the likely pathogenic or pathogenic variant are, however, regularly followed up by the cardiologist and timely treatment may prevent or delay the development of disease-related features.

In general, genotype does not guide treatment in genetic cardiomyopathies with a few exceptions. The most important exception is the gene encoding lamin A/C gene (*LMNA*). Disease-associated *LMNA* variants can be identified in ≈6% of DCM patients.[3–6] Patients with *LMNA* related cardiac disease generally demonstrate a highly penetrant phenotype characterized by sinus node dysfunction, atrioventricular conduction disease, atrial fibrillation or flutter, DCM, and in some cases muscular dystrophy.[6–8] Early observations already showed high rates of sudden cardiac death in patients with an *LMNA* disease-causing variant with a pacemaker for associated conduction disease, which suggested a tachyarrhythmic mode of death.[9,10] Implantable cardioverter defibrillator implantation in *LMNA* gene positive patients who were in need of a pacemaker seemed effective in treating possibly lethal tachyarrhythmias.[10]

Later studies demonstrated that male sex, a nonmissense mutation, nonsustained ventricular tachycardias, and a left ventricular ejection fraction <45% are important predictors for either dismal outcome or earlier presentation.[11,12]

This important *LMNA* gene is the subject of study by Cowan et al[13] in this issue of *Circulation Genomic and Precision Medicine*. They report on a follow-up study of 19 pedigrees with apparent *LMNA* related DCM. In 6 of these families studied, there was at least 1 individual that did not have the putative disease-causing *LMNA* variant even though a diagnosis of idiopathic DCM was made. Because of reduced or age-dependent penetrance in cardiac genetic disease, we are used to identify family members with the disease-causing variant, yet without any signs of associated disease. However, in relatively rare diseases, like inherited cardiomyopathies,

Key Words:: Editorials ■ cardiomyopathy, dilated ■ gene ■ genetic testing ■ risk

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identifying affected family members without the ostensible disease-causing variant is an unusual observation.

The current study by Cowan et al[13] is a follow-up study on an original article published a decade ago, in which limited segregation in sometimes large pedigrees was observed.[4] Of interest, this nonsegregation has also been observed by others for example in a large multigenerational pedigree, because of an exon-1 *LMNA* deletion underlying the cardiomyopathic phenotype.[14]

After the initial observation of nonsegregation in 6 *LMNA* families, Cowan et al[13] collected additional evidence for pathogenicity of the variants in those families.[15] In 4 of the 6 *LMNA* variants from the nonsegregating pedigrees, in vitro studies of nuclear morphology and green fluorescent protein-lamin A localization demonstrated abnormalities underscoring the pathogenicity of the variants. This conclusion argued for the existence of a second unidentified causative factor in the majority of families showing nonsegregation. Because next-generation sequencing methods were not readily available at that time, only a limited number of additional genes were studied. In their discussion, they already suggested that either misclassification of the variant identified, additional subtle environmental influences (toxins, viruses, etc), or genetic factors, such as a mutation in an additional DCM gene, could underly this nonsegregation.[4]

In the present study, Cowan et al[13] used exome sequencing methods to hunt for an additional genetic cause of DCM in these 6 families. In addition, they were able to enrich the initial data with additional clinical follow-up and family data. They identified at least 1 additional rare variant in a known DCM gene that could plausibly contribute to disease in the *LMNA* variant-negative individuals and confirmed bilineal inheritance in 3 of 5 families studied and was possible or presumptive in 2. The identified variants were rare, occurred in conserved regions of genes (with strong biological associations with DCM), yet did not fulfill the formal American College of Medical Genetics criteria for being pathogenic or likely pathogenic.[16]

When observing the different pedigrees, one has to stay critical about the interpretation of the different genetic variants. It is well known that variant interpretation is difficult with substantial discordance in classification.[17] In family L, for example, an 18-year earlier presentation of DCM as compared to a family member was explained by the presence of the additional LMNA p.(Ala318Thr) variant which did not show any in vitro abnormalities in their previous study and was classified as a variant of unknown significance.[15] To our opinion, the truncating titin *(TTN)* A band variant is more likely to be the genetic culprit as it was also identified in the family, and these variants have a high prior likelihood of being disease-causing.[18] This also holds true for pedigree O where a truncating A band TTN mutation was associated with disease, and the additional *LMNA* variant p.(Arg399Cys); (also without abnormalities in an in vitro assay) was present in the index patient with a 15- to 20-year earlier onset as compared to family members. Two family members with only the *LMNA* variant did not show any signs of *LMNA* related disease at 43 and 76 years, respectively. Attributing earlier disease onset to and additional *LMNA* variant of unknown significance without abnormalities in functional tests warrants a careful approach, particularly in *TTN* related disease because environmental factors are known to contribute to *TTN* related disease development: we and others have recently shown that people with TTN truncating variants respond well to therapy and in those with additional factors, including environmental factors, develop disease more easily.[19–22] So potential exogenic factors could also explain the variability in the age of onset rather than a variant of unknown significance without functional evidence for a pathogenic effect. In general, this illustrates that with current large-scale genomic analyses one is likely to run into a large number of truncating variants in TTN that may upset the initial assessment of a pedigree. With a few percent of the population carrying such truncating TTN variants, this will be a frequently encountered phenomenon that stirs up the clinical appearance and apparent mode of inheritance in a given DCM pedigree.[18]

Also for families N and P, the 2 identified variants do not explain the full clinical picture in the family because N.5, N.22 and N.28, and P.22 show more or less signs of DCM in the absence of the 2 identified variants in *LMNA/RBM20* and *LMNA/NEXN* respectively.

The detailed observations by Cowan et al[13] illustrate that even variants in the highly penetrant *LMNA* gene sometimes fail to segregate and may be influenced by more variants leading to more severe disease. The former has also been shown, for example, in Brugada syndrome. Probst et al[23] have shown that in a substantial number of pedigrees the Brugada syndrome phenotype was present in several family members without the pathogenic SCN5A mutation. In the latter, the presence of additional variants that may aggravate disease, is a well known phenomenon in cardiogenetic disease as, for example, in hypertrophic cardiomyopathy, long QT syndrome, or arrhythmogenic right ventricular cardiomyopathy, a few percent of patients have additional disease-associated variants, and this is associated with earlier presentation, or worse disease outcome.[24–26] This oligogenic pattern of inheritance cannot be ignored in cardiogenetic disease and has, in addition to the families in this study, also been observed in other families with *LMNA* related disease.[27,28] In addition, environmental factors, like hypertension, obesity, and vigorous exercise have also been shown to aggravate inherited cardiac disease.[29,30]

What does this mean for everyday clinical practice? These observations emphasize that we have to think beyond the borders of simple monogenic disease, to carefully take a family history and evaluate all close fam-

ily members and may be keep them under surveillance or at least instruct them well on the signs and symptoms associated with disease even in the absence of the culprit genetic lesion in the family. Like suggested by the authors, we should avoid to say that lack of an identified family variant, even the one considered pathogenic, relieves family members who do not carry that variant (or variants) of any future disease liability. It is to be preferred to state that the risk is decreased, as other genetic and environmental risk factors may still be present.

Future studies with large series of families, deeply-phenotyped and genotyped, and interviewed for environmental factors are needed to fully understand clinical variability and nonpenetrance and nonsegregation of disease and the extent of the oligogenic or multifactorial nature in genetic cardiomyopathies. This detailed study by Cowan et al[13] is a great initiative to be followed up.

ARTICLE INFORMATION

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Acknowledgments

We acknowledge the support from the Netherlands Cardiovascular Research Initiative, an initiative with support of the Dutch Heart Foundation (CVON2014-40 DOSIS).

Disclosures

None.

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