The Genetics of Left Ventricular Non-Compaction

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LVNC Genetics

Abstract

Purpose of review: This article summarises current understanding of the genetic architecture underpinning left ventricular non-compaction (LVNC) and highlights the difficulty in differentiating LVNC from hypertrabeculation seen in normal, healthy individuals, that caused by physiological adaptation or that seen in association with cardiomyopathy phenotypes.

Recent findings: Progress has been made in better defining the LVNC phenotype and those patients who may benefit from genetic testing. Yield of diagnostic genetic testing may be low in the absence of syndromic features, systolic dysfunction and a family history of cardiomyopathy. Sarcomeric gene variants are most commonly identified but a wide-range of genes are implicated, emphasising the high degree of heterogeneity of studied cohorts.

Summary: More accurate phenotyping and genotype-phenotype correlation is required to better characterise the genetic architecture of LVNC.

Keywords: non-compaction, genetics, hypertrabeculation, cardiomyopathy

Introduction

Left ventricular non-compaction (LVNC) is a complex, heterogenous, morphological abnormality of the myocardium, defined by the presence of prominent left ventricular trabeculae and deep inter-trabecular recesses.[1] Originally described as a consequence of arrested cardiac morphogenesis,[2] LVNC occurs as a genetic trait often in association with congenital heart defects and different cardiomyopathy phenotypes. A lack of robust criteria for differentiation from normal anatomic variants and secondary increases in trabecular mass mean that it has also been reported in normal healthy individuals or as an acquired reversible trait. When differentiation between pathology, normal variation and physiological adaptation is uncertain, the term hypertrabeculation rather than non-compaction is often used. When associated with LV hypertrophy, LV dilatation with systolic

impairment, or restrictive physiology, the term non-compaction cardiomyopathy is sometimes used but again lacks a universal definition.

Definitions

Non-compaction as a concept refers to an abnormal ventricular morphology characterised by the coincidence of a thin outer layer of normal or 'compacted' myocardium with an inner 'non-compacted' layer consisting of prominent muscular trabeculae and intra-trabecular recesses that communicate with the ventricular cavity. The term derives from the normal processes by which muscular trabeculae, formed during early embryogenesis, coalesce and form mature myocardium. Non-compaction, in this sense, implies a premature arrest of this process that results in persistence of an abnormal trabecular architecture which is present at birth. As cardiomyocytes are terminally differentiated cells, new trabecular formation cannot occur in the adult heart, but increases in ventricular mass can occur by hypertrophy of existing cardiomyocytes.

Non-compaction is most frequent in the apex of the left ventricle (**figure 1**) but may occur in both ventricles or, rarely, be confined to the right ventricle. Numerous diagnostic criteria for LVNC using different imaging modalities have been proposed, but with no accepted gold standard.[3–5] As a consequence, reported prevalence rates differ substantially.

In a recent systematic review and meta-analysis of studies reporting LVNC prevalence in adults, a total of 59 papers reporting 67 distinct cohorts were analysed.[6] Two-thirds reported patients with cardiac disease and the rest were population-representative volunteers, healthy controls, athletes and patients with no known cardiac disease. The prevalence of LVNC varied as a function of imaging modality, diagnostic criteria and study population. Cardiac magnetic resonance imaging was more sensitive than transthoracic echocardiography with a 12-fold higher reported frequency across all cohorts. Irrespective of the imaging modality, athletes, healthy controls and patients without cardiac disease had the highest prevalence of LVNC.

The occurrence of a supposedly abnormal phenomenon in so many normal healthy people and individuals with non-cardiac disease seems implausible and illustrates the extreme caution required when diagnosing LVNC, particularly in situations where physiological remodelling of the ventricle results in prominent rather than architecturally abnormal ventricular trabeculae. Normal ethnic variation in ventricular morphology also needs to be considered.[7]

The Genetic Architecture of LVNC

In spite of nosological confusion, LVNC is described as a familial trait and causative genetic variants are described in approximately one-third of patients with the condition.[8–11] Variants in a wide-range of genes have been reported in association with hypertrabeculation **(table 1**[12]**).** Recent studies have served to clarify these findings and provide insight into genetic risk factors.

A recent systematic review assessed 561 LVNC patients with pathogenic variants across 172 studies.[13] Variants in 66 genes were implicated in the development of LVNC with *MYH7* the most frequent, seen in one quarter of patients. Children were more likely to have LVNC associated with syndromes with complex genotypes, X-linked inherited conditions, mitochondrial and chromosomal defects. They were more likely to have congenital heart defects, neuromuscular symptoms and major adverse cardiovascular events (MACE). In comparison, adults displayed autosomal dominant inheritance of predominantly sarcomere genes with lower rates of MACE. These findings underline important aetiological and clinical differences between paediatric and adult populations with LVNC.

In a recent retrospective cohort of 327 unrelated adults and children with LVNC, 32% were found to carry a pathogenic variant.[14] These variants ranged across 22 known cardiomyopathy genes, 82% of which affected cardiac sarcomeric proteins; *MYH7*, *MYBPC3* and *TTN* variants were most frequent. Sixteen percent of the cohort reported a family history of cardiomyopathy without a pathogenic variant found, suggesting that yield may increase with better genotype-phenotype correlation to guide pathogenicity classification. Conversely, a third of patients with a pathogenic variant had no family history, highlighting the potential importance of genetic testing in apparently sporadic

disease. No difference in risk of MACE was observed among adults with genetic versus sporadic disease. However, not all patients received the full 45-gene panel and the panel lacked potentially important genes such as *FLNC* and *NKX2-5*. A significant difference in MACE was observed between children with and without a pathogenic variant.

A third study aimed to understand the yield of genetic testing in family members, reviewing 473 relatives of 113 index patients with LVNC.[15] Fifty eight percent of relatives of probands with a pathogenic variant also carried a pathogenic variant, with 63% of these demonstrating a cardiomyopathy phenotype. Using clinical screening alone, only 29% of relatives had features of cardiomyopathy. This shows the value of genetic testing over and above clinical tests in identifying a population at risk as well as the variable and incomplete disease penetrance of pathogenic variants. Affected relatives had less severe phenotypes than probands, offering the opportunity to initiate early, potentially preventive therapy.

The yield of genetic testing in adults outside the context of family screening was examined in a smaller study.[16] Thirty five unrelated patients underwent genetic testing with a 193-gene panel including mitochondrial genes. Only 3 patients (9%) were found to have a pathogenic variant, two in *NKX2-5* and one in *TBX5*. No pathogenic variants were found in patients with isolated LVNC in the absence of cardiac dysfunction or syndromic features. The absence of sarcomeric genes is notable and may be linked to the exclusion of patients presenting for family screening. A further 26% of the cohort were identified as having variants of unknown significance, again highlighting the need for better genotype-phenotype correlation to guide variant classification.

<u>New candidate genes for LVNC</u>

A prospective study of 95 unrelated adult LVNC patients underwent a 107-gene panel with a yield of 52 pathogenic or likely pathogenic variants in 40 patients using American College of Genetic Medicine criteria (42%).[17,18] As in other cohorts, a significant proportion of pathogenic variants were found in *TTN* (19%) and in *MYH7* (10%). However, 10% of variants were seen in *HCN4* and 8% in

RYR2. Both genes have been implicated in the development of hypertrabeculation [19–21], but they are not consistently part of the gene panels used in cohort studies.

HCN4 encodes the hyperpolarisation-activated cyclic nucleotide-gated channel 4, responsible for the 'funny' current in the sinoatrial node. Initially associated with familial sinus bradycardia, there is increasing evidence of its involvement in a syndrome of bradycardia, mitral valve defects, aortic dilatation and hypertrabeculation.[19,20] Hypertrabeculation may, be a physiological response to bradycardia and, of note, 3 of the 5 patients with *HCN4* variants in the cohort reported by Richard et al. presented with bradycardia. One family underwent segregation analysis with 2 family members found to have isolated bradycardia and 2 with bradycardia in addition to LVNC.

RYR2 exon 3 deletions have been reported in association with LVNC in patients with catecholaminergic polymorphic ventricular tachycardia (CPVT) and bradycardia.[21] The cardiac ryanodine receptor is an essential Ca2+ release channel of the sarcoplasmic reticulum, playing a central role in excitation-contraction coupling in cardiomyocytes. The patients identified by Richards et al. are classified as isolated LVNC but the presence or absence of features of bradycardia, LV systolic dysfunction or CPVT are not specifically discussed. The possibility remains that hypertrabeculation is a response to bradycardia, arrhythmia or systolic dysfunction in these patients.

ACTC1 is a sarcomeric gene encoding for the cardiac protein α-actin. Variants in *ACTC1* are associated with a non-compaction phenotype, seen in 7% of patients in a recent large systematic review. *ACTC1* pathogenic variants resulted in less LV dilatation than other sarcomere variants and, along with *MYH7*, conferred the lowest risk of MACE..[13] A novel variant in *ACTC1* was recently reported as co-segregating in a family with co-existent hypertrophic cardiomyopathy, non-compaction and transmural crypts.[22] *ACTC1* variants have also been associated with LVNC and septal defects[23] and LVNC and arrhythmias.[24]

<u>Phenotypic Overlap</u>

In 2006, The American Heart Association defined LVNC as a genetic cardiomyopathy.[25] However, more recent guidelines do not treat it as a disease entity independent from other cardiomyopathies, suggesting, instead, that clinicians use gene panels for the cardiomyopathy identified in association with the LVNC phenotype.[26] Phenotypic overlap with other cardiomyopathies is reflected in the genetic basis of specific disease subtypes. For example, truncating mutations in the *TTN* gene are the most frequent genetic cause of dilated cardiomyopathy (DCM)[27,28] and a high prevalence of *TTN*tv mutations is therefore unsurprising where LVNC cohorts are, for example, comprised of patients with left ventricular systolic dysfunction and dilatation.[17,29]

Recent studies have attempted to subclassify LVNC patients by associated cardiac phenotype. A paediatric LVNC cohort of 65 individuals were sub-categorised as isolated LVNC (30/65), LVNC co-occurring with cardiomyopathy (25/65) or LVNC co-occurring with cardiovascular malformation. The overall yield of a cardiomyopathy gene panel was 9% (6/65) with no pathogenic variants found in the isolated LVNC group.[30]

Precision Medicine

Genetic research offers a glimpse of a future where both clinical and genetic features combine to produce tailored management strategies and personalised risk assessment. Mutations in certain genes may be predictors of MACE in LVNC and thus require individualised management and follow-up.

In a small cohort of LVNC patients diagnosed using cardiac MRI or at post-mortem, left ventricular function improved over time with optimal-medical therapy in patients without a pathogenic variant but worsened in those with a pathogenic variant. The suggestion that genotype-negative patients may have reversible disease could have important consequences for patients in whom an implantable cardiac defibrillator is considered.[31]

Pathogenic *MYH7* variants are consistently prevalent in LVNC cohorts and, overall, seem to carry a lower risk of MACE.[14] Non-compaction with a DCM phenotype was associated with variants in the tail domain of *MYH7*. Variants in this location interfere with the binding site for *TTN* and thus may be more likely to cause a dilated phenotype**.** Right ventricular dysfunction was also more common in *MYH7* tail-domain variants versus those in the head domain. Variant location in *MYH7* therefore seems to influence phenotype. This is important as LVNC with a dilated phenotype seems to result in poorer outcomes.[15]

Improved Phenotyping

Endocardial trabeculae are a feature of the normal left ventricle and their extent varies among healthy subjects of different racial backgrounds. Current imaging criteria for LVNC do not account for this normal spectrum and are mostly semi-quantitative, relying on subjective delineation of endocardial borders that fails to consider the complex three-dimensional architecture of the myocardium. As a consequence, existing methods for diagnosis of LVNC correlate poorly and have low reproducibility. By using a box-counting method on CMR short-axis cine stacks, fractional dimension (FD), a unitless index that measures how completely a complex structure fills space, has been shown to have a characteristic pattern from base to apex along the LV (**figure 2**[32]). In healthy volunteers, black individuals have a higher FD than whites in the apical third of the left ventricle and when comparing LVNC (defined using existing CMR criteria) with healthy volunteers, maximal apical FD is higher in LVNC. Importantly, the fractal method appears more accurate and reproducible than other CMR criteria for LVNC.[7]

In a recent study of image-derived cardiac phenotypes of 18,096 UK Biobank participants, fractal analysis of cardiac MRI data was used to define trabecular morphology. A genome-wide association study was then performed, identifying 16 significant loci containing genes associated with haemodynamic phenotypes and regulation of cytoskeletal arborisation.[33] Trabeculae-associated loci were analysed for relationships with cardiovascular disease with linkage disequilibrium techniques

used to screen for genetic correlations between trabecular complexity and patient traits. Diagnosed vascular or heart problems were strongly correlated with decreased complexity of trabeculations while hypertension phenotypes were associated with increased trabecular complexity. Curiously, loci associated with decreasing trabecular complexity were found to be associated with increased susceptibility to both dilated cardiomyopathy and heart failure phenotypes; the locus around *GOSR2* (involved in cytoskeletal actin dynamics) showed a particularly strong association (yet to be replicated). The relationship of this observation to the increased propensity to cardiomyopathy and heart failure in patients with an embryonic non-compaction phenotype remains unclear. A number of identified loci overlapped with sarcomeric genes, such as *TTN* and *TNNT2*, previously reported in association with LVNC.

Observational data from the same UK Biobank study was used to show that increasing fractal dimension was associated with higher stroke volume, stroke work and contractility. This correlated with a biomechanical simulation which suggested a relationship between trabecular complexity and ventricular performance. Fractal analysis techniques were applied to cardiac MRI scans from DCM patients, showing greater trabecular complexity than in control participants. The authors suggested that trabeculae maintained cardiac performance in both healthy and failing hearts by increasing contractility and stroke work, but given the inability of the heart to generate new trabeculae, this seems counterintuitive and may simply reflect hypertrophy of existing structures.

Conclusions

Where hypertrabeculation is extreme, LVNC is easily diagnosed. Where it is mild, caution should be exercised when labelling a patient with the diagnosis. Emphasis should be placed on clinical assessment, family history and the elucidation of other signs consistent with myocardial abnormalities.[34] When the diagnosis is clear, genetic counselling and testing should be considered. New diagnostic methods such as fractal analysis may offer more accurate phenotyping and facilitate the creation of clearly defined LVNC study cohorts.

Bullet points:

1) Caution must be exercised when diagnosing left ventricular non-compaction.
2) Genetic testing may be indicated where the diagnosis is clear and particularly in the presence of additional myocardial abnormalities, syndromic features or a family history of cardiomyopathy.
3) More accurate phenotyping and genotype-phenotype correlation is required to better characterise the underlying genetic architecture of LVNC.

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References

1. Towbin, J. A., Lorts, A. & Jefferies, J. L. Left ventricular non-compaction cardiomyopathy. in *The Lancet* vol. 386 813–825 (Lancet Publishing Group, 2015).

2. Bleyl, S. B. *et al.* Neonatal, lethal noncompaction of the left ventricular myocardium is allelic with Barth syndrome. *Am. J. Hum. Genet.* **61**, 868–872 (1997).

3. Petersen, S. E. *et al.* Left ventricular non-compaction: Insights from cardiovascular magnetic resonance imaging. *J. Am. Coll. Cardiol.* **46**, 101–105 (2005).

4. Jenni, R., Oechslin, E., Schneider, J., Attenhofer Jost, C. & Kaufmann, P. A. Echocardiographic and pathoanatomical characteristics of isolated left ventricular non-compaction: A step towards classification as a distinct cardiomyopathy. *Heart* **86**, 666–671 (2001).

5. Chin, T. K., Perloff, J. K., Williams, R. G., Jue, K. & Mohrmann, R. Isolated noncompaction of left ventricular myocardium. A study of eight cases. *Circulation* **82**, 507–513 (1990).

6. **Ross, S. B. *et al.* A systematic review and meta-analysis of the prevalence of left ventricular non-compaction in adults. *European Heart Journal* vol. 41 1428-1436b (2020).

A systematic review and metanalysis that serves to highlight the large discrepencies between rates of diagnosis of LVNC across different imaging modalities and the high rates of diagnosis in cohorts of healthy individuals, athletes and patients without cardiac disease.

7. Captur, G. *et al.* Fractal analysis of myocardial trabeculations in 2547 study participants: Multi-ethnic study of atherosclerosis1. *Radiology* **277**, 707–715 (2015).

8. Oechslin, E. & Jenni, R. Left ventricular non-compaction revisited: A distinct phenotype with genetic heterogeneity? *European Heart Journal* vol. 32 1446–1456 (2011).

9. Probst, S. *et al.* Sarcomere gene mutations in isolated left ventricular noncompaction cardiomyopathy do not predict clinical phenotype. *Circ. Cardiovasc. Genet.* **4**, 367–374 (2011).

10. Klaassen, S. *et al.* Mutations in sarcomere protein genes in left ventricular noncompaction. *Circulation* **117**, 2893–2901 (2008).

11. McNally, E. & Dellefave, L. Sarcomere Mutations in Cardiogenesis and Ventricular Noncompaction. *Trends in Cardiovascular Medicine* vol. 19 17–21 (2009).

12. Finsterer, J. & Stöllberger, C. Left Ventricular Noncompaction Syndrome: Genetic Insights and Therapeutic Perspectives. *Current Cardiology Reports* vol. 22 (2020).

13. **van Waning, J. I., Moesker, J., Heijsman, D., Boersma, E. & Majoor-Krakauer, D. Systematic Review of Genotype-Phenotype Correlations in Noncompaction Cardiomyopathy. *Journal of the American Heart Association* vol. 8 (2019).

A systematic review of LVNC patients with pathogenic variants aimed at assessing genotype-phenotype correlations. Key differences between paediatric and adult cohorts are demonstrated highlighting the need to adjust diagnostic and management strategies by age.

14. van Waning, J. I. *et al.* Genetics, Clinical Features, and Long-Term Outcome of Noncompaction Cardiomyopathy. *J. Am. Coll. Cardiol.* **71**, 711–722 (2018).

15. *van Waning, J. I. *et al.* Cardiac Phenotypes, Genetics, and Risks in Familial Noncompaction Cardiomyopathy. *J. Am. Coll. Cardiol.* **73**, 1601–1611 (2019).

A study examining the yield of genetic testing in family members of LVNC patients that highlights the

potential value of genetic testing above that of clinical screening alone in this population.

16. **Ross, S. B. *et al.* Genetic architecture of left ventricular noncompaction in adults. *Hum. Genome Var.* **7**, 33 (2020).

A small but important study emphasising the low yield of genetic testing in patients presenting outside of family screening, without concurrent syndromic features or other myocardial abnormalities.

17. *Richard, P. *et al.* Targeted panel sequencing in adult patients with left ventricular non-compaction reveals a large genetic heterogeneity. *Clin. Genet.* **95**, 356–367 (2019).

A prospective study that highlights the potential value of using large gene panels to evaluate cohorts diagnosed with LVNC, yielding variants in less frequently implicated genes such as HCN4 and RYR2

18. Richards, S. *et al.* Standards and guidelines for the interpretation of sequence variants: A joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. *Genet. Med.* **17**, 405–424 (2015).

19. Schweizer, P. A., Koenen, M., Katus, H. A. & Thomas, D. A Distinct Cardiomyopathy: HCN4 Syndrome Comprising Myocardial Noncompaction, Bradycardia, Mitral Valve Defects, and Aortic Dilation. *Journal of the American College of Cardiology* vol. 69 1209–1210 (2017).

20. Milano, A. *et al.* HCN4 mutations in multiple families with bradycardia and left ventricular noncompaction cardiomyopathy. *J. Am. Coll. Cardiol.* **64**, 745–756 (2014).

21. Ohno, S. *et al.* Exon 3 deletion of RYR2 encoding cardiac ryanodine receptor is associated with left ventricular non-compaction. *Europace* **16**, 1646–1654 (2014).

22. Frustaci, A. *et al.* Novel α-actin gene mutation p.(Ala21Val) causing familial hypertrophic cardiomyopathy, myocardial noncompaction, and transmural crypts. clinical-pathologic correlation. *J. Am. Heart Assoc.* **7**, (2018).

23. Monserrat, L. *et al.* Mutation in the alpha-cardiac actin gene associated with apical hypertrophic cardiomyopathy, left ventricular non-compaction, and septal defects. *Eur. Heart J.* **28**, 1953–1961 (2007).

24. Yoshida, Y. *et al.* A novel ACTC1 mutation in a young boy with left ventricular noncompaction and arrhythmias. *Hear. Case Reports* **2**, 92–97 (2016).

25. Maron, B. J. *et al.* Contemporary definitions and classification of the cardiomyopathies: An American Heart Association Scientific Statement from the Council on Clinical Cardiology, Heart Failure and Transplantation Committee; Quality of Care and Outcomes Research and Functional Genomics and Translational Biology Interdisciplinary Working Groups; and Council on Epidemiology and Prevention. *Circulation* **113**, 1807–1816 (2006).

26. *Musunuru, K. *et al.* Genetic Testing for Inherited Cardiovascular Diseases: A Scientific Statement From the American Heart Association. *Circ. Genomic Precis. Med.* **13**, e000067 (2020).

Contains a change to previous guidance, no longer treating LVNC as a distinct disease entity, suggesting clinicians use a gene panel appropriate for the associated cardiomyopathy instead

27. Mcnally, E. M. & Mestroni, L. Dilated cardiomyopathy: genetic determinants and mechanisms HHS Public Access. *Circ Res* **121**, 731–748 (2017).

28. Akhtar, M. M. *et al.* Clinical Phenotypes and Prognosis of Dilated Cardiomyopathy Caused by Truncating Variants in the TTN Gene . *Circ. Hear. Fail.* (2020)

doi:10.1161/circheartfailure.119.006832.

29. Pinto, Y. M. *et al.* Proposal for a revised definition of dilated cardiomyopathy, hypokinetic non-dilated cardiomyopathy, and its implications for clinical practice: a position statement of the ESC working group on myocardial and pericardial diseases. *Eur. Heart J.* **37**, 1850–8 (2016).

30. Miller, E. M. *et al.* Genetic Testing in Pediatric Left Ventricular Noncompaction. *Circ. Cardiovasc. Genet.* **10**, (2017).

31. Lorca, R. *et al.* Characterization of Left Ventricular Non-Compaction Cardiomyopathy. *J. Clin. Med.* **9**, 2524 (2020).

32. Captur, G. *et al.* Quantification of left ventricular trabeculae using fractal analysis. *J. Cardiovasc. Magn. Reson.* **15**, (2013).

33. **Meyer, H. V. *et al.* Genetic and functional insights into the fractal structure of the heart. *Nature* **584**, 589–594 (2020).

A large study using fractal analysis cardiac MRI techniques to characterise trabecular complexity in a large Biobank cohort and perform a genome-wide association study to identify loci of interest and link them to population-level cardiac traits.

34. Protonotarios, A. & Elliott, P. M. Left ventricular non-compaction: Have we reached the limits of conventional imaging? *European Heart Journal* vol. 41 1437–1438 (2020).