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Genetic Dilated Cardiomyopathy Due to TTN Variants without Known Familial Disease

Running title: *Brown et al.; TTN pathogenic variants in isolated DCM*

Emily E. Brown, MGC[1]; Brittney Murray, MS[1]; Joban Vaishnav, MD[1];

Emmanouil Tampakakis, MD[1]; Lili A. Barouch, MD[1]; Cynthia James, MS, PhD[1];

Anne M. Murphy, MD[2]; Daniel P. Judge, MD[1,3]

[1]Division of Cardiology, [2]Division of Pediatric Cardiology, Johns Hopkins University, Baltimore, MD; [3]Division of Cardiology, Medical University of South Carolina, Charleston, SC

Correspondence:
Emily E. Brown
Division of Cardiology
Johns Hopkins University
600 N. Wolfe St., Blalock 572
Baltimore, MD 20278
T: (410)-502-2578
Email: ebrow102@jhmi.edu

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Dilated cardiomyopathy (DCM) is characterized by left ventricular (LV) enlargement with reduced LV ejection fraction[1]. Among those without coronary artery disease, approximately 35% have a familial or genetic cause to their cardiomyopathy[2]. The majority of familial DCM is thought to be inherited in an autosomal dominant manner, and therefore, although it lacks specificity, family history is often used as a tool to both identify individuals who likely have a genetic etiology and guide genetic testing, despite recent guidelines which highlight the insensitivity of this approach[3]. Isolated nonischemic DCM can have a genetic etiology[4] due to a variety of factors including environmental contributions, *de novo* variants, limited availability of family history, recessive inheritance, and reduced penetrance. Genetic testing is often not considered in these individuals, especially if they present at age over 50 years. Therefore, there is limited information available regarding the frequency of genetic etiologies in these isolated cases.

After obtaining Institutional Review Board approval, we reviewed the family history and clinical characteristics of 83 probands with nonischemic DCM seen at the Johns Hopkins Center for Inherited Heart Disease between 2008-2020 whose genetic test results indicated a pathogenic or likely pathogenic variant. Participants either agreed to participate in a prospective registry (2018-2020) or fell into a retrospective review (2008-2017). Patients with known *de novo* variants were excluded. Twenty-seven of the identified individuals had pathogenic or likely pathogenic variants in the *TTN* gene. We report a case series of six patients presenting with nonischemic DCM with negative family histories in which subsequent genetic testing identified likely pathogenic variants in *TTN*. Because of the sensitive nature of the data collected for this study, requests to access the dataset from qualified researchers trained in human subject confidentiality protocols may be sent to the corresponding author.

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Patient demographics and clinical features are listed in the table. Most commonly, patients presented with dyspnea and/or chest pain. Age of presentation ranged from 14 years to 64 years. Cardiac evaluations for all six individuals were consistent with nonischemic DCM. Stress tests and/or coronary angiographies were negative for ischemia for all except Patient 6 who did not undergo specific evaluation for coronary artery disease given her young age. Of note, Patient 6 did present in the context of marijuana use, but she denied use of other illicit drugs. Five of the six individuals had prominent arrhythmia, with either paroxysmal ventricular tachycardia or atrial fibrillation.

Three-generation family histories were obtained by a cardiac genetic counselor. None of the individuals had a known family history of cardiomyopathy, heart failure, or sudden death. Patient 4 did have a family history of coronary artery disease with her father undergoing coronary artery bypass surgery. Unfortunately, familial records were not available for review in order to confirm this negative history.

Patients 2, 3, 4, and 6 were referred for genetic counseling and genetic testing because they presented before the age of 50 years old without an alternative etiology for heart failure which is standard procedure at our center. Patients 1 and 5 were self-referred for genetic counseling based on personal concern of the etiology for their cardiomyopathy. Genetic analysis was performed on either saliva or whole blood samples and included next generation sequencing and deletion/duplication analysis. The specific genetic panel ordered varied between two commercial clinical laboratories, with a minimum of 50 genes associated with cardiomyopathy. Variants were classified using the American College of Medial Genetics Guidelines. Results are listed in the table. In all six individuals, a pathogenic or likely pathogenic variant was identified

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in *TTN*, confirming a genetic etiology for their cardiomyopathy. If a variant of uncertain significance was identified, these are listed in the table as well.

Truncating variants in *TTN* are a common cause of DCM, particularly those occurring in the encoded A-band of titin, or in exons with constitutive expression and those spliced into a high proportion of left ventricular mRNA[5]. Additionally, they contribute to several other forms of DCM, including peripartum and those associated with alcohol and cancer chemotherapy[4]. Since approximately 1% of people have truncating TTN variants, development of cardiac dysfunction probably involves genetic or environmental modifiers in most of those with DCM[4]. Although familial DCM is considered a Mendelian trait, genetic DCM probably includes a much larger percentage of those whose cardiomyopathy would otherwise be considered idiopathic or due to prior viral infection.

Positive genetic test results not only have important implications for the patient but also have significant implications for family members[3]. In the six families reported here, genetic testing provided important information regarding familial risk, and allowed for cascade screening. This case series illustrates that a negative family history should not exclude the possibility of a genetic etiology for DCM as almost of quarter (22%) of patients with a pathogenic variant in *TTN* had an unremarkable family history. The older ages at the time of diagnosis for patients 1 and 5 suggest that neither age nor family history should exclude the consideration of genetic testing for nonischemic DCM.

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References:

1. Maron BJ, Towbin JA, Thiene G, Antzelevitch C, Corrado D, Arnett D, Moss AJ, Seidman CE, Young JB. 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*. 2006;113:1807-16.

2. Pugh TJ, Kelly MA, Gowrisankar S, Hynes E, Seidman MA, Baxter SM, Bowser M, Harrison B, Aaron D, Mahanta LM, et al. The landscape of genetic variation in dilated cardiomyopathy as surveyed by clinical DNA sequencing. *Genet Med*. 2014;16:601-8.

3. Hershberger RE, Givertz MM, Ho CY, Judge DP, Kantor PF, McBride KL, Morales A, Taylor MR, Vatta M, Ware SM. Genetic evaluation of cardiomyopathy—a Heart Failure Society of America practice guideline. *J Card Fail*. 2018;24:281-302.

4. Ware JS, Cook SA. Role of titin in cardiomyopathy: from DNA variants to patient stratification. *Nat Rev Cardiol*. 2018;15:241.

5. Herman DS, Lam L, Taylor MR, Wang L, Teekakirikul P, Christodoulou D, Conner L, DePalma SR, McDonough B, Sparks E, et al. Truncations of titin causing dilated cardiomyopathy. *N Engl J Med*. 2012;366:619-28.

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Table. A) Patient demographics and clinical features either. **B**) Pathogenic or likely pathogenic *TTN* variant identified. **C**) Other variants of uncertain significance identified.

A

Study ID	Gender	Ethnicity	Age at Presentation	Presenting feature	NYHA Class	Ejection Fraction	LVIDd (cm)	ECG Findings	Arrhythmias
1	Male	Caucasian	55	Dyspnea	II	20%	7.1	LBBB	--
2	Male	Caucasian	28	Dyspnea	II	45%	4.6	PVCs, AV block	VT
3	Male	Indian	48	Chest pain	I	40%	5.18	NSR	--
4	Female	Caucasian	64	PVCs	II	37%	5.68	PVCs, NSVT	--
5	Female	Caucasian	46	Dyspnea	IV	25%	5.9	PVCs	VT
6	Female	African American	14	Dyspnea, chest pain	IV	25%	5.45	PVCs, tachycardia	VT

B

Study ID	Nucleotide change	Predicted amino acid change	Exon	PSI	ACMG Criteria Met	ACMG Classification
1	c.98134G>T	p.Glu32712*	352	100	PVS1, PM2	Likely Pathogenic
2	c.53901dupG	p.Arg17968AlafsX12	280	100	PVS1, PM2	Likely Pathogenic
3	c.63025C>T	p.Arg21009*	304	100	PVS1, PM2	Likely Pathogenic
4	c.96937C>T	p.Gln32313*	348	100	PVS1, PM2	Likely Pathogenic
5	c.86627del	p.Pro28876fs	326	100	PVS1, PM2	Likely Pathogenic
6	c.70162C>T	p.Arg23388*	326	100	PVS1, PM2	Likely pathogenic

C

Study ID	Gene	Nucleotide change	Predicted amino acid change	ACMG Criteria Met	ACMG Classification
1	*MYBPC3*	c.56T>C	p.Val19Ala	PM2, BP4	VUS
2	*TXNRD2*	c.591+1 G>T	IVS7+1 G>T	None	VUS
2	*KCNH2*	c.2854 C>T	p.Pro952Ser	BP4	VUS
2	*NKX2-5*	c.89 C>A	p.Ala30Asp	BP4	VUS
3	*LAMA4*	c.3796 T>G	p.Phe1266Val	PM2	VUS
3	*LAMA4*	c.3646 G>C	p.Val1216Leu	PM2	VUS
5	*DSG2*	c.1038_1040delGAA	p.Lys346del	BP3	VUS
6	*RBM20*	c.1024 C>A	p.Pro342Thr	PM2, BP4	VUS
6	*DMD*	c.652 G>A	p.Val218Ile	PM2,PM5,BP4	VUS

Ejection fraction is either at the time of diagnosis or report available closest to the time of diagnosis. The *TTN* gene transcript used is NM_001267550.2. PVC = premature ventricular contraction; LBBB = left bundle branch block; VT = ventricular tachycardiac; NSVT = nonsustained ventricular tachycardia; Afib = atrial fibrillation; NSR = normal sinus rhythm; -- = no history of arrhythmias; PSI = percent spliced in; PVS = pathogenic very strong; PM = pathogenic moderate; BP = benign pathogenic; VUS = variant of uncertain significance