Genetic Predictors and Remodeling of Dilated Cardiomyopathy in Muscular Dystrophy

John L. Jefferies, MD, MPH; Benjamin W. Eidem, MD; John W. Belmont, MD, PhD; William J. Craigen, MD, PhD; Stephanie M. Ware, MD, PhD; Susan D. Fernbach, RN, BSN; Steven R. Neish, MD; E. O’Brian Smith, PhD; Jeffrey A. Towbin, MD

Background—Dystrophin gene mutations cause 2 common muscular dystrophies, Duchenne muscular dystrophy (DMD) and Becker muscular dystrophy (BMD). Both are frequently associated with dilated cardiomyopathy (DCM) and premature death. We hypothesized that early diagnosis and treatment of DCM in DMD/BMD patients would lead to ventricular remodeling and that specific dystrophin gene mutations would predict cardiac involvement.

Methods and Results—Sixty-nine boys with DMD (n=62) and BMD (n=7) (mean age, 12.9 and 13.7 years, respectively) were referred to our Cardiovascular Genetics Clinic for evaluation, including echocardiography and DNA analysis. Follow-up evaluations were scheduled yearly until the first abnormal echocardiogram indicative of DCM and quarterly thereafter. After the first abnormal echocardiogram, angiotensin-converting enzyme inhibitor or β-blocker therapy was started. β-Blockers were added if echocardiography showed no ventricular remodeling in angiotensin-converting enzyme inhibitor–treated patients after 3 months. DCM was diagnosed in 31 subjects (DMD, 27/62, 44%; BMD, 4/7, 57%) (mean age at onset, 15.4±2.8 years; range, 10.4 to 21.2 years). All 31 subjects were begun on pharmacological therapy after diagnosis. On follow-up (n=29), 2 subjects (both DMD) showed stable DCM, 8 subjects (all DMD) showed improvement, and 19 subjects (16 DMD; 3 BMD) showed normalization of left ventricular size and function (total improvement, 27/29 [93%]). DNA analysis in 47 cases (68%) revealed a significant association between DCM and exon 12 and 14 to 17 mutations, possible protection against DCM by exon 51 to 52 mutations, and a trend toward significant association between onset of DCM and exon 31 to 42 mutations. Statistical significance was based on nominal probability values.

Conclusions—Early diagnosis and treatment of DCM may lead to ventricular remodeling in DMD/BMD patients. Specific dystrophin gene mutations appear to be predictive of cardiac involvement, while other mutations may protect against or inhibit development of DCM. Further studies evaluating the impact of early intervention strategies on left ventricular geometry and function in muscular dystrophy patients seem warranted. (Circulation. 2005;112:2799-2804.)

Key Words: cardiomyopathy ■ genetics ■ muscular dystrophy ■ remodeling ■ risk factors

Duchenne muscular dystrophy (DMD), a severe form of skeletal myopathy, and the less severe Becker muscular dystrophy (BMD) are 2 common forms of muscular dystrophy, occurring in 1 of every 3500 liveborn males. Both diseases result from mutations in the dystrophin gene,1 a 2.5-Mb gene located on chromosome Xp21.1. Dystrophin is among the largest known genes, encoding a 14-ki transcript and a 427-kDa protein.2 Clinically, both DMD and BMD are characterized by skeletal myopathy associated with elevated serum creatine kinase (muscle isoform CK-MM) and calf pseudohypertrophy.3 Boys afflicted with DMD often become wheelchair-bound before 12 years of age; those afflicted with BMD usually experience later onset and slower progression of myopathic disease, typically becoming wheelchair-bound after 16 years of age.

The role of cardiac involvement in muscular dystrophy has been recognized for decades. Although the extent of involvement varies, cardiac disease is present by 20 years of age in essentially all boys with DMD and ~70% of those with BMD.4,5 Conduction system disease and tachyarrhythmias may occur in some individuals,6 but the electrocardiographic changes, ventricular arrhythmias, and ventricular late potentials associated with such conditions appear to be of little value in predicting mortality.7 The most common cardiac...
abnormality in both DMD and BMD is dilated cardiomyopathy, which in its symptomatic phase is usually associated with congestive heart failure and significantly associated with premature death.2,8

Cardiac symptoms typically appear late in the course of the cardiomyopathy, in part because affected individuals are usually wheelchair-bound and often physically inactive. Cardiac symptoms appear more often in patients over 18 years of age.9 Disease tends to progress rapidly, leading to premature death, often before 25 years of age. No successful treatments for cardiomyopathy in these individuals have been reported. The purposes of this study were (1) to determine whether early diagnosis and treatment of dilated cardiomyopathy leads to ventricular remodeling and (2) to determine whether genetic predictors of cardiac involvement exist in boys with DMD and BMD.

Methods

Study Design and Patient Recruitment
The study design was a retrospective chart review. Subjects included boys with the diagnosis of DMD or BMD who were referred by request to the Texas Children’s Hospital Cardiovascular Genetics Clinic by local pediatricians, neurologists, or cardiologists.

Study Protocol
Blood and tissue samples were obtained for laboratory studies, including creatine kinase measurement and DNA analysis on 47 patients. The remaining patients did not have DNA samples available for review. DNA analysis was performed at the Kleberg DNA Diagnostic Laboratory in the Department of Molecular and Human Genetics at Baylor College of Medicine in Houston, Texas. Blood samples from the study subjects were examined by dystrophin multiplex amplification and by Southern blot analysis of HindIII DNA with the 8 cDNA probes that cover the full extent of the dystrophin gene cDNA sequence, following standard procedures. The dystrophin gene sequence was examined for deletion and duplication mutations.

At each subject’s initial clinic visit, a past medical history, medication history, and family history were obtained, and a physical examination was performed. Echocardiography was performed with patients in the supine position when possible and upright in a wheelchair when necessary. No patients required MRI for imaging. Echocardiographic measurements included left ventricular (LV) end-diastolic diameter, LV end-systolic diameter, LV posterior-wall diastolic diameter, interventricular septal-wall diastolic diameter, LV fractional shortening, LV ejection fraction, LV myocardial performance index (MPI), and LV sphericity. LV sphericity was calculated by dividing the LV short-axis dimension by the LV long-axis dimension. LV MPI was calculated by dividing the sum of the isovolumic relaxation and contraction times by the ejection time. Doppler echocardiography of atrioventricular valve inflow was also performed. An abnormal echocardiogram was defined as one showing either depressed LV systolic function (normal LV ejection fraction ≥55%) or LV dilation (>2 Z scores from normal values for body surface area). A normal MPI was defined as <0.35±0.03.10 ECG and chest radiography were performed in all patients.

Follow-up clinic visits and echocardiography were scheduled once yearly until the time of the first abnormal echocardiogram. After that, angiotensin-converting enzyme (ACE) inhibitor therapy was initiated, and follow-up clinic visits and echocardiography were scheduled quarterly. Response to therapy was assessed by monitoring serial echocardiographic changes in LV dimensions and systolic function. Serial echocardiographic monitoring of LV diastolic function was assessed via mitral and pulmonary venous Doppler inflow patterns, Doppler tissue imaging, and LV MPI when available. The ACE inhibitor dose was titrated upward when possible, and in many cases there was at least no deterioration of cardiac function while on ACE inhibitory therapy alone. If ventricular remodeling (ie, improved or normalized LV dimensions, systolic function, or both) did not occur within 3 months after initiation of ACE inhibitor therapy, then β-blocker therapy was added. All drug therapies were instituted on an outpatient basis.

Statistical Analysis
Standard descriptive techniques were used to estimate incidence rates and CIs. We used χ² analysis to compare groups with respect to nominal outcomes. ANOVA was used to compare groups with respect to continuous outcomes. Exon-specific associations were determined by relating mutation (yes/no) to cardiac involvement (yes/no) in a 2×2 table with the use of Fisher exact test. Comparison of parameters before and after medical therapy was performed by a paired t test. Probability values <0.05 were considered significant.

Results
Sixty-nine consecutive subjects with a confirmed muscular dystrophy (62 with DMD and 7 with BMD) were included in this study (Table). Of these, 31 subjects (27 with DMD and 4 with BMD) showed cardiac involvement (44% versus 57%; P=0.69). Overall, the mean age at onset of cardiac disease was 15.4±2.8 years (range, 10.4 to 21.2 years). However, subjects in the BMD group were slightly younger at disease onset than those in the DMD group (13.9±3.0 versus 15.5±2.8 years; P=0.69). The average total follow-up was 3.3 years from time of initial evaluation (BMD group follow-up, 2.9 years; DMD group follow-up, 3.3 years).

Genetic analysis of DNA from 47 of 69 subjects (68%) showed several specific exon mutations to be associated with cardiac involvement. Mutations in exons 12 (P=0.03), 14 (P=0.01), 15 (P=0.03), 16 (P=0.03), and 17 (P=0.03) were all shown to be associated with onset of cardiomyopathy. Differences between BMD and DMD involving mutations at exons 12 and 14 to 17 were as follows: exon 12 (BMD 0%, DMD 9.8%; P=0.57); exon 14 (BMD 33.3%, DMD 7.3%; P=0.12); and exons 15, 16, and 17 (BMD 33.3%, DMD 4.9%; P=0.07). A trend toward statistical significance was seen for the association between mutations in exons 31 to 42 and cardiomyopathy (P=0.08). Cardiomyopathy occurred in all subjects who had mutations in exons 10, 11, and 18 to 30, although this finding was not statistically significant (P=0.19) because of sample size limitations. Mutations in exons 51 and 52 appeared to protect against cardiac involvement (P=0.02).
and \( P=0.05 \), respectively). None of the subjects who had mutations in exons 53, 54, and 68 to 71 manifested cardiac disease; however, this finding was not statistically significant \( (P=0.99) \).

All 31 subjects with cardiac disease were managed on a progressive heart-failure therapy regimen of ACE inhibitors or a combination of ACE inhibitors plus \( \beta \)-blockers. No patient was intolerant of ACE inhibitor or \( \beta \)-blocker therapy, and all drug therapy institution and titration were performed on an outpatient basis. No significant adverse clinical effects secondary to any single or combination of medications were reported. ACE inhibitors were used as single therapy in 13 of 31 patients (42%); combination ACE inhibitor/\( \beta \)-blocker therapy was used in the remaining 18 of 31 patients (58%). In 2 cases, no follow-up echocardiographic data were available at study termination. The remaining 29 subjects each had at least 1 follow-up clinic evaluation and echocardiogram during the study period. Two of 29 subjects (8%) (both with DMD) had stable dilated cardiomyopathy and showed no deterioration in LV size or function, 8 (26%) (all with DMD) showed improvement in LV size or function or both, and 19 (66%) (16 with DMD and 3 with BMD) showed normalization of LV size or function or both. The mean (±SD) LV end-diastolic dimension was 5.2±0.9 cm before therapy and 4.8±0.9 cm after therapy \( (P=0.001) \) (Figure, A). The mean (±SD) LV ejection fraction was 36±11% before therapy and 53±12% with therapy \( (P<0.001) \). All patients showed improvement in ejection fraction (Figure, B). The mean (±SD) LV MPI was 0.53±0.2 before therapy and 0.38±0.1 with therapy \( (P<0.001) \). All patients showed improvement in LV MPI with therapy; in 1 outlier case, the improvement was dramatic (Figure, C). The mean (±SD) LV sphericity index was 0.73±0.1 before therapy and 0.59±0.1 with therapy \( (P<0.001) \). All patients also showed improvement in LV sphericity with therapy (Figure, D).

The mean age at initiation of ACE inhibitor therapy in these 29 subjects was 14.1 years for patients with BMD versus 16.1 years for those with DMD. Enalapril was the most frequently used ACE inhibitor (86%, 25/29), although dosages varied depending on drug tolerance and disease extent. Captopril (10%, 3/29) and lisinopril (4%, 1/29) were used much less frequently. The average doses for enalapril, captopril, and lisinopril were 3.6 mg (range, 2.5 to 10 mg) orally twice daily, 7.6 mg (range, 6 to 10 mg) orally 3 times daily, and 5 mg (range, 5 mg) orally once daily, respectively. The mean age at initiation of \( \beta \)-blocker therapy was 16.0 years for patients with BMD versus 18.1 years in patients with DMD. Carvedilol (57%, 8/14) and
metoprolol (43%, 6/14) were the most frequently used β-blockers. The average doses for metoprolol and carvedilol were 21 mg (range, 5 to 50 mg) orally twice daily and 4 mg (range, 3.125 to 6.25 mg) orally twice daily, respectively. The mean duration of ACE inhibitor therapy was 2.7 years, and the mean duration of β-blocker therapy was 1.6 years. The need for hospitalization secondary to cardiac problems was not assessed in this study.

**Discussion**

The results of the present study indicate that early noninvasive diagnosis and treatment of dilated cardiomyopathy can lead to ventricular remodeling in boys with DMD and BMD. The results also indicate that specific mutations of the dystrophin gene may result in predisposition to or protection against dilated cardiomyopathy.

Dystrophin gene mutations are well-documented causes of cardiac disease. Molecular genetic approaches have been used to identify the dystrophin gene as the gene responsible for DMD, BMD, and X-linked cardiomyopathy. Dystrophin is believed to provide structural support for the myocyte and cardiomyocyte sarcolemmal membrane by its linking of actin at the amino-terminus with the dystrophin-associated protein complex and sarcolemma at the carboxyl-terminus and the extracellular matrix of muscle. Mutations in dystrophin or dystrophin-associated protein subcomplexes result in skeletal cardiomyopathies in humans and in mice. Disruption of the amino-terminus of the dystrophin molecule occurs in adults and children with end-stage cardiomyopathy (dilated or ischemic) and in individuals with enterovirally induced cardiomyopathies. This suggests the existence of a final common pathway for cardiomyopathies in which the links between sarcolemma and sarcomere are critical. This dystrophin-linkage abnormality at the amino-terminus has been shown to be reversible by treatment with LV assist devices.

DMD is characterized by progressive muscular weakness that typically begins in early childhood. Many affected individuals die of respiratory failure (75%), but most of the remainder die of heart failure (20%). However, recent advances in respiratory management in this setting have made respiratory issues less problematic, making cardiac issues more important. Thus, dilated cardiomyopathy is becoming a major cause of morbidity and mortality in patients with muscular dystrophy.

Our study findings indicate that afterload modification with drugs (ie, ACE inhibitors alone or in combination with β-blockers) can significantly improve LV size and systolic function in patients with cardiomyopathy. The incidence of dilated cardiomyopathy in our study was lower than previously reported by others, a finding that we believe is likely due to earlier referral for cardiac evaluation in our study population. Our findings also suggest that early cardiac evaluation and aggressive noninvasive imaging of young patients with muscular dystrophy can identify those patients most likely to benefit from heart failure therapy. In the 29 patients for whom complete echocardiographic follow-up data were available, therapy for LV dysfunction was begun after a first abnormal echocardiogram. All 29 of these patients tolerated therapy without adverse events and in most cases showed partial or complete LV remodeling. Indeed, 19 (66%) showed normalization, 8 (26%) showed improvement, and 2 (8%) showed stable cardiac disease with no deterioration in LV size or function. The echocardiographic finding of a decrease in LV end-diastolic diameter was the initial change suggesting LV remodeling (from 5.2 to 4.8 cm; \( P = 0.001 \)). All 29 of these patients also demonstrated improvement in LV systolic function as shown by improvement in mean ejection fraction from 36% to 53% \(( P < 0.001 )\). The LV sphericity index, a 2-dimensional index of LV geometry, has been used to document ventricular remodeling in the surgical, electrophysiological, and medical treatment of dilated cardiomyopathy. The LV sphericity index in our cohort improved from 0.73 to 0.59 \(( P < 0.001 )\). Along with the improvement in LV ejection fraction, this is compelling evidence that drug therapy with ACE inhibitors or β-blockers or both can result in significant remodeling of the diseased LV. In addition, all patients showed improvement in LV MPI, with a dramatic benefit in 1 case. Because the MPI is a well-documented global measure of combined systolic and diastolic function, this suggests that drug therapy likely affects the diastolic function of these patients as well. Therefore, early echocardiographic identification of cardiac dysfunction, combined with institution of aggressive medical management, appeared to result in either the normalization or stabilization of dilated cardiomyopathy. That only 2 of our study subjects died during the study (of noncardiac causes in both cases) supports this concept.

All of the subjects with BMD who underwent drug therapy (a combination of digoxin and ACE inhibitors) ultimately achieved normalization of LV dimension and systolic function compared with the DMD group, which did not have as dramatic improvement. Our results also delineate the age at onset of cardiac disease in both populations. Knowing this should allow for more timely evaluation and aggressive screening and earlier institution of therapy. In addition, as reliable echocardiographic assessment of LV diastole becomes more consistent and widespread, this knowledge may facilitate beneficial therapeutic intervention at even earlier stages of isolated diastolic dysfunction when LV systolic performance may still be normal.

In general, data regarding the use of diuretics, ACE inhibitors, and β-blockers in patients with muscular dystrophy are limited. Our study findings indicate that, in the outpatient setting, ACE inhibitor therapy was safely initiated either alone or in combination with β-blocker therapy. Although the use of carvedilol and metoprolol in our study was not randomized, both drugs appeared to have the desired effects on cardiac function and were tolerated equally well. The choice of which β-blocker to use depended primarily on physician preference and cost to the patient.

Our findings regarding dystrophin gene mutations are noteworthy. Although we did not assay for all dystrophin-
associated protein deficiencies that are reportedly abnormal in individuals with muscular dystrophy,34 we did assay for specific mutations in the dystrophin gene. These assays revealed an association between mutations involving exons 12 and 14 to 17 and onset of dilated cardiomyopathy. In addition, they revealed a trend toward a significant association between mutations involving exons 31 to 42 and heart disease. Interestingly, mutations involving exons 51 or 52 appeared to decrease the risk of cardiac involvement. However, our relatively short follow-up did not allow us to establish this with certainty. Nevertheless, the identification of specific exon mutations and their association with dilated cardiomyopathy as reported here have not been previously noted. The impact of these findings on clinical management remains to be determined.

There are several limitations to our study. First, its design was not prospective, randomized, controlled, or blinded. Second, as mentioned, our study sample size was relatively small. However, for a single-institution study of muscular dystrophy patients, ours is a sizable cohort with good follow-up rates. Third, our study cohort included no females. Although we did identify 1 female with cardiac involvement, she was lost to follow-up. Fourth, the study period follow-up was relatively short. It is possible that the changes seen with therapy may not be maintained over time. However, our findings document that the progression of cardiomyopathy seen in muscular dystrophy can be favorably altered. Because this is a quickly progressive cardiac disease with fatal outcomes, we believe that there may be a significant impact with drug therapy on the natural history of this disease. Extended follow-up will be needed to determine the changes in cardiac size and function as well as life expectancy. Fifth, our genetic analyses were subject to accepted limitations, including sample mix-ups and genotyping errors. Finally, exon-specific mutations were determined by utilizing the Fisher test. We recognize the multiple comparison problem, but because of the small sample size and retrospective design, we did not correct for this. Any multiple comparison procedure in which a conservative α level is used would make all of the tests nonsignificant. Therefore, the results should be viewed as preliminary indications that these exons may be involved. We believe that other regions may also be associated with cardiomyopathy, as well as with the response to medical therapy. However, our small sample size allowed only limited statistical power and did not allow us to establish this notion with certainty. Future studies involving larger cohorts will be needed to further delineate this relationship.

Conclusion

The present findings suggest that early diagnosis and treatment of dilated cardiomyopathy in boys with DMD and BMD can lead to ventricular remodeling. These results also indicate that specific mutations of the dystrophin gene may lead to cardiomyopathy or exert cardioprotective effects in this population. Further studies in both directions are warranted.

References


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