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Myocardial fibrosis imaging based on T1-mapping and extracellular volume fraction (ECV) measurement in muscular dystrophy patients: diagnostic value compared with conventional late gadolinium enhancement (LGE) imaging

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Aim Cardiac involvement with progressive myocardial fibrosis leading to dilated cardiomyopathy is a major cause of death in muscular dystrophy patients. Extracellular volume fraction (ECV) measurement based on T1-mapping pre- and post-contrast promises the detection of early 'diffuse' myocardial fibrosis that cannot be depicted by conventional contrast-imaging based on late gadolinium enhancement (LGE). With this study, we evaluated the presence of diffuse myocardial fibrosis in regions of 'normal' (LGE-negative) and 'diseased' (LGE-positive) appearing myocardium as well as its relation to the extent of left ventricular (LV) dysfunction and the occurrence of arrhythmias in Becker muscular dystrophy (BMD) patients.

Methods and results Twenty-seven BMD patients (35 ± 12 years) and 17 matched male healthy controls (33 ± 8 years) underwent cardiovascular magnetic resonance (CMR) studies including ECV measurement and LGE-imaging. Ambulatory monitoring of arrhythmic events was performed by means of an external event loop recorder. Twenty BMD patients (74%) demonstrated cardiac involvement as detected by typical inferolateral presence of LGE. Twelve patients (44%) had an impaired LV ejection fraction—all being LGE-positive. Global myocardial ECV was significantly higher in the BMD group (29 ± 6%) compared with the control group (24 ± 2%, $P = 0.001$). Patients with cardiac involvement demonstrated higher global ECV (31 ± 6%) as well as significantly increased regional ECV not only in LGE-positive segments (34 ± 6%), but also in LGE-negative segments (28 ± 6%) compared with BMD patients without cardiac involvement and to controls, respectively (24 ± 3 and 24 ± 2%, $P = 0.005$). Global ECV in patients with cardiac involvement substantially correlated to LV ejection fraction ($r = -0.629$, $P = 0.003$) and to the number of LGE-positive segments ($r = 0.783$, $P < 0.001$). On univariable analysis, global ECV–but not the categorical presence of LGE *per se*––was significantly associated with arrhythmic events (OR: 1.97, CI: 32.22–1.21, $P = 0.032$).

Conclusion ECV measurement by CMR is a useful tool in assessing the total extent of myocardial fibrosis as well as in depicting subtle diffuse fibrosis in areas of normal appearing myocardium on LGE-images. Thus, myocardial ECV is a potential additional quantitative tool for accurate detection of cardiac involvement and risk stratification in muscular dystrophy patients.

Keywords Muscular dystrophy • Cardiomyopathy • CMR • Mapping • LGE

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Introduction

Becker muscular dystrophy (BMD) is a genetic, X-linked recessive, dystrophin deficiency disease with similar, but more benign clinical course compared with Duchenne muscular dystrophy (DMD). BMD and DMD belong to the broader, heterogeneous group of muscular dystrophies, inherited neuromuscular disorders characterized by progressive skeletal muscle wasting and weakness.[1] Cardiac involvement with myocardial fibrosis leading to dilated cardiomyopathy (DCM), progressive heart failure, and arrhythmias has been described in up to 70% of BMD patients and represents an important cause of morbidity and mortality in this population.[1–5]

In the last years, cardiovascular magnetic resonance (CMR) has been increasingly used for diagnosis as well as for follow-up of cardiac involvement in BMD/DMD patients.[2,6–8] CMR offers an accurate and reproducible tool for left ventricular (LV) systolic function assessment together with the possibility of myocardial tissue characterization and fibrosis detection based on late gadolinium enhancement (LGE) imaging. Thus, early detection of cardiomyopathy and timely initiation of heart failure therapy is facilitated with proven benefits on cardiac dysfunction progression and potential reverse remodelling.[7–11]

Contrast-imaging based on LGE-CMR after i.v. administration of a gadolinium-based compound is currently considered the gold standard technique for imaging of 'focally accentuated' myocardial fibrosis in ischaemic as well as non-ischaemic cardiomyopathies.[12,13] However, the presence of 'diffuse interstitial' myocardial fibrosis (a feature commonly encountered in cardiac pathology with important contribution to adverse LV remodelling) cannot be evaluated by LGE-CMR alone. Recently, equilibrium contrast CMR techniques for the quantification of myocardial extracellular volume fraction (ECV) as estimate for the extent of the myocardial interstitial space have been developed and validated against histological studies.[14,15] In brief, ECV measurements rely on assessing the volume of distribution of gadolinium by performing T1-mapping before and after contrast administration.[16–20]

With this study, we evaluated the presence of 'diffuse' myocardial fibrosis (based on ECV measurement) in regions of 'normal' (LGE-negative) and 'diseased' (LGE-positive) appearing myocardium as well as its relation to the extent of LV dysfunction and the occurrence of arrhythmias in BMD patients.

Methods

Study population

Twenty-nine patients with known BMD were prospectively enrolled between July 2012 and May 2013 and underwent comprehensive CMR studies. BMD had been previously diagnosed in a specialized Neurology Centre based on clinical data, skeletal muscle pathology with dystrophin analyses and/or genetic testing.[11] From this population, two patients were excluded due to impossibility of giving contrast and thus, the final study group (BMD group) consisted of 27 patients. In addition, 17 age-matched healthy male volunteers were enrolled between July 2012 and March 2013 and represented the control group. All BMD patients and controls underwent venous blood sampling for haematocrit measurements on the same day as the CMR scan. The local ethics committee approved the study protocol and all the participants gave written informed consent for participation in the study.

CMR data acquisition

ECG-gated CMR studies were performed on a 1.5-T Aera (Siemens Medical Solutions, Erlangen, Germany) using commercially available cardiac software, electrocardiographic triggering, and cardiac-dedicated surface coils. Cine-imaging was performed using a steady-state-free precession (SSFP) sequence in three long-axis slices (four-, three-, and two-chamber) and a stack of short-axis slices completely encompassing the LV. LGE-imaging was performed using a T1-weighted inversion recovery sequence 10–15 min after i.v. contrast administration (0.15 mmol/kg Magnevist®) in the same imaging planes.

For ECV-imaging, T1 measurements were made using an modified look-locker inversion recovery (MOLLI) sequence in two short-axis (basal and mid-ventricular) and in at least two long-axis slices (usually four- and three-chamber) before and 15–20 min after i.v. contrast administration (following LGE-imaging). Image planning was carefully done in order to exclude LV outflow tract in the basal short-axis slice. Typical imaging parameters were non-selective inversion pulse, SSFP single-shot read-out in late diastole, matrix of 256 × 144, slice thickness of 8 mm, time to repetition/time to echo of 325/1.1 ms, minimum inversion time of 110 ms, inversion time increment of 80 ms, flip angle of 35°, parallel acquisition technique factor of two, number of inversions two, images acquired after first inversion three, pause three heart beats, and images acquired after second inversion five (in total of eight images acquired during 11 heart beats).

CMR data analysis

CMR analysis was performed off-line by two experienced readers. Ventricular volumes, ejection fraction, and LV mass were derived by contouring endo- and epi-cardial borders on the short-axis cine images. LGE presence and pattern were visually assessed on the short- and long-axis images using the same AHA 17-segment model.[21] First, BMD patients were dichotomously categorized as having cardiac involvement when non-ischaemic LGE was present in at least one myocardial segment. Second, in BMD patients with cardiac involvement, the six short-axis segments in the LGE-image acquired in the same slice position as the analysed ΔR1 map were classified as being positive or negative according to LGE presence/absence. Negative LGE-segments in BMD patients with cardiac involvement are also referred as 'normal appearing' myocardium. LGE readings were blinded to ECV data.

For myocardial ECV calculations, motion-corrected pre- and post-contrast T1 pixel maps (automatically generated after each MOLLI acquisition) were used for processing with the Image J (http://rsbweb.nih.gov/ij/) software as previously described.[20,22] R1 maps were generated by taking the reciprocal of each T1 map on a pixel-by-pixel basis and ΔR1 maps were computed by subtracting the pre-contrast R1 map from the post-contrast R1 map. For each patient/healthy volunteer, one short-axis slice (preferably basal) served for analysis. Average pixel signal intensity (ΔR1 value) was measured in the chosen short-axis ΔR1 map using manually delineated regions of interest for each one of the six myocardial segments corresponding to the AHA 17-segment model. Additionally, a region of interest was placed in the blood pool, carefully avoiding papillary muscles and LV trabeculations. Segmental myocardial ECV was then computed according to the formula: $ECV = \lambda \times (1 - \text{haematocrit})$, where the partition coefficient $\lambda = \Delta R1\ (\text{myocardium})/\Delta R1\ (\text{blood})$.[15] To additionally report global and regional (interventricular septum vs. lateral wall) myocardial ECV, segmental ECV values were averaged in each individual accordingly.

Monitoring of arrhythmic events

In the same month as the CMR scan, all BMD patients underwent ambulatory monitoring of potential arrhythmias during a 5-day period by

means of an external event loop recorder (SpiderFlash-t, Sorin Group). This device records electrocardiographic tracings in two different leads during and up to 15 min after arrhythmia detection (auto-triggered) and/or patient activation. Subsequently, all ECG recordings were assessed for the presence of ventricular arrhythmias (triplets, non-sustained ventricular tachycardia), supraventricular arrhythmias (supraventricular tachycardia, atrial fibrillation), and conduction disturbances.

Statistical analysis

Continuous variables are expressed as means ± SD. Skewed variables are expressed as median and inter-quartile range (IQR). Categorical variables are expressed as frequency with percentage. Student's *t*-test was used for comparison of normally distributed characteristics between BMD and controls. Levene's test was used for testing equality of variances. One-way ANOVA with Bonferroni *post hoc* correction was used for the subgroup multiple comparison. Dunnett's *post hoc* was used in the case of inequality of variances. Pearson correlation (*r*) was used to assess the relationship between different normally distributed CMR measurements. Spearman's non-parametric correlation was used for not normally distributed variables. The Chi-square test with Yate's correction was used to compare non-continuous variables expressed as proportions. To find independent predictors for arrhythmic events, univariable analysis was performed first. Statistical analysis was performed using the SPSS software for Windows (version 18, SPSS, Chicago IL, USA). A *P*-value ≤0.05 was considered statistically significant.

Results

Patient characteristics

Table 1 summarizes the characteristics of the two study groups, BMD patients ($n = 27$), and controls ($n = 17$). The mean age was 35 ± 12 years (IQR 26–45 years) in the BMD group, whereas in the controls mean age was 33 ± 8 years (IQR 27–41 years), and all the patients in both groups were males. In the BMD group, median disease duration at inclusion was 14 years (IQR: 9–23 years). None of the BMD patients had a history of coronary artery disease, arterial hypertension, or diabetes. All control group volunteers had no prior known cardiovascular disease or any current medication.

CMR findings: BMD patients vs. controls

The volumetric CMR findings for the two groups are presented in *Table 1*. BMD patients showed significantly higher LV end-systolic volume indexes ($P < 0.001$) and significantly lower left ($P < 0.001$) and right ventricular ejection fraction ($P = 0.032$) compared with controls. Regarding the presence of LGE, 20 (74%) BMD patients were LGE-positive with a median number of 2 (IQR 1–4) segments. All of these 20 BMD patients demonstrated a non-ischaemic LGE-pattern and consequently formed the BMD subgroup with cardiac involvement. The remaining seven (26%) patients were assigned to the NO cardiac involvement subgroup. LGE distribution patterns were as follows: 11 (55%) showed subepicardial LGE, five (25%) showed mid-wall LGE and four (20%) showed a combination of the two. The most frequently involved LV segments were basal inferolateral (in 90% of cases) and basal anterolateral (in 60% of cases). The least affected basal segments were anterior (25%) and inferoseptal (15%). In all but one of the eight patients with septal LGE, extensive lateral wall LGE co-existed. None of the controls demonstrated ischaemic or non-ischaemic LGE.

Table 1 Patient characteristics and volumetric findings

	Controls ($n = 17$)	BMD ($n = 27$)	*P*-value
Age, years	33 ± 8	35 ± 12	0.522
Male, *n* (%)	17 (100)	27 (100)	NA
Age at diagnosis, years	–	17 ± 14	NA
Heart rate, b.p.m.	60 ± 9	63 ± 11	0.392
Haematocrit, %	44 ± 2	45 ± 3	0.667
ACE-inhibitor, *n* (%)	0 (0)	16 (59)	<0.001
β-Blocker, *n* (%)	0 (0)	10 (37)	0.004
LV end-diastolic volume index, mL/m^2	91 ± 11	93 ± 25	0.679
LV end-systolic volume index, mL/m^2	32 ± 5	48 ± 21	<0.001
LV ejection fraction, %	65 ± 4	50 ± 11	<0.001
LV mass index, g/m^2	65 ± 11	60 ± 13	0.180
RV end-diastolic volume index, mL/m^2	86 ± 13	75 ± 16	0.022
RV end-systolic volume index, mL/m2	36 ± 9	35 ± 9	0.677
RV ejection fraction, %	58 ± 8	54 ± 6	0.032

BMD, Becker muscular dystrophy; LV, left ventricle; RV, right ventricle; ACE, angiotensin-converting enzyme; LGE, late gadolinium enhancement; NA, non-applicable.
**Post hoc P* < 0.05 vs. BMD patients.

CMR findings: BMD patients with vs. without cardiac involvement

BMD patients with cardiac involvement had higher LV end-diastolic and end-systolic volumes and lower LV and RV ejection fraction compared with BMD patients without cardiac involvement and to controls (*Table 2*). Twelve (60%) BMD patients with cardiac involvement had an impaired LV ejection fraction. In none of the other two subgroups, LV ejection fraction impairment was encountered. Notably, there were no significant differences in disease duration between patients with and without cardiomyopathy (median, IQR: 11, 9–21 years vs. 15, 8–23 years, $P = 0.470$).

ECV findings

Global myocardial ECV was 29 ± 6% (IQR: 24–32%) in the BMD group and 24 ± 2% (IQR 22–26%) in the control group ($P = 0.001$). In the subgroup analysis, BMD patients with cardiac involvement had significantly higher global ECV compared with BMD patients without cardiac involvement and to CONTROLS ($P < 0.001$) (*Table 3*). No significant differences in global ECV were found between BMD patients without cardiac involvement and controls. The mean segmental ECV values in BMD patient subgroups are illustrated in *Figure 1*. Considering segmental ECV according to the presence of LGE revealed substantially increased ECV values not only in LGE-positive segments (34 ± 6%), but also in LGE-negative segments (28 ± 6%) of BMD with cardiac involvement compared with both BMD

Table 2 Volumetric findings in patients with and without cardiac involvement

	Controls ($n = 17$)	BMD NO cardiac involvement ($n = 7$)	BMD with cardiac involvement ($n = 20$)	*P*-value
LV end-diastolic volume index, mL/m^2	91 ± 11§	74 ± 14*	100 ± 24	0.010
LV end-systolic volume index, mL/m^2	32 ± 5*	30 ± 7*	55 ± 21	<0.001
LV ejection fraction, %	65 ± 4*§	59 ± 3*	46 ± 10	<0.001
LV ejection fraction < 50%, *n* (%)	0 (0)	0 (0)	12 (60)	<0.001
LV mass index, g/m^2	65 ± 11	59 ± 8	60 ± 15	0.435
RV end-diastolic volume index, mL/m^2	86 ± 11	71 ± 14	76 ± 17	0.054
RV end-systolic volume index, mL/m^2	36 ± 9	32 ± 9	36 ± 9	0.555
RV ejection fraction, %	58 ± 8*	57 ± 3	52 ± 7	0.032

LV, left ventricle; RV, right ventricle; ACE, angiotensin-converting enzyme; LGE, late gadolinium enhancement; NA, non-applicable.
**Post hoc* $P < 0.05$ vs. LGE + patients.
§*Post hoc* $P < 0.05$ vs. LGE − patients.

Table 3 Myocardial ECV results

	Controls ($n = 17$)	BMD NO cardiac involvement ($n = 7$)	BMD with cardiac involvement ($n = 20$)	*P*-value
ECV global, %	24 ± 2*	24 ± 3*	31 ± 6	<0.001
ECV interventricular septum, % (no. of analysed segments)	24 ± 2 (34)	24 ± 3 (14)	27 ± 7 (40)	0.088
ECV LV lateral wall, % (no. of analysed segments)	24 ± 3* (34)	24 ± 3* (14)	34 ± 8 (40)	<0.001
ECV in LGE-pos. segments, % (no. of analysed segments)	−(0)	−(0)	34 ± 6 (51)	NA
ECV in LGE-neg. segments, % (no. of analysed segments)	24 ± 2* (102)	24 ± 3* (42)	28 ± 6 (69)	0.005

ECV, extracellular volume fraction; LV, left ventricle; LGE, late gadolinium enhancement; NA, non-applicable.
**Post hoc* $P < 0.05$ vs. LGE + patients.

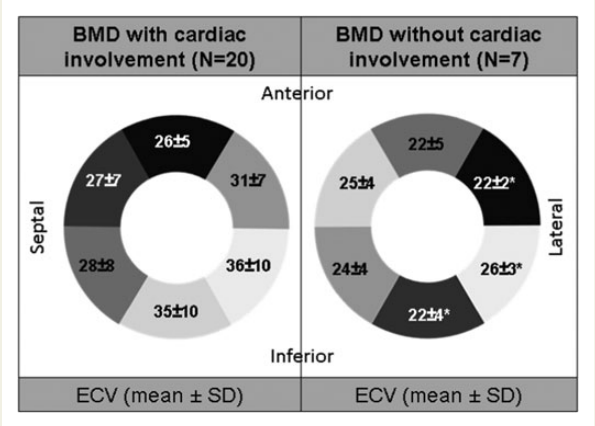

Figure 1: Schematic representation of six-segment bulls eyes for the analysed basal short-axis slice showing: (*A*) ECV segmental values (means ± SD) in the 20 Becker muscular dystrophy patients with cardiac involvement; (*B*) ECV segmental values in the seven patients without cardiac involvement. Note that even in segments with a low prevalence for late gadolinium enhancement (infero-septum) average ECV was increased compared with patients without cardiac involvement.

without cardiac involvement and controls (24 ± 3 and 24 ± 2%, $P = 0.005$) (*Figure 2*).

Relation between ECV and the extent of cardiac involvement

The relation between ECV and established parameters reflecting the severity of cardiomyopathy (LV ejection fraction and extent of LGE) were assessed in the 20 BMD patients with cardiac involvement. A significant inverse correlation between global ECV and LV ejection fraction ($r = -0.629$, $P = 0.003$) (*Figure 3A*) and as a significant positive correlation between global ECV and the number of LGE-positive segments (Spearman's $r = 0.783$, $P < 0.001$) were found. In addition, LV ejection fraction significantly correlated with regional ECV in LGE-positive segments ($r = -0.515$, $P = 0.02$), but not with regional ECV in normal appearing (LGE-negative) myocardium ($r = -0.364$, $P = 0.115$) (*Figure 3B–C*). LV ejection fraction related well to the number of LGE-positive segments reflecting the extent of cardiac involvement in BMD patients (Spearman's $r = -0.473$, $P = 0.035$). Moreover, a significant positive correlation was found between ECV in LGE-positive segments and ECV in normal appearing (LGE-negative) myocardium ($r = 0.480$, $P = 0.032$) (*Figure 3D*).

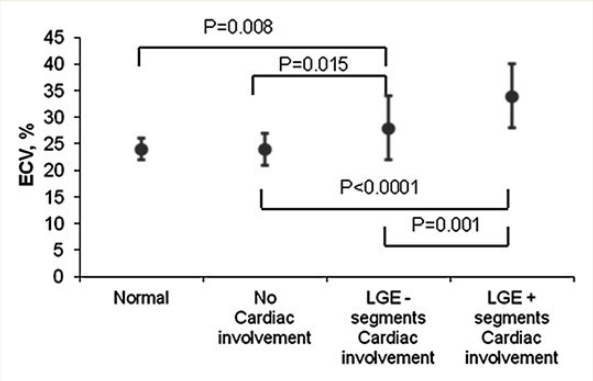

Figure 2: In Becker muscular dystrophy patients with cardiac involvement (LGE present) ECV in normal appearing myocardium (LGE-negative segments) was significantly higher compared with patients without cardiac involvement and to normal volunteers.

Relation between CMR parameters and the location and type of dystrophin gene mutation

A genetic diagnosis regarding the type and location of dystrophin gene mutation was available in all 27 BMD patients. *Table 4* illustrates the conventional and the ECV-CMR findings according to dystrophin gene mutation category. Owing to the small number of patients in the respective subgroups, a proper and meaningful statistical analysis was not possible.

Association between myocardial abnormalities and occurrence of arrhythmic events

Arrhythmic events as defined in the methods section were recorded in eight (30%) BMD patients during monitoring for 5 days with an external event recorder. Non-sustained ventricular tachycardia episodes were detected in four (15%) patients, ventricular triplets in five (20%) patients and runs of supraventricular tachycardia in two (7%) patients. In addition, an isolated episode of atrial fibrillation was seen in one patient. All the above-described events were exclusively encountered in BMD patients with cardiac involvement. On univariable analysis LV ejection fraction (OR: 0.79, CI: 0.64–0.97, $P = 0.022$), global ECV (OR: 1.97, CI: 32.22–1.21, $P = 0.032$) and the number of LGE-positive segments (OR: 5.34, CI: 1.19–23.91, $P = 0.029$)—but not the categorical presence of LGE *per se*—were significantly related to arrhythmia occurrence. Owing to the small study group and the limited number of arrhythmic events, a meaningful multivariable analysis could not be performed.

Discussion

To the best of our knowledge, this is the first study conducted in muscular dystrophy patients showing that ECV measurement based on non-invasive CMR imaging does not only allow a 'quantitative' assessment of myocardial fibrosis, but can also depict subtle diffuse abnormalities in myocardial areas that appear normal on conventional LGE-images. In addition, ECV measurements correlated well with both the severity of cardiac involvement and the occurrence of arrhythmic events, suggesting a possible future role in cardiovascular risk prediction—not only in muscular dystrophy patients.

In accordance with recently published data, the great majority (74%) of BMD patients in the present study demonstrated cardiac involvement detected by the presence of non-ischaemic LGE with a predominant distribution in the subepicardium of the LV lateral wall. Occurrence of LGE is believed to precede LV systolic dysfunction in patients with cardiac involvement, since none of the LGE-negative patients showed a reduced LV ejection fraction, while 60% of the LGE-positive patients presented with an impaired LV ejection fraction. Hence, LGE-CMR is a more sensitive tool compared with cine-CMR to detect cardiac involvement in BMD.[6,7] However, ECV measurement is even more sensitive than LGE-CMR considering the aforementioned unique depiction of subtle diffuse myocardial fibrosis in LGE-negative segments (*Figure 4*).

Major ECV findings

Significantly, higher global ECV values were found in BMD patients with cardiac involvement compared with patients without cardiac involvement or matched healthy controls. Thus, global ECV measurement can serve as a unique 'quantitative' tool for assessing (as well as monitoring) early myocardial fibrosis—not only in BMD patients.

The most striking finding of this study was the measurement of significantly increased ECV values (suggestive of subtle diffuse myocardial fibrosis) even in myocardial areas that appear normal on LGE-images. The detailed molecular pathomechanism leading to cardiac disease in muscular dystrophy patients is still not known. The underlying genetic dystrophin defect theoretically causes a diffuse disease process with ubiquitous or randomly distributed alterations in cell metabolism and signal transduction in the myocardium, which pre-dispose to morphological changes comprising early cardiomyocyte cell death and replacement fibrosis. These morphological changes precede functional impairment and comprise the entire myocardium. Myocardial damage depicted by LGE-imaging and preferentially located in the inferolateral wall[6,11,23,24] is possibly due to exaggerated mechanical stress in this region and reflects a more severe and advanced damage compared with the subtle and more diffuse replacement fibrosis[7,11,25] that may only be depicted by ECV measurement and is ongoing in LGE-negative segments.

Moreover, the good correlation between ECV in segments with and without LGE further supports the existence of a progressively spread disease process with coexistence of severely damaged focal areas (predominantly in the inferolateral wall) and more subtle and diffuse abnormalities in the remaining myocardium. As a parallel, in the histologically validated study by Miller *et al.*[15] performed in end-stage heart failure patients requiring cardiac transplantation, significantly increased ECV values were found both in LGE-positive and -negative areas. In another comparable population of patients with familial DCM and mild disease expression (average LV ejection fraction 53%), Sado *et al.*[19] report ECV values (among others in LGE-negative areas) that were similar to ours.[19] The respective ECV values were significantly higher compared with the ones in their control population (28 ± 4 vs. $25 \pm 4\%$) even though only 35% of the DCM patients had detectable LGE.

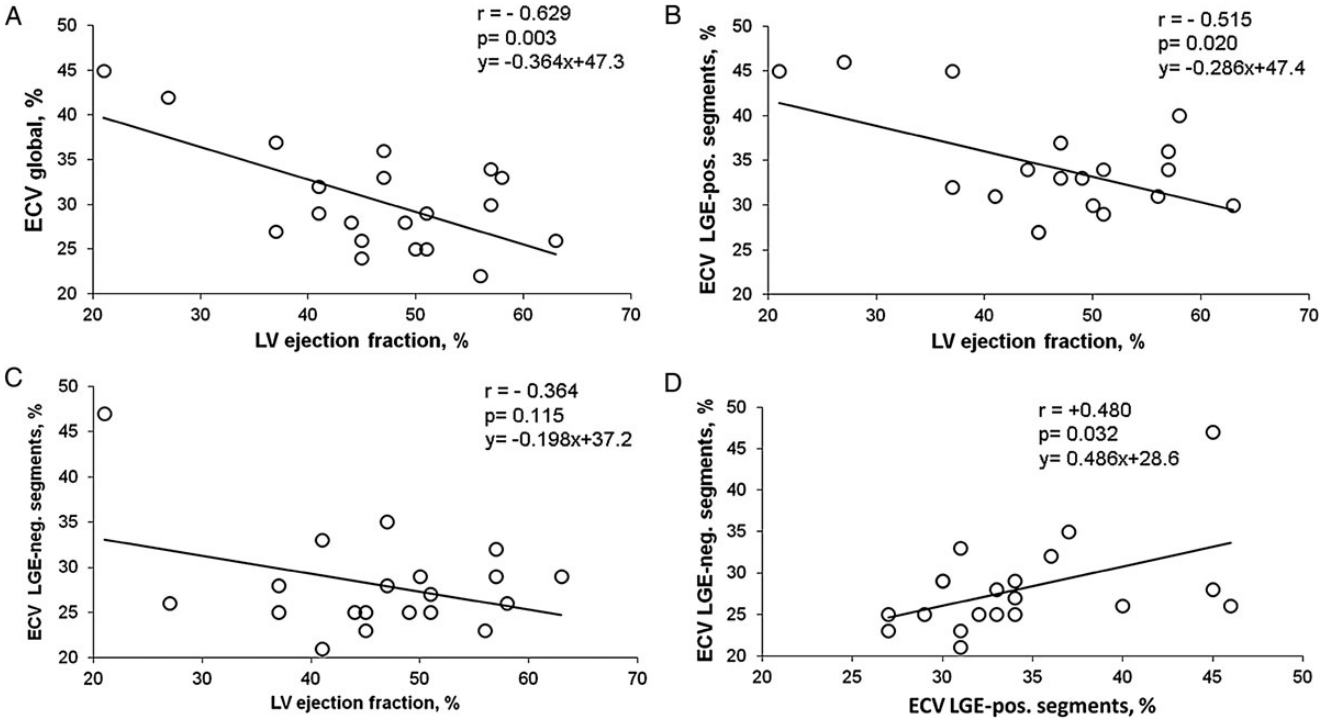

Figure 3: Scatter plots showing the correlation between different ECV measurements and the extent of cardiac involvement. (*A*) Global ECV inversely related to ejection fraction in patients with cardiac involvement. (*B*) ECV in LGE-positive myocardial segments inversely related to ejection fraction in patients with cardiac involvement. (*C*) ECV in LGE-negative myocardial segments was not related to ejection fraction in patients with cardiac involvement and (*D*) a good correlation between ECV in LGE-negative and LGE-positive segments was seen.

Table 4 CMR results according to dystrophin gene mutation type

Dystrophin gene mutation type	*n*	Age	LV-EDV (mL/m^2)	LV-EF (%)	LGE-pos. (%)	ECV global (%)	ECV in LGE-pos. (%)	ECV in LGE-neg. (%)
Central rod domain deletion	16	34 (16–53)	84 (63–110)	57 (44–63)	10 (63)	25 (21–34)	31 (27–40)	24 (21–32)
N-terminal domain deletion	3	15 (11–39)	106 (50–114)	57 (37–63)	2 (67)	30 (27–30)	33 (32–34)	29 (25–30)
Duplication	3	37 (28–43)	98 (83–113)	37 (27–41)	3 (100)	37 (29–42)	45 (31–46)	26 (21–28)
Point mutation	5	45 (23–56)	133 (67–155)	47 (21–49)	5 (100)	33 (28–45)	33 (31–35)	33 (25–47)

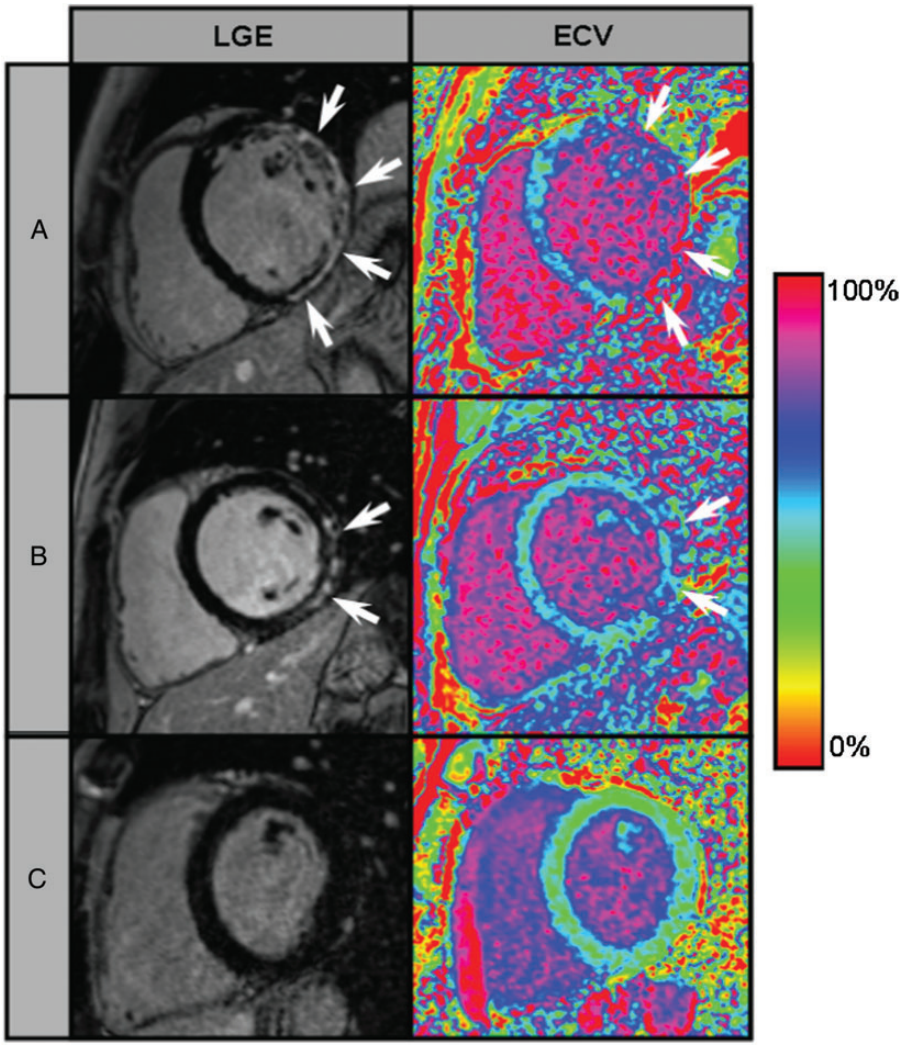

Figure 4: Late gadolinium enhancement (left) and quantitative ECV images (right) in three Becker muscular dystrophy patients: the first (*A*) showed a typical picture of dilated cardiomyopathy with extensive subepicardial to transmural enhancement in the lateral wall (arrows); the second (*B*) had preserved left ventricular dimensions and systolic function, showing limited subepicardial enhancement in the lateral wall (arrows) and the third (*C*) had no detectable cardiac involvement. Notably, in the first two cases higher ECV values are noticeable also in the 'normal appearing' non-enhanced myocardial areas when compared with the third patient. The colour scale ranges from 0 to 100% extracellular volume.

Relation between ECV and the extent of cardiac involvement

In the present study, we also evaluated the relationship between the presence and extent of structural abnormalities in the myocardium and the severity of LV dysfunction and the risk for arrhythmic events in BMD, respectively. We showed that the total extent of myocardial fibrosis measured either by 'global' ECV or by the number of LGE-positive segments was inversely related to LV ejection fraction. Obviously, increased myocardial fibrosis leads to impaired LV systolic function. Interestingly, an inverse correlation between 'regional' ECV and LV ejection fraction was only detected in LGE-positive segments—but not in LGE-negative ones. Therefore, one can argue that (segmental) ECV measurement allows the detection of subtle 'interstitial fibrosis' (not detected by LGE-imaging) 'prior' to having an effect on LV systolic function. Consequently, ECV measurement may possibly enable not only an earlier diagnosis of cardiac involvement, but also an earlier institution of targeted therapies in some patients.

Moreover, it is well known that in BMD/DMD patients the risk for arrhythmia is related to the presence of DCM and LV systolic dysfunction.[5,26] In our study population, the occurrence of arrhythmic events inversely correlated with LV ejection fraction, global ECV, and the number of LGE-positive segments, respectively, but not with the categorical presence of LGE *per se* which was suggested as an independent risk predictor in other cardiomyopathies in the past.[27,28] Hence, considering the well-known difficulties in 'quantifying' the extent of LGE, the simple and quick measurement of global ECV could serve as a welcome quantitative tool for arrhythmic risk prediction—among others for patients with milder degrees of LV systolic dysfunction.

A question that naturally arises is why (mean) ECV measurement was 'normal' in the seven BMD patients without overt cardiac abnormalities (no LGE and normal LV systolic function) in both septal as well LV lateral wall segments (*Table 3*). Considering the aforementioned explanations, one would rather expect increased ECV values at least in the LV lateral wall segments, since the future occurrence of LGE is expected first in this area. And indeed, there was one BMD patient in the group of seven patients without LGE and normal LV function who had a (regional) ECV value of 29% in the LV lateral wall. Follow-up studies are currently performed (*Figure 5*) and will show whether LGE occurs first in those areas demonstrating the highest ECV values (in the absence of LGE) in preceding studies.

Relation between ECV and the location as well as type of dystrophin mutation

Based on previous data showing that the location of dystrophin gene mutation is associated with the presence/onset of skeletal as well as cardiac disease, we categorized our patients in four subgroups: (i) deletions in the central rod domain of the dystrophin gene (exons 45–49)—suggested to be associated with a later onset of cardiomyopathy, (ii) deletions in the N-terminal domain (exons 2–9)—suggested to be associated with an early onset of cardiomyopathy, (iii) duplications, and (iv) point-mutations.[29,30] Owing to the small number of patients in the respective subgroups, a proper and meaningful statistical analysis was unfortunately not possible. Nevertheless, it appears that in our study group, BMD patients

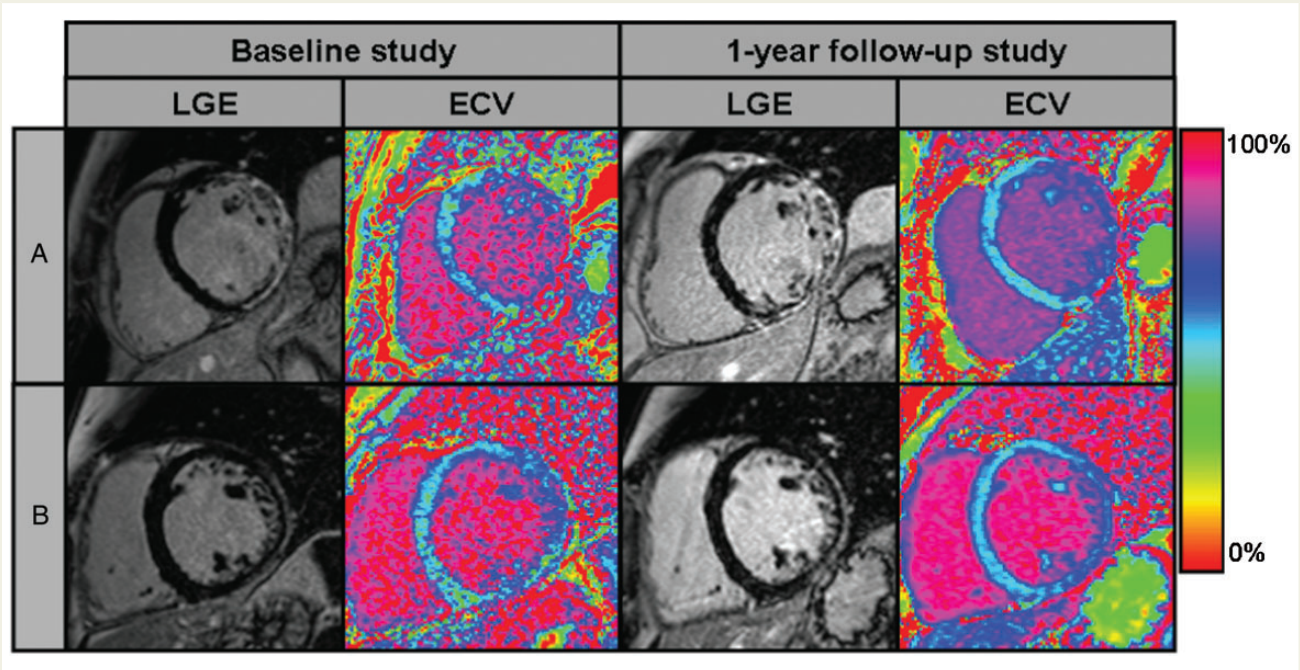

Figure 5: LGE and quantitative ECV images in two Becker muscular dystrophy patients with cardiac involvement in whom 1-year follow-up examinations were performed. In both patients, there was no relevant progression in the extent of LGE. In addition, no change in global ECV as well as ECV in LGE-positive segments (lateral wall) and LGE-negative segments was observed.

with mutations other than deletions had a more severe cardiac involvement considering the parameters LV-EF and global ECV (*Table 4*). However, these data need to be confirmed in a larger study group.

Study limitations

Obviously, the group of patients without any detectable cardiac involvement ($n = 7$) was rather small. However, BMD is a rare orphan disease and unfortunately, cardiac involvement is a frequent finding in particular in adults with BMD—as shown by the results of this study. Second, endomyocardial biopsy data for proof and correlation of CMR findings to histopathological results were unfortunately not available. However, considering the straightforward diagnosis based on CMR in these patients and the invasive character of biopsy with a non-neglectable risk of complication, ethical aspects prevented us to routinely perform biopsy in this patient population.

Conclusions

ECV measurement by CMR is a useful tool in assessing the total extent of myocardial fibrosis as well as in depicting subtle diffuse fibrosis in areas of normal appearing myocardium on LGE-images. Thus, myocardial ECV is a potential additional quantitative tool for accurate detection of cardiac involvement and risk stratification in muscular dystrophy patients.

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