J Vet Intern Med 2014;28:386–392

Dobutamine Stress Echocardiography for Assessment of Systolic Function in Dogs with Experimentally Induced Mitral Regurgitation

R. Suzuki, H. Matsumoto, T. Teshima, Y. Mochizuki, and H. Koyama

Background: Systolic dysfunction is associated with poor outcomes in dogs with myxomatous mitral valve disease. However, assessment of systolic variables by conventional echocardiographic methods is difficult in these dogs because of mitral regurgitation (MR).

Hypothesis: We hypothesized that assessment of systolic function by dobutamine stress may identify systolic dysfunction in dogs with MR, and that 2-dimensional speckle-tracking echocardiography (2D-STE) could quantitatively evaluate myocardial function.

Animals: Anesthetized dogs with experimentally induced MR.

Methods: Dogs were examined for systolic myocardial deformations using 2D-STE during dobutamine infusion before and 3 and 6 months after MR induction. We evaluated peak systolic rotation and rotation rate in each basal and apical view; peak systolic torsion and torsion rate were also calculated.

Results: Invasive peak positive first derivatives of left ventricular pressure (dp/dt) were significantly decreased in dogs 6 months after induction of MR compared with pre-MR results. After 3 and 6 months of MR, dogs had diminished peak systolic torsion values and torsion rates in response to dobutamine infusion compared with pre-MR results (3 months, $P < .001$ and $P = .006$; 6 months, $P = .003$ and $P = .021$). These results were significantly correlated with overall invasive dp/dt ($r = 0.644$, $P < .001$; $r = 0.696$, $P < .001$).

Conclusions and Clinical Importance: Decreased torsion during dobutamine infusion in dogs with MR may reflect latent systolic dysfunction. Dobutamine infusion, therefore, may be useful for the assessment of systolic function in dogs with MR.

Key words: Mitral valve disease; Myocardium; Speckle-tracking; Torsion.

Myxomatous mitral valve disease (MMVD) is the most common cardiac disease of dogs, and affected dogs suffer from volume overload because of mitral regurgitation (MR).[1] Dogs with moderate heart failure caused by MMVD have decreased systolic function,[2] which is associated with decreased survival time.[3] We recently demonstrated that dogs with MMVD may have latent myocardial dysfunction, as assessed by 2-dimensional speckle-tracking echocardiography (2D-STE), corresponding to progression of disease severity.[4,5] We also hypothesized that these myocardial functional abnormalities correspond to MMVD severity. However, assessment of systolic function, by conventional echocardiographic methods, is difficult owing to altered hemodynamic loading conditions in dogs with MR.[1]

Dobutamine infusion is an alternative to exercise for inducing cardiovascular stress in patients with cardiac disease.[6,7] Because high-quality echocardiographic images are more easily obtained during pharmacologically induced stress than those generated during exercise-induced stress, dobutamine stress tests can be performed repeatedly and safely in conscious dogs to assess myocardial function.[8,9] Furthermore, previous studies in dogs have indicated that dobutamine stress tests are sensitive and reliable methods for the early detection of cardiac dysfunction.[10,11] Cardiac function, however, has not been assessed using dobutamine infusion in dogs with MR.

Recently, the 2D-STE method has been found to be as reliable as sonomicrometry for the assessment of left ventricular (LV) function during dobutamine

Abbreviations:

2D-STE	2-dimensional speckle-tracking echocardiography
dp/dt	peak positive first derivatives of the left ventricular pressure
EDVI	end-diastolic volume index
EF	ejection fraction
ESVI	end-systolic volume index
FS	fractional shortening
IVSd	end-diastolic interventricular septum thickness
LA/Ao	left atrial-to-aortic root ratio
LVIDd	end-diastolic left ventricular internal dimension
LVIDs	end-systolic left ventricular internal dimension
LV	left ventricular
LVWd	end-diastolic left ventricular free-wall thickness
MMVD	myxomatous mitral valve disease
MR	mitral regurgitation
PEP/ET	pre-ejection period-to-ejection time ratio
RF	regurgitant fraction
RWT	relative wall thickness
TSVI	total stroke volume index

From the Division of Veterinary Internal Medicine, Department of Veterinary Science, Faculty of Veterinary Medicine, Nippon Veterinary and Life Science University, Tokyo, Japan (Suzuki, Matsumoto, Teshima, Mochizuki, Koyama).

Corresponding author: R. Suzuki, DVM, Division of Veterinary Internal Medicine, Department of Veterinary Science, Faculty of Veterinary Medicine, Nippon Veterinary and Life Science University, 1-7-1 Kyonan-cho, Musashino-shi, Tokyo 180-8602, Japan; e-mail: ryoheisuzuki0130@gmail.com.

Submitted May 12, 2013; Revised November 6, 2013; Accepted November 26, 2013.

10.1111/jvim.12293

infusion.[12] Although assessment of systolic function by conventional echocardiography is often subjective, 2D-STE can quantitate myocardial deformations during dobutamine infusion. We hypothesized that an inotropic challenge with dobutamine, as assessed by 2D-STE, could reveal early occult cardiac dysfunction not evident at rest.

To our knowledge, assessment of systolic function by a dobutamine stress test, with 2D-STE, in dogs with experimentally induced chronic MR has not been reported previously. Therefore, the purpose of this study was to quantitatively measure myocardial deformations during a dobutamine stress test in dogs with experimentally induced chronic MR.

Materials and Methods

Animals

Five healthy male Beagles (body weight, 10.0 ± 1.5 kg; age, 15.6 ± 0.5 months) were used in this study. Before the first assessment, all dogs were judged to be healthy, based on the absence of a history of cardiac signs, as well as normal findings on complete physical examination, CBC, serum biochemistry evaluation, ECG, thoracic radiography, and transthoracic echocardiography. All procedures were performed according to the Guide for Care and Use of Laboratory Animals and approved by the ethical committee for laboratory animal use of Nippon Veterinary and Life Science University, Japan.

Protocol

All dogs were premedicated with butorphanol tartrate[a] (0.2 mg/kg IM), induced by thiopental sodium[b] (25 mg/kg IV), maintained with 1–1.5% isoflurane[c] mixed with 100% oxygen, and manually ventilated. The left or right lateral neck region was clipped, aseptically prepared, and draped. An approximately 5-cm surgical cut-down was performed over the jugular furrow to expose the carotid artery. A microtip catheter with a pressure transducer[d] was directly inserted into the carotid artery and fixed within the mid-LV cavity to measure LV pressure under echocardiographic and fluoroscopic guidance. Hemodynamic measurements along with dobutamine stress echocardiography were performed to provide control data.

After the hemodynamic measurements to acquire pre-MR (control) data, each dog was placed in right lateral recumbency. The left lateral thoracic region was clipped, aseptically prepared, and draped. A left lateral thoracotomy was performed in the fifth intercostal space and the pericardium was opened by standard procedures. The left atrium was purse-string sutured and a small incision was made at the center of the purse-string suture. The suture was loosened and Halsted mosquito forceps were inserted through the small incision to grasp and rupture the mitral valvular chordae tendineae. The position of the chordae tendineae and the degree of induced MR were monitored by transthoracic echocardiography, and these procedures were repeated until visible MR (regurgitant jet signals were approximately 30% relative to left atrial area by color Doppler method) was identified. The chest then was closed in layers and air was evacuated by standard procedures. Bupivacaine hydrochloride[e] (2 mg/kg) was injected at the incision area and in the surrounding intercostal muscles during the operation. After the operation, cefalexin[f] was administered (25 mg/kg PO, q12h) for 7 days and postoperative pain was treated with butorphanol tartrate[a] (0.2 mg/kg, IM), as needed, and meloxicam[g] (0.2 mg/kg PO, q24h) on the day of, and for 3 days after, surgery. Echocardiographic and hemodynamic examinations were performed before surgery to collect pre-MR (control) and 3- and 6-month postsurgical data. Each measurement and anesthetic protocol were performed in the same manner. After the hemodynamic measurements were obtained, the catheter was removed and the carotid artery was closed with 7-0 nylon sutures. Standard surgical closure of the incision was performed.

Hemodynamic Measurement

Continuous LV pressures and simultaneous ECG data, along with dobutamine stress echocardiography, were obtained before and 3 and 6 months after inducing MR. Peak systolic LV pressure and peak positive first derivatives of LV pressure (dp/dt) were calculated by appropriate software.[h] All hemodynamic data were calculated as mean values from 10 consecutive high-quality pressure measurements.

Dobutamine Stress Echocardiography

We performed dobutamine stress echocardiography on anesthetized dogs before and 3 and 6 months after surgery. Baseline echocardiographic data were obtained, and dobutamine[i] was infused at a dosage of 5 μg/kg/min for 5 minutes and then 10 μg/kg/min for 10 minutes to allow plasma dobutamine concentration to reach a steady state. During infusion of dobutamine, we performed echocardiography measurements including 2D-STE along with previously described hemodynamic measurements. Instrument settings, including pulse and repetition rate, transducer frequencies, and gain, were kept constant throughout all echocardiographic studies.

Standard Echocardiography and 2D-STE

Conventional 2D, M-mode, and Doppler examinations were performed under anesthesia by a single investigator (RS) by an echocardiographic system (Vivid7 dimension[j]) and transducer (M7S probes[j]), and a simultaneous ECG limb lead II was recorded and displayed on the images. All data were obtained for at least 9 consecutive cardiac cycles in sinus rhythm and at end-expiration. During the procedure, each dog was placed on a table designed to allow echocardiographic examination during cardiac catheterization. The surgical neck region of all dogs was draped and packed with antiseptic dressing, and the dogs were carefully and repeatedly repositioned in right and left lateral recumbency. All hemodynamic and echocardiographic measurements were performed in closed-chest procedures. Images were analyzed by 1 observer using an off-line work station (EchoPac PC, version 108.1.4).[j] The left atrial-to-aortic root ratio (LA/Ao) was obtained from the right parasternal short-axis view by the B-mode method.[13] M-mode measurements were obtained from the right parasternal short-axis view at the level of the chordae tendinae, according to the leading edge-to-leading edge method. These included end-diastolic interventricular septum thickness (IVSd), end-diastolic LV free-wall thickness (LVWd), end-diastolic LV internal dimension (LVIDd), end-systolic LV internal dimension (LVIDs), and fractional shortening (FS). As indicators of LV geometrical remodeling, relative wall thicknesses (RWT) and sphericity indexes were calculated. RWT is the ratio of the sum of IVSd and LVWd to LVIDd,[14] and the sphericity index is the ratio of the LV long axis-to-short axis diameter both in end-systole and in end-diastole.[15] For LV volume measurements, the biplane method of disks (modified Simpson's rule) was used, based on high-quality left-apical 4- and 2-chamber views, according to the American Society of Echocardiography.[16] These

included end-diastolic volume, end-systolic volume, and total stroke volume indexed to body surface area (end-diastolic volume index [EDVI], end-systolic volume index [ESVI], and total stroke volume index [TSVI]). Global ejection fraction (EF) was calculated. Body surface area was derived from each animal's body weight using a previously published equation.[17] The LV outflow was obtained from the left-apical 5-chamber view, and pre-ejection period (PEP) and ejection time (ET) were measured; LV PEP/ET was calculated.[11] Forward SV was also calculated as the product of the aortic valve cross-sectional area (using a circular assumption for the aortic valve annulus) and the aortic valve outflow time-velocity integral[18] for subsequent calculation of regurgitant fraction (RF) with the formula, RF = ([total stroke volume − forward stroke volume]/total stroke volume) × 100. In our analyses, we used the mean of 3 consecutive cardiac cycles in sinus rhythm for each measurement.

After completion of standard echocardiographic examinations, high-quality images for 2D-STE analyses were carefully obtained by the same investigator, using the same protocol. We used previously published procedures for the speckle-tracking analysis.[4,5,19] For analysis of 2D-STE results, all views were recorded at rates of 76.1–139.2 frames/s. To evaluate torsional deformations, proper basal and apical short-axis views were carefully obtained using the following anatomic landmarks: at the basal level, mitral valve and at the apical level, LV cavity alone with no papillary muscle visible. Images were analyzed by 1 trained observer using an off-line work station.[j] We obtained peak systolic rotation and rotation rates in each of the apical and basal short-axis planes. Moreover, we calculated peak systolic torsion and torsion rate. The mean values of 3 consecutive cardiac cycles in sinus rhythm were used in all analyses.

Statistical Analysis

Data are expressed as mean ± SD. All statistical analyses were performed by commercially available computer software.[k] We used the Shapiro-Wilk test to evaluate the normal distribution of the variables. We used the Tukey HSD test for testing equality of the means at the 3 points (presurgical and after 3 and 6 months of MR) at rest and during dobutamine infusion. To examine correlations between torsional parameters and overall invasive dp/dt, a linear regression analysis was performed. A P value of <.05 was regarded as significant. Intra-observer reproducibility of 2D-STE measurements was assessed by repeating measurements 3 times for 3 dogs selected at random.

Results

Hemodynamic and Standard Echocardiography Data

Baseline (without dobutamine infusion) hemodynamic and standard echocardiography data, before and 3 and 6 months after MR induction, are summarized in Table 1. LA/Ao ($P < .001$), LVIDd ($P = .001$), LVIDs ($P = .012$), EDVI ($P < .001$), ESVI ($P = .033$), TSVI ($P = .035$), and RF ($P < .001$) were significantly increased 3 months after induction of MR compared with pre-MR measurements. Three months after induction of MR, the dogs had significantly decreased diastolic and systolic sphericity indexes, and RWT compared with presurgical data ($P = .001$, $P < .001$, and $P = .045$, respectively). LA/Ao ($P < .001$), LVIDd ($P < .001$), LVIDs ($P = .006$), EDVI ($P < .001$), ESVI ($P = .008$), TSVI ($P = .002$), and RF ($P < .001$) were significantly increased after 6 months of induced MR compared with presurgical data. Dogs with a 6-month history of induced MR had also significantly decreased invasive dp/dt ($P = .022$), diastolic ($P < .001$) and systolic ($P < .001$) sphericity indexes, and RWT ($P = .022$) compared with presurgical data. The systolic sphericity index was also decreased after 6 months compared with after 3 months of induced MR ($P = .044$). In contrast, heart rate, FS, EF, and PEP/ET were not significantly different at any stage of MR in this study. Selective echocardiography data for baseline (without dobutamine infusion) and dobutamine infusion (10 μg/kg/min) at each time point relative to MR induction are summarized in Table 2. Dogs at 3 and 6 months after MR induction had decreased diastolic sphericity ($P < .001$ and $P < .001$) and systolic sphericity ($P = .016$ and $P = .005$) in response to dobutamine infusion compared with presurgical dobutamine data. Three months after induction of MR, the dogs had significantly decreased RWT in response to dobutamine infusion compared with presurgical data ($P = .03$).

Table 1. Hemodynamic and standard echocardiography data for baseline (without dobutamine infusion) at before and 3 and 6 months after induction of mitral regurgitation.

	Pre	3 Months	6 Months
Heart rate (bpm)	91.1 ± 26.3	127.3 ± 48.6	102.2 ± 27.5
Peak systolic LV pressure (mmHg)	102.2 ± 20.6	92.4 ± 21.5	80.6 ± 8.0
Invasive dp/dt (mmHg/s)	2, 207.2 ± 377.4	1, 630 ± 552.9	1, 331.4 ± 156.5[a]
LA/Ao	1.2 ± 0.1	2.0 ± 0.2[a]	2.2 ± 0.2[a]
LVIDd (mm)	26.3 ± 2.5	38.7 ± 4.9[a]	41.8 ± 2.8[a]
EDVI (mL/m^2)	47.6 ± 11.1	90.0 ± 9.1[a]	117.9 ± 11.0[a]
TSVI (mL/m^2)	60.5 ± 12.2	113.8 ± 42.9[a]	153.0 ± 20.8[a]
RF (%)	N.A.	43.7 ± 20.9[a]	56.8 ± 10.1[a]

Data are expressed as mean ± SD.

LV, left ventricular; dp/dt, peak positive first derivatives of the left ventricular pressure; LA/Ao, left atrial-to-aortic root ratio; LVIDd, end-diastolic left ventricular internal dimension; EDVI, end-diastolic volume index; TSVI, total stroke volume index; RF, regurgitant fraction; N.A., Not applied.

[a]Within a row, values differed significantly from pre values.

2D-STE

Dobutamine stress tests were feasible in the anesthetized dogs at any stage of induced MR in this study. All myocardial segments for speckle-tracking images were analyzed and included in the statistical analyses. The mean of the standard deviation and coefficient of variation were 6.9° and 6.4% for peak systolic torsion, and 9.1°/s and 11.2% for peak systolic torsion rate.

Table 2. Standard echocardiography data for baseline (without dobutamine infusion) and dobutamine infusion (10 μg/kg/min) before and 3 and 6 months after induction of mitral regurgitation.

	Pre	3 Months	6 Months
LVIDs (mm)			
Baseline	19.7 ± 3.6	27.0 ± 3.9[a]	28.3 ± 1.2[a]
Dobutamine	14.3 ± 2.3	22.0 ± 6.6	21.8 ± 0.9
FS (%)			
Baseline	25.1 ± 11.8	29.9 ± 8.6	32.2 ± 2.6
Dobutamine	49.9 ± 11.1	43.3 ± 18.5	42.9 ± 8.6
LV PEP/ET			
Baseline	0.24 ± 0.07	0.29 ± 0.05	0.28 ± 0.07
Dobutamine	0.15 ± 0.03	0.20 ± 0.05	0.23 ± 0.10
EF (%)			
Baseline	59.5 ± 6.1	57.3 ± 16.4	62.3 ± 2.7
Dobutamine	62.0 ± 11.1	62.5 ± 14.5	69.3 ± 6.1
ESVI (mL/m^2)			
Baseline	19.8 ± 6.8	37.9 ± 13.5[a]	44.6 ± 6.5[a]
Dobutamine	17.2 ± 5.9	32.1 ± 9.3	30.7 ± 3.8
Diastolic sphericity			
Baseline	1.72 ± 0.09	1.36 ± 0.15[a]	1.29 ± 0.07[a]
Dobutamine	1.73 ± 0.07	1.38 ± 0.12[c]	1.29 ± 0.10[c]
Systolic sphericity			
Baseline	1.85 ± 0.22	1.51 ± 0.25[a]	1.32 ± 0.13[ac]
Dobutamine	1.95 ± 0.20	1.56 ± 0.21[c]	1.44 ± 0.10[c]
RWT			
Baseline	0.58 ± 0.06	0.42 ± 0.14[a]	0.42 ± 0.08[a]
Dobutamine	0.64 ± 0.08	0.47 ± 0.12[c]	0.49 ± 0.07

Data are expressed as mean ± SD.

LVIDs, end-systolic left ventricular internal dimension; FS, fractional shortening; LV PEP/ET, left ventricular pre-ejection period-to-ejection time ratio; EF, ejection fraction; ESVI, end-systolic volume index; RWT, Relative wall thickness.

[a]Within a row, values differed significantly from pre values at rest.

[b]Within a row, values differed significantly from 3-month values at rest.

[c]Within a row, values differed significantly from dobutamine responsive values at pre (control).

The 2D-STE data for baseline (without dobutamine infusion) and dobutamine infusion (10 μg/kg/min) at each time point relative to MR induction are summarized in Table 3. All 2D-STE variables for baseline (without dobutamine infusion) were not significantly changed at any stage of MR in this study. Dogs at 3 and 6 months after MR induction had decreased peak systolic apical rotation ($P = .005$ and $P = .006$), apical rotation rate ($P = .002$ and $P = .044$), torsion ($P < .001$ and $P = .003$), and torsion rate ($P = .006$ and $P = .021$) in response to dobutamine infusion compared with presurgical dobutamine data. Peak systolic basal rotation and basal rotation rates were not significantly changed during any stage of the MR model in this study. Peak systolic basal rotation ($r = -0.41$, $P = .009$), basal rotation rate ($r = -0.667$, $P < .001$), apical rotation ($r = 0.399$, $P = .009$), apical rotation rate ($r = 0.66$, $P < .001$), torsion ($r = 0.644$, $P < .001$), and torsion rate ($r = 0.696$, $P < .001$) were significantly correlated with invasive dp/dt. Correlations between peak systolic torsion and torsion rate, and invasive dp/dt are shown in Figure 1.

Table 3. Peak systolic 2-dimensional speckle-tracking echocardiography data for baseline (without dobutamine infusion) and dobutamine infusion (10 μg/kg/min) before and 3 and 6 months after induction of mitral regurgitation.

	Pre	3 Months	6 Months
Basal rotation (°)			
Baseline	−3.0 ± 2.3	−1.6 ± 2.1	−2.8 ± 2.7
Dobutamine	−4.9 ± 2.8	−2.5 ± 1.3	−4.4 ± 3.2
Basal rotation rate (°/s)			
Baseline	−78.4 ± 45.5	−43.5 ± 22.4	−67.1 ± 29.3
Dobutamine	−128.8 ± 53.3	−112.6 ± 40.2[a]	−112.0 ± 75.2
Apical rotation (°)			
Baseline	12.1 ± 12.0	4.9 ± 6.2	4.6 ± 4.3
Dobutamine	20.6 ± 2.8	12.1 ± 7.3[a]	10.5 ± 4.8[a]
Apical rotation rate (°/s)			
Baseline	122.2 ± 85.8	57.7 ± 46.3	62.6 ± 51.2
Dobutamine	230.0 ± 64.4	104.8 ± 77.1	124.7 ± 59.7
Torsion (°)			
Baseline	12.7 ± 6.7	6.2 ± 4.5	7.6 ± 3.2
Dobutamine	23.8 ± 3.6	15.9 ± 8.1[a]	14.2 ± 2.4[a]
Torsion rate (°/s)			
Baseline	158.8 ± 79.1	67.8 ± 36.5	78.0 ± 30.7
Dobutamine	272.4 ± 66.9	178.1 ± 69.3[a]	152.5 ± 60.8[a]

Data are expressed as mean ± SD.

[a]Within a row, values differed significantly from dobutamine responsive values at pre (control).

Discussion

Our findings confirmed that myocardial deformations during dobutamine stress testing could be assessed quantitatively using 2D-STE in dogs with MR. We also confirmed that 2D-STE–derived torsional motions, during dobutamine stress tests, were diminished in anesthetized dogs with severe MR compared with the pre-MR (control) state. Furthermore, these torsional motions were significantly correlated with the overall invasive dp/dt.

Systolic function has been suggested to decrease in dogs with naturally acquired MMVD[2,3] and in dogs with experimentally induced MR.[20,21] However, identification of systolic dysfunction is challenging in dogs with MMVD[22]; EF, FS, and PEP/ET at rest and dobutamine infusion could not detect systolic dysfunction. In addition to being dependent on intrinsic contractility, these parameters are also known to be influenced by hemodynamic load and sympathetic tone, which may mask significant myocardial dysfunction in dogs with MR.[22] In contrast, the sphericity index, in both systole and diastole, and RWT were observed to be significantly decreased during disease progression at rest and after dobutamine infusion, findings that agree with previous studies.[23,24] These geometrical variables may be useful in dogs with MR as part of disease stratification.

Twisting of the left ventricle is determined by the oblique orientation of the myocardial fibers, such that

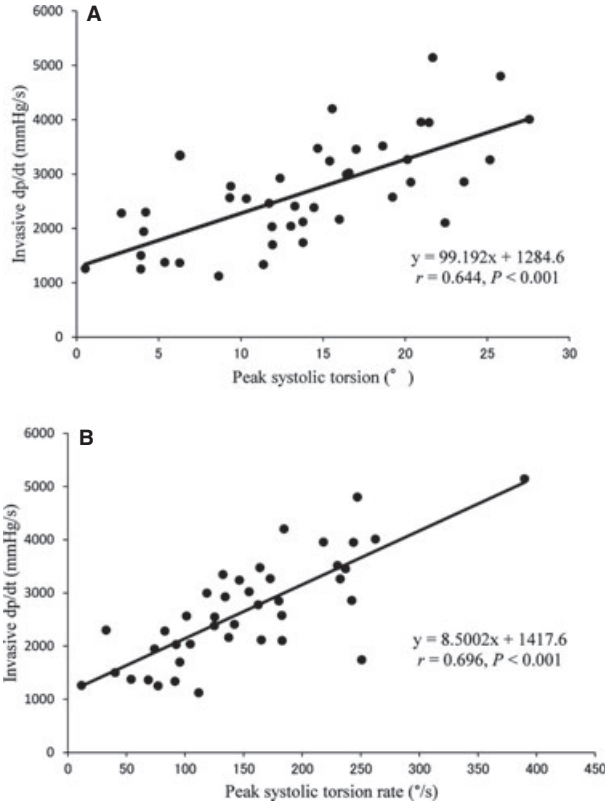

Fig 1. Correlations between peak systolic torsion **(A)** and torsion rate **(B)**, and invasive peak positive first derivatives of the left ventricular pressure (dp/dt) in overall dogs in this study.

2D-STE–derived torsional parameters are more sensitive than conventional echocardiographic parameters for detecting subtle myocardial functional abnormalities.[5,25] Previous studies have demonstrated that systolic torsion is decreased in animal models of chronic MR, compared with the acute phase in the same individuals.[26,27] Furthermore, we recently reported that systolic torsion was lower in dogs with advanced MMVD compared with weight- and age-matched healthy controls.[5] The decrease in torsional deformations at baseline (without dobutamine infusion), which agree with previous studies,[5,26,27] may reflect deterioration in myocardial contractility with disease progression, although these changes did not reach statistical significance. Small sample size, anesthesia effect, and myocardial compensation remodeling that differ from naturally acquired MMVD may have affected the results of this study.

Given that increased LVIDs and ESVI, and decreased invasive dp/dt, are associated with disease progression, systolic function was decreased in the MR model in this study. During dobutamine infusion, peak systolic torsion and torsional rates were decreased after 3 and 6 months of induced MR in the dogs compared with the presurgical (control) state. Such small responses during dobutamine infusion are believed to reflect systolic dysfunction in dogs with MR. In humans with severe MR, systolic function, as assessed by exercise echocardiography, predicts preservation of LV function and good clinical outcome after medical treatment.[6] Decreased systolic function may also contribute to limitations in exercise capacity in these patients more than the finding of large regurgitant volumes.[7] Although we could not assess exercise capacities and clinical outcomes in this study and the results of this study were not clearly superior to results of resting echocardiography, dobutamine infusion–assessed systolic function in dogs with MR may be an additional tool for detection of the systolic dysfunction. Furthermore, small responses during stress echocardiography might be useful before mitral valve surgery to identify those patients at risk of developing postoperative LV dysfunction.[28]

Dobutamine is a known agonist of the beta-adrenergic receptor, and inability to respond to beta-adrenergic stimulation is a hallmark of the failing human heart.[29,30] Isolated muscles and myocytes from failing human hearts show decreased inotropic response to beta-adrenergic agonists[31–33] and down-regulation of beta-adrenergic receptors.[29–31] Although we could not obtain histopathologic data and the dogs included in this study did not have obvious evidence of heart failure, the responsiveness of dobutamine stress to MR disease progression may suggest the presence of these functional abnormalities.

Left ventricular torsion is also thought to be a mechanism by which the ventricle equalizes transmural oxygen demand gradients, thus minimizing oxygen consumption and optimizing myocardial energetics and efficiency.[34,35] Increased myocardial oxygen consumption during the dobutamine stress test and decreased oxygen supply because of reduction in forward stroke volume in dogs with severe MR[23] may also contribute to decreased myocardial responsiveness. Therefore, response to dobutamine infusion in dogs with MR may provide additional information for the assessment of progression of disease, treatment responses, and prognosis.

Our study has several limitations. Because of the study design, the effects of anesthesia on myocardial function could not be eliminated. Therefore, our findings should not necessarily be extrapolated to conscious dogs. In this study, MMVD was modeled using young Beagle dogs with a history of up to 6 months of induced MR. However, dogs with naturally occurring, chronic MMVD might have different types of myocardial damage and myocardial function. Although previous studies in dogs demonstrated that dobutamine stress echocardiography was feasible, safe, and repeatable for conscious dogs,[8–11] older and diseased dogs may experience some adverse effects, such as arrhythmias. Future studies to assess safety in older conscious dogs are needed. Nevertheless, our low-dose protocol, instead of a high-dose dobutamine stress protocol as used in other previous studies, might decrease the development of adverse effects. Calculations of LV torsion depend on the nonsimultaneous measurements of 2 segments (base and apex), which may have affected our results. In addition, myocardial rotation has been

reported to be dependent on transducer angulation,[36] and acquiring the true apex of the left ventricle needed for 2D-STE analysis might not be technically feasible. Speckle-imaging, as determined by 3-dimensional echocardiography in a future study, may help to resolve these limitations. Finally, future work incorporating pressure-volume loop analyses for more accurate evaluation of cardiac function is warranted.

In conclusion, indices of geometrical variables at rest (sphericity index and RWT) and during a dobutamine stress test (2D-STE–derived torsional deformations) were decreased with the progression of MR severity. The 2D-STE method can quantitate myocardial deformations during dobutamine infusion, and decreased torsion during dobutamine infusion in dogs with MR may reflect latent systolic dysfunction. Nevertheless, the clinical importance of these findings and their relationship with survival time should be investigated further.

Footnotes

[a] Vetrphale; Meiji Seika, Tokyo, Japan
[b] Ravonal; Tanabe Mitsubishi Pharma, Osaka, Japan
[c] Isoflu; Dainippon Sumitomo Pharma, Osaka, Japan
[d] Model SPR-350, 5Fr; Millar Instruments, Houston, TX
[e] Marcain 0.5%; AstraZeneca, Osaka, Japan
[f] Larixin; Taisho Toyama Pharma, Tokyo, Japan
[g] Methacam 0.15%; Nippon Boehringer Ingelheim, Tokyo, Japan
[h] SBP2000; Softron, Tokyo, Japan
[i] Dobutrex; Shionogi & Co, Osaka, Japan
[j] GE Healthcare, Tokyo, Japan
[k] SPSS version 15.0J for Windows; SPSS, Tokyo, Japan

Acknowledgments

The authors thank Mayu Hamano, Norikazu Katagiri, Rei Okunaka, Satoko Ueno, and Takeshi Shintani for their technical assistance. This study was not supported or presented.

Conflict of Interest Declaration: The authors disclose no conflict of interest.

References

1. Kittleson MD. Myxomatous atrioventricular valvular degeneration. In: Kittleson MD, Kienle RD, eds. Small Animal Cardiovascular Medicine. St. Louis, MO: Mosby; 1998:297–303.

2. Borgarelli M, Tarducci A, Zanatta R, et al. Decreased systolic function and inadequate hypertrophy in large and small breed dogs with chronic mitral valve insufficiency. J Vet Intern Med 2007;21:61–67.

3. Borgarelli M, Savarino P, Crosara S, et al. Survival characteristics and prognostic variables of dogs with mitral regurgitation attributable to myxomatous valve disease. J Vet Intern Med 2008;22:120–128.

4. Suzuki R, Matsumoto H, Teshima T, et al. Clinical assessment of systolic myocardial deformations in dogs with chronic mitral valve insufficiency using two-dimensional speckle-tracking echocardiography. J Vet Cardiol 2013;15:41–49.

5. Suzuki R, Matsumoto H, Teshima T, et al. Noninvasive clinical assessment of systolic torsional motions using two-dimensional speckle-tracking echocardiography in dogs with myxomatous mitral valve disease. J Vet Intern Med 2013;27:69–75.

6. Lee R, Haluska B, Leung DY, et al. Functional and prognostic implications of left ventricular contractile reserve in patients with asymptomatic severe mitral regurgitation. Heart 2005;91:1407–1412.

7. Leung DY, Griffin BP, Snader CE, et al. Determinants of functional capacity in chronic mitral regurgitation unassociated with coronary artery disease or left ventricular dysfunction. Am J Cardiol 1997;79:914–920.

8. McEntee K, Amory H, Clercx C, et al. Physiologic response to dobutamine infusion during cardiac stress testing of dogs. Am J Vet Res 1998;59:1160–1165.

9. Spasojević Kosić L, Trailović DR, Matunović R. Resting and dobutamine stress test induced serum concentrations of brain natriuretic peptide in German Shepherd Dogs. Res Vet Sci 2012;93:1446–1453.

10. Mc Entee K, Clercx C, Soyeur D, et al. Usefulness of dobutamine stress tests for detection of cardiac abnormalities in dogs with experimentally induced early left ventricular dysfunction. Am J Vet Res 2001;62:448–455.

11. Minors SL, O'Grady MR. Resting and dobutamine stress echocardiographic factors associated with the development of occult dilated cardiomyopathy in healthy Doberman Pinscher dogs. J Vet Intern Med 1998;12:369–380.

12. Reant P, Labrousse L, Lafitte S, et al. Experimental validation of circumferential, longitudinal, and radial 2-dimensional strain during dobutamine stress echocardiography in ischemic conditions. J Am Coll Cardiol 2008;51:149–157.

13. Rishniw M, Erb HN. Evaluation of four 2-dimensional echocardiographic methods of assessing left atrial size in dogs. J Vet Intern Med 2000;14:429–435.

14. Dini FL, Capozza P, Donati F, et al. Patterns of left ventricular remodeling in chronic heart failure: Prevalence and prognostic implications. Am Heart J 2011;161:1088–1095.

15. Gomez-Doblas JJ, Schor J, Vignola P, et al. Left ventricular geometry and operative mortality in patients undergoing mitral valve replacement. Clin Cardiol 2001;24:717–722.

16. Zoghbi WA, Enriquez-Sarano M, Foster E, et al; American Society of Echocardiography. Recommendations for evaluation of the severity of native valvular regurgitation with two-dimensional and Doppler echocardiography. J Am Soc Echocardiogr 2003;16:777–802.

17. Jacobs G, Mahjoob K. Multiple regression analysis, using body size and cardiac cycle length, in predicting echocardiographic variables in dogs. Am J Vet Res 1988;49:1290–1294.

18. Lewis JF, Kuo LC, Nelson JG, et al. Pulsed Doppler echocardiographic determination of stroke volume and cardiac output: Clinical validation of two new methods using the apical window. Circulation 1984;70:425–431.

19. Suzuki R, Matsumoto H, Teshima T, et al. Influence of heart rate on myocardial function using two-dimensional speckle-tracking echocardiography in healthy dogs. J Vet Cardiol 2013;15:139–146.

20. Carabello BA, Nakano K, Corin W, et al. Left ventricular function in experimental volume overload hypertrophy. Am J Physiol 1989;256:H974–H981.

21. Urabe Y, Mann DL, Kent RL, et al. Cellular and ventricular contractile dysfunction in experimental canine mitral regurgitation. Circ Res 1992;70:131–147.

22. Bonagura JD, Schober KE. Can ventricular function be assessed by echocardiography in chronic canine mitral valve disease? J Small Anim Pract 2009;50(Suppl):12–24.

23. Suzuki R, Matsumoto H, Teshima T, et al. Left ventricular geometrical differences in dogs with various stages of

myxomatous mitral valve disease. J Small Anim Pract 2013;54:234–239.

24. Paraskevaidis IA, Dodouras T, Adamopoulos S, et al. Effects of dobutamine on left ventricular shape and geometry: An easy way to detect the functional status of chronic heart failure in patients with dilated cardiomyopathy. J Am Soc Echocardiogr 2003;16:132–139.

25. Chetboul V, Serres F, Gouni V, et al. Noninvasive assessment of systolic left ventricular torsion by 2-dimensional speckle tracking imaging in the awake dog: Repeatability, reproducibility, and comparison with tissue Doppler imaging variables. J Vet Intern Med 2008;22:342–350.

26. Ennis DB, Nguyen TC, Itoh A, et al. Reduced systolic torsion in chronic "pure" mitral regurgitation. Circ Cardiovasc Imaging 2009;2:85–92.

27. Tibayan FA, Yun KL, Fann JI, et al. Torsion dynamics in the evolution from acute to chronic mitral regurgitation. J Heart Valve Dis 2002;11:39–46.

28. Donal E, Mascle S, Brunet A, et al. Prediction of left ventricular ejection fraction 6 months after surgical correction of organic mitral regurgitation: The value of exercise echocardiography and deformation imaging. Eur Heart J Cardiovasc Imaging 2012;13:922–930.

29. Bristow MR, Ginsburg KS, Minobe W, et al. Decreased catecholamine sensitivity and beta-adrenergic receptor density in failing human hearts. N Engl J Med 1982;307:205–211.

30. Engelhardt S, Bohm M, Erdmann E, et al. Analysis of beta-adrenergic receptor mRNA levels in human ventricular biopsy specimens by quantitative polymerase chain reactions: Progressive reduction of beta1-adrenergic receptor mRNA in heart failure. J Am Coll Cardiol 1996;27:146–154.

31. Bristow MR, Anderson FL, Port JD, et al. Differences in b-adrenergic neuroeffector mechanisms in ischemic versus idiopathic dilated cardiomyopathy. Circulation 1991;84:1024–1039.

32. Davies CH, Davia K, Bennett JG, et al. Reduced contraction and altered frequency response of isolated ventricular myocytes from patients with heart failure. Circulation 1995;92: 2540–2549.

33. Feldman MD, Copelas L, Gwathmey JK, et al. Deficient production of cyclic AMP: Pharmacologic evidence of an important cause of contractile dysfunction in patients with end-stage heart failure. Circulation 1987;75:331–339.

34. Beyar R, Sideman S. Left ventricular mechanics related to the local distribution of oxygen demand throughout the wall. Circ Res 1986;58:664–677.

35. Ohayon J, Chadwick RS. Theoretical analysis of the effects of a radial activation wave and twisting motion on the mechanics of the left ventricle. Biorheology 1988;25:435–447.

36. Grabskaya E, Spira C, Hoffmann R, et al. Myocardial rotation but not circumferential strain is transducer angle dependent: A speckle tracking echocardiography study. Echocardiography 2010;27:809–814.