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EXPERIMENTAL STUDIES

Impact of Volume Loading and Load Reduction on Ventricular Refractoriness and Conduction Properties in Canine Congestive Heart Failure

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Objectives. **This investigation was undertaken to examine the alteration of electrophysiologic properties, including refractoriness, strength–interval relations and conduction, with the development of heart failure and to characterize the impact of volume loading on these indexes in the cardiomyopathic setting.**

Methods. **Electrophysiologic properties in eight dogs with pacing-induced dilated cardiomyopathy were compared with those in six control dogs before and after rapid infusion of 800 ml of intravenous saline.**

Results. **The right ventricular (RV) and left ventricular (LV) effective refractory period (ERP) and absolute refractory period (ARP) were significantly longer in dogs with pacing-induced cardiomyopathy than in control dogs: RV ERP 181 ± 11 ms versus 138 ± 7 ms (mean ± SD) ($p < 0.0001$) and anterior LV ERP 177 ± 13 ms versus 128 ± 11 ms ($p < 0.0001$), respectively; ARP 159 ± 14 ms versus 114 ± 7 ms ($p < 0.0001$) at the RV site and 153 ± 12 versus 117 ± 5 ms ($p < 0.0001$) at the anterior LV site. After volume loading in cardiomyopathic animals, posterior and anterior LV ERPs became prolonged to 178 ± 5 ms ($p = 0.004$) and 189 ± 14 ms ($p = 0.065$), respectively, shifting the strength–interval relation in the direction of longer S_1S_2 coupling intervals. Anterior LV monophasic action potential durations at 90% repolarization also became prolonged from 192 ± 10 ms to 222 ± 23 ms ($p < 0.012$) with volume loading. These findings were not altered by subsequent sodium nitroprusside. Local conduction times parallel and perpendicular to fiber orientation were not altered by development of cardiomyopathy or volume alterations.**

Conclusions. **The development of dilated cardiomyopathy results in significant prolongation of refractoriness and repolarization that is increased further by volume augmentation but is not reversed by pharmacologic load reduction. Although these abnormalities may contribute to the environment needed for a nonreentrant, triggered or stretch-mediated arrhythmogenic process in cardiomyopathic states, additional studies will be required to demonstrate such a focal mechanism conclusively.**

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The relation between general structural and electrophysiologic characteristics of the heart has been explored through a variety of models (1–14). Transient aortic occlusion (4), papillary muscle stretch (5) and left ventricular (LV) free wall traction (6) produce prolongations in effective or functional refractory periods. In contrast, volume-induced distortion of cardiac geometry, while producing little effect in the normal canine ventricle (7–10), significantly decreases refractoriness in the isolated normal rabbit heart (11) and the cross-circulating canine heart (12). Prominent reductions in refractoriness have also been described with volume stress in an ex situ canine infarct model (8) and in an isolated rabbit ventricle model of reentrant arrhythmia (13). In general, changes in refractoriness have paralleled changes in recovery (3,12,14). In some preparations, these pressure and volume interventions produce extrasystolic activity (2,10,14). In contrast, Li et al. (14) demonstrated prolongation of refractoriness in a canine model of pacing-induced dilated cardiomyopathy. Nevertheless, the electrophysiologic response to volume loading and load reduction has not been studied. This investigation was therefore undertaken to characterize the impact of volume loading and load reduction in the cardiomyopathic setting.

Methods

Pacemaker implantation. Eighteen mongrel dogs (12 in the study group and 6 in the control group) were studied after a protocol approved by the Mayo Foundation Institutional Animal Care and Use Committee. Each animal underwent permanent epicardial pacemaker implantation for generating tachycardia-induced cardiomyopathy (15–17). The animals were anesthetized with sodium pentobarbital (usually 30 mg/kg

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Abbreviations and Acronyms

ARP = absolute refractory period
CHF = congestive heart failure
CL = cycle length
EF = ejection fraction
ERP = effective refractory period
LV = left ventricle, left ventricular
MAP = monophasic action potential
RV = right ventricle, right ventricular

body weight), ventilated with supplemental oxygen and electrocardiographically monitored. Clindamycin (300 mg subcutaneously) and Combiotic penicillin (2 ml intramuscularly) were given prophylactically. A 53-cm epicardial, unipolar, screw-in lead was positioned at the apex of the heart through a fifth left intercostal space thoracotomy and tunneled to a modified Medtronic 8320 or 8239 pulse generator secured in an infrascapular pocket. The chest wall was closed with multiple layers of 3.0 Vicryl suture after the pleural cavity was evacuated with a chest tube. The dogs were allowed to recover from anesthesia for several days and were treated liberally with butorphanol as needed for discomfort.

Prepacing evaluation. Forty-eight hours postoperatively, neurohumoral indicators of congestive heart failure (CHF) were measured. Cardiac dimensions, wall thickness and ejection fraction (EF) were assessed by two-dimensional and M-mode echocardiographic studies. After 24-h ambulatory recordings were made, the pacemaker in study animals was programmed to the VOO mode at a rate of 250 beats/min. The paced group was examined daily, with periodic electrocardiographic monitoring to ensure appropriate pacemaker capture. After 4 weeks, the pacemakers were reprogrammed to the VVI mode (40 beats/min), and blood for neurohumoral factors, echocardiographic images and 24-h ambulatory ECG data were collected again. Pacemakers in control dogs remained in the VVI mode (40 beats/min).

Open chest study. *Instrumentation.* After repeat data acquisition, the dogs were again anesthetized with intravenous sodium pentobarbital and ventilated. To avoid potentially confounding the effects of pentobarbital on repolarization, constant anesthesia was initiated before and maintained throughout data collection. Body temperature was maintained at 37°C with a water-circulating heating pad and overhead lamps. A 7F Swan-Ganz catheter was advanced through an external jugular vein for measurement of intracardiac pressures and cardiac output. A 6F pigtail catheter was inserted through a femoral artery sheath for measurement of LV pressures.

After median sternotomy, three custom-made, 12 × 12 mm Silastic conforming plaques, with three pairs of recording electrodes (4 mm interpair spacing) arranged in a "cross" configuration around a central pacing site, were sutured to the epicardial surface of the right ventricular (RV) outflow tract and the anterior and posterior LV walls. Coaxial bipolar pacing was used to eliminate virtual cathode effects (18). The plaques were positioned with two electrode arms parallel and two transverse to the epicardial fiber orientation. Resulting electrograms were converted digitally and recorded on a 56-channel cardiac mapping system for subsequent analysis. Epicardial monophasic action potentials (MAPs) were recorded from near the RV outflow tract and anterior LV plaques with a table-mounted, spring-loaded cantilever system (19). Myocardial temperature was monitored using an intracardiac thermistor and an intramyocardial thermocouple positioned in the apex of the heart. The pH, partial pressure of oxygen, partial pressure of carbon dioxide and potassium concentrations were also serially monitored.

Refractoriness, action potential duration and conduction examination. After hemodynamic data were again acquired, refractoriness was evaluated from each epicardial plaque with twice threshold diastolic scanning after an 8-beat drive train (cycle length [CL] = 300 ms). The S_2 coupling interval was decreased by 10 ms until capture no longer occurred. The final 10 ms was rescanned in 2-ms steps to the longest S_1S_2 interval without capture, which was taken as the effective refractory period (ERP). The stimulus current was then increased in 0.5- to 1.0-mA increments until capture again occurred. The S_1S_2 interval was decreased in 2-ms intervals until capture was lost. This sequence was repeated to a 10-mA current. The absolute refractory period (ARP) was defined as the longest coupling interval at which a current of 10 mA failed to produce a propagated response.

A stable MAP configuration and diastolic segment over the course of the experiment were required for data analysis. The onset of the action potential was defined as the earliest point of abrupt rise of phase zero and the point of 90% MAP recovery referenced to the maximal amplitude of the plateau phase of the action potential. Typically, measurements from three consecutive action potentials during sinus rhythm or pacing at a CL of 300 ms were averaged for analysis. Local conduction times from the proximal to distal recording electrodes, as referenced to the stimulus artifact, were determined during coaxial pacing (CL = 300 ms).

Study protocol. After thoracotomy, hemodynamic status, refractoriness, local conduction times and epicardial MAP durations were assessed. Data were again rapidly acquired after 800 ml of 0.9% saline administered over a 20-min period through two central venous access sites and after an intravenous sodium nitroprusside infusion titrated to effectively unload the ventricle as indicated by a decrease in systolic blood pressure to ≤80 mm Hg or a decrease in LV EDP to a value below baseline, or both. After the blood pressure had recovered following sodium nitroprusside administration, the inducibility of ventricular tachyarrhythmias as a secondary consideration was determined using up to four extrastimuli from two LV sites.

At the end of the study, the position of each recording plaque was marked, and the heart was rapidly excised after induction of ventricular fibrillation and exsanguination. After the heart was perfused with 10% formaldehyde through a low

Table 1. Hemodynamic Measurements in Control and Cardiomyopathic Dogs

	HR (beats/min)	BP (mm Hg)	LVEDP (mm Hg)	RAP (mm Hg)	PAP (mm Hg)	PCWP (mm Hg)	CO (liters/min)
Before thoracotomy							
Control group (n = 6)	160 ± 18	146 ± 13/101 ± 10	11 ± 5	4 ± 2	22 ± 3/8 ± 2	6 ± 2	3.2 ± 0.4
CHF group (n = 8)	127 ± 22	113 ± 21/72 ± 14	34 ± 7	10 ± 4	40 ± 9/26 ± 10	25 ± 8	1.8 ± 0.5
p value	0.01	0.006/0.001	0.0001	0.008	0.002/0.002	0.0003	0.0003
After thoracotomy							
Control group (n = 6)							
Baseline	159 ± 22	108 ± 18/76 ± 17	5 ± 4	3 ± 1	18 ± 5/8 ± 1	4 ± 1	2.4 ± 0.5
Volume	135 ± 11	148 ± 19#/98 ± 20‖	12 ± 6§	8 ± 3#	26 ± 6¶/11 ± 4‖	10 ± 2#	3.7 ± 0.6#
Nitroprusside	158 ± 29	63 ± 8‡‡/46 ± 11‡‡	3 ± 2††	4 ± 1††	17 ± 3††/17 ± 2††	6 ± 2‡‡	1.6 ± 0.4‡‡
Paced group (n = 8)							
Baseline	130 ± 8†	91 ± 16/57 ± 12*	17 ± 9†	4 ± 2	25 ± 6*/13 ± 5*	10 ± 5†	1.7 ± 0.6*
Volume	119 ± 18	112 ± 22§†/60 ± 11‡	29 ± 8‡¶	11 ± 5¶	36 ± 6†¶/20 ± 7†	17 ± 4‡§	2.4 ± 0.7‡
Nitroprusside	127 ± 13*	62 ± 4‡‡/39 ± 6††	6 ± 4‡‡	5 ± 5††	18 ± 3‡‡/10 ± 3**	7 ± 3††	1.8 ± 1.0

Comparisons made between control and paced groups: *$p < 0.05$; †$p \leq 0.02$; ‡$p \leq 0.01$. Comparisons made within the control or paced groups: §$p < 0.05$ compared with baseline; ‖$p \leq 0.02$ compared with baseline; ¶$p \leq 0.01$ compared with baseline; #$p < 0.001$ compared with baseline; **$p < 0.02$ compared with volume; ††$p < 0.01$ compared with volume; ‡‡$p < 0.001$ compared with volume. BP = blood pressure; CO = cardiac output; CHF = congestive heart failure; HR = heart rate; LVEDP = left ventricular end-diastolic pressure; PAP = pulmonary artery pressure; PCWP = pulmonary capillary wedge pressure; RAP = right atrial pressure.

pressure perfusion pump for 24 h, weight, ventricular volumes and LV and RV free wall and interventricular septum thickness were measured. The myocardium underlying the electrode plaques was stained with hematoxylin-eosin for verification of epicardial fiber orientation and analysis of microscopic tissue changes.

Statistical analysis. Data are presented as mean value ± SD. Using a one-way analysis of variance technique, the differences were analyzed between control and paced animals and between baseline and each subsequent state within each study group in which each animal served as its own control. After incorporating a Bonferroni correction for multiple comparisons, $p < 0.0167$ was considered significant.

Results

Clinical, echocardiographic and neurohumoral features. All six of the control dogs remained symptom free throughout the observation period. Typically, study dogs developed findings of CHF after 3 weeks of pacing. One dog progressed to terminal CHF, and three developed electrical-mechanical dissociation or hypotension precluding study with anesthesia induction or sternotomy. Consequently, 8 of 12 paced dogs were evaluated. Under baseline conditions, the eight remaining study dogs were echocardiographically comparable to the control dogs. With the development of CHF in paced animals, the LV end-diastolic dimension increased from 39 ± 5 mm to 47 ± 3 mm ($p < 0.0001$) and the end-systolic dimension increased from 24 ± 4 mm to 40 ± 3 mm ($p < 0.0001$). This was accompanied by a reduction in LV EF from 62 ± 3% to 26 ± 4% ($p < 0.0001$). Interventricular septum and posterior diastolic wall thicknesses decreased significantly after prolonged rapid pacing, and the increase in wall thickness with contraction diminished.

The baseline neuroendocrine values in the study animals were also comparable to the control animals. After 4 weeks of continuous pacing, serum norepinephrine levels increased from 286 ± 74 pg/ml to 1073 ± 729 pg/ml ($p < 0.01$); atrial natriuretic peptides increased from 17 ± 6 pg/ml to 285 ± 151 pg/ml ($p < 0.0001$); plasma renin increased from 1.4 ± 1.5 ng/ml per h to 7.1 ± 5.7 ng/ml per h ($p < 0.01$); and aldosterone increased from 4.8 ± 2.3 ng/dl to 74.0 ± 82.7 ng/dl ($p < 0.02$). Serum endothelin levels did not change with the development of heart failure.

Hemodynamic changes. Measurements made before and after sternotomy and pericardiotomy, as well as after study interventions, are summarized in Table 1. Before sternotomy, the heart rate was slower in the study dogs than in the control dogs and tended to decrease with volume loading and increase with load reduction in both study and control groups. The systolic and diastolic blood pressures in the closed-chest state were lower, and the cardiac output was significantly lower in the study animals than in the control animals. Intracardiac pressures were significantly higher with CHF. A general reduction of cardiac pressures in both groups accompanied pericardial opening. Still, blood pressure and cardiac output remained lower and intracardiac pressures higher in the animals with CHF (Table 1).

Abnormalities of refractoriness and repolarization. In the setting of marked dilated cardiomyopathy, refractory periods were significantly longer in the study group than in the control group (Fig. 1). The mean RV ERP was 181 ± 11 ms in the study group and only 138 ± 7 ms in the control group ($p < 0.0001$). Similarly, the ARP was 159 ± 14 ms in dogs with CHF, which was significantly longer than the 114 ± 7 ms seen in control dogs ($p < 0.0001$). Anterior and posterior LV ERPs and ARPs were similarly prolonged in the study group (Table 2). As a result, the strength–interval curves in the presence of the dilated cardiomyopathy were shifted in the direction of longer S_1S_2 coupling intervals, as illustrated in Figure 2.

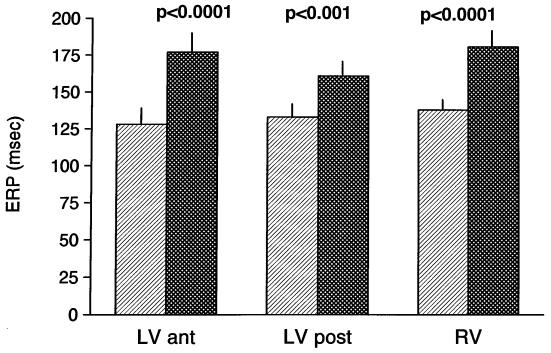


Figure 1. Comparison of ERPs in six dogs with CHF (**crosshatched bars**) and eight control dogs (**hatched bars**). With the development of CHF, the ERP in study dogs was significantly longer at the anterior (ant) and posterior (post) LV and RV sites than that observed in the control dogs. Similarly, ARPs were substantially longer in the paced than in the control animals.

Apparent dispersion of ventricular ERP and ARP, defined as the difference between the longest and the shortest ventricular ERP or ARP at three different ventricular sites, was comparable in both groups of dogs: ERP 17 ± 14 ms in study dogs vs. 15 ± 7 ms in control dogs; ARP 13 ± 7 ms in study dogs vs. 7 ± 9 ms in control dogs.

Similarly, MAP duration was significantly longer at both RV and LV positions in dogs with CHF, as shown in Figure 3. During normal sinus rhythm, the MAP duration of 192 ± 10 ms determined at the LV site was significantly longer than the 166 ± 12 ms value at equivalent sinus rates in control animals ($p < 0.01$). Similar findings were noted at the RV measurement site (Table 3). During pacing at a CL of 300 ms, the 192 ± 17 ms value in animals with CHF was significantly longer than the 165 ± 20 ms duration in control animals ($p = 0.016$). The LV MAP duration at a CL of 300 ms was also longer in CHF than in control groups, but this did not reach significance ($p = 0.078$).

Change in repolarization with volume loading. Volume loading with normal saline over 20 min produced significant increments in the intracardiac pressures in the dogs with CHF (Table 1). In paced animals, the end-diastolic pressure increased from 17 ± 9 mm Hg to 29 ± 8 mm Hg ($p < 0.007$), and the cardiac output increased from 1.7 ± 0.6 liters/min to 2.4 ± 0.7 liters/min, although this was not significant. Myocardial temperatures and pH were not appreciably altered by the fluid load.

Further prolongation of ventricular ERP and ARP with the volume challenge was observed in the dogs with CHF but not in control dogs, as shown in Figure 4. The anterior LV ERP increased from 177 ± 13 ms to 189 ± 14 ms ($p = 0.065$), and the ARP from 153 ± 12 ms to 169 ± 9 ms ($p < 0.005$) (Fig. 5). The posterior ERP prolonged from 161 ± 10 ms to 178 ± 5 ms ($p = 0.004$), and the ARP increased from 147 ± 6 ms to 162 ± 9 ms ($p = 0.014$). With fluid loading, the RV ERP increased to a lesser degree with volume loading—from 181 ± 11 ms to 192 ± 21 ms (p = NS)—and the ARP increased from 159 ± 14 ms to 172 ± 18 ms ($p = 0.098$) in dogs with CHF. No consistent change in the same variables was noted during volume loading in control dogs, although the anterior LV ERP increased from 128 ± 11 ms to 145 ± 10 ms. Dispersion of ventricular refractoriness was not altered.

MAP durations prolonged with volume loading in dogs with CHF but not in control dogs. In animals with adequate baseline and postvolume LV action potentials, a prolongation ≥20 ms with saline was seen in three dogs during pacing and in four during sinus rhythm. Representative action potentials are shown in Figure 6. Three additional animals showed MAP prolongation of at least 10 ms. Overall, the LV sinus rhythm MAP duration increased from 192 ± 10 ms to 222 ± 23 ms ($p = 0.012$) after fluid loading at equivalent heart rates (baseline 131 ± 10 beats/min vs. volume 125 ± 13 beats/min, p = NS). During pacing at a CL of 300 ms, the average MAP duration prolonged from 178 ± 15 ms to 196 ± 14 ms ($p = 0.047$). In contrast, only a trend toward MAP prolongation was seen at the RV site during sinus rhythm and pacing at a CL of 300 ms (Table 3). Quantitatively, MAP durations in control animals were not altered, but qualitatively showed a rapid

Table 2. Impact of Volume Alteration on Ventricular Refractoriness

	LV Ant		LV Post		RV	
	ERP (ms)	ARP (ms)	ERP (ms)	ARP (ms)	ERP (ms)	ARP (ms)
Control group (n = 6)						
Baseline	128 ± 11	117 ± 5	133 ± 9	120 ± 8	138 ± 7	114 ± 7
Volume	145 ± 10‖	121 ± 5	139 ± 11	120 ± 10	148 ± 15	124 ± 8
Nitroprusside	144 ± 12‖	123 ± 11	135 ± 13	122 ± 11	147 ± 7	122 ± 8
CHF group (n = 8)						
Baseline	177 ± 13†	153 ± 12†	161 ± 10*	147 ± 6*	181 ± 11†	159 ± 14†
Volume	189 ± 14§†	169 ± 9†#	178 ± 5†#	162 ± 9†¶	192 ± 21*	172 ± 18†‡
Nitroprusside	188 ± 11†	166 ± 13†‖	180 ± 6*#	160 ± 8*‖	188 ± 16*	168 ± 17†

Comparisons made between the control and paced groups: *$p \leq 0.001$; †$p < 0.0001$. Comparisons made within the control or paced groups: ‡$p = 0.098$; §$p = 0.065$ compared with baseline; ‖$p < 0.05$ compared with baseline; ¶$p = 0.0141$ compared with baseline; #$p < 0.01$ compared with baseline. ARP = absolute refractory period; CHF = congestive heart failure; ERP = effective refractory period; LV Ant = left ventricular anterior recording site; LV Post = left ventricular posterior site; RV = right ventricular outflow tract recording site.

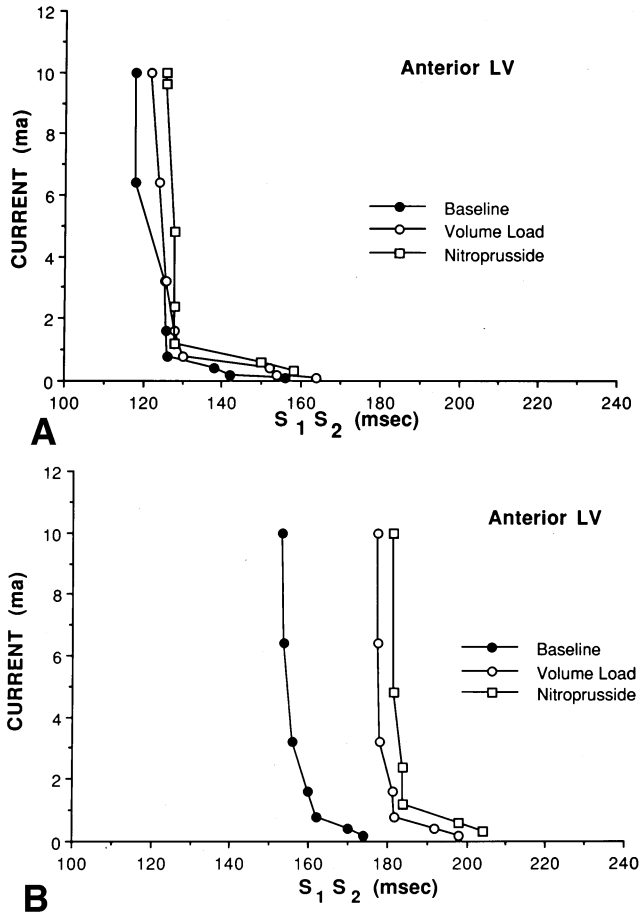


Figure 2. Strength–interval relationship and response to volume interventions. **A,** The anterior LV strength–interval relationship from a representative control animal showed no change with volume loading or subsequent sodium nitroprusside infusion. **B,** The strength–interval relationship in this cardiomyopathic dog revealed marked prolongation of both the ERP and ARP recorded at the anterior LV site with volume loading, although nitroprusside had little effect.

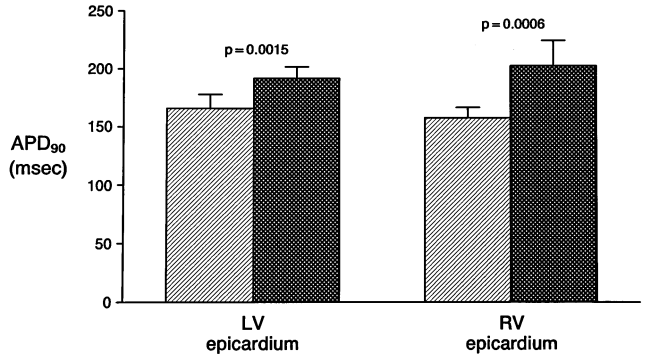


Figure 3. Impact of development of dilated cardiomyopathy on action potential duration at 90% repolarization (APD_{90}). The APD_{90} values in the paced animals (**crosshatched bars**) developing dilated cardiomyopathy were significantly longer at the LV and RV epicardial sites than APD_{90} values in control animals (**hatched bars**).

phase I repolarization yielding a spike-and-dome configuration without any effect on volume loading. This configuration was only infrequently observed in cardiomyopathic dogs.

These animals showed a marked sensitivity to sodium nitroprusside. A low dose infusion of this agent resulted in marked reductions in the systemic and intracardiac pressures and returned the cardiac output back to baseline values. This preload and afterload reduction had little impact on the ventricular ERP, ARP, MAP duration or dispersion of refractoriness in dogs with CHF and control dogs, as shown in Figures 4 and 5.

Conduction time with heart failure. Conduction time from the proximal to distal electrodes in control animals at the LV anterior site was 18 ± 3 ms longitudinal and 29 ± 6 ms transverse to myocardial fiber orientation. Similarly, conduction times in cardiomyopathic animals were 17 ± 6 ms and 24 ± 6 ms in the longitudinal and transverse directions, respectively. Volume administration had no effect on RV or LV conduction in directions perpendicular or parallel to fiber orientation at any site.

Ventricular arrhythmias in cardiomyopathic dogs. In control and study animals, initial 24-h ambulatory monitoring showed rare ventricular ectopy. One dog had frequent premature ventricular complexes and nonsustained ventricular tachycardia. After 4 weeks of pacing, five dogs had ventricular ectopy, including nonsustained tachyarrhythmia in one. No animal had spontaneous, sustained ventricular tachycardia or fibrillation during reevaluation. With the introduction of four extrastimuli, ventricular tachycardia or fibrillation was induced in one cardiomyopathic dog and five control dogs with the aggressive pacing protocol.

Pathologic findings. The hearts of the paced dogs showed marked four-chamber dilation. The mean LV and RV volumes were 63 ± 24 ml and 72 ± 15 ml in study dogs, respectively, whereas the comparable volumes were 25 ± 14 ml and 44 ± 14 ml in control dogs ($p < 0.01$). Compared with the control dogs, the posterior LV wall (9 ± 2 vs. 15 ± 3 mm, $p < 0.001$) and the interventricular septum thickness (10 ± 1 vs. 14 ± 1 mm, $p < 0.001$) were significantly reduced in the study group, although the average weight of the cardiomyopathic hearts was similar to that of the control hearts (208 ± 16 g vs. 194 ± 16 g, p = NS). On histologic examination, myocyte length and nuclear size were qualitatively increased in the cardiomyopathic animals. Mid-myocardial and epicardial regions were otherwise unchanged from control animals. In the subendocardial region, areas of fibrosis, myocyte vacuolization and areas of granulation tissue were present to a mild to moderate degree. Occasional macrophages and round cell infiltration were present.

Discussion

These results document the presence of significant electrophysiologic abnormalities in dogs with clinical, hemodynamic, neurohumoral and pathologic manifestations of dilated cardiomyopathy. Specifically, consistent prolongation of ventricular

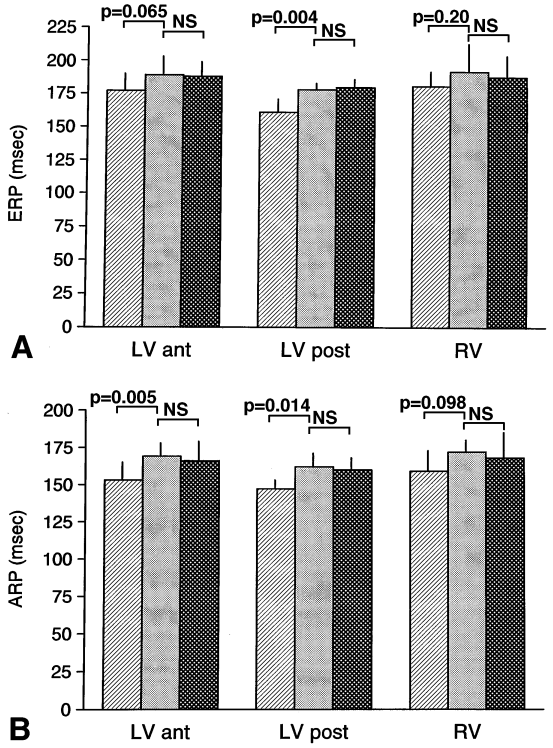
Table 3. Impact of Volume Alteration on Epicardial Monophasic Action Potential Duration at 90% Repolarization

	LV Ant		RV	
	NSR	PCL 300 ms	NSR	PCL 300 ms
Control group (n = 6)				
Baseline	166 ± 12	159 ± 21	158 ± 9	165 ± 20
Fluid	170 ± 13	176 ± 12	165 ± 9	172 ± 10
Nitroprusside	170 ± 21	178 ± 15	172 ± 14‖	165 ± 24
CHF group (n = 8)				
Baseline	192 ± 10‡	178 ± 15	202 ± 21§	192 ± 17†
Fluid	222 ± 23¶§	196 ± 14‖*	213 ± 34‡	193 ± 16†
Nitroprusside	217 ± 26‖‡	199 ± 20‖	232 ± 27‡	199 ± 12‡

Comparisons made between control and paced groups: *$p < 0.05$; †$p < 0.02$; ‡$p < 0.01$; §$p < 0.001$. Comparisons made within control or paced groups: ‖$p < 0.05$ compared with baseline; ¶$p < 0.012$ compared with baseline. NSR = normal sinus rhythm; PCL = pacing cycle length; other abbreviations as in Table 2.

repolarization times and refractoriness were present in both RV and LV regions, without evidence of substantive dispersion of refractoriness. Local anisotropic conduction patterns were unaltered. Volume loading further prolonged refractoriness and shifted the strength–interval relation in the direction of longer S_1S_2 coupling intervals in this model.

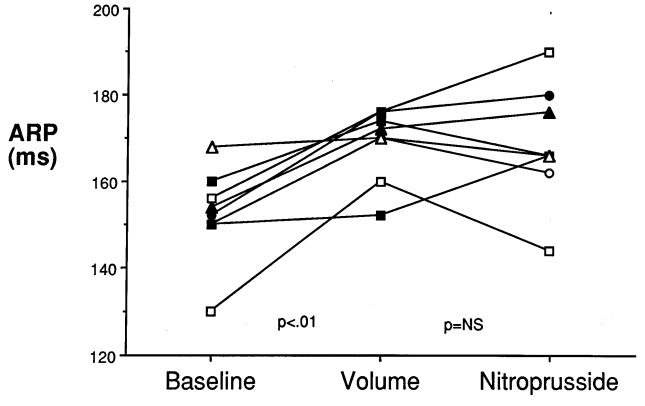
Figure 4. Effective (ERP) and absolute (ARP) refractoriness alteration with volume loading in cardiomyopathic dogs. **A,** The increase in the ERP at each of the recording sites with administration of the 800-ml volume **(gray bars)**. In contrast, no relevant change in refractoriness was noted with nitroprusside **(crosshatched bars)**. **B,** Similar prolongation of the ARP at each of the recording sites in the paced dogs. **Hatched bars** = baseline.



Prolongation of refractoriness with cardiomyopathy. Our in vivo findings are an important extension of previous in vitro observations. Several groups have documented action potential prolongation occurring with the development of CHF or hypertrophy (20–27). These data also confirm the presence of prolonged ventricular refractoriness in a similar canine pacing-induced cardiomyopathy model (14). The mechanism of such prolongation of repolarization and refractoriness may be related to 1) significant reductions in the density of inwardly rectifying potassium currents (I_{k1}) (21); 2) a decrease in transient outward current (I_{to}) (21,23,27); or 3) abnormalities of calcium currents, including slowing of inactivation with a net increase in inward ion flux (28–30).

It is less likely that the electrophysiologic changes observed in the presence of heart failure resulted from accompanying neurohumoral or paracrine abnormalities. Although several investigators have demonstrated an $alpha_1$-adrenergic mediated prolongation of the cardiac action potential in syncytial myocardial preparations (31,32) related to decreases in I_{to} (33–35), I_{k1} (36) or I_{kach} (37), other investigators have failed to

Figure 5. Effect of volume loading and nitroprusside load reduction on absolute refractoriness (ARP) in eight dogs with CHF. With volume loading, the ARP increased significantly but was unchanged by load reduction.



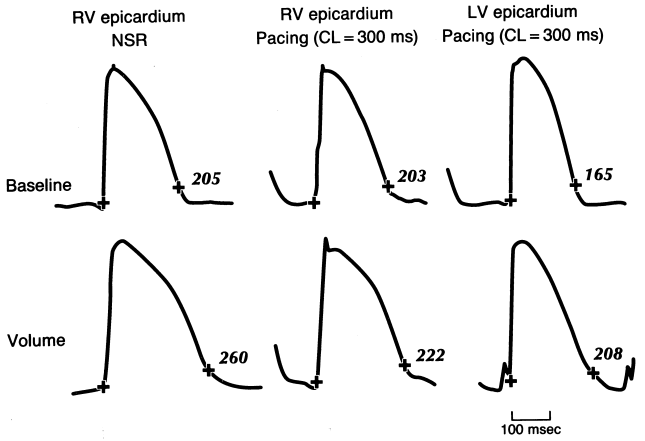


Figure 6. Effect of volume loading on MAP duration in a sample cardiomyopathic dog. APD_{90} from the RV epicardial site during sinus rhythm and from both RV and LV sites during pacing at a CL of 300 ms is shown. With volume administration, the APDs prolonged appreciably. NSR = normal sinus rhythm.

observe comparable changes in refractoriness in the intact canine heart (37,38), or have seen decreases in action potential duration with $alpha_1$ receptor agonists (39,40).

The effects of elevated angiotensin II, aldosterone or natriuretic peptides on repolarization in CHF are less clear. I_{kr} may be modified by angiotensin II (41) or its mediator, protein kinase C (42,43), although these effects appear to be modest. Whether such effects offset the increase in I_{ks} also seen with angiotensin II provocation (41) is unknown. Opposite increases in outgoing calcium, activated potassium currents (44,45) and decreases in I_{to} (46) due to natriuretic peptides further suggests that a neurohumoral mechanism of repolarization prolongation is unlikely.

Impact of volume loading in dilated cardiomyopathy. The significant prolongation of ERP or MAP duration with additional volume in the presence of preexisting global ventricular dysfunction has not been described previously, and appears to be opposite that seen in normal canine myocardium. Although Franz et al. (19) demonstrated an increase in late phase MAP duration with volume administered to normal canine hearts, predominantly due to an increase in the magnitude and duration of afterdepolarizations, these findings are at variance with other studies. Decreases in refractoriness with acute volume dilation have been observed in isolated normal rabbit (11) and canine heart models (12), at the discontinuation of cardiopulmonary bypass with increasing heart volumes (47) and after balloon valvuloplasty for congenital pulmonary stenosis (48). Similarly, Reiter et al. (13) observed a decrease in refractoriness accompanied by an overall increase in dispersion of refractoriness in residually normal tissue in Langendorff-perfused rabbit hearts.

The acute decrease in refractoriness in these studies or absence of MAP duration change with increases in end-diastolic volumes in these studies (8–10) was seen in largely normal preparations. Consistent with this, we saw only insignificant changes in refractoriness and MAP duration with volume administered to control dogs. Conceivably, a greater decline in these values might have been present with larger infused volumes producing higher filling pressures. In contrast, our consistent findings of an increase in refractoriness or repolarization times occurred in the myopathic setting, accompanied by significantly prolonged refractoriness and repolarization *before* volume dilation. In each dog with CHF, marked chamber enlargement presumably produced greater myofilament stretch than might occur with smaller volume loads in normal tissue. Enhanced RV accommodation of the volume load may have blunted the electrophysiologic changes in RV but not LV tissue. It is also possible that the differences between studies could be a function of the rate of volume dilation. In previous studies, experimental methods allowed instantaneous changes in LV volumes (8,11,12,24) with continued dilation in normal isolated, perfused hearts (11). In the present study, the 800-ml volume was delivered over 20 min.

The mechanism responsible for this further prolongation of MAP duration or refractoriness is unclear. Further alterations of I_{to} or I_{k1} through a feedback mechanism, triggering of a stretch-activated channel, or further autonomic or neurohumoral modulation are possibilities. It is also possible that the increase in blood pressure observed with volume loading produced sympathetic withdrawal, leading to lengthening of tissue refractoriness and MAP duration. Given the hemodynamics of the paced dogs, no attempt was made to test this possibility by blocking beta-adrenergic receptors.

Modulation of refractoriness with load reduction. In this study, sodium nitroprusside did not normalize ventricular refractoriness, despite complete ventricular unloading, as indicated by both systemic and end-diastolic pressure decreases to a level at or below baseline. This is in contrast to a previously reported increase in ARP in response to nitroprusside in normal porcine hearts subjected to acute aortic cross clamping (3). It remains consistent with observations of Hansen (49), who found no measurable effect of extreme changes in afterload on MAP duration in an isolated heart model where preload and afterload were controlled independently. In turn, this lack of a consistent electrophysiologic effect of load reduction may explain the absence of beneficial effects of oral load-reducing agents on sudden death mortality in patients with chronic CHF, although reductions in ventricular ectopy have been reported.

Spontaneous and induced ventricular tachyarrhythmias. There was no increase in spontaneous arrhythmia occurrence with additional volume administration in this model of CHF. Similarly, programmed stimulation after volume loading and nitroprusside administration failed to induce ventricular arrhythmias in the cardiomyopathic dogs. These findings are compatible with both animal and human studies. Franz et al. (50) demonstrated that although membrane depolarization could be produced by both gradual and rapid ventricular stretch, ventricular ectopy was typically elicited by rapid stretch, due to faster and larger volume changes. Hansen et al. (10) also found that the probability of extrasystole occurrence was a function of the magnitude of volume augmentation

during diastole. These effects were blocked by gadolinium but not calcium channel antagonists (50), consistent with arrhythmia mediation through a stretch-activated mechanism. Thus, the absence of appreciable arrhythmias in our animals could be related to "subthreshold" volume expansion, the slower rate of volume change or the absence of accompanying cellular hypertrophy. Furthermore, these data are consistent with low arrhythmia inducibility rates in patients with dilated cardiomyopathies, which along with the increased repolarization times and absence of relevant dispersions of refractoriness with such volume shifts, are against but do not exclude a reentrant mechanism for ventricular arrhythmias in this cardiomyopathic state.

Consistent with this is the increased prevalence of early afterdepolarizations in an in vivo canine model of left ventricular hypertrophy (51), with acute volume loading in the rabbit heart (52) and with substantial increases in afterload (49). In addition, Pogwizd (52) recently delineated a focal, nonreentrant mechanism for ectopy observed in a similar dilated myopathic setting. Although these data suggest the presence of the necessary cellular substrate for the occurrence of triggered automaticity or stretch-activated depolarizations as a mechanism of sudden death in dilated cardiomyopathic states, additional studies will be required for confirmation.

Study limitations. Several limitations should be borne in mind when interpreting these data. First, as was the case in previous studies (11,14), the volume load was monitored only in terms of changes in hemodynamic variables such as end-diastolic pressures. Nevertheless, ventricular dilation was evident in each case. The specific changes in ventricular systolic or diastolic volumes and their relation to electrical variables or the occurrence of arrhythmia are not available in this in situ model.

This study was designed to examine changes in local electrophysiologic properties occurring with the generation of cardiomyopathy and subsequent volume loading. To avoid previously reported animal demise precluding data acquisition (14), programmed stimulation was only performed after the administration of volume and nitroprusside. It is possible that the low inducibility rate was a consequence of volume loading or that load reduction had a beneficial effect on the inducibility of ventricular arrhythmias. Furthermore, it is possible that assessment from more epicardial sites would have detected relevant dispersions of refractoriness.

Clinical implications. Despite these limitations, the significant prolongation of effective and absolute refractoriness and repolarization in this CHF model has several potential clinical implications. First, these data suggest the potential for further electrical instability occurring with more acute hemodynamic decompensation, as frequently observed in patients with chronic CHF. Second, the infrequent occurrence of inducible ventricular arrhythmias, the absence of detectable dispersions of effective or absolute refractoriness and the significant prolongation of refractoriness in parallel to the increase in MAP duration decrease the likelihood of reentrant arrhythmias in the cardiomyopathic setting. Finally, these data provide early insight into the increased incidence of serious arrhythmias in patients with CHF receiving action potential–prolonging antiarrhythmic therapy (53,54). Further action potential prolongation, whether altering the same or different ion channels modified by underlying pathology, could aggravate preexisting repolarization abnormalities and more readily lead to torsade de pointes. Obviously, additional studies will be required to confirm this hypothesis.

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