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Asynchrony of ventricular activation affects magnitude and timing of fiber stretch in late-activated regions of the canine heart

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Abstract

Abnormal electrical activation of the left ventricle results in mechanical dyssynchrony, which is in part characterized by early stretch of late-activated myofibers. To describe the pattern of deformation during “prestretch” and gain insight into its causes and sequelae, we implanted midwall and transmural arrays of radiopaque markers into the left ventricular anterolateral wall of open-chest, isoflurane-anesthetized, adult mongrel dogs. Biplane cineradiography (125 Hz) was used to determine the time course of two- and three-dimensional strains while pacing from a remote, posterior wall site. Strain maps were generated as a function of time. Electrical activation was assessed with bipolar electrodes. Posterior wall pacing generated prestretch at the measurement site, which peaked 44 ms after local electrical activation. Overall magnitudes and transmural gradients of strain were reduced when compared with passive inflation. Fiber stretch was larger at aortic valve opening compared with end diastole ($P < 0.05$). Fiber stretch at aortic valve opening was weakly but significantly correlated with local activation time ($r^2 = 0.319$, $P < 0.001$). With a short atrioventricular delay, fiber lengths were not significantly different at the time of aortic valve opening during ventricular pacing compared with atrial pacing. However, ejection strain did significantly increase ($P < 0.05$). We conclude that the majority of fiber stretch occurs after local electrical activation and mitral valve closure and is different from passive inflation. The increased shortening of these regions appears to be because of a reduced afterload rather than an effect of length-dependent activation in this preparation.

Keywords

ventricular pacing; mechanical activation; fiber stretch; dyssynchrony

Ventricular epicardial activation results in a heterogeneous pattern of contraction such that the timing of the onset of local shortening mirrors the spread of the electrical depolarization wave front. As the region near the pacing site shortens, remote late-activated regions lengthen. Previous studies have observed this lengthening in late-activated regions, which has been termed “prestretch” by several investigators (2, 6, 16). These studies have used a wide variety of techniques, including ultrasonic crystals (2), surface markers (16), and magnetic resonance imaging (MRI; see Ref. 17) to quantify the prestretch. Most of these studies have shown augmented ejection shortening at late-activated sites relative to early activated sites, implying that prestretch may improve performance via muscle fiber length-dependent activation. However, they did not determine if prestretch is different from passive

distension or assess its timing relative to ventricular pressure generation. Moreover, none have examined the transmural gradient in prestretch or muscle fiber length changes in the prestretched, late-activated areas.

Thus the aim of the present study was to test the hypothesis that the late-activated regions of the left ventricle distend in the same manner as passive filling, and that the functional consequences of prestretch are the result of changes in length-dependent activation. To achieve this, midwall and transmural arrays of radiopaque markers and bipolar electrodes were implanted in the left ventricular (LV) free wall of open-chest dogs to describe myofiber deformation at high temporal and spatial resolution during atrial and ventricular activation.

In this study we show that, in contrast to passive distension, prestretch is uniform transmurally. The majority of prestretch occurs during isovolumic contraction, after the prestretched tissue has been electrically activated and the mitral valve has closed. Both of these findings support the conclusion that prestretch occurs in tissue that is actively generating force. We confirm that, at aortic valve opening, late-activated tissue is at a longer muscle length than early activated tissue. This dyskinesis is widely thought to produce the decrease in function observed in epicardial pacing. However, at least at short atrioventricular (A-V) delays, the muscle length in late-activated tissue is not significantly different from normal contraction, whereas early activated fibers are shorter. This not only provides insight into the decrease in function occurring with epicardial activation but also emphasizes the importance of A-V delay for clinical approaches such as cardiac resynchronization therapy.

MATERIALS AND METHODS

All animal studies were performed according to the National Institutes of Health *Guide for the Care and Use of Laboratory Animals*. All protocols were approved by the Animal Subjects Committee of the University of California, San Diego, which is accredited by the American Association for Accreditation of Laboratory Animal Care.

Surgical preparation

Ten adult mongrel dogs (21–24 kg) were anesthetized with intravenous propofol (6 mg/kg), intubated, and mechanically ventilated with isoflurane (0.5–2.5%), nitrous oxide (2 l/min), and medical oxygen (2 l/min) to maintain a surgical plane of anesthesia. Five dogs were implanted with midwall marker arrays, and five were implanted with transmural marker arrays. All underwent the same surgical procedure, as follows. The heart was exposed via a median sternotomy and a left thoracotomy at the fourth intercostal space and then placed in a pericardial cradle. An 8-Fr pigtail micromanometer catheter with a lumen (Millar Instruments, Houston, TX) was inserted through a 9-Fr arterial introducer placed in the left femoral artery, and the catheter tip was advanced into the LV. LV pressure was recorded with the pigtail micromanometer catheter, and the calibration was adjusted such that the end-diastolic pressure and peak LV pressure matched those recorded from a fluid-filled transducer connected to the lumen of the catheter. LV end-diastolic pressure (LVEDP) was calculated by finding $dP/dt_{\max}$ and working backward in time until dP/dt reached 5% of the maximum value. In the case of atrial pacing, this coincided closely with the notch in the pressure trace and the peak of the R-wave on the electrocardiogram (ECG). A flow probe (model #T208; Transonic Systems, Ithaca, NY) was placed on the ascending aorta to assess the onset and end of ejection. Stroke volume was obtained by integration of the aortic blood flow signal.

In one group of five dogs, radiopaque markers were used as follows to measure two-dimensional myocardial deformation in each heart. Lead beads (2 mm diameter) were

sutured to the apical dimple (apex bead) and on the epicardium at the bifurcation of the left anterior descending (LAD) and left circumflex coronary (base bead) arteries to provide end points for a LV long axis. Gold (0.9 mm) or lead (1.2 mm) beads were plunged in the anterolateral wall in a rectangular pattern, forming a surface approximately parallel to the epicardium (see Fig. 1).

Bipolar plunge electrodes were constructed by passing two 36-gauge dual-insulated magnet wires (Belden CDT, Richmond, IN) through the end of a 23-gauge needle and bending back the tips to form hooks. The wires were electrically isolated except at the tips, with interelectrode spacing varying between 1 and 5 mm, depending on how the hooks (and noninsulated wire tip) engaged the myocardium. Eight electrodes were evenly spaced around the edges of the array, and a ninth was placed in the center, all at the approximate depth of the beads (see Fig. 1). Bipolar electrodes were also used to pace the LV from the posterior wall epicardium, near the equator. Finally, a pair of pacing wires was sutured to the left atrium (LA).

Experimental protocol

Atrial pacing was performed by stimulating LA electrodes, and LV pacing was performed by stimulating both LA and LV electrodes (A-V delay = 35 ms), using a square-wave constant-voltage electronic stimulator (model SD9; Grass Instruments, Quincy, MA) at a frequency 20% above baseline heart rate to suppress native sinus rhythm. Stimulation parameters (voltage 10% above threshold, duration 8 ms, and frequency) were kept constant in each animal. Each animal was positioned in a biplane radiography system, and synchronous biplane cineradiographic images (125 frames/s) of the bead markers were digitally acquired with mechanical ventilation suspended at end expiration. The LV pressure, arterial pressure, surface ECG, and bipolar plunge electrograms were recorded simultaneously with the cineradiographic images. Recordings were obtained during atrial pacing and then following ~2–5 min of ventricular pacing (to achieve a steady hemodynamic state). Atrial pacing was continued between ventricular pacing runs.

Histology

After the functional data acquisition was complete, a 22- to 26-Fr cannula with side holes was inserted in the ascending aorta through a brachiocephalic artery, the animals were killed with pentobarbital sodium, and the heart was brought to arrest. The coronary arteries were perfused with normal saline, and the LV pressure was adjusted to the end-diastolic pressure observed during the study before fixing the heart with 2.5% buffered glutaraldehyde, as described previously (1). The fixed, Silastic-filled heart was skewered from the apical dimple to the mitral aspect of the aortic valve to define the LV long axis (18). It was then sectioned as shown in Fig. 2, yielding a total of ~25 transmural samples from within the bead array. The transmural thickness of the block was measured, and the block was sliced into 1-mm-thick sections parallel to the epicardial tangent plane, forming a series of sections from the epicardium to the endocardium used to measure the fiber angles across the LV wall. After measuring all of the angles in one transmural section, least-squares linear regression was used to fit the transmural data. This process was repeated in each of the samples, for use in the strain analysis described below.

Data analysis

Nine electrode pairs were used in each experiment, but an average of one per study was excluded because the time of activation could not be determined due to system noise or recording failure. Electrical activation time was defined as the time from the ventricular stimulus to the initial voltage deflection (see Fig. 3, dotted line). This definition was used

rather than other common definitions to remain consistent in the analysis of all electrograms, since some waveforms looked monophasic while others were biphasic.

Three-dimensional coordinates of markers were reconstructed from the cineradiographic images (1). The basic data analysis uses a homogenous strain calculation based on a triad of markers (13). This technique assumes a homogeneous distribution of strain within a triangle formed by three material markers (8). Myocardial markers were grouped automatically to form triangles with minimum height of 2.5 mm and maximum area of 60 mm². Additional triangles of larger area were then added manually in areas of low bead density to ensure coverage of the array. A single marker was often used in multiple adjacent triangles. These constraints resulted in ~50 triangles (functional data points) per array. The coordinate axes were defined individually in each triangle, using the following method. First, the outward-facing normal vector (local radial direction) of the triangle was determined. Next, the circumferential direction was determined by finding a vector perpendicular to both the radial direction and the LV long axis, pointing toward the septum. Finally, the longitudinal direction was determined by finding a vector perpendicular to the radial and circumferential directions, pointing toward the apex. This process ensures that the circumferential and longitudinal directions lie in the plane of the triangle (see Fig. 4).

Two-dimensional Lagrangian finite strains could then be calculated with respect to cardiac coordinates at the centroid of each triangle, as reported previously (13, 19). Strains were also transformed into fiber coordinates by rotating the strains by the fiber angle α at the location of that triangle's centroid. This angle was calculated as a weighted average of the three nearest measurements to the centroid (weighting inversely proportional to distance). Thus the fiber and cross-fiber strains within each triangle were found.

Three-dimensional triangle centroid locations were then projected on a two-dimensional grid such that the plane of the grid was perpendicular to the radial direction of an arbitrary triangle located near the center of the array. By representing all the strains from a single time point at their respective centroid locations on the grid, a spatial distribution of strain could then be fit using the Matlab function `griddata` (The Mathworks, Natick, MA) and visualized as a color map. Cubic interpolation was used to ensure smooth gradients between adjacent strain measurements.

Areas were considered prestretched if the maximum fiber strain before aortic valve opening (relative to the pacing stimulus) exceeded 0.02, the approximate resolution limit of the technique. Aortic valve opening strain was defined as the strain at the time of aortic valve opening during ventricular pacing with respect to that same hemodynamic reference during atrial pacing. This quantity was only defined in areas that were prestretched with ventricular pacing. Similarly, ejection strain was defined as the strain at the time when the aortic valve closes with respect to the time it opens, in the same areas used to define aortic valve opening strain.

Transmural arrays

A second group of five dogs was used to measure three-dimensional myocardial deformation. The same surgical preparation was used, except that the midwall bead array was replaced with three transmural columns of four to five 0.8-mm-diameter gold beads and a 1.7-mm-diameter surface gold bead above each column. The beads were placed within the anterior wall between the first and the second diagonal branches of the LAD. The experimental protocol was also the same, with the addition of epicardial pacing at the site of the array. The data analysis was performed by determining a continuous polynomial position field that mapped the beads in the undeformed reference configuration to those in the deformed configuration. Differentiation of this field with respect to undeformed position

yields a continuous, nonhomogeneous transmural distribution of three-dimensional finite strains (1, 5).

Statistical analysis

Values are means $\pm$ SD unless otherwise specified. A one-factor repeated-measures ANOVA was used to assess the effect of time during the cardiac cycle on fiber strain. Tukey's test was used for post hoc comparisons. For aortic valve opening strains, a one-sample *t*-test with two-tailed *P* value was used. For all other statistics, a paired *t*-test with two-tailed *P* value was used. Statistics were performed using Sigmaplot version 3.0 (SPSS, Chicago, IL). Significance was accepted at $P < 0.05$.

RESULTS

Midwall arrays

The results of this study are from five open-chest, isoflurane-anesthetized dogs with bead arrays implanted in the LV free wall. Table 1 summarizes the hemodynamic results. LVEDP was not significantly different following LV pacing. Peak LV pressure, average systolic pressure, stroke volume, $dP/dt_{\max}$, and $dP/dt_{\min}$ were all significantly reduced following ventricular pacing.

Bead depths and fiber orientation

Beads [25 ± 2 (range 23–28); gold, 0.9 mm or lead, 1.2 mm] were plunged to an average depth of 7.0 ± 1.6 mm ($64.3 \pm 14.6\%$ wall depth) spanning from 31.4 ± 5.0 to $81.9 \pm 3.5\%$ of the apex-base distance with a width of 5.0 ± 0.7 cm at the widest point. Myocardial markers were grouped to form triangles of area 41.6 ± 16.9 mm² (range 10.6–115.9 mm²). In all five animals, the number of triangles ranged from 37 to 59, with an average of 48 ± 10 triangles.

To understand the deformation data in the context of the fiber anatomy, fiber angles throughout the array were measured histologically. At the depth of the array, myofibers were found to be oriented at an average of $+27.5 \pm 22.1^\circ$ from circumferential, consistent with previous findings of fiber angle at this depth when compared at apical (1, 5) and basal sites (5). The average range of fiber angle within the array was $72.0 \pm 5.0^\circ$.

Timing of prestretch

Figure 3 shows a typical time course of fiber strain in one triangle and the bipolar electrogram of the nearest electrode. Fiber strain data from the whole array for this animal are shown in Fig. 5, where the location of the triangle shown in Fig. 3 is outlined. The strain maps shown in Fig. 5 were obtained at the times marked by vertical lines in Fig. 3. Referring to Fig. 3, 85% of the prestretch occurs between end diastole (dashed line) and aortic valve opening (solid line); it peaks 59 ms after the time of local activation.

Figure 6 shows the average values of strain components in areas that were prestretched. Fiber strain was significantly increased, with an average of 68% of total prestretch occurring during isovolumic contraction. Crossfiber strain and fiber-crossfiber shear strain were not significantly different. In each animal, peak prestretch occurred an average of 44 ± 23 ms after activation of the nearest bipolar electrode. The spatial distribution of prestretch varied from animal to animal; figure 5 shows a typical result. The majority of prestretch occurred along the septal border and was somewhat patchy in distribution.

Activation time vs. prestretch

Previous studies such as those performed with MRI (21) have shown that there appears to be a relationship between distance from the pacing site and midwall circumferential strain. We sought to quantify this regional pattern of stretch in terms of fiber function. As shown in Fig. 7, there was a general relationship between time of electrical activation and magnitude of maximum prestretch. Linear regression yielded a correlation coefficient $r^2 = 0.319$, with a slope of 1.2935/s, which was significantly different from zero ($P < 0.001$). The intercept, -0.0041 , was not significantly different from zero.

To determine if prestretch provides a functional advantage, we next addressed the question of whether fiber length at the time of aortic valve opening in prestretched areas was longer than at aortic valve opening during atrial activation. The bead configuration at the time of aortic valve opening during atrial activation is taken as the reference configuration, and the bead configuration at the time of aortic valve opening in ventricular pacing is taken as the deformed configuration. From the example in Fig. 8, there are negative aortic valve opening strains near the apex and slightly positive strains toward the base. As shown in Fig. 9A, on average there were no significant aortic valve opening strains with a short A-V delay. This is likely because of a shorter initial fiber length at the time of ventricular stimulation compared with atrial-paced end diastole (mean fiber strain = -0.058 ± 0.034 , $P < 0.05$). This suggests that, in this preparation, prestretched tissue provided no functional advantage relative to atrial pacing; however, fiber ejection strain was increased significantly as shown in Fig. 9B. Overall, stroke volume was reduced by ventricular pacing, suggesting a net decrease in ejection strain over the entire left ventricle. In addition, there was a relationship between magnitude of peak prestretch (PP) and ejection strain (EJS): $EJS = -0.6654 \times PP - 0.0541$ ($r^2 = 0.499$, $P < 0.001$). In light of the insignificant aortic valve opening strains, this functional improvement of prestretched tissue may instead be related to the 15.4% decrease in average systolic pressure observed in this preparation.

Transmural arrays

A second set of animals with transmural arrays was used to compare the pattern of three-dimensional deformation during prestretch with that of passive inflation. Strains were calculated at 2% ("epi"), 41% ("mid"), and 79% ("endo") of the wall depth from epicardium to compare with literature values. The magnitudes of the cardiac strain components at peak prestretch are listed in Table 2. Note the limited transmural variation. Peak prestretch occurred at 35.8 ± 37.5 mmHg, at a greater pressure (on average) than would occur as a result of a physiological passive inflation.

To assess the degree of asynchrony, we calculated aortic valve opening strains between early activated and late-activated tissue. Because simultaneous measurements at early and late-activated areas were not possible, the pacing stimulus was moved to vary the activation time of the measurement site. Pacing at the epicardium over the bead array resulted in early activation, whereas the normal posterior wall pacing location resulted in late activation. At the same depth as the midwall array, the fiber component of aortic valve opening strain was 0.080 ± 0.023 (wall depth = 64%, $P < 0.05$), suggesting that the fibers in the late-activated region are indeed longer than their early activated counterparts.

DISCUSSION

In this study, the mechanics of late-activated myocardium during ventricular pacing were investigated in an in situ canine heart preparation. Electrical activation and mechanics were measured in the late-activated region by use of midwall material markers that covered ~20–25% of the LV ($n = 5$) and transmural arrays ($n = 5$). The results show that prestretch mostly

occurs after time of end-diastolic pressure and differs from passive inflation. At short A-V delays, fiber length in prestretched regions was longer than early activated regions but not compared with atrial pacing, suggesting an overall decrease in preload that is consistent with a decrease in stroke volume. Ejection strain was increased, however, potentially as a secondary result of diminished pressure generation caused by the dyskinetic contraction.

Badke et al. (2) were among the first to recognize prestretch, which we and others have defined as tissue lengthening occurring before aortic valve opening. Since then, it has been studied by several investigators using various techniques such as epicardial video markers (6, 16), MRI (17, 20), radionuclide angiography (3, 4), tissue Doppler imaging (11), and finite element modeling (9). The hemodynamic effects of ventricular activation are well known, and the results in this study are similar to many early publications. LV systolic pressure was reduced, within the range of 12–25% found in most previous studies (2, 6, 14). A few studies showed no change in systolic pressure, but this was probably because of unmatched heart rate (15) or differences in anesthesia (16). LVEDP was reduced in at least one study (14), but LVEDP was unchanged in most studies of A-V sequential pacing, when there is a component of atrial filling (2, 6, 15). These studies also found a decrease in stroke volume of between 13 and 23%, consistent with the 18% decrease seen here. In the current study, $dp/dt_{\max}$ was reduced by 18%, also similar to these other studies (8–27%).

The time course of prestretch has been reported but, with lower temporal resolution (15, 16, 21), has not been expressed in terms of fiber direction (20, 21) and has not been described relative to phases in the cardiac cycle. Because activation is asynchronous, the pattern of regional contraction relative to LV pressure generation is altered. During normal activation, the majority of the ventricle is activated within 20–30 ms of end diastole. With ventricular pacing, however, some areas are activated much earlier than this (during late diastole), and some areas are activated much later (during isovolumic contraction). Compared with other studies of ventricular activation, the delay to reach the latest activated region in the present study was comparable, on the order of 100 ms (15, 16). In this study, we compared the magnitude of stretch at the time of activation, the end of diastole, and the time of aortic valve opening. An MRI study by Wyman et al. (20) showed that the prestretch begins in late diastole and continues through the isovolumic contraction phase, reaching magnitudes of as much as 20%. However, the time of end diastole was not available. Also, midwall circumferential strains were used, which may have a different time course than fiber strain, with significant regional variation. In studies with epicardial markers (15, 16), ~10% fiber stretch was seen during isovolumic contraction, the lower number most likely reflecting that late diastole was not included in those results. Our results clearly show that very little prestretch occurs before the time of ventricular end diastole, and perhaps more importantly when the tissue is lengthening it is also electrically depolarized and presumably capable of generating force.

From the current data, it is not possible to define the nature of the forces lengthening the prestretched tissue. However, it seems likely that the high intracavitary pressures during isovolumic systole play a major role in lengthening activated tissue. To determine if the three-dimensional deformation of prestretched tissues differs from passive inflation, we compared our transmural results with those of Omens and Cove11 (12). In those experiments, isolated, arrested canine hearts were passively inflated to a normalized volume change of 0.20 ml/g, corresponding to a pressure change from 0 to 10 mmHg. In the current experiments, the pressure changed from 3.9 mmHg (at stimulus) to 35.8 mmHg (at peak prestretch). Despite this larger pressure change, six of nine normal cardiac strain components were significantly smaller in magnitude, as shown in Fig. 10. The transmural gradients were also reduced in prestretch. For example, the gradient of radial strain (E33) was not significantly different from zero. Moreover, the transmural gradient during passive

inflation ($-0.088\% \text{ depth}^{-1}$) is >2 SD than during prestretch ($-0.010 \pm 0.033\% \text{ depth}^{-1}$), further emphasizing the difference in the mode of deformation. The reduced transmural gradients suggest that the activation sequence in these late-activated regions may progress transmurally from endocardium to epicardium, beginning the stiffening of these endocardial cells earlier and preventing them from stretching as much. In summary, the results of this study do not support the concept that prestretched tissue is passively lengthened in the same manner as diastolic filling of the ventricle.

Another possible mechanism of prestretch is that contracting early activated areas could directly stretch late-activated tissue. First, the stretch is predominately in the fiber direction (Fig. 6), whereas passive filling at this wall depth has a larger crossfiber component (12). Second, as time from the stimulus increases, the force developed by the early activated fibers increases and the total number of force-generating fibers also increases. Thus the overall deformation is likely the result of a combination of intracavitary pressure and fiber connectivity.

In the present study, peak lengthening in prestretched areas occurred 44 ms following local depolarization. There was an increased magnitude of prestretch that was roughly proportional to the delay in local activation time. It has been previously shown that the activation time correlates with the local ejection strain (9) and mechanical activation time (20), but to our knowledge no one has shown the relationship with magnitude of stretch.

The question of whether ejection strain is augmented in late-activated areas has received substantial attention. Badke et al. (2) showed that late-activated segments had unchanged or depressed ejection strain, but larger “actual” shortening (total shortening regardless of phase in the cardiac cycle) when there was no contribution of atrial contraction to filling. Prinzen et al. (16), using A-V sequential pacing with a 30-ms delay, showed that ejection strain was larger in the late-activated region than the same areas during atrial pacing. However, the 150% increase over atrial pacing is larger than the 32% increase seen in the present study, likely because of the hemodynamic differences between the two studies discussed above. Despite these differences in magnitude, an increase was seen in both studies. The mechanism of this increase is unknown. However, several authors have suggested there is a length-dependent activation effect because of increased fiber/sarcomere length in the prestretched areas (9, 15, 17).

To investigate this possibility, we compared the change in fiber length at aortic valve opening in prestretched areas with the same areas during atrial activation. At an A-V delay of 35 ms, there was no statistically significant change in fiber length (mean fiber aortic valve opening strain = 0.005). It therefore seems unlikely that length-dependent activation is responsible for the enhanced ejection strain in this preparation. In fact, of the four well-known regulators of ventricular performance (heart rate, preload, inotropic state, and afterload), only afterload changed significantly. Although average systolic pressure is not a true measurement of local tissue afterload, this may be the best explanation for the observed increase in ejection strain.

Although fiber length at aortic valve opening was not different from atrial pacing, there was still a difference in fiber length between early and late-activated regions. This reflects the heterogeneous distribution of stretch in the ventricle. Coupled with the aortic valve opening strain results, this shows a net reduction in overall LV preload that is at least partially responsible for diminished hemodynamic function.

Aortic valve opening strain, the change in muscle fiber length from atrial to ventricular pacing in prestretched regions, was not significantly different from zero. This can be explained by the preexcitation that occurs at short A-V delays. As a result, at the time of

ventricular stimulation, the fibers were shorter than at atrial-paced end diastole. We have anecdotal evidence in one dog with complete A-V block that shows that aortic valve opening strain increases with A-V delay, to a maximum of 100 ms. Despite this, the ejection strain was unchanged over this range of A-V delays. This likely reflects the balance between preload and afterload, both of which increase with A-V delay.

Study limitations

The present study was designed to investigate the magnitude and timing of stretch in the late-activated region of the ventricular paced canine LV. The approach has several limitations. First, beads were plunged into the myocardium with the use of a trocar, which likely causes some local injury. However, the overall deformation of the array region is unlikely to be affected by small areas of injury. No evidence of ischemia was seen in the ECG in any of the animals. Second, the interpretation of these data was made with the assumption that all the beads formed a surface at a fixed transmural depth, while in reality there was a small variation in bead depth. Third, although the equations to calculate strain are exact, there are errors involved in bead digitization (the resolution of the radiographic system is ~0.02 mm), as well as the three-dimensional reconstruction from the two-dimensional images. Fourth, the measured fiber orientations were registered with the strain data based on bead locations, which may have produced some inaccuracies. Averaging of the three closest fiber angle measurements presumably reduced these errors. Additionally, changes in segment length along the fiber direction were used for all fiber length comparisons rather than any direct measure of fiber or sarcomere length. Last, with the current technique, simultaneous strain measurements of the entire ventricle are not possible. Therefore, it is possible that the location of maximum prestretch is not encompassed by the array.

In this study, we used a fixed A-V delay of 35 ms. Because the experiments were performed in the presence of a functioning A-V node, a relatively low A-V delay was chosen to ensure that the activation wave front spread from the pacing site and activated the ventricles exclusive of native conduction. In a similar model, Faris et al. (7) showed that such fusion of activation sources did not occur at A-V delays <60 ms. However, this choice of delay likely results in suboptimal filling and a reduction of end-diastolic volume compared with atrial pacing (10).

In conclusion, during LV pacing, the mechanics of the late-activated tissue was studied using midwall and transmural marker arrays in the canine left ventricle. In late-activated tissue, the majority of prestretch was found to occur during isovolumic systole, probably because of a combination of intracavitary pressure and series and parallel force generation by early activated fibers. The deformation was different from passive inflation, reflecting the lengthening of already electrically activated tissue that resulted in reduced transmural gradients of strain. At short A-V delays, muscle fiber length in prestretched areas was unchanged compared with atrial activation, whereas fiber length in early activated areas was reduced. Although ejection shortening in late-activated areas was increased compared with atrial activation likely due to a decrease in afterload, overall ejection shortening (as reflected by stroke volume) was reduced due presumably to dyskinesis and to an overall reduction in muscle length.

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Fig. 1.

The bead array consisted of alternating gold and lead beads to simplify the matching of points in the anteroposterior (AP) and lateral (LAT) radiographic views. Beads were plunged into the midwall, ~60% through the transmural thickness. The array spanned longitudinally from the apex to the first diagonal of the left anterior descending artery (LAD) and circumferentially from the LAD to the posterior descending artery (data not shown). Circles with two straight lines protruding indicate location of bipolar electrodes for electrical activation recordings, which were at the same depth as the heads.

Fig. 2.

Histological procedure for measuring fiber angle. **A:** the heart was skewered along the long axis and cut into 5 or 6 rings perpendicular to this axis. **B:** each ring was then further sectioned into blocks in the area of the beads. **C:** each block was sliced into 1-mm-thick sections cut perpendicular to the epicardial tangent plane. Fiber angle (α) was determined under a dissection microscope with reference to the positive circumferential axis.

Fig. 3.

Left ventricular (LV) pressure, fiber strain in one triangle, and nearest bipolar electrogram plotted as a function of time during ventricular pacing for *dog 1*. Time = 0 represents the ventricular pacing stimulus, and the data depict a full cardiac cycle. Vertical lines indicate time of local activation (dotted), end diastole (dashed), and opening of the aortic valve (solid).

Fig. 4. Schematic representation of one triangle, illustrating the coordinate directions. X_c and X_l the circumferential and longitudinal direction in terms of cardiac coordinates. X'_c and X'_l represent the equivalent coordinate directions in terms of a local triangle coordinate system, used in strain calculations. Projections of X'_c and X'_l align with X_c and X_l .

Fig. 5. Spatial distribution of fiber strain in *dog 1* at the following 3 time points: average time of activation (ACT), end diastole (ED), and aortic valve opening (AO). Red (positive) strains indicate stretching, whereas blue (negative) strains indicate shortening. X's represent locations of beads. The outlined triangle has the time course shown in Fig. 3. B, base; A, apex; S, septum; L, lateral.

Fig. 6.

Average values of strain components in prestretched areas. Fiber strain (E_{ff}) at aortic valve opening was significantly larger than at the time of local activation or end diastole; crossfiber strain (E_{cc}) and fiber-crossfiber shear strain (E_{fc}) were not significantly different. $P < 0.05$ vs. local activation (*) and vs. end diastole (†).

Fig. 7. Maximum regional fiber strain vs. the local activation time of that region. All regions with a quantifiable electrogram were used, for a total of 33 points.

Fig. 8.

Example of aortic valve opening strains in fiber coordinates. All strain components are homogeneous and small.

Fig. 9.

A: aortic valve opening strains in prestretched regions, average of 5 animals. A value of zero indicates no change between atrial and ventricular pacing. None of the strain components were significantly different at the time of aortic valve opening during ventricular pacing compared with atrial pacing. Statistical comparisons were made with a 1-sample *t*-test against a hypothesized mean of zero. *B*: ejection strains, average of 5 animals. E_{ff} and E_{cc} are enhanced by ventricular pacing. E_{fc} was not significantly different. * $P < 0.05$.

Fig. 10.

Cardiac strains during prestretch (“Current”) and passive inflation (“JO 1991”). Average pressure changes were from 3.9 to 35.8 mmHg and 0 to 10 mmHg, respectively. Strain magnitudes were reduced relative to passive inflation, and transmural gradients were reduced or eliminated. E11, circumferential strain; E22, longitudinal strain; E33, radial strain. * $P < 0.05$ vs. passive inflation at same depth (with Bonferroni correction).

Table 1

Hemodynamic results

	HR, beats/min	LVEDP, mmHg	LVP _{peak} , mmHg	+dP/dt _{max} , mmHg/s	-dP/dt _{max} , mmHg/s	Average Systolic Pressure, mmHg	Stroke Volume, ml
Atrial pacing	104.2±4.1	6.8±1.7	97.3±11.4	1776±505	-1387±343	87.7±10.1	16.5±3.1
LV pacing	104.1±4.1	6.2±4.5	84.2±9.0*	1451±350*	-1050±277*	74.3±9.3*	13.6±2.2*
Change, %	-0.1%	-8.8%	-13.4%	-18.3%	-24.3%	-15.4%	-18.0%

Values are means ± SD.

HR, heart rate; LVEDP, left ventricular end-diastolic pressure; LVP_{peak}, peak left ventricular pressure; LV, left ventricle.

* $P < 0.05$ vs. atrial pacing.

Table 2

Cardiac strains at peak prestretch

	Epi	Mid	Endo
E11	0.051±0.019	0.077±0.024	0.079±0.037
E22	0.029±0.002	0.040±0.013	0.058±0.040
E33	-0.071±0.034	-0.069±0.026	-0.065±0.028
E12	-0.007±0.022	0.016±0.016	0.015±0.008
E23	-0.012±0.028	-0.010±0.011	-0.007±0.014
E13	-0.035±0.011	-0.016±0.009	-0.002±0.031

Values are means ± SD.

Strains during prestretch are of lesser magnitude and exhibit less transmural variation than during passive inflation.

E11, circumferential strain; E22, longitudinal strain; E33, radial strain; E12, circumferential-longitudinal shear strain; E23, longitudinal- radial shear strain; E13, circumferential-radial shear strain.