Relation of Ultrastructure to Function in the Intact Heart: Sarcomere Structure Relative to Pressure Volume Curves of Intact Left Ventricles of Dog and Cat

By Henry M. Spotnitz, B.A., Edmund H. Sonnenblick, M.D., and David Spiro, M.D., Ph.D.

The sarcomere is the basic ultrastructural unit of contraction in striated muscle. Huxley and Hanson have shown that the alternating band patterns of the sarcomere in skeletal muscle derive from the longitudinal disposition of two sets of interdigitating contractile protein filaments. On the basis of these and other studies, the sliding filament hypothesis for muscle contraction was proposed. The ultrastructure of heart muscle has also been examined and its sarcomeres shown to be similar in structure to those of skeletal muscle. More recently sarcomere structure has been studied in relation to the length-tension curve of both skeletal and cardiac muscle, and despite previous objections, it has been demonstrated that the sliding filament hypothesis explains the band pattern changes at varying muscle lengths as well as the relation between active tension and muscle length for both types of striated muscle. The relevance, however, of sarcomere length-tension relations for linear samples of myocardium (papillary muscle) to physiological performance of the intact left ventricle has not been reported. The purpose of these experiments was, therefore, to examine sarcomere length as a function of filling pressure and volume under passive conditions in the intact mammalian left ventricle.

Methods

Fresh hearts were used after removal from 27 dogs and 10 cats anesthetized with sodium pentobarbital (25 mg/kg) and sacrificed acutely. The mitral orifice of the hearts from dogs was sealed with a clamp, and the right ventricle was opened widely. A Gregg cannula was inserted into the left main coronary artery and the aorta was sealed around the proximal portion of the cannula. For studies including the range of normal left ventricular end diastolic pressure (0 to 15 mm Hg), two cannulas with plastic guards to prevent leakage were introduced through the interventricular septum into the cavity of the left ventricle. Intraventricular pressure was measured with a Statham P23G pressure transducer attached to one cannula and was recorded on a Sanborn oscillograph. The second cannula was utilized for the introduction and withdrawal of fluid increments. When passive intraventricular pressures beyond the physiological range were to be investigated (15 to 55 mm Hg), the canine ventricles were prepared with an intraventricular balloon attached to a large bore cannula which was placed through and sealed into the mitral valve orifice. The balloon employed was large so that stretching of its wall and development of tension were avoided over the volumes explored. Four or five curves relating pressure and volume were obtained for each left ventricle to ensure reproducibility. Satisfactory ventricular sealing, demonstrated by the quantitative recovery of the fluid introduced into the chamber, was established as a criterion for the acceptability of each preparation. These procedures were completed within fifteen to thirty minutes after the excision of the heart from the animal.

Fixation for electron microscopy followed ad-
justment of intraventricular conditions to a desired position on the pressure-volume curve. The preparation was perfused with fixative through the previously cannulated left coronary artery, employing glutaraldehyde, 6.25%, in phosphate buffer, pH 7.6. Previous studies have shown that this fixative does not change resting muscle tension and does not significantly alter filament dimensions or sarcomere length. The ventricles from cats, after similar preparation, were fixed by immersion and replacement of the intraventricular fluid volume with fixative. A fluid exchange device, consisting of a pair of matched syringes placed back-to-back, assured that the fluid removed from the ventricle was replaced simultaneously with an identical volume of glutaraldehyde. Fixation continued for three hours.

The left ventricular weight was then determined after removing the atria at the atrioventricular groove and separating the entire free wall of the right ventricle. Slices of the left ventricular wall were obtained and washed in cold phosphate buffer for 24 hours. With the aid of a dissecting microscope, these slices were trimmed to thin plates, less than 0.5 mm in thickness, cut parallel to the planes containing the muscle fibers, and fixed for an additional three hours in buffered osmium tetroxide. Tissues were then dehydrated in alcohol and embedded in Araldite. Details of the process of fixation and tissue preparation for electron microscopy have been described elsewhere. Care was taken in sectioning by orienting muscle fiber direction parallel to the knife edge to avoid compression artifacts. Postfixation staining of thin sections with potassium permanganate or lead citrate was used to heighten contrast. Left ventricles from both the dog and the cat were sampled from the inner, middle, and outer muscle layers in the region of the anterior descending branch of the perfused left coronary artery. In addition, portions of the interventricular septum and papillary muscles were obtained from the left ventricles of cat heart.

Thin sections were examined in a carefully calibrated RCA EMU-3 electron microscope operated at a single magnification (tap 6) with care to normalize microscope lenses at 50 kv. Five or six micrographs at an initial magnification of 8,000 were obtained for each sample, with about forty sarcomeres per field. In the several samples from each of the 37 hearts, sarcomere structure was examined and the average sarcomere length and band widths were determined, employing 25 or 30 measurements per sample. Sarcomere length was taken as the center-to-center distance between adjacent Z-lines. The variation in sarcomere length between adjacent fields was generally less than 0.05 μ. The data were treated statistically and sarcomere length was plotted relative to ventricular pressure and volume. Phase contrast microscopy of thick sections embedded in Araldite and optical microscopy of hematoxylin and eosin sections were also employed as an aid to tissue orientation and as a check for sampling errors.

### Results

Data were obtained for 27 canine left ventricles averaging 96.4 ± 2.6 (se) g in weight from dogs weighing an average of 17.1 kg (range: 13 to 28 kg). The combined weight of right and left ventricles in this series averaged 125 g.

A representative example of a passive pressure-volume curve for the canine left ventricle is illustrated in figure 1. A filling pressure of 0 mm Hg corresponds in this instance to an intraventricular volume of 12 cc. Intraventricular pressure rises at an increasing rate as a function of filling volume, reaching 12 mm Hg with an intraventricular volume of 47 cc. Further increases in volume are associated with gradual stiffening of the ventricular wall.
such that small increments in volume cause increasingly marked pressure elevations. As illustrated, negative pressure is required to remove all fluid from the ventricle. Average intraventricular volume at a pressure of 0 mm Hg for the 27 canine ventricles was 12.5 ± 0.9 (se) cc. At a pressure level of 12 mm Hg, intraventricular volume averaged 53 ± 3.6 cc (fig. 2 and table 1).

Figure 3 shows the appearance of a typical low-power field for the canine left ventricle as observed in the electron microscope. The excellent preservation of all cytoplasmic structures and uniform distribution of sarcomere lengths, with dilatation of the capillaries, suggest that perfusion of the coronary arteries with glutaraldehyde produces rapid, uniform fixation. The overall tissue preservation was excellent. This has been found to hold true even in hearts fixed as long as one hour after sacrifice of the animal.

The relationship of passive filling pressure to sarcomere length for the middle or sino-spiral layer of myocardium in the canine left ventricle is illustrated in figure 4. Each of the twenty-seven points is derived from a separate heart, and the best approximate curve is drawn between these points. Average sarcomere length increases gradually from 1.92 ± 0.05 μ at an intraventricular pressure of 0 mm Hg (fig. 5) to 2.25 ± 0.04 μ at 12 mm Hg (fig. 6). Beyond this point, sarcomere length changes less with increasing pressure. Thus, at an intraventricular pressure of 35 mm Hg, observed sarcomere length approximates 2.35 μ. Some scatter of experimental data relative to the plotted line in figure 4 exists at all filling pressures, but decreases with higher pressures, as indicated in table 2. Figures 7

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**TABLE 1**

<table>
<thead>
<tr>
<th>P* (mm Hg)</th>
<th>Vol. (cc)</th>
<th>R1 (mm)</th>
<th>R2 (mm)</th>
<th>R3 (mm)</th>
<th>h (mm)</th>
<th>R1'</th>
<th>R2'</th>
<th>R3'</th>
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*P: transmural pressure in mm Hg. Vol: intraventricular volume, cc. R1: calculated radius to inner surface of ventricular wall, mm. R3: calculated radius to outer surface of ventricular wall, mm. R3: calculated mean radius, mm. h: calculated wall thickness, mm.

†SE of mean.

‡Indicates estimated value.

Mean animal weight 17.1 kg, mean left ventricular weight 96.4 g.
FIGURE 3

Survey electron micrograph of inner layer of canine left ventricle fixed at 1 mm Hg filling pressure and filling volume of 24 cc. All cellular components are well preserved and the capillaries are widely patent as a result of the perfusion fixation. Sarcomeres measure 2.1 μ in length. Paired arrows at the level of the Z line delimit a sarcomere. Nuclei are indicated by N and capillaries by C.

and 8 show similar relationships of sarcomere length to filling pressure in the outer (subepicardial) layer and the inner (subendocardial) layer. Sarcomere length appears to change most in relation to intraventricular pressure or volume in the innermost regions of the ventricular wall (fig. 9). Over the range studied in the canine left ventricles, sarcomeres average 0.04 μ longer in the inner layer of myocardium than in the middle layer, and these in turn average 0.05 μ longer than those in the outer layer. Applying statistical analysis for significance of paired data, t values of 1.94 and 2.25 support these differences in sarcomere length as significant at 0.07 and 0.04 levels respectively.

Figure 10 relates sarcomere length to intraventricular volume; the values correspond to those plotted in figure 4 for sarcomere length related to intraventricular pressure. The dashed line in figure 10 represents for the “middle layer” of the wall, the theoretical relation of the changes in volume in a sphere of uniform wall thickness to the changes in circumference. The weight of the wall in the model is taken to be 96.4 g with a density of 1.00 g/cc. Calculated data for ventricular radii and wall thickness, used in preparing the theoretical plot, are presented in table 1. The theoretical and experimental curves diverge clearly in the region encompassing sarcomere lengths greater than 2.2 μ. Sarcomere length changes being smaller in this range than anticipated from the theoretical curve (fig. 10). For sarcomere lengths less than 2.2 μ and intraventricular volumes less than 40 cc. the observed and theoretical curves are correlated more closely. Despite efforts to
empty the left ventricle completely, and despite resulting negative intraventricular pressures, sarcomeres less than 1.85 \( \mu \) in length have not been observed generally in the non-activated ventricle.

The sarcomere lengths observed at varying filling pressures in the ventricles from cats are illustrated in figure 11. In the ten cats studied, left ventricular weight averaged 6.4 g. Intraventricular volume at 10 mm Hg averaged 4.0 cc. Despite a more than tenfold difference in ventricular volume and weight, a similar curve relating sarcomere length to left ventricular filling pressure pertains for both the cat and the dog. In both species sarcomere lengths of 2.2 \( \mu \) are associated with filling pressures of 10 to 12 mm Hg.

Sarcomere band pattern changes are a function of sarcomere length in the myocardium of both the cat and dog. While A-band width remains constant at 1.5 \( \mu \), I-band width varies linearly with the length of the sarcomere (figs. 5 and 6). A central dark band termed the M-line traverses the A-band and is closely flanked by two light areas termed the L (or para-M)-lines. This M-L complex is constant in width at all sarcomere lengths. In sarcomeres less than 2.2 to 2.0 \( \mu \) in length, however, the L (or para-M)-lines are less clearly defined (fig. 5) than those observed in longer sarcomeres (fig. 6). In sarcomeres measuring 2.30 to 2.40 \( \mu \), H zones may or may not be present (figs. 12, 13, 14). However, in sarcomeres measuring more than 2.4 \( \mu \), H zones are generally seen. Reference to figures 4 to 6 indicates that sarcomere lengths associated with the appearance of H zones are observed with ventricular filling pressures in excess of 15 to 20 mm Hg. The distribution of data in figure 15 indicates generally that H-zone width increases with sarcomere length.

**Discussion**

The pressure-volume curves as presented here for left ventricles from dogs resemble those previously reported. The present methods have allowed the delineation of reproducible passive pressure-volume curves with fixation of each ventricle for electron microscopic analysis at a known point along its curve. Reproducibility is indicated by the superimposition of successive curves from the same heart and the quantitative recovery of added fluid. The constancy of observed data also indicates that under the conditions described heart muscle does not develop.
rigor in the first half hour after removal from the animal, consistent with findings reported by others. Stress relaxation was found to be a factor in pressure-volume relations of the left ventricle only if filling pressure exceeded 15 mm Hg. Even in this circumstance, a tendency to return to the original curve was observed if the ventricle was allowed to recover at a lower pressure-volume point.

Previous studies of the isolated cat papillary muscle have shown that sarcomere length, band patterns, and actively developed tension are functions of overall muscle length and are in accord with a sliding filament model for muscle contraction. A reproducible curve for sarcomere length vs. active tension has been derived from these data. Active tension is zero at sarcomere lengths of approximately 2.9 and rises along the ascending portion of the curve to a maximum at initial sarcomere lengths of 2.18 to 2.24 μ. With further increments in muscle and sarcomere length, the descending limb of the active tension curve appears, and developed tension declines. Also, in accordance with the sliding filament model, progressive penetration of thin actin filaments...
into the M-L complex occurs at sarcomere lengths less than 2.2 to 2.0 \( \mu \), with the appearance of A-contraction bands due to a double overlap of thin filaments at sarcomere lengths of less than 1.8 \( \mu \).

The present results, including an A-band of constant width and I-band width proportional to sarcomere length, are in full accord with the sliding mechanism. The relative darkening of the L (or para-M)-lines at the shortest sarcomere lengths observed in this study, again reflects the passage of thin actin filaments into the center of the sarcomere. However, no A-contraction bands were observed, since sarcomeres shorter than 1.85 \( \mu \) were not encountered. Further evidence for the sliding model is afforded by the presence of H zones\(^{22, 21}\) which are consistently observed in sarcomeres measuring more than 2.4 \( \mu \). These H zones are created by the progressive withdrawal of thin actin filaments from the A-band at long sarcomere lengths, but it is difficult to ascertain a linear relationship between H-zone width and sarcomere length. There is no simple explanation for the fact that H zones are not consistently present.
Experimental volume vs. sarcomere length curve for the middle layer of the canine left ventricle (solid line) is compared to a theoretical curve for the middle layer of a spherical model of total mass identical to the average for the canine left ventricles studied (dashed line, see Table 1). Theoretical curve is chosen to intersect the experimental curve at a point where sarcomere length is 1.9 μ and filling volume is 12 cc.

In figure 11, the relation of sarcomere length to actively developed tension in the cat papillary muscle is correlated with the curve for sarcomere length vs. filling pressure for the middle layer of the left ventricle of both the cat and the dog. The upper limit of normal for left ventricular end diastolic pressure in the intact heart is approximately 12 mm Hg. This corresponds to a sarcomere length of 2.2 μ in the middle layer of the ventricular wall for both animals. The same sarcomere length also corresponds to the apex of the active length-tension curve of the isolated papillary muscle. Thus, the tension developed by the left ventricle during active contraction (systole) increases with increased end diastolic filling pressure and reaches a maximum as ventricular filling pressure approximates the upper limit of the normal range. These observations serve to support the view that the normal left ventricle functions only along the ascending portion of the length tension curve, when end diastolic sarcomere lengths measure 2.2 μ or less.27

The fundamental importance of the relation of sarcomere length to passive left ventricular filling pressure is reflected in the constancy of this relation for the dog and the cat, despite a tenfold difference in ventricular mass and volume. A mathematical basis for the generality of this relationship, independent of absolute chamber size, is given by a modified Laplace equation, which expresses intramural stress as a function of intraventricular pressures.

In figure 11, curve of active tension vs. sarcomere length for isolated cat papillary muscle is superimposed on the curve of left ventricular pressure vs. sarcomere length incorporating data obtained for the intact left ventricle of both the dog and the cat. The apex of the active length-tension curve for papillary muscle corresponds to a sarcomere length of 2.2 μ, a sarcomere length observed as well in the middle layer with a filling pressure of 12 mm Hg in the intact left ventricle of both animals.
pressure. If the ventricle is treated as an elastic sphere of uniform wall thickness, tangential stress is related to transmural filling pressure by this modified formula:

$$S_t = \frac{RP}{2h}$$  \hspace{1cm} (1)

where $S_t$ is tangential stress (force per unit area), $R$ is the mean radius of the ventricle, $P$ is transmural pressure, and $h$ is wall thickness. For any two given left ventricles, regardless of absolute size, tangential stresses in the wall will be approximately equal at a
Relation of sarcomere length to H zone width in sarcomeres from canine left ventricles filled to high intraventricular pressures. Dashed lines indicate the anticipated relation suggested by the sliding filament hypothesis with thin actin filaments measuring 0.9, 1.0, or 1.1 μ in length.

Given transmural pressure if the ratio of radius to wall thickness is a constant. Thus, if $R/h = k$, equation 1 reduces to

$$S_t = \frac{1}{2}kP = k'P \quad (2)$$

Therefore, if the relative dimensions of the left ventricle are all increased or decreased proportionately in hearts of varying sizes, sarcomere length will remain a constant function of filling pressure, provided that the passive sarcomere length-resting tension curve for elements of the myocardium is unchanged. A more exact treatment has been given by Timoshenko for elastic spheres of uniform wall thickness,

$$S_t = \frac{(Pa^3)}{(2R^3)} \cdot \frac{(2R^3 + b^3)}{(b^3 - a^3)} \quad (3)$$

where $a$ is the radius to the inside of the wall, $b$ is the radius to the outside of the wall. Other variables are as described above. This expression reaches a maximum when $R = a$, indicating that the distribution of stress is not uniform, increasing to a maximum at the inner surface of the chamber wall and decreasing toward the outer surface.

In line with this analysis, the present study has shown that at a given ventricular filling pressure sarcomeres are longest in the inner layers of the left ventricular wall, decreasing in length toward the more superficial layers. This trend, confirmed by statistical methods, is suggested as well by the superimposition of

Normalized fixation volume vs. sarcomere length. Relation of sarcomere length in the middle layer of the canine left ventricle to fixation volume normalized as a decimal fraction of the filling volume for each preparation corresponding to an intraventricular pressure of 10 mm Hg. Several cubic functions which intersect the experimental curve at arbitrary points are illustrated. A cubic function describes the relation of circumference (c) changes to volume (v) changes in a sphere or in a symmetrically expanding ellipsoid of revolution or cylinder if sections are examined in planes perpendicular to the long axis. Marked divergence between the slopes of the theoretical and experimental curves is evident at large filling volumes. A similar divergence is shown in figure 11.
tracings in figure 9. This gradient, which is particularly evident at elevated filling pressures, reflects the maximization of tangential stresses at the inner surface of the ventricular wall (equation 3). It also reflects the geometric relation noted by others\textsuperscript{30-32} that any given volume change results in maximal relative changes in circumference in those regions of the wall with the shortest radius (table 1).

The present experimental findings are consistent with the concept that the ratio of ventricular radius to wall thickness \((R/h)\) in individual left ventricles is adjusted to produce a relation of sarcomere length to transmural pressure independent of absolute size of the ventricle. As a result, transmural filling pressure represents a more general and reproducible index of sarcomere length (figs. 4 and 11) than either absolute or normalized filling volume (figs. 10 and 16). This finding, consistent with the Laplace equation, is even more noteworthy in the face of deviations from theory. Thus, structural factors, such as the content and disposition of connective tissue in the chamber wall, dictate that normal ventricles of similar size and weight may have dissimilar pressure-volume relations (fig. 17), while sarcomere lengths remain comparable at comparable filling pressures. A further complication is introduced by the true shape of the left ventricle which is complex and is not given accurately by any simple geometric model, spherical or otherwise.\textsuperscript{33-36} Nonetheless, experiments have indicated that variation in wall thickness of similar size and weight may have dissimilar pressure-volume relations (fig. 17), while sarcomere lengths remain comparable at comparable filling pressures. The relation of cross-sectional stresses to sarcomere length in the wall of the intact ventricle is more complex than that observed in papillary muscle. Resting tension in papillary muscle approaches zero at sarcomere lengths of 1.5 \(\mu\). The structure of the left ventricular wall, however, is such that sarcomere lengths of 1.87 to 1.95 \(\mu\) are observed in the various muscle layers (figs. 4, 5, 7, 8) when intraventricular pressure (and, therefore, tangential wall stress) is zero. The difference between papillary muscle and the intact ventricle is, in all probability, a consequence of differences in three-dimensional structure. In the papillary muscle, principal fiber direction is parallel to the long axis of the muscle while the left ventricular wall is an intricate system in which the long axes of fibers in the various layers are arranged obliquely with respect to one another.\textsuperscript{15,16} Thus, while forces sufficient to extend sarcomeres beyond 1.5 \(\mu\) may be exerted on individual contractile
units, the three dimensional arrangement of muscle fibers in the intact wall results in cancellation of such forces, resultant overall tension summatiing to zero.

This suggested stressing of the ventricular wall is in accord with the observation reported above, that even when negative pressures are employed to extract all fluid from the left ventricular cavity, sarcomeres shorter than 1.85 μ are not observed in any of the principal layers of myocardium. The observed rate of change of sarcomere length relative to passive filling volume in approaching this lower limit of sarcomere length, moreover, is not theoretically adequate for ejection of physiological stroke volumes from the actively contracting canine left ventricle, as indicated in table 1. Indeed, observations in beating hearts that have been fixed in systole have shown that sarcomere lengths as short as 1.5 to 1.6 μ may be observed in the wall of the actively contracting ventricle. With the augmented tension available in activated muscle, the lower limit of sarcomere length is shorter than that observed under passive conditions. Therefore, the general shape of the curve relating sarcomere length to intraventricular volume differs for activated and nonactivated heart muscle. In fact, the geometry of active systolic contraction has been found to be significantly different from that of diastolic filling. Also consistent with this concept are the findings of Hort and Linzbach who have utilized optical interferometric methods and observed, in many species, very short sarcomeres in hearts during rigor. Hort has reported for preparations of the canine left ventricle in rigor (average residual volume 6 cc) that sarcomere length averages 1.6 μ in the middle layer. Locker previously had reported the observation of varying degrees of sarcomere shortening with the onset of rigor in skeletal muscle.

The ventricular model employed in evaluation of the present results is defined in table 1 as a simple sphere of uniform wall thickness. For purposes of calculation, the wall was assumed to displace a volume of 96.4 cc, equivalent to the average weight of the canine ventricles used in the present study. In addition to predicting the expected degree of change in sarcomere length for any given volume change, the model illustrates clearly the very different degrees of shortening required in the various myocardial layers. Thus, a stroke volume of 23 cc, representing 43% of an end diastolic volume of 53 cc would require changes in circumferences of 17, 10, and 5.5% in the inner, middle, and outer layers, respectively. Experimentally observed changes in external cardiac dimensions during systole have thus been smaller than predicted in calculations neglecting the effect of the thickness of the left ventricular wall. The wall thicknesses calculated for the model agree well with experimental data. Calculated thicknesses of 9.6 mm at 53 cc left ventricular volume, and 11.8 mm at 30 cc volume, may be compared to experimental values of 8.4 mm and 10.3 mm recently reported for minimal diastolic and maximal systolic wall thickness respectively in the canine left ventricle. In weighing the left ventricles employed in this study, nonmural musculature (trabeculae carneae, papillary muscle) was not distinguished from mural myocardium. Wall thickness, as calculated for the model is augmented by this additional nonmural muscle, and this accounts partially for differences between theoretical and experimental values for mean wall thickness.

The use of a spherical model is adequate for the approximate evaluation required here and is subject to relatively simple mathematical treatment. Others have found that an ellipsoid of revolution is a more accurate description of the shape of the left ventricle. Symmetrical expansion of such a model for the left ventricle would lead to the same relation between changes of circumference and volume as that derived from the spherical model, when considering sections including fibers of the middle layer which form circular rings.
of muscle in planes perpendicular to the long axis of the ventricle.\textsuperscript{15, 16} The same relation obtains also when considering the ventricle as a cylinder, again allowing symmetrical expansion of all dimensions and considering only cross sections including the “ring fibers” of the middle layer. As has been noted, systolic contraction involves, in all likelihood, a somewhat different geometric relationship.\textsuperscript{35}

Figure 10 shows that the experimental curve for sarcomere length vs. intraventricular volume diverges from a theoretical curve, based on the spherical model defined in table 1, for changes in ventricular circumference relative to chamber volume. Figure 16 illustrates that this divergence is not decreased significantly by normalizing the volume data in order to compensate for variations in absolute ventricular size. The use of alternate theoretical curves, similarly, has little effect. Confirmation for the observation of relatively short sarcomers, 2.3 to 2.4 μ in length, in overdistended left ventricles is available in the work of Hort.\textsuperscript{16}

Certain relevant complexities of left ventricular geometry must be considered. The presence of papillary muscles and trabeculae carneae in the ventricular lumen reduces effective space\textsuperscript{33, 45} and may make volume changes appear disproportionately large. Thus, if 8 cc of the chamber volume were occupied by incompressible muscle, a twofold expansion in chamber size from 10 to 20 cc would appear to be a sixfold change from 2 to 12 cc. A similar effect would appear were collapse and buckling of the inner region of the ventricular wall to occur, converting smooth concentric layers of sarcomeres to involuted and serrated rings. Such infoldings at the inner surface of the ventricle can reduce the effective circumference without corresponding decreases in sarcomere length. Another form of collapse, appearing as a transition in shape from an ellipsoid to a sphere, similarly would change internal volume but not circumference. A minor degree of such transition of ventricular form from the ellipsoidal toward the spherical has been noted in passive filling.\textsuperscript{32} The significance of these mechanisms, however, in the divergence of theoretical and experimental curves for sarcomere length vs. intraventricular volume is questionable. This divergence is most prominent at filling volumes of such great magnitude (greater than 53 cc at 12 mm Hg pressure for left ventricles in the present study) that the relative volume occupied by nonmural musculature becomes insignificant, and the importance of wall collapse, infoldings, or changes in ventricular shape appears similarly reduced. At smaller chamber volumes, however, these mechanisms may become effective, serving in varying degrees to permit maximum ventricular emptying with minimal changes in sarcomere length.

The observation that changes of sarcomere length are apparently incommensurate with linear changes in the dimensions of the ventricular wall resembles observations in studies of the sarcomere length-tension curve for papillary muscle.\textsuperscript{9} These studies have shown that high intramuscular tension is required to extend sarcomeres beyond 2.3 μ and that under conditions of such high stress expansion of sarcomeres ceases to be proportional to changes in overall muscle length. The association of this finding with high levels of stress in both the papillary muscle and the intact ventricular wall is consistent with possible structural distortion resulting in relative slippage of myocardial elements. As an analogy to one type of distortion considered for the intact ventricle, if a large coil spring is twisted back upon itself, the circumference, and thus the enclosed volume, will increase while the length of wire forming the spring remains unchanged. In this connection, gross torsional motion of the left ventricular wall during overdistention has not been observed, although a few degrees of such motion have been noted for the left ventricle of the beating heart during systole.\textsuperscript{35, 39} Furthermore, major rearrangements of shell-like masses of myocardium in the left ventricle have been opposed on the basis of observations recently reported by Hort.\textsuperscript{16} Thus it would appear that structural distortion, if occurring, can be observed only at microscopic levels. The failure to observe marked changes in the con-
figuration of the intercalated disc under load suggests that within a given myocardial fiber, at least, relative sarcomere position remains constant. Slippage of columns of fibers, however, might account for the apparent failure of sarcomere expansion. Thus, while gross rearrangement of whole shells of muscle is unlikely in the intact ventricle, slippage of columns of fibers within those shells, relative to one another, may occur.

The finding that the apex of the sarcomere length-tension curve is reached with ventricular transmural pressures of 10 to 15 mm Hg, and its relevance to the Frank-Starling principle, has been discussed recently. Ventricular filling pressures significantly greater than 15 mm Hg cause sarcomeres to expand beyond 2.3 μ resulting in partial withdrawal of actin filaments from the A-bands with the appearance of an H zone. This places the myocardium at a relative disadvantage by forcing it onto the descending limb of the active length-tension curve, and may contribute to myocardial failure.

Consideration of the modified Laplace formula (equation 1) reveals the effect of pathological hypertrophy or ventricular dilatation on the relations discussed above. In the hypertrophied heart, abnormally high filling pressures may exist without extending sarcomeres to the point at which myocardial performance is compromised. But in the dilated ventricle, even ordinary filling pressures may extend sarcomeres beyond normal limits.

Summary
Reproducible pressure-volume curves have been obtained from fresh left ventricles removed from 10 cats and 27 dogs. Electron microscopic observations of sarcomere length and structure in the walls of these chambers have been correlated with the parameters of passive filling. In both the dog and the cat a similar relation of sarcomere length to filling pressure is observed, a filling pressure of 12 mm Hg corresponding to a sarcomere length of 2.2 μ. Sarcomeres shorter than 1.85 μ are not observed under conditions of passive filling. Sarcomeres tend to be longest in the inner layer of the ventricular wall. Filling pressure is observed to be a more general and reproducible index of sarcomere length than absolute or normalized filling volume. H zones are often present in the sarcomeres of the ventricular wall with filling pressures greater than 15 mm Hg and sarcomeres greater than 2.3 μ in length. These findings are discussed in relation to previous studies of papillary muscle and in relation to mathematical models for the left ventricle. The present results indicate that the normal left ventricle functions along the ascending portion of the length-tension curve, where the end diastolic sarcomere lengths are 2.2 μ or less.

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CARDIAC ULTRASTRUCTURE AND FUNCTION


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Henry M. Spotnitz, Edmund H. Sonnenblick and David Spiro

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