Recurrent sustained ventricular tachycardia: structure and ultrastructure of subendocardial regions in which tachycardia originates

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ABSTRACT Surgical resection of the endocardium and subendocardium often abolishes chronic recurrent sustained ventricular tachycardia in patients with healed myocardial infarcts or ventricular aneurysms, presumably by interrupting the reentrant pathway. To define the morphologic characteristics of cells in the reentrant pathway, we studied the histology and ultrastructure of the endocardial resections of 23 patients who underwent this procedure. Bundles of apparently viable myocardial fibers embedded in dense fibrous tissue were identified throughout the endocardial resections from all patients. These bundles of cells were separated from one another by fibrous tissue but extended uninterrupted to the margins of the surgical resection. In 14 patients Purkinje fibers were identified beneath the thickened endocardium whereas the remaining bundles were composed of ventricular muscle. The Purkinje fibers appeared to have normal ultrastructure and ventricular cells with both normal and abnormal ultrastructures were present. The abnormal muscle cells were characterized by loss of contractile elements, aggregates of dilated sarcoplasmic reticulum, and osmiophilic dense bodies. The sarcolemma was intact and the nuclear chromatin was evenly dispersed suggesting that these cells were still viable. The abnormal structure and arrangement of the surviving cardiac fibers in the endocardium may cause the abnormal electrophysiologic function that results in ventricular tachycardia.


CHRONIC recurrent ventricular tachycardia may occur in patients with healed or healing myocardial infarcts and ventricular aneurysms.1-2 Results of ventricular mapping studies indicate that these tachycardias often originate in the subendocardium of the infarcted region adjacent to the aneurysm.3-6 Surgical resection of the endocardium and subendocardium around the aneurysm may prevent tachycardia whereas aneurysctomy alone often does not.7-10

The fact that tachycardia is prevented by surgical resection of subendocardial regions suggests that cardiac fibers causing the arrhythmia are transected or removed by this procedure. It is assumed that these fibers have abnormal morphologic and electrophysiologic characteristics that are somehow related to the myocardial infarct, or perhaps to chronic ischemia, and that may be instrumental in causing tachycardia. We therefore have investigated the structure and ultrastructure of subendocardial regions, the excision of which abolished chronic recurrent ventricular tachycardia, and describe in this report the appearance of the cardiac fibers that may cause this arrhythmia.11

Methods

Clinical characteristics of patients. Twenty-three patients with a history of chronic, recurrent, medically refractory ventricular tachycardia who underwent surgery for arrhythmia are included in this study. The patients ranged in age from 39 to 69 years (mean age 58 years) and included 22 men and one woman. Each patient had a documented myocardial infarct 1 month to 15 years before surgery (mean interval 70 months) and 17 had ventricular aneurysms (table 1).

Before surgery each patient underwent a complete electrophysiologic study in the catheterization laboratory, including programmed ventricular stimulation and catheter endocardial mapping, as previously described.5 In all patients sustained
TABLE 1
Clinical and pathologic findings

<table>
<thead>
<tr>
<th>Patient No.</th>
<th>Agea</th>
<th>Sex</th>
<th>Intervalb</th>
<th>Sitesc</th>
<th>Resectiond</th>
<th>Aneurysme</th>
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<td>5</td>
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<td>6</td>
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Group 1 = patients without tachycardia after surgery; group 2 = patients with inducible tachycardia controlled with drugs after surgery; group 3 = patients with recurring tachycardia after surgery; ant pap = anterior papillary muscle; Deg = degenerative changes in ventricular muscle; inf pap = inferior papillary muscle; PF = Purkinje fibers; VM = ventricular muscle.

aAt the time of surgery.
bBetween myocardial infarct and surgery.
cOf earliest activation of arrhythmia. Numbers indicate location either adjacent to numbered site or between numbered sites (see figure 1).
dAnatomic location of endocardial resection and aneurysm.
eStructural findings of the endocardial resection.
fHistologic and ultrastructural study. The regions of endo-
cardium and subendocardium that were resected measured approximately 7 to 15 cm² and were 1 to 3 mm thick. In 14 of the patients two or more regions of excised endocardium were studied (table 1). These specimens were either from different sites of earliest activation that were separated by regions activated later, or were from large endocardial resections that were divided at the time of surgery. A total of 40 specimens were studied. Sutures were placed in the endocardial resections at the time of surgery and their sites approximated those of earliest activation so that these sites could be identified after the tissue was fixed. Sections were cut from the endocardial resections for histologic study and studies of ultrastructure both at sites marked by sutures and at sites distant from the regions of early activation so that the structure at both locations could be compared.

Both the endocardial resections and aneurysms from 17 of the patients were fixed “en bloc” in 10% formalin for a week or longer. Areas of about 1 cm² around the sutures were then cut from the endocardial resections and processed for electron microscopic examination. Other 1 cm² regions of the resections that were unrelated to early activation were also prepared for electron microscopic examination, as were 1 cm² subendocardial areas of the aneurysms. The tissue for electron microscopic study was divided into rows of uniform blocks (1 to 2 mm²) with a razor blade. The blocks were not separated at the endocardial surface so that they retained their spatial relationship to one another until they were embedded. The formalin-fixed tissue was rinsed twice with 0.1M phosphate buffer, pH 7.4, for 2 hr and then fixed in 2.5% phosphate-buffered glutaraldehyde. Tissue from six other patients was immediately fixed at the time of surgery in the 2.5% phosphate-buffered glutaraldehyde (pH 7.4) and this tissue included that from sites of early activation as well as sites not activated early. To obtain good preservation, areas of both the endocardial resections and aneurysms were divided before fixation into rows of uniform blocks of 1 to 2 mm², as described above. After fixation in 1% phosphate-buffered OsO₄ and dehydration in graded acetones, all tissue blocks were completely separated from one another by cutting through the endocardial surface. They were then embedded in Swiss araldite. The location of each block in relation to the suture marking earliest activation and to each other was carefully noted. During embedding the blocks were oriented so that the surface for sectioning was perpendicular to the endocardium. Fifty to 100 blocks were processed from each 1 cm² specimen obtained for electron microscopic study. Thin sections were cut with a diamond knife, stained with acetate and lead citrate, and studied with a Philips 300D electron microscope at 80 kV.

In order to assess cell size, thick sections (0.5 μ) were also cut from selected blocks and stained with toluidine blue. Measurements were made with a micrometer fitted to the eyepiece of a binocular microscope that was calibrated with a standard micrometer mounted on a glass slide. Cell diameters were measured at the nuclear level of longitudinally sectioned cells. Cell length was measured between the two most distinct segments of the intercalated discs in longitudinally sectioned cells. The amount of endocardial connective tissue and the width of bundles of myocardial cells were also measured with the micrometer.

Specimens for histologic study were taken from areas adjacent to the tissue processed for electron microscopy in all cases. Blocks were obtained from both the endocardial resections and the aneurysms and were refixed overnight in 10% neutral-buffered formaldehyde. The blocks for the histologic study included the entire length of the specimen. After paraffin embedding 8 μ sections were stained with hematoxylin-phloxine-safran and Masson’s trichrome.

Results

Histology. In general, 50% to 90% of each resection removed from the endocardial surface comprised non-muscular tissue including collagen bundles, fibroblasts, macrophages, mast cells, and collapsed blood vessels (figure 2). By far the majority of each resection was made up of collagen. The endocardium of all resections was also thickened, measuring from 90 to 1100 μ.

One or more bundles of myocardial cells were embedded in the dense connective tissue of every resection that we examined, both immediately beneath the endocardium and deep to the endocardium (figure 2). The bundles of myocardial cells extended the entire length of the endocardial resection in every patient. These muscle bundles varied in thickness and arrangement. In all patients there were one to four bundles less than 100 μ in width, each consisting of fewer than 10 cells across the diameter. When more than one bundle of this kind was observed, they were separated completely by connective tissue. In 17 patients there were also thick bundles (>100 μ) that comprised more than 10 cells (range 10 to 100 cells) across their diameter. Thick bundles were also separated from each other and from the thin bundles by connective tissue. Thick and thin bundles were randomly distributed throughout the
thickness of the endocardial resection and connections between bundles were not observed.

The cells within both thick and thin bundles were of various diameters and lengths and were arranged in several different ways. There were no apparent differences in cell size, shape, or arrangement between thick and thin bundles. The diameter of the cells ranged from 3 to 62 μ at the level of the nucleus and their lengths varied from under 100 to about 200 μ even within a single bundle. Some bundles (figure 3, A) consisted of cylindrically shaped cells of varying size that were closely apposed to each other along their length (tightly packed bundles). Other bundles (figure 3, B) were composed of spindle-shaped cells that were widely separated from each other by connective tissue (loosely packed bundles). Cells in these loosely packed bundles were connected to one another by thin cytoplasmic extensions.

It was apparent on light microscopic examination that various degrees of degenerative changes were present in many of the muscle fibers, in both the loosely packed and tightly packed bundles. Although many cells appeared normal and were filled with myofibrils with distinct striations (figure 3, A), other cells of varying size and shape were vacuolated and had large inclusions (figure 3, B). These abnormalities were present in individual cells within bundles of normal-appearing cells or throughout all the cells of a bundle and are described in more detail in the section on ultrastructure.

There were no noticeable differences between the histologic characteristics of endocardial resections from the patients in whom surgery prevented the occurrence of tachycardia and of those from the patients in whom surgery was not successful (table 1). Similarly, there were no histologic differences between sites adjacent to or far removed from the sutures marking the sites of earliest activation.

The amount of connective tissue, the arrangement of muscle bundles, and the histologic appearance of car-

FIGURE 2. The endocardial resections consisted of dense connective tissue (c) within which were embedded bundles of myocardial cells that were widely separated from one another (arrows). The bundles of myocardial cells were of varying size and arrangement. In some bundles the myocardial cells were separated by abundant connective tissue (open arrow) while in other bundles the cells were tightly packed together (solid arrow). The endocardium (E) of the resections was thickened. (Masson's trichrome stain, original magnification × 94.)
closely apposed cells with irregular shapes were found embedded in dense connective tissue on the epicardial surface.

**Ultrastructure.** The ultrastructures of the endocardial resections at marked sites of earliest activation and at sites distant from regions of early activation were similar. Purkinje fibers and ventricular muscle cells were found at both locations.

**Purkinje fibers.** Cells that had ultrastructural features of Purkinje fibers were found immediately beneath the endocardium in the endocardial resections from 14 patients (table 1). Although the ultrastructures of Purkinje fibers from postmortem specimens have previously been reported there are no ultrastructural studies on well-fixed peripheral Purkinje fibers in normal human hearts. Therefore, our conclusion that the cells in the endocardial resections are in fact Purkinje fibers is based on their similar structure to normal Purkinje fibers in the many animal species that have been investigated (see below). Purkinje fibers were not found away from the endocardial surface. The Purkinje fibers were present in groups of two to three fibers either in thin bundles consisting entirely of Purkinje fibers or in thick bundles consisting of both Purkinje fibers and ventricular muscle cells (figure 4). In bundles consisting of both Purkinje fibers and ventricular muscle cells the Purkinje fibers were separated from the underlying ventricular muscle by small amounts of connective tissue. Bundles containing Purkinje fibers were identified in specimens from the septum and from the base of the anterior and inferior papillary muscle but were not found in resections from the free wall. Purkinje fibers were not identified in any of the aneurysms.

The Purkinje fibers (figure 5) measured 20 to 45 μm in diameter and were surrounded by a 45 to 50 nm thick basement lamina that closely followed the sarcolemma. The basement lamina was separated from the sarcolemma by a 10 to 25 nm irregular space. The sarcolemma itself was intact around the entire margin of each cell. Pinocytic vesicles formed by microinvaginations of the sarcolemma were abundant. Deep invaginations of the sarcolemma and basement lamina forming a transverse tubular system were not found in the Purkinje cells. The Purkinje fibers were joined to each other by side-to-side and end-to-end intercalated discs that were composed of fascia adherens, macula adherens, and nexi (figure 5, B). The detailed structure of the discs was identical to that previously described for normal Purkinje fibers in other species. We did not find connections between Purkinje fibers and ventricular muscle cells.
Internally, Purkinje fibers contained myofibrils with well-defined sarcomeres but the myofibrils were smaller and less compact than those of ventricular muscle (see below) and did not fill the entire interior of the fiber (figure 5, A). The myofibrils were located at the periphery of the cells but were frequently not in close contact with the sarcolemma. Cylindrical mitochondria, 1.5 to 2.0 μ long and with numerous cristae, were located between myofibrils in areas in which the myofibrils were aligned. In areas devoid of myofibrils, round, oval, and branched profiles of mitochondria with diameters of 0.1 to 1.2 μ were predominantly found. Small mitochondria (<0.4 μ in diameter) usually had only a single cristae. The sarcoplasmic reticulum, consisting of highly branched tubes of irregular size (100 to 200 nm lumen), also coursed among the myofibrils; anastomoses of these tubules regularly occurred at the Z band of the myofibrils. Peripheral cisterns of the sarcoplasmic reticulum were also in contact with the sarcolemma. Abundant glycogen particles (exclusively the β type) occurred in irregular clusters around myofibrils and mitochondria and throughout the sarcoplasm (figure 5, C). Numerous lipid droplets, 0.5 to 1.2 μ in diameter, were also randomly distributed between glycogen particles, mitochondria, and myofibrils (figures 5, C and 6).

Most Purkinje cells had a single centrally located nucleus with several nucleoli (figure 5, A), although occasionally large cells had two nuclei. The nucleoplasm had homogeneously dispersed chromatin but infrequently large clumps of chromatin were observed at the nuclear membrane. A well-developed Golgi apparatus consisting of several stacks of flattened sacules and numerous vesicles was located at the nuclear poles. Profiles of cisternae of rough endoplasmic reticulum, as well as lipofuscin pigment, were present at the nuclear poles.

The ultrastructural appearance of the Purkinje fibers, including their intact sarcolemmas and nuclei with dispersed chromatin, strongly suggests that these cells were viable, an important observation when considering possible mechanisms for arrhythmias (see Discussion).

Ventricular muscle. The majority of the bundles of muscle fibers in the endocardial strips and aneurysms contained only ventricular muscle cells. The ultra-
FIGURE 5. Ultrastructurally the Purkinje fibers were well preserved. Characteristic of Purkinje fibers are (shown in A), the myofiber bundles were haphazardly arranged and the cells lacked T tubules. Aggregates of mitochondria (M) and glycogen were present both in the perinuclear region and between myofiber bundles. The nuclei (N) were centrally located and lipid droplets were abundant (arrows, A and C). The Purkinje fibers were joined by obliquely oriented intercalated discs. The intercalated discs (B) were intact and consisted of macula adherens, fascia adherens, and nexi (large arrow). (Magnifications: A, × 4000; B and C, × 22,400.)

structure of the ventricular muscle in the endocardial strips and that of the aneurysms were identical. Only a small percentage of these fibers (less than 5%) had a completely normal appearance, i.e., ultrastructure identical to muscle from ventricles of normal human or other mammalian hearts.25 These normal-appearing cells had a diameter of 15 to 30 μ and were present in both thick and thin bundles (figure 7). They were surrounded by a basement membrane, and the sarcolemmas were intact. There were numerous invaginations of the sarcolemmas forming an extensive T tubular system. The lumen of the T tubules varied in size and shape. Small tubules were located at the Z bands and large ones overlapped the A and I bands (figure 7, C). The intercalated discs, consisting of fascia adherens, macula adherens, and nexi, were more extensive than...
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FIGURE 6. The lipid droplets (L) were distributed throughout the Purkinje fibers but were most frequently apposed to mitochondria (M). The sarcoplasmic reticulum (arrows) was clearly identified in Purkinje cells but T tubules were not found. (Magnification × 22,400.)

those of the Purkinje cells (figure 7, B). The sarcoplasm contained abundant myofibrils that filled each fiber in all except the central region; the central region was occupied by a normal-appearing nucleus. Oval-shaped mitochondria, 1.5 to 2.0 μ in diameter, were more numerous than in Purkinje cells, and were aligned between myofibrils (figure 7, A). There were also many at the nuclear poles. Glycogen particles were scattered between myofibrils and mitochondria, and were less prominent than in Purkinje cells. The sarcoplasmic reticulum formed an extensive network around the myofibrils.

Mild-to-severe degenerative changes (figures 8 and 9) were found in the majority of ventricular muscle fibers in both thick and thin bundles in all patients.30,31 Cells with mild-to-moderate degenerative change measured 15 to 45 μ in diameter. In the cells with mild degenerative changes the basement laminae, sarcolema, intercalated discs, nuclei, Golgi apparatus, and mitochondria retained a normal appearance, but there was focal loss of myofibrils and a few irregular, large thickenings of the Z bands. No other abnormalities were present. In cells with moderate degeneration (figure 8) the basement laminae, sarcolema, nuclei, and Golgi apparatus were normal, but there was a marked loss of myofibrils with disorganization of the remaining sarcomeres (figure 8, B). There was also an apparent reduction in the number of mitochondria. Accumulations of Z-band material, although smaller in size, were more numerous than in fibers with only a mild amount of degenerative changes. The intercalated discs were focally widened and irregular (figure 8, C). It is possible that both mildly and moderately degenerating cells were viable since their sarcolema were intact and their nuclei appeared normal.

The cells with severe degenerative changes had diameters ranging from 3 to 12 μ. Severe degeneration was characterized by almost complete loss of the myofibrils and mitochondria (figure 9). Only a few small myofibrils remained at the periphery of the cells, and
large numbers of glycogen (α) particles, aggregated segments of dilated sarcoplasmic reticulum, aggregates of cytoskeletal filaments, and lysosomal inclusion bodies were present in areas of myofibril loss. The sarcoplasmic surface was irregularly thickened by deposits of Z band-like material. The basement membranes were focally thickened (figure 9, C). Sarcolemmal cisternae and T tubules could not be identified. Side-to-side intercalated discs were extensive and consisted mostly of desmosomes. The nonspecialized segments of the end-to-end intercalated discs were focally distended and contained particulate material (figure 9, B), although the discs were intact.

In addition to the degenerative changes many of the

FIGURE 7. A small percentage of the ventricular muscle cells in the muscle bundles in both the endocardial resections and aneurysms had a normal appearance. These cells (A) were filled with myofibrils and the sarcomeres appeared to be in registry across the width of the cell. Mitochondria (M) were present adjacent to sarcomeres; the nuclei (N) were centrally located, and sarcolemmas were intact. Many of these cells contained numerous lipid droplets (arrows, A and C) usually adjacent to mitochondria. The intercalated discs (B) were intact and oriented in step-fashion across the cells. Characteristic of ventricular muscle cells (C) T tubules (T) were present adjacent to the Z band. (Magnifications: A, × 8250; B, × 16,500; C, × 20,400.)
ventricular muscle cells were hypertrophied (figure 10). The hypertrophied cells measured up to 60 μ in diameter and were as long as 230 μ. The majority of the hypertrophic cells also demonstrated mild or moderate degenerative changes. In addition to cell size, hypertrophy was characterized by increased numbers of myofibrils across the cells, large lobulated nuclei, accumulations of Z-band material, increased amounts of rough endoplasmic reticulum, and irregular invaginations of the sarcolemmas. Many of the hypertrophic cells contained increased numbers of lipid droplets.

**Discussion**

Electrophysiologic studies in patients with ischemic heart disease and chronic sustained ventricular tachycardia have provided evidence suggesting that this arrhythmia is caused by reentry.4, 5, 9, 12-17, 32-34 Excitation maps obtained during tachycardia have shown that earliest activation during tachycardias usually occurs on the endocardial surface of the left ventricle in an area of endocardial scarring and often in an area adjacent to an aneurysm.3-6, 9, 19 The site of earliest activation with respect to the QRS is most likely the exit point at which impulses conduct from the reentrant circuit into the rest of the ventricle, indicating that either a part or all of the circuit is located in the subendocardium. Electrograms recorded from these subendocardial regions have low amplitudes and are fragmented,4, 23, 24, 34 characteristics that suggest inhomogeneous conduction,20-22, 34 further supporting the supposition that this is the region in which reentry is occurring.33, 34 However, the exact size of the reentrant circuits has not yet been defined. In some instances, discrete electrograms have been recorded throughout the diastolic interval at a number of sites around the borders of an aneurysm, with a sequence of activation suggesting that the circuit was located here (see figure 9 of Horowitz et al.35). When this occurs the circuit is probably relatively large (10 to 12 cm²). In other instances activity occurring throughout the diastolic interval may occur within 2 to 3 cm of the site of earliest activity and the circuit may be about this size.33, 34 The final proof that the arrhythmias originate in these endocardial regions is their abolition by endocardial resection; aneurysctomy alone is often not effective. It is assumed that resection disrupts the reentrant circuit and endocardial resection may affect the entire circuit or only part of it.

Since ventricular tachycardia is abolished by endocardial resection, it is obvious that viable cardiac fibers are removed by this procedure because arrhythmia genesis by reentry or other mechanisms requires electrical activity in cardiac cells.36 Yet, the presence of muscle fibers is not apparent on gross inspection of the resected endocardium, which appears like a piece of glistening white shoe leather (see figure 2 of Josephson et al.8). In fact, we found that 50% to 90% of the material of each resection is connective tissue and only a few bundles of cardiac fibers were found within them. Therefore, the total amount of cardiac fibers removed in any size resection is small.

The pathologic process that leads to the histologic changes in the endocardium of the left ventricle in patients with ventricular tachycardia may be caused by acute coronary artery occlusion. A marked decrease or total absence of blood flow in a major coronary artery leads to extensive muscle death and infarction that may result in the formation of a ventricular aneurysm, especially if the infarct is transmural.37 Myocardial fibers survive on the endocardial surface of either extensive infarcts or aneurysms,26, 27, 38 probably because they receive some blood supply from the ventricular cavity or because of retrograde perfusion through sinusoidal channels.26 When the necrotic myocardial cells are replaced by fibrous tissue, the surviving cells in the subendocardium are trapped in the scar.38-40 The endocardial resections that we studied were often from infarcted areas of the ventricle adjacent to an aneurysm and contained these surviving trapped myocardial cells. In areas adjacent to regions of extensive infarction coronary blood flow to the subendocardium may also be reduced, causing subendocardial ischemia.41 This process may result in focal subendocardial necrosis of myocardial cells, subendocardial fibrosis, and the trapping of myocardial cells that are surviving in the fibrous tissue replacing the necrotic cells.41 Thus, the pathologic changes in the endocardial resections that came from regions adjacent to the infarcts were similar to those in resections from the regions of the infarct. Chronic subendocardial ischemia may also occur in the ventricle without the electrocardiographic or other clinical evidence of infarction but producing subendocardial necrosis and fibrosis and the related histologic picture.41

The ultrastructure of the cardiac fibers embedded in the connective tissue of the subendocardial region provides some further clues to the pathophysiologic processes that lead to structural changes. We found bundles of cells structurally identical to the peripheral Purkinje fibers that have been described in normal and infarcted canine hearts.25, 36 Purkinje fibers were found in resections from the areas of the septum in which fibers of the specialized ventricular conducting system are expected to be abundant. In a previous study, Purkinje fibers were also found on the endocardial surface of trans-
mural anteroseptal infarcts in humans.\(^{27}\) Despite the extensive ischemia that we believe was present in the subendocardium and the dense connective tissue in which these Purkinje fibers were embedded, they appeared to be structurally normal except for the presence of increased numbers of lipid droplets, and we therefore assume that these cells were viable. In normal Purkinje fibers of the nonischemic canine heart, lipid droplets are not numerous.\(^{25,26}\) Purkinje fibers may not undergo the extensive ultrastructural changes that occur in ischemic ventricular muscle because their metabolic requirements are lower than those of ven-

**FIGURE 8.** The majority of ventricular muscle cells demonstrated some degree of myofiber loss, characteristic of degeneration. In cells with a mild or moderate (A) degree of degeneration there was focal loss of myofibers and accumulation of mitochondria (M) and glycogen (G) in areas of myofiber loss. Lipid droplets (L, A and C) and osmiophilic dense granules were prominent. The nuclear chromatin (N) was evenly dispersed and the sarcolemma was intact. In areas of partial myofiber loss many of the remaining myofibers were distorted (B) and thick filaments were lost. The intercalated discs (C) were intact (arrows) but focally widened by accumulations of fibrillary (F) and Z-band material. (Magnifications: A, \(\times\) 6000; B, \(\times\) 16,400; C, \(\times\) 15,600.)
FIGURE 9. In severely degenerating cells (A) only remnants of myofibrils were preserved, usually at the periphery of the cell. The cells were filled with aggregates of abnormal mitochondria (M), glycogen (G), dilated segments of sarcoplasmic reticulum, and osmiophilic dense granules. The nuclear chromatin (N) was evenly dispersed and the sarcolemma intact in spite of the extensive degenerative changes. The intercalated discs (B) were intact although the nonspecialized portions (arrows) were focally distended and contained particulate material. Accumulations of Z-band material (Z) were present beneath the sarcolemma. Scattered degenerating cells (C) had markedly thickened basement membranes (open arrows). Many of these degenerating cells contained numerous lipid droplets (L). (Magnifications: A, × 9900; B, × 32,000; C, × 16,500.)
tricular muscle, enabling them to withstand more ischemia. Diffusion of oxygen from the ventricular cavity may be sufficient to supply most of their metabolic needs. Nevertheless, the presence of lipid suggests that ischemia probably exerted some effect on the metabolism of these cells since lipid readily accumulates in ischemic cardiac fibers.26, 42, 43 Purkinje fibers with normal ultrastructural characteristics except for lipid deposits have been found to survive on the endocardial surface of experimental canine infarcts and are instrumental in the genesis of ventricular tachycardia.26, 40, 44-46

Unlike the Purkinje fibers, a large number of the ventricular muscle fibers in the endocardial resections underwent significant ultrastructural alterations, although some muscle fibers had a normal appearance. Ultrastructural abnormalities ranged from focal loss of myofibrils and irregular, large thickenings of the Z band to almost complete loss of the myofibrils and mitochondria. Most of these cells had intact sarcolemmas and normal-looking nuclei with well-dispersed chromatin, suggesting the possibility that some were viable. The presence of lipid droplets also suggested that they were ischemic. These degenerative changes are probably caused by cellular injury and have been described for several different types of cardiac disease, including cardiomyopathies and valvular heart disease.30, 31 The ultrastructural changes in the muscle fibers in the endocardial resections and aneurysms might be caused by the initial ischemic event or may represent ongoing ischemic damage because of chronic ischemia. These changes are similar to the structural alterations previously described in the ventricular muscle of patients with severe coronary artery disease or acute coronary insufficiency.47, 48

It is not possible to provide direct evidence showing how the structure of the subendocardial region is related to the genesis of chronic sustained ventricular tachycardia, but we can present some suggestions and hypotheses. Tachycardia might be caused by the abnormal geometric arrangement of muscle bundles trapped in fibrous tissue, by abnormal transmembrane potentials related to changes in intracellular ultrastructure, or a combination of both. In the subendocardium

**FIGURE 10.** Many of the ventricular muscle cells were hypertrophied. The cell in this photomicrograph measures 45 μ in diameter, had increased numbers of mitochondria (M), and myofibrils and multilobated nuclei (N). The T tubules of hypertrophied cells were prominent and scattered lipid droplets (arrows) were present. The sarcolemmas and intercalated discs were intact. (Magnification × 3150.)
of patients with tachycardia, muscles bundles are separated from each other by dense connective tissue. If there are interconnections between bundles, they might form anatomically discrete reentrant circuits with an anatomic obstacle. Since the subendocardial region with abnormal structure usually extends over a wide area, the possibility for relatively large as well as small reentrant circuits exists. These circuits in the subendocardium are probably connected to the normal ventricular subendocardium only at the lateral borders of the ischemic region since they were totally separated from the intramural ventricular myocardium beneath them by fibrous tissue. We observed that the muscle bundles traversed the entire length of the resections, toward areas of normal myocardium at its lateral borders. In normal ventricular subendocardium such anatomic circuits do not exist because of the tight packing of the muscle fibers and their numerous interconnections. Direct demonstration of anatomic circuits composed of these bundles would require serial sectioning and reconstruction of the endocardial resections. This approach might also be required to demonstrate differences in structure between the site of earliest activation during the arrhythmia and other sites in the endocardial resection. With the methods we used we could not distinguish any differences in arrangement of the bundles at these different sites that might be related to the mechanism of origin of the arrhythmia. Our data also do not indicate whether the reentrant circuits comprise Purkinje fiber bundles, ventricular muscle bundles, or both. However, we have shown that Purkinje fiber bundles are present in some resections, thus offering the possibility that they are involved. The absence of Purkinje fibers in other specimens does not necessarily mean they were not present, only that we did not find them.

We also must consider the possibility that abnormalities of the transmembrane potentials of the cardiac fibers in the resections contribute to the occurrence of tachycardia, and that these abnormalities may be related to some of the ultrastructural changes that we described. Purkinje fibers appeared ultrastructurally normal but contained significant cytoplasmic lipid droplets. In experimental studies in canine infarcts, Friedman et al. have shown that Purkinje fibers surviving on the endocardial surface of extensive infarcts also have large deposits of lipid, and that the accumulation of lipid in these cells is related temporally to the occurrence of abnormalities in the transmembrane action potential. In particular, these Purkinje fibers have reduced resting potentials and abnormally prolonged action potential durations that facilitate the occurrence of reentry in vitro. Action potentials return to normal when the lipid droplets disappear. Purkinje fibers with markedly reduced resting potentials and very slow upstrokes have also been found in canine infarcts and on the endocardial surface of resected human ventricular aneurysms.

Unlike the Purkinje fibers, most of the ventricular muscle fibers in the subendocardium had very abnormal ultrastructures. Although ultrastructural changes have not yet been causally related to changes in transmembrane potentials, similar ultrastructural changes in muscle cells in canine infarcts are associated with abnormalities in resting and action potentials. Bundles of ventricular muscle survive on the epicardial aspect of experimental transmural canine infarcts in the same way that muscle survived on the endocardial aspect the human infarcts described in this study. The surviving ventricular muscle fibers in the canine infarcts show loss of myofibrils and mitochondria, Z-band thickening, and significant lipid deposits, but intact sarcolemmas and normal nuclei 2 weeks after coronary occlusion. The cells have reduced resting potentials and upstroke velocities and markedly shortened action potential durations. Similar transmembrane potentials have been recorded from muscle fibers surviving in chronic feline infarcts. These alterations in membrane potentials might be related to ultrastructural changes and may contribute to the occurrence of tachycardia.

One additional extrapolation can be made from the structure of the subendocardium that we have described. The presence of individual, widely separated bundles of muscle fibers would be expected to give rise to the low-amplitude, usually fragmented extracellular electrograms that are often recorded from this region with bipolar electrodes. The low amplitude of the potentials may be caused by the paucity of surviving muscular tissue and fragmentation may be related to asynchronous activation of muscle bundles even within regions smaller than 1 mm². Asynchronous activation may occur because the muscle bundles are not interconnected along much of their length but are widely separated by connective tissue.

References
4. Josephson ME, Horowitz LN, Farshidi A, Spear JF, Kastor JA,
26. Friedman PL, Fenoglio JJ Jr, Wit AL: Time course for reversal of electrophysiologic and ultrastructural abnormalities in subendocardial Purkinje fibers surviving extensive myocardial infarction in dogs. Circ Res 36: 127, 1975
43. Ferrans VJ, Roberts WC: Myocardial ultrastructure in acute and chronic hypoxia. Cardiology 56: 144, 1972
44. Wit AL, Bigger JT: Possible electrophysiologic mechanisms for lethal arrhythmias accompanying myocardial ischemia and infarc- tion. Circulation 61: 11-96, 1975
48. Schwarz F, Flameng W, Thiedemann K-U, Schaper W, Schlepper M: Effect of coronary stenosis on myocardial function, ultrastruc-

49. Friedman PL, Stewart JR, Wit AL: Spontaneous and induced cardiac arrhythmias in subendocardial Purkinje fibers surviving extensive myocardial infarction in dogs. Circ Res 33: 612, 1973


52. El-Sherif N, Lazzara R: Reentrant ventricular arrhythmias in the late myocardial infarction period. Effects of verapamil and D600 and the role of the slow channel. Circulation 60: 605, 1979


58. Spach MS, Miller WT, Dolber PC, Kootsey JM, Sommer JR, Mosher CE: The functional role of structural complexities in the propagation of depolarization in the atrium of the dog: Cardiac conduction disturbances due to discontinuities of effective axial resistivity. Circ Res 50: 175, 1982
Recurrent sustained ventricular tachycardia: structure and ultrastructure of subendocardial regions in which tachycardia originates.
J J Fenoglio, Jr, T D Pham, A H Harken, L N Horowitz, M E Josephson and A L Wit

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