Decreased Myocardial Contractility in Papillary Muscles from Atherosclerotic Rabbits

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SUMMARY To determine the effect atherosclerosis has on myocardial contractility, we studied the contractile properties of right ventricular papillary muscles from 34 atherosclerotic and 17 control rabbits. We produced atherosclerosis by feeding for 2 to 8 months a diet of 5% lard, 5% peanut oil, 0.5% cholesterol, and 89.5% rabbit pellets. The controls received only rabbit pellets during the same time interval. Contracting isometrically 12 times per minute at 25°C, muscles from the atherosclerotic rabbits developed tension at a lower maximum rate (max dT/dt), had a longer latency, and required longer to develop tension at the maximum rate and to develop peak tension. In isotonic contractions, they shortened with lower minimum velocities and required longer to accelerate to minimum velocity and to shorten maximally. We found no evidence that developed tension or distance shortened differed between the two groups of muscles. Raising the contraction frequency to 24 contractions per minute brought performance of the two groups of muscles closer in both types of contraction. Norepinephrine (1.5 x 10⁻⁴ M) nearly abolished differences between performance of the two groups. The loss of contractility correlates poorly with coronary and aortic atherosclerosis. It occurred early in the feeding of the atherogenic diet. We think it was due to a lipid-induced defect in the cardiac cell's handling of calcium. Circ Res 45: 338-346, 1979

MYOCARDIAL contractility is low in coronary artery disease. As measured during cardiac catheterization, the loss is proportional to the severity of the disease (Hamby et al., 1973; Moraski et al., 1975; Rackley and Russell, 1975). Some of the loss may be due to ischemia, because bypassing plaques with arterial grafts improves exercise tolerance and lessens angina (Kouchoukos et al., 1975). However, perhaps there is also a loss that is independent of the immediate presence of ischemia. To test for it, we performed the research described in this paper. Our results were obtained from isolated papillary muscles. Oxygen supply to the muscles depended, therefore, on diffusion, not on blood flow through atherosclerotic arteries. Some of our results have been reported in an abstract (Peterson et al., 1977).

Methods

Animals and Diets

We assigned male New Zealand laboratory rabbits randomly to two groups. One group received rabbit pellets for 2–8 months. Those in the other group received an atherogenic diet composed of 89.5% rabbit pellets, 5% lard, 5% peanut oil, and 0.5% cholesterol for the same length of time. At the start of the study the control rabbits weighed 2.42 ± 0.12 kg (mean ± se); those receiving the atherogenic diet weighed 2.42 ± 0.09 kg. We randomized the order the rabbits were used in experiments. A rabbit selected for an experiment was brought to a surgical plane of anesthesia with sodium pentobarbital (mean of 54 ± 1.7 mg/kg) given intravenously. We then quickly removed the heart, isolated a...
ventricles separately. We preserved the ventricles for our study of contractility, and weighed the left and right ventricles midway between the base and apex of the heart. Had the papillary muscle we used in studying contractility been left in place, a large part of it would have been included in the block. Duplicate slides stained with hematoxylin and eosin were prepared from 5-μm sections of each block. We examined each slide for coronary atherosclerosis, necrosis, and fibrosis. In grading coronary atherosclerosis, we calculated the percentage of the arteries that contained plaque and visually estimated the percentage occlusion of each atherosclerotic artery. In making these determinations, we included only those arteries that were cut in cross-section, an average of 120 ± 6 in the left ventricle and 75 ± 5 in the right ventricle. We classified each artery as epicardial or intramural.

We stained the aorta with Sudan IV, then photographed it. From 20-cm by 25-cm photographic prints, we cut out and weighed the images of the entire aorta and of just the stained atherosclerotic lesions. Dividing the weight of the latter by the weight of the former, we calculated the percentage of the intimal surface that contained lesions.

**Papillary Muscle Preparation**

We suspended the papillary muscle between short sections of aluminum tubing and mounted it between a force transducer and a lever in a myograph. Other investigators have described the isolating procedure and similar equipment (Noble et al., 1969; Donald et al., 1976). During the isolation and while in the myograph, the muscle was immersed in a modified Krebs-Ringer superfusate composed, in millimoles per liter, of Na+, 145.0; K+, 4.2; Ca2+, 2.5; Mg2+, 1.2; Cl−, 125.5; HCO3−, 25.0; H2PO4−, 2.4; SO42−, 1.2; and glucose, 5.6. The superfusate was equilibrated with 95% O2 and 5% CO2, which made its pH 7.4 and its P02 greater than 600 mm Hg. It was pumped through the muscle bath at 9 ml/min. Immediately before the superfusate entered the muscle bath, a thermoelectric module (heat pump), functioning in a feedback circuit with a temperature controller and a power supply, brought it to 25°C. The muscle was stimulated by supramaximal rectangular pulses 5 msec in duration that were delivered to the muscle bath through large stainless steel wire electrodes placed parallel to the muscle. A micrometer, by contacting the lever, determined initial muscle length.

Force was applied to the lever electromagnetically. A copper wire attached to the lever was connected electrically through two pools of mercury to a programmable power supply (Kepco model BOP 15-20M) that provided current. Passing current through the wire, which was in a strong magnetic field, applied a proportional force to the lever.

During experiments, a LINC computer calculated the load for isometric and isotonic contractions and sent appropriate signals through a digital-to-analog converter to the current source. Length and tension signals, calibrated against known values, passed through analog-to-digital converters to the computer. The computer sampled the signals every 2 msec, obtaining 900 points from each per contraction. A stimulator initiated the sampling sequence by sending to the computer a signal coincident with the stimulus it sent to the stimulating electrodes. The computer averaged, point by point, the data it obtained from four contractions against a given load. It recorded the average on digital magnetic tape.

The force transducer consisted of a strain gauge bonded to a stainless steel bar. The length transducer was a linear differential transformer. Signals from the transducers were linear over the ranges of force and length encountered in our research. They remained flat and in phase to 200 cycles/sec. Compliance of the force and length transducing systems was about 0.5 μm per gram of force applied to the lever. The lever assembly had less than 400 mg of equivalent mass.

**Protocol**

After calibrating the length and tension signals, approximately 3 hours after installing the muscle in the myograph, when contractile performance had stabilized, we recorded data for the length-tension curve, starting at a length at which resting tension was zero and stopping when developed tension reached a plateau or dropped. The muscle was permitted to equilibrate to a new length during 2 minutes before data were recorded. We took as Lmax the shortest length at which developed tension was maximum. At the lower end of the length-tension curve, we increased length in increments that approximated 2% of Lmax. As muscle length approached Lmax at about 98% of Lmax, we decreased the increments in length to 1%. The contraction frequency was 12 per minute.

We next recorded one series of isotonic contractions at 12 contractions per minute, one at 24 contractions per minute, and one at 12 contractions per minute with the bathing solution containing norepinephrine (1.5 × 10−6 M). Before recording these data, we waited 5 minutes after changing the contraction frequency and 15 minutes after the isotropic effects of the norepinephrine first became apparent. Contractile performance stabilized during these intervals. Each series began with contractions against a load equal to, or slightly greater than, the preload and ended with isometric contractions.
between these extremes, contractions were recorded at 0.5 g/mm² increments in load. The initial muscle length for each contraction was \( L_{\text{max}} \).

**Data Analysis**

In analyzing the average of the contractions at each load, we ran a program that read the data from tape into the computer and analyzed it. Using least squares routines to smooth and differentiate the length and tension data (Savitzky and Golay, 1964), the computer calculated from each averaged contraction resting tension, maximum developed tension, maximum rate of tension development (max \( dT/dt \)), maximum distance shortened, and maximum velocity of shortening (max \( dL/dt \)). It also measured the time intervals from the stimulus to each of these events. In addition, from tension data, it measured latency, duration of contraction, and relaxation time. The last property was measured from maximum developed tension to the end of the contraction; the preceding two were measured from the stimulus.

We compared the results from the atherosclerotic and control groups statistically by Student's t-test for unpaired samples, using the pooled or unequal variance, as appropriate (Snedecor and Cochran, 1967).

**Results**

**Papillary Muscles**

Papillary muscles from the two groups of rabbits were nearly the same size. Cross-sectional area, which we calculated assuming the muscle to be circular in cross-section and using diameter measured through a dissecting microscope, averaged 0.63 ± 0.04 (mean ± SE) mm² in the control group \( n = 34 \) and 0.64 ± 0.06 mm² in the atherosclerotic group \( n = 17; P = 0.4 \). To assure adequate oxygenation throughout the muscle, we used no muscle for which cross-sectional area exceeded 1 mm²; this value is well below the area at which the center of rabbit papillary muscle becomes hypoxic under the conditions used in this research (Snow and Bressler, 1977). At \( L_{\text{max}} \), muscle length averaged 5.2 ± 0.3 and 5.3 ± 0.2 mm in muscles from the control and atherosclerotic rabbits, respectively.

**Isometric Contractions**

Figure 1 illustrates the length-tension relationships of the two groups of muscles. Muscles from atherosclerotic rabbits tended to be the weaker, but the differences, averaging only 457 mg/mm² (range: 346 to 626 mg/mm²), are significant only at lengths shorter than 94% of \( L_{\text{max}} \). At \( L_{\text{max}} \), muscles from the atherosclerotic rabbits developed 4.12 ± 0.24 g/mm²; those from the controls developed 4.46 ± 0.39 g/mm² (Table 1). We found no statistical evidence that resting tension differed between the two groups at any length.

At all the muscle lengths we studied, papillary muscles from atherosclerotic rabbits developed tension at slower maximum rates than those from the controls (Fig. 2). These differences are evident in the time intervals measured from the tension-development part of the contraction. Throughout the length-tension curve, time-to-peak tension was significantly longer in contractions of muscles from atherosclerotic rabbits \( P < 0.001 \); average difference: 64 msec), and so were latency \( P \leq 0.02 \); average difference: 14 msec) and time to max \( dT/dt \)
Isotonic Contractions

Results of the isotonic contractions corroborate those of the isometric contractions. We found no evidence that distance shortened, plotted as a function of force, differed between the two groups of muscles at 12 contractions per minute, at 24 contractions per minute, or when contractility was augmented with norepinephrine. In each instance the force-shortening relationships from the two groups of muscles are superimposable. However, when contracting 12 times per minute, muscles from the atherosclerotic rabbits shortened more slowly than those from the controls (Fig. 3). Raising the contraction frequency to 24 per minute decreased the differences. Norepinephrine abolished them and made the curves superimposable at all but 10% of the isometric force. At this force, shortening of muscles from the atherosclerotic rabbits was sig-

![Figure 3](https://example.com/figure3.png)
nificantly slower (0.91 ± 0.03 vs. 1.06 ± 0.05 muscle lengths/sec, \( P \leq 0.01 \)). As for time to max dT/dt, time to max dL/dt was longer at each force at 12 contractions per minute in contractions of muscles from the atherosclerotic rabbits (Fig. 4). At 24 contractions per minute, the differences were smaller but still significant. When norepinephrine was added to the superfusate, time to max dL/dt differed significantly only at 10% of the isometric force, where the average from contractions of muscles from atherosclerotic rabbits were longer by 10.4 msec \( (P \leq 0.01) \). Throughout the range of forces, the difference between the means averaged 95 msec at 12 contractions per minute, 29 msec at 24 contractions per minute, and 4 msec when contractility was increased by norepinephrine.

Time to maximum shortening was longer in contractions of muscles from the atherosclerotic rabbits. The average difference between the means was 95 msec at 12 contractions per minute (Fig. 5). As for time to max dL/dt, it was smaller in contractions of the two groups of muscles at 24 contractions per minute, the difference averaging 83 msec. Norepinephrine lowered the difference to 32 msec, but at each force the muscles from the atherosclerotic rabbits still required significantly more time to shorten maximally.

Correlations with Pathology and Time on Diets

As is common in atherosclerotic rabbits, intramyocardial arteries were affected more than were the larger epicardial ones. However, neither the percentage of the intramyocardial arteries that contained plaque (left ventricle = 11%; right ventricle = 40%) nor the percentage of each affected artery was occluded (left ventricle = 78%; right ventricle = 49%) correlates well with any properties of the isometric contraction. In each case the correlation coefficient is 0.35 or smaller. The highest correlation is between max dT/dt and the percentage of the small coronaries in the right ventricle that contained plaque. Although this correlation coefficient is significant \( (P \leq 0.05) \), it is small and positive. It therefore clearly does not explain the lower max dT/dt in contractions of muscles from our atherosclerotic rabbits.

Six sections from the atherosclerotic hearts and none from the control hearts revealed fibrosis, probably the result of ischemia. Damage was confined to the left ventricle: in five it was in a papillary muscle, and in one it was in the wall. The six sections came from rabbits that had been fed the atherogenic diet for 131-248 days. The severity of atherosclerosis in the left ventricle did not differ significantly between these hearts and the 28 in which we found no damage. Of all the properties we measured in the isometric and isotonic contractions, only relaxation time differed significantly between contractions of muscles from the fibrotic and nonfibrotic hearts. Simultaneously, papillary muscles from fibrotic hearts relaxed significantly more slowly \( (P \leq 0.05) \), by an average of 87 msec. They also relaxed more slowly when contracting 24 times per minute at \( L_{\text{max}} \), the average difference being 143 msec \( (P < 0.05) \). There was no statistically significant difference in relaxation when contractility was increased by norepinephrine.

The correlations between the mechanical properties and aortic atherosclerosis are also small and nonsignificant statistically. The largest correlation...
We have presented evidence that an atherogenic diet lowers myocardial contractility in the rabbit. The diet did not change distance shortened against a given isotonic load or developed tension in isometric contractions. However, it did depress the force-velocity relationship, lower max dT/dt, and lengthen time intervals in the tension development and shortening parts of the isometric and isotonic contractions. For the most part, the results corroborate those we obtained in preliminary research (Peterson and Griffith, 1977).

Evidence indicates that the loss of contractility is real. The sample size in each of the groups is adequate, and the levels of significance with which almost all the null hypotheses were rejected make extremely small the likelihood that the differences occurred by chance. Few papers describing the results of experiments with rabbit papillary muscles contain averages with which our control data can be compared, and in those that do, the data were obtained at combinations of contraction frequency, temperature, and calcium concentration different from ours. Nevertheless, when these differences are taken into account, our control muscles performed comparably to rabbit papillary muscles used by others (Bodem and Sonnenblick, 1975; Edman and Nilsson, 1972).

Two points that have bearing on performance deserve comment. First, the ratio of resting to total tension was 0.26 as the muscles were stretched to the point of the length-tension curve (Fig. 1 and Table 1; 12 contractions per minute, 25°C), higher than the 0.19 ratio Bodem and Sonnenblick (1975) found in rabbit muscles that were contracting 36 times per minute at 30°C. However, stress relaxation always occurred when the muscles were stretched to Lmax. The rate was most rapid initially. By the end of the first isotonic series, which was recorded under the same conditions as the length-tension curve, resting tension had fallen from 1.53 ± 0.19 to 0.96 ± 0.10 g/mm² in the control muscles and from 1.45 ± 0.08 to 0.95 ± 0.06 g/mm² in muscles from the atherosclerotic rabbits. Yet, developed tension had changed little, averaging 4.38 ± 0.43 g/mm² in contractions of control muscles and 4.14 ± 0.26 g/mm² in contractions of muscles from atherosclerotic rabbits. Thus, in these contractions, the ratio of resting tension to total tension was 0.18. At 24 contractions per minute it was 0.10 (Table 1). Both values are smaller than the average found by Bodem and Sonnenblick (1975) and below the upper limit some investigators use in selecting suitable cat papillary muscles (Henderson et al., 1973; Paulus et al., 1976). Second, the force-velocity relationship has little curvature (Fig. 3); this, too, is typical of data obtained from afterloaded isotonic contractions of rabbit papillary muscle (Bodem and Sonnenblick, 1975; Edman and Nilsson, 1972). Velocity of shortening at the preload is lower than values reported by others (Bodem and Sonnenblick, 1975; Edman and Nilsson, 1972), but appropriate for the temperature and contraction frequency we used (unpublished observations).

The loss of contractility seems unrelated to both coronary and aortic atherosclerosis. Although coronary atherosclerosis can cause ischemia and

<table>
<thead>
<tr>
<th>Table 2 Ventricular Weights</th>
<th>Control</th>
<th>Atherosclerotic</th>
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<tr>
<td>n</td>
<td>17</td>
<td>34</td>
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<tr>
<td>Left ventricle g/kg</td>
<td>4.76 ± 0.18</td>
<td>5.22 ± 0.17*</td>
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<tr>
<td>g/kg</td>
<td>1.34 ± 0.05</td>
<td>1.49 ± 0.03†</td>
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<td>Right ventricle g/kg</td>
<td>1.47 ± 0.06</td>
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<td>g/kg</td>
<td>0.42 ± 0.02</td>
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Each entry is an average wet weight ± SE.
* Significantly greater than control, 0.01 < P < 0.05.
† Significantly greater than control, 0.001 < P < 0.01.

coefficient, 0.03, is in the relationship between time-to-peak tension, the time interval that changed most during the experiment, and percentage of the intima that was covered by plaque.

Expressed in grams or grams per kilogram body weight, the left ventricles of the atherosclerotic rabbits hypertrophied by 10-11% (Table 2). As calculated from observations made by Ho and Taylor (1968), the increase in weight is too large to have been due solely to stored cholesterol. Our calculations do not correct for changes in water content, an unknown in our study that, depending upon the content of lipids and fatty acids in the myocardium, could have increased or decreased weight (Ho and Taylor, 1968). The hypertrophy does not explain with each mechanical property.

Feeding the atherogenic diet from day 63 to day 248 of the study produced little of the change in the time intervals of the isometric contraction and no change in developed tension or max dT/dt. Of those time intervals that were slowed significantly by the atherogenic diet, only time-to-peak tension correlates significantly with the number of days the atherogenic diet was fed, and that correlation coefficient is small (r = 0.37, P ≤ 0.05). The major increase in this time interval occurred before the first experiment, 63 days, and from then on, values of this property from the two groups overlapped little. Like time-to-peak tension, neither latency nor time to max dT/dt correlates with the length of time we fed the diet (P > 0.05 in each case); the largest increase in each occurred before our first experiment. The three time intervals changed little during the second half of the study. By 2-4 months (n = 17), the values were: latency, 60.0 ± 1.9 msec; time to max dT/dt, 208.1 ± 7.9 msec; time-to-peak tension, 418.6 ± 12.5 msec. All are significantly larger than corresponding measurements from controls and nearly the same as averages obtained for the entire study (Table 1).

Discussion

We have presented evidence that an atherogenic diet lowers myocardial contractility in the rabbit. The diet did not change distance shortened against a given isotonic load or developed tension in isometric contractions. However, it did depress the force-velocity relationship, lower max dT/dt, and lengthen time intervals in the tension development and shortening parts of the isometric and isotonic contractions. For the most part, the results corroborate those we obtained in preliminary research (Peterson and Griffith, 1977).

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The loss of contractility seems unrelated to both coronary and aortic atherosclerosis. Although coronary atherosclerosis can cause ischemia and
thereby lower myocardial contractility, we found no
evidence that it contributed significantly to our
findings. All of the mechanical properties we mea-
sured correlate poorly with coronary atheroscle-
rosis, and only relaxation time, which was not changed
by atherosclerosis, differed between contractions of
muscles from fibrotic and nonfibrotic hearts. These
results, and the fact that the changes in mechanical
function occurred largely during the first 4 months
we fed the diet, when coronary atherosclerosis is
mild and the rabbit myocardium shows no histologi-
cal evidence of hypoxia (this study; Prior et al.,
1961), argue against coronary atherosclerosis hav-
ing lowered contractility.

The left ventricle in the atherosclerotic rabbits
was hypertrophied by 10–11% from an unknown
cause. This, too, is not likely to have lowered con-
tractility in right ventricular papillary muscles; it
correlates poorly with each mechanical property.
Although hypertrophy and failure produced in one
ventricle by severe pressure overload lower Ca2+-
ATPase activity and contractility in the other ven-
tricle, hypertrophy alone does not (Raszkowski et
al., 1977). That the left ventricle was subjected to
severe pressure overload seems highly unlikely, and
we found no evidence of either left or right ventric-
ular failure in any rabbit. Also, if it had been pro-
duced by pressure overload, the amount of hyper-
trophy we found should not have lowered contrac-
tility even in the left ventricle (Bing et al., 1971).
Volume overload does not lower contractility
(Cooper et al., 1973).

The nature of the change in contractility could
indicate that the atherogenic diet slowed the rate
at which calcium was delivered to the sarcosome. A
diet-induced alteration in the composition, and
thereby in the fluidity and function of the sarco-
lemma or sarcoplasmic reticulum or both, is the
most likely cause. It might have included both
cholsterol and phospholipids, both of which are
natural components of cellular membranes in mam-
mals.

Fed to rabbits for several months, cholesterol
produces a lipid storage disease in addition to ath-
erosclerosis. Practically all organs but the pancreas
and brain accumulate additional cholesterol (Ho
and Taylor, 1968; Clarkson et al., 1974). When the
diet is 0.25% cholesterol and 10% Wesson oil (Corey
and Zilversmit, 1977) or 2% cholesterol (Ho et al.,
1974; Ho and Taylor, 1968), the heart doubles its
content in 2–3 months. Continuing the cholesterol
feeding for another 7 months does not force the
heart to store additional amounts (Ho and Taylor,
1968; Ho et al., 1974). The time course of the ac-
cumulation and the saturation at 60 days correlate
nicely with our observation that the biggest loss of
myocardial contractility occurred during the first 2
months we fed the diet.

Although some of the additional cholesterol
could be in the form of intracellular lipid droplets
(Jackson and Gotto, 1976), most of it must be in
cellular membranes (Sabine, 1977). The effects cho-
lesterol has on the sarcolemma and sarcoplasmic
reticulum of control or cholesterol-fed rabbits have
not been studied, but research with phospholipid
vesicles and membranes from other tissues indi-
cates that there it could be expected to decrease
permeability (Papahadjopoulos et al., 1973a, 1973b;
Sabine, 1977) and to lower the activity of mem-
brane-bound enzymes (Bloj et al., 1973a, 1973b;
Fiehn and Seiler, 1975; Kimelberg and Papahadj-
poulos, 1974; Kroes et al., 1972). For example, re-
placing cholesterol in skeletal muscle sarcolemma
with desmosterol increases the activities of (Na+ +
K+)-ATPase and Ca2+-ATPase, but not that of
Mg2+-ATPase (Peter and Fiehn, 1973). Cholesterol
is not necessary for the sarcoplasmic reticulum in
skeletal muscle to take up calcium (Drabikowski et
al., 1972; Flaherty et al., 1975), but excess choles-
terol bound to the sarcoplasmic reticulum decreases
fluidity of this membrane and inhibits both its
uptake of calcium and Ca2+-ATPase activity (Dra-
 bikowski et al., 1972; Seeilig and Hasselbach, 1971).

Phospholipids, like cholesterol, can alter the
fluidity and function of membranes. Fluidity varies
with the quantity of phospholipid in the membrane
and with the nature of the phospholipids’ polar
head groups; it decreases when the acyl chains
attached to the head groups are longer or more
saturated (Sabine, 1977). This mechanism may be
a less likely cause of the loss we found in contrac-
tility than the one involving cholesterol. A variety
of diets fed to rats does not change the fatty acid
composition of lipids in the erythrocyte (Bloj et al.,
1973a, 1973b), a cell that, like the sarcoplasmic
reticulum, readily exchanges cholesterol with nutri-
ent fluids and in which cholesterol modifies fluidity
and the activity of membrane-bound enzymes (Bloj
et al., 1973a, 1973b; Kroes and Ostwald, 1971; Kroes
et al., 1972).

Another of the possible mechanisms also involves
lipids and calcium flux. Light and electron micros-
copy have shown many membrane-enclosed drop-
lets of neutral lipid, probably triglycerides (Rouser
et al., 1968) but possibly also esterified cholesterol
(Jackson and Gotto, 1976), in cardiac fibers from
rabbits that had been fed an atherogenic diet for 2
months (Wellman and Volk, 1970). The droplets
were between myofibrils and, often, near mitochon-
dria. Perhaps they slow the diffusion of calcium to
the regulatory proteins. Changes one could expect
this mechanism to produce in mechanical function
are consonant with our data. From the results we
report here, this mechanism is, therefore, indisting-
uishable from, although in our opinion less likely
than, those involving a change in function of the
sarcolemma or sarcoplasmic reticulum.

Perhaps the atherogenic diet lowered myocardial
contractility indirectly through metabolism. ATP
the cardiac cell uses as fuel for contraction appears
to come from a specific intracellular pool (Gudbjar-
nason et al., 1975; McDonald et al., 1971; Scheuer,
This pool may contain only ATP that is produced by oxidative metabolism (McDonald et al., 1971), which occurs in mitochondria, primarily at the inner membrane. Both phospholipids and cholesterol may modify mitochondrial membranes and thereby both lower activity of enzymes in them and decrease ATP production. These membranes normally contain little cholesterol (Jain, 1975; Parsons and Yano, 1967; Rouser et al., 1968), and information obtained from other membranes indicates that, because they contain more protein than lipid (Rouser et al., 1966; Sabine, 1977) and because acyl chains in their phospholipids are highly unsaturated (Jain, 1975), they should not accumulate large quantities. Some mitochondrial enzymes require phospholipids for activity (Coleman, 1973), but we know of no research showing that excess phospholipids or different phospholipids will hamper their activity. However, because cholesterol or phospholipid could upset metabolism by being incorporated into just the area of the membrane where a specific enzyme is located, small increases in the amounts of these lipids may have produced the changes we found in contractile function.

There are also the possibilities that the dietary lipids altered the regulatory proteins or contractile proteins, that they made series elasticity more compliant, or that the increase in lipids intracellularly interfered physically with contraction. A decrease in the affinity of troponin C to calcium reportedly develops during ischemia (Schwartz et al., 1973), but, as noted above, we found little evidence of ischemia. Cholesterol plays a role in protein synthesis (Sabine, 1977), but the production of abnormal proteins has not been linked to the presence of excess cholesterol. We think our data eliminate as research applications: I. Atherosclerosis research. In The Biology of the Laboratory Rabbit, edited by WH Weisbroth, DE Platt, AL Kraus. New York, Academic Press, pp 155-165. Coleman R. 1973. Membrane-bound enzymes and membrane ultrastructure. Biochim Biophys Acta 300: 1-30.


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