# Effect of Catecholamine Depletion on an Index of Myocardial Contractility in Isolated Rabbit Hearts.* (29340)

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The role played by the endogenous catecholamines of the left ventricle in maintaining normal contractility is not established. It is well known that administered norepinephrine and related catecholamines increase the contractility of the mammalian myocardium. Thus, norepinephrine and epinephrine increase the peak force attained by segments of the myocardium which contract isometrically against a strain gauge arch(1); they increase the amplitude of contraction of isolated heart preparations(2,3); and they enhance ventricular function curves(4). Furthermore, injected norepinephrine and related catecholamines increase the rate of shortening and rate of tension development by the myocardium(5). Aspects of cardiac function which are dependent on these derivatives, *e.g.*, rate of change of ventricular pressure, ejection velocity and stroke power, are also increased by catecholamines(6).

Lee and Shideman(7) reported that the amplitude of isotonic contractions of electrically driven, isolated cat papillary muscles is reduced following treatment with reserpine or following sympathetic denervation. They considered the reduction in contractility of the papillary muscle to be a consequence of the depletion of endogenous catecholamine stores that these procedures induce. Lee and Shideman interpreted their findings as indicating that in normal hearts small quantities of norepinephrine are continuously released from catecholamine stores—presumably independently of neuronal activity—to maintain a tonic level of ventricular myocardial contractility.

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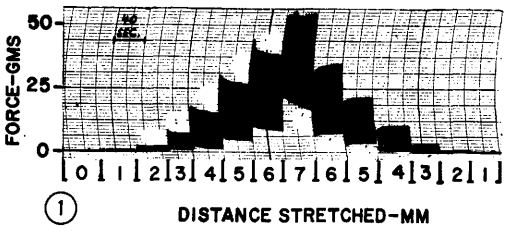
Withrington and Zaimis(8) also reported that reserpine reduces myocardial contractility in intact cats. The reduction in contractility may be so great that heart failure ensues. These authors hold the opinion that the reduction in contractility is due to the marked degeneration in the heart muscle which reserpine produces and is not due to catecholamine depletion. They consider it unlikely that the severe histological changes found in the myocardium following reserpine treatment could be caused by depletion of endogenous catecholamines.

We have examined the relationship between catecholamine stores and an index of myocardial contractility in isolated perfused rabbit hearts. Guanethidine and reserpine were used to deplete amine stores.

*Material and methods.* Rabbits weighing 5 to 6 lb were selected without regard to sex and sacrificed by cervical dislocation. Their hearts were excised and mounted in the Langendorff apparatus of Anderson(9). Hearts were perfused with Ringer-Locke solution at 37.5°C at a pressure of 55 mm Hg. A silver clip was attached to the apex of the left ventricle and was connected by a fine silver chain to a Grass FT-03 force transducer mounted in a Harvard isometric tension clamp. Signals from the transducer were recorded on a Grass Polygraph. The heart was allowed to equilibrate for 25 minutes before stretching was started. After the equilibration period, the slack in the chain was taken up until tension was first detected. The chain was then slackened to the nearest mm marking on the scale of the isometric tension clamp. This marking was considered as the point of zero stretch. The heart was then stretched in increments of 1 mm at 40 second intervals. Readings were taken at the end of the 40 second intervals. Stretching was limited to 7 mm since in preliminary experiments stretching in excess of 7 mm often resulted in left ventricular standstill or



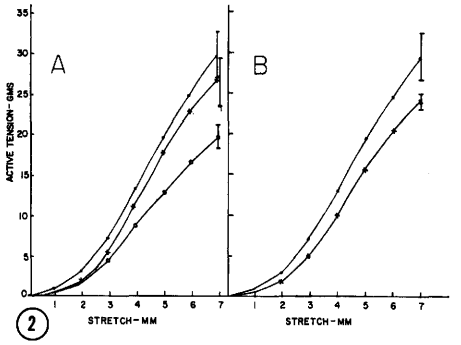




FIG. 1. Typical polygraph recording showing relationship between stretch and tension development in untreated isolated rabbit heart. Heart was stretched in 1 mm increments up to 7 mm total stretch and then relaxed in 1 mm increments.

FIG. 2. Peak active tension developed by left ventricle of isolated rabbit heart as a function of stretch applied to apex of heart. A. Effect of guanethidine. Solid circles—untreated hearts. Crosses—hearts from animals treated I.V. with 20 mg/kg guanethidine. Open Circles—hearts from animals treated I.V. with 10 mg/kg guanethidine. Vertical bars represent standard errors. B. Effect of reserpine. Solid circles—untreated hearts. Crosses—hearts from animals treated I.V. with 3 mg/kg reserpine phosphate.

tearing of the apex of the ventricle. Each heart was stretched and relaxed as many times as was necessary for the active tension occurring at various lengths during stretch to agree within 15% of each other with less than 1 mm change in the point of zero stretch. This usually required two trials and occasionally three. Readings were taken from the last trial. Ten minutes were allowed to elapse between rounds of stretching.

Rabbits were pretreated with 10 mg/kg guanethidine, 20 mg/kg guanethidine or 3 mg/kg reserpine. The drugs were given by vein 16 hours before sacrifice. Frequently, but not always, hearts from treated animals beat at a considerably slower rate than hearts from untreated rabbits. These slow hearts were driven by electrical impulses via platinum electrodes clipped to the right atrium at a rate equal to the median value for the rate of untreated hearts. It was considered necessary that the slow hearts be driven in order to rule out possible effects of reduced heart rate on strength of contraction.

The norepinephrine content of left ventricular tissue was estimated by the fluorometric assay described by Sourkes and Murphy(10).

*Results: Untreated hearts.* A typical record relating active tension to mm stretch in untreated hearts is presented in Fig. 1. Tensions were recorded as the ventricle was stretched and as it was returned to normal length. The active tension developed at any given length during stretch exceeded slightly the active tension which developed as the muscle was permitted to relax. A summary of the tensions developed during stretch in 11 similar experiments is plotted in Fig. 2. There is a sharp rise in active tension from 0 to approximately 30 g as the heart is stretched. The average heart rate $\pm$S.E. for controls was $39\pm3$ beats/15 sec. Heart rates did not change during stretch. The average value $\pm$S.E. for the norepinephrine content of the left ventricles of 5 untreated hearts was $1.59\pm.22$ $\mu$g/g of tissue.

*Treated hearts.* Ten mg/kg and 20 mg/kg of guanethidine reduced the norepinephrine content of the left ventricle by approximately 70%; average values $\pm$S.E. for groups of 5 animals were $.49\pm.05$ $\mu$g/g and $.52\pm.03$ $\mu$g/g, respectively. These values were significantly different from control (p values$<.01$). Hearts from 9 animals which had been pretreated with 10 mg/kg guanethidine exhibited a moderate reduction in the active tension developed during stretch when compared to that developed during stretch by control hearts (Fig. 2A). Average heart rate $\pm$S.E. was $41\pm1.2$/15 sec which was not significantly different from control ($p>.5$). The curve for 12 hearts from animals which had been treated with 20 mg/kg guanethidine was only slightly depressed as compared to the curve from untreated animals (Fig. 2A). Average heart rate $\pm$S.E. was $44\pm.70$/15 sec which did not differ significantly from control ($p=.1$).

Three mg/kg of reserpine phosphate lowered the norepinephrine content of the left

ventricle by approximately 94%. The content of norepinephrine per gram of left ventricle following reserpine pretreatment of 5 rabbits was .09±.02 μg/g which was significantly different from controls ($p<.001$). The active tension developed during stretch of 6 hearts was only slightly depressed by reserpine pretreatment (Fig. 2B). Average heart rate ± S.E. was 43 ± 1.3/15 sec, and did not differ significantly from control ($p>.3$).

*Discussion.* Although our data show that reduction in norepinephrine content of the left ventricle and reduction of tension development are concurrent events following 10 mg/kg of guanethidine, a higher dose of guanethidine produces equal reduction in catecholamine stores but only slight reduction in tension development. Apparently the reduction in tension development following 10 mg/kg guanethidine is independent of the depletion of the amines. One might postulate that depression of tension development is related to amine depletion, but that in the case of the higher dose of guanethidine, a second and dose-dependent action increases tension development. This might result in a nearly normal curve. Our evidence cannot rule out such a possibility. However, drastic reduction of the norepinephrine content of the left ventricle with reserpine leads to only minor reduction in the tension development by the myocardium. We conclude that as judged by our index of myocardial contractility, the left ventricle of isolated rabbit heart can maintain near normal contractility despite a marked diminution in its content of norepinephrine.

These results are in apparent conflict with those of Lee and Shideman(7) who hold the view that loss of catecholamines from the ventricle depresses the contractility of the cat papillary muscle. We cannot explain the discrepancy between our results except to assume that it must reside in the fact that we are using a different species, preparation and index of contractility in our studies. Our data help to support the view of Withrington and Zaimis(8) that the catecholamine depletion induced by reserpine is not responsible for its suppressant action on contractility. The data offer no comment on the opinion of these authors that doses of reserpine which reduce contractility do so by a cardiotoxic action.

*Summary.* Isolated hearts from 11 untreated rabbits, 9 pretreated with 10 mg/kg guanethidine, 12 pretreated with 20 mg/kg guanethidine and 6 pretreated with 3 mg/kg reserpine were subjected to graded, moderate stretch by means of a clip attached to the apex of the left ventricle. Active tension dedeloped by the ventricles as a function of stretch was measured. The low dose of guanethidine depressed tension development but reserpine and the higher dose of guanethidine did not interfere conspicuously with tension development. The drugs produced 70 to 94% reduction in catecholamine content of the left ventricle. It is concluded that drastic reduction in catecholamine content of the left ventricle of the rabbit does not necessarily result in severe impairment of its contractility.

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