UNIVERSITY OF OKLAHOMA MEDICAL CENTER

PRESERVATION OF MYOCARDIAL FUNCTION DURING CROSS-CIRCULATION IN TERMINAL ENDOTOXIN SHOCK

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13. ABSTRACT

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The pathogenesis of cardiovascular insufficiency in shock remains controversial and recent attention has been focused on a possible primary role of the heart. The question of a shock toxin initiating circulatory failure has been debated since the reports of Cannon[3], and the cardiotoxic theory was revived by the reports of Solis and Downing[18], and Lefer and co-workers[14,15] who described a circulating myocardial depressant factor. However, studies from this laboratory[7,9,10] and others[6] have shown normal myocardial performance in the early phase of endotoxin shock and no adverse effects on cardiac work or metabolism during cross-circulation with animals in intermediate stages of endotoxin shock. There is agreement on the ultimate depression of myocardial function which has been demonstrated in experimental animals following prolonged hemorrhagic[16] or endotoxin[18] shock. Similar observations have been reported in patients who show signs of left ventricular failure in the later stages of clinical septic shock[17]. These experiments were designed to measure cardiac performance in a normal heart perfused with blood from a dog in the later or terminal stages of endotoxin shock since the concentration of the depressant factor is believed to increase with time[15].

METHODS

Twenty-two healthy adult mongrel dogs were administered an LD_{60} of purified E. coli endotoxin (Difco, Detroit) the afternoon of the day prior to study and returned to their cages with access to water but without further treatment. The nine animals alive the following morning (18-21 hours later) were anesthetized with small amounts of pentobarbital (10-20 mgm/kg). An endotracheal tube was inserted and cannulae were positioned in the aorta and inferior vena cava through the femoral artery and vein. Body temperature

was maintained by circulating blankets and maintenance fluids were administered intravenously. Four normal untreated dogs were similarly prepared and served as control animals.

Thirteen normal dogs weighing 19-28 kg were anesthetized with pentobarbital (30 mgm/kg) and ventilated with a volume-cycled respirator. The great vessels in the mediastinum were isolated through a median sternotomy and the azygous vein and left subclavian artery ligated. Each animal was heparinized (300u/kg), the vagi were divided in the neck, and the brachio-cephalic artery was cannulated for retrograde perfusion of the aorta (Fig. 1). The right ventricle was cannulated through the right atrium and blood was collected to prime the perfusion circuit. Cross-circulation was commenced after ligation of the pulmonary artery, distal aorta, and both superior and inferior vena cavae. The isolated heart was perfused through the brachio-cephalic cannula and coronary perfusion pressure was maintained at mean levels of 120-130 mmHg by use of an atraumatic occlusive roller pump. With evidence of good cross-circulation without pressure drop in either the host dog or the isolated heart aorta, the thoracic contents were removed en bloc and suspended on an external frame by the tracheal cannula. The lungs of the isolated preparation were not ventilated and the host dog was allowed to breathe spontaneously.

The mitral valve of the isolated heart was excised through a left atriotomy and a fenestrated plug inserted with several interrupted sutures. A cannula was inserted in the left atrium during closure and drained to the venous reservoir along with coronary return from the right ventricle. Coronary blood flow was measured, warmed and returned by pump to the host animal. Isovolumetric left ventricular function was measured by a latex balloon cannula inserted through a small incision in the apex of the left ventricle and connected to two pressure transducers for measurement of end-diastolic (0-40 mmHg

scale) and left ventricular pressures (0-200 mmHg scale). The first derivative of left ventricular pressure (dp/dt) was recorded simultaneously using a resistance-capacitance differentiating network. The sino-atrial node was crushed and heart rate controlled using a pacemaker at a rate of 170/min. Temperature of the isolated heart was measured by a venacaval thermistor probe and maintained at 37° ± 1°C.

Coronary arterial and venous blood PO_2, PCO_2, and pH were measured by an Instrumentation Laboratories blood gas analyzer (Model 113) calibrated prior to each determination with known gas mixtures. Oxygen content was measured directly in a Lex-O_2-con analyzer with periodic confirmation by the Van Slyke manometric technic. Arteriovenous oxygen and CO_2 differences were used to determine myocardial O_2 uptake (MVO_2) and CO_2 output based on measured coronary flow drained from both left and right ventricles while the left ventricular balloon was distended with 5 ml of saline. Coronary perfusion pressure was maintained by pump adjustment to mean values above 120 mmHg throughout the study which was consistantly higher than the intraventricular isometric peak pressure.

Calculations of force-velocity and length-tension curves were based on the studies of Enright et. al[5] assuming the ventricle to be a thick-walled sphere:

$$F = \frac{Pr_e^2}{(r_o^2 - r_e^2)} \times 1.36$$ where F = force (g-wt/cm^2), P = LV pressure (peak systolic) or end-diastolic), r_e = endocardial ventricular radius (cm), r_o = epicardial radius (cm) and 1.36 = conversion factor. Internal ventricular radius (r_e) was determined from the volume of the intraventricular balloon using the equation $V = 4/3 \quad r^3$ and the maximal volume used to determine

maximal radius for plotting the length-tension curve as a percent of the maximum length. The external radius r_o was calculated from the sum of the volumes of the balloon and left ventricular muscle mass assuming the specific gravity of the muscle mass to be one. Velocity of the contractile element (V_{CE}) was calculated from the equation $V_{CE} = \frac{dp/dt}{2\delta p} \times 2\ r\ (mid)$ where P = ventricular pressure (mmHg).

Assessment of myocardial "efficiency" was made by the relationship between pressure "work" (systolic pressure-diastolic pressure) and oxygen uptake/100gm LV mass measured directly by wet weight of the trimmed left ventricle at the termination of each study:

$$\text{"efficiency"} = \frac{P_S - P_D}{(2.05)\ (MVO_2/100gm\ LV)}$$

Data were analyzed statistically by Student's t-test and the criterion for significance based on $p < 0.05$.

RESULTS

Survivors of the LD_{60} of purified endotoxin showed blood diarrhea, rapid respiration, and were unable to stand. Of 22 dogs injected, only 9 survived long enough to be connected to the isolated heart preparation and 2 died during the period of study. Arterial blood gas and pH analysis showed a compensated metabolic acidosis with mean pH of 7.33 ± .03, Pa CO_2 of 27.3±1.7 mmHg and PaO_2 75.5±2.6 mmHg (Table I). These values did not change during perfusion of the isolated heart in animals surviving the study period but there was a fall in PaO_2 and pH in dogs which died during the study. Mean systemic arterial pressure (MSAP) in the endotoxin group averaged 95 mmHg in contrast to MSAP of 130 mmHg in the normal donor dogs prior to cross-circulation. After 60 minutes of cross-circulation, MSAP increased to 101 mmHg but this improvement was not

sustained with MSAP falling to 85 mmHg at 120 minutes and 75 mmHg at 180 minutes (Fig. 2).

Control animals showed moderate hypoxia under pentobarbital anesthesia without ventilatory assistance (Table I). No significant change in blood gases or pH occurred during the perfusion study and MSAP was maintained except for one dog where a fall from 103 mmHg to 70 mmHg was noted after three hours.

Cardiac performance in response to stepwise distention of the intraventricular balloon showed improvement in peak systolic force in control dogs after one hour of perfusion which then stabilized for the remainder of the study (Table II). Peak systolic isometric force in the endotoxin group showed a gradual but steady improvement over the three hour perfusion period increasing from 61.3 ± 9.1 to 81.5 ± 21.2 g-wt/cm^2. Force-velocity curves in control dogs showed no significant change and coronary blood flow did not change during the study period.

Isometric length-tension curves showed consistently better myocardial function in the endotoxin group although the differences were not statistically significant (Fig. 3). There was little deterioration with time and at 180 minutes, the endotoxin group function curves remained better than control although the number of determinations was too low for statistical significance (Fig. 4). Similar stability of the force-velocity curves was noted in the endotoxin group which did not differ from each other or from control dogs (Fig. 5). Measured coronary blood flow tended to increase with time during the study period in the hearts perfused with blood from dogs in endotoxin shock but the increase was not statistically significant.

Calculated pressure work decreased slightly in controls while myocardial

O_2 uptake (MVO_2) remained unchanged for a slight but insignificant decrease in calculated myocardial efficiency at the end of the three hour period of study (Fig. 6). In the endotoxin group, both pressure work and MVO_2 increased during the study resulting in no significant change in myocardial efficiency.

DISCUSSION

Assessment of the role of the heart in irreversible shock following a lethal dose of endotoxin is complicated by peripheral vascular events which decrease venous return and reduce cardiac output[8,12]. These effects can be removed by employing an isolated heart preparation to which venous return from a host animal is maintained constant by pump perfusion. Using this preparation, we demonstrated in previous studies that the heart shows no adverse effects in response to direct administration of endotoxin[7]. Similarly, Weil and others[20] found no evidence of myocardial damage after endotoxin, Kutner and Cohen[13] reported no alteration of myocardial contractility after endotoxin, and Alican and co-workers[1] found resistance of the myocardium to endotoxin when arterial pressure was maintained. Even when cardiac output and mean aortic pressure were decreased by pump adjustment to match the host dogs' hypotension for three hours[7], cardiac power resumed control values after restoration of control flows and aortic pressure. These findings are in agreement with Siegel and Downing[16] who reported myocardial damage only after prolonged hemorrhagic hypotension. No evidence has been obtained to support recent observations by Lefer and co-workers[14,15] in hemorrhagic and endotoxin shock that a circulating myocardial depressant factor is released from the splanchnic bed which impairs cardiac function directly.

Further studies of a normal heart cross-circulated with a dog in later stages of endotoxin shock (6-9 hours) similarly showed no alteration of cardiac performance in response to afterloading[10]. The possibility that the adrenergic response of the host dog masked a depressive effect on the heart was tested by beta-adrenergic blockade which also failed to reveal a deleterious effect[9]. However we have observed left ventricular failure in animals in endotoxin shock after a minimum of 4-6 hours of hypotension[11]. Therefore, this group of animals in later shock was selected in the present study to provide exposure of the isolated normal heart to the highest levels of the postulated myocardial depressant factor.

Instead of a decrease in cardiac performance, a trend towards improved cardiac performance was noted in length-tension curves and pressure work consistent with the effects of circulating catecholamines from the host dog as proposed by Goodyer[6]. However there was no improvement in contractility demonstrated in the force-velocity curves and no alteration in myocardial efficiency.

The possibility of a circulating toxic substance depressing myocardial function early in shock is an important therapeutic consideration which would imply restriction of infusion fluids. However we find no evidence that the heart cannot respond to a fluid load early in shock. After 4-6 hours, the combination of prolonged coronary underperfusion, neural dysfunction from cerebral hypoxia[2], interference with cellular metabolism[19] and coronary vascular obstruction[4] presumably acts to overcome compensatory mechanisms and finally results in cardiac failure.

SUMMARY

Cross-circulation between normal isolated hearts and dogs in terminal shock 18-21 hours after administration of endotoxin was performed for a period of 3 hours. Of 22 dogs injected with endotoxin, only 9 survived long enough to be studied and 2 died during the study. The results were compared to four normal control dogs. Isometric cardiac performance measured by intraventricular balloon distention was not impaired in the endotoxin group which showed consistently better length-tension curves and pressure work than control dogs. No alteration in force-velocity curves was noted in either group. Increases in both oxygen uptake and pressure work in the endotoxin group resulted in no change in calculated myocardial efficiency, both effects presumably a result of increased circulating catecholamines. Results fail to show any deleterious effect on a normal heart perfused with blood from a dog in terminal shock and do not substantiate a primary role for the reported myocardial depressant factor.

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TABLE I

CORONARY ARTERIAL AND VENOUS BLOOD GAS AND pH DETERMINATIONS DURING CROSS-CIRCULATION WITH DOGS IN TERMINAL ENDOTOXIN SHOCK

CONTROL (N = 4)

		TIME (MIN.) 0	60	120	180
Arterial	pH	7.43±.08	7.41±.06	7.43±.02	7.43±.02
	P_{O_2}	55.3±4.4	51.0±4.0	53.0±4.9	52.5±11.5
	P_{CO_2}	28.2±7.5	28.7±7.7	25.5±6.8	28.7±4.2
	C_{O_2}	18.0±0.5	17.7±0.8	17.3±0.6	16.8±0.8
	C_{CO_2}	31.4±3.9	3.10±3.7	30.4±4.9	32.9±2.6
Venous	pH	7.40±.07	7.39±.05	7.40±.02	7.39±.01
	P_{O_2}	33.3±5.2	32.3±3.3	30.3±5.2	32.5±4.5
	P_{CO_2}	30.3±6.3	32.0±6.1	28.3±4.2	31.5±2.5
	C_{O_2}	13.0±0.8	12.4±1.1	11.1±1.8	11.3±0.5
	C_{CO_2}	36.5±2.8	35.8±3.3	36.3±3.6	37.4±2.8

ENDOTOXIN (N = 9)

Arterial	pH	7.33±.02	7.35±.04	7.38±.05	7.28±.06
	P_{O_2}	75.5±2.6	74.5±1.6	74.0±3.3	70.3±5.4
	P_{CO_2}	27.3±1.7	27.0±1.7	25.3±0.6	27.0±3.9
	C_{O_2}	17.9±1.2	17.9±1.4	16.8±2.7	20.3±2.2
	C_{CO_2}	28.7±1.9	28.5±2.2	28.8±2.3	25.2±0.9
Venous	pH	7.32±.03	7.31±.03	7.33±.03	7.26±.05
	P_{O_2}	36.8±1.7	35.6±2.5	34.5±2.8	37.7±5.5
	P_{CO_2}	32.5±2.0	32.3±1.9	31.0±1.0	34.2±2.0
	C_{O_2}	11.7±0.9	10.9±0.8	9.5±1.9	12.6±1.9
	C_{CO_2}	33.4±2.0	34.0±2.3	34.2±2.6	32.5±1.9

Values = mean ± S.D

P = gas tension in mmHg

C = gas capacitance in ml.

TABLE II

PEAK SYSTOLIC ISOMETRIC FORCE (Fs) GENERATED DURING MAXIMAL BALLOON INFLATION IN ISOLATED HEARTS CROSS-CIRCULATED WITH NORMAL OR ENDOTOXIN SHOCKED DOGS

	TIME (MIN.)			
Fs (gm-wt/cm^2)	0	60	120	180
Control (N = 4)	38.7±11.1	57.0±15.2	51.5±10.6	44.6±12.0
Endotoxin (N = 9)	61.3± 9.1	64.1± 9.0	66.7±12.8	81.5±21.2

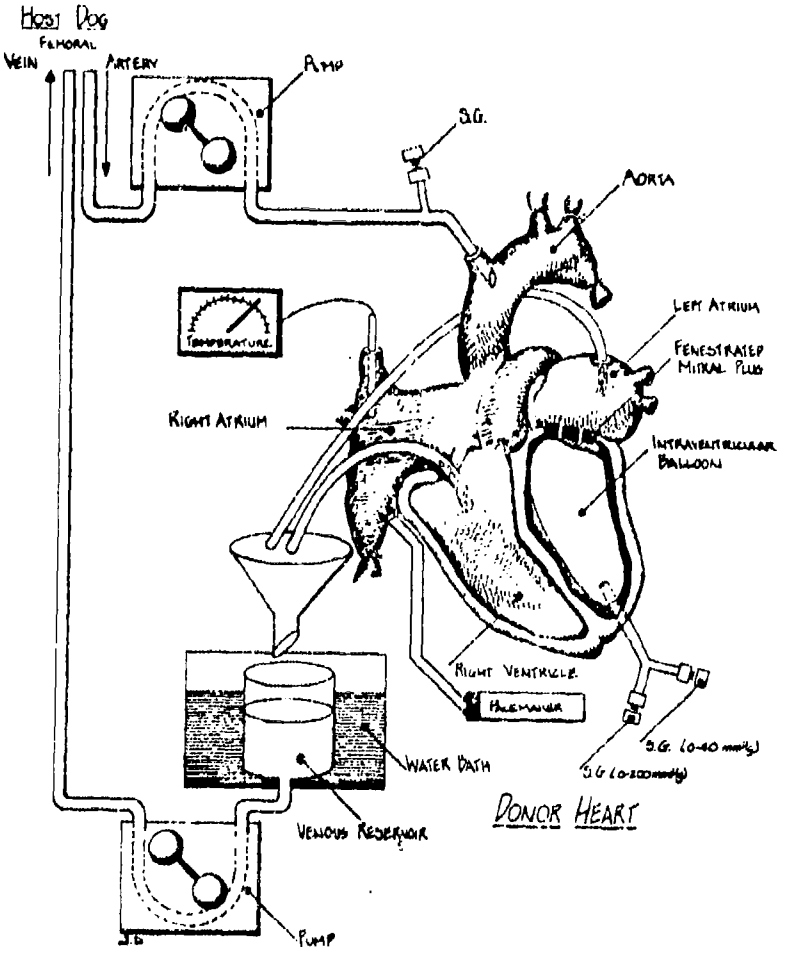

Figure 1. Schematic diagram of the isolated heart preparation permitting cross-circulation with a dog in endotoxin shock through the femoral artery and vein. Retrograde perfusion of the aorta is shown with coronary venous return from the right and left heart returned to a reservoir and then to the host dog. Left ventricular function is assessed by stepwise distention of the intraventricular balloon.

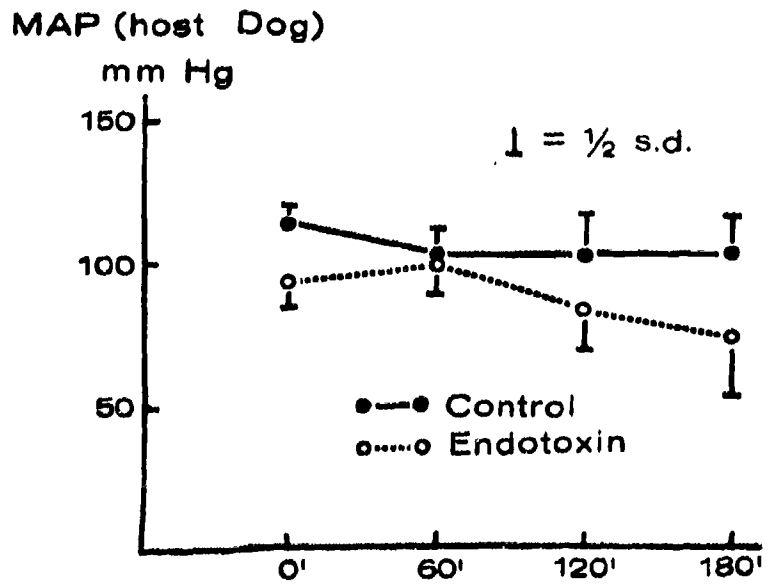

Figure 2. Mean systemic arterial pressure in control and endotoxin shocked dogs during the study period. After transient improvement during cross-circulation in the endotoxin group, there was a progressive fall in mean pressure.

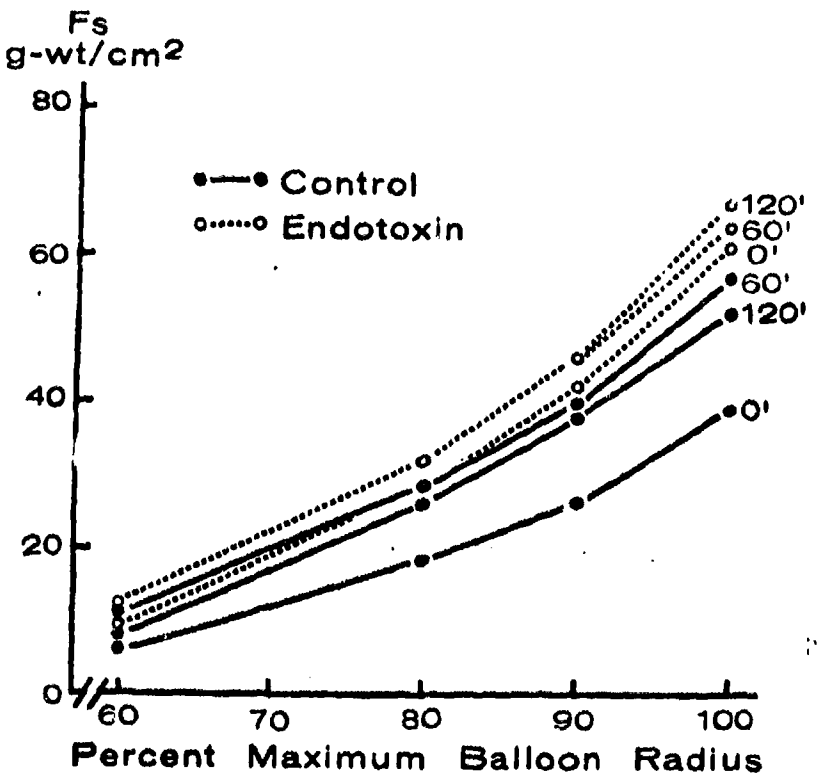

Figure 3. Length-tension curves showing the relationship between systolic forces (Fs) on the ordinate and increasing balloon size on the abscissa repeated at hourly intervals during cross-circulation. Although the endotoxin group (n = 9) shows higher peak isometric force than control (n = 4) the differences were not statistically significant.

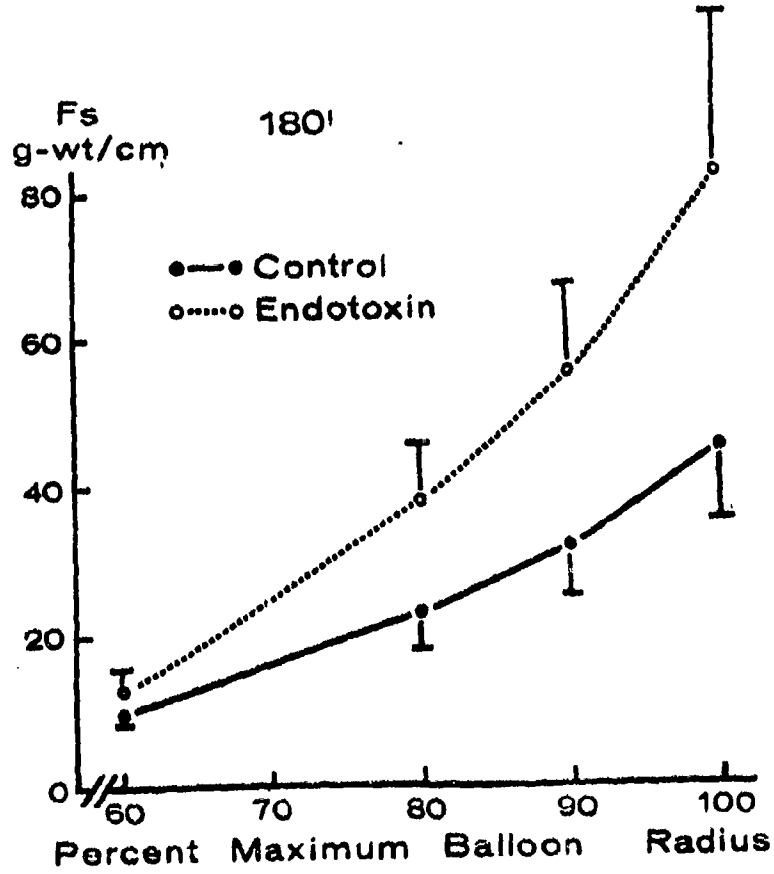

Figure 4. Length tension curves showing the relationship between peak systolic force (Fs) and increasing balloon radius after 180 minutes of perfusion. The mean Fs is consistantly higher in the endotoxin group than in the control dogs although the number of studies is too small for statistical significance.

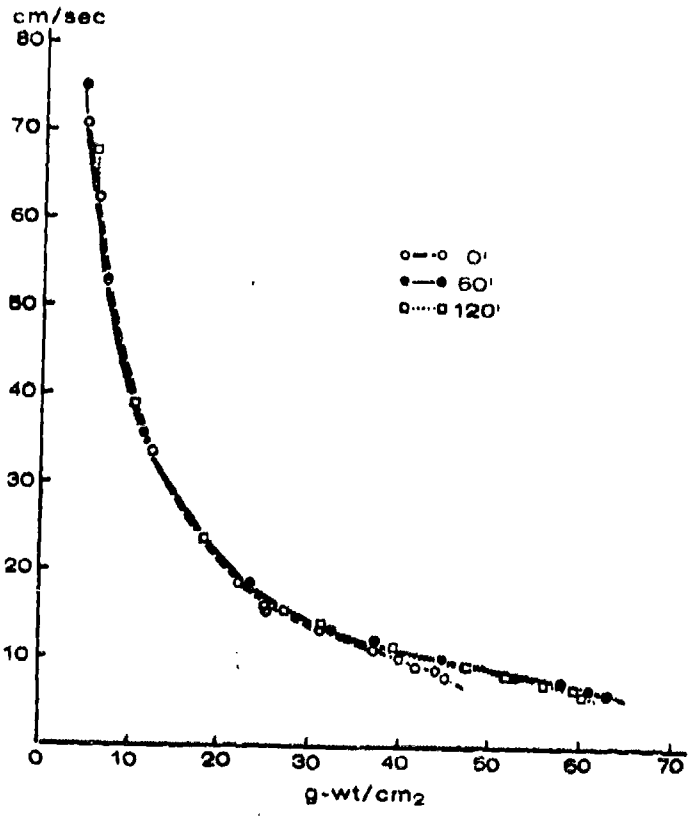

Figure 5. Force-velocity curves inscribed in a typical isolated heart preparation perfused with blood from a dog in terminal endotoxin shock. No significant change is noted at the same intraventricular volume (15cc) during the study period.

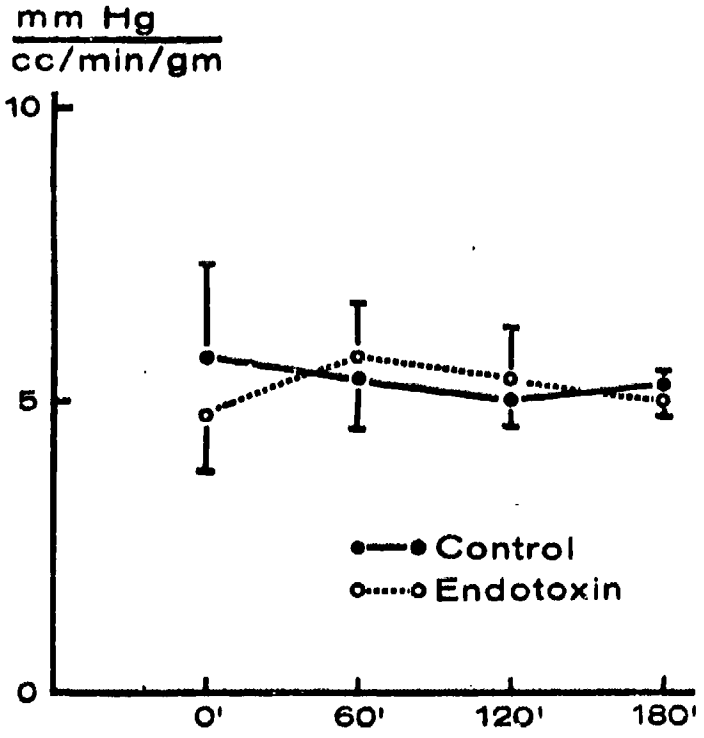

Figure 6. Calculated myocardial efficiency based on the ratio between pressure work and oxygen uptake of the ventricle in control and endotoxin perfused hearts. No significant differences were noted between the groups or with time although a proportional increase in both pressure work and oxygen uptake was noted in the endotoxin group.