A MECHANICAL MODEL OF MITRAL INSUFFICIENCY USING CANINE PAPILLARY MUSCLE PREPARATION

KOUICHI TAMIYA, M.D., MOTOAKI SUGAWARA, Ph. D.
AND RONALD W. MILLARD, Ph.D.*

A canine papillary muscle is loaded to mimic the load of the myocardium in the wall of the left ventricle with atrio-ventricular valvular insufficiency. This mechanical model which simulates the atrio-ventricular valvular regurgitation is based on two simple assumptions. The assumptions arranged for the papillary muscle experiment are as follows: 1) the force that the myocardium encounters during muscle shortening is proportional to the muscle shortening velocity due to regurgitation through a narrow regurgitant orifice; and 2) the myocardium exerts a constant force while the aortic valve is open. The muscle shortening except the isotonic phase is ascribed solely to regurgitation since the aortic valve is closed during these phases. In the combined antegrade/retrograde ejection phase, which is characterized by a constant muscle force, the shortening velocity due to regurgitation is constant because of the assumed functional relation between the muscle force and shortening velocity. The amount of shortening assigned to regurgitation in this phase is given by the product of the velocity at the beginning point of the isotonic phase and the duration of this phase. The present in-vitro studies offer an alternative explanation for decrease in the regurgitant fraction as total load was reduced at a constant preload. The regurgitant fraction decreased as preload was increased at a constant total load in the present study. The regurgitant fraction also decreased by either isoproterenol or $CaCl_2$ administration via the coronary artery.

IN the management of clinically significant atrioventricular valvular insufficiency, the degree of regurgitation has frequently been assumed to depend solely on the anatomical dimensions of the regurgitant valve orifice and the pressure gradient across the valve. Recently, the beneficial effects of reduction in afterload by the administration of vasodilators on the degree of valvular regurgitation have been demonstrated in both acute experimental studies[1-2] and clinical studies[3-7] Salutary effects of positive inotropic agents on the degree of valvular regurgitation have also been studied[1-3] The administration of these agents produces simultaneous changes in hemodynamic parameters including preload, total load, ventricular volume and so on. Thus a comprehensive understanding of mechanisms that regulate the degree of mitral regurgi-

Key Words:
Mitral regurgitation
Regurgitant fraction
Papillary muscle experiment
"Isometroid" contraction
"Isometroid" relaxation

(Received May 12, 1983; accepted October 27, 1983)
Department of Surgical Science, Heart Institute Japan, Tokyo Women's Medical College, Tokyo Japan
**Department of Pharmacology and Cell Biophysics, University of Cincinnati, College of Medicine Cincinnati, Ohio 45267, U.S.A.*
This study was supported in part by grants-in-aid, Project 57570361, 1982, from the Ministry of Education of Japan
Mailing address: Kouichi Tamiya, M.D., Department of Surgical Science, Heart Institute Japan, Tokyo Women's Medical College, 10 Kawadacho, Shinjukuku, Tokyo 162, Japan

NII-Electronic Library Service

tation is difficult.

In a normal heart, ventricular contraction is divided into two phases. The first phase of ventricular contraction should be isovolumic. In atrioventricular valvular incompetence, however, substantial changes in the ventricular volume due to regurgitation occur in this phase. This phase was named "an isometroid contraction phase" by Rodbard and Williams.[8] The second phase, where ventricular pressure exceeds aortic or pulmonary arterial pressures, is characterized by a combined antegrade/retrograde ejection when atrioventricular valvular insufficiency exists.

In terms of fluid dynamics, the retrograde ejection into the atrium (regurgitation) is quite different from that directed antegrade into the aorta or pulmonary artery. In contrast to antegrade ejection, which occurs with negligible pressure gradient between the ventricle and the aorta, the retrograde blood flow through the narrow regurgitant valve orifice requires a substantial pressure gradient between the ventricle and the atrium throughout systole. In the presence of atrioventricular valvular insufficiency, if the ventricle were to contract very slowly, the intraventricular pressure might never exceed aortic pressure due to substantial retrograde blood flow into the atrium. Therefore, in such an extreme case, it is possible that the blood in the ventricle would be ejected only into the atrium and no antegrade ejection would occur even if the regurgitant valve orifice were small. This idea was originally introduced by Wiggers and Feil[9] and led us to consider an analytical model of atrioventricular valvular insufficiency using the isolated heart muscle preparation. Sonnenblick and Parmley[10] proposed an in vitro model of mitral regurgitation and ventricular aneurysm using cat papillary muscles. In their model, however, a spring was placed in series with the papillary muscle, connecting it to the tip of the isotonic lever system. This model simulates the expected increase in series elasticity of the myocardium in mitral regurgitation or ventricular aneurysm in the whole ventricle, but does not simulate the hemodynamic situation of mitral regurgitation. The pressure loss across a mitral regurgitant orifice is produced by an energy dissipating process in which kinetic energy is transformed into heat after all. However, a spring does not dissipate kinetic energy, but store it as potential energy. In the present simulation of the valvular regurgitation, two assumptions are required: 1) the force that the myocardium exerts during the isometroid contraction and isometroid relaxation phases is proportional to the velocity of muscle shortening; and 2) the force that the myocardium develops (encounters) during the combined antegrade/retrograde ejection phase is constant. According to the fundamental notions of fluid mechanics, the pressure gradient in a steady flow across an orifice is proportional to a constant power of the flow velocity. This power increases from 1 to 2 as the flow velocity, i.e. the Reynolds number, increases. This power depends also on the shape of the orifice (divergent angle, area of the narrowest portion, etc.). In the present experiment, a linear relation between the pressure gradient and the flow velocity (that must have a direct connection with muscle shortening velocity) through the incompetent valve during the isometroid contraction and isometroid relaxation phases was adopted, as a first order approximation.

The second assumption, derived from the analogy between quasi-isobaric contraction phase of the ventricle in vivo and the isotonic contraction in the papillary muscle experiment, introduces some problems. In an atrioventricular valvular insufficiency, there would be substantial changes in ventricular diameter during contraction due to the increased stroke volume. Such a ventricular volume change would lead to decreasing muscle stress, even if the intraventricular pressure remained constant.

Despite these considerations we adopted the isotonic contraction as the analogue of ventricular ejection phase with atrioventricular valvular insufficiency. The reasons for it are: 1) excessive complexity diminishes the value of the experimental mechanical model; 2) the isotonic contraction has been conventionally adopted as an analogue of the ventricular ejection phase in the field of cardiac muscle mechanics. Thus, the present study was undertaken to evaluate the effects of changes in myocardial contractility and loading conditions on the relationship between the extent of muscle shortening ascribed to antegrade ejection and that ascribed to retrograde ejection.

METHODS

Blood perfused papillary muscles of the dog right ventricle were used for all the experiments. The specimen muscles were mounted vertically in a double walled glassware to insure constant

NII-Electronic Library Service

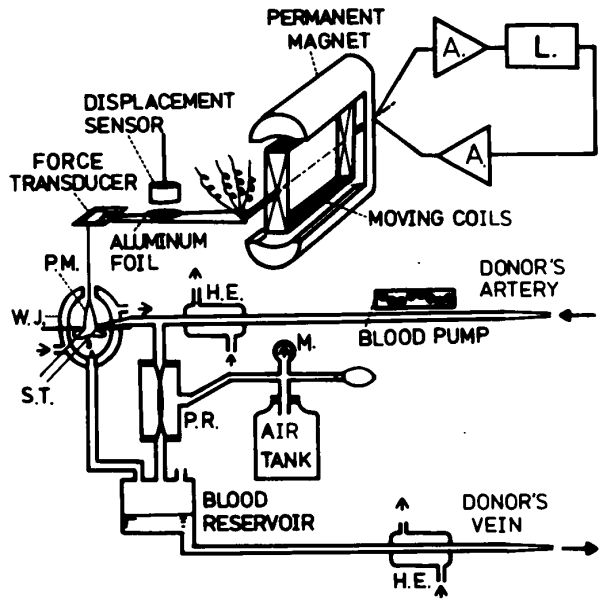

Fig.1. Experimental setup.
A. = amplifier; H.E. = heat exchanger; P.M. = papillary muscle; L = signal limiter; M. = manometer; P.R. = pneumatic resistor; S.T. = stimulator; W.J. = water jackets.
Aluminium foil was used as a target for the non-contacting displacement sensor.

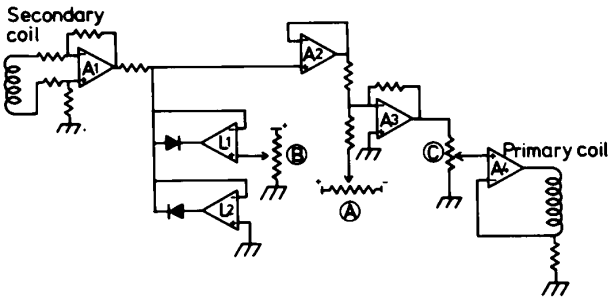

Fig.2. Circuit diagram of the force control system. A1–A4, L1–L2 = operational amplifiers; Ⓐ = preload adjusting potentiometer; Ⓑ = total load adjusting potentiometer; Ⓒ = feed-back gain adjuster.

muscle temperature (37°C) and to maintain adequate muscle surface hydration. The muscle was perfused with arterial blood from a donor dog at a constant pressure (13.3 kPa ≒ 100 mmHg) by a crosscirculation technique.[11] The tendinous end of the muscle was connected to the strain gauge force transducer (compliance $< 6 \times 10^{-4}$ $m \cdot N^{-1}$) by a short segment of fine copper wire (diameter 0.08 mm) and the force transducer was attached at the tip of the aluminium lever. A non-contacting displacement measuring system that senses the position of the aluminium lever was used to measure the changes in muscle length. The aluminium lever was attached to the shaft of the electromagnetic force generating system as described more in detail in the previous publication of ours.[12]

To impose a velocity proportional force on the muscle during the isometroid contraction phase and the isometroid relaxation phase, a secondary coil was installed in the moving coil that was suspended in a strong magnetic field. The output signal of the secondary coil (proportional to the angular velocity of the coils and shortening velocity of the muscle) was returned to the force generating system. To provide a constant force on the muscle during the isotonic contraction phase (combined ejection phase) and the isotonic relaxation phases (ventricular filling phase), two signal limiting controls were utilized.

Electromagnetic Lever System: A simplified diagram of the equipment used in this study is shown in Fig. 1. The aluminium lever, permanent magnet, moving coils and the measuring system for muscle force and muscle length are the same as that described in the previous report.[12] The total equivalent mass of the lever system including thin aluminium target (thickness 0.1 cm, diameter 10 mm) for non-contacting displacement measuring system (KAMMAN KD-2300 2S SPL) was measured. In free oscillation with a light spring,[12] the frequency of oscillation f (Hz), amplitude of displacement S (cm) and force F (N) acting on the tip of the lever system were 13.2 Hz, 0.22 cm and 9.8×10^{-3} N, respectively. The moving equivalent mass of the total system ($F/4\pi^2 f^2 S$) was calculated as 650 mg (mass). This moving equivalent mass is available only during both the isotonic contraction and isotonic relaxation phases. During the isometroid contraction and isometroid relaxation phases, the movement of the lever is controlled to achieve the proportional relation between the muscle shortening velocity and the force imposed on the muscle.

Control Unit: The control unit consists of a current amplifier and two signal limiters. The circuit diagram of the system is shown in Fig. 2. No signal is generated in the secondary coil when the moving coil is quiescent. The DC level off-set imbalance of the current amplifier provides an adjustable output current for the primary coil. In other words, the current through the primary coil when the moving coil is stationary provides an adjustable preload through an adjustment of

NII-Electronic Library Service

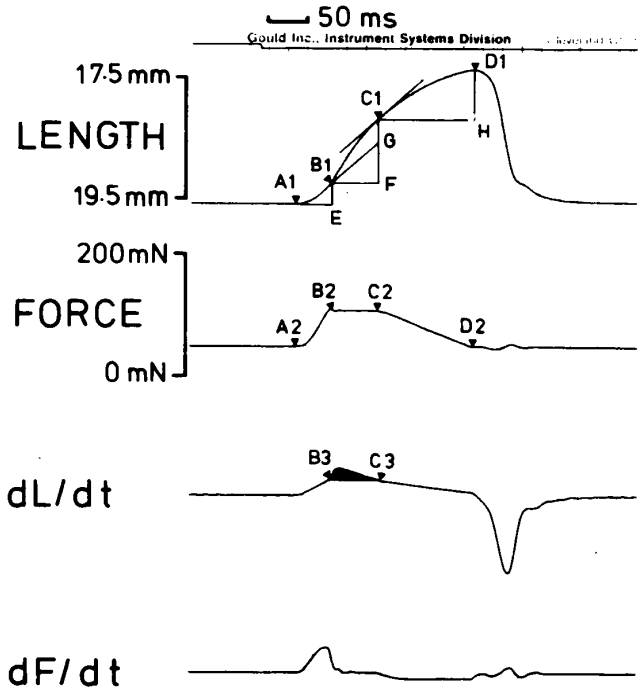

Fig.3. Representative tracings of muscle length, force and their first derivatives with respect to time (dL/dt and dF/dt).
Key to lettered/numbered points: A1 = initiation of muscle shortening; A2 = preset muscle preload; B1, B2, B3 = end point of isometroid contraction phase and beginning point of isotonic contraction phase on length, force and velocity records, respectively; C1, C2, C3 = end point of isotonic contraction phase and beginning point of isometroid relaxation phase on each record; D1 = shortest length of muscle; B1-E = amount of muscle shortening during isometroid contraction phase; C1-G = amount of muscle shortening corresponding to aortic ejection during isotonic contraction phase; G-F = amount of muscle shortening corresponding to mitral regurgitation during isotonic contraction phase; D1-H = amount of muscle shortening during isometroid relaxation phase

the off-set null potentiometer of the current amplifier. The initial lever position (initial fiber length of the muscle) is determined by both the force generated by the current amplifier DC imbalance and the stress-strain relationship of the resting cardiac muscle.

Figure 3 shows representative recordings of muscle length, force and their first derivatives with respect to time. Electrical stimulation causes the muscle to contract against a predetermined preload and begin to shorten (Al in Fig. 3). At the instant the muscle begins to shorten, the secondary coil in the strong magnetic field begins to move and generate a current (force) proportional to the angular velocity of the coils. This velocity signal is supplied to the current amplifier in order to add the velocity proportional force to preload. Thus the electromagnetic lever system provides afterload (developed force) that is directly proportional to the shortening velocity of the muscle during the isometroid contraction and relaxation phases. The proportionality constant between the velocity and the force was set at $4.9\ N \cdot m^{-1} \cdot s$ in the present study. The amount of muscle shortening (B1-E in Fig. 3. See the next section about points E, F, G, H.) against the velocity proportional force (A2-B2 in Fig. 3) during the isometroid contraction phase corresponds to the regurgitant volume flow in this phase since there is no ejection of blood into the aorta this phase.

The maximum signal level from the secondary coil is restricted to the preset level by the signal limiter, thus the current through the primary coil is maintained constant while the shortening velocity of the muscle exceeds the preset level. In other words, when the force imposed on the muscle (total load) reaches the preset level, isotonic contraction begins. Thus, isotonic shortening corresponding to the combined ejection phase in a whole ventricle with an incompetent atrioventricular valve occurs.

In the case of our mitral valvular insufficiency model, the left ventricular systolic pressure is nearly equal to the aortic pressure and relatively constant. Changes in left atrial pressure is disregarded because of enormous elasticity of the atrial wall. By this analogy, as long as the left ventricular (and left atrial) pressure is constant, the retrograde blood flow velocity through the regurgitant mitral orifice must be regulated to be constant by the functional relation between the pressure gradient and the flow velocity. Hence, the product of the rate of ventricular volume change at the beginning point of the combined ejection phase and the duration of this phase yields the amount of blood volume assigned to regurgitation during this phase. On the other hand, the total amount of blood ejection (that consists of blood ejected into the aorta and regurgitated into the left atrium) during the combined ejection phase is measured as ventricular volume change during this phase. Therefore, we can obtain the amount of antegrade blood ejection by subtracting regurgitant blood volume from the total amount of blood ejected from the

NII-Electronic Library Service

ventricle during the combined ejection phase.

In the case of the papillary muscle experiment that simulates the mitral valvular insufficiency, the muscle force is controlled to be constant during the isotonic (combined) ejection phase. While the force imposed on the muscle is kept constant, the amount of muscle shortening assigned to regurgitation is considered to be linear with respect to time, because the muscle shortening velocity assigned to regurgitation during that phase must be constant in order to simulate the constant retrograde blood flow velocity through the regurgitant mitral orifice. Thus the amount of muscle shortening that corresponds to the amount of regurgitation during combined ejection phase (G-F in Fig. 3) is given by the product of the isotonic time (B2-C2 in Fig. 3) and the shortening velocity of the muscle at the beginning point of the isotonic contraction phase (B1 in Fig. 3). The rest of the amount of muscle shortening (C1-G in Fig. 3) must be assigned to antegrade ejection.

At the instant that muscle shortening velocity becomes equal to initial velocity at the beginning point of the isotonic contraction phase (C1 in Fig. 3), the isometroid relaxation phase begins (C2 in Fig. 3). During the isometroid relaxation phase (C2-D2 in Fig. 3), the force imposed on the muscle declines inspite of the muscle shortening, and the amount of muscle shortening during this phase (D1-H in Fig. 3) is ascribed to regurgitation. At the instant that muscle shortening reaches its peak (muscle attains its shortest length, D1 in Fig. 3), the shortening velocity of the muscle becomes zero and velocity proportional force is no longer added to preload. Accordingly, the force imposed on the muscle at that point is precisely equal to the preset preload (D2 in Fig. 3).

At the instant the muscle begins to lengthen, a second signal limiter acts as a rectifier and restricts the output signal from the secondary coil to a positive range, such that no signal is applied to the electromagnetic lever system during the isotonic relaxation phase. Thus, the lever system keeps the force imposed on the muscle at preset preload level during the muscle lengthening in a fashion analogous to the intact ventricle without a stenotic atrioventricular valve lesion. In this way, the contraction and relaxation cycle of the simulated atrioventricular valvular regurgitation in the isolated cardiac muscle is accomplished.

Calculation of the Amount of Muscle Shortening: Since the force imposed on the muscle is proportional to the muscle shortening velocity, i.e. the first derivative of the muscle lenght with respect to time (dL/dt) during the isometroid contraction and relaxation phases, the shape of dL/dt curve is similar to that of muscle force curve during both the isometroid contraction and relaxation phases. A slope of the assumed linear shortening of the muscle that corresponds to regurgitation during the isotonic combined ejection phase (B1-G in Fig. 3) is equal to the height of point B3 in Fig. 3. Therefore the rest of muscle shortening that is considered to correspond to antegrade ejection (C1-G in Fig. 3) during the isotonic phase is equal to the area above line B3-C3 in Fig. 3 (hatched area). In the practical determination of the amounts of the muscle shortening that correspond to forward ejection into the aorta and the regurgitation to the left atrium during the isotonic combined ejection phase, we first find the end-point of the isometroid contraction phase (B3 in Fig. 3) and that of the isotonic contraction phase (C3 in Fig. 3) on the recording of dL/dt since they are easier to identify than those on the muscle length curve. Then we identify the corresponding points on the muscle length (B1 and C1 in Fig. 3). A tangential line is now drawn at point B1 and a perpendicular line at point C1 in Fig. 3. The intersection of the tangential and perpendicular lines is identified as point G. Intersections constructed by (1) horizontal extension of the muscle length curve at rest and the perpendicular line at point B1 in Fig. 3, (2) horizontal extension at the end of the isometroid contraction phase and the perpendicular line at point C1, (3) horizontal extension at the end of the isotonic contraction phase and the perpendicular line at point D1 are named points E, F and H, respectively. The distance between points B1 and E shows the amount of muscle shortening during the isometroid contraction phase. The distance between C1 and F denotes the amount of muscle shortening during the isotonic contraction phase. The distance between D1 and H denotes the amount of muscle shortening during the isometroid relaxation phase.

In the isotonic contraction phase, the distance between points C1 and G in Fig. 3 indicates the amount of muscle shortening corresponding to the forward ejection into the aorta. The distance between G and F is considered to be the amount

NII-Electronic Library Service

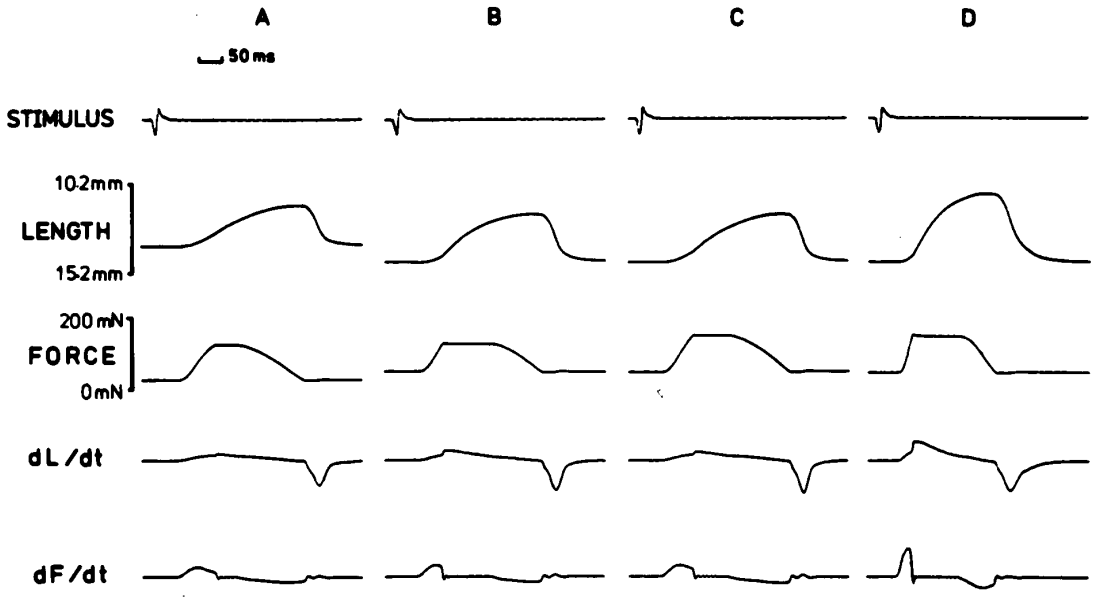

Fig.4. Representative recordings at (A) low preload and low total load, (B) high preload and low total lead, (C) high preload and high total load, and after (D) bolus injection of *l*-isoproterenol (4×10^{-9} mol) into perfusing blood. Note that there is almost no change in the total amount of muscle shortening including regurgitation between B and C.

of muscle shortening corresponding to the regurgitation into the atrium.

Stimulation of the Muscle: Most muscles were stimulated at a rate of 60 pulses per minute (1 Hz) with rectangular pulses of one msec duration, 10% above threshold (4 ± 2 V), while some were stimulated at higher rates in order to examine the effects of changes in contraction frequency on the mechanical parameters in the simulated atrioventricular valvular insufficiency. The stimuli were provided through two parallel electrodes at both sides of the base of the papillary muscle.

Preparation of the Papillary Muscle and Blood Donor Dog: The preparation of the papillary muscle and blood donor dog is similar to that of Endoh and Hashimoto[11] and have been described in detail previously.[13] Briefly, two dogs were used for each experiment, one to obtain the papillary muscle and its blood supply and the other as a blood donor dog. Both dogs were anesthetized (25 mg/kg body weight of pentobarbital), heparinized (1 mg/kg body weight of sodium heparin) and ventilated artificially. The carotid artery and external jugular vein of the donor dog were cannulated and connected to the arterial and vanous cannulas of the blood circuit, respectively, as shown in Fig. 1.

The dog to provide the papillary muscle specimen was thoracotomized, then the heart was excised and immediately plunged into cold saline at 4°C to inhibit spontaneous contraction. The anterior septal artery was exposed and cannulated with a fine polyethylene tube, and the right and left ventricular free walls were removed. All coronary artery branches except those perfusing the anterior papillary muscle region were ligated. The base of the papillary muscle was secured on the supporting platform by piercing the base of papillary muscle with parallel needles and the tendinous end of the muscle was connected to the strain gauge transducer with a short segment of fine copper wire (0.08 mm in diameter).

Blood Circuit: The blood circuit for the perfusion of the papillary muscle is shown in Fig. 1. A pneumatic resistor was utilized to obtain a constant perfusion pressure of the muscle. Water circulated double walled glassware and two heat exchangers were used to maintain the temperature of the perfusion blood and muscle at 37°C and prevent dryness of the surface of the muscle and hypothermia of the donor dog.

Recording System: The muscle force, muscle length and their electronically differentiated first

NII-Electronic Library Service

TABLE I PHYSICAL CHARACTERISTICS OF THE MUSCLES

Muscle	L max (mm)	Volume of muscles (mm^3)	Average cross sectional area (mm^2)	Developed stress at L max (kPa)	Maximum ΔL/L max
1	11.8	121.1	10.3	29.2	0.11
2	13.0	128.7	9.9	35.5	0.15
3	11.6	116.9	5.0	48.6	0.21
4	15.2	151.2	10.0	36.9	0.21
5	8.8	29.0	3.3	38.7	0.17
6	11.2	96.1	8.6	51.4	0.22
Mean ± SD	11.9 ± 2.1	107.2 ± 42.2	7.9 ± 3.0	40.1 ± 8.4	0.18 ± 0.04

ΔL is an amount of muscle shortening. L max is a resting muscle length at which isometrically developed force is maximum. (1 kPa = 1 mN/mm^2 ≒ 0.1 g/mm^2)

TABLE II FUNCTIONAL CORRELATES IN PAPILLARY MUSCLES

Muscle	Preload constant			Total load constant		
	Preload (kPa)	Total load (kPa)	Regurgitant fraction	Total load (kPa)	Preload (kPa)	Regurgitant fraction
1	8.0	10.8 – 17.9	0.63 – 0.90	13.0	4.5 – 9.4	0.90 – 0.58
2	7.0	10.9 – 14.9	0.77 – 0.93	15.4	7.4 – 11.0	0.93 – 0.78
3	8.6	10.4 – 21.5	0.46 – 0.92	19.6	6.3 – 14.9	0.83 – 0.56
4	4.9	9.6 – 17.2	0.53 – 0.94	9.3	2.6 – 5.1	0.90 – 0.54
5	9.5	12.0 – 20.9	0.29 – 0.90	20.9	6.6 – 17.2	0.92 – 0.34
6	8.5	10.9 – 17.2	0.55 – 0.94	17.2	7.0 – 10.2	0.97 – 0.86
Mean ± SD	7.8 ± 1.6	10.8 ± 0.8 – 18.3 ± 2.5	0.54 ± 0.16 – 0.92* ± 0.02	15.9 ± 4.3	5.7 ± 1.8 – 11.3 ± 4.3	0.91 ± 0.05 – 0.61** ± 0.19

** = statistically significant ($p < 0.001$); ** = statistically significant ($p < 0.01$)*
The mean regurgitant fraction values at lower total load (preload) and higher total load (preload) were compared. t-test for two mean values was employed for statistical analysis.

derivatives (dF/dt and dL/dt) with respect to time were recorded on a multi-channel strip chart recorder at a paper speed of 200 mm/sec (Hewlett Packard 7758A or Gould 200). The differentiators used have a time constant of 0.5 ms. The calibration procedures of the force and displacement transducer and their frequency characteristics were discussed in a previous report.[12]

Experimental Protocol: Electrical stimulation of the muscle was begun thirty minutes after the beginning of coronary perfusion of the specimen muscle with arterial blood from the donor dog. Muscle length, muscle force and their first derivatives with respect to time were recorded at various combinations of preload and total load in all the muscles demonstrating stable contractions (Fig. 4, A, B, C). The amount of muscle shortening, in terms of the fraction of the muscle length at which the muscle developed maximum isometric force (ΔL/L max), was measured and the portions assigned to 1) regurgitation during the isometroid contraction phase, 2) regurgitation during the combined ejection phase, 3) ejection into aorta during the combined ejection phase, 4) regurgitation during the isometroid relaxation phase were measured and calculated according to the procedures mentioned above. These experiments were carried out under conditions of 1) constant preload and varied total load, 2) varied preload and constant total load, 3) constant afterload. The effects of changes in the frequency of muscle contraction were evaluated over a range of 1.5–3 Hz in five muscles.

NII-Electronic Library Service

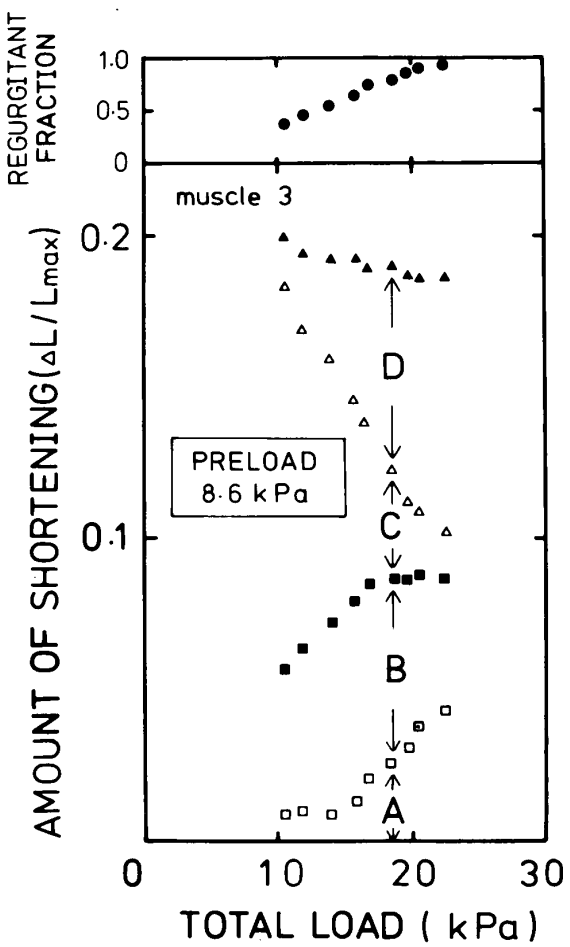

Fig.5. Relation between total load and the amount of muscle shortening assigned to: A = regurgitation during the isometroid contraction phase; B = regurgitation during the isotonic ejection phase; C = antegrade ejection during the isotonic ejection phase; D = regurgitation during the isometroid relaxation phase, under conditions of constant preload. Solid circles denote the value of regurgitant fraction (the ratio of the amount of muscle shortening assigned to regurgitation to the total amount of muscle shortening).

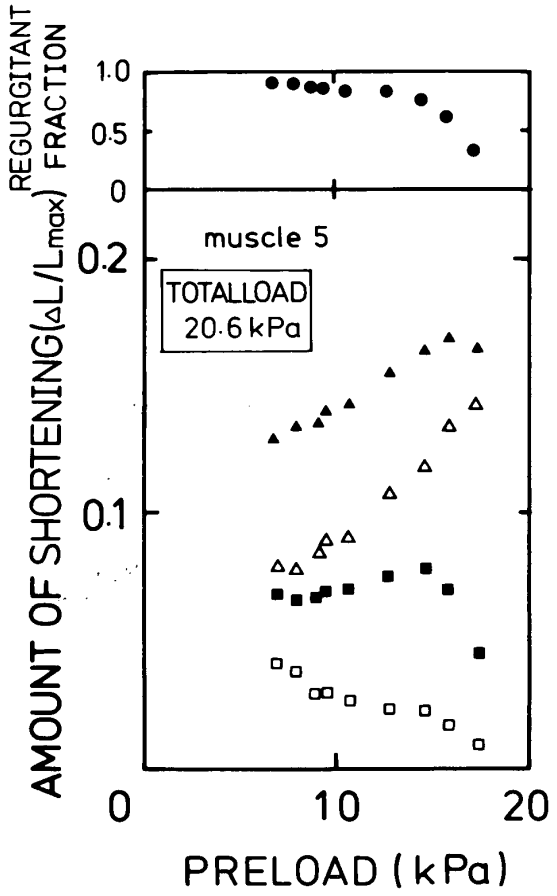

Fig.6. Relation between preload and the amount of muscle shortening under conditions of constant total load. Symbols are the same as those in Fig. 5.

To evaluate the effects of changes in contractile state of the myocardium on regurgitant fraction under conditions of constant frequency of contraction, bolus injections of *l*-isoproterenol (4×10^{-9} mol) and $CaCl_2$ (2.7×10^{-5} mol) were made into the perfusion blood through the arterial cannula under conditions of constant loads and contraction frequency of 1 Hz in four muscles. Representative recordings when contractility was enhanced by the administration of *l*-isoproterenol are shown in panel D of Fig. 4. Regurgitant fraction was calculated as the total amount of regurgitation divided by the total amount of muscle shortening.

Statistical Treatment of Data: Data obtained in this study were examined for statistical significance using Student's paired t-test with probabilities of less than 5% allowing rejection of the null hypothesis.

RESULTS

The physical characteristics of papillary muscles used in the present study are shown in Table I. The average crosssectional area (mean ± SD = 7.9 ± 3.0 mm^2) of each papillary muscle was calculated with the assumption that muscle in an elliptical cone. Representative tracings of muscle length, muscle force and their first derivatives with respect to time are shown in Fig. 4.

Effects of Changes in Total Load Under Conditions of Constant Preload on the Amount of Muscle Shortening Assigned to Regurgitation:

NII-Electronic Library Service

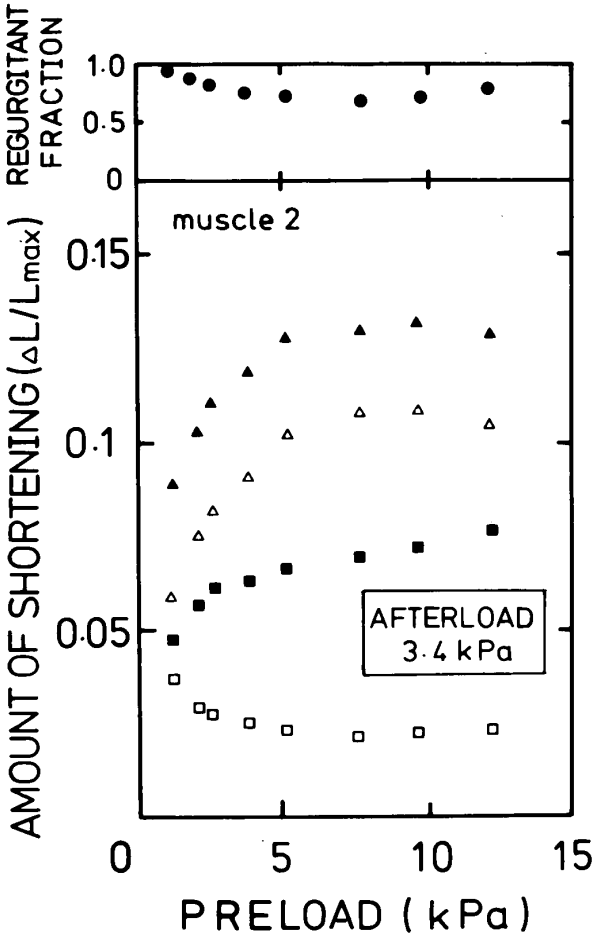

Fig.7. Relation between preload and the amount of muscle shortening under conditions of constant afterload (developed force). Symbols are the same as those in Fig. 5. Afterload = Total load – Preload.

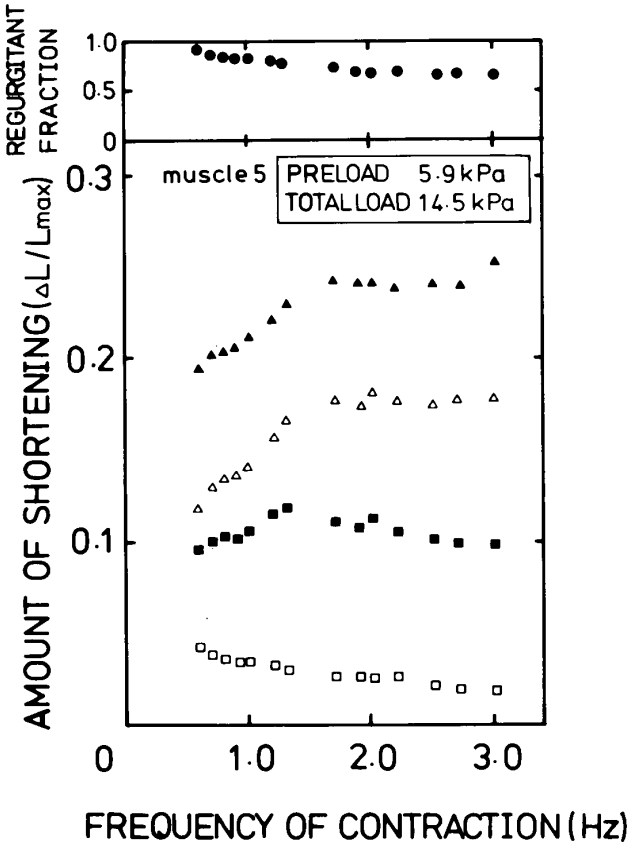

Fig.8. Relation between the frequency of contraction and the amount of muscle shortening under conditions of constant preload and total load. Symbols are the same as those in Fig. 5.

The representative data in Fig. 5 show the effects of changes in total load on these four portions. The changes in regurgitant fraction due to total load change are also shown in left side of Table II. When total load rose to 69% above control, regurgitant fraction increased by 70% as shown in the left side of Table II.

Effects of Changes in Preload Under Conditions of Constant Total Load on the Amount of Muscle Shortening Assigned to Regurgitation: Under conditions of constant total load, the increase in preload causes both a decrease in afterload (difference between total load and preload) and an increase in the maximum rate of force development (dF/dt_{max}). The former caused the decrease in the regurgitation during both the isometroid contraction and relaxation phases as shown in Fig. 5. The latter causes the decrease in regurgitation during the isometroid contraction phase and the increase in the antegrade ejection as shown in Fig. 9. Thus a two-fold increase in preload was accompanied by a 33% reduction in regurgitant fraction as shown in the right side of Table II. The representative results of increasing preload are shown in Fig. 6.

Effects of Changes in Preload Under Conditions of Constant Afterload on the Amount of Muscle Shortening Assigned to Regurgitation: When developed force was kept constant (that is, the slope of the line G-B1 in Fig. 3 was constant), the increase in preload produced an increase in total load that tended to increase the regurgitant fraction. On the other hand, the increased preload tended to reduce the regurgitation during the isometroid contraction phase and increase the regurgitation during the combined ejection phase (Fig. 6). As a net result, the regurgitant fraction may be reduced in a certain range of preload (Fig. 7).

Effects of Contraction Frequency on the Amount of Muscle Shortening Assigned to Re-

NII-Electronic Library Service

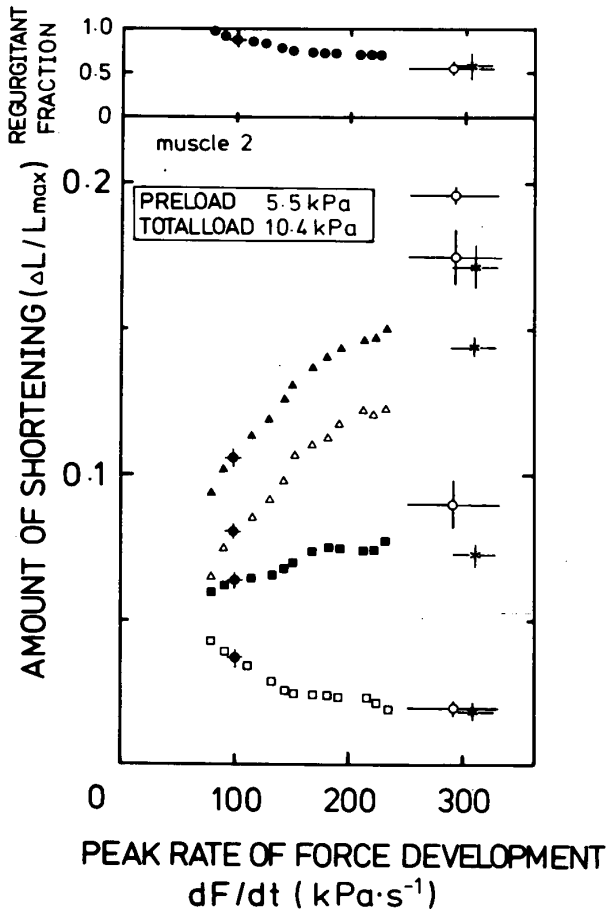

Fig.9. Relation between dF/dt_{max} and the amount of muscle shortening. ✦ denotes the control at a frequency of contraction of 1 Hz. ○ and ∗ denote the amount of muscle shortening at 1 Hz contraction after the administration of L-isoproterenol (4×10^{-9} mol) and $CaCl_2$ (2.7×10^{-5} mol), respectively. Bars indicate standard deviation.

gurgitation: An increase in contraction frequency enhances contractile state of the cardiac muscle as evidenced by an increase in the maximum rate of force development (dF/dt_{max}). The relationship between the contraction frequency and the shortening assigned to regurgitation is shown in Fig. 8. The control data were obtained at a contraction frequency of 1 Hz. A stimulation frequency of 2 Hz was chosen to examine the effects of increased frequency of contraction as some muscles could not follow 3 Hz stimulation. Regurgitant fraction was significantly reduced by increasing the contraction frequency from 1 Hz to 2 Hz (Table III).

Effects of Changes in Contractility on the Amount of Muscle Shortening Assigned to Regurgitation: The effects of inotropic interventions were examined as mentioned in the method section. An increase in the contraction frequency up to 3 Hz brought about 150% increase in dF/dt and 10% decrease in regurgitant fraction. Administration of *l*-isoproterenol increased dF/dt_{max} up to 290 $kPa \cdot s^{-1}$ and decreased regurgitant fraction from 0.89 to 0.60. The administration of $CaCl_2$ caused results similar to *l*-isoproterenol. The effects of these interventions on regurgitant fraction are summarized in Table III.

DISCUSSION

An examination and reconsideration of the

TABLE III VALUES OF REGURGITANT FRACTION AFTER VARIOUS INTERVENTIONS

Muscle	*Preload (kPa)*	*Total load (kPa)*	*Regurgitant fraction*			
			Control (1 Hz)	*Increased frequency of contraction (2 Hz)*	*l-isoproterenol (4×10^{-9} mol)*	*$CaCl_2$ (2.7×10^{-5} mol)*
1	*4.2*	*11.8*	*0.86*	*0.80*	—	—
2	*5.5*	*10.4*	*0.89*	*0.73*	*0.60*	*0.48*
3	*10.9*	*15.6*	*0.73*	*0.69*	—	—
4	*5.9*	*14.5*	*0.76*	*0.71*	*0.65*	*0.64*
5	*6.6*	*14.9*	*0.76*	—	*0.60*	*0.61*
6	*8.5*	*16.4*	*0.91*	*0.75*	*0.68*	*0.73*
Mean	*6.9*	*13.9*	*0.82*	*0.74**	*0.63***	*0.62***
± SD	*± 2.4*	*± 2.3*	*± 0.08*	*± 0.04*	*± 0.04*	*± 0.10*

** = statistically significant ($p < 0.05$) vs. control;*
*** = statistically significant ($p < 0.01$) vs. control*
The mean regurgitant fraction values at control and at each interventions were compared. t-test for two mean values was employed for statistical analysis.

NII-Electronic Library Service

mechanical basis of the concepts of atrioventricular valvular insufficiency were made on simple and clear assumptions. These hemodynamic situations have in common ventricular ejection in two opposing directions, one against higher pressure through relatively wide aortic or pulmonary valve with a negligible pressure gradient, and the other to lower pressure through relatively narrow regurgitant channel with a significant pressure gradient.

In a normal heart, most of the total amount of pressure rise in the ventricle is attributed to an isometric contraction. The in vitro analogue of the normal ventricle during contraction phase using an isolated cardiac muscle has been realized as the so-called afterloaded contraction. In the present study, a mechanical mitral regurgitation model in which the ventricular pressure rises to exceed the aortic pressure solely as a result of isometroid contraction was adopted. This analogue of the hemodynamic situation is realized using isolated cardiac muscles by substituting a linear muscle force – fiber shortening velocity relation for the ventricular pressure – flow velocity relation in the mitral regurgitation. The quantitative validity of this substitution must be subjected to criticism. However, the similarity of responses of the heart, observed in both clinical and acute experimental mitral insufficiency and in the present study, to the physiological or pharmacological interventions is striking. Recently, Yoran et al.[2] has studied the dynamic aspect of the degree of mitral regurgitation using dogs with partially excised anterior leaflet of the mitral valve. This experimental mitral regurgitation model was expected to simulate more fully the mitral insufficiency in human. However, human mitral insufficiency involves the abnormalities of various parts of valve apparatus including valve leaflets, chordae tendinae and the papillary muscles. These structural deformations cause various hemodynamic types of mitral regurgitation. They stressed the effects of changes in ventricular volume and contractility on the effective regurgitant orifice area, and concluded that changes in regurgitant volume was due to changes in that area.[2] However they did not carry out any direct measurement of the regurgitant orifice area but calculate it from Gorlin's equation.[14] The only theoretical basis of their argument was that the regurgitant volume depended only on the mean systolic pressure gradient between the left ventricle and the left atrium, the mean mitral regurgitant orifice area, and the duration of the regurgitation. However, even though all these factors remain constant, the regurgitant volume can change according to each change in the amount of regurgitation during the isometroid contraction or relaxation phase and the regurgitation during the combined ejection phase as is seen in our results. Yellin and Yoran et al.[15] developed the hypothesis that the mitral regurgitant orifice area decreases with time as ventricular volume decreases during systole. The validity of their hypothesis also depends totally on the validity of the equation which they used for the calculation of mitral regurgitant orifice area (modified Gorlin's equation). The mitral regurgitant flow waveform which they used for their argument is a peaky one. It reaches the summit in very early systole and falls rapidly throughout the remaining part of systole. However, in a previous paper,[16] Yellin and Laniado et al. have shown mitral regurgitant flow waveforms which have virtually a flat plateau (constant flow rate) over most of the regurgitant period. Furthermore, Kalmanson et al.[17] have also shown regurgitant velocity traces which have flat plateaus in patients with mild or moderate regurgitation using directional Doppler ultrasonic catheter tip velocimeter placed at the site of the mitral annulus. If these tracings were used for the calculation of the mitral regurgitant orifice area from the equation of Yellin and Yoran et al.,[15] the conclusion would be that the mitral regurgitant orifice area increases with time during systole. Although the method for creating mitral regurgitation in the experiment of Yellin and Laniado et al.[16] was not the same as that in the experiment of Yellin and Yoran et al.,[15] this conclusion would not be accepted. Therefore, we consider that the use of modified Gorlin's equation for the calculation of mitral regurgitant orifice area may be adequate in some cases of experimental mitral regurgitation, but may be inappropriate in other cases of experimental or clinical mitral regurgitation. Therefore, because of the underlying faults we could not rely on the hypothesis of Yellin and Yoran et al.[15] in the explanation of the responses of the heart with mitral regurgitation to various interventions.

More recently, Greenberg et al.[7] reported the beneficial hemodynamic changes by both intravenous and oral administration of hydralazine without any significant changes in ventricular volume in patients with mitral insufficiency. They ascribed this responses bo hydralazine

NII-Electronic Library Service

mainly to the reduction in aortic impedence. A decrease in ventricular volume might improve the valvular incompetence by improving the coaptation of valve leaflets as is hypothesized by Yoran and Yellin et al[2] and Yellin and Yoran et al[15] However, the beneficial responses of the heart to lowered total load can fully be explained by the assumption that the developed ventricular pressure has a functional relationship to the regurgitant flow velocity.

In our mechanical model, reduced total load under conditions of constant preload (reduced afterload) requires lower regurgitant flow velocity to overcome the aortic pressure. This means that early onset and late cessation of forward ejection of blood are expected. Furthermore, the amount of regurgitation during both the isometroid contraction and relaxation phases is reduced by the reduction in total load under conditions of constant preload, because of the curtailment of both periods. Consequently, the overall regurgitant fraction is expected to fall even though the increase in the duration of the isotonic (ejection) phase has an effect to increase the regurgitation during this phase.

An increase in preload under conditions of constant total load reduces afterload, which has similar effects to a decrease in total load under conditions of constant preload. At the same time, increased preload brings about the rise in the rate of force development, which acts to reduce the duration of the isometroid contraction phase and hence it reduces the regurgitation during that phase even under conditions of constant afterload. Both of them are supposed to act in concert to reduce the regurgitation. On the other hand, Yoran and Yellin et al[2] observed increased regurgitant volume (not regurgitant fraction) when the animals were infused by 200–400 ml of blood or dextran. The regurgitant fraction showed an insignificant increase by 8%. At the same time, the left ventricular systolic pressure rose by 12% during the volume expansion. This means that this maneuver was accompanied by the increment in total load. In the experiment using a whole ventricle, contrary to the experiment using a papillary muscle, changes in single hemodynamic parameter cause multiple hemodynamic changes. On their argument about the effect of the increase in preload on the regurgitation, the influence of the increase in total load accompanying their experimental maneuver was not taken into account. Therefore, their conclusion that the increase in preload causes the greater regurgitant fraction seems to arouse a question, though our experimental study could not exclude the possibility of regurgitant orifice area change due to increased preload.

The hemodynamic changes caused by the volume infusion resemble our experiments in which preload is increased under conditions of constant afterload (Fig. 7) rather than that under conditions of constant total load. Under conditions of constant afterload, increase in preload is accompanied by a concomitant increase in total load which opposes the effect of increase in preload. The regurgitant fraction took a minimum value at a certain preload between 5 kPa and 10 kPa as is shown in Fig. 7.

The effects of enhanced ventricular function curve due to the administration of mephentermine sulfate (0.4 mg/kg) on the hemodynamic parameters of the dog with an external conduit with a valve between the left atrium and the left ventricle were reported by Braunwald et al[1]. This intervention must have caused both the enhancement of myocardial contractility and the elevation of systemic arterial pressure (total load) due to the increased forward cardiac output. According to their data, the overall effects of this pharmacological maneuver on the degree of regurgitation were an increase in total cardiac output, an increase in the regurgitant volume and a decrease in the regurgitant fraction (0.70 to 0.54 according to our calculation) despite the fixed regurgitant orifice area. On the other hand, Yoran et al[2] reported the effect of 4–10 μg/min infusion of norepinephrine on the regurgitation. They observed an increase in stroke volume, decrease in regurgitant volume and a decrease in regurgitant fraction. Additionally, the clinical study by Jose et al[3] showed that the positive inotropism of norepinephrine canceled the predicted increase in the regurgitant fraction caused by the rise in arterial pressure. In our mechanical model of mitral regurgitation, the left ventricular systolic pressure is maintained by the pressure difference between the left ventricle and the left atrium which increases with the regurgitating blood flow velocity. Under such circumstances, it is obvious that a higher rate of ventricular contraction curtails the duration of the isometroid contraction phase and consequently the regurgitation during that phase. In our experiments, the decrease in regurgitant fraction was achieved when myocardial contractility was enhanced either by the administration

NII-Electronic Library Service

of positive inotropic agents or by the increase in the frequency of contraction under conditions of constant loads.

One may expect that no substantial regurgitation takes place during the isometroid contraction phase that lasts 40 ms (from our data in this study) to 100 ms (by Yoran et al.[2]) since the prolonged forward flow through the mitral valve may continue for several tens of miliseconds after the reversal of atrio-ventricular pressure gradient due to fluid inertia. However, the timing and mechanism of the final closure of the mitral valve is still controversial.[18] By Noran et al.[19] the mean deceleration of the mitral valve flow after the reversal of the pressure gradient is estimated to be of the order of 40 $m \cdot s^{-2}$ at most. The deceleration estimated from the data of Laniado et al.[20] and Kalmanson et al.[21] is much smaller than this. This value of deceleration gives an adverse gradient of the order of 40 $kPa \cdot m^{-1}$, which is consistent with the measured value (1.7 kPa ≒ 12.8 mmHg) of the ventricular pressure at mitral closure published by Tsakiris et al.[22] assuming that the distance between the points where the pressures were measured is about 4 cm. Thus the true mitral closure is estimated to take place at the time that the left ventricular pressure exceeds the left atrial pressure by 1.7 kPa, that is 6 ms after the reversal of atrio-ventricular pressure gradient when the mean rate of left ventricular pressure rise is assumed to be about 270 $kPa \cdot s^{-1}$. Therefore the effect of fluid inertia on the amount of regurgitation during the isometroid contraction phase was disregarded in the present study.

The present study also suggests a possible explanation for relative insensitivity of total cardiac output including regurgitation to changes in arterial pressure. As total load was increased about two-fold, only a 10% reduction in the total amount of muscle shortening occurred under conditions of constant preload in the present experiment (Fig. 5). This observation is consonant with that obtained by Braunwald et al.[1] using a whole ventricle and an external conduit with a valve between the left atrium and the left ventricle. They saw only a slight change in total cardiac output including regurgitation when aortic pressure was changed widely from 110 mmHg (14.6 kPa) to 230 mmHg (30.6 kPa). On the contrary, the total amount of ventricular volume change (stroke volume) is inversely related to the level of total load in a normal ventricle. In the present mechanical model of mitral insufficiency as well as in a mitral insufficiency in the whole ventricle, the end-systolic fiber force (force imposed on the muscle at the shortest muscle length) corresponds to preload. According to the unique end-systolic force length relationship of the cardiac muscle reported by Suga et al.[2,3] the end-systolic fiber length, therefore, the total amount of muscle shortening under conditions of constant preload i.e., initial fiber length (end-systolic force in this case) is expected to be constant (Fig. 5). In other words, total stroke volume including regurgitation is constant under conditions of constant preload and contractility. Thus afterload determines regurgitant fraction in a mitral regurgitation.

Thus, a mechanism for the improvement of cardiac function under reduced arterial pressure in mitral regurgitation has been suggested by the present study, however, papillary muscle experiments do not simulate true mechanical conditions of the ventricle in situ, and the present model does not imitate the full range of factors that have been identified in human mitral insufficiency. Therefore, the application of the data obtained in the papillary muscle experiment is necessarily limited. The extension of the basic ideas in the present study to the intact heart should enhance our present understanding of mechanics of the mitral insufficiency.

Acknowledgments

The authors are grateful to Professor Haruka Okino at the Department of Physiology, Tokai University School of Medicine for his advice and encouragement in the completion of this work. This study was supported in part by reserch grants-in-aid, Project 5757031, 1982, from the Ministry of Education of Japan.

REFERENCES

1. BRAUNWALD E, WELCH GH, SARNOFF SJ: Hemodynamic effects of quantitatively varied experimental mitral regurgitation. *Circ Res* **5:** 539, 1957
2. YORAN C, YELLIN EL, BECKER RM, GABBAY S, FRATER RWM, SONNENBLICK EH: Dynamic aspects of acute mitral regurgitation: Effects of ventricular volume, pressure and contractility on the effective regurgitant orifice area. *Circulation* **60:** 170, 1979
3. JOSE AD, TAYLOR RR, BERNSTEIN L: The influence of arterial pressure on mitral incompetence in man. *J Clin Invest* **43:** 2094, 1964
4. CHATTERJEE K, PARMLEY WW, SWAN HJC, BERMAN G, FORRESTER J, MARCUS HS: Beneficial effects of vasodilator agents in severe mitral regurgitation due to dysfunction of

NII-Electronic Library Service

subvalver apparatus. *Circulation* **48:** 684, 1973
5. GOODMAN DJ, ROSSEN RM, HOLLOWAY EL, ALDERMAN EL, HARRISON DC: Effect of nitroprusside on left ventricular dynamics in mitral regurgitation. *Circulation* **50:** 1025, 1974
6. HARSHAW CW, GROSSMAN W, MUNRO AB, MCLAURIN LP: Reduced systemic vascular resistence as therapy for severe mitral regurgitation of valvular origin. *Ann Int Med* **83:** 312, 1975
7. GREENBERG BH, MASSIE BM, BRUNDAGE BH, BOTVINICK EH, PARMLEY WW, CHATTERJEE K: Beneficial effects of hydralazine in severe mitral regurgitation. *Circulation* **58:** 273, 1978
8. RODBARD S, WILLIAMS F: The dynamics of mitral insufficiency. *Am Heart J* **48:** 521, 1954
9. WIGGERS CJ, FEIL H: The cardio-dynamics of mitral insufficiency. *Heart* **9:** 149, 1921
10. SONNENBLICK EH, PARMLEY WW: Mechanical effects of increased series elasticity: An in vitro model of mitral regurgitation and ventricular aneurysm. *Am J Cardiol* **27:** 376, 1971
11. ENDOH M, HASHIMOTO K: Pharmacological evidence of autonomic nerve activities in canine papillary muscle. *Am J Physiol* **218:** 1459, 1970
12. TAMIYA K, SUGAWARA M, SAKURAI Y: Maximum lengthening velocity during isotonic relaxation at preload in canine papillary muscle. *Am J Physiol* **237:** H83, 1979
13. TAMIYA K, KIKKAWA S, GUNJI A, HORI M, SAKURAI Y: Maximum rate of tension fall during isometric relaxation at end-systolic fiber length in canine papillary muscle. *Circ Res* **40:** 584, 1977
14. GORLIN R, DEXTER L: Hydrauric formula for the calculation of cross-sectional area of the mitral valve during regurgitation. *Am Heart J* **43:** 188, 1952
15. YELLIN EL, YORAN C, SONNENBLICK EH, GABBAY S, FRATER RWM: Dynamic changes in the canine mitral regurgitant orifice area during ventricular ejection. *Circ Res* **45:** 677, 1979
16. YELLIN EL, LANIADO S, PESKIN CS, FRATER RWM: Flow studies of experimental mitral stenosis and regurgitation. *In* The Mitral Valve, ed by KALMANSON D, Edward Arnold, London, p 195
17. KALMANSON D, VEYRAT C, BERNIER A, SAVIER CH, CHICHE P, WITCHTZ S: Diagnosis and evaluation of mitral valve disease using transseptal Doppler ultrasound catheterization. *Br Heart J* **37:** 257, 1975
18. LITTLE RC: The mechanisms of closure of the mitral valve: A continuing controversy. *Circulation* **59:** 615, 1979
19. NORAN SP, DIXON SHJr, FISHER RD, MORROW AG: The influence of arterial contraction and mitral valve mechanics on ventricular filling. *Am Heart J* **77:** 784, 1969
20. LANIADO S, YELLIN EL, MILLER H, FRATER RWM: Temporal relation of the first heart sound to closure of the mitral valve. *Circulation* **47:** 1006, 1973
21. KALMANSON D, BERNIER A, VEYRAT C, WITCHITZ S, SAVIER CH, CHICHE P: Normal pattern and physiological significance of mitral valve flow velocity recorded using transseptal directional Doppler ultrasound catheterization. *Br Heart J* **37:** 257, 1975
22. TSAKIRIS AG, GORDON DA, PADYAR R, FRÉCHETTE D: Relation of mitral valve opening and closure to left atrial and ventricular pressures in the intact dog. *Am J Physiol* **234:** H146, 1978
23. SUGA H, SAEKI Y, SAGAWA K: End-systolic force-length relationship of nonexcised canine papillary muscle. *Am J Physiol* **233:** H711, 1977

NII-Electronic Library Service