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Research Article

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The Mechanics of Esophageal Muscle Contraction

EVIDENCE OF AN INOTROPIC EFFECT OF GASTRIN

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ABSTRACT To compare the mechanical properties of lower esophageal sphincter (LES) and esophageal circular smooth muscle, force-velocity determinations were made under various physiological conditions. Isotonic and isometric recordings of opossum circular muscle were used to obtain the velocity of shortening and force, respectively, during alterations in: (*a*) initial muscle length (preload), (*b*) afterload, (*c*) calcium concentration, and (*d*) gastrin I. Muscle contraction was elicited to the neurogenic response at the termination of electrical stimulation. A change in preload (muscle length) altered the peak force (Po) developed during an afterloaded contraction, but had only a minor effect on the maximum velocity of shortening (V max). At the length of optimal tension development, Lo, (preload, 1.5 g), the LES muscle had a V max of 6.1±0.2 mm/s and a Po of 17.7±0.7 g. The esophageal muscle at its Lo (preload, 2.0 g) had a V max of 6.3±0.5 mm/s and a Po of 18.1±1.2 g. A decrease in calcium from 2.5 mM to 1.0 mM significantly reduced the V max and Po of all muscle, but an increase in calcium to 5.0 mM increased these parameters only minimally. At a calcium of 1.0 mM, gastrin I increased both V max and Po of all muscle. This inotropic effect of gastrin I occurred at lower concentrations in LES muscle than in muscle from the upper esophagus. The power (force × velocity) and work (force × muscle shortening) of esophageal and LES muscle were calculated from these data. Both the work and power generated during esophageal and LES muscle contraction were determined by: (*a*) the initial muscle length as produced by the preload, (*b*) the afterload against which the muscle was contracting, and (*c*) the contractility of inotropism of the muscle, that is, the force-velocity curve on which the muscle was operating.

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INTRODUCTION

In recent years, there has been considerable investigation into the function of the esophagus and the lower esophageal sphincter (LES) [1] in man and in animal models (1–11). These studies have described multiple mechanisms by which the esophagus and the LES responded to the various demands placed upon it. The esophagus may have the capacity to change its propulsive force in response to bolus size (5) and neurohumoral agents (3, 4). The LES can change its strength in response to humoral stimuli (1, 11) and alterations in intraabdominal pressure (2, 8, 9). Despite the description of these mechanisms that alter esophageal and LES performance, there has been no investigation of the mechanics of muscle function to characterize these adaptive responses. The purpose of this study is fourfold. First, the mechanics of isolated esophageal and LES muscle are described by the general schema developed by Hill for skeletal muscle (12–15) and subsequently used for cardiac muscle (16–20) and smooth muscle (21–26). Second, alterations in the mechanics of muscle function in terms of the force-velocity relationship are evaluated under varying conditions of both preload and afterload. Third, an attempt to characterize the contractility or inotropism of the muscle in terms of muscle mechanics is made. Fourth, the extent to which these studies on the mechanics of isolated muscle, contracting under afterloaded conditions, can be applied to the responses of the intact esophagus and LES is discussed.

[1] *Abbreviations used in this paper:* ΔL, distance the load was moved; dl, change in length; dt, change in time; LES, lower esophageal sphincter; Lo, length of optimal tension development; P, isometric force; Po, peak force; V max, maximum velocity of shortening.

METHODS

Studies were performed on 30 adult opossums of both sexes, weighing 2.5–5.0 kg. The method of obtaining muscle, outlined below, had previously been described in detail (10). The animals were anesthetized with 40 mg/kg of intraperitoneal pentobarbital and strapped supine to an animal board for studies in vivo. Esophageal manometric studies utilizing an infused, open-tipped catheter system were performed in all animals. A triple-lumen polyvinyl tube (1.4 mm ID with three side orifices 1.2 mm in diam) was passed through the mouth into the stomach. Each catheter was continuously infused with distilled water by an infusion pump at the rate of 1.2 ml/min. Intraluminal pressure was transmitted to external transducers (Statham P23BB, Statham Instruments, Inc., Oxnard, Calif.), whose output was graphed on a Beckman curvilinear, ink-writing recorder (Beckman Instruments, Fullerton, Calif.). After all orifices were in the stomach, the tube assembly was withdrawn at 0.5-cm intervals, and pressures were recorded at each level for a 1-min period. After completion of the manometric study, the recording tube was anchored at the lower jaw and positioned with the middle recording orifice in the LES. All animals were then killed by intravenous pentobarbital. The LES, as determined by manometry, was identified and marked. The LES was within 2 cm of the anatomic gastroesophageal junction and below the diaphragm.

The esophagus, 8 cm proximal to the anatomic gastroesophageal junction, the entire stomach, and the duodenum were mobilized and freed from the surrounding tissues. The midesophagus and distal duodenum were ligated, and the upper gastrointestinal tract from midesophagus to distal duodenum was excised, washed in Krebs-Ringer solution (composition in millimoles per liter: Na^+, 138.6; K^+, 4.6; Ca^{++}, 2.5; Mg^{++}, 1.2; Cl^-, 126.2; HCO_3^-, 21.9; PO_4^-, 1.2; glucose, 49.6, at 37°–38°C, transferred to an organ bath of Krebs-Ringer solution bubbled with 95% O_2 and 5% CO_2, and maintained at 37°–38°C. The esophagus was separated from the stomach at the anatomic gastroesophageal junction where the narrow esophagus flares into the stomach. The mucosa from each region was removed to the level of the submucosa. Circular smooth muscle strips, 0.5 cm wide and 1.0 cm long, were cut from each anatomic region. All muscles were lightly blotted and weighed at the termination of the experiment.

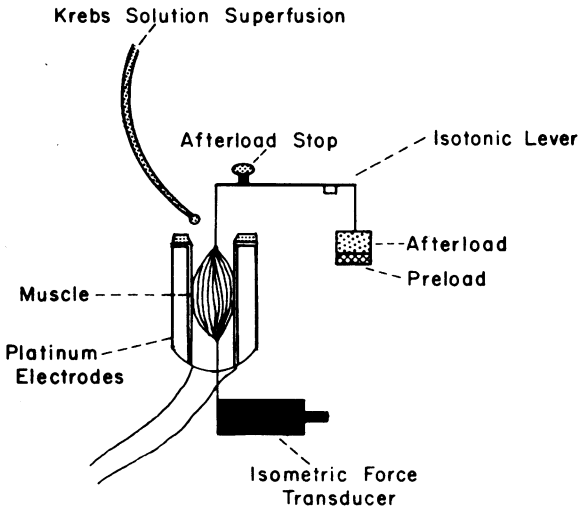

FIGURE 1 Schematic diagram of apparatus used to measure afterloaded muscle shortening. The muscle was mounted between an isometric transducer and an isotonic transducer. A stop allowed the muscle to receive both a preload and an afterload. The muscle was superfused with heated and oxygenated Krebs solution to which agents were added. The muscle was electrically stimulated across two platinum wires.

Each muscle was studied with the apparatus diagrammatically illustrated in Fig. 1. One end of the muscle was connected to an inflexible wire attached to an isometric force transducer (Grass model FT-03C, Grass Instrument Co., Quincy, Mass.). The other end was attached to the lever of an isotonic transducer (Harvard model 387, Harvard Apparatus Co., Inc., Millis, Mass.). Muscle length was adjusted with a micrometer. Muscle loading was accomplished through calibrated balance weights placed on the isotonic lever. A stop allowed the muscle to receive both a preload and an afterload. Each muscle was stimulated electrically across two platinum wires adjusted to juxtapose the lateral surfaces of the muscle. Stimuli were delivered by a stimulator (Grass model S44 with stimulus isolation unit SIU I). The muscles were superfused by a heated and oxygenated Krebs solution with the composition noted above. A thermistor was used to monitor temperature at the muscle. The superfusate was maintained at 36°–38°C. All recordings were graphed on a rectilinear, ink-writing polygraph (Beckman model R411).

After a 30-min equilibration period, the muscle length was reduced until the passive tension had first reached zero. This length was termed the initial length. From this initial length, graded preloads in 250-mg increments were placed on the muscle. The change in muscle length caused by each preload was measured directly by the excursion of the isotonic lever as seen on the recorder. At each preload, a force-velocity curve was constructed utilizing afterloads in either 0.5-g or 1.0-g increments.

To obtain the force-velocity relationship of the esophageal and LES muscle, the neurogenic response elicited at the termination of electrical stimulation was utilized. This muscle, as previously demonstrated, did not develop active tension during stimulation, but at the end of stimulation a prominent neurogenic contraction ("off" response) occurred after a brief latent period (6). Stimulus parameters were initially selected to elicit the maximal amplitude of this contraction under pure isometric recording conditions. Subsequently, muscles were stimulated by a 30-s train of square wave pulses of 1.0 ms duration, 10 Hz at 50–60 V.

Force-velocity curves were constructed for each muscle under various physiological conditions using the off response as a basis of nontetanic muscle contraction. The isometric force generated before shortening was measured directly from its respective tracing. The maximal velocity of shortening was determined from the initial phase of the isotonic tracing. The maximal phase of shortening was expressed as the change in length (dl) per unit change in time (dt). The degree of shortening of the muscle during an afterloaded contraction was obtained directly from the isotonic recording. The load at which shortening first became zero was denoted by Po. The muscle length at which the maximal Po was achieved was denoted by Lo.

Force-velocity curves were also constructed during the superfusion of Krebs solution at different calcium concen-

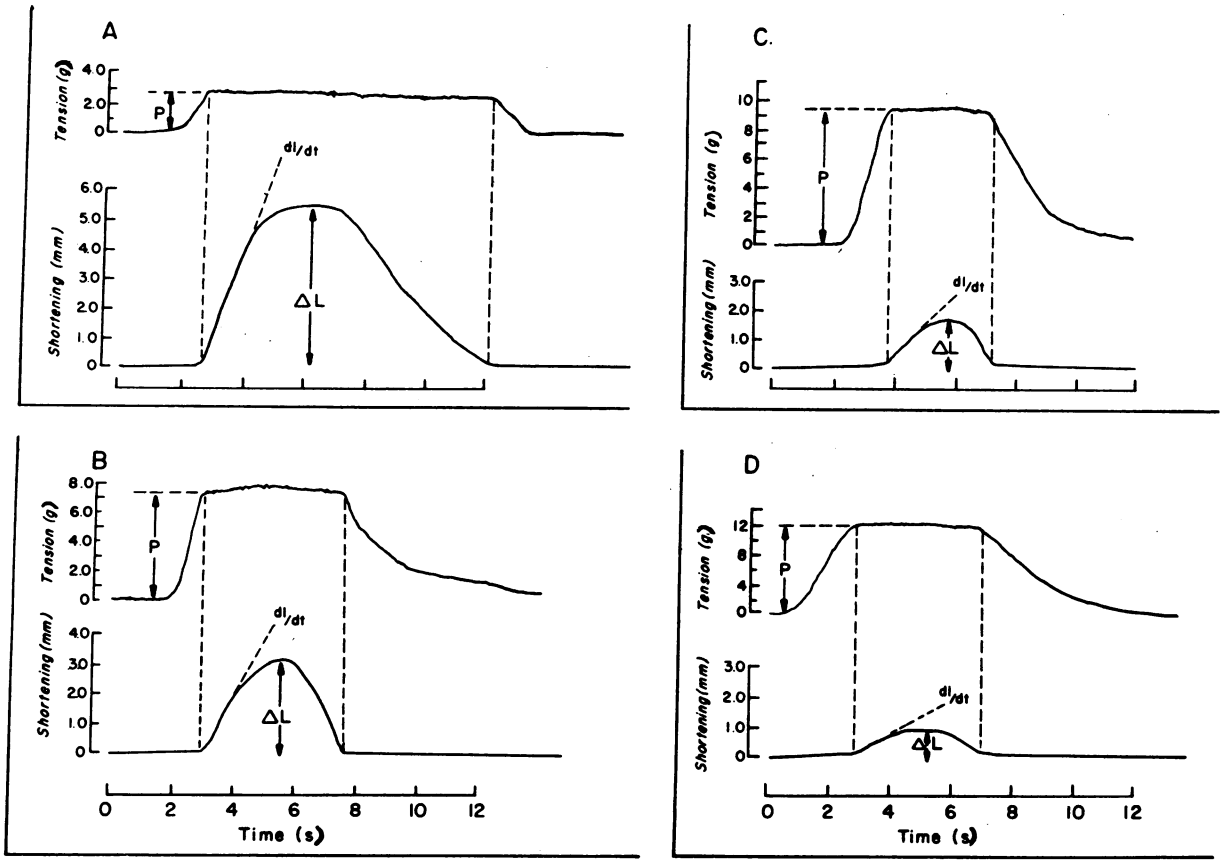

FIGURE 2 Simultaneous isometric and isotonic recordings obtained for a single strip of LES circular muscle at 1.5 g preload (not shown on force tracing) and afterloads of 1.5, 6, 8, and 11 g in panels A–D, respectively. When isometric force equaled the total load on the muscle (first vertical line), isotonic shortening began. The velocity of shortening was calculated by measuring the change in length (dl) per unit change in time (dt). Total muscle shortening was measured directly, ΔL. As load increased, velocity of shortening decreased.

trations. In addition to the usual 2.5 mM Ca^{++} Krebs solution, studies were done at 1.0 mM Ca^{++}, 1.5 mM Ca^{++}, and 5.0 mM Ca^{++}. After the determination of the force-velocity relationship at different calcium concentrations, gastrin I (residue 2-17, Imperial Chemical Corp., Alderly Park, Cheshire, England) was added to the superfusate at concentrations of 0.1 ng–10.0 ng/ml. Gastrin I was evaluated in Krebs solution containing either 2.5 or 1.0 mM Ca^{++}. Force-velocity measurements were made after a minimum of 10 min of superfusion with the Krebs solutions containing different calcium concentrations or gastrin I.

The effect of muscle depolarization by superfusion with Krebs solution containing 143.6 mM KCl on the isometric force was also evaluated. KCl depolarization was compared in 2.5 and 1.0 mM calcium solutions.

Measurements of velocity of shortening in millimeters per second, shortening in millimeters, load in grams, and Po were placed into a computer program. The constants of the Hill equation as determined from a linearized plot of the data were used to determine the extrapolated V max. The best fit of the line for the displaced rectangular hyperbola of the force-velocity relationship and the standard deviation of that line were directly plotted by the computer using the least square regression analysis. Calculations of work and power were also made at each load. Statistical analysis was made for paired comparisons using the Student *t* test.

RESULTS

In Fig. 2 are shown the simultaneous isotonic and isometric recordings of LES circular muscle at 1.5-g preload and increasing afterload. After a brief latent period at the termination of the electrical stimulus, force development began. When isometric force (P) equaled pre- and after-load at the first vertical line, isotonic shortening began at a maximal rate (dl/dt or V). The net shortening of the muscle (ΔL) equaled the distance the load was moved. As the afterload was increased, the initial velocity of shortening (dl/dt) diminished. ΔL was also diminished as the afterload was increased.

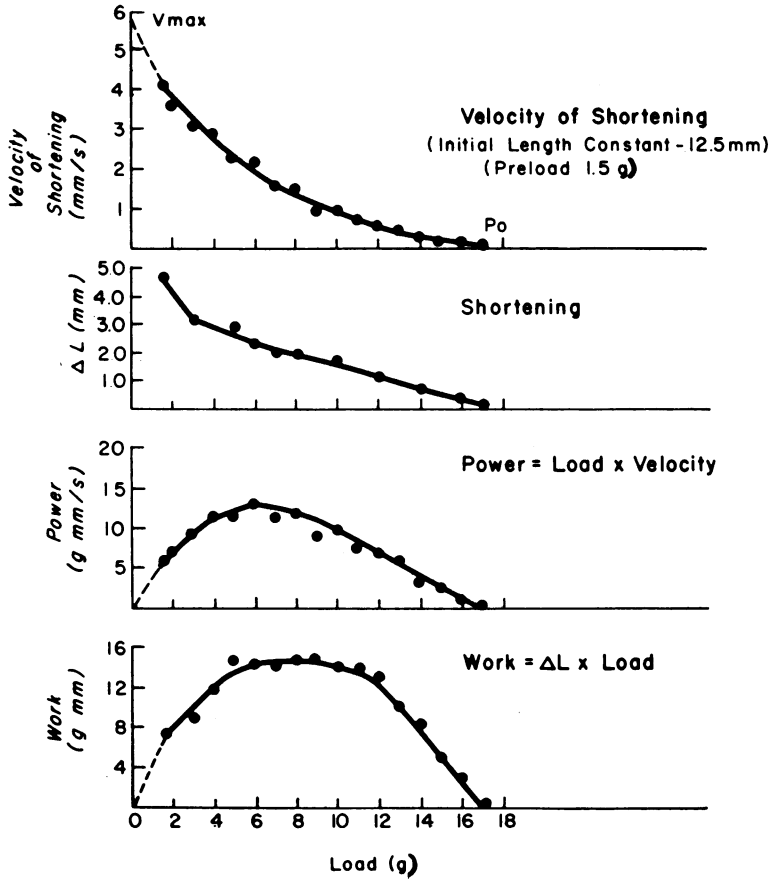

FIGURE 3 Force-velocity relation of opossum LES circular muscle. Muscular length was constant, 12.5 mm, at 1.5 g preload. Velocity of shortening in millimeters per second, shortening in millimeters, power (load × velocity), and work (load × ΔL) were plotted as functions of increasing total load (preload plus afterload). Velocity of shortening and shortening decreased with increasing load. When velocity was extrapolated to zero load, the theoretical maximum velocity of shortening (V max) was obtained. When load was increased until no shortening occurred, peak isometric force (Po) was recorded. Work and power were functions of the above relation with maximum values obtained at about 40–50% of Po.

In Fig. 3 are shown the force-velocity, shortening, power, and work relations of LES circular muscle. The initial muscle length was constant (12.5 mm) at a preload of 1.5 g. The velocity of shortening plotted as a function of load decreased with increasing load. Theoretically, at zero load on the muscle the velocity of shortening was maximal (V max). However, under experimental conditions a preload was necessary to establish the initial length of the muscle. Therefore, V max was approximated by extrapolation of the force-velocity curve to zero load. As the load was increased until no muscle shortening occurred, the velocity of shortening was zero and isometric tension (Po) was recorded. Po, as used here, indicated the maximum tension achieved during a contraction elicited at the termination of electrical stimulation. In skeletal muscle, Po refers to maximum isometric force during tetanus (14, 15). Inherent in the force-velocity relation of muscle was the statement of power (load × velocity) and work (load × ΔL) shown at the bottom of Fig. 3. Work and power formed parallel relations. Both were zero at either end of the force-velocity curve and approximated their maximum at about 40–50% of Po.

Since the force-velocity curve was a rectangular hyperbola similar to that described for skeletal muscle, Hill's equation was used to describe it: $(P + a)(V + b) = (Po + a)b$, where P represents load, V the velocity of shortening, Po the maximum load which the muscle is first unable to move, a is a constant with units of force, and b is a constant with units of velocity (12–14). To prove that the relationship was hyperbolic, a linearized transformation of the Hill

equation was applied as shown in Fig. 4 (20, 21, 24). The points fell along a straight line with an excellent correlation coefficient ($r = 0.97$). From this linearized plot of the force-velocity curve, it was deduced that the relationship for this circular muscle was hyperbolic and the Hill equation can be applied to it. From the linearized plot, the slope ($1/b$) and the y intercept (a/b) provided the constants for the equation. The theoretical V max was calculated from the equation: V max = Pob/a.

After demonstrating that esophageal circular muscle was characterized by a force-velocity relationship similar to other muscles, we evaluated this function under various physiologic conditions. In Fig. 5*a* the force-velocity curves of LES circular muscle obtained at different initial lengths are shown. The initial muscle length was altered by increasing the preload. The Po was increased with an increase in initial length or preload up to a preload of 1.5 g. The V max (as determined from the Hill equation) differed only slightly at each preload. At a preload of 2.0 g (force-velocity curve not shown), the Po was less than that achieved at 1.5 g. The length-tension relationship is shown in the insert. The length of optimal tension development at 1.5-g preload was termed Lo.

In Fig. 5*b*, the force-velocity curves for esophageal muscle, 4.0 cm above the LES, are shown. The V max was approximately the same at each preload but Po differed. The length-tension relationship illustrated that esophageal muscle developed its greatest Po at 2.0 g preload and thus differed from the LES muscle. Otherwise, the force-velocity parameters, V max and Po, were similar.

All force-velocity data during change in initial muscle length (preload) are given in Table I. For all muscle, increase in preload or muscle length gave slight changes in V max, but marked changes in Po. At Lo (1.5 g LES, 2.0 g esophagus), the V max and Po values were similar ($P > 0.05$). Only the V max values at 0.3 g for LES muscle and 0.5 g for esophageal muscle differed significantly from the V max at the respective Lo ($P < 0.01$). Both the V max and Po of the esophageal and LES muscle were expressed further in terms of millimeters per second per centimeter tissue length (lengths/second), and kilograms per square centimeter, respectively. These measures allowed comparison with other muscles where force-velocity curves were made (21, 26). The cross-sectional area was calculated from the mass (grams) divided by the specific gravity (1.056) times the length (centimeters) (21). The V max and Po of LES muscle studied at Lo and 2.5 mm calcium was 0.41 lengths/s and 0.64 kg/cm^2, respectively. The V max and Po of esophageal muscle studied at Lo and 2.5 mm calcium was 0.37 lengths/s and 0.68 kg/cm^2, respectively.

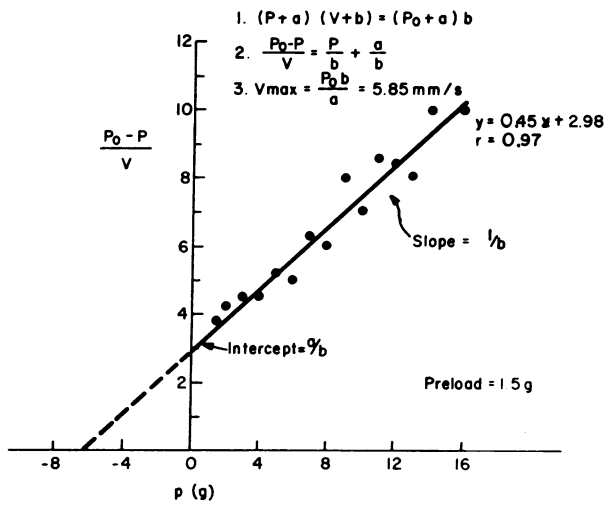

FIGURE 4 Constants of Hill's equation obtained by rearrangement to obtain a linear plot. Points were obtained from the force-velocity curve shown in Fig. 3. When (Po − P/V) was plotted as a function of P, the slope of the line was $1/b$, the intercept on the ordinate was a/b and the intercept on the abscissa was minus a. V max was calculated as $(Po/a)b$.

To further evaluate the characteristics of LES and esophageal muscle function, studies were done to determine the contractility of the muscle as defined by the force-velocity curve. In Fig. 6 the force-velocity curves of LES circular muscle are shown as obtained in Krebs' solution of 2.5 mM and 1.0 mM calcium. The muscle was studied at Lo using a preload of 1.5 g. Under the experimental condition of decreased calcium (1.0 mM), the force-velocity curve was altered. Both the V max and Po were diminished. Additional studies done at 5.0 mM calcium indicated that the circular muscle was operating at near-maximal contractility at 2.5 mM calcium. All data for alterations in calcium concentration are given in Table II. Reductions in both V max and Po from the control values at 2.5 mM calcium were achieved at calcium concentrations of 1.0 mM and 1.5 mM ($P < 0.001$). V max determinations at 5.0 mM calcium were slightly greater than at 2.5 mM ($p < 0.05$).

To determine whether the effect of calcium was primarily on neural or contractile elements, KCl muscle depolarization was performed at 1.0 mM and 2.5 mM calcium concentration. In 10 studies on both esophageal and LES muscle, KCl depolarization gave an active tension of 18.6±2.1 g at 2.5 mM calcium, and 8.1±1.6 g at 1.0 mM calcium ($P < 0.001$). Thus, at Lo, the maximal isometric force of the muscle to direct stimulation

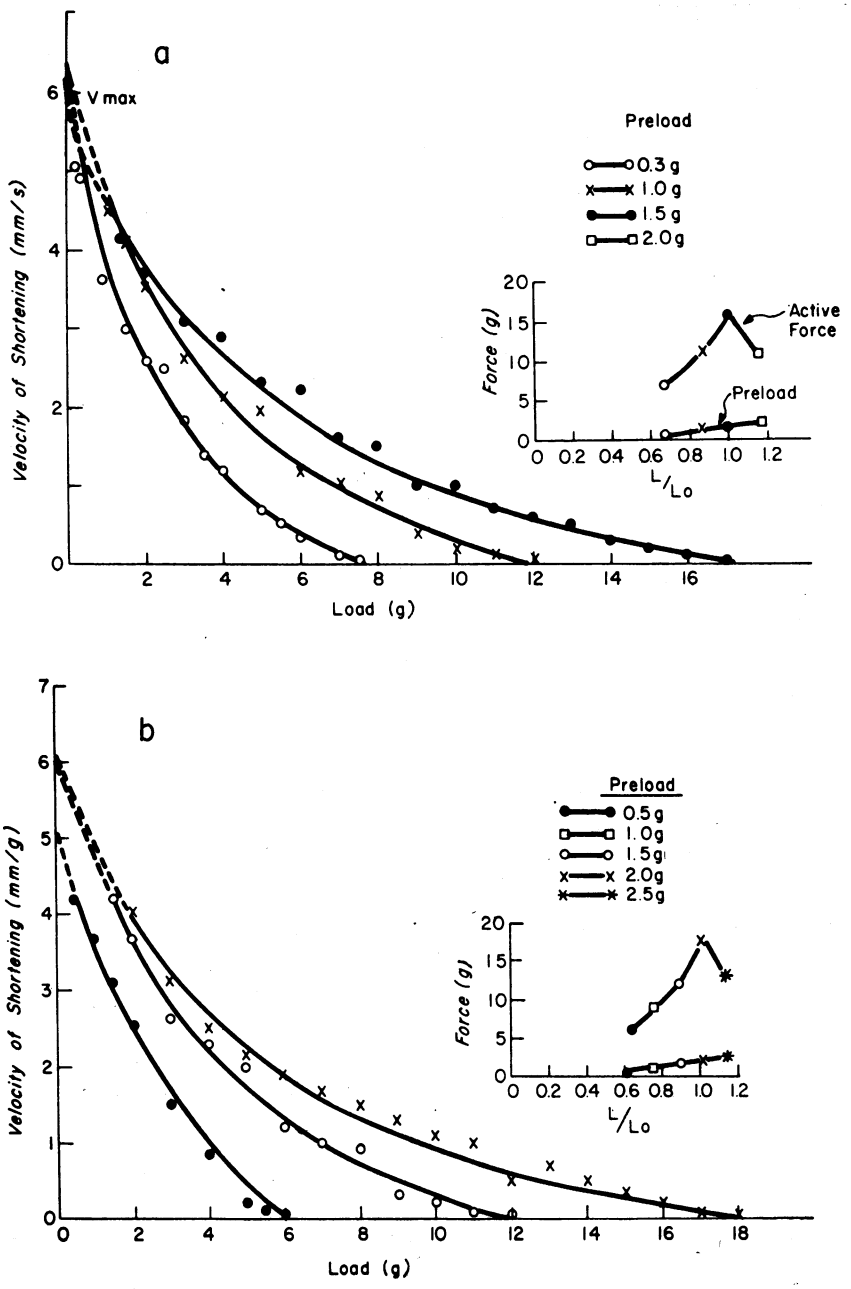

FIGURE 5 The effect of increasing initial muscle length (preload) on the force-velocity curves of LES (above) and esophageal (below) circular muscle. As preload was increased, Po also increased, but V max was only minimally altered. As shown in the inserts, the Lo or length of optimal tension development for LES muscle occurred at a lower preload than the esophageal muscle. The Po and V max at Lo were similar for both muscles.

by KCl depolarization was similar to the Po value (Table I) achieved during the off response at the termination of electrical stimulation. This direct muscle response to KCl depolarization was reduced during a low calcium superfusion.

The demonstration that force-velocity curves of the esophageal and LES smooth muscle could be altered by changing the calcium concentration suggested that this muscle may also alter its contractility in response to physiologic agents known to act upon the esophagus.

TABLE I

*Effects of Preload on Force Velocity Determinations**

Preload	LES		Esophagus (1–2 cm above LES)		Esophagus (4–5 cm above LES)	
	V max	Po	V max	Po	V max	Po
g	*mm/s*	*g*	*mm/s*	*g*	*mm/s*	*g*
0.3	5.0±0.1	9.5±1.3	—	—	—	—
0.5	—	—	5.1±0.2	7.2±0.7	5.0±0.3	7.7±0.8
1.0	5.7±0.3	12.1±0.9	5.2±0.4	10.7±0.4	5.1±0.4	11.2±0.6
1.5	6.1±0.2	17.7±0.7	5.5±0.3	13.8±0.8	5.9±0.5	14.1±0.9
2.0	3.9±1.0	10.3±1.6	5.9±0.3	17.9±0.9	6.3±0.5	18.1±1.2
2.5	—	—	5.7±0.2	16.1±0.8	5.6±0.4	15.1±0.8

Each number represents observations made on a minimum of eight muscle strips.
* All studies were performed at 2.5 mM Ca^{++}.

Since contractility as defined by the force-velocity relation was near-maximal at the usual Krebs solution (2.5 mM calcium), further studies were carried out at 1.0 mM calcium. In the top half of Fig. 7 are shown the force-velocity curves of LES circular muscle at Lo (1.5 g preload) superfused with Krebs solution containing 1.0 mM calcium. The gastrointestinal hormone, gastrin I, (amino acid residue 2–17) was added to the superfusate at a concentration of 0.1 ng/ml. The Krebs solution containing gastrin I gave a brief contraction of the muscle at the onset of superfusion. No prolonged change in muscle tension was recorded. After 5 min of the gastrin I superfusion, the force-velocity characteristics were determined. The curve was shifted toward that obtained with the 2.5 mM calcium (Fig. 6). Both the V max and Po were significantly increased by gastrin I. This change in the force-velocity curve suggested that gastrin I may have an inotropic effect on the circular smooth muscle of the distal esophagus.

To evaluate the specificity of this observation, muscle from the body of the esophagus was evaluated. In the lower half of Fig. 7 are shown the responses of the esophageal circular muscle (4 cm above the LES) at Lo (2.0 g preload) superfused by Krebs solution containing 1.0 mM calcium. The esophageal muscle force-velocity curve was shifted in the presence of gastrin I (10.0 ng/ml). In Table III, all data utilizing gastrin I are given. Studies were carried out at 1.0 mM calcium. Significant increase in V max and Po were first achieved on LES muscle at 0.1 ng/ml gastrin I. Higher gastrin concentrations were required for esophagus 2 cm above the LES and 4.0 cm above the LES. The higher esophageal strips were least sensitive to the effect of gastrin I on contractility.

At 2.5 mM calcium, gastrin I had only a slight effect on V max and Po. Gastrin I (1.0 ng/ml) increased the V max and Po of LES muscle from 6.1±0.2 mm/s and 17.7±0.7 g to 7.4±1.0 mm/s and 18.1±1.2 g, respectively. Only the change in V max was statistically significant ($P < 0.05$).

The parameters of power and work, noted earlier (Fig. 3), were evaluated further during changes in the force-velocity relationship brought about by alterations in muscle length and muscle contractility. In Fig. 8 are shown the work and power determinations for LES circular muscle at different muscle lengths (preloads 0.3 g and 1.5 g) and different states of contractility (1.0 mM calcium alone, and with gastrin I). At each muscle length, both power and work were the functions of load. With an increase in initial muscle length, power and work were increased. The maximum power and work at each muscle length were achieved at approximately 40–50% of Po. The power and work of the muscle were also increased by changing the

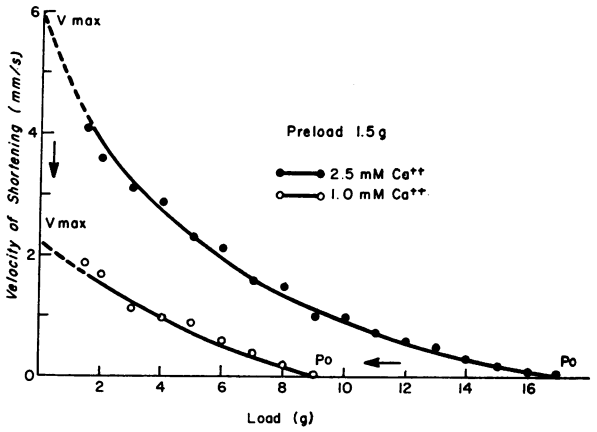

FIGURE 6 Alteration in force-velocity relation of LES circular muscle by a decrease in calcium concentration. The muscle studied at Lo (1.5 g preload) showed a decrease in both V max and Po when the calcium concentration of the superfusate was reduced from 2.5 mM to 1.0 mM.

TABLE II

*Effect of Calcium on Force Velocity Determinations**

Calcium	LES		Esophagus (1–2 cm above LES)		Esophagus (4–5 cm above LES)	
	V max	Po	V max	Po	V max	Po
mM	*mm/s*	*g*	*mm/s*	*g*	*mm/s*	*g*
1.0	2.4±0.2	8.0±0.6	2.8±0.3	8.5±0.4	2.3±0.3	9.1±0.5
1.5	3.5±0.4	11.3±0.8	3.8±0.3	10.2±0.6	3.5±0.5	10.8±0.8
2.5	6.1±0.2	17.7±0.7	5.9±0.3	17.9±0.9	6.3±0.3	16.8±1.1
5.0	7.0±0.4	18.1±1.1	6.7±0.4	17.7±1.1	7.1±0.3	17.9±1.3

Each number represents observations made on a minimum of eight muscle strips.
* All studies were performed at Lo.

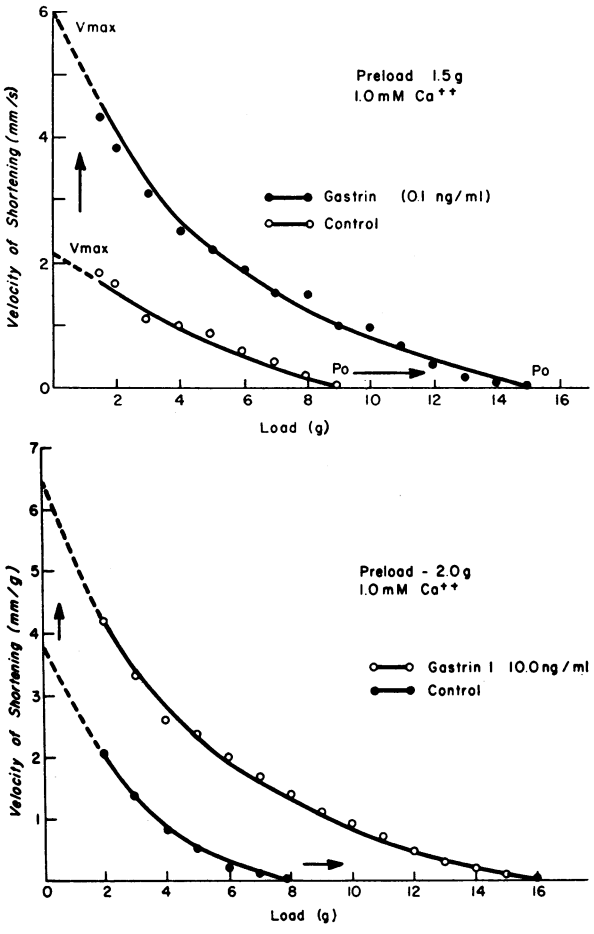

FIGURE 7 Effect of gastrin I on LES (above) and esophageal circular muscle (below) force-velocity relation. Each muscle was studied at its Lo (1.5 g preload for LES, 2.0 g preload for esophagus) at a calcium concentration of 1.0 mM. Gastrin I shifted the entire force-velocity curve of both muscles. The V max and Po of each were markedly increased. Note that LES muscle was studied at 0.1 ng/ml of gastrin I whereas esophageal muscle required 10.0 ng/ml of gastrin I to change the force-velocity curve to a similar degree.

contractility of the muscle with gastrin I. Gastrin I and initial muscle length increased both power and work at any one load and also increased the maximum power and work of the muscle.

DISCUSSION

The purpose of this study was to determine the force velocity characteristics of the circular smooth muscle of the esophagus and LES of the opossum. These studies indicated that the work and power of this muscle, as determined from the force-velocity measurements, were dependent upon at least three factors: first, the initial muscle length as determined by the preload, second, the afterload at which these parameters were measured, and third, the inotropic state or contractility of the muscle, that is, the force-velocity curve on which the muscle was operating.

Hill initially described the basic features of skeletal muscle contraction utilizing the force-velocity relationship (12–14). In the study of skeletal muscle during tetanic stimulation, the characteristics of the contractile apparatus were described. These features, as defined by the Hill equation, are now well accepted in muscle physiology. The Hill equation, $(P + a)(V + b) = (Po + a)b$, expresses the force-velocity relationship as a displaced hyperbola with asymptotes at minus a and minus b. The constants a and b, have dimensions of force and velocity, respectively. These constants have been calculated only for frog sartorius and equal the extra heat liberated with shortening (a) and the rate of heat liberation (b) (12–14). Po equals tetanic tension for skeletal muscle, but has been used as the maximum force developed during a single twitch for cardiac muscle (17–20). There are certain limitations in using the Hill equation to obtain constants for esophageal muscle from mechanical measurements. As noted above, the constants have not been derived for this smooth muscle. In addition, there are potential limitations in using a single twitch to obtain force-velocity relations. These measurements were originally formulated for skeletal muscle in terms

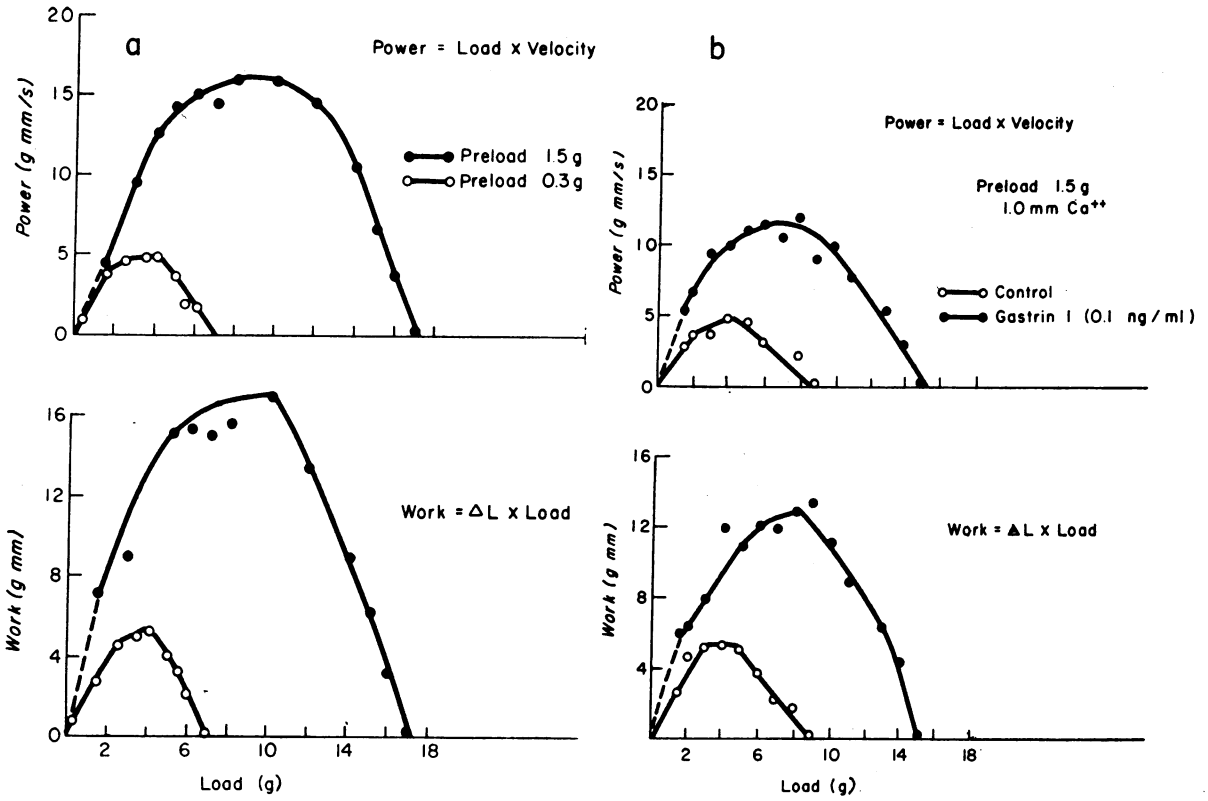

FIGURE 8 Effect of change in muscle length (left) and inotropic intervention by gastrin I (right) on the work and power of LES circular muscle. Work and power formed parallel relations. A change in initial muscle length by an increase in preload from 0.3 g to 1.5 g increased the work and power at any one afterload. Gastrin I produced a similar effect. Both the muscle length and gastrin I also increased the load at which the maximum work and power were achieved.

of tetanus tension. It was possible that because of limitation of activation time and the slow development of the active state in smooth muscle, the twitch tension did not approach tetanus tension. However, as discussed later, the maximum force (Po) generated during the single twitch did not differ significantly from tetanus tension achieved during KCl depolarization. Thus, as cautioned in other studies utilizing smooth and cardiac muscle, the derivation of the constants *a* and *b* for the Hill equation must be viewed as being potentially hazardous (15–26). The data, when evaluated using the Hill equation, did form a linear relationship with a

TABLE III
*Effects of Gastrin I on Force Velocity Determinations**

Gastrin I	LES		Esophagus (1–2 cm above LES)		Esophagus (4–5 cm above LES)	
	V max	Po	V max	Po	V max	Po
	mm/s	*g*	*mm/s*	*g*	*mm/s*	*g*
1.0 mM Ca^{++} (Control)	2.4±0.2	8.0±0.6	2.8±0.3	8.5±0.4	2.3±0.3	9.1±0.5
0.1 ng/ml	6.7±0.6	14.4±1.1	4.8±0.2	14.1±0.7	2.1±0.4	8.6±0.6
1.0 ng/ml	7.9±0.5	16.5±0.5	6.6±0.8	14.2±1.0	3.1±0.7	12.2±1.8
10.0 ng/ml	7.3±0.9	17.1±1.2	5.4±1.6	13.7±1.2	7.4±0.8	18.5±1.0

Each number represents observations made on a minimum of eight muscle strips.
* All studies were performed at 1.0 mM Ca^{++} and at Lo.

high correlation coefficient (all above 0.90), suggesting that the force-velocity relationship could be described as a displaced hyperbola similar to that of other muscles (26).

In all studies described here, Po represents the maximum force of a single twitch produced during a neurogenic response. Thus, Po differed from the tetanic response of skeletal muscle and the single twitch elicited during direct electrical stimulation of cardiac muscle. The use of a neurogenic-mediated muscle contraction as a basis for the force-velocity determinations was necessitated by the response of the muscle to electrical stimulation (6). Esophageal circular muscle could not be tetanized during electrical stimulation and responded only at the termination of stimulation with a prominent contraction. This contraction, called the off response, had been thought to represent the peristaltic response to swallowing in the esophageal body (27) and the sphincteric contraction after its relaxation (28). The use of this neurogenic response to evaluate force-velocity characteristics may, therefore, evaluate the mechanical characteristics of peristalsis and LES contraction. However, there are several possible objections to using a neurogenic response to quantify characteristics of the contractile apparatus. First, the neurogenic response may elicit only a portion of the maximal force that the muscle may generate. Second, intervention of agents to modify contractility may simply alter the neural mechanism. These objections were resolved by determining the response to direct muscle depolarization with KCl. During muscle depolarization, the maximum tension achieved did not differ significantly from the maximum Po developed during an afterloaded contraction. In addition, a diminution in muscle contractility, as defined by a decrease in V max and Po during 1.0 mM calcium superfusion, was also seen with direct muscle stimulation by KCl. Therefore, although a neural response was utilized, these findings suggested that maximum force was achieved and muscle contractility was being affected. Realizing these inherent potential limitations in using a neurogenic-elicited twitch for smooth muscle and having discussed the potential objections in using the Hill equation, we performed studies to further characterize the mechanical properties of esophageal and LES circular smooth muscle. It was felt that, despite these limitations, a reasonable determination of the entire force-velocity curve, a calculated V max, and a measured Po could be compared during conditions of loading and inotropic intervention.

The initial studies on the force-velocity characteristics of the muscle indicated that by increasing length (greater preload), the force developed by the muscle (Po) could be increased. At a length (Lo), the maximum force achieved by the muscle was recorded. Lo represented the length of optimal tension development at which different calcium and gastrin I concentrations were studied. The Lo of esophageal muscle was greater than that of the LES muscle. This finding confirmed previous studies in which the length-tension properties were evaluated under strict isometric conditions in response to acetylcholine (10). The force-velocity curves indicated that peak force (Po) differed markedly with length but the V max varied only minimally with length. This finding suggested that an increase in length produced an increase in force-generating sites within the contractile apparatus, with only a slight change in basic contractility of the muscle (14, 16, 20). Other smooth muscle had also been shown to change both V max and Po with change in initial muscle length or preload (21). Thus, the esophageal and LES muscle generated greater power and performed more work when muscle length was increased toward Lo. However, at any muscle length or preload, both work and power were dependent upon the afterload against which the contraction was generated. These changes with preload and afterload were those generally described for skeletal and cardiac muscle, and represented basic mechanisms by which muscle could alter its function in response to changing needs.

In addition to the alterations in muscle function with preload and afterload, the muscle demonstrated a change in contractility or inotropism in response to gastrin I. The definition of inotropism in force-velocity relationships is that an agent can increase the V max with or without a change in the Po (16, 20, 22). In cardiac muscle, inotropic intervention with chemicals such as norepinephrine, digitalis, or calcium increase both the V max and the Po (16–20). The demonstration that the gastrointestinal hormone gastrin I changed the contractility of the muscle represented a new action for a hormone on gastrointestinal smooth muscle. It had been demonstrated previously that glucagon, a hormone that may be released from the gut and that had a structure similar to secretin, altered the contractility of the heart (29, 30). However, the finding that gastrin I had an apparent inotropic effect on esophageal circular muscle must be considered in the context of the experimental design. First, the effect could be demonstrated most conclusively during a low calcium (1.0 mM) superfusion. Second, the effect of gastrin I and diminished calcium were tested on the neural response of the muscle. Previously, the inotropic effect of norepinephrine on cardiac muscle was demonstrated best in a low-calcium bath (17). In the studies described here, gastrin I had a slight effect on V max during the 2.5 mM calcium superfusion, and a marked effect on both V max and Po during 1.0 mM calcium superfusion. At 2.5 mM calcium, both V max and Po were close

to maximum and could be increased only slightly. The evaluation of muscle contractility during a neurogenic response had been discussed earlier. It would seem that gastrin I had altered muscle contractility and had not simply changed the response of the neural component. However, since a means of performing force-velocity determinations on this muscle without an intermediary neural mechanism was not available, this question cannot be totally resolved. The more sensitive effect of gastrin I on the force-velocity characteristics of the LES was consistent with previous studies (10). The changes in muscle force-velocity properties to a given hormone may be reflected in the specific response to each hormone upon isometric tension without intervening electrical stimulation.

Until these esophageal muscle mechanics can be definitively tested in vivo, they must be interpreted conservatively. It remains possible that the force-velocity properties of opossum esophageal muscle studied during the off response have little relevance to the function of the human esophagus in vivo. The method of in vitro study and muscle excitation may not simulate intact organ function. However, the study was designed to elucidate the properties of the circular muscle component of the esophageal wall. Further study will determine whether the intact esophagus demonstrates the properties of its circular muscle component. Certain analogies between the muscle mechanics described in this study and esophageal performance in vivo can be drawn. These analogies are purely speculative at this time. The distending force within the esophageal lumen may be analogous to the preload. Thus, the volume of a bolus may in part determine the propulsive force of the esophagus. The afterload may be analogous to the impedance against which the bolus must be transported. Since the esophagus moves material from a negative intrathoracic pressure to a positive intra-abdominal pressure, the latter may in part determine the impedance. Thus, the force developed within the esophagus may in part be determined by the impedance against which the bolus is being moved. The inotropic effect of gastrin I altered the force-velocity characteristics of the muscle and thus increased the contractility at any given condition of loading. The power and work of the esophagus may therefore be functions of initial muscle length (preload), impedance to movement (afterload), and endogenous inotropic intervention (gastrin I).

The analogies to intact organ function based on the mechanics of the circular muscle are more easily made for esophageal than for LES function. The LES, being tonically contracted, shows an off response only after termination of the relaxation after deglutition. The correlates of work and power for the LES are not easily made. Work, in the sense of movement, is not really a major function of the LES. However, the concept of power may be applied to LES performance. It would be interesting to suggest that the LES response to increased intra-abdominal pressure was the change in power in response to increasing afterload (2, 8, 9). It also remains possible that these findings in vitro may not be applicable to sphincter function in vivo.

Esophageal and LES muscle can be studied under similarly regulated conditions as cardiac muscle. The application of these concepts to esophageal function should be made. The assessment of esophageal propulsive force should incorporate the parameters of work and power since primary esophageal function involves them. Evaluating esophageal propulsion by measuring amplitude of peristalsis may be analogous to assessing cardiac function solely by measuring blood pressure. In fact, esophageal propulsive force in man correlated to only a fair degree with peristaltic amplitude when both were measured simultaneously (5). LES function should be evaluated to determine the mechanical factors that alter its performance. These studies may provide greater insight into the function of this organ system.

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