THE ANISOTROPIC ELASTIC PROPERTIES OF THE SARCOLEMMA OF THE FROG SEMITENDINOSUS MUSCLE FIBER

STANLEY I. RAPOPORT

From the Laboratory of Neurophysiology, National Institute of Mental Health,
Bethesda, Maryland 20014

ABSTRACT Tension and curvature of the sarcolemmal tube of the frog muscle fiber were measured at different extensions and were used to calculate the anisotropic elastic properties of the sarcolemma. A model was derived to obtain the four parameters of the elasticity matrix of the sarcolemma. Sarcolemmal thickness was taken as 0.1 μm. Over the range of reversible sarcolemmal tube extension, the longitudinal elastic modulus $E_L = 6.3 \times 10^7$ dyn/cm², the circumferential modulus $E_c = 0.88 \times 10^7$ dyn/cm², the longitudinal Poisson's ratio $\sigma_L = 1.2$, and the circumferential Poisson's ratio $\sigma_c = 0.18$. At tubular rest length $E_L = 1.2 \times 10^7$ dyn/cm². The sarcolemma is less extensible in the longitudinal direction along the fiber axis than in the circumferential direction. It can be extended reversibly to 48% of its rest length, equivalent to extending the intact fiber from a sarcomere length of 3 μm to about 4.5 μm. The sarcolemma does not contribute to intact fiber tension at fiber sarcomere lengths <3 μm, and between 3 and 4 μm its contribution is about 20%. It also exerts a pressure on the myoplasm, which can be calculated by means of the model. The longitudinal elastic modulus of the whole fiber is 1×10^6 dyn/cm² at a sarcomere length of 2.33 μm.

INTRODUCTION

The sarcolemma is a four-layered structure on the surface of the striated muscle fiber and is composed, from inside to outside, of a plasma membrane, a basement membrane, a collagen layer, and an outer reticular layer, having together a thickness δ of about 0.1 μm (Jones and Barer, 1948; Wang, 1956; Mauro and Adams, 1961; McCollester, 1962) or more (Koketsu et al., 1964). The elastic properties and contribution of the sarcolemma to whole fiber tension at different extensions have not been definitely established. The sarcolemma can exert a tensile force equal to that of the fiber under some conditions (Street and Ramsey, 1965), but appears not to exert tension at sarcomere lengths S below 3 μm (Podolsky, 1964; Rapoport, 1970, 1972).

At $S > 3.2$ μm, experiments on stripped single fibers indicate that the sarcolemma contributes about 80% of passive fiber tension (Podolsky, 1964). Comparison of the length-tension curve of the sarcolemmal tube with that of the intact fiber, however, suggests that the sarcolemma contributes little to passive fiber tension (Casella, 1950). The sarcolemmal tube is formed by damaging a single fiber so as to produce a sarcolemmal region free of myoplasm, and probably does not contain the intact plasma membrane (Mauro and Adams, 1961). Results using the elastimeter method suggest also that the sarcolemmal contribution to fiber tension at $S < 3.75$ μm is probably small (Rapoport, 1970, 1972).

Any theory on myoplasmic elasticity depends on knowing relative myoplasmic and sarcolemmal contributions to passive fiber tension (e.g., Hill, 1968). It is difficult to interpret previous experiments on the sarcolemmal tube because either (*a*) the reference sarcomere length of the intact fiber to which tubular extension could be referred was not determined, or (*b*) length-tension studies on the tube were probably made after it was deformed irreversibly (see below) (Casella, 1950; Sten-Knudsen, in Buchthal et al., 1951; Fields, 1970). In the present experiments, these two factors were taken into account so as to estimate the role of the sarcolemma in passive muscle tension.

Four elastic coefficients are needed to describe the tubular surface, a longitudinal and circumferential elastic modulus, and a longitudinal and circumferential Poisson's ratio (cf. Love, 1944; Westergaard, 1952). We formulated a model in Appendix I to calculate these coefficients from the tension, extension, and curvature of the sarcolemmal tube. An abstract of this work has been published (Rapoport and FitzHugh, 1971).

LIST OF SYMBOLS

a, b	Parameters defined by Eq. A 9.
c_{ij}	$i, j = 1, 2$; elastic constants of Eq. A 1 in centimeters per dyne.
ds	Distance between two points on sarcolemmal surface; ds_0 equals the initial separation at sarcolemmal rest length.
e	Extension, or $(S - S_0)/S_0$; $(ds - ds_0)/ds_0$.
e_2	$(S - 2\ \mu\text{m})/2\ \mu\text{m}$.
E_L, E_c	Longitudinal and circumferential elastic moduli in dynes per square centimeter.
E	Elastic modulus in dynes per square centimeter (Eq. 6)
F, G	Longitudinal and circumferential stresses in dynes per centimeter.
H	Horizontal force (tension) on tube or fiber in dynes.
k_{2n}	$n = 1, 2, \ldots$; constants of polynomial of Eq. 1.
L	One-half length of tube; $L = L_0$ at rest length.
P	Pressure exerted by sarcolemma on myoplasm of intact fiber in dynes per square centimeter.
r	Radius of intact fiber in centimeters; $r = r_2$ when $S = 2$ μm.
S	Sarcomere length of fiber in microns; S_0 equals the initial value; $S = S_3$ at 3 μm.
x	Horizontal distance from center of tube, $-L \leq x \leq L$.
y	Radius of sarcolemmal tube in centimeters; $y = y_0$ at $L = L_0$; $y = y_3$ at $S = 3$ μm.

δ Sarcolemmal thickness equals 0.1 μm.
σ_L, σ_c Longitudinal and circumferential Poisson's ratios.

METHODS

A single fiber of the semitendinosus of female *Rana pipiens* was held, at room temperature (about 22°C), by its tendinous ends with two forceps, one of which was movable and the other of which was fixed to a Pickering Core LVDT transducer (model no. 7411, Pickering and Company, Plainview, N.Y.). The experimental apparatus (Fig. 1) was designed by Mr. James Bryan. The transducer output was fed to a bridge, whose output was 5.5 mV/dyn and linear to 200 dyn. The bridge output was analyzed with an averaging digital voltmeter and could be read, over a 1 min period, to ± 1.1 mV. The fiber was kept in Ringer's solution, whose composition is, in millimolars, K^+, 2.5; Na^+, 120; Cl^-, 121; Ca^{2+}, 1.8; $(HPO_4)^{2-}$, 2.15; $(H_2PO_4)^-$, 0.85.

Regions on the intact fiber were marked initially by dispersing graphite particles in the bathing solution. The tension-length analysis of the whole fiber was made by stretching it in quick steps, starting first at a rest length at which the fiber was just horizontal and waiting 5–10 min before making a measurement. Tension was recorded and photographs of the muscle at high power were compared with photographs of an immersed micrometer scale in order to calculate fiber radius and sarcomere length S. The radius is not exact because the fiber is not a perfect cylinder (Blinks, 1965). The muscle was stimulated three times before and after an analysis, so as to reduce hysteresis effects from stretching (cf. Buchthal et al., 1951, p. 40).

The sarcolemmal tube was formed at rest length or at about 1.4 times rest length of the whole fiber by squeezing the fiber with forceps or by raising it out of solution for 1 min. The fiber first was photographed along its entire length at low power. 1 h after damage, regions of clot and sarcolemmal tube had stabilized and the damaged fiber was photographed again along its entire length. Positions of the identified graphite markers and connective tissue attachments permitted, in some cases, evaluation of the rest length and degree of elongation of a tube (cf. Casella, 1950).

The damaged muscle was stretched in quick steps and photographs taken and tensions obtained after 10–15 min. After a quick stretch, tension rose quickly and then decreased

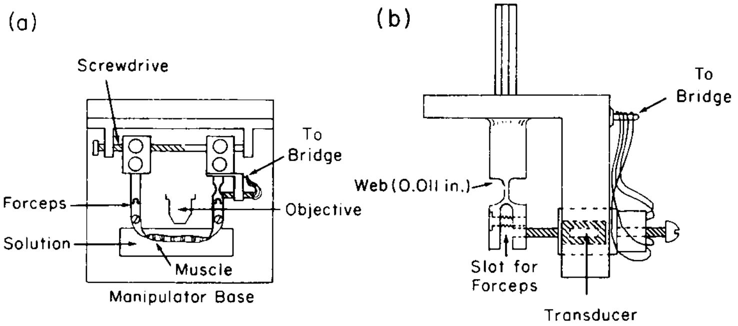

FIGURE 1 Muscle-holder and tension-recording device. The single fiber is held by two forceps in Ringer's solution and can be observed and photographed through the microscope objective. The left holder in *a* can be moved by a screwdrive to stretch the muscle. The right is fixed and will bend with tension at the web (*b*), moving the core of the transducer, whose output is fed to a bridge and then fed to an averaging digital voltmeter.

with time, possibly because of relaxation of clotted parts of the fiber as well as of the tubular sarcolemma. Tension stabilized after 10 min, and the dimensions of tubes photographed 10 and 15 min after a quick stretch did not differ. We did not photograph tubes within 10 min after a stretch. Any hydrostatic pressure differences after a quick stretch should disappear within 1 min, since the sarcolemma, even when attached to the fiber, is very permeable to water (Blinks, 1965).

Length-tension characteristics of individual sarcolemmal tubes were determined by measuring the distance between identified points, the tubular diameter, and curvature. Reversibility of extension was tested by returning the damaged fiber to zero tension after different extensions.

Analysis of Curvature

Photomicrographs of a sarcolemmal tube at different extensions (Figs. 6, 7) were enlarged and the tube was divided into four quadrants (Fig. A 1), $y > 0, x > 0$; $y > 0, x < 0$; etc. The origin ($x = y = 0$) and x axis were chosen by eye for symmetry, and the coordinates of the curve in each quadrant were obtained in digital form with a decimal converter model F (Benson-Lehner Corp., Van Nuys, California). Errors arose in the choice of origin and of x axis, and because of some asymmetry of the tube. In addition, the ends of the tube may not be exactly circular (Fields, 1970), as is assumed in the model of Appendix I. The coordinates of the curves in two or four quadrants were pooled as a function of x and were fit to an even polynomial by least squares on a remote time-sharing IBM digital computer (Call/360, Service Bureau Corporation, Statpack polynomial regression program). Two quadrants composing one entire side of the tube were used, rather than four, when the other side had pieces of carbon particles that obscured its surface outline.

The data were fit by an even polynomial of the form,

$$y(x) = k_0 + k_2x^2 + k_4x^4 + \cdots + k_{2n}x^{2n}, \quad (n = 1, 2, \cdots), \tag{1}$$

where the k_{2n} are constants. It was found that a fourth or sixth degree polynomial usually gave a satisfactory fit, and higher degrees did not give significantly better fits at the $P = 0.05$ level (Brownlee, 1960). The k_{2n} were usually positive for fourth or sixth degree fits.

Fig. 2 shows a sixth degree polynomial fit of data from four quadrants (both sides) of a tubular profile. Deviations from the curve are due to incomplete symmetry of the quadrants and errors in choices of origin and coordinate axes. The 12th degree polynomial fit was not significantly better than the 6th, but had variations in the first and second derivatives of y which prohibited application of the model to the curve (see below).

The polynomial of Eq. 1 was differentiated to give y' and y'' as a function of x, which together with the observed values of y_0, y, and H, were inserted into Eq. A 9 to obtain equations of the type,

$$yy'' = X_1(x)a + X_2(x)b, \tag{2 a}$$

or

$$a = -\frac{X_2(x)}{X_1(x)}b + \frac{y(x)y''(x)}{X_1(x)}, \tag{2 b}$$

where a and b are variables and $X_1(x)$ and $X_2(x)$ are functions of x, as defined in Eq. A 9

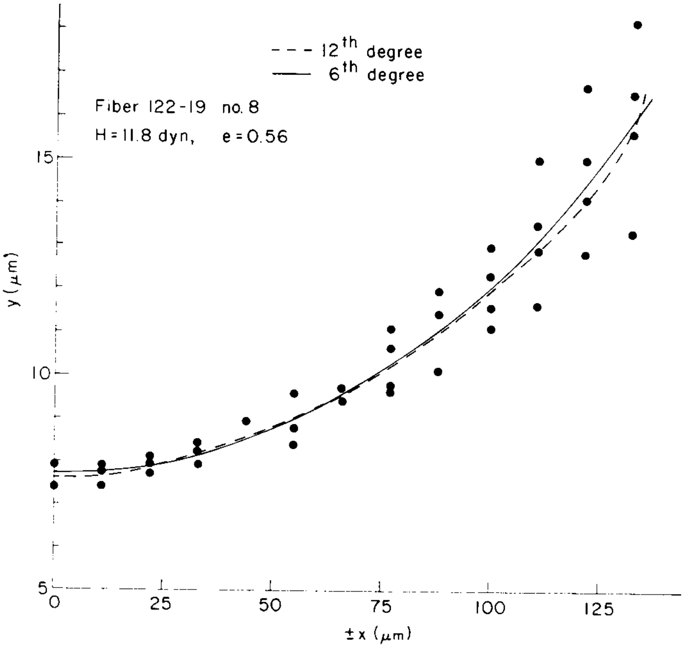

Figure 2 Fit of polynomial equation to data from four quadrants of tube. The y coordinates of each quadrant are plotted as a function of x, and the curves show the least square fits of a 6th and 12th degree polynomial equation (Eq. 1).

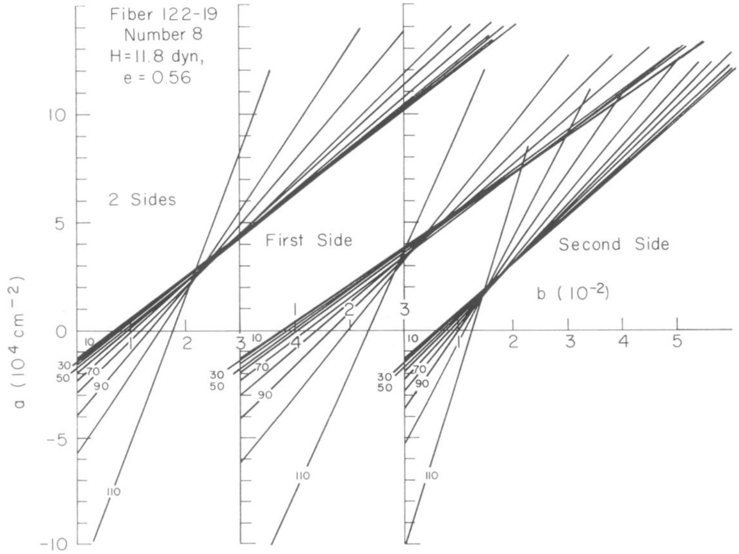

Figure 3 Plots of a against b as given by Eq. 2 b. The numbers associated with the lines are the values of x(μm) in Eq. 2 b. The analysis is for the tube of Fig. 2. The sixth degree polynomial (Eq. 1) gives good focusing if both sides of the tube are analyzed together (four quadrants), or if either side is analyzed independently. The values of a and b for both sides are bracketed by the values obtained from either side.

The variable a was plotted as a linear function of b for different values of x in 5–10 μm steps between $\pm L$. If a and b satisfy Eqs. 2 for each x, linear plots of b against a should overlap or focus at this point of agreement, from whence the parameters c_{21} and c_{22} can be obtained.

Fig. 3 shows plots of a against b for a sixth degree polynomial fit to four quadrants and to each of the two sides of the sarcolemmal outline of the fiber in Fig. 2. The region of overlap or focusing, in which a and b satisfy Eqs. 2 for all x, was apparent in all conditions. The values of a and b were 4.8×10^4 cm^{-2} and 3.6×10^{-2} for the first side and 2.4×10^4 cm^{-2} and 1.7×10^{-2} for the second side. They bracket the values from the four-quadrant fit, 3.3×10^4 cm^{-2} and 2.5×10^{-2} for a and b, respectively. In view of the large variance among tubes of the parameters calculated from a and b (Table I), the error in choosing one side or the other or all four quadrants together is unimportant for the general results.

RESULTS

Intact Fiber

The length-tension curves of 10 of 13 intact fibers and of their 13 respective sarcolemmal tubes were determined. Fiber extension e at sarcomere length S is defined by the following equation, where S_0 is fiber sarcomere length at rest

$$e = \frac{S - S_0}{S_0}. \qquad (3)$$

If $S_0 = 2$ μm, then $e = e_2$; if $S_0 = 3$ μm, $e = e_3$.

The length-tension curve of an intact fiber is shown in Fig. 4. The elastic modulus of the fiber can be obtained by fitting the following equation to the data (cf. Sten-Knudsen, 1953),

$$H/\pi r_2^2 = \frac{f_0}{A} \{\exp(A[e_2 - B]) - 1\} \text{ dyn/cm}^2. \qquad (4)$$

A, f_0, and B are constants, and H is the horizontal force on the fiber. r_2 is fiber radius at $S = 2$ μm, calculated by the following equation at constant fiber volume (Huxley, 1953),

$$r_2 = r\sqrt{S_0/2} \ \mu\text{m}, \qquad (5)$$

where r is fiber radius at S_0. πr_2^2 was chosen as the reference cross section because the minimal sarcomere length at which passive tension can be exerted is 2 μm (Huxey, 1964).

Figure 5 is a plot of ln $(H/\pi r_2^2)$ against e_2 for lumped observations, together with the least squares curve obtained by Eq. 4. The curve has the following constants: $A = 4.04$, $B = 0.166$, and $f_0 = 9.8 \times 10^4$ dyn/cm^2.

The elastic modulus at different extensions of the intact fiber is defined by the

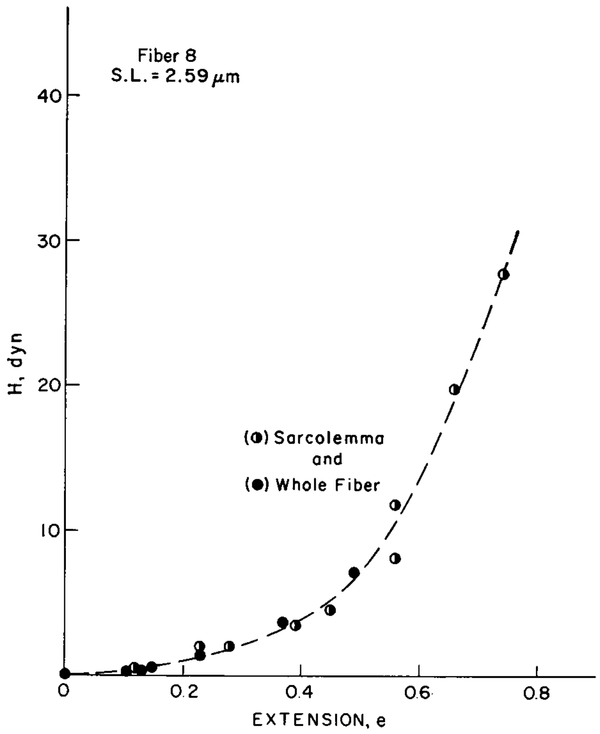

FIGURE 4 Tension-length curve of single fiber and of sarcolemmal tube formed from that fiber. The points can be fit roughly by the same curve. S. L., sarcomere length.

following equation (Condon and Odishaw, 1958; Sten-Knudsen, 1953):

$$E = \frac{d(\text{stress})}{d(\text{strain})} = \frac{d(H/\pi r_2^2)}{de_2} \text{ dyn/cm}^2. \tag{6}$$

Differentiating Eq. 4 with respect to e_2 and using the definition of Eq. 6 gives,

$$E = f_0 \exp (A[e_2 - B]). \tag{7}$$

By Eq. 4, $e_2 = B$ at fiber rest length, and from the value of B found above, the mean $S_0 = 2.33$ μm, in agreement with the value of 2.45 μm found by Edman (1966). The elastic modulus of the fiber at rest length is f_0, or 1×10^5 dyn/cm^2, agreeing with the value found by the elastimeter method (Rapoport, 1972), but less than reported by Sten-Knudsen (1953) and Buchthal et al. (1951). These latter authors used rest tensions $H > 0$ and therefore started out at rest sarcomere lengths >2.3 μm (see Discussion).

Sarcolemmal Tube

Figs. 6 and 7 are photographs of sarcolemmal tubes at different longitudinal ex-

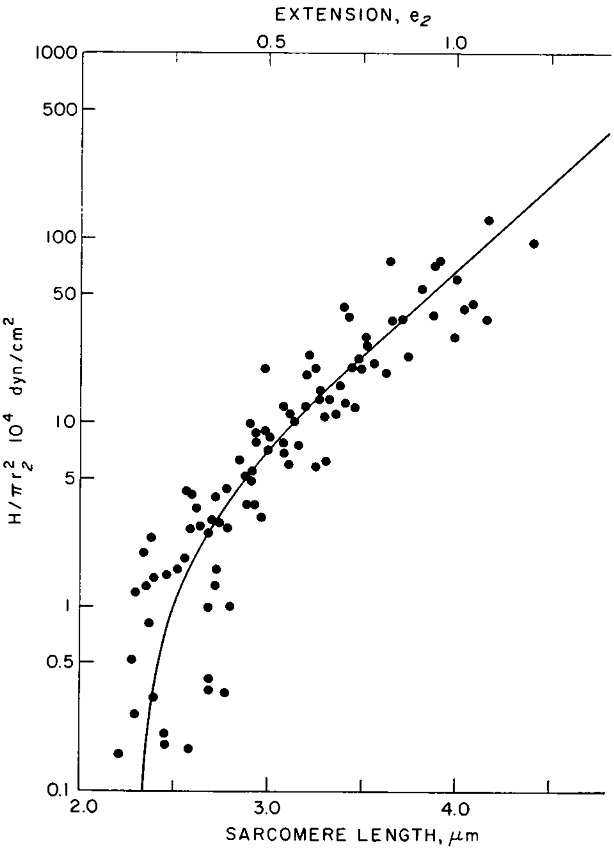

FIGURE 5 Lumped data on whole fibers in tension-length relation. r_2 is the calculated radius of the fiber at $S = 2$ μm, assuming constant volume with extension. H is the longitudinal force in dynes. The data are fit by least squares to Eq. 4.

tensions e, defined by the equation,

$$e = \frac{ds - ds_0}{ds_0}. \quad (8)$$

ds_0 is the distance along the x axis between two identifiable points at tube rest length (when the damaged fiber lies just horizontal), and ds the distance after a stretch (Fig. A 1). Not enough points could be identified to form a grid on the tubular surface so as to show a consistent relation between ds and the position x along the tubular axis. Because of this experimental limitation, we took e as the mean extension between pairs of identified marks along the axis and could not use Eq. A 10 to calculate σ_c. E_L was approximated by letting $c_{12}G = 0$, thereby neglecting circumferential elasticity in the equation. From the stress-strain (length-tension) observation on a sarcolemmal tube, $E_L = H/(2\pi y_0 \delta e)$ (cf. Eq. 6). Sarcolemmal thick-

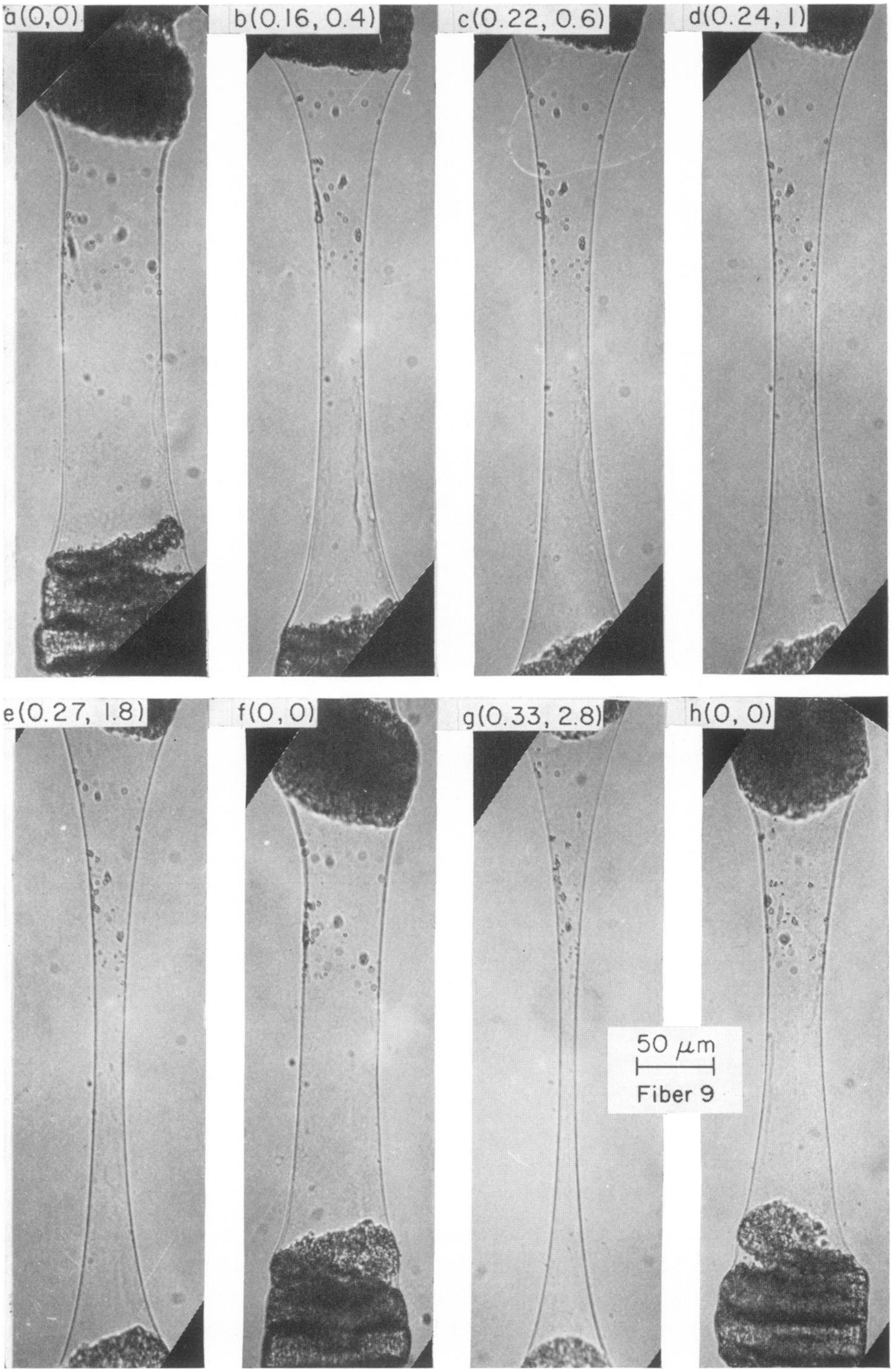

FIGURE 6 Sarcolemmal tube at different extensions. The numbers in parentheses are paired values of extension e and tension H. The figure shows reversible extension to e = 0.33, although radius y_0 has decreased (compare Figs. 6 *a*, *f*, *h*). A singular solution for σ_c and E_c could be found for each of the extensions in the figure by the method of Fig. 3.

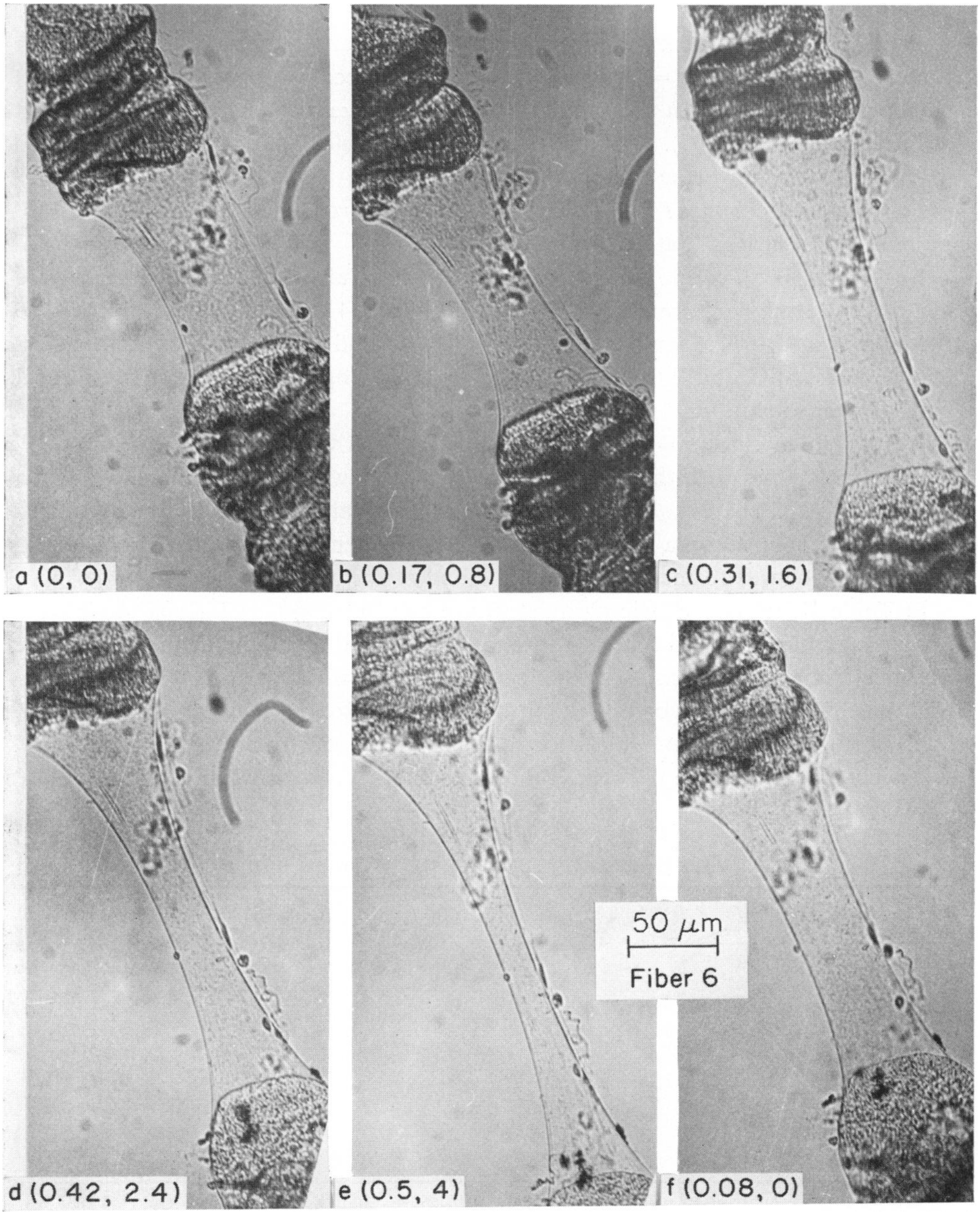

FIGURE 7 Sarcolemmal tube at different extensions. The numbers in parentheses are paired values of extension *e* and tension *H*. The figure shows that the tube is stretched irreversibly at Fig. 7 *f*. A singular solution for E_c and σ_c could be obtained only for the tubes in Figs. 7 *b*–*d* by the method of Fig. 3.

ness δ was taken as 0.1 μm. H is the horizontal force on the tube, and y_0 the radius at end of tube.

Table I illustrates observations and calculations at different extensions of the tube of fiber no. 8. At $e = 0.56$, y_0 is markedly reduced, probably because the soft clot masses holding open the tubular ends are squeezed by circumferential, purse-string tension (Figs. 6, 7). The median values of the calculated parameters are listed in the table without being corrected for changes of y_0, since it is not clear how E_c, σ_L, or σ_c would change. E_L in Table I can be normalized to $y_0 = 61$ μm at $e = 0$ by multiplying it by y_0 (at e)/61 μm.

Results on 13 tubes and fibers are summarized in Table II. Mean fiber sarcomere length before tubular formation is 2.45 μm. Fibers 1–4 and 11–13 were stretched to between 20 and 40% when the tubes were formed, and the others were kept at rest length. Tubes 1–5 and 9–10 were formed by squeezing, the others by withdrawal from Ringer's solution.

At its rest length at $e = 0$, the sarcolemmal tube is wider by 18% and longer by 26%, as compared with the corresponding region between two identified marks on the intact fiber. The 26% extension is somewhat smaller than the 46% initial extension found by Casella (1950), and does not depend on the fiber extension at which the tube was formed nor on the method of formation.

In Table II, tubular elastic parameters are calculated for extensions at which a unique a and b are found for the entire length of the tube by the method of Fig. 3. The maximal mean extension e(max), at and below which unique a's and b's are found, is less than 0.42, corresponding to a mean horizontal tension $H(\text{max}) = 8.2$ dyn. Furthermore, e(max) does not differ ($P < 0.05$) from the upper limit of reversible extension e(rev), which is the extension beyond which the tube does not

TABLE I

ANALYSIS OF SARCOLEMMAL TUBE OF FIBER NO. 8

e	H	y_0	E_L	E_c	E_L/E_c	σ_L	$E_L y_0/61$	σ_c
	dyn	*μm*	*dyn/cm² × 10⁷*				*dyn/cm² × 10⁷*	
0	0	61.0						
0.122	0.4	56.6	0.92	0.45	2.1	0.53	0.86	0.26
0.28	2.0	51.6	2.20	0.25	8.8	0.85	1.88	0.10
0.39	3.6	43.7	3.35	0.88	3.8	0.40	2.42	0.10
0.56	11.8	14.3	23.5	1.02	23.0	0.63	5.53	0.03
			2.78*	0.67*	6.3*	0.58*	2.15*	0.10*

$S_0 = 2.59$ μm. Extension (e) was determined for different tensions (H), and y_0, the end tubular diameter, was measured. The longitudinal and circumferential elastic moduli were calculated at different extensions as $E_L = H/(2\pi y_0 \delta e)$ and $E_c = a y_0 H/2\pi\delta$. The Poisson's ratios were calculated as $\sigma_L = bE_L/E_c$ and $\sigma_c = \sigma_L E_c/E_L = b$. $E_L y_0/61$ μm represents the longitudinal elastic modulus normalized to the sarcolemma radius of 61 μm at $e = 0$.

* Median.

TABLE II

ANALYSIS OF SARCOLEMMAL TUBES

Fiber no.	S_0	Initial widening of tube	Initial extension of tube	e(rev)	H(max)	e(max)	y_0 at $e = 0$	E_L	E_c	E_L/E_c	σ_L	σ_c
	μm	*Fractional change*			*dyn*		*μm*	*dyn/cm² × 10⁷*				
1	2.48	0.35	—	—	2.8	0.27	46.3	1.91	0.27	4.5	1.2	0.26
2	2.30	0.43	—	—	8.4	0.42	47.0	6.31	0.45	11.1	1.5	0.20
3	2.24	0.37	0.80	—	9.8	0.21	54.0	15.5	1.09	15.1	2.3	0.18
4	2.32	−0.23	0.18	0.15	1.2	0.05	30.2	16.4	0.16	108.0	3.5	0.04
5	2.36	−0.09	0.26	0.44	24.2	0.30	32.7	39.3	1.30	36.5	1.2	0.03
6	2.36	0.39	—	0.44	4.0	0.50	56.0	2.46	0.88	2.8	0.73	0.31
7	2.33	−0.23	0.45	0.47	3.8	0.34	25.2	6.70	0.53	26.0	0.92	0.06
8	2.59	0.43	0.10	0.65	11.8	0.56	61.0	2.78	0.67	6.3	0.58	0.10
9	2.23	0.20	0.25	0.38	2.8	0.33	53.0	2.15	0.22	8.2	0.99	0.12
10	2.24	0.15	0	0.70	9.6	0.46	39.0	9.04	1.30	8.2	1.5	0.15
11	2.49	0.12	0.05	0.64	8.1	0.63	58.9	2.91	1.05	2.9	0.70	0.27
12	2.30	0.18	0.14	0.58	10.4	0.63	55.0	4.24	2.26	2.0	0.74	0.36
13	2.27	0.30	0.34	0.37	9.4	0.32	48.0	8.56	1.27	6.8	1.4	0.18
Mean	2.35	0.18	0.26	0.48	8.2	0.42	46.6	6.31*	0.88*	8.2*	1.2*	0.18*
±SE	±0.03	±0.07	±0.07	±0.06	±1.6	±0.04	±3.2					

The rest sarcomere length of the intact fiber S_0 is given in the second column. The third column shows the initial widening of the tube as the fractional increase of y_0 with reference to the radius r of the intact fiber at S_0. The fourth column has initial elongation of a tubular region at tubular rest length as compared with its rest length in the intact fiber. The fifth column gives the maximum extension from which the tube returned to its original rest length, e(rev), in those tubes in which reversibility was studied. The sixth and seventh columns have the paired maximal tensions H(max) and extensions e(max) at or below which singular values of E_c and σ_c could be obtained by the model. y_0 (eighth column) is the radius of the sarcolemmal tube at $e = 0$. E_L (ninth column) is the median longitudinal elastic modulus for a tube, calculated as $E_L = H/(2\pi y_0 \delta e)$, where y_0 is the end radius at e. E_c (10th column) is the median circumferential elastic modulus for a tube, calculated as $E_c = a y_0 H / 2\pi\delta$ at different extensions. E_L/E_c is the median value of the ratio of the moduli, and σ_L and σ_c are the median longitudinal and circumferential Poisson's ratios. Medians were chosen to reduce the effect of extreme values on estimates. In the table, the median value for E_c does not necessarily correspond to that of E_L or of E_L/E_c. For this reason, E_L, E_c, and E_L/E_c may not agree exactly.

* Median.

return to rest length after tension is released. Equivalence of e(max) and e(rev) shows that the model of Appendix I can be used to find elastic parameters of the tube only for reversible tubular extensions.

Table II shows that the median value of E_L, uncorrected for changes in y_0, is 6.3×10^7 dyn/cm² for mean extensions up to $e(\max) = 0.42$. The 95% confidence interval for E_L is 2.9–15.5 × 10^7 dyn/cm². E_L, if normalized to y_0 at $e = 0$ as in Table I, is 4.9×10^7 dyn/cm².

E_L at $e = 0$ was estimated by extrapolating the following equation, which represents the tubular length-tension relation (cf. Eq. 4),

$$H = \frac{f_0'}{A'}\,[\exp\,(A'e) - 1]\ \text{dyn}. \qquad (9)$$

Fig. 8 is an example of a least squares fit to the logarithmic expression of Eq. 9 that gives the constants A' and f_0'. The longitudinal elastic modulus at tubular rest length ($e = 0$) equals $f_0'/2\pi y_0\delta$ (cf. Eq. 6), and was calculated from the best fit constants for the individual tubes where y_0 equals the tubular radius at $e = 0$. The median of E_L at $e = 0$ is 1.2×10^7 dyn/cm^2 (95% confidence interval is 0.5–7.0 $\times 10^7$ dyn/cm^2), smaller than the 6.3×10^7 dyn/cm^2 which is found for the entire region of reversible stretch. The difference is due to the arbitrary extrapolation and to an increase of E_L with extension (Table I, see Discussion).

Circumferential Elastic Modulus E_c and Poisson's Ratios σ_L and σ_c

The model of Appendix I was used to calculate these quantities (Tables I and II). From the definitions of Eqs. A 2 and A 9, it can be shown that $E_c = ay_0H/2\pi\delta$,

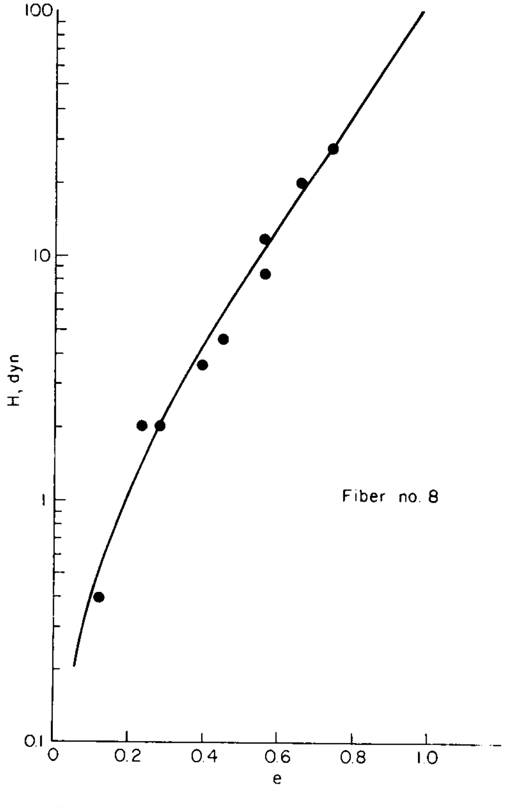

FIGURE 8 Least square fit of Eq. 9 to length-tension observations on sarcolemma of fiber 8.

$\sigma_L = bE_L/E_c$ and $\sigma_c = \sigma_L E_c/E_L = b$. σ_c was not calculated from Eq. A 10 because of the experimental limitation discussed above. In Table II, the median of $E_c = 0.88 \times 10^7$ dyn/cm^2 (95% confidence interval is from 0.3 to 1.3×10^7) and of $E_L/E_c = 8.2$ (95% confidence interval is from 2.9 to 26.0). Thus, the sarcolemma in the region of reversible extension is anisotropic, being less extensible in the longitudinal than in the circumferential direction. The median of σ_L is 1.2 (95% confidence interval is 0.7–1.5), and of $\sigma_c = 0.18$ (95% confidence interval is 0.06–0.36).

Relation of Elastic Properties of Sarcolemmal Tube to Its Properties in the Intact Fiber

The length-tension curve of an individual sarcolemmal tube may fall on, below, or above the corresponding fiber curve when not corrected for rest-length extension. Both curves may overlap, as in Fig. 4 (cf. Ramsey and Street, 1940), but if the tube is first stretched irreversibly to $e = 0.8$, its curve will fall below that of the fiber (see Sten-Knudsen, in Buchthal et al., 1951, p. 111). As the limit of reversible stretch is approached, the sarcolemmal tube becomes less extensible and E_L increases (Table I). The length-tension curve is hyperbolic.

The combined length-tension relation for the sarcolemmal tubes of Table II is shown in Fig. 9 as a function of sarcomere length, which was calculated for each tube by noting the rest sarcomere length of the intact fiber and the initial extension of the tube at $e = 0$. The figure shows that the rest length of the tube is equivalent to a fiber sarcomere length of about 3 μm, in agreement with observations by Podolsky (1964) and Rapoport (1972). When compared with the combined length-tension curve of intact fibers, it would appear that the sarcolemma contributes to fiber tension to a small degree only when $S > 3$ μm.

It should be recognized, however, that the length-tension relation for the sarcolemma around the relatively incompressible fiber myoplasm will differ from the relation for the empty tube. In Appendix II, we use the two-dimensional stress-strain matrix of Appendix I to derive the length-tension contribution of the sarcolemma to intact fiber tension from estimated elastic moduli and Poisson's ratios of the tube. The sarcolemmal radius in the fiber does not change along the longitudinal axis (y the same for all x), and can be calculated from the constant volume constraint on the fiber at different extensions. Fig. 9 gives one sarcolemmal length-tension (H) curve calculated with use of Eqs. A 19 and A 21 and with values of elastic parameters taken from Table II: $E_L = 6.31 \times 10^7$ dyn/cm^2, $E_c = 0.88 \times 10^7$ dyn/cm^2, $\sigma_L = 1.2$, $\sigma_c = 0.18$, $\delta = 0.1$ μm, and F was taken as zero at $S = 3$ μm in the intact fiber, when radius $y_3 = 40$ μm.

In Eq. A 19, the term $E_L\delta/(1 - \sigma_L\sigma_c)$ is the main scaling factor for calculating F and H; as the product $\sigma_L\sigma_c \rightarrow 1$, $H \rightarrow \infty$. The other term in Eq. A 19 depends on initial circumferential stress G_3, which is unknown. For $G_3 = 0$, Fig. 9 shows that the calculated sarcolemmal contribution to intact fiber tension is about 20%

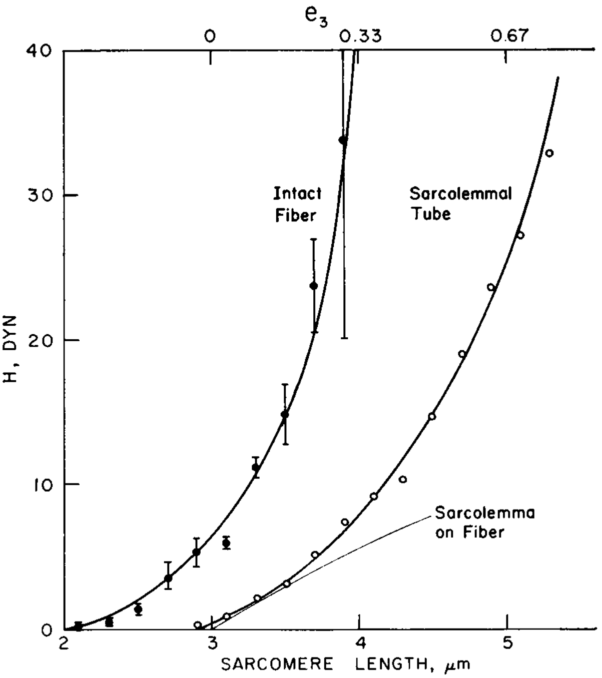

FIGURE 9 Calculated sarcolemmal contribution to whole fiber tension at different sarcomere lengths. The mean curve for the intact fiber was drawn by eye from observations, given as means ±SE. The curve for the sarcolemmal tube was taken from the median values of length-tension observations of individual tubes, corrected for initial extension and fiber sarcomere length. The curve for the sarcolemma as part of the intact fiber was calculated by Eqs. A 19 and A 21 for $G_3 = 0$, using median values for elastic parameters from Table II. It was assumed that $F = 0$ at $S = 3$ μm, $y_3 = 40$ μm, and $\delta = 0.1$ μm.

for S between 3 and 4.5 μm. It is less if $G_3 > 0$. Similarly, for these extensions the pressure exerted by the sarcolemma on the myoplasm can be estimated with Eqs. A 20 and A 23 under the same initial conditions used to calculate H in Fig. 9. $P < 7 \times 10^3$ dyn/cm² if $G_3 = 0$, and $P < 1.9 \times 10^4$ dyn/cm² if $G_3 = 20$ dyn/cm.

DISCUSSION

There are two ways to interpret length-tension observations on the sarcolemmal tube (Casella, 1950). The first is that the empty tube, when elongated, does not differ elastically from the sarcolemma in the intact fiber. If this were so, these experiments show that the sarcolemma does not contribute to whole fiber tension at $S < 3$ μm, and at $S > 3$ μm may contribute about 20%. The lack of a sarcolemmal contribution below 3 μm agrees with observations made with other methods (Rapoport, 1972; Podolsky, 1964). Between 3 and 4 μm, Rapoport (1972) estimated the contribution to be about 10% and Podolsky (1964) suggested it was about 80%.

Another interpretation of the results is that the longer rest length of the sarcolemmal tube, as compared with its length in the intact fiber, indicates an irreversible change of sarcolemmal structure. The tubular sarcolemma does not have an intact plasma membrane (Mauro and Adams, 1961), but the plasma membrane would be expected to contribute little to sarcolemmal tension (Höber, 1945). If the sarcolemma changed structure, reconstruction of sarcolemmal curves as a function of sarcomere length (Fig. 9) is not justified.

Stripping the sarcolemma from a fiber in oil may extend the fiber to $S = 4.5$ μm (Podolsky, 1964). Since this is the estimated limit of reversible tubular extension, and may require a tubular tension of 50 dyn (Fig. 9), it is possible that the sarcolemma is deformed when the tube is being formed. This possibility is not dealt with by our experiments, but the agreement of tube rest length with rest length by other estimates suggests that the sarcolemmal tube is not drastically changed when formed.

In these experiments, we specify the reference sarcomere length of the intact fiber, the rest-length extension of the sarcolemma, and the region of reversible extension. These three factors should be considered if tubular results are to be related to intact fiber physiology. Comparison with data when extension is not referred to sarcomere length of the fiber is difficult because the definition of rest length varies (Delèze, 1961; Sten-Knudsen, 1953; Buchthal et al., 1951), and studies at extensions $e \gg 0.48$ apply to irreversibly deformed tubes (cf. Sten-Knudsen, in Buchthal et al., 1951, p. 111). Fields (1970) worked at tensions > 1000 dyn/cm, while dividing the values of H in Table II or Fig. 9 by sarcolemmal circumference shows that 1000 dyn/cm would produce irreversible deformation. In addition, Fields did not refer sarcolemmal extensions to a reference sarcomere length. For these reasons, it is difficult to agree with his conclusion that the sarcolemma limits fiber length between 70 and 140% of fiber rest length (Fields and Faber, 1970).

According to the model of Appendix I, the tubular sarcolemma is characterized by four elastic coefficients, the circumferential and longitudinal elastic moduli, and Poisson's ratios. The model leads to equations which can be used to obtain these coefficients from the horizontal tension on the tubular ends and the curvature and diameter of the tube.

The data are not good enough to show if ds is related to x for the tube, so that only three coefficients could be calculated initially, E_L, E_c, and σ_L. σ_c was obtained from them because of the reciprocity relation of Eq. A 2 *d*. Had Eq. A 10 been amenable experimentally, it could have been used to check the value of σ_c.

The elastic coefficients vary from preparation to preparation, perhaps because of differing quantities of connective tissue or differing degrees of irreversible deformation. Even with this variability, in some cases it appears that E_L and E_c increase with extension (Table I). This is expected for E_L from the hyperbolic nature of the length-tension curve of the sarcolemma (Figs. 4, 9). The assumption that the tube is linearly elastic is not entirely correct, which suggests that the parameters calculated by means of the model should be considered as approximations. Also, they apply only to the region of reversible tubular extension.

For reversible extensions the sarcolemma is anisotropic, being less extensible in the longitudinal direction ($E_L \gg E_c$). This conclusion extends Fields's (1970) demonstration of anisotropy at high tensions and irreversible extensions to low tensions and reversible extensions. The longitudinal modulus E_L for S between 3 and 4 μm is 6.3×10^7 dyn/cm^2, close to but somewhat higher than the 5×10^6 dyn/cm^2 found by the elastimeter method (Rapoport, 1972). E_L calculated by extrapolation to $e = 0$ is 1.2×10^7 dyn/cm^2, and not significantly different from the elastimeter value.

Sarcolemmal extension to $S = 4$ μm does not represent intrinsic extension of collagen fibers which compose a large part of the sarcolemma, since the elastic modulus of collagen is about 10^{10} dyn/cm^2 (Harkness, 1968). The comparatively low sarcolemmal modulus may be due to elastic fibers, to a thick polysaccharide-protein basement membrane (cf. Mauro and Adams, 1961; Carton et al., 1962; Abood et al., 1966), or to progressive recruitment of slack collagen fibers, which are oriented mainly longitudinally in the sarcolemma (Boyde and Williams, 1968). If the sarcolemmal tube has a knit-stocking array (cf. Mulling and Guntheroth, 1965), it could be extended longitudinally and would contract circumferentially with a low longitudinal elastic modulus E_L before collagen is stretched. In addition, σ_L may be large for a loose fiber network, depending on the orientation of the knit-stocking array with respect to the direction of extension.

Poisson's ratio for natural rubber and gelatin is 0.5 and represents extension at constant volume (Condon and Odishaw, 1958). Since $\sigma_L \simeq 1$ for the sarcolemma, much of the circumferential shrinkage with tubular extension can be ascribed to lateral contraction as represented by Poisson's ratio. We plan to compare area measurements of the tube with those of a model tube with known elastic parameters.

APPENDIX I

STRESS-STRAIN PROPERTIES OF THE ELASTIC CYLINDER

RICHARD FITZHUGH *and* STANLEY I. RAPOPORT

From the Laboratory of Biophysics, National Institute of Neurological Diseases and Stroke, National Institutes of Health, and the Laboratory of Neurophysiology, National Institute of Mental Health, Bethesda, Maryland 20014

Fig. A 1 represents the stretched and nonstretched sarcolemmal tube. In Fig. A 1 *a*, the nonstretched tube is a cylinder with radius y_0 and length $2L_0$. When a horizontal force H dynes is applied to one end, the radius decreases from y_0 to y_m at $x = 0$ and the tube elongates (Fig. A 1 *b*). We write the relation between strain and stress as,

$$\text{strain vector} = \text{elasticity matrix} \times \text{stress vector}$$

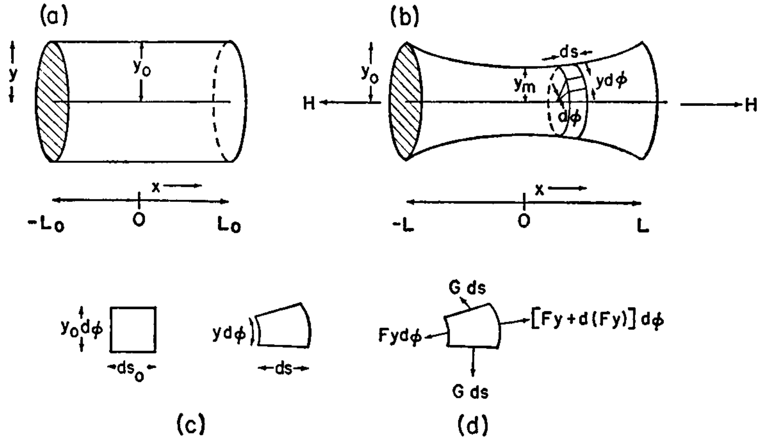

FIGURE A 1 Model of sarcolemmal tube in unstretched and stretched states. (*a*) Tube is cylindrical and unstretched, with radius y_0 and length $2L_0$. (*b*) Tube is stretched to $2L$, radius at end remains y_0 and decreases to y_m at $x = 0$. (*c*) A small region on the unstretched surface with sides $y_0\,d\phi$ and ds_0 is on left, which become $y d\phi$ and ds in stretched state. (*d*) The circumferential forces on the small stretched region are Gds and the longitudinal forces tangential to the surface in the x direction are $Fyd\phi$ and $[Fy + d(Fy)]\,d\phi$.

or

$$\begin{pmatrix} \dfrac{ds - ds_0}{ds_0} \\ \\ \dfrac{y - y_0}{y_0} \end{pmatrix} = \begin{pmatrix} c_{11} & c_{12} \\ \\ c_{21} & c_{22} \end{pmatrix} \times \begin{pmatrix} F \\ \\ G \end{pmatrix}, \qquad (\text{A } 1)$$

where $(ds - ds_0)/ds_0$ is the longitudinal extension and $(y - y_0)/y_0$ is the circumferential extension. In Fig. A 1, ds_0 and $y_0\,d\phi$ are unstretched differential lengths. In Fig. A 2, F is the longitudinal stress, G the circumferential stress (in units of dynes per centimeter), and they are shown vectorially in Fig. A 1 *d*. It is assumed that there is no pressure gradient across the membrane. The c_{ij} are constants in centimeters per dyne. They can be converted to elastic moduli (dynes per square centimeter) where δ is the sarcolemmal thickness, E_L and E_c are the longitudinal and circumferential moduli of elasticity, respectively, and σ_L and σ_c are the longitudinal and circumferential Poisson's ratios, respectively. c_{11} and c_{22} are defined by the following identities,

$$\begin{aligned} c_{11} &= 1/E_L\delta, \\ c_{22} &= 1/E_c\delta. \end{aligned} \qquad (\text{A } 2\,a)$$

The longitudinal Poisson's ratio is the ratio of circumferential contraction to longitudinal extension, and the circumferential ratio is the ratio of longitudinal extension to circum-

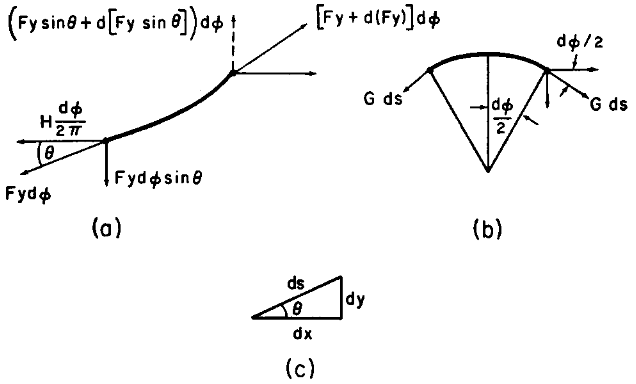

FIGURE A 2 Analysis of stress and curvature of model of sarcolemmal tube. (*a*) Part of longitudinal cross section of the tube. The longitudinal forces shown in Fig. A 1 *d* are given here, and the relation of *Fydϕ* to the horizontal and vertical forces is shown as a function of the angle *ϕ* between the horizontal force and the tangential surface in the *x* direction. (*b*) A circumferential cross section of the tube at its surface. The circumferential forces are *Gds*. *dϕ* is shown in Fig. A 1 *b*. (*c*) The right-triangular relation between *dx*, *dy*, and *ds*.

ferential contraction, defined by (Love, 1944, p. 106; Westergaard, 1952),

$$\frac{y - y_0}{y_0} = -\sigma_L \frac{ds - ds_0}{ds_0} \quad \text{for} \quad G = 0, F \neq 0,$$

$$\frac{ds - ds_0}{ds_0} = -\sigma_c \frac{y - y_0}{y_0} \quad \text{for} \quad F = 0, G \neq 0. \qquad (\text{A } 2\, b)$$

Insertion of Eqs. A 2 *a* and *b* into Eq. A 1 gives the following identities for the cross coefficients of Eq. A 1,

$$c_{12} = -\sigma_c / E_c \delta,$$

$$c_{21} = -\sigma_L / E_L \delta. \qquad (\text{A } 2\, c)$$

The c_{ij} satisfy the following equality (Love, 1944, p. 107):

$$c_{12} = c_{21}. \qquad (\text{A } 2\, d)$$

From Eq. A 1, we have

$$(y - y_0)/y_0 = c_{21}F + c_{22}G. \qquad (\text{A } 3)$$

The geometric relations of Fig. A 2 are used for the following derivations. In Fig. A 2 *a*, the horizontal stress for length $yd\phi$ is $(H/2\pi y)(yd\phi) = (H/2\pi)\, d\phi$. The longitudinal stress tangential to the surface and to the left is $Fyd\phi$. From the diagram

$$H\, d\phi/2\pi = Fyd\phi \cos\theta. \qquad (\text{A } 4)$$

Dividing by $d\phi$ and rearranging,

$$F = (H \sec \theta)/2\pi y. \tag{A 5}$$

The circumferential stress (Fig. A 2 *b*) is $G ds$, tangential to the surface. The net vertical force due to G is downward and is $2G ds \sin (d\phi/2)$, where the factor 2 means that stresses $G ds$ on the left and right have been taken into account. For $d\phi/2 \simeq 0$, $\sin (d\phi/2) \simeq d\phi/2$. The net upward force is obtained from Fig. A 1 *a* by subtraction, and is $d(Fy \sin \theta)\, d\phi$. For vertical force balance,

$$d(Fy \sin \theta) = G ds. \tag{A 6}$$

From Eq. A 5,

$$Fy \sin \theta = Hy'/2\pi, \tag{A 7}$$

since $y' = dy/dx = \tan \theta$. We note that

$$ds/dx = \sec \theta = (1 + y'^2)^{1/2}. \tag{A 8 a}$$

Substitute from Eqs. A 7 and A 8 *a* into Eq. A 6 to get the expression,

$$\frac{Hy''}{2\pi} = G(1 + y'^2)^{1/2}. \tag{A 8 b}$$

Eliminate F, G and $\sec \theta$ from Eqs. A 3, A 5, A 8 *a*, and *b* to obtain

$$yy'' - ay(y - y_0)(1 + y'^2)^{1/2} - b(1 + y'^2) = 0,$$

where

$$a = 2\pi/(c_{22} y_0 H) > 0,$$

$$b = -c_{21}/c_{22} > 0. \tag{A 9}$$

If y, y', and y'' are determined from the data as a function of x, a and b can be found graphically (see Methods).

Eq. A 1 gives,

$$(ds - ds_0)/ds_0 = c_{11}F + c_{12}G. \tag{A 10}$$

F and G are obtained from Eqs. A 5, A 8 *a*, and *b* and substituting into Eq. A 10 gives

$$(ds - ds_0)/ds_0 = c_{11}\frac{H(1 + y'^2)^{1/2}}{2\pi y} + c_{12}\frac{Hy''}{2\pi(1 + y'^2)^{1/2}}. \tag{A 11}$$

Rearrangement gives,

$$yy'' - \frac{2\pi(ds - ds_0)/ds_0}{c_{12}H}\, y(1 + y'^2)^{1/2} + \frac{c_{11}}{c_{12}}(1 + y'^2) = 0. \tag{A 12}$$

If $(ds - ds_0)/ds_0$ is known as a function of x, then this equation may be solved for c_{11} and

c_{12}. In practice, it was difficult to make this evaluation, so that Eq. A 12 could not be used. Instead, Eq. A 10 was solved for c_{11} by letting $F = H$, $G = 0$ (see text). Eq. A 9 was used to solve for c_{22} and c_{21}. c_{12} was calculated from the reciprocity relation of Eq. A 2 *d*.

APPENDIX II

LENGTH-TENSION PROPERTIES OF THE SARCOLEMMA IN THE INTACT FIBER

STANLEY I. RAPOPORT *and* RICHARD FITZHUGH

We assume that the sarcolemma has the same elastic coefficients in the intact fiber as in the tubular preparation and that it is linearly elastic. Its stress-strain relation is given by Eq. A 1, in which ds and ds_0 are replaced by S and S_0, because the sarcolemma is cylindrical (y the same for all x) and S is the length of a uniformly stretched section of fiber. Let $F = 0$ at $S = 3$ μm for the sarcolemma in the intact fiber; this approximately agrees with tubular observations. Let y_3 equal the sarcolemmal radius at $S = 3$ μm, and let y_0 be the radius of the sarcolemma alone (myoplasmic contents removed) for the condition $G = F = 0$. If G at 3 μm does not equal zero in the intact fiber, then by Eq. A 1,

$$\frac{y_3 - y_0}{y_0} = c_{22}G_3, \tag{A 13}$$

G_3 equals the circumferential stress at $S = S_3 = 3$ μm. Since the intact fiber stretches at constant volume it imposes a constraint on the sarcolemma (Huxley, 1953; Shear, 1969). The sarcolemmal radius y at a sarcomere length S is given in terms of S_3,

$$y = y_3\sqrt{S_3/S}. \tag{A 14}$$

We define extension from a rest length of 3 μm as e_3,

$$e_3 = (S - S_3)/S_3. \tag{A 15}$$

Inserting Eqs. A 14 and A 13 into A 1 and rearranging gives:

$$(\sqrt{S_3/S})(c_{22}G_3 + 1) - 1 = c_{21}F + c_{22}G. \tag{A 16}$$

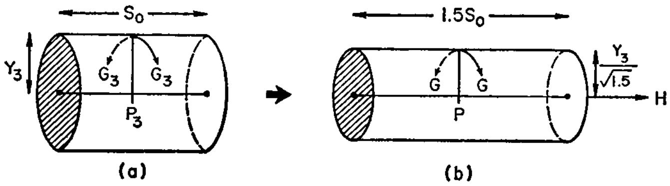

FIGURE A 3 Extension of fiber and sarcolemma at constant fiber volume. The fiber is stretched from a rest sarcomere length of S_0, at which $H = F = 0$ (*a*) to a length of $1.5S_0$ (*b*). Because fiber volume is constant with stretch, the radius of the stretched fiber is $y_3/\sqrt{1.5}$. The circumferential stress G_3 exerts a pressure $P_3 = G_3/y_3$ at rest and a pressure $P = G\sqrt{1.5}/y_3$ when the fiber is extended (see Eq. A 22). The horizontal force H operates to extend the fiber. For the purposes of calculation (see text), it is assumed that rest sarcomere length $S_0 = 3$ μm.

At $S = 3\ \mu$m and $F = 0$, from Eq. A 1 we have

$$(S_3 - S_0)/S_0 = c_{12}G_3. \tag{A 17}$$

Substituting Eq. A 17 into A 1, for any sarcomere length S we have,

$$\frac{S}{S_3}(c_{12}G_3 + 1) - 1 = c_{11}F + c_{12}G. \tag{A 18}$$

Eqs. A 16 and A 18 are simultaneous in F and G. Using the definitions of Eqs. A 2, A 14, and A 15, we have,

$$F = \frac{E_L\delta}{1 - \sigma_c\sigma_L}\left[(1 + e_3)\left(\frac{-\sigma_c G_3}{E_c\delta} + 1\right) - 1 + \sigma_c\left(\frac{\dfrac{G_3}{E_c\delta} + 1}{\sqrt{1 + e_3}} - 1\right)\right]\ \text{dyn/cm}, \tag{A 19}$$

$$G = \frac{E_c\delta}{1 - \sigma_c\sigma_L}\left[\sigma_L\left[(1 + e_3)\left(\frac{-\sigma_c G_3}{E_c\delta} + 1\right) - 1\right] + \frac{\dfrac{G_3}{E_c\delta} + 1}{\sqrt{1 + e_3}} - 1\right]\ \text{dyn/cm}. \tag{A 20}$$

These equations give F and G as a function of extension e_3 when the four constants of the elasticity matrix are known. One important restriction on Eq. A 20 is that G must be ≥ 0. For instance, if $\sigma_L = 0$ and $G_3 = 0$, the value of G will become negative, since y is reduced with extension by the constant volume constraint (Eq. A 14). The requirement that $G \geq 0$ implies that the sarcolemma will not contract below its rest circumference but will only fold up.

The horizontal force on the sarcolemma is $H = 2\pi yF$ (Eq. A 5). This definition and Eqs. A 14 and A 15 give:

$$H = \frac{2\pi y_3 F}{\sqrt{1 + e_3}}\ \text{dyn}. \tag{A 21}$$

The pressure P due to the circumferential tension of the sarcolemma is given by the Laplace equation for a cylinder (Condon and Odishaw, 1958; Rapoport, 1972),

$$P = G/y\ \text{dyn/cm}^2, \tag{A 22}$$

where y is the cylindrical radius. Eqs. A 20, A 14, and A 15 give P as a function of extension:

$$P = \frac{\sqrt{1 + e_3}\,G}{y_3}\ \text{dyn/cm}^2. \tag{A 23}$$

Since $G_3 \geq 0$, $P \geq 0$.

We thank Mr. Rufus Gunn, Mrs. Jeanette Bidinger, and Mr. Alvin Ziminsky for valuable technical assistance, and Dr. Harold Lecar for valuable help with the theory of elasticity.

Mr. James Bryan, Section of Technical Development, National Institute of Mental Health, designed and provided the tension measuring apparatus.

Mr. Richard Shrager, Division of Computer Research and Technology, aided with computer analysis.

Mrs. Karen Pettigrew, Section of Theoretical Statistics and Mathematics, National Institute of Mental Health, performed statistical analyses and made helpful suggestions as to their presentation.

Received for publication 20 December 1971 and in revised form 4 August 1972.

REFERENCES

ABOOD, L. G., K. KURAHASI, E. BRUNNGRABER, and K. KOKETSU. 1966. *Biochim. Biophys. Acta.* **112**:330.

BLINKS, J. R. 1965. *J. Physiol. (Lond.).* **177**:42.

BOYDE, A., and J. C. P. WILLIAMS. 1968. *J. Physiol. (Lond.).* **197**:10P.

BROWNLEE, K. A. 1960. Statistical Theory and Methodology in Science and Engineering. John Wiley and Sons, Inc., New York.

BUCHTHAL, R., E. KAISER, and P. ROSENFALCK. 1951. *Dan. Biol. Medd.* **21**: no. 7, p. 1.

CARTON, R. W., J. DAINAUSKAS, and J. W. CLARK. 1962. *J. Appl. Physiol.* **17**:547.

CASELLA, E. 1950. *Acta Physiol. Scand.* **21**:380.

CONDON, E. U. and H. ODISHAW. 1958. Handbook of Physics. McGraw-Hill Book Company, New York.

DELÈZE, J. B. 1961. *J. Physiol. (Lond.).* **158**:154.

EDMAN, K. A. P. 1966. *J. Physiol. (Lond.).* **183**:407.

FIELDS, R. W. 1970. *Biophys. J.* **10**:462.

FIELDS, R. W., and J. J. FABER. 1970. *Can. J. Physiol. Pharmacol.* **48**:394.

HARKNESS, R. D. 1968. *In* Treatise on Collagen. B. S. Gould, editor. Academic Press, Inc., New York. 2(A):247.

HILL, D. K. 1968. *J. Physiol. (Lond.).* **199**:637.

HÖBER, R. 1945. Physical Chemistry of Cells and Tissues. Blakiston Division, McGraw-Hill Book Company, New York.

HUXLEY, A. F. 1964. *Annu. Rev. Physiol.* **26**:131.

HUXLEY, H. E. 1953. *Proc. R. Soc. Ser. B. Biol. Sci.* **141**:59.

JONES, W. M., and R. BARER. 1948. *Nature (Lond.).* **161**:1012.

KOKETSU, K., R. KITAMURA, and R. TANAKA. 1964. *Am. J. Physiol.* **207**:509.

LOVE, A. E. H. 1944. A Treatise on the Mathematical Theory of Elasticity. Dover Publications, Inc., New York. 4th edition.

MAURO, A., and W. R. ADAMS. 1961. *J. Biophys. Biochem. Cytol.* **10**(4, Pt. 2):177.

MCCOLLESTER, D. L. 1962. *Biochim. Biophys. Acta.* **57**:427.

MULLING, G. L., and W. GUNTHEROTH. 1965. *Nature (Lond.).* **206**:592.

PODOLSKY, R. J. 1964. *J. Physiol. (Lond.).* **170**:110.

RAMSEY, R. W., and S. F. STREET. 1940. *J. Cell. Comp. Physiol.* **15**:11.

RAPOPORT, S. I. 1970. *Biophys. Soc. Annu. Meet. Abstr.* **10**:217a.

RAPOPORT, S. I. 1972. *J. Gen. Physiol.* **59**:559.

RAPOPORT, S. I., and R. FITZHUGH. 1971. *Biophys. Soc. Annu. Meet. Abstr.* **11**:200a.

SHEAR, D. B. 1969. *Physiol. Chem. Phys.* **1**:495.

STEN-KNUDSEN, O. 1953. *Acta Physiol. Scand. Suppl.* 104. **28**:1.

STREET, S. F., and R. W. RAMSEY. 1965. *Science (Wash. D.C.).* **149**:1379.

WANG, H. 1956. *Exp. Cell Res.* **11**:452.

WESTERGAARD, H. M. 1952. Theory of Elasticity and Plasticity. Harvard University Press, Cambridge, Mass.