ing along the opposite flank. The growth rate on the compressed side would increase, while that on the stretched side would decrease, resulting in a bend in the opposite direction, as has been observed.

Another important property of this growth response is that of adaptation; though the growth rate deviates from normal for 5 minutes after the change in load, it subsequently returns to normal even though the load remains constant. To test whether this adaptation is complete, we compared growth rates before and after the application of a load, using the following procedure. Before the hook was attached, the growth rate was measured for 10 minutes. Then the load was applied for 60 minutes, removed for 60 minutes, and finally reapplied for 60 minutes. Average growth rates were obtained for the 10-minute control period and the last 10 minutes of each of the 60-minute periods. We used only the last 10 minutes to reduce the contribution of the growth response at the beginning of each 60-minute period. Having combined the data for loads of 5, 8, 10, and 20 mg, we obtained the following average growth rates (percentage of control rate): load applied first time, $74 \pm 5$ percent; load removed, $80 \pm 7$ percent; load applied second time, $79 \pm 5$ percent (standard error of the mean given). Thus, no significant effect of load on growth rate was noted 50 minutes after the load was applied. This is consistent with the observation that the cell is mostly elastic, since one might expect a cell with a large plastic component to undergo continuous slow deformation when loaded and hence show a greater growth rate with the load applied than with it removed.

The ability of the sporangiophore to adapt completely to the applied load suggests that the deformation itself is the stimulus or is closely related to it. Since the sporangiophore is mostly elastic, most of the deformation occurs within a few seconds of the load's being changed. Since little deformation occurs thereafter, there will be no further stimulus acting to affect the growth rate as long as the load is constant.

We have also examined some of the factors affecting the magnitude of this growth response. We define the magnitude of the response as the ratio of the total growth during the 5-minute interval following the stimulus to the total growth during the 5-minute interval preceding the stimulus. Thus a response to an added load would be a number less than 1 (typically 0.6 to 0.8), and a response to a reduced load would be a number greater than 1 (typically 1.3 to 1.6).

Two factors affecting the response size are the length of the interval between stimuli and light. If the interval is shortened to 20 minutes, the responses are of smaller magnitude than those when there is an interval of 60 minutes between stimuli. Illumination with blue light causes a definite but highly variable reduction of the response's magnitude; this inhibition is largely removed by 60 minutes of darkness.

The weight of the load also affects the size of the response. In Fig. 3 are presented the mean response magnitudes at stimulus loads varying from 0.3 to 8 mg. Although variability is high, there is a rather sharp threshold at a load between 0.5 to 1.0 mg; at 0.5 mg and below there is no response, and at 1.0 mg and above the response is relatively large. The lack of response at 0.5 mg is not due to a lack of deformation, since at this load the total deformation of the cell is $0.13 \pm 0.02$ mm.

DAVID S. DENNISON
CAROLYN C. ROTH
*Department of Biological Sciences, Dartmouth College, Hanover, New Hampshire*

**References and Note**

1. E. S. Castle, *Science* **154**, 1416 (1966).
2. D. S. Dennison, *J. Gen. Physiol.* **45**, 23 (1961).
3. E. S. Castle, *ibid.* **42**, 697 (1959); R. Cohen and M. Delbrück, *J. Cell. Comp. Physiol.* **52**, 361 (1958).
4. Supported by an NSF grant.

27 March 1967

## Force, Shortening, and Work in Muscular Contraction: Relative Contributions to Overall Energy Utilization

Abstract. *The amounts of energy used during muscular contraction under different loads have been compared by measurement of the total amount of disturbance of the concentration of reduced nicotinamide-adenine dinucleotide during the period of oxidative recovery. The results are in quantitative agreement with the concept that three parameters determine the energy utilization: namely the time-integral of the force development, the time-integral of the shortening process, and the mechanical work.*

When a muscle shortens against a lever bearing a load, the amount of energy expended in the process is highly dependent on the load that is moved. This was first pointed out by Fenn in his studies on the heat developed during and after twitches and short tetanuses (*1*). Until relatively recently, three processes were thought to account for this so-called Fenn effect: activation (and its maintenance), shortening, and the performance of mechanical work (*2*). We have found that two parameters, the time-integrals of the developed force ($\int P_o$) and of shortening ($\int S_o$) suffice for the quantitative description of energy used in isometric and minimally loaded isotonic contractions, respectively. In addition these parameters, together with the mechanical work performed, describe the Fenn effect quantitatively. This finding is at variance with the concept that the activation energy is a major contribution, independent of load, to the total energy utilization (*2–5*). It is also in disagreement with the concept that the energy requirement for shortening is linearly related, or very nearly so, to the distance of shortening (*2, 5*). But in contrast to other recent results (*3, 6*), we find there is a major contribution by the shortening process.

Energy utilization in various types of contractions was compared on the basis of changes in the amount of mitochondrial reduced nicotineamide-adenine dinucleotide (NAD) within the muscle at rest. During oxidative metabolism the average redox steady-state of several members of the respiratory chain varies depending on the rate of electron transport (*7*). A close correlation exists between the amount of reduced NAD at equilibrium, the rate of oxygen uptake, and the rate of oxidative phosphorylation. Thus, it is possible to monitor the metabolism during recovery in an intact muscle after contractile activity if the amount of reduced mitochondrial NAD can be measured. When the reduced form of the coenzyme (NADH) is excited with light with a wavelength of 366 m$\mu$, the main part of the fluorescent light is emitted in the region of 400 to 500 m$\mu$, with a broad maximum around 460 m$\mu$. The intensity of the

emitted light is related to the total amount of NADH and, therefore, to the reduced fraction of the total concentration of this coenzyme (*8*).

Excised sartorius muscles of tropical toads (*Bufo marinus*) and bullfrogs (*Rana catesbiana*) were prepared and mounted in a fluorometer constructed for the purpose (*9*). Rectangular current pulses delivered between two stainless steel bars parallel to the muscle provided massive, transverse stimulation. The mechanical performance was registered with an after-loaded lever fitted with strain gages for the recording of force and displacement. The stretching of the series-elastic component was estimated by the quick-release method of Jewell and Wilkie, and results from both preparations were closely comparable to their published values for frog sartoriuses (*10*). The initial length of the muscle was 100 to 105 percent of its length in the body.

A temporary and partial oxidation of NADH is noted when electron transport is accelerated during the period of increased oxidative metabolism that constitutes the recovery from one twitch. At 12°C, the entire cycle takes about 10 minutes. Extensive experimentation has shown that fluorescence from mitochondrial NADH is greatly enhanced, so that the contribution from other NADH fractions and from reduced NAD phosphate can be neglected (*11*). The area under the fluorescence cycles ($\int\Delta Fl$), measured from the moment the trace departs from the baseline to its return, varies directly with the amount of recovery metabolism. We proved this by comparing the recovery cycles in response to series of different numbers of twitches in rapid succession. Such experimentation at 12°C showed that the $\int\Delta Fl$ is directly related to the number of contractions up to five twitches at one per second (*11*).

We studied the relative energy utilization for two different types of contractions, isometric and isotonic. In the first case, the muscle exerts force against an immovable attachment point, and tension rises and falls. In the second condition, the muscle lifts a load; that is, tension is constant while the muscle shortens and lengthens again. We compared the relative amounts of energy used in contractions resulting from different numbers of stimuli delivered at a tetanic rate.

When separate trains of increasing numbers of stimuli at constant frequency are delivered to a muscle under isometric conditions, twitch-like contractions result; these gradually become larger until flat-topped, tetanic contractions are achieved. The isometric force ($P_o$) and the area under the myogram ($\int P_o$) are shown in the top of Fig. 1 as a function of the number of shocks in each train. Maximum tetanic tension was about twice that of a single twitch and was approached in response to approximately eight stimuli. Save for a small region near the origin, the area of the myogram ($\int P_o$) is linearly related to the number of stimuli for up to 3 seconds of stimulation—the maximum interval tested. The region of curvature for the very small numbers of stimuli is rather variable but has been noticed in all experiments. Frequently, only the first two points are nonlinear. The disappearance of the curvature does not coincide with the attainment of maximum tetanic tension.

In the bottom graph of Fig. 1 is shown the correlation between the $\int P_o$ and the relative amount of recovery metabolism, measured as the $\int\Delta Fl$. We checked the linear relation of the $\int\Delta Fl$ to the amount of energy expended by measuring the recovery cycles induced by one, two, or three twitches in rapid succession. These points are identified by the open circles.

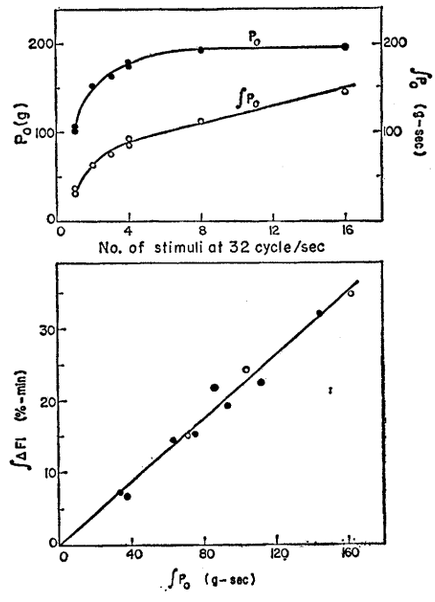


Fig. 1. The effect of various numbers of stimuli at a tetanic rate on mechanical and energetic parameters of isometric contractions. (Top) The isometric force ($P_o$, filled circles) developed by the muscle, and its time-integral ($\int P_o$, open circles) as a function of the number of stimuli. (Bottom) The time-integral of the fluorescence cycle ($\int Fl$) as a function of the $\int P_o$. Filled circles: the same contractions as shown in the top graph. Open circles: cumulative effect of two, three, and four single twitches in rapid succession (1 cycle/sec).

The relation between the two variables plotted in the lower part of Fig. 1 adheres to a straight line through the origin. In other experiments this held true regardless of the frequency of stimulation, the animal used (frog or toad), or the temperature. Since the relation between $\int P_o$ and the number of stimuli is not linear, the aforementioned result is neither trivial nor redundant. It is also important to note that the $\int P_o$ takes the entire contraction cycle into account, the contraction and relaxation phases contributing in proportion to their contribution to the total $\int P_o$.

The use of the $\int P_o$ eliminates activation as a significant, fixed contribution to the overall energy utilization. Since the "activation heat" supposedly reflects a separate, initial process preparatory to the mechanical contraction, the straight line connecting the experimental points should transect the ordinate at a value greater than zero. This value should be that of the fixed amount of energy that is postulated as required for the achievement of the "active state" (*2*, *5*). Myothermic estimates of the activation heat range from eight- to nine-tenths of the total heat of a single isometric twitch (*3*), to 33 to 50 percent (*2*, *5*) and to 40 to 45 percent (*4*). Interpretation of the results of biochemical analysis of splitting of high-energy phosphate by the frog sartorius has yielded another range of values: 80 to 90 percent (*3*) and 60 percent (*12*). In the frog rectus abdominis muscle no evidence was found for any breakdown of high-energy phosphate by activation (*13*), whereas it was also shown that in the frog sartorius some delayed hydrolysis of adenosine triphosphate (ATP) occurred that was not related to the development of tension (*6*, Fig. 2). This amounted to about 30 percent of that hydrolyzed in a 1.5-second isometric tetanus. Interestingly enough, under slightly different conditions all hydrolysis of ATP appeared to be related to the duration of the tetanic stimulation: no evidence for a high rate of ATP hydrolysis was apparent early in the contraction (*6*, Fig. 1). The distribution of the data in experiments such as shown in Fig. 1 might possibly allow a variation in the intercept with the ordinate of $\pm$ 10 percent of the twitch value. This is due to the experimental error in the data, and it may conceal a contribution from an activation process. With this limitation in mind, we can

make the generalization that adoption of the $\int P_o$ as the energy parameter of muscle contraction eliminates activation as a process contributing separately to the overall energy utilization. Since a sizable burst of heat preceding the actual contraction (*2*) can be measured, the possibility does remain that this "activation heat" may be compensated by a reaction that later in the cycle absorbs heat, presumably one in the relaxation phase. However, no evidence for such a reaction has been found as yet in myothermic studies.

A typical result of similar experiments under isotonic conditions is seen in Fig. 2. The top graph shows the total distance shortened ($S_o$) and the area under the displacement curve ($\int S_o$) as a function of the number of stimuli. Both curves are fairly representative of this type of experiment. There is some variation, however, in the degree of concavity of the lower portion of the $\int S_o$ curve. In a small number of preparations the entire range adhered to a straight line, but in none of the experiments was the curve convex upward. The lower graph of Fig. 2 shows the relative energy utilization ($\int \Delta Fl$)

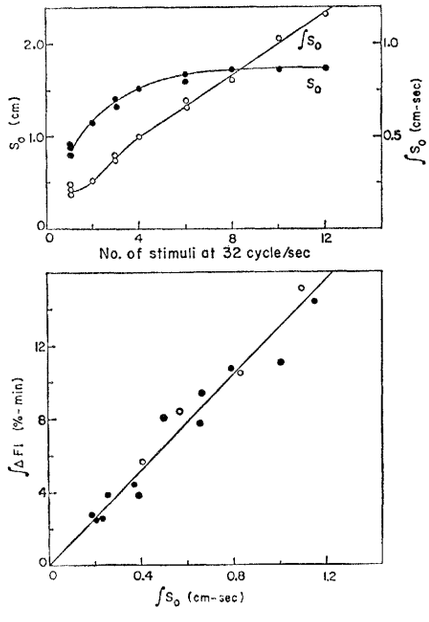


Fig. 2. The effect of various numbers of stimuli at a tetanic rate on the mechanical and energetic parameters of minimally loaded isotonic contractions. (Top) The shortening distance ($S_o$, filled circles) and its time-integral ($\int S$, open circles) as a function of the number of stimuli. (Bottom) The $\int \Delta Fl$ as a function of the $\int S_o$. Filled circles: the same contractions as depicted in the top graph. Open circles: cumulative effect of two, three, four, and six single twitches in rapid succession.

as a function of the time-integral of the shortening cycle, $\int S_o$. The determinations for short tetanuses were randomly interspersed with a few series of two to four single twitches in rapid sequence. The results of the latter are represented by the open circles. It is clear that a linear relation exists for both multiple twitches and short tetanuses. The scatter of the experimental points is usually greater than that in isometric experiments, a fact that may be partially caused by the smaller value of $\int \Delta Fl$ for these isotonic conditions. Nevertheless, the direct correlation between the two parameters appears to be unmistakable. As in the previous experiment, the straight line fitting the points goes through the origin; that is, no initial contribution of an activation process is discernible. Within the limitations set by the experimental error, the $\int S_o$ suffices as a direct, linear index to the total energy turnover in a minimally loaded isotonic contraction.

Our data and conclusions are limited to the transition region between twitch and tetanus. During tetanuses that are maintained for a long time other factors come into play and decrease the rate of energy turnover (*14*). On a plot of $\int \Delta Fl$ as a function of the $\int P_o$, the data suddenly deviate from the linear relation through the origin. The new relation is still linear with the $\int P_o$, but it has a lesser slope. Extrapolation to the zero ordinate yields, therefore, an intercept with a sizable value of $\int \Delta Fl$. This later phase has been explored thoroughly over the years. Aubert's monograph (*15*) contains perhaps the most extensive myothermal records, and these appear to agree fully with our interpretation based on the $\int P_o$. Maréchal and Mommaerts, and Sanberg and Carlson have presented detailed studies of phosphocreatine utilization in tetanic contractions (*16*). Their data show the same general result. In long tetanuses (> 1 second) it was demonstrated that a linear correlation exists between breakdown of high-energy phosphate and the $\int P_o$ during the maintenance of stable tension, but an extra amount of breakdown was found to occur in the rising phase. The latter is the region we have investigated. Together with these published studies, our data show that the $\int P_o$ is an essential parameter having two distinct values in the range between the single twitch and a long-lasting tetanus.

In Fig. 3, the variation of the total energy utilization as measured from

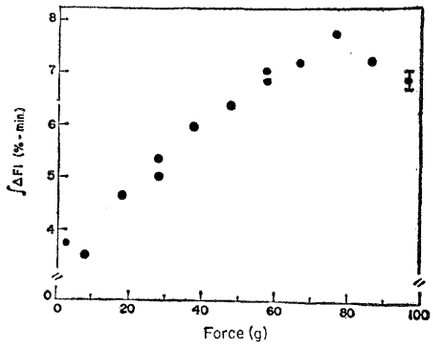


Fig. 3. Relative energy utilization in the single twitch as a function of the force developed during the contraction of toad sartorius at 11°C with an initial load of 2.0*g*; all other loads are after-loaded. Four cycles were recorded for the isometric twitch; the bar shows the spread of the data. Two cycles were recorded for the minimally loaded isotonic condition; the $\int \Delta Fl$ was virtually identical for these two cycles.

fluorometric cycles is shown as a function of the load lifted by the muscle. The curve is typical for single twitches of the toad sartorius at about 12°C. It is closely similar to a number of published records of metabolic and heat measurements under similar conditions (*11, 12*) but disagrees with others (*1, 2, 5, 12*). In the latter, the left side of the curve is shifted upward, the energy utilization at minimal loads being about equal to that at the highest load (that is, the isometric condition). The difference appears to be related to the use of different muscles and experimental temperatures.

The three parameters depicted in Fig. 4 are the total work and the time-integrals of shortenings ($\int S$) and force ($\int P$) registered in the same experiment as shown in Fig. 3. For each twitch the outputs of the force and displacement transducers were recorded separately. At intermediate loads the force in the muscle rises until a tension equal to the load has been generated, whereupon shortening commences. The tension remains constant until the load returns to its original, supported position; it subsequently disappears. The areas under the two parts of the myogram were measured with a polar planimeter.

The curve for the total work is obtained by adding the external and internal work. The former consists not only of the product of the load and the distance through which it is lifted, but also of the work performed in bending the lever (the stretching of the chain and of other mechanical compli-

ances is negligible in this apparatus). The lever has a linear stress-strain relation, and so the work performed on it by the muscle is one half the product of total force and displacement of the lever tip. The stretching of the series-elastic component of the muscle, however, is not linear: relatively more shortening occurs at low tension than at high. From this stress-strain relation it was calculated that the internal work amounts to approximately one-fourth of the product of the final force and the stretching of the series-elastic component. We obtained the total work curve ($W_T$) of Fig. 4 by adding the two types of external work and the internal work obtained for each load and plotting this as a function of load.

It is our hypothesis that the three parameters $\int S$, $\int P$, and $W_T$ account for the total energy expended in the contraction. Since these parameters, as well as the $\int \Delta Fl$, are expressed in units that cannot be interconverted at the present time, it is necessary to find a method of scaling them. As a first step, we normalized the $\int \Delta Fl$, designating the averaged isometric value as 100. These measurements of total energy utilization are shown as the solid circles in the top curve of Fig. 5.

At three points in the $P$ scale (2.0, 48.3, and 97.5$g$) the numerical value of $\int \Delta Fl$ was expressed as the sum of the parameters:

$$\int_{t=0}^{t=\infty} \Delta Fl_{2.0}\, dt = \int_{t=0}^{t=\infty} S_{2.0}\, dt + W_{T2.0} = 56 \qquad \left(\int_{t=0}^{t=\infty} P_{2.0}\, dt \cong 0\right)$$

$$\int_{t=0}^{t=\infty} \Delta Fl_{48.3}\, dt = \int_{t=0}^{t=\infty} S_{48.3}\, dt + W_{T48.3} + \int_{t=0}^{t=\infty} P_{48.3}\, dt = 93$$

$$\int_{t=0}^{t=\infty} \Delta Fl_{97.5}\, dt = W_{T97.5} + \int_{t=0}^{t=\infty} P_{97.5}\, dt = 100 \qquad \left(\int_{t=0}^{t=\infty} S_{97.5} \cong 0\right)$$

From these three equations the scaling factors for $\int S$, $\int P$, and $W_T$ can readily be calculated. When these factors are applied to each value of the points of Fig. 4, they are scaled as shown by the hollow squares in Fig. 5. The line has been drawn from the addition of the three curves fitting the experimental points in the lower part of the figure. The closeness of fit of the experimental $\int \Delta Fl$ points (filled circles) to the calculated line indicates that the hypothesis is well borne out.

We have performed similar experiments with the bullfrog sartorius at 15°C using a four-stimulus tetanus. In this case, the curve of energy utilization as a function of the load (Fig. 3) shows a more pronounced maximum (about 1.4 times the isometric value), and it occurs at a lower load (approximately 0.6 $P_o$). Thus the curve resembles more closely that of the frog at low temperatures. Using the same scaling techniques as before, we found that the $\int P$, $\int S$, and $W_T$ curves fit satisfactorily to this relation. A larger relative contribution of the work curve, because of greater shortening (especially at low loads), underlies both the more pronounced maximum in the total energy curve and its shift toward a lower load as compared to the data on the single twitch of the toad sartorius (*14*).

The use of the $\int P$ as a parameter of energy utilization is reminiscent of the tension-time index of cardiac physiology (*17*). In heart tissue, the tension-time index apparently suffices as the sole determinant of energy utilization of cardiac muscle, as judged from the studies of $O_2$ consumption. In skeletal muscle, however, the total energy used in an isotonic contraction is a complex function of the work and the time-integrals of shortening and tension production. In 1921, Hartree and Hill applied the notion of a tension-time index to isometric contractions of the frog's sartorius muscle and correlated it with heat development (*18*). The relations appear similar in general, but the transition region between twitch and tetanus was not studied. Since then, the tension-time parameter has been neglected almost completely in studies of skeletal muscle.

Our results show that the time-integrals of shortening and of force together with the total work can account for the total energy expended in single twitches and in short tetanuses. Several important points emerge from this novel account of the energy relations.

The concept of a sizable, fixed amount of energy expended in "activation" of the muscle could not be verified by our experiments. If such a term accounted for 80 to 90 percent of the energy utilized in the isometric twitch (*3*), a straight line would need to be drawn across Fig. 5 at that level; no significant contribution from $\int P$ could be allowed, and it would be entirely impossible to fit the total energy curve. The same holds to a slightly smaller

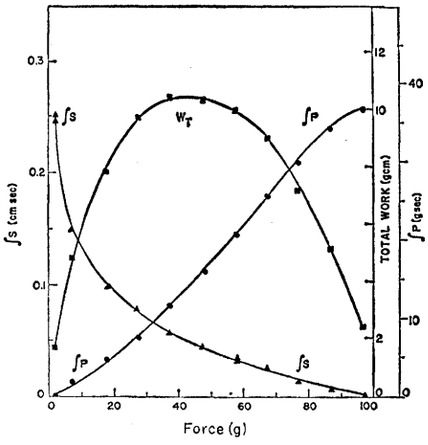


Fig. 4. Total work ($W_T$) and the time-integrals of shortening ($\int S$) and of force ($\int P$) as a function of the force developed during the contraction. Data from the same experiment as Fig. 3. The measurements of most duplicate points were virtually identical and could not be differentiated in the drawing.

degree for other estimates of the activation energy: 60 percent (*12*), 40 to 45 percent (*4*), 33 to 50 percent (*2*, *5*), and 30 percent (*6*). Only if the activation energy were 10 percent or less, would a reasonable fit be maintained. Since there is considerable resemblance in the shape of the shortening ($S$) and the $\int S$ curves when plotted as a function of the force, substitution of $\int S$ for the more conventional $S$ relation is not the basis for the inadequacy of the concept of a large, fixed "activation" heat in the approach presented above.

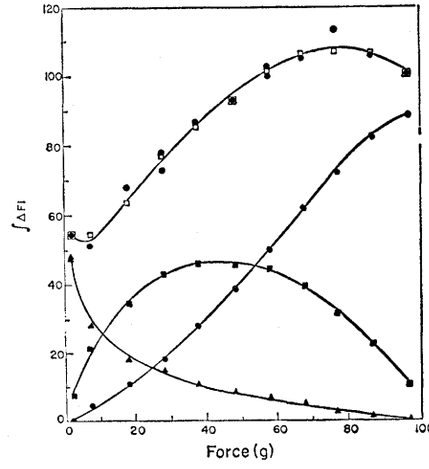


Fig. 5. Comparison of the energy utilization (filled circles in upper curve) with the summed $W_T$, $\int S$, and $\int P$ parameters after scaling (open squares). Same experiment as Figs. 3 and 4. Filled squares, $W_T$; filled triangles, $\int S$; filled circles in lower curve, $\int P$.

The substitution of $\int S$ for the linear shortening term ($S$) as a parameter of energy utilization is not as self-evident as the introduction of the $\int P$. The main difference between the two curves is a greater degree of curvature in the case of $\int S$. If shortening ($S$) is used in Fig. 5 instead of $\int S$, the small trough at $7g$ is eliminated in the top curve, and a continuously convex relation results.

Our evidence is in agreement with the notion that muscle contraction can be regarded as a series of sequential chemical reactions (*14*). Force arises and is maintained as one step in this sequence. Shortening may constitute another such step occurring either in series or in parallel to the reaction producing the force. In the case of an isometric contraction, in which shortening is practically zero and the internal work is no more than 10 percent of the total energy utilization, force production is virtually the sole energy-consuming activity. Thus, tension may be looked upon as the index to the intensity and the time-course of one step in the sequence of reactions initiated by the action potential. For unbranching systems of sequential, chemical reactions it can be shown that the time-integral of the increase and decrease in concentration of one of the intermediates is linearly related to the total amount of the final product resulting from the overall reaction (*14*). In the case of muscular contraction, it is proposed that shortening and production of force can be treated as mechanical manifestations of the time-course of the concentration of two sequential intermediates or two reaction pathways originating from a common intermediate.

FRANS F. JÖBSIS
JAMES C. DUFFIELD
*Department of Physiology and Pharmacology, Duke University Medical School, Durham, North Carolina*

### References and Notes

1. W. O. Fenn, *J. Physiol. (London)* **58**, 175 (1923).
2. A. V. Hill, *Proc. Roy. Soc. London Ser. B* **126**, 136 (1938); *ibid.* **136**, 195 (1949); *ibid.* **137**, 330 (1950).
3. F. D. Carlson, D. J. Hardy, D. R. Wilkie, *J. Gen. Physiol.* **46**, 851 (1963).
4. C. L. Gibbs, N. V. Ricchiutti, W. F. H. M. Mommaerts, *ibid.* **49**, 517 (1966).
5. A. V. Hill, *Proc. Roy. Soc. London Ser. B* **159**, 297 (1964).
6. A. A. Infante, D. Klaupiks, R. E. Davies, *Biochim. Biophys. Acta* **88**, 215 (1964).
7. B. Chance and G. R. Williams, *J. Biol. Chem.* **217**, 383 (1955).
8. F. F. Jöbsis, in *Handbook of Physiology* W. O. Fenn and H. Rahn, Eds. (American Physiological Society, Washington, D.C., 1964), sec. 3, vol. 1, p. 63.
9. F. Jöbsis, V. Legallais, M. O'Connor, *Inst. Electron. Elec. Eng. Trans. Bio-Med. Eng.* **13**, 93 (1966).
10. B. R. Jewell and D. R. Wilkie, *J. Physiol. (London)* **143**, 515 (1958).
11. F. F. Jöbsis and J. C. Duffield, *J. Gen. Physiol.* **50**, 1009 (1967).
12. S. Distèche, in *The Biochemistry of Muscular Contraction*, J. Gergely, Ed. (Little, Brown, Boston, 1964), p. 458.
13. D. F. Cain, A. A. Infante, R. E. Davies, *Nature* **196**, 214 (1962).
14. F. F. Jöbsis, in *Proceedings of an International Symposium on Muscle*, J. Tigyi, Ed. (Hungarian Academy of Science, Budapest, 1966), in press.
15. X. Aubert, *Le Couplage Énergétique de la Contraction Musculaire* (Arscia, Bruxelles, 1956).
16. G. Maréchal and W. F. H. M. Mommaerts, *Biochim. Biophys. Acta* **70**, 53 (1963); J. A. Sanberg and F. D. Carlson, *Biochem. Z.* **345**, 212 (1966).
17. G. Weiss, *J. Physiol. Pathol. Gén.* **15**, 999 (1913); S. J. Sarnoff, E. Braunwald, G. H. Welch, Jr., R. B. Case, W. N. Stainsby, R. Macrusz, *Amer. J. Physiol.* **192**, 148 (1958).
18. W. Hartree and A. V. Hill, *J. Physiol. (London)* **55**, 133 (1921).
19. Supported by grants from the Muscular Dystrophy Association of America, Inc.

7 September 1966

## Oxygen Tension Changes Evoked in the Brain by Visual Stimulation

Abstract. *Localized changes in oxygen tension were recorded with platinum cathodes placed in the lateral geniculate nucleus in both anesthetized and awake cats. The amplitude of the responses increased with increasing stimulus intensity, but decreased with increasing flash rate. Both increases and decreases in cathode current were produced by steady illumination. The characteristics of the responses suggest that the responses reflect localized variations in blood flow, produced in turn by changes evoked in the tonic neural activity of the lateral geniculate nucleus.*

Localized temperature changes can be evoked in the cat's brain by natural stimulation (*1, 2*). Light flashes evoke thermal changes in the lateral geniculate nucleus but not in the inferior colliculus, while auditory stimuli evoke thermal changes in the colliculus but not in the geniculate nucleus (*1*). Somatic stimulation evokes temperature changes localized in the ventrobasal nucleus in the thalamus (*2*). Elevations in the temperature of the entire brain during paradoxical sleep and arousal have also been reported (*3*). Such temperature changes may result from variations in cerebral blood flow and metabolic heat output, and they are probably accompanied by variations in the oxygen tension of the tissue. The oxygen tension of tissue can be indirectly assessed with bare oxygen cathodes consisting of platinum wire negatively biased with respect to a nonpolarizable reference electrode. Such cathodes have been used to determine variations in the concentration of oxygen in brain tissue during hypoxia and hyperoxia, stimulation of the hypothalamus, and during avoidance conditioning in unrestrained cats (*4*). We here report changes in oxygen tension in the lateral geniculate nucleus (LGN) of cats. The changes were evoked by visual stimulation and were recorded by platinum oxygen cathodes.

Twenty-one cats were anesthetized with moderate doses (30 mg per kilogram of body weight) of sodium pentobarbital injected intraperitoneally; supplementary doses were given when necessary during the experiments. The cats were held in a stereotaxic instrument, and the cathodes were inserted through a hole, 1 cm in diameter, trephined through the skull. Recordings were also taken from two unanesthetized cats with cathodes permanently implanted in the LGN. The unanesthetized cats were allowed 1 week to recover from surgery before they were tested. Their line of vision was directed toward the stimulus source by means of a restraining box. In all animals, the pupils were dilated with Cyclogyl during testing, and the cathode placements were verified by standard histological techniques.

The oxygen cathodes were made of platinum–iridium wire (10 percent Ir), 0.5 mm in diameter, and insulated, except at the tip, with Insul-X. The reference electrodes usually consisted of large-area silver electrodes inserted under the temporal muscles, and the cathode bias ranged from 0.5 to 0.8 volt. The current through the electrodes was measured with Grass (7P1A) d-c amplifiers. Light flashes were produced by a Grass photostimulator (PS2D), with flash rates in the range of 2 to 50 per second, flash duration of 10 $\mu$sec, and estimated intensities at the cat's eyes of about $2.5 \times 10^5$ lux and $2 \times 10^6$ lux. Background intensity was about 200 lux.

Consistent increases in the cathode current were evoked in the LGN by flashing light in 11 of the anesthetized cats and in both unanesthetized animals. The responses were approximately 5 percent above the base current, with latencies of 5 to 10 seconds (Fig. 1). The cathode current increases were usually maintained throughout the pe-