AN ENERGETIC MODEL OF MUSCLE CONTRACTION

J. B. CHAPMAN *and* C. L. GIBBS

From the Department of Physiology, Monash University, Clayton, Victoria, Australia

ABSTRACT Initial energy utilization in the twitch is visualized as the result of the activity of two distinct processes. The first is the calcium-pumping activity of the sarcoplasmic reticulum, which has a constant energy requirement under normal conditions. The second is the chemomechanical transduction process consisting of a variable number of quantal contractile events, each with a fixed enthalpy equal to the molecular enthalpy of adenosine triphosphate (ATP) hydrolysis in vivo. This enthalpy appears either as heat or as contractile element work. Total enthalpy varies according to the number of quantal contractile events that occur in the twitch cycle. The basis of the variation is suggested to be velocity-dependent activity of the actomyosin ATPase, allowing more quantal events to occur in a contraction cycle when shortening occurs. The classical designation "activation heat" is held to be appropriate for the first process. The partition of the enthalpy of the second process that is currently in vogue is held to be misleading and a new formulation is suggested in which the properties of the quantal contractile event are reflected in general terms. The formulation of the proposed transduction model represents a conceptual return to the viscoelastic theory, but at a quantal level. The model can explain the results of the preceding paper and is adaptable to different muscles without having to postulate fundamental differences in energy utilization.

INTRODUCTION

In the preceding paper (Chapman and Gibbs, 1972) results were presented which were not readily explainable in terms of current approaches to muscle energetics. In this paper it is shown that the energy utilization of a muscle twitch can be accounted for in terms of a fixed component identified as the enthalpy of ATP hydrolysis associated with the sarcoreticular calcium pump together with a variable component identified as the enthalpy of ATP hydrolysis associated with the contractile proteins. The basis of variability in the latter component is explained in terms of a descriptive model similar to the mathematical model developed by A. F. Huxley (1957). This treatment is shown to be adequate to explain not only the results presented in the previous paper (Chapman and Gibbs, 1972) but also the results obtained by other workers on both twitches and tetanuses using various preparations and experimental conditions.

Throughout this paper the muscle is regarded as a closed thermodynamic system exchanging energy but not matter with its environment. This situation is closely approximated during the few seconds over which initial enthalpy or initial heat measurements are made. Thus recovery processes are not included in the discussion.

THE ENERGETICS OF INTRACELLULAR CALCIUM TURNOVER

According to current theories of excitation-contraction coupling (Sandow, 1965; Ebashi and Endo, 1968) contraction is induced by release of calcium ion, Ca^{++}, from its confinement in the sarcoplasmic reticulum such that the sarcoplasmic free Ca^{++} concentration rises from its resting value of less than 10^{-7} M to greater than 10^{-4} M (Heilbrunn and Wiercinski, 1947; Portzehl et al., 1964; Bendall, 1969). Relaxation occurs when the Ca^{++} pump of the sarcoplasmic reticulum sequesters the Ca^{++} so that the free sarcoplasmic Ca^{++} level returns to its resting value. It is known that mitochondria can also accumulate Ca^{++} (Poe, 1969), but the radioautographic studies of Winegrad (1968, 1970) would suggest that in amphibian skeletal muscle the sarcoplasmic reticulum is the prime Ca^{++} transport system. It seems reasonable to assume that the nature of the constraint on the Ca^{++} is a low resting permeability of the sarcoreticular membranes to Ca^{++} such that Ca^{++} is retained in the sarcoreticular space until depolarization, conducted via the transverse tubules, causes a sudden increase in sarcoreticular membrane permeability, thus allowing Ca^{++} to diffuse relatively freely into the sarcoplasm. It is also likely that some sort of Ca^{++} binding occurs within the sarcoreticular space either by precipitation of calcium phosphates or by chelation of Ca^{++} with structural mucopolysaccharides (Marler and Davidson, 1965; Philpott and Goldstein, 1967; Dougherty, 1967; see also Sommer and Johnson, 1968). Such binding will reduce the chemical gradient which calcium has to be pumped and therefore the work per mole of Ca^{++} transported. The binding process itself, however, will not affect the energetic balance sheet over a complete contraction cycle because any heat of dissociation of calcium after stimulation will be thermally balanced by an opposite heat of binding during relaxation. It is also possible that a resting membrane potential exists across the sarcoreticular membranes (see discussion of Birks and Davey, 1969) and, if so, this would increase the work per mole of Ca^{++} transported.

Although the mechanism of ATP-dependent Ca^{++} pumping by the sarcoplasmic reticulum is at present unknown, the process can be expected to involve a series of stoichiometrically fixed chemical reactions involving Ca^{++}, ATP, and the enzymic transport mechanism. The essential properties of the system must be the presence of asymmetry and a high free energy for one of the chemical reactions (in this case ATP hydrolysis) to make the back reaction unlikely. These thermodynamic principles are clearly illustrated for Na^{+} transport by frog skin (Ussing, 1960) and have been demonstrated more recently for Ca^{++} transport by the sarcoplasmic reticulum.

Weber et al. (1966) showed that two calcium ions are transported by the sarcoplasmic reticulum for every ATP molecule hydrolyzed, and that this ratio is independent of the external Ca^{++} concentration. This independence of the Ca^{++}: ATP ratio means that the free energy of ATP hydrolysis is utilized with more or less thermodynamic efficiency depending on the electrochemical gradient against which the Ca^{++} is transported, rather than that more or less ATP is hydrolyzed. Whenever ATP is hydrolyzed during calcium pumping the enthalpy change will be approximately -11 kcal/mole (Wilkie, 1968). Because the work of Ca^{++} pumping is all "internal" work as far as the muscle mass is concerned, it follows that the ATP-dependent Ca^{++}-pumping activity of the sarcoplasmic reticulum will never be thermally neutral and the heat evolved will be equal to the enthalpy of hydrolysis of ATP associated with the pumping. This conclusion contradicts earlier statements in the literature (see Davies, 1963; Gibbs, 1967). The statement that heat will be evoked during such pumping is strongly supported by the differential calorimetric studies on the Ca^{++} accumulation of mitochondria (Poe, 1969). Poe has shown that when Ca^{++} is added to a medium containing mitochondria and oxidizable substrate and oxygen (or ATP), then the mitochondria rapidly accumulate Ca^{++}, consume substrate and oxygen (or ATP), and produce heat. Procedures which eliminate the development of tension or shortening, and thereby minimize the hydrolysis of ATP by actomyosin, will leave an initial heat component per activation that will be largely due to ATP hydrolysis by the Ca^{++} pump. Hill's definition (1949) of this component as the activation heat is therefore entirely appropriate and there should be no debate about its existence. The problem remains of establishing its magnitude.

The work of Hill (1949) and Gibbs et al. (1966), together with the present work, indicates that the activation heat component is about 1 mcal/g of muscle in the twitch. This indicates that 10^{-7} moles of ATP are hydrolyzed by the Ca^{++} pump, and, therefore, that approximately 2×10^{-7} moles of Ca^{++} are transported per gram of muscle per activation. Weber et al. (1963) indicate that about 10^{-6} moles of Ca^{++} must be removed from 1 g of Ca^{++}-saturated myofibrillar protein to achieve relaxation. As the myofibrillar proteins comprise about 12% of muscle wet weight (Bendall, 1969), this corresponds to a Ca^{++} removal of 1.2×10^{-7} moles/g of muscle per activation. Heilbrunn and Weircinski (1947) found that intracellular injection of 1.25×10^{-7} moles of Ca^{++}/g of muscle was sufficient to produce maximal contraction of pieces of frog muscle fibers. Bendall (1969) has established the exchangeable Ca^{++}-binding capacity of the contractile proteins to be 3.8×10^{-7} moles of Ca^{++}/g of muscle. Bendall (1969) also includes the statement that the apparent binding capacity of troponin could be 2 Ca^{++}/myosin molecule. Taking the density of myosin cross-bridges as 5.4×10^{16}/g (Huxley, 1960) this gives a Ca^{++}-binding capacity for troponin of 1.8×10^{-7} moles/g of muscle. This would constitute the Ca^{++} load to be borne by the sarcoreticular pump in completely extracting all the exchangeable Ca^{++} from Ca^{++}-saturated myofilaments. Thus, four

independent biochemical estimates of the amount of Ca^{++} cycled in each contraction give a range of 1.2–3.8 $\times$ 10^{-7} moles/g of muscle in close agreement with the value of 2 $\times$ 10^{-7} moles/g deduced from most myothermic experiments to date.

Further experiments in progress in this laboratory using double twitches and stretched muscles indicate that the true activation heat in toad sartorius muscle may lie in the range 0.5–1.0 mcal/g when all interference from myofibrillar ATPase activity has been eliminated. This suggests that in toad sartorius the amount of Ca^{++} transported corresponds to the lower end of the range predicted biochemically.

The onset of calcium pumping should occur immediately after stimulation as is consistent with the experimental results of Jöbsis and O'Connor (1966) and the theoretical model of Taylor (1969). Any attempt to follow the time course of Ca^{++} pumping by examining the time course of the activation heat, however, would be complicated by entropic transients associated with the release and rebinding of Ca^{++}. The radioautographic studies of Winegrad (1968, 1970) suggest that the released calcium returns to the terminal cisternae from the sarcoplasm after it has been bound by the tubular reticulum and actively transported into the lumen of the reticulum. We are of the opinion that the active transport of the calcium ions takes place early in the contraction cycle and that heat production accompanies this transport in an exactly analogous fashion to mitochondrial heat production (Poe, 1969). This suggestion of early active transport seems to be quite in accord with a possibility proposed by Winegrad (1970), namely, "Since the movement of calcium from the tubular reticulum to the terminal cisternae does not involve a process with a high activation energy, the splitting of ATP associated with the active accumulation of calcium by the reticulum may occur during the initial binding of calcium by the reticulum." The activation energy referred to in the above quotation is the physicochemical entity and not the myothermic one.

THE ENERGETICS OF THE TRANSDUCTION MECHANISM

Properties of the Model

It now appears that, upon physiological activation of a skeletal muscle fiber, the following sequence of events occurs (see reviews by Ebashi and Endo, 1968; Bendall, 1969; H. E. Huxley, 1969; Mommaerts, 1969):

(*a*) Calcium is liberated from the sarcoplasmic reticulum, probably from the region of the triads, and diffuses to the myofilaments.

(*b*) Calcium is bound to the troponin molecules of the thin filaments, thereby neutralizing the electrostatic repulsion that probably normally prevents interaction between the thick and thin filaments in the resting state.

(*c*) Interaction of thick and thin filaments occurs by direct combination of the globular head of the H-meromyosin cross-bridges emanating from the thick filaments with specific sites on the thin filaments.

(*d*) A conformational change occurs within the cross-bridge–actin complex such that tension is developed and ATP is hydrolyzed by the Mg^{++}-ATPase of the H-meromyosin cross-bridges with a stoichiometry of 1 ATP/cross-bridge per interaction.

To these four points the following speculations could be reasonably added:

(*a'*) A dynamic equilibrium exists between sarcoplasmic free Ca^{++} and troponin such that the dissociation constant of the Ca^{++}-troponin complex is lower when a local cross-bridge combination is intact than when the combination is broken or has not yet formed.

(*b'*) The cross-bridge–actin combination, on coming into existence, behaves like a stretched spring which will develop tension if the intramolecular conformational change is opposed as in isometric contraction, or will shorten if allowed to do so as in isotonic contraction.

(*c'*) The integrity of the cross-bridge–actin combination is dependent on the presence of ATP, intact, in the molecular combination; when ATP is hydrolyzed the combination disintegrates.

(*d'*) The hydrolysis of ATP by H-meromyosin ATPase is inhibited if the conformational change cannot take place, or facilitated if molecular "shortening" is allowed.

(*e'*) The total energy available to a cross-bridge–actin combination is quantized and is equal to the molecular enthalpy of ATP hydrolysis.

Before developing these suppositions further it is worth noting that points *b'* and *e'* taken together describe a viscoelastic system (Gasser and Hill, 1924), but this time at a quantal level, and with the reservation that "viscosity" becomes an obscure concept at the quantal level. Furthermore, points *c'* and *d'* taken together bear a close resemblance, at a descriptive level, to the model of muscle contraction developed by A. F. Huxley (1957).

The important aspect to be emphasized about the descriptive model just outlined is that it can explain a diversity of experimental observations on whole muscles by considering the summed behavior of a large number of fundamental quanta of contractile machinery. The variation of initial enthalpy with mechanical conditions is due entirely to the variation of the number n of quantal contractile events that results in the muscle developing tension or shortening against a load.

Therefore the problem of the self-regulation of muscle energetics can be restated as follows: What is the mechanism whereby, given a fixed amount of Ca^{++} liberated in the twitch to activate the contractile mechanism, the number of quantal contractile events per twitch can be varied? The answer is contained in the suggestions of points *a'* to *d'* listed above. This will now be shown by considering first a tetanic response and then by extending the discussion to the twitch response.

Suppose a muscle is maintaining a force of P_1 g wt. In the steady state this means that a certain number n_1 of cross-bridges are combined with actin. These n_1 bridges are disintegrating with a limiting rate constant k_1, so that n_1 is an instantaneous

value which remains constant for the whole muscle in an isometric steady state. Note that the limiting constant is here identified as the velocity constant k of the actomyosin ATPase enzyme. The enthalpy rate due to the maintenance of force will be $k_1 n_1 \times \delta H$, where δH is the molecular enthalpy of ATP hydrolysis; however, P_1 is determined by n_1 provided the average extension x of the actomyosin links is a certain value, say x_1. Suppose the muscles were to shorten against a load of P_1; then x would become less than x_1 and, by point b', the n_1 actomyosin links would now determine a force P less than P_1. In order for the muscle to bear the load P_1 while shortening, the instantaneous value of n must increase to n_2. Furthermore, by points c' and d' the mean rate constant k will increase to k_2 so that with the muscle shortening against a load P_1 the enthalpy rate will become $k_2 n_2 \times \delta H$ which is clearly greater than the isometric maintenance enthalpy rate. This argument is quite general and so it follows that a tetanized muscle will have a greater power consumption if it shortens against a load P_1 than if it maintains an isometric force of P_1.

By symmetry, using points b', and c' it follows that a muscle lengthening against a load P_1 will have a smaller power consumption than a muscle maintaining an isometric force of P_1. Thus the problem of how a muscle can apparently utilize external work performed on it (Abbott and Aubert, 1951; Abbott et al., 1951; Aubert, 1956; Hill and Howarth, 1959; Maréchal, 1964; Infante et al., 1964; Wilkie, 1968) disappears altogether. The work is not utilized by the muscle; the decreased enthalpy consumption is a consequence of the postulated properties of the model.

The description of the behavior of a tetanized muscle can be extrapolated, as a first approximation, to describe the energetics of twitches. The initial enthalpy of an isometric contraction in which $(\int P \cdot dt)_1$ units of force-time integral are generated will be given by $(\delta H \int k_1 n_1 dt)_1$ where k_1 is a function of internal shortening against the series elastic element, and n_1 is a function of force and rate of shortening against the series elastic element. The initial enthalpy of an isotonic contraction in which $(\int P \cdot dt)_1$ units of force-time integral are generated will be given by $(\delta H \int k_2 n_2 dt)_2$. Now k_2 and n_2 are both functions of contractile element shortening which now includes external shortening, and n_2 is also a function of force. The relative enthalpy consumptions of an isometric and an isotonic twitch each generating $(\int P \cdot dt)_1$ units of force-time integral can be appreciated qualitatively by considering the effect of force alone (in the absence of shortening) and then by taking into account the effect of shortening. If the effects of shortening are ignored (i.e., k_1 is held approximately invariant), then $(\int P \cdot dt)_1$ will be determined exactly by $(\int n_1 dt)_1$, independently of the shape of the force wave form. In practice, however the effect of shortening will be to increase $(\int n_1 dt)_1$ to $(\int n_2 dt)_2$ and to increase k_1 such that the number of quantal events to sustain an isotonic force-time integral of $(\int P \cdot dt)_1$ units will be $(\int k_2 n_2 dt)_2$. Thus the influence of shortening will make $(\delta H \int k_2 n_2 dt)_2$ considerably greater than its isometric counterpart when the integral is evaluated up to the time of peak shortening; during the lengthening (relaxation)

phase ($\delta H \int k_2 n_2 dt)_2$ will become less than its isometric counterpart. This effect is well demonstrated by the typical results shown in Fig. 2. The time involved in isotonic shortening during a twitch is much longer than the time involved in isotonic lengthening. Thus, when the effects of shortening and lengthening are integrated over the entire isotonic twitch, the model predicts than an isotonic contraction generating a given quantity of force-time integral will consume more enthalpy than an isometric contraction generating the same quantity of force-time integral.

At this stage one can replace equations 1–3 of the preceding paper (Chapman and Gibbs, 1972) with the following expression for the initial enthalpy of a muscle twitch:

$$E = A + \delta H \int kn \cdot dt, \tag{4}$$

where A = activation enthalpy, being the energy consumption of the sarcoreticular Ca^{++} pump; δH = molecular enthalpy of ATP hydrolysis in vivo; k = rate "constant" for disintegration of cross-links between actin and myosin (ATPase velocity constant); n = instantaneous number of cross-links in existence; and the integral is evaluated between the moment of stimulation ($t = 0$) and the completion of relaxation ($t = \infty$). Now n and k, as described earlier, are dependent on force and velocity of shortening, both of which are dependent upon each other according to the "characteristic equation" (Hill, 1938).

To develop equation 4 any further requires explicit quantitative working hypotheses to be developed about the points a'–d' listed above, particularly the relationship between ATPase activity and molecular shortening (point d'); however, in its general form the model, together with equation 4, is still useful to account descriptively for observed mechanical and myothermic data.

Mechanical and Energetic Corollaries of the Transduction Model

As shown above, a muscle lengthening against an afterload of P_1 will have, say, n_1 cross-links in existence at any instant, where n_1 is less than n_2, the instantaneous number of cross-links necessary to maintain an isometric force equal to P_1. Hence, in an isotonic twitch, when the isotonic lever hits the afterload stop during relaxation, n_1 suddenly becomes inadequate to sustain P_1 isometrically and so there should be a sharp collapse of tension at this instant as shown in the P record of Fig. 3 of the previous paper and in the superimposed records of Fig. 1. An increase in n at this instant is not possible because there is insufficient free Ca^{++} available to reactivate the contractile mechanism.

The effect of shortening in reducing the duration of the "active state" (Jewell and Wilkie, 1960) would be expected from points a', b', c', and d' listed above. The increased turnover of cross-links would increase the rate of dissociation of Ca^{++} from the troponin molecules. Thus the rate of loss of calcium from the myofilaments would be expected to be greater in a muscle that is shortening. The converse of the

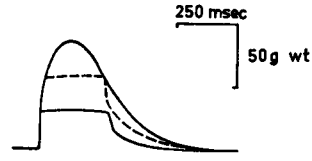

FIGURE 1 Contractile force developed isometrically (upper solid line) and isotonically against loads of 60 g (dashed line) and 30 g (lower solid line).

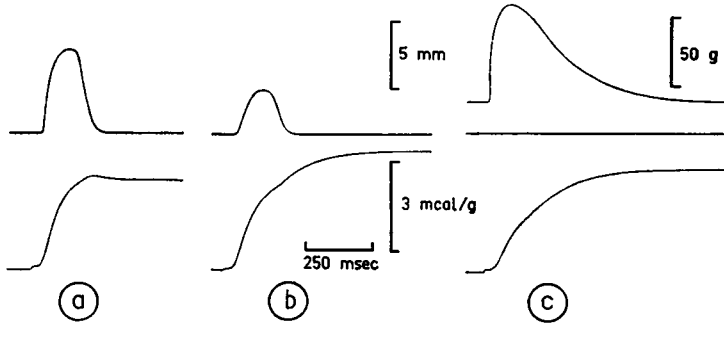

FIGURE 2 Original records: mechanical (top) and heat (bottom) of: (*a*) an isotonic contraction, load 10 g wt; (*b*) an isotonic contraction, load 35 g wt; (*c*) an isometric contraction at l_0. The muscle weighed 157 mg and had a resting length of 3.8 cm. Temperature, 7°C.

effect of shortening on the duration of the active state is also predictable from points a', b', c', and d'; the persistence of tension will prolong the active state duration (cf. Hill, 1964 *a*) because the myofilaments will retain hold of the Ca^{++} for a longer period during the twitch.

The rate of heat production will depend on the rate of Ca^{++} accumulation by the sarcoreticular pump and on the velocity of shortening as discussed previously. In an isometric twitch the heat production should be greatest just after stimulation where Ca^{++} is being accumulated most rapidly and the velocity of internal shortening is greatest. This is indeed the case for frog sartorius (Hill, 1949) and appears to hold for toad sartorius under the present experimental conditions (see Fig. 2).

An important property of the model arising from equation 4 is that the extra enthalpy consumed in isotonic twitches, relative to isometric enthalpy consumption, is not directly related to the work performed but is rather related to sensitivity of the velocity constant of the actomyosin ATPase to shortening. Thus it is not surprising that the isotonic heat production reported in the present work (Chapman and Gibbs, 1972) should fall below the isometric heat. It is important to make a clear distinction between the *magnitude* of the isotonic heat production, which is less in our experiments than in its isometric counterpart, and the *rate* of isotonic heat production, which initially is higher than in the isometric case (see Fig. 2). The increase in total initial enthalpy associated with shortening varies considerably in magnitude between frog sartorius at 0°C (Hill, 1949, 1964 *b*) and toad sartorius

at 6–12°C (Jöbsis and Duffield, 1967; Chapman and Gibbs, 1972). These differences are possibly associated with differences in ATPase activity of the two muscles. Bárány (1967) has correlated ATPase activity with maximum velocity of shortening between different muscles at different temperatures. It could be that faster muscles have a wastefully high actomyosin ATPase activity when shortening occurs at certain temperatures, e.g., frog muscle at 0°C. On the other hand, the absolute increase in actomyosin ATPase activity in frog muscles at higher temperatures (Bárány, 1967) could reduce the sensitivity of k (see equation 4) relative to velocity of shortening, resulting in a relative absence of "shortening heat" in frog muscle at 11–12°C (Fischer, 1931; McCarter and Ramsey, 1968). This is to say that the rate constant k of the term $\int kn \cdot dt$ in equation 4 displays different sensitivities to shortening at different temperatures in different species.

The model, as developed in this paper, does not necessitate partition of energy transduction by actomyosin into contractile element work, shortening heat, and tension-maintenance heat. It merely states that the fundamental quantum of energy transduction is the enthalpy of hydrolysis of one molecule of ATP for every actomyosin link that is made and broken, and this molecular enthalpy summed over the number of linkages that are made and broken in a contraction cycle accounts entirely for the total enthalpy (heat plus work) of the contraction after the heat due to the Ca^{++} pump has been subtracted. The self-regulating properties of muscle energy consumption are due to the intrinsic properties of the quantal contractile event which has a turnover rate sensitive to the velocity of shortening.

We thank Mr. M. Hart and Mr. H. Vogelsanger for technical assistance, and Miss D. Harrison and Miss S. Woolley for preparation of diagrams.
This paper is based on part of a thesis submitted by J. B. Chapman for the degree of Doctor of Philosophy.

This work was supported by a grant (68/2403) from the National Health and Medical Research Council of Australia.

Received for publication 14 July 1970 and in revised form 27 October 1970.

REFERENCES

ABBOTT, B. C., and X. M. AUBERT. 1951. *Proc. Roy. Soc. Ser. B. Biol. Sci.* **139**:104.
ABBOTT, B. C., X. M. AUBERT, and A. V. HILL. 1951. *Proc. Roy. Soc. Ser. B. Biol. Sci.* **139**:86.
AUBERT, X. 1956. Le couplage énergétique de la contraction musculaire. Arscia, S. A., Brussels.
BÁRÁNY, M. 1967. *J. Gen. Physiol.* **50**:197.
BENDALL, J. R. 1969. Muscles, Molecules and Movement. Heinemann Ltd., London.
BIRKS, R. I., and D. F. DAVEY. 1969. *J. Physiol. (London).* **202**:171.
CHAPMAN, J. B., and C. L. GIBBS. 1972. *Biophys. J.* **12**:215.
DAVIES, R. E. 1963. *Nature (London).* **199**:1068.
DOUGHERTY, W. J. 1967. *J. Cell Biol.* **35**:34A.
EBASHI, S., and M. ENDO. 1968. *Progr. Biophys. Mol. Biol.* **18**:123.
FISCHER, E. 1931. *Amer. J. Physiol.* **96**:78.
GASSER, H. S., and A. V. HILL. 1924. *Proc. Roy. Soc. Ser. B. Biol. Sci.* **96**:398.
GIBBS, C. L. 1967. *Aust. J. Exp. Biol. Med. Sci.* **45**:379.

GIBBS, C. L., N. V. RICCHIUTI, and W. F. H. M. MOMMAERTS. 1966. *J. Gen. Physiol.* **49**:517.
HEILBRUNN, L. B., and F. J. WIERCINSKI. 1947. *J. Cell. Comp. Physiol.* **29**:15.
HILL, A. V. 1938. *Proc. Roy. Soc. Ser. B. Biol. Sci.* **126**:136.
HILL, A. V. 1949. *Proc. Roy. Soc. Ser. B. Biol. Sci.* **136**:195.
HILL, A. V. 1964*a*. *Proc. Roy. Soc. Ser. B. Biol. Sci.* **159**:589.
HILL, A. V. 1964*b*. *Proc. Roy. Soc. Ser. B. Biol. Sci.* **159**:596.
HILL, A. V., and J. V. HOWARTH. 1959. *Proc. Roy. Soc. Ser. B. Biol. Sci.* **151**:169.
HUXLEY, A. F. 1957. *Progr. Biophys. Biophys. Chem.* **7**:255.
HUXLEY, H. E. 1960. *In* The Cell. J. Brachet and A. E. Marsky, editors. Academic Press, Inc., New York. **4**:365.
HUXLEY, H. E. 1969. *Science* (*Washington*). **164**:1356.
INFANTE, A. A., D. KLAUPIKS, and R. E. DAVIES. 1964. *Science* (*Washington*). **144**:1577.
JEWELL, B. R., and D. R. WILKIE. 1960. *J. Physiol.* (*London*). **152**:30.
JÖBSIS, F. F., and J. C. DUFFIELD. 1967. *Science* (*Washington*). **156**:1388.
JÖBSIS, F. F., and M. J. O'CONNOR. 1966. *Biochem. Biophys. Res. Commun.* **25**:246.
MARÉCHAL, G. 1964. Le métabolisme de la phosphorylcreatine et de l'adenosine triphosphate durant la contraction musculaire. Arscia, S. A., Brussels.
MARLER, E., and E. DAVIDSON. 1965. *Proc. Nat. Acad. Sci. U.S.A.* **54**:648.
MCCARTER, R. J. H., and R. W. RAMSEY. 1968. *Int. Congr. Physiol. Sci. Symp. Spec. Lect. 24th.* **7**:286. (Abstr.)
MOMMAERTS, W. F. H. M. 1969. *Physiol. Rev.* **49**:427.
PHILPOTT, C. W., and M. A. GOLDSTEIN. 1967. *Science* (*Washington*). **155**:1019.
POE, M. 1969. *Arch. Biochem. Biophys.* **132**:377.
PORTZEHL, H., P. C. CALDWELL, and J. C. RUEGG. 1964. *Biochim. Biophys. Acta.* **79**:581.
SANDOW, A. 1965. *Pharmacol. Rev.* **17**:265.
SOMMER, J. R., and E. A. JOHNSON. 1968. *J. Cell Biol.* **36**:497.
TAYLOR, C. P. S. 1969. *Biophys. J.* **9**:759.
USSING, H. H. 1960. *J. Gen. Physiol.* **43**:135.
WEBER, A., R. HERZ, and I. REISS. 1963. *J. Gen. Physiol.* **46**:679.
WEBER, A., R. HERZ, and I. REISS. 1966. *Biochem. Z.* **345**:329.
WILKIE, D. R. 1968. *J. Physiol.* (*London*). **195**:157.
WINEGRAD, S. 1968. *J. Gen. Physiol.* **51**:65.
WINEGRAD, S. 1970. *J. Gen. Physiol.* **55**:77.