The absorption of work by a muscle stretched during a single twitch or a short tetanus

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When a stimulated muscle is stretched fairly quickly during the active phase of contraction, it resists strongly and mechanical work must be done in stretching it. What happens to this work? If the length to which the muscle is stretched is not too great no significant part of the work remains as mechanical (elastic) energy after the muscle has relaxed. The total heat produced up to the end of relaxation is greater than it would have been had no work been performed on the muscle, but the excess is too small to account for all the work done. It is concluded that the missing work, about half of the whole, is absorbed, presumably as chemical energy.

If a stretch is applied entirely during the relaxation phase, when activity is over but tension is still present, the whole of the work performed reappears as heat.

If the view is accepted that the missing work is absorbed in chemical synthesis, it appears that the physical system responsible for mechanical work is reversibly coupled, during the active state, with a chemical system providing the necessary energy; and that this coupling is broken when activity passes off. Other possible hypotheses, however, are discussed.

The application to ordinary muscular movement is referred to.

It was reported some years ago (Hill 1938, p. 177) that a muscle maintaining a steady isometric contraction, and then subjected to a constant load rather greater than it can bear, is capable of absorbing a large part, or the whole, of the mechanical work done on it by the load during the slow lengthening that ensues. Indeed, it appeared, though the evidence was not then conclusive, that after the initial readjustment of tension, and provided that the load was not so large as to cause the muscle to 'give' irreversibly, not only did the whole of the work disappear, but also a negative 'heat of lengthening' accompanied the process. The work which disappeared was presumably absorbed in driving backwards the chemical changes which normally provide mechanical work when a muscle is able to shorten, while the negative heat of lengthening was naturally assumed to be the converse of the positive heat production which accompanies shortening.

It was shown in 1938 for maintained contractions, and since confirmed for single twitches (Hill 1949 *b*), that mechanical work is performed by muscle without any extra liberation of heat beyond that associated with shortening as such. By varying the load and the manner of its application the work can be varied, but provided that the amount of shortening is kept constant the heat remains constant also. The situation can be expressed by the equation

$$E = A + ax + \int P \, dx,$$

where all the quantities except a are functions of the time, E is the total energy liberated, A the heat of activation, x the amount of shortening, a a constant, P the

load and $\int P\,dx$ the mechanical work. The results obtained by stretching appeared to show that the relation described by the equation applies just the same to lengthening as to shortening, that x can be negative as well as positive. It seemed, therefore, that the physical framework by which mechanical effects are produced when activated by stimulation is reversibly coupled to chemical processes which provide the work and the heat.

These conclusions were so fundamental to an understanding of the energetics, and so of the chemistry, of muscular contraction that they needed, if possible, to be confirmed and extended. Unfortunately, the war distracted attention from them for six years, and two more years were needed for the laboratory to be reinstated and the new equipment constructed which was needed for an adequate study of the problem. At first, attention was devoted to a closer analysis of the energetics of shortening, particularly in single twitches; but during the last two years a concentrated effort has been made to obtain decisive results on the energetics of lengthening. Experimentally, this proved more difficult than expected, but we believe that firm conclusions can now be drawn.

The present paper refers only to experiments involving a single twitch or a short tetanus, in which—owing to the limited time available—a stretch has to be rather quick if effects are to be produced which are large enough for accurate measurement. In a quick stretch, however, a muscle is always liable to 'give' irreversibly, and some of the mechanical work done is then degraded into heat. This obscures the heat of lengthening which will not be referred to further here. In the following paper, Abbott & Aubert (1951) return to the older method of a slow isotonic lengthening; their results, obtained with a more adequate technique than was then available, confirm the conclusions of 1938.

There were two special reasons for investigating the single twitch or the short tetanus: (a) It has long been known that in ordinary relaxation heat is produced which can be attributed to mechanical work, or elastic potential energy, degraded (Hill 1938, p. 167; 1949a). We expected to find, and, in fact, found, that the mechanical work done in stretching a muscle during relaxation appears quantitatively as heat. If work performed during full activity was absorbed, should we find an intermediate stage—as activity merged into relaxation—in which less work was absorbed and more degraded? (b) The interpretation of the earlier experiments, on slow isotonic lengthening, required an assumption the validity of which might be questioned. During a maintained contraction a considerable amount of heat is produced—the summated effect of the heats of activation resulting from successive elements of the stimulus. From the heat given out during a slow stretch it was necessary to subtract the heat that would have been produced in the same interval had no stretch occurred; the effect of the stretch, therefore, was the small difference between two larger quantities. If the operation of stretching as such somehow affected the processes required to maintain the active state, the subtraction might lead to erroneous results. There was no reason to suppose that it did, but it would be better to explore the problem in some way requiring no such assumption. In a twitch, or a short tetanus, the whole of the heat could be measured, and compared

for the cases of stretch and no stretch. If the work done considerably exceeded the difference (heat for stretch) − (heat for no stretch), some of it must have been absorbed. This proved to be so; about half the work done in stretching a muscle during its active phase disappeared, that is, could not be found afterwards—when the muscle had finished its contraction and relaxed—either as heat or as mechanical potential energy. (There are good reasons, which will be discussed later, why the whole of the work cannot disappear when stretches are made in this way.) The result gave us confidence in returning to the other experiments, those on the slow isotonic stretch, which are described by Abbott & Aubert (1951).

The chief experimental difficulty has been to make sure that measurements of the heat production are valid under the rather drastic conditions affecting the muscle during a powerful stretch. The sensitivity and speed of the myothermic equipment (see Hill 1949c) are entirely adequate, indeed at 0° C it is possible to dispense with any analysis allowing for time-lag in heat flow and galvanometer response, and to apply nothing more than a small and easy allowance for heat loss. The danger in these experiments is that, under a powerful stretch, different regions along the length of the muscle may behave differently (for example, by reason of a slightly different cross-sectional area), one region 'holding' and lengthening reversibly, another 'giving' and lengthening irreversibly. The difference of strength need not be large, since even a slightly weaker region is bound to 'give' first and so relieve the strain on the remainder when the whole is being stretched at a constant velocity. An undue proportion of the work done by the stretch would then appear in the part that 'gave', and if some of this region were not on the thermopile too low a heat deflexion would result. Just as dangerous is the possibility of one region relaxing rather earlier than another; too large a share of the mechanical energy still present as potential energy of elastic stretch would then be released as heat in the part which relaxed first.

For six months doubtful or erroneous results were obtained in this way, some of them even appearing to show that the whole of the work done in stretching a muscle during a twitch could be absorbed. This last result seemed incredible, because some of the work was certainly carried over as potential energy of elastic stretch into the relaxation phase, when it should have been dissipated as heat. A new thermo-pile, therefore, was designed in order to obviate the errors suspected to be present. This was constructed by Mr A. C. Downing, to whose skill we are indebted for a masterpiece of instrument making without which the present reliable results could not have been obtained. All the experiments described below were made with this thermopile, the results with the earlier ones being discarded; it has the advantage, not only that errors are largely avoided, but that a ready means is available of detecting them if present.

Thermopile

The earlier experiments were all made with one or other of the 'protected' thermopiles described in previous papers (Hill 1937, 1938, 1949c). These had been designed for contractions in which the muscle was allowed to shorten, sometimes to a considerable extent, and the 'protecting' region ensured that no part of the

muscle came on to the active elements which had not previously been subject to the same thermal conditions as the part already on them. The active region of the thermopile, therefore, had to be considerably shorter than the whole muscle, and measured the rise of temperature only in a limited region towards one end (the pelvic end in the case of a frog's sartorius). In a good muscle, under normal conditions of contraction, there was no evidence that significant errors are caused by 'sampling' the rise of temperature in this way; indeed, the new thermopile has given direct evidence that they are not. In a muscle, however, subjected to the drastic procedure of a rapid stretch during a maximal contraction, in which very high tensions are developed, a slight inequality of strength between the two ends may lead to a considerable difference in the extent to which they are stretched. The part which 'gives' more will take up irreversibly more than its proportional share of the work, and its rise of temperature will be greater.

With the existing thermopiles there was no way of avoiding this possible error, nor even of detecting whether it occurred. It was necessary (a) to use a longer thermopile, sampling the temperature of the muscle over more of its length, and (b) to provide means by which the rise of temperature of one end could be compared with that of the other. Since shortening was not to occur, but only lengthening, a 'protecting' region was unnecessary, so a longer active thermopile could be used. Another requirement, in order to ensure equality of response of the two halves of the muscle, was a central electrode to serve as a stimulating cathode, with anodes at the two ends. With this arrangement one could reasonably expect that all the fibres of the muscle would be effectively stimulated. Finally, it was desirable, since heat had to be measured over periods of seconds, that the rate of heat loss should be as low as possible, so that metals of low thermal conductivity were preferred for the thermo-elements.

These requirements were met by the instrument shown in figure 1. Its elements are of constantan-manganin wire, originally of 42 s.w.g. (102μ), rolled out to a thickness of 15μ and a width of 0·34 mm. The insulation, as in earlier instruments by mica and Bakelite varnish, is very good, a resistance of hundreds of megohms being found from muscle to thermopile after soaking for hours in Ringer's fluid. This ensures freedom from direct leak of stimulating current. The total thickness is about 45μ, which is equivalent in heat capacity to about 34μ of muscle, or 17μ on each side, since two muscles are used. This instrument is referred to as D_1, a second one (slightly modified) as D_2.

The thermopile is built in two symmetrical halves, normally used in parallel; when required, however, either half can be connected separately to the galvanometer. The three platinum electrodes, rolled to 45μ (the same thickness as the complete thermopile), lie flush with the surface of the mica, sealed to it by Bakelite varnish. Their arrangement (the central cathode being close to both terminals, + and −, of the thermopile, the two anodes being close to the + and the − respectively) helps further to reduce leak of the stimulus to the galvanometer through the rather high capacity of the mica insulation. All other details are similar to those described for previous instruments. For thermopile D_1, with 24 couples in each half, the resistance in parallel is 19·5 ohms and the sensitivity

956 μV/1° C. For D_2, with 22 couples in each half, the resistance in parallel is 18·8 ohms and the sensitivity 877 μV/1° C. Each half, used alone, has the same sensitivity in μV/1° C but twice the resistance. If unnecessary sensitivity was available the galvanometer could be quickened and stabilized by feed-back (Hill 1948b).
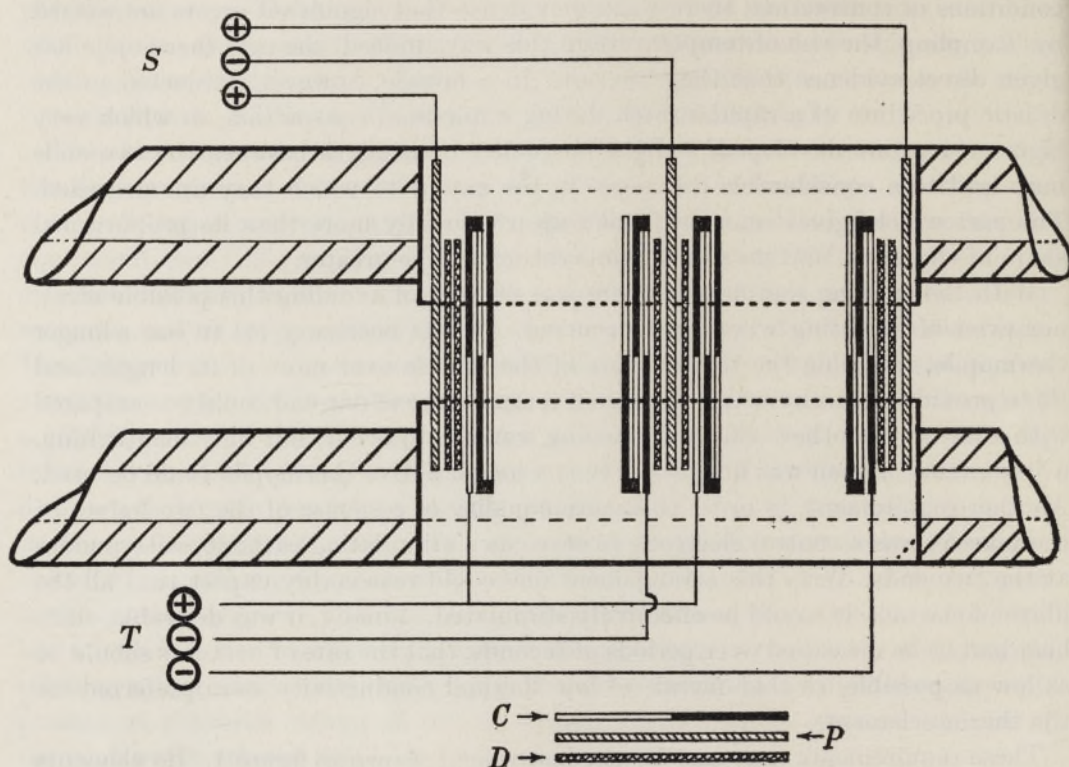

FIGURE 1. Double thermopile D_1 with central stimulating cathode. Overall length between outer edges of electrodes 22·5 mm.; length between outer thermocouples 19·2 mm. D_1 consists of two similar thermopiles with three terminals (one common +), capable of being used in parallel or separately. Each thermopile has twenty-four couples of constantan-manganin (rolled wire), 15 μ thick, 0·34 mm. wide, mica and Bakelite insulation about 15 μ each side, resistance 39 ohms, sensitivity 956 μV/1° C. Resistance in parallel 19·5 ohms. 'Hot' junctions in line down the middle; 'cold' junctions alternately on either side, 'loaded' with solder and in thermal proximity to brass plates soldered to the frame. Stimulating electrodes, rolled platinum wire, 45 μ thick, flush with the face, cathode in the middle, anodes at the two ends. The leads to the thermopile and electrodes pass up through the insulated brass tubes forming the frame. There are two 'dummy' thermocouples of constantan at each end, and one on each side of the central electrode. The sensitivity was determined with rolled constantan-manganin thermocouples of the same wire, with junctions at a measured difference of temperature in the neighbourhood of 0° C. C, manganin-constantan thermocouple; P, platinum electrode; D, dummy thermocouple; T, leads from thermopiles to galvanometer; S, leads from electrodes to stimulating circuit.

Recording

The thermopile was connected to a rapid galvanometer of the type used for all recent myothermic experiments in this laboratory (Downing 1948; Hill 1948a, b; 1949a); its deflexion operated one beam of a large cathode-ray tube. Photographic

records were made in a fixed camera, usually of several linear sweeps (with negligible fly-back time (Attree 1949)) in order to expand the time-scale. The tension of the muscle produced a proportional deflexion of the other beam by the method recently described (Hill 1950). The photo-electric recorder was carried on the arm of a Levin–Wyman ergometer which could travel at any velocity, start at any moment and move through any distance required. The simultaneous recording of heat and tension on a single sheet of paper ($10\cdot8 \times 8\cdot2$ cm.) was a great convenience and added largely to the speed and accuracy of the analysis. The beginning and end of the stretch were indicated on the record. The ergometer reached its terminal velocity so quickly that a tension-time record of the stretch was effectively the same as a tension-distance record, and the work done could be simply calculated from the area of the tension-time curve between start and stop, multiplied by the appropriate constant. Thus a single photographic record, continued until the muscle had relaxed, gave complete information of the magnitude and time course of the heat and tension and of the work done in the stretch. The only correction of any importance was that for heat loss, which was simple to apply later, being the area of the temperature-time curve up to the point considered, multiplied by a constant determined directly in each experiment.

RESULTS

The results are best introduced by describing in detail a single rather complete experiment (24 March 1950). A pair of frog's sartorii weighing 176 mg., 30·5 mm. long in the body with the legs laid out in line ('standard length'), was mounted on thermopile D_1 and soaked in Ringer's fluid at 0° C. The Ringer was replaced by oxygen and four preliminary contractions were recorded; then a series of twitches was begun, with a 3·5 mm. stretch from 27·5 to 31 mm. The results are given in figure 2, and in figure 3 on an enlarged time-scale, the beginning and end of stretch being marked by arrows.

Record (5) was for a stretch starting at 40 msec. after the shock and ending at 212 msec. It is not shown in the figures, being practically indistinguishable from record (8). The heat in record (5) was 50·4 g.cm., the work done in the stretch 40·5 g.cm. Record (6) was for a stretch during relaxation, starting at 455 msec. and ending at 624 msec.; the heat was 41·1 g.cm., the work 15·5 g.cm. Record (7) was for a stretch starting at 246 msec., just at the peak of the isometric tension, and ending at 415 msec. The heat was 55·7 g.cm. and the work 34·3 g.cm. Record (8) was for an early stretch, 44 to 216 msec., ending just before the peak of an isometric contraction. The heat was 52·0 g.cm. and the work 40·7 g.cm. Record (9) was for an isometric contraction in the long position, 31 mm.; the heat was 29·9 g.cm., the maximum tension 95 g. Record (10) was for an isometric contraction in the short position, 27·5 mm.; the heat was 28·4 g.cm. and the maximum tension 89·5 g. The maximum tensions were very high for a twitch ($Tl_0/M \approx 1600$) showing that the muscles were in excellent condition. (Tl_0/M is about equal to the force exerted per cm.² of muscle cross-section, T being tension, l_0 standard length and M weight of muscle.)

At the end of the experiment the muscles were heated electrically and the rate of heat loss recorded, viz. 3·23 % per sec. The heat records of figures 2 and 3 have been corrected for heat loss—a small correction, having no significant effect before 200 msec. No other analysis was applied; owing, therefore, to galvanometer lag and lag in heat flow to the thermopile, all the heat curves are slightly delayed and the sharpness of their changes is slightly blunted. A numerical analysis could have
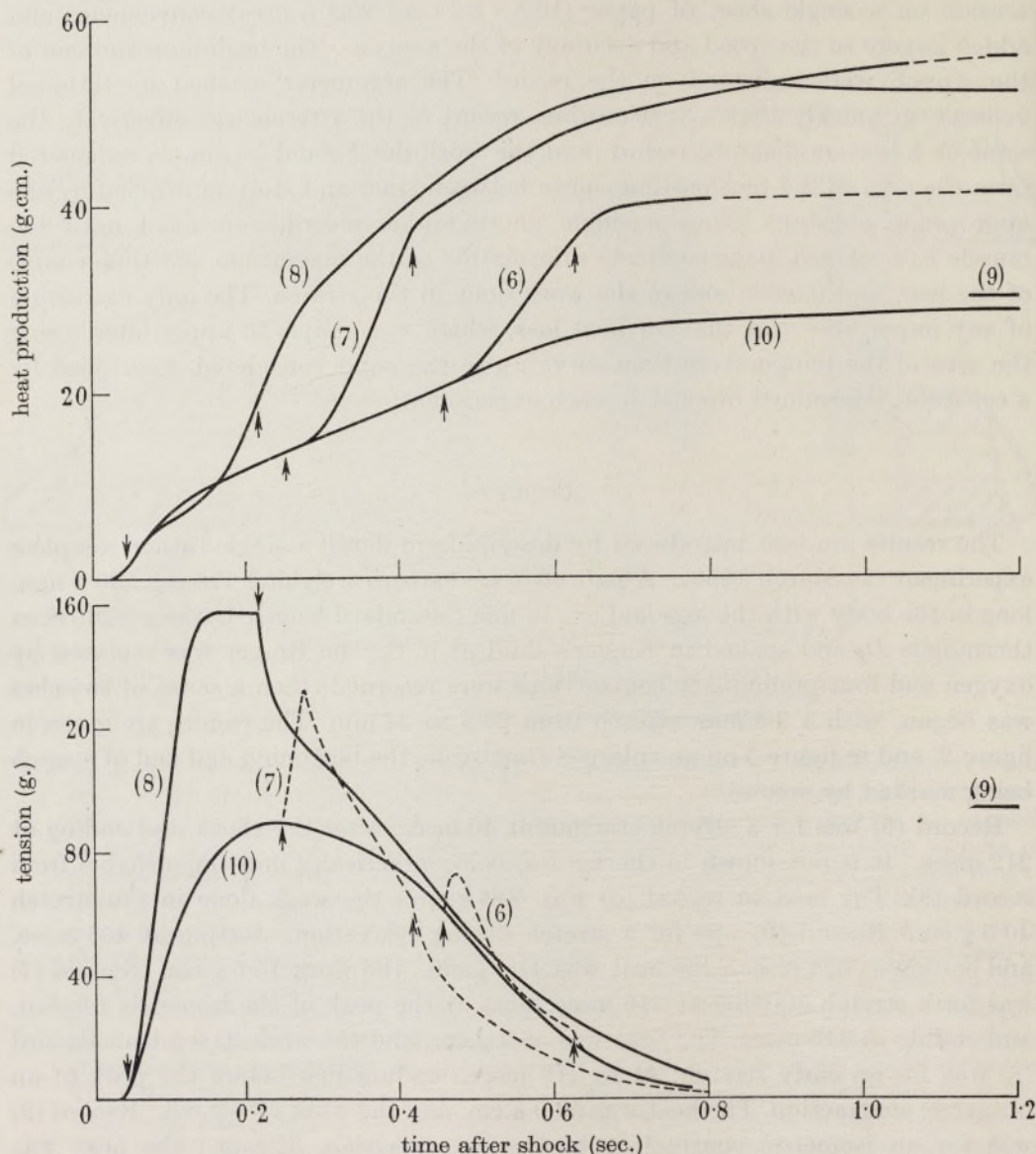

FIGURE 2. Twitch of frog's sartorii at 0° C. To show tension (below) and heat (above) with a stretch of 3½ mm. applied at various phases of contraction (the beginnings and ends of the three stretches are indicated by arrows). Record (10), isometric (short); record (8), stretch from 44 to 216 msec. after the stimulus; record (7), stretch from 246 to 415 msec.; record (6), stretch from 455 to 624 msec. Record (9), isometric (long) is not shown, except (on the right) for total heat and maximum tension. For fuller details see text. For records (8) and (10) during the first 0·3 sec. see figure 3.

been carried out, but it would have been very laborious, it would not have affected total quantities, and the picture of the heat changes would not have been significantly altered. Moreover, for the following reason it would not really have improved the accuracy.
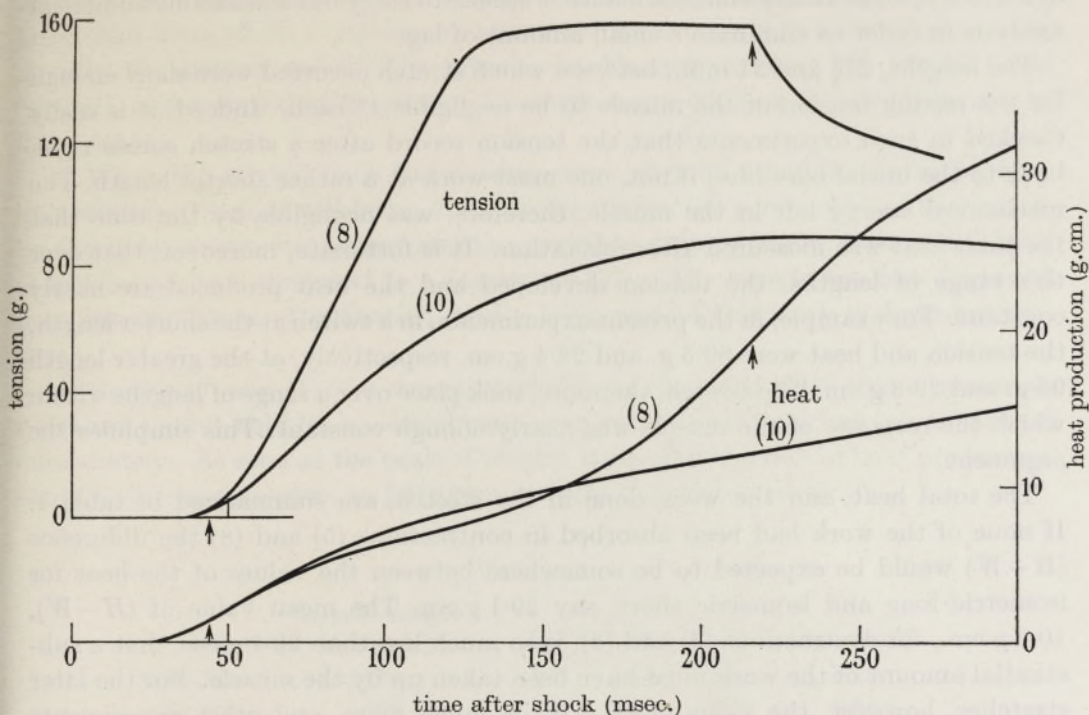

FIGURE 3. Twitch of frog's sartorii at 0° C. Heat and tension. Comparison of early stretch (record (8)) and isometric (record (10)). The same experiment as in figure (2) but on a fourfold time-scale. Beginning and end of stretch shown by arrows. For details see legend of figure 2 and text.

In such experiments as these, which put a considerable strain on a muscle (in record (8) a tension of nearly 3 kg./cm.² was registered), there is always apt to occur a slight inequality of heat production in the different regions of its cross-section. This is probably due to different groups of fibres 'giving' at different stages of a stretch, or relaxing earlier, and so receiving rather different quantities of energy from the work done in the stretch, or degraded in relaxation. These inequalities show themselves by delay in attaining a final steady level of the rise of temperature due to contraction, since 1 to 2 sec. is usually required for differences of temperature to be equalized by conduction (Hill 1938, p. 151). The allowance for heat loss in 1 to 2 sec. is small but not negligible; when it is made the corrected rise of temperature usually becomes constant by 2 sec. Record (8) in figure 2 shows an example of this: by 1·2 sec. the rise of temperature corrected for heat loss had reached its final value, but at 0·8 sec. it was 2 % higher than the final value, due to the part of the muscle near the thermopile being slightly warmer than the outer part. This effect is more liable to occur with a stretch, and did not appear in the isometric contractions of the same experiment. If it is large it strongly suggests

a muscle in poor condition, and the results had better be discarded. The muscle in the present experiment was very good, and continued to give consistent results in spite of being stretched. There is no reason to suppose that the temperature recorded at any moment was very far from the true average; but the possibility of a 2 to 5 % error at any moment makes it useless to carry out a laborious numerical analysis in order to eliminate a small amount of lag.

The lengths, $27\frac{1}{2}$ and 31 mm., between which stretch occurred were short enough for the resting tension of the muscle to be negligible at each. Indeed, it is easily checked in such experiments that the tension record after a stretch comes right back to the initial base-line; if not, one must work at a rather shorter length. The mechanical energy left in the muscle, therefore, was negligible by the time that the total heat was measured after relaxation. It is fortunate, moreover, that over this range of lengths, the tension developed and the heat produced are nearly constant. For example, in the present experiments, in a twitch at the shorter length, the tension and heat were 89·5 g. and 28·4 g.cm. respectively, at the greater length 95 g. and 29·9 g.cm. The stretch, therefore, took place over a range of lengths within which the response of the muscle was nearly enough constant. This simplifies the argument.

The total heat, and the work done in the stretch, are summarized in table 1. If none of the work had been absorbed in contractions (5) and (8) the difference $(H - W)$ would be expected to be somewhere between the values of the heat for isometric long and isometric short, say 29·1 g.cm. The mean value of $(H - W)$, 10·6 g.cm., for contractions (5) and (8), is so much less than 29·1 g.cm. that a substantial amount of the work must have been taken up by the muscle. For the later stretches, however, the value of $(H - W)$ is much more, and other experiments reported below show that in late enough stretches the whole of the work is turned into heat.

TABLE 1

record no.	interval of stretch (msec.)	heat H (g.cm.)	work W (g.cm.)	$H - W$ (g.cm.)
5	40 to 212	50·4	40·5	9·9
6	455 to 624	41·1	15·5	25·6
7	246 to 415	55·7	34·3	21·4
8	44 to 216	52·0	40·7	11·3
9	isometric long	29·9	—	29·9
10	isometric short	28·4	—	28·4

The curves of figures 2 and 3 show several interesting relations:

(a) The first effect of an early stretch is slightly to diminish the rate of heat production (cf. curves (8) and (10) in figures 2 and 3 and also curves (1a) and (2a) in figure 5 below). This is explained as being due to a decrease of the heat of shortening. During an 'isometric' contraction the active elements shorten and stretch the series elastic elements. As the tension is raised by a stretch the shortening of the contractile elements diminishes in speed and soon ceases altogether. The missing heat in record (8) has a maximum value of the order of 1 g.cm., which corresponds to about 0·36 mm. of internal shortening prevented by the stretch up to about 0·12 sec. At that stage another factor begins to come in.

(b) Heat curve (8) for stretch (figures 2 and 3) continues to deviate downwards from heat curve (10) for isometric until about 120 msec., which is just at the end of the nearly linear portion of the curve of rising tension during stretch. The muscle was able to bear a tension of about 130 g. before it began to 'give' irreversibly. As the stretch continued and the tension rose beyond 130 g., the rise ceased to be linear and bent off to a plateau at about 157 g., from which it dropped rapidly directly the stretch ended. The muscle, unable now to bear the tension, was 'giving', and a considerable part of the work being done on it was being turned into heat.

(c) When the stretch ends the tension curve (figures 2 and 3) begins to drop immediately. Work is still being done on the contractile component of the muscle by the series elastic component and the recording system (chain, lever, etc.), both of which had been stretched to a tension of 157 g. The muscle has now begun to pass into the phase of relaxation, and a large part of this work is degraded into heat.

(d) Curves (6) and (7) of a stretch started later (figure 2) show that a muscle so stretched cannot hold its tension during even early relaxation, but 'gives' almost immediately. As soon as the peak of tension is reached the rate of heat production starts to increase at once, to fall again directly the stretch stops.

TABLE 2

date	muscle weight (mg.)	H_s	W	H_i	$(H_s - H_i)/W$
20 Jan.	188	40·7	33·6	25·4	0·46
14 Feb.	206	68·7	58·9	30·8	0·64
15 Feb.	168	64·5	47·3	25·8	0·82
17 Feb.	143	37·2	25·4	19·6	0·69
13 Mar.	102 (toad)	22·5	18·6	13·5	0·48
24 Mar.	176	50·4	40·5	29·1	0·53

An experiment is shown in figure 4 in which a pair of toad's sartorii was stretched during a twitch. These muscles also were very strong, as shown by the high values of Tl_0/M for an isometric twitch. The stretch started rather later, and internal shortening had mostly occurred before the stretch began; consequently little of the early heat of shortening was prevented and the heat curve for stretch did not go detectably below that for isometric. The value of H_s (heat for stretch) was 22·5 g.cm., that for H_i (heat isometric) had a mean value (long and short) of 13·5 g.cm.; thus $H_s - H_i = 9·0$ g.cm., which is about half the work done in the stretch, 18·6 g.cm. Again about half the work done in the stretch disappeared.

Four other reliable experiments were performed on early stretches during a twitch of frogs' sartorii at 0° C, starting about 50 msec. after a shock and ending well before the peak. The results, including those of the two experiments described already, are given in table 2. If the whole of the work done in the stretch were transformed into heat, and if the stretch had no other effect on the energy liberated by the muscle, we should expect $(H_s - H_i)$ to equal W. In fact, however, $(H_s - H_i)$ is always less than W; with rather a wide scatter, the average value of $(H_s - H_i)/W$

is 0·6. Thus, about 40% of the work done in the stretch, on the average, disappeared. We tried in every way to increase the fraction of work absorbed. The magnitude and speed of the stretch, the moment at which it started relative to the stimulus, the initial length of the muscle, could all be varied, but no means was found by which more than about half the work was absorbed.
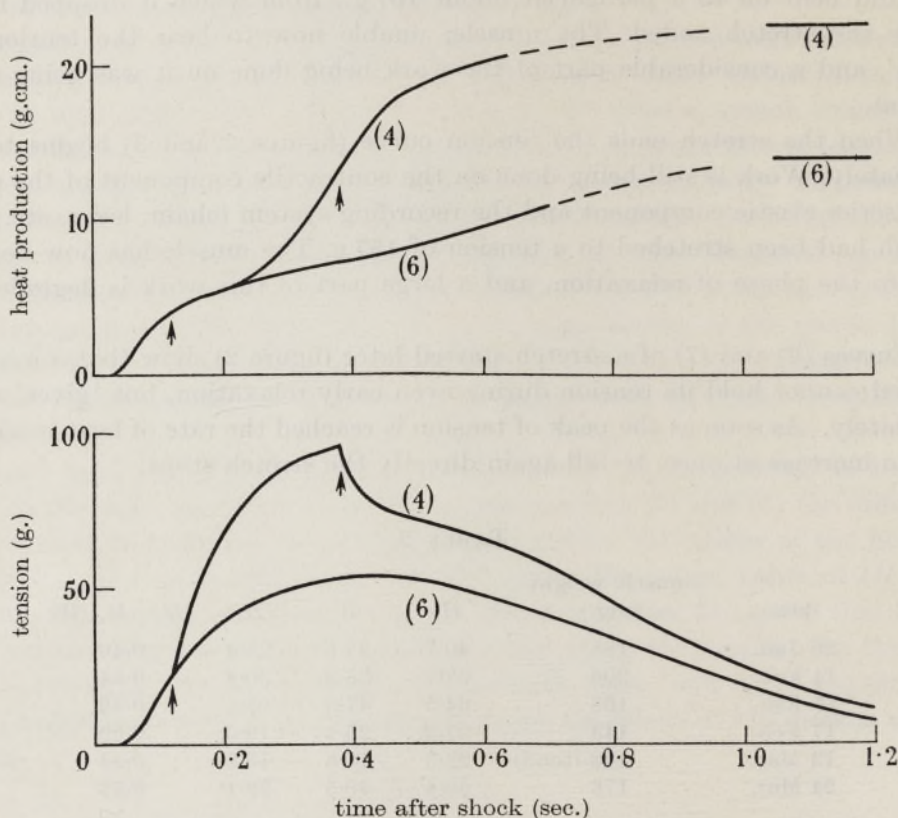

FIGURE 4. Stretch during twitch. Experiment of 13 March 1950. Toad's sartorii, standard length 24 mm., 102 mg., on thermopile D_1 at 0° C. Preliminary soaking in Ringer, then three contractions, then soaked again in Ringer, then records (4), (5) and (6) (record (5) not shown). Record (4), stretch $2\frac{1}{2}$ mm., $22\frac{1}{2}$ to 25 mm., 120 to 380 msec., heat 22·5 g.cm., work 18·6 g.cm.; tension at start of stretch 23 g., at end 96 g. Record (5), isometric long (25 mm.), tension 62 g., heat 13·2 g.cm. Record (6) isometric short ($22\frac{1}{2}$ mm.). tension 54 g., heat 13·8 g.cm. The muscle was very strong ($Tl_0/M = 1400$ and 1200 in the two isometric twitches). The heat curves were corrected for heat loss, the final values being shown at the top right. Beginning and end of stretch shown by arrows.

In the last five experiments tabulated above later stretches also were applied. Taking account only of those which started not earlier than 225 msec. after the shock (for the frogs) or 450 msec. (for the toad), which are near the peaks of the isometric twitches, the average values of $(H_s - H_i)/W$ were, in order, 0·95, 1·08, 1·10, 0·77, 0·78; mean 0·94. While about 40% of the work done in a stretch during the active stage of contraction disappeared, in a stretch during relaxation practically the whole of the work reappeared as heat. Intermediate values were found for stretches which came partly in each phase.

There are obvious reasons why a greater fraction of the work cannot be absorbed during a twitch: (a) If a stretch of sufficient magnitude is to be applied during the short interval between the beginning of contraction and the beginning of relaxation it has to be very rapid, far more rapid than the maximum rate of 'reversible' lengthening; parts of the muscle 'give', and the work done in stretching them is turned into heat. (b) When the stretch ends the tension is very high and a considerable part of the work done is carried over as mechanical potential energy in the series elastic elements of muscle or recording system. Some of this potential energy is absorbed by the contractile elements as those slowly lengthen and the tension falls, but relaxation soon sets in and all the mechanical energy remaining is now dissipated as heat.

Both difficulties can to some degree be avoided by working with fairly short tetanic contractions instead of single twitches; these allow slower stretches to be applied, and permit them to end before relaxation begins. There is a further reason for using tetanic contractions for this purpose; one can avoid the following logical difficulty in the interpretation of the results. We have assumed hitherto that work not absorbed is simply added to the heat which would have been produced had no stretch taken place. That, however, requires that the energy liberated by the muscle in consequence of stimulation should not itself be affected by stretching. In principle this assumption is clearly untrue. Stretching a muscle early during an isometric twitch, by raising the tension, diminishes and finally prevents altogether the internal shortening of the contractile component against the series elastic component of tendon and recording system. Not only, therefore, is heat of shortening reduced (see figures 2 and 3 above and figure 5 below), but also the work done by the contractile component in stretching the series elastic bodies is diminished. This work, like other work, is presumably turned into heat in relaxation. Thus, as the result of a stretch, the total energy given out by the muscle itself is reduced. It is possible to allow for this (see below), and calculation shows that the correction is not so large as seriously to affect the main conclusion; but an allowance of this kind is rather uncertain, and it is better, if possible, to obviate its necessity.

The difficulty can be avoided by stretching a muscle, during a tetanus, after it has developed its full isometric tension. The internal shortening of the contractile component has then been completed before the stretch begins, so the stretch cannot reduce either the heat of shortening or the internal mechanical work. In the absence of any other known effect of a stretch on the energy liberated by a stimulated muscle it is then logically permissible to subtract H_i from H_s and compare the difference with W. We shall see below that the result is the same as before.

Many experiments were made on tetanic contractions by the methods described above; in some of these the stretch started early and the results were similar to those with twitches. Such an experiment on a very strong muscle is shown in figure 5. The stretch was much slower than was possible in a twitch, and ended at about the same moment as the last stimulus. The value of $(H_s - H_i)$ was 20·8 g.cm., W was 55·4 g.cm., so that $(H_s - H_i)/W = 0·38$; thus about 62 % of the work done in the stretch did not reappear. In fifteen such experiments on frogs'

muscles at $0°$ C, with 3 to 4 mm. stretches starting usually about 100 msec. after the beginning of the stimulus and finishing by the time it ended, and with stimuli lasting from 0·5 to 2 sec., the following values were obtained for $(H_s - H_i)/W$:

0·38, 0·51, 0·45, 0·32, 0·61, 0·75, 0·47, 0·60,
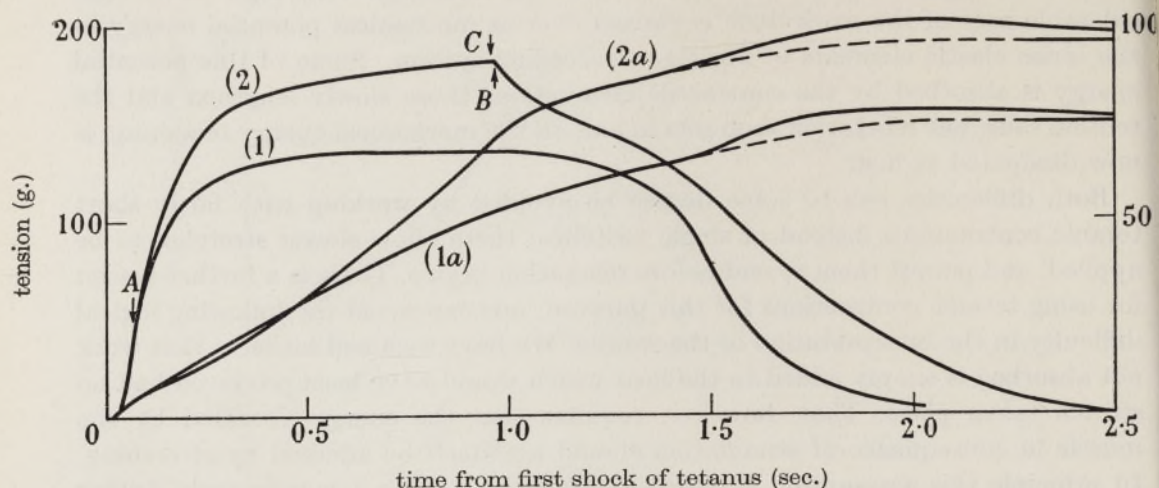0·51, 0·67, 0·41, 0·34, 0·54, 0·69, 0·49; mean 0·52.

FIGURE 5. Stretch during a short tetanus at $0°$ C. Experiment of 28 March 1950. Frog's sartorii, 150 mg., standard length 28·5 mm., on thermopile D_1. Stimulus, alternating condenser discharges, 12 shocks one way, 13 shocks the other, last shock (C) 0·955 sec. after the first (at time zero). Stretch $3\frac{1}{2}$ mm., $25\frac{1}{2}$ to 29 mm., starting (A) 64 msec. after the first shock, ending (B) 10 msec. after the last shock. There was no significant tension at rest at either length. Curve (1) tension, curve ($1a$) heat, in isometric contraction at $25\frac{1}{2}$ mm.; maximum tension 135 g., total heat 74·7 g.cm. after relaxation. Later in the experiment two consecutive records of isometric short ($25\frac{1}{2}$ mm.) and isometric long (29 mm.) gave nearly identical heat and tension. The muscle was very strong ($Tl_0/M = 2550$ in an isometric tetanus). Curve (2) tension, curve ($2a$) heat, during a stretch between A and B; final tension 135 g. ($Tl_0/M = 3450$), total heat 95·5 g.cm.; work 55·4 g.cm. The heat curves have been corrected for heat loss throughout (3·23 % per sec.). The final levels of the heat are shown by the ends of the broken lines. The small humps observed on the heat curves between 1·5 and 2 sec. were due to slight inequalities in the heat distribution, caused by unequal rates of relaxation in different regions of the muscles' cross-section; they even out and the levels corrected for heat loss became constant by 3·5 sec. The broken lines show what the curves should have been had the heat been produced evenly throughout the muscles.

From this series no valid result is omitted. The mean value 0·52 of $(H_s - H_i)/W$ corresponds to 48 % of the work absorbed; in seven out of the fifteen experiments the mean was 0·41, corresponding to 59 % of the work absorbed. The absorption of work was higher than in the twitches, because the stretches were less rapid and started later, and there was no risk of their running into the early stage of relaxation.

It is possible to make an approximate allowance for the amount of heat and work which a muscle is prevented, by a stretch, from producing. We apply the characteristic equation (Hill 1938), $(P+a)v = (P_0 - P)b$, which may be written

$$(P+a)\,dx = (P_0 - P)\,b\,dt,$$

where dx is the small amount of shortening in the short interval dt. On integrating this between any limits the left-hand side is the sum of work and heat of shortening, the right-hand side is b times the area of the tension-time curve below P_0. We need to know b, but b can be taken as about $\frac{1}{4}v_0$, where v_0 is the velocity of shortening under zero load. In frogs' sartorii at $0°$ C v_0 is not far from $1.7\ l_0$ sec. (Hill 1951). This is near enough for the present purpose and the calculation is simple.
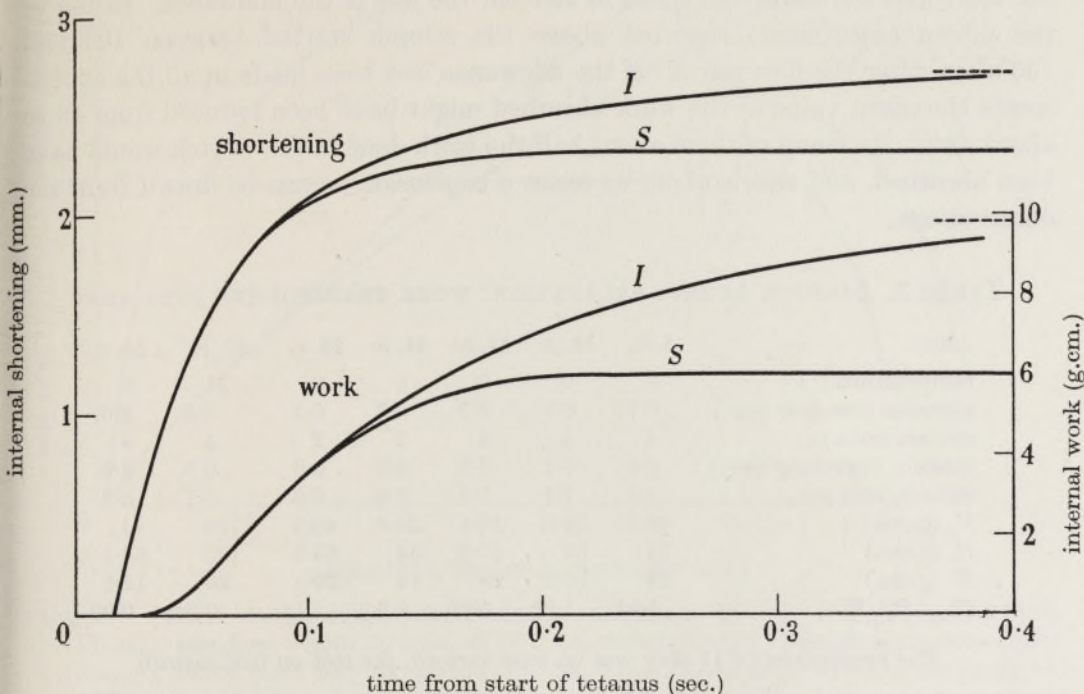

FIGURE 6. Internal shortening and internal work calculated from the tension curves of figure 5, as described in the text. *I*, isometric contraction; *S*, stretch. The final levels of the two curves *I* are shown by the broken lines at the right.

The amount of shortening, and the work, can be separately calculated if desired (taking $a = \frac{1}{4}P_0$), and compared for the cases of 'stretch' and 'no stretch'. This has been done for the experiment of figure 5 and the results are shown in figure 6. Soon after the stretch begins curves *S* (for stretch) begin to drop below curves *I* (for isometric), because of the increase of tension, and cease to rise any further as the tension approaches P_0. The total amount of shortening prevented by the stretch is 0.40 mm., corresponding to a heat of shortening prevented of 1.4 g.cm. The work prevented is 3.8 g.cm. The sum of these is 5.2 g.cm. which should be subtracted from H_i (74.7 g.cm.) for comparison with H_s (95.5 g.cm.). Thus the corrected value of $(H_s - H_i)/W$ is $(95.5 - 69.5)/55.4 = 0.47$, which corresponds to 53% of the work being absorbed instead of 62%.

In another experiment a similar calculation was made. During a 0.75 sec. tetanus a stretch of 4 mm. was applied to a muscle at $0°$ starting 0.11 sec. after the first shock and ending 0.04 sec. before the last one. H_s was 81.1 g.cm., H_i 58.5 g.cm., and W 67 g.cm. The value of $(H_s - H_i)/W$ is 0.34. Numerical integration

showed that the internal work prevented by the stretch was 3·1 g.cm., the heat of shortening prevented 1·3 g.cm., total 4·4 g.cm. With a corrected value of $H_i = 54·1$ g.cm. the corrected ratio $(H_s - H_i)/W$ is 0·40; about 60% of the work, instead of 66%, was absorbed. In both cases the correction, although significant, does not affect the main conclusion.

The magnitude of the allowance for heat of shortening and work prevented by stretch depends on the moment at which stretch begins and on its speed. The later the start and the lower the speed of stretch, the less is the allowance. In ten of the fifteen experiments reported above the stretch started between 100 and 150 msec. after the first shock. If the allowance had been made in all the experiments the mean value of the work absorbed might have been reduced from 48 to about 43%. In many of them about half the work done in the stretch would have been absorbed, and this is about as exact a conclusion as can be drawn from the observations.

TABLE 3. STRETCH DURING RELAXATION: WORK TRANSFORMED INTO HEAT

date	1. iii	19. iv	21. iv	21. iv	25. iv	27. iv	15. v
temperature (° C)	0	0	0	0	0	11	0
stimulus duration (sec.)	0·75	0·7	0·7	0·7	0·5	0·5	2·0
stretch (mm.)	4	4	4	2	2	4	4
stretch, beginning (sec.)	0·6	0·7	0·8	0·8	0·6	0·5	2·0
stretch, end (sec.)	1·1	1·1	1·2	1·0	0·8	0·7	3·5
H_i (g.cm.)	36·5	24·6	21·6	21·6	40·7	106	41
H_s (g.cm.)	71	55	45·5	34	63·7	125	53·1
W (g.cm.)	34	30·2	25	13	20·1	20	12·2
$(H_s - H_i)/W$	1·01	1·01	0·96	0·95	1·14	0·95	0·99

The experiment of 15 May was on toad sartorii, the rest on frog sartorii.

In six experiments stretches were applied during relaxation. Details are given in table 3. The average of seven values of $(H_s - H_i)/W$ is almost exactly unity, showing that when work is done in stretching a muscle during relaxation it is transformed quantitatively into heat.

It should be remarked that one of the stretches started slightly before the stimulus ended, while three others began at exactly that moment; this needs comment. In comparatively slow stretches such as these the first effect of a stretch is to increase the mechanical potential energy present in tendon and recording system; the muscle does not begin to 'give' at once, and the reversible lengthening process is slow. The stored mechanical energy is later either absorbed or transformed into heat. In the experiments of table 3 the mechanical energy was clearly held over long enough for relaxation to set in, when it was all turned into heat.

It is possible to avoid altogether the necessity for allowing for shortening heat and work prevented by stretch, by applying the stretch only after the full isometric tension has been developed. Three experiments of this kind were made, of which one is shown in detail in figure 7. The stretch began about 0·2 sec. after the full tension was reached, continued for 2·8 sec. at 1·07 mm./sec., and ended 0·45 sec. before the last shock of the stimulus. The records of heat and tension for isometric

long and isometric short were nearly identical. The amount of work unaccounted for as heat was 42 %. In two similar experiments (1 June and 28 July) the work absorbed was 50 and 21 % respectively.
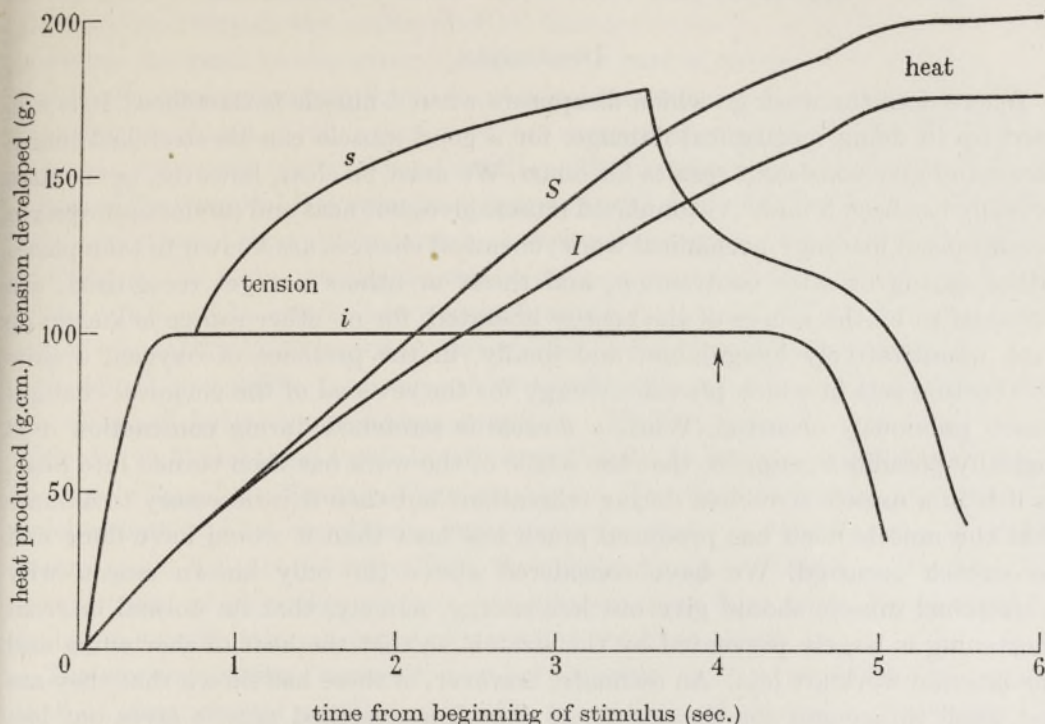

FIGURE 7. Stretch during a maintained tetanic contraction, 18 July 1950. Frog's sartorii, 192 mg., standard length 30 mm., at 0° C on thermopile D_2. Stimulus, condenser discharges, 10/sec. each way for 4·0 sec. Stretch 3 mm., from 28 to 31 mm., beginning at 0·75 sec. and ending at 3·55 sec. The heat curves, corrected for heat loss, are S for stretch, I for isometric short (28 mm.); the tension curves are s and i respectively. The curves for isometric long (31 mm.) are not shown, being nearly identical with those for isometric short. Total heat to end of relaxation, stretch 199 g.cm., isometric short 174 g.cm., isometric long 171'g.cm. Maintained tensions, isometric short 100 g., isometric long 105 g. Work done in stretch 45·9 g.cm. Taking H_i as the mean for short and long, $(H_s - H_i)/W = 0·58$; work absorbed 42 %. The arrow shows the end of stimulus.

If the characteristic equation could be applied to lengthening as well as to shortening, the muscle would in theory resist with an infinite force a stretch at velocity b; which in frogs' sartorii at 0° C would be about 12 mm./sec. The stretches applied in the twitches described above were rather faster than this, and no possibility exists of reversible lengthening at such speeds. Abbott & Aubert (1951) describe experiments with very slow stretches, in which lengthening seemed to be completely reversible; the force used in the stretch was not usually greater than 1·5 P_0, for which, from the equation, the rate of stretch should be 0·28b or about $3\frac{1}{2}$ mm./sec. Actually, the observed velocity of stretch was much less than this, in keeping with Katz's conclusion (1939) that the observed velocity of isotonic lengthening, for a load in excess of the full isometric tension, is considerably less than calculated from the characteristic equation. The matter is considered in

detail by Abbott & Aubert; it is mentioned here to show how slow 'reversible' lengthenings have to be, so slow that none of the stretches described in this paper fall nearly within that category. It is striking, therefore, that even at these much higher speeds a substantial part of the work was in fact absorbed.

DISCUSSION

Where does the work go which disappears when a muscle is stretched? It is not used up in doing mechanical damage, for a good muscle can be stretched many times and give consistent results for hours. We must be clear, however, as to what actually has been found. A stimulated muscle gives out heat and (under appropriate conditions of loading) mechanical work; chemical changes are known to take place, either during or after contraction, and these, or others not yet recognized, are assumed to be the source of the energy liberated, for no other source is known or even quantitatively imaginable; and finally, in the presence of oxygen, a slow combustion sets in which provides energy for the reversal of the chemical changes which previously occurred. When a muscle is stretched during contraction it is logically possible to suppose that the whole of the work has been turned into heat, as it is in a muscle stretched during relaxation; but then it is necessary to assume that the muscle itself has produced much less heat than it would have done had no stretch occurred. We have considered above the only known reason why a stretched muscle should give out less energy, namely, that its normal internal shortening is largely prevented by the stretch, so that the heat of shortening and the internal work are less. An estimate, however, of these has shown that they are too small to account for the deficit, so that if a stretched muscle gives out less total energy it must do so for reasons at present unknown.

The methods of thermodynamics cannot by themselves reveal the details of a mechanism, and without additional information of a more specific kind it is impossible to decide between the alternatives. So far as measurements of energy go, the muscle behaves *as though* a considerable fraction of the work done in the stretch was absorbed. This is merely the simplest way of expressing the facts, but other ways are possible. When a muscle shortens against a load and does mechanical work, the work appears as a separate term in the energy equation; presumably it is derived from the energy of chemical change. It is natural, therefore, to suppose that when work disappears it is taken up in the reversal of chemical processes, as the free energy provided by recovery oxidation is taken up. But it remains possible to suppose that the work is wholly degraded into heat, and that the apparent absorption is due to the stretch somehow preventing the occurrence of, not merely reversing later, the chemical changes which normally follow stimulation. Indeed, three possibilities exist: (a) that the work is absorbed in driving backwards chemical processes which have actually occurred as a normal part of contraction; (b) that the work is absorbed in some other chemical or physical process at present unknown; and (c) that the work is wholly degraded into heat, but that chemical processes normally occurring in contraction are prevented by the stretch. Without specific evidence such as thermodynamics cannot provide it is impossible logically to decide between such hypotheses.

We have adopted the simplest way of expressing the facts, namely, that during a stretch the work is absorbed in reversing chemical processes which normally provide work when the muscle is able to shorten; the conditions of 'reversibility' in the stricter thermodynamic sense apply only, if at all, to those stretches that are conducted very slowly, as described by Abbott & Aubert (1951). In the more rapid stretches discussed in the present paper only a part of the work is absorbed. This sort of relation is commonly found in practical processes by which chemical energy is liberated or stored. Only when they are conducted very slowly is the thermo-dynamic efficiency high; if conducted rapidly in the forward direction merely a part of the chemical free energy appears as useful work, in the backward direction only a fraction of the applied work is stored as free energy. Looking at the present problem in this way, one might even be rather astonished that so large a fraction of the work can be retained in a stretch which is so much faster than one which might reasonably be regarded as 'reversible' in the thermodynamic sense.

It would be possible to discuss these results in terms of current hypotheses of the chemical basis of muscular contraction. Pryor (1950), for example, sees no difficulty in fitting them to his kinetic hypothesis of adenosine triphosphate acting in muscle as a 'plasticizer' of protein chains. The present results, however, are independent of any hypothesis, and to discuss them in relation to any special theory might distract attention from their factual nature. Chemical theories must take account of physiological facts, but physiological facts might be obscured by trying to express them in terms of a specific chemical theory.

The present results, however, can be discussed from a very different aspect, that of their general application to the physiology of muscular movement. In ordinary life every movement of skeletal muscles involves the lengthening of some, the shortening of others. The lengthening need not be forcible but it frequently is, as in lowering a heavy weight or the body itself. A large amount of mechanical energy is taken up by a gymnast's arms when he uses them to lower himself on a rope. Is this energy dissipated entirely as heat, or is some of it absorbed by the muscles?

In practical life it can seldom happen that a muscle is forcibly stretched as slowly as appears to be necessary if the whole of the work is to be absorbed. Everyday movements in which weights, or the body, are lowered, or the muscles yield to superior force, come rather in the class of contractions investigated in this paper, in which large amounts of work have to be got rid of in contractions of limited duration. If so, we might expect that a substantial part of the work would be dissipated in the muscles as heat, but some of it retained and absorbed. The problem, however, is not necessarily as simple as this. The maintained contrac-tions investigated in this paper have all been produced by stimuli of high enough frequency to give a complete tetanus. With a lower frequency and an incomplete tetanus the muscle fibres would alternate between full activity and a state of partial relaxation, and during the latter the work done in stretching them would appear mainly as heat. Only when the muscles were resisting very strongly in a complete tetanus could we confidently expect them to absorb a substantial amount of work in yielding to superior force.

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Changes of energy in a muscle during very slow stretches

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When a muscle, during a maintained isometric contraction, is suddenly subjected to a load between 1·2 and 1·5 times its full isometric tension, the immediate effect is a brief stretch, expressing readjustment of the elastic parts of the muscle and lever system to the new tension. This is followed by a gradual fall of the load, progressively decreasing in speed, accompanying a slow lengthening of the contractile part of the muscle. Although a substantial amount of work is done by the load in falling, the heat production of the muscle is actually diminished; thus during the slow lengthening the whole of the work disappears. During a single stretch the heat deficit is proportional throughout to the amount of lengthening; the 'heat of lengthening' is negative. The constant of proportion is less, in general, the greater the load. The reason for this is considered.

The tension during a rapid stretch at constant speed (as described by Abbott, Aubert & Hill 1951) rises much higher than the full isometric tension. As soon as the stretch ends the tension begins to fall, the elastic component shortening and stretching the contractile component. If the stimulus is continued this 'after-stretch' takes place very slowly; as in isotonic stretches work is absorbed and a heat deficit occurs.

Whenever any part of a stretch occurs during relaxation, all the work performed during that period reappears as heat.

These results are consistent with reversal of the physical and chemical events associated with shortening, and the conditions necessary for thermodynamic reversibility are considered. An analogy with the cold-drawing of an extruded thread of a long chain polymer is discussed. Other possible hypotheses described in the preceding paper are referred to.

In the preceding paper (Abbott, Aubert & Hill 1951), experiments were described in which stretches at constant speed were applied to muscles during a twitch or a short tetanus. Although the stretches were fast—in order to produce large and decisive energy changes—much of the work was absorbed, which suggests that the mechanism producing the work is partially reversible. There are several disadvantages to this method, however: some work may be dissipated as heat through