

THE INFLUENCE OF DIAMETER OF MEDULLATED NERVE FIBRES OF CATS ON THE RISING AND FALLING PHASES OF THE SPIKE AND ITS RECOVERY

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SUMMARY

1. The relation of conduction velocity, i.e. fibre diameter (Hursh, 1939*a*) to certain temporal dimensions of the nerve impulse recorded monophasically was studied in medullated fibres of cats *in vivo* at temperatures mostly ranging between 21 and 37° C.

2. Contrary to existing belief, it was demonstrated unequivocally that spike duration varies inversely with the conduction velocities of the fibres; so also the durations of the rising and the falling phases (rise-time and fall-time) of the impulse. The fall-time is linearly related to conduction velocity at all recorded temperatures. The rise-time varies steeply with conduction velocity at the lower levels of conduction rate, and very gradually at the higher conduction rates.

3. The spike duration of preganglionic sympathetic fibres is identical with that of somatic medullated fibres with similar conduction velocities. There is therefore little justification for classifying them separately as so-called B fibres.

4. The rate of recovery of spike amplitude following a preceding impulse also varies inversely with conduction velocity, and in the same manner as the absolute refractory period (ARP). In fact the relation of time for 40% recovery of spike amplitude to conduction velocity is identical with the relation of conduction velocity to ARP. The Q_{10} for 40% recovery of spike amplitude is 4.7 between 13 and 29° C.

5. Rise-time increases exponentially with fall in temperature in all medullated fibres, fast (say 64 m/sec) and slow (say 16 m/sec), the Q_{10} being 2.5. Fall-time varies exponentially with temperature in slow fibres ($Q_{10} = 3.5$); in fast fibres it varies linearly. The Q_{10} for spike duration is the same in all fibres between 27 and 37° C only, its value being about 3.4. Below 27° C the Q_{10} depends on the conduction velocity of the fibres. Only in slow fibres does spike duration tend to vary exponentially with temperature.

6. Only abortive spikes are generated during the interval between the end of a preceding spike and the end of the ARP which is about $1\frac{1}{2}$ times spike duration in fast fibres and about twice spike duration in slow fibres.

INTRODUCTION

In a recent paper (Paintal, 1965) it was demonstrated that in medullated nerve fibres the absolute refractory period (ARP) varies inversely with the conduction velocities of nerve fibres. As pointed out in that paper, it therefore follows that there should be a similar relation between spike duration and conduction velocity, since the prevailing view, originating with Adrian (1921), is that the ARP lasts as long as the spike. However, this conclusion contrasts with the existing belief that spike duration is the same in all medullated nerve fibres regardless of their conduction velocities, a belief that is based exclusively on one paper published by Gasser & Grundfest (1939). In that paper, Gasser & Grundfest made certain extrapolations in their records of the compound action potential of the saphenous nerve and from the results obtained concluded that the spike durations of all medullated fibres were the same. They supported this conclusion by recording impulses in single nerve fibres which as stated by them '... They all appear alike in duration and the measurements all fall between 0.4 and 0.5 msec, without a systematic difference with respect to velocity. It would be difficult to measure them closer than to 0.1 msec'.

Indeed, as shown in the present paper, at about 37–38° C which must have been the temperature at which they recorded their impulses (cf. Grundfest, 1939), this statement is quite true if one approximates the measurements to the first decimal place. However, the present results also show beyond doubt that if the measurements are approximated to the second or third decimal place, there exists a clearly demonstrable inverse relation between spike duration and conduction velocity, i.e. fibre diameter (Hursh, 1939*a*), a relation that becomes more obvious and unequivocal at lower temperatures.

METHODS

All the experiments were carried out on adult cats anaesthetized with chloralose (75 mg/kg). Impulses in individual fibres were recorded from thin filaments dissected from the vagus and saphenous nerves; a few observations were also made on fibres of the cervical sympathetic. The filaments were dissected as described earlier (Paintal, 1953). As done by Gasser & Grundfest (1939), the impulses were recorded monophasically under paraffin by placing the cut injured end of the filament on the distal recording electrode. The high frequency response of the whole recording set-up, i.e. recording electrodes, preamplifier and amplifier was about 37 kc (Fig. 1*A*); the low frequency response was 0.06 c/s. It is therefore certain that since the minimum rise time of the impulses recorded was 0.036 msec and their maximum duration at low temperatures was about 15 msec, that these impulses were recorded without any distortion. The distance between the recording electrodes (silver-silver chloride) was 1 mm and their thickness, 0.2 mm.

Apart from exceptional cases, the fact that an impulse is all or none is a satisfactory criterion that it originates in a single fibre. This criterion was therefore used in the present investigation. However, in order to take care of the exceptional case in which two fibres may have almost exactly the same conduction velocity and exactly the same threshold, the contour of the spike was always carefully examined for any inflexions making either the rising or the falling phase of the impulse non-uniform; if there was an inflexion the fibre was ignored. This had to be done in only a few fibres.

For measurements of spike duration, the nerve impulses were recorded on as fast a sweep as possible and the duration measured with a calibrated magnifier ($\times 12$). In order to eliminate any guessing about the end of the spike, a difficulty experienced by earlier workers (e.g. Blair & Erlanger, 1933), a straight line was drawn over the down-slope of the mono-

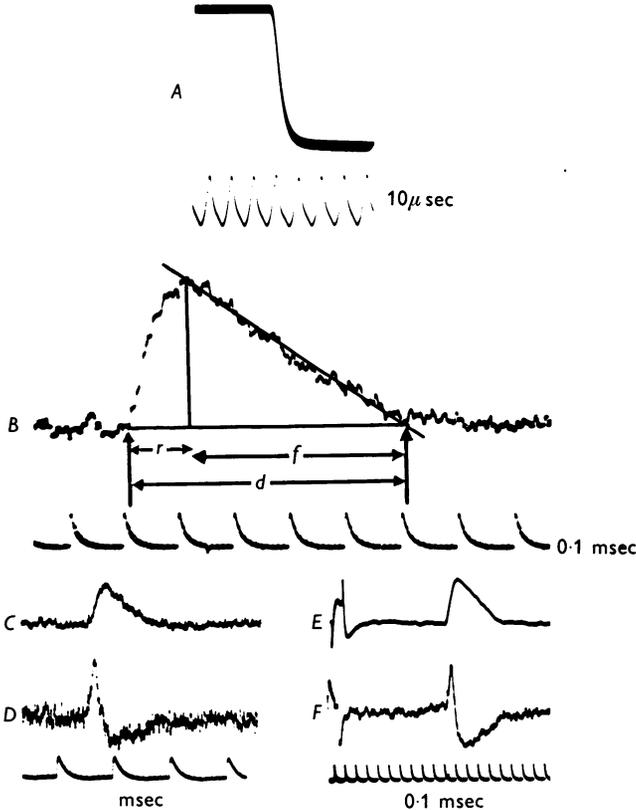


Fig. 1. Records showing that the recording set-up provided a faithful record of monophasic impulses that are identical in duration, rise-time and fall-time to those recorded in uninjured nerve fibres. *A*, shows the frequency response of the recording set-up. *B*, the procedure used for measuring spike duration (*d*), and duration of the rising (*r*) and falling phase (*f*) of the impulse. *C* shows a monophasic impulse in a slow fibre with a normal conduction velocity of 9 m/sec. *D* shows a diphasic impulse in an uninjured stretch of the same fibre at the same temperature (33° C) recorded at an amplification $2\frac{1}{2}$ times greater than that in *C*. *E* and *F* are similar records at 34° C obtained from a fast fibre with a normal conduction velocity of 70 m/sec; amplification in *F* is 4 times greater than that in *E*. Time marks for *C* and *D*, 1 msec; those for *E* and *F*, 0.1 msec.

phasic spike and the duration reckoned as the interval between the beginning of the spike (which was easy to define) to the point where the superimposed straight line cut the base line (Fig. 1*B*). The difference between any two measurements of the same spike duration was less than 2% of the total spike duration. However, only one measurement of each spike was made, because several impulses of each fibre were recorded so that the recorded value of the spike duration of a fibre represents the average value obtained from several impulses, usually 10 (range 5 to 16). This procedure therefore helps to rule out random errors arising from variation in the noise level of the amplifier, or from the appearance of small stray potentials influencing the rising or falling phases of the impulse.

The duration of the rising phase of the impulse (rise-time) was measured from the start to the peak of the impulse; the duration of the falling phase (fall-time) was obtained by subtracting the rise-time from the total duration of the impulse (Fig. 1*B*). As in the case of spike duration each recorded value of rise-time or fall-time represents the average value obtained from about ten impulses.

Following a suggestion by Dr B. Katz (personal communications) the monophasic impulses of a few fast and slowly conducting fibres were compared with the diphasic impulses recorded over an uninjured stretch of the same fibres. For diphasic recording an interelectrode distance of about 0.1 mm was used as described by Katz & Schmitt (1940). In most fibres it was found that spike duration, rise-time [indicated by the interval between the start of the diphasic impulse and the moment its first phase crosses the base line (cf. Schmitt, 1939)] and fall time of the diphasic impulse were roughly similar to the corresponding intervals measured in the monophasic records (Fig. 1*C-F*), but in some fibres they were shorter and in others longer. This applied to both fast and slow fibres, showing thereby that the method of monophasic recording employed had no differential effect on spike duration, rise-time and fall-time of fast and slowly conducting fibres respectively. As expected, the size of the diphasic impulse was much less than that of the monophasic impulse; in fact, these relative sizes of the two depended on the conduction velocity of the fibre (Fig. 1) (cf. Hill, 1934). From these observations, it may be concluded that the method of recording monophasic impulses provided a faithful record of the impulses in uninjured nerve fibres, at least as far as the intervals measured were concerned.

The crystal controlled time maker was calibrated against a 1 kc frequency standard broadcast by the National Physical Laboratory, New Delhi.

The temperature of the nerve filament was always measured. In a few experiments, it was measured with a thermocouple and in the remainder with a thermistor and a suitable Wheatstone bridge circuit. The null method was used since temperatures were recorded only when they were stable. The temperatures could be read accurately to 0.1° C. The tip of the thermistor (type F 2311/300 made by Standard Telephone and Cables, Ltd), which was about 1 mm in diameter including the thin glass case, was kept in contact with the filament but a portion of its surface was in contact with liquid paraffin. Under these conditions, if the body temperature is very different from that of the paraffin, the temperature recorded must represent some sort of an average temperature of the nerve and the liquid paraffin. This state of affairs obtained only in experiments in which measurements of recovery of spike height were made (Fig. 9). In these experiments cold liquid paraffin was poured into the recording pool continuously (the excess being sucked away), so as to lower the temperature of the nerve filament on the recording electrodes to say 20° C, while the temperature of the rest of the nerve between the stimulating and recording electrodes was at body temperature (Fig. 9*A*). Therefore, in order to get an estimate of the actual temperature of the filament, in one experiment the temperature was recorded in the usual way and also by surrounding the whole tip of the thermistor with a thick nerve filament. It was found that the difference between the two readings was less than 1° C at a time when the temperature recorded in the usual way was 20° C and the temperature of the rest of the vagus was 36° C. In those experiments therefore, in which the recovery of spike amplitude was studied at a recorded tem-

perature of about 19.5° C (average of all experiments), 1° C was added to the reading in each case

In all the other experiments, i.e. those concerning measurement of spike duration, rise-time or fall-time, no such allowance was necessary because in these experiments it was ensured that body temperature, paraffin pool temperature and ambient temperature were nearly all the same. For example, while recording the impulse at about 21.5° C, the body temperature was kept at 21.5° C, and so also was the ambient temperature and that of the liquid paraffin in the recording pool.

The thermometer against which the thermistor was calibrated was itself calibrated against a standard thermometer of the National Physical Laboratory, New Delhi.

The conduction velocity was determined from measurements of the shock response time as described before (Paintal, 1953). At temperatures higher or lower than 37° C, the conduction velocity so determined was corrected to 37° C using the appropriate temperature coefficients, e.g. a Q_{10} of 1.6 for temperatures between 27 and 37° C (Paintal, 1965).

Several sources of errors give misleading information about spike amplitude with the kind of recording arrangement used in the present investigation (Tasaki, 1949; Tasaki & Spyropoulos, 1957). Therefore, no measurements of spike height *per se* were made. However, all these errors are common to all the spikes of the same fibre recorded a few milliseconds apart. Thus there are not likely to be any errors worth considering in a study of the relative spike amplitude of a second impulse relative to that of the first, recorded a few milliseconds after the latter. The present results, therefore, relating to the recovery of spike amplitude of the second impulse may be regarded as being free from errors.

Except in the case of recovery of spike amplitude, the Q_{10} was determined as done previously, i.e. from measurements 10° C apart (Paintal, 1965). In the case of recovery of spike amplitude, it was determined from two or more observations about 5–6° C apart using the appropriate logarithmic formula $10(\log x_{t_1} - \log x_{t_2})/(t_1 - t_2)$, where x_{t_1} and x_{t_2} are the intervals for 40% recovery of spike amplitude (after a preceding impulse) at temperatures t_1 and t_2 .

RESULTS

The monophasic spikes recorded in vagal and saphenous nerve fibres at different temperatures were similar to those recorded by Gasser & Grundfest (1939) with a quick rise and a more gradual fall and they showed no evidence of diphasicity (e.g. Fig. 1*B, C, E*).

Relation of spike duration to conduction velocity. Figures 2 and 3 show the relation of spike duration to conduction velocity recorded at four different temperatures. That at 37.1° C (Fig. 2*B*) relates to fibres of the saphenous nerve obtained from three cats; those at the remaining temperatures to vagal fibres (Fig. 2*A* and Fig. 3). In the case of vagal fibres all the measurements were made in the same cat at a particular temperature which varied by less than 0.5° C from one fibre to the other. However, even slight variations from the mean temperature of the experiment (e.g. 32.9° C in the case of Fig. 2*A*) were taken into account by making a correction using a Q_{10} of 3.5 (Inman & Peruzzi, 1961).

The results show conclusively that at all temperatures the spike durations of the fibres vary inversely in a systematic manner with the conduction velocities of the fibres, but the actual relation between the two is

not exactly the same at all temperatures (compare curve at 21.5° C in Fig. 3 with that at 37.1° C in Fig. 2*B*).

A noteworthy feature of the spike durations at 37.1° C is the considerable scatter of the points which is not due to variations in temperature. The results confirm the observations of Gasser & Grundfest (1939) that in general the spike durations of medullated fibres lie between 0.4 and

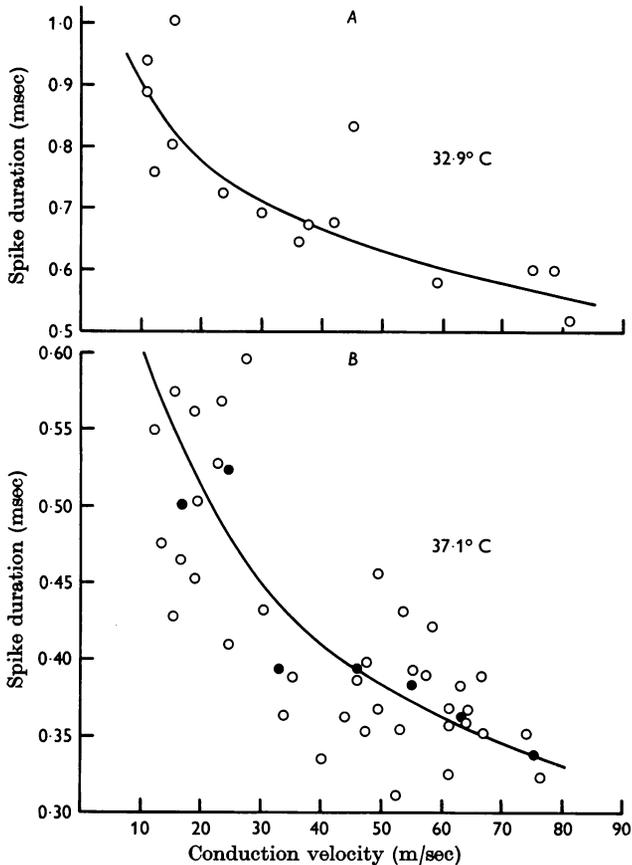


Fig. 2. Relation of spike duration to conduction velocity. The observations in *A* were obtained in one cat from vagal fibres at a temperature of about 32.9° C. Those in *B* from saphenous fibres from three cats at a temperature of 37.1° C. The filled circles in *B* represent the mean spike durations in the different ranges of conduction velocity; the curve has been drawn by eye to fit these points as closely as possible.

0.5 msec if the values are approximated to the first decimal place. As shown in Fig. 2*B* the spike durations of some fibres were much smaller than those reported so far, e.g. 0.32 msec. This is to be expected con-

sidering the degree of scatter observed (Fig. 2*B*). This scatter becomes smaller as the temperature becomes lower (Figs. 2, 3).

All the curves in Figs. 2 and 3 were drawn by eye so as to fit the points as closely as possible.

The spike durations at 21.5° C (Fig. 3) show that there is a large difference between the spike durations of fast and slow fibres. That this

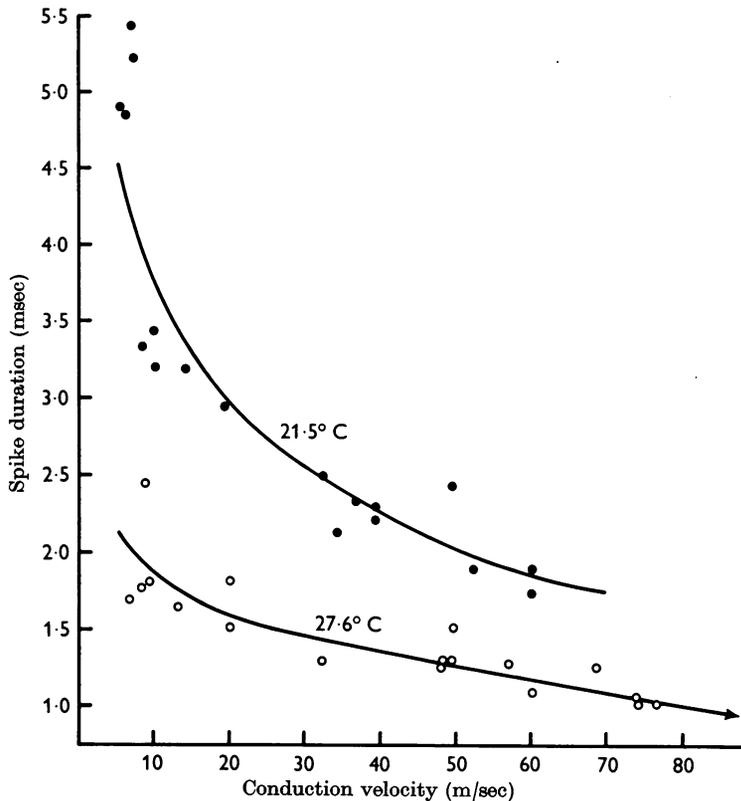


Fig. 3. Relation of spike durations of vagal nerve fibres to their conduction velocities recorded in two cats at two different temperatures, filled circles at 21.5° C and open circles at 27.6° C. The body temperature was maintained at the same temperature as the temperature of the nerve fibres.

difference is real and free from artifacts is clear from the following consideration: Tasaki has shown that a second spike cannot be initiated before the end of the preceding one (Tasaki, 1949). Therefore, since a small spike could be initiated at the end of the preceding one in the fast fibres (Fig. 9), there can be no doubt that the relatively small spike duration of the fast fibre is not due to an artificial abbreviation of the spike. Conversely, since it has never been possible to initiate a second spike before the end of the

relatively longer duration spikes of the slower fibres, it follows that the larger spike durations of these fibres (Fig. 3) have not been artificially exaggerated.

The effect of temperature on spike duration was studied by plotting the relation of spike duration to temperature at different levels of conduction velocity 10 m/sec apart or less. Some of these are shown in Fig. 4. The data for all the curves of Fig. 4 were obtained from the four original curves shown in Figs. 2 and 3. There is no doubt that at every level of conduction velocity (including those not shown in Fig. 4), the slope of the curves between 27 and 37° C is almost the same, the Q_{10} being about 3.4.

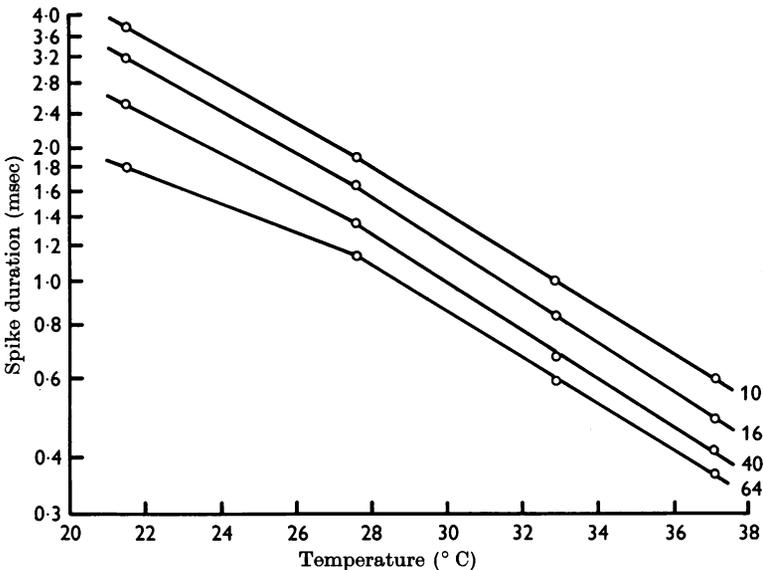


Fig. 4. Graphs showing the relation of spike duration to temperature in fibres of different conduction velocities indicated on the right. These results were obtained from the curves shown in Figs. 2 and 3. The slope of the curves between 37 and 27° C is the same in slow and fast fibres; at lower temperatures it is different. Spike duration varies logarithmically with temperature in the slowest fibres (10 m/sec.). The ordinate scale is logarithmic.

This agrees with the value of 3.5 reported by Inman & Peruzzi in their normal preparation and with that for frog fibres between 5 and 20° C (Tasaki & Fujita, 1948). In the case of the squid axon the Q_{10} is 3.2 between 10 and 20° C (Hodgkin & Katz, 1949).

As shown in Fig. 4, below 27° C the slope is different in fibres of different conduction velocities. Thus, whereas the slope below 27° C is almost the same as that above 27° C in the slowly conducting fibres (10 and 16 m/sec), it is clearly much less in the faster fibres (64 m/sec). This

behaviour is due to the fact that the relation between temperature and fall-time of the spike is markedly different in fast and slow fibres (cf. below).

In the case of the slowly conducting fibres (10 and 16 m/sec (Fig. 4)) a common linear regression line could be drawn, thus showing that for practical purposes the relation between temperature and spike duration is logarithmic between 20 and 37° C. This, as shown below, is due to the fact that in the slower fibres the relation between temperature and fall-time (and also rise-time) is logarithmic.

Relative spike durations of functionally different fibres. The spike durations of fibres of the cervical sympathetic nerve (B fibres) were compared with those of the vagus and the saphenous. Since the conduction velocities of so-called B fibres is less than 14 m/sec (Grundfest, 1939), fibres with conduction velocities ranging from 10 to 20 m/sec in the three nerves (vagus, saphenous and sympathetic) were selected for comparison with one another as far as their spike durations were concerned. All the measurements were made at about 33·6° C and corrections for small deviations in temperature made as described in Methods.

Spike durations of twelve sympathetic fibres with conduction velocities less than 14 m/sec were recorded. The average conduction velocity of these fibres was 11 m/sec and at 33·6° C their average spike duration was 0·72 msec. Similarly, the mean conduction velocity of thirteen fibres of the saphenous was 17 m/sec; these had an average spike duration of 0·75 msec. The six fibres of the vagus with a mean conduction velocity of 16 m/sec had a mean spike duration of 0·75 msec. No information about variance can be provided. This could have been given only if fibres with exactly the same conduction velocity were studied. The results, however, are clear-cut and they demonstrate beyond doubt that the spike durations of sympathetic fibres do not differ from those of other medullated fibres. Therefore, at 37° C the spike duration of sympathetic fibres will range from 0·45 to 0·6 msec for fibres with conduction velocities of 10–14 m/sec (Fig. 2*B*); for the slower fibre it will be greater. In view of these results there is no need for putting the preganglionic fibres in a separate class.

Duration of rising and falling phases of the impulse. Figure 5 shows the relation of the duration of the rising phase (rise-time) of the impulse to conduction velocity at two different temperatures. Here too there appears to be a consistent relation between rise-time and conduction velocity. The significant feature of the curves is that there is a steep inverse relation between rise-time and conduction velocity below about 40 m/sec and that the curve flattens out beyond 40 m/sec (Fig. 5). However, at 32·9° C the scatter of the points for conduction velocities greater than 40 m/sec is too large for drawing definite conclusions; the nature of the trend, which is

clear-cut at 21.5° C, is obvious. Qualitatively similar results were reported earlier by Blair & Erlanger (1933), but their values for rise-time were rather large, due partly to the poor high frequency response of their amplifier. Perhaps for this reason their observations were ignored by subsequent investigators, including the authors themselves.

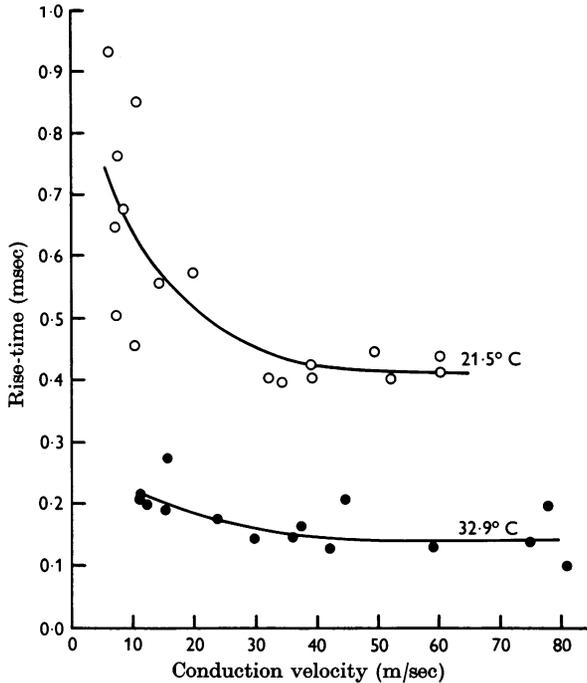


Fig. 5. Relation of rise-time (duration of rising phase) to conduction velocity of the fibres (vagal) at two temperatures, 21.5 and 32.9° C.

In contrast to rise-time, the relation between the duration of the falling phase (fall-time) and conduction velocity is linear (Fig. 7). The correlation coefficient between conduction velocity and fall-time is -0.80 at 32.9° C; at 27.6° C it is -0.88 and at 21.5° C it is -0.93 for fibres with conduction velocities greater than 10 m/sec. The straight lines in Fig. 7 were drawn from regression equations.

Figure 6 shows the relation of rise time to temperature in slowly (16 m/sec) and rapidly (64 m/sec) conducting fibres. From these results one can be certain that rise-time decreases exponentially with temperature in slow and fast conducting fibres alike. The Q_{10} for rise-time is also the same in both, being 2.5. This value is in reasonable agreement with those of Hodgkin & Katz (1949) found in the squid axon and those of Inman & Peruzzi (1961) found in the fibres of the Pacinian corpuscle of the cat.

In contrast to rise-time, the relation of fall-time to temperature varies in fibres of different conduction velocities; in slowly conducting fibres the relation between the two is exponential, while in fast fibres it is linear (Fig. 8). The fibres with intermediate conduction velocities show some intermediate relation, i.e. the relation is neither linear nor logarithmic as observed by Hodgkin & Katz (1949) in the squid axon. Figure 8 suggests

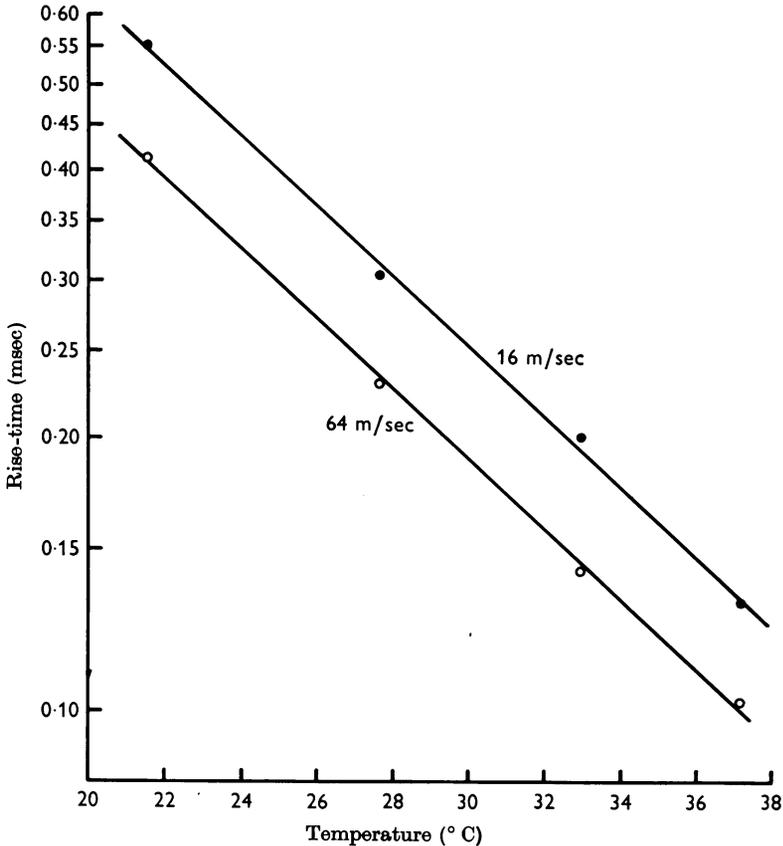


Fig. 6. Graphs showing that logarithmic relation of rise-time of the impulse to temperature is similar in fast (64 m/sec) and slow (16 m/sec) fibres. The points were obtained from the graphs of Fig. 5 and from similar results obtained at 27.6 and 37.1° C. The regression lines were computed. The ordinate scale is logarithmic.

that between 27 and 37° C the fast fibres are affected in nearly the same way as the slower fibres since the Q_{10} is about 3.6 in the fast fibres and 3.5 in the slow fibres. Below 27° C the Q_{10} in the fast fibres falls markedly, being about 1.9 between 20 and 30° C.

Mechanism underlying ARP. The relation of conduction velocity to ARP at 20° C has been described in Fig. 8C of an earlier paper (Paintal, 1965). This should be consulted for details about the method used for

determining ARP and how artifacts inherent in earlier work (cf. Erlanger & Gasser, 1937; Schoepfle & Erlanger, 1941) were eliminated. Except for one aberrant point at 84 m/sec all the other points at 20° C (of Fig. 8C, Paintal, 1965) have been shown in Fig. 11. In this figure the relation of

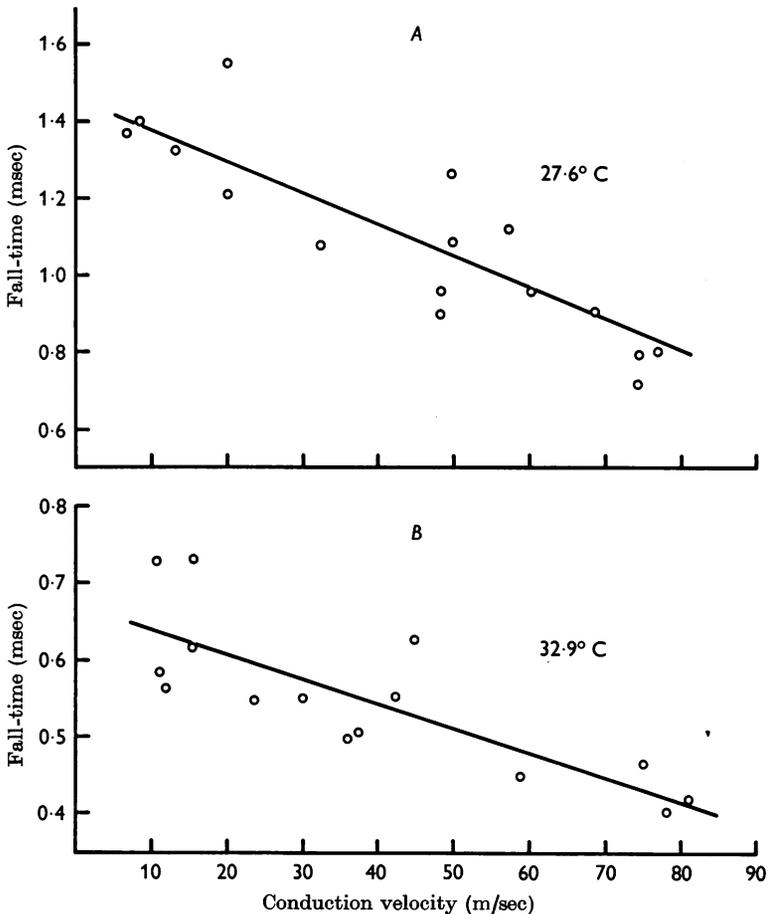


Fig. 7. Relation of duration of falling phase (fall-time) of the impulse to the conduction velocities of vagal nerve fibres at 27.6° C (A) and 32.9° C (B). The regression lines were computed assuming a linear relation to exist between the falling phase and conduction velocity, which seems to be the case.

spike duration at 20° C to conduction velocity is also shown. This curve for spike duration was obtained by extrapolating the curves in Fig. 4 to 20° C. It is clear from Fig. 11 that the spike duration at 20° C is by itself too short to account for the ARP at 20° C. Thus, at a conduction velocity of about 40 m/sec the ARP is about one and a half times spike duration, and at a conduction velocity of 10 m/sec it is about twice spike duration.

It is therefore certain that some other factor (or factors) must be responsible for the excess interval (forming part of ARP) over the spike duration. In this connexion Tasaki has estimated that conduction of the second impulse should cease at a time when, during the recovery period, the amplitude of the second spike is below 40% of normal (Tasaki, 1953). It therefore seemed worth while to determine whether the observed relation of ARP to conduction velocity can be accounted for by this factor.

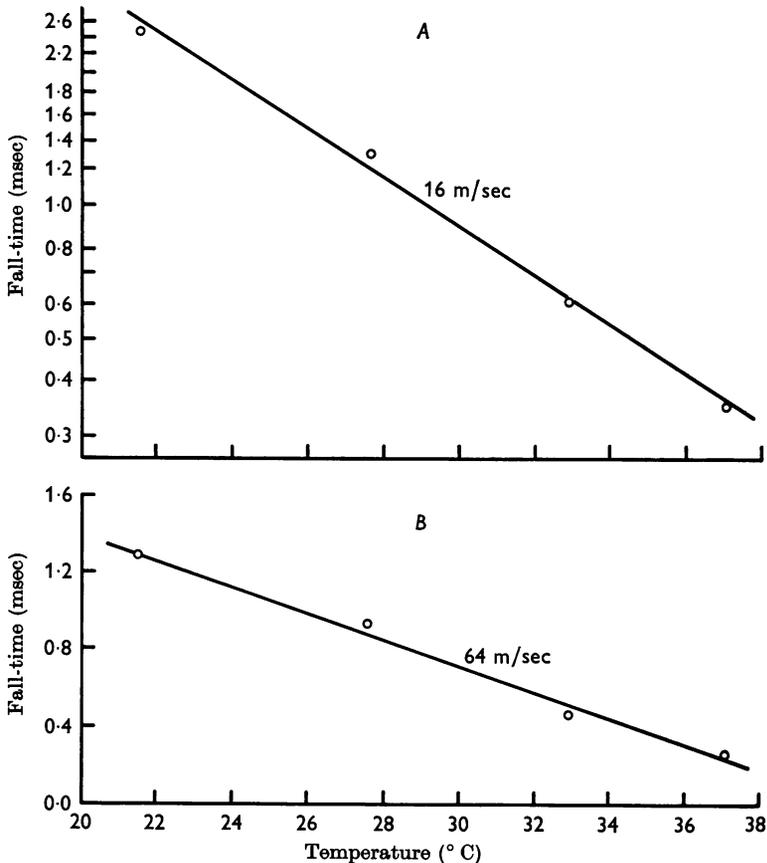


Fig. 8. Relation between fall-time and temperature. *A*, slow fibres, 16 m/sec; *B*, fast fibres, 64 m/sec. In slow fibres the relation of the falling phase to temperature is logarithmic; in fast fibres it is linear. Ordinate scale in *A* is logarithmic.

In order to do this the nerve fibres were cooled at the recording end of the fibre only (Fig. 9*A*). This ensured that the second impulse was able to propagate up to the recording electrodes because the temperature of the fibre was lowest at the recording electrodes. The remainder of the fibre was at body temperature (Fig. 9*A*) (see Methods) except for a few millimetres of the fibre in the transition zone between body temperature

and the cooled filament which must have been at some intermediate, but higher, temperature than the filament on the recording electrodes.

Figure 9*B-D* provides samples of records obtained from three fibres with this experimental arrangement. These show the manner in which the

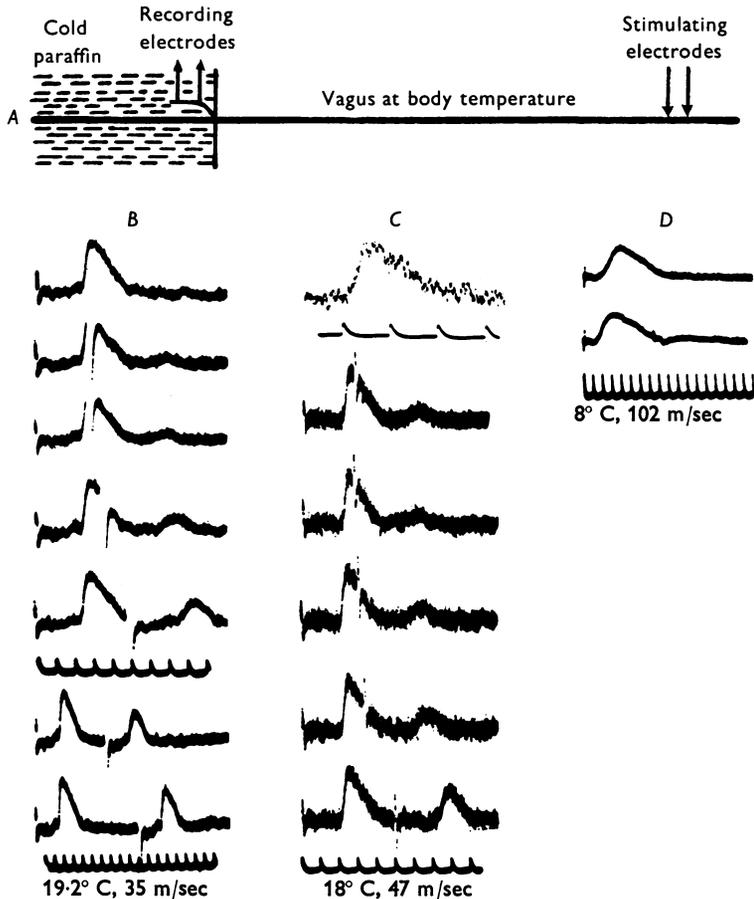


Fig. 9. Records showing recovery of amplitude of the second impulse at varying intervals after the first. *A* shows the arrangement for recording impulses from the cooled end of the nerve fibre while the rest of the vagus is at body temperature which was 37, 31 and 26° C in series *B*, *C* and *D* respectively. Conduction velocities of the three different fibres were 35, 47 and 102 m/sec respectively in *B*, *C* and *D*; the temperatures of nerve fibres at the recording electrodes were respectively 19.2, 18 and 8° C. The first sweep in each series shows the first impulse. The second and the third sweeps (only second in *D*) show that a small impulse is generated at the end of the first impulse. All time marks represent msec. Those in fifth sweep in series *B* apply to the first four sweeps as well; those in the seventh sweep also to the sixth sweep; similarly in series *C*. The graphs in Fig. 10*A* were plotted from the sweeps in series *B* as well as other sweeps.

amplitude of the second impulse recovers with increasing interval after the first impulse. The recovery curve at 19.2° C shown in Fig. 10A has been plotted from such records, and those in Fig. 10B from similar records in another fibre. Many of the curves obtained showed a logarithmic relation between recovery of spike amplitude and time. In ten fibres it was found that the time for recovery of spike height from 40 to 100 % was about

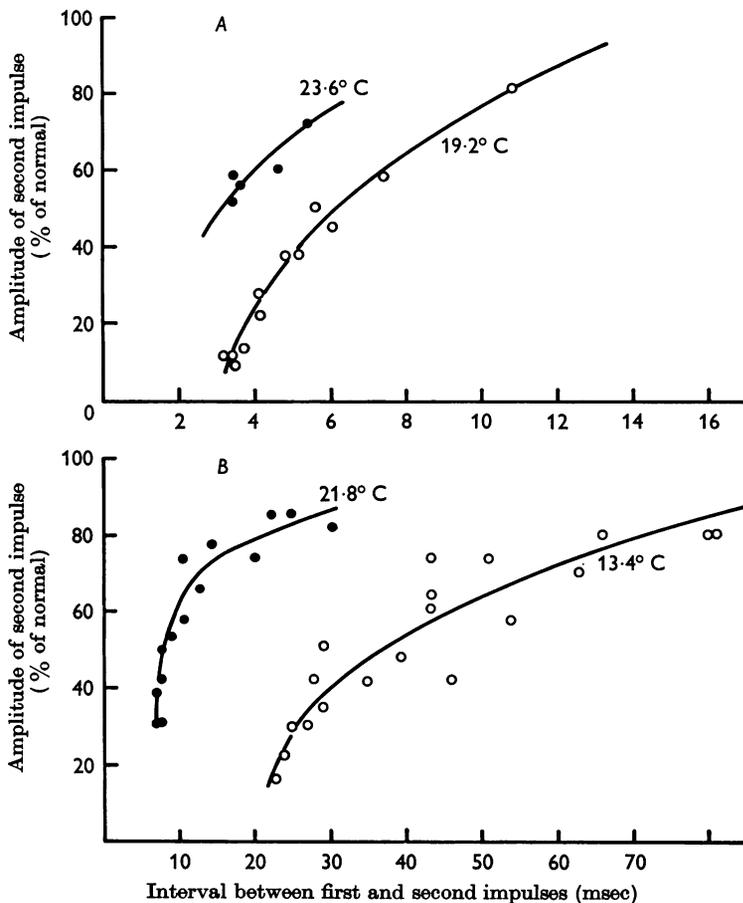


Fig. 10. Graphs showing recovery of amplitude of the second impulse at varying intervals after the first. The graph at 19.2° C in A was plotted from sweeps such as those shown in series B of Fig. 9. The conduction velocity of the fibre was 35 m/sec. B shows similar results at two temperatures obtained from another fibre with a conduction velocity of 12 m/sec.

four times the interval for recovery up to 40% without any obvious relation to temperature or conduction velocity. Comparison of the two curves in Fig. 10A (and those of Fig. 10B) shows that the rate of recovery of the second impulse is greatly slowed with fall in temperature. At very

low temperatures it is very slow indeed, even in fast-conducting fibres. For example at about 8° C, the second impulse of the fast fibre shown in Fig. 9D had barely recovered up to 50 % of its normal amplitude 30 msec after the first impulse.

The most noteworthy new finding in the present context is that the rate of recovery depends on the normal conduction velocity of the fibre; the greater the velocity, the quicker the recovery of the second impulse. Thus recovery in the fibre with a normal conduction velocity of 35 m/sec at 19.2° C (Fig. 10A) is faster than that in Fig. 10B with a conduction velocity of 12 m/sec even though the temperature of the latter (21.8° C) is 2.6° C higher than that of the former.

From curves such as those shown in Fig. 10 the time for 40 % recovery of spike amplitude was determined at different temperatures in different fibres. The mean Q_{10} for 40 % recovery of spike amplitude determined in six fibres was 4.6 between 13 and 29° C (s.e. 0.45; range, 3.3–5.9). Since the Q_{10} for ARP is 3.6 between 18 and 28° C, and 12.2 between 8 and 18° C (Paintal, 1965) it is very likely that there is no difference between the temperature coefficients of ARP and time for 40 % recovery of spike amplitude.

The interval for 40 % recovery at about 20° C was determined in thirteen fibres with different conduction velocities; these have been plotted in Fig. 11 which shows unequivocally that the interval after a preceding impulse for 40 % recovery of spike amplitude is inversely related to the normal conduction velocities of the fibres. Similar curves were obtained at other levels of recovery, e.g. at 50 % recovery, the only difference being that the curves are displaced vertically upward in the case of 50 % recovery.

The curve in Fig. 11 has been drawn by eye to fit the points as closely as possible. The interesting point is that it also fits the points for ARP rather well and so one can say that the curve for 40 % recovery of spike amplitude at 20° C is almost identical with that for the ARP at the same temperature. This temporal coincidence between ARP and time for 40 % recovery and also the similarity of their respective temperature coefficients shows that the ARP lasts as long as it takes the amplitude of the second impulse to return to 40 % of its normal amplitude. Simultaneous changes in other factors are also undoubtedly concerned, e.g. changes in threshold of the fibre during the ARP (Tasaki, 1953), but whatever these are, they all add up to make the fibre unable to *conduct* an impulse at a time when the spike amplitude of the second impulse is less than 40 % of normal. However, Figs. 9 and 10 show that spikes with amplitudes less than 40 % can be *initiated*. These must all be abortive, because (1) there is a clear difference between spike duration and ARP at all rates of conduction

(Fig. 11), and (2) small spikes can be generated at the end of the first spike (Fig. 9). Although these spikes are abortive, it has been confirmed that they nevertheless leave behind a period of refractoriness similar to that following the first spike.

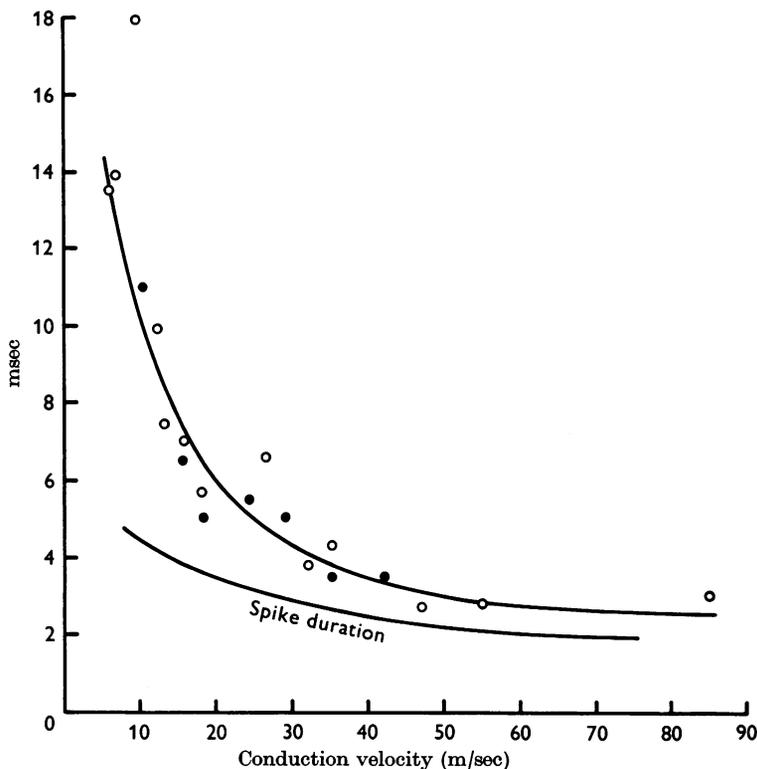


Fig. 11. Relation of time of recovery of the amplitude of the second impulse to 40% of normal following a preceding impulse, to the normal conduction velocity of the fibres; temperature, 20° C. The filled circles are the values for ARP at 20° C reproduced from Fig. 8C of an earlier paper (Paintal, 1965). The lower curve shows the relation of spike duration at 20° C to normal conduction velocity. This was obtained by extrapolating the curves shown in Fig. 4 (and other similar curves for other values of conduction velocities) to 20° C. Note the identity of the ARP with the interval for 40% recovery of spike amplitude and the marked disparity between spike duration and ARP at the lower levels of conduction velocity.

DISCUSSION

The results of Gasser & Grundfest (1939) have been confirmed in so far as that the spike duration of the majority of the fibres is 0.4–0.5 msec at 37° C. Contrary to accepted belief (that originated with them) it has been demonstrated unequivocally that the spike duration of medullated nerve fibres is inversely related to their conduction velocities at various tem-

peratures. It appears that Gasser & Grundfest did not notice this relation because they approximated their measurements to the first decimal place. This conclusion is borne out by a re-analysis of their records of ten impulses shown in Fig. 3 of their paper which reveals a relation between spike duration and conduction velocity qualitatively similar to that found in the present investigation. However, it is not certain what criterion they used for defining the end of the spike.

The second important conclusion of this paper is that, as far as spike duration is concerned, preganglionic nerve fibres are identical with somatic nerve fibres having the same conduction velocities. Their spike durations at 37° C will therefore range from 0.45 to 0.6 msec for fibres with conduction velocities between 10 and 14 m/sec (Fig. 2*B*); for the still slower fibres they will be greater. Here it is important to point out that Grundfest's conclusion, that the spike duration of preganglionic fibres is about 1.2 msec, is largely based on the fact that their ARP (determined in the whole nerve) is 1.2 msec (Grundfest, 1939, 1941), which is to be expected from the present results (i.e. twice spike duration as shown in Fig. 11). And since he wrongly assumed, as nearly all workers have done so far since Adrian's paper (1921), that the ARP lasts as long as the spike, he concluded that the spike duration of these fibres was 1.2 msec. As pointed out by Grundfest (1939) it is not easy to record impulses in single fibres dissected from filaments of the cervical sympathetic.

The classification of preganglionic fibres into a separate B group has stood so far on two considerations, their longer spike durations and their after-potentials (Grundfest, 1939, 1941). In view of the present results the desirability of retaining this distinction needs to be reconsidered.

The duration of the rising phase of the impulse varies exponentially with temperature in both fast and slow fibres, the Q_{10} being 2.5 between 20 and 37° C. This is in agreement with the observations of Inman & Peruzzi (1960) on mammalian nerve fibres in the same temperature range. Further, Frankenhaeuser & Moore (1963) have found recently that the rate constant α_m , on which the rate of rise of the impulse depends, varies exponentially with temperature in toad nerve fibres. On the other hand, in the squid axon the results of Hodgkin & Katz (1949) suggest that the relation of temperature to rise-time is not logarithmic because the Q_{10} is different at different ranges of temperature.

The present conclusions concerning fall-time are in agreement with certain casual observations of earlier workers. Blair & Erlanger (1933) observed that the duration of the falling phase, in general, seemed to increase as conduction rate slowed. Unfortunately they could not make a firm statement because they could not be certain of the end of the spike and because their records were partly diphasic (Blair & Erlanger, 1933).

Similarly, I. Tasaki in a personal communication to Rushton (1951) hinted that the duration of falling phase of the spike may be greater in smaller fibres, but the matter received no further attention, presumably because the idea conflicted with the established belief that spike duration is the same in all A fibres!

The most interesting point concerning fall-time is that its actual relation to temperature depends on the conduction velocity of the fibres. In the slow fibres it is exponential, and in the fast fibres, linear. This, therefore, suggests that there are (at least) two or more factors that determine the magnitude of fall-time and that these factors vary in quantitatively different ways in the fast and slow fibres with variation in temperature.

In connexion with ARP, now that the concept of constancy of spike duration has fallen, it is important to draw attention to the papers by Blair & Erlanger (1933) and by Hursh (1939*b*). Blair & Erlanger showed that in frog nerve fibres the ARP varied with conduction velocity in a manner somewhat similar to that demonstrated recently (Paintal, 1965). They also showed that the rate of recovery of threshold varied with the speed of conduction of the fibre. Similarly, Hursh's results in cat nerves on ARP (Hursh, 1939*b*) closely resemble those of the present author and they also show that the properties of A and so-called B fibres form a continuous series as far as ARP is concerned. This view is compatible with the demonstration in the present paper that A and so-called B fibres are similar in their spike durations. However, Hursh's results were totally ignored because they ran contrary to the established belief of constancy of spike duration in A fibres.

There can no longer be any doubt that abortive spikes are produced during part of the ARP, i.e. the ARP as defined by Adrian's method of determining it (Adrian, 1921). This definition of ARP (cf. Paintal, 1965) should be retained because of the vast amount of literature associated with it and because it is based on the method most frequently used for determining it (Amberson, 1930; Gasser, 1931; Blair & Erlanger, 1933; Gasser & Grundfest, 1936, 1939; Erlanger & Gasser, 1937; Hursh, 1939*b*; Schoepfle & Erlanger, 1941). The interval lasting about as long as the spike during which no impulse can be initiated might be termed as absolute refractory period (initiation) (ARP_i) since abortive spikes are initiated at the end of the spike.

It is now possible to give a more suitable explanation for the mechanism underlying the complete block of a train of impulses in a cooled stretch of nerve fibre described earlier (Paintal, 1965). In Fig. 13*A* of the earlier paper (Paintal, 1965), it was shown that all the impulses of a train (except the first) with a frequency only a little higher than the maximum frequency that could pass through the cooled stretch of nerve (determined by the

ARP following the second impulse (Paintal, 1965)) were blocked. This behaviour is now best explained as follows: it is known from the present investigation that at low temperatures there is a large difference between ARP and ARP_1 (equal to spike duration) especially in slowly conducting fibres (Fig. 11). It is also known from the present results that abortive spikes generated during the ARP leave behind a state of refractoriness similar to that following the first impulse. Therefore it follows that if one abortive spike follows another and so on [as would happen if the frequency is a little higher than the maximum permissible frequency (Fig. 13A in Paintal, 1965)], total block must result. Precisely the same sort of thing will happen at higher frequencies in the transition zone, as discussed earlier. In the earlier explanation (Paintal, 1965) the entire mechanism for block of a train of impulses was supposed to lie in the transition zone because it was assumed (wrongly, since nothing was known about abortive spikes at that time) that an impulse arriving at the cooled region during the ARP will have no depressant effect. No doubt the earlier explanation attributed to the transition zone still holds for block of still higher frequencies but here too it follows that the role of abortive spikes must be predominant.

The present results may be helpful for explaining certain observations of earlier workers. For example, Frankenhaeuser (1960) observed that there was a variation in α_h in different fibres. This variation can now be explained if it is assumed that α_h and fibre diameter are related [since rate of recovery of spike height and fibre diameter are related (Fig. 11)], and that there was a variation in the diameter of his different fibres.

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