

**SYNAPTIC STIMULATION SUPERIMPOSED
ON MOTONEURONES FIRING IN THE 'SECONDARY RANGE'
TO INJECTED CURRENT**

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SUMMARY

1. This paper extends the work on the 'primary range' of firing (Granit, Kernell & Lamarre, 1966), in lumbar motoneurones to the 'secondary range'. By definition the latter begins when, with stronger currents, the linear curve relating firing rate to injected current in the primary range undergoes a fairly sudden increase of slope.

2. It was shown that motoneurones firing at the higher frequencies of the secondary range were partially inactivated. Yet such firing rates were within the physiological range.

3. Algebraical summation of firing rates, when present in the secondary range, implied at the same time that the synaptic amount added was diminished by comparison with what it had been within the primary range.

4. Superimposed synaptic excitatory stimuli did not (as in the 'primary range') regularly add their effect algebraically on to the rate of firing achieved by injected currents alone. More commonly the synaptic effect of the constant input underwent a progressive increase throughout the secondary range.

5. Superimposed inhibitory stimuli regularly reduced the slope constant as determined by trans-membrane current alone and, by counteracting inactivation, made the motoneurone approach the mode of firing characteristic of the 'primary range'.

6. The latter finding emphasizes the significance of analysing firing motoneurones with the aid of 'slope constants' and provides inhibition with a new role in the integrative behaviour of motoneurones, as considered in the Discussion.

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INTRODUCTION

At the upper limit of the primary range of firing, discussed in our previous paper (Granit *et al.* 1966), the linear curve relating firing rate to injected current in cat motoneurons undergoes an increase of slope, as described by Kernell (1965*a*, 1966). This is the secondary range of firing, to which also can be fitted a straight line, with the consequence that the primary and secondary ranges in the graphs meet at an angle. Several examples of this are found in the Figures of this paper. The mechanism of after-hyperpolarization tends to be disturbed in the secondary range (Kernell, 1965*a, b* and Fig. 7 below). In the preceding paper (Granit *et al.* 1966), which from now on we shall refer to as Paper I, it was shown that firing within the secondary range is likely to involve a measure of inactivation of the regenerative capacity of the cell. With the aid of inhibition we shall demonstrate this process below.

The main concern of the present paper is with synaptic superposition of excitation and inhibition on the activity of motoneurons firing within the secondary range to injected currents. In this respect it is a direct sequel to Paper I dealing with the primary range and it also makes use of the motoneurons collected in Table 1 of that paper. The experimental arrangements and procedures are those reported there. When a sufficient number of readings were available the regression lines presented were calculated by the method of least squares.

RESULTS

Curves, parallel within secondary range. The curve of superimposed facilitation, which in the primary range runs parallel with that of the control representing the effect on firing rate of injected current by itself, may occasionally also be parallel with it in the secondary range. There were two such cases, one of which is illustrated in Fig. 1. The slope constants of the two curves in the primary range were identical to within one decimal point, $1.4 \text{ impulses sec}^{-1} \text{ nA}^{-1}$. In the secondary range the values were few but it suffices for the present purpose to draw the curves parallel there, too, with slope constants of $7\text{--}8 \text{ impulses sec}^{-1} \text{ nA}^{-1}$.

There is undoubtedly an element of arbitrariness in deciding how many readings should be included within the primary range and thus excluded from the secondary. The best we could do was to calculate the slope of the primary range with the aid of the method of least squares. To judge by the high degree of precision obtained in Paper I on the basis of a statistical evaluation of the slope constants obtained in this manner, the upper limit of the primary range, i.e. the last reading to be included in the calculations, must have been correctly estimated. However, the exact frequency

of discharge at the turning point of the curves could not be determined with sufficient accuracy, as this would have required a greater number of readings from this particular region than were obtained in our experiments. In them its location could not become known until after the actual measurements had been performed. For this reason relative shifts in the turning points of the controls and the facilitated curves cannot be given any significance. These points are largely a consequence of treating our data by two linear approximations.

The difference added by facilitation in Fig. 1 was 7.4 impulses/sec in the primary range and around 8.5 impulses/sec in the secondary range. The same synaptic stimulus was, of course, added throughout the experiment. It is easily calculated that the difference of 7.4 impulses/sec in the primary range corresponds to a current of 5.3 nA. With an increase of the slope constant to 7–8 impulses $\text{sec}^{-1} \text{nA}^{-1}$ this amount of current—had its effect been constant—ought to have delivered around 40 impulses/sec instead of the 8.5 actually recorded.

In Fig. 1 the synaptic effect was reduced in the secondary range, possibly, by a block produced by the current inactivating the region around the electrode tip. If the reduction had been due to a diminution of the excitatory post-synaptic potentials (EPSPs) with stronger currents, the process ought to have developed gradually and not, as here, suddenly, afterwards to become constant within the secondary range. The weakness of the latter explanation of the reduction of the synaptic effect in the secondary range is best exposed by an experiment—the counterpart to that of Fig. 1—in which the superimposed test stimulus is inhibitory. What happened is shown in Fig. 2 in which the number of readings in the secondary range guarantee greater accuracy. Again parallelism prevailed in both ranges of firing. We know, however, that the inhibitory post-synaptic potentials (IPSPs) should increase with the increasing depolarization produced by the current (Eccles, 1957). Thus the question of why an effect of subtraction in the secondary range is constant poses the same problem as was encountered above for the effect of an addition.

In the experiments of Fig. 2 the slope constant was 1.9 impulses $\text{sec}^{-1} \text{nA}^{-1}$ in the primary, 2.8 impulses $\text{sec}^{-1} \text{nA}^{-1}$ in the secondary range. The corresponding differences were -8 and -6 impulses/sec. A calculation similar to the one carried out above for excitation reveals that if the synaptic effect actually had been the same in the secondary as it was in the primary range, the difference in the former range ought to have been -12 impulses/sec instead of the -6 recorded. Thus, also with inhibition there is a reduction of the synaptic effect in the secondary range of firing. Since the depolarizing current has opposite effects on excitatory and inhibitory post-synaptic potentials, and, indeed, fairly small ones on the

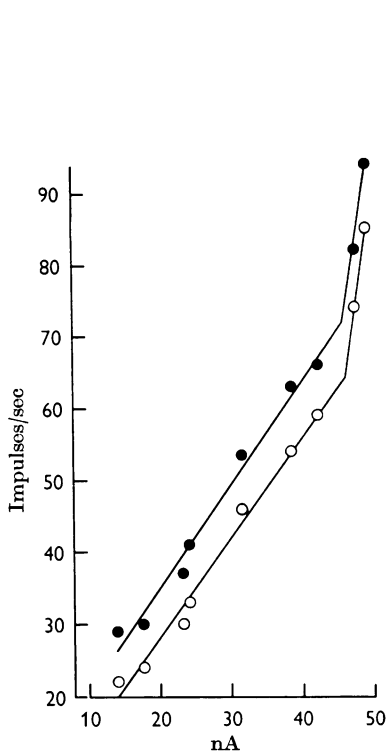


Fig. 1

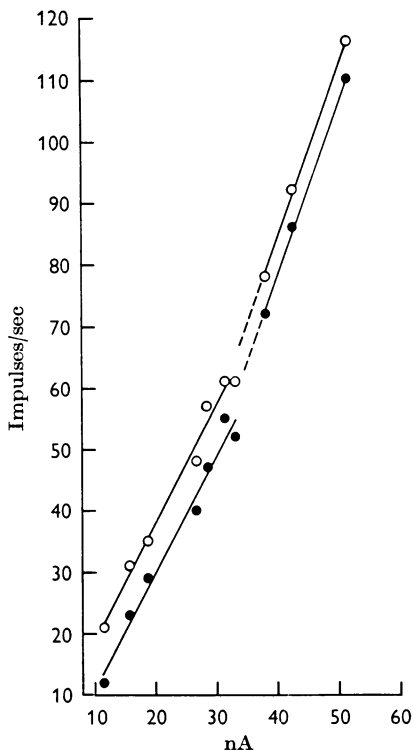


Fig. 2

Fig. 1. Anaemically-narcotized cat. Ankle extensor motoneurone of spike height 70 mV (2 M potassium citrate electrode). Curve drawn through *open circles* in this and subsequent Figures is the control showing the relation between steady discharge frequency (measured for 0.5 sec before onset of stretch) and trans-membrane stimulation alone. *Filled circles*, same with facilitation by 10 mm stretch of the gastrocnemius-soleus muscle. Stretch rose in 1.4 sec and was maintained for about 2 sec. Facilitated discharge frequency was measured from 0.5 to 2.5 sec after onset of stretch. Maximal firing rate within the primary range was at a frequency of about 65 impulses/sec. In the primary range $k_A = 1.38$ impulses $\text{sec}^{-1} \text{nA}^{-1}$ for the 'control', 'facilitated' $k_A = 1.44$ impulses $\text{sec}^{-1} \text{nA}^{-1}$. In the 'secondary range' the slope constants of the two curves are identical within 7–8 impulses $\text{sec}^{-1} \text{nA}^{-1}$. The difference added by facilitation is 7.4 ± 0.77 (s.d.) impulses/sec in the primary range and 8.5 impulses/sec in the secondary range.

Fig. 2. Anaemically-narcotized cat. Unidentified motoneurone of spike height 80 mV and steady resting membrane potential of 62 mV (2 M potassium citrate electrode) inhibited by high-frequency stimulation of cut hamstring nerve. 'Control' measured for 0.5 sec before onset of nerve stimulation; 'inhibited' readings during the first 0.5 sec of hamstring nerve stimulation. The maximal discharge frequency within the primary range is around 60 impulses/sec. In the primary range, control $k_A = 1.90$ impulses $\text{sec}^{-1} \text{nA}^{-1}$; for the 'inhibited' curve $k_A = 1.89$ impulses $\text{sec}^{-1} \text{nA}^{-1}$; the difference between the two curves is -8.1 ± 1.52 (s.d.) impulses/sec. In the secondary range, $k_A = 2.83$ impulses $\text{sec}^{-1} \text{nA}^{-1}$ for the two curves and the difference is -6.0 impulses/sec (by method of least squares).

EPSPs (Eccles, 1957), we conclude that the similar reduction of the synaptic effect in both cases may well be explained by a block induced by strong currents preventing distant synaptic effects from reaching the firing site. In such cases the electrode tip would have to be near the firing site to produce a zone of local inactivation between it and the synapses concerned.

Curves, diverging within secondary range. Alternatively the parallel curves in the secondary range may be supplanted by divergent ones, both with excitation or inhibition added.

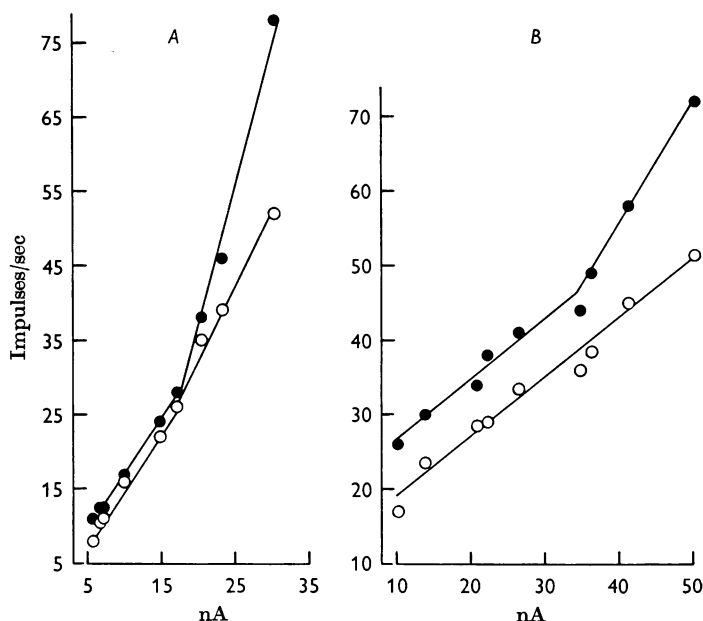


Fig. 3. *A*, pentobarbitone-chloralose cat. Hamstring motoneurone of spike height 62 mV (2 M potassium citrate electrode) facilitated by steady tetanic contraction of gastrocnemius-soleus muscle as elicited from peripheral stump of cut ventral root. Maximal discharge frequency within the primary range at about 30 impulses/sec. In the primary range 'control' $k_A = 1.51$ impulses $\text{sec}^{-1} \text{nA}^{-1}$; 'facilitated' $k_A = 1.59$ impulses $\text{sec}^{-1} \text{nA}^{-1}$; difference, 2.0 impulses/sec. In the secondary range 'facilitated' $k_A = 3.79$ impulses $\text{sec}^{-1} \text{nA}^{-1}$ ('control' $k_A = 1.9$) indicating increase of the synaptic excitatory effect. *B*, anaemically-narcotized cat. Ankle extensor motoneurone of spike height 65 mV (2M potassium citrate electrode) facilitated by 10 mm stretch of the gastrocnemius-soleus muscle rising in 1.4 sec. Control curve is the mean of values measured before and after stretch (1.0 sec before onset of stretch and from 0.5 to 1.0 sec after cessation of stretch). Facilitated curve measured from 0.5 to 2.5 sec after onset of stretch. 'Control' stays within the primary range ($k_A = 0.74$ impulses $\text{sec}^{-1} \text{nA}^{-1}$). Facilitated curve is parallel with 'control' in the primary range ($k_A = 0.80$ impulses $\text{sec}^{-1} \text{nA}^{-1}$). Difference = 7.9 ± 1.35 (s.d.) impulses/sec. Synaptic excitation pushed the cell into the secondary range where $k_A = 1.63$ impulses $\text{sec}^{-1} \text{nA}^{-1}$.

A. Excitation. Figure 3*A* and *B*, shows that this effect is obtained both with a contraction making use of the Golgi tendon organs and their polysynaptic circuits, as well as with a stretch reflex which has a large monosynaptic component. Thus Fig. 3*A* is from a hamstring motoneurone in a chloralose-pentobarbitone cat. The cell was facilitated by a gastrocnemius-soleus contraction from the cut ventral root. The experiment was carried out twice with a pause in between. The outcome was the same in both cases. In this experiment there was actually some irregularity of firing rate in the secondary range but this fact can hardly be significant as, in the large majority of our experiments, firing within the secondary range was as regular as in the primary (cf. Kernell, 1965*a*, 1966).

In Fig. 3*B*, the facilitation arrives from an autogenetic stretch reflex and influences a gastrocnemius-soleus motoneurone. It is one instance of several (cf. Fig. 4 below and 4*B* of Paper I) in which the facilitated curve runs into the secondary range before that range has been reached with the aid of injected current alone. The effect of the latter on the cell was not always easy to estimate by listening-in to the firing rate so that the turning point (for the secondary range) could have been misjudged and the experiment interrupted too soon.

When strong currents were needed for the secondary range, say, owing to leakage around the electrode tip, the compensation might have been insufficient and electrode polarization might have created trouble. When for such reasons it became impossible to push the cell into the secondary range by means of injected current, the motoneurone itself could be shown to be in perfect condition by adding a large enough synaptic stimulation which in that case achieved the desired effect. Very high frequencies were then reached. Figure 4*A* illustrates the largest difference studied in the primary range, 56.2 impulses/sec. It was multiplied to 100 impulses/sec in the final value of the secondary range. The experience of Kernell (1965*a*) was that impulse frequency, and not current as such, is the decisive factor in bringing on firing within the secondary range. Our findings with synaptic excitation are in general agreement with this conclusion (but see section below on Inhibition).

Figure 4*B* is a case in which the injected current—as nearly always with rat motoneurones (Granit, Kernell & Shortess, 1963)—led in the end to a block preventing any further increase in the frequency of discharge. All three curves flattened out at the same current strength. The uppermost one, running into the secondary range, shows the immediate effect of the added synaptic stimulus; that in the middle, the effect after 1 sec of adaptation. To judge by the range of increase in spike frequency in the uppermost curve, the cell, regarded as a spike generator responding to

synaptic currents, must have been in good condition up to the very moment the increasing current produced the blocking effect.

In the secondary range there is clearly great variability of behaviour from cell to cell. There is, of course, also a corresponding—and essentially

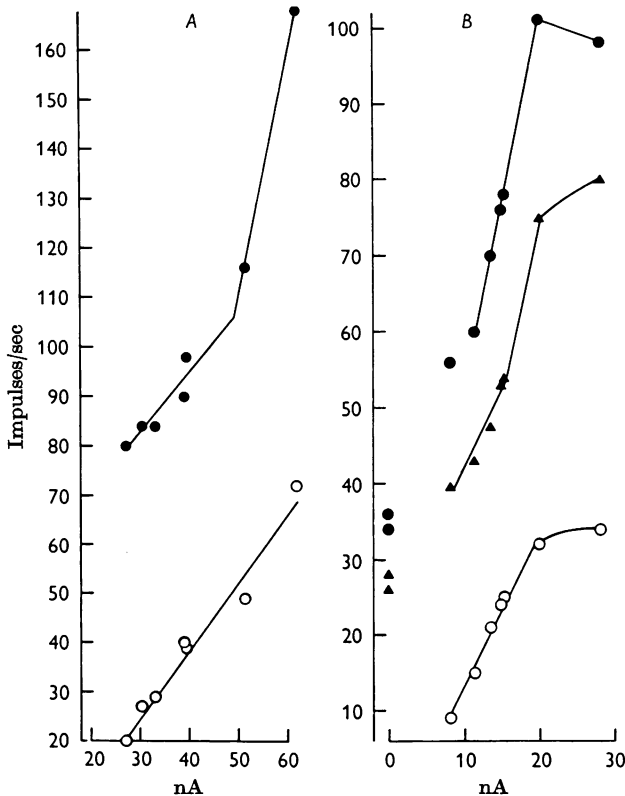


Fig. 4. *A*, anaemically-narcotized cat. Hamstring motoneurone of spike height 87 mV (3 M potassium chloride electrode) facilitated by high-frequency stimulation of cut hamstring nerve. Control and facilitated curves measured for 0.25 sec before and 0.25 sec after onset of nerve stimulation. 'Control' stays within the primary range ($k_A = 1.39$ impulses $\text{sec}^{-1} \text{nA}^{-1}$). In this range 'facilitated' $k_A = 1.20$ impulses $\text{sec}^{-1} \text{nA}^{-1}$ with a difference of 56.2 ± 3.54 (s.d.) impulses/sec. Synaptic excitation pushed the cell into the secondary range in which k_A is about 5 impulses $\text{sec}^{-1} \text{nA}^{-1}$. *B*, anaemically-narcotized cat. Ankle extensor motoneurone of spike height 70 mV (3 M potassium chloride electrode) facilitated by high-frequency stimulation of popliteal nerve. 'Control' measure for 0.5 sec before onset of stimulation; facilitated curves for 0.5 sec after onset of stimulation (phasic, filled circles) and from 1.0 to 1.5 sec after onset of stimulation (tonic, filled triangles). Control and 'tonic' curves are parallel in the primary range ($k_A = 2.01$ and 2.02 impulses $\text{sec}^{-1} \text{nA}^{-1}$ respectively) with a difference of 28.6 ± 1.31 (s.d.) impulses/sec. The 'phasic' curve is almost entirely confined to the secondary range where $k_A = 4.79$. In this cell, the highest current strength used (28 nA) led to a block preventing any further increase in the frequency of discharge.

uninterpretable—variability of geometry of the penetrated structure both as such as well as relative to the site of the electrode tip. But some findings stand out as common for the superposition of synaptic excitation in this range: (i) the synaptic effect is often increased when the motoneurons are forced to fire at fast rates, (ii) this effect is in a more definite manner tied to frequency of discharge than to current strength, (iii) synaptic stimulation tends to have a considerably stronger effect on the firing rate than can be achieved by injected current alone, (iv) evidence of blocking is not uncommon at the current strengths needed for the secondary range (Fig. 4*B*).

B. Inhibition. With inhibition, divergent curves in the secondary range is the rule. Figure 5*A* and *B* illustrates two experiments in which the secondary range could be followed into high frequencies of discharge for trans-membrane stimulation alone. These 'controls' are the upper curves in the graphs. In Fig. 5*A* the constant difference of -5 impulses/sec within the primary range underwent a steady increase in the secondary range amounting in the end to -54 impulses/sec. The slope of the inhibited curve, which was 1.9 impulses $\text{sec}^{-1} \text{nA}^{-1}$ in the primary range, fell below this value to 1.7 impulses $\text{sec}^{-1} \text{nA}^{-1}$ in the secondary range. Fundamentally the same result is shown in Fig. 5*B*, but the slope in the secondary range, 1.7 impulses $\text{sec}^{-1} \text{nA}^{-1}$, stayed above the value found in the primary range which was 1.3 impulses $\text{sec}^{-1} \text{nA}^{-1}$.

Smaller ranges of firing rate are covered by the experiments reproduced in Fig. 6*A* and *B*, but otherwise the outcome of them is the same. In Fig. 6*A* inhibition in the secondary range produced what is virtually an extension of the primary range, while in Fig. 6*B* with inhibition, the slope in the secondary range again fell below the value obtained in the primary range. These results suggest that, when experimental curves with greatly extended primary ranges are found (Fig. 1, Paper I), the explanation may well be that the cell in question has been under tonic inhibition.

Inactivation revealed by inhibition. From the quantitative work on the inactivation (*h*) of the sodium mechanism in the vertebrate nerve fibre (Frankenhaeuser, 1963; Vallbo, 1964; Frankenhaeuser & Vallbo, 1965) it is possible to obtain precise values demonstrating its dependence on the degree of depolarization. In principle these results must be applicable also to the motoneurons but in actual practice it is necessary to show by experimentation that inactivation occurs in the secondary range. A complication in our type of experiment is that the same electrode is used for stimulation and recording. With the strong currents needed for the secondary range, electrode polarization and deficient compensation may interfere with the measurements of spike height. This is where superimposed inhibition becomes a valuable tool, because the spike should

increase in size in an inactivated cell subjected to inhibition. Whenever spike size was kept inside the proportionality range of the amplifier, it proved possible to demonstrate this increase. In the primary range, inhibition did not influence spike height, except occasionally at the upper limit of it.

Figure 7A is an example. This motoneurone, silent from the beginning, was stimulated by a current of 36.5 nA corresponding to a value of about

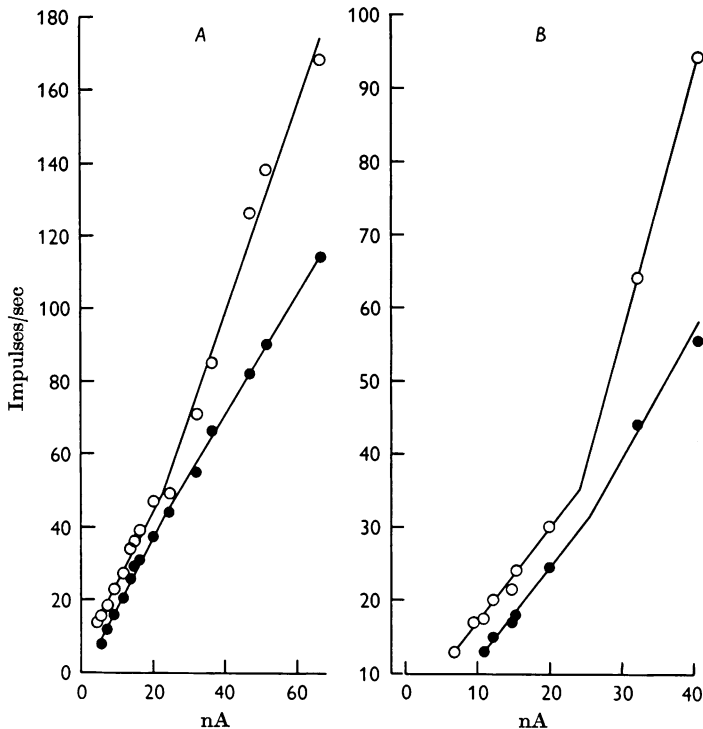


Fig. 5. *A*, pentobarbitone cat (35 mg/kg). Hamstring motoneurone of spike height 75 mV (2 M potassium citrate electrode), inhibited by high-frequency stimulation of the deep peroneal nerve. 'Control' and inhibited curves measured for 0.5 sec before and 0.5 sec after onset of nerve stimulation. In the primary range the two curves are parallel; 'control' $k_A = 1.89$ impulses $\text{sec}^{-1} \text{nA}^{-1}$; 'inhibited' $k_A = 1.90$ impulses $\text{sec}^{-1} \text{nA}^{-1}$ and the difference is -7.3 ± 0.94 (s.d.) impulses/sec. In the secondary range, 'control' $k_A = 2.86$ impulses $\text{sec}^{-1} \text{nA}^{-1}$ and 'inhibited' $k_A = 1.66$ impulses $\text{sec}^{-1} \text{nA}^{-1}$. *B*, pentobarbitone cat (35 mg/kg). Popliteal motoneurone of spike height 88 mV (2 M potassium citrate electrode) inhibited by high-frequency stimulation of deep peroneal nerve. Control and inhibited curves measured for 1.0 sec after and 1.0 sec before cessation of nerve stimulation respectively. In the primary range, 'control' $k_A = 1.25$ impulses $\text{sec}^{-1} \text{nA}^{-1}$ and 'inhibited' $k_A = 1.23$ impulses $\text{sec}^{-1} \text{nA}^{-1}$. The difference is -5.1 ± 0.58 (s.d.) impulses/sec. In the secondary range 'control' k_A about 3.5, the 'inhibited' k_A about 1.7 impulses $\text{sec}^{-1} \text{nA}^{-1}$.

3 times the minimal firing rate. In the graph of the experiment (Fig. 7B) the point illustrated in Fig. 7A is marked by an arrow and seen to be the first recording after the shift into the secondary range. At the onset of the current the spike height was 63 mV. Just before the onset of inhibition it had decreased to a steady value of 43 mV; during inhibition it again increased to 63 mV. The form of the spike before the onset of inhibition also demonstrates inactivation.

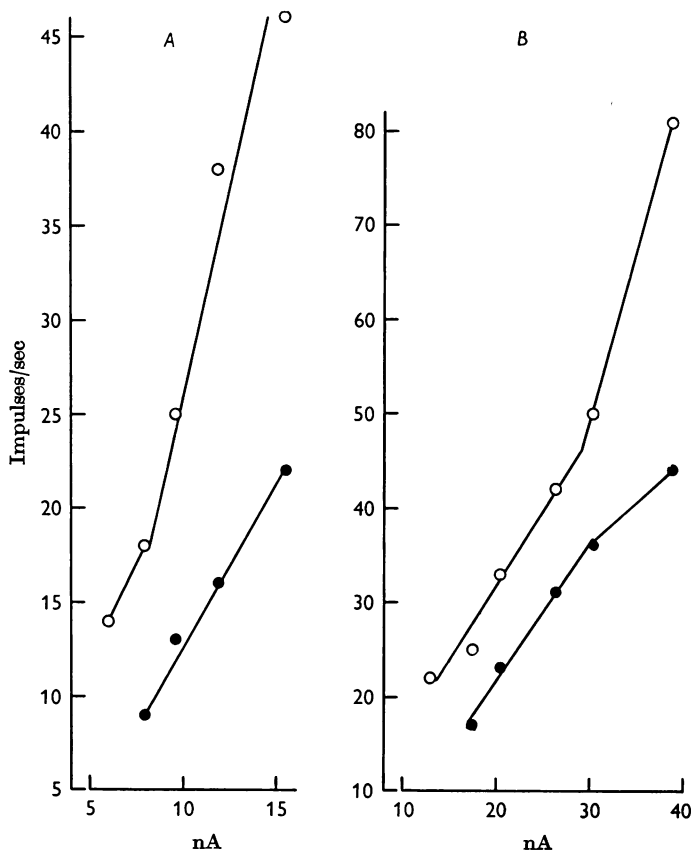


Fig. 6. *A*, anaemically-narcotized cat. Popliteal motoneurone of spike height 88 mV (2 M potassium citrate electrode) inhibited by high-frequency stimulation of deep peroneal nerve. Control curve, measured for 0.5 sec before onset of nerve stimulation, falls mostly in the secondary range where $k_A = 3.76$ impulses $\text{sec}^{-1} \text{nA}^{-1}$. Inhibited curve (measured from 0.5 to 1.0 sec after onset of nerve stimulation) remains in the primary range with $k_A = 1.67$ impulses $\text{sec}^{-1} \text{nA}^{-1}$. *B*, same as *A* but with another popliteal motoneurone of spike height 81 mV. In the primary range, 'control' $k_A = 1.57$ impulses $\text{sec}^{-1} \text{nA}^{-1}$ and 'inhibited' $k_A = 1.46$ impulses $\text{sec}^{-1} \text{nA}^{-1}$. The difference is 9.7 ± 1.65 (s.d.) impulses/sec. In the secondary range, 'control' $k_A = 3.8$ impulses $\text{sec}^{-1} \text{nA}^{-1}$.

In the upper ranges of inactivation the discharge rate diminishes and then it actually benefits from the restorative effect of inhibition upon the membrane potential. Suitably adjusted inhibitory stimulation may then succeed in increasing the firing rate. This is a source of error to watch out for.

Figure 7C has been included to demonstrate diminution of spike height and the break-down of the mechanism of after-hyperpolarization (see legend) in the secondary range, noticeable as an irregularity also in Fig. 7A, but here more clearly visible in spite of the relatively low amplification used. This is the effect found by Kernell (1965*a, b*, 1966) to be a characteristic of firing within the secondary range.

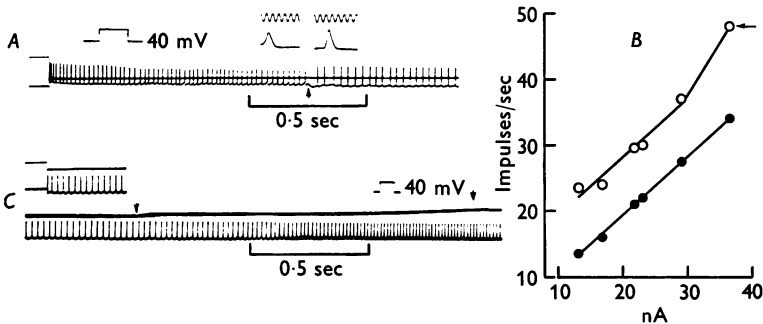


Fig. 7. *A* and *B*, pentobarbitone cat (35 mg/kg). Popliteal motoneurone of spike height 87 mV (2 M potassium citrate electrode) inhibited by high-frequency stimulation of the cut hamstring nerve. In *B*, control and inhibited curves measured for 0.5 sec before and 0.5 sec after onset of nerve stimulation respectively. In the primary range 'control' $k_A = 0.88$ impulses $\text{sec}^{-1} \text{nA}^{-1}$, 'inhibited' $k_A = 0.89$ impulses $\text{sec}^{-1} \text{nA}^{-1}$. Actual records at the current strength which is indicated by an arrow are shown in *A*. Repetitive discharge initiated by trans-membrane current at 36.5 nA, as indicated by downward deflexion of upper trace. High-frequency stimulation of hamstring nerve added about 1.0 sec later, at arrow. Note, increase of spike height during synaptic inhibition. Above, samples of spikes recorded simultaneously at faster sweep speeds (time, 1000 c/s) before and after inhibition. *C*, anaemically-narcotized cat. Gastrocnemius-soleus motoneurone of spike height 78 mV (2 M potassium citrate electrode) facilitated by a 10 mm stretch of the gastrocnemius-soleus muscle. Spikes recorded on upper beam together with current indicator (30 nA), on lower beam with myogram. Lower records begin 1.0 sec after end of upper record. Stretch from zero to 10 mm between arrows. Note, disappearance of after-hyperpolarization when facilitation pushes cell into secondary range with inactivation reducing spike height.

DISCUSSION

The superimposed synaptic effects show that the slope constant in the secondary range is greatly variable depending on the synaptic inflow and that it can be regarded as approximately constant only with respect to the specific conditions of a given experiment. This is the range within which

the motoneurone in the long run can add but little to the strength of the tetanic muscle contraction (Kernell, 1966). Its high spike frequencies would seem better adapted for brief maximal efforts. A high initial firing rate may be useful also as a starting device and leave the motor end-plates in a state of post-tetanic potentiation. An alternative view according to which the secondary range would be an electrical artifact of little physiological importance seems excluded by the experiments presented (cf. also below). With sufficiently large synaptic additions the secondary range is reached with very modest trans-membrane currents producing low basic firing rates. A lesser addition to the discharge rate would then have held the cell to the algebraical summation characteristic of the primary range.

The two concepts used above, i.e. 'ranges' of firing and 'slope constants', have served the major purpose of drawing attention to integrative properties of the firing cell which are essential in any discussion of neural control. From this point of view two disclosures seem particularly important: (i) the perfect algebraical addition of plus and minus terms within the primary range, and (ii) the capacity of superimposed inhibition to stabilize firing rates so as to extend the primary range upwards into higher frequencies. This provides inhibition with a hitherto unknown role as a neural instrument of control and raises the question of by what kind of mechanism synaptic inhibition acts in this context.

In the primary range, stability of firing rate is achieved by the potent negative feed-back mechanism of after-hyperpolarization. In the secondary range the increase of the synaptic excitatory effect may be a consequence of the break-down of this mechanism (Kernell, 1965*a, b*, 1966; cf. also Fig. 7 above). A final explanation of what takes place when a motoneurone is firing within the secondary range cannot be given at the moment, but we shall try to present a brief summary of our own line of reasoning.

Earlier experiments with rat and cat motoneurones (Granit, Kernell & Smith, 1963; Kernell, 1964) showed that antidromic spikes were often followed by delayed depolarization and sometimes by little if any after-hyperpolarization. The delayed depolarization was interpreted as a sign of invasion of the dendrites and this view has since been confirmed by Grampp (1966) studying the slow stretch receptor of the crayfish in which it is possible to record directly from the dendrites (cf. also Purpura & McMurthy, 1965). Granit, Kernell & Smith (1963) found that slight depolarization by injected currents sufficed to make all motoneurones fire with after-hyperpolarization to antidromic stimulation and from this it was concluded that a measure of depolarization facilitates access to the dendrites and that a large activated area, including the latter structures, is required for firing with after-hyperpolarization.

It seems clear that discharging within the secondary range implies

partial inactivation of the cell and it is suggested that this process is brought about directly through heavy depolarization of soma and dendrites. Inactivated dendrites would be incapable of firing at high rates (cf. Grampp, 1966) and thus effectively blocked. For this reason we assume that the essential process in establishing a secondary range of firing is successive blocking of the dendrites with consequent shrinkage of the active area of the motoneurone. On the line of reasoning we have followed, this reduces the after-hyperpolarization and thereby the amount of restitution of the membrane potential. The two factors, blocking of the dendrites and inactivation consequent upon depolarization, are thus interconnected by the need for a large active area in order to maintain firing with restoration of the membrane potential. In the absence of after-hyperpolarization the motoneurone reaches the high rates of discharge of which the secondary range of our curves is an expression. This process can be quickly prevented by an intercurrent inhibition, or, more slowly, by giving the generative mechanism time for the necessary recovery.

The idea that a modest amount of depolarization by injected current favours invasion of the dendrites finds some support in the work of Nelson & Frank (1964). Our view, that at higher current strengths the dendrites are blocked, may seem to imply an unwarranted double action of the depolarizing currents, opening the dendrites at low, closing them at high current strengths, but the same notion has recently been advanced by Grinnell (1966) studying mechanisms of antidromic transmission in frog motoneurones.

The effect achieved by inhibition in the secondary range would follow from the increase in the inhibitory post-synaptic potentials with increasing depolarization (Eccles, 1957). Testing supra-spinal post-synaptic potentials in motoneurones stimulated by injected currents Shapovalov, Kurchavij & Stroganova (1966) found the EPSPs very little influenced by augmentation of current strength while the IPSPs underwent a large increase, as the work of Eccles and his co-workers (1957) has led one to expect. This property of inhibition will automatically counteract inactivation and throw the cell back towards the primary range of firing. The view set forth above would at the same time lead to an expansion of the active area of the motoneurone membrane by giving access to the dendrites. It remains to be found by further experimentation whether inhibition has an additional favourable effect on the preservation of an intact mechanism of after-hyperpolarization as seems likely from the way in which it may expand the primary range upwards.

'Multiplicative behaviour' of units in the inferior colliculus of rats was mentioned by Bureš & Burešova (1965). The neurones were stimulated from the outside and some of them were found to add, others to multiply the effect of this stimulus pitted against their spontaneous activity. In their extracellular type of experiment the two effects could not be related

to events in the cell membrane. However, Shapovalov *et al.* (1966) comparing the effect of injected current with that of superimposed supraspinal inhibition in the manner of our own experiments noted divergence of the two curves. Our results have made such effects dependent upon firing within the secondary range but, as pointed out above, we have used segmental reflexes only.

There are several reports in the literature on high frequencies of discharge from motoneurons beginning with that of Cooper & Denny-Brown (1927) who observed driving at 120/sec from the motor area. Adrian & Bronk (1929) found firing rates of up to 100/sec though values below 50/sec were more common. In flexor reflexes the *d'embrée* opening of the response suggests high initial frequencies and this surmise has proved to be correct (see e.g. Perl, 1962). In intracellular work on flexor motoneurons Wilson & Talbot (1964) recently noted brief initial discharges at rates as high as 400–500 impulses/sec, definitely implicating the secondary range of firing.

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