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AN INVESTIGATION OF PRIMARY OR DIRECT INHIBITION

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In deriving the first curves for the time course of direct inhibition Lloyd (1946) used very weak stimuli for setting up the single inhibitory volleys and hence felt justified in assuming that there was little contamination by group II impulses. The inhibitory action of the group I afferent volley was tested by the depression of a monosynaptic reflex spike which was evoked by a single excitatory volley at various intervals after the inhibitory volley. As the test interval was progressively increased from zero, the inhibition increased rapidly to a maximum at about 0·5 msec test interval and then more slowly declined along a curve that had approximately an exponential decay with a half-time of about 2·8 msec. More recently, Laporte & Lloyd (1951, 1952) have found that a group I inhibitory volley often produces an inhibitory curve that in its decaying phase deviates widely from this exponential form, but they attributed this deviation to the superimposed disynaptic facilitatory action of a group I*b* volley, the assumption being that the uncontaminated group I*a* volley always gave the inhibitory curve decaying exponentially with a half-time of 2·8 msec. However, it appears that only in a few experiments were Laporte & Lloyd able to demonstrate an inhibitory curve having this exponential decay, a disability which they attributed to the great difficulty in discriminating between the thresholds of afferent fibres belonging to groups I*a* and I*b* respectively. While this situation obtains for their field of investigation—the afferent nerves from leg muscles—there is a remarkable threshold discrimination between the group I*a* and I*b* fibres in the afferent nerves from thigh muscles (Bradley & Eccles, 1953*b*). In the present investigation advantage has been taken of this very favourable condition for discriminating between the central actions of impulses in I*a* and I*b* fibres. Single and repetitive volleys in I*a* fibres or in I*a* plus I*b* fibres of quadriceps nerve have been employed to inhibit monosynaptic reflexes by a maximum group I volley in biceps-semitendinosus nerve. The inhibition produced by a pure I*a* volley also proved

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a very favourable situation in which to investigate the nature of the convulsant action of strychnine. A preliminary report of this latter investigation has already appeared (Bradley & Eccles, 1953*a*).

METHOD

The general experimental procedure resembled that previously described (Brock, Eccles & Rall, 1951; Bradley & Eccles, 1953*b*). The animal (cat) was under light pentobarbitone sodium anaesthesia and the spinal cord was severed in the lower thoracic region. With the usual investigation on the inhibitory action of afferent volleys in quadriceps nerve, the testing reflexes were evoked by volleys in the nerve to the antagonist muscles, biceps femoris posterior plus semitendinosus, and recorded in S 1 v.r. (1st sacral ventral root). In some experiments a single volley produced a testing reflex that was too small and variable, so it was enhanced by a small excitatory volley preceding it by about 1·5 msec in the same nerve. So long as this facilitating volley itself failed to produce a significant reflex discharge, it was deemed improbable that it would cause any distortion of the inhibitory curve, and this expectation was borne out by experimental test. For example, single test volleys were employed in the curves of Fig. 2, whereas in Fig. 5 a facilitating volley preceded the test volley by a constant interval of 1·6 msec.

RESULTS

Time course of inhibition for single volley

Typical series of records which have formed the basis of our inhibitory curves are illustrated in Fig. 1. Each record was formed by the superimposition of twenty faint traces so that random variations were fairly effectively smoothed

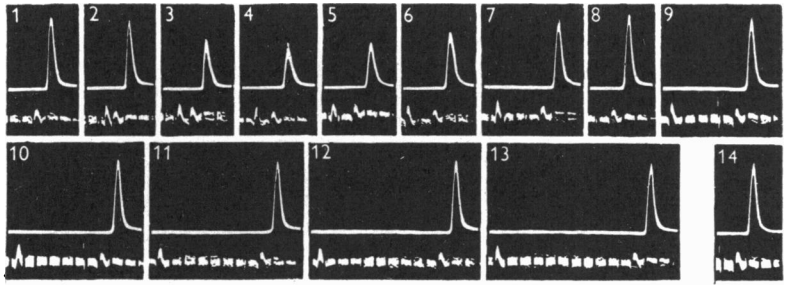

Fig. 1. Inhibition by an afferent volley in quadriceps nerve of monosynaptic reflex discharge evoked in S 1 v.r. by a volley in the nerve to biceps-semitendinosus. Records 1, 8 and 14 show reflex spike evoked by biceps-semitendinosus volley alone; other records show the reflex inhibited by a preceding quadriceps volley at intervals indicated by the dorsal root spikes in the lower records. Stimulus to quadriceps nerve subliminal for group I*b* and only exciting about 40% of group I*a*. Time in milliseconds registered by gaps in lower line.

out when the reflex spikes were measured to the summit height as indicated by the maximum density of the image. The fastest repetition at which a monosynaptic reflex maintains its size is one every three seconds (Eccles & Rall, 1951; Jefferson & Schlapp, 1953), so this frequency has been uniformly adopted in this investigation. Dorsal root volleys have been simultaneously recorded below the ventral root record, so that the volley interval as well as volley size

and composition could be directly assessed (cf. Bradley & Eccles, 1953*b*). A more accurate measure of the mean reflex spike has been secured in our later experiments by employing a device which provided a faster time base for the superimposed records of the reflex spikes and a single trace at slower sweep speed for measurement of the volley interval. This procedure has always been adopted for volley intervals longer than 10 msec. Except for a few preliminary experiments the device of twenty superimposed images has been employed throughout this investigation. Every plotted point in Figs. 2–6 has been so

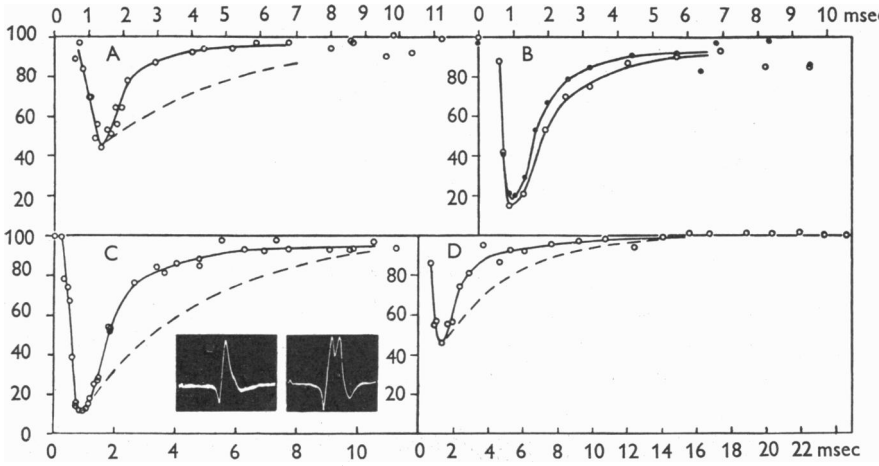

Fig. 2. A: plotting of points from experimental series partly shown in Fig. 1, i.e. for an inhibitory curve generated by a pure group I*a* volley. Ordinates, inhibited reflexes as percentages of control reflex spike; abscissae, intervals between inhibitory and excitatory volleys as recorded by an electrode making contact with rostral part of L6 D.R. as it enters spinal cord. Broken line shows exponential decay from summit with a half-time of 2·8 msec. B: as in A and same experiment, but for open circles inhibitory volley was maximum for group I*a*, group I*b* being negligible, while for filled circles the volley was maximum for both groups I*a* and I*b*. C: as for open circles in Fig. 2B, but in another experiment, the group I*a* volley in quadriceps nerve being virtually maximum and group I*b* negligible. Inset shows action potential of the quadriceps volley employed in contrast to a volley that was maximum for both groups I*a* and I*b*. D: Inhibitory curve as in A, but for another experiment, the inhibitory volley being a pure group I*a* volley of about half maximal size in quadriceps nerve.

derived, and has taken about 1 min to record. Controls of the uninhibited reflex were similarly recorded after every three or four test intervals. Thus the curve of Fig. 2A is derived from 700 responses and Fig. 2C from almost 1000.

In Fig. 2A the plotted points have been derived from the series partly shown in Fig. 1. The inhibitory volley in quadriceps nerve was about 40% maximum, and, so far as could be observed from the triphasic spike in the dorsal root, it had no group I*b* component (cf. Bradley & Eccles, 1953*b*). With the open circles in Fig. 2B the quadriceps volley was maximum for group I*a* and had a very small group I*b* component, while with the filled circles the quadriceps

volley was maximum for both groups I*a* and I*b*. In Fig. 2C, as shown by the inset, there was a very small group I*b* component. The curves deviate from those of Lloyd (1946) and Laporte & Lloyd (1952) in the following features.

The first sign of inhibition occurred with a volley interval of about 0·5 msec in Fig. 2 as against the zero interval reported by Lloyd. This difference is doubtless attributable to the different bases for the measurement of volley intervals. In Fig. 2 the volley interval was measured between the afferent volley spikes as recorded by an electrode on the rostral fraction of L6 D.R. at the locus of its entry into the cord, while the reflex spike was recorded in S1 V.R. Thus the inhibitory volley had a descending intramedullary path of about 2 cm before it exerted its inhibitory action. Presumably the additional delay of 0·5 msec is thus explicable, for in Lloyd's curves the plotted intervals represent actual intervals in the nucleus of the motoneurones that are being inhibited (Laporte & Lloyd, 1952). It has usually been impossible to detect any trace of the descending quadriceps impulses in the biceps-semitendinosus nucleus that they are inhibiting, so this method of measuring testing interval has been impracticable in most of our experiments. However, in several a very small spike has been detected immediately preceding (by about 0·3 msec) the hyperpolarization that is induced in a motoneurone by an inhibitory volley (Coombs, Eccles & Fatt, 1953, unpublished observations). If the inhibitory curve is timed by intervals between this spike and the biceps-semitendinosus spike similarly recorded, zero time in Fig. 2 would be moved almost 1 msec to the right. The first sign of inhibition would occur with the excitatory preceding the inhibitory volley by about 0·5 msec, and maximum inhibition would almost be attained with simultaneous volleys.

After the summit the inhibitory curve in Fig. 2A declined very rapidly, the inhibition being halved at 0·65 msec later, but thereafter it slowed progressively and became very slow indeed after 5 msec. With the larger group I*a* volleys in Fig. 2B the inhibition was greater, but the time courses were similar. Possibly the large I*b* volley in the curve with filled circles caused the slight deviation after about 1 msec from the curve with open circles, in conformance with the disynaptic facilitation described by Laporte & Lloyd (1951, 1952). That effect would be expected to be small in our experiments on account of the depression of synaptic transmission by pentobarbitone. Fig. 2C is the most accurately defined inhibitory curve from our experiments, the successive half-times in milliseconds being 0·95, 0·95, 2·4. 6·1. Figs. 5A_1, A_2 and A_3 show inhibitory curves from another experiment that conform closely to Figs. 2A and B. Finally, in Fig. 2D the inhibition has decayed to zero by about 15 msec and no further development of inhibition can be detected with intervals almost as long as 25 msec. The initial rapid decay (half-time 0·65–1·3 msec) and the progressively slower decay thereafter have occurred in eleven of our twelve experiments where the inhibitory effect of quadriceps volley on biceps-

semitendinosus motoneurones has been carefully plotted. In the remaining experiment the decay was approximately exponential with a half-time of about 1·5 msec. In the ten of these experiments where a pure group I*a* volley was employed, the progressive slowing was invariably observed (cf. Figs. 2A and D, and 5A_1). Thus in our experiments the monosynaptic inhibitory curves for pure group I*a* volleys have shown an initial rapid decline which progressively became much slower. The deviation from the standard curve described by Lloyd (1946) and Laporte & Lloyd (1951, 1952) is revealed in Fig. 2A, C and D, where the broken lines depict this curve decaying exponentially with a half-time of 2·8 msec. In Fig. 2A, and also in Figs. 2D and 5A_1 this deviation is certainly not attributable to the disynaptic excitatory action of a group I*b* volley, and in Figs. 2B (open circles) and 2C it is most improbable that the small contaminating group I*b* volley would have any significant effect.

Time course of inhibition for repetitive volleys

Since it now appears probable that inhibition is mediated by a chemical transmitting substance (Brock, Coombs & Eccles, 1952*a*, *b*; Eccles, 1953), it is important to investigate the possibility of prolonged inhibitory actions attributable to the accumulation and consequent slow dissipation of this hypothetical substance. This has been attempted by investigating the time course of inhibition following repetitive inhibitory volleys. However, only in a relatively few experiments was it possible to employ group I*a* afferent volleys that had no appreciable I*b* admixture. During repetitive stimulation at a frequency sufficiently fast for summation of the inhibitory effects of successive volleys, i.e. above 200 per sec, there is a progressive increase in the threshold of the stimulated fibres; hence it was usually impossible to find a strength of stimulus selective for exciting only group I*a* at the outset (cf. Bradley & Eccles, 1953*b*) and yet strong enough to excite sufficiently large group I*a* volleys for as many as ten stimuli. In some five experiments the inhibitory curve has been determined for volleys which have had throughout little or no contamination by group I*b* impulses. The progressively increasing threshold of the stimulated group I*a* fibres always caused a considerable decline in the group I*a* volley during repetitive stimulation, but these repetitive volleys were always directly recorded so that the degree of this decline could be assessed. Usually the volley size was almost halved by the tenth volley, so this was about the longest tetanus that was practicable.

The inhibitory curves determined in this way have differed very much at the longer intervals. Prolonged inhibition was largest in the experiment of Fig. 3B and persisted for as long as 80 msec. Even after two or four volleys (Fig. 3A) there was a large prolonged inhibition. Unfortunately in this experiment the inhibition after a single volley was not tested at a sufficiently long interval (continuous line, Fig. 3B). Repetitive group I*a* volleys produced

much less prolongation of inhibition in Fig. 4A, but the inhibitory curve (Fig. 4B) resembled that of Fig. 3B when the stimulus was increased so that the inhibitory volleys were maximum for both groups I*a* and *b*. Finally, Fig. 4C shows the least prolonged inhibition (about 15 msec) after repetitive

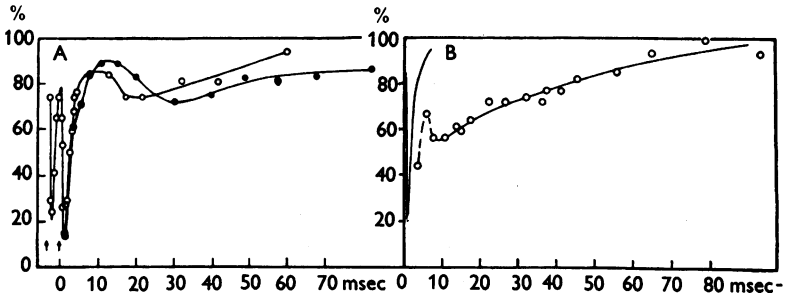

Fig. 3. A: inhibitory curves obtained as in Fig. 2, but subsequent to two (○) and four (●) volleys at 3·6 msec apart. With the 2-volley series the curve is also shown for the first of the two volleys. Both these volleys are marked by arrows, which also signal the last two volleys of the 4-volley series. Volleys submaximal for group I*a* with no detectable group I*b* component. B: same series as A, but for single inhibitory volley (—) and ten inhibitory volleys at 280 per sec (○—○) respectively. Abscissae, intervals after last inhibitory volley.

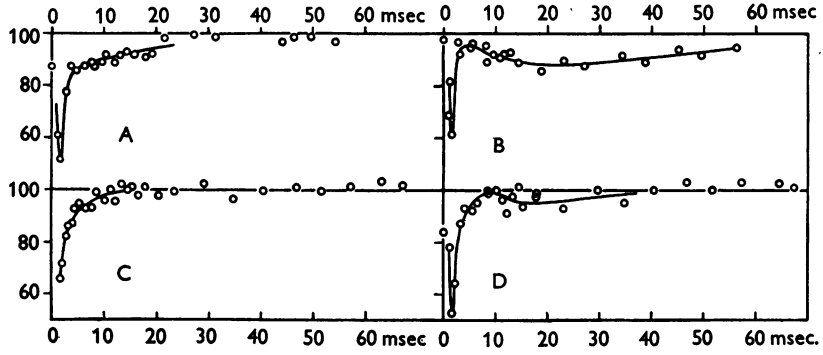

Fig. 4. A and B: inhibitory curves as in Fig. 3B, but in another experiment and after nine inhibitory volleys in quadriceps nerve at 210 per sec. In A the first volley was almost maximum for group I*a* and virtually uncontaminated by group I*b*, but progressive decline occurred during each repetition, the ninth volley being about 60% maximum for group I*a*. In B the repetitive stimuli were 1·7 times stronger so that group I*a* was maximum throughout and group I*b* was almost maximum even with the ninth volley. C and D: as with A and B but in another experiment, the volleys in C corresponding to A and in D to B. Nine volleys at 210 per sec.

group I*a* volleys, but as shown in Fig. 4D the addition of maximum group I*b* volleys caused little sign of prolonged inhibition, though there was probably the same hump just before 10 msec and a trace of the later prolonged inhibition, much as occurred in Figs. 3B and 4B.

In Figs. 3 and 4 it will be seen that, when there was prolonged inhibition, it was usually separated from a brief initial inhibition by a hump. This hump

has usually been observed when there was a considerable admixture of group I*b* in the repetitive volleys (cf. Fig. 4B). It seems probable that such humps would be attributable to the disynaptic facilitatory action of group I*b* fibres (Laporte & Lloyd, 1951, 1952), but in Fig. 3 such an explanation could hardly obtain because no group I*b* component was detectable in the afferent volley (cf. Bradley & Eccles, 1953*b*). In one other experiment, too, repetitive group I*a* volleys evoked an early and late inhibition separated by a hump. The extreme variability in the duration of inhibition following repetitive inhibitory volleys suggests that there is no appreciable accumulation of the inhibitory transmitting substance, for such an effect would be expected to vary little from experiment to experiment. Rather does it seem that the prolonged inhibition is attributable to the continued bombardment of the motoneurones by inhibitory impulses for as long as 50 msec. Such prolonged actions would be vulnerable to the effect of the anaesthetic, pentobarbitone sodium used throughout our experiments, hence perhaps the variability between experiments. It is possible that group I*b* impulses contribute to the prolonged inhibition, and even group II impulses could not be excluded in Fig. 4B, for example, because it would be difficult to distinguish between the thresholds of groups I*b* and II (cf. Brock *et al.* 1951).

Action of strychnine

In Fig. 5 quadriceps afferent volleys were set up by three strengths of stimuli, and A_1, A_2 and A_3 are the inhibitory curves produced by volleys giving the spike potentials shown in the insets, being respectively about half maximum for group I*a*, maximum for group I*a* and maximum for groups I*a* and I*b*. After intravenous injection of 0·09 mg strychnine/kg the inhibition was greatly diminished (B_1 and B_2), and a further diminution occurred after a second injection (0·08 mg/kg in C_1 and C_2). Unfortunately a progressive decrease in the excitability of quadriceps nerve caused the inhibitory volleys to decrease from A_1 to B_1 to C_1 and from A_2 to B_2 to C_2. When allowance is made for this decrease, there is still a large depression of inhibition (cf. Fig. 7A). The maximum I*a* plus I*b* volley was not tested after 0·09 mg strychnine, but in the C_3 curve the initial small inhibitory effect appears to be reversed to facilitation after about 1 msec. The absence of this late facilitation in the C_1 and C_2 curves would suggest that it is due to the group I*b* volley. This explanation has been further tested in Fig. 6 where the inhibitory curve shown in A was changed to curves B and C by dosages of 0·08 and a further 0·12 mg strychnine/kg respectively. The hump in curve A is presumably attributable to the disynaptic facilitatory action of the group I*b* volley (Laporte & Lloyd, 1951, 1952), an effect which progressively became more prominent as the initial inhibition was depressed by strychnine.

The effect of strychnine in diminishing the direct inhibitory action of

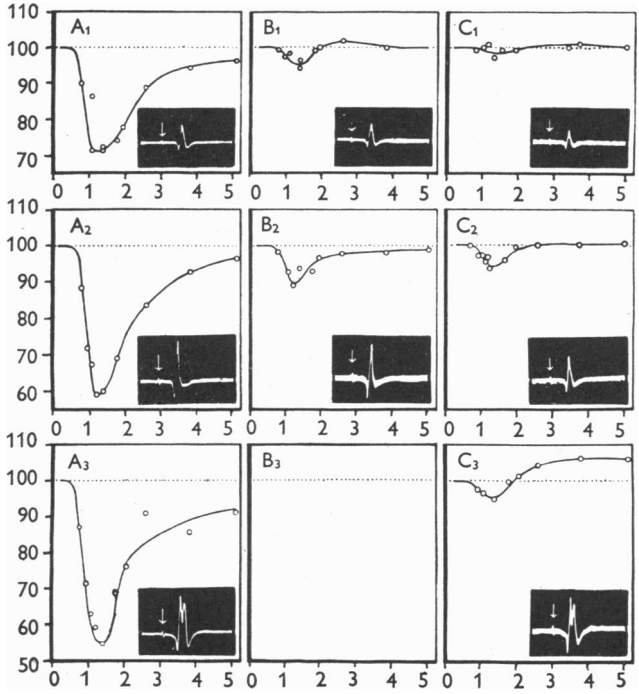

Fig. 5. A_1, A_2 and A_3 are inhibitory curves plotted as in Fig. 2 and for the quadriceps volleys giving the action potentials shown in the insets being respectively about half maximum for group I*a*, maximum for group I*a* and maximum for both groups I*a* and I*b*. Strychnine (0·09 mg/kg) given between series A and B and a further 0·08 mg/kg between B and C. Note progressive decline in size of I*a* spike in two upper series, due presumably to an increase in threshold produced by strychnine. The combination B_3 was not tested.

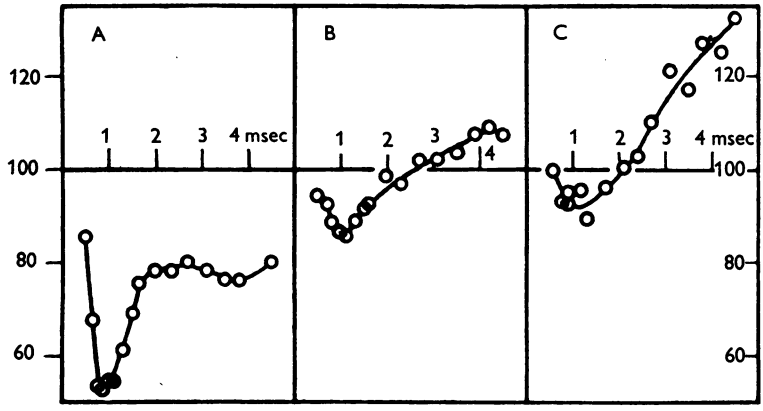

Fig. 6. A, B and C are inhibitory curves for a quadriceps volley maximum for both groups I*a* and I*b*, A being before strychnine and B and C after 0·08 and 0·12 mg strychnine/kg respectively, i.e. corresponding to A_3, B_3 and C_3 of Fig. 5.

group I*a* afferent impulses has been observed in all our experiments (fifteen in all), while in nine experiments the secondary facilitation (presumably of group I*b* impulses) became more prominent as the inhibition was depressed. The size of the control value of the testing reflex spike was of importance in evaluating the significance of the changes in the inhibitory curves. If large changes had occurred, the inhibitory effect would have been signalled by quite different populations of motoneurones, which would at least reduce the

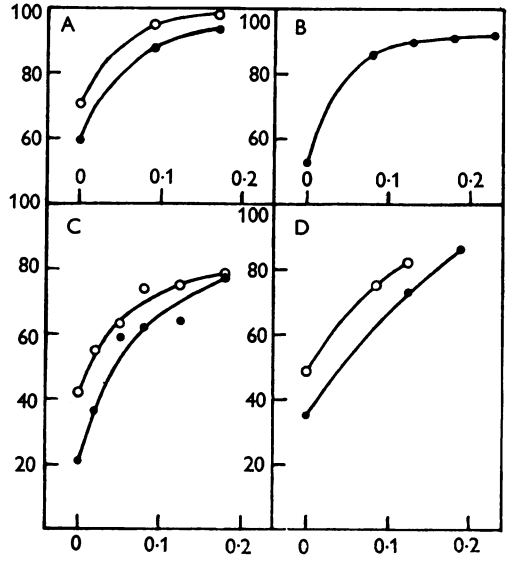

Fig. 7. Plots of test reflexes taken at the interval after the inhibitory volley giving maximum effect (cf. Figs. 5 and 6) and expressed as percentages of control test reflexes (ordinates) against dosages of strychnine (mg/kg body weight: abscissae). A–D show four experiments (A is from Fig. 5 A_1, B_1, C_1 and A_2, B_2, C_2), in three of which two response-dosage curves are drawn, that with open circles being for a submaximum group I*a* inhibitory volley with negligible I*b* contamination, while with filled circles I*a* is maximum and usually significantly contaminated with group I*b*. The decrease produced by strychnine in the size of the submaximum inhibitory volley (cf. Fig. 5) is approximately allowed for in A by proportionally increasing the inhibition.

significance attaching to quantitative changes. However, in some experiments, e.g. those of Figs. 7A, B and D, strychnine produced virtually no change (less than 10%) in the size of the testing reflex spike, while in others there was a relatively small diminution, e.g. to about 65% for the whole range of dosage in Fig. 7C, hence our curves should suffer little distortion by this complication.

Our most extensive dose-response curves are shown in Fig. 7, corrections being made for any changes that occurred in the size of the inhibitory volley (cf. Fig. 5). Attempts as in Figs. 7B and C to record the effect of a more finely graded dosage were distorted considerably in their later stages by the pro-

gressive decay of the earlier doses, i.e. with the heavier doses the curves should have approached much closer to the 100% line. A distortion in the opposite direction may be introduced by the secondary facilitation of group I*b* impulses (cf. Fig. 6B and C). Since this facilitation apparently was not depressed by strychnine, it would be relatively much more effective in antagonizing the inhibition, though it is doubtful if its onset was sufficiently early to affect the peak of the inhibitory curve. In summary, Fig. 7 shows that a subconvulsive dose of strychnine (0·08 mg/kg) suppressed the direct inhibitory action by more than half, while with the heaviest dosage (up to 0·20 mg/kg) very little inhibition survived.

Preliminary experiments have shown that strychnine also depressed the secondary inhibition exerted by muscular and cutaneous afferent impulses operating through polysynaptic paths, but often this depression was relatively less than with direct inhibition. Further investigation is desirable.

DISCUSSION

The deviation of our inhibitory curves from the exponential form of decay cannot be ascribed to distortion produced by the method of sampling that is implicit in the construction of an inhibitory curve. The testing reflex responses occurred in a considerable fraction of the biceps-semitendinosus motoneurones, and even at maximum inhibition a considerable fraction of motoneurones still responded in such curves as Figs. 2A and $5A_1$. Under such conditions the inhibited population of motoneurones would be distributed over an approximately linear range of reflex excitation (Lloyd, 1945; Rall, 1952), and the number inhibited would be approximately in direct proportion to the intensity of inhibition, i.e. distortion would be negligible. It should further be pointed out that in such curves as Figs. 2C and $5B_1$, where a very deep inhibition could be introducing considerable distortion through sampling by the responses of only a small fraction of motoneurones remote from the linear range, the deviation so introduced would cause the change in inhibition to be more than the change in reflex response, i.e. the decay of the inhibitory process would be actually steeper than is indicated by inhibitory curves such as Figs. 2C and $5B_1$. It is therefore justifiable to assume that our inhibitory curves plot approximately the time course of the inhibitory process on a motoneurone and show that the decay of this process usually deviates considerably from the exponential form.

Laporte & Lloyd (1951, 1952) were able in some experiments to plot an inhibitory curve which decayed exponentially with a time constant of about 4 msec (cf. Lloyd, 1946). It appears that in most of their experiments this was not possible, and even the smallest inhibitory volleys produced inhibitions which decayed from the summit with time courses similar to those of Figs. 2A, D and $5A_1$. They attributed this deviation from the exponential form of decay

to the disynaptic excitatory action of impulses in group I*b* fibres, which had usually the same threshold range as the group I*a* fibres and which acted as disynaptic excitors of antagonist motoneurones. Doubt can now be thrown on this explanation, at least in its entirety, because the inhibition generated by a pure group I*a* volley usually showed just such a deviation from the presumed exponential curve (cf. Figs. 2A, C and D). Doubtless the disynaptic excitatory actions of group I*b* impulses also contributed to the deviation from exponential at least in some of the curves of Laporte & Lloyd (cf. Figs. 2B, filled circles, and Fig. 6A above), but, in the absence of direct demonstration of an exponentially decaying inhibitory curve (only available in their fig. 13), it is no longer justifiable to extrapolate an anticipated inhibitory curve from the summit (as in Figs. 2A, C and D above), and ascribe all the deviation therefrom to disynaptic excitatory action (cf. figs. 11, 12 and 14 of Laporte & Lloyd, 1952).

In investigating the nature of the inhibitory process, direct or primary inhibition is of particular significance because there is good evidence that it is generated directly on the motoneurone by the virtually synchronous action of impulses of the group I*a* volley (Lloyd, 1941, 1946; Laporte & Lloyd, 1952; Brock *et al.* 1952*a*, *b*; Eccles, 1953). Thus the time course of direct inhibition would give a mean index of the development and decay of the inhibitory process generated by a single inhibitory impulse operating at a single inhibitory synapse. It is, for example, probable that the more prolonged rise and decline of secondary inhibition is attributable to a prolonged temporal dispersion of the actual inhibitory bombardment of the motoneurones.

There is now good evidence that both direct and secondary inhibition are caused by the hyperpolarization which inhibitory impulses induce in motoneurones (Brock *et al.* 1952*a*, *b*; Eccles, 1953). The hyperpolarization associated with direct inhibition showed an approximately exponential decay with a time constant of 2·2–7 msec (mean about 4). There is thus a problem in explaining how this exponentially decaying hyperpolarization could account for the initially fast and later slowly decaying curves of direct inhibition. A possible explanation is that the inhibitory synaptic knobs are not distributed uniformly over the motoneurone, but are concentrated strategically close to the excitatory knobs whose action they inhibit. Under such conditions, in the region where inhibitory action was occurring, the inhibitory hyperpolarization would run a time course deviating from the exponential decay in just the way that has been observed for the inhibitory curve (Fig. 2A). The exponential decay would obtain for the hyperpolarization as a whole, as is observed with the intracellular electrical record, but not close to the foci where it was generated, as is excellently illustrated by the depolarization of the end-plate potential recorded at the motor end-plate (cf. Fatt & Katz, 1951, fig. 8), which initially decayed along a time course much faster than the electric time constant of the

membrane. On the other hand, if there were a completely random distribution of excitatory and inhibitory knobs over the surfaces of each motoneurone, the inhibitory curve would be expected to conform to the exponential form, decay occurring with the electric time constant (about 4 msec) of the membrane, as is sometimes observed (cf. Lloyd, 1946; Laporte & Lloyd, 1952).

Sherrington (1905) first described and later analysed (Owen & Sherrington, 1911) the reversal which strychnine produced in spinal reflexes, an inhibitory reflex thereby becoming excitatory. It appeared that this effect was the cause of the convulsant action of strychnine. Sherrington recognized that all the afferent volleys which he employed had mixed excitatory and inhibitory actions on the motoneurones of the test muscle. Hence he proposed two explanations, but was unable to distinguish between them at that time: that the excitatory impulses were rendered prepotent by strychnine favouring their central action and depressing the inhibitory; that strychnine actually 'transformed the process of central inhibition into one of central excitation'. Subsequent investigations have been concerned with attempts to distinguish between these two alternatives by employing afferent inputs in which the inhibitory impulses were as uncontaminated as possible by excitatory. In convincing experiments Liddell & Sherrington (1925) found that a subconvulsive dose of strychnine (0·08 mg/kg) did not reverse the inhibitory effect which was exerted on quadriceps motoneurones by stretching biceps muscle. The inhibition was made relatively a little smaller. Similarly, Cooper & Creed (1927) found that strychnine did not reverse the inhibitory effect exerted on quadriceps motoneurones by the afferent impulses set up by the active contraction of the hamstring muscles. There have been, in addition, several other reports of the failure of strychnine to reverse inhibitory actions which were believed to be virtually uncontaminated by excitatory (Magnus & Wolf, 1913; Bremer, 1925; Creed & Hertz, 1933; Denny-Brown, 1932; Bremer & Bonnet, 1942). Thus there has been convincing experimental evidence that strychnine does not transform central inhibition into central excitation (cf. the comprehensive review by Bremer, 1944). The general conclusion has been that the convulsant action of strychnine is attributable to the enhancement which it produces in synaptic excitatory action.

However, there has been neglect of another possibility suggested by Owen & Sherrington (1911), namely that strychnine reversal may be caused at least in part by a depression of the central inhibitory process. Doubtless this neglect is attributable to the large inhibitory effects evoked in the strychninized nervous system by a relatively pure inhibitory input (cf. Liddell & Sherrington, 1925). On the other hand, the simplest excitatory reflexes (the monosynaptic) are also very little affected by strychnine in subconvulsive doses, being often not significantly changed (cf. above), though there may be a small decrease or increase, effects which contrast with the large increase always produced in

the polysynaptic reflexes (Hoffmann, 1922; Kaada, 1950; Bradley & Schlapp, 1950; Naess, 1950; Bernhard, Taverner & Widén, 1951; Scherrer, 1952; Brooks & Fuortes, 1952). Thus the simple hypothesis that strychnine causes an enhancement of all synaptic excitatory action must be rejected.

The attempts to formulate explanations of the various changes which strychnine causes in the activity of the central nervous system will now be re-examined in relationship to the simplest effect it is known to have, namely depression of direct inhibition (cf. Figs. 5–7). How far is it possible to explain its various actions as resulting from a general depression of the central inhibitory process? Some illustrative examples will suffice to show that this simple explanation is plausible.

Monosynaptic reflexes would be changed only in so far as there was a change in the background barrage of impulses on motoneurones. A general depression of inhibition would be expected to increase this barrage, hence the increase sometimes observed in the reflex. In our experience depression of monosynaptic reflexes only comes on at a late stage or after high dosage and may be ascribable to deterioration of the condition of the animal.

The large increase in polysynaptic reflexes (Kaada, 1950; Bradley & Schlapp, 1950; Naess, 1950; Scherrer, 1952) would be attributable to the depression of inhibitory control at each successive synaptic relay in the reflex pathway. For example, cutaneous afferent impulses have mixed excitatory and inhibitory actions on both flexor and extensor motoneurones (Hagbarth, 1952; Eccles, 1953). Depression of the inhibitory actions would cause increase in the flexor reflex, together with reversal of the inhibition usually exerted on extensor motoneurones (Owen & Sherrington, 1911), and in polysynaptic reflexes this effect would be cumulative at each stage of synaptic relay.

In comparison with direct inhibition, indirect or secondary inhibition appears to be less depressed by strychnine or even enhanced (cf. Bremer & Bonnet, 1942), and thus the possibility arises that strychnine selectively depresses direct inhibition. However, the depression of indirect inhibition is exerted on motoneurones through polysynaptic paths, i.e. by way of several excitatory synaptic relays. If it is assumed that each of these synaptic relays on polysynaptic inhibitory paths is the site of a release by strychnine from inhibitory depression (cf. above), the total inhibitory bombardment of motoneurones would be intensified and hence the indirect inhibition may be but little decreased or even increased despite the depression of the inhibitory effect exerted by any one impulse. Thus any changes produced in indirect inhibition must be interpreted with caution.

Since there is now conclusive evidence against the Golgi-cell hypothesis of central inhibition (cf. Brock *et al.* 1952*a*, *b*; Eccles, 1953), the effect of strychnine need be discussed only in relationship to the action of an inhibitory synaptic transmitter. In view of the rapid onset of the strychnine action

(a very few seconds), the most probable explanation is that strychnine and the inhibitory transmitter compete for the same steric configurations on the inhibitory post-synaptic membrane, i.e. that the action is analogous to that generally believed to occur for curarine at the motor end-plate. Alternatively, strychnine could diminish the hyperpolarization produced by inhibitory impulses by acting further downstream, e.g. by antagonizing in some way the outward movement of cations which has been suggested as the cause of the inhibitory hyperpolarization (cf. Eccles, 1953). Whatever the eventual explanation of its action, it is evident that strychnine will provide a valuable means of investigating the nature of central inhibition.

SUMMARY

1. Direct inhibition has been investigated by using afferent volleys from the cat's quadriceps nerve to inhibit monosynaptic reflex discharges from biceps-semitendinosus motoneurones. A single inhibitory volley in group I*a* fibres only (the afferent fibres from muscle spindles) exerts an inhibitory action which decays with a time course that is very different from the exponential form with a time constant of about 4 msec, being initially much faster and later slower. As a consequence much of the evidence for the disynaptic facilitatory action of group I*b* impulses is no longer acceptable. The relationship of the actual decay curve to the electric time constant of the neuronal membrane is discussed.

2. In some experiments repetitive inhibitory volleys were followed by an inhibition persisting for as long as 80 msec, but usually this effect is not attributable to group I*a* fibres. There is thus no good evidence for the accumulation and persistence of the inhibitory transmitter substance that is assumed to cause direct inhibition.

3. In subconvulsive doses, strychnine depressed selectively direct inhibitory action, 0·08 mg/kg reducing it to about one-third, while monosynaptic excitatory action was virtually unaffected. Larger doses almost abolished the inhibition. Strychnine also depressed polysynaptic or indirect inhibition. It is suggested that the convulsant action of strychnine is caused by this selective depression of inhibition. The possible ways in which strychnine could exert this depression are discussed.

REFERENCES

BERNHARD, C. G., TAVERNER, D. & WIDÉN, L. (1951). Differences in the action of tubocurarine and strychnine on the spinal reflex excitability of the cat. *Brit. J. Pharmacol.* **6**, 551–559.

BRADLEY, K. & ECCLES, J. C. (1953*a*). Strychnine as a depressant of primary inhibition. *Nature, Lond.*, **171**, 1061–1063.

BRADLEY, K. & ECCLES, J. C. (1953*b*). Analysis of the fast afferent impulses from thigh muscles. *J. Physiol.* **122**, 462–473.

BRADLEY, K. & SCHLAPP, W. (1950). Effect of strychnine on spinal reflexes of decapitated cats. *J. Physiol.* **111**, 62*P*.

BREMER, F. (1925). Recherches sur le mécanisme de l'action de la strychnine sur le système nerveux. I. La strychnine et les phénomènes d'inhibition. *Arch. int. Physiol.* **25**, 131–152.

BREMER, F. (1944). Le mode d'action de la strychnine à la lumière de travaux récents. *Arch. int. Pharmacodyn.* **69**, 249–264.

BREMER, F. & BONNET, V. (1942). Contributions à l'étude de la physiologie générale des centres nerveux. II. L'inhibition réflexe. *Arch. int. Physiol.* **52**, 153–194.

BROCK, L. G., COOMBS, J. S. & ECCLES, J. C. (1952*a*). The recording of potentials from motoneurones with an intracellular electrode. *J. Physiol.* **117**, 431–460.

BROCK, L. G., COOMBS, J. S. & ECCLES, J. C. (1952*b*). The nature of the monosynaptic excitatory and inhibitory processes in the spinal cord. *Proc. Roy. Soc.* B, **140**, 170–176.

BROCK, L. G., ECCLES, J. C. & RALL, W. (1951). Experimental investigations on the afferent fibres in muscle nerves. *Proc. Roy. Soc.* B, **138**, 453–475.

BROOKS, C. MCC. & FUORTES, M. G. F. (1952). Potential changes in spinal cord following administration of strychnine. *J. Neurophysiol.* **15**, 257–267.

COOPER, S. & CREED, R. S. (1927). More reflex effects of active muscular contraction. *J. Physiol.* **64**, 199–214.

CREED, R. S. & HERTZ, D. H. (1933). The action of strychnine on Hering-Breuer reflexes. *J. Physiol.* **78**, 85–95.

DENNY-BROWN, D. (1932). Quoted in CREED, R. S., DENNY-BROWN, D., ECCLES, J. C., LIDDELL, E. G. T. & SHERRINGTON, C. S., *Reflex Activity of the Spinal Cord*, p. 83. Oxford: Clarendon Press.

ECCLES, J. C. (1953). *The Neurophysiological Basis of Mind.* Oxford: Clarendon Press.

ECCLES, J. C. & RALL, W. (1951). Effects induced in a monosynaptic reflex path by its activation. *J. Neurophysiol.* **14**, 353–376.

FATT, P. & KATZ, B. (1951). An analysis of the end-plate potential recorded with an intracellular electrode. *J. Physiol.* **115**, 320–370.

HAGBARTH, K. E. (1952). Excitatory and inhibitory skin areas for flexor and extensor motoneurones. *Acta physiol. scand.* **26**, Suppl. 94, 1–58.

HOFFMANN, P. (1922). *Untersuchungen über die Eigenreflexe (Sehnenreflexe) menschlicher Muskeln.* Berlin: Springer.

JEFFERSON, A. A. & SCHLAPP, W. (1953). Some effects of repetitive stimulation of afferents on reflex conduction. In *The Spinal Cord*, pp. 99–117. Ciba Foundation Symposium. London: Churchill.

KAADA, B. R. (1950). Site of action of myanesin (mephenesin, tolserol) in the central nervous system. *J. Neurophysiol.* **14**, 89–114.

LAPORTE, Y. & LLOYD, D. P. C. (1951). Disynaptic reflex linkage between muscles and a myotatic unit. *Fed. Proc.* **10**, 78–79.

LAPORTE, Y. & LLOYD, D. P. C. (1952). Nature and significance of the reflex connections established by large afferent fibres of muscular origin. *Amer. J. Physiol.* **169**, 609–621.

LIDDELL, E. G. T. & SHERRINGTON, C. S. (1925). Further observations on myotatic reflexes. *Proc. Roy. Soc.* B, **97**, 267–283.

LLOYD, D. P. C. (1941). A direct central inhibitory action of dromically conducted impulses. *J. Neurophysiol.* **4**, 184–190.

LLOYD, D. P. C. (1945). On the relation between discharge zone and subliminal fringe in a motoneuron pool supplied by a homogeneous presynaptic pathway. *Yale J. Biol. Med.* **18**, 117–121.

LLOYD, D. P. C. (1946). Facilitation and inhibition of spinal motoneurones. *J. Neurophysiol.* **9**, 421–438.

MAGNUS, R. & WOLF, C. F. L. (1913). Weitere Mitteilungen über den Einfluss der Koffstellung auf den Gleidertonus. *Pflüg. Arch. ges. Physiol.* **149**, 447–461.

NAESS, K. (1950). The effect of D-tubocurarine on the mono- and polysynaptic reflex of the spinal cord including a comparison with the effect of strychnine. *Acta physiol. scand.* **21**, 34–40.

OWEN, A. G. W. & SHERRINGTON, C. S. (1911). Observations on strychnine reversal. *J. Physiol.* **43**, 232–241.

RALL, W. (1952). Input-output relation of a monosynaptic reflex arc. *Proc. Univ. Otago med. Sch.* **29**, 17–18.

SCHERRER, J. (1952). Action de strychnine sur l'activité spinale réflexe. *J. Physiol. Path. gén.* **44**, 29–44.

SHERRINGTON, C. S. (1905). On reciprocal innervation of antagonistic muscles. Eighth note. *Proc. Roy. Soc.* B, **76**, 269–297.