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SUPRASPINAL INFLUENCES ON SPINAL MECHANISMS ACTIVATED PRIOR TO LEARNED MOVEMENT

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Abstract. Dogs were trained to perform an instrumental avoidance reflex of lifting and holding a limb above a pre-determined level to prevent or to interrupt painful stimulation of the same limb. A tone preceding the stimulation by 0.5 sec served as the CS. During the CS the excitability of the motoneuronal pools involved in the execution of the instrumental motor response was monitored as the magnitude of the response of knee flexors to single stimuli applied to sciatic nerve. For 70–80 msec preceding the onset of the EMG of the conditioned instrumental movement, the amplitude of the muscle response progressively increased. This phenomenon is apparently analogous to that of "motor presetting" described by others for man. Bilateral pyramidotomy at the medullary level was performed following the elaboration of instrumental reflexes. It produced obvious changes in the motor performance: the latencies of conditioned movements became longer and the speed of limb lifting decreased. It did not, however, alter the preparatory increase in motoneuronal excitability. On the other hand pyramidotomy prior to elaboration of the conditioned reflex strongly affected the preparatory changes, Destruction of the red nucleus contralateral to the conditioned limb in pyramidotomized dogs resulted in disappearance for 1.5 months of the conditioned motor response to the CS. After the response had been restored, the latency of movement showed a further increase, and the presetting of the spinal system was essentially absent.

INTRODUCTION

The present experiments were designed to study the role of the pyramidal and rubrospinal systems in the alteration of excitability which occur in motoneurons in association with the performance of a learned movement. Gurfinkel and Kots (6) in experiments on man have shown that before the beginning of voluntary movements the excitability of the motoneuronal pool of the agonist muscles, measured by the amplitude of the monosynaptic H-reflex, progressively increased during the last 60–70 msec before the onset of activity recorded in the EMG. It was suggested that this alteration of motoneuronal excitablity was caused by supraspinal influences. It was of considerable interest to test this hypothesis in experiments with animals, comparing the course of fluctuations in efferent excitability in normal animals and in animals with pyramidal or rubral lesions.

MATERIAL AND METHODS

Experiments were done on nine dogs trained to perform local avoidance reflexes: to lift and to hold the right hind limb in order to prevent or to remove electrical stimulation applied to the same limb for 4.5 sec. Conditioned stimulus (CS) was a tone of 90 db which preceded the onset of the unconditional stimulus (UCS) by 0.5 sec and continued throughout the period of possible electrocutaneous stimulation.

The excitablity of the motoneuronal pool involved in the execution of the conditioned movement was tested through the magnitude of the response of flexors of the knee (*m. semitendinosus* or posterior portions of *m. biceps femoris*) to a stimulation of the sciatic nerve. In some cases the response of the gastrocnemius to stimulation of the tibial nerve was recorded. Bipolar stimulation and electromyographic (EMG) recording was accomplished with nichrome wire electrodes of 100–200 μ diameter, inserted into the nerve or muscle (1). Detailed description of the method is given elsewhere (23). Single shocks, 0.1–1.0 msec, were generated by an ESU-1 stimulator or from the stimulator of the "Medicor" electromyograph. The amplified muscle response was displayed on an oscilloscope (Fig. 1) and also fed into an ink recording device (Fig. 2).

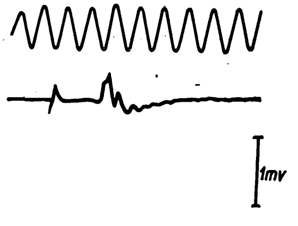

Fig. 1. Reflex response of *m. semitendinosus* to stimulation of sciatic nerve; upper trace, time 200 Hz.

Stimuli were delivered to the nerve at different times after the CS, during the latency of the conditioned EMG. The amplitude of a muscle response to nerve stimulation was measured from the photographic re-

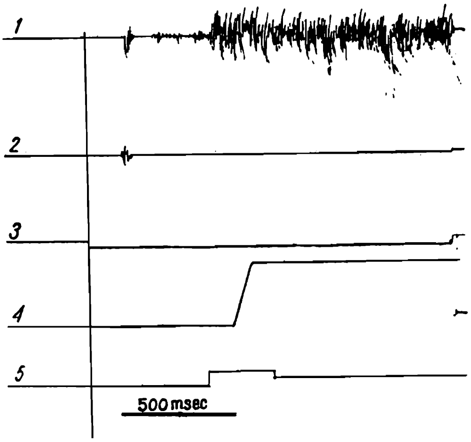

Fig. 2. Partial records. *1*, electromyogram of *m. semitendinosus; 2*, nerve stimulation; *3*, conditional stimulus; *4*, mechanogram; *5*, electrocutaneous stimulation.

cord and was expressed in per cent of the mean baseline amplitude of muscle responses in a given experiment obtained in the intervals between CS presentations. The time between the muscle response and onset of the EMG of the conditioned movement was measured from the ink records. For statistical treatment of the data, Wilkinson's and Chi-square tests were used (22).

The bulbar pyramids were sectioned by Starlinger's method (19) usually 2–3 mm below the caudal margin of the trapezoid body. The red nuclei were coagulated by the positive current of 5 ma for 40–60 sec via an electrode 0.3 mm in diameter with 0.5 mm exposed tip. The coordinates of red nuclei were determined from the atlas of Lim, Liu and Moffitt (15). Flexion of the contralateral hindlimb to rubral stimulation was used to locate the proper position of the electrode in the red nucleus (4, 8). Terminally the animals were perfused with saline, followed by 10% formalin, and the position of the lesion verified histologically.

RESULTS

Muscle responses to nerve stimulation

When the sciatic nerve was stimulated, two kind of synchronized responses were obtained from the knee flexors. Stimuli at or near threshold usually elicited only one response with a latency of about 10 msec. This response disappeared following section of the nerve central to the stimulation site, thus suggesting its reflex nature. With enhanced stimulation, the amplitude of the response increased and there appeared a direct motor response with a latency of about 3–5 msec. With a further increase of stimulation the amplitude of the direct response increased; at the same time the reflex response, reached its maximum and began to decrease. This pattern of muscle response to nerve stimulation is similar to that observed in man. In the present experiments threshold or near-threshold stimuli were used which, in the resting state, elicited a reflex response of 10–20% of the maximal value. The animal manifested no sign of pain or discomfort with this level of stimulation.

Effect of nerve stimulation on conditioned reflex responses

The experiments showed that the nerve stimulation per se was not indifferent for the time of appearance of the conditioned EMG. The latency of the conditioned EMG, as shown in Fig. 3*A*, became much shorter when the CS was followed by test stimulation of the nerve. Detailed study of this phenomenon revealed the dependence of the effect of nerve stimulation on the time of stimulation. When the stimulation was applied in the first 30–50 msec of the CS interval, it did not usually alter the

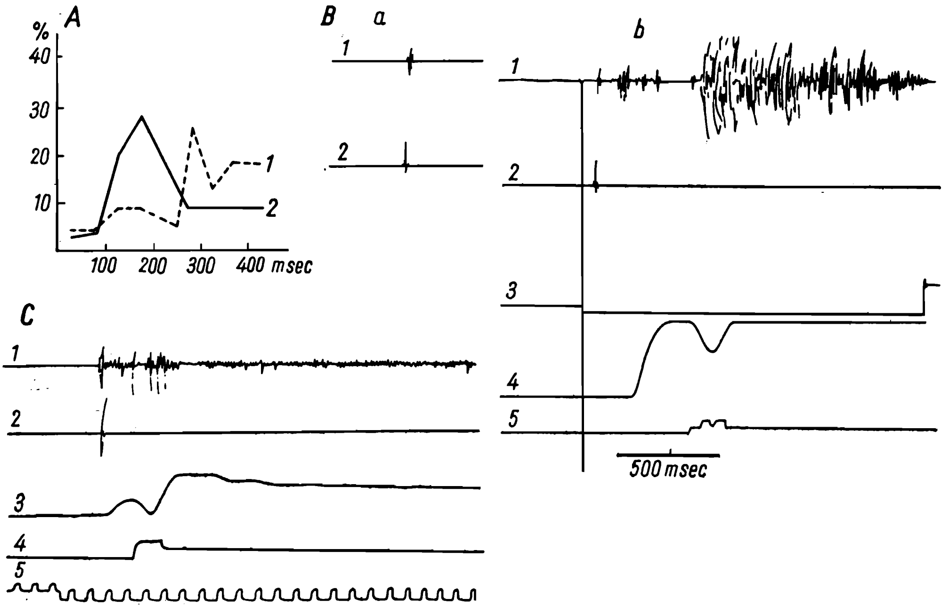

Fig. 3. Response to nerve stimulation before onset of EMG. *A*, distribution curves of latencies of conditioned reflex EMG (in msec); *1*, without nerve stimulation; *2*, with nerve stimulation. *B*, response to nerve stimulation; *a*, in interval between conditioning trials; *b*, during latent period of conditioned reflex EMG. "Tail" of desynchronized EMG seen after reflex response. Designations as in Fig. 2. C, appearance of EMG immediately after reflex response to the nerve stimulation. *1*, *2*, as in Fig. 2; *3*, mechanogram; *4*, electrocutaneous stimulation; *5*, time (0.1 sec) and conditioned stimulus.

latency of the conditioned reflex EMG. However, when the stimulation was applied closer to the time of the occurrence of the conditioned EMG, the reflex response not infrequently was followed by the "tail" of a desynchronized EMG (Fig. 3*B*). Sometimes this EMG was accompanied by a slight movement of the limb. A conditioned reflex EMG and a conditioned movement in these cases occurred after a pause. With a further shortening of the interval between nerve stimulation and onset of the EMG, the phenomenon of "attraction" was observed in a number of cases, i.e. the conditioned reflex EMG occurred just after the reflex response (Fig. 3*C*).

However, these effects were not invariable: occasionally, nerve stimulation prolonged the latency of the conditioned reflex EMG instead of shortening it.

Changes in the amplitude of reflex muscle responses prior to conditioned reflex movement

The setting of the motoneuronal pools prior to the avoidance movement was studied in five normal dogs. These experiments showed that 120 msec or even longer before the occurrence of the conditioned reflex EMG, the magnitude of the reflex muscle response to nerve stimulation on the average increased twofold. During the last 60–80 msec before onset of the EMG, the response amplitude showed a furher increase (Fig. 4).

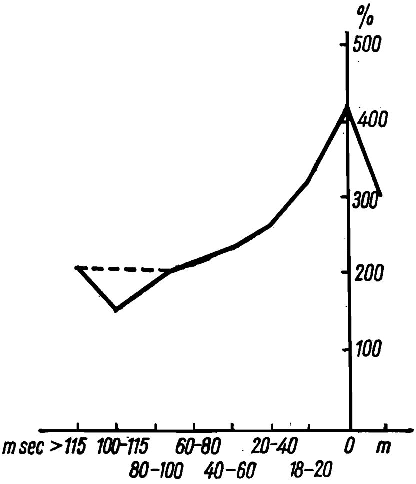

Fig. 4. Magnitude of reflex response to nerve stimulation prior to onset of conditioned movement in dog "Kiy". Abscissa, interval to beginning of movement in msec (*m*, during the movement). Ordinate, magnitude of reflex response in per cent of values at rest.

The absolute value of the increases varied from one dog to another reaching an average amplitude from 350–400% in some dogs to 900% in others, relative to the baseline amplitude, but statistically significant in all animals. When the movement had already started, the average amplitude of the reflex response decreased somewhat. However, changes in reflex response at different intervals after onset of movement were not analyzed in detail.

In order to define whether the increase of motoneuronal excitability was wholly associated with the onset of conditioned movement or was also partly associated with a non-specific effect of the acoustic signal used as a CS, changes in amplitude of the reflex muscle response were studied in two naive dogs using the same acoustic stimulation as in the experiment with conditioned reflexes. The results are presented in Fig. 5.

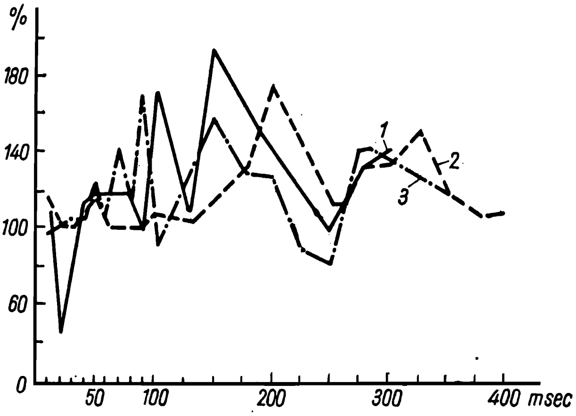

Fig. 5. Magnitude of reflex response to nerve stimulation during nonsignificant acoustic stimulation. *1*, dog "Chernaja", first 500 presentations of sound; *2*, same dog, the next 500 presentations; *3*, dog "Topka", all presentation. Abscissa, time after onset of sound stimulus; ordinate, reflex response in per cent of control level.

Several increases in motoneuronal excitability with maximal amplitudes of 180–200% of the baseline level were observed in the 400 msec interval after onest of acoustic stimulation. After a number (500 and more) of presentations of the tone, the early increases were reduced, but the magnitude of the late waves with the peak at 200 msec or more remained unchanged. Taken together these results indicate that an early (120 msec or more before onset of the conditioned reflex EMG) increase of motoneuronal excitability represents a non-specific component of the response to the auditory signal. The magnitude of this increase was nearly the same as the response to a sound devoid of signal meaning.

In the presence of this early increase of excitability there occurred a further rise of excitability during the period immediately preceding the movement which significantly differred from the early increase and appears to be specifically associated with the onset of movement.

Presetting of spinal cord excitability in pyramidotomized dogs

Section of the bulbar pyramids resulted in certain general motor disturbances similar to those described elsewhere (3, 5, 16, 19). Avoidance responses in the form of hind limb lifting and holding were observed as early as 8–12 days after the operation and could be elaborated again. The tonic form of this relatively simple motor response persisted but its latency noticeably increased and the speed of limb lifting decreased.

The presetting in pyramidotomized dogs was different, depending on whether the pyramidal section was performed prior to or after the conditioned avoidance reflex had been established. In three dogs with preoperatively elaborated reflexes pyramidotomy had no significant effect on the presetting of the motoneuronal pool. The pre-movement increase of segmental excitablity was not significantly different from the preoperative increase (Fig. 6). A prolonged (8–16 months) interval did not alter the general shape of the presetting curve, although the increase in excitability during the last 80 msec before movement was somewhat less.

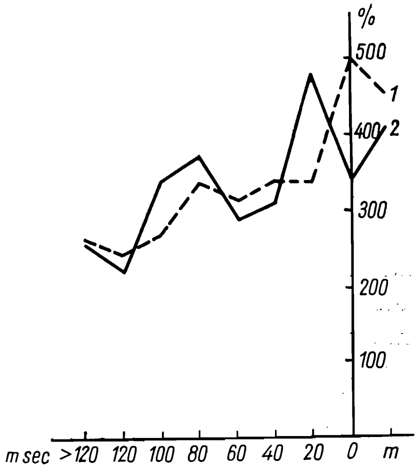

Fig. 6. Magnitude of reflex response prior to beginning of movement before (*1*) and after (*2*) section of bulbar pyramids (dog "Dick"). Designations as in Fig. 4.

In four dogs with conditioned reflexes elaborated after pyramidotomy the curve of pre-movement alteration of excitability was significantly different from that in normal dogs. According to the pattern of segmental setting, these dogs could be divided into two groups, two dogs in each (Fig. 7). The dogs of group I showed an early increase of motoneuronal excitability which peaked 50–60 msec prior to onset of the EMG. During the last 30–40 msec prior to onset of the conditioned EMG, the excitability showed some decrease (Fig. 7 *2*).

The time-course of excitability changes in the pyramidotomized dogs

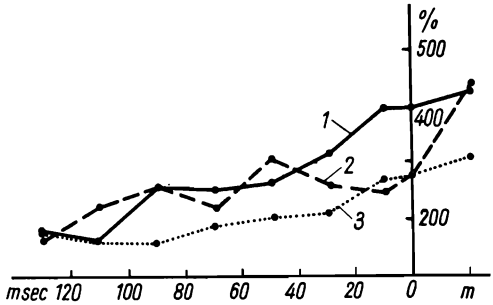

Fig. 7. Averaged curves of reflex response in normal dogs (*1*) and dogs pyramidotomized before elaboration of instrumental conditioned reaction (*2* and *3*). Designations as in Fig. 4.

of group 2 was in general similar to that of the intact dogs, i.e. excitability progressively rose as the time interval before onset of movement diminished, but this rise was significantly lower than in normal dogs (Fig. 7 *3*).

Pre-movement excitability of the motoneuronal pool following destruction of red nuclei in pyramidotomized dogs

In two dogs their pyramides sectioned after reflex elaboration, the nucleus ruber was destroyed on the left side, contralateral to the limb performing the instrumental response. After the rubral lesion these dogs showed a number of postural disturbances. When walking they leaned to one side. One dog rotated the head around the sagittal axis, the affected limbs turned under and dragged up. The conditioned response to presentations of the CS at first (for 1–1.5 months) was absent and appeared only when electrocutaneous stimulation was also applied. When the conditioned response was restored, its latency was considerably increased so that movements usually occurred only after painful stimulation (Fig. 8). Thus, conditioned movements could be observed only in special tests in which the CS was prolonged.

An early (apparently non-specific) increase of excitability of the motoneuronal pool in these animals was not altered. This increase was up to 200% of the amplitude in the control intervals, as in intact dogs. The same was true for the excitability increase occurring 80–120 msec before the onset of the EMG. At the same time the late, most prominent excitability changes during the last 60 msec before movement did not occur at all. The excitability rose slightly only during movement. The respective curves are given in Fig. 9.

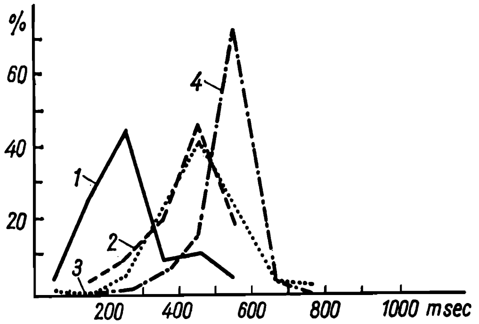

Fig. 8. Distribution of latencies of conditioned responses (according to EMG) of dog "Dick" before operation (*1*), a short time after pyramidotomy (*2*), a long period after pyramidotomy (*3*) and after added destruction of red nucleus (*4*). Designations as in Fig. 3*A*.

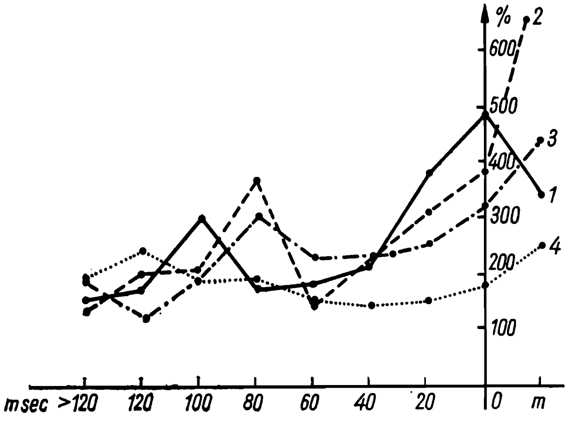

Fig. 9. Magnitude of response to nerve stimulation prior to onset of conditioned movement by dog "Seraya" before operation (*1*), a short time after pyramidotomy (*2*), a long period after pyramidotomy (*3*) and after added destruction of red nucleus (*4*). Designations as in Fig. 4.

DISCUSSION

The results show that prior to onset of an instrumental motor response in dogs the excitability of the motoneuronal pool involved in the execution of the conditioned movement increased. This phenomenon is similar to the motor setting prior to voluntary movement observed in man (6, 11). According to Kots (11) this increase of excitability of motoneurones is caused by several supraspinal mechanisms, different in origin and pathways, among which he distinguished:

1. A "baseline" nonspecific increase reflecting the general state of attention and readiness to perform a movement.
2. A "presetting" rise of excitability specific for the motoneuronal nuclei of the agonist muscles, occupying the greater part of the CS period except for the last 60 msec.
3. A "setting" increase of excitability of the same motoneurons starting 55–60 msec prior to movement.
4. A "triggering" rise starting 25–30 msec before the onset of the EMG, and associated with the excitability of the motoneurons only of fast, phasic muscles.

The present study revealed three phases of altered motoneuronal excitability preceding the onset of the instrumental motor response:

1. An early stage of increase in motoneuronal excitability, starting over 120 msec prior to the beginning of the conditioned EMG, which seemed to be associated with a non-specific facilitating effect of the acoustic stimulation used as a CS;
2. A pre-movement increase of excitability starting 100–80 msec before the onset of the EMG, often succeeded by a fall of excitability;
3. A period immediately preceding the onset of movement and starting 70–80 msec before the conditioned EMG. This period was characterized by a rise of excitability of the motoneuronal pools up to the start of the conditioned EMG.

Analysis of the role of the various supraspinal mechanisms in the setting of motoneurons prior to the onset of the conditioned activity suggests that the very last phase of altered excitability, occupying 60–80 msec before the appearance of the EMG, is associated with the pyramidal and rubro-spinal systems. Following pyramidal section, these changes seem to become wholly determined by rubro-spinal mechanisms. The unilateral destruction of the red nucleus in pyramidotomized dogs completely eliminated the setting on the contralateral side. These data are in full agreement with the evidences on the unidirectional influences of the pyramidal and rubro-spinal systems on flexor motoneurons (7, 10, 20) and interchangeable function of the pyramidal and rubro-spinal systems in the execution of general behavioral (2) and local instrumental (9, 13, 14) responses. Direct somatotopic projections of the motor cortex to the red nucleus are known (e.g., 18) corresponding to the somatotopic organization of the descending projections of the red nucleus itself (17). The collateral connections of axons of the motor cortical pyramidal cells with cortico-rubral neurons in the motor cortex and with neurons of the red nucleus have investigated (21). The cortico-rubro-spinal system appears to duplicate the pyramidal system in the transmission of cortical influences, ensuring adequate performance of a local instrumental response.

In man, the rubro-spinal tract is poorly developed, and this may account for the data on the absence of the segmental setting in patients with pyramidal paralysis. (11).

However, the pyramidal and cortico-rubro-spinal systems cannot also be considered as equivalent in animals. As reported elsewhere (9), following destruction of the red nuclei learned motor coordinations can still be performed, e.g., controlling the position of a feeding dish by maintaining a specific level of foreleg flexion while eating. On the other hand such coordinations are strongly altered by pyramidotomy even when the red nuclei are left intact (9). Pyramidotomy also influenced the latency of conditioned movements (5, 12). The present study showed, that in dogs, which were subjected to pyramidotomy before conditioning, the shape of the setting curve of the motoneuronal pools, apparently depending on cortico-rubro-spinal influences, was different from that of non-operated animals. This fact indicates that cortico-rubro-spinal influences are not equivalent to those carried out by the pyramidal tractes and cannot fully substitute for them in the process of compensation.

These results reveal certain differences in segmental setting when pyramidal sections are performed prior to or after conditioning. While a pyramidotomy performed after the reflex has been elaborated did not noticeably affect the time-course or the degree of pre-movement increase of motoneuronal excitability, the surgery performed before elaboration of the instrumental reflex, effected significant changes. This suggests that the characteristic pattern of segmental setting evolves during the formation and specialization of a movement and that the leading role in the development of this pattern belongs to the pyramidal system. The fact that pyramidal section after conditioned response elaboration does not affect the segmental setting indicates that after consolidation of the response, the meaning of pyramidal influences is diminished and can be entirely fulfilled by the cortico-rubro-spinal system. Thus, in this case, there seems to be the enlarger of a memory trace determining the pattern of excitability in the pre-movement motoneuronal pools. Such traces are apparently localized at the cortical level since loss of the second of the main descending cortical systems, i.e. of the cortico-rubro-spinal system, completely eliminates them, although the movements themselves could be and were performed.

A question arises as to the relationship between the changes in setting of motoneuronal excitability and the possibility of executing the motor response itself. It would be natural to suppose that an instrumental movement can be performed only, when a certain level of motoneuronal excitability is attained. Yet, in fact, in animals with loss of pyramidal tracts and red nuclei the occurrence of the conditioned EMG and movement are possible in the complete absence of specific setting of the motoneuronal pool (in the last 70–80 msec before movement). On the other hand, following pyramidal transection some parameters of the motor response: the latency, the speed of movement were altered but the setting curve of motoneuronal excitability did not change appreciably. This is possibly due to the heterogeneity of the motoneuronal pools of the agonist muscles. It is obvious that near-threshold testing stimulation does not activate all motoneurons but only those with the lower thresholds. Prior to movement, in an intact animal, the pool excitability and the number of motoneurons activated by the testing stimulation both increase, and this is reflected in the amplitude of reflex muscle response and, of course, in the segmental setting curve. It may be assumed that in the absence of pyramidal and cortico-rubro-spinal influences those motoneurons are involved in the execution of the instrumental response which are not activated by the testing stimulation and their changes in excitability are not reflected in the curve. On the other hand, pyramidotomy may affect motoneurons whose activity is not tested and thus produce changes in movement without altering the apparent setting of spinal excitation.

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