SYNAPTIC PROCESSES IN SPINAL NEURONS ACTIVATED MONOSYNAPTICALLY BY THE PYRAMIDAL TRACT

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In the study of the mechanism of cortical motor function and the mechanism of voluntary movements, one of the more important aspects is determination of the nature of interaction between the pyramidal system and the spinal neurons. Microelectrode investigations so far undertaken have shown that a special system of internuncial neurons is present in the spinal cord, connected specifically with the pyramidal fibers [2, 6]. These neurons possess special functional properties which differ from the properties of other spinal internuncial neurons, and their presence on the pathway of pyramidal impulses to the motoneurons is essential for interaction between descending pyramidal activity and the activity of segmental reflex arcs [4]. Obviously a detailed study of synaptic processes in such neurons, which can only be done by intracellular recording of their potentials, must provide particularly valuable information concerning the nature of corticospinal interaction. Data relating to synaptic processes in internuncial neurons are still very limited because of the difficulty of making successful intracellular recordings from them; no investigations whatever have been made of the neurons connected monosynaptically with the pyramidal tract by means of this method. The present investigation was therefore carried out with the special aim of studying this group of spinal internuncial neurons.

EXPERIMENTAL METHOD

Intracellular recordings of the activity of internuncial neurons in the lumbar division of the cat's spinal cord were made with glass microelectrodes filled with 2.5 M KCl solution by the usual method [5]. The animals were anesthetized with Nembutal. A descending wave was evoked in the pyramidal tract by means of a coaxial electrode [2]. The number of stimuli varied from 1 to 30-50, and their frequency in the series was up to 600 per second. The internuncial neurons activated monosynaptically by the pyramidal tract were identified from their localization in a transverse section of the spinal cord, the nature of their responses to stimulation of the cortex, as described below, and the absence of activation during stimulation of the peripheral nerves.

EXPERIMENTAL RESULTS

Focal recordings of the potentials arising in the spinal cord after stimulation of the motor cortex [2], together with morphological and electrophysiological data indicating the distribution of nerve endings of the pyramidal tract in the spinal cord [9, 14], show that the internuncial neurons activated monosynaptically by the rapidly conducting fibers of the pyramidal tract should be sought mainly in the most lateral part of Rexed's 5th layer—in the external basilar region of Cajal (EBR). The neurons of this region had no endings of peripheral afferent fibers [11, 12], but they have many racemose synaptic endings of pyramidal fibers [13].

The search for these neurons with the microelectrode was therefore carried out deliberately in the EBR. Both in the lower (L6-L7) and middle (L3-L4) lumbar segments in this region, activity of many neurons activated by the pyramidal wave only and not responding to afferent influences of any form from the peripheral nerves was recorded. In both these segments successful intracellular recordings were made of the potentials of these neurons, but a significant difference was that in the lower lumbar segments all the recordings were purely intraaxonal, whereas in the L3-L4 segments some recordings were intrasomatic (intraaxonal and intrasomatic recordings were differentiated by the absence or presence of postsynaptic potentials). Responses of the neurons in the EBR (which all the evidence showed to be intraaxonal) could also be recorded in the white matter of the lateral column throughout the extent of the lumbar segments.

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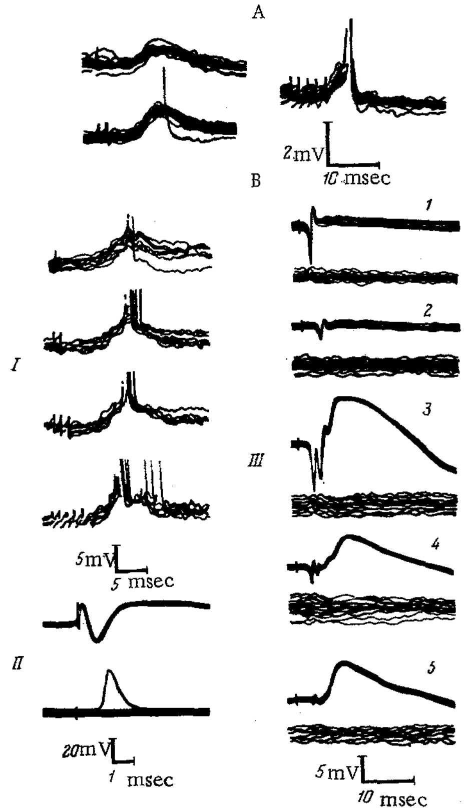

Fig. 1. Intrasomatic recording from neurons of the EBR. A—Neuron in EBR situated in segment L3, EPSP and generation of AP during stimulation of the cortex by an increasing number of stimuli; B—neuron in EBR situated in segment L4: I— EPSP and generation of AP during stimulation of cortex with an increasing number of stimuli; II—antidromic activation of neuron during stimulation of lateral column in segment L7 (distance between points of stimulation and recording 5.5 cm); III—absence of activation of neuron during stimulation of peripheral nerves: 1) posterior biceps—semitendinosus + anterior biceps—semimembranosus muscles; 2) gastrocnemious muscle; 3) tibial nerve; 4) common peroneal nerve; 5) sural nerve. Strength of stimulation 5 times above threshold level. All oscillograms obtained by superimposing several sweeps of the beam. In oscillograms II and III (1-5) the potential of the dorsal surface of the spinal cord recorded in segment L7 is shown by the top beam.

Altogether,11 neurons in the EBR were investigated by intrasomatic recording, and 35 neurons by intraaxonal recording. In addition, recordings were taken from 33 fibers of the pyramidal tract in the lateral column, identified from the generally accepted signs [2, 10].

In the case of both intraaxonal and intrasomatic recording from the neurons of the EBR at level L3-L4, these neurons could be excited by stimulation of the lateral column with needle electrodes in segment L7 (Fig. 1, B, II and Fig. 2, C). The latent period of this response varied for different cells from 0.7 to 1.2 msec, and was exceptionally stable for each neuron. The intrasomatic recordings showed that the action potential (AP) evoked by such stimulation was not preceded by slow waves of potential (Fig. 1, B, II). This suggests that these responses of the EBR neurons may be regarded as the result of antidromic activation. It follows, therefore, that the bodies of the EBR neurons are located mainly in the middle lumbar segments and their axons descend in the lateral columns, passing through several segments, to terminate on other spinal neurons. The conduction velocity in these axons, taking into consideration the distance covered (about 5-6 cm), is 50-70 m/sec.

Single stimulation of the cortex evoked an excitatory postsynaptic potential (EPSP) in the soma of the EBR neuron. The latent period of the EPSP in the group of neurons investigated ranged from 6-12 msec, with a mean value of 8 msec. For comparison, the conduction time of pyramidal impulses in segment L3 may be calculated on the basis of measurements of the conduction velocity in the pyramidal fibers of the investigated group (the investigated group consisted mainly of rapidly conducting pyramidal fibers with a

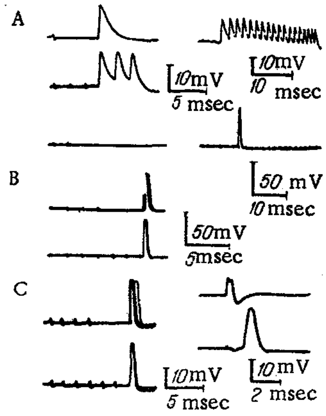

Fig. 2. Intraaxonal recording from fiber of the pyramidal tract and neurons of the EBR. A–pyramidal fiber. Lateral column of segment L1. Velocity of conduction about 65 m/sec. Apparent increase in frequency of responses due to the somewhat nonlinear sweep. B–neuron in the EBR. Lateral column of segment L2. Responses to stimulation of the cortex with different numbers of stimuli; C–neuron in the EBR. Lateral column of segment L4. Left oscillograms–responses to stimulation of cortex by different numbers of stimuli. Right oscillogram–response to stimulation of lateral column in segment L7. The top beam records the potential of the dorsal surface in segment L7. Distance between points of stimulation and recording 6 cm. All oscillograms obtained by superimposing several sweeps of the beam.

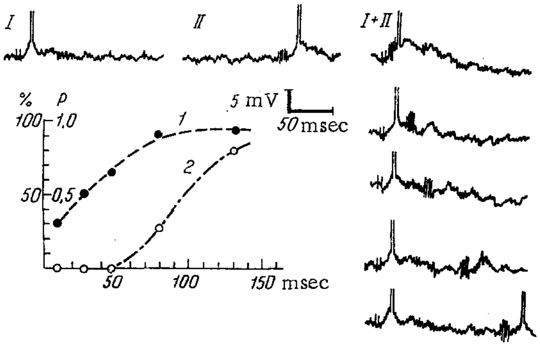

Fig. 3. Inhibition of response of EBR neuron evoked by the second of 2 successive stimuli applied to the cortex. Intrasomatic recording. I–response to preliminary stimulation (series of 2 stimuli): II–response to test stimulation (series of 5 stimuli); I + II–responses to combination of preliminary and test stimuli separated by different intervals; 1) change in amplitude of EPSP evoked by test stimulation, depending on interval between test and preliminary stimulation (amplitude of EPSP adequate to cause generation of AP taken as 100%); 2) change in probability of AP generation after test stimulation; probability (P) in the absence of preliminary stimulation taken as 1. All points on the graph are the mean values of results of 10 measurements.

conduction velocity of between 65 and 24 m/sec). The minimal time was 4.9 msec and the mean value 7.5 msec. These results thus agree fully with the hypothesis that activation of EBR neurons by fast-conducting fibers of the pyramidal tract is monosynaptic in character.

With relatively weak stimulation, when only a single descending impulse appeared in the pyramidal fibers, the EPSP had a shape similar to that of the monosynaptic EPSP evoked in segmental neurons by activity of the peripheral nerve fibers (Fig. 1, A), but its build-up time was longer (about 2.0–2.5 msec). The total duration of the EPSP was about 15 msec. With stronger single stimulation of the motor cortex, when several descending impulses appeared in some pyramidal fibers [4], the EPSP was more complex in character and consisted of several successive waves (Fig. 1, B, I), evidently reflecting the arrival of successive presynaptic impulses at the cell. The amplitude of the first 2 or 3 synaptic waves successively increased.

With an increase in the number of stimuli the amplitude of the EPSP rose by summation of the successive waves (Fig. 1, A and B, I). On reaching the critical level the EPSP were transformed into a spreading AP. The latter was accompanied by well marked after-hyperpolarization, although this did not completely abolish the synaptic depolarization, so that in some cases 2 successive AP were formed from the EPSP, separated by an interval of 5-10 msec.

The latent period of generation of the AP was noticeably dependent on the number of summated presynaptic waves. Since the build up time of the single EPSP, as shown above, was about 2 msec, if the frequency of descending waves was 600/sec the successive EPSP were superimposed on the ascending phase of their predecessors, thus greatly increasing the steepness of build-up of synaptic depolarization. The latent period of generation of the AP was correspondingly shortened (Fig. 1, A, I), as was clearly seen with intraaxonal recording (Fig. 2, B and C), amounting to 2.4 msec. The minimal latent period of generation of the AP measured during a series of stimuli of adequate duration, when recordings were made in the middle lumbar segments, varied from 6.3 to 20.4 msec (mean value 10.76 msec). On the other hand, the mean latency of the AP measured by the authors previously [4] in a lower lumbar segment (L6) had a mean value of 11.54 msec. This delay in appearance of the AP in the lower lumbar segments can be fully explained by the duration of intersegmental conduction of the impulse along the axons of the EBR neurons.

A characteristic feature of the activity of the EBR neurons, previously observed when intraaxonal recordings were made [4], was the long period of suppression of the discharge arising in response to a repeated descending wave in the pyramidal tract. This aspect of functioning of the EBR neurons led to sharp differences between their impulse activity and the activity of the pyramidal fibers. Fast-conducting pyramidal fibers reproduce a frequency of stimulation of up to 600 per second (Fig. 2, A). Meanwhile, the EBR neurons respond by only 1 or 2 AP even to a prolonged series of stimuli applied to the motor cortex (compare Fig. 1, B, I and Fig. 2, B and C).

Intrasomatic recordings from the EBR neurons showed that the EPSP developing in these neurons in response to the second of two successive descending waves is reduced in amplitude if the intervals between them are from 10 to 100-150 msec (Fig. 3, 1). This also leads to a decrease in probability of a discharge from the EBR neuron evoked by the second descending wave (Fig. 3, 2).

The results obtained clearly show that the pyramidal tract is connected monosynaptically in the spinal cord with a special system of propriospinal neurons covering several segments. The axons of these neurons, possessing relatively high conduction velocity, evidently run mainly in a descending direction and cover a distance equal to at least 2-3 segments before reentering the gray matter and terminating in synapses on other spinal neurons. This conclusion agrees fully with the morphological data of Szentagothai [13].

A particularly interesting question is whether these propriospinal neurons can transmit pyramidal influences directly to motoneurons. The minimal duration of the latent period of the EPSP evoked by cortical stimulation in the motoneurons of segment L7, according to the results of measurements made earlier in the authors' laboratory [1], was 8 msec while the mean latent period for a group of 72 motoneurons was 12.63 msec. If these results are compared with the minimal and mean latencies of discharge of the EBR neurons measured in the present investigation, with recordings taken in segments L3-L4 (6.3 and 10.76 msec) and the same values measured previously for segment L6 (6.7 and 11.54 msec respectively [4]), and if the conduction velocity along the axons of EBR neurons is taken into account, the monosynaptic character of activation of the motoneurons by the EBR neurons becomes very probable.

It thus follows that the pyramidal tract may evidently evoke excitation of motoneurons through a system of propriospinal neurons, with a very simple, disynaptic pathway. This pathway is adapted for transmission only of fast influences from the motor cortex, arriving as a short series of high-frequency impulses. These fast influences are transmitted by fast-conducting fibers of the pyramidal tract— the fibers which form synaptic connections with the EBR neurons. Transmission through the EBR neurons of tonic impulse activity is suppressed by the subsequent depression of their synaptic excitability. The reason for this depression is probably complex. On the one hand, the EPSP and the AP evoked in the EBR neuron by the pyramidal wave is followed by development of hyperpolarization of small amplitude but considerable duration (over 100 msec). This is particularly clearly seen if the cortex is stimulated by a relatively long series of stimuli. Its initial phase is evidently one of true after-hyperpolarization after the AP. Its subsequent development is possibly attributable to synaptic influences. This hyperpolarization may lead to a decrease in amplitude of the EPSP (on account of an increase in the ionic conductivity of the membrane) and an increase in the critical level for AP generation. On the other hand, the decrease in amplitude of the EPSP may be associated with presynaptic changes in the endings of the pyramidal fibers on the EBR neurons.

It must not, however, be supposed that the system described above is the only system connecting the pyramidal tract with the motoneurons. The possibility of activation of internuncial neurons of segmental arcs by the pyramidal fibers has been demonstrated by several authors [6-8]; however, the motor pyramidal effects evoked through these neurons must have longer latent periods than the effects arising through the system of propriospinal neurons. The connection between slow-conducting pyramidal fibers, excitation of which evokes prolonged synaptic changes of tonic character in motoneurons [3], and the motoneurons must be of a special character.

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