Facilitation of the dorsal root potentials by volleys spreading caudally and cranially in the spinal cord

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Summary. Conditioning volleys spreading along 3 segments of the cord produce larger facilitation of bilateral dorsal root potentials in sacral cord than volleys in neighbouring dorsal roots used to evoke the testing depolarization. Contralateral potentials are more facilitated by volleys descending from lumbar than ascending from caudal segments.

Dorsal root potentials (DRPs) indicate depolarization of primary afferent terminals, which is the postulated mechanism for presynaptic inhibition. Depolarization produced by single volleys in the lower lumbar cord is facilitated by preceding stimuli, and the time course of facilitation was found to be different in ipsilateral and contralateral dorsal roots[1,2]. Since transmission of depolarization to the other side of the cord occurs through synaptic relays, it is hypothetized that the pattern of facilitation is related to the organization of pathways mediating presynaptic inhibition. DRPs also spread along the cord and may be recorded several segments from the point of entry of the afferent volley[3,4]. Our recent experiments have shown that ipsilateral depolarization spreading in caudal direction has a shorter latency, decreases to a lesser extent in more distant segments and displays a smaller increase when produced by an increasing number of volleys than DRP spreading cranially[5]. These findings suggest various properties of pathways subserving the spread of DRPs in both directions. In the present communication, we describe facilitation of bilateral DRPs evoked in the sacral cord and conditioned by volleys spreading caudally and cranially along 3 segments of the spinal cord.

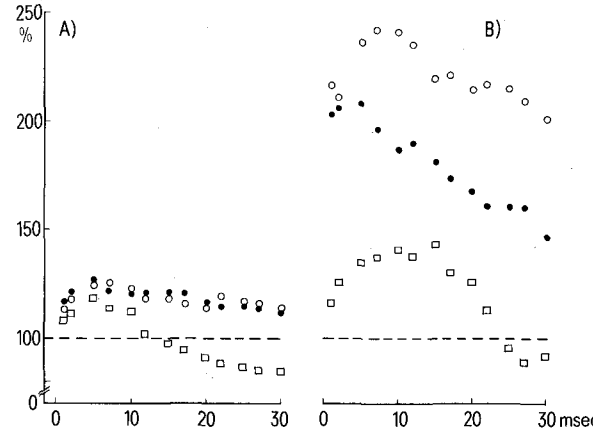
Material and methods. 14 cats were anaesthetized with thiamylal sodium and spinalized at the 1st lumbar segment (L1). After exposing the spinal cord, dorsal and ventral roots were cut bilaterally from L6 to Ca2. The most caudal rootlets of S2 dorsal root were separated on both sides up to their entries into the cord. Each rootlet was mounted on recording electrodes, one close to its entry into the cord and the other on the cut end. Dorsal roots L6, S1 and Ca2 on the right side were placed on bipolar stimulating electrodes. Single square wave pulses of 0.1 msec duration were used to produce conditioning and testing volleys. The pairs of pulses were repeated every 12-15 sec. The temperature of the oil covering the cord and of the animal was kept at 37±1 °C.

Results and discussion. Facilitation of the DRPs produced by conditioning volleys spreading from lumbar and caudal segments is shown in the figure. It may be seen that these changes are supplemented by and compared to facilitation occurring when both conditioning and testing volleys are applied to the same dorsal root. On the ipsilateral side of the cord, facilitation of the DRPs produced by 2 volleys in the S2 dorsal root rapidly increases and attains a maximum of 118% when the testing interval is 5 msec. Lengthening the interval between the volleys decreases the facilitation and at 12 msec the DRP returns to its control value. Then the potential undergoes depression, and when the testing interval is 30 msec it reaches 84% of the initial level. Following conditioning stimulation of L6 dorsal root facilitation of the DRP is larger, reaching 125% at the interval of 7 msec. The decrease of facilitation at longer intervals is slow and at the longest testing interval used depolarization is enhanced to 116% of control. The course of facilitation produced by conditioning volleys in Ca2 dorsal root very much resembles that seen during conditioning by volleys descending from L6 dorsal root. The largest increase of the DRP occurs at an interval of 5 msec, and it amounts to 125%. The size of depolarization slowly decreases at longer testing intervals and at 30 msec its amplitude is about 115%.

Facilitation of the contralateral DRP is much larger and depends to a greater extent on the place where the conditioning volley enters the spinal cord. With both stimuli applied to S1 dorsal root, a facilitation of 142% is attained at the testing interval of 10-15 msec. The decrease of the depolarization is slower than on the ipsilateral side and the DRP regains its control value at the interval of 22-25 msec. The rise of facilitation evoked by volleys in L6 dorsal root is very steep, and at the interval of 7-10 msec the DRP reaches 242% of the control. Then facilitation gradually decreases but at the interval of 30 msec it still amounts to 209%. Maximum facilitation of the DRP evoked by volleys ascending from Ca2 dorsal root is 208% and is attained at the interval of 5 msec. The decrease in the size of the DRP observed at longer testing intervals parallels changes in facilitation of presynaptic inhibition evoked by volleys descending from L6 dorsal root.

Our findings show that, in the 2nd sacral segment, facilitation of bilateral DRPs produced by 2 volleys in S1 dorsal root displays much the same maximum and decline as that in the lower lumbar cord[2]. These data indicate basically similar organization of pathways producing presynaptic inhibition close to the place where afferent volleys enter the spinal cord at both segmental levels.

Longer time course of facilitation in polysynaptic than in monosynaptic pathways has been ascribed to activation of additional interneurones[1,6-8]. Comparison of the DRPs on



Facilitation of the dorsal root potentials produced in S2 dorsal root by stimulation of S1 dorsal root and preceded by conditioning volleys in S1 dorsal root (□), in L6 dorsal root (○) and in Ca2 dorsal root (●). *A* shows ipsilateral and *B* contralateral depolarizations. Abscissa: intervals between conditioning and testing volleys in msec. Ordinate: amplitude of potentials in percentages in relation to the size of control DRPs not preceded by conditioning stimuli and taken as 100. Strength of stimulation was 50% of that producing maximum DRPs in the ipsilateral S2 dorsal rbot. DRPs were recorded simultaneously on both sides of the cord. In every preparation control depolarizations were evoked several times at the beginning and then throughout the experiment. The mean size of control DRPs was calculated from 20 records. At each testing interval the DRPs were elicited 4-6 times. Different testing intervals were studied in random sequence. Each point in the figure is the arithmetic mean of measurements obtained from 14 animals.

both sides of the lumbar cord suggests that the size of facilitation is related to the number of synapses depolarizing primary afferent terminals. Increased facilitation of the DRPs conditioned by volleys spreading along the cord corroborates this hypothesis. Larger maximum and slower decay of facilitation of contralateral DRPs evoked by volleys spreading caudally implies that pathways descending from lumbar cord activate more synapses than those ascending from caudal segments. This finding gives evidence for different organization of pathways transmitting depolarization in both directions. The spread of the DRPs along the cord most probably occurs via substantia gelatinosa and Lissauer tract. However, anatomical studies did not disclose different numbers of synapses involved in transmitting depolarization in both directions[9]. We have found that ipsilateral DRPs spreading caudally decrease in more distant segments while contralateral potentials are increased[5]. The enhancement may be produced by volleys which traverse the cord from the ipsilateral side a few segments below a stimulated dorsal root. These volleys would activate additional synapses and increase facilitation of contralateral DRPs despite the lack of known accessory synaptic relays in descending pathways of the substantia gelatinosa.

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Supernormal responses to premature stimulation in Ca-dependent action potentials

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Summary. The responses of Ca-dependent action potentials to premature stimulation were studied in the isolated canine ventricular muscle. When very premature stimuli were applied, supernormally augmented responses with propagation occurred, while responses with long preceding intervals were small and not conducted.

In general, recovery from inactivation of slow Ca current is known to be delayed[2-4] and refractoriness of slow responses[5], whose depolarization depends on slow inward current, is thought to be prolonged. Sano et al.[6] reported that the effective refractory period was increased to more than 1 sec in ventricular muscle preparation in which slow responses were activated by isoproterenol in high K solutions. Recently, however, we examined the responses of Ca-dependent action potentials[7] (AP) to premature stimulation and revealed that supernormal responses occurred when very premature stimuli were applied, and this augmented premature response showed supernormal conduction. This agreed with our previous report[8] that Ca-influx might be enhanced during premature depolarization.

Material and methods. A small portion of the trabecular muscle was dissected from the right ventricle of dogs and mounted in a tissue bath perfused with tris buffer physiological solution. Premature stimuli (S_2) were applied at variable intervals after every 10th driving stimulus (S_1) of 0.5 Hz. APs (R_1 and R_2) elicited by S_1 and S_2 of equal strength and duration were recorded with conventional microelectrode methods as described previously[9] at the proximal (P) and distal points (D) which were at intervals of a few mm. Measurements were made of the amplitude (Amp), duration (APD), the maximal rising velocity of APs recorded at P and conduction time for P and D. The ratio of these parameters of R_2 to those of R_1 were calculated.

Results and discussion. In control solutions (K 4.5 mM, Ca 1.6 mM), both Amp_2/Amp_1 and APD_2/APD_1 were near unity at any S_1S_2-interval (figure 1). In high K (21 mM)-high Ca (10 mM) solutions, the basic responses were small and had a low rising velocity (8 to 23 V/sec). Both their amplitude and duration were depressed by verapamil (10^{-5} M), and were augmented by isoproterenol (10^{-6} g/ml). Moreover, the resting membrane potential in this solution decreased to -49 ± 1 mV (mean + SE) (ranged from -45 to -54 mV), resulting in complete inactivation of fast inward Na current. Thus, they were thought to be Ca-dependent APs. When a very premature stimulus was applied, R_2 greater than R_1 occurred. The rising velocity of R_2 was also greater. Figure 1 shows the relation between the degree of the augmentation of R_2 and S_1S_2-intervals. Amp_2/Amp_1 and APD_2/APD_1 were 1.9 ± 0.3 and 2.8 ± 0.4, respectively at a S_1S_2-interval of 180 msec. They declined progressively to unity with increasing the coupling interval to 500 msec. Asterisks above the value show the significance of change. This supernormal premature response, R_2 elicited by S_2 applied 200 msec after S_1, was conducted from P to D, while a basic beat (R_1) was not (figure 2, A). When the

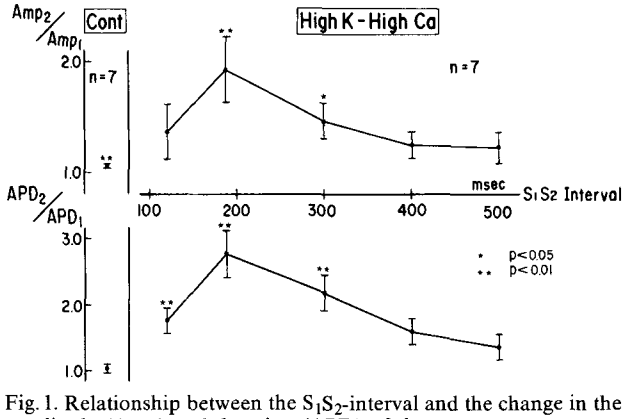

Fig. 1. Relationship between the S_1S_2-interval and the change in the amplitude (Amp) and duration (APD) of the premature response. On the left, the maximal ratio in the control solution is shown. Vertical bars indicate SEM. See text for additional explanation.