Activation of Central Neurons by Ventral Root Afferents

Abstract. *It is a fundamental principle of vertebrate neuronal organization that sensory fibers are restricted to dorsal roots and motor fibers to ventral roots. Recent evidence, however, indicates that there are many sensory fibers in ventral roots. The present report shows that stimulation of these fibers activates neurons in the dorsal horn. This provides evidence at the single-cell level for the importance of ventral root afferents and provides an explanation for the clinical phenomenon of recurrent sensibility.*

We report that stimulation of the distal stump of a cut ventral root activates neurons in the dorsal horn of the mammalian spinal cord. These findings bear on the function of ventral root afferents and thus on the law of separation of function of the spinal roots.

Five adult cats (2.5 to 3.5 kg) were anesthetized with α-chloralose (70 mg/kg, intravenously) and then paralyzed with gallamine triethiodide (Flaxedil). The lumbosacral spinal cord was exposed and ventral roots L7 and S1 were cut near the spinal cord. The peripheral ventral root stump was placed on stimulating electrodes. Unitary activity of cells in the dorsal horn was monitored with a carbon filament–filled microelectrode (*1*), while the ipsilateral sciatic nerve was stimulated at a frequency of 1 Hz. Once isolated single-cell activity was found, the distal stump of the cut ventral root of the same segment was stimulated. Recorded activity was led into a window discriminator whose output was led to a computer to compile peristimulus time histograms.

Fourteen cells from five cats were excited by ventral root stimulation. An example is illustrated in Figs. 1 and 2. This neuron responded to noxious stimuli (Fig. 1A) and had a small cutaneous receptive field on the foot (Fig. 1B). The location of the cell body is shown in Fig. 1C. All tested cells excited by ventral root stimulation had cutaneous receptive fields and some had an additional input from muscle. The excited cells responded strongly to noxious mechanical stimuli and some also responded weakly to innocuous stimuli such as light touch and pressure. The response of the cell illustrated in Fig. 1 to ventral root stimulation is shown in Fig. 2A. The stimulus intensity was high, and the excitation of the neuron occurred after a significant delay. The response was no longer observed when the ventral root was crushed just distal to the site of stimulation (Fig. 2B), and the response reappeared if the root was stimulated distal to the crushed area (Fig. 2C). This response could then be abolished by cutting the dorsal root of the same segment (Fig. 2D). Crushing and stimulation were done for four other neurons, and the results were the same.

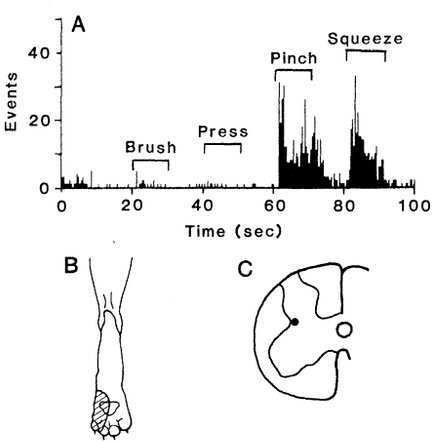

Fig. 1. Responses of a dorsal horn cell to mechanical stimuli. (A) Single-pass peristimulus histogram (bin width, 400 msec) made while the receptive field (B) was stimulated mechanically with four different intensities. The pinch and squeeze stimuli were painful to humans. Note that the cell responded only to noxious stimuli. (C) Location of the recorded cell.

These results strongly suggest that afferent fibers in the ventral root enter the spinal cord through the dorsal root and activate dorsal horn cells. From the crush experiments, it is apparent that the responses are not due to current spread. The activated afferents in the ventral root are unmyelinated, as evidenced by their conduction velocities (judged from the latencies of dorsal horn neuron excitation and the distance from stimulating electrode to dorsal root ganglion to recording site), which ranged from 0.41 to 1.73 m/sec (mean ± standard deviation, 0.75 ± 0.35 m/sec). In addition, high-intensity trains of stimuli were needed to elicit excitation, suggesting that temporal summation is necessary. This is characteristic of central neuron activation by unmyelinated fibers (*2*).

The classic view of mammalian spinal roots is that they have separate functions, the dorsal roots mediating sensory input and the ventral roots carrying motor commands (*3*). The explanation for this in anatomical terms is that only sensory axons are found in the dorsal root and only motor axons in the ventral root. This is an extremely important generalization. For example, acceptance of this view led to the use of dorsal rhizotomy as an operation to alleviate intractable segmental pain without affecting the motor fibers for that segment.

Ever since the functions of the roots were clearly delineated, however, there have been suggestions that some types of sensory activity are mediated by the ventral root. Magendie (*4*), for example, noted that stimulation of the ventral root gave rise to pain-like responses and that cutting the appropriate dorsal root abolished these responses. This was confirmed in humans by showing that ventral root stimulation led to a deep, diffuse, dull ache that was much more unpleasant than stimulation of the dorsal root of the same segment and that the

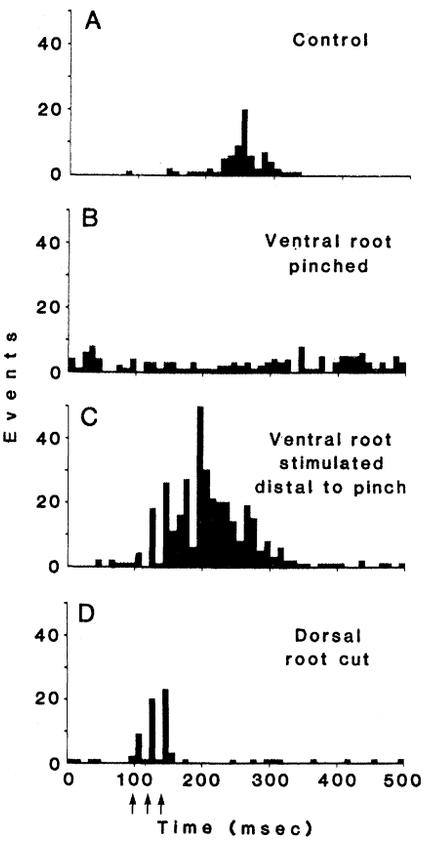

Fig. 2. Responses of the same cell to electrical stimulation of the distal stump of the ventral root. Peristimulus time histograms were made of responses to 20 consecutive stimuli (bin width, 10 msec) to the S1 ventral root (A). The stimuli consisted of a train of three pulses (each 15 V for 0.5 sec) separated by 20-msec intervals. The response was abolished after the ventral root was pinched just distal to the stimulation site (B). An even bigger response was then elicited by stimulating the ventral root distal to the pinch (C). This response was abolished when the S1 dorsal root was sectioned (D). Conduction distance (from stimulating electrode to dorsal root ganglion to recording site) was 74 mm. The arrows indicate the times at which electrical stimuli were applied. Note that in (C) and (D) some of the shock artifacts triggered the window discriminator.

pain was relieved when the dorsal root was inactivated by a local anesthetic (*5*). The phenomenon was termed recurrent sensibility. Thus it seems clear in both animals and humans that ventral root stimulation leads to pain and that interruption of dorsal root conduction abolishes that pain.

In 1894 Sherrington (*6*) noted a few sensory myelinated ventral root fibers and suggested that these were the morphological basis of recurrent sensibility. The suggestion was not accepted, however, because large fibers are thought not to carry nociceptive information. In recent years it has been shown that there are many unmyelinated ventral root afferents and that most of these carry nociceptive information (*7*). Evidence was obtained to indicate that some of the unmyelinated ventral root fibers enter the spinal cord directly through the ventral root (*8, 9*). These fibers may well be responsible for the failure of dorsal rhizotomy to relieve pain, and the observations imply that it is impossible to provide sensory denervation by dorsal rhizotomy alone (*10*).

However, fibers that enter the spinal cord directly through the ventral root would not explain the pain elicited by stimulating the distal stump of a cut ventral root and abolished by inactivating the fibers in the dorsal root. Thus there may be other types of afferent fibers in the ventral root. The present study provides, as far as we are aware, the first physiological data at the single-unit level showing that ventral root afferents can modify the activity of neurons in the dorsal horn. Furthermore, measurements of latency indicate that the afferent information entering the spinal cord is carried by unmyelinated fibers. Since we find, in confirmation of earlier work, that interruption of conduction in the dorsal root abolishes the phenomenon, it would seem that the information ultimately enters the spinal cord through the dorsal root. Thus some of the unmyelinated fibers that have recently been discovered in the ventral root are presumably the fibers carrying the noxious information to the spinal cord through the dorsal root and are thus the explanation of recurrent sensibility.

There seem to be two populations of ventral root afferents. First, there are those that enter the spinal cord directly through the ventral root. These probably explain the failure of dorsal rhizotomy to relieve pain, and their presence blurs the previously perceived clear separation of function of the spinal roots. Second, there are the ventral root afferents that enter the spinal cord through the dorsal root. These might be further subdivided into those with receptive fields in the meninges on the ventral side of the spinal cord (*11*) and those whose fibers loop into the ventral root and then enter the dorsal root (*12*). It remains an important task to determine the proportions of each and to see whether different functional modalities are transmitted by the different groups.

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Inhibition of Gastric Acid Secretion in Rats by Intracerebral Injection of Corticotropin-Releasing Factor

Abstract. *Intracisternal injection of ovine corticotropin-releasing factor (CRF) into the pylorus-ligated rat or the rat with gastric fistula resulted in a dose-dependent inhibition of gastric secretion stimulated with pentagastrin or thyrotropin-releasing hormone. When injected into the lateral hypothalamus—but not when injected into the cerebral cortex—CRF suppressed pentagastrin-stimulated acid secretion. The inhibitory effect of CRF was blocked by vagotomy and adrenalectomy but not by hypophysectomy or naloxone treatment. These results indicate that CRF acts within the brain to inhibit gastric acid secretion through vagal and adrenal mechanisms and not through hypophysiotropic effects.*

Neuropeptides originally characterized from the hypothalamus because of their ability to regulate pituitary hormone secretion—thyrotropin-releasing hormone (TRH), luteinizing hormone-releasing hormone (LHRH), and somatostatin—have been implicated in various regulatory processes in addition to their specific hypophysiotropic action (*1*). For example, TRH and somatostatin act both peripherally and within the brain to regulate gastrointestinal functions (*2*). Vale *et al.* used ovine hypothalami to characterize a 41–amino acid peptide called corticotropin-releasing factor (CRF), which stimulates the secretion of corticotropin and β-endorphin in vitro and in vivo (*3*). We showed that CRF injected into the cisterna magna or the lateral hypothalamus inhibits basal and pentagastrin-stimulated gastric acid secretion in rats through modulation of the autonomic nervous system.

Male Sprague-Dawley CD rats weighing 200 to 250 g were given free access to Purina Laboratory Chow and tap water and were housed under conditions of controlled temperature and lighting. All experiments were performed in rats deprived of food for 24 hours but with free access to water until the beginning of treatment. Ovine CRF was synthesized by the solid-phase method and purified by high-performance liquid chromatography (HPLC) after cleavage and deprotection by hydrofluoric acid (*4*). The peptide in lyophilized form was freshly dissolved in 0.9 percent saline just before each experiment. Animals were lightly anesthetized with ether for injection of CRF into the cisterna magna and were anesthetized with urethane (1.25 g/kg, intraperitoneally) for injection of CRF bilaterally into the lateral hypothalamic area or frontal cortex (*5*). Gastric secretions were collected by means of a 2-hour pylorus ligation or a short-term gastric fistula (*6*). Gastric secretions ob-