INTRODUCTION

The dorsal column (DC) plays an important role in relaying noxious visceral information to the ventral posterolateral (VPL) nucleus of the thalamus (Al-Chaer et al. 1996a,c, 1998b; Feng et al. 1998). A study done on primates has shown that a lesion of the DC blocks much of the colorectal input into the VPL nucleus (Al-Chaer et al. 1998b). Furthermore, recent results obtained using functional magnetic resonance imaging (fMRI) show that a DC lesion placed at the T10 spinal level dramatically reduces the increases in blood volume observed in thalamic and other brain areas in response to noxious colorectal distension (CRD) (Al-Chaer et al. 1998a,e). Earlier studies done on rats demonstrated that the DC-mediated colon input is largely conveyed in a newly identified component of the postsynaptic dorsal column (PSDC) system, whose cells of origin are in the vicinity of the central canal and whose axons project in the DC (Al-Chaer et al. 1996a,b, 1997b; Hirshberg et al. 1996; C. C. Wang, W. D. Willis, and K. N. Westlund, unpublished observations). Neurons of this newly identified component of the PSDC can be activated by CRD or colon inflammation (Al-Chaer et al. 1996b, 1997b).

Spinothalamic tract (STT) neurons are also known to be involved in visceral sensory processing. The spinothalamic tract arises largely from neurons in the dorsal horn of the spinal cord. The locations of these cells have been mapped in rats, cats, and monkeys using retrograde tracing and antidromic activation methods (see reviews by Hodge and Apkarian 1990; Willis and Coggeshall 1991). Most STT cells studied have cutaneous receptive fields and usually respond to noxious and often also to innocuous mechanical stimulation of the skin (Katter et al. 1996; Owens et al. 1992; Willis et al. 1974). Visceral afferent fibers can activate many STT cells (Ammons et al. 1985; Foreman et al. 1981; see Foreman 1989). STT neurons can be excited by distension of the gall bladder (Ammons et al. 1984), kidney (Ammons 1989a), ureter (Ammons 1989b), or urinary bladder (Milne et al. 1981). Milne et al. 1981) also recorded responses of STT cells in the upper lumbar and sacral segments of the monkey spinal cord to noxious testicular stimulation. These observations form the experimental basis for the assumption that the STT is the major ascending spinal cord tract that conveys information about noxious visceral events to lateral thalamic nuclei.

On the other hand, several research groups have argued for an important role of the DC in visceral input into the VPL nucleus (Apkarian et al. 1995; Berkley and Hubscher 1995; Chandler et al. 1998). In fact, Al-Chaer et al. (1996a, 1997a,c, 1998b) have argued that the DC plays a more important role than that of the STT in relaying information about noxious and innocuous visceral activity into the VPL nucleus in rats and monkeys, because a DC lesion largely blocks colonic input into the VPL nucleus of the thalamus, whereas a lesion that interrupts the STT has much less effect (Al-Chaer et al. 1996a, 1998b).

The purpose of this study was to compare the responses of PSDC and STT neurons in the monkey lumbar-sacral spinal cord to visceral and somatic inputs. To do this, recordings were made from single, randomly isolated neurons in the L6–S1 spinal segments. These neurons were examined for projections in the DC or the STT and were tested with graded CRD and colonic distension (CRD) (Al-Chaer et al. 1998d).

METHODS

Experiments were done on four adult male monkeys (Macaca fascicularis) weighing between 2 and 2.5 kg. The monkeys were...
The visceral stimulus used was CRD. It was adapted from the model used in rats (Gebhart and Sengupta 1996; see also Al-Chaer et al. 1996a,b). The stimulus was applied using an inflatable balloon inserted rectally. The balloon was constructed from a latex glove finger attached to a length of tygon tubing (10 cm). CRD consisted of consecutive inflations of the balloon to pressures ranging between 20 and 80 mmHg, applied in increments of 20 mm for 20 s every 4 min. CRD stimuli having an intensity above 40 mmHg are considered noxious in rats and painful in humans (Ness and Gebhart 1988; Ness et al. 1990).

The cutaneous stimuli employed were brushing (BR) of the receptive field using a camel hair brush, an innocuous stimulus; pressure (PR), using a large arterial clip applied to a fold of skin, a stimulus that causes a sense of near painful pressure if applied to human skin; and pinch (PI) using a small arterial clip that exerts a force of 550 gm/mm², a distinctly painful stimulus if applied to human skin.

Central stimulation consisted of electrical pulses (200 μs in duration and up to 500 μA in intensity) at a frequency of 2 Hz applied to the CNS to activate projection neurons antidromically. The VPL nucleus was stimulated to identify STT cells. The upper cervical DC or the nucleus gracilis (NG) was stimulated to identify PSDC cells. For VPL nucleus stimulation, a tungsten microelectrode was introduced stereotaxically into the VPL nucleus while recording from the DC or the VPL nucleus and also for peripheral somatic and visceral inputs. During the search process, the recording electrode was driven into the spinal cord while electrical stimuli were simultaneously applied in the VPL nucleus (to activate STT neurons antidromically) and to the DC (to activate PSDC neurons antidromically). Every neuron encountered was examined for possible antidromic activation from either the VPL nucleus or the DC separately. All neurons recorded in this sample were checked for input from the colon using CRD as the stimulus. Then all neurons were examined for cutaneous input using three different modalities (Brush, Press, and Pinch). Units that responded to CRD were classified as viscerosensitive, units that were antidromically activated from the DC or the DCN were considered to be PSDC neurons, and units antidromically activated from the VPL nucleus were regarded as STT neurons. Units that were not antidromically activated from either the DC or the VPL nucleus were considered to be unidentified neurons.

The extracellular action potentials recorded were fed into a window discriminator and displayed on an oscilloscope screen. The output of the window discriminator was fed to a data collection system (CED 1401+) and a personal computer to compile rate histograms or wavernark files using the Spike 2 software program. The response to each intensity of CRD was stored separately. Twenty seconds of baseline activity preceded the application of a distention stimulus. Each stimulus lasted 20 s. Four minutes were allowed to elapse between two consecutive CRDs. The responses are expressed as the average rate of firing of the cell during a particular stimulus minus the average baseline rate.

Histology

At the end of each experiment, a continuous current (1 mA for 20 s) was passed to mark the stimulation sites in the VPL nucleus and in the NG. The tip of the carbon fiber glass microelectrode was cut off and left at the recording site. The spinal cord at the level of the recordings and the brain were removed and put into a neutral Formalin solution (4%). The tissues were stored in 20% sucrose before frozen sectioning at 50 μm. The recording and stimulation sites were later identified histologically.

Statistics

The CRD data were analyzed using a repeated measures ANOVA. A model with the repeated factor of intensity, group, and the (intensity × group) interaction showed that there was a significant intensity effect (P < 0.001), no group effect (P = 0.67) and no (intensity × group) interaction (P = 0.12). The interpretation of these results is that there was a significant increase in the magnitude of neuronal responses across levels of CRD for all three groups. The nonsignifi-
cant group and interaction effects mean that responses in all three
groups increased in a similar fashion.

The responses to cutaneous stimuli (Brush, Press, and Pinch) were
first analyzed using an overall test (Hotelling’s $T^2$ test) to assess the
extent to which the distributions of these three variables differed
across all three groups (PSDC, STT, and Unidentified), when consid-
ered simultaneously. This test showed a significant difference across
groups ($P = 0.038$), and subsequent one-way ANOVAs showed that
only the variable Press showed an overall difference among the three
groups ($P = 0.002$). Tukey’s Studentized range test showed that
groups STT and PSDC differed significantly, with a difference of
means of 33.6 spikes/s (95% CI: 12.9, 54.2).

RESULTS

A total of 100 neurons were isolated in the L6–S1 segments
of the spinal cord. The neurons were distributed between 400
$\mu$m and 2 mm in depth measured from the surface of the spinal
cord. The mediolateral distribution extended from near the
dorsal median septum to the edge of the dorsolateral funiculus.
The recording sites were reconstructed by extrapolating their
locations from distance parameters recorded in relation to
marked sites as shown in Fig. 1.

Neuronal characteristics

All isolated neurons were tested for projections into the DC
and into the VPL nucleus. They were also tested for responses
to cutaneous stimulation and to CRD. The neurons were then
grouped into three categories based on whether they could be
antidromically activated by DC stimulation, by stimulation of
the VPL nucleus or by neither. The responses of neurons
within each category were analyzed separately. Table 1 illus-
trates the number of neurons in each category according to
their responses to CRD.

Neurons activated by CRD exhibited an increase in their re-
sponses that correlated with the increase in stimulus intensity. The

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<th>TABLE 1. Types and responses of neurons recorded in this study</th>
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The number of neurons isolated in each monkey is shown in the respective
columns. The number of neurons identified as belonging to a specific category
is shown in the respective rows. The total number of neurons within each
category is shown in the right column. PSDC, postsynaptic dorsal column; STT, spinothalamic tract; E, excited by colorectal distension (CRD); I, inhib-
ited by CRD; NR, no response to CRD.

FIG. 1. Drawing of a cross-section through S1 illustrating recovered re-
cording sites. , postsynaptic dorsal column (PSDC) neurons that responded to
colorectal distension (CRD); , PSDC neurons that did not respond to CRD; ,
spinothalamic tract (STT) neurons that responded to CRD; , STT neurons that
did not respond to CRD.

FIG. 2. Approximation of the population stimulus response by a linear
regression for all the excited PSDC neurons (A; $n = 21$; $r = 0.3$), STT neurons
(B; $n = 7$; $r = 0.5$), and unidentified neurons (C; $n = 9$; $r = 0.6$). The linear
regressions indicate a correlation between the response and the stimulus
intensity; $r$ is the correlation coefficient. $r^2$ is sometimes called the coefficient
of determination and is a measure of the closeness of fit of a scatter graph to
its regression line. Confidence intervals, also called the confidence interval for
regression, describe the range where the regression line values will fall 95% of
the time for repeated measurements. Prediction intervals, also called the
confidence interval for the population, describe the range where the data values
will fall 99% of the time for repeated measurements.
population stimulus response curve was approximated by a linear regression for all the neurons in each category. These regression lines show that the rate of change (slope) was similar across neuronal types (Fig. 2: PSDC, STT, and Unidentified). This graded response was observed regardless of the threshold of activation of the VPL unit by CRD. The threshold of activation was defined as the lowest intensity of CRD that evoked a neuronal response and was determined with increasing steps of 20 mmHg of intracolonic pressure for the cells tested. The accuracy is estimated to be 10 mmHg across the pressure spectrum (20–80 mmHg). The average thresholds of activation for neurons of each category are illustrated in Fig. 3.

PSDC NEURONS. Of 100 neurons isolated, 48 were antidromically activated by stimulation of the DC. In one preparation, nine PSDC neurons were antidromically activated by low-intensity stimulation in the NG (as low as 10 μA). The PSDC neurons could be classified into two clearly separable populations. One group of relatively shallow neurons ($n = 24$; $<1,400 \, \mu m$ in depth) located mostly near the lateral edge of the pia hole was unresponsive to CRD. A second group of relatively deep neurons ($n = 24$; $>2 \, \text{mm}$) located mostly near the midline responded to CRD. Twenty-one neurons were excited and three neurons were inhibited (Table 1). Of the 24 viscerosensitive PSDC neurons, 22 had cutaneous receptive fields located on the inner and posterior aspects of the thigh, on the rump, or on the scrotum. Two PSDC neurons excited by CRD could not be activated by skin stimulation. Of the 24 PSDC neurons that did not respond to CRD, 12 neurons had cutaneous receptive fields located over the outer aspect of the leg, the upper thigh, and the rump, and 12 could not be activated by somatic stimulation. The responses of viscerosensitive PSDC neurons to CRD were graded with stimulus intensity (Fig. 2). The mean threshold for activation of PSDC neurons was 34 ± 3.9 mmHg for PSDC neurons ($n = 24$), 31 ± 5.9 mmHg for STT neurons, and 43.4 ± 3.4 mmHg for unidentified neurons ($n = 30$); (mean ± SE: range 20–80 mmHg). The vertical box plot displays the median, 10th, 25th, 75th, and 90th percentiles as the boxes with error bars and the 5th and 95th percentiles as small circles (the percentiles increase downward).

STT NEURONS. Seventeen isolated neurons were antidromically activated by stimulation in the VPL nucleus. The depth of these neurons ranged between 1 and 2 mm from the surface of the pia hole, and most of them were best isolated at the lateral edge of the pia hole near the dorsolateral funiculus. Of the 17 STT neurons, 7% were excited by CRD, 4% were inhibited, and 6% did not respond to CRD. Fifteen STT neurons could be activated by cutaneous stimulation applied to the back and inner aspect of the thigh, to the scrotum, and to the perineal area. The responses of viscerosensitive STT neurons to CRD were graded with stimulus intensity (Fig. 2). The mean threshold for activation of STT neurons was 31.4 ± 5.9 mmHg (Fig. 3). The average response to 20 mmHg was 6.8 ± 4 spikes/s, and the average response to 80 mmHg was 20.6 ± 7.2 spikes/s (Fig. 4). The STT cells responded better to Press or Pinch than to Brush stimuli applied to the cutaneous receptive field (Fig. 5). Figure 7 illustrates the site of recording of an STT neuron, the site of stimulation for antidromic activation in the NG, the antidromic spikes, and the responses of the PSDC neuron to CRD and cutaneous stimulation.

UNIDENTIFIED NEURONS. Thirty-five neurons encountered could not be activated antidromically from either the VPL nucleus or the DC and were classified as unidentified neurons. Twenty-three of

FIG. 3. Graph illustrates the distribution of threshold intensities of CRD for excited PSDC, STT, and unidentified neurons. The mean thresholds were 34 ± 3.9 mmHg for PSDC neurons ($n = 24$), 31 ± 5.9 mmHg for STT neurons, and 43.4 ± 3.4 mmHg for unidentified neurons ($n = 30$); (mean ± SE: range 20–80 mmHg). The vertical box plot displays the median, 10th, 25th, 75th, and 90th percentiles as the boxes with error bars and the 5th and 95th percentiles as small circles (the percentiles increase downward).

FIG. 4. Line graphs illustrating the average responses, in spikes/s, of 13 PSDC neurons, 8 STT neurons, and 9 unidentified neurons excited by CRD. No significant differences were seen between the 3 groups.

FIG. 5. Bar graphs illustrating the average responses, in spikes/s, of 13 PSDC neurons, 12 STT neurons, and 4 unidentified neurons to cutaneous stimulation of their respective receptive fields. * Significant difference between the responses of PSDC and STT neurons to pressure.
PSDC neurons that respond to CRD may underlie the greater role of the DC in transmitting colonic input into the VPL nucleus than the STT as was shown in a recent study by Al-Chaer et al. (1998b). A lesion of the DC at the T10 segmental level dramatically reduced the responses of VPL neurons to CRD, whereas the effects of anterolateral or dorsolateral spinal lesions were not as consistent and dramatic as those of a DC lesion. Earlier observations in the rat (Al-Chaer et al. 1996a) showed that the visceral input in the DC is conveyed largely by the axons of PSDC neurons (Al-Chaer et al. 1996b), whose cell bodies are located mainly around the central canal (Hirshberg et al. 1996). These axons converge onto gracilothalamic neurons (Al-Chaer et al. 1996b), which presumably relay the majority of pelvic visceral input from the colon to the VPL nucleus (Al-Chaer et al. 1997a).

In most mammalian species, the VPL nucleus receives sensory information arising from stimulation of the skin, deep tissues, and viscera and conveyed over two major ascending tracts: the dorsal column pathway and the spinothalamic tract. Some of this information can also be conveyed through the spinocervical tract and also other indirect pathways relaying through neurons of the brain stem reticular formation that receive input from fibers traveling in the ventrolateral quadrant (Willis and Coggeshall 1991). Although spinoreticular neurons may respond to visceral stimuli (e.g., Blair et al. 1984), it is not known whether visceral input can be transmitted indirectly by this pathway to the VPL nucleus.

A considerable amount of research has been done on the response characteristics of STT neurons and of PSDC neurons and neurons of the DC nuclei (Willis and Coggeshall 1991). However, very little has been done to characterize potential differences in the response properties of these different popu...
lations of projection neurons (Brown et al. 1986; Chandler et al. 1998). In a recent study in monkeys, Chandler et al. (1998) described differences in the evoked discharge rates, latencies to activation and duration of peristimulus histogram peaks between cuneothalamic and STT neuronal responses to cardio-pulmonary sympathetic input. They suggested that dorsal and ventrolateral pathways to the VPL nucleus of the thalamus play different roles in the transmission and integration of nociceptive cardiac information. Cuneothalamic neurons are not the anatomic equivalent of STT neurons. Therefore the differences seen by Chandler et al. may be due to different organizations of a spinal pool of neurons (STT) and a supraspinal pool (DCN).

In this study, we compare two different populations of projection neurons, both of which are located in the spinal canal, at the level of convergence of colon afferents. In addition, there may be differences in the nature of the input received by each pathway. These may arise out of differences between the stimuli used (natural colon stimulation vs. electrical stimulation of cardiothoracic afferents) or differences in the type of receptors innervating the colon and the heart.

In the present study, the location of viscerosensitive PSDC neurons corresponded well with the location of viscerosensitive PSDC neurons observed previously in the rat (Honda 1985; Ness and Gebhart 1987). The vast majority of these neurons were in and around the central canal. PSDC neurons isolated elsewhere in the spinal cord, mainly in more superficial layers of the dorsal horn, were not responsive to CRD. The distributions of visceroceptive PSDC neurons correlate with the site of convergence of pelvic nerve terminals (Nadelhaft et al. 1983). The STT neurons encountered were not as abundant as PSDC neurons. The distribution of STT neurons corresponded with findings of previous studies from this laboratory (Willis et al. 1979). Unidentified viscerosensitive neurons were also distributed throughout the area of gray matter explored.

Careful analysis of the response characteristics of each group of neurons to CRD did not reveal any differences in the attributes of these responses. The responses were stimulus bound and graded with the stimulus intensity. No significant differences in the amplitude of the responses to each individual stimulus were seen; in addition, the slopes of the stimulus-response curve for each type of neuron were not significantly different. These observations reduce the likelihood that the predominant role of the DC in visceral input into the thalamus can be attributed to stronger responses of individual viscerosensitive PSDC neurons and argues for a stronger collective input of the PSDC system compared with other ascending spinal pathways. Even though the characteristics of the neuronal responses could differ were we to use a different stimulus (Chandler et al. 1998), the effect of a DC lesion remains consistent with a stronger DC input.

Putative roles for each system

These various pathways may be anatomically separate, but they are by no means independent (Zhang et al. 1996). Disruption of the information flow in one of them may alter the function of the other (Saadé and Jabbur 1984). So how could one extrapolate a limited physiological function into a global sensory role? In a situation where the perceived stimulus is localized in time and brief in duration, a linear flow of information is a plausible hypothesis even though it may be incomplete. However, when dealing with situations of chronic pain or sensory loss, many variables come into play, among them the dynamic interplay between the various neuronal components (Berkley et al. 1993; Chandler et al. 1996; Le Bars et al. 1979; see also Bouhassira et al. 1998), the wind-up of activity within each component (Al-Chaer et al. 1996c, 1997a,b; Roza et al. 1998); and the rewiring of the nervous hardware when necessary. The abundance of PSDC neurons in lumbosacral segments of the cord affords the PSDC system the ability to manage the peripheral input at that level and to relay it to other neurons within those segments and to higher neuronal structures.

In summary, the results of this study show that the PSDC system plays an important role in the processing of innocuous and noxious colorectal information in the monkey. This role may be based largely on the number of viscerosensitive PSDC neurons encountered at L₆–S₅ segments. This is not to discount the role of other ascending pathways such as the STT, but the data obtained suggest that STT cells are less abundant in those segments than PSDC cells. We can only speculate about the role of unidentified neurons. Therefore similar to what we have seen in rats (Al-Chaer et al. 1996b), we conclude that the neuronal basis for the role of the DC in colon pain (Al-Chaer et al. 1998b) rests largely on the PSDC system.

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