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Differential presynaptic control of the synaptic effectiveness of cutaneous afferents evidenced by effects produced by acute nerve section

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Key points

- We investigated, in the anaesthetized cat, the effects of acute section of the saphenous and superficial peroneal nerves on the synaptic effectiveness of the sural nerve afferents.
- We found that acute section of these nerves produced a long-lasting increase of the sural-evoked field potentials. At the same time, sural afferents ending within Rexed's laminae III–IV showed a reduced tonic primary afferent depolarization, while sural afferents projecting deeper into the dorsal horn (Rexed's laminae V–VI) instead showed increased tonic primary afferent depolarization.
- It is suggested that a differential control of the synaptic effectiveness of the low-threshold cutaneous afferents according to their sites of termination within the dorsal horn provides means for a selective processing of sensory information in response to tactile and nociceptive stimulation or during the execution of different motor tasks.

Abstract In the anaesthetized cat, the acute section of the saphenous (Saph) and/or the superficial peroneal (SP) nerves was found to produce a long-lasting increase of the field potentials generated in the dorsal horn by stimulation of the medial branch of the sural (mSU) nerve. This facilitation was associated with changes in the level of the tonic primary afferent depolarization (PAD) of the mSU intraspinal terminals. The mSU afferent fibres projecting into Rexed's laminae III–IV were subjected to a tonic PAD that was reduced by the acute section of the SP and/or the Saph nerves. The mSU afferents projecting deeper into the dorsal horn (Rexed's laminae V–VI) were instead subjected to a tonic PAD that was increased after Saph and SP acute nerve section. A differential control of the synaptic effectiveness of the low-threshold cutaneous afferents according to their sites of termination within the dorsal horn is envisaged as a mechanism that allows selective processing of sensory information in response to tactile and nociceptive stimulation or during the execution of different motor tasks.

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Abbreviations CDP, cord dorsum potential; DRP, dorsal root potential; GABA, gamma aminobutyric acid; IFP, intraspinal field potential; mSU nerve, medial branch of the sural nerve; n, negative; np, negative–positive; PAD, primary afferent depolarization; PAH, primary afferent hyperpolarization; Saph, saphenous; SP, superficial peroneal; SU, sural.

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Introduction

It has been known for a long time that peripheral nerve injury expands the sensory fields of dorsal horn neurones in response to mechanical stimulation of the skin (Devor & Wall, 1978, 1981*b*). In addition, other studies have indicated that inactive synapses of the saphenous nerve terminals on dorsal horn neurones in the sciatic territory are also activated after chronic transection or acute anaesthesia of the sciatic nerve (Markus & Pomeranz, 1987; Biella & Sotgiu, 1995). These findings led to the proposal that peripheral nerve injury or anaesthesia triggers a complex series of events. Among these events is a reduction of tonic primary afferent depolarization (PAD) and of the ensuing presynaptic inhibition exerted on the spinal projections of other cutaneous (intact) afferents on dorsal horn neurones. A reduction in the tonic presynaptic inhibition has been assumed to contribute to the potentiation of synaptic actions of low-threshold afferent fibres following peripheral nerve injury or anaesthesia (Biella & Sotgiu, 1995; see also Devor & Wall, 1981*a*; Wall & Devor, 1981, 1984; Devor, 1983).

Recent studies in the spinal cord of the anaesthetized cat have disclosed the generation of synchronized spontaneous negative (n) and negative–positive (np) cord dorsum potentials (CDPs) in the lumbosacral spinal cord. The spontaneous npCDPs, unlike the nCDPs, appeared to be preferentially associated with negative dorsal root potentials (DRPs) generated by PAD. Acute section of the sural and of the superficial peroneal nerves was found to increase the correlation between paired sets of npCDPs, leading to further activation of the pathways mediating PAD and presynaptic inhibition of cutaneous afferents (Chávez *et al.* 2012).

At first glance, these findings appear not to support the proposal of Biella & Sotgiu (1995) that the unmasking of the cutaneous nerve-evoked spinal responses is partly due to a reduced presynaptic inhibition. Nevertheless, it is possible that the changes in presynaptic effectiveness of the cutaneous afferents induced by acute nerve injury depend, to some extent, on the segmental projections and functional relations between the tested and damaged cutaneous afferents, in a similar way as for group II muscle afferents (Riddell *et al.* 1995; Jankowska *et al.* 2002). Differences in the degree of presynaptic inhibition within the regions of projection of group II muscle afferents in the dorsal horn and in the intermediate zone of the same segments suggest that a stronger presynaptic inhibition at one of these sites might result in a preferential recruitment of interneurones and in changes in the pattern of motor reactions evoked by group II muscle afferents.

We now report observations aimed to explore more directly the contribution of presynaptic mechanisms to the long-lasting increase of the synaptic actions of cutaneous afferents triggered by the acute nerve sections. We were particularly interested in the analysis of the effects of the acute section of cutaneous nerves because, besides being a common accident in humans, there is evidence that acute skin incisions affect the subsequent responses to nociceptive stimulation in a variety of spinal pathways (Lam *et al.* 2008; Vernon *et al.* 2009) and may have some bearing in the pre-emptive analgesia (produced by local anaesthesia of skin) that reduces postoperative pain (Dahl & Møiniche, 2004).

To this end, we examined, in the anaesthetized cat, the effects of the acute section of the saphenous and superficial peroneal nerves on the PAD of the intraspinal terminals of the medial sural afferents ending within the dorsal horn. Primary afferent depolarization was inferred from the changes in the antidromic responses of afferent fibres produced by constant-current intraspinal stimulation (Wall, 1958; Eccles *et al.* 1962). The results obtained suggest a differential control of the synaptic effectiveness of low-threshold cutaneous afferents according to their sites of termination within the dorsal horn that is envisaged as a mechanism that allows selective processing of cutaneous sensory information during nociceptive stimulation or during motor performance.

Some preliminary results have been reported in abstract form (García & Rudomin, 2006) and as part of a review on presynaptic inhibition (Rudomin, 2009).

Methods

Ethical approval

All experiments were approved by the Institutional Ethics Committee for Animal Research (protocol number 126-03) and comply with the ethical policies and regulations of *The Journal of Physiology* (Drummond, 2009) and of the National Institutes of Health, Bethesda, MD, USA (Animal Welfare Assurance #A5036-01). Protocols contained in the *Guide for the Care and Use of Laboratory Animals* (National Research Council, 2010) were followed in all cases.

General procedures

The experiments were carried out in 13 young adult cats of either sex. The animals were initially anaesthetized with pentobarbitone sodium (40 mg kg^{-1} I.P.). The carotid artery, radial vein, trachea and urinary bladder were cannulated. Additional doses of pentobarbitone sodium (5 mg kg^{-1} h^{-1}) were given intravenously to maintain an adequate level of anaesthesia, tested by assessing that withdrawal reflexes were absent, that the pupils were constricted and that arterial blood pressure was

between 100 and 120 mmHg. A solution of 100 mM of sodium bicarbonate with glucose 5% was given I.V. (0.03 ml min^{-1}) to prevent acidosis (Rudomin *et al.* 2004a). When necessary, dextran 10% or ethylephrine (Effortil; Boehringer Ingelheim, International GmbH Ingelheim am Rhein, Germany) was administered to keep blood pressure above 100 mmHg.

The lumbosacral and low-thoracic spinal segments were exposed by laminectomy and opening of the dura mater. The medial sural (mSU), superficial peroneal (SP) and saphenous (Saph) nerves in the left leg were dissected free and kept in continuity (see Fig. 1). After the surgical procedures, the animals were transferred to a stereotaxic metal frame, allowing immobilization of the head and spinal cord. Pancuronium bromide (0.1 mg kg^{-1}) was administered for muscle relaxation. The animals were artificially ventilated, and tidal volume was adjusted to maintain a concentration of 4% CO_2 in the expired air. Adequacy of anaesthesia was subsequently tested by assessing that the pupils remained constricted and that heart rate and blood pressure were not changed following a noxious stimulus (paw pinch). To prevent desiccation of the exposed tissues, pools were made with the skin flaps, filled with paraffin oil and maintained between 36 and 37°C by means of radiant heat.

Recordings and stimulation

The intact mSU nerve was mounted on two pairs of bipolar hook electrodes, one for stimulation and the other to record the antidromic responses produced by intraspinal stimulation. The intact SP nerve was mounted on a bipolar hook electrode pair for stimulation only (see Fig. 1).

Cutaneous nerve-evoked CDPs were recorded with a series of ball electrodes placed on the dorsum of the L4–L7 spinal segments against an indifferent electrode placed on nearby paravertebral muscles, using separate preamplifiers with bandpass filters 0.3 Hz to 10 kHz. Simultaneous recordings of CDPs were used to select the segments with the strongest mSU projections. Glass micropipettes (1–2 MΩ, 2–3 μm tip diameter) filled with a 2 M solution of NaCl were positioned in these segments (usually middle L6 segments) to record the mSU-evoked intraspinal field potentials (IFPs).

Experimental procedures

Once the recording micropipette was properly placed over the spinal surface, the mSU nerve was stimulated once per second with single pulses of fixed strength (5–20 microamps, 0.1 ms) while recording the evoked IFPs at different depths. Several tracks were made to locate the sites where stimulation of the mSU nerve produced the largest IFPs. Once in position, the mSU nerve was stimulated with different strengths (usually 1.1–2.0 times the threshold of the most excitable fibres in the nerve, T), and the averages of the evoked CDPs and IFPs ($n = 32$) were displayed on the oscilloscope and stored in the computer memory.

After recording the mSU IFPs evoked at different depths, the glass micropipette was connected to a constant-current pulse generator for intraspinal stimulation and displaced upwards or downwards along the same track to find the site that produced the largest antidromic population responses in the mSU nerve. Once found, the micropipette was left in place throughout the experiment. The strength of intraspinal stimulation was also expressed as multiples of the intensity required to produce detectable antidromic responses. This allowed comparison of data from different experiments in terms of the stimulated fibres.

For each stimulus strength (2–30 μA), recordings were made of the unconditioned antidromic responses, as well of the antidromic responses preceded by conditioning stimulation of the SP nerve (usually one pulse of 2T strength applied 35 ms before the test stimulus). These procedures were repeated before and after the acute section of the Saph or SP nerves.

The CDPs and IFPs evoked by mSU nerve stimulation, as well as the mSU antidromic responses produced by intraspinal stimulation, were displayed on the oscilloscope, digitized with a sampling rate of 10 kHz and stored in the computer disk memory for subsequent processing. Changes in the mSU antidromic responses produced by the different experimental procedures were estimated from the area of the evoked potentials using a specially designed program that allowed integration between two preset time intervals (Wall, 1958; Eccles *et al.* 1962; Rudomin & Dutton, 1969).

Histology

At the end of the experiment, the animal was killed with an overdose of I.V. pentobarbitone, perfused with 10% formalin, and the spinal cord removed while leaving the shank of the glass micropipette in place. After complete fixation and dehydration, the lumbosacral segments were placed in a solution of methyl salicylate for clearing and subsequently cut transversely to obtain a section containing the electrode, and photographed (Wall & Werman, 1976).

Once the histological sections were available, the microelectrode track within the spinal cord was reconstructed, taking into account the marks left in the tissue by the micropipette, combined with depth measurements from the cord surface made during the experiment. In addition, the position of the stimulated sites was referred relative to the amplitude of the mSU IFPs recorded at different depths in that particular track (see Figs 8 and 9 and Jankowska *et al.* 1981).

Results

Changes produced by acute section of the saphenous nerve

As stated in the Introduction, Biella & Sotgiu (1995) proposed that the unmasking of the saphenous nerve-evoked responses that follows anaesthesia of the sciatic nerve is due, at least in part, to reduction of tonic PAD exerted on the distant (inappropriate) saphenous nerve spinal projections by the activity transmitted through the sciatic nerve afferents. Therefore, in the initial series of experiments we tried to measure the changes in the intraspinal threshold of the Saph terminals projecting caudally towards the sciatic projection regions (see Methods). However, with the cat fixed in a prone position, recording the antidromic responses produced in the Saph nerve by intraspinal stimulation was rather difficult. After several unsuccessful attempts, we instead examined the changes in the intraspinal threshold of the mSU nerve terminals following the acute section of the Saph and/or the SP nerves. We expected that sectioning smaller nerves instead of the whole sciatic nerve would produce similar changes in the tonic PAD exerted on intact cutaneous afferents.

In six experiments, we examined the effects of the acute Saph nerve section on the IFPs evoked by stimulation of the peripheral (intact) mSU nerve and also on the intraspinal threshold of the mSU nerve terminals. Figure 2 shows data from one of these experiments. Before sectioning the Saph nerve, stimulation of the mSU nerve with stimuli as low as 1.1 *T* already produced detectable IFPs that were increased with stronger stimuli up to saturation (see Fig. 2*A* and *C*, black bars).

Recordings made 17 min after the acute section of the Saph nerve showed a clear increase of the mSU-evoked IFPs (Fig. 2*B* and *C*, blue bars). This increase was proportionally larger for the responses evoked with the weakest stimuli (1.1 *T*) than for responses evoked with strengths above 1.6 *T*. The potentiation of the mSU-evoked IFPs was still significant 2 h after the Saph nerve section.

After retrieving the mSU IFPs evoked by stimuli of different strengths, the recording micropipette was switched to the stimulating mode. Intraspinal stimuli were then applied while recording the antidromic responses from the intact mSU nerve. As shown in Fig. 2*D* and *F*, black bars, intraspinal stimulation produced mSU antidromic compound action potentials whose amplitude

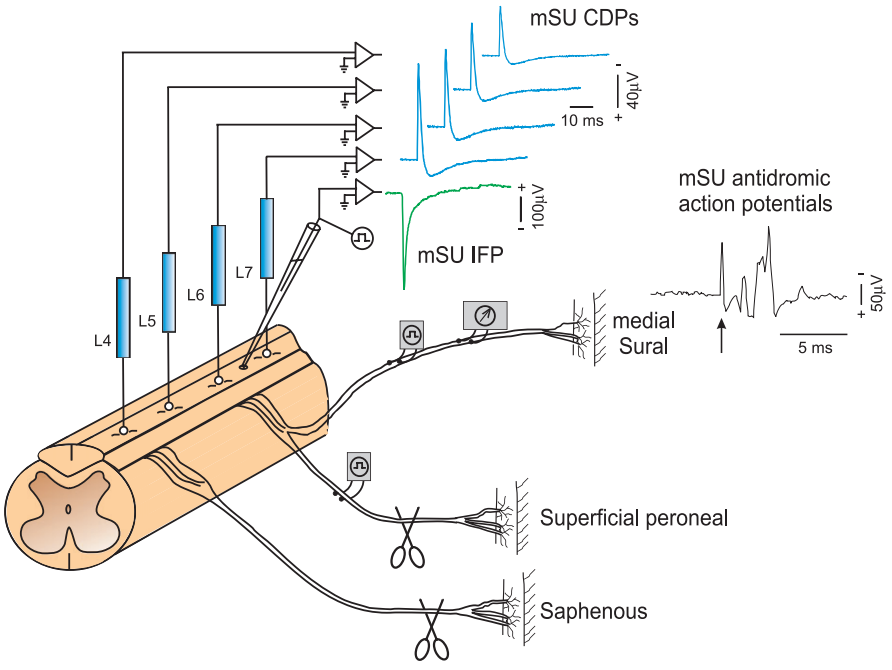

Figure 1. Diagram of the method
Cord dorsum potentials (CDPs) and intraspinal field potentials (IFPs) were simultaneously recorded from the lumbosacral spinal cord. The inset shows CDPs in the medial branch of the sural nerve (mSU) recorded from several spinal segments and the mSU IFPs recorded within the dorsal horn following stimulation of the medial sural nerve with single pulses of two times threshold (*T*) strength. By means of a switch, the recording micropipette was subsequently connected to a constant-current pulse generator for intraspinal stimulation. Antidromic responses were then recorded from the intact mSU nerve, as shown. Arrow indicates stimulus artifact. The superficial peroneal and saphenous nerves were dissected, left intact and prepared for acute transection during the experiment. See text for further explanations.

increased with stimulus strength (from 1.1 to 4 *T* relative to the strength producing detectable antidromic responses). These responses had onset latencies between 1.7 and 2.1 ms, suggesting that they were produced by direct activation of the intraspinal terminals of afferent fibres (Jankowska *et al.* 1981).

The same procedure was repeated 32 min after sectioning the Saph nerve, and we could observe a clear reduction of the mSU antidromic responses evoked throughout the whole range of intraspinal stimuli (Fig. 2*E* and blue bars in Fig. 2*F*), a finding suggesting reduced tonic PAD and presynaptic inhibition that would partly contribute to the generation of larger mSU field potentials (see Discussion).

In the experiment of Fig. 3, sectioning the Saph nerve produced a mild facilitation of the IFPs elicited by 1.6 *T* stimulation of the mSU nerve (Fig. 3*A* and *B*). This facilitation was observed throughout the whole range of stimulus strengths (1.2–2 *T*; Fig. 3*D*, black and blue bars).

Intraspinal stimuli applied through the glass micropipette produced antidromic responses in the mSU nerve. These responses could be elicited with stimuli as low as 1.2 *T* and became larger with increasing strengths (Fig. 3*E* and *G*, filled bars). Twenty-five minutes after the acute section of the Saph nerve the mSU antidromic responses elicited with the weakest stimuli (1.2–1.6 *T*) were practically abolished (Fig. 3*H*), and stronger stimuli (1.8 and 2.0 *T*) produced small responses (Fig. 3*J*, filled blue bars), indicating an increase in the threshold for activation of the intraspinal mSU nerve terminals.

We also examined the changes in phasic PAD of the mSU afferents that followed the acute section of the Saph nerve. Figure 3*F* shows that the mSU antidromic responses produced with intraspinal stimuli of 1.6 *T* were facilitated when conditioned with single pulses of 2 *T* strength applied

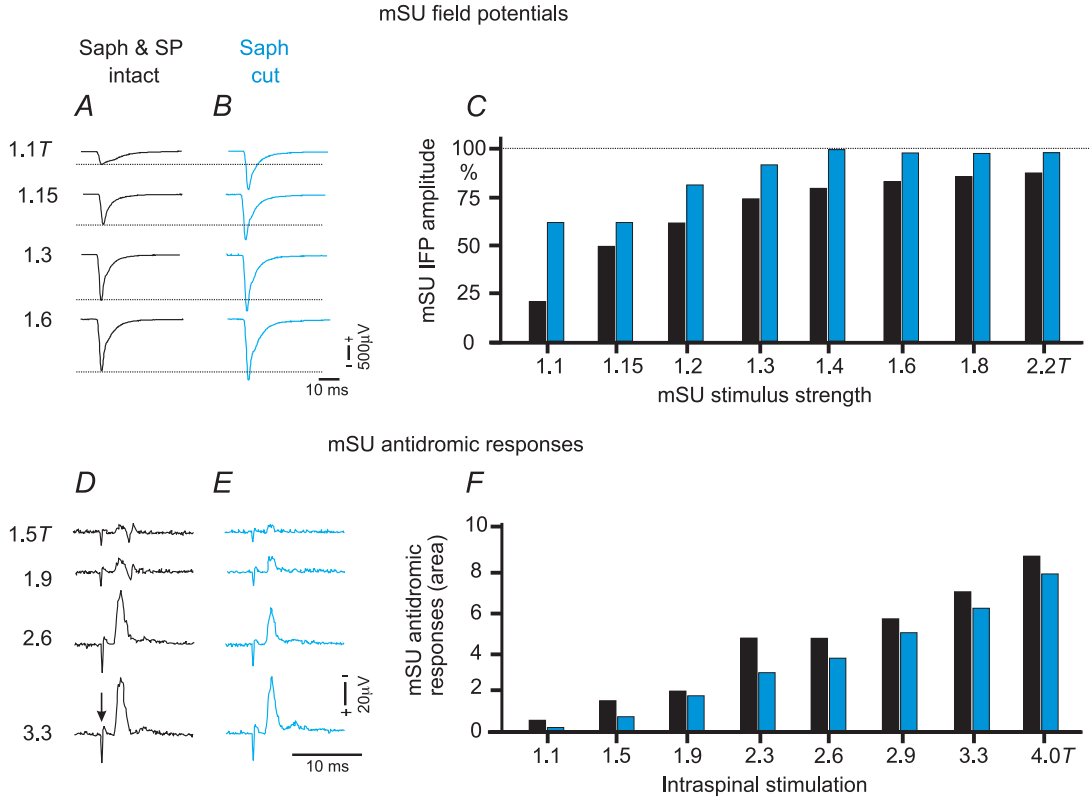

Figure 2. Acute sectioning of the saphenous nerve increases the mSU IFPs and reduces the tonic primary afferent depolarization of mSU intraspinal terminals
A, IFPs recorded in the dorsal horn following mSU nerve stimulation with single pulses of 1.1–1.6 *T*, with the Saph and SP nerves intact. *B*, the same, 17 min after the acute section of the left Saph nerve. *C*, amplitude of the mSU IFPs evoked with different strengths before (black bars) and after (blue bars) the acute section of the Saph nerve. Abscissae, mSU stimulus strength expressed as multiples of the threshold of the most excitable fibres (*T*). Ordinates, percentage change relative to the maximal response amplitude attained after the Saph nerve section. *D*, antidromic responses recorded from the intact mSU nerve following intraspinal stimulation with different stimulus strengths. Arrow indicates stimulus artifact. *E*, the same, but 32 min after the acute section of the Saph nerve. *F*, changes in the area of the mSU antidromic responses partly illustrated in *D* and *E*. Black bars, before and blue bars, after the acute section of the Saph nerve. The tip of the micropipette used to record the mSU IFPs and for intraspinal stimulation was in the L6 segment at 1.4 mm depth. Further explanations can be found in the main text.

to the SP nerve 35 ms earlier. This facilitation was seen throughout a wide range of test responses (1.2–1.8*T*; see open bars in Fig. 3*G*).

Although sectioning the Saph nerve reduced quite markedly the mSU antidromic responses, SP conditioning still produced phasic PAD, particularly when using the weakest intraspinal stimuli (Fig. 3*I* and *J*, open bars). However, the facilitation produced by SP conditioning stimulation, even with the highest strengths (2*T*), failed to compensate for the reduction of the mSU antidromic responses that followed the acute section of the Saph nerve.

Altogether, we found in four experiments that the acute Saph nerve transection increased the mSU IFPs and reduced the mSU antidromic responses elicited by intraspinal stimulation, i.e. reduced the tonic PAD (see Fig. 4*A*, blue bars), while in two experiments sectioning the Saph nerve instead increased the antidromic SU responses throughout the whole range of intensities, particularly in the low-threshold range (1.1–1.3*T*; see Fig. 4*A*, red bars). In these experiments, the IFPs produced by mSU nerve stimulation were also facilitated after the nerve transection (not illustrated).

Changes in IFPs and PAD produced by the acute section of the SP nerve

The main spinal projections of the Saph afferents are clearly rostral to the projection of the SU afferents (Bernhard, 1953). This led to the question of whether the same effects on the SU IFPs and on the intraspinal threshold of the fibres would be attained by sectioning the SP afferents that project more caudally, in regions partly overlapping with the projections of the SU afferents.

In two of six experiments, we found that the acute section of the SP nerve reduced the mSU antidromic potentials evoked by intraspinal stimulation. Figure 5 illustrates data obtained in one of these experiments.

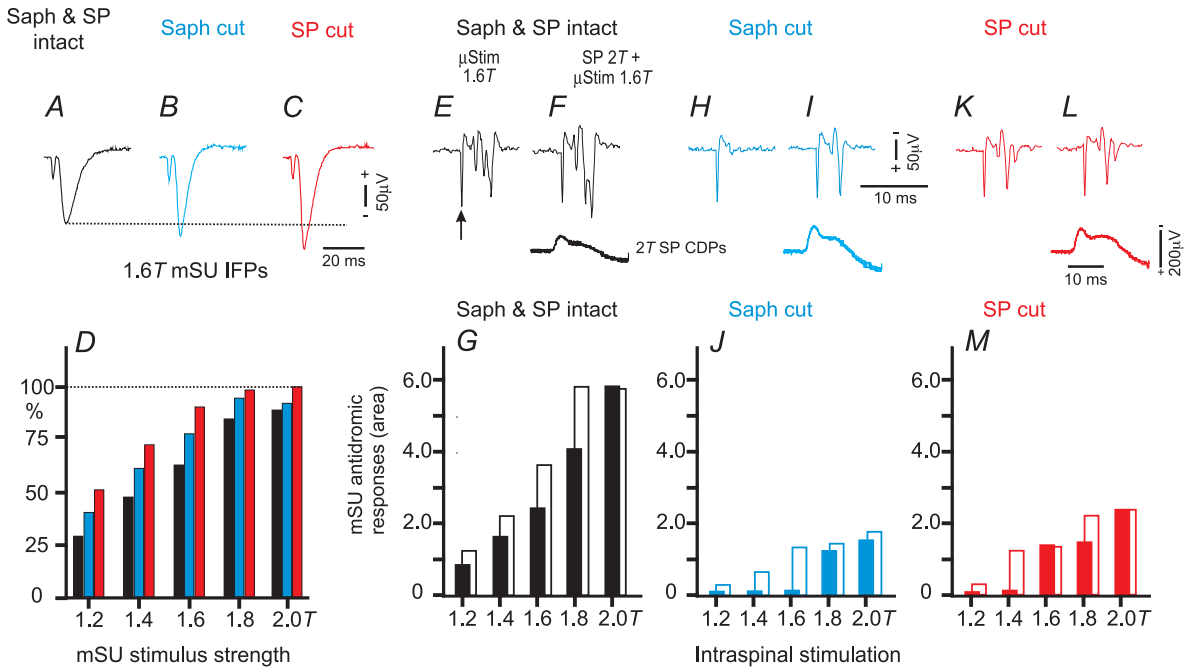

Figure 3. Facilitation of the mSU IFPs is associated with a reduced tonic PAD of mSU terminals
A, IFPs produced by stimulation of the mSU nerve with single pulses of 1.6*T*. *B*, the same, 15 min after the acute section of the Saph nerve. *C*, 45 min after Saph nerve transection and 15 min after the additional SP nerve section. *D*, graph relating mSU IFP amplitude *versus* mSU stimulus strength obtained from data partly shown in *A*–*C*. Black bars, mSU IFPs recorded with the Saph and SP nerves intact; blue bars, the same, after Saph nerve section; and red bars after SP acute section. Abscissae, mSU stimulus strength measured before the nerve lesions. Ordinates, amplitude of IFPs as a percentage of the maximal response attained after the SP nerve section. *E*, antidromic mSU nerve responses produced by intraspinal stimulation (μStim) with single pulses of 1.6*T*. Arrow indicates stimulus artifact. *F*, the same, but intraspinal test stimuli preceded by SP conditioning stimulation (single pulses of 2*T* applied 35 ms before). Traces below show 5 superposed CDPs produced by the SP conditioning stimulus. *G*, area of unconditioned (black bars) and of SP-conditioned mSU antidromic responses (open bars) produced with intraspinal stimuli of different strengths. *H*–*J* the same, but 25 min after the acute section of the Saph nerve. *K*–*M*, the same, but 45 min after the additional section of the SP nerve. The tip of the micropipette used for recording and for intraspinal stimulation was placed in the L6 segment at 0.8 mm depth. See main text for further explanations.

The recordings displayed in Fig. 5*A* show the mSU antidromic potentials generated by intraspinal stimulation with different strengths, as indicated (filled bars in Fig. 5*C*). These potentials were facilitated when preceded by single 2*T* pulses applied to the SP nerve 35 ms before the intraspinal stimuli (Fig. 5*B* and *C*, open bars).

Twenty-five minutes after the acute section of the SP nerve, the mSU antidromic potentials produced with intraspinal stimuli of 1.2–2*T* strength showed a significant reduction (Fig. 5*D* and *F*, filled bars), but even so conditioning stimulation of the SP nerve facilitated the already depressed mSU antidromic responses, particularly in the low-threshold range (Fig. 5*E* and *F*, open bars).

In four experiments, the acute section of the SP nerve was found instead to increment the tonic PAD elicited in the lowest threshold mSU intraspinal terminals. One example of this situation is illustrated in Fig. 6. Thirty minutes after the SP nerve section, the mSU IFPs were increased to 118% of control values, remained so for about 30 min more, and increased to 163% 20 min later (Fig. 6*B*–*D*). The antidromic SU responses generated by intraspinal stimulation 40 min after the SP nerve section were increased throughout a wide range of intensities (1.5–5*T*; see Fig. 6*G* and *H* and black and red bars in Fig. 6*J*).

Figure 7 provides another example where the acute section of the SP nerve also increased the mSU IFPs (see Fig. 7*A* and *B* and black and red bars in *E*) as well as the mSU antidromic potentials produced by low strengths of intraspinal stimulation within the dorsal horn (1.2–1.3*T*; Fig. 7*F*, red traces and *L*, filled bars), but reduced the responses produced with stronger stimuli (1.8 and 2.0*T*; see Fig. 7*H*, red trace, and *L*, filled bars).

Before the SP nerve section, conditioning stimulation of the SP nerve facilitated the mSU antidromic responses elicited with intraspinal stimuli of 1.2–2.0*T* (upper black traces in Fig. 7*G* and *I* and open bars in *K*). After the acute section of the SP nerve, conditioning stimulation of the SP nerve still increased the mSU antidromic responses produced with stimuli of 1.2–1.8*T* to about the same extent as before the nerve section (Fig. 7*G* and *I*, red traces, and *L*, open bars).

Altogether, these observations indicate that acute section of the Saph and/or of the SP nerves may either increase or reduce the tonic PAD exerted on the SU terminals, as evidenced by changes in their excitability, while at the same time there is a general increase of the SU-evoked field potentials, although the magnitude and duration of this potentiation was not always the same. For example, in the experiment of Fig. 6, the potentiation was relatively small 30 min after the SP nerve section and became large by 80 min later, while in the experiment of Fig. 7 it was already large 20 min after the SP nerve lesion.

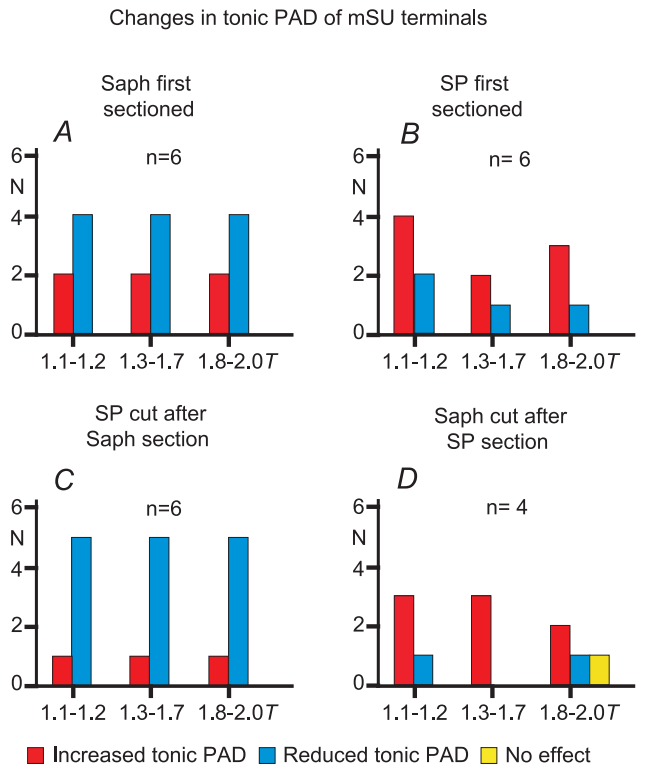

Figure 4. Summary of effects produced by acute nerve sections on tonic PAD of mSU terminals within the dorsal horn
The graphs show the number of experiments in which the acute section of the Saph and/or SP nerves increased (red bars), reduced (blue bars) or had no effect (yellow bars) on the tonic PAD of mSU intraspinal terminals, plotted against the strength of the intraspinal stimuli used to generate the antidromic mSU responses, as indicated. Further explanations can be found in the main text.

Changes produced by a second nerve section

As described in the preceding sections, in some experiments the acute section of the Saph or the SP nerves reduced the tonic PAD of the SU afferent terminals, as suggested by Biella & Sotgiu (1995), while in other experiments these nerve sections had the opposite effect. Given that the SP spinal projections are closer to the SU nerve projections than those of the Saph fibres, it seemed possible that local events triggered by the SP nerve section contributed to the increased tonic PAD of the SU afferents. If this were the case, we expected that the effects produced by sectioning the SP nerve after a preceding Saph nerve section would be similar to those observed when the SP nerve was sectioned without being preceded by the Saph nerve section.

The acute transection of the SP nerve made after the Saph nerve was cut further incremented the already potentiated mSU IFPs (Fig. 3*C* and *D*, red bars). In contrast, the mSU antidromic responses produced by

intraspinal stimulation with weak stimuli (1.2 and 1.4*T*) remained practically the same, while the responses produced with higher strengths (1.6 and 2.0*T*) were clearly increased (Fig. 3*K* and *M*, filled bars). After the acute SP nerve section, conditioning stimulation of this nerve still increased the antidromic mSU responses evoked by the weakest stimuli (1.2 and 1.4*T*).

In the experiment of Fig. 6, sectioning the Saph after the SP nerve slightly reduced the already facilitated mSU IFPs and CDPs produced with stimuli of 2*T* (the effects on IFPs produced with weaker stimuli were not tested). Yet, the SU antidromic responses showed a clear additional increase relative to the values attained after the SP nerve section throughout the whole range of tested strengths of intraspinal stimuli (from 1.5 to 5*T*; see Fig. 6*I* and *J*, blue bars). However, in other experiments, the additional section of the Saph nerve significantly increased the mSU IFPs elicited with stimuli up to 2.0*T* (see Figs 7*C* and *D* and blue bars in *E*). This facilitation lasted more than 1.5 h and was associated with a significant increase in the tonic and phasic PAD of the mSU intraspinal terminals, particularly when activated with stimuli of 1.8 and 2*T* (see Fig. 7*H* and *I*, blue traces, and *M*).

Figure 4 summarizes the effects on the tonic PAD of mSU nerve terminals produced by the acute section of the Saph and SP nerves. The graphs show the number of experiments in which the tonic PAD was increased (red bars), reduced (blue bars) or unaffected (yellow bars) by the first or second nerve section, as indicated. These effects were separated into three groups according to the strength of the intraspinal stimuli used to generate the antidromic responses of the mSU afferents. It may be seen in Fig. 4*A* that after the acute Saph nerve section the tonic PAD was increased in two and reduced in four

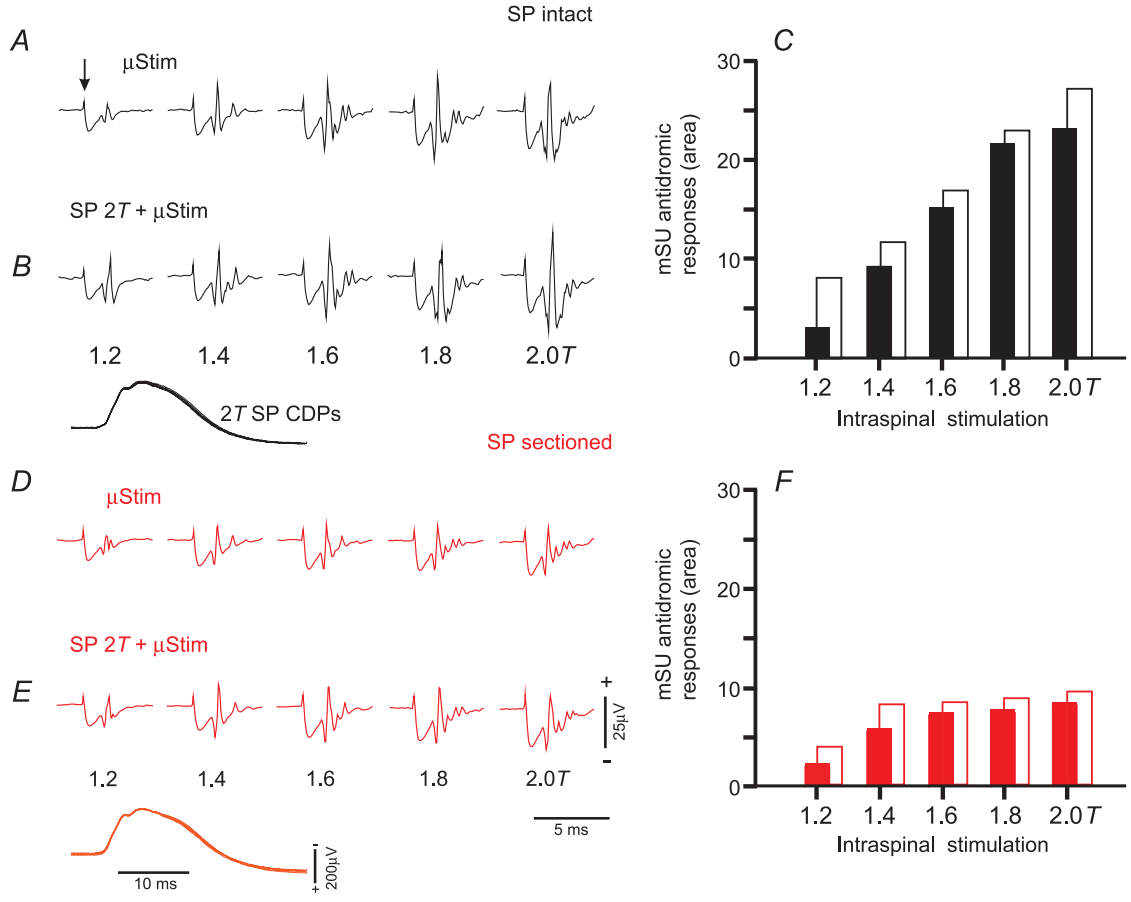

Figure 5. Acute sectioning of the SP nerve reduces the tonic PAD of mSU intraspinal terminals
A, antidromic mSU responses produced by dorsal horn intraspinal stimulation (µStim) with different strengths, as indicated. Arrow shows stimulus artifact. *B*, test antidromic responses preceded by SP conditioning stimulation with single pulses of 2*T* applied 35 ms before. Traces below show 5 superimposed CDPs produced by the SP conditioning stimulus. *C*, area of the mSU antidromic responses in arbitrary units *versus* stimulus strength of data depicted in *A* and *B*. Black bars, unconditioned and open bars, SP-conditioned antidromic responses. *D–F*, the same as in *A–C*, but 25 min after the acute section of the SP nerve. Note reduced tonic PAD of mSU intraspinal terminals. The tip of the micropipette used for recording and for intraspinal stimulation was placed in the L6 segment at 0.8 mm depth. See main text for further explanations.

experiments and that this proportion remained the same for antidromic responses produced with low and with stronger intraspinal stimuli. The acute section of the SP nerve increased the tonic PAD of the most excitable mSU afferents in four experiments and reduced the mSU tonic PAD in two experiments (Fig. 4*B*).

Figure 4*C* and *D* shows that the effects produced by the second nerve section resembled those produced by the first nerve section. In most experiments, sectioning the SP nerve after the Saph nerve reduced the tonic PAD (Fig. 4*C*), while sectioning the Saph nerve after the SP nerve increased the tonic PAD (Fig. 4*D*).

These findings suggest that the changes in the tonic PAD produced by the acute section of the Saph and SP nerves were to some extent independent of the segmental location of the projection of these two nerves relative to the projection of the SU afferents.

Changes in the synaptic efficacy of the SP conditioning volleys

We found in several experiments that although the acute section of the Saph and SP nerves reduced the tonic

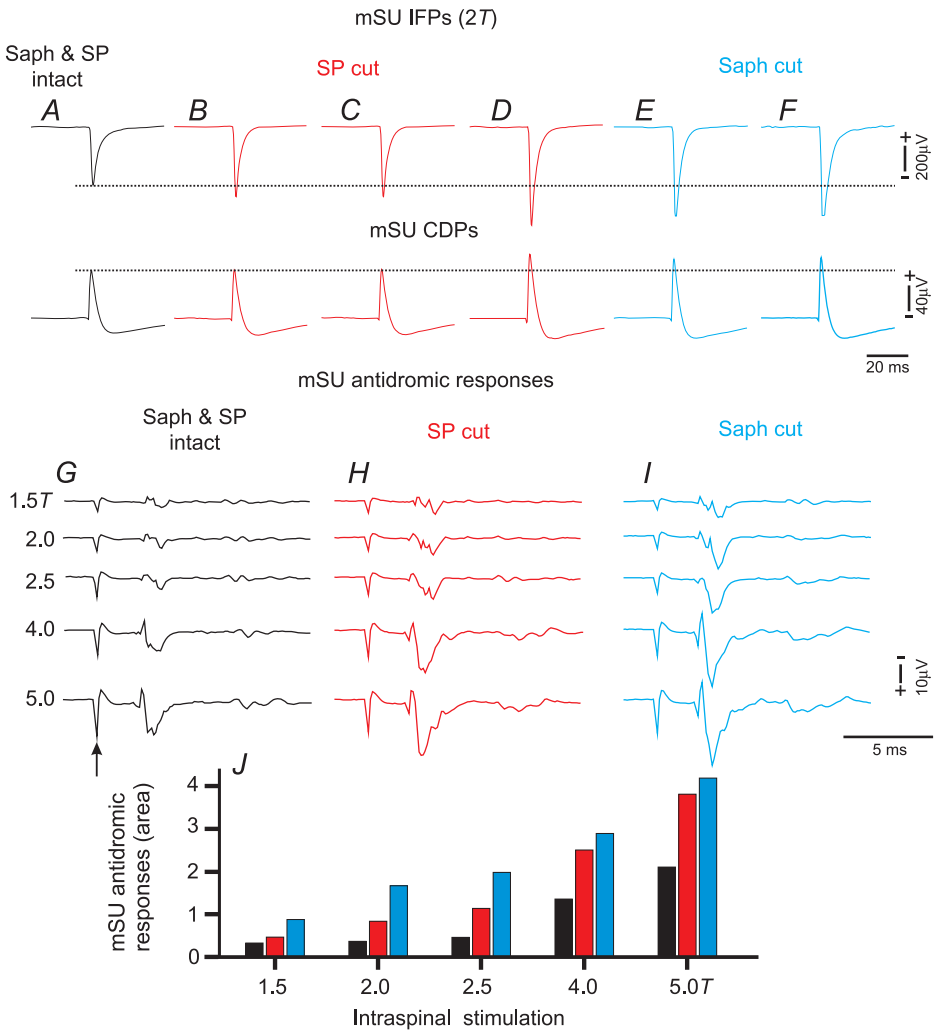

Figure 6. Long-lasting potentiation of the mSU IFPs and increased tonic PAD produced by SP and Saph nerve transection
A–F, intraspinal mSU IFPs and CDPs produced by single pulses of 2*T* strength, recorded before (black traces), 30, 60 and 80 min after SP nerve transection (red traces) and 30 and 60 min after the additional Saph nerve section (blue traces). *G–I*, antidromic mSU responses produced by intraspinal stimulation with different strengths, before (*G*), 40 min after SP nerve section (*H*) and 75 min after the Saph nerve section (*I*; made 90 min after SP nerve transection). Arrow indicates stimulus artifact. *J*, area of antidromic responses illustrated in *G*, *H* and *I*. Black bars before and red and blue bars after SP and Saph nerve transection, respectively. The tip of the micropipette used for recording and for intraspinal stimulation was placed in the L6 segment at 2 mm depth. See main text for further explanations.

PAD of the mSU terminals, the phasic PAD produced by conditioning stimulation of the SP nerve appeared not to be reduced. As the acute nerve section increased the mSU IFPs, it seemed possible that the effectiveness of the SP afferents was also increased after the nerve lesions. In order to evaluate this possibility, we examined the changes produced by the nerve lesions on the CDPs evoked by the SP conditioning stimuli used to generate phasic PAD of the mSU afferents.

The lower sets of traces in Fig. 3*F*, *I* and *L* show superimposed CDPs produced by single pulses of 2*T* applied to the SP nerve. It may be seen that after the acute section of the Saph nerve, these potentials were increased to about 160% of their control amplitude (Fig. 3*I*), and so were the SP-conditioned antidromic mSU responses produced with low-strength stimuli (1.2–1.6*T*). Although the subsequent section of the SP nerve produced no additional effects on the SP CDPs peak amplitude (Fig. 3*L*), SP conditioning still facilitated the mSU antidromic responses produced by the weakest stimuli (Fig. 3*M*, open bars).

In the experiment of Fig. 5, the acute section of the SP nerve had a very small effect, if any, on the SP CDPs evoked with single pulses of 2*T* (lower traces in Fig. 5*B* and *E*). Yet, the increment in the area of the SP-conditioned mSU antidromic responses remained about the same, suggesting antidromic activation of a similar number of

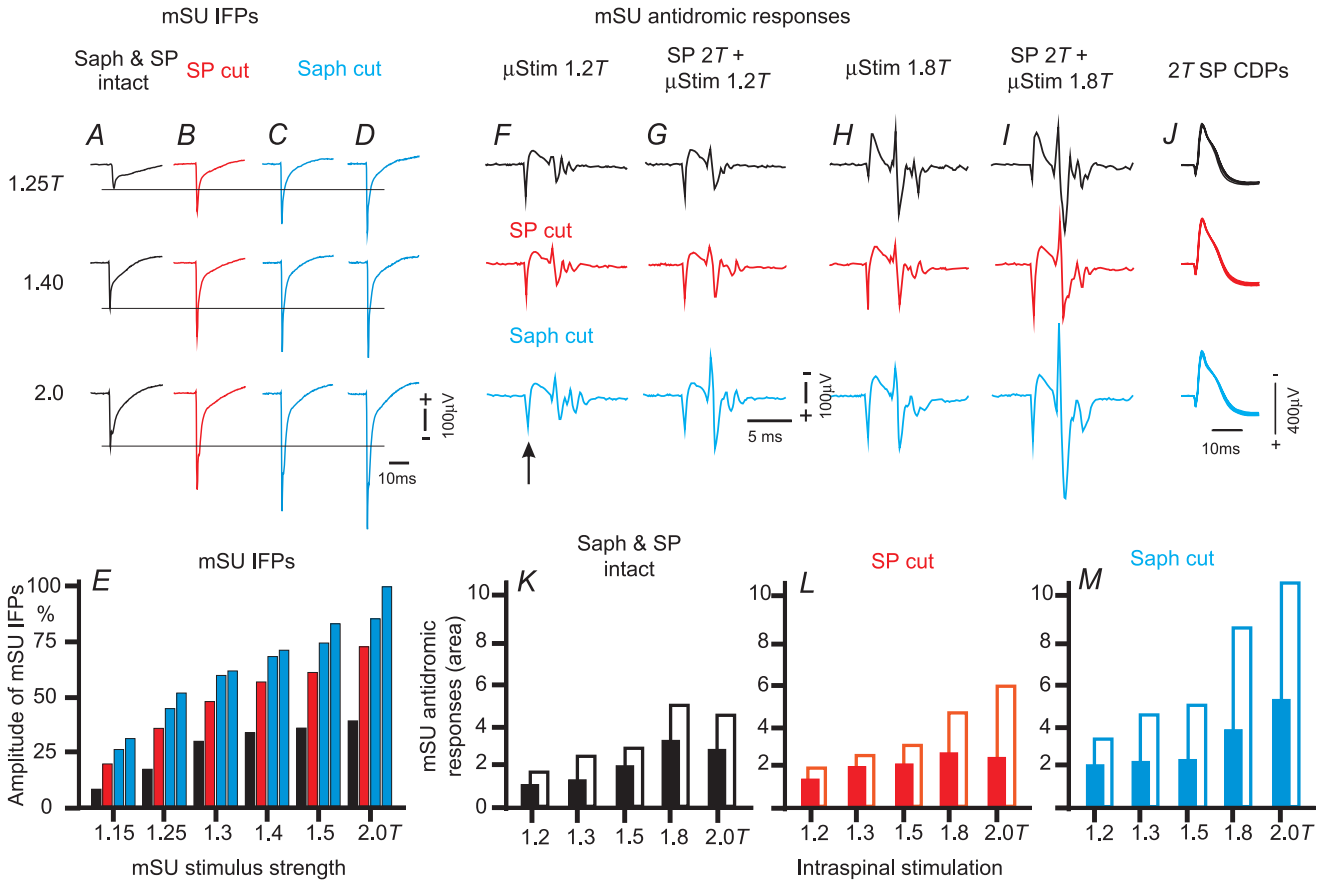

Figure 7. Acute transection of the SP and Saph nerves increases mSU IFPs as well as tonic PAD in lowest threshold mSU afferents
A–D, mSU IFPs produced with 3 different strengths, before (*A*), 20 min after the acute section of the SP (*B*) and 30 and 60 min after the additional section of the Saph nerve (*C* and *D*). *E*, amplitude of mSU IFPs produced with graded stimulation. Graph obtained from recordings partly illustrated in *A–D*. Black bars, before and red and blue bars after acute SP and additional Saph nerve transections, respectively. *F*, mSU antidromic responses produced by intraspinal stimulation (μStim) with pulses of 1.2*T*, before (black trace), 35 min after SP nerve section (red trace) and 75 min after the additional section of the Saph nerve (blue trace). Arrow indicates stimulus artifact. *G*, same as *F*, but antidromic responses preceded by single conditioning SP stimuli of 2*T* applied 35 ms before. *H* and *I*, same as *F* and *G* for antidromic responses produced by intraspinal stimulation with pulses of 1.8*T*. *J*, 5 superimposed CDPs produced by the SP 2*T* stimuli used for conditioning stimulation in *G* and *I*, before and after SP and Saph nerve section as indicated. *K*, area of unconditioned (filled bars) and SP conditioned antidromic mSU responses (open bars) produced by intraspinal stimuli applied before the SP and Saph nerve sections. *L* and *M*, same after the SP and the Saph nerve transections, as indicated. The tip of the micropipette used to record the IFPs and for intraspinal stimulation was placed in the L6 segment at 2 mm depth. See main text for further explanations.

afferent fibres. In the experiment of Fig. 7, the SP-induced CDPs were only slightly increased after the acute SP and Saph nerve section (113 and 115%, respectively; Fig. 7*J*). Yet, there was a significant increase of the phasic PAD, particularly after the acute section of the Saph nerve (Fig. 7*M*, open bars).

These observations suggest that the acute nerve lesions may increase the synaptic effectiveness of the SP afferents, thereby counteracting, to some extent, reduced impulse transmission along the pathways mediating the phasic PAD of the mSU terminals (see Discussion).

Intraspinal sites of SU afferent excitability testing

Based on the finding of differential presynaptic inhibition in group II muscle afferents ending at the intermediate zone and dorsal horn (Riddell *et al.* 1995; Jankowska *et al.* 2002), we questioned the extent to which the similarity of the effects on the tonic PAD produced by the first and second nerve lesion could be related to the location within the dorsal horn of the tested mSU afferents.

As discussed in the Methods section, the intraspinal location of the sites used to test the excitability of the

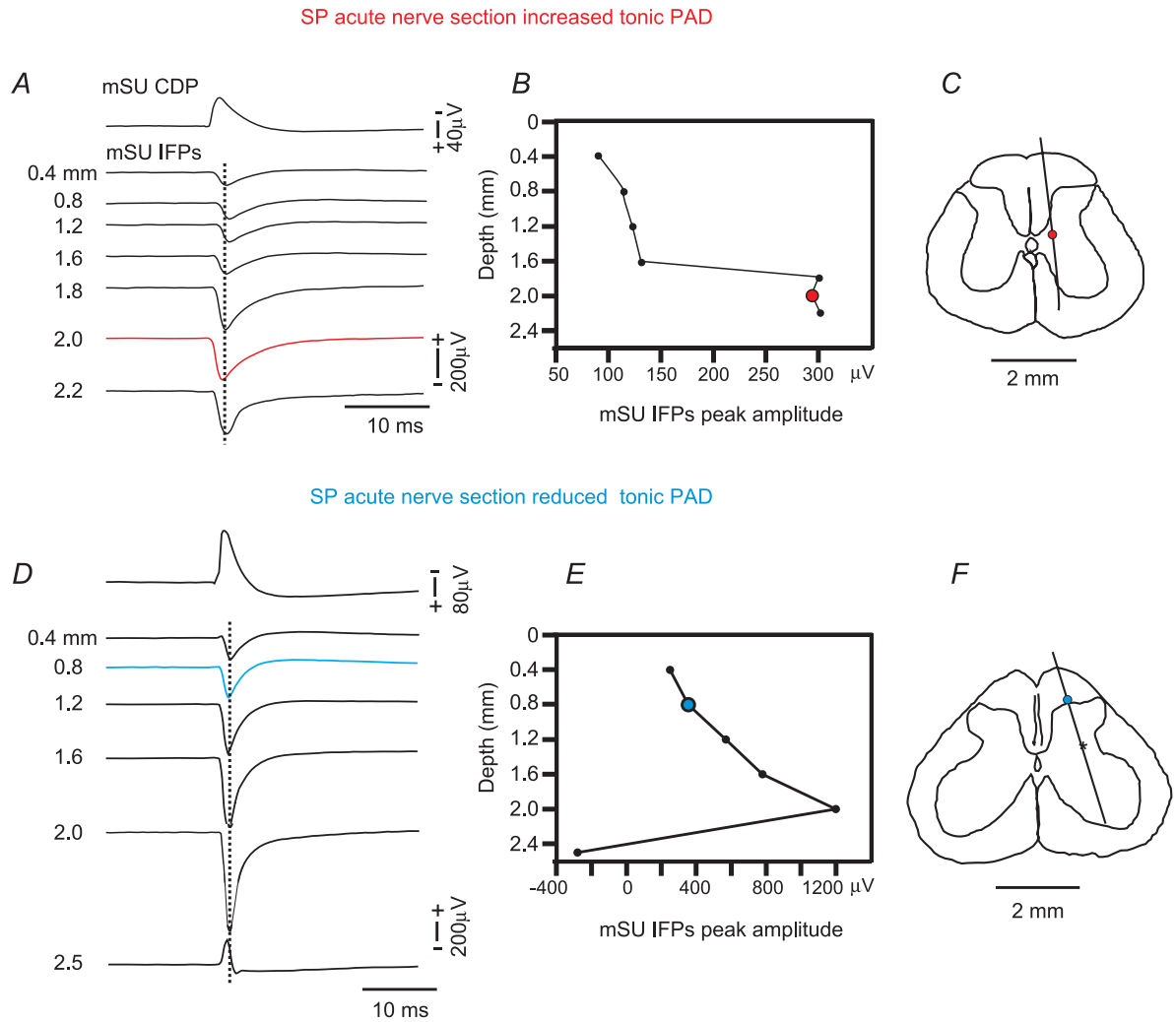

Figure 8. Intraspinal sites of excitability testing
A–C, data derived from an experiment where the acute section of the SP nerve increased the tonic PAD of the mSU afferents. Same experiment as that of Fig. 6. *A*, L6 CDP and mSU IFPs produced by single pulses of 1.4*T* recorded at different spinal depths, as indicated. Trace in red shows the IFPs recorded at the site of intraspinal threshold testing. *B*, peak amplitude of the IFPs displayed in *A versus* recording depth. Red circle shows site of intraspinal threshold testing. *C*, histology and estimated intraspinal location of threshold testing site. *D–F*, data from an experiment where the acute section of the SP nerve reduced the tonic PAD of mSU terminals. Same experiment as that of Fig. 5. Same format as in *A–C*. The mSU IFPs produced with single pulses of 1.6*T* were recorded 0.8 mm from the cord surface at the site indicated with the blue circle. Asterisk shows recording site of largest IFPs. See main text for further explanations.

mSU afferents was established by taking into account the marks left in the tissue by the micropipette, combined with depth measurements from the cord surface made during the experiment. In addition, the position of the stimulated sites was referred relative to the mSU IFPs recorded at different depths in that particular track.

Figure 8*A*–*C* shows data obtained in one experiment (same as that of Fig. 6) where the acute section of the SP nerve increased the antidromic mSU potentials, that is, the tonic PAD. The traces displayed in Fig. 8*A* show the L6 CDP and the IFPs produced by mSU nerve stimulation with single pulses of 1.4*T* recorded at different depths, as indicated. In that particular track, the mSU IFPs were relatively small in the superficial layers and attained their highest amplitude deeper, at a depth between 1.8 and 2.2 mm.

The plot of the peak amplitudes of the mSU IFPs recorded at different depths based on readings made during the experiment is shown in Fig. 8*B*. The red circle shows the location of the tip of the micropipette used for afferent threshold testing that was positioned at a site where the mSU IFPs were largest in that same track. The histological section depicted in Fig. 8*C* shows the electrode

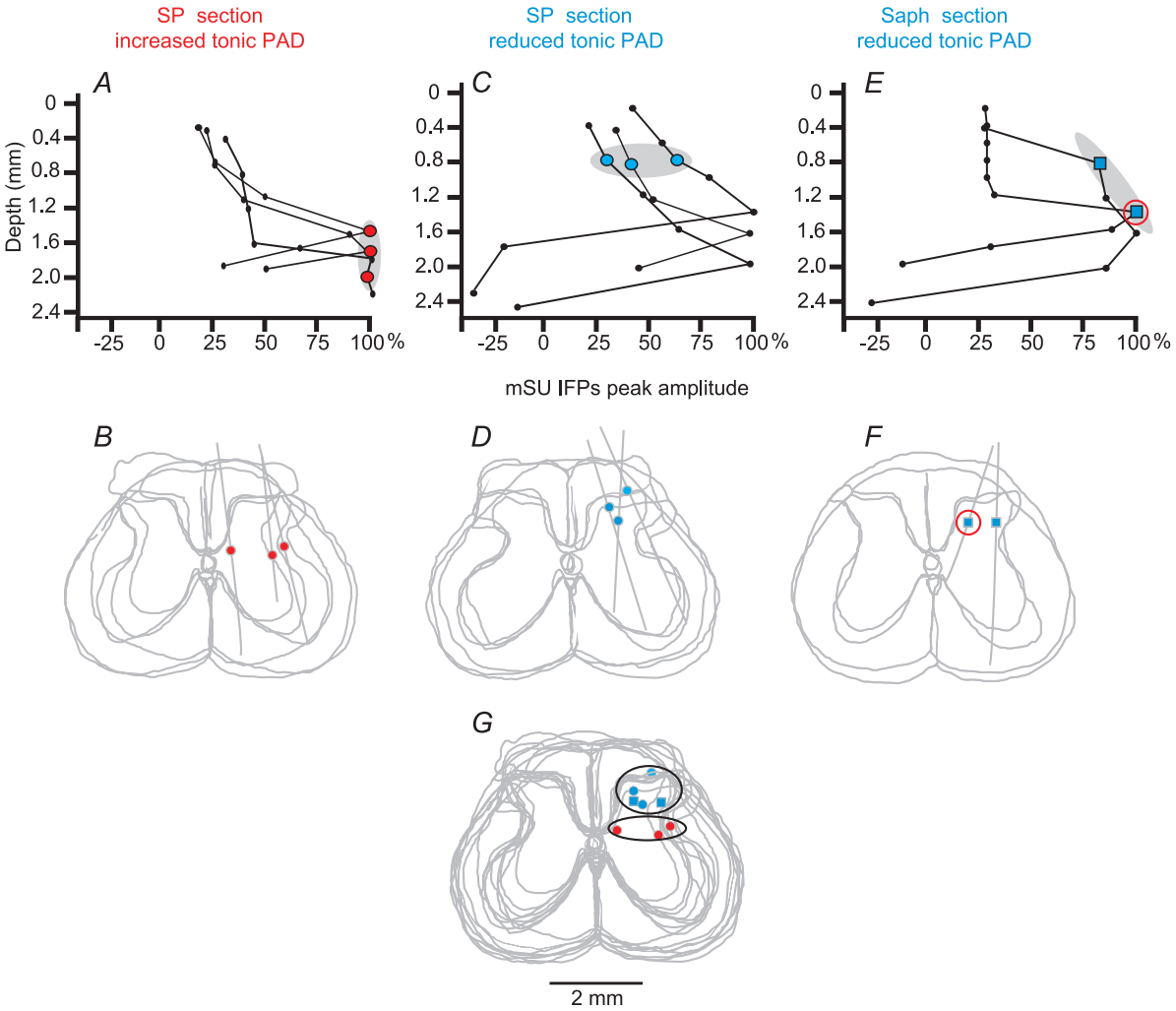

Figure 9. Intraspinal threshold testing sites and changes in tonic PAD induced by acute Saph and SP nerve transections
A and *B*, data derived from 3 different experiments where the acute (first) section of the SP nerve increased the tonic PAD of the mSU afferents. *A*, ordinates, depth distribution of mSU IFPs; abscissae, mSU IFPs peak amplitude plotted as a percentage of maximal responses. *B*, superimposed drawings of spinal sections showing electrode tracks. Red symbols indicate location of threshold testing sites. *C* and *D*, data from 3 experiments where the acute (first) section of the SP nerve instead reduced the tonic PAD of the mSU afferent terminals. Same format as in *A* and *B*. Blue circles, location of threshold testing sites. *E* and *F*, data from 2 experiments where the acute (first) section of the Saph nerve reduced the tonic PAD of the mSU terminals. Same format as in *C* and *D*. *G* shows superimposed drawings of spinal cord sections displayed in *B*, *D* and *F*. Further explanations can be found in the main text.

track with the presumed location of the excitability testing site.

Similar plots were made for data obtained in one experiment where the acute section of the SP nerve instead reduced the mSU antidromic responses produced by intraspinal stimulation in the low-threshold range (1.2–1.6*T*; same experiment as that of Fig. 5). It may be seen in Fig. 8*D* and *E* that in this case the site of afferent testing was rather dorsal, well above the site where the orthodromic mSU IFPs attained their maximal amplitude (blue trace and blue dot). This location agrees well with the presumed location obtained from histological reconstruction indicated by the blue circle in Fig. 8*F*. The site of the largest mSU IFPs recorded in the same track is indicated with an asterisk.

Figure 9 summarizes observations made in eight experiments where the effects produced by the first acute section of the Saph or SP nerves on the excitability of the low-threshold (1.2–1.6*T*) mSU afferents could be documented in detail and the histology was available. The format is similar to that shown in Fig. 8. In this case, the data were grouped according to the effects produced by the acute section of the SP and Saph nerves, as indicated. The graphs in Fig. 9*A*, *C* and *E* show the peak amplitudes of the mSU IFPs recorded at different depths, normalized relative to the maximal values (100%). The red circles, blue circles and blue squares show the sites of intraspinal threshold testing in each experiment. Figure 9*B*, *D* and *F* shows the corresponding histological sections with the presumed location of the stimulation sites drawn together with the electrode tracks, superimposed for proper comparison. The histology and stimulation sites depicted in Fig. 9*G* show the different locations of the intraspinal sites in which the acute section of the Saph and of the SP nerves reduced the mSU antidromic responses (blue symbols) from those sites where the nerve sections increased the antidromic responses (red symbols).

In the experiments depicted in Fig. 9*A* and *E*, we performed a second nerve lesion. Only in one of them were the effects produced by the second lesion opposite to those produced by the first lesion. That is, the Saph nerve lesion reduced the tonic PAD, while the subsequent lesion of the SP nerve increased the tonic PAD. The location of the excitability testing site is marked in Fig. 9*E* and *F* with a red circle surrounding the blue square.

It thus seems reasonable to propose that in those experiments where sectioning the SP nerve increased the mSU antidromic potentials, the stimulated sites were located deeper within the dorsal horn (around Rexed's laminae V–VI) than those sites where the acute section of the SP and of the Saph nerve reduced the mSU antidromic responses, which were located more superficially, around Rexed's laminae III–IV.

Discussion

Long-lasting facilitation of SU IFPs

The observations reported herein indicate that the acute section of the SP and Saph nerves produces a long-lasting facilitation of the intraspinal responses evoked by stimulation of low-threshold SU nerve afferents (1.1–1.6*T* strength), i.e. by stimulation of fibres in the Aβ range that mainly convey tactile information (Eccles *et al.* 1963; Schmidt, 1971). It is most likely that this facilitation results from the same mechanisms as those involved in the unmasking of the Saph nerve responses produced by local anaesthesia and/or chronic injury of the sciatic nerve described by other investigators (Markus & Pomeranz, 1987; Biella & Sotgiu, 1995), seen also after the intradermal injection of capsaicin and of other irritating substances (Neugebauer & Schaible, 1990; Winter *et al.* 1995; Cervero & Laird, 1996*a*; Cervero *et al.* 2003), or after acute injury (incision, burn) to the skin (see Lam *et al.* 2008; Vernon *et al.* 2009), which leads to a state of central sensitization related to the development of secondary hyperalgesia (Cervero & Laird, 1996*a*; Sandkühler & Gruber-Schoffnegger, 2012).

The finding that a second cutaneous nerve lesion produces an additional increment of the IFPs evoked by stimulation of the mSU nerve (Figs 3*D* and 7*E*) suggests further that the magnitude of the central facilitation induced by this procedure is not an all-or-none phenomenon, but rather an additive process that may well depend on the degree and extension of the induced damage.

Changes in tonic and phasic PAD

One of the purposes of this investigation was to examine the changes in PAD of the mSU intraspinal terminals occurring during the facilitation of the IFPs induced by the acute section of the Saph and SP nerves. We found in several experiments that, together with the facilitation of the mSU IFPs, the acute nerve sections reduced the mSU antidromic responses elicited by intraspinal stimulation. We have attributed the increased threshold that follows the acute nerve transection to a reduced tonic PAD. A reduction of the concurrent presynaptic inhibition would partly contribute to the potentiation of the responses evoked by stimulation of the mSU afferents, in agreement with the proposal of Biella & Sotgiu (1995).

We have assumed further that these effects result from changes in impulse transmission along the neuronal pathways mediating a $GABA_A$ PAD (see also Cervero *et al.* 2003), but this remains to be tested, because there is evidence suggesting that noradrenaline (Jeftinija *et al.* 1981) and 5-hydroxytryptamine (Carstens *et al.*

1981), as well as substance P (Randić *et al.* 1982), reduce the excitability of the cutaneous nerve terminals, and are released by the concurrent activation of descending pathways (Vanegas & Schaible, 2004).

However, in several experiments the facilitation of the mSU IFPs was associated with opposite effects on tonic PAD, because the acute section of the Saph or of the SP nerve was found to increase rather than decrease the SU antidromic responses produced by intraspinal stimulation. To the extent that these changes resulted from incremented tonic $GABA_A$ PAD, this would lead to an increased presynaptic inhibition and may not have contributed to the potentiation of the mSU-evoked orthodromic responses, which should instead be attributed to postsynaptic mechanisms (Biella & Sotgiu, 1995).

The possibility that the increase in tonic PAD was not due to activation of $GABA_A$ receptors must also be considered. In fact, there is evidence that iontophoretically injected 5-HT, noradrenaline or substance P may reduce the threshold of sural afferents in the cat spinal cord (Carstens *et al.* 1981; Jeftinija *et al.* 1981; Randić *et al.* 1982). These actions would also lead to a reduced synaptic effectiveness of the affected afferents (Carstens *et al.* 1981).

Are changes in tonic and phasic PAD mediated by the same mechanisms and/or pathways?

It is generally accepted that primary afferent hyperpolarization (PAH) in muscle and articular afferents results from inhibition of the tonic PAD generated by the sustained activity of the pathways that mediate the phasic PAD (Rudomin *et al.* 2004*a*,*b*; Rudomin & Lomelí, 2007). If the same situation holds for mSU afferents, we would expect that the acute section of the Saph and SP nerves had similar effects on the tonic as well as on the phasic PAD. That is, both should be either increased or decreased following the acute nerve sections. However, as shown in Figs 3 and 5, this seems not to be the case, because, although the acute section of the cutaneous nerves significantly reduced the tonic PAD, SP conditioning stimulation still produced a significant phasic PAD.

The persistence, or even increase, of the phasic PAD despite the reduction in the tonic PAD observed in these experiments could be due, at least in part, to an increased effectiveness of the SP afferents produced by the acute nerve section, similar to that seen for the mSU afferents. This appears to be the case for the Saph nerve section in the experiment of Fig. 3 and perhaps also in the experiment of Fig. 7, where the SP CDPs were increased after the Saph nerve section, but it was probably not the case in the experiment of Fig. 5, where the SP CDPs and the phasic PAD remained about the same even though there was a strong reduction of the tonic PAD.

Seen in retrospect, a more reliable procedure would have been to produce the phasic PAD of mSU afferents by stimulating some descending pathways (e.g. rubrospinal and reticulospinal), because their synaptic efficacy appears not to be affected by presynaptic $GABA_A$ mechanisms (Rudomin *et al.* 1975, 1980, 1981).

Nevertheless, it should be noted that the phasic PAD produced in muscle and articular afferents by stimulation of cutaneous nerves can be fairly selective. It may affect some, but not all, intraspinal terminals of individual fibres, or even produce PAD in one collateral and PAH in another collateral of the same fibre (Quevedo *et al.* 1997; Rudomin & Lomelí, 2007). To the extent that this selectivity is also a feature of the mSU afferents, it could explain those situations where the acute nerve section changed the tonic and phasic PAD in different directions. Even so, the possibility that different mechanisms and/or pathways mediate the changes of the tonic and phasic PAD produced by the acute nerve section should be left open, particularly in view of recent evidence in the turtle spinal cord suggesting that extrasynaptic α_5-$GABA_A$ receptors are involved in the generation of tonic PAD (Loeza *et al.* 2012).

Some functional considerations

We have previously assumed that the correlation between the spontaneous nCDPs and npCDPs simultaneously recorded from different spinal segments represents, to some extent, the segmental distribution of the functional connectivity between the dorsal horn neuronal ensembles involved in the generation of these potentials (Chávez *et al.* 2012). The functional connectivity between these sets of dorsal horn neurones was changed after the acute section of the cutaneous nerves. Although there was a general increase in the correlation between paired sets of spontaneous npCDPs, the observed changes were not homogeneous. The correlation between the npCDPs recorded from L5 and L6 segments was relatively high and showed smaller increments following the acute nerve section than the correlation between CDPs recorded from pairs of distant segments (e.g. L4–L7, L3–L6). It thus seems that the acute section of cutaneous nerves would generate differences in the magnitude of the tonic and phasic PAD exerted on the cutaneous afferents ending in different segments or at different spinal depths.

As shown in Figs 8 and 9, the location of the threshold testing sites relative to the intraspinal distribution of the mSU-evoked IFPs, together with the locations derived from the histological sections, suggest that in those experiments where the acute section of the Saph or the SP nerves reduced the mSU antidromic responses, the stimulated sites were within Rexed's layers III–IV, while in

those experiments where the nerve acute section increased the tonic PAD, the stimulated sites appeared to be located deeper within the dorsal horn (Rexed's layers V–VI).

These findings support the proposal that cutaneous afferents are subjected to a differential control of the tonic PAD exerted according to their intraspinal sites of termination within the dorsal horn. This differential control would be similar to that exerted on group II muscle afferents, where there are differences in the degree of presynaptic inhibition within the regions of projection in the dorsal horn and in the intermediate zone of the same segments (Riddell *et al.* 1995; Jankowska *et al.* 2002).

We have assumed that the mSU antidromic potentials elicited by intraspinal stimulation were due mostly to activation of low-threshold afferent fibres that converge, together with group II muscle and articular afferents, onto the same sets of spinal neurones (Lundberg *et al.* 1987). This being the case, it is not at all surprising that, like the group II muscle afferents, cutaneous afferents projecting to different spinal regions are also subjected to a differential presynaptic control. This possibility raises a question concerning the extent to which the presynaptic control of the information transmitted by cutaneous, joint and group II afferents is mediated by the same or by different sets of interneurones. Involvement of separate neuronal sets could be an efficient mechanism, allowing a selective and dynamic adjustment of the information arriving at the receiving neurones from different sources (muscle, skin or joints) according to the task to be executed.

We have assumed further that the cutaneous afferents projecting dorsally are subjected to a tonic presynaptic inhibition that reduces their synaptic efficacy. Sectioning the SP and/or the Saph nerves would remove this inhibition and increase their synaptic effectiveness. This would promote the activation by cutaneous inputs of neurones in a wide range, many of them non-nociceptive, that could possibly contribute to the development of secondary hyperalgesia (Cervero & Laird, 1996*b*).

The pathways mediating the PAD of the cutaneous afferents projecting to deeper layers in the dorsal horn would instead be subjected to an inhibitory control of their synaptic effectiveness that would be reduced following the acute section of the cutaneous nerves. It seems reasonable to assume that an increased tonic presynaptic inhibition could prevent undesired interference from cutaneous afferents in the programming of reflex movements, as seems to be the case in monkeys during performance of specific tasks (Seki *et al.* 2003, 2009). It may also lead to an 'anti-allodynic' action by reducing the activation of wide-dynamic-range interneurones by low-threshold afferents (Traub, 1997).

An alternative explanation for the finding that the changes in the intraspinal threshold of the mSU terminals produced by a second nerve lesion resembled those produced by the first nerve lesion could be that the 'state' of the neuronal connectivity ensued by the first nerve lesion determined the response of the PAD-mediating pathways to a subsequent nerve lesion. This is in line with recent evidence (Lam *et al.* 2008; Vernon *et al.* 2009) showing that a previous skin incision influences the nociceptive responses produced by stimulation of the trigeminal nerve (see also Li *et al.* 2000) and with the observations on pre-emptive analgesia, where a local anaesthetic applied prior to the skin injury decreases the nociceptive response (Dahl & Møiniche, 2004). At this stage, we cannot disregard this possibility.

In this context, it seems important to compare the effects of local anaesthesia of the SP or Saph nerves with the effects of acute nerve section on the tonic and phasic PAD of the mSU afferents as well as on their synaptic efficacy, because these two procedures are not necessarily equivalent. The effects of local anaesthesia can be attributed to the suppression of ongoing sensory information of peripheral origin, while the acute nerve section, in addition to the suppression of sensory information, generates a barrage of C fibre activity, which leads to central sensitization and enhancement of responses produced by stimulation of low-threshold afferents (Sandkühler, 2009).

It will also be important to have a more precise appraisal of the sites of afferent excitability testing within the dorsal horn. One possible approach would be to examine the differential modulation of PAD produced by acute section or local anaesthesia of cutaneous nerves simultaneously on the intraspinal threshold of pairs of collaterals belonging to the same mSU fibre ending in different regions of the dorsal horn, using the approach developed for group I muscle (Quevedo *et al.* 1997; Lomelí *et al.* 1998) and articular afferents (Rudomin & Lomelí, 2007).

Concluding remarks

The present set of observations complements previous studies on potentiation of orthodromic responses of cutaneous nerves caused by anaesthesia of the whole sciatic nerve (Biella & Sotgiu, 1995; Sotgiu *et al.* 1996). They show that the acute section of the Saph or the SP nerves also produces a long-lasting potentiation of the intraspinal responses of the mSU nerve. These nerve lesions change the level of tonic PAD of the mSU intraspinal terminals, a finding which suggests that the long-lasting potentiation of the orthodromic mSU responses produced by nerve section also has a presynaptic component. The effects of acute nerve lesions on the tonic PAD of the mSU nerve terminals appear to be related to the location of the tested afferents within the dorsal horn. A differential presynaptic control of the synaptic effectiveness of the low-threshold cutaneous afferents according to their sites of termination within the dorsal horn is envisaged as a mechanism

that allows selective processing of sensory information either during nociceptive stimulation or during motor performance.

References

Bernhard CC (1953). The spinal cord potentials in leads from the cord dorsum in relation to the peripheral source of afferent stimulation. *Acta Physiol Scand* **29**, 1–29.

Biella G & Sotgiu ML (1995). Evidence that inhibitory mechanisms mask inappropriate somatotopic connections in the spinal cord of normal rat. *J Neurophysiol* **74**, 495–505.

Carstens E, Klumpp D, Randić M & Zimmermann M (1981). Effect of iontophoretically applied 5-hydroxytryptamine on the excitability of single primary afferent C- and A-fibers in the cat spinal cord. *Brain Res* **220**, 151–158.

Cervero F & Laird JM (1996*a*). Mechanisms of allodynia: interactions between sensitive mechanoreceptors and nociceptors. *NeuroReport* **7**, 526–528.

Cervero F & Laird JM (1996*b*). Mechanisms of touch-evoked pain (allodynia): a new model. *Pain* **68**, 13–23.

Cervero F, Laird JM & García-Nicas E (2003). Secondary hyperalgesia and presynaptic inhibition: an update. *Eur J Pain* **7**, 345–351.

Chávez D, Rodríguez E, Jiménez I & Rudomin P (2012). Changes in correlation between spontaneous activity of dorsal horn neurones lead to differential recruitment of inhibitory pathways in the cat spinal cord. *J Physiol* **590**, 1563–1584.

Dahl JB & Møiniche S (2004). Pre-emptive analgesia. *Br Med Bull* **71**, 13–27.

Devor M (1983). Plasticity of spinal cord somatotopy in adult mammals: involvement of relatively ineffective synapses. *Birth Defects Orig Artic Ser* **19**, 287–314.

Devor M & Wall PD (1978). Reorganization of spinal cord sensory map after peripheral nerve injury. *Nature* **276**, 75–76.

Devor M & Wall PD (1981*a*). Effects of peripheral nerve injury on receptive fields of cells in the cat spinal cord. *J Comp Neurol* **199**, 277–291.

Devor M & Wall PD (1981*b*). Plasticity in the spinal cord sensory map following peripheral nerve injury in rats. *J Neurosci* **1**, 679–684.

Drummond GB (2009). Reporting ethical matters in *The Journal of Physiology*: standards and advice. *J Physiol* **587**, 713–719.

Eccles JC, Magni F & Willis WD (1962). Depolarization of central terminals of group I afferent fibres from muscle. *J Physiol* **160**, 62–93.

Eccles JC, Schmidt RF & Willis WD (1963). Depolarization of the central terminals of cutaneous afferent fibers. *J Neurophysiol* **26**, 646–661.

García CA & Rudomin P (2006). Changes in primary afferent depolarization during the unmasking of sural responses following acute section of cutaneous nerves in the anesthetized cat. *Abs Soc Neurosc* **146.10.**

Jankowska E, McCrea D, Rudomín P & Sykova E (1981). Observations on neuronal pathways subserving primary afferent depolarization. *J Neurophysiol* **46**, 506–516.

Jankowska E, Slawinska U & Hammar I (2002). Differential presynaptic inhibition of actions of group II afferents in di- and polysynaptic pathways to feline motoneurones. *J Physiol* **542**, 287–299.

Jeftinija S, Semba K & Randić M (1981). Norepinephrine reduces excitability of single cutaneous primary afferent C-fibers in the cat spinal cord. *Brain Res* **219**, 456–463.

Lam DK, Sessle BJ & Hu JW (2008). Surgical incision can alter capsaicin-induced central sensitization in rat brainstem nociceptive neurons. *Neuroscience* **156**, 737–747.

Li Y, Dorsi MJ, Meyer RA & Belzberg AJ (2000). Mechanical hyperalgesia after an L5 spinal nerve lesion in the rat is not dependent on input from injured nerve fibers. *Pain* **85**, 493–502.

Loeza Alcocer JE, Canto-Bustos J, Aguilar J, González Ramírez R, Felix R & Delgado-Lezama R (2012). α5-$GABA_A$ receptors modulate primary afferent excitability in the turtle spinal cord. *Abs Soc Neurosc.* **138.05.**

Lomelí J, Quevedo J, Linares P & Rudomin P (1998). Local control of information flow in segmental and ascending collaterals of single afferents. *Nature* **395**, 600–604.

Lundberg A, Malmgren K & Schomburg E (1987). Reflex pathways from group II muscle afferents. 3. Secondary spindle afferents and the FRA: a new hypothesis. *Exp Brain Res* **65**, 294–306.

Markus H & Pomeranz B (1987). Saphenous has weak ineffective synapses in sciatic territory of rat spinal cord: electrical stimulation of the saphenous or application of drugs reveal these somatotopically inappropriate synapses. *Brain Res* **416**, 315–321.

National Research Council (2010). *Guide for the Care and Use of Laboratory Animals*, 8th edn. The National Academic Press, Washington, DC.

Neugebauer V & Schaible HG (1990). Evidence for a central component in the sensitization of spinal neurons with joint input during development of acute arthritis in cat's knee. *J Neurophysiol* **64**, 299–311.

Quevedo J, Eguibar JR, Lomelí J & Rudomin P (1997). Patterns of connectivity of spinal interneurons with single muscle afferents. *Exp Brain Res* **115**, 387–402.

Randić M, Carstens E, Zimmermann M & Klumpp D (1982). Dual effects of substance P on the excitability of single cutaneous primary afferent C- and A-fibers in the cat spinal cord. *Brain Res* **233**, 389–393.

Riddell JS, Jankowska E & Huber J (1995). Organization of neuronal systems mediating presynaptic inhibition of group II muscle afferents in the cat. *J Physiol* **483**, 443–460.

Rudomin P (2009). In search of lost presynaptic inhibition. *Exp Brain Res* **196**, 139–151.

Rudomin P & Dutton H (1969). Effects of muscle and cutaneous afferent nerve volleys on excitability fluctuations of Ia terminals. *J Neurophysiol* **32**, 158–169.

Rudomin P, Engberg I, Jankowska E & Jiménez I (1980). Evidence of two different mechanisms involved in the generation of presynaptic depolarization of afferent and rubrospinal fibers in the cat spinal cord. *Brain Res* **189**, 256–261.

Rudomin P, Engberg I & Jiménez I (1981). Mechanisms involved in presynaptic depolarization of group I and rubrospinal fibers in cat spinal cord. *J Neurophysiol* **46**, 532–548.

Rudomin P & Lomelí J (2007). Patterns of primary afferent depolarization of segmental and ascending intraspinal collaterals of single joint afferents in the cat. *Exp Brain Res* **176**, 119–131.

Rudomin P, Lomelí J & Quevedo J (2004*a*). Differential modulation of primary afferent depolarization of segmental and ascending intraspinal collaterals of single muscle afferents in the cat spinal cord. *Exp Brain Res* **156**, 377–391.

Rudomin P, Lomelí J & Quevedo J (2004*b*). Tonic differential supraspinal modulation of PAD and PAH of segmental and ascending intraspinal collaterals of single group I muscle afferents in the cat spinal cord. *Exp Brain Res* **159**, 239–250.

Rudomin P, Núñez R & Madrid J (1975). Modulation of synaptic effectiveness of Ia and descending fibers in cat spinal cord. *J Neurophysiol* **38**, 1181–1195.

Sandkühler J (2009). Models and mechanisms of hyperalgesia and allodynia. *Physiol Rev* **89**, 707–758.

Sandkühler J & Gruber-Schoffnegger D (2012). Hyperalgesia by synaptic long-term potentiation (LTP): an update. *Curr Opin Pharmacol* **12**, 18–27.

Schmidt RF (1971). Presynaptic inhbibition in the vertebrate central nervous system. *Ergeb Physiol* **63**, 20–101.

Seki K, Perlmutter SI & Fetz EE (2003). Sensory input to primate spinal cord is presynaptically inhibited during voluntary movement. *Nat Neurosci* **6**, 1309–1316.

Seki K, Perlmutter SI & Fetz EE (2009). Task-dependent modulation of primary afferent depolarization in cervical spinal cord of monkeys performing an instructed delay task. *J Neurophysiol* **102**, 85–99.

Sotgiu ML, Biella G & Lacerenza M (1996). Injured nerve block alters adjacent nerves spinal interaction in neuropathic rats. *NeuroReport* **7**, 1385–1388.

Traub RJ (1997). Spinal modulation of the induction of central sensitization. *Brain Res* **778**, 34–42.

Vanegas H & Schaible HG (2004). Descending control of persistent pain: inhibitory or facilitatory? *Brain Res Rev* **46**, 295–309.

Vernon H, Sun K, Zhang Y, Yu XM & Sessle BJ (2009). Central sensitization induced in trigeminal and upper cervical dorsal horn neurons by noxious stimulation of deep cervical paraspinal tissues in rats with minimal surgical trauma. *J Manipulative Physiol Ther* **32**, 506–514.

Wall PD (1958). Excitability changes in afferent fibre terminations and their relation to slow potentials. *J Physiol* **142**, 1–21.

Wall PD & Devor M (1981). The effect of peripheral nerve injury on dorsal root potentials and on transmission of afferent signals into the spinal cord. *Brain Res* **209**, 95–111.

Wall PD & Devor M (1984). The effects of peripheral nerve injury on dorsal root potentials. *Brain Res* **209**, 95–111.

Wall PD & Werman R (1976). The physiology and anatomy of long ranging afferent fibres within the spinal cord. *J Physiol* **255**, 321–334.

Winter J, Bevan S & Campbell EA (1995). Capsaicin and pain mechanisms. *Br J Anaesth* **75**, 157–168.

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Conception and design of experiments: P.R. and I.J.; Collection, analysis and interpretation of data D.Ch., I.J. and P.R. Drafting of the article and reviewing it critically for important intellectual content: P.R. and I.J. All of the authors have contributed to this study. Experiments were performed at the Department of Physiology, Biophysics and Neurosciences, Center of Research and Advanced Studies of the Instituto Politécnico Nacional, México. All authors approved the final version of the manuscript for publication.

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