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Heterogeneity of contraction-induced effects in neurons of the cat dorsal spinocerebellar tract

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1. Clarke's column neurons of the dorsal spinocerebellar tract (DSCT) were recorded intracellularly in anaesthetized cats during weak sustained contractions of triceps surae (TS) produced by direct electrical stimulation of the muscle.

2. Of 145 DSCT neurons, 77 (53%) were contraction sensitive suggesting that information about weak contraction of a limited number of muscles is widely distributed among DSCT neurons. Four types of effects were observed in individual neurons during TS contractions.

3. In the first group of 11 DSCT neurons (14% of the contraction-sensitive cells), the effect was excitation persisting throughout the duration of contractions. These responses were ascribed to actions of afferents from contraction-activated tendon organs.

4. In a second group of 15 neurons (20% of the contraction-sensitive cells), quickly declining excitatory potentials were recorded during sustained TS contractions. By analogy with previous observations of contraction-induced effects in motoneurons, the decline of excitation might be explained by contraction-induced presynaptic inhibition of group I afferents in Clarke's column.

5. Declining inhibitions, resembling those previously observed in homonymous and synergic motoneurons, were recorded in 49% of contraction-sensitive DSCT neurons. This appears in keeping with the fact that interneurons mediating Ib inhibition to motoneurons project axon collaterals to DSCT neurons. Presynaptic inhibition of Ib fibres might therefore cause parallel reductions of inhibitory potentials in motoneurons and in DSCT neurons.

6. In a final group of 13 neurons, mixed excitatory and inhibitory effects were observed during TS contractions. Such DSCT neurons might monitor the excitability of Ib interneurons by integration of information about input to and output from these neurons.

7. The non-uniform patterns of DSCT responses to TS contractions suggest complex processing of information on ankle extensor activity in cerebellum. Phasic signalling of contraction onset is observed in many DSCT neurons while others carry messages about duration and strength of contraction.

Inhibitory potentials appearing in homonymous and synergic motoneurons at the onset of sustained contractions of gastrocnemius medialis muscle quickly subside and may disappear before the end of contraction (Zytnicki, Lafleur, Horcholle-Bossavit, Lamy & Jami, 1990). *A priori*, such contraction-induced inhibition could be ascribed to Ib inputs because tendon organs are activated by contraction and Ib afferents produce di- or trisynaptic inhibition in homonymous and synergic motoneurons (see Jami, 1992). In support of this assumption, repetitive electrical stimulation of Ib afferents in the muscle nerve did elicit declining inhibitions resembling those induced by contraction in motoneurons, and this suggested further that the decline was due to a central mechanism (Zytnicki *et al.* 1990). Intra-axonal recordings of primary afferent depolarizations in Ib afferent fibres from the contracting muscle later pointed to a contribution of presynaptic inhibition of Ib afferents to this mechanism (Lafleur, Zytnicki, Horcholle-Bossavit & Jami, 1992).

The present study was designed to examine the effects of triceps surae (TS) contractions on other targets of muscle afferents, the neurons of origin of the dorsal spinocerebellar tract (DSCT) in Clarke's column. This uncrossed tract conveys muscle proprioceptive information not only to the cerebellum, but also to the sensorimotor cortex via nucleus Z and thalamus (Landgren & Silfvenius, 1971; McIntyre, Proske & Rawson, 1985; Low, Mantle St John & Tracey, 1986; see also Mackel & Miyashita, 1993). The

neurons of the DSCT proprioceptive component are monosynaptically excited from either Ib afferents or Ia plus group II muscle afferents, but lack excitatory connection with cutaneous and high threshold muscle afferents (Lundberg & Oscarsson, 1960; see also Bloedel & Courville, 1981). In addition, DSCT cells are disynaptically inhibited by group I fibres via axon collaterals of interneurons mediating Ib inhibition of motoneurons (Hongo, Jankowska, Ohno, Sasaki, Yamashita & Yoshida, 1983*b*). The variety of muscle afferent connections with DSCT neurons suggested three questions. (i) What is the net effect on DSCT neurons of afferent inputs generated by muscle contraction? (ii) Do all the DSCT neurons receive uniform information during contraction? (iii) Is the information to DSCT neurons filtered out as was found to be the case in motoneurons?

Available data suggest that DSCT neurons receive non-uniform information during contraction (Lundberg & Oscarsson, 1956; Osborn & Poppele, 1983, 1989). Intra-axonal recordings of DSCT cell discharges in Flechsig's tract have shown that, in some cells, quadriceps contractions elicited an acceleration of discharge ascribed to the action of Ib afferents, whereas in other cells, the same contraction produced a pause in the discharge. It was not clear, however, whether this pause was due to a disfacilitation of Ia and group II excitation resulting from spindle unloading during contraction or to an inhibitory action of Ib afferents (Lundberg & Oscarsson, 1956). Non-uniform effects of muscle contractions were also suggested by cross-correlograms of DSCT unit activities with ventral root stimuli evoking TS contractions. A reduction in the probability of DSCT unit discharges, occasionally preceded or followed by a transient increase, was the most common response observed during contraction (Osborn & Poppele, 1983, 1989). However, as the postsynaptic events elicited in DSCT neurons by contraction-induced afferent inputs were not directly observed by intracellular recording in these studies, distinction between disfacilitation and inhibition was again impossible.

The present study reinvestigated the problem of contraction signalling in DSCT neurons by intracellular recording of the membrane potential variations during subtotal TS contractions: contraction-induced effects on DSCT neurons were analysed in terms of excitatory and inhibitory postsynaptic potentials. The afferent input arising during weak contractions was found to affect more than half of DSCT neurons, but in most cases, contraction-induced information to DSCT cells, signalled by excitatory and/or inhibitory potentials, was filtered out. However, non-filtered contraction-induced excitatory inputs were observed in a small but significant fraction of DSCT neurons.

A preliminary report of this work has been published in abstract form (Zytnicki, Lafleur & Kouchtir, 1991).

METHODS

Experiments were carried out on nineteen adult cats (2·5–3·5 kg) anaesthetized with an initial intraperitoneal dose of 45 mg kg^{-1} pentobarbitone sodium (Sagatal; May & Baker). Body temperature was maintained at 38 °C; heart rate and blood pressure were monitored. A permanent intravenous infusion (4–12 ml h^{-1}) of a solution of glucose (5%) containing $NaHCO_3$ (1%) and Plasmagel (14%; Roger Bellon, Paris, France) was used to maintain blood pressure above 80 mmHg (Edgley & Jankowska, 1987). A catheter allowed evacuation of urine from the bladder. Animals were paralysed (Flaxedil, 8 mg kg^{-1} h^{-1}; Specia, Paris, France) and artificially ventilated (end-tidal CO_2 maintained around 4%). Additional intravenous doses of pentobarbitone sodium (4 mg kg^{-1}) were delivered whenever necessary in order to maintain a deep level of anaesthesia as assessed by: (i) miotic pupils, (ii) regularity of heart rate (110–150 beats min^{-1}) and (iii) stability of blood pressure (90–130 mmHg). In addition, it was systematically checked that electrical stimulation of peripheral nerves at strengths eliciting potentially noxious stimuli did not modify blood pressure and heart rate.

The gastrocnemius medialis (GM) muscle (in 6 experiments), or the whole triceps surae (TS, in 13 other experiments) were dissected out without disturbing their blood supply; their tendons were cut and tied to a force transducer (Celaster; compliance, 60 μm $(5\ kgf)^{-1}$, i.e. the full range of the transducer) connected to an amplifier. The hindlimb was immobilized, and the muscle length was set close to its physiological maximum. Except for GM or TS muscles, all the rest of the hindlimb was denervated. The nerves to quadriceps, posterior biceps-semitendinosus, anterior biceps-semimembranosus, pretibial flexors (i.e. tibialis anterior and extensor digitorum longus), plantaris and the sural nerve were dissected out, cut, and their proximal ends were mounted on a pair of electrodes for bipolar stimulation. GM or TS nerves were dissected in continuity with their muscles and placed on a single electrode for monopolar stimulation. The dissected muscles and nerves were covered with a pool of mineral oil kept at 38 °C.

The main difficulty of the present experiments was to design a preparation in which stable intracellular recordings of DSCT cells in the Clarke's column could be obtained while gastrocnemius medialis or triceps muscles were contracting. Stable intracellular recordings were achieved by: (i) paralysis of the preparation associated with a bilateral pneumothorax to eliminate passive ventilation movements, and (ii) preservation of an intact dura mater except for small patches allowing insertion of the microelectrode into the spinal cord. Muscle contractions had to be produced by direct electrical stimulation of the muscle(s) through one or two pairs of intramuscular steel hook electrodes. In all experiments one pair was inserted in the GM muscle and an additional pair was inserted in the gastrocnemius lateralis muscle in the thirteen experiments where contractions of the whole triceps surae were elicited. Great care was taken to avoid direct electrical stimulation of afferent fibres in the muscle, first, by inserting the electrodes in the most distal part of the muscle belly that is at a distance from the point of nerve entry in the muscle, and second, by using weak electrical stimulations which produced contractions developing less than 5% of the maximal muscle force. Verification that direct stimulation did not excite afferent fibres was carried out by averaging large numbers of records from the spinal cord surface near the entry of L7 dorsal root. Action potentials at short constant latencies had to be absent from such records during muscle stimulation.

This method was sensitive enough to detect volleys due to direct excitation of a few afferent fibres. As illustrated in Fig. 1, averaging of five successive sweeps allowed the detection of a small afferent volley (indicated by the filled arrow) induced by direct excitation and occurring with a latency of 2·3 ms after the stimulation. The amplitude of this volley was less than 5% of the maximal group I volley suggesting that only a few fibres were excited. In Fig. 1 the number of averaged traces was only five and the direct volley was quite easily detected; an improved signal-to-noise ratio was obtained by averaging larger numbers of records and this allowed detection of very small afferent volleys. If, in spite of these precautions, some afferent fibres were still directly excited, they could be expected to induce responses only in a limited population of DSCT cells because individual group I afferents are known not to project extensively to the DSCT cell population (see Oscarsson, 1973). Moreover, effects elicited by direct afferent stimulation onto DSCT neurons were easily recognized in intracellular records as exemplified in Fig. 1. A sharp excitatory potential occurred 1·2 ms after the direct afferent volley entered the spinal cord and before the onset of contraction (vertical dashed line). This early excitatory potential was therefore caused by afferents engaged by direct muscle stimulation. The short central latency of this potential suggested a monosynaptic action by group I afferents. On the other hand, a second wave of excitatory potentials (indicated by the open arrow) appeared 16 ms after the onset of contraction. This latency was too long for excitatory potentials due to the effect of direct excitation of muscle afferents: the new excitation was very probably induced by the contraction itself. The example in Fig. 1 shows that effects of direct afferent stimulation were readily recognized, and for this reason, we are confident that effects appearing at relatively long latencies after the onset of contraction indeed represent contraction-induced effects. All cases where direct activation was seen were excluded from the present results.

Frequencies of muscle stimulation were kept between 10 and 40 s^{-1}, that is, within the rates of activation of hindlimb-muscle motor units observed in freely moving cats (Hoffer, Sugano, Loeb, Marks, O'Donovan & Pratt, 1987). Stimulation sequences lasted 0·4 s and were separated by intervals of 1–5 s to avoid the too rapid development of fatigue. The experiment was discontinued if contractile force fell below 30% of its initial level.

The vertebral column was held immobile with clamps on Th8–Th9 spinous processes and L1 vertebral body, and with pins applied to the rostral parts of coxal bones. The L3 and L4 spinal segments were exposed by a laminectomy and small patches were made through the dura and pia mater covering the dorsal column at a distance of 0·5–2 mm from mid-line. Conventional glass micropipettes filled either with a mixture of 2 M potassium acetate and 0·6 M potassium chloride, or 3 M potassium chloride (2–5 MΩ) were driven through the patches in a sagittal plane. Micropipettes filled with potassium chloride were used to test whether hyperpolarizations recorded in neurons could be reversed by intracellular injection of chloride ions (see Results). Two additional

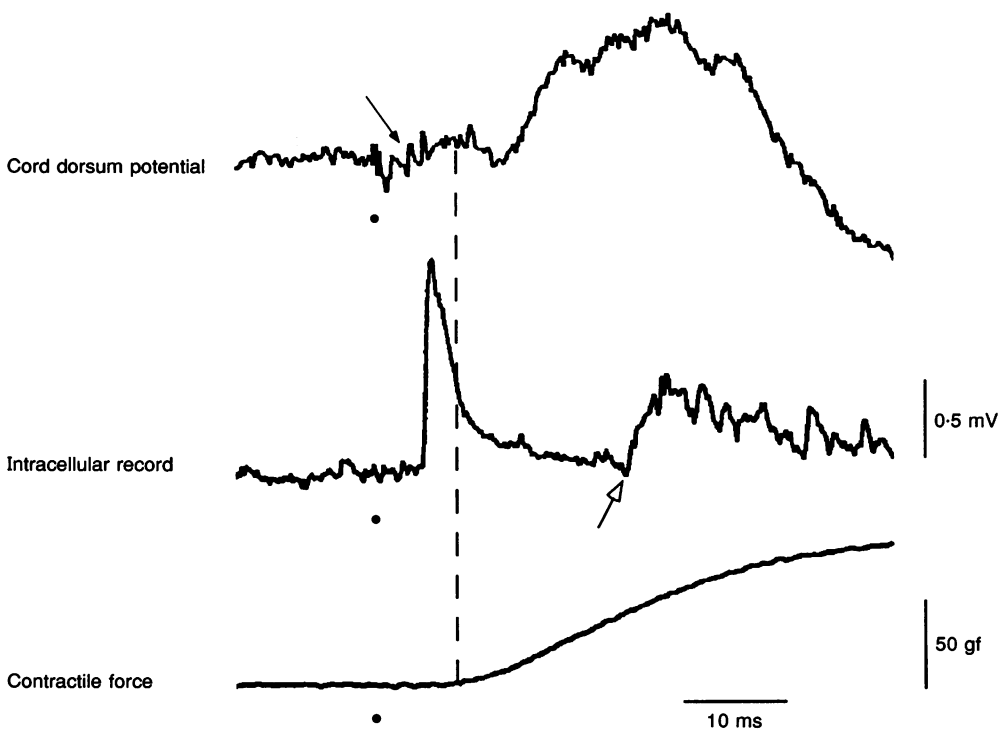

Figure 1. Difference between effects elicited in a DSCT neuron by direct intramuscular excitation of afferent fibres and by contraction

Responses to a single shock of stimulation indicated by the points under force profile, intracellular record and cord dorsum potential. All records are averages of 5 traces. Filled arrow points to the small 2-component afferent volley induced by direct muscle stimulation (the preceding event on cord dorsum potential corresponds to stimulation artifact). Open arrow points to the onset of contraction-induced excitatory potentials (see text for further details). Vertical dashed line indicates the onset of contraction.

Table 1. Types of responses among contraction-sensitive DSCT neurons

	Number of neurons	Contraction-sensitive neurons	Declining inhibitions	Declining excitations	Persistent excitations	Mixed actions
Sample A	94	46	22 (48%)	9 (20%)	7 (15%)	8 (17%)
Sample B	51	31	16 (52%)	6 (19%)	4 (13%)	5 (16%)
Total	145	77	38 (49%)	15 (20%)	11 (14%)	13 (17%)

Numbers of neurons displaying each type of response in samples in which neurons were identified with the 'simplified procedure' (A), with the procedure including antidromical activation from cerebellum (B) and in the total sample. Percentages indicate the proportion of contraction-sensitive cells displaying a given type of response.

laminectomies allowed exposure of: (i) Th10–Th11 spinal segments where two pairs of bipolar stimulation electrodes were placed on the surface of the dura mater covering ipsi- and contralateral dorsolateral funiculi, respectively; and (ii) the caudal part of the L5 vertebra where a silver ball electrode was placed on the surface of the spinal cord close to the entry of the ipsilateral L7 dorsal root in order to record afferent volleys. A bath of mineral oil kept at 38 °C covered the exposed parts of the spinal cord.

In the first series of fourteen experiments identification of group I-coupled DSCT cells in Clarke's column rested on three criteria and will be referred to as the 'simplified identification procedure': (i) antidromic activation from thoracic ipsilateral funiculus and absence of response from contralateral funiculus, (ii) monosynaptic excitatory postsynaptic potentials (EPSPs) evoked by group I inputs from at least one muscle nerve, and (iii) absence of excitatory input from cutaneous afferents. Lundberg & Oscarsson (1960) demonstrated that these criteria allow identification of the proprioceptive component of the DSCT neuron population in Clarke's column. In a further series of five experiments, identification of group I-coupled DSCT cells also included antidromic activation from cerebellum in order to check that the selection of DSCT neurons was not biased by the simplified identification procedure. For the sake of simplicity, group I-coupled DSCT cells in Clarke's column will be referred to as 'DSCT cells'.

The procedure for placement of the cerebellum-stimulating electrode was similar to that used by Edgley & Gallimore (1988). An occipital craniotomy allowed exposition of an area in the anterior lobe of the cerebellum, 1–2 mm rostral to the fissura prima and 3–5 mm lateral to the mid-line. A concentric bipolar electrode (Rhodes Medical Instruments, Woodland Hills, CA, USA) was inserted in the cerebellum via a hole made in the dura mater, pointing in the rostral direction at an angle of 30–40 deg. The electrode was driven through the cerebellum while field potentials elicited by stimulation of ipsilateral thoracic spinal cord were recorded. The final location of the electrode was at the site of maximal field potential, usually at depths of 9–12 mm. The electrode was then connected to a stimulator through an isolation unit.

Simultaneous records of membrane potential, cord dorsum potential and muscle force were fed into the channels of a Nicolet 4094A digital oscilloscope performing on-line response averaging. DC coupling was used for membrane potential and muscle force recordings through specific amplifiers with low-pass filters of 20 kHz and 2 kHz cut-off frequencies, respectively. AC coupling, through a resistance–capacitance high-pass filter with a cut-off frequency of 1·5 Hz, was used for cord dorsum potential recordings. Single sweeps or averages of three to ten responses were stored on a floppy disk (Nicolet CF-44 unit) and subsequently displayed on a HP 7550A digital plotter.

Finally, in two further experiments, triceps surae nerves were cut and mounted on a pair of electrodes for bipolar stimulation. In this case, AC coupling was used to record the responses of DSCT neurons to electrical stimulation of the TS nerve at 20 s^{-1} at strengths activating either group I afferents alone (1–1·5 times the threshold (T) or group II fibres in addition (1·5–5 T; see Fig. 5).

RESULTS

Effects of ankle extensor contractions were analysed in two samples of ninety-four neurons identified with the simplified procedure (sample A in Table 1) and fifty-one additional neurons (sample B in Table 1) whose identification was confirmed by antidromic activation from the cerebellum.

Samples A and B were considered to represent similar populations of DSCT neurons for three reasons. (i) Means and S.D.s of axonal conduction velocities were very close in the two samples: 72 ± 11 and 67 ± 10 m s^{-1}, respectively, for samples A and B. They were in a range of 42–107 m s^{-1}, which is comparable to the data of Lundberg & Oscarsson (1960). Their distribution was unimodal with a peak between 65 and 75 m s^{-1}, and 90% of axonal conduction velocities were in the 55–85 m s^{-1} range. (ii) When the effects of TS contractions of similar amplitude (100–450 gf) were tested in both samples, a similar proportion of neurons were contraction sensitive: 56% in sample A (42 out of 75 neurons; in addition 4 of 19 neurons tested with weak GM contractions were also contraction sensitive) and 61% in sample B (31 out of 51 neurons). (iii) The four main types of contraction-induced effects (see below) occurred with similar incidences in samples A and B (see Table 1).

Taken together, these data suggest that the simplified identification procedure did not bias the selection of DSCT neurons and the results obtained in both samples were therefore pooled. In the total sample of 145 DSCT neurons, 77 (53%) were sensitive to ankle extensor contractions.

Figure 2 illustrates the simplest type of responses elicited in DSCT cells by afferent inputs arising from TS during contractions. When stimulated at a rate of 10 s^{-1}, the muscle produced a series of twitches eliciting excitatory potentials (EPSPs) in the DSCT neuron (Fig. 2*A*). These twitches, developing relatively small forces, were unlikely to activate a large number of muscle afferents, but nevertheless, the amplitude of excitatory potentials reached 4·5 mV (Fig. 2*A*) which is in keeping with the large amplitudes of unitary group I EPSPs found in DSCT neurons by Kuno & Miyahara (1968). During unfused tetanic contractions developing higher force (Fig. 2*B*), EPSPs with amplitudes reaching discharge threshold occurred (arrows in Fig. 2*B*), either in isolation or in short trains. The latency of the first component of this response with respect to the onset of contraction was 12 ms. In the whole sample of contraction-sensitive neurons the corresponding latencies were in the 10–48 ms range. Such long latencies support the view that the observed responses were elicited by contraction and not by direct excitation of muscle afferents (see Methods and Fig. 1). It is difficult to know with certainty which afferents were responsible for the excitatory potentials induced during contraction in DSCT neurons, but it is very likely that Ib inputs *contributed* to the contraction-induced excitations of DSCT neurons for two reasons (see also Discussion). First, the DSCT neurons in which contractions induced excitatory potentials were also excited monosynaptically by the electrical stimulation of TS group I fibres (not illustrated, but see Fig. 5). This observation suggests that contraction-induced EPSPs could be due to effects of group I fibres known to establish monosynaptic excitatory connections with DSCT cells (see Oscarsson, 1973). Second, the contraction-induced excitations occurred most often during the rising phase of force oscillations (see Figs 2 and 3), which is in keeping with the fact that tendon organs are known to discharge on the rising phase of force oscillations during unfused tetanus (see Jami, 1992).

In eleven (14%) of the seventy-seven contraction-sensitive neurons, series of EPSPs were observed throughout the period of contraction ('Persistent excitations' in Table 1). However, this was not the case for all the EPSPs observed in DSCT neurons, and an example of excitation declining during TS contractions is shown in Fig. 3*A*. At the onset of contraction, i.e. during the first oscillation developing 80 gf, a first peak of excitation of 4·6 mV amplitude was recorded. However, the second EPSP was much smaller, with an amplitude of only 1 mV, although it appeared while force had increased to 130 gf. Subsequent EPSPs of about 1·2 mV appeared with each force oscillation. Electrical testing of connectivity showed that this neuron had monosynaptic excitatory connections with group I afferents from TS. This neuron was classified as exhibiting declining excitatory potentials throughout contractions as was also the case for all neurons in which excitatory potentials

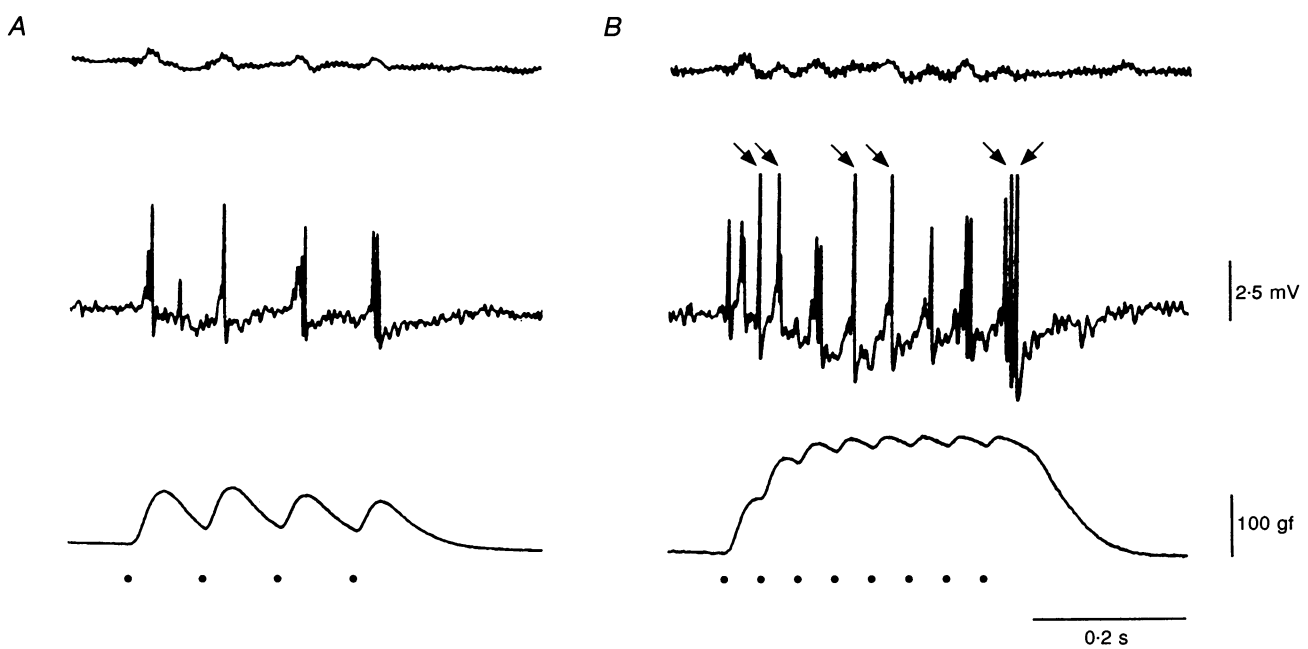

Figure 2. Excitatory potentials during TS contractions in a DSCT neuron

Top traces, cord dorsum potential; middle traces, intracellular recording of the DSCT neuron; bottom traces, contractile force. Points under force profile indicate stimulation shocks. *A*, response of the neuron to a series of twitches evoked by a stimulation rate of 10 s^{-1}. Three responses averaged. *B*, single trace response of the same neuron to unfused contractions elicited by a stimulation rate of 20 s^{-1} (arrows point to cut action potentials). The earliest EPSP, appearing before development of force, could be a spontaneous EPSP.

declined by 30–80% during contractions. A reduction of less than 30% was not taken into account, and the response was considered as persistent excitation (this occurred for a single neuron only).

In agreement with data from previous reports (see e.g. Eide, Fedina, Jansen, Lundberg & Vyklicky, 1969), fourty-nine DSCT cells, i.e. 34% of the total sample, had resting discharges. Figure 3*B* shows a neuron with a spontaneous discharge rate in the 25–60 s^{-1} range. This rate clearly increased at the onset of contraction, where two spikes occurred with an interval of 7 ms during the rising phase of the first force oscillation, followed, during the second oscillation, by a burst of eight spikes at 150–200 s^{-1}. Subsequently, the neuron recovered its resting discharge frequency with a brief pause towards the end of tetanus (arrow in Fig. 3*B*). The increase in discharge frequency at the onset of contraction indicated a temporary supplement in excitation, increasing the firing probability of the neuron for a short period. However, the excitation was not maintained since the discharge frequency very quickly subsided and it is therefore likely that this neuron received declining excitatory influences similar to those received by the neuron illustrated in Fig. 3*A*. Altogether, fifteen DSCT neurons (20% of the contraction-sensitive sample) displayed signs of declining excitation during TS contractions (see Table 1).

Figure 4 illustrates the most frequently observed effect of an unfused TS tetanus on contraction-sensitive DSCT cells, that is, a declining inhibition, similar to those previously observed in motoneurons (Zytnicki *et al.* 1990). Figure 4*A* shows a record from a DSCT neuron in which inputs generated by TS contractions first induced an hyperpolarization of 0·4 mV amplitude. This hyperpolarization declined while force was increasing and almost disappeared during the plateau; by the end of contraction the hyperpolarization retained less than 20% of its initial amplitude. In another DSCT neuron (Fig. 4*B*), which had a resting discharge frequency of 60 s^{-1}, the onset of contraction was marked by a pause lasting about 100 ms (arrow in Fig. 4*B*). Subsequently, the discharge resumed at a slower rate before the end of contraction and a transient acceleration occurred during the relaxation.

Intracellular injections of chloride ions by hyperpolarizing current carried through potassium chloride-filled microelectrodes were attempted in DSCT neurons to verify whether the observed inhibitions represented genuine inhibitory potentials. However, injections of large amounts of current in these neurons often caused oscillations of membrane potential and loss of the cell. In four instances, as exemplified in Fig. 4*C*, injection of weak currents elicited distinct reductions in the amplitude of hyperpolarizations. Declining inhibitory potentials observed before current injection (Fig. 4*Ca*) were reduced after a 2 nA current had hyperpolarized the membrane potential by 16 mV (Fig. 4*Cb*). This reduction was interpreted as an 'incomplete reversal', suggesting that inputs arising from muscle contraction elicited declining inhibitory potentials in this DSCT neuron rather than a disfacilitation of some permanent excitatory influence. Support for this interpretation was obtained by systematically testing each neuron for the pattern of effects elicited by electrical stimulation of group I muscle afferents in the prepared muscle nerves (see Methods). The DSCT

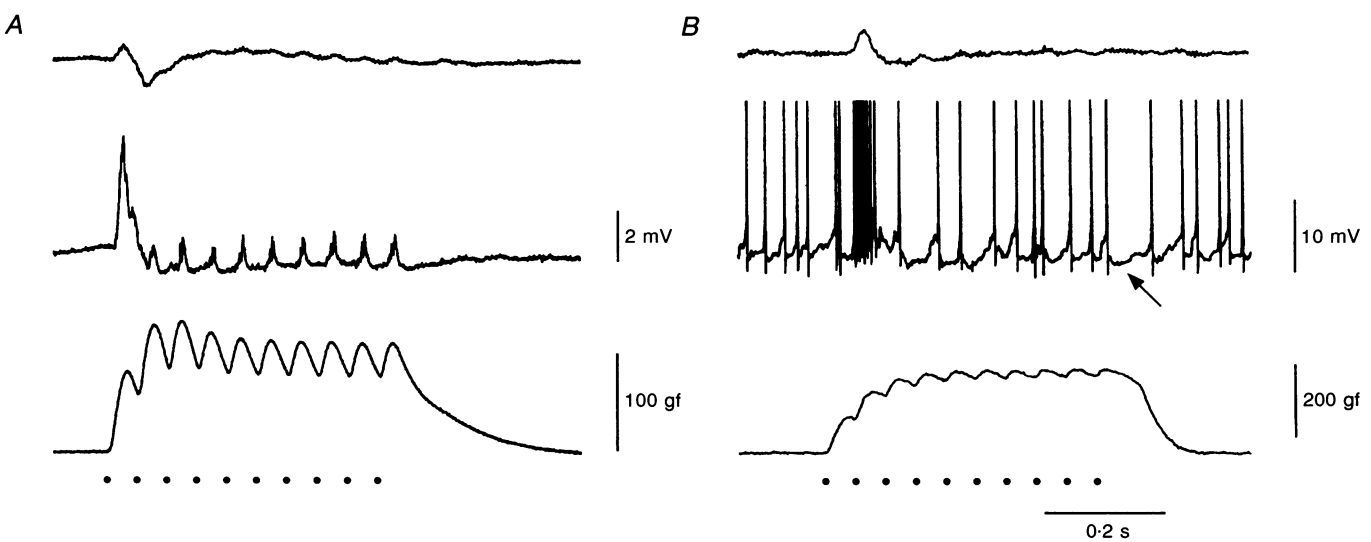

Figure 3. Declining excitatory effects recorded in two DSCT neurons (*A* and *B*) during TS contractions

Same arrangement as in Fig. 2. The neuron in *B* had a spontaneous discharge (action potentials were cut to fit within the limits of the figure); note the acceleration at the onset of contraction and the absence of hyperpolarization after the burst. The arrow points to a pause in discharge near the end of contraction. Five traces averaged in *A*, single trace in *B* (the same response pattern was recorded several times).

neurons illustrated in Fig. 4*A* and *C* were found to lack mono- or polysynaptic excitation from TS group I or group II afferents, which was not compatible with their contraction-induced hyperpolarization being due to a disfacilitation of excitation. These DSCT neurons were therefore considered to display declining inhibitory potentials during the contraction.

In only four out of thirty-eight instances, contraction-induced inhibitions occurred in DSCT cells receiving monosynaptic EPSPs from TS group I fibres. In such cases, it was difficult to decide whether the slowing of discharge was due to a decrease in Ib input or to unloading of Ia discharges by contraction. The two mechanisms, namely disfacilitation and inhibition, were not mutually exclusive.

Altogether, declining inhibitory potentials (i.e. reduction by 30–80% during contraction plateaus lasting 0·4 s) or transient reductions in discharge frequency during contractions were observed in thirty-eight DSCT neurons, that is, 49% of the seventy-seven contraction-sensitive neurons (see Table 1). The question then, was whether the decrease in inhibitory potentials had a peripheral or a central origin. Repetitive electrical stimulation of group I afferents in the TS nerve was used to test whether a constant input would elicit declining inhibitory post-synaptic potentials in DSCT neurons as it did in synergic motoneurons (see Zytnicki *et al.* 1990). As illustrated in Fig. 5*A* (left), electrical stimulation at a constant strength of 1·5*T*, recruiting only group I afferents, inhibited some DSCT neurons with a central latency of 2·2 ms, compatible with a disynaptic linkage through inhibitory interneurons located in lower lumbar segments (see Hongo, Jankowska, Ohno, Sasaki, Yamashita & Yoshida, 1983*a*). The inhibitory potentials had an initial amplitude of 1 mV, which decreased by 36% after four repetitions of the stimulation at 20 s^{-1} (arrow in Fig. 5*A*). Small random EPSPs (known to occur in DSCT cells; see Kuno & Miyahara, 1968) partly obscured the decrease, but the last IPSP in the series was clearly smaller than the first one. When the sequence of stimulation was repeated with a strength of 5*T*, recruiting group II fibres in addition to group I, the amplitude of the inhibitory potential and its

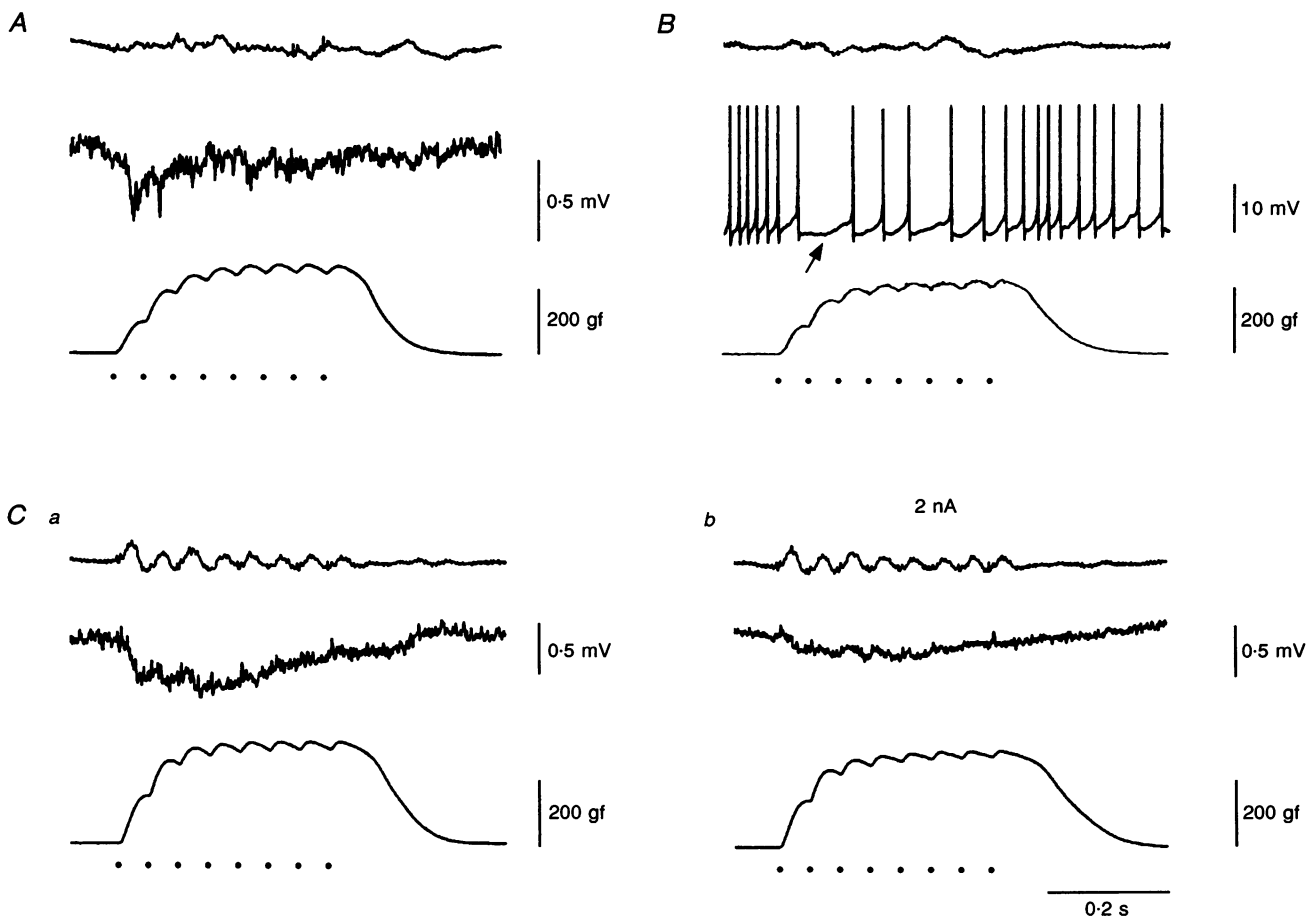

Figure 4. Declining inhibitory potentials recorded in three DSCT neurons during TS contractions

In each panel, same arrangement as in Fig. 2. The neuron in *B* had a resting discharge. Arrow points to a pause in discharge. *Ca* and *b* are responses recorded in the same neuron before and after an injection of 2 nA hyperpolarizing current. Five traces averaged in *A* and *C*, single trace in *B*.

decline were similar. This observation, repeated in five other DSCT neurons, suggests that group I afferents could be responsible for the contraction-induced inhibitory potentials in DSCT neurons and that contribution of group II afferents to inhibitory potentials was negligible. Since group I input was constant throughout the sequence of repetitive stimulation, the decline of inhibition during the sequence had a central origin. A plausible conclusion might be that the decline of contraction-induced inhibition also had a central origin. The decline of inhibitory potentials might be explained by transmission of inhibitory inputs to DSCT neurons via axon collaterals of the same Ib interneurons which mediate the autogenetic inhibition of motoneurons. Since it was demonstrated that Ib autogenetic inhibition declined during contraction because of presynaptic inhibition of Ib fibres (Zytnicki *et al.* 1990; Lafleur *et al.* 1992; Zytnicki & L'Hôte, 1993), it was not surprising also to observe declining inhibitions in DSCT cells. However, in Clarke's column, group I fibres were not all submitted to presynaptic inhibition, and Fig. 5*B* (left), shows an instance where monosynaptic excitatory potentials (central latencies, 1·1 ms) in a DSCT neuron, elicited by repetitive electrical stimulation of group I TS afferents, fluctuated in amplitude but without decline. Recruitment of group II afferents did not modify the effects (Fig. 5*B*, right). The same observation was repeated in another DSCT neuron.

Taken together, the present results indicate that responses of DSCT neurons during ankle extensor contractions are not uniform as is the case in motoneurons. Three relatively simple patterns of contraction-induced responses in DSCT neurons were observed: persistent excitations, declining excitations or declining inhibitions. However, complex responses also occurred, and in a final group of thirteen DSCT neurons, TS contractions evoked a mixture of excitatory and inhibitory potentials (see Table 1). The four patterns occurred in the same experiment: for instance, among fifteen contraction-sensitive neurons (out of 23) recorded in a single experiment, eight exhibited declining inhibitions, one declining excitations, two persistent excitations and four mixed actions. This result indicates that a single contraction simultaneously elicits the four types of responses among the DSCT neuron population.

DISCUSSION

The present work shows, first, that 77 (53%) of 145 DSCT neurons were sensitive to weak contractions of ankle extensor muscles suggesting that information about TS contraction is widely distributed across the population of DSCT cells and not restricted to a limited number of neurons (see also Osborn & Poppele, 1992). The responses of DSCT neurons were tested with forces developing less than

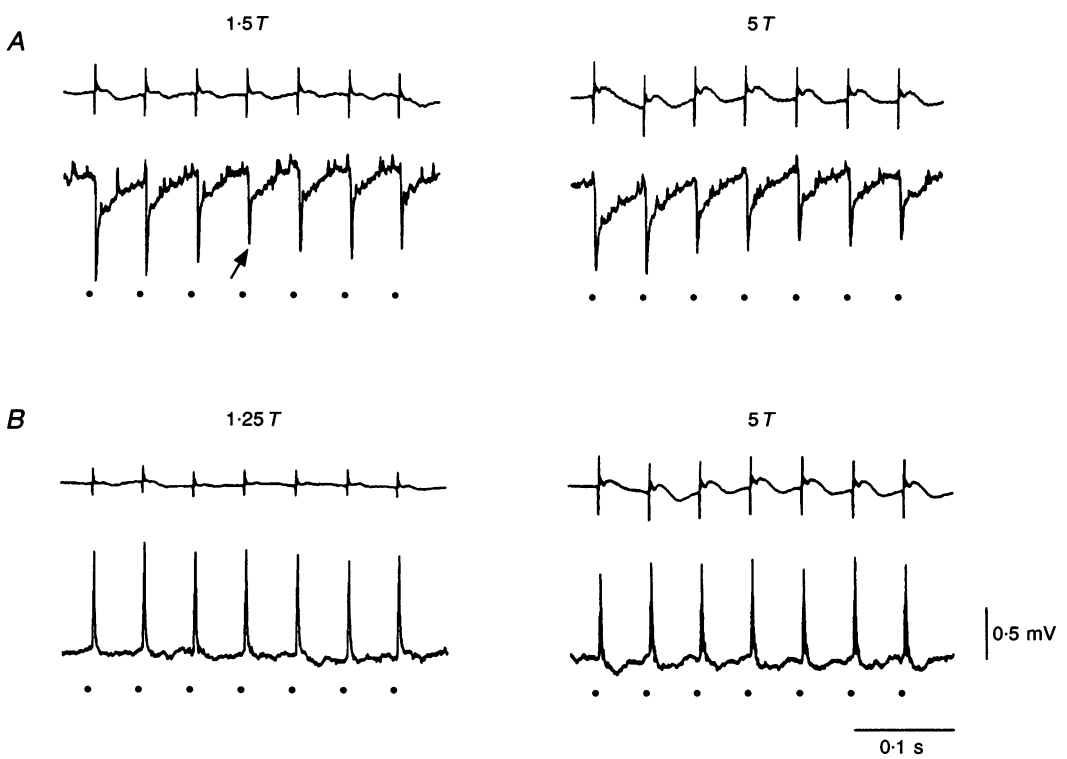

Figure 5. Responses of two DSCT neurons to electrical stimulation of TS nerves

Top traces, cord dorsum potential with superimposed afferent volleys; bottom traces, intracellular recordings. Points under the record indicate a stimulation rate of 20 s^{-1}. The amplitude of the IPSP indicated by the arrow was 36% less than the amplitude of the initial IPSP. Stimulation strengths are indicated in multiples of group I threshold. Five traces averaged in *A* and 10 in *B*.

5% of the total muscle force and which were unlikely to activate a large proportion of the muscle mechanoreceptor population. Stronger contractions, possibly producing more powerful afferent inputs, might influence higher proportions of DSCT neurons. Another factor restricting the number of responsive DSCT neurons might be the fact that the receptive field of Ib input in these neurons is most often limited to a single muscle (Eccles, Oscarsson & Willis, 1961). Afferent inputs generated by co-contractions of several muscle groups, as occurs under natural conditions, would be expected to affect substantial fractions of the DSCT neuron population. Moreover, our study was restricted to those neurons of the DSCT proprioceptive component excited by group I muscle afferents in Clarke's column, but it is likely that group I-coupled DSCT neurons located outside Clarke's column (Aoyama, Hongo & Kudo, 1973) also convey information about contraction.

Second, four types of responses were observed during contractions: (i) the most frequent responses, found in 26% of the total sample, were subsiding inhibitions similar to those observed in homonymous motoneurons (Zytnicki *et al.* 1990), (ii) in another 10% of DSCT neurons, declining excitation occurred during sustained TS contractions, (iii) excitatory potentials persistently repeated throughout contractions were found in a small but significant fraction (8%) of DSCT neurons, and finally, (iv) responses observed in 9% of the sample displayed a mixture of inhibitory and excitatory potentials.

It is known that tendon organs are specific contraction sensors and that, in addition, primary and secondary spindle endings plus non-specific mechanoreceptors innervated by group II and III fibres can also be activated during contractions (see the discussion in Lafleur, Zytnicki, Horcholle-Bossavit & Jami, 1993). However, group I-coupled DSCT neurons in Clarke's column have few connections with group II fibres and none with group III (see Oscarsson, 1973; Bloedel & Courville, 1981), and it was shown in the present study that recruitment of group II fibres did not significantly change the response, whether excitatory or inhibitory, elicited in DSCT cells by repetitive electrical stimulation of group I fibres in TS nerves (Fig. 5). The present results could be explained in the light of data about pathways from group I muscle afferents to DSCT neurons, as summarized in the diagram of Fig. 6.

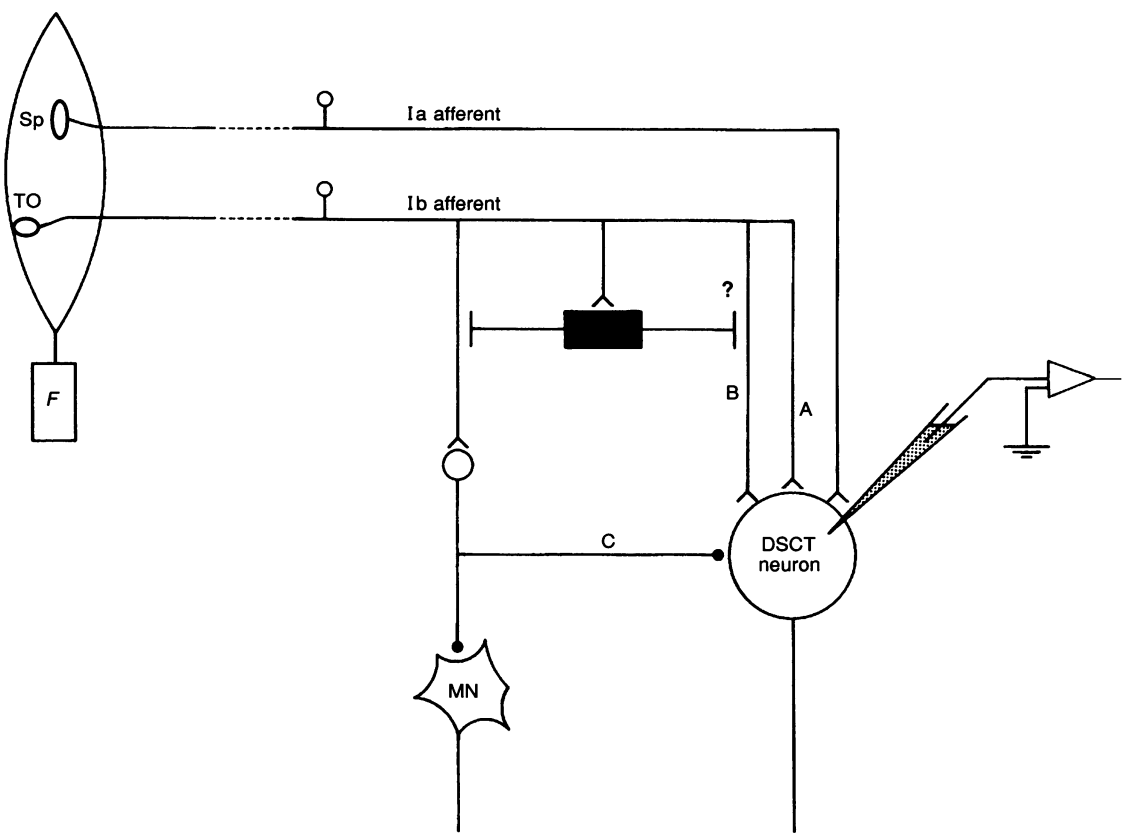

Figure 6. Schematic diagram of connections that might produce the different effects observed in DSCT neurons during TS contractions (see text)

Sp, spindle; TO, tendon organ; *F*, contractile force recorded at muscle tendon; MN, motoneuron. The filled box represents the pathways mediating presynaptic inhibition of Ib afferents, and the question mark indicates that contraction-induced presynaptic inhibition of Ib afferents in the upper lumbar segments has not been demonstrated so far.

In the first fraction of DSCT neurons, contractions were signalled by a series of EPSPs with non-declining amplitudes that could be due to Ib or/and Ia inputs. However, while it is well known that tendon organs are very efficiently activated throughout contractions (see Jami, 1992), it is not certain whether spindle primary endings are consistently activated during weak contractions induced by direct muscle stimulations that are unlikely to elicit intrafusal contractions. In addition, in our experiments, the contraction-induced EPSPs appeared on the rising phases of twitches or force oscillations during unfused tetanus rather than on the relaxation phases (Figs 2 and 3) suggesting Ib rather than Ia effects. Rapid muscle stretches of TS muscles would allow the demonstration of possible excitatory connections from Ia fibres to contraction-sensitive DSCT, but this test was not attempted because muscle stretch might have endangered the stability of intracellular recording. Moreover, classical experiments using selective adequate stimulation of tendon organs or/and spindles suggested that excitatory Ib and Ia inputs do not converge onto the same DSCT neurons (Lundberg & Winsbury, 1960). For these reasons, we favour the assumption that contraction-induced effects on DSCT cells were mainly elicited by Ib inputs. If this is the case, excitatory connections from Ib fibres (pathway A in Fig. 6) would be responsible for excitations persisting throughout contractions as exemplified in Fig. 2. A fraction of DSCT neurons might therefore forward to the cerebellum the information encoded in the ensemble discharge from tendon organs, concerning not only duration of contractions but also variations in contractile force (see Jami, 1992).

The rapid decrease of EPSPs observed during contractions in another fraction of DSCT neurons might be due to either peripheral or central mechanisms. However, the reduction of EPSPs is unlikely to reflect a reduction in tendon organ discharge rates because, in a given experiment, the same contraction-induced inputs could elicit persistent excitation in some DSCT neurons and declining excitation in others. The possibility of a reduction of efficacy in the transmission of group I impulses to DSCT cells had therefore to be considered. A possible explanation for this reduction might be that contractions induce presynaptic inhibition of Ib fibres (pathway B in Fig. 6). Two sets of data would support this assumption. First, in similar experimental conditions, contraction-induced primary afferent depolarizations were demonstrated to occur in L7–S1 intraspinal portions of Ib fibres (Lafleur *et al.* 1992) and second, impulses from Ib and high threshold afferents are known to elicit presynaptic inhibition of both Ia and Ib terminals in Clarke's column (Eccles, Schmidt & Willis, 1963; Jankowska, Jukes & Lund, 1965; see also Walmsley, Wieniawa-Narkiewicz & Nicol, 1987). Our results are compatible with the assumption that some but not all of the collaterals of Ib fibres terminating in Clarke's column could be affected by contraction-induced presynaptic inhibition. Observations indicating a non-uniform distribution of presynaptic inhibition among the terminal branches of individual group I afferents were recently reported by Quevedo, Eguibar, Jimenez & Rudomin (1993).

In a third fraction of DSCT neurons, impulses arising from muscle contractions caused inhibitory potentials, which declined in a manner resembling that observed in motoneurons (Zytnicki *et al.* 1990). Repetitive electrical stimulation of group I fibres also elicited declining disynaptic inhibitory potentials in DSCT cells (Fig. 5). Differences between contraction-induced declining inhibitions and inhibitions induced by electrical stimulation of nerves may be explained by recruitment of fewer Ib fibres during weak contractions than during electrical stimulations and/or by the asynchrony of tendon organ discharges during contractions contrasting with the synchronous excitation of Ib fibres by electrical stimulation. Whatever may be the case, it is known that Ib interneurons mediating the autogenetic inhibition of motoneurons can also inhibit DSCT neurons via axon collaterals (Hongo *et al.* 1983*b*). Contraction-induced inhibition of DSCT cells through pathway C of Fig. 6 might be expected to decline, as was the case in motoneurons, because transmission from Ib terminals is inhibited presynaptically during contraction (Lafleur *et al.* 1992; Zytnicki & L'Hôte, 1993). Other categories of lower and upper lumbar interneurons might also contribute to contraction-induced inhibition of DSCT neurons (see Hongo *et al.* 1983*a*).

A 'copy' of the contraction-induced autogenetic inhibition of motoneurons appears to be forwarded to some DSCT neurons via Ib interneuron collaterals. The resulting slowdown of DSCT cell discharge rates (Fig. 4*B*) might convey to the cerebellum negative information signalling reduction in excitability of homonymous and synergic motoneurons during contractions. Even though negative, such information should be relevant for cerebellar operations of motor control. In non-discharging DSCT neurons, contraction-induced IPSPs (Fig. 4*A* and *C*) reduced the excitability, thereby lowering the efficiency of any concurrent excitatory input.

Finally, some DSCT neurons were both excited and inhibited during contractions. If transmission from caudal Ib terminal branches located in L7–S1 segments and from rostral branches in the upper lumbar segments is presynaptically inhibited, some DSCT neurons might receive a 'copy' of the filtered Ib input to interneurons mediating autogenetic inhibition of motoneurons. DSCT neurons in which contraction elicited a mixture of excitatory and inhibitory potentials might thus be able to integrate information about inputs to (pathway B) and outputs from (pathway C) Ib interneurons, allowing computation of the excitability level of these inhibitory

interneurons. A similar hypothesis was proposed in the case of the ventral spinocerebellar tract which is thought to monitor transmission in inhibitory pathways to motoneurons (Lundberg, 1971). The Ib interneurons are known to play a crucial role in the spinal pathways of motor control and it was suggested that they could be involved in segmental processes of hindlimb muscle co-ordination (see Harrison, 1985). It is therefore important for the central nervous system to monitor their excitability.

The present study demonstrates that different types of contraction-induced messages are encoded by subpopulations of DSCT neurons, so that the cerebellum receives elaborate information about muscle contractions. This is in keeping with extracellular recordings of Purkinje cells in decerebrate cats, showing that mossy fibre discharges during TS contractions may evoke either excitation or inhibition in these cells (Iosif, Pompeiano, Strata & Thoden, 1972). It remains to be understood how the cerebellum uses information about muscle contraction supplied by the DSCT during execution of movements or muscle tone regulation. In addition, group I inputs from hindlimb muscles might participate in kinaesthesia because they were demonstrated to reach the cerebral cortex via DSCT axon collaterals to nucleus Z (Landgren & Silfvenius, 1971; McIntyre *et al.* 1985). Similarly, it was recently shown that midlumbar spinocerebellar neurons specialized in the processing of muscle group II information also project axon collaterals to this nucleus (Asif & Edgley, 1992). In this context it would be important to know whether all four types of contraction-sensitive DSCT neurons project axon collaterals towards the nucleus Z area. Interestingly, the observations of McIntyre *et al.* (1985) suggest that it might be the case for DSCT neurons with persistent excitation. These authors reported a 'Golgi tendon organ-like' pattern of discharge during TS contractions in some nucleus Z neurons.

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