

## CENTRAL CONNECTIONS OF SENSORY NEURONES FROM A HAIR PLATE PROPRIOCEPTOR IN THE THORACO-COXAL JOINT OF THE LOCUST

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### Summary

The hair plate proprioceptors at the thoraco-coxal joint of insect limbs provide information about the movements of the most basal joint of the legs. The ventral coxal hair plate of a middle leg consists of group of 10–15 long hairs (70  $\mu\text{m}$ ) and 20–30 short hairs (30  $\mu\text{m}$ ). The long hairs are deflected by the trochantin as the leg is swung forward during the swing phase of walking, and their sensory neurones respond phasically during an imposed deflection and tonically if the deflection is maintained. Selective stimulation of the long hairs elicits a resistance reflex that rotates the coxa posteriorly and is similar to that occurring at the transition from the swing to the stance phase of walking. The motor neurones innervating the posterior rotator and adductor coxae muscles are excited, and those to the antagonistic anterior rotator muscle are inhibited. By contrast, selective stimulation of the short hairs leads only to a weak inhibition of the anterior rotator. The excitatory effects of the long hairs are mediated, in part, by direct connections between their sensory neurones and particular motor neurones. A spike in a sensory neurone

elicits a short-latency depolarising postsynaptic potential (PSP) in posterior rotator and adductor motor neurones whose amplitude is enhanced by hyperpolarising current injected into the motor neurone. When the calcium in the saline is replaced with magnesium, the amplitude of the PSP is reduced gradually, and not abruptly as would be expected if an interneurone were interposed in the pathway. Several sensory neurones from long hairs converge to excite an individual motor neurone, evoking spikes in some motor neurones. The projections of the sensory neurones overlap with some of the branches of the motor neurones in the lateral association centre of the neuropile. It is suggested that these pathways would limit the extent of the swing phase of walking and contribute to the switch to the stance phase in a negative feedback loop that relieves the excitation of the hairs by rotating the coxa backwards.

Key words: *Schistocerca gregaria*, locust, motor control, mechanoreceptor, hair plate, motor neurone.

### Introduction

Transitions between the swing and stance phases of walking depend on both proprioceptive signals reporting the position, velocity, acceleration and load on the leg and on the activity of a central pattern-generating circuit (Pearson, 1993). Although the central nervous system is sufficient for producing patterns of activity in the motor nerves that resemble the walking pattern (Grillner and Zangger, 1979; Sillar and Skorupski, 1986; Ryckebusch and Laurent, 1993), sensory input is necessary for adapting the motor output to the actual progression of the animal with respect to the substratum and the load on each leg. Thus, passive movement of the leg, or stimulation of a specific proprioceptive organ, can evoke a transition from the stance (or power) phase of walking to the swing (or recovery) phase (Grillner and Rossignol, 1978) and entrain an ongoing rhythm (Andersson *et al.* 1978; Sillar *et al.* 1986).

In insects, groups of hairs arranged in tight clusters called hair plates provide proprioceptive information for the

transition from swing to stance during walking (Cruse *et al.* 1984; Dean and Schmitz, 1992; Graham and Wendler, 1981; Wong and Pearson, 1976). Each hair plate consists of a group of short, closely spaced hairs between segments of either the body or a leg. As the joint is closed, the hairs are deflected by folds of the joint membrane, or by sclerotized cuticle, and the resulting excitation of their sensory neurones provides proprioceptive information about the state of the joint. In the legs, hair plates occur at the more proximal joints between the thoraco-coxal and coxo-trochanteral segments (locusts, Pflüger *et al.* 1981; cockroaches, Pringle, 1938; stick insects, Wendler, 1964; honeybees, Markl, 1963; moths, Kent and Griffin, 1990). Earlier work in cockroaches has described the morphology (French and Sanders, 1979) and signalling properties (French and Wong, 1976) of hair plate sensory neurones, and their synaptic connections with motor neurones and unidentified interneurones (Pearson *et al.* 1976). In the locust, where much is known of the network of synaptic connections underlying

the processing of other types of proprioceptive signals (Burrows, 1987; Burrows *et al.* 1988; Laurent and Hustert, 1988), relatively little is known about the reflex effects and central synaptic connections of hair plate receptors, although the central projections of some have been described (Pflüger *et al.* 1981). This paper shows that the sensory neurones of the ventral coxal hair plate excite motor neurones that innervate two coxal muscles and thus form a negative feedback loop which could be used to limit the excursion of the leg during the swing phase of locomotion.

### Materials and methods

Adult male and female locusts, *Schistocerca gregaria* Forskål, were used from our crowded colony. A locust was fixed ventral side uppermost in Plasticine with the legs restrained. The ventral coxal hair plate of the right mesothoracic leg was exposed by rotating the leg dorsally and pulling the coxa and trochanter posteriorly. Electromyograms were recorded from selected thoraco-coxal muscles with pairs of stainless-steel wires, 30  $\mu\text{m}$  in diameter and insulated to their tip, inserted through holes in the overlying cuticle.

To make intracellular recordings from motor neurones in the mesothoracic ganglion, the sterna of the meso- and metathoracic segments were removed and the meso- and metathoracic ganglia were stabilised on a wax-covered silver platform. The sheath of the ganglion directly over the cell bodies of the target neurones was softened by applying crystals of protease (Sigma type XIV) for 30 s, and then rinsing with saline. The body cavity and central nervous system were then continuously perfused with locust saline (Usherwood and Grundfest, 1965). In experiments designed to investigate synaptic transmission, calcium ( $4\text{ mmol l}^{-1}$ ) was replaced by magnesium. The intracellular microelectrodes were usually filled with  $2\text{ mol l}^{-1}$  potassium acetate and had resistances of 40–60 M $\Omega$ , but for intracellular staining they were filled with hexamminecobaltic chloride (6% w/v). Motor neurones were identified either by recording an antidromic spike evoked by electrical stimulation of a specific muscle or by correlating their intracellular spikes with those in a myogram. All data were stored on a four-channel FM tape recorder (Racal Instruments) for later analysis on a microcomputer using the SPIKE2 and SIGAVG programs (Cambridge Electronic Design).

Hairs belonging to the hair plate were stimulated mechanically by a small pin mounted on the central cone of a small loudspeaker or on a piezo-electric device driven by a ramp-and-hold generator (Matheson and Ditz, 1991). The long hairs were sometimes stimulated electrically through the broken tip of a saline-filled microelectrode pressed against their bases. The sensory spikes were recorded extracellularly by a monopolar oil-hook electrode on nerve 3B (N3B), or by an intracellular electrode from this nerve close to where it enters the mesothoracic ganglion. The specificity of the electrical stimulus was attested by the sudden disappearance of the sensory spike in nerve 3B when the electrode was moved

from long hairs to the space between the long and short hairs. Sensory neurones from the hairs of the hair plate were stained by back-filling with hexamminecobaltic chloride (6% w/v). The tips of the long hairs were shaved and the hair plate surrounded with a wall of petroleum jelly that was then filled with the cobalt solution. The cobalt was allowed to diffuse for 18–24 h before the ganglion was dissected out of the locust, the stain developed and then intensified with silver (Bacon and Altman, 1977). Drawings of the stained neurones were made from whole mounts of the mesothoracic ganglion and the projections of the sensory neurones were analysed further from 10  $\mu\text{m}$  serial transverse sections.

Muscle activity resulting from the stimulation of the hair plate was recorded electromyographically in 13 locusts, and the connections of the sensory neurones with the motor neurones were determined in 29 locusts. Intracellular recordings were made from the sensory neurones in 11 locusts and they and the motor neurones were stained in 23 locusts.

### Results

#### *Anatomy of the thoraco-coxal joint*

The coxa articulates with the thorax by a monocondylic joint between its dorsal rim and the pleuron (Fig. 1A) (Hustert, 1983; Snodgrass, 1929). We have focused on the more accessible ventral muscles in a search for the effects and central connections of the ventral hair plate. In the mesothorax, three muscles originate on the sternum and insert on the ventral margin of the coxa (Fig. 1B): the anterior rotator coxae (muscle 92 of Snodgrass, 1929), the posterior rotator coxae (muscle 100) and the adductor coxae (muscle 93). The posterior rotator and adductor muscles are synergists and cause the leg to swing backwards. They are active during the stance phase of walking and are antagonised by the anterior rotator, which swings the leg forwards and is active during the swing phase (Hustert, 1982).

The ventral coxal hair plate (cxHP1) is a compact group of hairs on the proximal rim of the coxa near the insertion of the anterior rotator muscle (Fig. 1A,B). It consists of two types of hairs (Fig. 1C); first, a group of 10–15 closely spaced, slender hairs, approximately 70  $\mu\text{m}$  long, and second a group of 20–30 widely spaced, shorter and thicker hairs, approximately 30  $\mu\text{m}$  long. Except where noted, our experiments are concerned only with the long hairs.

#### *Response properties of sensory neurones from the ventral coxal hair plate*

The long hairs are sensitive to the position and velocity of anterior rotation of the coxa about its dorso-ventral axis. During walking on a horizontal surface, the tibia is tilted backwards so that the femoro-tibial joint is posterior to the tarsus, and the ventral margin of the coxa swings in an anterior-medial direction during the swing phase. This motion causes the joint membrane to buckle, pushing the trochantin (a small cuticular sclerite in the membrane of the coxal joint that articulates with the coxa) over the hair plate and bending the

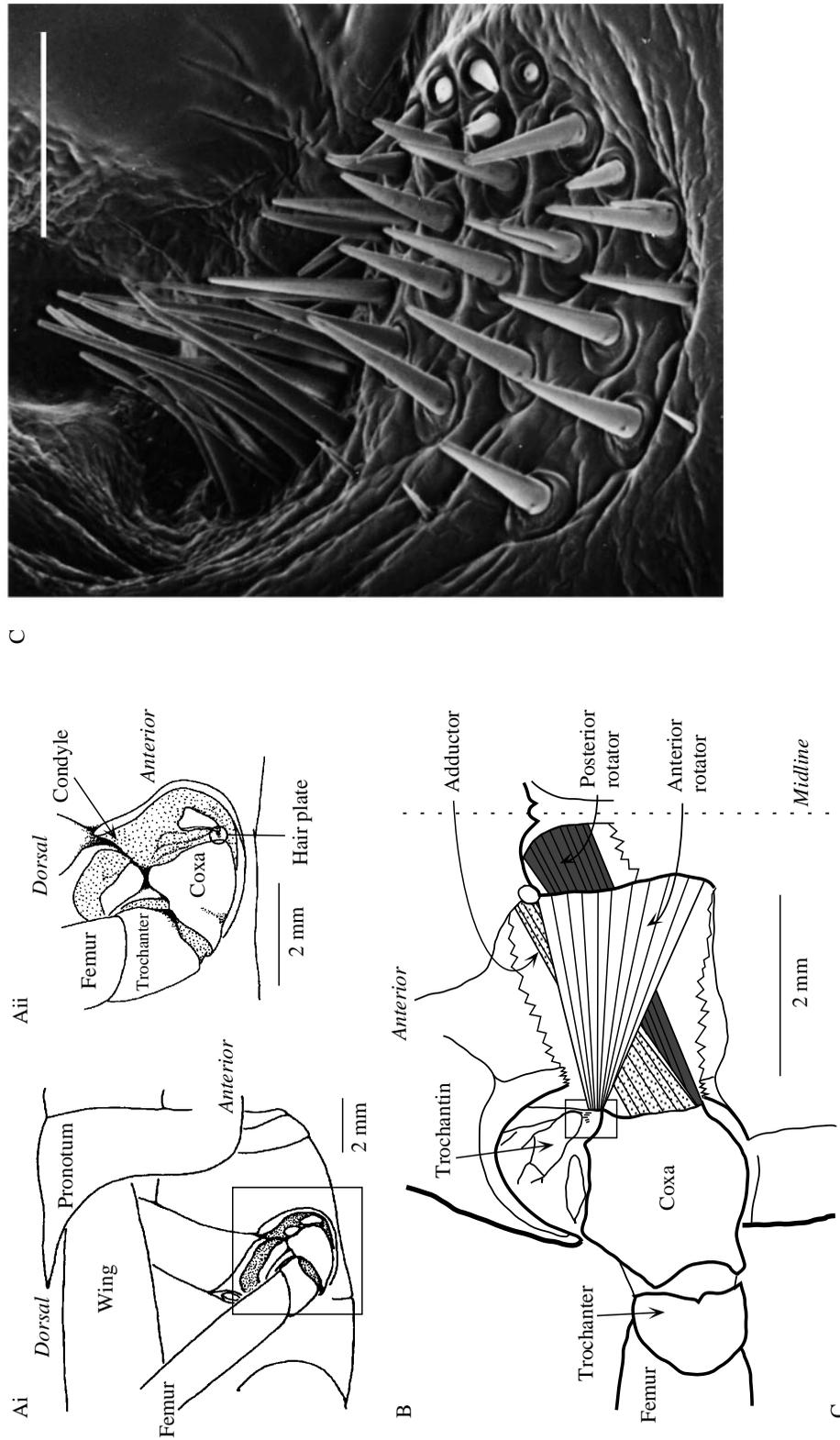


Fig. 1. Anatomy of the thoraco-coxal joint and the ventral coxal hair plate (cxHPI) of a middle leg. (Ai) View of the right side of the mesothorax to show the articulation of the right middle leg with the thorax. (Aii) Enlarged view of the area boxed in Ai showing the position of the ventral coxal hair plate and the condyle of the thoraco-coxal articulation. (B) Ventral view to show the three muscles that connect the mesothoracic sternum with the coxa. The anterior rotator coxae originates on a cuticular ridge that extends posteriorly from the mesosternal apophysis and inserts on the coxal margin near the ventral coxal hair plate. The posterior rotator coxae originates on the mesosternal spina and inserts on the posterior coxal margin. The adductor coxae originates on the apophysis and has a broad insertion on the ventral coxal margin. The dotted line indicates the ventral midline of the animal. (C) Scanning electron micrograph (equivalent to the boxed area in B) of the hair plate showing the tightly packed long hairs and more widely spaced short hairs. Scale bar, 50  $\mu\text{m}$ .

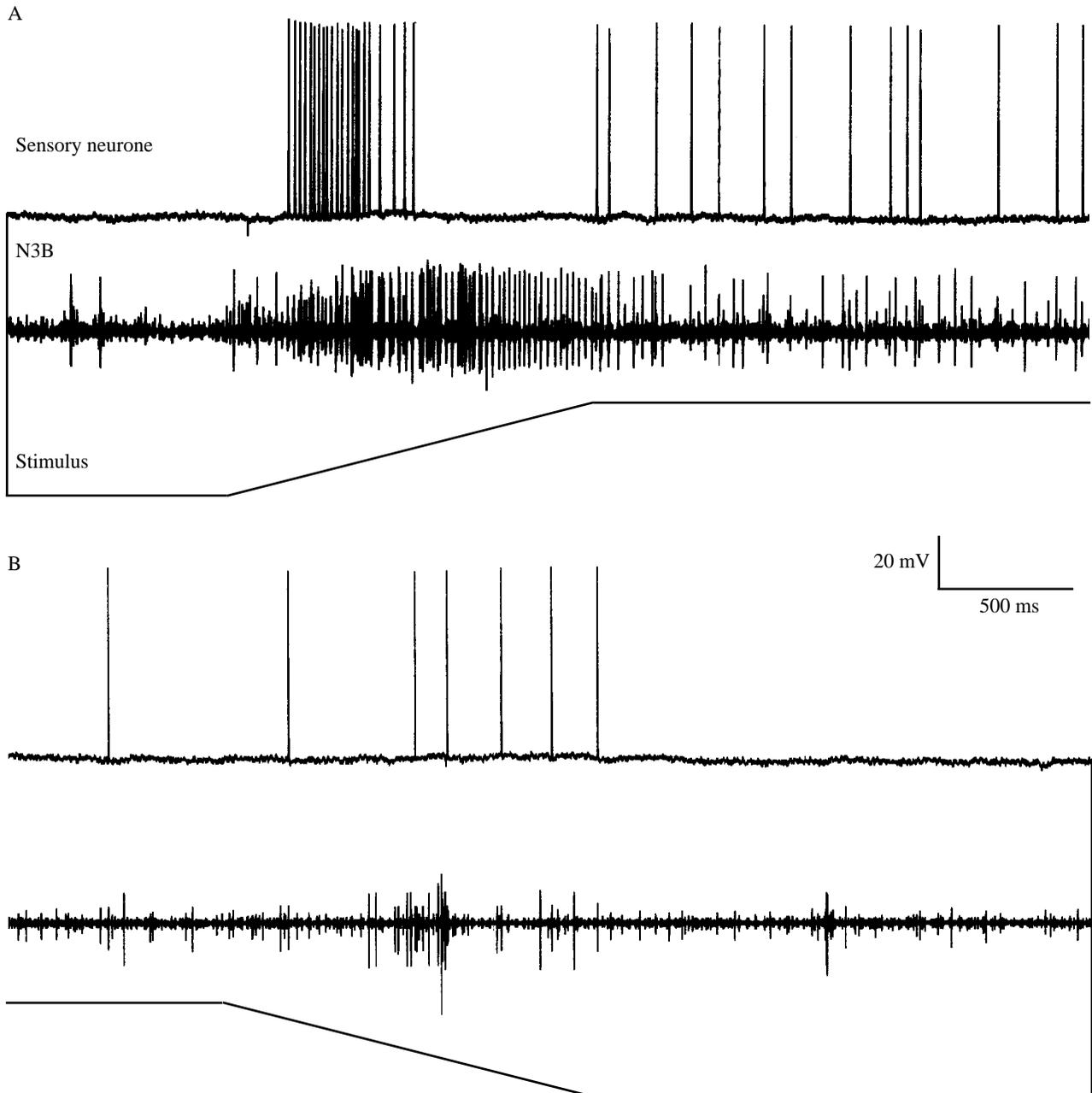


Fig. 2. Response of the hair plate to a mechanical stimulus. (A) An intracellular recording from a single sensory neurone and the summed extracellular recording from the axons of hair plate sensory neurones in N3B. When the hairs are bent in the direction they would normally be pushed by the trochantin, there is a phasic response, and during sustained displacement there is a tonic response of lower frequency. (B) When the stimulus is removed the sensory neurone stops spiking.

hairs towards the periphery. When the leg is in a more natural position than in Fig. 1C, the long hairs rest against the trochantin, even when the leg is fully retracted. Deflecting the hairs in a similar manner with a pin increased the spike activity of their sensory neurones transiently during the movement and to a lower sustained level while the hairs remained deflected. In most extracellular recordings from N3B, the spikes of individual sensory neurones could not be differentiated reliably, but the summed activity clearly showed both phasic

and tonic components. Both these components were, however, also apparent in intracellular recordings of single sensory neurones (Fig. 2A,B). Thus, as in the cockroach trochanteral hair plate (French and Wong, 1976), sensory neurones from the locust coxal hair plate can code coxal movement and provide some information about static position. The spikes are conducted in the axons of these sensory neurones at a velocity of  $1.3 \pm 0.1 \text{ m s}^{-1}$  (mean  $\pm$  s.d.,  $N=3$ ) as measured between an intracellular electrode placed close to the entry of N3 to the

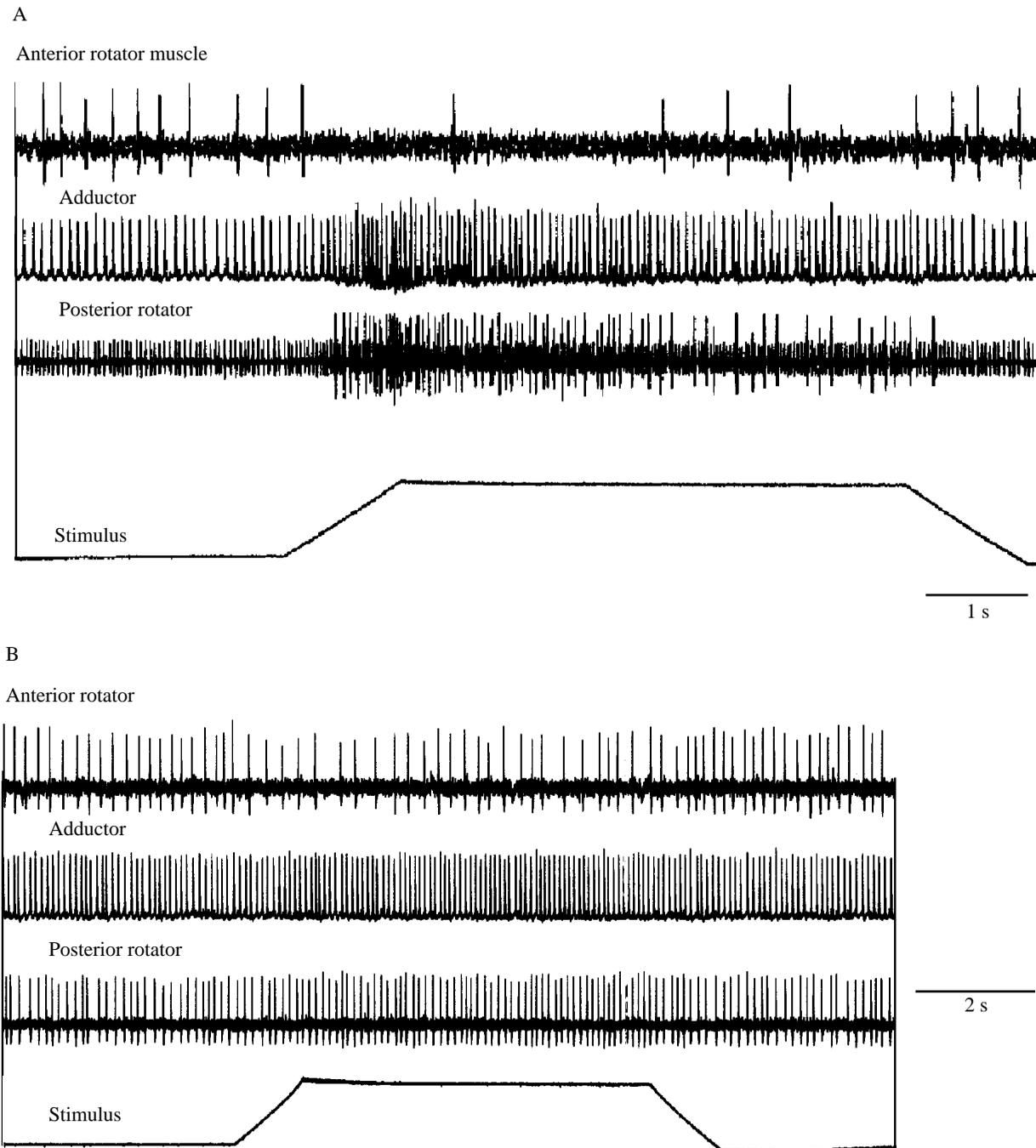


Fig. 3. Mechanical stimulation of the hair plate causes a negative feedback reflex in three thoraco-coxal muscles. (A) Mechanical stimulation of the long hairs of the hair plate excites motor neurones innervating the posterior rotator and adductor of the coxa and inhibits those to the anterior rotator. (B) Stimulation of the short hairs of the hair plate leads only to slight inhibition of the anterior rotator muscle.

mesothoracic ganglion and an extracellular electrode close to the hair plate. This average value for conduction velocity makes the assumption that all the sensory neurones conduct spikes at the same velocity.

*Reflex responses in coxal muscles to stimulation of the hair plate*

In a quiescent locust, stimulation of the long hairs of the hair

plate activated the ventral thoraco-coxal muscles in a way that was consistent with a negative feedback response (Fig. 3A). The frequency of spikes in a tonically active motor neurone to the posterior rotator muscle was increased, and spikes of other motor neurones to the same muscle were also recruited whose frequency declined as the deflection of the hairs was maintained (Fig. 3A). At the same time, the frequency of tonically active motor neurones innervating the adductor

muscle was also increased, while the spikes in the anterior rotator were decreased in frequency. Thus, there is a consistent excitation of motor neurones to two muscles and inhibition of those to a third. No effects of the stimulation were apparent in other coxal muscles such as the protractor and abductor coxae ( $N=4$ ), in the trochanteral depressor muscle ( $N=3$  for the thoracic branches,  $N=2$  for the coxal branches) or in the extensor and flexor tibiae muscles ( $N=2$ ). The trochanteral levator muscle received weak excitation ( $N=2$ ). The net effect of these changes during walking would be to lift and rotate the leg posteriorly and thus reduce the excitation of the hairs as the coxa rotates posteriorly. The changes in the motor spikes occurred within 10 ms of a step displacement of the hairs, so that, after allowing 2.7 ms for the conduction of the sensory spikes over a distance of 3.5 mm from the hair plate to the ganglion and  $5.4 \pm 1.5$  ms (mean s.d.,  $N=11$ , as measured from a spike in a soma to a spike in a myogram) for the motor spikes, only some 2 ms remains for the central processing. Stimulation of the equivalent hair plates in an ipsilateral front or hind leg had no effect on these muscles of a middle leg, suggesting that if intersegmental effects are present they must be weak in a quiescent locust. Stimulation of the short hairs generally did not elicit a response, but in one of five preparations a weak inhibition of the anterior rotator was observed (Fig. 3B).

#### *Innervation of ventral thoraco-coxal muscles*

The posterior rotator is innervated by at most six motor neurones ( $N=17$  backfills) which have their somata in the posterior ventral cortex of the mesothoracic ganglion. A medial cluster contains three large somata, whereas a more posterior and lateral cluster contains two smaller somata (numbers are modal values). Intracellular recordings from the medial neurones showed that they have the properties of fast motor neurones whereas the smaller lateral somata belong to slow motor neurones. The primary neurites of all the neurones converge in the dorsal neuropile before exiting the ganglion in nerve 4. The somata of two motor neurones of the adductor coxae muscle, and two motor neurones innervating the anterior rotator, are in the anterior region of the ganglion ventro-medial to the root of N1 (numbers are modes of four and six backfills respectively).

#### *Central connections between hair plate sensory neurones and motor neurones*

All the posterior rotator motor neurones from which recordings have been made responded to a ramp mechanical stimulus to the hair plate with a depolarisation, and some also spiked (Fig. 4). Slow motor neurones typically responded with an initial high-frequency burst of spikes followed by more sporadic spikes as the stimulus continued (Fig. 4A). By contrast, a fast motor neurone often showed only a depolarisation that did not lead to spikes, although motor neurones did spike as indicated by the activity of the myogram (Fig. 4B). At least part of the depolarisation of both types of motor neurones can be attributed to direct, excitatory synaptic inputs caused by the release of chemical transmitter from the sensory neurones for the following four reasons.

Fig. 4. Motor neurones to the posterior rotator muscle are excited by mechanical stimulation of the hair plate. (A) A slow motor neurone is depolarised and spikes rapidly during the stimulus and more sporadically as the stimulus is maintained. The motor neurone was held slightly hyperpolarised throughout the recording. (B) A fast motor neurone is also depolarised by the mechanical stimulus but does not spike. An extracellular recording from N3B shows the summed sensory discharge, and the myograms show the activity of other motor neurones innervating the posterior rotator muscle.

First, a sensory spike was followed by a depolarising postsynaptic potential (PSP) in the motor neurone that could only be discriminated from the background synaptic activity after averaging 20–60 sweeps triggered from the sensory spikes in N3B (Fig. 5A). The latency between the sensory spike in the periphery and the beginning of the synaptic response was  $4.1 \pm 1.1$  ms ( $N=7$ ) in the slow motor neurones and  $3.4 \pm 0.9$  ms ( $N=10$ ) in the fast motor neurones. Electrical stimulation of the hair plate that excited several sensory neurones simultaneously evoked a PSP in all fast motor neurones tested (Fig. 5B) that consistently followed each stimulus with a latency of  $3.3 \pm 0.7$  ms ( $N=14$ ). The average latency from both methods of activation in all motor neurones was  $3.3 \pm 1.0$  ms ( $N=25$ ). It took the sensory spike approximately 1.5 ms to travel the 2.3 mm from the recording site on N3B to the mesothoracic ganglion, so that the central processing time was approximately 1.8 ms, which is within the range expected for monosynaptic connections in insects. To test if more than one of the 10–15 sensory neurones from the long hairs converged on the motor neurones, the electrical stimulus was gradually increased in strength so that the current spread progressively to more hairs. The compound action potential recorded in N3B then increased in a stepwise fashion and parallel increases occurred in the compound PSP in a posterior rotator motor neurone, indicating the progressive recruitment of sensory neurones and their common connection to the motor neurones (Fig. 5C).

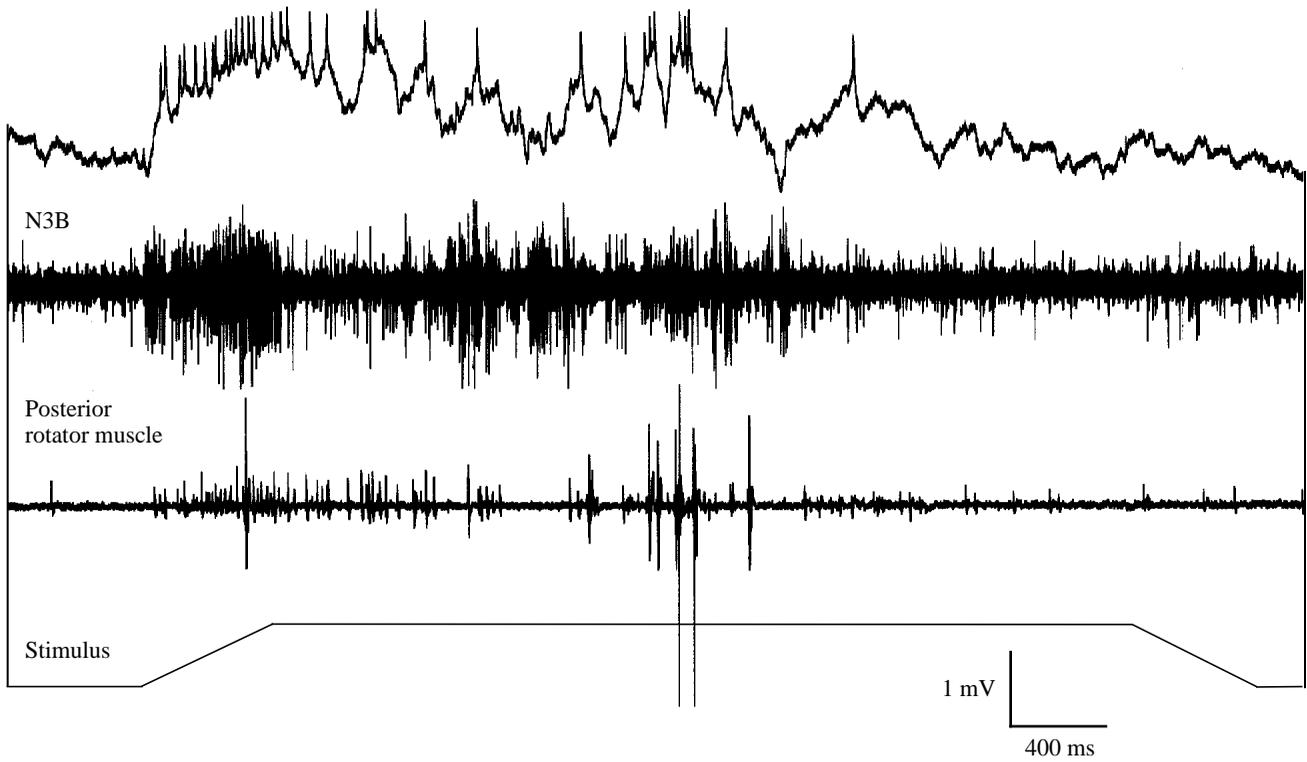
Second, when the calcium in the saline was replaced by magnesium, the amplitude of the evoked PSP fell gradually, sometimes to only 20% of its original value, without the sudden failure that would be expected if spiking interneurones were interposed in the pathway (Fig. 5D).

Third, the amplitude of the PSP was increased by applying a hyperpolarizing current to a postsynaptic motor neurone (Fig. 5B). The summed PSPs could also evoke spikes in the motor neurones, indicating that they were excitatory and thus excitatory postsynaptic potentials (EPSPs).

Finally, the branches of both the motor neurones and the sensory neurones overlap in the lateral association centre (nomenclature of Pflüger *et al.* 1988) of the neuropile. The primary neurites of both slow and fast motor neurones extend dorsally from their somata and then course laterally through the neuropile, sending some branches into the lateral association centre (Fig. 6A). Hair plate sensory neurones send branches dorsally into the lateral association centre and other

A

Slow posterior rotator motor neurone



B

Fast posterior rotator motor neurone

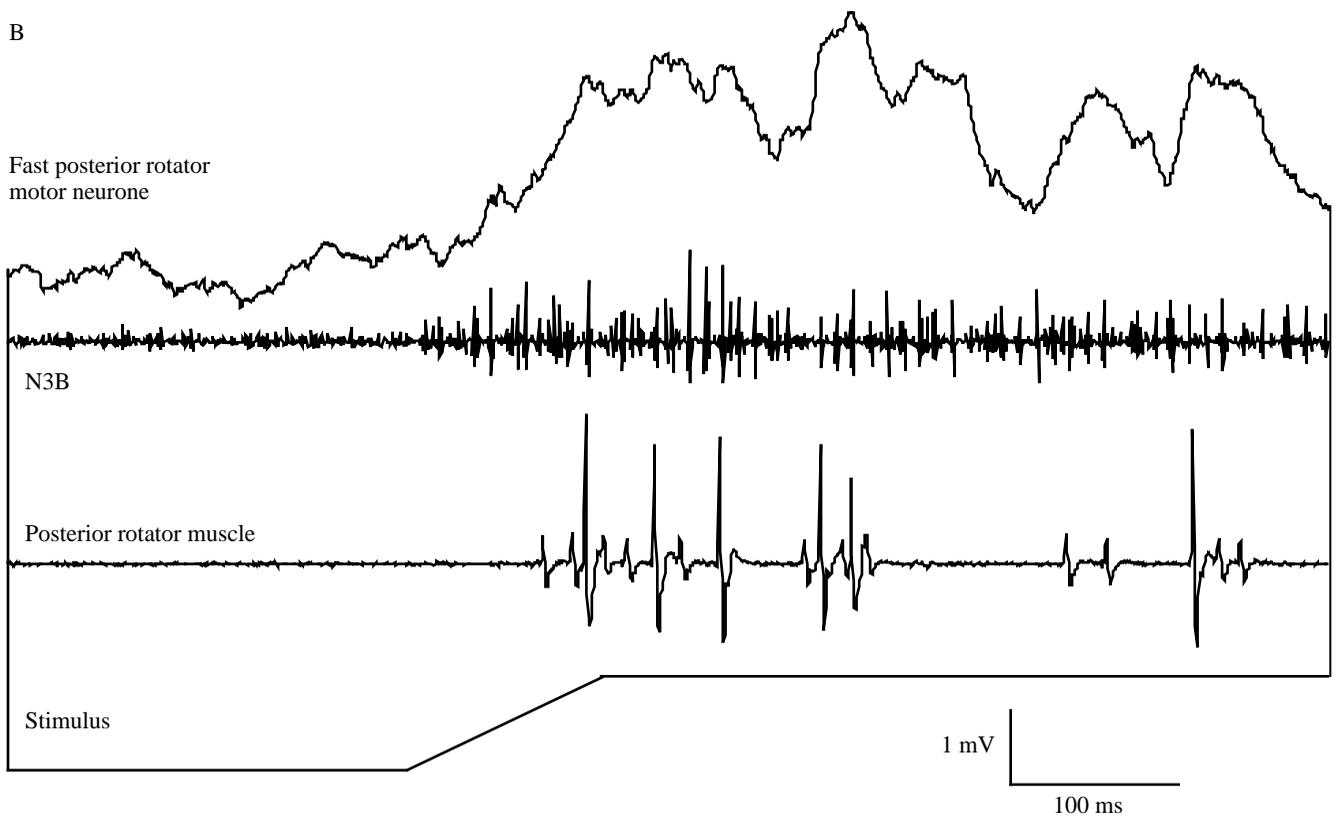


Fig. 4

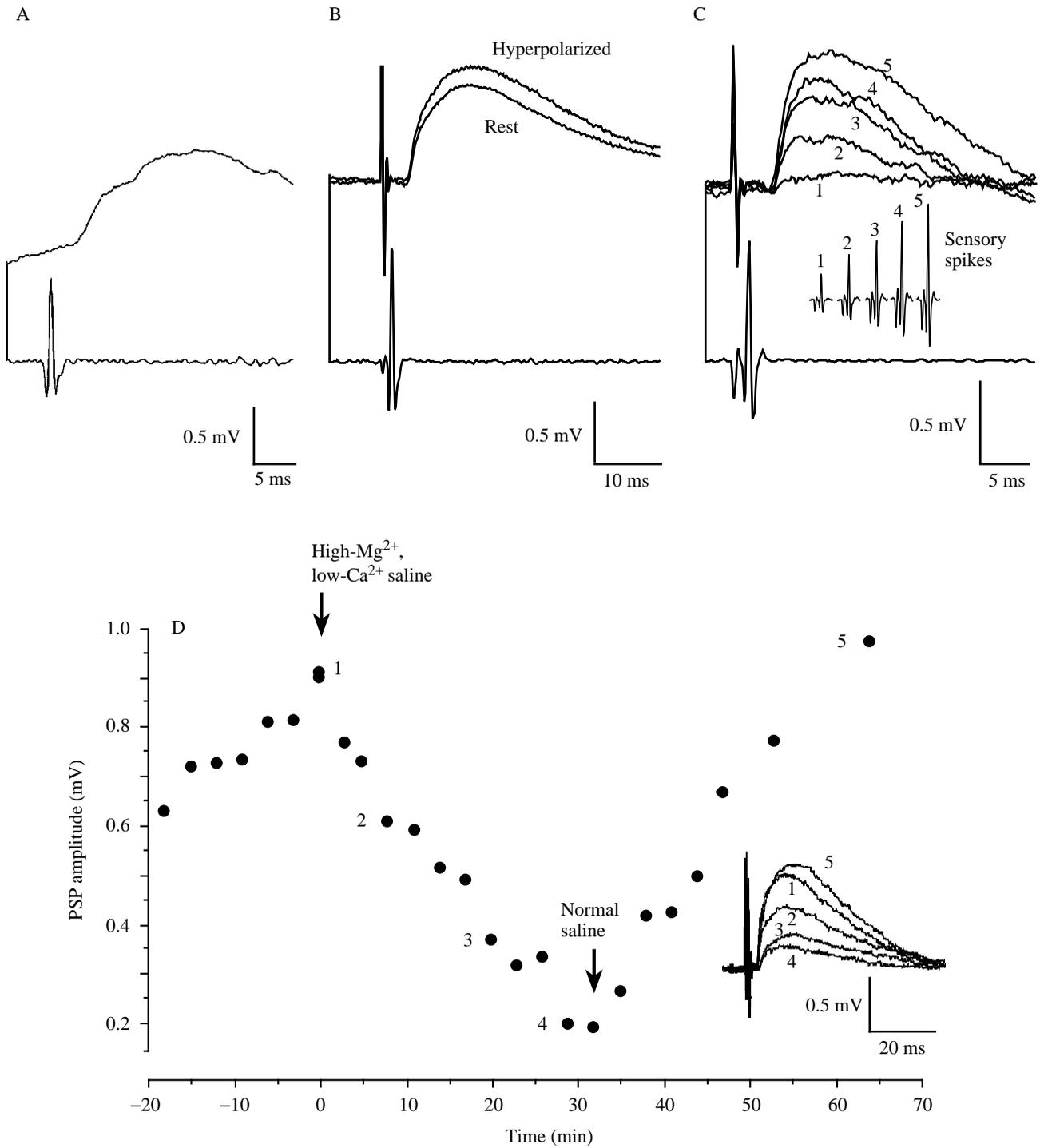


Fig. 5

branches into the more ventral neuropile, where there are no branches of the motor neurones (Fig. 6B).

Motor neurones to the adductor coxae muscle also received direct input from the hair plate sensory neurones. Mechanical stimulation of the hair plate caused depolarisation and spiking of motor neurones (Fig. 7A), as recorded in two different locusts. Signal averaging from electrical stimuli delivered to the hair plate revealed a depolarising PSP with a central delay

of 1.1 ms (Fig. 7B), which again suggests a monosynaptic connection.

### Discussion

This paper has shown that mechanical stimulation of the long hairs of the ventral coxal hair plate excites motor neurones that innervate posterior coxal rotator and adductor muscles of

Fig. 5. Sensory neurones from the hair plate make chemical synapses directly onto posterior rotator motor neurones. (A) An average of 60 responses, triggered from extracellularly recorded sensory spikes and selected by a window discriminator, reveals a depolarizing potential in the motor neurone with a latency of 2.8 ms. The extracellular spikes were recorded 2.3 mm from the mesothoracic ganglion, so that the central latency is 1.3 ms. The spikes were evoked in bursts by repeated mechanical stimulation of the hair plate, and the average includes spikes that occur late in these bursts. This means that the baseline shows a slow depolarization before the sensory spike, and the high frequency of spikes means that successive PSPs are captured in each sweep, causing the second notch in the depolarisation. (B) The amplitude of the evoked PSP is increased by hyperpolarising current injected into the motor neurone. The PSPs were evoked by electrical stimulation of the hair plate, and each trace represents an average of 90 sweeps. (C) Several sensory neurones converge on a posterior rotator motor neurone. Increasing the intensity of an electrical stimulus progressively recruits more sensory neurones so that the amplitude of their summed extracellular spikes increases (inset), and there is a parallel increase in the amplitude of the compound PSP in the motor neurone. Each trace is an average of 30 sweeps. (D) Replacing the  $\text{Ca}^{2+}$  in the saline with  $\text{Mg}^{2+}$  caused the PSP to decrease gradually to 20% of its original amplitude in normal saline. The stimulus was adjusted to just above threshold for a constant-latency spike in N3B. Upon return to normal saline, the amplitude recovered. Each point is the result of 50 averaged sweeps. The inset shows averages from the numbered data points. In this example, the initial increase in amplitude is due to recovery from a previous treatment with high- $\text{Mg}^{2+}$ , low- $\text{Ca}^{2+}$  saline.

a middle leg and inhibits anterior rotator motor neurones. The excitation of a motor neurone is due in part to the direct and convergent connections that they receive from sensory neurones of the long hairs. These connections form part of a negative feedback loop that would swing the coxa backwards and thus reduce the deflection of the hairs.

At the end of the swing phase of walking, the motor neurones innervating the anterior rotator muscle stop spiking and those innervating the posterior rotator and adductor muscles begin spiking (Hustert, 1982, 1983). Mechanical or electrical stimulation of the ventral coxal hair plate produces the same pattern of activity in these motor neurones, and the receptors will normally be stimulated towards the end of the swing phase. These observations suggest that the reflex effects of the hair plate on these motor neurones could limit the extent of the swing phase and contribute to the transition to the stance phase of walking. Ablation of the ventral coxal hair plate of stick insects or of the trochanteral hair plate in cockroaches causes the swing phase movements to be exaggerated (Dean and Schmitz, 1992; Cruse *et al.* 1984; Wong and Pearson, 1976), whereas tonically deflecting the hairs with wax brings the swing phase movements to a premature end (Bässler, 1977). In other animals, proprioceptors in the proximal joints also have a particularly strong effect on shaping the motor pattern for walking. For example, the walking pattern of low spinal cats can be entrained by forced movements of the femur or the hip joint (Andersson *et al.* 1978; Grillner and Rossignol, 1978), and

the fictive locomotion produced by the isolated ganglia and proximal leg segments of the crayfish can be entrained by thoraco-coxal proprioceptors (Sillar *et al.* 1986; Elson *et al.* 1992). If the distal segments of a stick insect's leg are removed and replaced with a prosthesis, the proprioceptive information from the remaining part of the proximal limb is sufficient to maintain normal intersegmental coordination (Wendler, 1966). In locusts, evidence for a central pattern generator for walking has been elusive, but Ryckebusch and Laurent (1993) have shown that a motor pattern similar to that expressed during locomotion can be induced in isolated thoracic ganglia with the muscarinic agonist pilocarpine. In isolated ganglia of stick insects showing similar rhythms, stimulation of the femoral chordotonal organ can evoke changes that resemble the transition between stance and swing in forward walking (Büschges and Böhme, 1994).

#### *Connections made by sensory neurones from hairs*

The resistance reflex mediated by the ventral coxal hair plate results in part from the direct excitatory connections between its sensory neurones and coxal motor neurones. Motor neurones of the posterior rotator and adductor muscles responded consistently to hair plate stimulation with short-latency depolarisations which could sum to produce spikes. The central latencies of these connections are within the range 1–2 ms measured for monosynaptic connections in insects, for example, from the wing hinge stretch receptor to flight motor neurones (Burrows, 1975) and from other cuticular receptors to leg motor neurones (Laurent and Hustert, 1988). Much of this central latency is explained by the reduction in conduction velocity when the sensory spikes enter a ganglion and the differences in path length to the synapses with different postsynaptic neurones. The gradual decline in the amplitude of a PSP evoked by a sensory neurone from the hair plate in a posterior rotator motor neurone when magnesium ions are substituted for calcium ions is further indication that the connection is direct. If a spiking interneurone were interposed between the sensory neurone and the motor neurone, the PSP would be expected to fail when spike threshold was no longer exceeded. This experiment, however, does not eliminate the possibility that a nonspiking interneurone is interposed. Nevertheless, branches of the posterior rotator motor neurones and branches of the sensory neurones from the hair plate occur in the same dorsal and lateral region of neuropile, so that direct synaptic connections are anatomically possible. Finally, the convergence of hair plate sensory neurones onto motor neurones showed that although the PSP amplitude increased as more sensory neurones were recruited, there was no change in latency, as would be expected if the response depended on excitation of an interneurone.

Similar patterns of connections have been found between the sensory neurones from the trochanteral hair plate of the cockroach and coxal depressor motor neurones (Pearson *et al.* 1976), and between the tegula hair plate and wing elevator motor neurones of the locust (Pearson and Wolf, 1988), but the sensory neurones from hair plates at the base of the wings in

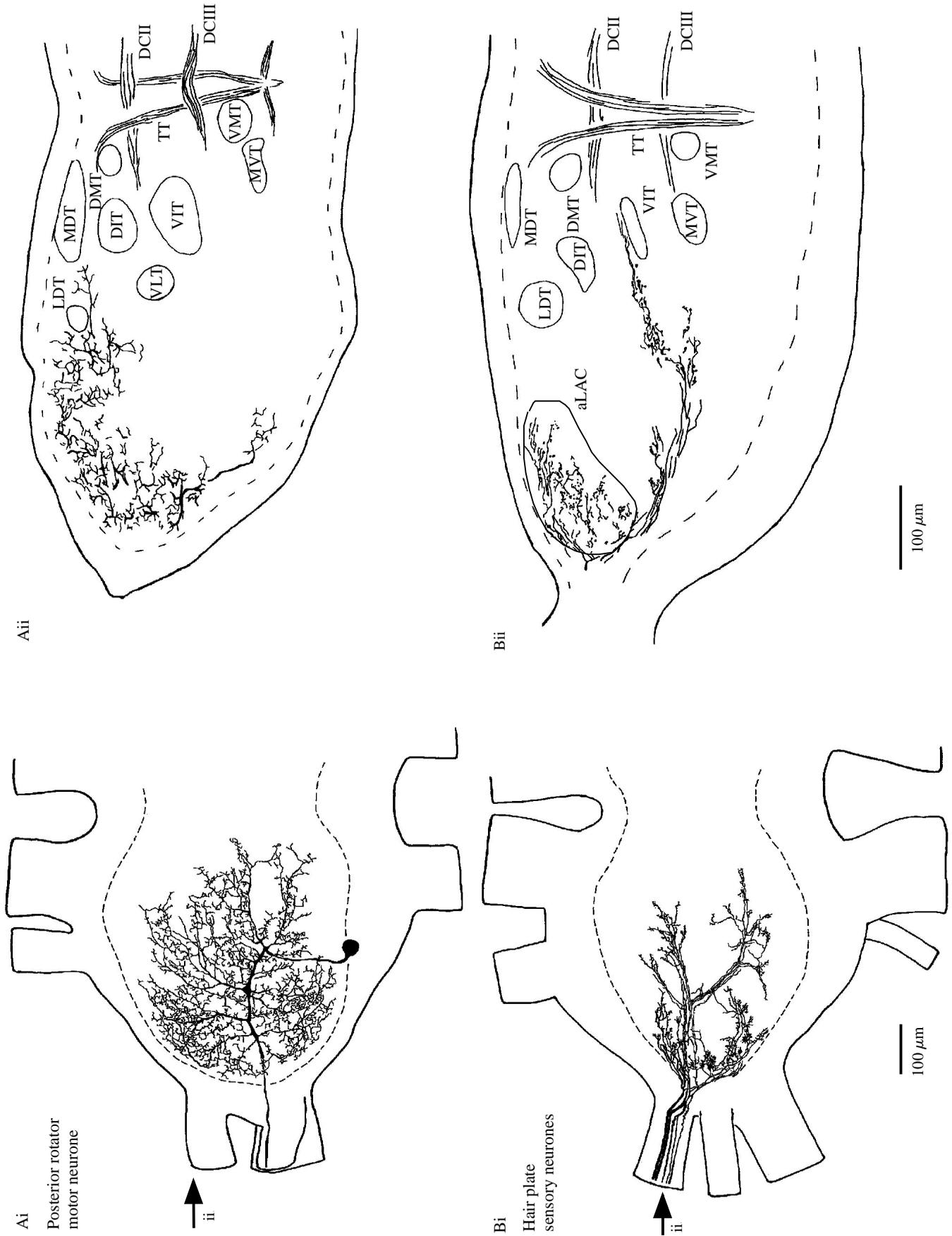


Fig. 6

Fig. 6. The branches of a posterior rotator motor neurone and sensory neurones from the hair plate overlap in the lateral association centre. (Ai) Drawing of a posterior rotator motor neurone stained intracellularly and viewed ventrally in a whole mount of the mesothoracic ganglion. (Aii) Three transverse  $10\ \mu\text{m}$  sections taken at the level indicated in Ai (arrow) and combined to show the anterior branches of the motor neurone in lateral and dorsal areas of neuropile. (Bi) Drawing of the projections of sensory neurones of long hairs from the hair plate stained by back-filling. (Bii) Three transverse  $10\ \mu\text{m}$  sections at the same level as for the motor neurone (arrow in Bi) show the presence of branches in the same area of lateral and dorsal neuropile, and branches in ventral neuropile where there are no branches of the motor neurone. Abbreviations (after Tyrer and Gregory, 1982): aLAC, anterior lateral association centre; DCII, DCIII, dorsal commissures II and III; DIT, dorsal intermediate tract; DMT, dorsal median tract; LDT, lateral dorsal tract; MDT, median dorsal tract; MVT, median ventral tract; TT, T-tract; VIT, ventral intermediate tract; VLT, ventral lateral tract; VMT, ventral median tract.

crickets excite motor neurones by polysynaptic pathways (Elliott, 1983). These latter hair plates differ from those at the base of the leg in that the hairs are longer and more widely scattered and may have more in common with the long tactile hairs scattered over the cuticle, which generally do not synapse with motor neurones either in insects (Siegler and Burrows, 1983) or in spiders (Milde and Seyfarth, 1988).

Taken together, these results suggest a difference between the processing of hair receptor information for proprioception and for tactile exteroception. The hairs with a proprioceptive function, signalling movements and positions of joints, mediate resistance reflexes that include a monosynaptic component, whereas the hairs with an exteroceptive function, signalling contact of the leg with an external object, mediate compensatory reflexes (Pflüger, 1980; Siegler and Burrows, 1986) that do not include direct connections with the motor neurones. This distinction also extends to other proprioceptors such as campaniform sensilla (Burrows and Pflüger, 1988; Laurent and Hustert, 1988), strand receptors (Skorupski and Hustert, 1991) and chordotonal organs in locusts (Burrows, 1987; Laurent and Hustert, 1988; Laurent, 1987) and crayfish (El Manira and Clarac, 1991), which all make direct connections with particular motor neurones.

The inhibition of the anterior rotator coxae motor neurones that results from stimulation of the ventral coxal hair plate is not caused by direct connections, as the inhibitory postsynaptic potentials that are evoked have a much longer latency than the EPSPs in posterior rotator motor neurones. This also accords with the results of Pearson *et al.* (1976) on cockroaches. Moreover, no mechanosensory neurones are known to make inhibitory connections within the central nervous system of an insect. The most likely interneurons to mediate the inhibition are those with cell bodies in a ventral midline population which receive direct inputs from hairs and make inhibitory output connections with other interneurons and with motor neurones (Burrows and Siegler, 1982; Siegler and Burrows, 1983). Our preliminary experiments suggest

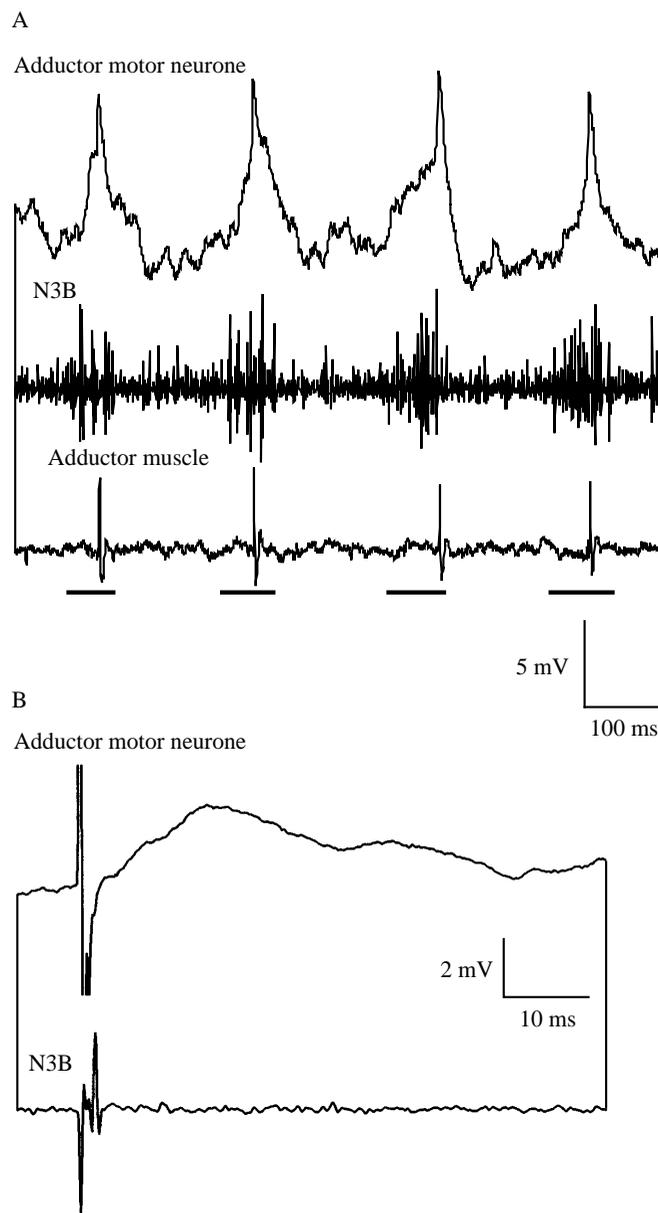


Fig. 7. An adductor motor neurone receives a direct synaptic input from hair plate sensory neurones. (A) The motor neurone depolarises and spikes in response to repeated mechanical stimulation of the hair plate (horizontal bars). (B) Electrical stimulation of the hair plate causes a compound PSP (average of 40 sweeps) with a latency of 2.5 ms from the sensory spike recorded in N3B.

that the sensory neurones from the hair plate synapse directly on certain interneurons within this population, and our further experiments are directed towards revealing details of this circuitry.

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