SWIMMERET PROPRIOCEPTORS IN THE LOBSTERS *NEPHROPS NORVEGICUS* L. AND *HOMARUS GAMMARUS* L.

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SUMMARY

1. The morphology, sensory responses and reflex effects of two proprioceptive systems in the swimmerets of the Norway lobster *Nephrops norvegicus* are described.

2. Two bipolar cells embedded in an elastic strand (strand B) which spans from the sternal rib to the proximal edge of the basipodite respond to stretch of the strand, applied directly or through swimmeret protraction. Powerstroke motoneurones are excited by a negative feedback reflex, and the transition from returnstroke to powerstroke movement is thereby sharpened. When protraction movements of the swimmeret are blocked, the intensity of beating is reduced both in the blocked swimmeret, and in neighbouring (particularly posterior) swimmerets.

3. A second receptor strand, the twisting muscle receptor (TMR), stretches from the sternal rib wall to the proximal end of the twisting muscle M10 in both the lobsters *Nephrops norvegicus* and *Homarus gammarus*. It contains the sensory endings of two cells which have somata in the abdominal ganglion. The axons of these cells convey conventional spikes in response to strand stretch, which occurs on release of M10 from imposed extension or following active M10 contraction. They produce a specific activation of M10 motoneurones, which represents a positive feedback reflex. This reinforces the twist of the swimmeret blade, so that the beat is directed laterally to its greatest extent throughout the powerstroke.

4. It is suggested that the TMR is homologous with the crayfish non-spiking swimmeret receptors, which also have central cell bodies. However, the receptors differ in their location, mode of afferent transmission and reflex actions. The discovery of these differences resolves anomalies between previous studies on lobsters and crayfish.

5. The results are discussed in terms of the homologies of all limb proprioceptors with central cell bodies in decapod crustaceans, and of the proprioceptive control of swimmeret beating.

INTRODUCTION

The study of swimmeret beating in decapod crustaceans has contributed much to our knowledge of how central pattern generators (CPGs) control rhythmic limb movements, both individually and as a linked chain of hemisegmental oscillators.

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The majority of this work has been carried out on crayfish (Hughes & Wiersma, 1960; Ikeda & Wiersma, 1964; Stein, 1971, 1974), while other studies have been made on lobsters and have concentrated on rather different aspects, such as details of the neuromuscular organization (Davis, 1968), motor patterns (Davis, 1969a) and possible 'command fibre' effects (Davis & Kennedy, 1972a,b,c). For a long time, the work of Davis (1969b) on *Homarus americanus* has also provided the only available information about the role of proprioceptive feedback in the regulation of swimmeret beating. He described two elastic strands which span the coxal joint, and concluded from sensory recordings that the primary effect of their proprioceptive input was to provide a positive feedback reinforcement of the powerstroke. These data were interpreted as support for the concept of central pattern generation, as developed by Wiersma for the crayfish, with proprioceptive reflexes relegated to the role of 'subservient amplifiers' for the centrally programmed motor pattern (Davis, 1969b).

Recent work by Heitler (1983) and Paul (Paul, 1981; Paul & Mulloney, 1985a,b) on the crayfish *Pacifastacus* has greatly increased our knowledge of the neuronal circuitry controlling swimmeret beating in this species. It is now established that motoneurones form part of the CPG (Heitler, 1978, 1981) and, from a detailed examination of the timing of motoneurone activities in the swimmeret motor programme, that the phases and relative durations of the powerstroke and return-stroke are independent of beating frequency (Heitler, 1983). In order to make adjustments to each part of the beat cycle within the generated rhythm, information has to be available about the position and movement of the swimmeret during both phases of its beat. Heitler (1983) therefore concluded that proprioceptive feedback must play a crucial role in the timing and coordination of the swimmeret rhythm. The source of this positional information has been identified as a pair of non-spiking neurones associated with an elastic strand which spans the coxal joint of the crayfish swimmeret (Heitler, 1982).

This recent emphasis on the importance of proprioceptive timing cues in the production of rhythmic motor patterns, both in the crayfish swimmeret and in other well-studied cyclical activities, e.g. locust flight (Pearson, 1985; Möhl, 1985a,b,c) and dipteran flight (Heide, 1979; Miyan & Ewing, 1984), brings into question the conclusions and interpretations of Davis (1969b) regarding the lobster swimmeret system. It also highlights the lack of comparative information about swimmeret systems in decapods, which show great variability both in their structure and their performance.

The present study re-examines the proprioceptors of the swimmeret of nephropid lobsters, and their involvement in the regulation of beating, and compares them with swimmeret receptors in other crustaceans. It was carried out in the course of a study of the contribution of swimmerets to righting behaviour, as expressed in a redirection of their beat when the animal is tilted (Neil & Miyan, 1986). Opportunity was therefore provided to study proprioceptive effects not only on the powerstroke and returnstroke timing, but also on the control of lateral twisting of the swimmeret. Our results suggest that proprioceptive feedback has a strong influence on the timing of events in the swimmeret beat, and on the coordination of movements of adjacent

swimmerets. We also describe a new proprioceptor, associated with the lateral twisting muscles, that exerts reflex control over their activity.

MATERIALS AND METHODS

General procedures for maintaining animals, and for performing the morphological and neurophysiological studies, are described in the preceding paper (Neil & Miyan, 1986). For experiments involving controlled movements of the swimmeret, the appendage was cut distal to the basipodite and a thin Perspex rod glued to the stump using cyanoacrylate adhesive. This was attached to the drive arm of a galvanometer (General Scanning Inc.) driven by a signal generator. The swimmeret was blocked in its movement by placing a Perspex rod in its path. Stretches were applied to specific muscles or receptor strands by grasping their cut ends either with forceps or with a small hook which was attached to a micromanipulator or to the galvanometer drive arm.

RESULTS

Anatomy of proprioceptors and associated muscles

The details of the anatomy of the swimmeret muscles in *Nephrops norvegicus* and *Homarus gammarus* follow essentially the same pattern as those in *Homarus americanus* described by Davis (1968). These are dealt with fully in the preceding paper (Neil & Miyan, 1986), and the nomenclature used by Davis (1968) is adopted here.

Each swimmeret is located in a socket of the ventral abdominal sternal rib, and articulates about a cuticular peg-and-hook joint which is surrounded by arthrodial membrane (see fig. 1 in Neil & Miyan, 1986). Two muscle bundles span the abdominal/coxobasal joints, numbered M9 and M10 by Davis (1968) (Fig. 1A). Muscle M10 comprises separate lateral and medial bundles of fibres, which originate on a posterior–medial area of the sternal rib socket, cross the joint diagonally and insert onto the lateral area of the basipodite cuticle, just ventral to the sclerite of M13. Muscle M9, which has an origin alongside M10, is a single bundle of fibres which inserts onto the anterior cuticular rim of the basipodite, medial to the sclerite of M13.

Receptor strand B

Lying alongside and originating between M9 and M10 is an elastic strand (strand B of Davis, 1968) which inserts laterally next to M9 on the basipodite rim. The orientation of strand B dictates that it will be stretched by protraction of the swimmeret (i.e. during the returnstroke movement), and unloaded during retraction (i.e. the powerstroke) (see fig. 3 in Neil & Miyan, 1986). The innervation of strand B in *Homarus americanus* was not clearly identified by Davis (1968, 1969b). In our serial sections of strand B in *Nephrops* we were also unable to make an unequivocal identification of a nerve supply or terminal structures. Furthermore, in cobalt backfills of the swimmeret nerve no innervation was seen along the length of the
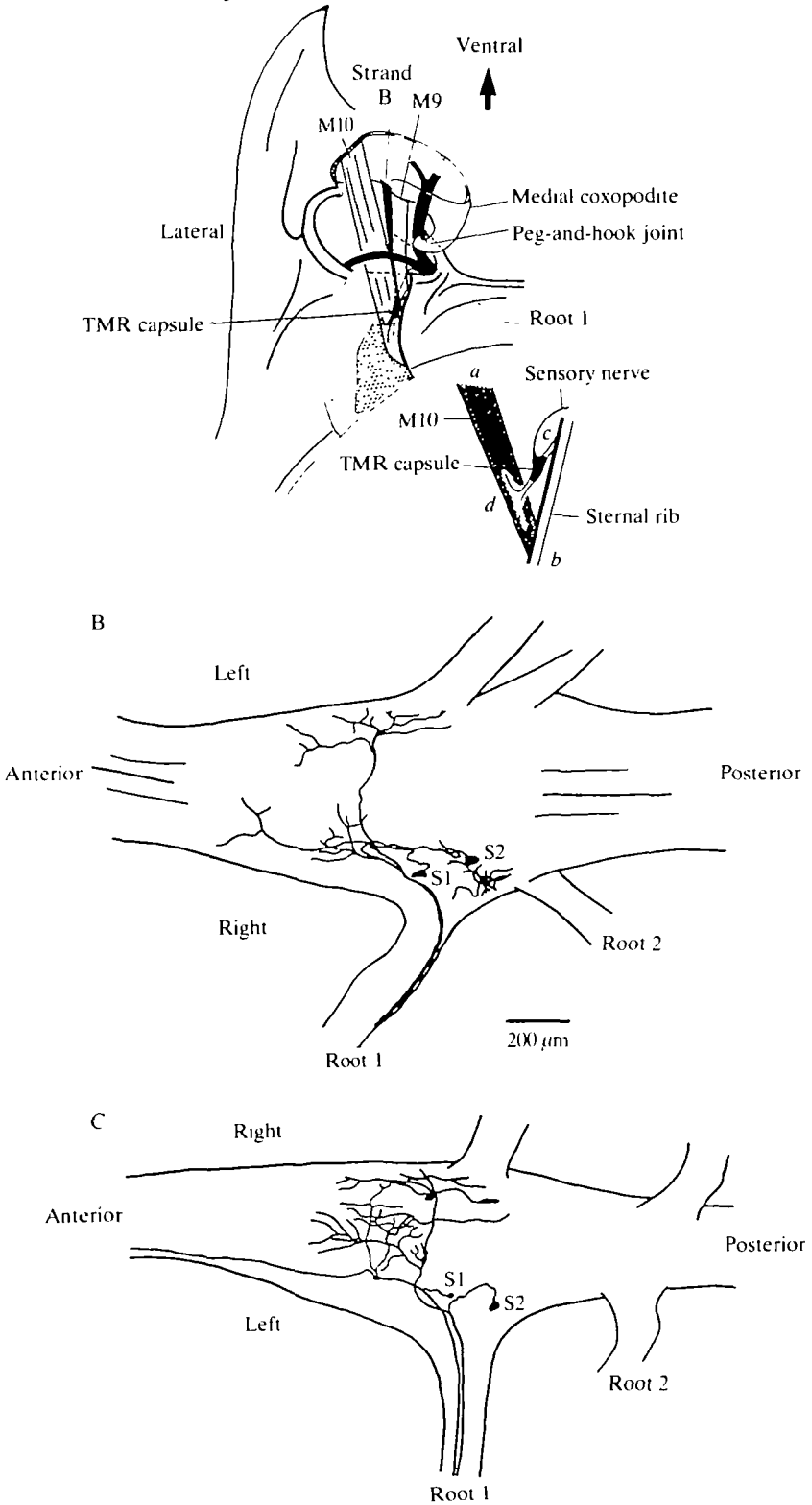
Fig. 1

strand. However, in these same fills two bipolar cells were observed at the base of strand B, encased in some form of capsule (Fig. 2). They send fibres of less than 5 μm diameter along a branch of the anterior first root, which may enter the posterior root before reaching the ganglion. These are the most prominent, and probably the only, receptors associated with strand B.

Twisting muscle receptor (TMR)

In both *Nephrops* and *Homarus gammarus* a second, and previously undescribed, receptor structure was found to be associated with the lateral twisting muscles. It will be called here the Twisting Muscle Receptor (TMR). The TMR has a similar location and structure in the two species: it is contained within an elastic strand which stretches from an origin at the base of the lateral bundle of M10 to the ventral rim of the sternal rib socket (Fig. 1A). This second strand is not homologous with strand A described by Davis (1969*b*) in *Homarus americanus*, which was not located in this study. Geometrical considerations indicate that M10 will be stretched at full protraction, thereby unloading the TMR. An imposed retraction will release M10 from stretch, and the consequent recoil of the connective tissue and other elastic elements at the base of the muscle will stretch the TMR (Fig. 1A, inset). Active contraction of M10 during retraction will also stretch the TMR, since the sarcomeres proximal to its insertion on the muscle will be shortening.

The TMR receptor strand has a clear innervation which is stained heavily by Methylene Blue and cobalt techniques (Fig. 3). The strand is innervated by two nerve fibres of 8–10 μm diameter which leave at its midpoint. A thick sheath encloses both fibres along their length, forming a capsule. Within the capsule fine dendritic processes extend to both ends, and these form complexes with very compact chains of some material (Fig. 4), which resemble the 'vacuolated strings' observed in the thoracicocoxal proprioceptors of decapod crustaceans (Whitear, 1965). The repeat pattern seen in the fibres in longitudinal section has the characteristic dimensions of collagen (i.e. a band length of 64 nm).

Fig. 1. (A) Diagram of the base of the right ($N = 3$) swimmeret, with the basipodite removed at the fused joint with the coxa, viewed from the posterior aspect to show the arrangement of the twisting muscles and the two main receptor systems (Strand B and TMR). The two bipolar cells described in the text, and shown in Fig. 2, lie at the proximal end of strand B. Inset: detailed view of the geometrical relationship between the lateral bundle of M10 (*a–b*) and the TMR (*c–d*). The twisting muscle receptor (TMR) strand attaches to the elastic connective tissue at the proximal end of the muscle (*d*). When the muscle is stretched (as by swimmeret protraction) the distance *b–d* will increase, and the receptor length *c–d* will decrease. When the muscle shortens (due either to release from stretch, or to active contraction) the distance *b–d* will decrease, and the receptor length *c–d* will increase. When contraction ends, the distance *b–d* will return to its resting value. (B,C) *Camera lucida* drawings of intensified centripetal cobalt fills from the TMR sensory nerve, showing the central somata and the pattern of arborization within the ganglion. (B) Preparation from right swimmeret, viewed from the ventral side. The cell bodies, S1 and S2, of the TMR are indicated to distinguish them from similar profiles which are dendritic concentrations. Note contralateral arborizations. (C) Preparation from left swimmeret, viewed from the dorsal side. Note contralateral projection, and the anterior projection of cell S1.
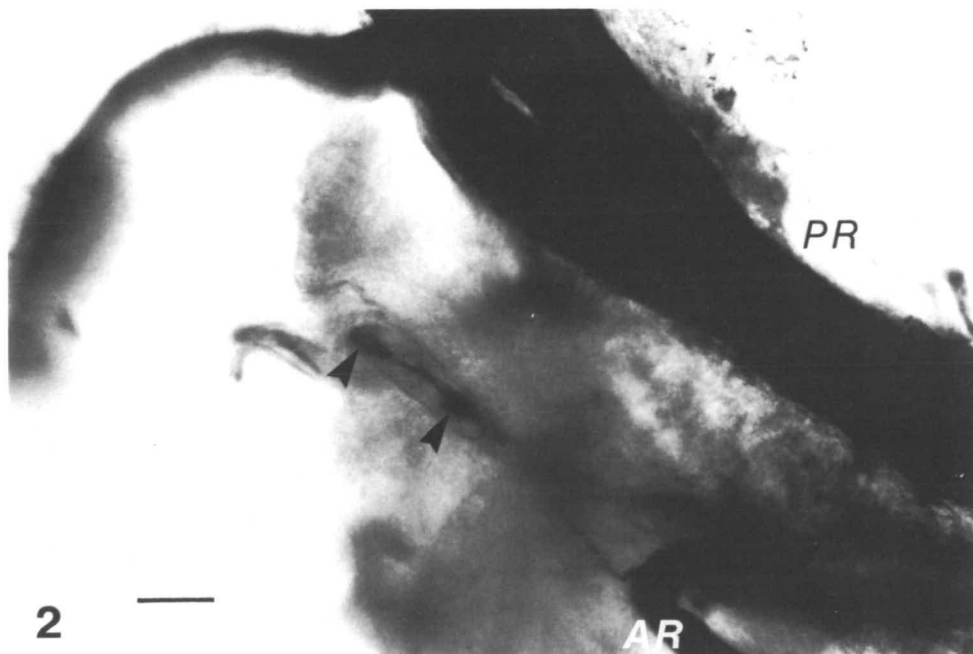
Fig. 2. Photomicrograph of a peripheral cobalt fill of the swimmeret nerve in *Nephrops norvegicus* showing two bipolar cell bodies (arrowheads) at the base of strand B. They are encased in a capsule within the matrix of the strand. *PR*, posterior branch; *AR*, anterior branch of first abdominal nerve root. Scale bar, 50 μm.

No cell bodies were found within the capsule or along the length of two nerve fibres as they traverse to the P1 branch of root 1. However, a centripetal cobalt fill of the fibres in *Nephrops* revealed the location of the two cell bodies within the ipsilateral hemiganglion (Fig. 1B). Extensive arborizations were found both here, and also in the contralateral hemiganglion. Additionally, some fills showed evidence of an anterior projection from one of the cells, although this could not be traced as far as the adjacent ganglion (Fig. 1C).

Sensory responses

The physiological experiments were all carried out on *Nephrops*. No attempt was made to investigate the responses of the ramal setae in this study (but see Davis, 1969b; Cattaert, 1984) and so the rami were removed to leave a stump of the basipodite. Recordings from the peripheral cut end of the posterior branch of the first abdominal root revealed two populations of sensory units, responsive to different directions of movement and held positions of the swimmeret.

Units sensitive to swimmeret protraction: strand B

These units were tonically active when the swimmeret was protracted (small unit in Fig. 5A and Fig. 5B, upper panel). Selectively stretching strand B produced similar activation (Fig. 5C), suggesting that receptors associated with this strand are
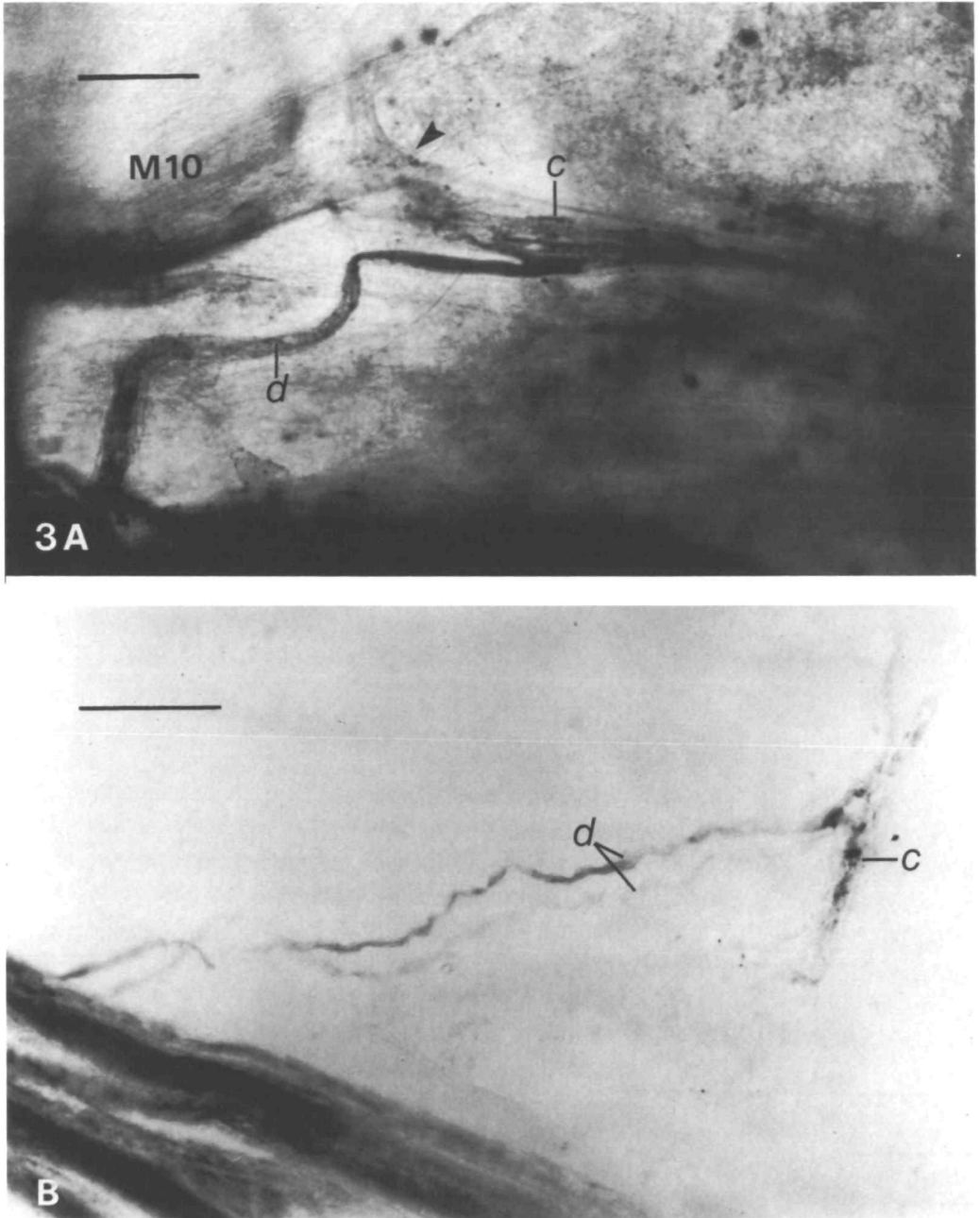
Fig. 3. Photomicrographs of the lobster twisting muscle receptor (TMR). (A) *Nephrops norvegicus*, stained with Methylene Blue. The attachment of the receptor strand to the base of the lateral bundle of M10 is indicated (arrowhead). (B) *Homarus gammarus*, stained with cobalt sulphide. This preparation shows the two dendrites which innervate the capsule. *d*, dendrites; *c*, TMR capsule. Scale bars, 50 µm.
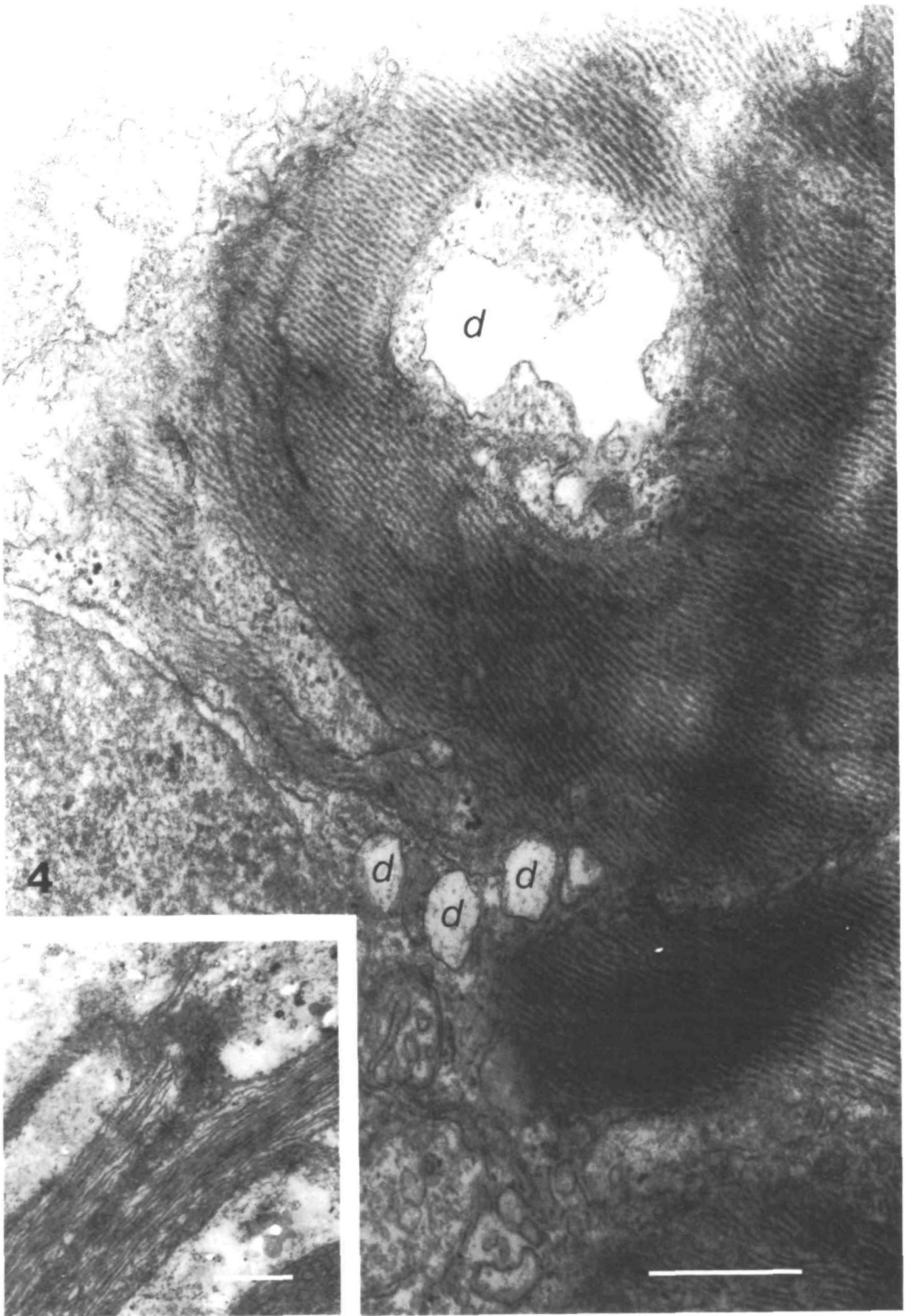
Fig. 4. Transmission electron micrograph of a transverse section through the capsule of the twisting muscle receptor (TMR). Branches of one of the receptor dendrites (d) are seen within the 'vacuolated string' tissue. The inset shows a longitudinal section through the TMR capsule. The banding pattern of the fibres seen in this section has a repeat of 64 nm, which is characteristic of collagen. Scale bars, 1·0 μm.

involved. This is clearly demonstrated by the absence of firing when imposed movements do not reach the fully protracted position (Fig. 5B, lower panel).

Units sensitive to swimmeret retraction: the TMR

These units were also found in the posterior branch of the first root, and were often recorded together with the protraction units (Fig. 5A). At least two units could be recognized with different, and variable sensitivities (Figs 5B, 6A). The smaller,
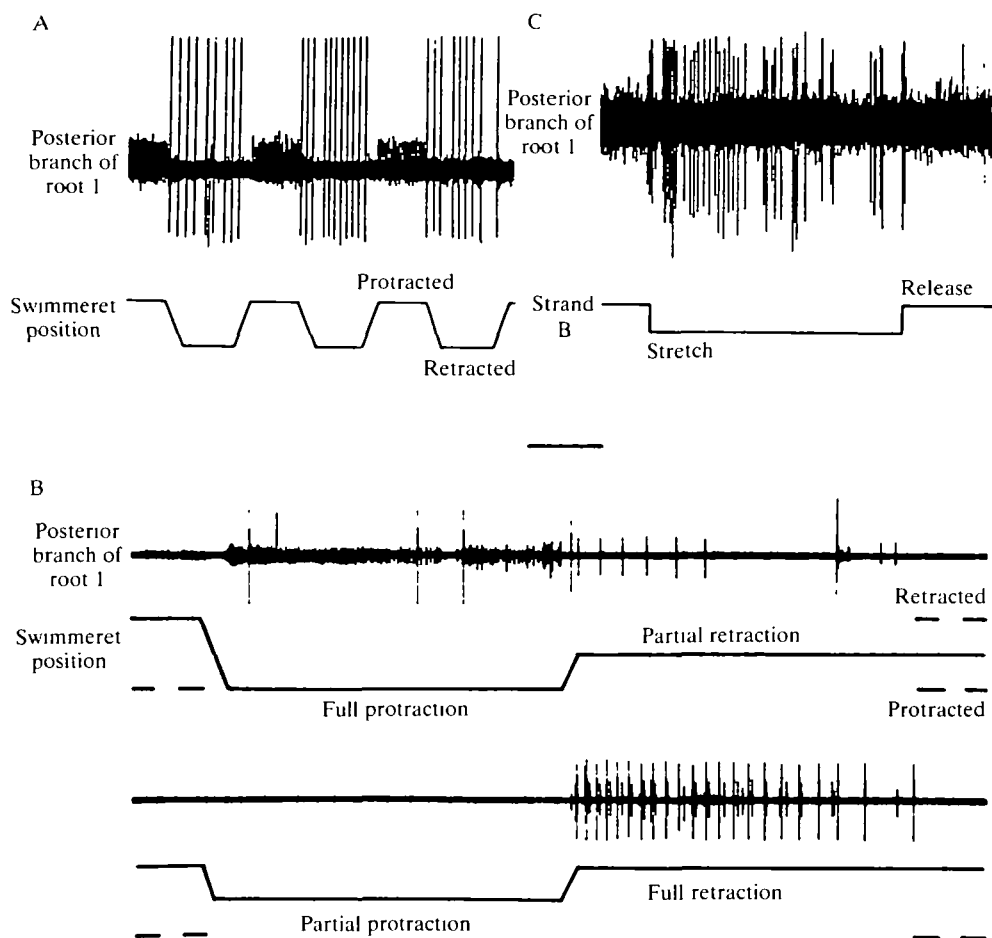

Fig. 5. Extracellular recordings from the distal cut end of the posterior branch of the first abdominal root. Results from three different preparations. (A) Responses of two sensory units to full protraction (downward ramps) and retraction (upward ramps) of the swimmeret. (B) Responses of protraction-sensitive units to a full protraction (upper panel) and to a partial protraction (lower panel) from the fully retracted position. Responses of retraction-sensitive units to a partial retraction from a fully protracted position (upper panel) and from a partially protracted position (lower panel). Broken lines on movement traces indicate fully protracted and retracted positions. (C) Responses of units to stretch (downward step) and release (upward step) of strand B. Time bar, 4 s (A); 2 s (B); 1 s (C).

A

B

Posterior branch
of root 1

Swimmeret
position   Protracted   Retracted

Posterior
branch
of root 1

2 s

Retracted

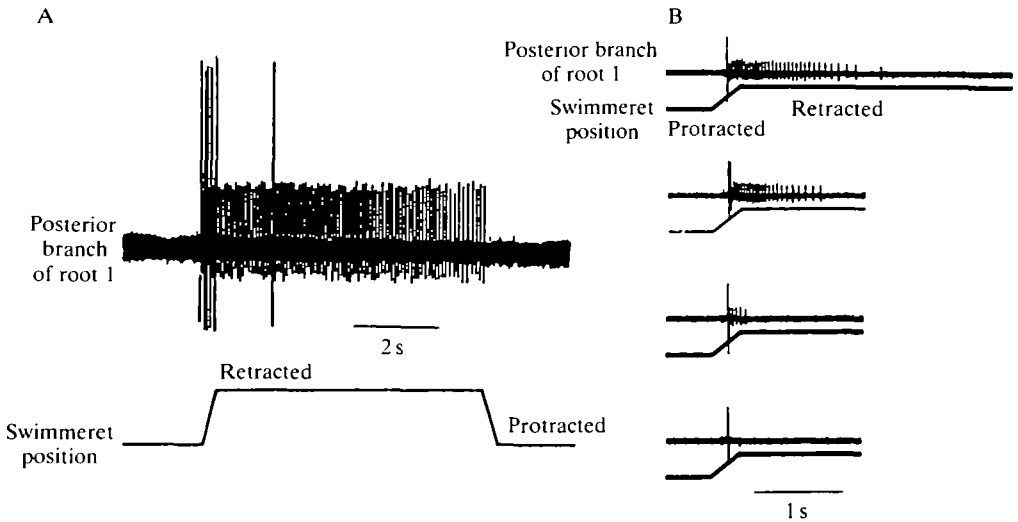Swimmeret
position   Protracted

1 s

Fig. 6. Responses of retraction-sensitive units in the posterior branch of root 1 to repeated stimulation. (A) An initial response to retraction of the swimmeret (upward ramp) demonstrating two units with different dynamic sensitivities. (B) A series of retraction movements. There is a reduction in the response of the smaller, more tonic unit with repetition, although the phasic unit persists. 4 s separates each record, during which the protraction movement was imposed. No response occurred to these intervening protractions.

more tonic, unit responded weakly to partial retraction, and more strongly to full retraction. The larger, more phasic, unit showed maintained discharge only at the fully retracted position (Fig. 5B). Over a number of cycles of stimulation the smaller unit showed a strong reaction to the first retraction, which became progressively weaker over subsequent cycles (Fig. 6B). The larger unit in this case was strongly phasic. This variability of the response was demonstrated in a number of other ways. A mechanical stimulus applied with a blunt seeker to the overlying membrane produced a large burst of activity in retraction-sensitive units, which was further enhanced by an imposed retraction and subsequently inhibited by an imposed protraction of the swimmeret (Fig. 7A). Further repetitions of the imposed movements produced a smaller response on the second retraction, and no response at all on the third. A similar result, clearly involving two units, was obtained when a sharp squeeze to the ipsilateral uropod preceded (by 20 s) imposed swimmeret movements (Fig. 7B).

Such diverse mechanosensory stimuli most probably act by producing reflex muscle contraction in the swimmeret. These results therefore suggest that retraction-sensitive units are associated with some muscle system whose state of contraction affects the level of the sensory response. The anatomical relationship of the TMR to M10 makes it an obvious candidate for such modifiable responsiveness, and this was tested physiologically by applying direct stretch and release to M10. Fig. 8A,B shows typical records from such experiments, which demonstrate that the release of M10 from stretch activates a unit with characteristics very similar to those seen on
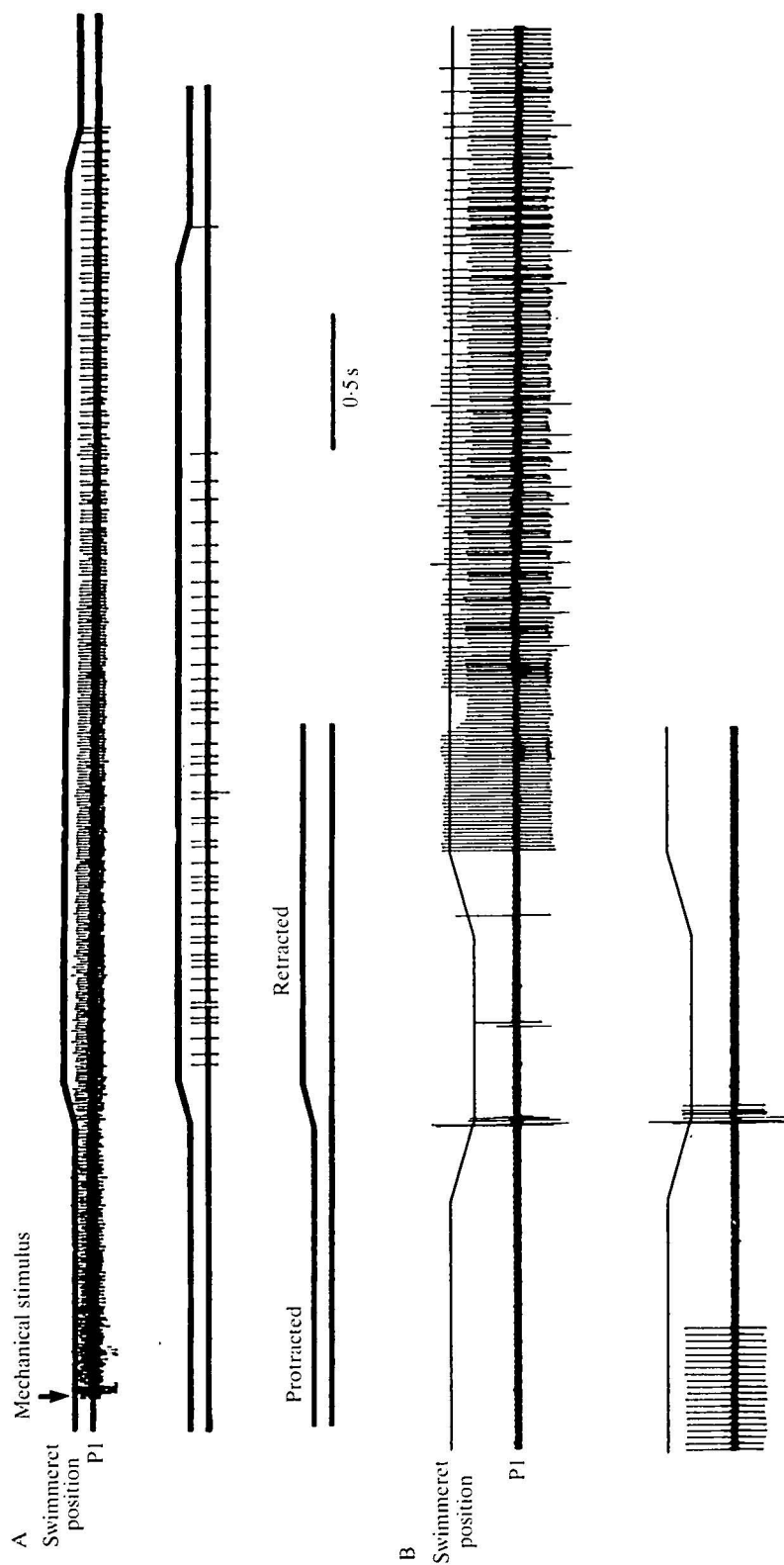
Fig. 7. Responses of retraction-sensitive units in the distal branch, P1, of the posterior root to swimmeret retraction following mechanosensory stimulation. (A) A single unit discharges strongly when the arthrodial membrane of the swimmeret basal joint overlying M10 is depressed (arrow), and subsequently shows a decreasing strength of response to repeated retraction movements (upward ramps). (B) Response to retraction movements of the swimmeret 20 s after a squeeze of the ipsilateral uropod. An intense response occurs to the first retraction (upward ramp), but no response to the second.
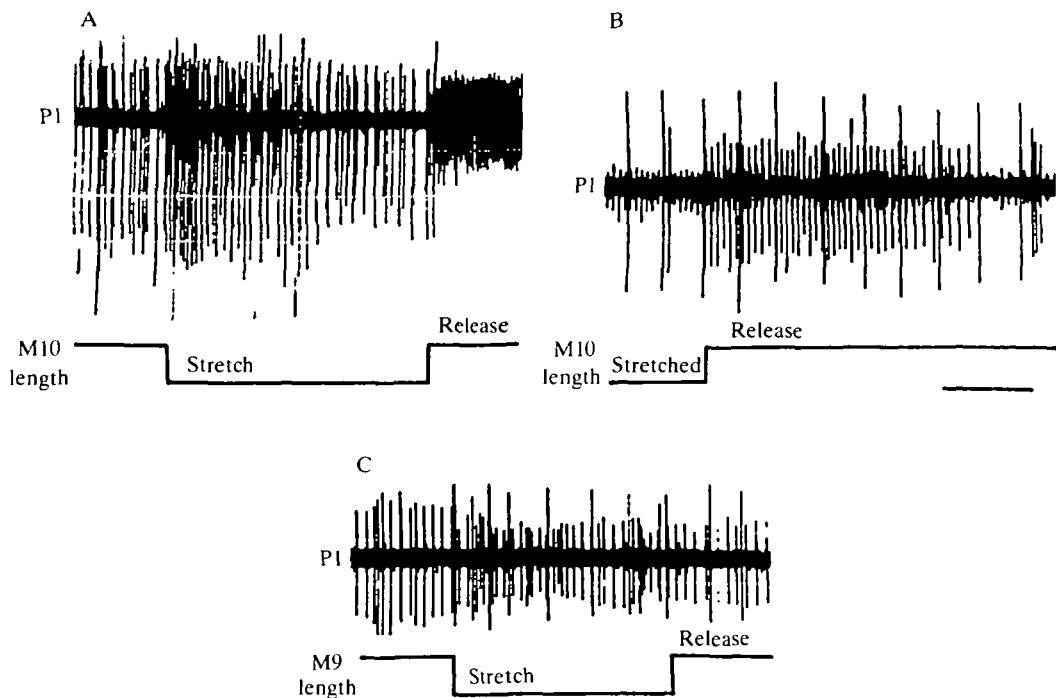
Fig. 8. Responses in the distal branch, P1, of the posterior root to applied stretch (downward step) and release (upward step) of the twisting muscles; A,B, M10; C, M9. Stretches of both muscles, which are equivalent to protractions, elicit activity in several units, but the response of a release-sensitive unit is specific to M10. Time bar, 1 s (A); 0·6 s (B); 2 s (C).

swimmeret retraction. Since similar stimuli applied to M9 or to any other swimmeret muscle were ineffective in activating such units (Fig. 8C), we conclude that they originate from the TMR associated with M10. Stretching M10 also, on occasion, produced sustained excitation in clearly different units (Fig. 8A), similar to the effect of strand B stretch (Fig. 5C). This is not an inconsistent finding since there is some mechanical linkage between this muscle and strand B.

Reflex effects of strand B stimulation

Reflex effects of strand B stimulation were investigated by recording motor activity in branches of the first swimmeret root, and intracellular activity in particular swimmeret muscles. The most convenient muscle to study was M9, due to its tonic activation by at least two units in the absence of swimmeret beating. All other powerstroke muscles are silent under these conditions (Neil & Miyan, 1986). M9 was therefore used as a representative, if not completely typical, member of the powerstroke muscle group.

With the animal in an inverted position, stretches to strand B caused a frequency increase in units to M9 (Fig. 9A), indicating a negative feedback reflex effect. When the animal was tilted about its longitudinal axis to elevate firing levels in M9 (Neil & Miyan, 1986), stretching strand B further increased discharges, and in many cases

induced a pattern of cyclical bursting of EPSPs in the recorded muscle (Fig. 9B). Bursting persisted for as long as the strand was stretched, and upon release the activity subsided abruptly to levels below that recorded prior to the stretch stimulus. A similar bursting frequency was observed in M9 of the monitored swimmeret when beating occurred spontaneously in adjacent swimmerets (Fig. 9C). This occurred even if the swimmeret was held in a fully retracted position, unloading strand B. It seems likely that this bursting in M9, induced by strand B stretch, reflects the rhythmical output of the swimmeret CPG. The main powerstroke (PS) and return-stroke (RS) muscles must also be activated under these conditions, since M9 bursting is only expressed in conjunction with beating activity (Neil & Miyan, 1986).

Blocking the movement of the swimmerets at different positions provided additional information about the influence of strand B receptors on swimmeret beat parameters. Animals were tilted to induce swimmeret beating, and motor activity was recorded in M13, a phasic member of the powerstroke muscle group. Blocking one swimmeret did not suppress the metachronal activity, but caused systematic changes in certain parameters of its beat (Fig. 10). Blocking in the fully retracted position caused a significant reduction in the burst duration and number of spikes in M13; burst period, however, was only slightly affected. Blocking in the fully protracted position had, by contrast, no clear effect on beat parameters.

Intersegmental effects were studied in a similar way, by recording myograms from M13s of three adjacent ipsilateral swimmerets while blocking the middle one at
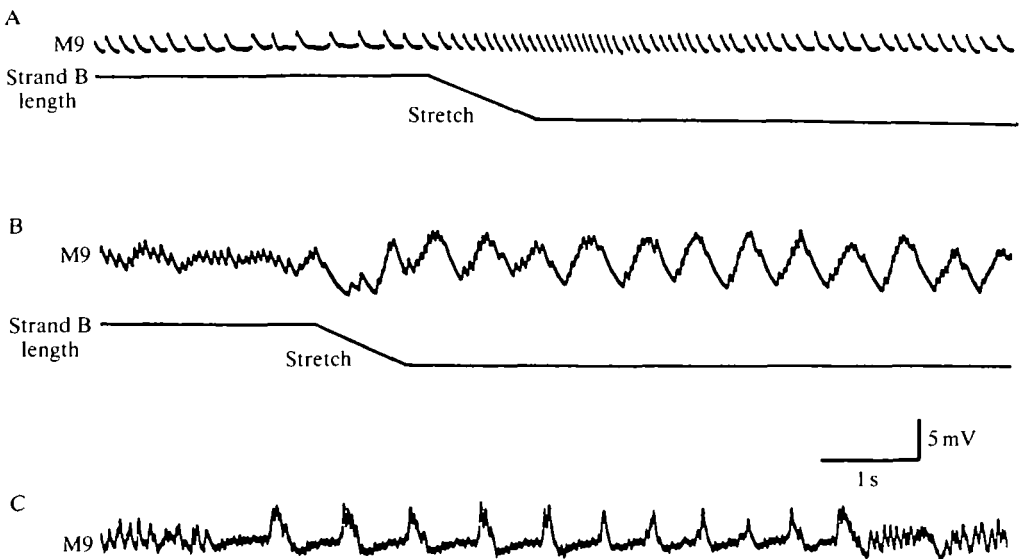

Fig. 9. Intracellular recordings from fibres of M9. (A) In an inverted animal, stretch of strand B (downward ramp) produces a transient frequency increase in the EPSPs. (B) Following a side-up tilt of the body, which induces a high frequency of EPSPs in the recorded M9 fibre, stretch of strand B induces a bursting pattern which has a frequency of approximately 2 Hz. (C) In an inverted animal, during a period of spontaneous beating in adjacent swimmerets, bursts of EPSPs occur in the recorded M9 fibre which, although less intense than in B, have a similar frequency.
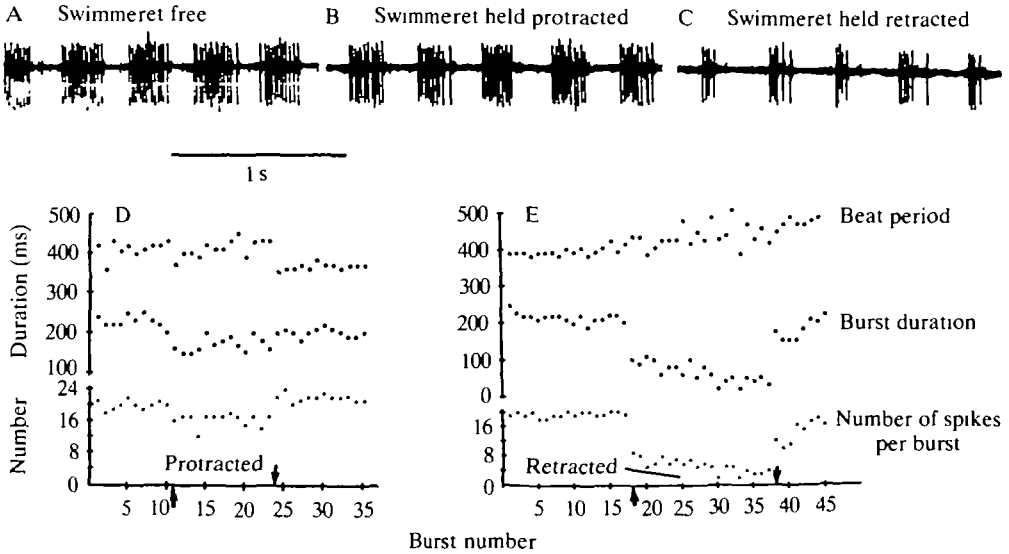
Fig. 10. Effects of blocking swimmeret movement on the activity of M13 in beating induced by tilting the animal to the right side up. Myographic records obtained from the third right swimmeret while it was unrestrained (A), held in the fully protracted position (B) and held in the fully retracted position (C). All other swimmerets continued to beat during these procedures. D and E show plots of beat period (upper), burst duration (middle) and number of spikes per burst (lower) during periods of restraint (upward arrow, held; downward arrow, release). (D) Held fully protracted; (E) held fully retracted.

different positions (Fig. 11A). Compared with the free-moving condition, blocking a swimmeret in the fully retracted position resulted in a reduction in burst duration in all monitored swimmerets. The intersegmental effect was directed more powerfully to the posterior, and resulted in a later onset of the powerstroke in swimmeret 4 relative to 3. Effects on the anterior swimmeret 2 were less marked. Blocking a swimmeret in a fully protracted position had no measurable effect on neighbouring appendages. Intracellular recording of M9 activity during imposed movements of the adjacent posterior swimmeret, showed an inhibition on retraction and an excitatory effect on protraction (Fig. 11B).

Reflex effects of TMR stimulation

Reflex effects of TMR stimulation were studied by recording from different muscles while stretch and release movements were applied to M10. No reflex effects to the adequate stimulus of M10 release were detected in any of the swimmeret muscles, except for M9. However, constraints of experimental method prevented records being obtained from M10 itself. To measure the direction of impulse traffic, simultaneous recordings were made from two different points along the posterior nerve root which supplies the lateral twisting muscles. The characteristic tonic activity of two units known to supply M9 (Neil & Miyan, 1986) is seen in the more proximal record (lower traces in Fig. 12). The larger unit (L), but not the smaller

unit (S), shows a phasotonic response to M10 release with a time course which closely matches that of the TMR sensory response (see Figs 6, 8). A similar excitation of an otherwise silent unit is seen in the more distal record (upper trace in Fig. 12). On the basis of its timing relationships to the M9 motor unit it is likely to be another motor unit, rather than a sensory unit, and expanded records demonstrate that these two units are tightly, but not completely coupled (Fig. 12). From the known firing relationships between units to the twisting muscles (Neil & Miyan, 1986), it seems probable that this second, distally-recorded unit is a motoneurone to M10 itself. There is thus a positive feedback excitation onto the twisting muscles during retraction, which may be mediated by the TMR.

Species comparison

Amongst published data there are a number of accounts of swimmeret anatomy, in particular for the hermit crab, *Pagurus pollicarus* (Bent & Chapple, 1977*a,b*), the lobster, *Homarus americanus* (Davis, 1968, 1969*b*) and the crayfish, *Pacifastacus leniusculus* (Heitler, 1982). The musculature responsible for the twisting of the swimmeret (as identified in lobsters) is shown for three species in Fig. 13. In the hermit crab, most of the basipodite muscles are absent, and there is no coxal receptor system. The remaining muscle acts to maintain a medial twist, allowing the swimmerets to beat within the shell, for the purposes of aeration. In the crayfish swimmeret the basipodite muscles are reduced, and M10 is absent. There is a two-point hinge joint at the basipodite (compared with the universal joint in lobsters)
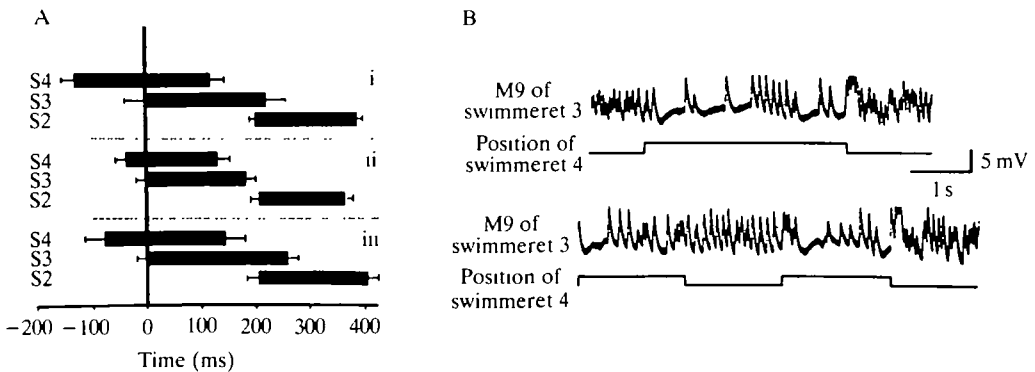

Fig. 11. (A) Effect of blocking a swimmeret on the beating activity of adjacent swimmerets. Data obtained from myograms of M13 activity in swimmerets 2, 3 and 4 of the right side, and expressed as burst duration averaged over 20 consecutive cycles of beating. Timing is expressed relative to the onset of the burst in swimmeret 3. Horizontal lines show standard deviations. (i) All swimmerets unrestrained. (ii) Swimmerets 3 held in fully retracted position. There is a clear effect on the onset and duration of activity in swimmeret 4. (iii) Swimmeret 3 held in fully protracted position. There is an increase in the burst duration in the held swimmeret (cf. Fig. 10), but no clear intersegmental effects. (B) Intracellular recording from M9 of the third right swimmeret showing a response to imposed retraction movements (upward steps of movement trace) and protraction movements (downward steps) of the fourth right swimmeret. Retraction of the posterior swimmeret produces an inhibition of activity.
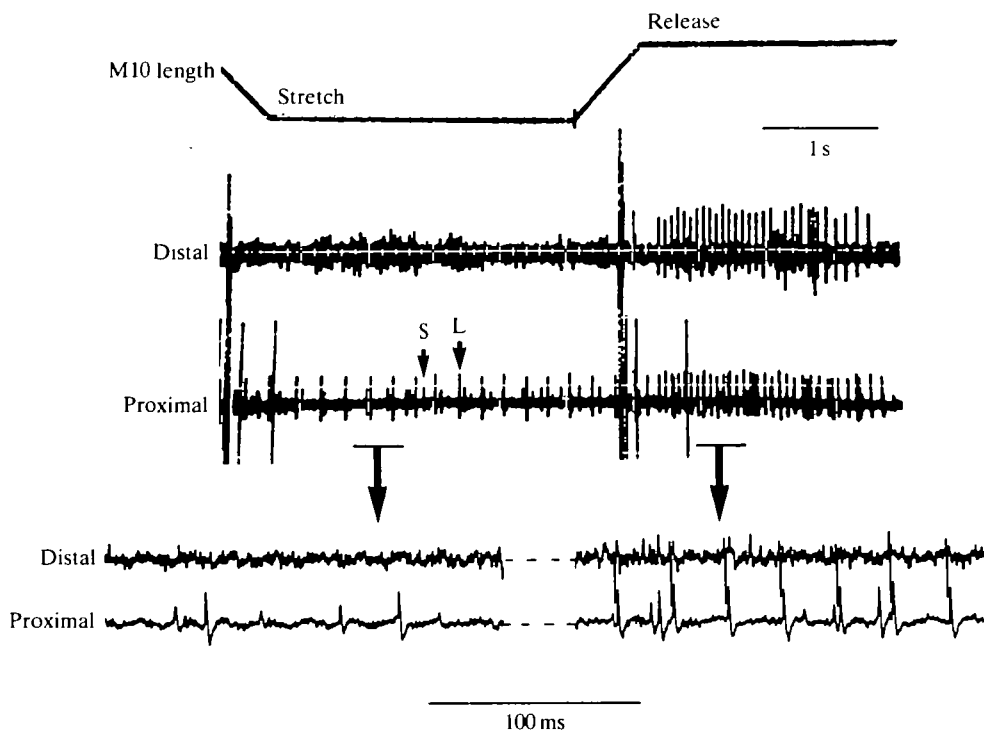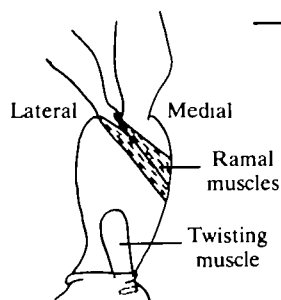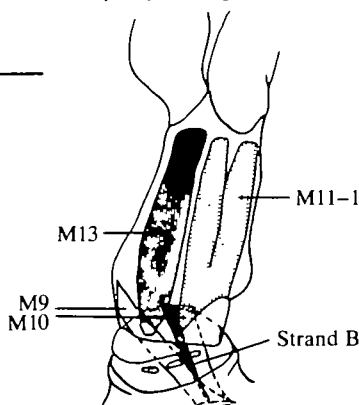
Fig. 12. Extracellular suction-electrode recordings at distal and proximal points along the posterior branch of the first abdominal nerve root, during stretch (downward deflection of movement trace) and release of M10. The proximal record contains two spontaneously active units (S and L) which are most probably those known to innervate M9 (Neil & Miyan, 1986). Expanded records (from sections marked with arrows) show the firing relationships between the units in the proximal and distal recordings.
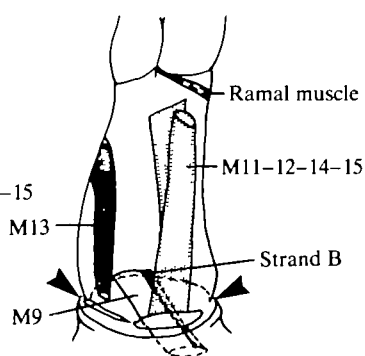
Fig. 13. Comparison of swimmeret musculature in three species of decapod crustacean. (A) The hermit crab *Pagurus pollicarus* (from Bent & Chapple, 1977a). (B) The lobster *Nephrops norvegicus*. (C) The crayfish *Astacus leptodactylus*. The twisting muscles in the hermit crab and crayfish are reduced compared to the lobster. The basipodite of the crayfish is constrained from rotating laterally by a two-point articulation (arrowheads). Scale bar, 2 mm (A,C); 5 mm (B).

which restricts lateral twisting movements. The righting reaction of the crayfish to body tilt is generated primarily by rotation of the whole abdomen at its articulation with the thorax (Suzuki & Hisada, 1979).

Heitler (1982) has described the receptor systems in the swimmeret of the crayfish *Pacifastacus leniusculus* and reports that they differ in several respects from those of the lobster, *Homarus americanus* (Davis, 1969*b*). In an attempt to ascertain whether differences between the anatomy of the lobster swimmeret system and that reported for crayfish (Heitler, 1982) are indeed real, and not a function of staining procedures, we have examined the crayfish *Astacus leptodactylus*, using the same staining techniques as for the lobsters *Nephrops* and *Homarus gammarus*.

The main receptor strands are arranged differently in the two groups, strand B being divided into distinct S1 and S2 branches in the crayfish, whereas the lobsters possess only the equivalent of the S1 branch (Fig. 14A). In crayfish, S1 will be stretched by retraction and, as a result of its suspension *via* S2, its distal portion will also be stretched during protraction. In the lobsters, strand B is stretched only by protraction movements.

The innervation of strand B also shows striking differences. Clearly stained in all our crayfish preparations were two large (60 μm diameter) receptor fibres which ran from central branch points to both ends of the S1 strand (Fig. 15), essentially as described by Heitler (1982) for *Pacifastacus leniusculus*. No comparable structures were ever found in the lobsters. On the other hand, no evidence was found in *Astacus* for a TMR associated with the twisting muscle M10, although this routinely stained in lobster preparations.

Study of the central anatomy of *Astacus* has confirmed the finding of Heitler (1982) that crayfish strand B fibres have central cell bodies. Differential cobalt/nickel staining of the anterior and posterior roots reveals that, after entering into the anterior root, one fibre passes to a soma in association with the PS motoneurone somata while the other remains with the anterior (RS) group (Fig. 14B). There are very few contralateral arborizations or intersegmental projections. The two sensory cells of the lobster TMR share this feature of a central location, but differ in having clear contralateral and intersegmental projections (Fig. 1C,D). They also support spikes (Figs 5–7), in contrast to the non-spiking properties of the crayfish receptor cells described by Heitler (1982).

DISCUSSION

Proprioceptive homologies

There is accumulating evidence from a number of decapod crustacean species that proprioceptors with central somata form a homologous segmental series in the set of biramous appendages. These have now been described in the second maxilla of lobsters, as the oval organ of the scaphognathite (Pasztor, 1969, 1979), in the walking legs of brachyurans and macrurans, as the TCMRO (Bush, 1976, 1981), in the swimmerets, as the crayfish NSSR (Heitler, 1982) and nephropid lobster TMR (present study), and in the uropods of the anomuran crab *Emerita* (Paul, 1972,
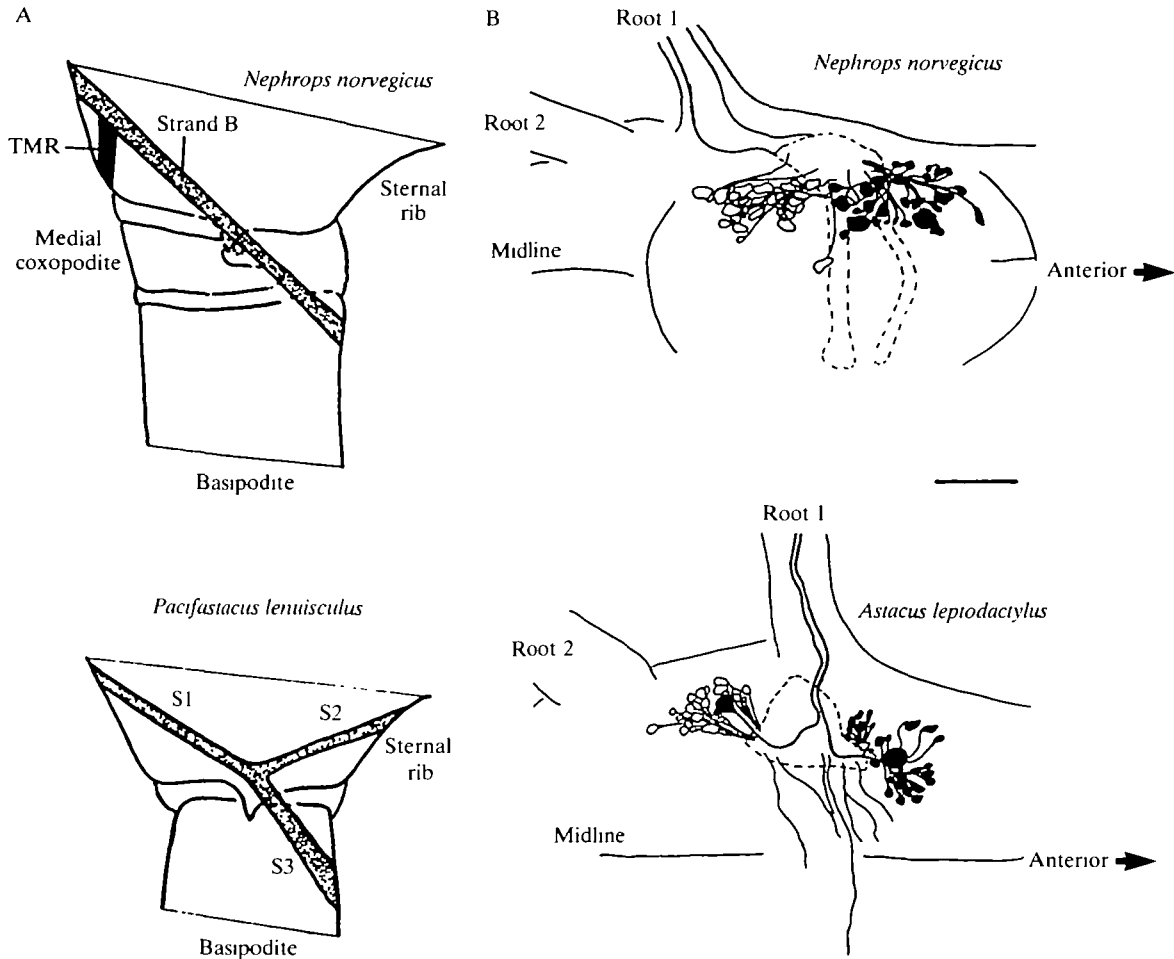
Fig. 14. (A) Comparison of strand B swimmeret receptors in the lobster *Nephrops norvegicus* (above) with the probably homologous receptor strands (S1, S2) in the crayfish *Pacifastacus lenuisculus* (below: after Heitler, 1982, with the permission of the author). The swimmeret is viewed from the medial aspect. The position of the twisting muscle receptor (TMR) in the lobster is also indicated, although it passes behind strand B to insert on M10. Scale bar, 5 mm for lobster; 2 mm for crayfish. (B) Comparison of central anatomy of swimmeret neurones in the lobster *Nephrops norvegicus* (above) and the crayfish *Astacus leptodactylus* (below). *Camera lucida* drawings, viewed ventrally, of differential cobalt/nickel backfills of the anterior branch (filled cell outlines) and posterior branch (open cell outlines) of the first right abdominal root. In the lobster, the two groups of somata are arranged closely together, and there are two large transganglionic tracts (dashed outlines). In the crayfish, there are two widely-spaced groups of somata, and very few projections across the ganglion. The two largest somata entering from the anterior root lie separately within the two main groups. These are most probably the somata of the non-spiking receptors (cf. Heitler, 1982). Scale bar: 300 μm for the lobster; 200 μm for the crayfish.

1976), the squat lobster *Galathea* (Maitland, Laverack & Heitler, 1982) and the lobster *Homarus gammarus* (M. S. Laverack, in preparation). However, an interesting divergence in physiological properties appears to have developed. The

lobster TMR bears spikes, while others conduct decrementally, e.g. crayfish swimmeret NSSR, crab TCMRO, uropod receptors in *Emerita* and squat lobsters. The scaphognathite oval organ represents an intermediate category, since it is able to conduct in either mode (Pasztor & Bush, 1982). The functional advantages of non-spiking transmission have been discussed in terms of its graded output signal, which obviates the need for encoding and decoding a frequency code of spikes (Bush, 1981; Heitler, 1983). Limitations exist in terms of the decrement of the signal [although it is transmitted over many millimetres in TCMRO (Bush, 1976)], the loss of high-frequency responsiveness, and the susceptibility to contamination with 'noise' of various kinds. In considering what factors determine the mode of transmission adopted by these receptors, it is particularly interesting to find in the swimmerets of two such closely related species as lobsters and crayfish equivalent, and probably homologous, receptors which share basic common features of position and gross morphological organization, and yet differ in their method of coding and the reflex effects produced. It remains to be determined if there is a causal relationship between the mode of transmission adopted and the proprioceptive function performed.

Proprioceptive monitoring of power- and returnstrokes

In the lobster *Homarus americanus*, Davis (1969*b*) reported two distinct proprioceptive effects on the swimmeret motor programme during retraction: a positive feedback from ramal setae which reinforces the PS movement, and an excitatory reflex from coxal proprioceptors simultaneously onto PS and RS motoneurones. Cattaert (1984) has demonstrated a similar phase-specific reflex from coxal proprioceptors to the opener muscle of the swimmeret rami in *Homarus*. We have found a specific negative feedback excitation of M9 during strand B stretch (which occurs on protraction), which represents a resistance reflex. This is most probably mediated by the bipolar cells associated with strand B, which are conventional spiking neurones, are normally silent, and are unidirectionally sensitive (Fig. 12A). In the beating swimmeret of crayfish, negative feedback resistance reflexes from the NSSR act to sharpen the transitions at the ends of both power- and returnstrokes, and provide amplitude control (Heitler, 1982, 1986).

Our results also demonstrate more general effects of stretching strand B or blocking of the basipodite movement on the intensity of swimmeret beating (Figs 10, 11). These effects are more consistent than the rather diffuse and variable perturbations to swimmeret beating found by West, Jacobs & Mulloney (1979) in the crayfish. The finding that strand B stretch can initiate rhythmic motor activity with certain characteristics of the pattern in voluntary beating (Fig. 12) suggests that proprioceptive input has access to the swimmeret oscillator circuitry, and has powerful effects upon it.

Proprioceptive monitoring of lateral beating

The lobster TMR proprioceptor, which is probably homologous to the crayfish NSSR, is weakly stimulated by retraction of the swimmeret, but has no reflex action on the majority of swimmeret motoneurones. It therefore does not contribute to

15 A

B
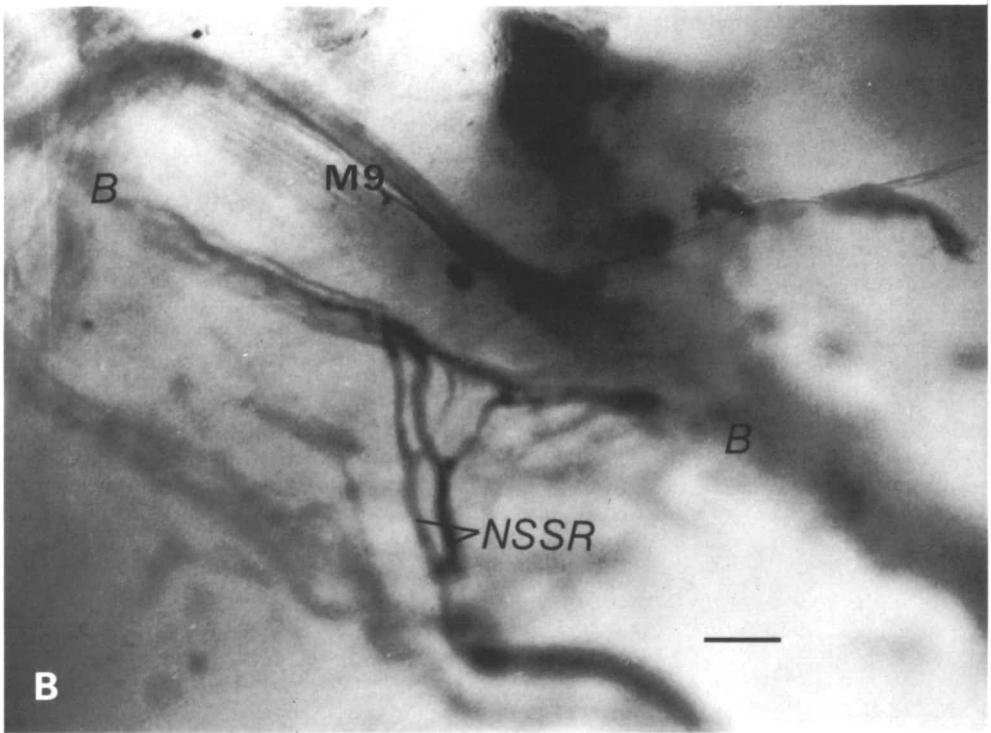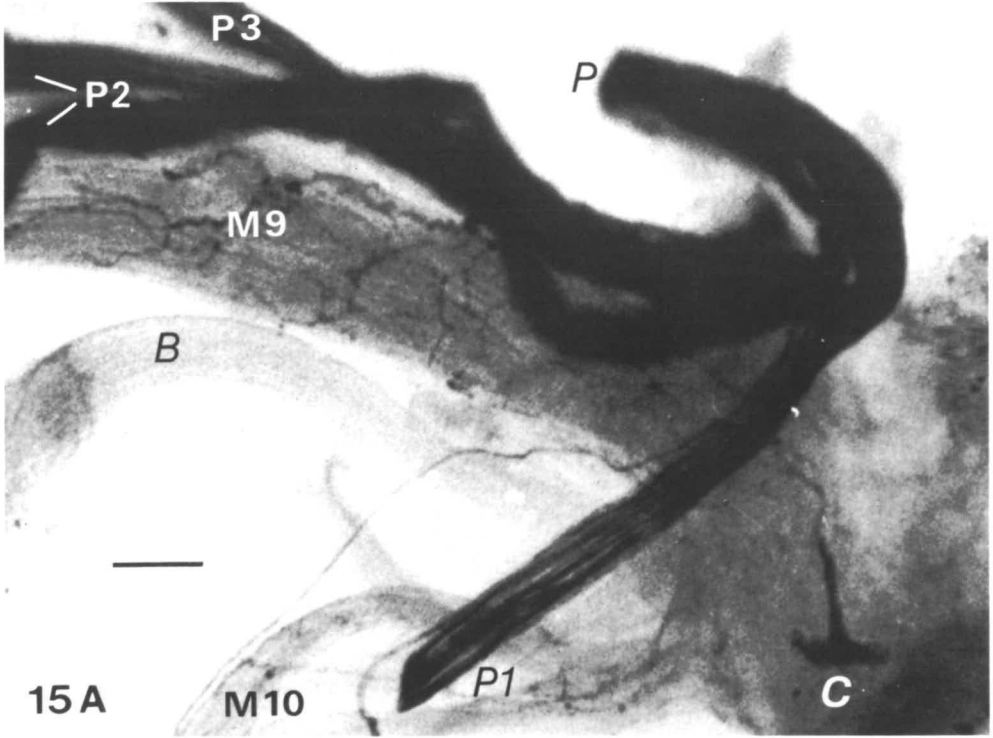
normal, rearward beating but is only brought into play when the tilt of the body, as detected by the statocysts, dictates lateral beating (Neil & Miyan, 1986). The TMR has a particular association with the twisting muscle M10, and we have shown that on muscle shortening the TMR elicits contraction specifically in the lateral twisting muscles. Since this results in further muscle shortening, the reflex involves positive feedback, and its function seems to be to ensure that lateral twisting is always to the maximum extent permitted by the anatomy of the joint. Direct measurements of this movement during body tilts confirm this prediction (D. M. Neil, unpublished observations), rather than a proportionality between the extent of twist and tilt angle. Small deviations from the upright detected by the statocysts therefore recruit the swimmeret righting reaction in its fullest form. In this respect this lobster righting reaction differs from the steering reactions in locust flight (Möhl, 1985*a,b,c*; Reichert & Rowell, 1985) and dipteran flight (Heide, 1979; Miyan & Ewing, 1984), which exhibit variable output. It remains to be determined if these differences are a function of the different media through which the locomotion is performed, or of the different requirements of steering as opposed to righting manoeuvres. Whatever the case, it is clear that, in addition to the combined effects of statocyst interneurones and the swimmeret oscillator on the lateral twisting muscles (Neil & Miyan, 1986), the TMR positive feedback makes an essential contribution to the formation of the motor pattern for lateral swimmeret beating.

The role of proprioceptive feedback

All these forms of proprioceptive modulation demonstrate the potentially important role played by sensory feedback in forming the natural pattern of swimmeret beating. This aspect of control has previously received little attention, since the CPGs in a deafferented preparation have been found to produce rhythmic output in the swimmeret motoneurones (Ikeda & Wiersma, 1964) with coordination both bilaterally (Heitler, 1981) and intersegmentally (Stein, 1971, 1974). Although the CPG appears to set the basic rhythm and dictate its frequency, the proprioceptive modulation will serve to adapt the precise timing and amplitude of the movements to the functional demands made upon the system (Heitler, 1986). Clear evidence for such an integration of sensory input into the pattern-generating system has been found in other arthropod locomotory systems, notably those for walking and flying. The TCMRO of the crayfish produces positive feedback assistance reflexes onto retractor motoneurones during the stance phase of walking, and negative feedback resistance reflexes onto protractor motoneurones during the swing phase (Sillar & Skorupski, 1985). In addition, imposed movements of the TCMRO entrain the

Fig. 15. Peripheral cobalt fills of swimmeret receptors in the lobster *Nephrops norvegicus* (A) and the crayfish *Astacus leptodactylus* (B). The most striking difference is the apparent lack of innervation to strand B in the lobster, in contrast to the clear innervation seen in the crayfish. In A, *c*, twister muscle receptor capsule; *B*, strand B; *P*, posterior trunk of the first abdominal root. This nerve trunk divides to form branch P1 to the main powerstroke muscles, branch P2 to the basipodite muscles and branch P3 to the rami (see Neil & Miyan, 1986). Scale bar, 120 μm. In B, *NSSR*, non-spiking stretch receptor, which innervates strand B (the ends of which are marked *B*). Scale bar, 300 μm.

centrally-generated rhythm, and K. T. Sillar, R. C. Elson, P. Skorupski & B. M. H. Bush (in preparation) imply that this receptor is properly regarded as an element in the pattern-generating circuit. Similar conclusions have been reached by Bässler (1985) regarding the status of sensory feedback in stick insect locomotion, and Pearson (1985) has further challenged conventional views by concluding that a CPG for walking may not exist as a functional unit in the intact insect. Even where CPGs are well documented, as for locust flight (Wilson, 1961; Kutsch, 1974; Robertson & Pearson, 1982), the rhythms produced by isolated nervous systems differ significantly from the normal patterns in intact animals, and Pearson (1985) suggests that the characteristics of the CPG are fundamentally altered by sensory inputs, primarily those from the wing stretch receptor (Pearson, Reye & Robertson, 1983; Möhl, 1985a,b,c). It therefore seems appropriate to re-examine in detail the manner in which the macruran swimmeret beating is influenced by the various sensory inputs which are now known to act upon it.

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