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MODULATION OF REFLEX AND CENTRALLY GENERATED MOTOR ACTIVITY BY TENSION RECEPTOR AFFERENCE IN *CANCER PAGURUS* (DECAPODA, BRACHYURA)

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Abstract—1. Electrical stimulation of the tension receptor nerve of the flexor of the carpus inhibits not only resistance reflex activity in the flexor motor nerve but also motor activity evoked by a variety of other external stimuli.

2. Not all units in the flexor motor nerve respond in the same way to a given level of flexor tension nerve stimulation.

3. A resistance reflex to imposed extension occurs as a result of MC1 chordotonal organ input alone. A modified resistance reflex to extension also occurs in the presence of isometric contraction of the flexor muscle produced by centrally generated activity in the flexor motor nerve.

4. The resistance reflex present when there is no centrally generated activity in the flexor motor nerve is abolished by cutting the MC1 nerve; at least part of the reflex which occurs during centrally generated activity remains after the MC1 nerve has been cut. This remaining part of the reflex is abolished when the tension receptor nerve is cut.

5. These results indicate that the sign of the effect that tension receptor afference has on the flexor motor neurons depends on whether the neurons are activated reflexly or centrally. Ways in which this could occur are discussed.

INTRODUCTION

There is now a considerable amount of information on the structure and function of the sense organs in the limbs of decapod crustaceans. As the whole area has been reviewed recently it will not be considered further here (Mill, 1976). Of late, there has been increasing interest in the role that the afference from some of the sense organs plays in modulating the motor output to the limbs. The work described here deals with modulation of motor output by chordotonal organs and apodeme tension receptors.

Prosser (1935) and Eckert (1959) demonstrated reflex motor responses to imposed movements of crustacean limbs. Bush (1962) showed that the chordotonal organs are responsible for these reflexes which he termed "resistance reflexes" because their sign is such that they resist imposed movements. Subsequent work (Bush, 1963; Cohen, 1963; Evoy & Cohen, 1969; Macmillan *et al.*, 1981; Spirito, 1970; Spirito *et al.*, 1972; Wales *et al.*, 1970) has elaborated further the characteristics of the resistance reflexes at a variety of joints and involving several different chordotonal organs and the myochordotonal organ. Analysis of resistance reflex activity during patterned motor activity suggests that they stabilise the joint angles of the resting limb but are suppressed and do not have an important functional role during locomotion (Barnes *et al.*, 1972; Cohen, 1965; Evoy & Cohen, 1969; Evoy & Fournier, 1973; Fournier & Evoy, 1973).

The receptors that lie in series with the limb muscles and monitor muscle tension (Macmillan & Dando, 1972; Macmillan, 1976; Parsons, 1980) have been found to modify resistance reflexes (Clarac &

Dando, 1973; Dando & Macmillan, 1973; Parsons, 1981).

The work here examines further the effect of tension receptor afference on both non-reflex motor neuron activity and also on resistance reflexes.

MATERIALS AND METHODS

Specimens of the crab *Cancer pagurus* of 12–20 cm carapace width were collected from around St. Andrews and stored in fresh running sea water. The experimental animal was induced to autotomise its chelipeds and the body was then secured to a perspex holder with elastic bands. To keep the animal damp and cool, paper towels moistened with sea water were laid over the carapace and ice packs placed loosely in contact with these. The first pereopod on the right side was firmly secured to a perspex platform with its anterior surface upwards. The apodeme of the levator muscle was severed to reduce the possibility of autotomy. A bath was constructed around the meropodite of the leg with plasticine and that portion of the anterior wall of the meropodite lying within the confines of the bath was removed with a dental drill. The bath was perfused with cold crab saline (Macmillan & Dando, 1972). Following dissection, the exposed nerves were lifted onto bipolar, platinum, hook electrodes and the level of the saline lowered until the nerves were clear of the surface. Drying was prevented by covering the nerves with a mixture of vaseline and glycerine. Stimulating and recording methods were conventional, the activity being photographed directly from the oscilloscope. Controlled movements were applied to the carpus with a servo-motor driven by a waveform generator.

Anatomy and dissection

Characteristically, the crab *Cancer pagurus* rests on its ventral surface with its legs folded beside and partially beneath the carapace. In this position the broad, flat sur-

faces of the leg segments face approximately dorso-ventrally with respect to the body. Morphologically, however, the broad surfaces represent the anterior and posterior aspects of the legs, the dorsal and ventral surfaces being reduced to narrow, convex edges. Because of the rotation allowed at the most proximal joints, the morphologically anterior surface can face anywhere between anterior and dorsal with respect to the body. To avoid ambiguity in this description the morphological nomenclature will be adhered to irrespective of leg position.

The merus is the largest of the leg segments. Its interior is almost completely occupied by two large muscles, the flexor and extensor of the carpus. The extensor muscle originates over most of the inner surface of the anterior wall of the merus, its fibres attaching to an apodeme which lies antero-dorsally in the merus and inserts onto the proximal, dorsal edge of the carpus. The flexor originates on the posterior surface and part of the ventral edge of the merus and attaches to an apodeme which lies postero-ventrally and inserts onto the proximal, ventral edge of the carpus. There is a small accessory flexor muscle which originates ventrally in the proximal part of the merus just anterior to the flexor muscle and inserts, by means of a tendon running almost the entire length of the merus, onto the anterior edge of the flexor apodeme proximal to its insertion onto the carpus.

The main leg nerve and a blood vessel run together mid-ventrally between the two large muscles. To expose the main leg nerve, the extensor apodeme was severed close to its insertion and the muscle folded proximally. The blood vessel was dissected away from the nerve, some care being necessary to avoid damage to fine branches of the flexor motor nerve. Removal of small pieces of connective tissue associated with the nerve and blood vessel allowed access to the nerves from the two chordotonal organs associated

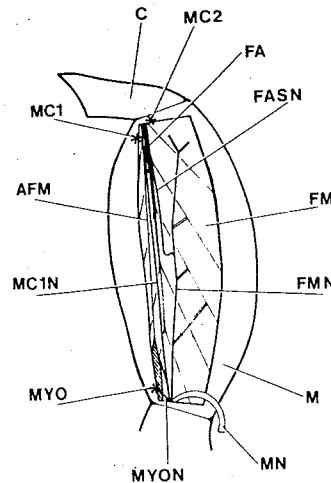


Fig. 1. Diagram of the merus (M) of the first pereiopod on the right side. Part of the anterior wall of the merus and the extensor muscle have been removed to show the underlying structures. The MC1 chordotonal organ is situated on the carpal end of the accessory flexor muscle (AFM) and its nerve (MC1N) runs anterior to the flexor apodeme (FA) along which the flexor apodeme sensory nerve (FASN) originates. AFM: Accessory flexor muscle; C: Carpus; FA: Flexor apodeme; FASN: Flexor apodeme sensory nerve; FM: Flexor muscle; FMN: Flexor motor nerve; M: Merus; MC1: position of MC chordotonal organ; MC1N: Nerve from MC1 chordotonal organ; MC2: Position of MC2 chordotonal organ; MN: Main leg nerve which has been cut and retracted proximally; MYO: Position of myochordotonal organ complex; MYON: Nerve to myochordotonal organ complex.

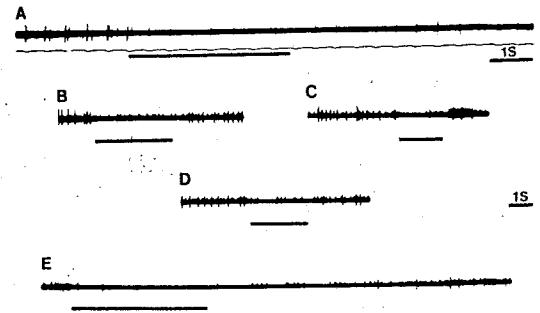


Fig. 2. Records made from the FMN. Stimulation of the FASN indicated by bar below trace. (A) Downward movement of trace corresponds to extension of carpus. Note that at this level of stimulation the larger unit was inhibited but not the smaller unit. Note also that the inhibition persisted for a period after the stimulation stopped. (B) Inhibition of FMN activity evoked by stroking the carapace. (C) Inhibition of FMN activity evoked by moving other legs. Note the apparent post-inhibitory rebound in the smaller unit. (D) Inhibition of FMN activity evoked by visual stimuli. (E) Inhibition of a low level of spontaneous background discharge in the FMN. Note that the inhibition is complete during the latter part of the FASN stimulation. Upper time marker refers to trace A, lower time marker to traces B-E.

with the joint (Bush, 1965a; Whitear, 1962), the nerve from the myochordotonal organ receptor (Clarac, 1968; Evoy & Cohen, 1969) and the flexor motor nerve (FMN) with the associated flexor apodeme sensory nerve (FASN) (Macmillan & Dando, 1972; Parsons, 1980). The position and orientation of these elements is shown in Fig. 1.

RESULTS

Clarac & Dando (1973) showed that stimulation of the FASN inhibits resistance reflexes and we confirmed that result with our preparation (Fig. 2A). If one is to consider the functional significance of these reflexes it is important to know whether the tension receptor afference inhibits resistance reflexes specifically. To test this, we evoked motor activity in the flexor motoneurons using a variety of different stimuli including stroking the carapace, moving other joints and legs and visual stimulation. The activity evoked by these stimuli usually involved at least two FMN units. At least one unit was invariably inhibited by FASN stimulation (Fig. 2B-D). Where more than one unit was firing, we often found a difference in the response to FASN stimulation with one unit inhibited and one not (Fig. 2B). In some cases there were also signs of post-inhibitory rebound (Fig. 2C). Sometimes a low level of background discharge was seen in the FMN in the absence of applied stimuli and this was also inhibited by FASN stimulation (Fig. 2E).

In all cases, the strength of the inhibition depended on the level of activity in the motoneurons. In some cases the inhibition was only partial (Fig. 2A) whereas in others the inhibition was almost complete (Fig. 2E).

Although negative feedback from the FASN to the FMN was the rule for reflexly evoked activity, we found exceptions which suggest that there is a difference between the effect of tension receptor input on reflexly evoked motor activity and motor activity pro-

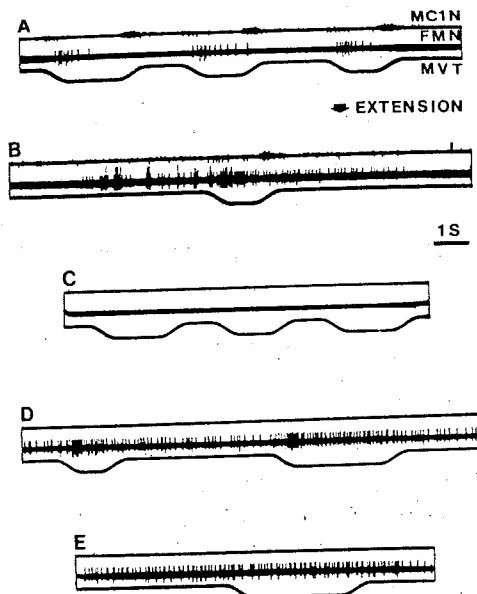


Fig. 3. (A) After cutting MC2N and MYON it was still possible to evoke a resistance reflex in the FMN to imposed extension of the carpus (movement trace, MVT, moves down during extension). (B) A modified resistance reflex was also evoked during spontaneously generated central activity in the FMN which increased the isometric tension in the flexor muscle. (C) Cutting the MC1N abolished the reflex in the passive animal. (D) Some resistance reflex activity to imposed extension was still present in the spontaneously active animal. (E) The resistance reflex to imposed extension in the active animal was abolished by cutting the FASN.

duced during self-activated, centrally patterned movements.

With the MC2N and the MYON nerves cut, a resistance reflex to imposed extensions was still present (Fig. 3A). This reflex was still discernable in the presence of centrally generated activity in the flexor motor nerve which would increase the isometric tension in the flexor muscle (Fig. 3B). There was increased background activity in the MC1N in this situation, presumably due to the effect of tension on MC1 (Macmillan *et al.*, 1981). Cutting the MC1N abolished the reflex in the non-active animal (Fig. 3C). In the active animal which was developing isometric flexor tension, some of the reflex response to an imposed extension still remained (Fig. 3D). We concluded that this part of the reflex was mediated by tension receptor afference because it was abolished when the FASN was cut (Fig. 3E).

DISCUSSION

Our findings that stimulation of the FASN inhibits FMN activity which is reflexly evoked by a variety of stimuli, suggests that the inhibition is occurring close to the motor side, perhaps even onto the motor-neurons themselves. In considering the implications of this result two limitations should be borne in mind. First, electrical stimulation of the FASN may represent an unphysiological input. Secondly, there are two possible sources supplying the flexor muscle and we cannot be sure that these nerves are recruited for the

resistance reflex or whether the tension receptor input affects them differently. Our finding, that different units of the FMN may be inhibited in differing degrees by the same FASN stimulus, suggests that this may have some functional significance. It is also possible that excitatory and inhibitory units are affected differently.

Our finding that tension receptor afference that is inhibitory to reflexly evoked motor activity can be excitatory during some centrally generated motor activity raises some interesting issues, but again, certain experimental limitations should be borne in mind. Inhibition by tension afference has only ever been demonstrated by electrical stimulation of the whole nerve and this is not exactly comparable with our physiological activation to reveal the positive feedback reflex. Furthermore, it should be remembered that the apodeme of the extensor muscle had been cut and this could alter the normal balance of sensory input found in the intact animal.

Bearing in mind these limitations, there are three ways in which the change of sign of the reflex could be accomplished. First, the tension receptors could be activated differently or different parts of them could be activated in the two different situations. This possibility seems most unlikely if one considers recent results for *Carcinus* (Parsons, 1980, 1981). These investigations found no evidence that the tension receptors code for specific motor units. The second possibility is that while the tension afference is the same in both cases it is being switched centrally under the influence of other forms of afference which are not the same in the two situations. There are examples of central switching controlled by proprioceptive organs in the locust walking system (Macmillan & Kien, 1981) and in the lobster swimmeret system (Neil & Miyan, pers. comm.), and it has also been shown recently that chordotonal organs may change their response characteristics with changes in the tension of related muscles (Macmillan *et al.*, 1981).

The final and most probable possibility is that the tension afference is the same in each situation but that part of the function of the interneurons of the central pattern generators is to redistribute the incoming signal and alter the gain of the signal onto succeeding elements. Evidence for modulation of this type can be found in several arthropod systems (Kien, 1976; Sandeman, 1969; Silvey & Sandeman, 1976).

Intracellular recordings will have to be made to determine which of the above mechanisms is operating in this system. The finding in its present preliminary form is significant however because a change in sign of the effect of tension afference is likely to be an important factor in load compensation and power output in the walking system.

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