

## REVERSAL OF A WALKING LEG REFLEX ELICITED BY A MUSCLE RECEPTOR

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### SUMMARY

Passive movement of the basal (thoracic-coxal) leg joint in the shore crab *Carcinus maenas* normally elicits a resistance reflex in the promotor and remotor motoneurons. Remotion of the joint excites promotor motoneurons and promotion excites remotor motoneurons. This reflex behaviour may reverse and become an assistance reflex, where movement of the joint excites the motoneurons innervating the muscle which would assist the passive movement. This reversal of reflex activity appears to be dependent upon the central state of activity of the animal.

### INTRODUCTION

Various studies of postural control and locomotion in vertebrate and invertebrate preparations have demonstrated that the integration of sensory information is subject to considerable modification when involved in centrally programmed movements. The most common type of reflex seen in crustacean preparations is the resistance reflex (Bush, 1962, 1963, 1965), where imposed movement of a joint causes the activation of motoneurons of an antagonistic muscle to resist the imposed movement. Several studies have also demonstrated that distributed or intersegmental reflexes operate in the same leg from one joint to another (Bush, 1962; Ayers & Davis, 1977; Bush, Vedel & Clarac, 1978; Clarac, Vedel & Bush, 1978). These studies have emphasized the complexity of proprioceptive reflexes and their role in the control of joint movement and position. Nevertheless these results appear contradictory if data from dissected preparations are compared to those from intact animals: only resistance reflexes are usually observed in the former case while more complex responses are recorded in the latter. It has been shown in the crab that resistance reflexes may be suppressed during normal spontaneous locomotor activity, although these reflexes are still active when there is a deviation from the intended movement pattern (Barnes, Spirito & Evoy, 1972). Resistance reflexes observed in intact crustacean preparations may also be completely blocked by spontaneous central activity (Bush *et al.* 1978). The present report describes a complete reversal of the reflex activity in the basal joint of the crab leg evoked by the thoracic-coxal muscle receptor organ (TCMRO).

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The TCMRO lies within the thorax at the base of each leg and is in parallel with the leg promotor muscle (Alexandrowicz, 1958). It consists of a long cylindrical bundle of muscle fibres enclosed in an elastic sheath, innervated by two large (50–70  $\mu\text{m}$ ) diameter and one smaller diameter sensory fibres at its proximal end and by two fine motor axons which run along the length of the muscle. The receptor organ attaches proximally on the medial rim of the exoskeletal septum separating thoracic compartments, and inserts distally into the coxal promotor tendon. It is stretched by remotion of the coxopodite and is shortened passively during promotion.

Passive remotion of the limb reflexly excites several promotor neurones in a typical resistance reflex (Bush & Roberts, 1968; Bush, 1977). In addition, passive promotion of the limb sometimes elicits a resistance reflex discharge in neurones innervating the antagonist remotor muscle. These reflexes are mediated by the TCMRO, which is the only stretch receptor in crabs responsive to movements of the thoracic-coxal (T–C) joint.

#### MATERIALS AND METHODS

All experiments were performed at room temperature on the posterior leg of the shore crab, *Carcinus maenas*. The chelipeds and legs were autotomized, the abdomen, carapace, viscera and brain were removed, and the ventral thorax pinned down in a dish containing crab saline. The saline composition was  $\text{Na}^+$ , 500;  $\text{K}^+$ , 12;  $\text{Mg}^{2+}$ , 20;  $\text{Ca}^{2+}$ , 12;  $\text{Cl}^-$ , 576 (mM): buffered with tris maleate to pH 7.2. The thoracic ganglion was perfused with aerated saline via a cannula inserted into the sternal artery. The nerves innervating the promotor and remotor muscles of the leg were cut and their activity monitored with polyethylene suction electrodes. All other nerves in this leg segment, with the exception of the TCMRO afferents, were cut. Passive movements of the coxa were made with an electromechanical puller (Pye-Ling) via a mechanical linkage to a pin glued to the coxa with Eastman 910 adhesive.

#### RESULTS

Passive remotion of the T–C joint normally evokes a resistance reflex, with promotor nerve discharge occurring during remotion of the joint, and remotor nerve discharge on promotion (Fig. 1). During some periods of joint movement, the pattern will spontaneously switch to an ‘assistance reflex’ type of discharge (Fig. 2). The remotor neurones are now excited when the joint is remoted and the promotor discharge occurs on promotion of the joint. During a continuous cyclic T–C movement, the modification usually occurs first in the promotor neurones, which first discharge in response to TCMRO stretch and then again to a release in the same cycle of stimulation; the next passive remotion elicits a remotor discharge. This behaviour persists for a variable length of time, in Fig. 2*a*, it lasts several seconds, but the discharge then returns progressively to a classical resistance reflex. The different patterns occur in a complimentary fashion in the two antagonistic groups of motoneurones at any given phase of T–C joint movement, the remotor discharge decreases in strength as the promotor activity returns to a resistance discharge.

While the reflex discharge evoked by movement may be either an assistance resistance pattern, the firing of both the remotor and promotor neurones may vary

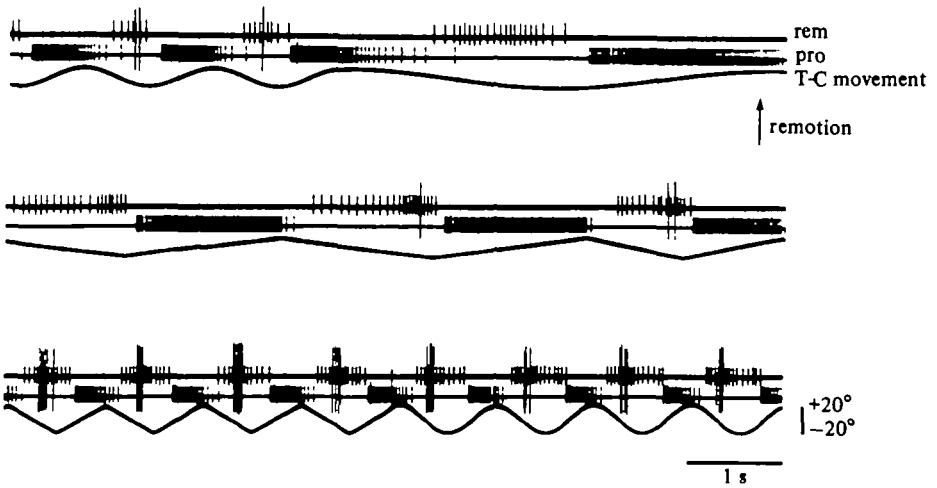


Fig. 1. Resistance reflex evoked by passive movement of the T-C joint with varying frequencies of two different types of joint movements. The top two traces in these three continuous records are extracellular recordings of promotor (pro) and remotor (rem) motoneurone activity. The bottom trace is the T-C joint angle. The zero of the movement trace is when the joint is midway between full remotion and promotion of the joint.

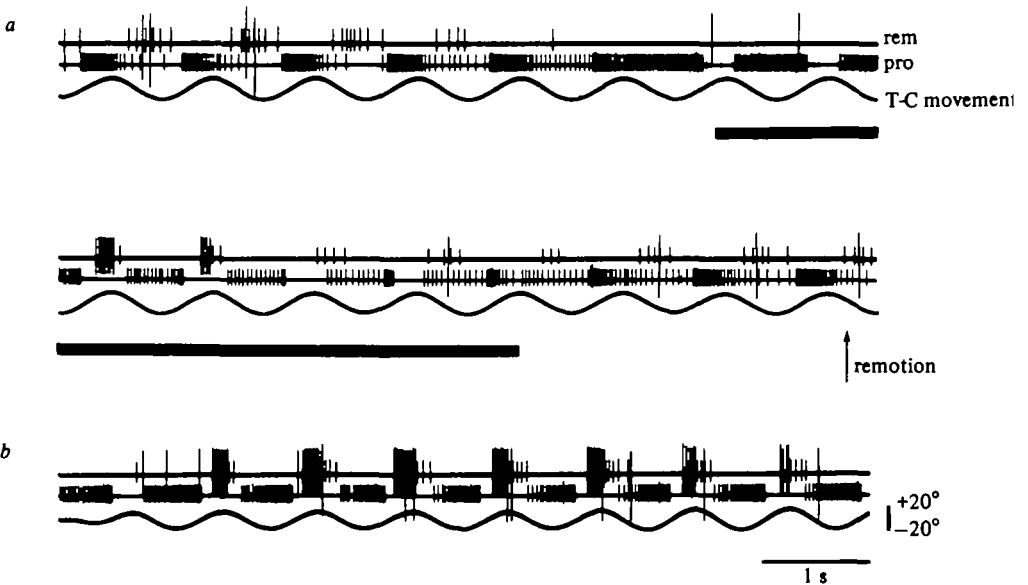


Fig. 2. Spontaneous reversal of reflex activity during movement of the T-C joint. (a) In these two continuous records, the solid bar beneath the movement trace indicates a period of reversed reflex activity. A normal resistance reflex is evoked in promotor and remotor neurones during the first 6 cycles of movement. The reflex pattern then abruptly switches to an assistance pattern, indicated by the solid bar, which progressively weakens and returns to a resistance reflex pattern. (b) This record illustrates a longer lasting assistance reflex pattern, which started as soon as the T-C joint was moved.

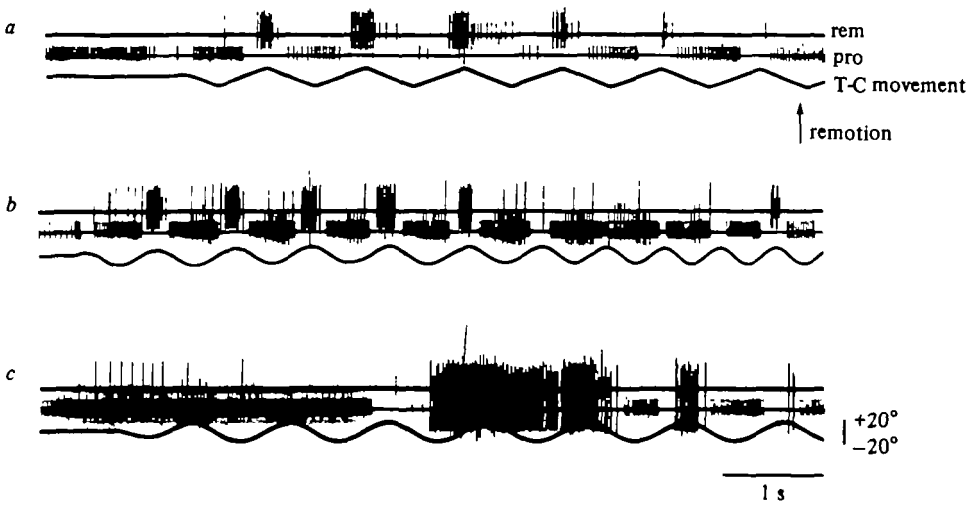


Fig. 3. Assistance reflexes recorded during and after periods of spontaneous motoneurone activity. (a) Movement of the T-C joint after a period of spontaneous activity in the promotor neurones immediately evokes an assistance reflex in remotor and promotor neurones. Note also the variation in reflex activity elicited from both motor nerves in any cycle of joint movement. After two cycles of reflex activity in both nerves, the promotor response almost disappears, while the remotor units fire normally, followed by a similar decrease in remotor firing while promotor activity is present. (b) An assistance reflex produced after spontaneous activity in the promotor nerve persists for several seconds, followed by five cycles of movement where there is an assistance reflex evoked in only the promotor nerve, with no reflex activity in the remotor nerve. (c) Joint movement elicits no reflex activity in either nerve during spontaneous firing of promotor neurones, and only a weak modulation of remotor activity during a burst in these nerves. An assistance reflex is evoked at the end of this period of spontaneous activity.

in phase with relation to the imposed limb movements. The variation in the reversal effect is demonstrated in Fig. 3. Sometimes the assistance reflex sequence is perturbed, with no promotor activity during promotion of the T-C joint and weak firing on remotion (Fig. 3a). At other times, an assistance reflex is manifested in, for example, strong promotor discharge during a passive promotion while only weak remotor activity is evident during the remotion movements (Fig. 3b).

This variability presumably corresponds to a variation in the activity of the central pathways controlling the afferent information from the TCMRO. The occurrence of the assistance reflex appears to be dependent on the level of central activity of the preparation. For example, when the animal has been stimulated by touching one of the leg stumps, imposed movements of the T-C joint will usually elicit an assistance reflex immediately. Following a period of rhythmic activity in the promotor and remotor neurones probably corresponding to walking or swimming movements, passive movement of the T-C joint elicits one of two possible responses: either the central discharge may continue as before, the T-C movement appearing totally ineffective (Fig. 3c); or alternatively, the motoneurone firing stops abruptly and a very strong assistance reflex occurs when the joint is moved with a discharge of a large

Number of remotor and promotor units (Fig. 3*a, b*). With continued repetitive T-C movement there is a progressive return to a classical resistance reflex. This result is also obtained after periods of non-rhythmic activity in either or both motoneurones.

#### DISCUSSION

A significant feature of this preparation is that a high level of central nervous system activity is maintained even though the animal is extensively dissected. It is clear in our experimental situation that variation of central activity greatly influences the proprioceptive reflex pattern, switching a resistance reflex to an assistance one. Bässler (1976) in studies on the stick insect, was the first to demonstrate that antagonistic reflex patterns could be evoked by the same proprioceptor, depending on the central state of the animal. Ayers & Davis (1977, 1978) demonstrated 'reinforcing reflexes' in the walking legs of *Homarus* but in their experiments the reflexes were elicited by moving the joint in an intact animal, thus all the receptors of each leg joint were simultaneously stimulated. In the present study, only the TCMRO was involved, all the other sensory nerves being cut. A complete switch between antagonistic reflex patterns was never described in their study, and assistance reflexes always occurred concurrently with resistance reflexes at the same joint. In the antenna of the rock lobster, Vedel (in press) showed in intact restrained animals a competition between a resistance and an assistance reflex. Here again the two reflex patterns were always co-active, although the strength of each reflex pattern could change depending on the state of the animal. In that case, as in the stick insect leg (Bässler, 1976), the reflex originates from a chordotonal organ, whereas in the present experiments it stems from a muscle receptor organ. It is unlikely that this effect can be explained by a variation in tension of the receptor muscle since the reversal effect persists when the receptor motoneurones are cut.

To obtain the two antagonistic reflex patterns described here, the sensory information from the TCMRO must be modified by central nervous mechanisms. The three possible neurones involved in these reflexes are the T, S and P fibres of the TCMRO (cf. Bush, 1977). The T and S fibres excite promotor motoneurones in inactive preparations, although only the T fibre appears to be able to excite these neurones at physiological levels of sensory fibre depolarization, and the functional significance of the P fibre is as yet unknown. The T fibre has been shown to make monosynaptic excitatory connections with promotor motoneurones (Blight & Llinas, 1978), and the mechanism which causes the reflex must alter this strong interaction. The occurrence of the assistance reflex may suggest a possible role for the S fibre input to the ganglion, but further studies of this system using intracellular recordings will be necessary to investigate these central effects.

The occurrence of reflex reversal in vertebrate studies has been known for some time. Sherrington (1900) demonstrated that the initial limb position may determine the reflex response to cutaneous stimulation. More recently, in studies of cat walking and dogfish swimming, tonic and phasic reversal reflex responses are known to occur in a given stereotyped behaviour pattern (Forssberg *et al.* 1976). For example, it has been demonstrated that during locomotion in the cat, an identical sensory stimulus

elicits a different motor response depending upon the timing of the stimulation in the step cycle.

The fact that this effect seems to appear as a general feature of central nervous mechanisms invites some speculation as to its role in different motor behaviour patterns. The same sensory information could be used in a resistance reflex to maintain a given posture or to oppose unpredictable passive limb disturbances, or in an assistance reflex to help the central motor score in maintaining an ongoing movement, and in load compensation during movement as suggested by Ayers & Davis (1977) and Kennedy & Davis (1978).

This switching between a resistance and assistance effect may explain why resistance reflexes are not recorded during a stereotyped behaviour. The present data highlights the complexity of the interactions between the sensory inflow and the central motor command (Clarac, 1977), and indicates the importance of central effects in the modification of sensory-motor relationships.

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