

## INFLUENCE OF COXA-THORAX JOINT RECEPTORS ON RETRACTOR MOTOR OUTPUT DURING WALKING IN *CARAUSIUS MOROSUS*

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### SUMMARY

The coxal leg-stump of a stick insect was rotated about the coxa-thorax joint, by means of a pen-motor, while the animal walked on a self-propelled double treadwheel. Motor activity in the retractor muscles of the legs was recorded for standing and walking animals with the stump of either the middle or hind leg moved forward and backward in a trajectory similar to that used in a walking step.

In a standing animal the movement of either leg evokes a weak resistance reflex. If the animal walks with the middle leg-stump held still, then short and weak motor bursts are generated with the periodicity of the walking legs. Front and hind legs alternate in a manner typical of the middle leg amputee and the retractor muscle of the amputated leg is most active during the power stroke of the leg behind.

When a middle leg-stump is moved at a different frequency from that of the walking legs, the motor output to the retractor is strongly modulated and depends on the relative timing of the stump and the walking legs.

Rearward movement of the hind leg-stump, during walking, is always accompanied by strong motor output in the retractor muscle. In addition, forward movement in this leg-stump produces a resistance reflex similar to that produced in the standing animal.

### INTRODUCTION

Fast-walking stick insects with an amputated middle leg fitted with a balsa prosthesis, step with 'normal' coordination (front and hind legs lifting synchronously) rather than with the amputee gait (front and hind legs alternate) (Wendler, 1966). This suggests that all sensory apparatus distal to the coxa-thorax joint is relatively unimportant when considering the gross aspects of inter-leg coordination. This has also been demonstrated in Crustacea (Grote, 1981) and it suggests that information on the position and stress of the leg during standing and walking behaviour is derived primarily from the organs situated at the base of the leg where it attaches to the thorax.

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From a recent review by Stein (1982), it is clear that the complexity of the primary receptor (muscle spindle) in mammals has made it difficult to assess the importance of position, velocity, acceleration and force in the control of mammalian movements. In insects and other arthropods different organs are used to provide information on the parameters of force and position (and their derivatives). This simplifies to some extent the investigation of the control of movement in these animals. The precision of control available to insects appears to be at least of the same order as that available to mammals. Studies by Wendler (1972) on the vertical loading of standing animals, by Baessler (1972) on the femur-tibia joint, and by Graham & Wendler (1981) for the coxa-thorax joint have demonstrated reflex control in standing animals, and more recent work by Cruse & Schmitz (1983) and D. Graham & D. Godden (in preparation) on walking animals shows that servo control related to position or velocity feedback is present in the femur-tibia joint and coxa-thorax joint respectively.

The present study was designed to examine the capabilities of the coxa-thorax joint in controlling the motor output of the leg when moved externally over a range and with a frequency comparable to the normal step during walking. The joint is still acted upon by its own muscles in this configuration and movements of the stump are applied during standing and walking behaviour.

#### METHODS

Adult female *Carausius morosus* were supported above a pair of light counter-balanced wheels as described in Graham (1981). The middle leg on the left side was cut away between the coxa-trochanter and coxa-thorax joints. The wound was then sealed by allowing the blood to clot around a small ball of fibre inserted into the open coxa. A Helliger pen-motor fitted with a drawing pen clamp was used to grasp the open end of the coxa and move the stump about the coxa-thorax axis as shown in Fig. 1. Thin copper wires (50–100  $\mu\text{m}$  in diameter), attached to miniature insect pins by conductive silver paint, were used to harpoon and differentially record the motor activity in the retractor muscles of all the legs. These signals were amplified and recorded on tape along with the ramp voltage supplied to the pen-motor. The pen-motor was driven by a voltage-dependent relaxation oscillator whose frequency and rate of rise and fall could be adjusted to give a rearward stance duration of between 0.5–1.5 s while the swing duration remained approximately constant at 100–200 ms. A position detector was used to compare the pen-motor output with the voltage input. The lag was 5° at 10 Hz corresponding to 8% of the full swing.

The following experiments were performed on four animals moving middle and hind legs while attempting to record from retractor muscles of all the other legs.

(1) Animal stands while the leg is moved forward and backward with the timing of a typical leg step (1 Hz).

(2) Animal walks with the leg held still at 90° to the body.

(3) Animal walks with the leg oscillating at a constant rate between 0.5 and 1.2 Hz. Occasionally movement of the wheel was blocked.

Unfortunately, it was not possible to move the front legs due to problems in immobilizing the prothoracic segment. Furthermore, the precise positioning of the

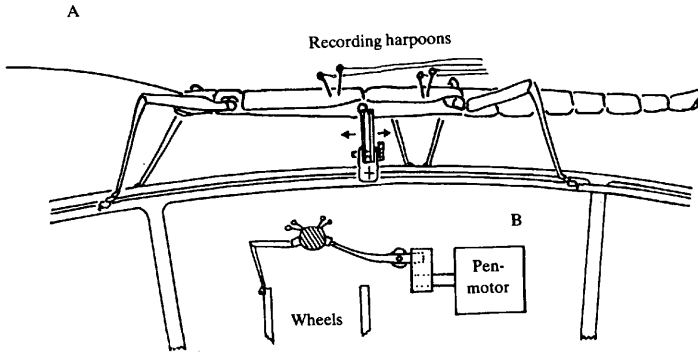


Fig. 1. Side view of a preparation showing the operated leg and 'harpoon' electrodes used to record activity in the muscles which move the coxa to the rear. (A) Side view; (B) front view.

electrodes to record from the required muscles is not known for the front legs and these records were often difficult to interpret, although in most instances the motor output was clearly related to rearward movement of the leg. Myograms and the driving voltage of the pen-motor were recorded on tape.

#### RESULTS

During thanatosis (stick mimesis) the retractor muscles of the stick insect show a strong and reliable resistance reflex during forward movement of the leg and are capable of following a sinusoidal input in the frequency range 0.01 to 10 Hz. The reflex appears to be associated with sense organs around the lower articulation of the subcoxal joint. Neither the caudal hair-rows parallel to the coxa long axis nor the upper joint provide a necessary input to this reflex (Graham & Wendler, 1981).

##### *Movement of the middle leg-stump*

In animals which are standing but have recently walked, the gain of the reflex is much lower than in cataleptic animals (Baessler, 1972; Godden, 1974). This weak reflex is shown in Fig. 2A where the leg-stump is moved forward and backward while the animal stands still. The other legs show no motor output as the operated leg is moved forward stretching the retractor muscle and eliciting a few muscle potentials. This may be compared with the much stronger responses reported by Graham & Wendler (1981) in cataleptic stick insects.

Fig. 2B shows the same animal walking with the leg-stump (L2) held still at approximately 90° to the body axis. The retractor is active in each step cycle with a short burst which is coordinated with the output of the ipsilateral walking legs. Comparison with leg R2 on the other side shows that the stump burst is much shorter and contains a much smaller number of muscle potentials. The timing of the

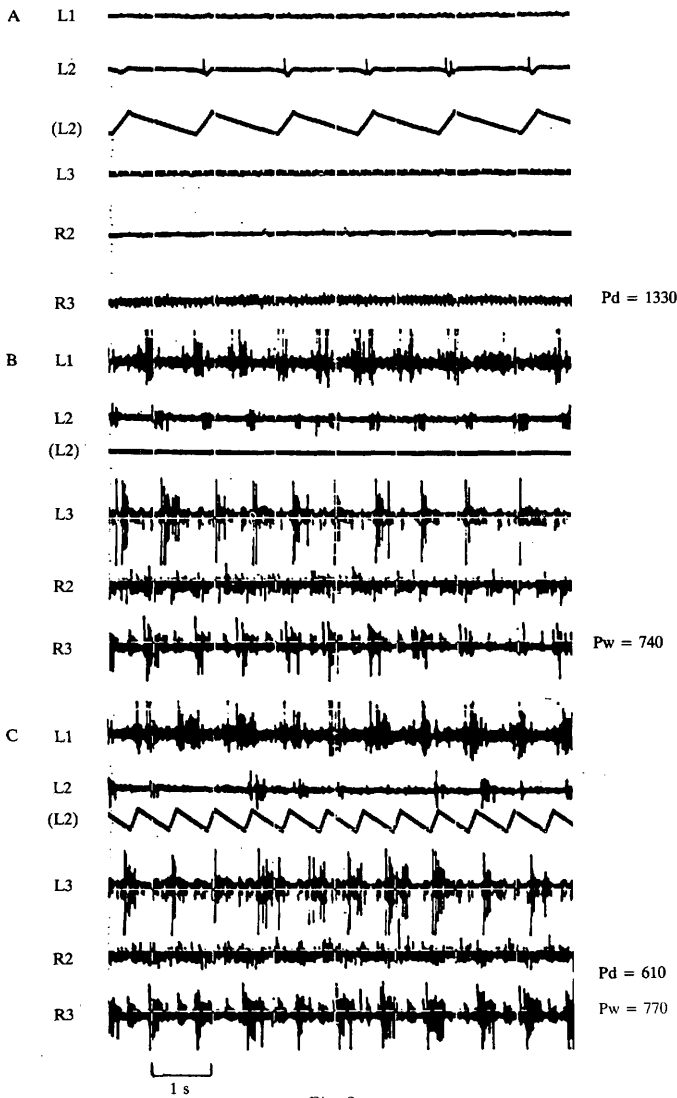


Fig. 2

burst alternates with the maximum retractor activity in leg L1 and is close to synchrony with the maxima in L3 which is alternating with L1.

Fig. 2C shows the same animal with the middle leg-stump moved forward and backward to resemble the normal movement in walking. The stump is moved with a constant period of 610 ms, while the walking period is 770 ms. Thus the driving rhythm and the step rhythm come into a particular phase relationship every five cycles of the stump movement. This beating effect can be seen in the sequence and it is when the middle leg is moving rearward in phase with the power stroke of the hind leg on the same side that the motor output is at a maximum. Front and hind legs on the same side still alternate and the maximum in the stump retractor muscle occurs close to the swing phase of the front leg.

#### *Movement of the hind leg-stump*

The hind leg has a similar static reflex, as shown in Fig. 3A, when the stump is moved in the standing animal. However, the imposed movement often causes the standing legs to show weak motor activity and the animal sometimes walks forward under this stimulus. If the animal is stimulated to walk by touching the abdomen while the stump is moved the motor output is much stronger than that of the middle leg and there is no pronounced 'beating' effect. The motor output to the retractor muscle is strong in each cycle of the stump movement and the firing frequency is similar to that of the contralateral leg R3, although some of the units appear to be absent from the stump retractor record.

Another difference is that forward movement of the hind leg-stump during a walk is usually accompanied by a brief burst of retractor potentials (Fig. 3B). This is similar to the normal reflex activity shown in Fig. 3A and has not been observed in the middle leg experiments (see Fig. 2C). Fig. 3C shows the result of stopping the wheel. The animal generates a prolonged burst in the retractor muscles of all the legs. This maintained activity is modulated by a forward movement of the stump which inhibits or suppresses the reaction to the increased horizontal load produced by stopping the wheel.

#### DISCUSSION

The experimental results show that rotation of the coxal stump about the coxa-thorax joint produces changes in the motor output to the muscles producing the powerstroke in walking. The influence of moving the leg-stump varies with both the state (standing or walking) and behaviour (walking freely or under load) of the animal and the leg examined. If the animal stands, a weak resistance reflex is present. If the animal walks with the stump held still, a weak motor burst coordinated with the walking legs appears. If the middle leg-stump is moved during walking, the motor output is only expressed when its movement bears a certain

Fig. 2. (A), (B) and (C). Simultaneous recording from the tergo-coxal retractors of legs L1, L2, L3, R2 and R3. The stump movement trace is labelled (L2) indicating that the left middle leg is being moved forward (up) and rearward (down) in this experiment. Pw denotes the mean step period of the walking legs and Pd is the period of the driven leg. During imposed movement the leg-stump rotates through an angle of 40–60°.

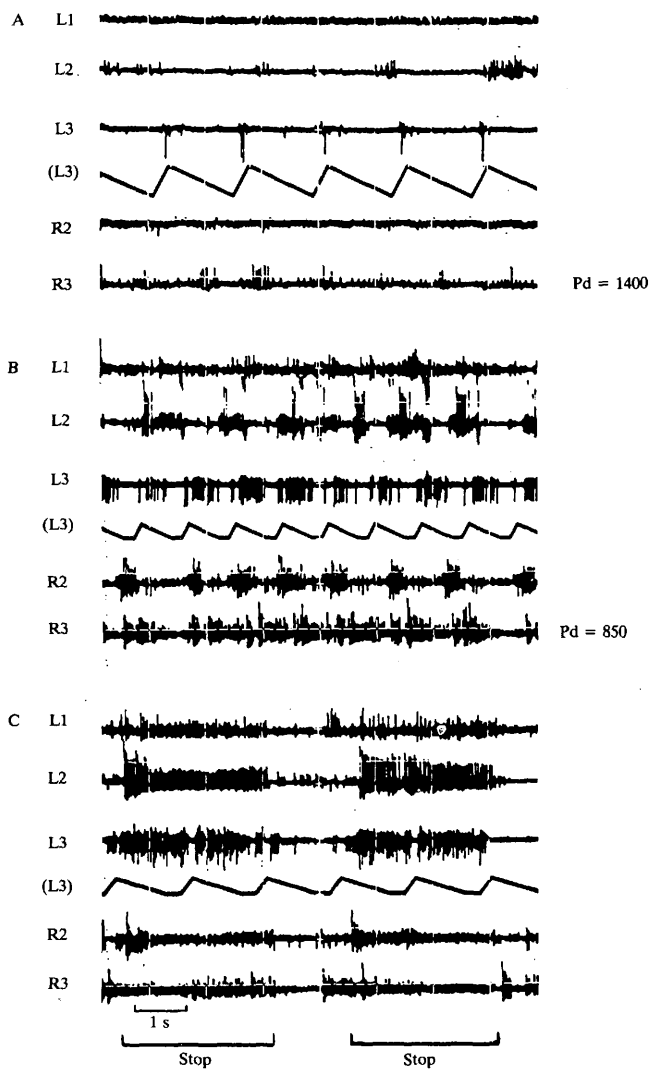


Fig. 3. (A), (B) and (C). As in Fig. 2 except here the driven leg-stump is (L3).

relationship to the leg in front and behind it. The phase relationship of the front and hind leg is one of alternation whether the stump moves or not. This suggests that the animal is not fooled into believing that the leg is intact when it is moved by the pen-motor as is thought to be the case when a 'peg-leg' is attached to the stump. The walking system appears to be aware that the moved stump cannot provide support but sensory input still modifies motor output in this experimental situation.

Considering the movements in the plane parallel to the walking surface containing the body axis, the sensory input to the leg in the standing experiment is primarily a position input with a brief stress transient as the leg changes direction (caused by inertia) and a weak tonic component as the retractor muscle attempts to resist the imposed movement. When the animal walks with the leg held still, the sensory input is primarily one of stress generated by the weak bursts of motor output with the rhythm of the walking legs. This stimulus is probably similar in sign to that experienced in normal walking during the powerstroke but is smaller in magnitude and may enhance the output generated endogenously or from the movements of the other legs. If this is the case then the initial output may be even weaker before this stress enhancement, suggesting that motor generation must be heavily dependent upon movement of the leg if the powerstroke burst is to approach that expected in a freely-moving leg. In the moving leg experiment, the stimulus situation is much more complex as both the imposed movement and the active motor output could cause stimulation of stress- and position-detecting organs. The modulation of motor output observed in a middle leg-stump shows that if stress and/or position are in some sense inappropriate, the weak motor output normally associated with amputation is suppressed. In this experimental situation the motor output of the stump is more closely synchronized with the hind leg rather than the leg in front, as predicted for the amputee from the work of Wendler (1966), Barnes (1975), Graham (1977) and Clarac & Chasserat (1979). It is possible that such a time shift could be caused by some unrealistic interaction of stress and position inputs derived from the pen-motor stimulator.

There is, however, some evidence that middle leg amputees can use the normal intact gait. Delcomyn (1971) found examples of such stepping patterns in fast walking cockroaches. More recently, Baessler & Wegner (1983) observed that stimulation of a partially denervated preparation (by squeezing the trochanter of a middle leg) could induce synchronized retractor motor output in both the middle and hind leg. The motor activity produced in their preparation differs from that found in normal walking behaviour but there is evidence for a reflex pathway directed towards the rear which could be responsible for the unusual timing observed in the leg movement experiment described here. This would not appear to be a path normally used in walking but might occur under special conditions such as starting or during rocking behaviour.

Considering the movements in the vertical plane of the leg, the pen-motor drives the leg along an arc similar to that produced during a normal power and recovery stroke (leg levation being mainly derived from the movement of the coxa-femur joint). This experimental situation gives a resistance to depression similar to that provided by the outer parts of the leg in the intact situation and should provide a similar stimulus to that found in the 'peg-leg' experiment.

The present study shows that the major reflex response to movement changes in sign (phase) and magnitude (gain) in going from a 'standing' to a 'walking' state and is another example of the reflex-reversals or phase-dependent responses reported by Grillner, Rossignol & Wallen (1975), Baessler (1976), Marrelli & Evoy (1976), Vedel (1980) and DiCaprio & Clarac (1981).

The middle leg experiments reported here are not sufficiently exact to show whether subtle coordinating influences exist between the driven and intact legs, but the modulation or beating phenomenon shows clearly that driving the middle leg does not force the other legs to operate at the leg-stump frequency.

In the hind leg, the periphery appears to have a general excitatory influence which tends to start a standing animal. In a walk there is a strong and specific excitatory influence producing a rhythmic motor output similar in intensity and duration to that found in the intact leg. This motor output is phase-locked to the stump movement and with a suitable choice of stimulus frequency some phase-locked coordination with other legs may be possible.

This new experimental configuration suggests an effective approach to the investigation of the mechanisms of reflex control of the muscles in the arthropod leg and the coordination between legs. Selective cutting of the thoracic muscle tendons acting on the sub-coxal joint should make it possible to eliminate stress as a variable and to concentrate on the role of position information and its derivatives in the control of motor output to the muscles of this joint. It may then be possible to evaluate the stress influences by a careful comparison of cut and uncut tendon experiments.

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