

THE ORGANIZATION AND ROLE DURING LOCOMOTION OF THE PROXIMAL MUSCULATURE OF THE CRICKET FORELEG

II. ELECTROMYOGRAPHIC ACTIVITY DURING STEPPING PATTERNS

BY GILLES LAURENT* AND DANIEL RICHARD

† *Laboratoire de Neuroethologie, UA CNRS 664, Université Paul Sabatier, 118 Route de Narbonne, 31062 Toulouse Cedex, France*

Accepted 27 February 1986

SUMMARY

A description is made of the patterns of electrical activity in the proximal muscles of the cricket foreleg during restrained locomotion and seeking movements, while the animal is held by the mesonotum, allowing the legs complete freedom of movement.

1. The initiation of the swing phase corresponds to the onset of the abductor muscle activity (Fig. 1). Its duration is matched by that of abduction–promotion and does not depend on the step frequency. Leg position is more variable at the end of the stance than at the end of the swing.

2. The promotor and abductor muscle activities are linked (Fig. 2). At least three units can be distinguished in each and the duration of their bursts is independent of the period (Fig. 3).

3. In the double depressors of the trochanter, muscles 77-1b,c (Fig. 4), one unit per muscle was identified, bursting during the swing phase. The duration of the burst is independent of the period. Some isolated potentials occasionally occur during the stance phase.

4. The overall activity in the lateral and medial remotors is coupled to the period; three main patterns can be described, depending upon the muscle bundle and the velocity of movement (Fig. 5).

5. In the coxal depressors two patterns of activity are described which depend on velocity of stepping (Fig. 6): (i) during regular and fast stepping (at frequencies greater than 2.5 Hz), the activity is coupled to that of the double depressors; (ii) during slow or irregular stepping, the activity is biphasic: an initial burst is followed after a latency correlated to the period by a second one in the second half of the stance phase. Conversely, the latency between the end of the second burst and the onset of the following abductor burst does not depend on the period. In most cases, a fast neurone (large amplitude, short phasic activation) is recruited when a slow one reaches high rates of discharge.

*Present address: University of Cambridge, Department of Zoology, Downing Street, Cambridge CB2 3EJ, UK.

† Reprint requests should be sent to D. Richard at this address.

6. The levators are active during the whole of the stance, and during the early swing phase, when a 45-ms period of coactivation with the abductors occurs (Fig. 8). Activity is correlated to the period.

7. During the transition from stance to swing phase there is almost no coactivation of direct anatomical antagonists. Conversely, during the transition from swing to stance phase (i.e. when the leg reaches the substrate), simultaneous activity is often seen in direct antagonists (Fig. 9).

8. The overall activity and the sequence of recruitment of the proximal musculature is summarized for a typical step cycle.

9. Records are shown to compare muscular activity during 'seeking' movement with actual locomotor patterns (Fig. 10). The period of the seeking movements is shorter than an average step, and this shortening mainly consists of a curtailing of the retraction duration. Also the level of activity is higher, the firing frequency being raised and fast units being recruited. The coxal depressors are only active in phase with the abductors.

10. The influence of the experimental procedure on the myographic patterns is discussed.

INTRODUCTION

Numerous descriptions of the walking patterns of insects have been made in the last two decades. Intra- and intersegmental coordination has been considered from the point of view of behaviour (Wilson, 1966; Delcomyn, 1971; Burns, 1973; Harris & Ghiradella, 1980), electrophysiology (Hoyle & Burrows, 1973; Pearson & Iles, 1973; Graham & Wendler, 1981) and model simulation (Graham, 1977; Cruse & Saxler, 1980). During 'normal' walking on a smooth, horizontal surface, an insect typically uses an alternating triangular gait in which a mesothoracic leg is promoted or remoted synchronously with the contralateral pro- and metathoracic ones, and in antiphase with the other three limbs. In a step cycle, each leg alternately goes through a stance and swing phase, during which the coxa, trochanter-femur, tibia and tarsus move relative to each other and to the thorax, in a more or less constant sequence. In Orthoptera, the stepping cycle of the hind leg mainly consists of alternating flexion and extension of the coxotrochanteral and femorotibial joints (Burns & Usherwood, 1979), whereas the middle and forelegs also undergo essential coxal rotations relative to the body (Burns, 1973; Harris & Ghiradella, 1980). Though considered as important in locomotion (Hustert, 1983), the elaborate proximal thoracocoxal, thoracotrochanteral and coxotrochanteral muscles have only rarely been considered from the point of view of their sequential recruitment during walking. The importance of the musculature of the proximal joints is further emphasized by the high complexity of their mechano- and proprioceptive control (Bräunig & Hustert, 1985*a,b*).

This paper analyses the normal locomotor activity of the muscles at the two proximal joints of the foreleg of the cricket, by providing a qualitative and quantitative description of their motor patterns during locomotion on a styrofoam ball. The muscles and their innervation as well as the morphology of the corresponding motor neurones have been described in the preceding paper (Laurent & Richard, 1986).

MATERIALS AND METHODS

Thirty-three intact adult crickets, *Gryllus bimaculatus*, were fixed to a holder by the posterior vertex and mesonotum after removal of the forewings. They were allowed to walk freely on a light styrofoam ball (diameter = 32 mm, mass = 400 mg; mass of cricket = 1020 ± 205 mg, $N = 15$ for the crickets) coated with varnish or paraffin. In some experiments, this ball was put on the surface of water, in order to counteract its weight and cancel this vertically directed load. The patterns of muscular activity were then compared to those when the animal held the ball (Fig. 7). Pairs of 20 or 50 μm diameter insulated copper electrodes were inserted through small holes in the cuticle, to record the electrical activity (electromyograms, EMGs) from selected muscles during walking. Since most of the muscles from which activity was recorded have only a small attachment to the cuticle, each successful recording was followed by the dissection of the animal, to locate the tips of the electrodes. Only data from identified recording sites were considered. Furthermore, the behaviour of the animal was described, as it was moving, on the voice channel of the tape recorder.

In several experiments, the movements of the distal end of the femur were recorded simultaneously with the EMGs using CCD line SCAN sensors (Fairchild CCD 111), with a rate of 10 images s^{-1} . The point of reference chosen was the first potential in the myographic recordings. When plotting the successive positions of a leg, the standard time error was estimated to be half the duration of an image, i.e. 25 ms. The exact occurrence of tarsal contact with the substrate, however, could not be determined with precision.

The step period is divided into two successive phases: the swing phase, during which the leg is lifted and brought forward, and the stance phase, when the leg is drawn posteriorly while the foot is in contact with the substrate.

For the analysis of the temporal parameters, we took the onset and offset values of the EMG activity when the bursts were clearly defined. The data presented in the histograms are pooled from 3 to 11 animals. The statistical relationships between the period duration (x) and several temporal parameters of the step cycle (y , burst durations, interburst intervals) were studied using linear regression analysis.

In the scatter diagrams, Figs 3C–D, 4C, 5E–F, 6E–F, 8C, the two regression lines (y on x and x on y) are presented. The y -intercepts and slopes are different since the means and standard deviations of both x and y variables are different. The correlation coefficients were calculated in each case by dividing the sample covariance by the product of the sample's standard deviations.

For the analysis of instantaneous frequency of motor spikes *versus* period of the step, only excitatory signals of relatively large amplitude could be used. These records were passed through a spike window discriminator before digitization, and then processed with an Apple II microcomputer. Spikes occurring at frequencies lower than 10 Hz were not represented. Each part of Fig. 9 was obtained by pooling 15 periods (each of 600 ms duration) from three to five different animals. Each dot represents the instantaneous firing frequency, i.e. the inverse of the time interval

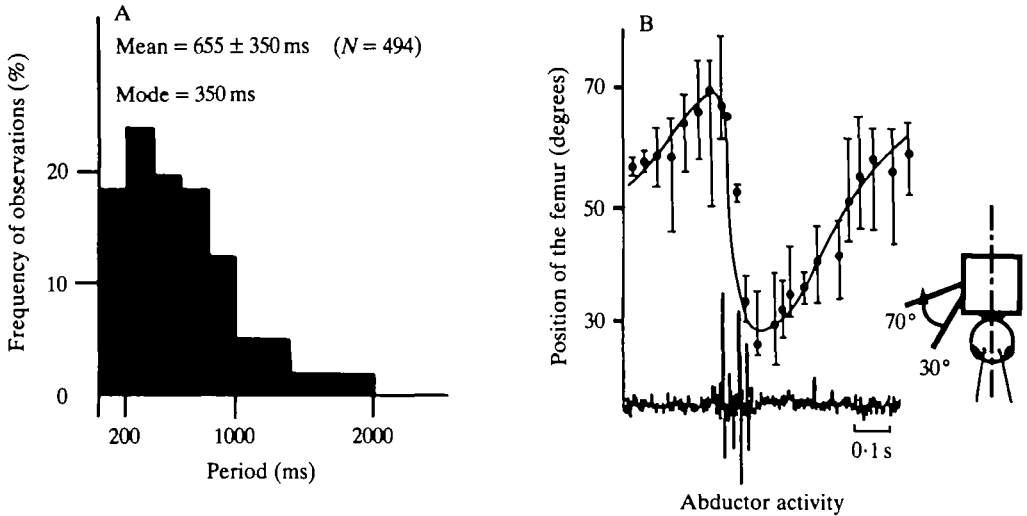


Fig. 1. Characteristics of the step cycle. (A) Histogram of distribution of the period durations over 494 steps (mean \pm S.D., number of observations) pooled from 11 animals. The mean period corresponds to a frequency of $1.52 \text{ steps s}^{-1}$. (B) Anteroposterior position of the femur, plotted against time, along a step cycle; the point of reference is the first potential in the abductor myographic recording (50 steps, see Materials and Methods). The activity in the coxal abductors corresponds to the swing phase, when the leg is brought from its posterior to its anterior position. Note that, for a given position of the femur, the distance between the two extreme positions is generally greater before than after the swing.

elapsed between two successive potentials. Apart from the coxal depressors, two successive potentials belonged most of the time to different units, the window of the discriminator having been set to count a potential of any size. Consequently, the density and distribution of dots represent the *general* activity in a particular muscle. All the data come from paired recordings, one of the traces being either the abductor or the promotor muscle; all traces were assembled by choosing as time 0 the first spike in either the abductor or the promotor.

RESULTS

Leg movements and electrical activity

To help in relating electrical signals to the actual limb movement that they induce, an experiment was first performed to determine an electrical parameter that would indicate reliably a constant phase of the step cycle, and could then be used as a landmark throughout the succeeding experiments.

The observed step periods mostly varied between 150 and 1000 ms, with a mean step frequency of 1.5 steps s^{-1} (Fig. 1A). The highest running speeds were observed after stimulation of the cerci, which triggered an escape reaction.

In Fig. 1B, the average horizontal positions of the femur (with extreme values represented by vertical lines) over some 50 steps of one animal are plotted as a function of time, correlated with the first potential appearing in a simultaneous

recording from the abductor muscle (muscle 74-1, see Laurent & Richard, 1986). This was possible because the steps chosen were of similar duration (600 ms), and because the time for which the abductors are active shows little variability (see below) whatever the step frequency.

Three main points emerge. (1) The swing phase began with the onset of the abductor activity, and slightly outlasted the burst duration: the abductor onset is therefore a useful indicator of the stance-to-swing transition. (2) The swing phase duration represented only one-fifth to one-quarter of the step cycle, depending on the velocity (ranging from 1.5 to 6 Hz: see below). It should be kept in mind that the restrained animals were propelling the substrate (that was less than half their weight) with their legs, which might explain the narrow range of velocities. (3) For a given position of the femur, the distance between its extreme positions was greater at the end than at the beginning of the stance phase.

Activity in the abductor and promotor muscles

The abductor and promotor muscles have been shown to be supplied by six and five motor neurones, respectively, with axon diameters greater than 15 μm (Laurent & Richard, 1986); consequently, it was often possible to distinguish several of the excitatory motor units from the EMGs (Fig. 2A).

The better records of the abductor activity during walking reveal with certainty at least two small junction potentials (Fig. 2A), and a large amplitude unit mainly seen during a fast motor activity (an escape reaction or seeking movements: Fig. 10). The three remaining units may not be normally recruited during walking, they may have been masked by the three larger ones, or they may only innervate distal muscle fibres and therefore their activity could not be recorded. The onset and termination of the abductor bursts were clearly delineated (Fig. 2A,B). The average burst duration was 101 ± 45 ms, $N = 200$ (mean \pm standard deviation, sample population) and the mode was 80 ms (Fig. 3A). Over the whole range of walking velocities, the duration of the activity was relatively constant, and not statistically correlated to the step period (Fig. 3C). During a burst, the average firing frequency of each of the two small units was 45 Hz. All units were silent during the interburst period.

The promotor muscles showed a similar pattern of activity. Three main units could be described without ambiguity (Fig. 2B,C): (i) the first, of small amplitude, fired during the phasic promotion burst, and occasionally tonically during a long stance phase (potential 1, Fig. 2B); (ii) a medium-sized unit which was limited to the promotion burst (potential 2, Fig. 2B); (iii) a large unit, also restricted to the phasic promotion, which fired from one to five times during rapid motion (potential 3, Fig. 2B), or during different behavioural activities (eye cleaning, Fig. 2C) that require sharp promotion-adduction movements.

During locomotion, the main duration of the phasic burst was 103 ± 43 ms, $N = 122$, and did not depend on the velocity, remaining almost constant regardless of the period (Figs 2B, 3B,D). The activities of both abductors and promotors were strictly phase-related, and the onsets of each burst coincided.

Activity in the double depressors

The pattern of activity in this muscular group has only been recorded from muscles 77-1b and 77-1c (Laurent & Richard, 1986) because muscles 77-1d and 77-2

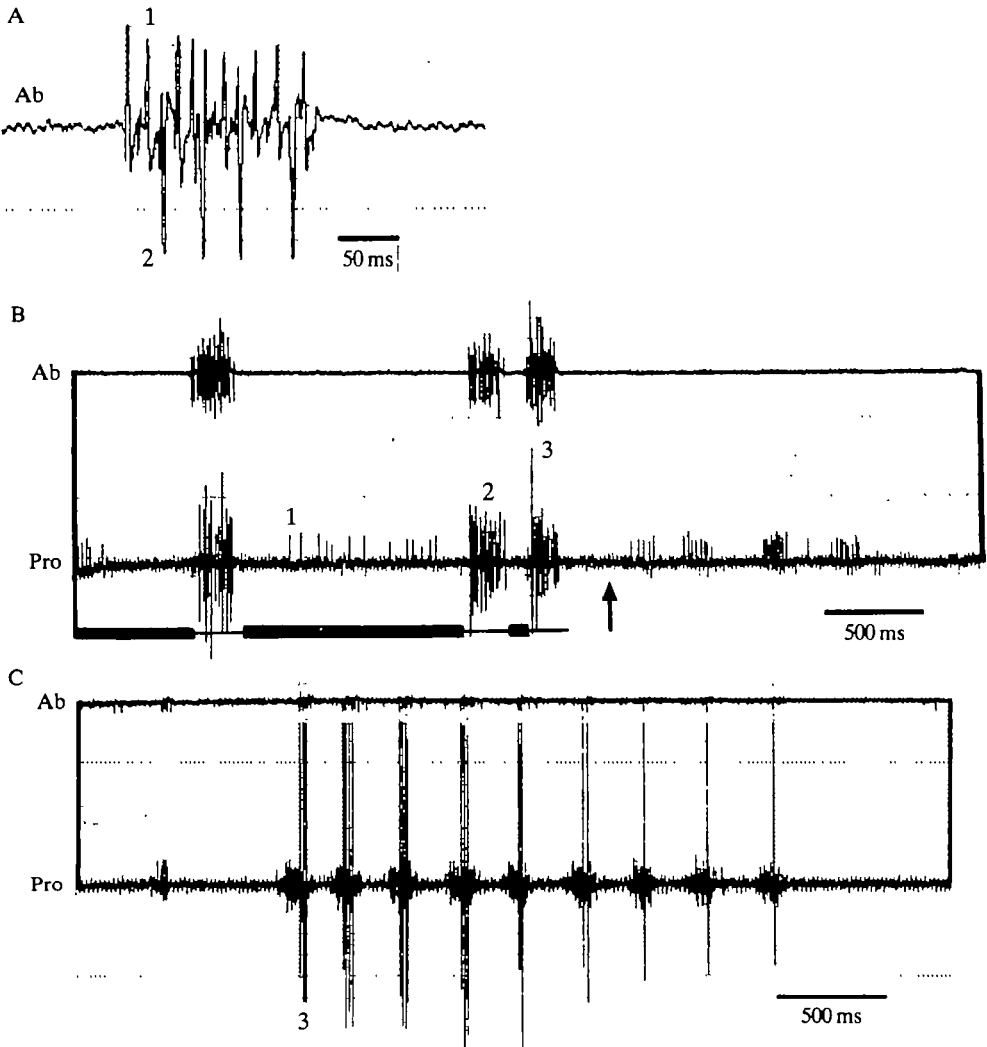


Fig. 2. Patterns of activity in the abductor (Ab) and promotor (Pro) muscles. (A) The two main potentials in an abductor burst. (B) Simultaneous recordings from the abductor and promotor muscles: the onsets of the two bursts are simultaneous, their durations are similar, and do not depend on the period. The two steps the animal makes before stopping (arrow) are of different duration, but the abductor and promotor bursts remain constant. When the animal stops, a phasitonic activity is seen in the promoters. This small amplitude unit also sometimes fires during the stance phase. Units 1, 2 and 3 are the three main ones in the promoters. The stance phase is indicated by the thickened line. (C) During a sequence of eye-cleaning behaviour, only the smallest and largest amplitude units are recruited; the firing frequency of the large unit decreases, and the abductors are silent.

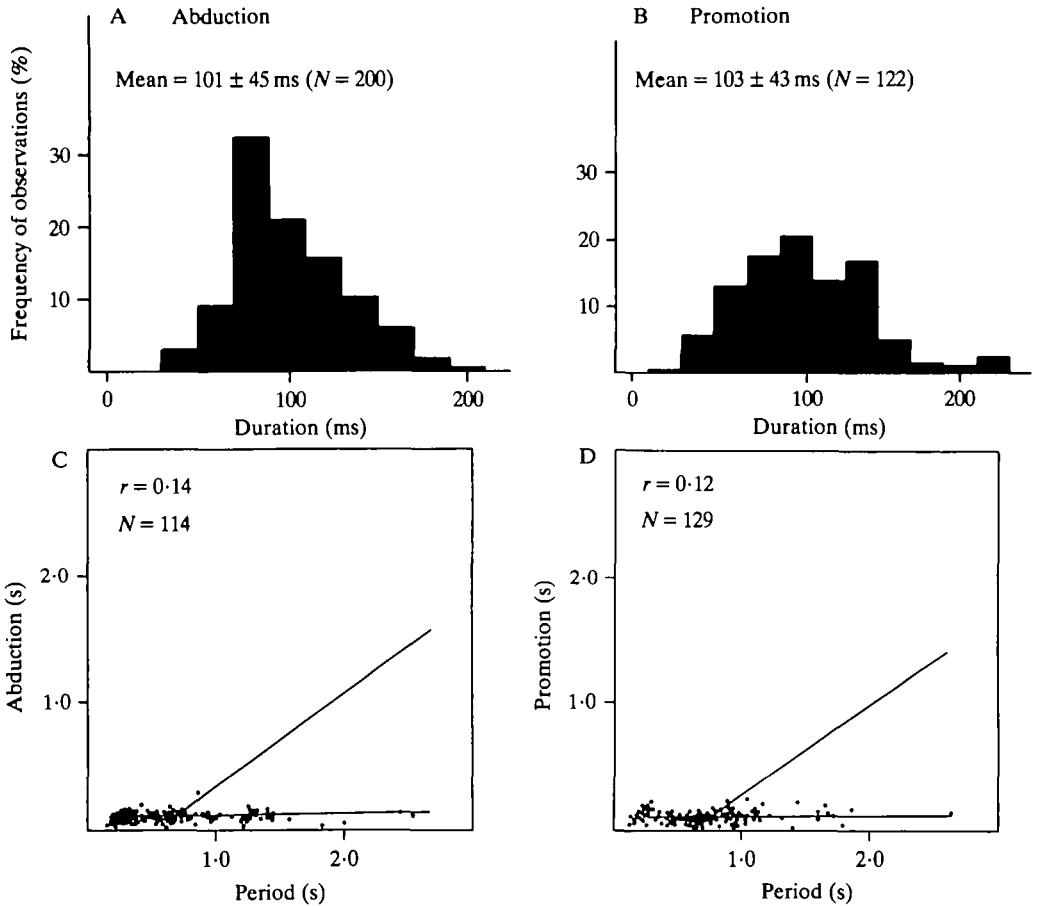


Fig. 3. Quantitative characteristics of the abductor and promoter activities. (A),(B) Histograms presenting the distribution of the abduction and promotion durations. The data are pooled from five animals for the abductors and three for the promoters (mean \pm S.D., number of observations). (C),(D) Correlation between the abduction/promotion and the period. r is the correlation coefficient, N is the sample population, and in each case the two regression lines are traced (see Materials and Methods).

were, for anatomical reasons, difficult to impale with the recording wires. Conversely, though easily accessible from its tergal origin, muscle 77-1a did not show any electrical activity, suggesting that only very few excitatory motor neurones terminate on the proximal fibres of this long muscle, from which activity was recorded.

The activity of the two studied muscles occurred in bursts, in each of which only one unit was identified (Fig. 4A,B). The bursts began abruptly, whereas in certain animals, the activity in muscle 77-1c terminated after a relatively long (half of the period) decrease of the frequency (peak value >120 Hz, Fig. 4B). Single or paired potentials sometimes occurred during the interburst period. The average and modal duration of the phasic burst (Fig. 4A) was 70 ± 45 ms, $N = 216$, and independent of the period (Fig. 4C,D). The double depressor burst always began after abductor

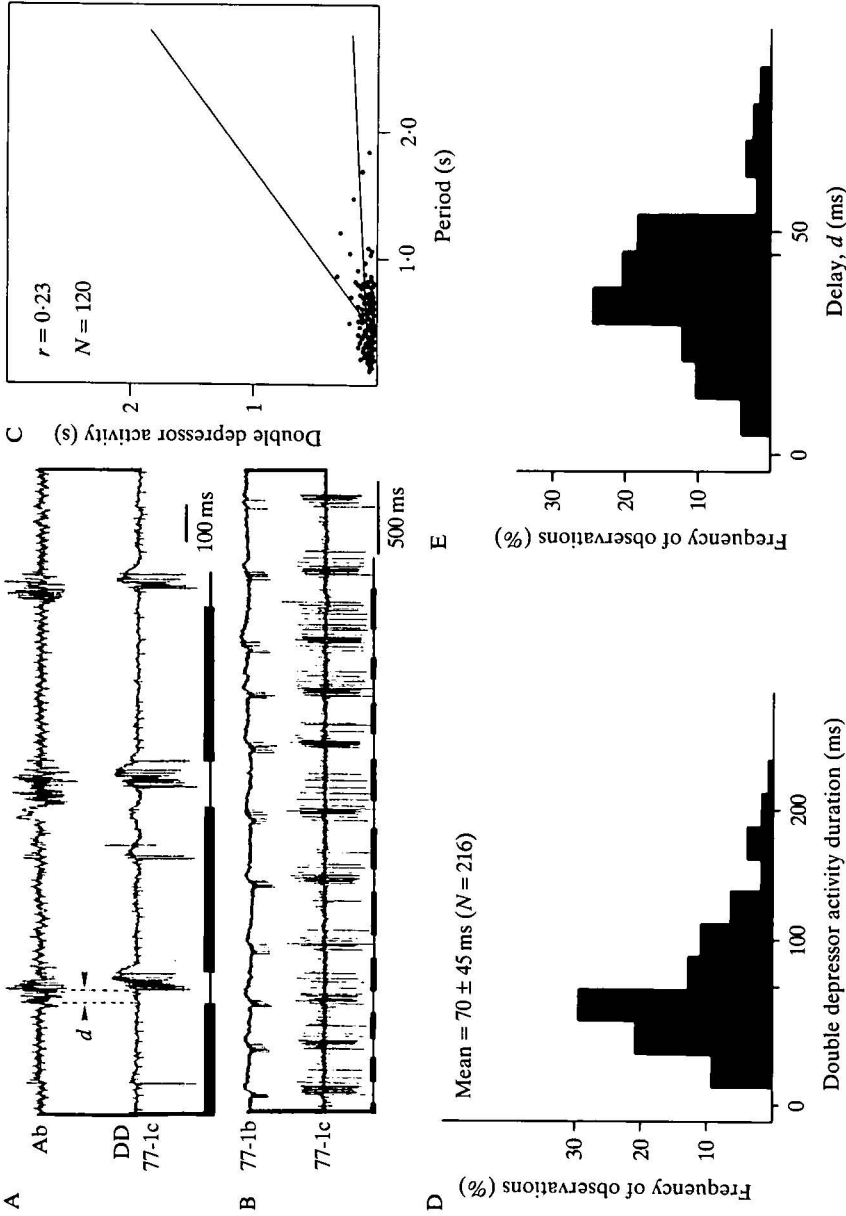


Fig. 4. Qualitative and quantitative analysis of the double depressor (DD) activity. (A) Simultaneous recordings from the abductors (Ab) and the double depressor 77-1c: the onset of the muscle 77-1c burst is delayed (d) relative to that of the abductors. The single unit seen in the double depressor recording occasionally fires in the second half of the interburst silence (end of the stance phase). (B) Simultaneous recordings from two double depressors, which are each activated by a different unit. (C) Correlation between the double depressor activity and the period duration. (D) Histogram presenting the distribution of the double depressor activity duration, pooled from five animals (mean \pm s.d., numbers of observations). (E) Histogram showing the distribution of the delay d , pooled from three animals: this delay hardly ever exceeds 55 ms.

activity had started. The delay between the two onsets (d , Fig. 4A) was 30–35 ms (Fig. 4E) whatever the period.

Activity in the remotor muscles

The electrical activity in the remotor muscles has been recorded from the lateral and medial subgroups. Three main patterns can be described.

(i) In the medial remotors, at least two units (potentials 1 and 2, Fig. 5A) were distinguished, both firing in two bursts restricted to the stance phase. No overlap between the activity of these two units and that of the phasic abductor–promotor burst was detected. An interburst interval (i , Fig. 5A), when almost no tonic activity could be seen, existed for step frequencies of less than 2 Hz and had a duration that was tightly coupled to the step period (correlation coefficient $r = 0.81$, Fig. 5F). For walking velocities greater than 2 steps s^{-1} , only a single burst was seen, whose firing rate increased until abruptly returning to zero. The interval between the end of remotor activity and the beginning of the next abductor burst showed little variability.

(ii) Other recordings, made in the medial and lateral remotors of different animals, showed sustained multi-unit activity during the whole stance phase (Fig. 5B,C). It is impossible, however, to establish whether a biphasic pattern was also present but masked by the overall activity. At least eleven excitatory motor neurones have been shown to innervate the remotors (Laurent & Richard, 1986) and, even if some are probably inactive during walking, the large number of different potentials recorded did not enable each to be reliably identified.

(iii) A third unit (potential 3, found so far only in the medial remotors) was occasionally recruited at the end (Fig. 5B) and, more rarely, at the onset of long-lasting periods of retraction. When active, this unit spiked from 1 to 10 times, with a peak frequency greater than 100 Hz. The first potential generally occurred about 30 ms after the end of the sustained activity of pattern ii, whereas the last one or two sometimes occurred during the first 50 ms of the next abductor–promotor bursts.

Over the entire range of velocities, the average duration of the overall activity in this complex remotor system was 480 ms (Fig. 5D), that is, approximately, three-quarters of the average period. This rate can be calculated because the remotion duration was linearly linked to the period ($r = 0.71$, Fig. 5E).

Activity in the intrinsic coxal depressors

We found two different units, and many different activation patterns, varying gradually with the walking velocity. There was no single step frequency at which there was a sharp transition between distinct patterns. Nevertheless, two broad patterns could be distinguished. First, when the step period was regular and less than 500 ms, a short (about 100 ms) burst of small potentials (1, Fig. 6A) was produced 50 ms ($50 \pm 25 \text{ ms}$, $N = 120$) after the abductor–promotor onset (Fig. 6A). This latency was never less than 20 ms. Rarely more than one to six large potentials (2, Fig. 6A) were produced when the smaller unit was at its highest frequency (Fig. 6D). A few isolated potentials of the small amplitude unit 1 were also seen

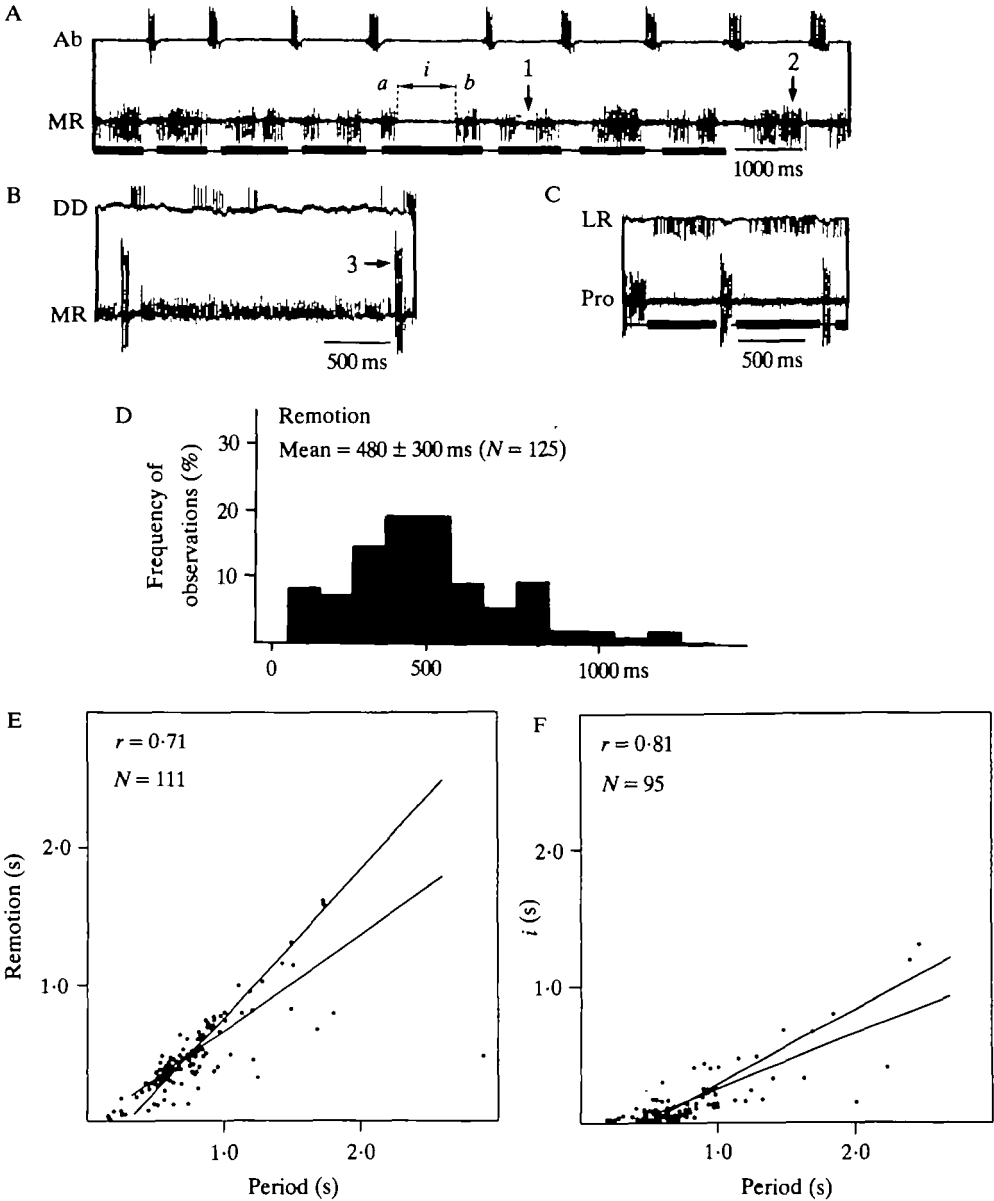


Fig. 5. Characteristics of the remotor activity. (A) Simultaneous recordings from the abductors (Ab) and medial remoters (MR): the units recorded in the remoters (1 and 2) fire in two bursts (a and b), delayed from each other by a variable latency (i). This latency decreases and disappears when the step frequency increases. (B) Another pattern of activity recorded in the medial remoters, during a long step cycle: a large amplitude unit fires at the end of a long stance phase, whilst the whole stance phase shows a sustained tonic activity of several small amplitude units; DD, double depressors. (C) Simultaneous recordings from the lateral remoters (LR) and promoters (Pro): the activity in the lateral remoters occupies the whole stance phase. (D) Histogram showing the distribution of the overall remotor activity; data pooled from three animals (mean \pm s.d., number of observations). (E) Correlation between remotion duration and period. (F) Correlation between the interburst latency, i (Fig. 5A), and the period.

during the stance phase (Fig. 6A); this pattern appeared to be very similar to that of the double depressors (see Fig. 4A). Second, during irregular walking and when the step period was greater than 750 ms, the pattern of activity was almost biphasic and usually involved the small unit only (Figs 6B, 10A). A first burst of potentials, similar to that occurring during rapid walking, was followed by a relatively quiet period (D in Fig. 6B) whose duration was coupled to the period ($r = 0.90$, Fig. 6E). A second burst then terminated the coxal depressor activity, some 100 ms before the next onset of the abductors (a , Fig. 6B,C). This latency was always greater than 30 ms, and independent of the period ($r = 0.25$, Fig. 6F). Many mixed and intermediate patterns were also seen for step frequencies ranging from 1 to 2 Hz: (i) the last step shown in Fig. 6A was rather long (1 s), but did not have a second burst of small potentials; (ii) all steps shown in Fig. 10A are of short duration (0.5 s), but some (the first and fourth) showed continuous activity of the small unit, even during most of the stance phase. It seems likely that these differences in the patterns were not only a function of the velocity of the animal, but of other parameters, such as its direction, which were not taken into account in these experiments. Moreover, when the styrofoam ball was put onto water, so that the animal did not have to support it, the activity in the coxal depressors (and the remotors) was reinforced (increase of the firing frequency) and often lasted for most of the stance phase (Fig. 7). This was interpreted as a consequence of surface tension forces that the animal had to counteract to make the ball move. The pattern of activity in the abductors, promoters and double depressors was not modified.

Activity in the coxal levators

Small potentials in the levators usually appeared at the end of double depressor activity, i.e. some 50 ms after the end of abductor activity (Fig. 8A). Levator activity usually ended with a burst of large potentials that appeared up to 50 ms before the next abductor onset (Fig. 8A,B), when all activity had stopped in the coxal depressors. The large potentials were sometimes recorded alone (Fig. 8B), which suggests that of the five levator muscles (Laurent & Richard, 1986) one might be supplied by these units only. The most striking feature of this composite pattern, however, was the short period of phasic coactivation (45 ± 25 ms) of the thoracocoxal abductors and the coxotrochanteral levators in the early part of the swing phase (Fig. 8B). The end of this final levator burst immediately preceded the onset of the depressor activity (Fig. 8A). The duration of the levator activity depended on the period ($r = 0.91$, Fig. 8C).

Overall activity during the step cycle

In Fig. 9, the instantaneous frequency has been plotted as a function of the phase for a 600-ms step cycle. The diagram is an average computed for 15 periods from several animals. The two units of the coxal depressors have been separated, but for clarity only the biphasic units of the remotors have been plotted. Three features are typical: (1) the long duration of the first burst in the slow coxal depressor unit; (2) the occurrence of a second burst in the two coxal depressor units; (3) the biphasic

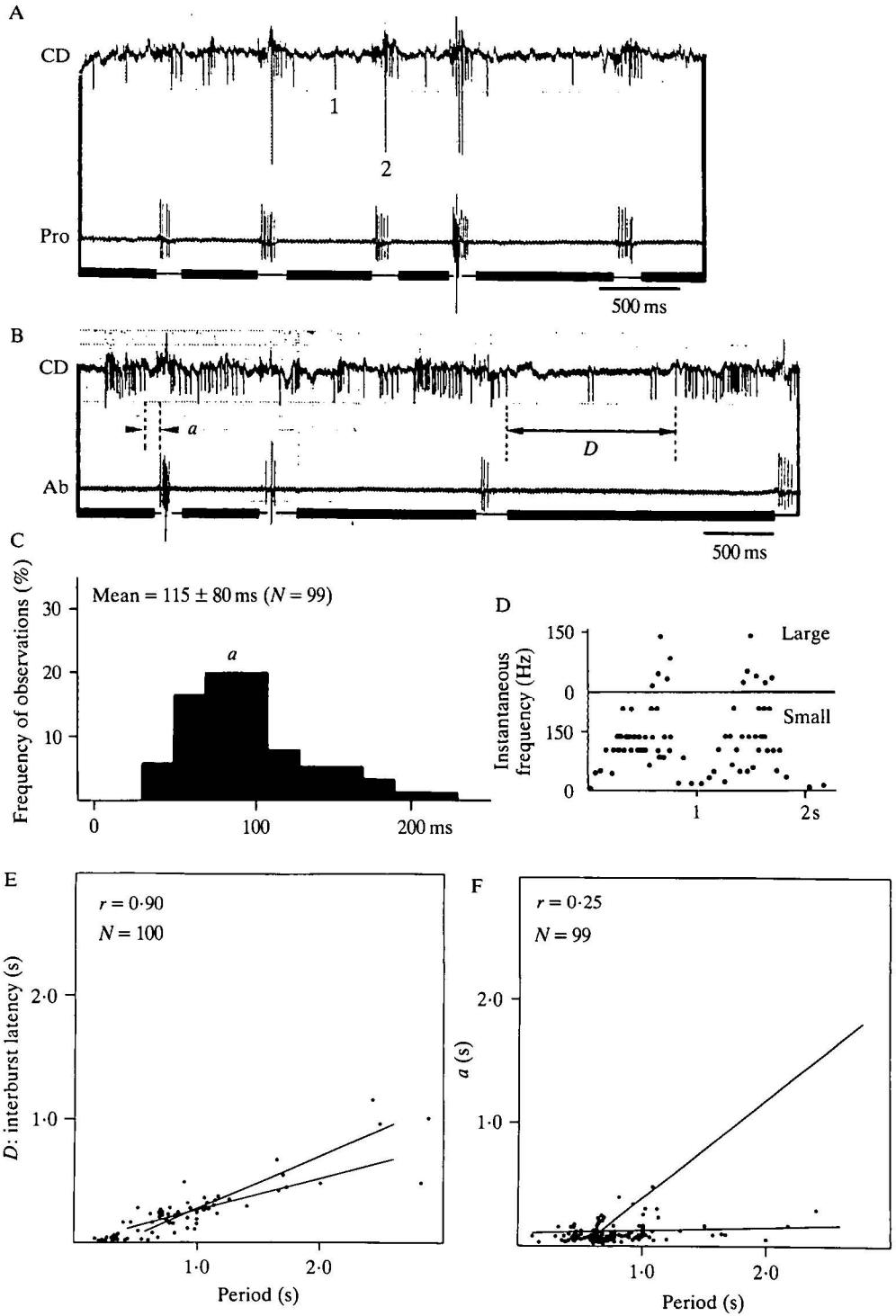


Fig. 6

pattern of activity in the remotors. For velocities greater than 2.5 steps s^{-1} , these three features did not usually appear, but the patterns of activity in the remaining muscles were not modified.

During slower stepping, however, the swing-to-stance phase and the reverse transitions showed an important difference: when the swing was initiated (beginning of the abductor activity), almost no overlap of excitation could be found between sets of anatomical antagonists (see Laurent & Richard, 1986).

At the thoracocoxal joint, the abductor and promotor activity was delayed until the remotor activity had ended [except in the rare cases when the fast unit was recruited (Fig. 5B)]; at the coxotrochanteral joint, the activity of the intrinsic and double depressors began only when the activity in the levators had ended (arrow, Fig. 9). Conversely, when the stance phase began, simultaneous activity was seen in the promoters and remotors as well as in the depressors and levators.

There was a short delay between the coxal and trochanteral stance-to-swing transition periods. The onset of activity in the two groups of depressors, and the end of that of the levators, was delayed by some 50 ms with respect to the onset of abductor activity (arrow, Fig. 9). However, there was an increase in firing frequency in the levators at the end of the stance phase, indicating that the swing phase might actually be initiated by the levators.

Comparison between locomotor activity and seeking electromyographic patterns

When the animal was deprived of its substrate, the forelegs and middle legs typically swept through the air in a way that was interpreted as a search for tarsal contact. During naturally interrupted locomotion, the animal would hold the ball with the meso- and metathoracic legs, and make similar 'seeking' or 'searching' movements with the forelegs for a few (1–4) seconds. These rapid movements comprised alternating flexion and extension of the axial joints combined with rotation about the thoracocoxal articulation. Neither swing nor stance phases could be defined since no contact was made with the substrate.

In Fig. 10, the abductor and coxal depressor activities in stepping (Figs 6, 10A) and seeking (Fig. 10B) are compared. Seeking had four characteristics: (1) its

Fig. 6. Activity in the coxal depressors. (A) Simultaneous recordings from the coxal depressors (CD) and the promoters (Pro) during regular and fast stepping; a short burst of potentials (one of small and one of large amplitude) overlaps with each burst of the promoters and stops at the beginning of the stance phase. During the second part of the stance phase, one or several small potentials occur in the depressors. (B) During irregular or slower stepping, the pattern of depressor activity is clearly modified: only the small unit fires, and its activity starts during the abductor (Ab) burst; it then goes on firing during the stance phase either regularly or, if the period increases, in a second burst restricted to the second half of the stance phase, and delayed from the first by a variable latency (*D*). Its activity totally stops before the beginning of the following swing phase (*a*). (C) Histogram showing the distribution of the depressor offset–abductor onset delay (*a*): *a* is never nil. Data pooled from three animals (mean \pm s.d., number of observations). (D) Instantaneous frequency plotted against time during two following steps in the small and the large units of the coxal depressors; the large amplitude unit is recruited when the small one reaches high frequencies. (E) Correlation between the interval *D* and the period. (F) Correlation between the interval *a* and the period.

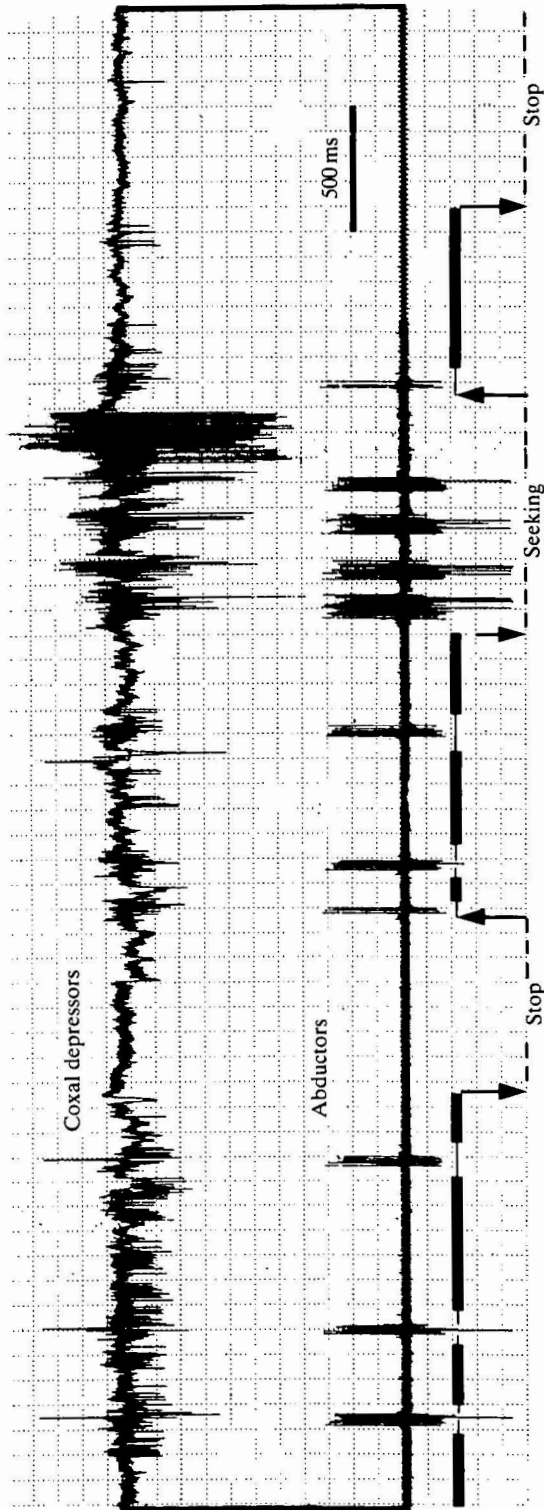


Fig. 7. Activity in the coxal depressors of the trochanter and the abductors during stepping, while the ball is being supported on the surface of water. Three steps are followed by a short stop, and the animal then starts again for another three steps. After four seeking movements (see Fig. 10), the animal makes a single step and stops. During stepping, the activity in the depressors is reinforced (see Fig. 6B for comparison), the firing frequency being increased.

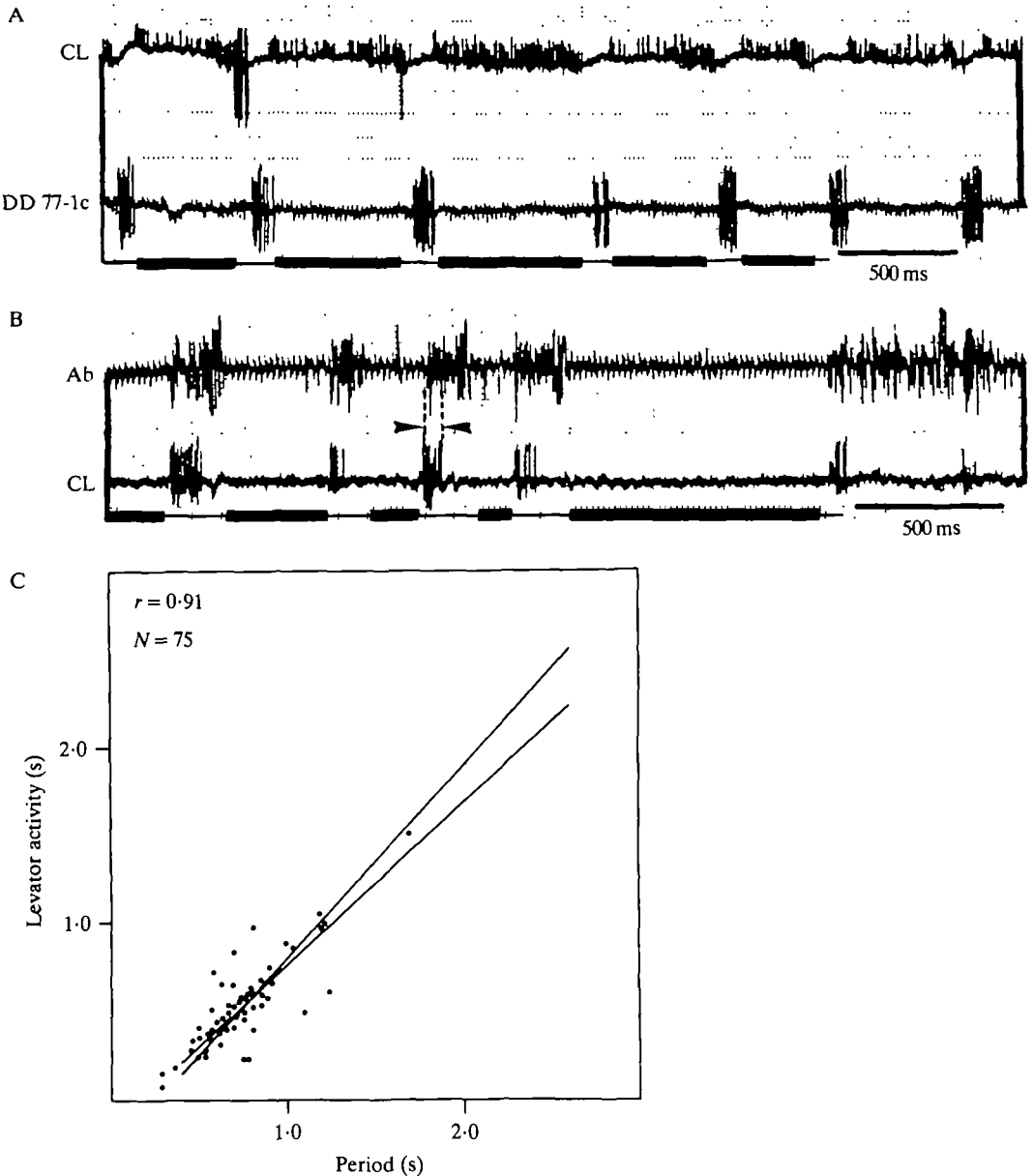


Fig. 8. Activity in the coxal levators (CL). (A) Simultaneous recordings from the levators and the double depressor (DD) 77-1c; in the levators, small units are tonically active during the whole stance phase whereas large amplitude potentials are only seen at the end of the small units' activity; the end of the large units' activity immediately precedes the onset of the double depressors: therefore, as shown in B, the terminal activity in the levators overlaps with that of the abductors at the beginning of the next swing phase. (C) Correlation between the levators' activity and the period.

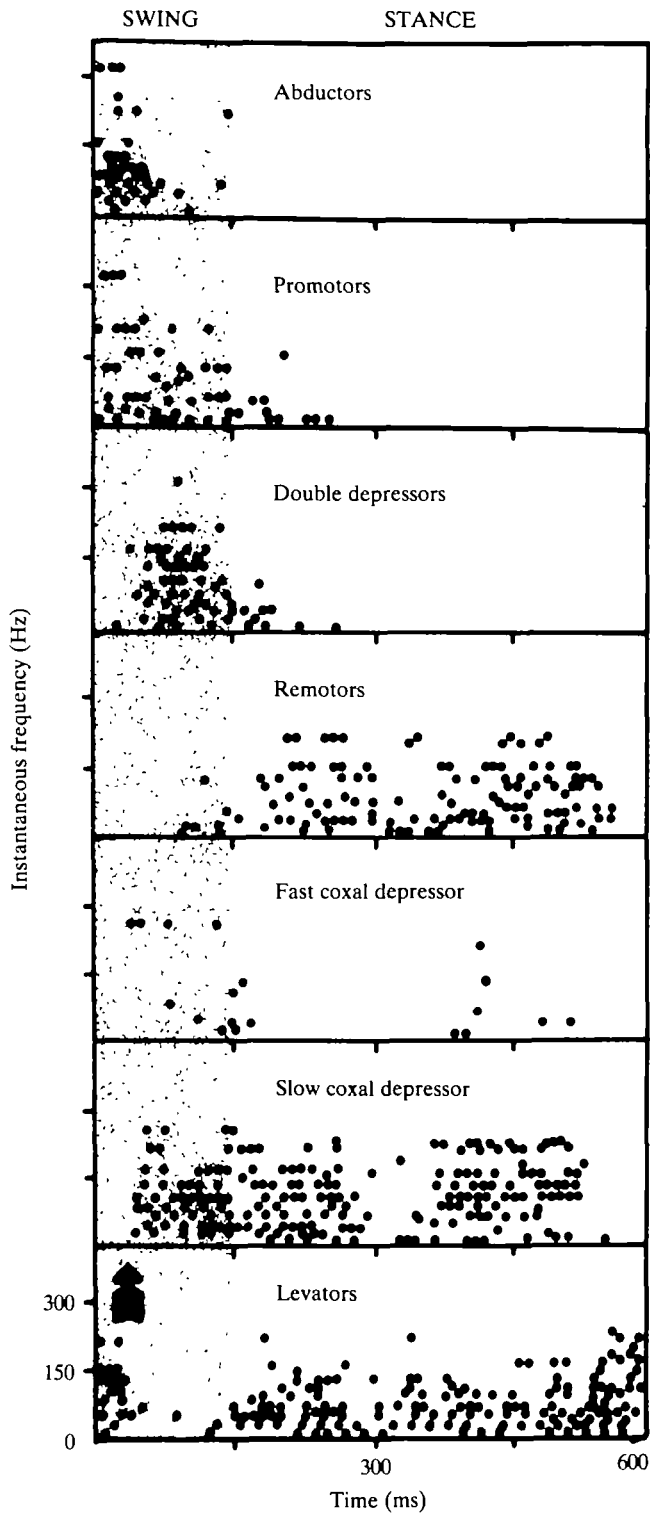


Fig. 9

movements occurred at higher frequency ($>2\text{Hz}$) than the step cycles (1.52Hz , Fig. 1A); (2) the small and the large intrinsic depressor units fired together; (3) a cycle was more or less symmetrical, in that the period of activity in either muscle was equal to the duration of the following quiet period; the latter corresponded to the activity of the antagonists. This was achieved by an increase of the abduction duration and also by a shortening of the following silence. During locomotion, the abductor muscles were active for only one-quarter to one-fifth of the period. (4) There was no depressor activity during the time between the abductor bursts, though this pattern reappeared when the leg touched the substrate (arrow, Fig. 10B).

In Fig. 10C, two steps are followed by a sequence of six seeking cycles: the remotors were activated in antiphase with the double depressors, and their level of activity was higher (recruitment of the fast units in particular) than during the preceding walking phase.

DISCUSSION

In our experiments the legs of the tethered crickets held a ball instead of supporting the animals' own weight, so that it might be suspected that the reversal of sign of the load afference (Pearson, 1972) could have modified the pattern of activity during walking (at least during the stance phase). Several arguments can be put forward against this. First, putting the ball onto water did not qualitatively alter the patterns of electrical activity; rather it resulted in an increase of general activity during the stance phase, probably due to the superimposed surface tension forces. Second, no reflex response to the movement of the substrate has been found in the promoters when the tarsus was brought back on the ball at the end of a swing phase. This indicates that, in contrast to a stick insect walking on a tread wheel (Graham & Wendler, 1981), the light (less than half the weight of a cricket) styrofoam ball had little inertia during walking. Third, the centre of gravity of the animal is somewhere between the meso- and metathoracic coxae (Harris & Ghiradella, 1980), and hence the front pair of legs probably normally plays a less important role in bearing the animal's weight.

Fig. 9. Instantaneous frequency analysis of the general muscular activity, in a 600-ms step cycle, computed from 15 steps in different animals (see Materials and Methods). The activity in the three muscle groups active during the swing phase (abductors, promoters and double depressors) is followed by the activity in the remotors during the stance phase. The coxal depressors become active simultaneously with the double depressors, but their activity goes on during the stance phase, stopping eventually before the next swing phase. The levators become active at the end of the swing phase, and go on firing till the first third of the next swing phase: consequently, there is a co-contraction of the two coxotrochanteral antagonists during most of the stance phase. At the end of the latter, however, the coxal depressors are turned off whilst the levators increase their activity, and the legs can be lifted and brought forwards by the contraction of the abductor muscles. Note that the stance-to-swing phase transition, which occurs at the beginning of the abductor/promotor activity for all thoracocoxal muscles, occurs later (arrow) in the thoracotrochanteral (double depressor) or coxotrochanteral (intrinsic depressor and levator) muscles.

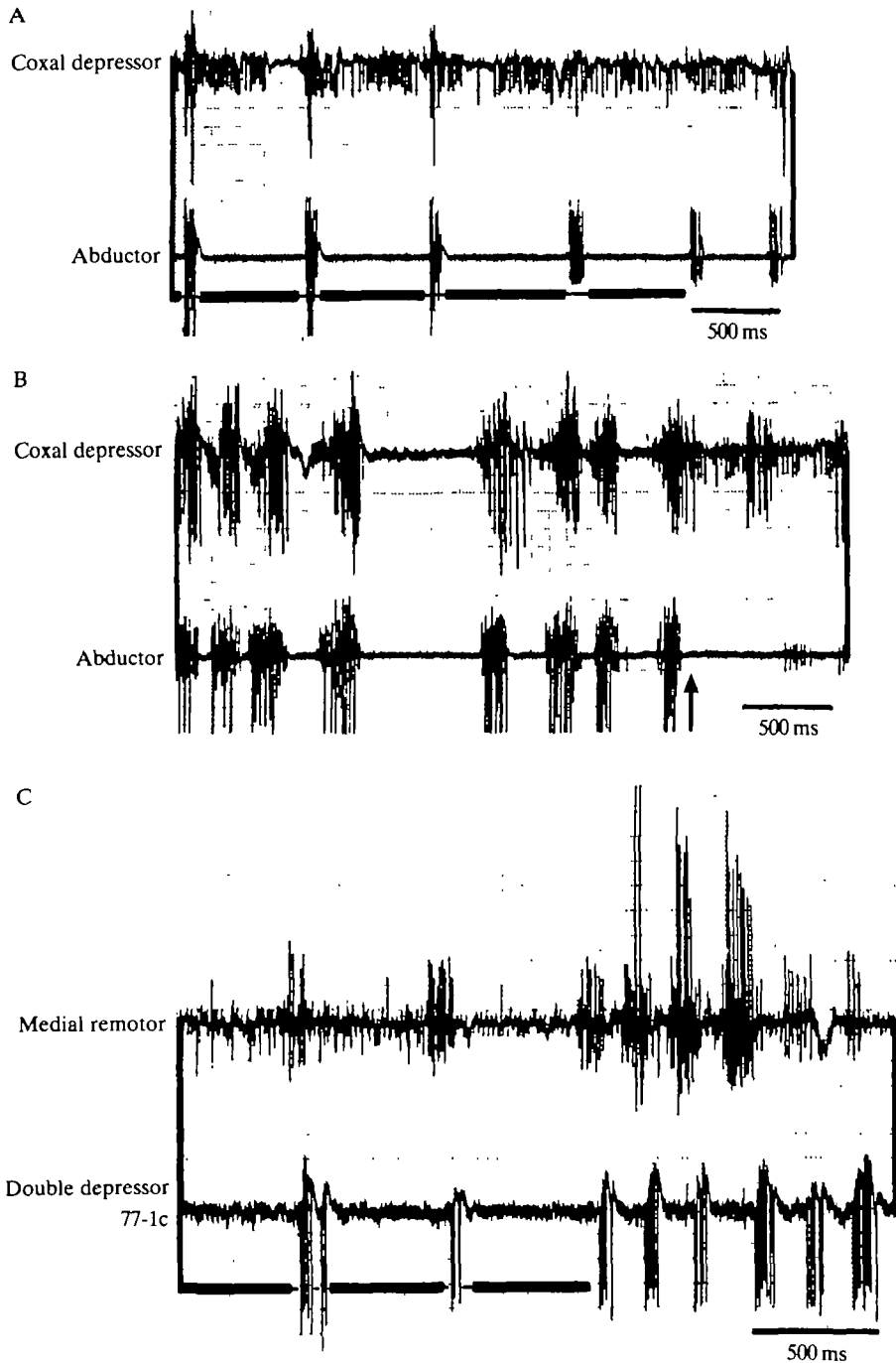


Fig. 10. Comparison of the patterns of muscular activity between stepping and 'seeking' movements. (A) Simultaneous recordings from the coxal depressors and the abductors during stepping. In B, the same animal holds the ball with its meso- and metathoracic legs, whilst the two prothoracic legs make two series of four seeking movements, before reaching back to the substrate (arrow). The periods of these movements are short and the coxal depressors burst only once, in phase with the abductors. In C, two different muscles are recorded from another animal which performs the same type of behaviour: two normal steps are followed by six seeking movements, during which the medial remotors and the double depressor 77-1c are activated in antiphase. The depressor bursts show no change, whereas the remotor activity is shortened by its first two-thirds.

With the animal having to move the ball instead of the body, the major difference from natural walking was that the sign of the imposed vertical load to the legs was reversed. Therefore, consequences in the electromyographic patterns should mainly be seen during the forelegs' stance phase, i.e. when the animal had to support the ball. The muscles active during the stance phase have been shown to be the coxal depressors, the remotors and the levators. Since the coxal depressors extend the femur, their contraction would push the ball away rather than carry it; their activity during the stance phase is therefore unlikely to depend upon the need to support the ball. Because the remotors act about the transverse axis of the coxa (Laurent & Richard, 1986), their contraction is also difficult to explain as a means to retain the ball. The levators are the muscles whose activation during the stance phase is the easiest to relate to the necessity for retaining the ball. However, as discussed above, this activity was not diminished by putting the ball on water, which removed the need for the animal to support it. This suggests that the long levator activity during the stance phase was not heavily influenced by the load. However, it is possible that the pattern of activity in this muscle would differ if the animal were freely walking on a flat surface.

The present observations of muscle activity during locomotion enable the following conclusions to be drawn.

Electrical activity and motion

During a step cycle, the durations of the abductor, promotor and double depressor bursts always correspond to the swing phase, which does not depend on the period. This result agrees with previous observations in the lobster (Clarac & Chasserat, 1983), the scorpion (Bowerman, 1981*b*), the free-walking adult stick insect (Graham, 1972), the cockroach (Delcomyn, 1971; Pearson, 1981), the locust (Burns & Usherwood, 1979) and the cricket (Harris & Ghiradella, 1980), where the swing is approximately constant over the whole range of speeds. In the cockroach, this constancy has been demonstrated to depend partly on afferents from the trochanteral hairplates (Wong & Pearson, 1976), whose role is to signal that the required amplitude of the swing phase has been reached. This would also explain the small observed variability in leg position at the end of protraction, compared to that at the end of retraction.

During the stance phase, however, the number of active muscles and their pattern of activity depend on the velocity and the regularity of locomotion. During fast locomotion, only the levators and remotors reach a high rate of discharge, and there is usually no second burst in the coxal depressors. During slower walking, however, there is a long period of coactivation in the coxal levators and depressors during the stance phase. Similarly, in the locust prothoracic leg, the beginning of the stance phase is marked by a coactivation of the slow extensor tibiae motor neurone and the tibial flexors (Burns & Usherwood, 1979); these authors interpret this co-contraction as an assistance to posture by an increased rigidity of the leg. It is possible, however, that a simultaneous coactivation of the depressors and levators trochanteris and extensor and flexor tibiae would result, at least during the early stance phase, in the

actual propulsive stroke in which the remotors probably provide the necessary rotative forces about the thoracocoxal joint.

The termination of the coxal depressor burst at the end of the stance phase probably releases the tension developed by the levators which continue to fire for some 70–90 ms and are generally assisted by the recruitment of their faster units. This is likely to result in the *actual* initiation of the swing phase, by lifting the leg off the substrate. The abductors and promotors are then activated and bring the whole leg forward. The activation of the two sets of depressors follows shortly (after some 50 ms) and the tarsus is consequently brought to the substrate for another power stroke.

Swing-to-stance and reverse transition

It has been shown (Fig. 9) that some anatomical antagonists (promotors *vs* remotors, depressors *vs* levators) are coactive during the swing-to-stance transition period, whilst such simultaneous recruitment was not found during the reverse transition. This asymmetrical pattern has also been found in the locust prothoracic flexors and extensors tibiae (Burns & Usherwood, 1979), in the stick insect mesothoracic promotors and remotors (Graham & Wendler, 1981) and in the scorpion trochanter–femur depressors and levators (Bowerman, 1981*a,b*). This probably results from the fact that bringing the leg back to the substrate, which occurs in the Anterior Extreme Position (AEP, Cruse, 1979), needs precise reflex adjustments of muscular tension: these are likely to be achieved by subtle and fine balancing of activity in the sets of antagonists that fire during this swing-to-stance transition period.

As well as proprioceptors in the femorotibial joint of the locust mediate resistance reflexes onto the flexor and extensor tibiae motor neurones (Field & Burrows, 1982), it must be expected that similar pathways exist between the sensory and motor units of the more proximal joints. In the locust metathoracic coxotrochanteral articulation, trochanteral hair plates and rows of hairs are sensitive to levation (Bräunig & Hustert, 1985*a,b*) and they increase the firing frequency of a depressor. In the locust mesothoracic thoracocoxal articulation, position and movement are encoded by an elaborate system of at least five types of proprioceptor which converge onto coxal motor neurones (Hustert, 1982, 1983). Though there is as yet no direct evidence, it is very probable that similar sense organs control the position and movement of the cricket prothoracic coxa and trochanter.

The depressor system

The activation pattern of the two-part depressor system depends on the period and the regularity of walking; it is also thought to depend upon other parameters such as the direction of walking. During fast and regular stepping or during the short periods of seeking movements, the coxal and double depressors are both activated some 50 ms after the abductors and promotors, and burst in the swing phase, spilling into the beginning of the stance phase. During slow or irregular stepping, only the coxal depressors continue firing in the second half of the stance phase. This suggests that

the coxal depressor motor neurones probably receive a great deal of sensory information during a step cycle since the motor neurones (the small unit mainly) show a firing pattern relatively independent with respect to the others, and very variable with the locomotor pattern. These sensory inputs are probably common to the slow and fast motor neurones, since the fast motor neurone is recruited mainly when the slow motor neurone reaches high rates of discharge. Another piece of evidence is that their central arborizations are in close contiguity in the dorsal neuropile (Laurent & Richard, 1986). Since the patterns of discharge are dependent upon the step frequency, it is possible that rapid movements (such as seeking or escape running) would involve a rather rigid central programme, whereas slower and irregular locomotion would be highly dependent on precise sensory readjustments.

A better understanding of prothoracic locomotion in the cricket will now rely on: (1) the description of the electromyographic patterns during walking on a flat surface; (2) the identification of the sense organs that monitor the position and movement of the proximal leg segments and (3) the search for a population of premotor interneurons such as those which control movement of the metathoracic tibiae and tarsi of the locust (Burrows, 1980).

We are grateful to Malcolm Burrows, Kay Seymour and Alan Watson for their many helpful comments on the draft and to Fiona Howarth for typing the manuscript. We thank Professor Bessou and Dr Leitner for kindly letting us use their electrostatic printer. This work has been supported by an ATP '*Dynamique du neurone et des ensembles neuronaux*' from the Ministère de l'Industrie et de la Recherche.

REFERENCES

- BOWERMAN, R. F. (1981a). An electromyographic analysis of the elevator/depressor muscle motor program in the freely walking scorpion, *Paruroctonus measensis*. *J. exp. Biol.* **91**, 165–177.
- BOWERMAN, R. F. (1981b). Arachnid locomotion. In *Locomotion and Energetics in Arthropods* (ed. C. F. Herried II & C. R. Fourtner), pp. 73–102. New York, London: Plenum Press.
- BRÄUNIG, P. & HUSTERT, R. (1985a). Actions and interactions of proprioceptors of the locust hind leg coxo-trochanteral joint. I. Afferent responses in relation to joint position and movement. *J. comp. Physiol.* **157A**, 73–82.
- BRÄUNIG, P. & HUSTERT, R. (1985b). Actions and interactions of proprioceptors of the locust hind leg coxo-trochanteral joint. II. Influences on the motor system. *J. comp. Physiol.* **157A**, 83–89.
- BURNS, M. D. (1973). The control of walking in Orthoptera. I. Leg movements in normal walking. *J. exp. Biol.* **58**, 45–58.
- BURNS, M. D. & USHERWOOD, P. N. R. (1979). The control of walking in Orthoptera. II. Motor-neurone activity in normal free-walking animals. *J. exp. Biol.* **79**, 69–98.
- BURROWS, M. (1980). The control of sets of motoneurons by local interneurons in the locust. *J. Physiol., Lond.* **298**, 213–233.
- CLARAC, F. & CHASSERAT, C. (1983). Quantitative analysis of walking in a decapod crustacean, the rock lobster *Jasus lalandii*. I. Comparative study of free and driven walking. *J. exp. Biol.* **107**, 189–217.
- CRUSE, H. (1979). A new model describing the coordination pattern of the legs of a walking stick insect. *Biol. Cybernetics* **32**, 107–113.
- CRUSE, H. & SAXLER, G. (1980). The coordination of force oscillations and of leg movement in a walking insect *Carausius morosus*. *Biol. Cybernetics* **36**, 165–171.
- DELCOMYN, G. (1971). The locomotion of the cockroach *Periplaneta americana*. *J. exp. Biol.* **54**, 443–452.

- FIELD, L. H. & BURROWS, M. (1982). Reflex effects of the femoral chordotonal organ on leg motor-neurons of the locust. *J. exp. Biol.* **101**, 265–285.
- GRAHAM, D. (1972). An analysis of walking in the first instar and adult stick insect *Carausius morosus*. *J. comp. Physiol.* **81**, 23–52.
- GRAHAM, D. (1977). Stimulation of a model for the coordination of leg movement in free walking insects. *Biol. Cybernetics* **26**, 187–198.
- GRAHAM, D. & WENDLER, G. (1981). Motor output to the protractor and retractor coxae muscles in stick insects walking on a treadwheel. *Physiol. Entomol.* **6**, 161–174.
- HARRIS, J. & GHIRADELLA, H. (1980). The forces exerted on the substrate by walking and stationary crickets. *J. exp. Biol.* **85**, 263–279.
- HOYLE, G. & BURROWS, M. (1973). Neural mechanisms underlying behaviour in the locust *Schistocerca gregaria*. I. Physiology of identified motor neurons in the metathoracic ganglion. *J. Neurobiol.* **4**, 3–41.
- HUSTERT, R. (1982). The proprioceptive function of a complex chordotonal organ associated with the mesothoracic coxa in locusts. *J. comp. Physiol.* **147**, 398–399.
- HUSTERT, R. (1983). Proprioceptor responses and convergence of proprioceptive influence on motoneurons in the mesothoracic thoraco-coxal joint of locusts. *J. comp. Physiol.* **150**, 77–86.
- LAURENT, G. & RICHARD, D. (1986). The organization and role during locomotion of the proximal musculature of the cricket foreleg. I. Anatomy and innervation. *J. exp. Biol.* **123**, 255–283.
- PEARSON, K. G. (1972). Central programming and reflex control of walking in the cockroach. *J. exp. Biol.* **56**, 173–193.
- PEARSON, K. G. (1981). Function of sensory input in insect motor systems. *Can. J. Physiol. Pharmacol.* **59**, 660–666.
- PEARSON, K. G. & ILES, J. F. (1973). Nervous mechanisms underlying intersegmental coordination of leg movements during walking in the cockroach. *J. exp. Biol.* **58**, 725–744.
- WILSON, D. M. (1966). Insect walking. *A. Rev. Ent.* **11**, 103–122.
- WONG, R. K. S. & PEARSON, K. G. (1976). Properties of the trochanteral hair plate and its function in the control of walking in the cockroach. *J. exp. Biol.* **64**, 233–249.