

DISCHARGES OF PYRAMIDAL TRACT AND OTHER MOTOR CORTICAL NEURONES DURING LOCOMOTION IN THE CAT

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SUMMARY

1. A method is described for chronically implanting fine flexible microwires into cat motor cortex, which permitted extracellular recordings to be made from 165 single neurones. Most units were recordable for 12 h and some for up to 2 days.

2. Of the neurones tested, 57% were shown to project to the medullary pyramid (pyramidal tract neurones, p.t.n.s). Antidromic latencies corresponded to a range of conduction velocities from 63 to 9 m/s.

3. In the animal at rest neurones discharged at rates from 0.5 to 44 impulses/s.

4. During locomotion at 0.5 m/s (a slow walk) 56% of cells discharged faster than at rest and 80% showed frequency modulations time-locked to the step cycle. Most fired one discrete burst of impulses per step or one peak period superimposed on a maintained discharge. In different cells peak activity occurred at widely different times during the step cycle. A few cells peaked twice per step.

5. Peak rates (averaged over twenty steps) ranged from 10 to over 120 impulses/s, the values for most slow-axon p.t.n.s (conduction velocity < 21 m/s) being lower than for any of the fast-axon p.t.n.s.

6. For locomotion at speeds between 0.37 and 1.43 m/s a roughly linear relationship existed between discharge rate and speed in 14% of cells. However, the changes were modest and in most cells both mean rate and peak rate were unrelated to speed. In some cells discharge phasing was fixed (relative to the step cycle in the contralateral forelimb); in others there were progressive phase shifts (or more complex changes) as speed increased.

7. During locomotion up a 10° incline discharge phasings were the same as on the flat in all of the twenty-seven neurones studied and most showed no substantial change in mean rate or peak rate (although there were substantial increases in limb muscle electromyogram amplitudes).

INTRODUCTION

Investigations of the central nervous mechanisms contributing to locomotor control in the cat have demonstrated that neural circuits or 'pattern generators' capable of maintaining stepping movements of the limbs exist within the spinal cord

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(see Grillner, 1974). However, studies using high decerebrate preparations capable of walking on a moving belt have provided evidence for substantial brain stem contributions to locomotor control. Orlovsky has shown, for example, that Deiter's neurones projecting via the lateral vestibulo-spinal tract to the lumbo-sacral cord discharge rhythmic bursts of impulses which are phase-locked to the locomotor cycle in the ipsilateral hind limb (Orlovsky, 1972*b*). Furthermore, lesions of Deiter's nucleus abolish or severely disrupt the limb movements (Orlovsky, 1972*a*).

Supraspinal influences on locomotion do not originate only from Deiter's nucleus: rubro-spinal (and reticulo-spinal) neurones also discharge rhythmically during walking movements (Orlovsky, 1970, 1972*c*). However, the red nucleus seems not to play an essential role since its destruction does not prevent locomotion, though there is some loss of movement precision (Orlovsky, 1972*a*).

Regarding the role of the motor cortex in locomotion much less is known, although the fact that decerebrate preparations are capable of locomotor movements must imply that it plays a facultative rather than an obligatory role. This is not, however, to argue it is unimportant. Indeed, Orlovsky (1972*a*) has shown that electrical stimuli applied to the medullary pyramid can interrupt and reset the hind-limb step cycle in the decerebrate cat. Such actions could not be duplicated from either Deiter's or the red nucleus, where stimulation influenced only the amplitude and not the timing of locomotor electromyograms (e.m.g.s) in hind-limb muscles (but cf. Russell & Zajac, 1979).

To obtain further information regarding the possible role of the motor cortex we have used chronically implanted microwire electrodes to study the discharge patterns of individual motor cortical neurones in cats trained to walk steadily at controlled speeds on a moving belt. Pyramidal tract neurones (p.t.n.s) have been identified by their antidromic responses to stimulation of the medullary pyramid. Approximate axonal conduction velocities have been calculated and this has given information regarding the selectivity of the micro-electrodes towards fast-axon and slow-axon p.t.n.s (cf. Evarts, 1965; Takahashi, 1965).

In agreement with a brief report by Palmer, Marks, Bak & Pedersen (1980) we find that many p.t.n.s (and other cortical neurones) discharge rhythmically during locomotion. The present paper mainly describes the discharges during walking at 0.5 m/s and the emphasis is on firing rates; discharge timings are described more extensively in the following paper (Armstrong & Drew, 1984). In addition, because Orlovsky (1973*b, c*) has found that neurones in brain-stem motor centres (i.e. vestibulo-spinal and rubro-spinal neurones) discharge more vigorously when the forcefulness of locomotion increases, the discharges have also been studied at other walking speeds and during locomotion on an uphill gradient. In view of previous reports that fast-axon and slow-axon p.t.n.s in the monkey discharge differently in relation to signal-initiated arm movements (Evarts, 1965; Lewis, 1974) the activities of these two categories of neurone have been compared when possible.

Two preliminary reports have been made (Armstrong & Drew, 1980*a, b*).

METHODS

In a preliminary experiment extracellular recordings could be obtained from single neurones in the cat motor cortex when glass-insulated tungsten micro-electrodes were advanced through the intact dura using a miniature micromanipulator mounted on the skull (cf. Evarts, 1965; Armstrong & Rawson, 1979). However, during vigorous locomotion some units were suddenly lost whilst others showed fluctuations in spike height. Because of these indications that recording conditions were unstable we turned to the use of chronically implanted microwire electrodes introduced individually into the cortex via a temporary cranial defect which was sealed after electrode insertion. Chronically implanted microwires have previously been employed for single-unit recording in the visual cortex of cats by Burns, Stean & Webb (1973) and in the motor cortex of monkey by Schmidt, Bak & McIntosh (1976): our method is essentially similar though our electrodes are simpler to fabricate.

Animals

Recordings were obtained from fifteen purpose-bred young adult cats which were contented and confident in the laboratory. They were trained without aversive techniques to walk for brief periods on a belt moving at controlled speeds between 0.37 and 1.43 m/s. Occasionally a pacing or a trotting gait was adopted but all the reported observations were made during periods when the animals used their usual walking gait in which the homologous limbs were 180° out of phase and the homolateral limbs were also out of phase, with hind limb preceding forelimb footfall by approximately 60°.

Surgery

All surgical procedures were carried out with full aseptic precautions and with general anaesthesia induced and maintained with pentobarbitone sodium (initial intraperitoneal dose 40 mg/kg; Sagatal; BDH). Post-operative recovery was uneventful and was speeded by placing the animals in a humidicrib. Recordings were begun 3 days after operation.

At operation, a small craniotomy was made and a bipolar electrode was inserted through the cerebellum and brain stem to reach the right medullary pyramid at stereotaxic plane AP-7. The electrode comprised two Teflon-insulated stainless-steel wires glued together with the tips staggered vertically by 1 mm. This craniotomy was closed with dental acrylic cement and sockets attached to the wires were cemented to the skull (cf. other electrodes below).

The right pericruciate cortex was exposed and up to five strips of nylon contact strip (Amphenol) were cemented to the skull nearby. Each strip carried five miniature sockets to each of which was soldered a 2–4 cm length of platinum-iridium wire 17 μm in diameter and insulated with Teflon (Clark Electromedical Instruments, 101R-IT). Each wire was grasped with watchmaker's forceps coated with Silastic rubber and under stereomicroscopic control pushed firmly through the intact pia to a depth of around 1.5 mm. The entry point was recorded on a photograph of the brain surface.

In addition, a bundle of six wires was inserted into the medial part of the posterior sigmoid gyrus to depths of 3–9 mm to sample neurones in the posterior wall of the cruciate sulcus. These bundles were prepared by temporarily glueing the wires to a fine tungsten introducer with polyethylene glycol. For full details see Palmer (1978).

After implantation of all cortical electrodes the pia was covered with pledgets of fibrin foam (Sterispon; Allen & Hanbury) and the craniotomy was sealed with acrylic cement. A small socket for an F.E.T. preamplifier was mounted behind the contact strips and the animal was earthed via a stainless-steel wire implanted under temporalis muscle.

A pair of recording leads constructed from nylon-insulated multi-strand stainless-steel wire (Bergen; Lodi, New Jersey) was inserted into each of several flexor and extensor muscles in each forelimb and the left hind limb. The muscles always included triceps brachii and brachialis in the left (i.e. contralateral) forelimb and vastus lateralis in the left hind limb. The e.m.g. leads were connected subcutaneously to sockets mounted on the skull.

Finally, additional acrylic was applied on the skull to produce a smooth boat-shaped head-piece (total weight 30 g) around which the skin margins were drawn up and sutured.

Recording

After post-operative recovery each cortical electrode was screened daily for single-unit action potentials for up to 6 weeks. When a unit was present recording was continued, otherwise the animal was returned to a large pen.

Preamplifier output was fed to a main amplifier via a flexible lead and was bandpass filtered (1 kHz to 20 kHz). Signals were displayed on an oscilloscope and stored on a Racal Store 7D tape-recorder. Neuronal activity was accepted as originating from a functional single unit only when the signal to noise ratio was more than 2:1 (usually > 3:1), when the interval between successive spikes was always > 1 ms and when spike height and wave form were invariable. The e.m.g. signals were led from the skull connectors via lightweight cables to a differential amplifier and were high-pass filtered (> 100 Hz) before oscilloscope display and tape storage.

Most cortical units were tested for the presence of an antidromic action potential in response to stimulation of the medullary pyramid with 0.1 ms pulses. Cells identified as p.t.n.s gave an action potential which displayed constant threshold current (T), invariant latency and the ability to follow a train of four stimuli of intensity $2T$ and frequency 1000 Hz. The average straight-line distance from pyramid to pericruciate cortex in three cats similar in size to the experimental animals was 44 mm.

Units usually emerged from a background of multi-unit activity or from a quiet background and persisted for 6–18 h before gradually decreasing in size. In general, units encountered late in the life of the preparation were more durable, often lasting a day or more (maximum 4 days). The growth and decay of the signals presumably resulted from small relative movements between electrode tip and signal source (cf. Burns *et al.* 1973; Schmidt *et al.* 1976). Since central neurones are mechano-sensitive the presence of such movement naturally raises the question as to whether the locomotor movements of the animal might produce 'spurious' discharges. We believe this did not occur: spike amplitudes never displayed any short-term fluctuations time-locked to the step cycle or to any other movements, nor did jumping, grooming or feeding movements lead to any injury discharge or loss of units.

Histological study of the electrode tracks (see below) indicated that any electrode movements were small. Each track was short and little wider than the microwire, being marked only by a minimal gliosis. Nevertheless, the phenomenon of electrode 'drift' was useful since many electrodes recorded successively from two or more cells (up to seven) which were presumably close together. When these were p.t.n.s their individuality was confirmable through their different antidromic thresholds and latencies; non-p.t.n.s recorded on a single electrode were identified from spike shape, over-all discharge rate, peripheral receptive field (see Armstrong & Drew, 1983) and discharge pattern during locomotion. Two discriminable units were very occasionally recorded simultaneously.

Unit discharges and e.m.g. were recorded whilst the animal sat and/or stood quietly and also during periods when it walked steadily to maintain constant position on a moving belt for at least twenty steps at each speed used.

Data analysis

E.m.g. signals. For each bout of walking in which cortical discharges were recorded the e.m.g. signals from the muscles studied were digitized, full-wave rectified and displayed on a visual display unit (Tektronix 611) provided with moveable cursors. These were used to measure the times of onset and offset of activity in each muscle relative to the start of the step cycle, which was taken by convention as occurring at the onset of activity in the lateral head of triceps brachii, an elbow extensor which produces one well-defined burst of activity per step cycle. The values for the individual steps were computer-averaged and print-outs showed the mean times and their standard deviations.

To determine the temporal correlation existing between the e.m.g. activities and the mechanical events of the step cycle cine films at 64 frames/s were taken during numerous bouts of steady walking in six of the animals and pulses synchronous with the opening of the camera shutter were taped along with the e.m.g. signals. The results will be presented in full elsewhere (D. M. Armstrong & T. Drew, unpublished), but it can be noted here that they were highly reproducible both within and between cats. During locomotion at all speeds and in all cats footfall in the forelimb occurred 1–2 frames after the onset of e.m.g. in the lateral head of triceps brachii (i.e. with a delay of 15–30 ms) whilst foot lift occurred 2–3 frames after the onset of activity in the elbow flexor brachialis (delay 30–45 ms). During locomotion at 0.5 m/s, when the mean duration of the step cycle was *ca.* 850 ms, the onset of brachialis e.m.g. occurred *ca.* 80 ms after the end of activity in triceps brachii, but this interval decreased progressively as speed increased reaching a value of *ca.* 25 ms at 1.43 m/s (step duration *ca.* 400 ms). Triceps activity lasted for 56% of the step cycle at 0.5 m/s decreasing slightly to 54% at 1.43 m/s.

Discharges of cortical neurones. Taped action potentials were discriminated with a voltage window; its output was always compared with the original data by photographing both signals from an oscilloscope screen. Discriminator output pulses and e.m.g. signals were displayed using an ink-jet recorder (Mingograf; Elema-Schonander) and also analysed with a digital computer (PDP 11/34; DEC). The computer was programmed to yield interspike interval histograms together with values for mean interspike interval and its reciprocal (referred to below as mean rate). A minimum of 1000 successive interspike intervals was used.

Programs were also available to produce displays which facilitated quantitative analyses of unit discharges during locomotion. These included post-event time histograms in which the spikes during twenty successive paces were overlap-averaged. Moveable cursors were used to measure discharge timings and the discharge rate corresponding to any desired time segment of the histogram. The timings of the e.m.g.s recorded simultaneously in the limb muscles could be shown below the p.e.t.h. as bars representing the mean active period in each muscle (see Figs. 7 and 8).

Raster displays were also computed in which each dot represented an action potential. Both for p.e.t.h.s and rasters the onset of e.m.g. in the lateral head of triceps brachii was used as the event marker for step cycle onset. This muscle was chosen for purely practical reasons: it invariably yielded large e.m.g. signals and its active/inactive periods were readily distinguishable so that a cursor could easily be set at the onset of the e.m.g. burst. Use of this particular marker is *not* meant to imply that a causal relationship necessarily existed between the discharge of any one cortical neurone and the e.m.g. onset.

Histology

After 2–3 months animals were deeply anaesthetized with pentobarbitone sodium and killed by perfusion *per aortem* with fixative (Holt–Hicks solution). All e.m.g. electrodes were inspected. The brain was removed and photographed and appropriate blocks were celloidin-embedded, sectioned at 80 μ m and stained with Cresyl Violet and Luxol Fast Blue. Brain stems were sectioned coronally to verify the position of the pyramidal electrode and the perieruciate cortex was sectioned sagittally.

RESULTS

Nature of the cortical units recorded

Extracellular recordings were made from 165 motor cortical units in fifteen cats. The number per animal ranged from three to twenty-two. Over-all, 150 of the units were recorded via seventy-four of the electrodes implanted singly (see Methods) so that the average yield was two units per successful electrode. However, a total of around three hundred electrodes was implanted so that only about one in four electrodes was successful. In one animal the position of each electrode tip as determined histologically was compared with its success or failure in recording units: all successful tips lay in the deeper layers of the grey matter (though not all such tips yielded units). The points of insertion of all twenty-three single wires implanted in this case are shown in Pl. 1*A* by circles superimposed on the brain photograph and the six wires yielding units are distinguished as filled circles. A bundle of wires was also inserted into the posterior sigmoid gyrus (see Methods) and the entry point is starred. In all, fourteen units were recorded by seventy-six wires implanted in bundles, so that this method was clearly less successful. However, most of the difference is attributable to the fact that all tips in a bundle often lodged in the subcortical white matter. In such cases no units were ever encountered. The track of one bundle is shown in Pl. 1*C*. In this case the electrodes splayed out during insertion and although five wires ended in the white matter one tip (white arrow) was located 4.3 mm below the surface in the grey matter forming the posterior wall of the cruciate sulcus.

The entry points for all successful electrodes are pooled onto a single cortical diagram in Pl. 1 *B* where filled circles represent single wires and open circles those implanted in bundles. Because the cortical surface is curved Pl. 1 *B* provides only an approximate indication of the surface projections of the units. However, since most

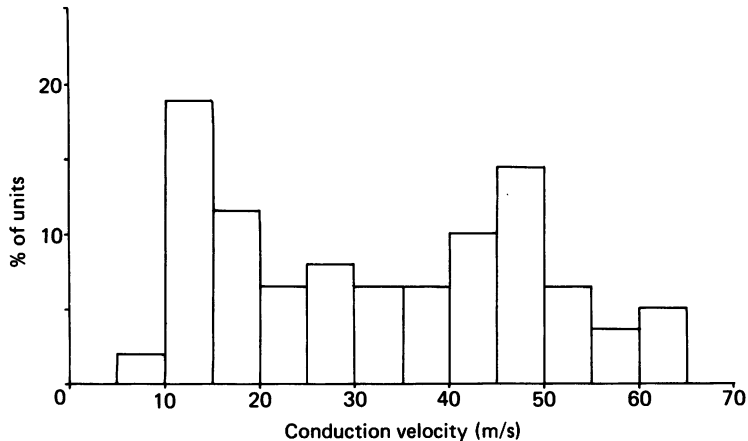


Fig. 1. Normalized frequency histogram of axonal conduction velocities for the sixty-five units which responded antidromically to pyramidal stimulation (see Methods.).

single wires were inserted radially for less than 2 mm the errors for these are not large. Clearly, most units were in the coronal gyrus (nomenclature as in Livingston & Phillips, 1957) with substantial minorities in the anterior and posterior sigmoid gyri. Pl. 1 *D* shows a sagittal section through the coronal gyrus in one brain. The terminal portion of the narrow track from a wire implanted singly is arrowed.

Unit action potentials were mostly negative-positive though a few were positive-negative. Amplitudes ranged from 300 μ V to 2.0 mV and durations from 0.4 to 0.8 ms. Rarely, brief (0.3 ms) triphasic spikes (positive-negative-positive) were encountered and were discarded on the assumption they were from axons. We assume the remaining 165 units were recorded from cell bodies and in fact some spikes showed a point of inflexion on the rising phase though it was never marked and fractionation of the spike did not occur (cf. Phillips, 1959).

Sixty-five out of 115 units tested (57%) responded antidromically to stimulation of the medullary pyramid (see Methods). Two units failed to follow at 1000 Hz but they would follow at 200 Hz and were therefore included as p.t.n.s. Collisions between antidromic and natural discharges were often observed.

Thresholds for antidromic invasion in response to 0.2 ms pyramidal shocks ranged widely (from 12 μ A upwards) but 50% were less than 600 μ A and 80% less than 2 mA. The wide variation presumably reflects variations both in axonal excitability and in the spatial relationship with the stimulating electrode. The latter was probably the major factor because antidromic latency did not correlate with threshold.

The range of antidromic latencies was 0.7-4.8 ms (measured at 2*T* stimulus intensity), and assuming a conduction distance of 44 mm (see Methods) these latencies correspond to a range of velocities from 63 to 9.2 m/s. The (normalized) frequency distribution of velocities is shown in Fig. 1, which confirms the classical

finding that there are maxima at 10–15 m/s and at 40–55 m/s. Sixty-seven per cent of p.t.n.s had latencies less than 2·1 ms (i.e. velocities > 21 m/s); following Takahashi (1965) these were classed as fast-axon p.t.n.s, the remainder as slow-axon p.t.n.s.

If any proportionality exists between cell body size and axonal conduction velocity then spike amplitude among p.t.n.s might be inversely related to antidromic latency. Among a random sample of forty-three p.t.n.s a loose inverse relationship did in fact exist: spike amplitude exceeded 1 mV in ten out of twenty-nine fast-axon p.t.n.s but in only two out of fourteen slow-axon p.t.n.s.

A more obvious relationship existed between the antidromic latency and the duration of the (orthodromic) spikes discharged by p.t.n.s. Units with latency less than 2·1 ms had spikes lasting 0·4–0·6 ms whilst for slow-axon p.t.n.s the range was 0·5–0·8 ms and most durations exceeded any of those for fast-axon p.t.n.s. This difference agrees well with results of Takahashi (1965) obtained by intracellular recording.

Discharges in the absence of locomotion

Animals which walked on the belt with a confident regular stride were the most adventurous and curious regarding their surroundings and it was therefore difficult to obtain recordings in the complete absence of movement. Nevertheless, data were collected during periods when the animals were sitting or standing and making a minimum of spontaneous movements and the rest of this report is based on 102 cells studied both during 'rest' and during locomotion. They included eighty-three cells tested for the presence of a projection to the pyramid and fifty of these (i.e. 60%) were p.t.n.s, a proportion similar to the 57% of p.t.n.s in the whole population.

Records from three typical p.t.n.s are shown in Fig. 2A–C. In each case the upper continuous record shows the impulse activity during several seconds without overt movement by the animal, whilst the lower record shows the activity during seven successive paces at 0·5 m/s and also the e.m.g. recorded simultaneously from triceps brachii muscle of the contralateral forelimb (T). In all three units an irregular slow discharge at rest altered during walking to rhythmic bursts of spikes entrained to the step cycle. For each unit a fast-swept record on the right shows the antidromic action potential elicited by stimulation of the pyramidal tract at intensity 2*T* (latencies 1·2, 0·8 and 1·1 ms in A, B and C respectively). In A and B responses to trains of four stimuli at 1000 Hz are also shown; although responses to the first three stimuli are in each case partly obscured by stimulus artifact it is clear that each shock elicited a spike.

In the stationary animal all 102 neurones were discharging although in some the spikes were interspersed with pauses lasting several seconds. Because the animal was encouraged to move during the initial screening of each electrode it is unlikely that cells inactive in the absence of movement were overlooked. Fig. 3A shows (normalized) frequency distributions for the discharge rates of the neurones in the absence of locomotion. The top histogram shows the distribution for all units whilst the others show in descending order the distributions for p.t.n.s, for non-p.t.n.s (i.e. units which failed to respond antidromically to pyramidal stimulation) and for untested units. Rates varied from 0·5 to 44 impulses/s over-all and also ranged widely in each sub-population, although the mode for p.t.n.s was faster than that for non-p.t.n.s.

To determine whether fast-axon and slow-axon p.t.n.s differed in discharge rate

the rate for each cell was plotted as a function of antidromic latency. The result is shown in Fig. 3*B* where some difference is evident: twelve of the thirty-four fast-axon p.t.n.s discharged at over 20 impulses/s whilst none of the sixteen slow-axon p.t.n.s discharged this rapidly.

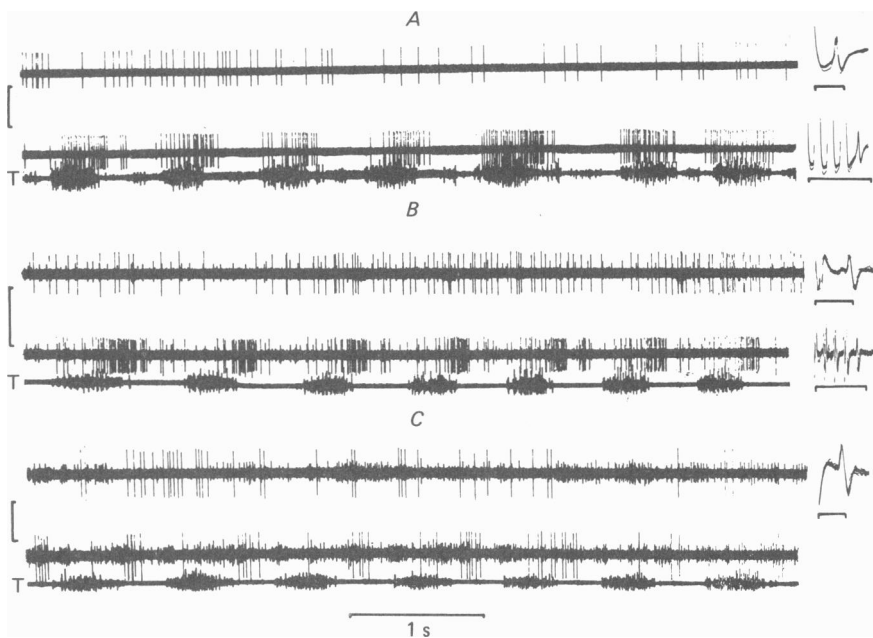


Fig. 2. *A*, *B* and *C* are similar presentations for three different fast-axon p.t.n.s. Upper continuous record shows unit discharge in absence of overt movement. Lower record shows discharge during locomotion at 0.5 m/s together with e.m.g. recorded from lateral head of triceps brachii muscle in contralateral forelimb (T). Upper fast-swept record to right shows several superimposed antidromic responses to pyramidal stimulation at intensity $2T$. Lower fast-swept record in *A* and *B* shows superimposed responses to trains of four stimuli at 1000 Hz. The 1 s time calibration applies to all continuous traces. Time bars for fast-swept records are 2 ms in *A* (upper), 5 ms in *A* (lower), 1 ms in *B* (upper), 5 ms in *B* (lower) and 2 ms in *C*. Voltage calibrations in *A*, *B* and *C* are all 1 mV.

Discharges during locomotion at 0.5 m/s

Discharge rates. The 102 neurones in Fig. 3*A* were all studied during locomotion at 0.5 m/s and the normalized frequency distributions for mean rate are shown in Fig. 3*C*. There is a wide range of values and comparison with Fig. 3*A* shows this is primarily due to substantial acceleration of some cells. In Fig. 5*A* the rates during locomotion and during rest are compared for eighty-five individual cells. During walking, rate increased (sometimes only slightly) in forty-seven units (56%) whilst in the rest it remained the same or decreased. Different categories of neurone are identified by different symbols and inspection reveals that each category included some cells which showed substantial increases or decreases in rate and others showing little or no change.

The relationship between axonal conduction velocity and discharge rate during

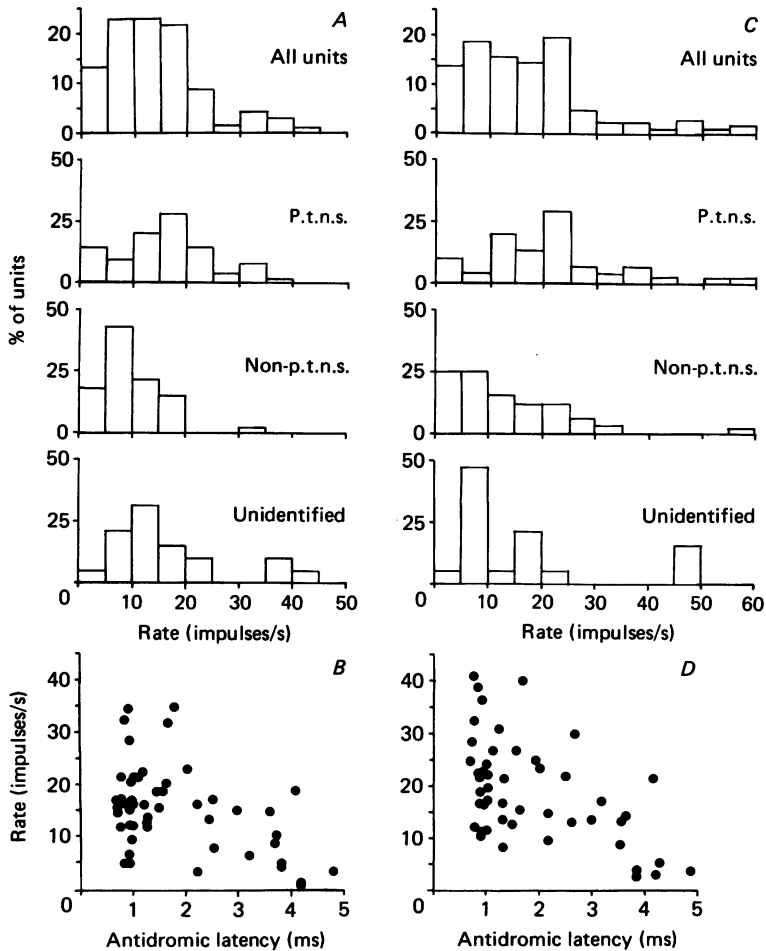


Fig. 3. Discharge rates of cortical neurones. *A* shows normalized frequency histograms for values of mean discharge rate in the resting animal. *A* is based on 102 neurones of which fifty were p.t.n.s., thirty-three were non-p.t.n.s. and nineteen were unidentified. *B*, scattergram relating mean discharge rate in the resting animal to antidromic latency in the fifty p.t.n.s. of which thirty-four were fast-axon neurones (i.e. latency < 2.1 ms). *C*, normalized frequency histograms for values of mean discharge rate during locomotion at 0.5 m/s, based on the same neurones as in *A*. *D*, scattergram relating discharge rate during locomotion at 0.5 m/s to antidromic latency in forty-eight of the p.t.n.s. Two fast-axon p.t.n.s. are omitted which had rates of 51 and 57/s and latencies of 0.8 and 1.7 ms respectively.

locomotion is shown in more detail for forty-eight of the p.t.n.s. in Fig. 3*D*, where it is noticeable that as many as nineteen out of thirty-two fast-axon p.t.n.s., but only three of the sixteen slow-axon p.t.n.s. discharged at faster than 20 impulses/s. Evidently, the difference between these groups which existed in the resting animal was retained during locomotion (cf. Fig. 3*B* and *D*).

Frequency modulation of unit discharges. During locomotion the three units in Fig. 2 were markedly frequency modulated, each discharging one burst of impulses per

step. However, they clearly differed significantly in the number of spikes per burst, in the peak instantaneous rate during the burst and also in the timing of their discharge relative to the step cycle.

To facilitate quantitative comparisons of such variables between different units (and between the same unit during different bouts of walking) use was made of computer-generated displays, particularly post-event time histograms (p.e.t.h.s) which demonstrated the average pattern of discharge during twenty successive paces (see Methods). The p.e.t.h.s for three different units are included in Fig. 4A–C, and other examples are given in Figs. 7 and 8. In these displays periods of accelerated discharge time-locked to the step cycle show up as peaks of discharge probability whilst periods of reduced activity or silence appear as troughs. Bin width is always 20 ms so that the count in any bin can be translated into an equivalent rate in impulses per second by multiplying by a factor of 2.5. Step cycle onset was at the start of e.m.g. activity in triceps brachii muscle in the contralateral forelimb so that the stance phase begins with foot placement two or three bins after the start of the display and continues until the swing or transfer phase is initiated at foot lift, approximately 67% of the way through the pace (see Methods).

Although bin width was fixed each p.e.t.h. was automatically adjusted so that the complete time axis spans a period equal to *twice* the mean duration of the step cycles during the sampling period (and the vertical broken line therefore indicates the average time of transition from one step to the next). This facilitates comparison of p.e.t.h.s obtained for different units (when step cycle duration may differ) and also for the same unit at different speeds of locomotion (when step cycle duration inevitably differs).

Each p.e.t.h. was accompanied by a raster display constructed using the same batch of data, and three examples are shown in Fig. 4A–C. On each line are shown the discharges during one pace (to the right of the vertical line) and during the preceding 750 ms (to left). Paces are not shown in their order of occurrence but are rank-ordered according to duration.

Two complete step cycles rather than one are displayed in the p.e.t.h.s because a clearer impression is provided of any rhythmic character in the discharge. Note, however, that because step cycles have considerable duration (400–900 ms depending on speed of locomotion) the impulses during the second displayed pace occur long after the triggering event at the start of the p.e.t.h. Because step duration inevitably varies somewhat from pace to pace, the longer the time elapsing after p.e.t.h. onset the more the display is subject to temporal ‘blurring’. Any peaks and troughs of discharge probability are therefore often smoothed down somewhat during the second pace (e.g. in Figs. 7A and 8A). For this reason measurements of discharge frequencies and timing were made only from the first displayed pace. Temporal blurring can of course occur even during the first pace and will be greatest when successive steps are most variable in duration.

Comparison of the p.e.t.h. with the raw record and raster for the unit of Fig. 4B shows how such blurring can lead to a p.e.t.h. which markedly over-estimates the duration of the burst discharged by the cell (and correspondingly underestimates the peak rate of discharge). Such distortions cannot be completely avoided because unrestrained animals rarely maintain pace duration absolutely constant. However, to limit such unwanted effects our practice has been to make measurements only from p.e.t.h.s for sequences of paces in which duration fluctuated about the mean by not more than 10%. In most cases the fluctuation was 5% or less.

An alternative approach to the problem would be to normalize the durations of the individual steps but this was not attempted because it is unlikely that long and short steps involve ‘stretching’ or ‘shrinkage’ which is uniform throughout the step cycle. Indeed there is evidence that variation occurs mainly in the stance phase, swing being relatively fixed in duration (e.g. Grillner, 1974).

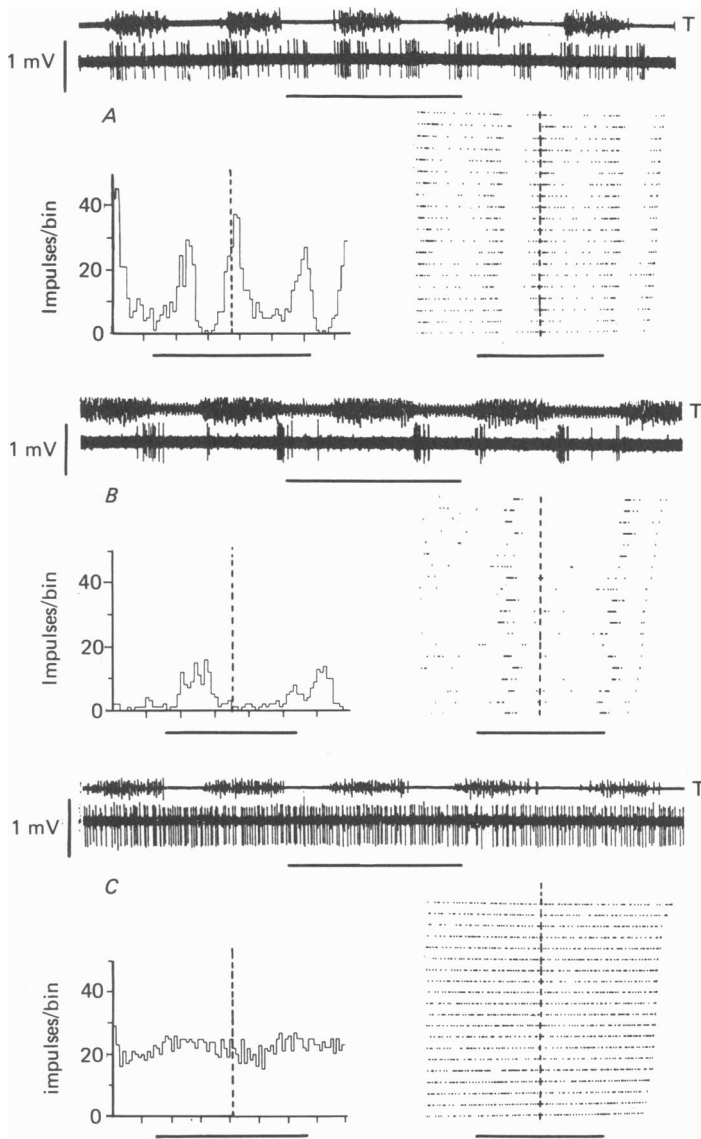


Fig. 4. Discharge patterns during locomotion at 0.5 m/s. *A* is an unidentified neurone, *B* a fast-axon p.t.n. and *C* a slow-axon p.t.n. In each case the raw record shows the unit discharge during several successive paces with above the e.m.g. recorded simultaneously from lateral head of contralateral triceps brachii (T). The post-event time histogram (p.e.t.h.) for each unit overlap averages the discharge during twenty successive paces. Vertical axis is total counts per bin, bin width 20 ms. Time axis begins at onset of locomotor e.m.g. in contralateral triceps brachii. Length of horizontal axis is twice the mean duration of the steps sampled so the vertical broken line indicates mean time of transition from one pace to next. Accompanying raster display is formed from the same data as the p.e.t.h. Each dot represents an action potential and each line shows one step cycle (beginning at vertical dashed line) plus the preceding 750 ms. Steps rank-ordered by duration. Time calibrations 1 s throughout.

Proportion of neurones showing frequency modulation. In most units which were frequency modulated during walking the rhythmic nature of the discharge was detectable (often obvious) from visual inspection of the spike train. However, in some neurones which discharged throughout the step cycle construction of a p.e.t.h. revealed a rhythmicity too weak to be obvious in raw records. An example is provided by Fig. 4C in which there is a slight tendency for fewer spikes to occur during the first than during the second half of the step cycle.

Such findings raise a question as to what may be considered a significant degree of modulation. In fact, we have chosen arbitrarily to consider as 'modulated' those units in which the peak of the p.e.t.h. exceeded the trough by at least four spikes per bin (equivalent to 10 impulses/s). On this basis the unit of Fig. 4C is at the lower limit of the modulated class.

Using this criterion eighty-two of the 102 units (80%) were frequency modulated during locomotion at 0.5 m/s. Eighty-eight cells were in the 'forelimb' motor cortex (i.e. coronal gyrus, lateral part of anterior sigmoid gyrus and lateral part of posterior sigmoid gyrus) and seventy-two of these (i.e. 82%) were modulated. Fourteen cells were in the 'hind-limb' area (i.e. medial part of the posterior sigmoid gyrus; see Nieoullon & Rispal-Padel, 1976) and a similar proportion was modulated (i.e. ten neurones).

Most modulated neurones (seventy-one out of eighty-two) resembled the cells of Fig. 2 in showing one peak of discharge probability per step whilst the remainder (eleven units) showed two peaks (see for example the unit of Fig. 4A). Of the seventy-two modulated cells in the 'forelimb' motor cortex only six showed two peaks per step whilst no fewer than five out of ten modulated cells in the 'hind-limb' area showed two peaks. Whilst this finding appears to suggest that biphasic responses are much commoner in the hind-limb area it should be noted that the number of cells is small (see Discussion).

Amongst the 102 neurones, fifty were identified as p.t.n.s and forty-one of these (i.e. 82%) were frequency modulated. Thirty-three were non-p.t.n.s and twenty-six of these (i.e. 79%) were modulated. The proportion showing frequency modulation was therefore the same irrespective of whether or not a projection to the pyramid could be demonstrated.

Frequency modulation was common in both classes of p.t.n., being present in thirty-one out of thirty-four fast-axon cells and ten out of sixteen slow-axon cells. Because some cells discharged discrete bursts separated by silent periods, whilst others gave a maintained though frequency-modulated discharge, the frequency of occurrence of these two patterns was compared in fast-axon and slow-axon p.t.n.s. Of the twenty-five p.t.n.s firing discrete bursts twenty were fast-axon and five were slow-axon p.t.n.s. By comparison, of the sixteen p.t.n.s showing a frequency-modulated maintained discharge eleven were fast-axon and five were slow-axon p.t.n.s. Evidently, discrete bursts occur more commonly among the cells in the fast-axon class but are nevertheless not uncommon in slow-axon p.t.n.s.

Peak discharge rates. For modulated cells maximum firing rates were measured from p.e.t.h.s (after smoothing by eye). Frequency distributions for the resultant values are shown in the histograms of Fig. 5B-E. It is clear from Fig. 5C that p.t.n.s ranged widely in peak rate from 10 to in excess of 120 impulses/s. Non-p.t.n.s (Fig.

5D) also showed a wide range of peak rates though the mode of the distribution was at 10–20 impulses/s as compared with 30–40 impulses/s for p.t.n.s.

In general, fast-axon p.t.n.s showed higher peak rates than slow-axon p.t.n.s and therefore a greater depth of frequency modulation. This is illustrated by Fig. 5F in

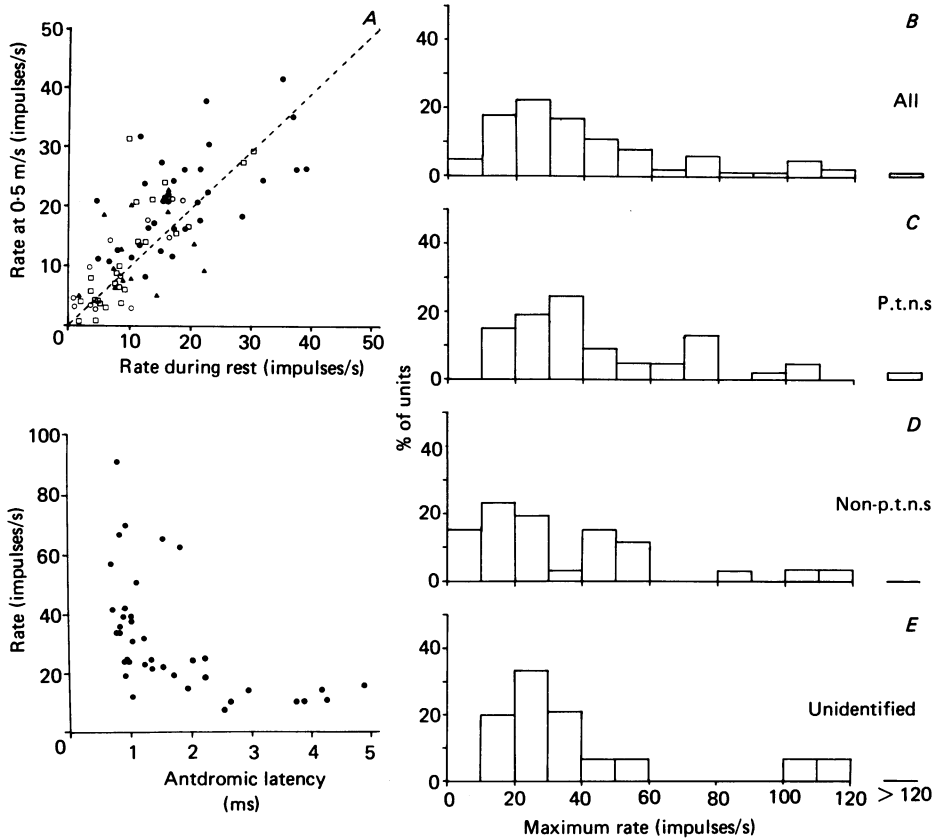


Fig. 5. Discharge rates of cortical neurones during locomotion at 0.5 m/s. *A*, scattergram comparing discharge rates during locomotion and rest for eighty-five individual neurones. ●, fast-axon p.t.n.s; ○, slow-axon p.t.n.s; □, non-p.t.n.s; ▲, unidentified neurones. *B*, *C*, *D* and *E*, normalized frequency histograms for peak discharge rates during the step cycle. *B* shows peak rate values for all eighty-two frequency-modulated neurones; *C*, *D* and *E* comprise forty-one p.t.n.s, twenty-six non-p.t.n.s and fifteen unidentified neurones respectively. Any cells with rates in excess of 120 /s are included in detached bin on right. *F*, scattergram which plots difference between peak rate and mean rate during step cycle against antidromic latency for thirty-nine frequency-modulated p.t.n.s.

which the difference between peak rate and mean rate is plotted against antidromic latency. None of the slow-axon p.t.n.s showed a difference greater than 25 impulses/s whilst this value was exceeded in seventeen out of twenty-nine fast-axon p.t.n.s.

It must be noted that the peak rates above represented averages obtained over twenty steps. The raw records in Figs. 2 and 4 and the rasters in Fig. 4 show that in some cells the peak frequencies fluctuated quite considerably from step to step, reaching values considerably higher than average in some steps and considerably lower in others. It is possible that some of the fluctuations correlate

with fluctuations in the stepping movements or the muscular activities producing them, but we have not attempted the step-by-step analysis needed to investigate such possibilities.

Phasing of discharges relative to the step cycle. It is clear from Figs. 2 and 4 (see also Figs. 7 and 8) that within the population of units the timing of the discharges varied widely so that (for example) some cells discharged during the stance phase when the

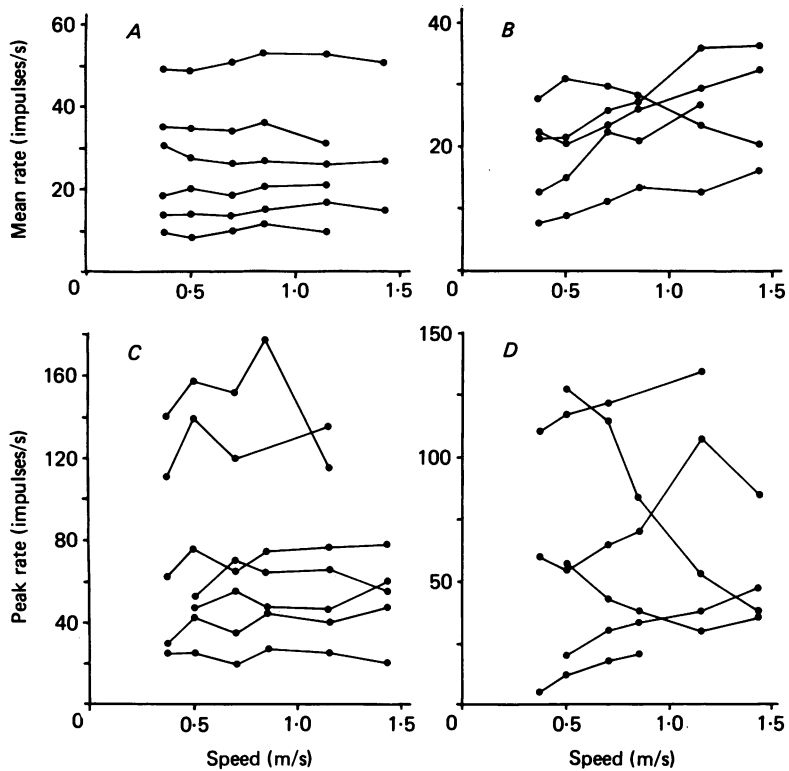


Fig. 6. Discharge rates during locomotion at different speeds. *A*, data for six neurones in which mean rate remained approximately constant over a range of walking speeds (see text for further explanation). *B*, data for five neurones in which mean rate changed systematically over a range of speeds (see text). *C* and *D* are similar plots of peak discharge rate (see text).

extensor muscles are active, whilst others discharged during the swing phase when flexors are active. This is not perhaps surprising because the units were distributed throughout the pericruciate cortex and presumably differed widely in their afferent and efferent connexions. Detailed consideration of individual phasings is deferred to the following paper (Armstrong & Drew, 1984) where they are discussed together with peripheral receptive field data for the neurones. However, it should be noted that no obvious correlation could be found between the discharge phasing of individual neurones and the locations of the points at which the relevant micro-electrodes entered the cortex.

In most modulated cells the discharges were tightly phase-locked to the step cycle in the contralateral forelimb. This was demonstrated by two observations. First,

the p.e.t.h. peaks for cells firing discrete bursts were similar in duration to the bursts fired during the individual steps (provided the steps were reasonably uniform in duration). Secondly, inspection of rasters showed that in paces with the same duration, the discharges occurred during the same part of the step. However, some cells showed looser phase-locking than others. Thus the raster in Fig. 4*B* shows that even in those paces with similar duration there was some fluctuation in the timing of the short burst of impulses discharged at around the cessation of activity in triceps brachii muscle.

It should be remembered that all displays were triggered from the onset of triceps brachii e.m.g. and it is possible that in cases like that of Fig. 4*B* the discharges occurred in fixed temporal relationship with some other aspect of the step cycle which fluctuated slightly in its timing relative to the onset of triceps e.m.g. Indeed, when a p.e.t.h. was constructed using e.m.g. onset in an alternative muscle as trigger, the p.e.t.h. peak in some cells was both shorter and higher, indicating better time-locking to that event. In future studies it may be worthwhile investigating systematically the extent to which different neurones are time-locked to different events in the step cycle.

Effect of speed of locomotion on the discharges

Discharge rates. Most cells were studied during locomotion at more than one speed and in twenty-seven cases recordings were made at five or six of speeds 0.37, 0.5, 0.7, 0.85, 1.15 and 1.43 m/s. These values span the whole range of possible walking speeds from a slow amble to a hurried walk verging on a trot. In twenty-two cells the mean rate of discharge remained nearly constant and Fig. 6*A* shows examples of such behaviour in six cells with different discharge rates. In the remaining five neurones rate changed more or less progressively with speed (in roughly linear manner) as shown in Fig. 6*B*. In one case (a non-p.t.n.) the rate declined as speed increased above 0.5 m/s, changing from 31 impulses/s to 20 impulses/s at 1.43 m/s (equivalent to a gradient of 12 impulses/s per m/s). In the other four cases there were modest increases in rate. These cells included two fast-axon p.t.n.s in which the increases amounted approximately to 12 and 17 impulses/s per m/s and a slow-axon p.t.n. and a non-p.t.n. with gradients of 18 and 7 impulses/s per m/s respectively.

A further thirty-eight cells were studied at four speeds within a narrower range (typically 0.37–0.85 m/s) and the results confirmed those for the cells studied most fully. In only four cases was there a progressive change (an increase) in the discharge as walking speed increased. In each of these the rate/speed relationship was approximately linear and the gradients involved were in the same modest range as for the five cells more thoroughly studied. Two of the four cells were fast-axon p.t.n.s and two were unidentified. In the remaining thirty-four cases rate remained constant or (in a few cells) fluctuated unsystematically. Over-all, therefore, discharge rate was unrelated to speed over a considerable range of walking speeds in fifty-six out of sixty-five cells (86%).

The effect of speed on peak rate was also explored and although the relationships in individual cells were somewhat less consistent the findings were broadly similar to those for mean rate. Among the twenty-seven cells studied over the widest speed range, peak rate remained approximately constant or fluctuated unsystematically in twenty-one cases; the results for seven of these neurones with different peak rates are shown in Fig. 6*C*. More or less progressive changes were found in six units of which four (two fast-axon p.t.n.s, one slow-axon p.t.n., one non-p.t.n.) showed an increase

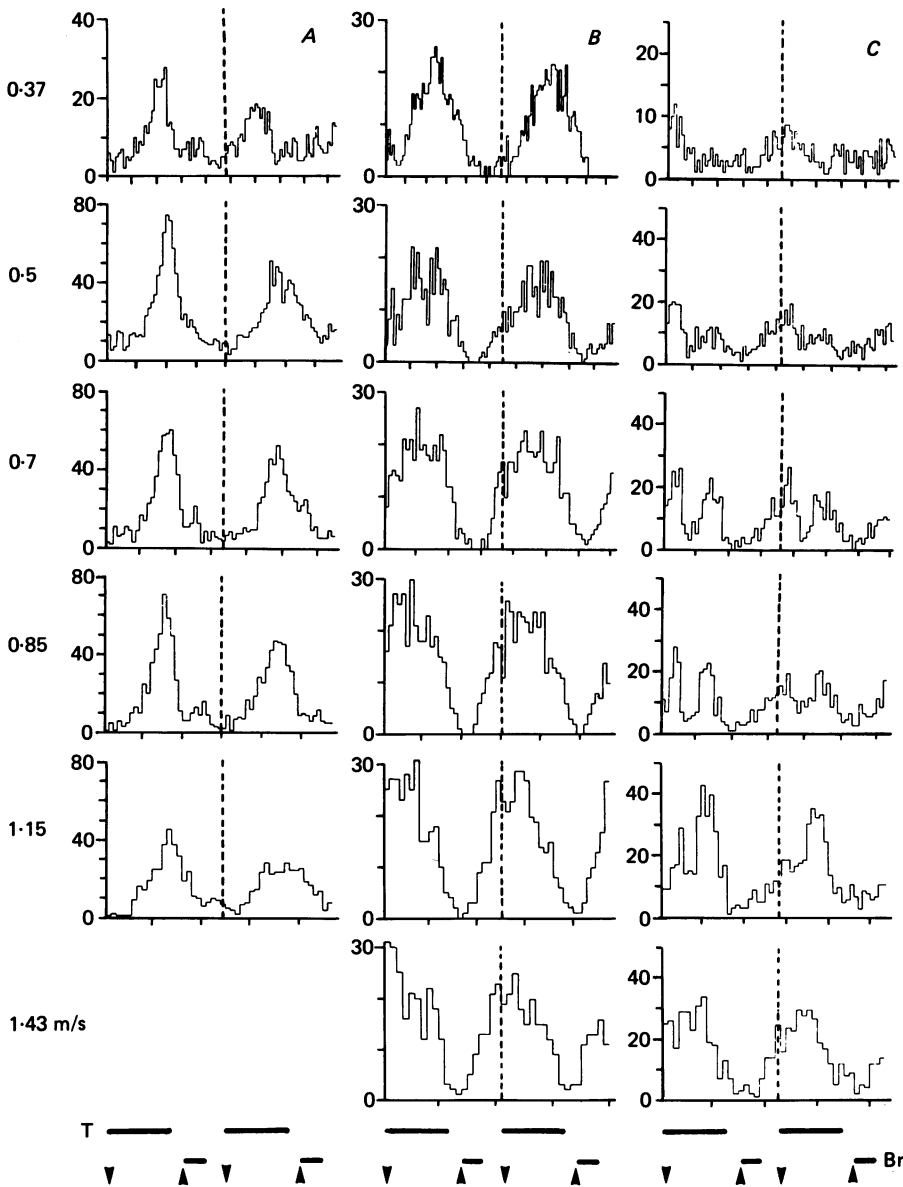


Fig. 7. Effect of walking speed on neuronal discharge patterns. *A*, *B* and *C* are three different neurones. Each p.e.t.h. has been formed from the discharges during twenty consecutive steps except at speed 0.37 m/s in *A* and *C* where only ten steps were used. In each p.e.t.h. the time axis has been scaled to equal the duration of two complete steps. The end of the first step and beginning of the second is therefore indicated by the vertical broken line. Note that bin width is 20 ms in all cases. Horizontal bars at bottom indicate the timing of the locomotor e.m.g.s in triceps brachii (T) and brachialis (Br) muscles of the contralateral forelimb during locomotion at 0.5 m/s. Approximate times for footfall and foot lift are indicated by downward and upward arrowheads respectively.

in peak rate as speed increased (accompanied in two cases by an increase in mean rate) whilst the remaining two (non-p.t.n.s) showed a decrease (accompanied in one case by a decrease in mean rate). These findings are shown in Fig. 6*D*. Depth of modulation behaved similarly because minimum rate remained constant or underwent small changes.

Similar findings were made in cells studied less thoroughly so that, over-all, peak rate and depth of modulation were unrelated to speed of locomotion in the large majority of neurones. In addition, it was noted that amongst twelve cells which were not significantly modulated at 0.5 m/s ten remained unmodulated at any higher speed. The other two showed a weak modulation at 0.7 m/s which was sustained, but did not increase, at still higher speeds.

Phasing of discharges. The effects of speed on the phasing of the discharges in frequency-modulated neurones were more complex. Findings from three units are illustrated by the p.e.t.h.s in Fig. 7*A–C*. In Fig. 7*A* the discharge timing remained substantially unchanged when velocity of locomotion increased by a factor of 3. However, in Fig. 7*B* and *C* phase changes are evident. In Fig. 7*B* maximum activity occurs in late stance at 0.37 m/s but as speed increases this peak moves progressively earlier until by 1.43 m/s it occurs at time zero (i.e. coincides with the onset of activity in triceps brachii). In Fig. 7*C* there is one peak of activity coincident with the onset of triceps activity at 0.37 m/s but by 0.85 m/s the pattern has changed so that one peak occurs just after step cycle onset and a second equally prominent peak occurs in late stance. Further increases in speed enlarge and spread this peak so that at 1.43 m/s the unit discharges from just before step cycle onset until just before the end of triceps activity.

Of sixteen well-modulated cells in the forelimb motor cortex which were studied over the widest speed range, seven showed no change in phasing (cf. Fig. 7*A*) whilst nine cells showed either a progressive phase shift (to the left as in Fig. 7*B* or to the right) or more complex changes like those in Fig. 7*C*. However, the changes in Fig. 7*B* and *C* were amongst the largest observed and most phase shifts were small, amounting to less than 10% of the step cycle for a speed change of 1 m/s. Unfortunately, the changes did not correlate in direction or extent with any other information available for the units, such as location in the cortex or whether or not they were p.t.n.s. Their existence does show, though, that in some cells timing is dependent on some aspect of locomotion which undergoes progressive temporal shift relative to the onset of triceps brachii e.m.g. as walking speed changes.

Effect of uphill gradient

For twenty-seven cells the discharges during locomotion on a level surface could be compared with those when the animal walked at the same speed (0.5 m/s) on a 10° uphill incline. The effects on locomotion were to increase step cycle duration by less than 5% but to increase markedly the peak amplitude of the locomotor e.m.g.s, particularly in hind-limb extensor muscles but also in forelimb flexors and extensors which on average increased by ca. 50%.

The effects on four neurones are illustrated by the p.e.t.h.s of Fig. 8, from which it is clear that there were no striking changes in over-all discharge rate or in depth of frequency modulation.

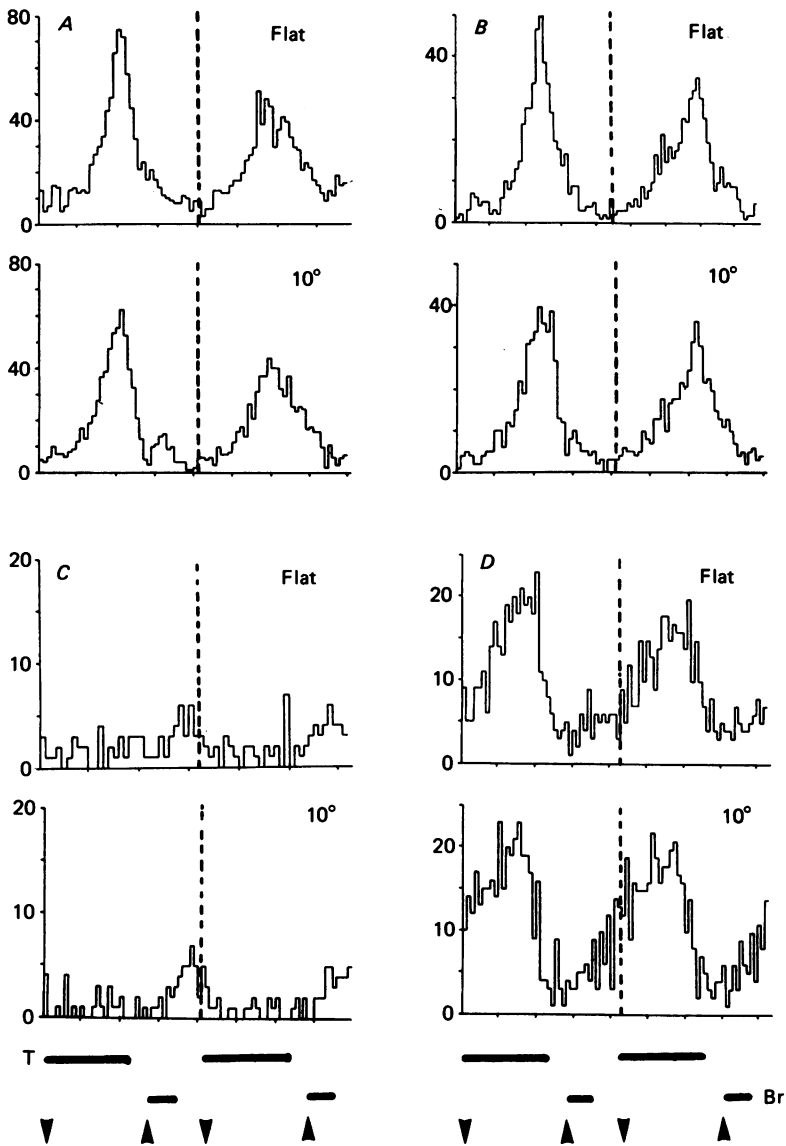


Fig. 8. Discharge patterns compared for locomotion at 0.5 m/s on the flat and on a 10° uphill incline. *A*, *B*, *C* and *D* are four different neurones. Each p.e.t.h. comprises twenty successive paces and each time axis spans two complete step cycles. Bin width 20 ms. Other conventions as in Fig. 7.

Similar findings were made in the other neurones and Fig. 9*A* compares the values for mean rate during uphill walking with the corresponding values obtained on the flat. The diagonal line shows the relationship expected in the absence of change and different symbols are used for p.t.n.s, non-p.t.n.s and unidentified neurones. It is clear that the two values were mostly the same or little different. The largest change was an increase of 11 impulses/s in one unidentified cell and the largest change for a p.t.n.

was an increase of 8 impulses/s (in a fast-axon p.t.n.). Twenty-one of the neurones were frequency modulated and in Fig. 9B peak rates uphill are compared with the corresponding values on the flat. Again, with the exception of one fast-axon p.t.n. which showed an increase of 25 impulses/s, all points lie on or close to the line of equality. There were no striking shifts of discharge timing in any of the cells, the slight phase advance visible in Fig. 8D being the largest change found.

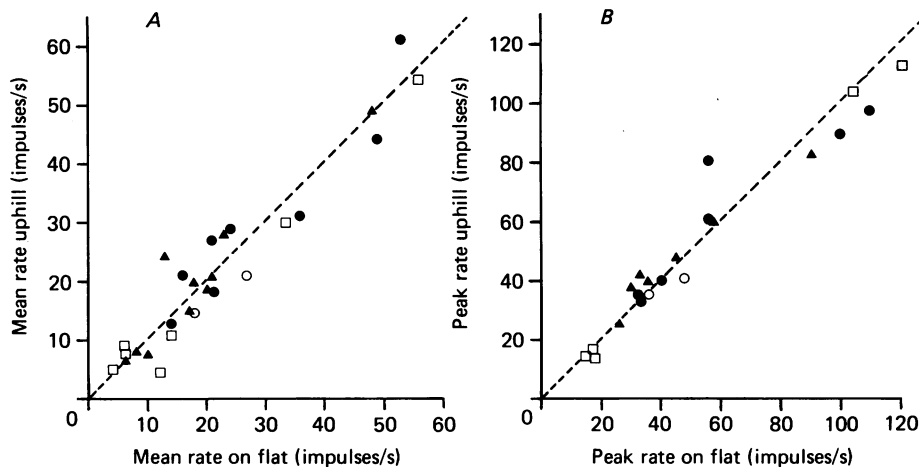


Fig. 9. Discharge characteristics compared for locomotion at 0.5 m/s on the flat and on a 10° uphill gradient. A includes mean rates for twenty-seven neurones. B includes peak rate values for twenty-one of the same neurones. ○, slow-axon p.t.n.s; ●, fast-axon p.t.n.s; □, non-p.t.n.s; ▲, unidentified neurones.

DISCUSSION

The present experiments amply demonstrate the feasibility of using chronically implanted micro-electrodes to obtain recordings from motor cortical neurones in the awake, actively moving cat. Recording conditions were so stable that it would be possible to monitor the neurones during virtually the whole movement repertoire of the animal (and during the learning of new movement tasks). A similar approach has previously been employed by Burns *et al.* (1973) to investigate activity in cat visual cortex and their success was comparable although the yield of units per electrode was lower (personal communication), most probably because the neurones are somewhat smaller. Microwires have also been used successfully in monkey motor cortex (Schmidt *et al.* 1976) and in the time since the present experiments were begun investigations have been briefly reported in which the discharges of motor cortical neurones in the cat were recorded during falls (Palmer, Bak, Dold & Schmidt, 1979) and during locomotion on a moving belt (Palmer *et al.* 1980). Few neurones were studied during locomotion because the emphasis was on investigating small groups of closely juxtaposed cells. However, the discharge patterns encountered seem to have been generally similar to those we describe.

The use of a stimulating electrode in the medullary pyramid has allowed us to distinguish cells which could be antidromically invaded (p.t.n.s) from those which

could not (non-p.t.n.s). Although strictly speaking failure to evoke an antidromic impulse shows only a failure of the stimulus to excite the axon it is nevertheless probable that the majority of 'non-p.t.n.s' did not project to the pyramid: such cells were presumably cortico-rubral, cortico-striate and other types of cell. The proportion of p.t.n.s in our sample was 57 %, which is very similar to the 54 % in the study by Brooks, Rudomin & Slayman (1961) who recorded extracellularly with glass micropipettes in locally anaesthetized paralysed cats.

Because implanted microwires have not been widely employed for cortical studies it is desirable that information should be available regarding their selectivity. This is particularly important because the tips were fairly large (though not grossly larger than those of glass-coated tungsten micro-electrodes). Our evidence suggests that most if not all the recordings were from cell bodies rather than axons (see Results). For p.t.n.s the over-all range of conduction velocities was from 9 to 63 m/s which is similar to that found in previous extracellular studies employing high-impedance glass micropipettes. Lance & Manning (1954), for example, found a range from 7 to 70 m/s and Armstrong (1965) found a range from 6 to 55 m/s in a sample of ninety-seven p.t.n.s. It is widely accepted (see Phillips & Porter, 1977) that the frequency spectrum of conduction velocities in the cat pyramidal tract shows one peak at 10–15 m/s and another at 40–55 m/s, and this and other evidence (see Takahashi, 1965) has suggested that p.t.n.s can be classified into a fast-axon group conducting at over 21 m/s (i.e. with antidromic latency < 2.1 ms) and a slow-axon group conducting at less than 21 m/s (i.e. with latency > 2.1 ms). In the present study cells in both groups were sampled (see Fig. 2C) but the proportion of slow-axon cells (33 %) was considerably smaller than in extracellular studies with micropipettes. However, it was rather similar to the proportions of 23 % (Armstrong, 1965) and *ca.* 35 % (Takahashi, 1965) found in two intracellular studies.

Unit discharges in the absence of overt movements were markedly irregular and sometimes paused for several seconds. Based on samples of 1000 successive impulses the rates ranged from 0.5 to 44 impulses/s. One surprising finding was that amongst p.t.n.s the fast-axon cells tended to show the highest levels of background activity. This is opposite to the finding reported by Evarts (1965) for monkey p.t.n.s among which the cells with fastest axons (antidromic latency < 1 ms) were usually almost silent in the absence of phasic movements whilst those with slow axons were usually tonically active. This could be a species difference but alternatively it may reflect a difference between the levels of muscle activity in the two studies. The monkeys were trained to assume a state of relaxed immobility whereas the cats here were alert, inquisitive animals which rarely relaxed fully.

During locomotion just over half the units discharged faster than during 'rest' (see Fig. 5A) and large increases most commonly occurred amongst fast-axon p.t.n.s. These findings closely resemble those made by Evarts (1965) when the activity of monkey p.t.n.s during natural movements of the hand and arm (i.e. movements of grooming, scratching and food handling) was compared with the activity in the resting animal. The actual discharge rates involved were also similar, ranging from 5 to 60 m/s (p.t.n.s only) in the present study and from 3 to 45 m/s in the monkey. The rates were also rather similar to the range (12–67 impulses/s) found by Lewis (1974) in a study of monkey p.t.n.s discharging in relation to a stereotyped arm

movement (lever pulling). Like Evarts and ourselves, Lewis found that p.t.n.s with higher conduction velocities tended to discharge at a higher rate, though with considerable scatter in the relationship.

The most striking finding relating to activity during walking was that no fewer than 80% of neurones discharged rhythmically during locomotion at 0.5 m/s. This agrees well with the statement by Palmer *et al.* (1980) that 82% of a sample of twenty-eight p.t.n.s from a small area of forelimb motor cortex were frequency modulated during locomotion. Our results also indicate that rhythmic discharges occur over a wide area of the motor cortex and in roughly similar proportions of p.t.n.s and non-p.t.n.s. Most frequency-modulated neurones generated one discrete burst of impulses per step and such bursts ranged in different cells from a few impulses at high frequency to a train lasting for a substantial fraction of the step cycle. Some cells discharged throughout the step and this could occur in both slow-axon and fast-axon p.t.n.s though the latter most often generated discrete bursts of impulses and were silent or almost silent between bursts. Like Palmer *et al.* (1980) we encountered a few neurones which generated two bursts per step and it is possible that this pattern is commoner amongst cells in the hind-limb motor cortex since it occurred in five out of ten units. However, the five units were recorded via only two electrodes so that no firm statement can be made. The modal value for peak rates amongst p.t.n.s lay between 30 and 40 impulses/s but amongst both these cells and non-p.t.n.s maximum rates varied considerably. The highest rates were found among the fast-axon p.t.n.s (though not all such cells achieved peak rates higher than the slow-axon p.t.n.s).

It is of interest briefly to compare the patterns of activity among motor cortical neurones with those described by Orlovsky (1972*b, c*) for neurones of brain-stem motor centres in decerebrate walking cats. The proportion of rubro-spinal neurones which were rhythmically active was similar (83%), and whilst some neurones discharged discrete bursts others displayed a maintained discharge with rhythmic fluctuations in frequency. Maximum discharge frequencies were slightly higher but, as in the present study, the discharges fluctuated step by step: indeed modulation sometimes ceased for a few steps then reappeared. Among vestibulo-spinal neurones 67% were rhythmically active and again some neurones developed discrete bursts whilst others were continually active. Peak frequencies varied widely but were usually in the range 50–100 impulses/s.

Both in red nucleus and in Deiter's nucleus Orlovsky (1972*b, c*) encountered a minority of cells (11% and 5% respectively) which generated two peaks of activity per step cycle. In those experiments movements of the forelimbs were usually absent or prevented and in subsequent experiments by Udo, Oda, Tanaka & Horikawa (1976) in which the walking involved both forelimbs and hind limbs, most lumbo-sacral-projecting Deiter's neurones were said to be biphasically active. In the present experiments, quadrupedal locomotion was involved but the proportion of cortical neurones showing two bursts was less than 10% in the 'forelimb' area.

For both vestibulo-spinal and rubro-spinal neurones Orlovsky (1972*b, c*) mentions that more forceful locomotion (produced by stronger stimulation of 'locomotor centres' in mid-brain or hypothalamus) was accompanied by increases in discharge rate and in the depth of frequency modulation. These changes were presumably substantial because they were detected without averaging techniques. In the present

study, over-all rate and/or peak rate changed progressively with speed in only *ca.* 14 % of motor cortical cells. Moreover, the changes were small, the largest increase in mean rate, for example, being only 18 impulses/s for an almost fourfold increase in walking speed. Most cells showed either no change or inconsistent changes in both mean and peak rate and these characteristics are the more striking and surprising because they occurred across virtually the whole range of possible walking speeds, when step duration ranged from *ca.* 950 ms (at 0.37 m/s) to *ca.* 400 ms (at 1.4 m/s).

The findings from locomotion on an uphill gradient when compared with walking on the flat were similar: twenty-five out of twenty-seven cells that included both p.t.n.s and non-p.t.n.s showed little or no change in either peak rate or mean rate.

These findings offer a marked contrast with those made in monkeys using the wrist or wrist and arm to move a lever against variable load (Evarts, 1968; Lewis & Porter, 1974). Under these conditions an appreciable proportion of those p.t.n.s which changed rate in relation to the movement exhibited discharge frequencies which correlated closely with the levels of muscle force required to overcome different loads. In our experiments changes in muscle force were not demonstrated directly but it is *a priori* likely that more vigorous locomotion will require increased levels of active force in many muscles and certainly the locomotor e.m.g.s from flexor and extensor muscles of the limbs increased progressively in amplitude with speed and were also markedly increased during uphill locomotion. Under such circumstances the probability of encountering 'force-related' (i.e. speed- or gradient-related) neurones should be substantially greater than for lever tasks in which fewer muscles are likely to show force levels strongly dependent on the external load applied to the manipulandum.

It follows that the paucity with which speed- or gradient-related changes were found requires some discussion. First, it could be argued that the rhythmic cortical discharges are an epiphenomenon which accompanies locomotion but plays no part in the causation of the locomotor movements. This proposition cannot be dismissed out of hand because pyramidal tract section does not grossly impair locomotion in the cat (see Eidelberg (1981) for a review). However, the evidence from lesions cannot be conclusive and the argument is difficult to sustain, particularly in the case of p.t.n.s because at least some of these must have been cortico-spinal neurones directly influencing the cord. In the cat there is of course at least one interneurone between the cortico-spinal terminals and the α -motoneurones (Lloyd, 1941; Lundberg & Voorhoeve, 1962; Illert, Lundberg & Tanaka, 1976) and it is conceivable that the excitability of these cells is so low as to restrict greatly the cortico-spinal influence on the motor output. However, cortical volleys artificially produced by weak electrical stimulation via the microwire electrodes exert profound short-latency effects on locomotion in progress at the time (D. M. Armstrong & T. Drew, unpublished), so this possibility seems remote.

Secondly, it is possible that the activity changes which were observed in a small proportion of units were sufficient to adapt the muscle forces to the prevailing conditions of speed or gradient. However, this would seem to imply that small changes in cortical output lead to rather substantial changes in the spinal motoneurone pools.

Thirdly, it may be speculated that the control of volitional movements at the primate wrist differs from that of semi-automatic movements such as those of steady walking: for the latter the responsibility for adjusting muscle force to meet mechanical requirements might to a much larger extent be delegated to subcortical (i.e. brain stem and/or spinal) motor centres and the cortical discharges might have some quite other role to play. If this were the case then most cortical neurones would not be expected to display the functional characteristics which would fit them for helping to determine the levels of muscle force developed under different conditions of speed or gradient.

Here it may be noted that in monkeys performing a lever-pulling task, the time of onset of agonist muscle contraction correlates very closely with the time of the maximum instantaneous frequency in some individual p.t.n.s discharging in association with the movement (Porter, 1972). This suggests strongly that, at least during precise volitional movements, p.t.n.s have an important role in determining the timing as well as the force of muscle contractions. It is therefore possible that during locomotion it is the former aspect of p.t.n. function which is most important and that these neurones are especially concerned with helping to determine the precise time at which some aspect (or aspects) of the stepping performance takes place.

In this connexion it is noteworthy that in decerebrate cats Orlovsky (1972*a*) was able to reset the rhythm of stepping in the hind limb by electrically stimulating the medullary pyramid. It may also be relevant that as walking speed increased in the present experiments some cortical units (including p.t.n.s) showed progressive advances or delays in their discharge timing (relative to the onset of triceps brachii activity) even when they showed little or no change in mean or peak rate. At the same time comparably sized phase shifts also occur between the locomotor e.m.g.s of some forelimb muscles. For example, as speed increases e.m.g. onset in the elbow flexors is advanced progressively whilst, by contrast, activity in dorsiflexors of the wrist is delayed, relative to triceps brachii onset (D. M. Armstrong & T. Drew, unpublished).

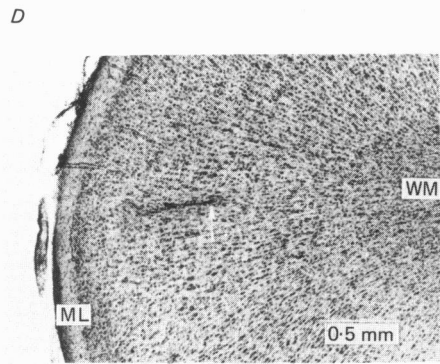
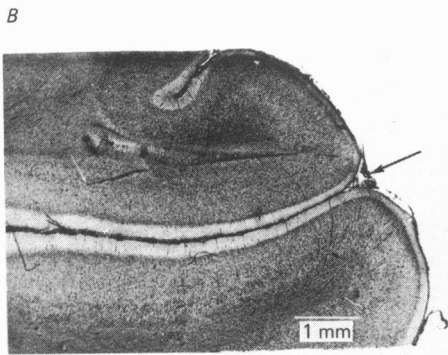
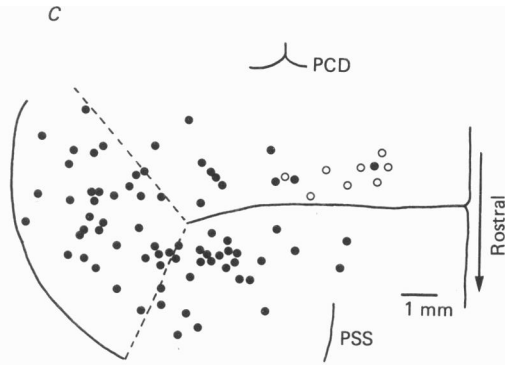
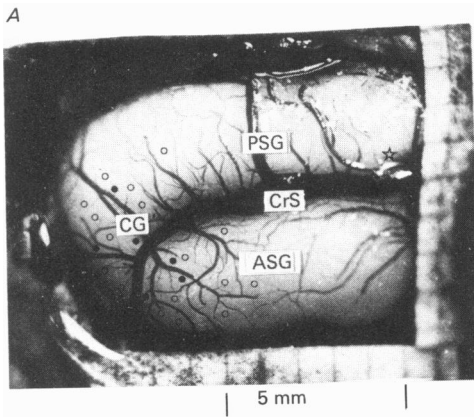
Further investigations will obviously be required in order to test the 'timing' speculation offered above and in particular it will be necessary to investigate the nature of the temporal correlations between the discharges of the motor cortical neurones and the e.m.g.s of different muscles under conditions when successive step cycles are less uniform in duration than in the present experiments.

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REFERENCES

- ARMSTRONG, D. M. (1965). Inhibitory actions on pyramidal neurones. B.Sc. Thesis, Oxford University.
- ARMSTRONG, D. M. & DREW, T. (1980*a*). A simple method for extracellular recording from motor cortex neurones during locomotion in the cat. *J. Physiol.* **303**, 14*P*–15*P*.
- ARMSTRONG, D. M. & DREW, T. (1980*b*). Motor cortex discharges during locomotion in the cat. *Abstr. XXVIII int. Congr. physiol. Sci.* **14**, 387 (1267).

- ARMSTRONG, D. M. & DREW, T. (1984). Locomotor-related discharges in cat motor cortex compared with peripheral receptive fields and evoked movements. *J. Physiol.* **346**, 497-517.
- ARMSTRONG, D. M. & RAWSON, J. A. R. (1979). Responses of neurones in nucleus interpositus of the cerebellum to cutaneous nerve volleys in the awake cat. *J. Physiol.* **289**, 403-424.
- BROOKS, V. B., RUDOMIN, P. & SLAYMAN, C. L. (1961). Sensory activation of neurons in the cat's cerebral cortex. *J. Neurophysiol.* **24**, 286-301.
- BURNS, B. D., STEAN, J. P. B. & WEBB, A. C. (1973). Recording for several days from single cortical neurones in the unrestrained cat. *J. Physiol.* **231**, 8-10P.
- EIDELBERG, E. (1981). Consequences of spinal cord lesions upon motor function, with special reference to locomotor activity. *Prog. Neurobiol.* **17**, 185-202.
- EVARTS, E. V. (1965). Relation of discharge frequency to conduction velocity in pyramidal tract neurons. *J. Neurophysiol.* **28**, 216-228.
- EVARTS, E. V. (1968). Relation of pyramidal tract activity to force exerted during voluntary movement. *J. Neurophysiol.* **31**, 14-27.
- GRILLNER, S. (1974). Locomotion in vertebrates: central mechanisms and reflex interactions. *Physiol. Rev.* **55**, 247-304.
- ILLERT, M., LUNDBERG, A. & TANAKA, R. (1976). Integration in descending motor pathways controlling the forelimb in the cat. I. Pyramidal effects on motoneurons. *Exp. Brain Res.* **26**, 509-519.
- LANCE, J. W. & MANNING, R. L. (1954). Origin of the pyramidal tract in the cat. *J. Physiol.* **124**, 385-399.
- LEWIS, M. MCD. (1974). Cited in Phillips, C. G. & Porter, R. (1977). *Corticospinal Neurones. Their Role in Movement*, p. 268. New York & London: Academic Press.
- LEWIS, M. MCD. & PORTER, R. (1974). Pyramidal tract discharge in relation to movement performance in monkeys with partial anaesthesia of the moving hand. *Brain Res.* **71**, 245-251.
- LIVINGSTON, A. & PHILLIPS, C. G. (1957). Maps and thresholds for the sensorimotor cortex of the cat. *Q. Jl exp. Physiol.* **42**, 190-205.
- LOYD, D. P. C. (1941). The spinal mechanism of the pyramidal system in cats. *J. Neurophysiol.* **4**, 525-546.
- LUNDBERG, A. & VOORHOEVE, P. (1962). Effects from the pyramidal tract on spinal reflex arcs. *Acta physiol. scand.* **56**, 201-219.
- NIBOULLON, A. & RISPAL-PADEL, L. (1976). Somatotopic localisation in cat motor cortex. *Brain Res.* **105**, 405-422.
- ORLOVSKY, G. N. (1970). Activity of reticulospinal neurons during locomotion. *Biophysics* **15**, 728-737.
- ORLOVSKY, G. N. (1972a). The effect of different descending systems on flexor and extensor activity during locomotion. *Brain Res.* **40**, 359-371.
- ORLOVSKY, G. N. (1972b). Activity of vestibulospinal neurons during locomotion. *Brain Res.* **46**, 85-98.
- ORLOVSKY, G. N. (1972c). Activity of rubrospinal neurons during locomotion. *Brain Res.* **46**, 99-112.
- PALMER, C. (1978). A microwire technique for recording single neurons in unrestrained animals. *Brain Res. Bull.* **3**, 285-289.
- PALMER, C., BAK, M. J., DOLD, G. M. & SCHMIDT, E. M. (1979). Stable simultaneous single unit recordings from groups of motor cortical neurons in the unanaesthetized and unrestrained cat. *Neurosci. Abstr.* **5**, 381 (1281).
- PALMER, C., MARKS, W. B., BAK, M. J. & PEDERSEN, G. (1980). The activity of closely spaced motor cortical pyramidal tract projecting neurons during locomotion. *Neurosci. Abstr.* **6**, 158 (56.10).
- PHILLIPS, C. G. (1959). Actions of antidromic pyramidal volleys on single Betz cells in the cat. *Q. Jl exp. Physiol.* **44**, 1-25.
- PHILLIPS, C. G. & PORTER, R. (1977). *Corticospinal Neurones. Their Role in Movement*. New York & London: Academic Press.
- PORTER, R. (1972). Relationship of the discharges of cortical neurones to movement in free-to-move monkeys. *Brain Res.* **40**, 39-43.
- RUSSELL, D. F. & ZAJAC, F. E. (1979). Effects of stimulating Deiter's nucleus and medial longitudinal fasciculus on the timing of the fictive locomotor rhythm induced in cats by DOPA. *Brain Res.* **177**, 588-592.



- SCHMIDT, E. M., BAK, M. J. & McINTOSH, J. S. (1976). Long-term chronic recording from cortical neurons. *Exptl Neurol.* **52**, 496-506.
- TAKAHASHI, K. (1965). Slow and fast pyramidal tract cells and their respective membrane properties. *J. Neurophysiol.* **28**, 908-924.
- UDO, M., ODA, Y., TANAKA, K. & HORIKAWA, J. (1976). Cerebellar control of locomotion investigated in cats: discharges from Deiter's neurones, EMG and limb movements during local cooling of the cerebellar cortex. *Prog. Brain Res.* **44**, 445-459.

EXPLANATION OF PLATE

A, photograph of cortical surface showing points of insertion of chronically implanted micro-electrodes in one animal. ●, successful electrodes; ○, unsuccessful. CrS, cruciate sulcus; ASG and PSG, anterior and posterior sigmoid gyri; CG, coronal gyrus.

B, diagram to show entry points of all successful electrodes. ●, electrodes inserted singly; ○, those inserted in bundles (see Methods). PCD, post-cruciate dimple; PSS, pre-sylvian sulcus. Broken lines divide off CG from ASG and PSG.

C, sagittal section through cerebral hemisphere to show tracks left by micro-electrodes inserted in a bundle into posterior sigmoid gyrus. Small white arrow shows a track ending in grey matter. Large arrow indicates cruciate sulcus.

D, sagittal section through coronal gyrus to show terminal portion of a microwire track (arrow). ML, molecular layer; WM, subcortical white matter.