Discharges of Single Hindlimb Afferents in the Freely Moving Cat

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SUMMARY AND CONCLUSIONS

1. Implanted semimicroelectrodes were used to record single afferent fiber discharges from L_7 dorsal roots during unrestrained walking in the conscious cat.

2. A series of tests were used to identify an afferent during a short period of anesthesia following each recording session. The majority of afferents were from muscle spindle primary endings in hindlimb muscles.

3. Ankle extensor spindle primaries generally showed their highest firing rates during that phase of stepping in which they were passively stretched. During active muscle contraction there was evidence of fusimotor drive, although this was not usually sufficient to entirely overcome the unloading effect of rapid muscle shortening. The variability of firing rate from cycle to cycle was considerably greater for the phase of active muscle contraction. The EMG response to brisk stretches of the ankle extensor muscle indicated a rapid (disynaptic or trisynaptic) reflex arc in the conscious animal.

4. Knee flexor spindle primaries showed similarly higher firing rates during passive muscle stretching in the step cycle. The shorter periods of presumed α-γ coactivation corresponded to the much more phasic role of these muscles in stepping.

5. Tendon organs in the physiological extensors of the toes were mainly active during stance, although some discharges were usually seen during the swing phase. It is suggested that previous experiments on mesencephalic preparations may have led to an exaggerated view of the degree of α-γ coactivation during normal stepping movements.

INTRODUCTION

The discharges of single mammalian first-order afferents have rarely been recorded in the conscious animal. It is, therefore, of considerable interest that the behavior of presumed muscle spindle afferents in the jaw muscles of conscious cats (27) and monkeys (5, 15) seems to differ from that expected from current theories. The bulk of the afferents showed an unexpectedly high firing rate during movements where their muscle was passively stretched, and generally a surprisingly low degree of apparent fusimotor drive during active contraction.

This is at variance with data from anesthetized (4, 23) and decerebrate preparations (24, 25) where the large majority of muscle spindles were active mainly during muscle contraction.

It is similarly at variance with the discharge patterns of presumed spindles in conscious humans (26, 28, 29). However very few records of spindle discharge during actual movement have been presented by these latter workers. The joint movements never exceeded more than a few degrees. It should be pointed out that in most of the conscious recordings to date, the identification tests have generally been rather cursory and, therefore, at least some of the afferents may have innervated other types of receptors.

In the present study, recordings of single fiber discharges were obtained from the L_7 dorsal roots of fully conscious cats during normal walking. This involved the implantation of dorsal root semimicroelectrodes, which have been described in preliminary reports (18, 20).

An important aspect of the experiments was the identification procedure, which took place during short periods of anesthesia immediately after each afferent recording session. This allowed physiological and pharmacological testing of the given afferent under controlled conditions.

Due to the relatively large electrode tips, the recordings heavily favored group I afferents. The electrodes were "free-floating" and, therefore, single-fiber recordings relied on chance movements of the electrode tips into stable locations near dorsal root axons. Fortunately, this was generally found to occur at least two or three times over the course of a few days. As a result, recordings were obtained of the activity

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of a population of carefully identified hindlimb afferents during normal large-scale movements.

METHODS

The discharges of 28 single afferent nerve fibers were recorded from spinal roots during walking in 10 cats (2.5–3 kg). Of these afferents, 20 were identified physiologically during short periods of anesthesia which immediately followed each walking session. The nerve fiber discharges and associated muscle length and electromyogram (EMG) were recorded using radio telemetry.

Dorsal root electrodes

Each electrode was made from a 15-mm length of 17-μm insulated Karma alloy wire (Driver Harris Ltd., Stockport, Cheshire, England). The insulation was stripped 2 or 3 mm from each end by applying pressure with a micrometer. One end was given three coats of varnish (Epoxylite 3000 M), with 30 min baking at 150°C between each coat.

One varnished end was now cut to a bevel using a scalpel blade mounted over a glass slide bearing the wire. The other end was wound around and soldered onto a shielded insulated connecting wire (Cooner AS632). This junction was insulated with a coating of silicone wax (Dow Corning 630) and a further coating of silicone elastomer (Dow Corning 384).

Smaller diameter (10 μm) wires were used in two implants, but their Teflon insulation proved to be insufficiently resilient.

Electrode stability

The maximum time after implantation for which single-fiber recordings could still be obtained was 10 days. Essentially, the technique relies on the chance juxtaposition of the dorsal root electrode tip and an afferent fiber. As the tip dimensions are relatively large, one would expect to record single-fiber discharge trains only from the larger afferents and this has, in fact, been borne out by the results.

Some electrodes were not selective enough, some were unstable, and some remained silent throughout. The usual cause of final failure was the withdrawal of the wire from the root. However, the number of times stable recordings were obtained was remarkable considering the presumably large relative movements of spinal structures in the freely behaving animal.

EMG electrodes

The EMG electrodes, modified from those described previously (19), were made from pairs of flexible multistrand insulated stainless steel wire; 1 mm of insulation was stripped from the ends. A 4-cm length of 5/0 silk suture thread was tied to each wire some 2 mm from the end, and the wire was folded back at the knot. The thread was passed through the muscle using a 21G needle, and the bared portion of wire was then pulled into the muscle. The thread was now sewn into a 2-mm-diameter, 200-μm-thick plastic button so that the button lay flush with the surface of the muscle.

Electrodes implanted in this way were found to remain in place for many weeks.

Muscle-length gauges

The mercury-in-rubber length gauges (19) were modified so that the contacts at either end simply consisted of tapered platinum rods thrust into the lumen of the tubing. The tubing at each end of the gauge was formed into a 3-mm-diameter loop and embedded in a few drops of dental acrylic. The loops reduced the direct forces tending to pull the rubber tubing away from the platinum contacts.

Implantation

A laminectomy (removal of L_7 spinous process and laminae) were performed with halothane anesthesia. Precautions were taken to ensure the maximum possible degree of sterility. Pairs of dorsal root electrodes were stuck to the dura with drops of isobutyl cyanoacrylate. The electrode tips were manually inserted through a small slit in the dura into L_7 dorsal root bundles just proximal to the ganglion. The connecting cable was passed subcutaneously to the head.

EMG electrodes were implanted in the biceps femoris and lateral gastrocnemius muscles. A length gauge was implanted between the calcaneum and the head of the tibia (19). Connecting cables were passed subcutaneously to the head, along with a heparinized catheter from the jugular vein.

The cable sockets and catheter hub were now embedded in acrylic around two skull screws. A prefabricated plastic bayonet socket was included in the skull cap. This provided a mounting for the detachable telemeter capsule.

The implantation procedure lasted from 2.5 to 3.5 h. The cat was treated with penicillin and local anesthetics and allowed to recover for 24 h before recording was attempted.

Telemetry devices

The plastic telemeter capsule (2 x 2.5 x 3 cm) held two battery-driven FM transmitters. One was a two-channel (EMG and length) device described previously (19). The second transmitter was a high-gain 200-MΩ input-impedance de-

vice, using essentially the same high-gain amplifier and oscillator as the first, but with a field effect transistor source-follower input stage.

Experimental procedures

Recording sessions commenced the day after implantation. The telemetry capsules were clipped onto the animal's head, and miniature plugs were mated with their appropriate sockets (Fig. 1).

The animal walked over a flat surface to retrieve food pellets. The three channels of transmitted recordings were decoded by FM receivers and recorded on magnetic tape using a Tandberg 100 FM tape recorder. In some experiments, a Statham pressure transducer measured the downward force exerted on a 2 cm x 2 cm thrust plate.

Single nerve fiber discharges were recorded for some 20 or 30 steps. The telemetry EMG connector was now changed over so as to record from the other pair of EMG electrodes, and another 20 or 30 steps were recorded. The cat was then quickly anesthetized via the indwelling jugular catheter using a short-duration anesthetic: methohexital sodium (Lilly). The afferent was then carefully identified.

Afferent identification

The receptor was located by its discharge response to palpation, muscle stretch, and discrete skin stimuli. The maximum elicitable dynamic index was obtained by rapid and maintained stretches of the appropriate muscle (Fig. 2*A*). Taps of graded amplitude and speed were applied over the receptor region (Fig. 2*D*). Vibration from 10 to 1,000 Hz was applied over the appropriate tendons and muscles (e.g., Fig. 2*E*: instantaneous firing frequency of an afferent during vibration at 360 Hz).

If on the basis of the responses to these stimuli the afferent was suspected of innervating a muscle receptor, the muscle was stimulated electrically using a surface electrode consisting of two sharp stainless steel wires 6 mm apart pressed down onto the skin. Spindle afferents are generally silenced during a small twitch contraction of their parent muscle, the primaries showing a rapid discharge in the descending phase of the twitch (Fig. 2*C*), whereas tendon organ afferents increase their firing in the ascending phase.

A dose of succinylcholine chloride (200 μg/kg) was administered via the jugular catheter. This selectively increases the dynamic index of spindle primaries (22). Figure 2*B* shows the dynamic

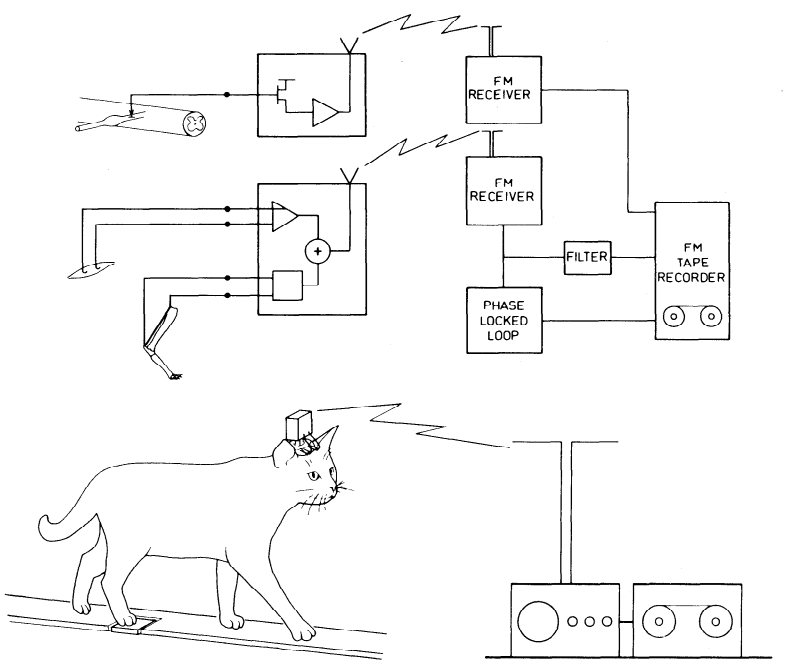

FIG. 1. Block diagram showing the telemetry system. Two separate FM transmitters were carried in the detachable capsule on the cat's head. One amplified and transmitted nerve spikes from the dorsal root microelectrode. The other was a two-channel device, handling both the length gauge and EMG signals. The transmitted signals were demodulated at the two receivers and recorded on magnetic tape. A small thrust-plate transducer was sometimes used to monitor downward force for one stance phase of a sequence of steps. The hub of a jugular catheter was included in the head socket. This allowed intravenous administration of short-term anesthetic.

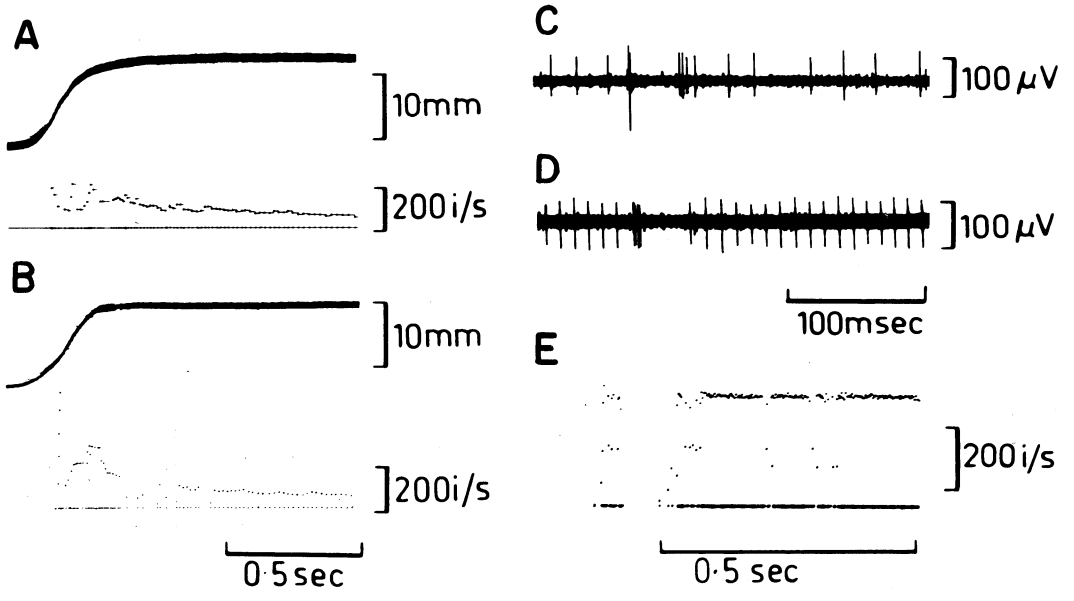

FIG. 2. Responses of a muscle spindle primary afferent to routine identification tests. *A*: instantaneous firing frequency (bottom trace) in response to rapid and then maintained muscle stretch (length gauge record: upper trace). *B*: same test after succinylcholine. *C*: response to electrically evoked muscle twitch. Stimulus artifact followed by a pause in discharge, then rapid resumption of firing at the end of twitch. *D*: discharge responses to light taps over muscle belly. Note high dynamic sensitivity. *E*: instantaneous firing frequency during vibration at 360 Hz over tendon. One-to-one following achieved after small adjustments of position of vibrator probe.

response of the same spindle afferent as in Fig. 2*A*, 1 min after a dose of succinylcholine.

The range of tests used on each afferent reduced to a minimum the possibility of false localization and identification. Doubts raised after one or two tests (e.g., a high vibration sensitivity and medium dynamic index) were dispelled by the responses to the further tests (e.g., no succinylcholine effects, rapid discharge during ascending phase of muscle twitch).

The criteria used to classify an afferent as a spindle primary were: silencing during small electrically evoked muscle twitches with a phasic response on the descending phase, high dynamic sensitivity to taps and stretches, high sensitivity to vibration over the muscle belly and tendon at frequencies above 100 Hz, a large increase in dynamic index after succinylcholine.

Spindle secondary criteria: silencing during small electrically evoked muscle twitches; low dynamic sensitivity; low vibration sensitivity, especially over the tendon; no increase in dynamic index after succinylcholine, but some increase in tonic firing rate.

Tendon organ criteria: accelerated discharge during muscle twitch; medium dynamic sensitivity to taps; low, medium, or high sensitivity to vibration; no change in dynamic index or tonic firing frequency after succinylcholine. The initial fasciculations caused by intravenous doses of succinylcholine may cause phasic increases in tendon organ firing rate, but this effect only lasts a few seconds. The passive stretch sensitivity of tendon organs increases at lighter levels of anesthesia when some muscle tone is present.

A technique using obliquely cut wire electrodes fixated at the L_7 spinous process has been developed independently by G. E. Loeb, National Institutes of Health, Bethesda, Md. This technique has yielded recordings of the discharges of single cells in the dorsal root ganglion during locomotion in the conscious cat (personal communication).

RESULTS

Step cycle correlates

The EMG activity of the main hindlimb muscle groups has been shown to relate closely to the various phases of the step cycle (2, 3).

Figure 3 shows a recording of the length of the triceps surae muscles (ankle extensors), lateral gastrocnemius muscle EMG, and the forces exerted downward by first the front, and then the hind paws on a small pressure plate during walking. The symbols F, E^1, E^2, and E^3 refer to the four phases of the step cycle defined by Philippson (17), and analyzed in detail by Goslow, Reinking, and Stuart (6).

Flexion (F) of the ankle joint begins as the foot

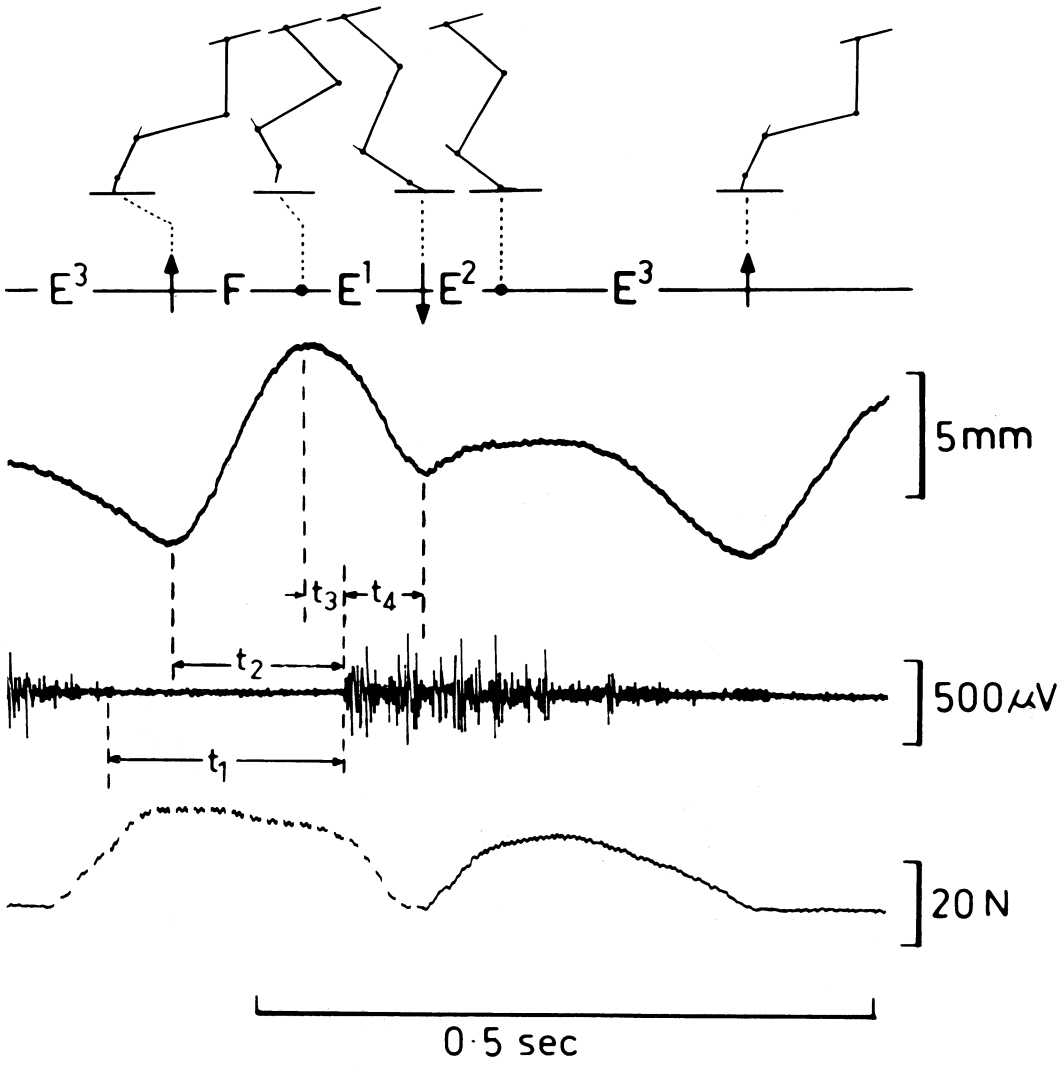

FIG. 3. Muscle length and EMG activity during the step cycle. Top trace: signal from length gauge in parallel with the ankle extensor muscles of the left hindlimb. Middle trace: EMG from lateral gastrocnemius muscle. Bottom trace: thrust-plate force signal for one stance phase only (first peak has been dashed to indicate that it refers to forepaw thrust). Phases of step cycle (F, E^1, E^2, and E^3) after Philippson (17). Hindlimb joint angles after Goslow et al. (6). Measurements of t_1, t_2, t_3, and t_4 were made for a large number of step cycles of different amplitude and duration.

is lifted from the ground for the forward swing. The triceps group is passively stretched during F. The first extension (E^1) is accompanied by electrical activity in these muscles, and corresponds to the lowering of the foot toward the ground. On foot contact, the triceps muscles are generally stretched slightly (E^2), and the active stance phase then continues into E^3 as the animal's center of gravity moves forward.

Because some of the afferent recordings lacked a muscle length record, it was necessary in these cases to estimate the relative timing of the phases of the step cycle from the onset and cessation of lateral gastrocnemius EMG.

Measurements of t_1, t_2, t_3, and t_4 as defined in Fig. 3, were made from filmed recordings of 120 step cycles from each of two cats walking normally over a flat surface. The onset and cessation of EMG were established using a criterion level of 5% of the average EMG during stance.

Figure 4 shows that t_4, the time from onset of EMG to foot contact, was relatively constant (70 ± 25 ms (SD)) for a large number of steps of widely differing duration. On the other hand, t_2

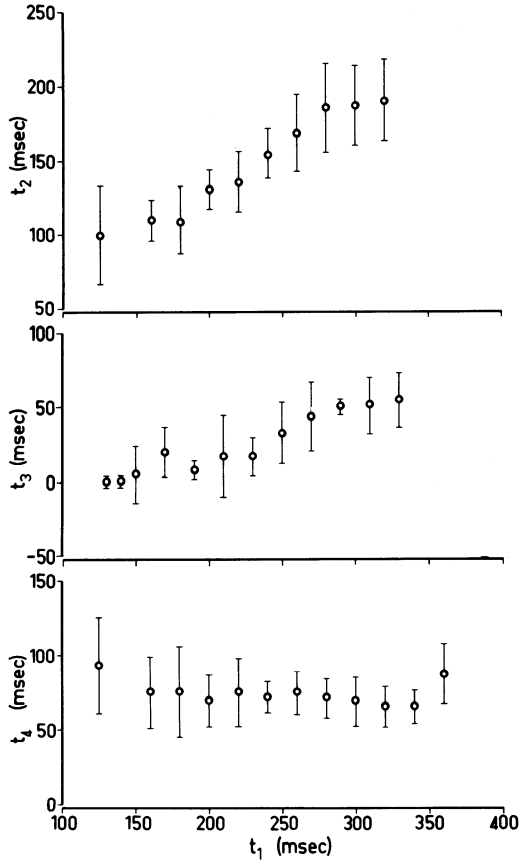

FIG. 4. Time relationships defined in Fig. 3 related to periods of silence of lateral gastrocnemius muscle (t_1) for 120 step cycles. Each ordinate is an average of values over a range of 50 ms of t_1. Standard deviations were calculated for an average of 12 values in each case. The top and middle graphs show that times from start and end of flexion to onset of EMG increase with increasing duration of EMG silence. The bottom graph shows a relative constancy of latency of EMG onset to foot contact. The low standard deviations (ca. 25 ms) allow estimates of t_2, t_3, and t_4 from a knowledge of t_1.

and t_3 increased with increases in step duration. Because of the constancy of t_4, it was found that for very short steps, the short duration of the E^1 phase meant that the EMG onset sometimes occurred during the F phase (negative values of t_3).

The variability in the above latencies was at least partially attributable to the different speeds and postural relationships during the steps. No attempt was made to standardize such parameters, although the mode of locomotion always remained walking (as opposed to trotting). Nevertheless, the standard deviation of latencies for any given value of t_1 was sufficiently low to allow relatively accurate predictions of phase.

It should be mentioned here that the range of values of t_1 (125–360 ms) was much larger than the range of durations of swing ($t_2 + t_4$): 165–290 ms. Relatively small increases in the duration of the swing phase are coupled with larger increases in the duration of the E^3 phase, and a correspondingly earlier reduction in EMG activity toward the end of stance.

In the subsequent recordings, the EMG of the lateral gastrocnemius and biceps femoris muscles were taken as representative of the activity of the combined ankle extensors and the combined biarticulate hamstring group, respectively. The validity of this comparison will be discussed later.

Types of afferents

Only those afferents whose discharges were recorded for at least 5 min are reported here. Of the 28 afferents held during walking, 5 were lost prior to or during the induction of anesthesia and 3 were lost during the identification tests.

Of the 20 afferents identified, 15 were spindle primaries, 2 were tendon organ afferents, 2 were from slowly adapting skin receptors, and 1 was from a rapidly adapting skin receptor. No recordings have yet been obtained from spindle secondaries.

The distribution of Ia afferents was as follows: dorsal femoral (hamstrings), 6; triceps surae, 5; gluteal muscles, 2; flexor digitorum longus, 1; intrinsic foot muscle, 1. Both tendon organs were located in the flexor digitorum longus tendon (in different cats). One slowly adapting skin receptor was located over the gluteal region and the other on a footpad, and the rapidly adapting skin receptor was located over the hamstring muscles.

There exists in the literature a vast amount of neurophysiological data on the ankle extensors, particularly the soleus muscle. For this reason, the ankle extensor spindle primary recordings will be treated in detail.

Triceps surae spindle primaries

The behavior of the five triceps spindles ranged from predominantly "passive" to significantly "active."

Figure 5 shows the discharge train of the most passive spindle primary during three consecutive step cycles. Certain features of the activity were very constant and common to all of the triceps spindle afferents. First, there was nearly always a reduction in firing rate toward the end of the (E^3) stance phase, followed by an acceleration of firing during passive muscle stretch in the flexion (F) phase of the swing. Generally a pause, or at least a reduction in firing rate, oc-

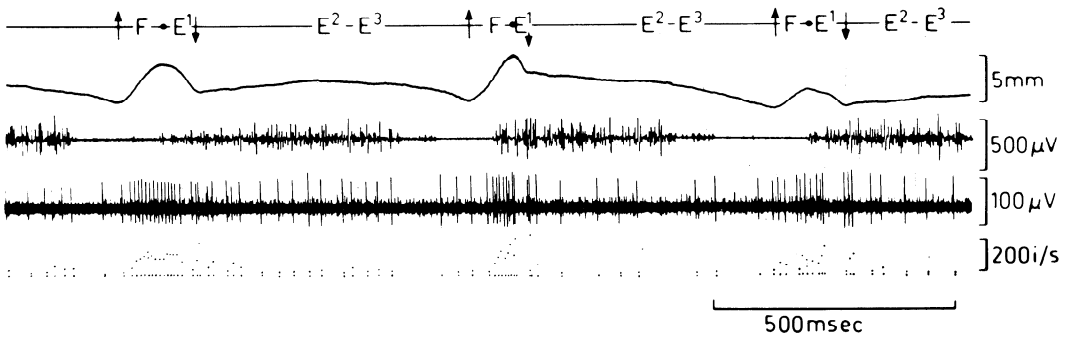

FIG. 5. Discharge train of passive ankle extensor spindle primary afferent (third line from top, dots in bottom trace indicating instantaneous firing frequency). Length and EMG of lateral gastrocnemius muscle in top traces. Consistent modulations in firing frequency included: reduction toward end of stance (E^3); increase during passive muscle stretch (F); pause during muscle shortening in latter half of swing phase (E^1); rapid firing on foot contact followed by lower firing rates during stance.

curred during the E^1 phase, just after the onset of lateral gastrocnemius EMG. Often a burst of discharges occurred at foot contact, which was followed by varying frequencies of discharge during stance.

In the record of Fig. 5 there was a consistent increase in firing rate just prior to flexion, despite the fact that the parent muscle group was shortening. This suggests that even this very passive spindle received at least some fusimotor drive.

Further evidence for fusimotor drive may be seen in the E^1 phases of the first and especially the third steps, where a high firing rate was maintained after muscle stretching had ceased.

The pause in discharge just prior to foot contact must naturally be partly attributed to muscle shortening. However, there may also have been some withdrawal of fusimotor drive because in some steps (e.g., the third) there was an abrupt cessation of firing without a corresponding change in the velocity of shortening.

The discharge rate of this spindle during the stance phase was consistently very low and rather variable. It showed little direct correspondence with the level of lateral gastrocnemius EMG activity.

During F, the firing rate occasionally peaked well before the end of flexion and sometimes before the muscle-lengthening velocity had reached a maximum. Similar spindle primary discharge patterns were reported by Goslow et al. (7) for passive muscle stretching within the locomotor range. These workers attributed the early peaking of firing rate to "stiction," a phenomenon presumably related to the forces set up by the breaking of intrafusal muscle cross bridges.

However, in the present study such peaks of discharge rate were relatively rare and might equally have been the result of phasic fusimotor activity.

Figure 6 shows the firing patterns of three futher triceps surae spindle primary afferents during single step cycles. In these cases the length gauge was not functional, and so the phases of the step cycle were estimated from the cessation and onset of lateral gastrocnemius EMG, using time relationships derived from Fig. 3.

The spindle primaries of Fig. 6*A*, *B* showed evidence of an intermediate amount of fusimotor drive, whereas Fig. 6*C* represents the most extreme degree of fusimotor drive of the most active spindle primary observed.

The main features of the response described for the most passive spindle may also be seen in these three discharge trains. There was a reduction in firing rate prior to the cessation of lateral gastrocnemius EMG toward the end of stance (E^3). An acceleration of firing rate occurred during EMG silence (flexion—F phase). A pause or reduction of firing rate occurred shortly after the onset of EMG and bursts of firing were observed at a latency consistent with foot contact.

The onset of lateral gastrocnemius EMG was rarely accompanied by any marked acceleration of firing beyond the rate attained during the passive stretch (F) phase. In fact, the pauses during E^1 suggest that there is very little excitation available to the homonymous muscle group via the afferent route in the first 30 or 40 ms of activation.

There seemed to be no direct relationship between spindle primary firing rate and amplitude of EMG. In particular, no significant fixed-latency EMG response was detectable after high-frequency bursts accompanying foot contact. The identification procedure did not permit the localization of spindles to particular muscles

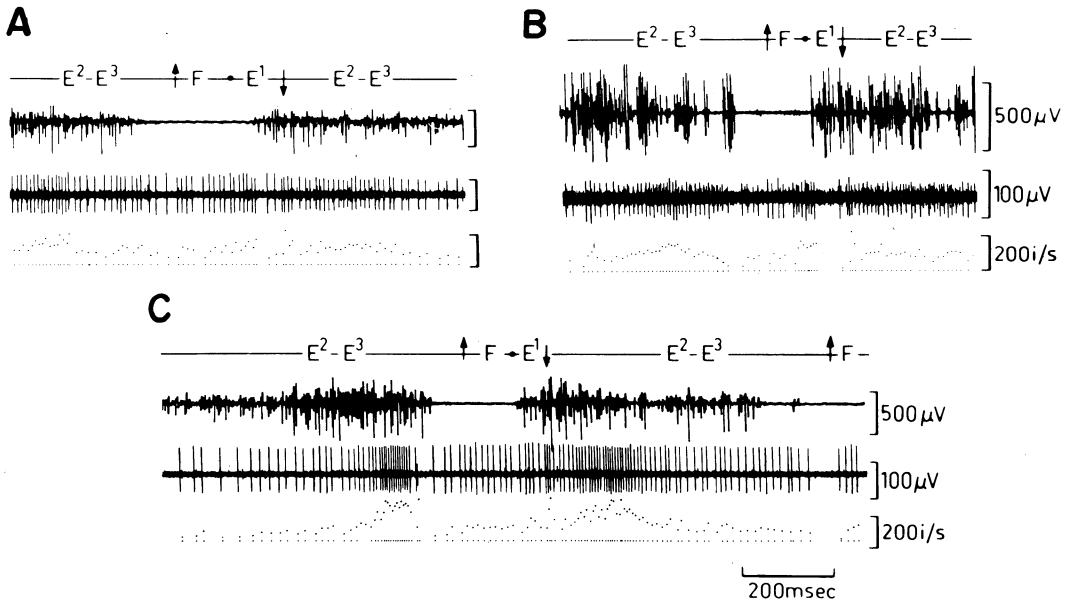

FIG. 6. Discharge trains of intermediately active (*A* and *B*) and very active (*C*) ankle extensor spindle primary afferents during step cycles. Phases of the step cycles were estimated from relationships shown in Fig. 3. Upper traces: lateral gastrocnemius EMG; lower traces: afferent discharges and their instantaneous firing rate. All three afferents showed the characteristic firing patterns described in Fig. 5, but with higher firing rates during stance. *C* represents the most extreme degree of presumed α-γ coactivation observed. Note lack of correspondence between primary afferent firing rate and EMG amplitude.

of the triceps group and it is, therefore, possible that the lateral gastrocnemius EMG would not be an exact measure of the spindle's homonymous extrafusal muscle's activity. On the other hand, one might reasonably expect all of the spindle primaries of the group to respond in a similarly phasic manner on foot contact and, therefore, any efferent responses to the afferent barrage would be common to the whole group.

ACTIVE AND PASSIVE BEHAVIOR. Figure 7 illustrates the firing behavior of three of the ankle extensor primary afferents ranging from the most passive to the most active. For each afferent, three step cycles have been chosen ranging from the most passive to the most active firing behavior of that particular afferent.

Spike trains were aligned with the onset of lateral gastrocnemius EMG prior to foot contact. The spike trains are represented by horizontal rows of dots. The times of foot lift-off and contact were estimated from the data of Fig. 3 for the second and third afferents and are shown as open arrows (the standard deviations of these estimates are 25 ms). EMG cessation and onset occurred at the times indicated by the vertical dashed lines.

To establish a quantitative measure of the relative degree of fusimotor activation in the stance phase, the maximum firing rates averaged over 100 ms were calculated for the E^2–E^3 and F phases, respectively. The ratios of these rates were then calculated for every step cycle, and the segments of Fig. 7 were selected as representing the minimum, mean, and maximum values of this ratio for each afferent.

It is evident from Fig. 7 that the main changes in relative firing rates occurred in the E^2–E^3 phases (stance). Table 1 shows that the average rates of firing in the F phases were less variable than those in the E^2–E^3 phases. This suggests that the largest variations in fusimotor drive tend to occur in the active contraction phase.

It was also found that after anesthesia, at ankle angles of about 90° the more active afferents had higher tonic firing rates (Table 1).

REFLEX BEHAVIOR. In one experiment, the cat was resting on its side but was otherwise quite alert. Brisk taps were applied to the footpad causing a small, rapid, passive flexion of the ankle. Figure 8 shows the resulting lateral gastrocnemius EMG and triceps spindle primary discharges. Although a length trace was obtained, it is not shown in this record because the transducer had an inherent phase lag for stretch frequencies above 100 Hz. The commencement of stretch was more accurately defined by the

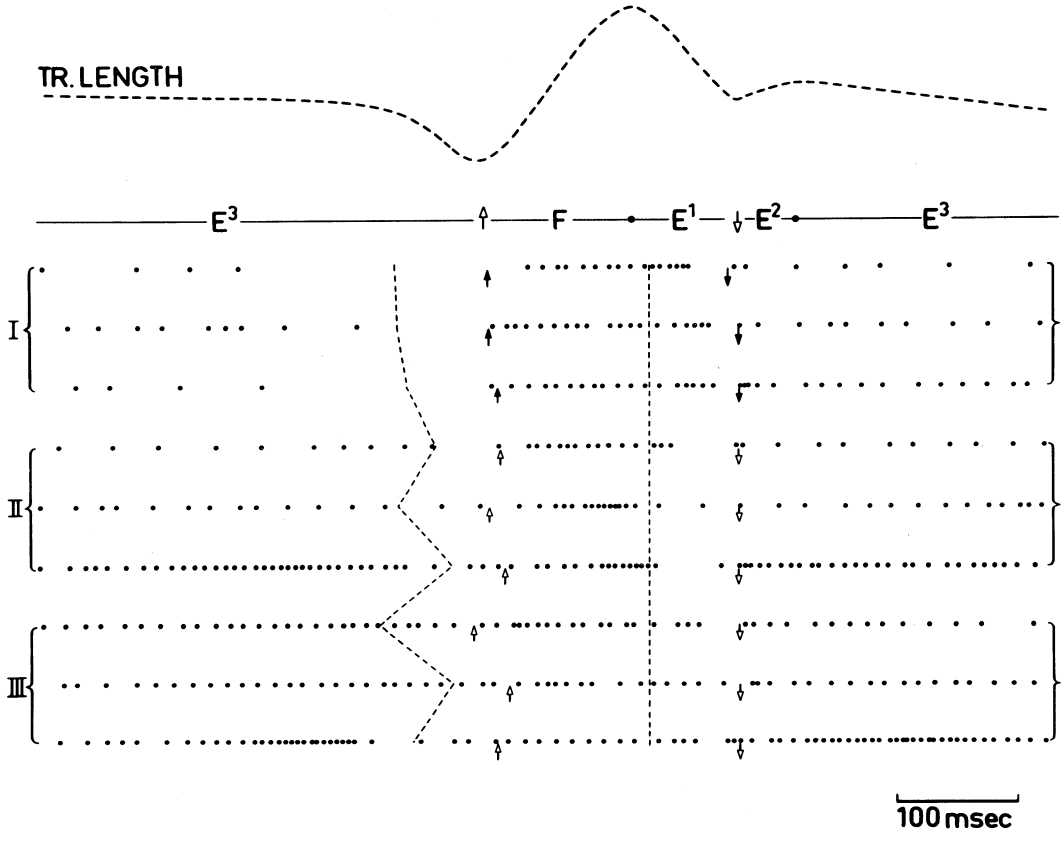

FIG. 7. Firing patterns of three ankle extensor spindle primaries ranging from most passive afferent (I) to most active afferent (III). For each afferent, three step cycles where chosen, ranging from the lowest to the highest relative firing rates during stance. The spike trains are represented by horizontal rows of dots. The left vertical dashed line indicates times of cessation of lateral gastrocnemius EMG, the right vertical line shows times of EMG onset. Times of foot liftoff and contact are shown as upward and downward arrows, respectively (open arrows being estimates). The approximate length of the ankle extensor muscles during the step cycles has been included as a dashed curve above the dot raster.

initial small EMG movement artifact (arrowed).

The shortest observed latency between the first afferent spike and the first small component of the polyphasic EMG response was 6.2 ms. The average latency of this component for six stretches was 6.5 ms.

In order to establish whether this latency also applied to large rapid stretches during stance, the cat was placed so that the appropriate hindlimb stood on a flat board which protruded over the side of the walking platform. A brisk downward blow to the protruding part of the board caused a rapid upward movement of 1 or 2 cm of that part supporting the hindlimb.

TABLE 1. *Comparison of firing rates of five triceps surae spindle primaries during stepping and after anesthesia*

Afferent	Max Rate in F	Max Rate in E^2–E^3	Avg "Drive" Ratio	Avg Anes Rate, imp/s (Ankle Angle ca. 90°)
1	100 ± 11	53 ± 19	0.5	1–5
2	127 ± 17	84 ± 28	0.7	10–15
3	128 ± 22	107 ± 34	0.8	5–15
4	124 ± 21	115 ± 36	0.9	15–20
5	92 ± 12	116 ± 38	1.3	20–25

Values are in impulses per second ± SD.

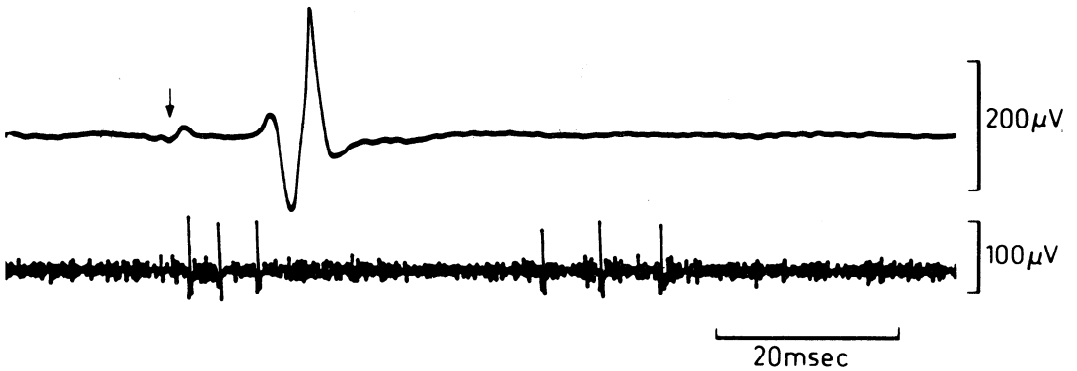

FIG. 8. Responses to a brisk stretch of the ankle extensors in the conscious cat. Top trace: lateral gastrocnemius EMG; bottom trace: discharges of ankle extensor spindle primary. The initial small wave in the EMG signal (arrowed) was a movement artifact. The arrow indicates the start of muscle stretch. The latency of the first subsequent EMG deflection from the first afferent spike was 6.2 ms. This corresponds to at most a trisynaptic delay.

The average latency of the first increase in EMG measured from the movement artifact was 8.4 ms. This experiment was done on a subsequent day, when the afferent fiber was no longer recordable. However, if the delay between rapid muscle stretch and the arrival of primary afferent impulses at the dorsal root is estimated at 2.4 ms (14), the average latency of the EMG for the large stretches was closely similar to that for the small stretches.

Knee flexor spindle primaries

The six hamstring spindles all responded to passive knee extension and hip flexion during the identification trials. They were, therefore, presumably located in the biarticulate division of this group: biceps femoris, semitendinosus, and semimembranosus posterior.

Figure 9 shows the discharge behavior of one of these primaries during two step cycles of different duration. During identification, this afferent had its lowest threshold to taps over the distal part of biceps femoris just dorsal to the proximal third of the tibia.

The EMG was recorded from the biceps femoris muscle. The length records refer to the triceps group, and the dashed lines are estimates of the corresponding length changes of the knee flexors from the data of Goslow et al. (6).

The hamstring spindle afferents nearly always showed two peaks of discharge, one associated with the burst of biceps femoris EMG at the end of E^3, just prior to flexion (F), and the other during the latter part of F and the whole of E^1, where the hamstring muscles are rapidly stretched. During the remainder of the step cycle, the firing rate was generally (but not always) lower.

The high rate at the end of E^3 may reasonably be attributed to fusimotor drive, assuming that there is relatively little muscle length change in this phase.

In the records of Fig. 9*A*, *B*, it is evident that the bursts of EMG activity during the latter part of F and during E^1 corresponded closely to peak rates of spindle firing. Furthermore, in the slower step of Fig. 9*C*, the firing rate during these phases are relatively low and no EMG activity occurred.

General observations

Most of the spindle primaries showed "bursting" firing patterns when the animal was relaxed, either sitting or lying. The conversion from steady firing to bursting, or vice versa, was generally abrupt. Bursting patterns could occasionally be observed during stepping, especially among the hamstring spindles in the E^2–E^3 phases.

The spindle primary response to slow and maintained stretching of a muscle in the conscious animal was an increase in firing rate roughly proportional to joint angle, although modulations of the firing rate could occur spontaneously at a given extension, usually associated with changes in the animal's level of arousal.

During one of the knee-flexor afferent recordings, at a time when the cat was lying on its side in a relaxed state, a few milliliters of water were squirted onto the hindblimb footpad, causing a rapid flexor withdrawal. The spindle afferent firing rate doubled simultaneously with the first onset of biceps femoris EMG. After 50 ms at this higher firing rate, there was a ca. 50-ms pause and then a resumed rapid firing for the duration of the flexion movement.

Assuming that the knee flexors started short-

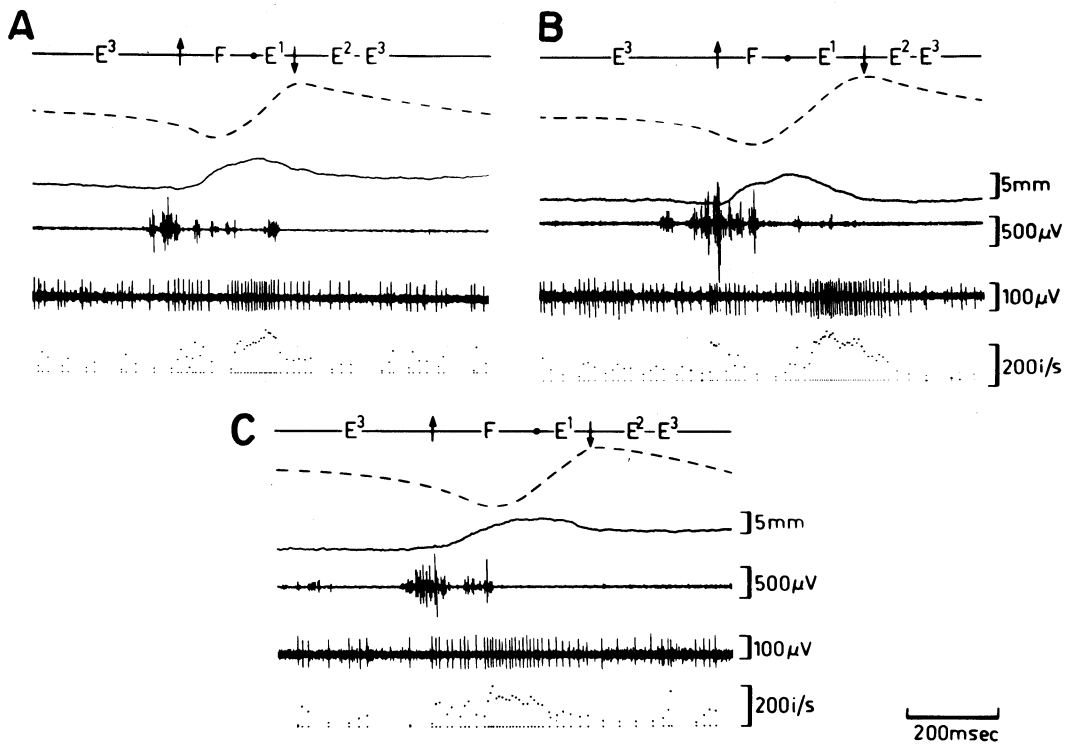

FIG. 9. Discharge trains of a hamstring muscle spindle primary afferent during three step cycles. Length-gauge signal (calibrated) refers to the length of the triceps surae group, giving phases of cycles (top). Dashed lines approximate length of hamstrings group (after Goslow et al. (6)). EMG was recorded from biceps femoris muscle. The afferent had peak firing rates (*1*) just prior to F when biceps femoris showed bursts of EMG, and (*2*) during F and E^1 when the hamstrings were rapidly stretched. In *A* and *B*, the latter peaks were associated with small bursts of EMG. In the slower step of *C*, the afferent firing rate during E^1 was lower and the EMG burst was absent.

ening some 40 or 50 ms after the first onset of EMG, it would seem that the fusimotor drive was insufficient to overcome at least the first short period of spindle unloading (no length record was available from the hamstrings group).

Tendon organ afferents

Only two afferents recorded during walking were subsequently identified as tendon organs. Their spike trains are shown for step cycles in Fig. 10*A*, *B*. The receptors were located in the flexor digitorum longus muscle, a physiological extensor of the toes lying deep to the triceps surae group. The recordings are from two different cats.

The length traces refer to the triceps surae group and the EMG was from the lateral gastrocnemius muscles. Both afferents fired mainly during stance, although generally there were one or two spikes in both the F and E^1 phases. The afferent of Fig. 9*B* was more phasic than that of Fig. 10*A*, this being most evident during the yield phase of E^2 after foot contact.

The more phasic afferent was found to follow vibration to 180 Hz after anesthesia, whereas the other did not follow at any frequency above 10 Hz. Both afferents had a very low threshold to passive stretch at light levels of anesthesia when muscle tone had developed.

DISCUSSION

Ankle extensor spindles

During normal stepping, hindlimb spindle primaries were generally active throughout the cycle of movement. This was true of extensor as well as flexor afferents. The ankle extensor spindles had consistently high discharge rates during passive muscle stretch and generally lower rates during muscle activity in the stance phase. This is in contrast to the mesencephalic results (24, 25) where the rates during stance were considerably higher.

It has been pointed out that the method of fixation of the lower spine in the mesencephalic preparation would distort the step cycle, limiting

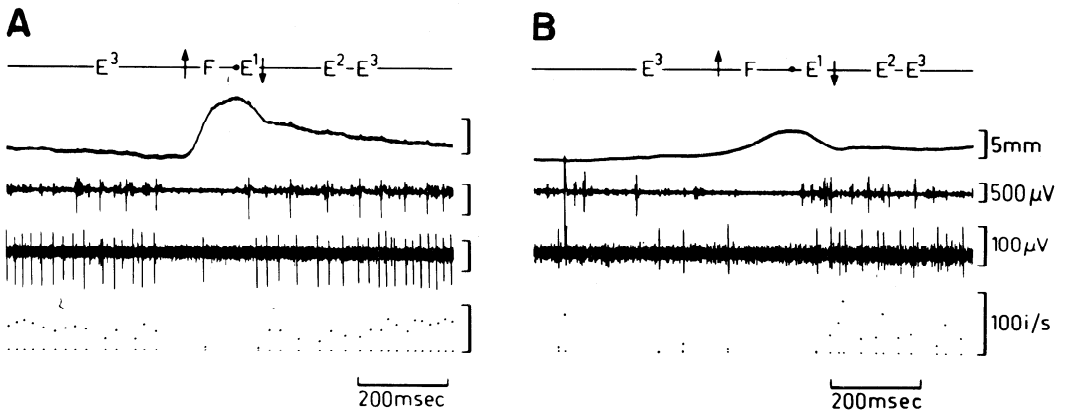

FIG. 10. Discharges of two toe extensor tendon organ afferents during stepping. The length and EMG traces refer to the lateral gastrocnemius muscle in each case. *A*: this afferent fired tonically during stance, although two discharges are evident during the swing phase (F–E^1). *B*: this afferent showed a more phasic firing behavior during stance, but also exhibited discharges during the swing phase.

the duration of the F phase and prolonging the E^2 phase (7). Furthermore, there is very likely to be a difference in the balance of the segmental reflex effects in the decerebrate animal compared to normals.

However, the deceleration of discharge just prior to flexion, the pause during the E^1 phase and the phasic response on foot contact which were so characteristic of our conscious recordings are also clearly evident in the two triceps surae spindle afferent records presented by the Moscow group. Apart from the exaggerated activity during stance, it would therefore seem that the basic pattern of fusimotor drive to spindle primaries during stepping in the mesencephalic preparation is basically similar to that in the conscious animal.

The firing patterns of our hindlimb spindle primaries showed much the same relationships to muscle length and EMG as did the jaw muscle afferents of Goodwin and Luschei (5). If horizontal jaw movements are neglected, the three phases of the chewing cycle in their records, namely, opening, fast close, and slow close are closely analogous to the F, E^1, and E^2–E^3 phases of the step cycle. On this basis, the high discharge rates during opening, the pauses during fast closing, and the resumed activity during slow closing are similar to the corresponding triceps surae spindle discharge trains presented above.

Some differences did exist, however. Even our most passive spindle (Fig. 5) was active throughout the cycle. Furthermore, the ratios of average discharge frequencies in the active contraction and passive stretch phases was generally higher in the present study. This may be due to higher velocities of stretching of the jaw muscles, or it may indeed reflect a higher relative potency of fusimotor drive to hindlimb spindle primaries.

In this context it should be pointed out that the identification procedure used by Goodwin and Luschei (5) was generally restricted to "gentle surface palpation of the temporalis and masseter muscle." As pointed out by these authors, it is not clear whether they were recording from spindle primaries or secondaries. The histological evidence that the trigeminal mesencephalic nucleus contains only first-order afferents derives from studies on the cat (1). The possibility of synaptic influences on these cells in the monkey cannot entirely be ruled out. Furthermore, the trigeminal mesencephalic nucleus is narrow and lies in the vicinity of populations of second-order cells whose responses can mimic those of the first-order afferents (13).

The lack of a tight coupling between ankle extensor spindle firing and EMG during stance reinforces the observations of Taylor and Cody (27). In EMG studies on reflexes in humans, it is usually necessary to average a large number of sweeps in order to enhance the stimulus-locked EMG response. This is because of the inherently stochastic nature of the summation of motor unit discharges. A reflex response to the spindle afferent barrage occurring after foot contact might, therefore, exist without any obvious corresponding change in the EMG.

However, large and relatively slow modulations in the spindle firing rate during muscle activity were clearly not accompanied by similar fluctuations in the homonymous EMG. It was also quite obvious that at the onset of contrac-

tion in both the triceps surae and hamstring muscles, there was very little supportive excitation available from the spindle primaries.

There is evidence from our recordings that fusimotor drive occurred both during active muscle contractions and passive muscle stretch. In the latter case, the low variability of firing rates suggests that the drive was fairly stereotyped and relatively weak. A similarly lower variance in firing rate during passive stretch was observed by Goodwin and Luschei (5).

The pause in triceps surae spindle firing just prior to foot contact may be due to a withdrawal of fusimotor drive. Alternatively, it may reflect a relatively high dynamic fusimotor component throughout F and E^1, although this would not explain those cases of abrupt cessation of firing at a constant velocity of shortening. It is interesting to note that the single flexor digitorum longus spindle from which we recorded also exhibited a pause at the same latency. This muscle, a physiological extensor, undergoes a slight lengthening in the E^1 phase (6).

The amount of fusimotor drive to a given spindle primary during the step cycle would seem to be related to the steady firing level of the afferent during moderate methohexital anesthesia. This may be the result of a residual amount of fusimotor drive after anesthesia, reflecting the relative degree of fusimotor innervation to the spindle. Alternatively, it may be a measure of the static stretch sensitivity of the primary ending both in the driven (conscious) and passive (anesthetized) states.

Stretch reflex

The average latency of the first EMG response to rapid stretches of the triceps surae was 6.5 ms after the arrival of the first spindle primary discharge at the dorsal root. Allowing 1.5-ms efferent conduction delay along the fastest α-fibers and a further 1.0 ms for the generation of the first motor-unit action potentials, this leaves about 4 ms attributable to spinal delay. This is a maximum figure because it is unlikely that the afferent discharges were sufficiently synchronized to recruit the largest motoneurons immediately.

The spinal delay was, therefore, consistent with a reflex arc involving, at most, three synapses. Furthermore, the EMG response was often subsequently maintained for some tens of milliseconds. This is in contrast to the EMG response to stretch in conscious humans (11) where the main component occurs some 30 or 40 ms after the monosynaptic latency.

To extend the present observations, it would be desirable to supplement the chronic recordings with subsequent acute experiments involving dorsal and ventral root stimulation in the spinalized animal.

Knee flexor spindles

In general the hamstring primaries showed similar firing patterns in relation to passive muscle stretch and active contraction to those of the ankle extensors. The longer periods of relative quiescence might well be attributable to the more phasic nature of the activity of this muscle group during stepping.

There was no evidence that the flexor spindles showed a markedly higher relative firing rate than the extensors during passive muscle lengthening, as was the case for the ankle flexor spindles in the mesencephalic preparation (25). However, this difference cannot be ruled out as the material in the present study precluded comparisons between flexors and extensors acting around the same joint.

Unlike ankle extensors, knee flexors show bursts of EMG activity during the phase of the step cycle in which they are stretched by their antagonists (3). In our recordings these EMG bursts corresponded in time to the peaks of firing rate of knee flexor spindle primaries, and it seems reasonable to suppose that the EMG was mainly the result of Ia excitation of the knee flexor motoneuron pool.

It should be pointed out that in this period of the swing, hip flexion and knee extension result in a rapid forward movement of the leg. The forces developed by the knee flexors (which also have a hip extensor role) would presumably aid in limb deceleration.

It is particularly interesting, for example, that during trotting, these EMG bursts are very well developed (3). The rates of stretching of the knee flexors during trotting are greatly increased (6) and, therefore, their spindle firing rates are presumably considerably higher.

The pause in hamstring spindle discharge associated with rapid flexion withdrawal indicates that in this case, as well as during stepping movements, fusimotor drive may not always be provided in sufficient measure to counter the unloading effect of muscle shortening.

The bursting behavior of spindle primaries seen in the relaxed animal and sometimes during the stance phases of stepping points to a continuous static fusimotor drive (21), even at low levels of arousal.

Tendon organs

The discharges during stepping of the two tendon organ afferents demonstrated that these receptors had a sufficiently low threshold to be active for much of the cycle.

The EMG activity of the flexor digitorum longus muscle has an almost identical time course to that of the lateral gastrocnemius muscle (ref 2; unpublished observations). It is, therefore, of interest that there were consistent early discharges in the E^1 phase after the onset of EMG but before foot contact.

Similar early discharges may be seen in the triceps surae tendon organ recordings of Severin et al. (25). The latency of these spikes would be consistent with an activation of the receptor by the earliest motor unit contractions (12). Additionally, there is evidence for a small amount of stretch of the flexor digitorum longus muscle during the E^1 phase (6) and this, coupled with the early muscle activity, would seem adequate to give rise to the early discharges.

Spinal organization of stepping

It would seem that for flexors as well as extensors, the main components of α-motoneuron activity during stepping are initiated centrally, with little indirect excitation via the γ-loop. From our recordings there is good reason to believe that fusimotor drive to spindles accompanies such efferent activity, but its relative potency would seem lower than previously suggested (25).

Rhythmical locomotor contractions have recently been obtained after complete transection of all dorsal roots in the mesencephalic cat (10). The basic locomotor program, therefore, exists centrally, but may presumably be modified by afferent input (8).

In the present study, the only clear example of such a modification was the close correlation of knee flexor EMG in the late swing phase with spindle primary discharge rate. Grillner and Zangger (10) claimed that deafferentation did not alter the EMG pattern of a knee flexor (semitendinosus) during induced locomotion. However, even before deafferentation, their records show a very late onset of this component of EMG compared with that in the conscious animal (3). Furthermore, in a very similar experiment, Perret and Cabelguen (16) recently reported that deafferentation did, in fact, abolish one of the two semitendinosus EMG bursts.

Apart from muscle spindles, many other receptors around the hip and knee joints would no doubt contribute excitation or inhibition to the generation of this late-swing component of knee flexor EMG, although presumably the knee flexor spindle primaries would have the most powerful excitatory effect.

It has been suggested that the role of the reflexes might be restricted to overcoming perturbations not predicted by the central locomotor program (9). Indeed the apparent lack of a tight coupling between spindle firing rate and EMG in the ankle extensors would support such a view. However, as in the instance of the knee flexors, there are presumably many cases where afferent input may combine with the spinal locomotor program during undisturbed movements.

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