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CHANGES IN DISCHARGE RATE OF FUSIMOTOR NEURONES PROVOKED BY FATIGUING CONTRACTIONS OF CAT TRICEPS SURAE MUSCLES

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SUMMARY

1. Changes in discharge rate of thirty-one fusimotor neurones to triceps surae muscles during long-lasting, fatiguing contractions of these muscles were studied in decerebrate cats. Discharges of fusimotor neurones were recorded from the nerve filaments. Muscle contractions were elicited by electrical stimulation of either the muscle nerves (twenty-one neurones) or the corresponding ventral roots (ten neurones), until the muscle tension fell to about 30% of its initial value.

2. Early and late changes could be recognized in fusimotor discharge rate during long-lasting muscle contraction. The early changes obviously not related to muscle fatigue, consisted of an initial increase at the onset of muscle contraction and a subsequent decrease to or below the resting discharge level. The late change in discharge rate, supposedly related to muscle fatigue, was an increase developing gradually towards the end of muscle contraction, ranging at its peak from 2 to 15 impulses/s (mean value 5·5 impulses/s, $n = 31$) and outlasting the contraction for 20–320 s.

3. When the contracting muscle was made ischaemic the late increase in fusimotor discharge rate started earlier and was maintained until the arterial clamp was removed. After severing the muscle nerves distal to the site of stimulation no changes, a slight sustained increase, or else a decrease in fusimotor discharge rate occurred during electrical stimulation of either muscle nerves or ventral roots. At its cessation the spontaneous firing rate was reassumed immediately. Stimulation of the distal stumps of the severed nerves elicited no changes in fusimotor discharge rate.

4. It is proposed that the late increase in fusimotor discharge rate may appear due to autogenetic excitation of fusimotor neurones by discharges from group III and IV muscle afferent fibres provoked and/or enhanced by metabolic products liberated in muscle tissue during the fatiguing contraction. The fusimotor firing was estimated to remain elevated to a level twice that of the spontaneous activity on average for approximately 120 s after the muscle contraction. Its functional role in muscle fatigue is discussed.

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INTRODUCTION

It has been shown that discharges from group III muscle afferents exert strong autogenetic reflex effects on fusimotor neurones when elicited by either electrical stimulation of muscle nerves (Appelberg, Hulliger, Johansson & Sojka, 1983*b*) or by more physiological stimuli such as isometric twitch contractions of the parent muscle (Ellaway, Murphy & Tripathi, 1982) or algesic agents and lactic acid applied by close arterial injection (Jovanović, Anastasijević & Vučo, 1990). Metabolic products liberated in muscle tissue during contraction have been shown to elicit discharges in group III and IV muscle afferents (Thimm & Baum, 1987; Rotto & Kaufman, 1988) or to enhance their responses to muscle contraction (Rotto, Schultz, Longhurst & Kaufman, 1990), while an increase in both responsiveness as well as in resting discharge of mechanosensitive group III afferents develops during muscle fatigue (Hayward, 1990). While group III and IV afferent discharges have been proposed to play an important role in muscle fatigue by optimizing firing rates of skeletomotor neurones (Bigland-Ritchie, Dawson, Johansson & Lippold, 1986), their reflex effects on fusimotor neurones in muscle fatigue have not been investigated. The slowing of skeletomotor discharge rate during muscle fatigue has been supposed recently (Bongiovanni & Hagbarth, 1990) to be provoked by a decrease in support by autogenetic excitation through the γ-loop due to intrafusal fibre fatigue and/or an autogenetic inhibition of fusimotor neurones by group III and IV afferent discharges. However, reflex effects on fusimotor neurones of these afferent discharges cannot be predicted with enough certainty. Fusimotor neurones responded with an increase in discharge rate to either mechanically (Ellaway *et al.* 1982) or chemically (Hong, Kniffki & Schmidt, 1978; Jovanović *et al.* 1990) induced discharges in these afferents. When elicited by electrical nerve stimulation group III discharges may provoke either autogenetic excitation or inhibition of fusimotor neurones (Ellaway *et al.* 1982; Appelberg *et al.* 1983*b*), or else may be supposed to elicit recurrent inhibition (Ellaway, 1971; Noth, 1971) by exciting Renshaw interneurones (Piercey & Goldfarb, 1974). Muscle ischaemia, shown to induce (Mense & Stahnke, 1983) or increase (Kaufman, Rybicki, Waldrop & Ordway, 1984) responses of some group III and IV muscle afferent fibres to muscle contraction, unexpectedly failed to provoke any changes in fusimotor responses to short-lasting muscle contractions (Anastasijević, Jocić & Vučo, 1987). The present experiments are therefore undertaken to establish actual changes in fusimotor discharge rate developing when muscle fatigue is provoked by long-lasting contractions.

Preliminary results have been reported in abstract form (Anastasijević, Jovanović & Ljubisavljević, 1990).

METHODS

Surgical preparation

Experiments were performed on nineteen adult decerebrate cats. Three of the cats were spinalized at the T9 level in the course of the experiments. The operative procedure until decerebration was carried out under halothane-in-oxygen anaesthesia. The right hindlimb was completely denervated except for the triceps surae muscles. The nerves to medial gastrocnemius (MG) and to lateral gastrocnemius and soleus (LGS) muscles were freed from the surrounding tissue to be mounted, one or both of them, on platinum wire bipolar stimulating electrodes. Decerebration was performed by intercollicular section of the brainstem and the nervous tissue rostral to the

section removed. In the experiments where muscle contractions were elicited by ventral root stimulation lumbar laminectomy was performed and the dura cut to allow access to ventral roots L7 and S1. These roots were freed from the pia and mounted intact on stimulating electrodes. Cats were fixed to the stand by clamps on the third lumbar spine and the iliac processes, and screws in the right tibia and femur. The exposed tissues were kept moist in paraffin pools. Blood pressure and the temperature of both the animal and the paraffin pools were monitored and maintained within physiologically desirable ranges.

Recording techniques

Spike discharges of functionally single fusimotor neurones were recorded from thin filaments, dissected free from desheathed fascicles cut out of otherwise intact MG and LGS nerves. The discharging neurones were identified as fusimotor if their conduction velocity was in the range of 10–45 m/s. It was determined by back-averaging of impulse traffic in the parent nerve, triggered by single impulses recorded from the filament (Bostock & Sears, 1976). As described in a previous paper (Jovanović *et al.* 1990) activity from the whole MG or LGS nerve was recorded with bipolar electrodes and spike-trigger averaged (10 ms sweep time, 128 sweeps) after being delayed by 5 ms; the triggering single-neurone spikes (recorded from the dissected filament) were then delayed and averaged in the same way (Fig. 1*A*). A Neurolog system (Digitimer) was used for signal amplification, filtering, delaying and averaging. Conduction velocity was calculated from the distance of the two averaged signals displayed on the cathode ray oscilloscope screen (Tektronix 5103N Oscilloscope System), i.e. conduction time, and the distance between the two recording sites (on the whole nerve and the dissected filament, respectively). Muscle tension changes were recorded by a tension transducer attached to the tendon of the triceps surae muscles. The compliance of the transducer was 20 μm/N.

Muscle contraction

Isometric contractions of the triceps surae muscles were elicited by electrical stimulation of either the muscle nerves (both MG and LGS or the heteronymous nerve with respect to the fusimotor neurone recorded) or the corresponding ventral roots (L7 and S1). In both cases 0·2 ms electrical stimuli at 1·3 times motor threshold were applied at a rate of either 25 or 40 Hz until the muscle tension fell to approximately one-third of the initial value. In some experiments 7 s periods of stimulation were applied at 1 s intervals (see Results). The motor threshold was determined by observing muscle tension changes on the oscilloscope screen while the stimulus strength was gradually increased. The smallest apparent tension changes occurred usually at the stimulus strength in the range 0·10–0·16 V. The estimated motor threshold was checked at the end of the experiments applying the same procedure after severing the muscle nerves proximal to the site of stimulation. The muscle was held extended by 3 mm from the length at which the slack was just taken up.

Analysis of data

Fusimotor discharges were recorded before (60–120 s), during and after (at least 60 s) muscle contraction, stored on magnetic tape and/or analysed on-line on a Hewlett–Packard 9817 computer. The action potentials were converted to voltage steps, which were sampled at 10 ms intervals. This procedure was adopted in order to achieve, with the available computer memory, an on-line record, at a sampling rate high enough to detect every action potential, during a 300 s period. This was the shortest period allowing an immediate visual inspection of the whole time course of muscle contraction and the related changes in fusimotor discharge rate, while on-line 300 s records of background discharges served to check whether any longer-lasting spontaneous oscillations occurred in firing rate. Fusimotor neurones showing such oscillations were discarded from further analysis. The output signal from the tension transducer was amplified, stored on magnetic tape, and A/D converted (sampling interval 40 ms) simultaneously with fusimotor discharges. Actual signals of both fusimotor spike discharges (Fig. 1*B*) and muscle tension were also monitored on an oscilloscope screen and their changes observed during recording. Further analysis of records (Fig. 1*C*) was performed off-line. The number of impulses per 1 s interval as well as per 10 s interval was counted during the whole recording period (thick and thin line, respectively).

Mean value and the standard deviation of the spontaneous discharge rate (horizontal continuous and dashed lines) were computed from the counts per 1 s interval during 60 or 120 s periods immediately preceding the onset of stimulation (horizontal arrow). Moments at which the line drawn through the mean values of the counts per 10 s period (expressed in impulses/s) and the line

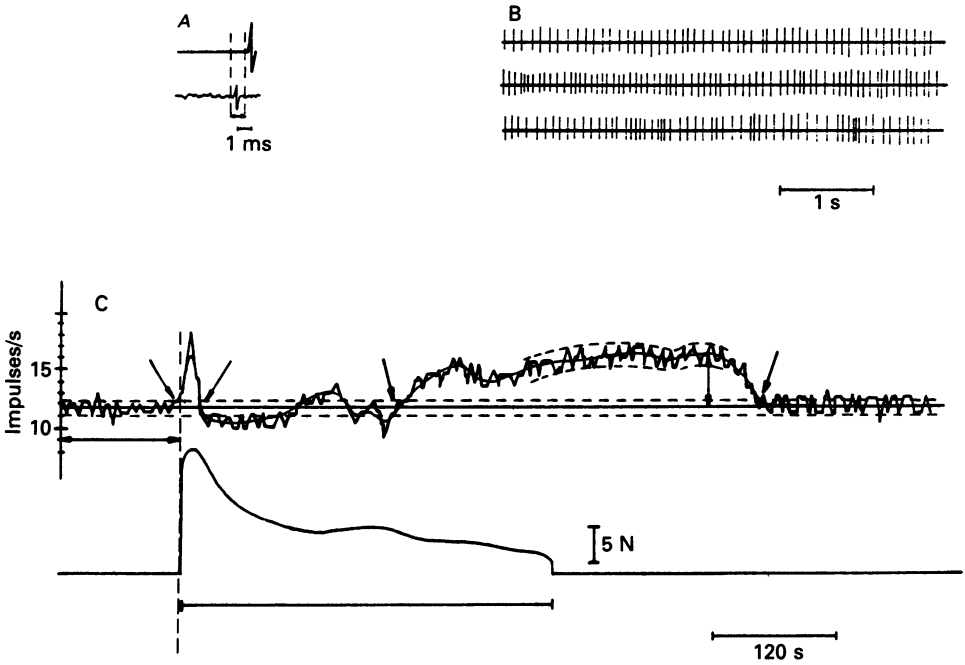

Fig. 1. An example illustrating identification of fusimotor neurones and estimation of changes in their discharge rate. *A*, records of a neurone spike (upper trace) from the nerve filament and the activity in the parent MG nerve (lower trace), both spike-trigger averaged and delayed. Conduction time (horizontal arrow) 1·2 ms; distance between the recording sites 24 mm; calculated conduction velocity 20 m/s. *B*, segments of actual records of spike discharges. From above downwards: immediately preceding the onset of LGS nerve stimulation; at the moment indicated by vertical arrow in *C*; at the end of the recording period shown in *C*. *C*, changes in discharge rate of the neurone during long-lasting muscle contraction provoked by continuous stimulation at 40 Hz of LGS nerve. Upper traces, fusimotor discharge rate (impulses/s). Interval of random fluctuations indicated by dashed lines during the period of the late increase in discharge rate. Lower trace, muscle tension changes (N). Vertical dashed line, onset of stimulation; the thick lowermost horizontal line, stimulation period.

drawn through the mean of the resting discharge rate intersect, depart or join (oblique arrows) were taken as the beginning and end of changes in fusimotor discharge rate. The magnitude of the changes was estimated from the segments of records at the moments of the largest departure of these two lines (vertical arrow). Whenever the actual discharge rate was maintained at a fairly constant level, different from the resting level, during a period lasting 60 or 120 s, statistical significance of the difference between their mean values was estimated using Student's *t* test. When there was no such segment, the difference between the actual and the spontaneous discharge rate was estimated roughly to be significant if the intervals limited by two standard deviations from the mean resting discharge rate and the interval covering the amplitudes of random fluctuations in the actual discharge rate did not overlap.

Control procedure

To check the reproducibility of changes in fusimotor discharge rate, the effects of muscle ischaemia, and the dependence on the preserved afferent inflow from the muscle, the following procedure was adopted: fusimotor discharges were recorded (a) while the same electrical stimulation was repeated; (b) while the contracting muscles were made ischaemic by clamping the femoral artery (throughout or towards the end of the stimulation period); (c) during stimulation after severing the muscle nerves distal to the site of recording. Time was allowed for the muscle to recover by introducing half-hour pauses between any two stimulations. The muscle was considered to have recovered when the tension changes provoked by the same stimulation applied after the period of rest were the same as before. Independent data exist to show that changes due to muscle fatigue in blood concentration of metabolites subside during this period of time (e.g. Duchateau, de Montigny & Hainaut, 1987; Juel, Bangsbo, Graham & Saltin, 1990).

RESULTS

The effects of long-lasting muscle contraction were studied on thirty-one fusimotor neurones from nineteen cats, including twenty-three MG and eight LGS units. Spontaneous discharge rate of the cells ranged between intermittent low rates (less than 5 impulses/s) to sustained rates of 23 impulses/s, and the conduction velocity of their axons ranged from 12 to 30 m/s.

Provoking of muscle fatigue

Muscle fatigue has been provoked in a variety of ways (e.g. Bigland-Ritchie *et al.* 1986; Garland, Garner & McComas, 1988; Hayward, Breitbach & Rymer, 1988) while either autogenetic reflex effects or those on the close synergist were observed. Relying mostly on the stimulation procedure used by Hayward *et al.* (1988) to provoke muscle fatigue in decerebrate cats, but trying also to make it more similar to a sustained maximal voluntary contraction (Bigland-Ritchie *et al.* 1986), we applied either continuous electrical stimulation of the muscle nerves at rates of 25 and 40 Hz, or 7 s trains of the stimuli at 1 s intervals. Either both nerves to triceps (to MG and LGS) or one nerve (to MG muscle if fusimotor discharges were recorded from a filament from LGS nerve and vice versa) were stimulated. Spike discharges of twenty-two fusimotor neurones were recorded during muscle contraction elicited by nerve stimulation, five of them while both nerves were stimulated and an additional sixteen cases during one-nerve stimulation. In order to avoid reflex effects of spike discharges in low-threshold muscle afferents elicited by necessity during muscle nerve electrical stimulation (though the possibility of provoking recurrent inhibition by antidromic impulses elicited by the stimuli in axons of skeletomotor neurones would remain), muscle contractions were provoked by electrical stimulation of the corresponding (either L7 or S1) ventral roots in five experiments, and changes in discharge rate of an additional ten fusimotor neurones recorded. Additional spinalization was performed in three cats in which the changes in fusimotor discharge rate during muscle contraction provoked by either muscle nerve (one experiment) or ventral root stimulation (two experiments) were rather small. It was expected that the reflex effects, if provoked by the discharges from high-threshold muscle afferents, would be enhanced after spinalization since the transmission in these reflex pathways might be suppressed in decerebrates (Eccles & Lundberg, 1959).

Changes in fusimotor discharge rate

Electrical stimulation of either both MG and LGS nerves, one nerve, or ventral roots, provoked an initial sharp increase in fusimotor discharge rate at the onset of the muscle contraction and a late slowly developing increase towards its end and

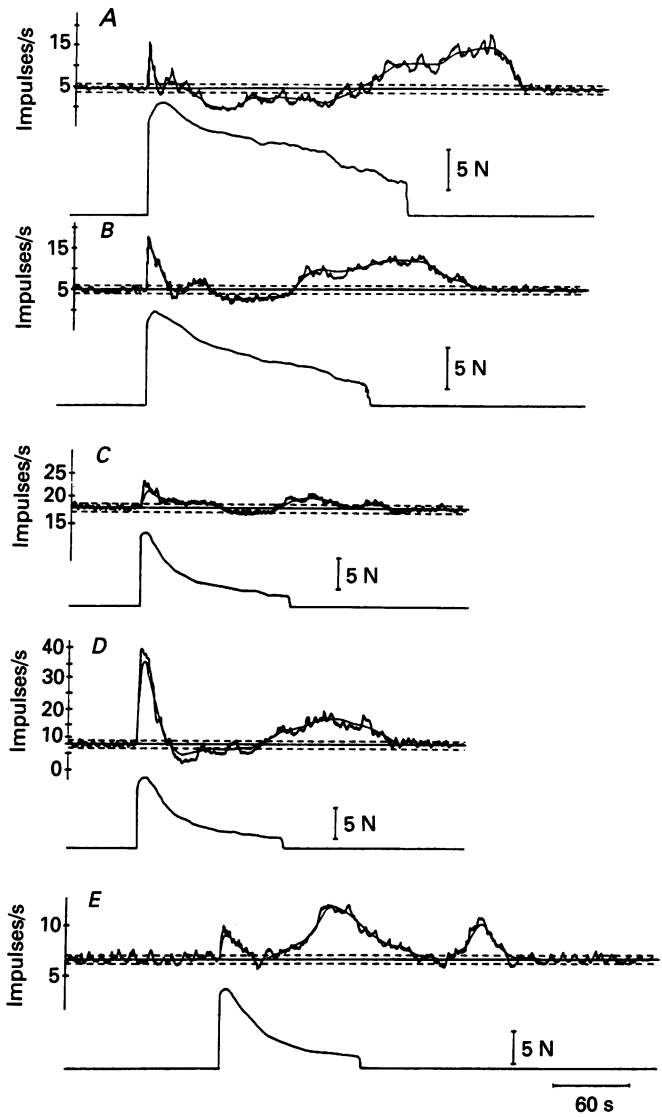

Fig. 2. Changes in discharge rate of several MG fusimotor neurones during long-lasting muscle contraction provoked in different ways: electrical stimulation, at a rate of 40 Hz, of both LGS and MG nerves (*A*), LGS nerve (*B*) and L7 ventral root (*C*, *D* and *E*). *D*, spinalized cat. Upper and lower traces same as in Fig. 1*C*. The line computed from 1 s periods smoothed by five-point averaging for the sake of clarity.

outlasting it. The initial increase, by 2–30 impulses/s (mean value 8·8 impulses/s, $n = 20$) with respect to the resting discharge level, appeared in all except one neurone at the onset of muscle nerve stimulation. In four of the neurones its peak, however, did not reach the values beyond the random fluctuation level. It lasted for up to 20 s in fifteen fusimotor neurones and was followed by a decrease below the resting level ranging from 3 to 6 impulses/s (twelve in one neurone), with a mean value of 3·6 impulses/s ($n = 14$). In the remaining five neurones the initial increase was prolonged (35–110 s), the discharge rate subsiding gradually to or slightly below the resting discharge level. The initial increase (by 4–35 impulses/s, mean value 16·1) during ventral root stimulation was prolonged in most neurones (10–60 s) and was followed by a gradual decrease to, but seldom below (two neurones, by 2 impulses/s), the resting discharge level. Whenever the initial increase was prolonged, the subsequent decrease did not reach the values below the resting discharge level beyond those of random fluctuations in discharge rate.

The late increase in discharge rate during muscle nerve stimulation ranged, with respect to the resting discharge level, from 2 to 11 impulses/s at its peak (mean 5·5 impulses/s, $n = 21$) and outlasted muscle contraction by 20–320 s. During ventral root stimulation it ranged from 2 to 15 impulses/s (mean 5·5 impulses/s, $n = 10$) and outlasted muscle contraction by 25–180 s. In four neurones (two cases during muscle nerve stimulation and two cases during ventral root stimulation) the increased discharge rate was within limits of random fluctuations. In three fusimotor neurones an additional increase, by 3–9 impulses/s at its peak, occurred after ventral root stimulation, starting 40–60 s after the end of muscle contraction and lasting for another 40–60 s. No differences were found in responses of MG and LGS neurones.

Examples of changes in fusimotor discharge rate during long-lasting muscle contractions provoked in different ways are shown in Fig. 2. A sharp short-lasting increase in discharge rate of a fusimotor neurone to medial gastrocnemius muscle occurred at the onset of muscle contraction elicited by continuous stimulation of either MG and LGS nerves or the LGS nerve only (*A* and *B*, respectively). It was followed, in this neurones, by oscillations around the mean resting firing level before falling below this level. Towards the end of muscle contraction a second increase developed in discharge rate and outlasted the contraction by about 90 s. The initial increase in discharge rate of another fusimotor neurone to MG muscle during muscle contraction provoked by ventral root stimulation (*C*) was prolonged, the subsequent decrease below the resting level almost lacking, while the late increase developed towards the end of muscle contraction and outlasted it. Both the early and late increase in discharge rate were rather small in this and another two fusimotor neurones from the same decerebrate cat. They were larger in a fourth fusimotor neurone recorded after spinalization (*D*). In the neurone shown in *E* an additional increase in discharge rate occurred after the end of muscle contraction provoked by ventral root stimulation.

Origin of the late increase in fusimotor discharge rate

While the early changes in fusimotor discharge rate are likely to correspond to already described reflex effects of discharges from mechanosensitive group III muscle afferents (Ellaway *et al.* 1982), and autogenetic (Ellaway & Murphy, 1980)

and recurrent inhibition (Ellaway, 1971; Noth, 1971), the late increase in discharge rate only might be related to muscle fatigue. To check whether these late changes in fusimotor discharge rate indeed represent reflex responses to changes in afferent inflow from the contracting muscle, and to differentiate the receptors responsible for

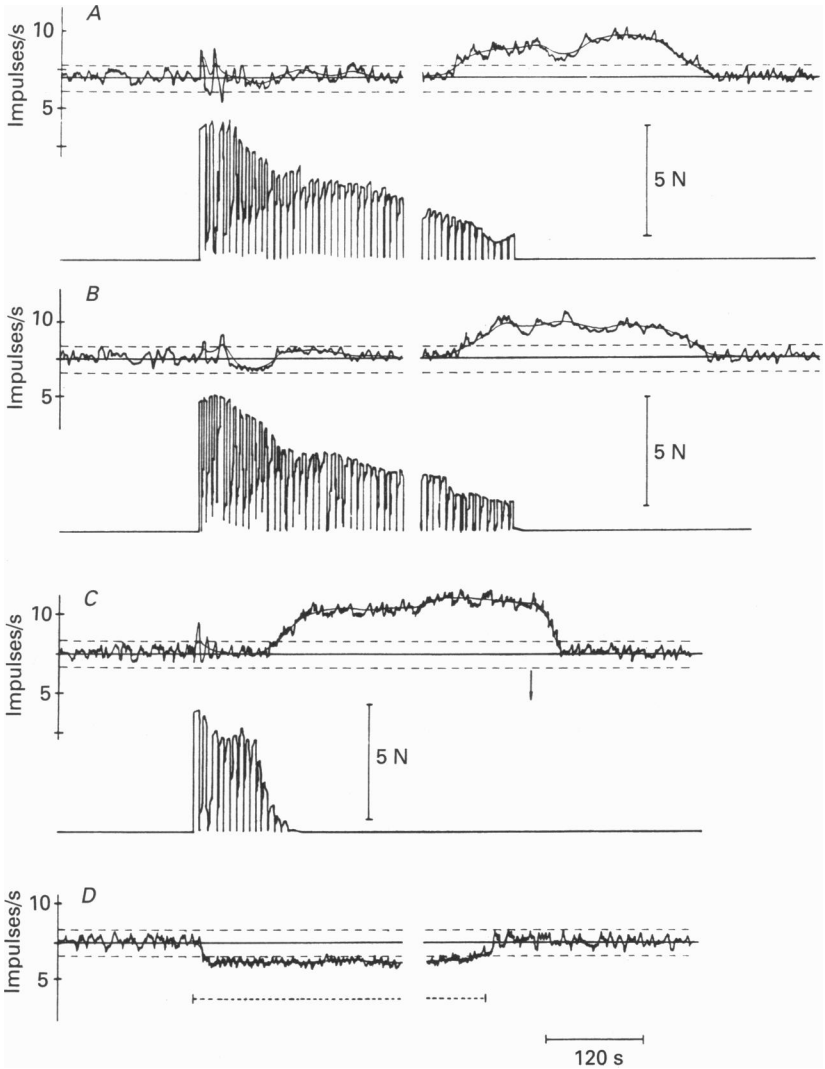

Fig. 3. Changes in discharge rate of a MG fusimotor neurone in control procedure. Muscle contraction provoked by a series of 7 s periods of electrical stimulation at 1 s intervals, at a rate of 25 Hz of the nerves to LGS muscles. Ischaemia in *C* started before the recording period, its end indicated by the arrow. Stimulation period in *D* indicated by the dashed line below the record. Upper and lower traces, same as in Fig. 2. In *A*, *B* and *D* right parts of the records start about 200 s after the end of the left parts.

the reflex effects, their reproducibility by repeating the same contraction after a period of rest, their dependence on the preserved reflex loop as well as the effects of muscle ischaemia were tested. An example is shown in Fig. 3. The late increase in

discharge rate of a fusimotor neurone developed towards the end of a series of short-lasting periods of muscle nerve stimulation (*A*). Similar muscle tension changes as well as an increase in fusimotor discharge rate of similar amplitude and duration occurred when the same stimulation was repeated after a period of rest (*B*). When the same stimulation was applied while the muscle was made ischaemic by clamping the femoral artery (*C*) both the muscle tension fall and the increase in fusimotor discharge rate started earlier. The increase was enhanced and maintained until the arterial clamp was removed. The same effect was encountered in another five fusimotor neurones when the muscle was kept ischaemic throughout the stimulation period. Muscle ischaemia *per se* did not provoke changes in discharge rate of fusimotor neurones (see also Anastasijević *et al.* 1987). In an additional six neurones the late increase in discharge rate seemed to be enhanced when the muscle was made ischaemic towards the end of the stimulation period. Transient increase in discharge rate, however, occurred occasionally at the moments of positioning and removing the arterial clamp while manipulating the skin. Though this increase could clearly be recognized on four occasions and similar changes were absent in the other trials, there was no way to prove or disprove the possibility that they might have been hidden within the presumed enhancement of the late increase in fusimotor discharge rate. Electrical stimulation of the proximal stumps after severing the muscle nerves was applied in eight fusimotor recordings and of the ventral roots in two more. It provoked no changes in discharge rate in three fusimotor neurones, a slight sustained decrease in another three units, and a slight sustained increase in four neurones. At the cessation of stimulation the discharge rate returned immediately to the resting level. The differences between the mean spontaneous firing rate and the firing rate during the period of stimulation were not statistically significant. The most marked effect was encountered in the neurone shown in the Fig. 3*D*. The stimulation applied to the distal parts of the severed muscle nerves provoked no changes in fusimotor discharge rate (not shown). Changes would have been expected if metabolic products liberated in muscle tissue during contraction were exerting reflex effects on fusimotor neurones by acting on receptors outside the contracting muscle (Gregory, Kenins & Proske, 1987).

Generality of the late increase in fusimotor discharge rate

While the late increase in discharge rate occurred in all the fusimotor neurones studied it varied considerably in both magnitude and duration among the neurones. In an attempt to assess the changes in overall fusimotor activity level, pooled data are presented in Fig. 4. Mean values of spontaneous discharge rates of individual fusimotor neurones together with the corresponding discharge rates during the period of late increase, as well as histograms of their distribution, are shown in *A* and *B*, respectively. Twenty-nine out of thirty-one fusimotor neurones studied (94%) fired at rest at rates ranging from 3 to 18 impulses/s, while their discharge rate during the period of late increase attained values from 8 to 23 impulses/s (*A*). With respect to the spontaneous discharge range the range of firing rates during the period of late increase was shifted by 5 impulses/s towards the higher frequencies. While for the spontaneous discharges the distribution of units was skewed to the left, 61% of units (nineteen out of thirty-one) firing at rates below 10 impulses/s, for the late increase the distribution was skewed to the right, twenty-four out of thirty-one units

(77%) firing at the rates above 10 impulses/s and almost half of the units above 15 impulses/s (*B*). The discharge rates of fusimotor neurones during the period of late increase normalized with respect to the spontaneous discharge rate (not shown) ranged from 1·11 to 6·0 impulses/s (mean value 1·91) indicating that during this

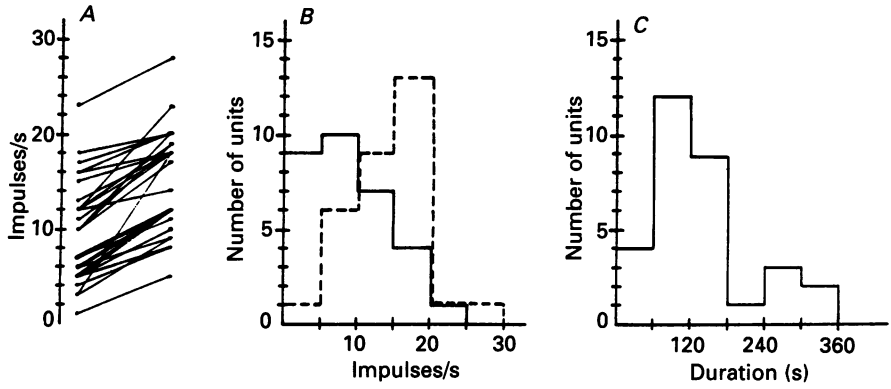

Fig. 4. *A*, mean values of the spontaneous discharge rates (left) and the discharge rates during the period of its late increase (right) for the thirty-one fusimotor neurone studied. *B* and *C*, histograms of distribution of the discharge rates (continuous line, at rest; dashed line, during the period of late increase) and of duration of the late increase respectively.

period the neurones fire at rates on average double that of their spontaneous firing. Distribution of the duration of the late increase in fusimotor discharge rate beyond the end of muscle contraction (Fig. 4*C*) shows that it lasted in the majority of the neurones (68%) from 60 to 180 s, and was longer in an additional six units (19%). Taking into account that the decline of the increased discharge rate towards the resting firing level was gradual, fusimotor activity would remain fairly elevated for at least 120 s after the end of muscle contraction while approximately 50% of the neurones fire at rates higher by 5 impulses/s on average than the resting discharge rate.

DISCUSSION

The results of our experiments show that changes in discharge rate of fusimotor neurones innervating muscle spindles in the contracting muscle and a close synergist do occur during long-lasting, fatiguing muscle contractions. In most cases it was an initial sharp increase, followed by a decrease to or below the resting discharge level, and a late slow increase outlasting the contraction. No consistent changes in fusimotor discharge rate have been provoked by electrical stimulation of either muscle nerves or ventral roots after severing the muscle nerves. When present, the changes were small, maintained at a constant level throughout the stimulation period and devoid of after-effects. It seems justified, therefore, to propose that the changes in fusimotor discharge rate, occurring during muscle contraction, represent mostly their reflex response to changes in afferent inflow from the muscle provoked by its contraction and the developing fatigue.

The early changes in fusimotor discharge rate, bearing obviously no relation to muscle fatigue and corresponding to those already described to be provoked during

muscle twitch contractions (Ellaway & Murphy, 1980; Ellaway *et al.* 1982), will be discussed only briefly. The initial increase in fusimotor discharge rate could safely enough be attributed to afferent discharges from mechanosensitive group III muscle afferents (Ellaway *et al.* 1982). Contribution of early discharges from muscle spindle secondary endings (Hunt, 1954) and/or of those from non-spindle group II afferents (Rymer, Houk & Crago, 1979) cannot be excluded since the discharges from group II muscle afferents have been shown also to exert strong reflex effects on fusimotor neurones (Noth & Thilmann, 1980; Appelberg, Hulliger, Johansson & Sojka, 1983*a*). The subsequent decrease, subsiding with muscle tension fall, might appear due to afferent discharges from Golgi tendon organs (Ellaway & Murphy, 1980), responding initially to a large increase in muscle tension and becoming depressed later on (Hutton & Nelson, 1986). Short-lasting bursts of Renshaw interneurone discharges, often encountered at the onset of muscle contraction provoked by ventral root stimulation (Anastasijević & Vučo, 1978), presumably due to their excitation by afferent inflow from high-threshold muscle afferents (Piercey & Goldfarb, 1974), may contribute initially to the decrease in fusimotor discharge rate (Ellaway, 1971; Noth, 1971). Since the decrease in fusimotor discharge rate was seldom substantial and the net inhibition practically absent on many occasions, the subsequent late increase in fusimotor discharge rate can hardly be ascribed to either a reflex or recurrent disinhibition. Muscle fatigue provoked no changes in either spontaneous discharge rate or sensitivity to muscle stretch of muscle spindle primary and secondary endings unless the fatigue was produced by electrical stimuli above the threshold for fusimotor axons (Nelson & Hutton, 1985). Thus, by exclusion, discharges from group III and IV muscle afferents remain to be considered as a probable cause of the late increase in fusimotor discharge rate.

Responses of mechanosensitive group III and IV muscle afferents to muscle contraction may be delayed in onset, developing gradually and often outlasting the end of contraction (Mense & Stahnke, 1983). Their responsiveness is enhanced by muscle ischaemia (Kaufman *et al.* 1984), fatigue (Hayward, 1990) and metabolic products liberated in muscle tissue during contraction (Rotto *et al.* 1990). Background activity in these fibres also increases during muscle fatigue (Hayward, 1990). Discharges in other group III and IV muscle afferents are provoked by metabolic products liberated in muscle tissue during contraction (Thimm & Baum, 1987; Rotto & Kaufman, 1988). Chemically induced discharges in these afferent fibres, as well as the consecutive increase in fusimotor discharge rate (Jovanović *et al.* 1990), last for several tens of seconds after close arterial injection of an algesic agent or metabolic product. When metabolic changes developing during static exercise have been mimicked by applying chemical substances by long-lasting perfusion of the muscle or its superfusion (Thimm & Baum, 1987) the resulting increase in discharge rate of group III and IV afferents lasted for several minutes. On the grounds of these findings it seems justified to assume that both the proportion of group III and IV muscular afferents discharging, as well as their firing rate, are increased and can be sustained at an elevated level during long-lasting fatiguing muscle contraction. Their discharges, shown to exert strong reflex effects on fusimotor neurones (Ellaway *et al.* 1982; Appelberg *et al.* 1983*b*; Jovanović *et al.* 1990), could elicit the late increase in fusimotor discharge rate developing during

fatiguing muscle contraction and outlasting it. The effects of muscle ischaemia speak also in favour of its origin ultimately related to metabolic products liberated in muscle tissue (Bigland-Ritchie *et al.* 1986).

The additional late increase in discharge rate starting 40–60 s after the end of muscle contraction was encountered in three fusimotor neurones only. Its enhancement by muscle ischaemia (not shown in the figures) speaks against it being a spontaneous oscillation in fusimotor activity occurring by mere coincidence in temporal relation to muscle contraction. Its appearance could be tentatively explained by supposing that the activity provoked in some group III and IV muscle afferents was intermittent, as described to occur in inflamed muscle (Berberich, Hoheisel & Mense, 1988). Though this pattern of firing was never encountered in normal muscle before induction of inflammation (but see Franz & Mense, 1975), it nevertheless may be supposed to appear in muscle fatigue since the responses of these afferents to metabolic products are rather similar to those elicited by algesic agents.

In our experiments we could not differentiate static from dynamic fusimotor neurones. Since the late increase in discharge rate was encountered in all the fusimotor neurones recorded in both decerebrate and, though few, spinal cats, it could be supposed to have appeared in both static and dynamic ones. Variability in magnitude and duration of the increase in discharge rate provided no evidence indicating separate groups of neurones behaving differently. It resembled rather the differences in responsiveness encountered among individual fusimotor neurones to afferent inflow of other origin (Johansson, Sjölander, Sojka & Wadell, 1989).

Functional implications

Muscle contraction elicited by long-lasting electrical stimulation of either muscle nerves or ventral roots should provoke changes in afferent inflow from muscle receptors similar to those occurring during sustained voluntary contraction (Garland *et al.* 1988). It seems therefore justified to assume that late autogenetic excitation of fusimotor neurones by an increased afferent inflow from group III and IV muscle afferents occurs also during sustained voluntary contraction and to consider its possible functional role.

Autogenetic inhibition of skeletomotor neurones by discharges provoked in group III and IV muscle afferents has been shown to provide reflexly a matching between the discharge rates of skeletomotor neurones and changes due to fatigue in contractile properties of the muscle during sustained voluntary contraction (Bigland-Ritchie *et al.* 1986). It has been proposed recently that a decrease in support of skeletomotor activity by autogenetic excitatory impulses mediated via the γ-loop may contribute to the gradual decrease in skeletomotor firing rate during muscle fatigue (Bongiovanni & Hagbarth, 1990). It would appear, according to these authors, due to fatigue of intrafusal muscle fibres or else due to co-inhibition of fusimotor neurones by group III and IV muscle afferent discharges. Our experiments show, however, that an increase rather than a decrease in fusimotor discharge rate can be expected to develop during muscle fatigue. Since the slowing of skeletomotor firing rates during muscle fatigue has a deep functional meaning, the simultaneous increase in fusimotor discharge rate, though in accordance with the described reflex effects of the inflow from small-diameter muscle afferents (Ellaway *et al.* 1982; Appelberg *et al.* 1983*b*; Jovanović *et al.* 1990), might seem paradoxical. Its role,

however, still may be to maintain the autogenetic excitatory inflow to skeletomotor neurones from muscle spindles at an appropriate level so as to optimize, in balance with the autogenetic inhibitory influences, their firing rates. Records of spike discharges from a few Ia afferents from the present experiments (not shown) indicate that fusimotor support through the γ-loop, preserved in these experiments, and its additional increase during muscle contraction, might be sufficient to prevent any larger decrease in afferent activity from muscle spindles.

It has been proposed recently (Llewellyn, Yang & Prochazka, 1990) that the low Hoffmann reflex gain coupled with a high level of fusimotor drive in a demanding motor task may serve to provide supraspinal centres with increased proprioceptive information avoiding simultaneous saturation and instability of skeletomotor neurones. The increased fusimotor activity during muscle fatiguing contraction, as well as its possible after-effects (e.g. Hutton, Smith & Eldred, 1973), may be supposed to play a similar role in muscle fatigue. It could preserve and/or enhance sensitivity of muscle spindle sensory endings providing more information to be conveyed to higher centres on the fatigued muscle. The history-dependent changes in muscle spindle resting discharge and responsiveness, supposed to be involved in both biasing of segmental reflex activity and in kinaesthesia (see Proske, Gregory & Morgan, 1990, for review), might be important for motor control of a fatigued muscle. Changes in sensitivity to stretch of muscle spindle sensory endings have been shown to occur in fatigued muscle (Nelson & Hutton, 1985). In the experiments of Nelson & Hutton, however, ventral roots were severed and electrical stimulation above threshold for fusimotor axons applied to their distal stumps. Further studies are needed therefore to prove whether the increase in fusimotor discharge rate, found in our experiments to develop during muscle fatigue, is sufficient to provoke similar effects. Nevertheless, since an apparent reduction in responsiveness of muscle spindle primary endings occurred when spontaneous fusimotor discharges were prevented from reaching the spindles (Matthews & Rushworth, 1958), an increase in responsiveness might be expected to follow a doubling of spontaneous fusimotor firing rates.

Since the discharges from group III muscle afferents elicit strong reflex effects on fusimotor neurones innervating muscle spindles in other muscles (Appelberg *et al.* 1983*b*), changes in discharge rate during muscle fatigue may not be limited to fusimotor neurones destined for the contracting muscle. The possibility that the increase in fusimotor activity is widespread and contributes to the spreading of activity to other muscles when the muscle performing voluntary contraction is fatigued (Lippold, Redfearn & Vučo, 1960) will be dealt with in a separate paper (M. Lubisavljević, K. Jovanović & R. Anastasijević, in preparation).

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