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THIN-FIBRE RECEPTORS RESPONDING TO MECHANICAL, CHEMICAL, AND THERMAL STIMULATION IN THE SKELETAL MUSCLE OF THE DOG

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SUMMARY

1. Unitary activities of muscular thin fibre afferents, which were not sensitive to muscle stretching, were recorded from the nerve of the medial gastrocnemius muscle of the dog. Responses to mechanical stimulation, intra-arterial injection and local application of chemical solutions, and thermal stimulation of the surface of the muscle were studied. It was observed that polymodal receptors which responded to all types of stimulation existed in the thin fibre afferents of the muscle.

2. The receptive area of these units tested by mechanical stimulation was spot-like and appeared to be located not only on the surface but in the midst of the muscle.

3. The mechanical response varied among these units with respect to the threshold and the pattern of discharges.

4. In these units, NaCl, KCl, and bradykinin consistently evoked responses, with differences in the latencies and discharge patterns, while solutions of histamine, acetylcholine and sodium citrate caused responses less consistently and less effectively. In the stretch receptors, chemical stimulation applied in the same way as tested in the thin fibre afferents produced quite different features in their responses.

5. Heating the receptive area of the muscle surface caused responses in twenty-five out of thirty-six units, which were sensitive both to mechanical and to chemical stimulations. The threshold varied from 38·0 to 48·3 °C, with a mean of 43·1 °C for C fibre units and 41 °C for A-δ fibre units.

6. The responses to heating were consistently obtained in the units responding to the surface application of chemical solutions. However, the above response was never obtained in the units which did not respond to surface chemical stimulation but responded to intra-arterial injection. These results suggest a large population of polymodal receptors in the muscular thin fibre afferents.

INTRODUCTION

The characteristics of the cutaneous unmyelinated (C) fibre receptors have been studied in detail and the classification of receptor types has progressed in this decade (see Burgess & Perl, 1973; Lynn, 1975). The majority of C fibre receptors in the human skin and in the skin of the posterior limb of the monkey were found to be of the polymodal type (Van Hees & Gybels, 1972; Torebjörk & Hallin, 1974; Kumazawa, Boivie & Perl, 1974), while the incidence of these receptors in the posterior limb of the cat was reported comparatively low (Bessou & Perl, 1969). This type of receptor responds to between moderate and strong mechanical stimuli, to acid applied topically, and to the heating of the skin to a noxious level (Bessou & Perl, 1969). Besides this 'polymodality', a low reproducibility of the response to repeated stimulation was another characteristic of this type of receptor, as shown by the 'sensitization' phenomena to repeated heat stimulation (Bessou & Perl, 1969; Perl, Kumazawa, Lynn & Kenins, 1976).

These features of the response suggest that the polymodal receptor is not well developed to send specific and accurate information about the changes of the environment. With a few exceptions (e.g. the carotid baroreceptor), deep tissues are generally considered to be innervated by afferents transmitting qualitatively and quantitatively less accurate information than those from the surface of the body. It seemed possible, therefore, that much of the innervation of deep tissues was by receptors of the polymodal type. However, other investigators have used only restricted types of stimuli. For example, the nature of the thin fibre receptors in the muscle has been investigated using mechanical stimulation (Paintal, 1960) and also recently using chemical stimulation (Mense & Schmidt, 1974; Franz & Mense, 1975; Fock & Mense, 1976; Hiss & Mense, 1976).

In the present experiment, the existence of the polymodal receptor in the muscle was investigated in the dog using heat, mechanical, and chemical stimulations.

A preliminary report has been published elsewhere (Kumazawa & Mizumura, 1976).

METHODS

Dogs were anesthetized with pentobarbitone sodium and artificially ventilated after immobilization with Flaxedil. The blood pressure was monitored and kept above 100 mmHg and the rectal temperature was maintained between 37 and 39 °C. The animal was kept in a prone position fixed with steel pins at the iliac bone, the knee, and the ankle. The skin of the posterior surface of the thigh and leg was cut from the gluteal region to the ankle. The medial gastrocnemius muscle was freed

from surrounding tissues except at the entry zone of the nerve and blood vessels to this muscle. Blood vessels in the posterior limb, except those supplying this muscle, were ligated as completely as possible. A polyethylene cannula was inserted into a small branch of the caudal femoral artery close to the branch to the gastrocnemius muscle and used for injection of chemical solutions. A snare ring was set at the site of bifurcation of the popliteal vessels from the femoral vessels, for transient interruption of the blood flow during injection of solutions. To ascertain the spread of the injected solution, Evans blue dye was injected through this cannula before or after the experiment. The units which had their receptive area beyond the spread of the dye were discarded.

The nerve to the gastrocnemius muscle was cut in the upper thigh and a fine filament of the nerve was dissected on a mirror plate in a paraffin oil pool, until a distinguishable single unit discharge could be isolated by testing the response to electrical stimulation of the nerve in the popliteal fossa. The conduction velocity of the unit was determined by the latency and the distance between the stimulating and recording electrodes measured *in situ* at the end of the experiment. In this experiment, the unit which was at first secured to be mechanoresponsive was studied further as an afferent unit. Exceptionally, in a few units on which only chemical responses were investigated, the existence of on-going discharges was regarded as a sign of an afferent unit.

The receptive area of the unit was searched by stroking and pressing the muscle surface with a camel's hair brush and a glass rod or by pinching or by squeezing with a forceps. For quantitative testing, a strain gauge-mechanostimulator with a tip diameter of 0·75 mm was used. The effect of stretching the muscle was tested by pulling the Achilles tendon with a weight of 60 g.

The chemical substances used in this experiment were bradykinin, acetylcholine, and histamine, dissolved in Ringer–Locke solution; and NaCl and sodium citrate dissolved in distilled water. KCl solution was made by mixing the isotonic KCl solution and Ringer–Locke solution. The concentrations of the solution were: bradykinin 1 μg/ml.; NaCl, 3·6 or 4·5 g % (w/v); KCl, 50 or 60 mM; acetylcholine, 100 μg/ml.; histamine, 10 μg/ml.; sodium citrate, 1·5 g % (w/v). In the majority of the cases, 5 ml. solution was injected in about 1 min through the arterial cannula. For a 120 sec period from 30 sec before to 30 sec after the injection, the femoral vessels were transiently ligated with a snare ring. Arresting the blood flow to the muscle alone did not cause any changes in the discharge of units for at least 2 min. Local effects of these solutions were tested by application of a cotton ball about 2 mm in diameter on the receptive area of the muscle surface for 1 min. The concentrations of the solutions were: bradykinin, 10 μg/ml.; NaCl, 4·5 or 9 g % (w/v); KCl, 60 or 100 mM; acetylcholine, 1 mg/ml.; histamine, 100 μg/ml. With either method of application, the Ringer–Locke solution alone did not cause changes of discharge except in two cases. In these two instances the slight increase of discharge was coincident with the increase of the pressure of injection.

Radiant heat was applied to the receptive area through an aperture 5 mm in diameter in a heat-reflection plate, which protected the surrounding part of the receptive area from heating. The surface temperature of the heat-exposed part of the muscle was measured with a thermistor. The muscle was heated to a surface temperature not exceeding 50 °C. In several units cooling the muscle surface was carried out by a thermode 5 mm in diameter which was cooled with a rate of about 10 °C per minute to between 7 and 22 °C by ether evaporation.

Impulses of units and signals from the stimulators were recorded on FM magnetic tape and analysed with the help of a computer.

RESULTS

Fifty-two C fibre units with conduction velocities between 0·4 and 2·3 m/sec (mean 1·2 m/sec) and 12 A-δ units between 2·7 and 20 m/sec (mean 9·9 m/sec) were studied. Since some stretch receptors are known to be connected with A-δ fibre (Paintal, 1960), the muscle was stretched with a force of at least 60 g on the Achilles tendon in order to differentiate the stretch receptors. Some units showed a transient and slight response on stretching the muscle and a larger response on localized pressing of the receptive area. Such units were not classified as stretch receptors.

Receptive areas of the unit were investigated by mechanical stimulation of the muscle surface. The receptive area was usually a few mm in diameter and one to four sensitive spots were found within the area. A few units had two receptive areas separated from each other by 1–2 cm. Receptive areas were found all over the muscle, but it seemed that they were more frequently found in the head, the tail, and the edge of the muscle. Though not in all cases, the mechanical threshold was measured from both sides of the muscle surface, some units had a definitely lower threshold on the outer or on the inner surface, suggesting the location of the receptive area close to that side of the surface, and some units did not, suggesting that the receptive site was located deep in the midst of the muscle.

An on-going discharge at a low rate, which seldom exceeded 1 impulses/sec, was observed in twenty-three out of twenty-six C fibre units and five out of seven A-δ fibre units isolated at the start of each experiment. After testing a set of stimulations on the muscle, an on-going discharges were found in all units.

Mechanical responses. Mechanically these units were activated by a light stroking with a glass rod or by pinching and squeezing with a forceps, while stroking with a camel's hair brush never evoked discharges. No clear systematic difference was found in the mechanical response between C fibre and A-δ fibre units. Many units were found to respond with a considerably long sustained discharge as well as a dynamic discharge. One typical example of this type of response is shown in Fig. 1. The static and dynamic discharge rates roughly paralleled the increase of pressure, within the range applied.

Some units responded to pressing only at the start of stimulation and also often at the end of stimulation. Though not systematically tested, units with high mechanical threshold tended to show a rapidly adapting discharge and less reproducible response to repeated stimulations and vice versa. Whether these differences in mechanical response among units depend on the nature of the receptor itself or the location of the receptor site remains unclear.

Chemical responses. Responses of C and A-δ fibre units to arterial injection and/or surface application of chemical solutions were obtained in thirty-three out of thirty-seven units tested with NaCl, thirty-three out of thirty-eight units with bradykinin, twenty-two out of twenty-three units with KCl, six out of twelve units with sodium citrate, thirteen out of twenty-six units with acetylcholine, and nine out of twenty units with histamine.

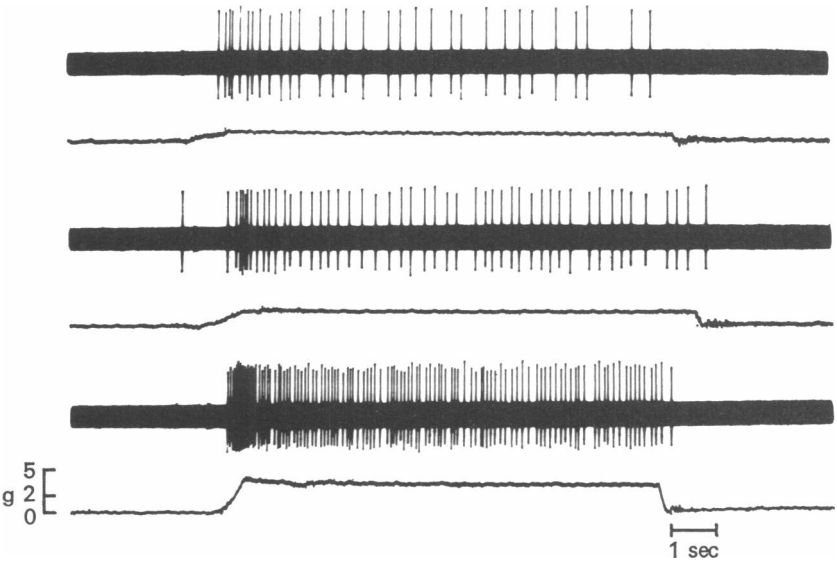

Fig. 1. Mechanical responses of an A-δ polymodal type unit. The receptive spot was pressed with 1, 1·6, and 3·2 g (from above downward), shown by the lower trace in each pair. Note clear dynamic and static responses. This unit with conduction velocity of 7·5 m/sec responded to heating and also to both arterial and local application of solutions of NaCl, KCl, and bradykinin.

In most cases more than two kinds of solutions were tested on the same unit, with at least 5 min intervals between the tests. Fig. 2 shows how many units among those which responded to a certain chemical solution could also exhibit a response to another chemical solution. Among the six chemicals employed, NaCl, bradykinin, and KCl elicited a response in almost all units which also responded to the other five chemicals; while sodium citrate, acetylcholine, and histamine caused responses in only about half of the units responsive to NaCl, bradykinin, and KCl. These observations reveal that solutions of NaCl, bradykinin, and KCl used in this experiment are consistent stimulants for muscular C and A-δ fibre afferents.

Examples of the discharge pattern of an A-δ and a C fibre unit are shown in Fig. 3. A slowly-rising and slowly-decaying discharge was

characteristic of the response to bradykinin. In the response to NaCl, a rather abrupt increase of discharge was followed by a slowly decaying phase with or without intermittent interruptions of impulses. The response to KCl was more variable, having multiple peaks in discharge rate, in the concentrations used in this experiment.

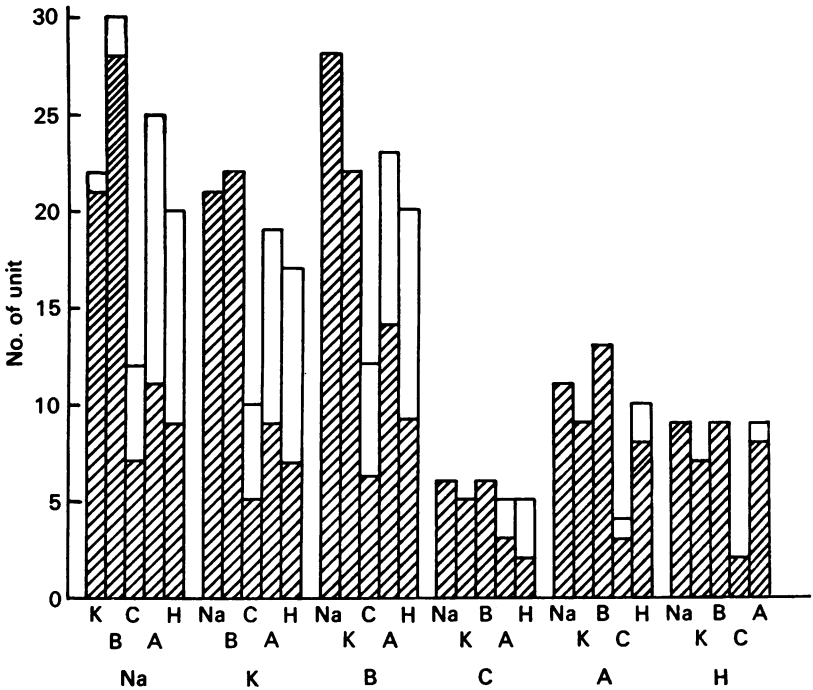

Fig. 2. Cross responsibility to chemicals. Units responding to a certain chemical (shown at the bottom of the Figure) are grouped in sets. In each group, cross-hatched columns show the number of units which further responded to another chemical (shown at the base of each column), and white columns show a lack of response. Na, NaCl; K, KCl; B, bradykinin; C, sodium citrate; A, acetylcholine; H, histamine. Cases using two ways of application of solution with all concentrations described in the method are included.

Averages of the latency, the mean discharge rate, and the peak discharge rate of the response of C and A-δ fibre units to NaCl, bradykinin, and KCl are summarized in Table 1. The most potent stimulant among the three was 4·5 g % (w/v) solution of NaCl. It produced responses with the shortest latency and the largest mean and peak discharge rate. Comparing the responses to bradykinin and KCl, the latency was shorter with KCl, and the discharge rate did not differ much, though with bradykinin the discharge rate had a slightly higher mean and a slightly lower peak.

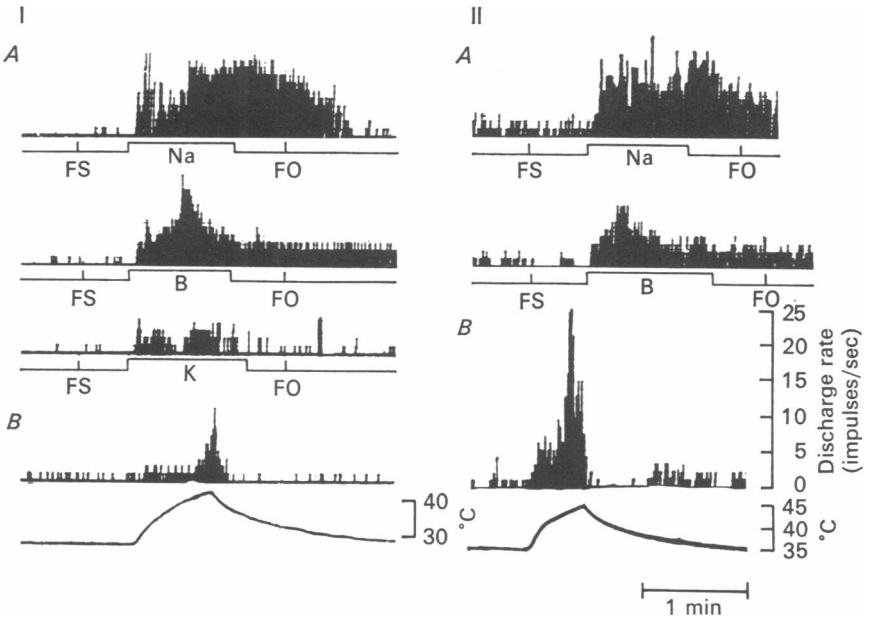

Fig. 3. Discharge pattern of an A-δ and a C fibre unit to chemical stimulation and heat stimulation. I, an A-δ fibre unit with conduction velocity of 12 m/sec. II, a C fibre unit with conduction velocity of 1·1 m/sec. *A* and *B*, chemical and heat responses respectively. Upper traces of each pair show the number of impulses/sec, the scale being indicated at the right of II*B*. This A-δ unit also responded to acetylcholine and histamine and this C fibre unit responded to histamine and KCl (not shown here). Lower traces are stimulus marks, upward deflexion in *A* shows the period of intra-arterial injection of 4·5 g % (w/v) NaCl (shown as Na), bradykinin 1 μg/ml. (B), and 60 mM-KCl (K). Arresting and reestablishing of the blood flow (see text) were indicated as FS and FO respectively. Time scale of 1 min is shown at the bottom right.

TABLE 1. Latency, mean and peak discharge rate of the responses to the arterial injection of the three chemicals in A-δ and C fibre units. Mean discharge rate: discharge rates during 90 sec period from the onset of injection were averaged. Number of units is shown in parentheses

Chemicals	Fibre type	Latency (sec)	Mean discharge rate (impulses/sec)	Peak discharge rate (impulses/sec)
NaCl (4·5 g % (w/v))	Aδ	4·6 ± 0·7 (S.E.) (5)	7·1 ± 3·5 (5)	15·2 ± 2·2 (5)
	C	8·4 ± 2·6 (17)	6·9 ± 1·3 (16)	15·2 ± 2·2 (16)
Bradykinin (1 μg/ml.)	Aδ	17·3 ± 7·0 (7)	3·7 ± 1·7 (5)	8·0 ± 1·3 (7)
	C	28·3 ± 1·0 (20)	1·6 ± 0·2 (20)	5·9 ± 0·7 (19)
KCl (60 mM)	Aδ	4, 16	0·5, 1·4	4, 6
	C	13·4 ± 4·2 (10)	1·3 ± 0·6 (9)	7·5 ± 1·6 (10)

Though the small number of A-δ fibre units tested in this way may not allow a conclusive description, the chemical response of A-δ fibre units did not differ from those of C fibre units, except that the A-δ units had a shorter latency.

In several units, the same dose of bradykinin was applied repeatedly several times with a 10 min interval. The discharge rate decreased gradually during successive trials, being accompanied by an increase of the latency. But in some cases an abrupt recovery of the response was observed without any detectable reason.

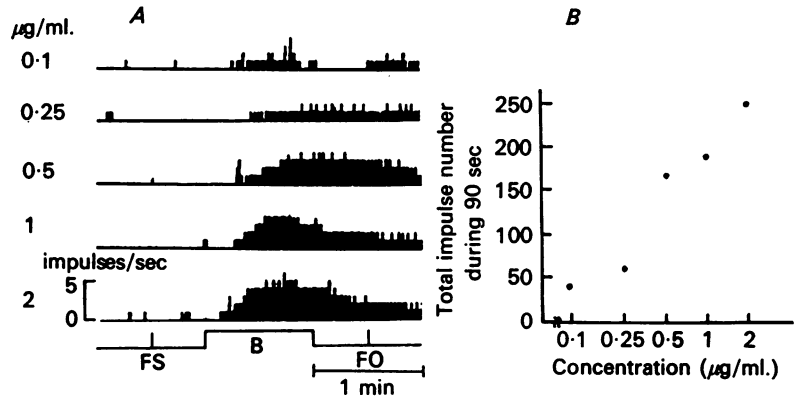

Fig. 4. Responses of a polymodal C fibre unit in the muscle to arterial injection of the bradykinin solutions of different concentrations. *A*, response pattern. The concentrations used were shown at the left side of the Figure. *B*, ordinate is the total number of impulses counted in the 90 sec period between the onset of injection and the reestablishment of the blood flow (marked as FO in the bottom line of *A*). Abscissa is the concentration of bradykinin in logarithmic scale.

Although the reproducibility of the response was thus not entirely consistent, the effect of different doses of bradykinin was tested on one unit. As shown in Fig. 4, the response augmented and the latency shortened with increase of the concentration of the solution. The number of the impulses during the 90 sec period after the start of the arterial injection of each solution shows an approximately linear relation with a logarithms of the concentration of solutions.

Thermal responses. The surface of the mechanically responsive area was heated from an adapting temperature of 28–35 °C to a temperature not exceeding 50 °C with a rate of about 10 °C increase per minute. A clear increase of discharge rate was observed in nineteen out of thirty C fibre units and all of six A-δ fibre units. The pattern of increase in discharge rate was not regular, but a more or less progressive increase of the discharge was seen as the temperature was raised (Figs. 3 and 5). In a few

units, before the appearance of a progressive increase of discharges, discharges at a low rate were seen even at the onset of the temperature increase (Fig. 3I*B*), these phenomena were often noted after repeated heating or with a rapid increase in temperature starting from a low adapting temperature.

Apart from the case of a rapid temperature increase and repeatedly tested cases, the temperature which evoked the increase of discharge ranged between 38·0 and 48·3 °C, and the mean value was 43·1 °C for C fibre units ($n = 11$), and 41 °C for A-δ fibre units ($n = 4$). The peak discharge rate observed in these cases were $14{\cdot}6 \pm 6{\cdot}5$ impulses/sec for C fibre units and $17{\cdot}0 \pm 3{\cdot}4$ for A-δ units. On cessation of heating, an after-discharge was frequently observed, especially on repeatedly heated cases, but a sudden suppression of discharge rate to less than the rate of the on-going activity was also observed.

When heat stimuli were applied repeatedly, the response of the unit changed variably. In many cases, augmentation of on-going activities and lowering of the threshold temperature and increase of discharge during heating period were observed, but sometimes decrease of discharge was also seen. These were similar to the findings observed in the cutaneous polymodal receptors (Witt & Griffin, 1962; Bessou & Perl, 1969; Beck, Handwerker & Zimmermann, 1974; Kumazawa *et al.*, 1974; Beitel & Dubner, 1976; Croze, Duclaux & Kenshalo, 1976).

One example of augmentation of the response in the second application of heat is shown in Fig. 5. In this case, existence of on-going activities made it difficult to detect the change of threshold temperature; the total number of impulses during heating period increased substantially, from 128 in the first trial to 215 in the second.

The effect of cooling was tested on seven units. In one unit, which did not respond to heating, a slight and irregular increase of discharge was seen in the cooling period from 31·5 to 8 °C, but quantitatively this increase of discharge did not parallel the temperature of the muscle surface. Among six heat-responding units, three units were not affected by cooling up to 22, 13, and 7 °C; and in three units on-going discharges were suppressed below 24 or 22 °C (in two units).

All of the heat-tested units were responsive to mechanical stimulation and either to local or arterial administration of chemical solutions. All units which responded to local application of chemicals were responsive to heating, while all units which did not respond to local chemical stimulation did not respond to heating either.

Chemical responses of the stretch receptors. In twenty-one stretch receptors which showed an increase of discharge on stretching the muscle and a transient suppression of on-going discharges on cessation of stretching,

the effects of arterial injection of chemical solutions were tested in the same way as used for thin fibre units. As shown in Table 2, the incidence of the response of these receptors to various chemical stimuli exhibited a striking contrast to that obtained in the above mentioned thin fibre receptors tested exactly in the same way.

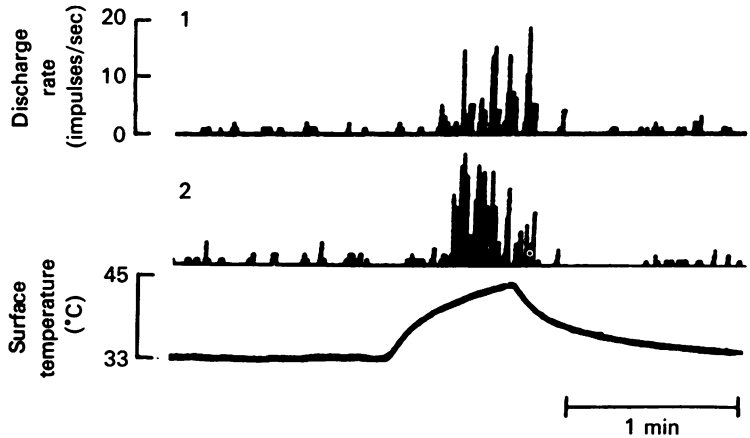

Fig. 5. Augmentation of the response to successive applications of heat. These responses were obtained by heating the same spot with the same pattern of temperature rise. *1* and *2*, the first and second trial of heating with a 20 min interval between them. This unit is the same unit shown in Fig. 1, and responded to both arterial injection and surface application of solutions of NaCl and KCl.

TABLE 2. Response of stretch receptors to arterial injection of various chemical solutions. Increase and decrease of the discharge rate during injection period are shown as 'increased' and 'decreased' respectively. *, unclear response

Chemicals	Increased	Decreased	No response	Total
NaCl (3·6, 4·5 g % (w/v))	4	6	11	21
Bradykinin (1 μg/ml.)	0	0	12	12
KCl (50, 60 mM)	7	1	2	10
Sodium citrate (1·5 g % (w/v))	8	0	2	10
Acetylcholine (100 μg/ml.)	0	0	7	7
Histamine (10 μg/ml.)	1*	0	9	10

A vigorous increase of discharges was consistently observed with sodium citrate, and in some units the increase of discharge more than 100 impulses/sec lasted for more than 10 min, and with intervals of 10–30 min it spontaneously reappeared several times. In the majority of the units, KCl caused responses, though far less when compared with that of sodium citrate, being followed frequently by a few minutes complete suppression of discharges. Among twenty-one units tested with NaCl,

increase of the discharges was obtained in only four units, and in six units a slight decrease of the on-going discharge was seen. In no instance, did bradykinin as well as acetylcholine and histamine act as a consistent

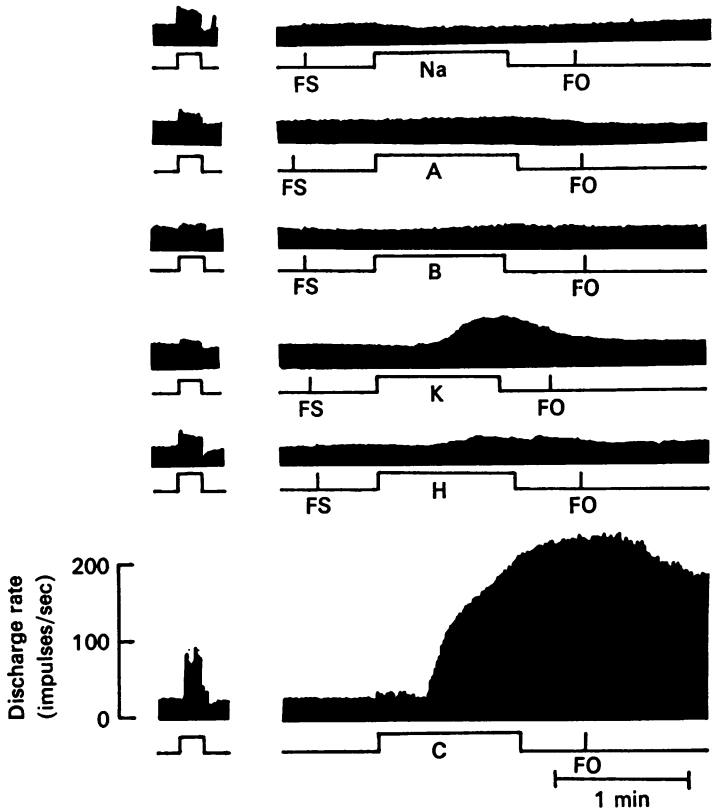

Fig. 6. Response of a stretch receptor in the muscle to various chemical solutions. At the left side, responses to stretch tested before injection were shown, stimulation period being indicated by upward deflexion of the lower trace. Note that NaCl and bradykinin caused no clear response, while sodium citrate caused a large increase of discharge rate. Na, NaCl, 4·5 g% (w/v); A, acetylcholine, 100 μg/ml.; B, bradykinin, 1 μg/ml.; K, KCl, 60 mM; H, histamine, 10 μg/ml.; C, sodium citrate, 1·5 g% (w/v).

stimulant for these receptors. An example of responses of the stretch receptor to various chemical stimuli is shown in Fig. 6. It may be pointed out that features of the chemical responses obtained in the above described thin fibre receptors definitely differ from those of the stretch receptors.

DISCUSSION

The present experiment demonstrated that thin fibre receptors which responded to all of the localized mechanical stimulation, chemical stimulation and thermal stimulation, were present in the muscle.

The intensities of mechanical stimulation required for evoking discharges of these thin fibre receptors varied greatly from simple contact to penetration of the muscle surface. If applied to the skin of a human being, the former is far from nociceptive, while the latter caused definite pain sensation. Though not systematically proven, there is an indication that the lower the threshold, the slower the adaptation rate. These differences in the threshold and the adaptation rate might reflect the location of the receptor site of the units.

With regard to the response to chemical stimuli, a remarkable difference could be detected between the stretch receptors and the polymodal receptors. In the stretch receptors, sodium citrate caused a consistent and vigorous increase of the discharge, while for the polymodal receptors it is not a consistent stimulant. On the other hand bradykinin and NaCl, which are the most potent excitants for the polymodal receptors, exerted slight or no effect on the stretch receptors.

Recently responses of muscular thin fibre receptors to chemical stimulations have been studied intensively by Mense and coworkers (Mense & Schmidt, 1974; Franz & Mense, 1975; Fock & Mense, 1976; Hiss & Mense, 1976). They considered the effect of KCl was non-specific (Fock & Mense, 1976). In our results KCl caused responses both in the stretch receptors and in the thin fibre receptors and thus is considered to be non-specific in this sense. The discharge patterns of the response to bradykinin observed in the present experiments are similar to the results observed by the group of Mense. A little difference found in the latency and in the least effective dose is probably due to the different method of arterial injection used by the two groups. A great difference between our results and theirs is the incidence of the response to bradykinin: about half of the units they tested gave a response (Franz & Mense, 1975), while almost all units responded in our results. According to their assumptions, the incidence of the KCl-responding units showed the accessibility of the afferent units via the blood stream, and this figure amounts to 42% (Fock & Mense, 1976). This might show that almost all C fibre units in their experiment would be excited by bradykinin, if access were secure.

A gradually decreased response to repeated application of bradykinin was observed in several C fibre units in the present experiment. This finding is similar to tachyphylactic behaviour to bradykinin observed on the cutaneous unmyelinated nociceptors and slowly adapting mechano-

receptors by Beck & Handwerker (1974). In muscular afferents, no tachyphylactic phenomenon had been reported in the discharges of unmyelinated fibre receptors (Hiss & Mense, 1976) and also in the vocalization response of puppies (Taira, Nakayama & Hashimoto, 1968). This difference in tachyphylaxis between ours and the two other groups might be due to the different method of administration of bradykinin solution, but this remains to be tested.

The responsiveness to heat is a characteristic and consistent feature of the polymodal receptor of the skin (Bessou & Perl, 1969). In the present experiment the response to heat was tested on thirty-six units which were both mechano- and chemoresponsive. Only 25 units were found to respond to heat. However, in the heat unresponsive units, surface application of chemical stimuli did not cause any clear response, while a consistent response to the same stimuli was observed in the heat responsive units. This clear parallelism between the responsiveness to heat and to surface chemical stimuli may suggest the possibility that the receptive sites of these heat unresponsive units might be located deep in the muscle, and therefore, if the local temperature of their receptor site were raised to a comparable level to that of the heat responsive units, these unresponsive units might be excited.

The observation that in some units, the mechanical thresholds measured from both sides of the muscle surface did not differ much, also suggests that the receptor site of some units was located deep in the midst of the muscle. Morphologically free-ending terminals have been observed associated with all types of tissues in the muscle except the capillary net work and not restricted to any specific region of the muscle (Stacey, 1969). These free-ending terminals are presumed to be the site of polymodal receptors.

The analysis described above might suggest that the majority of the thin fibre afferents reported here are polymodal receptors, and possibly at least a certain part of those reported previously (Paintal, 1960; Iggo, 1961; Mense & Schmidt, 1974; Franz & Mense, 1975; Fock & Mense, 1976; Hiss & Mense, 1976) might be included in polymodal receptors.

Though in the present experiment, it was suggested a large population of the muscular thin fibre afferent might be polymodal type, this did not exclude the existence of other types of receptors in the muscular thin fibre afferents. Hertel, Howaldt & Mense (1976) reported the definite existence of the thermosensitive receptors in the muscle of the cat, though in a small population. We failed to find units which were identified to be true thermoreceptor units in the present experiment. This might be possibly due to two reasons: one is the majority of the units we studied were secured to be mechanoresponsive, and the other is thermal

stimulation we used was limited to a small receptive area of the muscle surface.

The same kind of response to mechanical, chemical, and thermal stimulation of the surface of the testis has also been observed in the A-δ fibre receptors of the superior spermatic nerve of the dog and also in thin fibre afferents from the medial gastocnemius muscle of the monkey (unpublished, in this laboratory). This might lead to an assumption that there are receptors having similar properties among different animal species and also in the skin and deep tissues. Therefore, the physiological role of this type of receptor should be of great importance. Considering the nature of effective stimuli which cause responses in these receptors, it is quite probable that these polymodal receptors take part in transmitting nociceptive information from the muscle. Nociceptive functions of the thin fibre afferents from the muscle were suggested as a mechanononciceptor by Paintal and as a chemonociceptor by the group of Mense.

It has long been known that hyperpnea during exercise is partly caused by neural information from the muscle (Dejours, 1963; Kao, 1963), and several results suggest that thin fibre afferents from the muscle contribute to this phenomenon (Senapati, 1966; Hodgson & Matthews, 1968; McCloskey & Mitchell, 1972). However, it remains unclear whether any single change in the chemical, mechanical, and thermal environment occurring during muscle exercise could work as a stimulant causing such reflex hyperpnea.

With the same chemical stimulation of the muscle as used in this experiment, we observed reflex augmentation of ventilation of the dog. This response was similar to the response observed in C fibre unit discharge with respect to the latency and relative magnitude of the response to each chemical (Mizumura & Kumazawa, 1976).

A large population of polymodal receptors in the thin fibre afferents of the muscle; the receptors' ability to respond to thermal, mechanical and chemical stimuli; and the relative similarities of chemical response between unit discharge of the polymodal receptors and reflex hyperpnoea, suggest that polymodal receptors might participate in reflex modulation of the autonomic phenomena, i.e. exercise hyperpnea. For full clarification, however, further investigations are needed.

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