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THE HEAT SENSITIZATION OF POLYMODAL NOCICEPTORS IN THE RABBIT AND ITS INDEPENDENCE OF THE LOCAL BLOOD FLOW

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SUMMARY

1. The firing frequency of polymodal nociceptor units with C-fibre axons was found to increase exponentially with skin temperature as the latter was raised linearly at 1 °C/sec. Q_{10} values ranged from 3·6 to 120.

2. Following skin heating to 54–64 °C , average firing thresholds fell substantially. This sensitization was accompanied by a 46 % increase in interval by interval variability of firing during heat stimulation.

3. Stopping the blood flow to the saphenous area by ligaturing the femoral or saphenous arteries had no significant effect on heat thresholds, on firing patterns during heat stimulation or on variability of firing in sensitized units. There were no changes in continuous, background firing during periods of arterial ligation.

4. It is concluded that if the maintenance of nociceptor heat sensitization depends on the continuous local production of chemical substances, these substances must either be rapidly broken down locally or able to move only slowly into blood vessels.

INTRODUCTION

Nociceptors in the skin become more sensitive after injury to their receptive fields, a phenomenon that has been termed 'sensitization' by Bessou & Perl (1969). Nociceptor units recorded from skin nerves in a number of different species including frog (Echlin & Propper, 1937), rabbit (Perl, Kumazawa, Lynn & Kenins, 1976; Fitzgerald & Lynn, 1977), cat (Witt, 1962; Bessou & Perl, 1969) and monkey (Kumazawa & Perl, 1977) show sensitization. It has been suggested by Lynn (1977) that nociceptor sensitization is an important factor in producing hyperalgesia.

In 1933 Lewis & Hess put forward the idea that cutaneous hyperalgesia in injured regions was due to the local release of a hypothetical 'pain substance' that increased the sensitivity of pain nerve endings. Evidence for their view was that pain in injured areas increased if the blood flow was temporarily stopped, a fact attributed by Lewis to accumulation of the pain substance which under normal circumstances would be continually removed by the local blood flow.

This paper is concerned with experiments to see whether local blood flow has any influence on the heat sensitization of nociceptor units in the skin of rabbits. The responses to noxious heating of one class of nociceptors, the polymodal nociceptors with unmyalinated axons, have been quantitatively examined before, during and after stopping the local blood flow. In order to provide a secure basis for examining responses during periods of reduced blood flow, a detailed preliminary analysis was made of responses to one restricted pattern of skin heating where the skin temperature was raised at a steady rate. The same pattern of heating was used throughout the investigation reported here.

A preliminary account of these experiments has been published (Lynn, 1978).

METHODS

Experiments were carried out on rabbits anaesthetized with 25% urethane in 0·9% saline administered via the marginal ear vein. The initial dose was 1·6–1·8 g/kg and this was supplemented as required to maintain areflexia. A tracheal cannula was introduced and rectal temperature was monitored using a thermistor and was maintained between 36·5 and 38 °C by heating the table on which the animal lay.

Most unit recordings were made from the saphenous nerve although in a few experiments the sural nerve was used. There were no obvious differences between units from the two nerves. Fine filaments were dissected from the main nerve trunk after slitting the outer fibrous sheath. Filaments were placed across a platinum wire recording electrode and a second platinum wire was placed on the filament or on the nerve close to the point where the filament had been dissected free. Nerve action potentials were amplified with a conventional high gain AC amplifier, filtered to maximize the signal to noise ratio (usual bandpass 150–1500 Hz) and displayed on a storage oscilloscope. Nerve activity was also recorded on magnetic tape and on an ultra-violet chart recorder.

The whole nerve was mounted on a second pair of platinum wires positioned approximately 2 cm distal to the recording location and electric shocks (0·5 msec duration rectangular pulses; up to 10 mA current) could be applied to the nerve through these electrodes. This enabled all the fibres in the nerve and therefore all the fibres in a dissected filament to be excited. Only filaments containing small numbers of conducting C-fibres were used. Where more than one C-fibre spike was present recordings were only made from units with distinctive spike shape or amplitude. Conduction velocities of identified units were estimated from the delay at the recording site after a twice threshold shock to the nerve and the conduction distance.

In most experiments the femoral artery was cleared at the inguinal ligament and a loose nylon suture (0 size) was passed around it. The blood flow could then be stopped by applying a force of 0·5–1 N to the nylon loop. In a few experiments this procedure was applied to the saphenous artery in mid-thigh instead of the whole femoral artery in the groin. To check that these procedures did effectively stop blood flow to the skin in the saphenous region, two experiments were carried out in which blood flow was estimated by the xenon clearance method described by Lassen, Lindbjerg & Munck (1964). Small injections (20–100 μl.) of saline containing ^{133}Xe were made into the skin. A crystal scintillation detector was positioned over the injection region and a few mm from the skin surface as shown in Fig. 5*A*. The rate of fall in activity at the injection site was measured before and after pulling on the ligatures around the femoral or saphenous artery in the same way as was done in unit recording experiments.

Natural stimulation of the skin was carried out in a number of ways. Mechanical sensitivity was tested using a set of calibrated von Frey hairs and these were also used to map receptive fields. Cold sensitivity was tested by lightly pressing on the receptive field with a cold (0–15 °C) metal ball. Responses to this stimulus were compared with responses to pressure when the metal ball was at 30–35 °C. Heat stimuli were delivered using a slight modification of the radiant heat method described previously (Fitzgerald & Lynn, 1977). The stimulator consisted of a pre-focused projector bulb that illuminated and heated a circular area of skin approximately 6 mm in diameter. In the centre of the illuminated area a fine polished thermocouple was placed so that it pressed gently on the skin. The modified stimulator used for this study had a Chromel-Constantan thermocouple made from 0·2 mm wires. The thermocouple was stiffer than the copper-Constanton couple used previously and was not attached to an additional spring, but was self-supporting.

Feed-back control of the heat lamp from the skin thermocouple allowed the skin temperature to be raised in a controlled manner and throughout this study linearly rising temperature ramps have been used. A typical stimulus profile is shown in trace B, Fig. 1. Heat ramps were always preceded by a 15–30 s period where the skin temperature was held at 35 or 40 °C. Rates of increase of temperature were 0·8–1·05 °C/s except in a special series of experiments where rates of heating were varied from 0·2 to 2 °C/s. Peak temperatures ranged from 50 to 65 °C depending on the sensitivity of the unit under examination.

The use of ramp stimuli has several advantages. First it means that even when the peak temperatures reached with different stimuli varies, it is still possible to make direct comparisons between the early parts of responses. Secondly, it means that firing frequency at any point during a stimulus can be related to skin temperature without the problems of fluctuating rates of heating that occur with staircase stimulus profiles. Using the data from a single ramp, instantaneous firing frequency has been plotted against average temperature between successive pairs of spikes as shown in Fig. 1 *C*. Such an analysis has allowed more information to be obtained from each stimulus presentation. Further, it has also been possible to compare the data obtained under different conditions using standard regression and convariance methods (Snedecor, 1967, chaps. 6 and 14).

RESULTS

General characteristics of C-fibre units in the rabbit saphenous nerve

The data was obtained from a sample of forty-four C-fibres isolated successively from nine saphenous nerve preparations and one sural nerve preparation. Thirty-two of these units were sensitive to skin heating above 40 °C and all of these were also sensitive to moderate mechanical pressure but were insensitive to innoxious cooling. They were therefore classified as polymodal nociceptors (Bessou & Perl, 1969). Of the remaining twelve units, one was extremely sensitive to cooling and was classified as a specific cold unit, five were very sensitive to mechanical stimulation but were insensitive to heating and were classified as C-mechanoreceptors, 2 were very insensitive to all forms of stimulation and four were not adequately characterized.

Conduction delays were determined for thirty-nine units and calculated conduction velocities ranted from 0·72 to 1·13 m/sec, the average being 0·91 m/sec. The conduction velocities of polymodal units were spread throughout this range and averaged 0·90 m/sec. All units, with the possible exception of the two insensitive ones, had small receptive fields with just one central zone less than 1 mm across where sensitivity was highest. Receptive fields were spread wisely; most lay on the medial or anterior surface of the leg from the level about 1 cm above the knee down to a point about 2 cm below the ankle. Minimum pressure thresholds, assessed using von Frey hairs, were determined for all thirty-two polymodal units and averaged 3·1 mN; twenty-nine (91 %) had thresholds between 0·9 and 9 mN and three had thresholds of 0·3–0·8 mN. For comparison, the thresholds of the five C-mechanoreceptors ranged from less than 0·04 to 0·07 mN and the minimum thresholds of the two insensitive units were greater than 30 mN.

Responses of polymodal units to the first heat stimulus

Responses of polymodal units are known to depend markedly on the past history of the skin area in which their receptive field is situated. Care was therefore taken to avoid injuring the receptive field before applying the first heat stimulus. Further, the hair was only clipped short (to about 1 mm length) and depilatories were not used since other experiments had shown that depilatories often excite and sensitize polymodal units (M. Fitzgerald & B. Lynn, unpublished data).

Firing thresholds when the skin temperature was raised steadily at about 1°/sec were determined for twenty-six units. They varied from 41·1 to 62·8 °C (mean 52·5 ± 5·8 °C (S.D.)). This is a very considerable range, but there did not appear to be any qualitative difference between units with low and high thresholds and the frequency distribution of heat thresholds was unimodal.

A more detailed examination of heat response has been made by comparing the intervals between successive spikes (usually expressed as their reciprocal the instantaneous firing frequency) at different levels of skin temperature during each linearly rising ramp stimulus. A typical stimulus and response are shown in traces *A* and *B* of Fig. 1. Once the threshold has been exceeded, firing accelerates during the ramp from approximately 0·25 $\sec^{-1}$ to a peak rate of 8 $\sec^{-1}$ and stops shortly after the heating is turned off. Fig. 1*C* shows a plot of log instantaneous frequency against skin temperature. For this unit there is a linear relation between log frequency and temperature, i.e. frequency of firing increases exponentially with temperature. The responses of another thirteen units that fired at least nine spikes on the first heat trial have also been analysed in this way and all could be fitted well to an exponential function. Another example is shown in Fig. 3*B*. Correlation coefficients between log frequency and skin temperature ranged from 0·59 to 0·99 and eight out of fourteen were above 0·90.

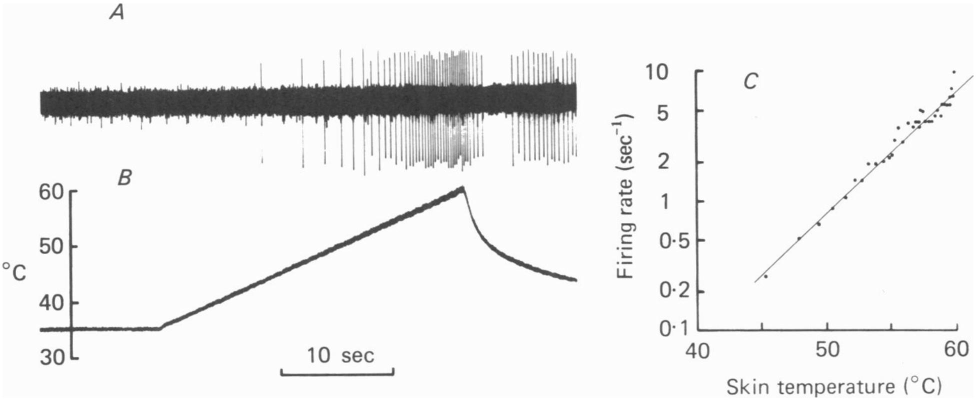

Fig. 1. Response of polymodal nociceptor unit to the first heat ramp. *A*, action potentials recorded from saphenous nerve filament. *B*, skin temperature pattern during heat stimulus. *C*, plot of instantaneous firing frequency (reciprocal inter-spike intervals for each successive pair of spikes) against average temperature during the interval. Note logarithmic frequency scale. Line through points is best-fitting straight line for the regression of log frequency on skin temperature.

For polymodal units it is thus possible to describe heat responses to temperature ramps of limited extent by four parameters: the threshold temperature, two parameters for the exponential function (its slope and elevation) and a paremeter that measures the scatter of the points about the best fitting exponential function. The last three parameters were determined by calculating the regression of log frequency upon skin temperature. The elevation of the function was expressed indirectly as the temperature required to produce average firing of 1 $\sec^{-1}$, T_1. The slope, *b*, was measured in decades/°C but was then converted to the more familiar Q_{10} ($= 10^{10b}$), the increase in firing rate for a 10 °C rise in skin temperature. The scatter about the straight line was expressed as the sample standard deviation from the best fitting regression line.

For the fourteen responses to the first heat stimulus that were analysed in this way there was a considerable range of parameters. T_1 values varied from 40·7 to 63.3 °C, the average being 53·8 °C ($\pm$ 6·06 °C, S.D.). There was a close correlation between firing thresholds and T_1 values, T_1 being on average 2·6 °C higher than the threshold. Exponents varied from 0·056 to 0·208 decades/° C, i.e. Q_{10} values ranged from 3·6 to 120. Twelve out of fourteen units had Q_{10} values between 3·6 and 17. There was no significant correlation between the values of Q_{10} and T_1 for individual units. Sample standard deviations about the regression line varied from 0·04 to 0·23 log sec^{-1} with an average of 0·123 log sec^{-1}. This within trial variability should be related to the fact that units increased in firing frequency about tenfold, i.e. by one log unit, during a typical ramp stimulus.

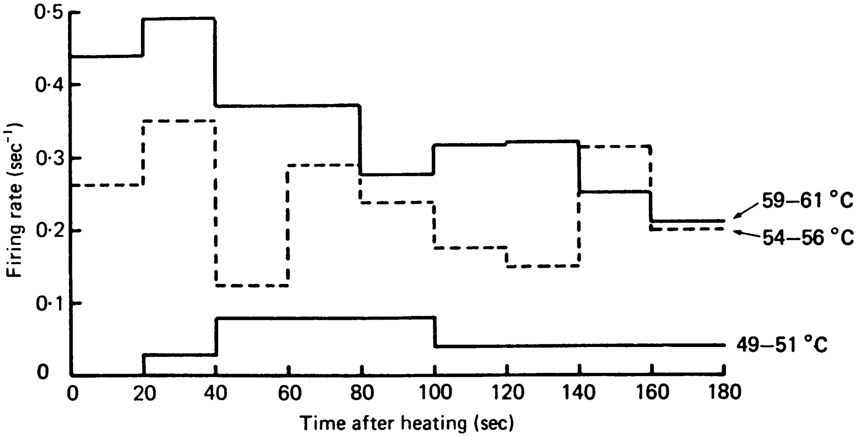

Fig. 2. Continuous background firing following initial heating to different peak temperatures. Time measured from the end of the ramp heating. Upper continuous line, average firing of ten units heated initially to 59–61 °C; interrupted line, average for four hours heated to 54–56 °C; lower continuous line, average for five units heated to 49–51 °C.

The reason for the wide range of paremeters between units is not clear. Variations from unit to unit within one animal were almost as great as the variations in the whole sample. There was also no tendency for parameters to vary systematically with the position of receptive fields.

Continuous firing following the first heat stimulus

Only two units out of twenty-nine fired in the absence of experimental stimulation before the first heating of the skin, and these only fired at low rates of 0·13 sec^{-1} or less. However, after one suprathreshold heat stimulus most units (twenty-four out of twenty-nine) became continuously active. Usually this firing started after a pause of 5–20 sec from the end of the ramp heating (e.g. see Fig. 1*A*) and then increased rapidly to a maximum 30–120 sec after the ramp whereupon it declined slowly (see Fig. 2). The frequency of ongoing firing appeared to be related to the extent of the preceding skin heating as shown in Fig. 2. The average maximum firing of five units that became continuously active after being heated to 50 °C was 0·08 sec^{-1} whilst the average maximum firing for ten units after heating to 60 °C was 6 times greater, nearly 0·5 sec^{-1}.

Heat responses following the first suprathreshold heat stimulus

Following the first suprathreshold heat stimulus, subsequent heat stimuli usually produced a greater response as shown by the representative discharges in Fig. 3*A*. In this section the responses to a second heating 4 min after the first will be described in detail.

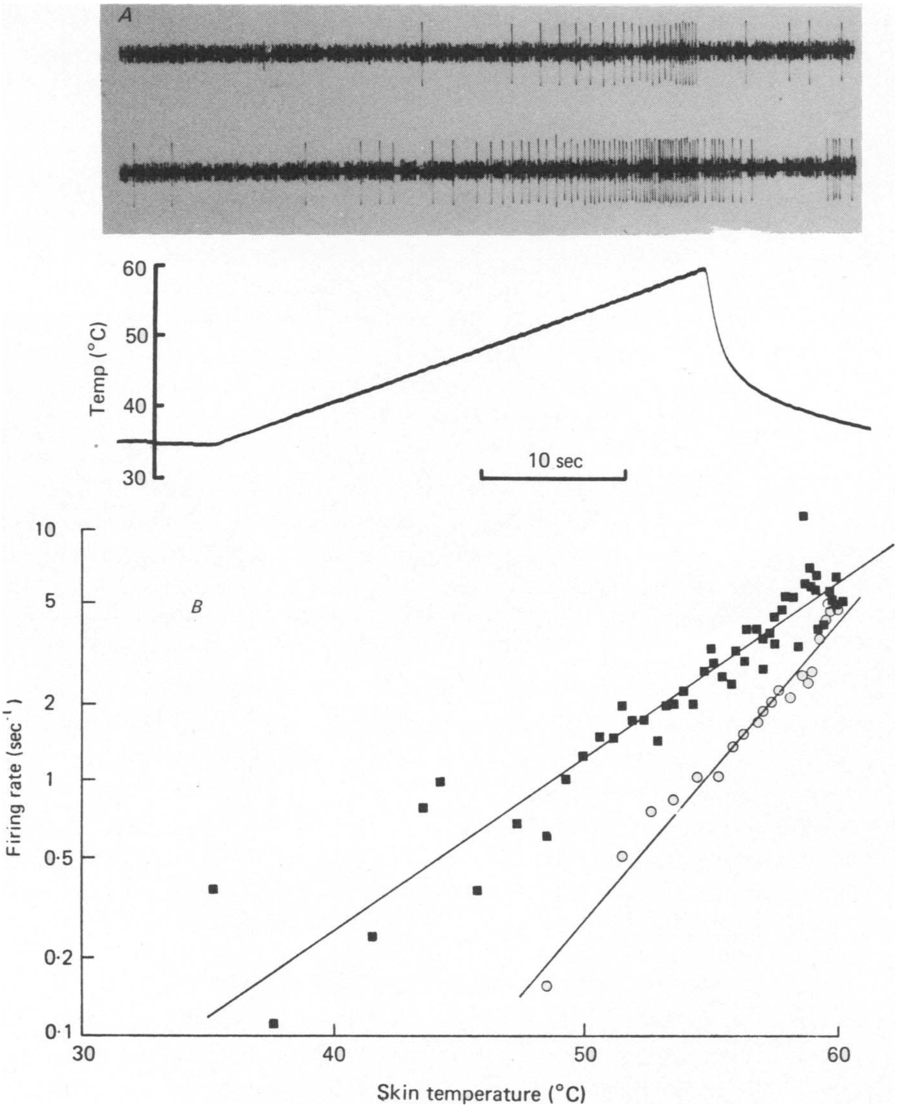

Fig. 3. Responses of nociceptor unit to first and second heat ramp. *A*, upper trace, action potentials recorded in response to the first stimulus; middle trace, responses to second stimulus 4 min after the first; lower trace, skin temperature profile during stimuli. *B*, instantaneous firing frequency against skin temperature for these responses; ○ first and ■ second ramp stimuli. Note log frequency scale. Lines through points are best-fitting regression lines for log frequency against temperature.

Despite the onset of low rates of continuous firing, clear-cut firing thresholds for heating could usually be determined. This was largely because any ongoing activity was often suppressed during the early, warming, part of the heat ramp. Thresholds measured for twenty-six units on the second heat ramp ranged from 2·9 °C above to 17·4 °C below the values for the first heating. The extent of the change was greater when the first heat stimulus was large than when it was small. Four units heated initially to 48–52 °C showed an average fall in threshold of only 1·37 °C, four units heated to 54–57 °C showed an average fall of 3·7 °C, thirteen units heated to 59–61 °C fell by 6·2 °C and five units heated to 63–67 °C had an average threshold fall of 10·2 °C. Within a group of units heated to the same temperature there was a lot of variability in the extent of threshold changes. For example, with the thirteen units heated initially to 59–61 °C the standard deviation of the threshold changes was 3·1 °C. Despite this variability, the trend for increased sensitization with increased heating was highly significant (one-way analysis of variance, linear trend, $t = 3{\cdot}3$, degrees of freedom 22, $P \simeq 0{\cdot}003$).

Regression analysis of the responses to second heat stimuli revealed some changes from the parameters for the first heat ramps although it was still possible to fit all the data to exponential functions. Fig. 3*B* shows a plot of log firing frequency against skin temperature for the first and second heat stimuli shown in Fig. 3*A*. Three differences are apparent between the first and second responses in Fig. 3. First, there is more scatter about the best fitting line, an increase that is highly significant statistically ($F = 2{\cdot}9$; $P = 0{\cdot}008$). A number of other units also showed an increase in the variability of firing when sensitized. For the thirteen units that fired at least six spikes on both the first and second heat runs the average increase in sample standard deviation was 45 % (± 30%, 5 % confidence interval). Inspection of the individual plots showed that this was a genuine increase in interval by interval variability, rather than an indirect reflexion of a poorer fit to the exponential relation.

The second difference between the first and second responses shown in Fig. 3 is that the best fitting exponent (i.e. the slope of the log frequency versus temperature relation) is smaller for the second heat run. The shift between runs in Fig. 3 is highly significant statistically, and a similar significant fall was shown by two other units out of the thirteen analysed in detail. However, one unit showed a significant rise in exponent on the second heat trial. On average the best fitting exponent on the second ramp was 84 % (± 24 %, 5 % confidence interval) of its value on the first, and so over-all there was no significant change in exponents from first to second heating.

As well as these relatively small changes in variability and exponent, the responses to second stimuli were usually shifted substantially towards lower temperatures and higher firing frequencies (e.g. see Fig. 3*B*). T_1 values for thirteen units fell by up to 12·4 °C. Eight of these units were heated to 59–61 °C on the first ramp and the average shift in T_1 for these units was 6·3 °C ± 3·9 °C (S.D.). Shifts in T_1 are clearly similar to the shifts in firing threshold although a paired comparison between ten units for which both measurements were made indicated that shifts in threshold were usually greater (on average by 2·8 ± 2·2 °C, 5 % conf. int.). The difference between the shifts in T_1 and in threshold was due to two factors, firstly the difference in exponents described above and secondly a tendency for units to start firing at a lower frequency on the second than on the first ramp.

After the second heat trial the peak stimulation temperature was lowered so that responses of five to twelve spikes were obtained for each ramp heating. When such stimuli were given at 4 min intervals responses were usually fairly stable with thresholds and regression parameters remaining close to those on the second trial.

Effects of different rates of heating

The effect of varying the rate of skin heating has been studied in six units over the range from 0·2 to 2 °C/sec. Thresholds tended to be lower with slower heating, as shown in Fig. 4. However, at relatively low skin temperatures the frequency of firing of units was higher with faster rates of heating. In a study of monkey cutaneous polymodal units Croze, Duclaux & Kenshalo (1976) found no change in heat thresholds for 4 units heated at 0·2 °C/sec and at 1·5 °C/sec. In view of the small numbers of units examined and the differences between the exact stimulus conditions of the two studies it is hard to evaluate the significance of this difference.

At all rates of heating studied frequency of firing rose exponentially with increasing skin temperature (see Fig. 4*B*). Best fitting exponents were lower when rates of heating were higher, as shown by the responses plotted in Fig. 4*B*.

The effect of stopping blood flow on nociceptor responses

Having now analysed the responses of polymodal units to controlled heating, we are in a position to investigate the effects of changing the local blood flow. Blood flow to the saphenous area was stopped by tightening a loose ligature around the femoral or saphenous artery in 8 experiments during which the responses of twenty-two units were examined.

In order to establish whether the ligation procedure was effective in stopping skin blood flow two additional experiments were performed. In these experiments small volumes of saline containing radioactive ^{133}Xe were injected into the saphenous skin and the rate of removal of tracer was determined by measuring the rate of fall in radioactivity at the injection site. A sketch of the experimental arrangements is given in Fig. 5*A*. A typical plot of log local radioactivity against time is shown in Fig. 5*B*. When both ligatures were loose activity fell steadily with a half time of approximately 17 min (equivalent to a local blood flow of 3 ml./100 g tissue/min (Lassen *et al.* 1964)). When the saphenous artery was blocked by applying a force of 0·5 N to the loose ligature, activity fell about 7 times less quickly and when the block was applied to the femoral artery, no fall in activity could be discerned at all, indicating a complete cessation of local blood flow. The injection for Fig. 5*B* was into the medial aspect of the lower leg. With injections into other parts of the saphenous area (e.g. dorsum of proximal foot; skin overlying the knee) a similar pattern was observed; activity fell steadily until one of the ligatures was tightened when activity remained constant, or nearly so, until the ligature was released. The rate of loss of activity when the arteries were not blocked was, however, very different at different sites and also often fell with time. During the first 30 mins after injection, half times at different sites ranged from 8·5 to 30 min.

Typical heat responses before, during and after a period of arterial block are shown

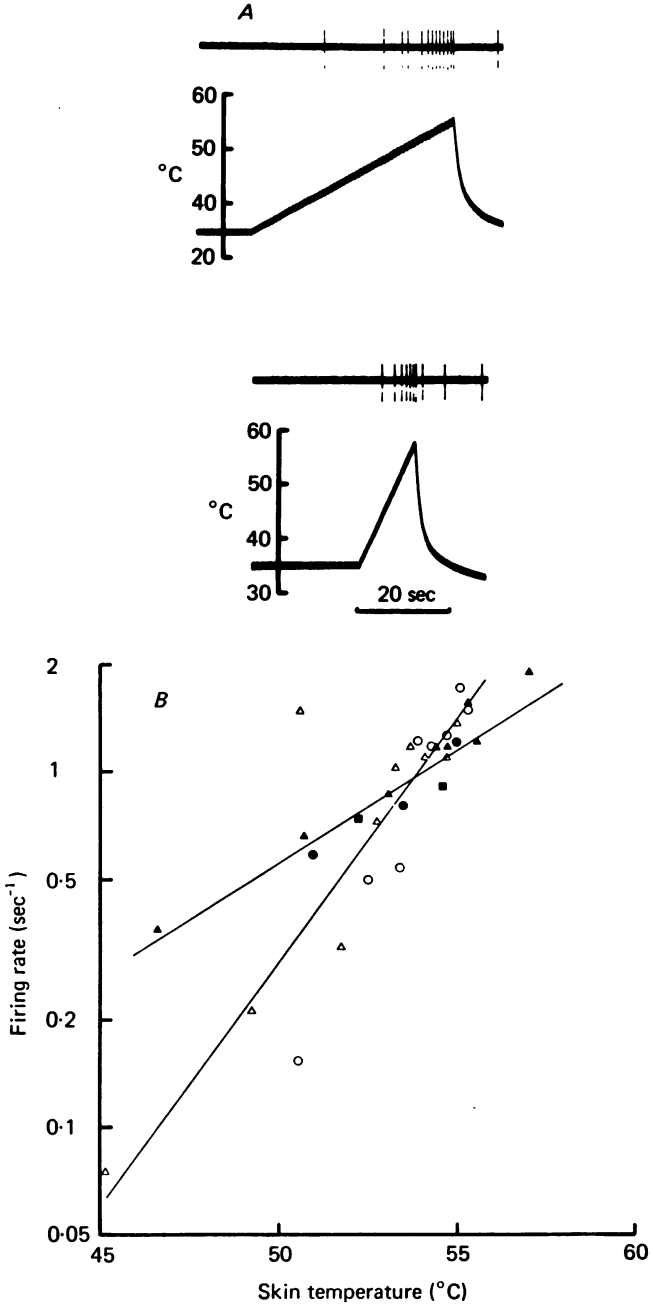

Fig. 4. Nociceptor responses to different rates of skin heating. *A*, upper traces, action potentials and stimulus profile for heating at 0·5°/sec; lower traces, action potentials and stimulus profile for heating at 2·0°/sec, same unit. Same time scale for all traces. *B*, log firing frequency versus skin temperature for three trials at 2°/sec (△, ○) and two trials at 0·5°/sec (△, ○). Same unit as in *a*. Lines are as in Figs. 1 and 3.

in Fig. 6*A*. There are no obvious effects of altering the blood flow in this unit. Similar experiments on twenty-one units where the blood flow was stopped for 8–12 min on one to four occasions failed to reveal any consistent pattern of changes. Average firing thresholds for six units during 12 min ligations are shown in Fig. 7*A*. Analysis of variance on this data indicates no significant differences in thresholds at different times. Thresholds of nineteen units were measured 2 and 6 min after blocking the saphenous or femoral artery. On average the thresholds of these nineteen units fell during the block by only 0·52 °C (± 0·99 °C, 5 % confidence limits). The number of spikes fired to a constant heating stimulus likewise showed no relation to blood flow changes. The average number of spikes fired during periods of block was increased by 7 % (± 20 %, 5 % confidence limits).

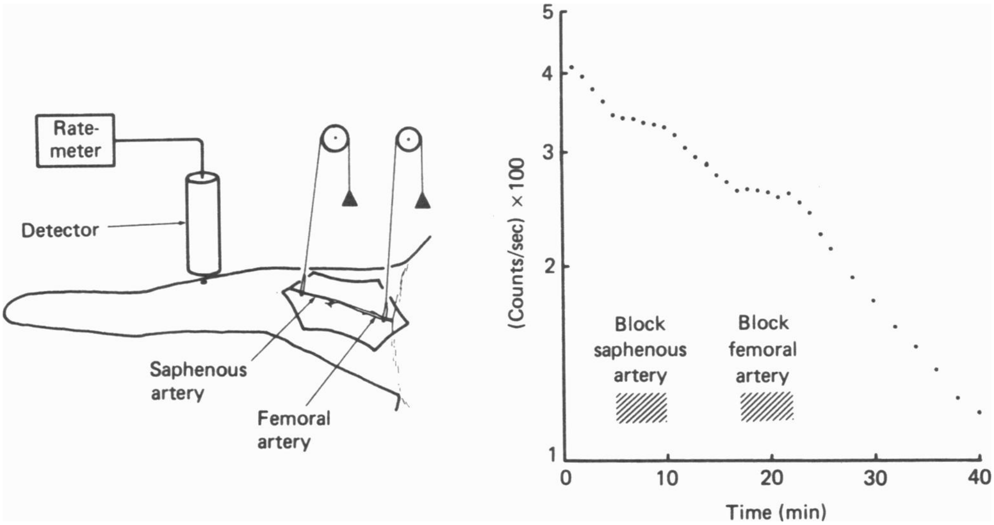

Fig. 5. Skin blood flow with and without ligating the femoral or saphenous arteries. *A*, arrangement of experiments. The site of injection of saline containing ^{133}Xe was immediately beneath the detector. A force of 0·05 N was applied to the nylon suture around the arteries to block blood flow. *B*, local radioactivity during a typical pair of blocks.

Regression analysis of log frequency versus skin temperature for ramp stimuli applied before, during and after the arterial block likewise showed no consistent trends. For example, the responses in Fig. 6*A* are plotted in Fig. 6*B* along with the best fitting regression lines for the ramps with and without arterial ligation. The best fitting lines are not significantly different under the two conditions. For all fifteen units whose responses were analysed in this way, simple standard deviations decreased 14 % (± 23 %, 5 % confidence interval), exponents fell 8 % (± 14 %) and T_1 values rose 0·21 °C (± 0·99 °C).

Continuous firing was also monitored during periods of arterial block. Average firing frequencies of seven units for the minute preceding and the minute following each heat stimulus are plotted in Fig. 7*B*. The rate of background firing fell steadily with time and does not appear to have been affected by altering the blood flow. The

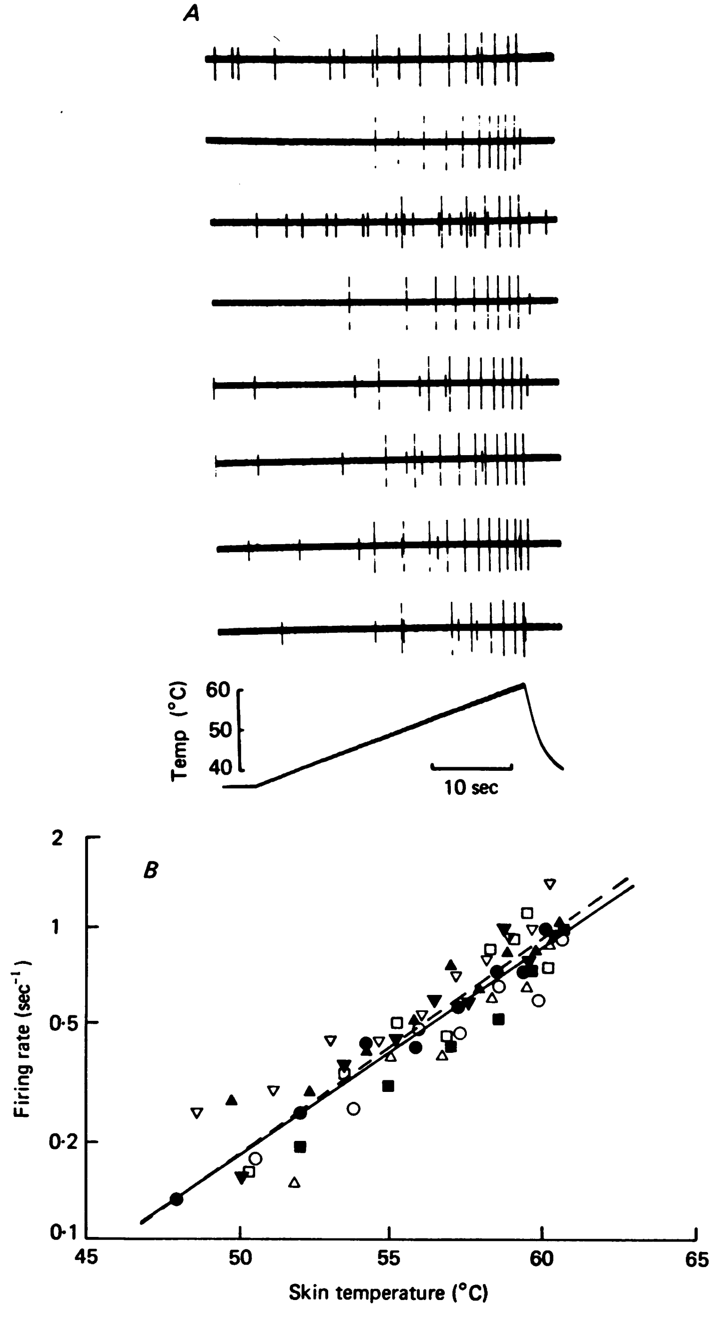

Fig. 6. Responses of a polymodal nociceptor unit (larger spikes) before, during and after blocking the femoral artery in the groin. *A*, action potentials to eight identical skin heat ramps (shown in the lowest trace) at 4 min intervals. Top two traces, before block; next four traces, during block, lowest two action potential traces, after releasing block. *B*, log firing frequency *vs.* skin temperature plots for the data in *a*. ○, 6 min before and □ 2 min before blocking the artery; ■ 2 min, ●, 6 min, ▼ 10 min and ▲ 14 min after starting block; ▽ 2 min and △ 6 min after releasing ligature. Lines through data are best fitting regression lines for log frequency against skin temperature. Continuous line is for heating before and after the block; interrupted line is for heating during the block.

average change in firing frequency for seventeen units during 8 minutes of block was a 3 % decrease (± 17·5 %).

A few units did show statistically significant trends on one parameter or another, but these units did not conform to any consistent pattern and in a number of cases the changes were not repeated on subsequent blocks. However, one unit did show a consistent set of changes on three repeated arterial blocks and these changes were in the same direction as the average trends for all units. A subsequent unit isolated in the same preparation did not show any similar trend.

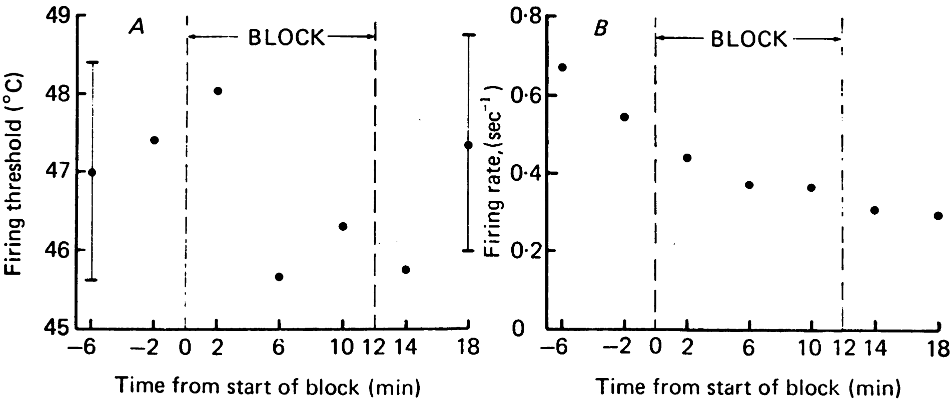

Fig. 7. Average effects of stopping skin blood flow. *A*, heat thresholds of 6 nociceptor units previously sensitized by skin heating. Error bars on first and last points indicate 5 % confidence limits for comparisons between different times. *B*, continuous background firing. Average for six units of firing during 1 min periods before and after each stimulus. Steady downward trend probably indicates recovery from large sensitizing stimuli given earlier in the experiments.

DISCUSSION

Heating thresholds in previously unstimulated skin

The threshold temperatures of polymodal units ranged from 41 to 63 °C when tested with heat ramps where the skin temperature was raised at 1 °C/sec. With slower rates of heating thresholds were lower; with higher rates they were higher. When considering the biological role of these receptors one wonders whether units with firing thresholds greater than 60 °C can contribute usefully to a warning system. In fact brief, localized heating to 60 °C with the stimulator employed in this study did not cause visible damage to the skin. With repeated heating the high threshold units rapidly sensitized and were than able to signal information about much lower temperatures. There was no indication of differences in other properties (e.g. mechanical sensitivity, receptive field size, conduction velocity) between units with high and low heat thresholds, and so it seems reasonable to treat this group of nociceptors as a single class, albeit one with a wide range of initial sensitivities.

The exponential relation between firing frequency and skin temperature

Input–output functions for sensory receptors are usually either linear or negatively accelerating (e.g. see Somjen, 1972, chap. 4). However, it is clearly appropriate for a nociceptor to have a rapidly accelerating firing rate with increasing stimulus intensity. The information being transmitted is presumably not about fine gradations in stimulus magnitude but rather about impending danger to the integrity of the body surface. A graded response over an extended stimulus range is not needed in a nociceptive system.

The exponential relation is not restricted to one set of stimulus conditions. Varying the rate of heating over a tenfold range did not alter the basic exponential pattern of firing, although there were systematic changes in the values of the best-fitting exponent. A few experiments with a contact heat thermode (B. Lynn, unpublished data) showed that under these conditions ramp heating also produced an exponentially rising firing frequency. A study of polymodal nociceptors in monkey skin has also described rapidly accelerating firing rates with increasing temperature during ramp stimulation at 0·2 °C/sec using a contact thermode (Croze *et al.* 1976). Studies of heat responses of feline C-fibre units to stepwise heating patterns have indicated an approximately linear relation between firing frequency and skin temperature (Beck, Handwerker & Zimmerman, 1973; Handwerker & Neher, 1976). However, even under these conditions average firing frequency over the population shows an increasing rate of change of firing frequency with increasing skin temperature (Handwerker & Neher, 1976).

It is clearly not possible for firing frequency to increase exponentially for long. In a few units subjected to large heat ramps firing stopped when frequencies of 10–20 sec^{-1} were reached, even though the skin temperature continued to rise. Up to the cut-off point such units fired at frequencies that fitted to the exponential relation.

The exponential relation between the firing rate and skin temperature is similar to the behaviour of many physiological (and chemical) processes. However the Q_{10} values (3·6–120) are unusually high. These high Q_{10} values may reflect the nature of the chemical processes that are induced by noxious heating. It has been suggested that the vascular permeability changes caused by thermally induced injury may be triggered by protein denaturation (Willoughby, Coote & Turk, 1969). If protein denaturation is also involved in producing nociceptor firing as suggested previously by Hardy (1953) then this would provide an explanation for the high Q_{10} values since rates of protein denaturation are known to increase unusually rapidly with increasing temperature (e.g. see Pauling, 1953, chap. 19).

The increased variability of firing following heat stimulation

It was known from previous studies that threshold temperatures fell substantially following a large heat stimulus. However an unexpected result from the analyses of the firing of polymodal units was that the variability of firing rate also increased with sensitization. This finding would be consistent with a model of sensitization that involved the formation of additional, independent routes by which excitation could occur.

Failure of stopping the blood flow to affect responses of sensitized units

Nociceptor units examined in this study were not significantly affected by stopping the local blood flow, although the results presented here do not exclude the possibility of a small effect over the 8–12 min period of arterial block. The isotope experiments showed that the concentration of injected xenon fell substantially over a 10 min period. This means that if a substance as free to move into the blood as xenon were being kept at an approximately constant level by loss in the blood stream, then it should have accumulated substantially when the blood flow was stopped for 8–12 min. Despite a number of uncertainties, for example about the possibility that synthesis of a 'pain substance' might slow when the blood flow was stopped, it seems unlikely in the light of the results presented here that the maintenance of sensitization is dependent on the continuous local production of a freely diffusible, stable 'pain substance'. Rather, the sensitization process may depend upon production of substances that are rapidly broken down locally or of substances that are not able to move quickly into the blood vessels.

How far can these results be compared with the results of Lewis & Hess (1933) which showed that hyperalgesia in man increased during short periods when the blood flow was stopped by a pressure cuff? In fact there are many differences between the experimental conditions. Lewis & Hess (1933) only reported in detail about hyperalgesia following mechanical trauma or exposure to ultra-violet radiation. It is clearly important to examine the changes in receptor endings under these conditions before drawing any conclusions about similarities or differences between human hyperalgesia and polymodal nociceptor sensitization.

This study was restricted to skin receptors of only one type, polymodal nociceptors with C-fibre axons. In fact the firing of 2 other types of cutaneous receptor unit in response to noxious heating is also increased by previous stimulation. These units are the high threshold mechanoreceptors with A-delta axons (Fitzgerald & Lynn, 1977) and the cold receptors (paradoxical firing) (Dubner, Sumino & Starkman, 1974). It is clearly possible that these receptor types might show increased sensitization during periods of zero blood flow and this possibility needs further investigation.

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