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INHIBITION OF CUNEATE NEURONES: ITS AFFERENT SOURCE AND INFLUENCE ON DYNAMICALLY SENSITIVE 'TACTILE' NEURONES

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SUMMARY

1. Responses were recorded in decereberate, unanaesthetized cats from individual cuneate neurones in order to determine firstly, the afferent sources of inhibition on cuneate neurones and secondly, the influence of afferent-induced inhibition on those response features of dynamically sensitive tactile neurones which determine their capacity to code information about parameters of tactile stimuli.

2. For all cuneate neurones which displayed afferent-induced inhibition from areas surrounding or within their excitatory receptive field (71 % of the sample) it was consistently found that 300 Hz vibration at low amplitudes (< 25–50 μm) which selectively engages Pacinian corpuscles was an effective source of inhibition. In contrast, steady indentation which activates slowly adapting tactile afferents was quite ineffective, as was low frequency vibration (30 Hz) at amplitudes of < 50–100 μm. The latter stimulus can be used to engage rapidly adapting receptors either within glabrous skin (presumed to be Meissners corpuscles) or in association with hair follicles. It is concluded that afferents from Pacinian corpuscles are the dominant or exclusive source of afferent-induced inhibition of cuneate neurones.

3. For dynamically sensitive neurones responsive to low frequency cutaneous vibration (30 Hz) there was a reduction in the slope of stimulus–response relations with afferent-induced inhibition, but no expansion of the range of stimulus amplitudes over which the neurone responded.

4. The influence of afferent-induced inhibition on the phase-locking of impulse activity to a cutaneous vibratory wave form was examined by constructing post-stimulus time histograms and cycle histograms. Measures of dispersion of impulse activity around the preferred point of firing in the vibratory waveform indicated that the capacity of individual cuneate neurones to code information about the *frequency* of the cutaneous

vibration was not systematically changed in the presence of afferent-induced inhibition.

INTRODUCTION

Afferent-induced inhibition of transmission through the dorsal column or trigeminal relays of the somatosensory pathways is primarily or exclusively evoked by *dynamic* tactile stimuli such as vibration or blowing (Andersen, Etholm & Gordon, 1968, 1970; Rowe & Carmody, 1970; Carmody & Rowe, 1974). Steady tactile stimuli which activate slowly adapting or tonically sensitive tactile afferent fibres were found to be ineffective, although such stimuli have been shown to induce primary afferent depolarization in the terminals of slowly adapting mechanosensitive afferent fibres at the level of the lumbar dorsal horn of the spinal cord (Jänig, Schmidt & Zimmermann, 1968). Jänig *et al.* (1968) also demonstrated that inputs from Pacinian corpuscles evoked primary afferent depolarization in the lumbar dorsal horn but they were unable to determine whether inputs from other rapidly adapting receptors also contributed. The studies within the dorsal column and trigeminal relays (Andersen *et al.* 1970; Carmody & Rowe, 1974) did not provide definite identification of the tactile receptors responsible for the afferent-induced inhibition as the tactile stimuli used were not controlled in a way which resulted in selective activation of known receptor types. In the present study sinusoidal vibration has been employed as a controlled and reproducible form of dynamic tactile stimulation. Account has been taken of the observations of Talbot, Darian-Smith, Kornhuber & Mountcastle (1968) and Merzenich & Harrington (1969) which demonstrated the differential sensitivity to sinusoidal vibration of different groups of tactile afferent fibres. They found that one class could be selectively activated at low amplitudes of cutaneous vibration at 200–300 Hz which indicated that these innervated Pacinian corpuscles (Hunt & McIntyre, 1960; Sato, 1961). Talbot *et al.* (1968) reported that Pacinian afferent fibres were typically unresponsive at 30 Hz unless vibration amplitudes exceeded 50–100 μm. In contrast, dynamically sensitive tactile receptors *within* the glabrous and hairy skin, whose afferents project centrally in the dorsal columns (Wall, 1960; Brown, 1968; Petit & Burgess, 1968), are most sensitive to low-amplitude cutaneous vibration at 20–50 Hz (Talbot *et al.* 1968; Merzenich & Harrington, 1969). Those within the glabrous skin have been presumptively identified as Meissners corpuscles (Talbot *et al.* 1968; Jänig, 1971), while those in the hairy skin are associated with the Type G hair follicles (Brown & Iggo, 1967; Burgess, Petit & Warren, 1968; Merzenich & Harrington, 1969).

In an earlier study on *slowly adapting* 'tactile' neurones at the first relay

level of the somatosensory pathways Carmody & Rowe (1974) examined the effects of afferent-induced inhibition on the capacity of those neurones to code information about the intensity of steady skin indentation. In the present study we have investigated the influence of afferent-induced inhibition on *dynamically sensitive* 'tactile' neurones of the cuneate nucleus, in particular its influence on response features which are important for coding information about the parameters of sinuosidal vibration applied to the skin. This form of stimulation is particularly suitable for investigating these neurones as it simulates the vibratory patterns normally set up in the skin in association with cutaneous texture and pattern discrimination. In the present experiments both the frequency and intensity parameters of the vibration could be controlled systematically and reproducibly.

METHODS

Animal preparation

Experiments were performed on twenty-one unanaesthetized adult cats, decerebrated surgically at the mid-collicular level. Initially the animal was anaesthetized with halothane, the trachea cannulated and the femoral vein and artery catheterized. The animal was then placed in a frame with conventional stereotaxic supports. A fronto-parietal craniotomy allowed access for the decerebration after which halothane administration was terminated. The cuneate nucleus was exposed by removing the atlas and a small part of the occipital region of the skull. Throughout the recording period animals were immobilized with gallamine triethiodide and artificially ventilated. Blood pressure and rectal temperature were monitored in all experiments. End tidal P_{CO_2} was monitored in most experiments and held at 3–4 %.

Recording and stimulation procedures

Impulse activity was recorded from individual neurones within the cuneate nucleus using either glass micro-electrodes (3–8 MΩ, filled with 2 M-NaCl) or tungsten micro-electrodes. A 4 % agar gel was placed over the exposed brain stem during recording periods to minimize cardiac and respiratory pulsations. Penetrations were made between 1 and 4 mm posterior to the obex where presynaptic inhibition is strongest (Andersen, Eccles, Schmidt & Yokota, 1964; Andersen *et al.* 1970) and where afferent inhibition of neurones in the adjacent gracile nucleus is most pronounced (Gordon & Jukes, 1964). Confirmation that neurones studied were within the cuneate nucleus was based on subsequent histological verification of the recording site in haematoxylin stained sections (50 μm in thickness).

Neurones principally selected for study had receptive fields on the ventral surface of the distal fore limb, which in all experiments was fixed, pads uppermost, in a Perspex trough with paraffin wax (Schmidt, Senges & Zimmermann, 1967; Leicht, Rowe & Schmidt, 1973). This stabilized the limb and permitted accurate positioning of the mechanical stimulator over the foot pads or nearby hairy skin. Prior shaving of the hair in this area ensured that stimuli applied to the glabrous skin of the foot pads did not directly displace nearby hairs.

When a single neurone was isolated electrophysiologically its excitatory receptive field was delineated by gentle tapping or brushing with a small probe. Precise and reproducible mechanical stimuli derived from a servo-controlled mechanical stimulator

(Darian-Smith, Rowe & Sessle, 1968; Carmody & Rowe, 1974) were applied as *test* stimuli to the point of maximum sensitivity within the excitatory receptive field of the neurone. From a second stimulator of the same type conditioning stimuli could be applied, usually to areas beyond the excitatory receptive field in order to engage particular afferent fibre types so that their inhibitory actions on cuneate neurones could be examined. Stimuli were delivered using circular probes 4–6 mm in diameter and were repeated at rates no faster than one per 10 seconds to allow recovery of skin position.

Neurones were tested for afferent-induced inhibition by comparing the average number of impulses evoked in ten or more responses to the test stimulus alone with the average number evoked when the test stimulus was accompanied by the conditioning input. A significant reduction (t-test) in mean impulse frequency with conditioning was taken as evidence of inhibition. Test and conditioning stimuli included a steady displacement of 'rectangular' form (1·4 sec duration; rise time 30 msec; amplitude $\leqslant 2$ mm) which was synchronized when both test and conditioning stimuli were operating. Where a rapidly adapting cuneate neurone was being examined the *test stimulus* included a train of sinusoidal vibration (usually 1 sec) superimposed on, and commencing 200 msec after the onset of the rectangular displacement (see Fig. 1). When the inhibitory influence of rapidly adapting afferents was being studied the *conditioning stimulus* also included a vibratory train commencing 170 msec after the onset of the rectangular displacement and lasting 1·03 sec. The 30 msec interval between the start of test and conditioning vibratory stimuli was to allow for the onset of inhibitory action before the start of the test response.

Impulse activity was displayed on an oscilloscope and fed to a differential amplitude discriminator from which constant output pulses could be relayed to a counter unit and a laboratory computer (PDP-8) which was programmed to produce post-stimulus time histograms and cycle histograms. The post-stimulus time histograms use a pulse associated with the start of each train of vibratory cycles as the stimulus marker and therefore show the probability of occurrence of impulse activity throughout the period of the test vibration train. The cycle histograms use a pulse associated with the onset of *each* successive vibratory cycle as the stimulus marker and therefore display the probability of an impulse occurring throughout the period of the vibratory cycle. Up to fifty repetitions of the vibratory train were used to construct the post-stimulus time histograms and a total of 1000–2000 cycles of vibration, usually delivered in 1 sec trains, for constructing the cycle histograms. These analyses permitted an evaluation of the effect of afferent-induced inhibition on the responses of cuneate neurones to vibratory cutaneous stimulation, in particular, on the extent to which impulse activity was entrained or phase-locked to the vibratory stimulus. However, because of conduction and synaptic delays, the actual phase relation is not known, particularly with faster vibratory frequencies.

RESULTS

Neurone types within the cuneate nucleus responding to tactile stimuli

The receptive fields of ninety-three neurones responding to light tactile stimulation of the skin were in most cases confined to areas of the distal fusilateral forelimb. Neurones were classified as slowly adapting or rapidly adapting on the basis of their response to a controlled 'rectangular' indentation of the skin lasting 1·4 sec. Of seventy-five 'tactile' neurones

whose functional properties were examined, seventeen (23 %) responded throughout this steady indentation and were designated slowly adapting neurones (Fig. 1*A*). The majority had receptive fields confined to the glabrous skin of one or more foot pads, while a smaller proportion of cells had receptive fields on the hairy skin or areas including both glabrous and hairy skin and were usually about 1 cm^2 in area.

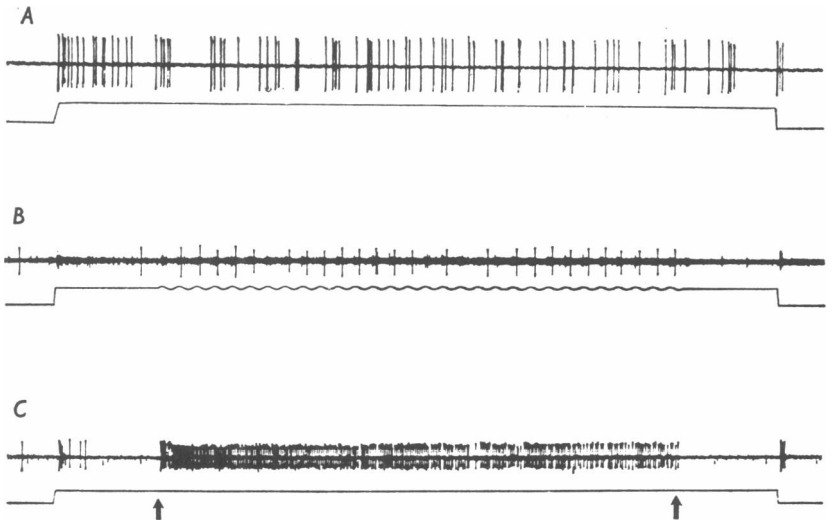

Fig. 1. Responses of cuneate neurones to tactile stimulation of the skin. *A*, response of a slowly adapting neurone to a 2 mm indentation of 1·4 sec duration applied to the pad of toe 3. The neurone's receptive field included the central foot pad and pads of toes 2 and 3. *B* and *C*, responses of dynamically sensitive cuneate neurones to sinusoidal vibration of the skin; in *B* 30 Hz (50 μm) vibration (1 sec duration) superimposed on a 270 μm rectangular indentation to the pad of toe 2; receptive field included the central pad and toes 2–5. *C*, response of cuneate neurone sensitive to high frequency vibration, 300 Hz, 25 μm; 1 sec duration, superimposed between the arrows on a 1·2 mm indentation to medial side of central foot pad; (the vibration wave form is not apparent here because of the small amplitude); receptive field extended from distal extremity of forelimb to shoulder. Upward deflexions in impulse traces are negative-going.

Neurones responding only to the dynamic ('on' and 'off') phases of the 1·4 sec rectangular stimulus were designated rapidly adapting or dynamically sensitive neurones and were divided into two functional subgroups based on their responsiveness to sinusoidal vibration. One subgroup, of thirty-nine neurones, was most sensitive to cutaneous vibration at frequencies of 20–50 Hz (Fig. 1*B*) whereas the nineteen neurones in the other subgroup were most sensitive at 200–300 Hz (Fig. 1*C*). There was little

overlap in the sensitivity of neurones in these two classes, particularly if low amplitudes ($< 50\ \mu m$) of vibration were employed. In Fig. 2 the average discharge rate during 1 sec periods of vibration (50 μm amplitude at all frequencies) is plotted at a series of different vibration frequencies for an individual neurone typical of each of these two classes. The neurone

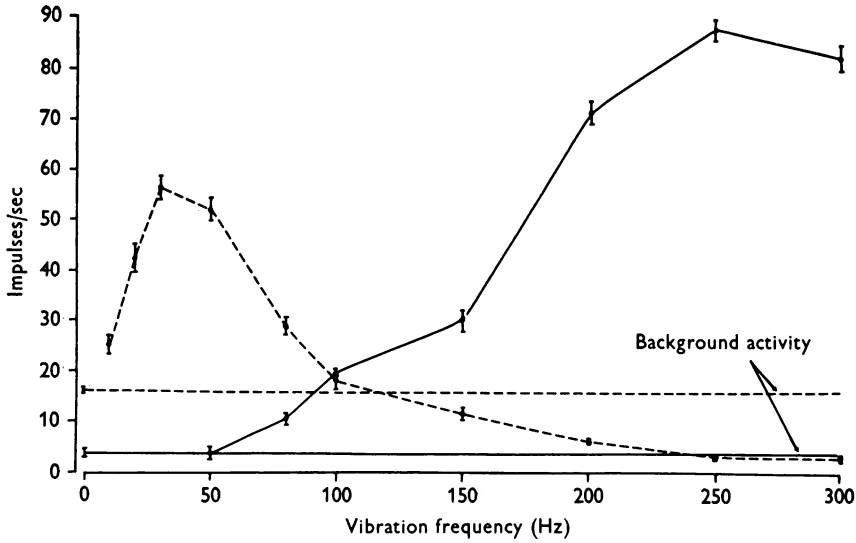

Fig. 2. Vibratory sensitivity of dynamically sensitive tactile neurones within the cuneate nucleus. Graphs plot mean discharge rate (± S.E.; $n = 10$) during 1 sec periods of sinusoidal vibration (50 μm at all frequencies, superimposed on 1·2 mm rectangular indentation) for two neurones; the broken line was obtained from responses of a neurone with receptive field on toes 2, 3 and 4 following stimulation of toe 3; the continuous line following stimulation of toe 2 for a neurone with receptive field which included all the distal forelimb. The two horizontal lines represent the average background discharge rate for the two neurones.

in the 'low frequency' class (interrupted line) was most sensitive at 20–50 Hz and with stimulation at high frequencies was inhibited from within its excitatory receptive field. In contrast, the neurone from the 'high frequency' class (continuous line) displayed an increased discharge rate only at frequencies above 50 Hz and was most sensitive at frequencies of 200–300 Hz. Neurones responsive to high frequencies usually had thresholds of < 1–$2\ \mu m$ at 300 Hz. As the only type of mechanoreceptor known to respond to cutaneous vibration at this frequency and amplitude is the Pacinian corpuscle (Hunt & McIntyre, 1960; Hunt, 1961; Sato, 1961; Talbot *et al.* 1968; Merzenich & Harrington, 1969) it may be concluded that cuneate neurones of this class derive their input from afferent fibres

of Pacinian corpuscles and will be referred to as Pacinian neurones. The receptive fields of the Pacinian neurones studied usually included the central foot pad region around which Pacinian corpuscles are known to be concentrated (Lynn, 1971) and often extended over the whole limb or large parts of it. On average they were considerably larger than receptive fields of neurones in other classes. However, because of their exquisite sensitivity to rapid mechanical disturbances it was more difficult to specify the extent of receptive fields for Pacinian neurones than for other neurone types.

Dynamically sensitive cuneate neurones responsive to low frequency cutaneous vibration had thresholds of $< 10\ \mu m$ at vibration frequencies of 20–40 Hz. Their receptive fields were usually 1–3 cm^2 in area on the glabrous skin of the foot pads or areas of hairy skin. For about a quarter of the cells the field included areas of both glabrous and hairy skin.

Sources of afferent-induced inhibition on different neurone types in the cuneate nucleus

Interactions of tactile inputs were studied satisfactorily on fifty-five cuneate neurones in order to test for afferent inhibition and, where it was present, to characterize the stimulus forms inducing it so that reliable inferences could be made about the receptors and afferent fibre types responsible for these inhibitory actions. Thirty-nine neurones (71 %) displayed afferent-induced inhibition, which was evoked from areas in the vicinity of, or within the excitatory receptive field of the neurone under study.

In order to investigate separately the capacity of different receptor types for inducing inhibition of individual cuneate neurones three forms of conditioning stimuli were routinely employed in testing each neurone; steady indentation up to 1·5 mm in amplitude, sinusoidal vibration at, firstly, 30 Hz and secondly 300 Hz, the vibration amplitude being usually less than 50 μm in amplitude.

A consistent observation for all neurone types studied was that steady indentation ($\leqslant 1{\cdot}5$ mm) and low frequency cutaneous vibration at amplitudes of 50 μm or less were ineffective sources of afferent-induced inhibition, whereas 300 Hz vibration at low amplitudes was very effective. This inhibition by high frequency cutaneous vibration was seen for eight out of fourteen slowly adapting cuneate neurones, for nineteen out of twenty neurones responding to low frequency vibration of glabrous skin and for nine out of fifteen neurones responding to low frequency vibration of the hairy skin. Three Pacinian neurones whose receptive fields did not occupy the whole forelimb were clearly inhibited by a conditioning 300 Hz vibration applied to the same limb. However, the larger excitatory

receptive fields of other Pacinian neurones prevented satisfactory evaluation of a possible inhibitory effect of the high frequency conditioning stimulation.

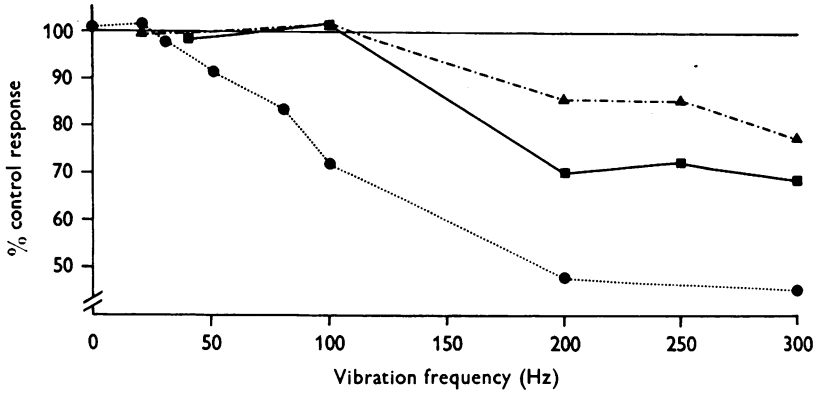

Fig. 3. Inhibition of cuneate neurones by vibratory conditioning stimuli. Response magnitude expressed as % control response (ordinate) for three classes of cuneate neurone in the presence of different frequencies of conditioning vibratory stimuli (abscissa). For the slowly adapting neurone (triangles and broken line) the test stimulus was 1000 μm steady indentation to receptive field on pad of toe 3; conditioning vibratory stimulus, 50 μm in amplitude at all frequencies, applied to lateral side of central pad. For the rapidly adapting cuneate neurone (squares and continuous line) the test stimulus was 30 Hz (25 μm) to receptive field on pad of toe 3; conditioning vibratory stimulus, 40 μm in amplitude at all frequencies, applied to pad of toe 5. For the Pacinian neurone (circles and dotted line) the test stimulus was 300 Hz (1 μm) applied near elbow (receptive field extended from wrist to elbow); conditioning vibratory stimulus applied to distal side of central pad, amplitude 25 μm at frequencies up to 100 Hz, 10 μm at 200 and 300 Hz.

The effectiveness of different vibratory frequencies was examined in more detail for each class of cuneate neurone (Figs. 3 and 4). In Fig. 3 graphs showing the inhibition of test responses (expressed as % control response) with different frequencies of conditioning vibratory stimuli are plotted for two neurones with inputs from glabrous skin and for one Pacinian neurone. The afferent inhibition was significant only at frequencies above 50–100 Hz for all three neurone types, firstly, the slowly adapting neurone with input from glabrous skin (triangles, Fig. 3; $P > 0{\cdot}5$ for 100 Hz; $P < 0{\cdot}01$ for 200 Hz), secondly, the rapidly adapting neurone also with glabrous skin input (squares, Fig. 3; $P > 0{\cdot}5$ for 100 Hz; $P < 0{\cdot}01$ for 200 Hz) and thirdly for the Pacinian neurone (circles, Fig. 3; $P > 0{\cdot}2$ for 50 Hz; $P < 0{\cdot}01$ for 80 Hz).

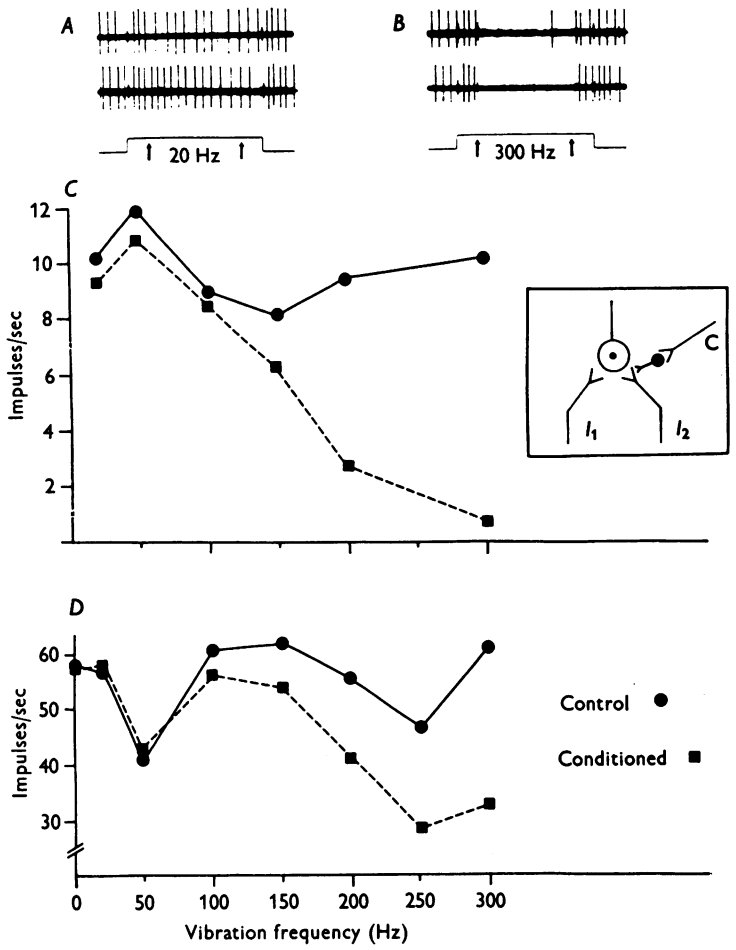

Fig. 4. Inhibition of cuneate neurones receiving input from hairy skin. *A* and *B*, two specimen traces of background impulse activity of a slowly adapting cuneate neurone. Conditioning vibratory stimulation (1 sec duration, between arrows, on 1·4 sec rectangular indentation of 1 mm) applied to the pad of toe 5 produced clear inhibition at 300 Hz (25 μm) in *B* but no effect at 20 Hz (50 μm) in *A*. *C*, effect of a range of conditioning vibratory frequencies (25 μm) on background firing rate; circles represent control background rate, squares the rate in the presence of conditioning stimulation. Inset shows schematically a possible explanation for differential inhibitory effect of conditioning stimulus on background firing and evoked responses; I_1 and I_2 represent inputs to the cuneate neurone, one of which (I_1) is responsible for evoked responses, the other (I_2) for background activity of the neurone. Only inputs over I_2 are subject to presynaptic inhibitory action from conditioning sources (*C*). *D*, effect of different conditioning vibratory frequencies (25 μm amplitude at all frequencies) on responses of a dynamically sensitive cuneate neurone to 20 Hz vibration applied to its receptive field on the hairs medial to the central pad; conditioning stimulation to pad of toe 5.

The inhibitory actions induced by high frequency cutaneous vibration appear to operate upon other classes of 'tactile' cuneate neurones as the pattern seen in Fig. 3 was also observed for both slowly adapting and rapidly adapting neurones whose excitatory input came from the hairy skin (Fig. 4). For the particular slowly adapting neurone for which data are shown in Fig. 4*A–C* the conditioning vibratory stimulus was without inhibitory action on the background discharge rate of the neurone at 20 Hz,

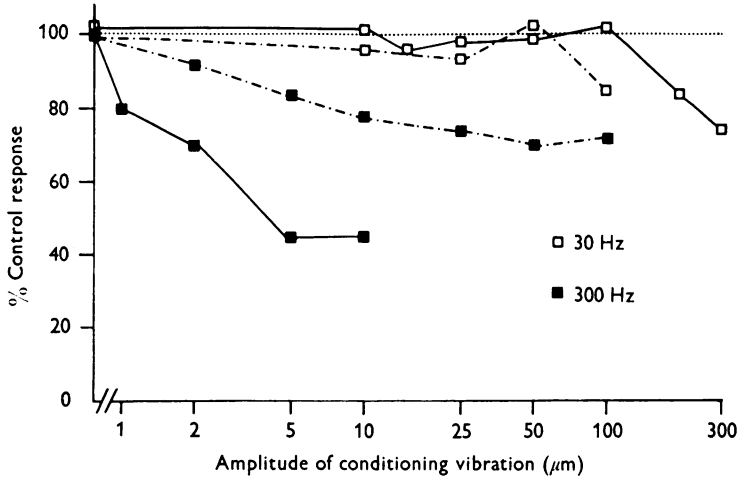

Fig. 5. Effect of variations in amplitude of conditioning vibratory stimulation at 30 and 300 Hz on the extent of inhibition of cuneate neurones. Response magnitude, expressed as % control response (ordinate) for a dynamically sensitive neurone responsive to low frequency vibration (interrupted line) and a Pacinian neurone (continuous line) at different amplitudes of conditioning vibration (abscissa-log scale). The dotted line indicates responses at the control level. The test stimulus for the Pacinian neurone was a 300 Hz (1 μm) vibration applied to the skin near the elbow; conditioning stimuli to distal side of central pad. For the other neurone the test stimulus was 30 Hz (50 μm) applied to the pad of toe 3; conditioning stimuli to lateral margin of central pad. Conditioning vibration was 30 Hz (open squares) or 300 Hz (filled squares).

50 μm (Fig. 4*A*) but was powerfully inhibitory at 300 Hz, 25 μm (Fig. 4*B*). When a range of frequencies was employed (Fig. 4*C*) only those above 100 Hz produced significant inhibition ($P > 0{\cdot}5$ for 100 Hz; $P < 0{\cdot}02$ for 150 Hz). This particular neurone was unusual in that the high frequency conditioning stimulus was without inhibitory effect ($P > 0{\cdot}5$) on its evoked responses, in this case following rectangular indentation applied to its excitatory receptive field. This apparently selective inhibitory action exerted by the afferent fibres engaged by high

frequency vibration may result from a presynaptic inhibitory action confined to inputs contributing to the background activity of the neurone (Fig. 4*C* inset). For the dynamically sensitive neurone (Fig. 4*D*) driven from the hairy skin, significant inhibition was also only seen at frequencies above 100 Hz ($P > 0{\cdot}2$ for 100 Hz; $P < 0{\cdot}05$ for 150 Hz). When the

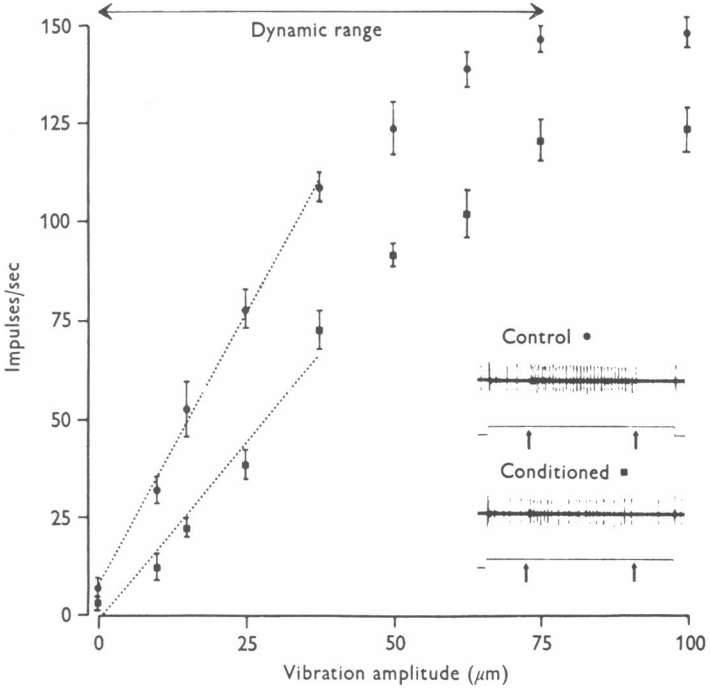

Fig. 6. Influence of afferent inhibition on stimulus–response relationship for a dynamically sensitive cuneate neurone. Control (circles) and conditioned (squares) stimulus–response relations were constructed from responses to 1 sec periods of 30 Hz vibration superimposed on 1·4 sec rectangular indentation of 0·2 mm applied to the hairy skin just proximal to the central pad (receptive field 8 mm in diameter). Each point represents mean ± S.D. for ten responses. The inset shows in the upper trace a control response to 30 Hz (15 μm) applied between the arrows and in the lower trace a conditioned response to the same stimulus. The conditioning stimulus (not shown) was 300 Hz (40 μm) vibration applied to toe pad 4. The slopes were determined for each curve over the steep initial sections (0–37·5 μm) and are indicated by the dotted lines.

amplitude of the conditioning vibratory stimulus was varied the 300 Hz frequency was usually an effective source of inhibition at amplitudes as low as 1–5 μm, while at 30 Hz the inhibition was only found at amplitudes above 50–100 μm. This is seen in Fig. 5 for two cuneate neurones, one a

Pacinian neurone (continuous line) with receptive field extending from the wrist to proximal areas of the forelimb and the other a low-frequency dynamically sensitive neurone (interrupted line) with receptive field on the pads of toes 3 and 4. For the 30 Hz conditioning vibration (open squares) significant inhibition was seen only at the 100 μm amplitude ($P < 0{\cdot}05$) for the 'low frequency' neurone and at amplitudes of 200 μm ($P < 0{\cdot}01$) and above for the Pacinian neurone. In contrast, with 300 Hz conditioning (filled squares) significant inhibition was seen at an amplitude of 5 μm ($P < 0{\cdot}05$) for the 'low frequency' neurone and at 1 μm ($P < 0{\cdot}01$) for the Pacinian neurone. In all thirty-nine cuneate neurones which displayed afferent inhibition a 30 Hz conditioning stimulus at amplitudes of 50 μm or less had no significant inhibitory action.

Influence of afferent-induced inhibition on response features of dynamically sensitive cuneate neurones

The capacity of individual cuneate neurones which respond to low frequency cutaneous vibration to code information about the intensity of vibration at a particular frequency presumably depends, in part, on the range of vibration amplitudes over which the neurone displays increments in its response, i.e. its dynamic range (Jänig *et al.* 1968; Carmody & Rowe, 1974) and on the sensitivity of the neurone to the vibration, a measure of which is provided by the slope of the neurone's stimulus–response relationship. There was no evidence of a change in dynamic range for these cuneate neurones in the presence of afferent-induced inhibition (Fig. 6). For the stimulus–response relations constructed in Fig. 6 from responses to 30 Hz vibration the dynamic range was approximately 75 μm for both unconditioned and conditioned curves. However, linear regression analysis showed that the slope over the steep portion of the stimulus-response relation was reduced from $2{\cdot}76 \pm 0{\cdot}11$ impulses/1 μm to $1{\cdot}86 \pm 0{\cdot}21$ impulses/1 μm, a 33% reduction which was highly significant ($P < 0{\cdot}01$). The conditioning stimulus in this case was a 300 Hz vibration at an amplitude of 40 μm.

The capacity of cuneate neurones responsive to low frequency vibration to code information about the *frequency* of the vibration will presumably depend on the extent to which their impulse activity is entrained or phase-locked to the wave form of the applied vibratory stimulus. In order to examine the effect of afferent-induced inhibition on the phase-locking of impulse activity, post-stimulus time histograms and cycle histograms (see Methods) were constructed, firstly from unconditioned responses of cuneate neurones to 30 Hz vibratory stimulation and then from a series of conditioned responses. Most (eight out of thirteen) cuneate neurones tested in this way showed no apparent change in their entrainment. This evalua-

tion was made by visual inspection of the post-stimulus time histograms and by computing and comparing an index of dispersion of impulse activity within cycle histograms for unconditioned and conditioned responses. In the remaining five neurones the phase-locking of impulse activity appeared to be poorer for one neurone and tighter for the other four in the presence of the conditioning inhibitory input. In Fig. 7 for

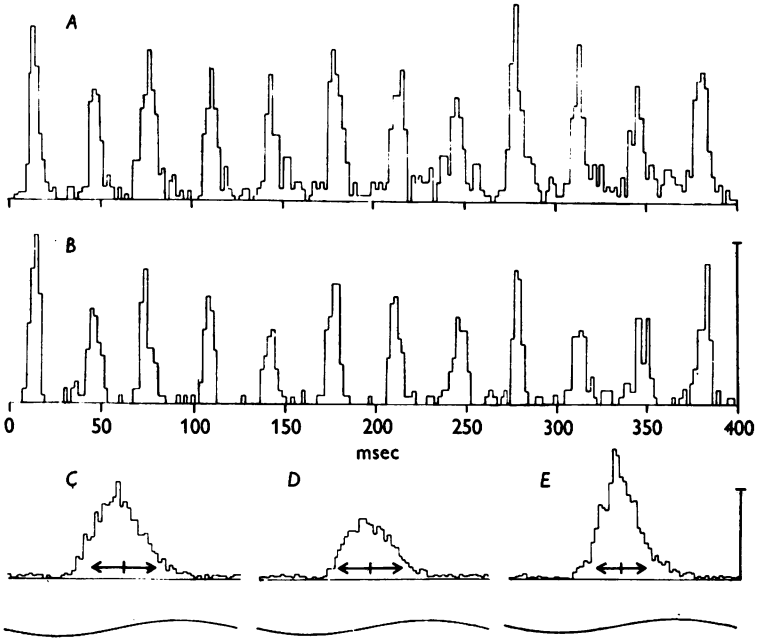

Fig. 7. Histograms for responses to 30 Hz sinusoidal vibration. *A* and *B*, post-stimulus time histograms showing the distribution of impulse activity for a cuneate neurone during 50 repetitions of a 400 msec segment of vibration (30 Hz) applied to the pad of toe 4. *A*, for unconditioned responses to 30 Hz (25 μm); *B*, for conditioned (300 Hz, 100 μm to central foot pad) responses to 30 Hz (75 μm) test stimulus. Calibration bar on right hand side of *B* represents 25 counts; applies to *A* and *B*. *C–E*, cycle histograms from responses of a different cuneate neurone to vibration (30 Hz) to the hairy skin medial to central pad. The duration of each cycle histogram was 32·5 msec. The wave form of the vibratory cycle is represented below each cycle histogram. Each histogram constructed from responses occurring in 1000 cycles of vibration delivered in 1 sec trains every 12 sec. *C*, distribution of activity for unconditioned responses to 30 Hz (25 μm) vibration; *D*, distribution of responses to the same test stimulus in the presence of a 300 Hz (100 μm) conditioning stimulus applied to the carpal pad; *E*, distribution of conditioned responses to an augmented test stimulus, 30 Hz (40 μm). Horizontal bars with arrows represent ± 1 S.D. unit about the mean for each distribution. Vertical calibration bar in *E* represents 100 counts; it applies to *C*, *D* and *E*.

example, the post-stimulus time histograms in *A* and *B* suggest that for this cuneate neurone the impulse activity was more tightly phase-locked to a preferred point in the 30 Hz vibration cycle for conditioned responses (*B*) than for unconditioned responses (*A*). For a different cuneate neurone the extent of phase-locking at 30 Hz is compared in a series of cycle histograms in Fig. 7*C–E*. A measure of the dispersion of impulse activity is indicated on each histogram by the horizontal bar which indicates the extent of ± 1 standard deviation unit around the mean for the distribution. Its value was similar for unconditioned responses in *C* (4·3 msec)

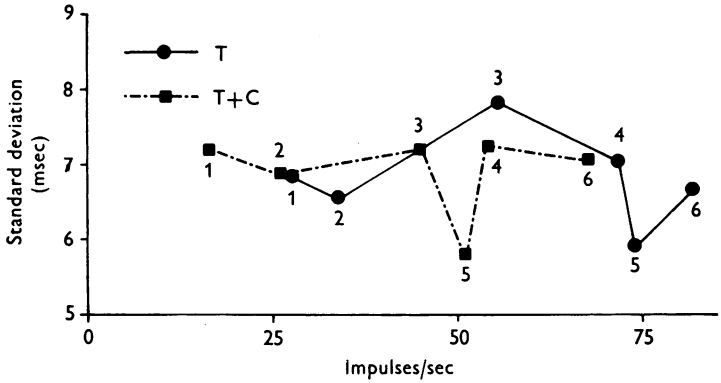

Fig. 8. Dispersion of impulse activity within individual vibratory stimulus cycles. The ordinate plots the standard deviation in msec for the distribution of impulse activity in cycle histograms constructed from responses of a cuneate neurone to 30 Hz vibration applied to the pad of toe 1. The dispersion measurements were made at six levels (1–6) of response (impulses/sec, on abscissa) to test stimulation alone (T) and for responses evoked in the presence of the conditioning stimulation (T + C). The paired test and conditioned points (obtained at the same test stimulus intensity) are indicated by the identity of associated numbers on the plots.

and conditioned responses in *D* (4·8 msec) to a 30 Hz (25 μm) test stimulus although the value was lower (3·6 msec) for the conditioned responses in *E* which were evoked by an augmented test stimulus (30 Hz, 40 μm). This observation emphasizes the importance of comparing the phase-locking of impulse activity in conditioned and unconditioned responses at a number of different response levels. However, detailed observations of this type on individual cuneate neurones are difficult to complete as stable recording conditions are required over quite long periods (1–2 h). One such detailed analysis is illustrated by the plot in Fig. 8 based on data obtained from cycle histograms constructed at six response levels (1–6) for both unconditioned (test) and conditioned responses. The dispersion (standard

deviation, on the ordinate) in the distribution of impulse activity within individual cycle histograms is plotted against response magnitude (abscissa) and reveals no consistent difference between unconditioned and conditioned responses.

On the assumption that frequency coding for cutaneous vibration depends on the phase-locking of impulse activity to a particular point of the vibratory wave form our observations on the thirteen cuneate neurones provide no convincing evidence for an alteration in the capacity of these neurones to code this information, at least at frequencies around 30 Hz, in the presence of conditioning inhibitory inputs.

DISCUSSION

Classes of dynamically sensitive tactile neurones

The functional properties of *dynamically sensitive* cuneate neurones indicate that considerable specificity exists in the linkage between primary afferent fibres and their target neurones within the dorsal column nuclei. Those responsive to high frequency cutaneous vibration appear to receive their afferent supply *exclusively* from the subcutaneous Pacinian corpuscles. Perl, Whitlock & Gentry (1962) and Gordon & Jukes (1964) earlier suggested that the vibration-sensitive neurones observed in their studies received Pacinian inputs.

Dynamically sensitive cuneate neurones responsive to low frequency vibration (10–50 Hz) applied to the glabrous skin presumably derive their input selectively from the Meissner corpuscles (Talbot *et al.* 1968; Jänig, 1971) and correspond to the rapidly adapting 'pad-sensitive' group studied in the gracile nucleus by Gordon & Jukes (1964) and in the medial lemniscus by Brown, Gordon & Kay (1974). Those sensitive to low frequency vibration with receptive fields confined to the hairy skin probably receive their input from afferents associated with hair follicle receptors, perhaps exclusively from the type G_2 quickly adapting alpha fibre (Brown & Iggo, 1967; Burgess *et al.* 1968) as its counterpart in the primate skin was sensitive to vibration at 20–40 Hz (Merzenich & Harrington, 1969). The type G_1 hair receptors are unresponsive to cutaneous vibration unless large amplitudes ($> 50\ \mu m$) are employed at high frequencies, e.g. 200 Hz (Burgess & Perl, 1973). Although the type D hair follicle receptors (Brown & Iggo, 1967) are highly sensitive to low frequency vibration (Merzenich & Harrington, 1969) they are innervated by the delta class of afferent fibres which do not project in the dorsal columns to neurones of the dorsal column nuclei (Wall, 1960; Brown, 1968; Petit & Burgess, 1968). Furthermore, no neurones were found within the cuneate nucleus in the present study that displayed the properties of the *delta* quickly adapting afferents

studied by Merzenich & Harrington (1969); namely, a broad tuning with about equal sensitivity to frequencies in the range from 5 to 100 Hz.

For cuneate neurones which responded to low frequency vibration and whose receptive fields included areas of both glabrous and hairy skin there was presumably a convergence from different receptor types as has been reported for a proportion of units in the gracile nucleus (Gordon & Jukes, 1964) and the medial lemniscus (Brown *et al.* 1974).

Inhibition of cuneate neurones

The present results are consistent with previous observations on the trigeminal relay of the somatosensory pathway that both slowly and rapidly adapting mechanosensitive neurones may be inhibited by afferent inputs (Carmody & Rowe, 1974). The absence of afferent-induced inhibition on slowly adapting neurones within the dorsal column nuclei in previous studies (Gordon & Jukes, 1964; Perl *et al.* 1962; Andersen *et al.* 1970) may have been due to the use of barbiturate anaesthesia.

The finding that inhibition of cuneate neurones was evoked by stimuli which excite dynamically sensitive tactile receptors but not by stimuli which activate slowly adapting tactile afferents is consistent with previous observations in both the cuneate and trigeminal nuclei (Andersen *et al.* 1970; Carmody & Rowe, 1974). However, by employing in the present study precisely controlled mechanical stimuli which permit the selective activation of different tactile receptor groups (Talbot *et al.* 1968; Merzenich & Harrington, 1969) we have demonstrated that the dominant and perhaps exclusive source of afferent-induced inhibition of cuneate neurones arises from Pacinian corpuscles, these being the only receptors activated by low amplitudes of cutaneous, sinusoidal vibration at 300 Hz. Although Andersen *et al.* (1970) suggested that the major part of the primary afferent depolarization and afferent inhibition seen in the cuneate nucleus was derived from rapidly adapting hair receptors they were careful to point out that their stimuli, blowing and brushing, were also capable of activating Pacinian corpuscles. Indeed, Burgess *et al.* (1968) observed such activation even when the stimulator engaged only the tips of the hairs. It was also found in the present study that cuneate neurones driven by Pacinian inputs could be readily activated by blowing on hairs.

The absence of inhibitory actions on cuneate neurones following steady indentation or low frequency vibration (20–40 Hz) at amplitudes of < 50–100 μm indicates that the receptor classes responsive to these forms of stimulation do not give rise to afferent-induced inhibition within the dorsal column nuclei, at least under the present experimental conditions. Although significant inhibition was found at high amplitudes (> 50–100 μm) of vibration at 30 Hz (see Fig. 5) this cannot necessarily be attributed

to these receptor classes as some Pacinian corpuscles will be activated by 30 Hz vibration at these high amplitudes (Talbot *et al.* 1968). We cannot be certain whether the *vibration-insensitive* hair follicle receptors (Merzenich & Harrington, 1969) contribute to afferent-induced inhibition, as stimuli which activate this group (movement across the receptive field) would also activate Pacinian afferent fibres.

Our conclusion that Pacinian inputs are the major source of afferent-induced inhibition of cuneate neurones is in accord with findings (Schmidt *et al.* 1967; Jänig *et al.* 1968) at the lumbar level of the spinal cord that Pacinian inputs were a major source of the primary afferent depolarization seen in mechanosensitive afferent fibres. The primary afferent depolarization at the lumbar level of the spinal cord was organized, however, in a specific manner with Pacinian inputs operating predominantly on phasically sensitive afferent fibres and a second system, arising from tonic or slowly adapting inputs operating selectively on slowly adapting units. In contrast, the inhibitory actions of the Pacinian inputs in the cuneate nucleus appear to be generalized, operating on all classes of 'tactile' neurones studied, whether slowly adapting or dynamically sensitive and whether driven from glabrous or hairy skin (see Figs. 3 and 4).

As Pacinian corpuscles are particularly responsive to rapidly changing mechanical stimuli the present observations suggest that when abrupt changes occur in the patterns of tactile stimulation the Pacinian inputs will initiate an inhibitory suppression of other channels for tactile inflow, thereby ensuring that information about the changing aspects of tactile inputs is given priority of access to higher centres.

The observation that cuneate neurones could be inhibited from within their excitatory receptive fields is consistent with the notion that the inhibitory fields for these neurones consist not merely of an area surrounding the excitatory field but also extend through the excitatory field (Gordon, 1973). Similar conclusions have been made for 'surround' inhibitory fields of retinal ganglion cells (Rodieck & Stone, 1965; Enroth-Cugell & Robson, 1966).

Influence of afferent-induced inhibition on coding by dynamically sensitive cuneate neurones

Dynamically sensitive cuneate neurones presumably contribute to tactile texture discrimination and other discriminations involving changing tactile stimulus parameters. However, in order to examine the capacity of these neurones to code sensory information it is necessary to use reproducible stimuli whose intensive and temporal parameters can be precisely controlled. The coding of information about the *intensity* of sinusoidal vibration at a particular frequency in the range below approximately

60 Hz may depend on *both* the average number of impulses discharged in individual neurones and the size of the neurone population responding to the vibratory stimulus (Johnson, 1974; Talbot *et al.* 1968). For individual cuneate neurones responding to low frequency vibration it was found that in the presence of afferent-induced inhibition there was a reduction in the slope of their stimulus-response relationship constructed for a series of different amplitudes at 30 Hz. Thus, there was a reduction in *gain*, defined as the ratio of output, or response increment to input increment (Jänig *et al.* 1968) as was earlier observed for the response of slowly adapting neurones which code information about steady stimulus intensity (Carmody & Rowe, 1974). The reduction in *gain* for these dynamically sensitive cuneate neurones was not accompanied by a change in their dynamic range.

For coding information about vibratory *frequency* the impulse trains of cuneate neurones will more accurately reflect the periodicity within the vibratory stimulus pattern when the activity is tightly phase-locked to a preferred point in the wave form of the applied vibratory pattern. In the present analysis the real phase relation is not known or relevant and therefore we place no importance on the value of the mean in the cycle histogram distributions (Fig. 7*C–E*). However, the standard deviation provides a measure of the dispersion of impulse activity around the preferred point of discharge in the sine wave cycle. The results indicate that systematic changes in the level of afferent-induced inhibition on cuneate neurones do not cause consistent shifts in the phase-locking of activity in response to 30 Hz vibration. This appears to contrast with observations in the visual system where lateral inhibition increases the extent of modulation of impulse activity of individual optic nerve fibres in response to a sinusoidally modulated light signal (Ratliff, Knight, Toyoda & Hartline 1967; Maffei, 1968).

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REFERENCES

ANDERSEN, P., ECCLES, J. C., SCHMIDT, R. F. & YOKOTA, T. (1964). Slow potential waves produced in the cuneate nucleus by cutaneous volleys and by cortical stimulation. *J. Neurophysiol.* **27**, 78–91.

ANDERSEN, P., ETHOLM, B. & GORDON, G. (1968). Presynaptic depolarization of dorsal column fibres by adequate stimulation. *J. Physiol.* **194**, 83–84*P*.

ANDERSEN, P., ETHOLM, B. & GORDON, G. (1970). Presynaptic and post-synaptic inhibition elicited in the cat's dorsal column nuclei by mechanical stimulation of the skin. *J. Physiol.* **210**, 433–455.

BROWN, A. G. (1968). Cutaneous afferent fibre collaterals in the dorsal columns of the cat. *Expl Brain Res.* **5**, 293–305.

BROWN, A. G., GORDON, G. & KAY, R. H. (1974). A study of single axons in the cat's medial lemniscus. *J. Physiol.* **236**, 225–246.

BROWN, A. G. & IGGO, A. (1967). A quantitative study of cutaneous receptors and afferent fibres in the cat and rabbit. *J. Physiol.* **193**, 707–733.

BURGESS, P. R. & PERL, E. R. (1973). Cutaneous mechanoreceptors and nociceptors. In *Handbook of Sensory Physiology*, vol. II, *Somatosensory System*, ed. IGGO, A. Berlin: Springer-Verlag.

BURGESS, P. R., PETIT, D. & WARREN, R. M. (1968). Receptor types in cat hairy skin supplied by myelinated fibres. *J. Neurophysiol.* **31**, 833–848.

CARMODY, J. & ROWE, M. (1974). Inhibition within the trigeminal nucleus induced by afferent inputs and its influence on stimulus coding by mechanosensitive neurones. *J. Physiol.* **243**, 195–210.

DARIAN-SMITH, I., ROWE, M. J. & SESSLE, B. J. (1968). 'Tactile' stimulus intensity: information transmission by relay neurons in different trigeminal nuclei. *Science, N.Y.* **160**, 791–794.

ENROTH-CUGELL, C. & ROBSON, J. G. (1966). The contrast sensitivity of retinal ganglion cells of the cat. *J. Physiol.* **187**, 517–552.

GORDON, G. (1973). The concept of relay nuclei. In *Handbook of Sensory Physiology*, vol. II, *Somatosensory System*, ed. IGGO, A. Berlin: Springer-Verlag.

GORDON, G. & JUKES, M. G. M. (1964). Dual organization of the exteroceptive components of the cat's gracile nucleus. *J. Physiol.* **173**, 263–290.

HUNT, C. C. (1961). On the nature of vibration receptors in the hind limb of the cat. *J. Physiol.* **155**, 175–186.

HUNT, C. C. & MCINTYRE, A. K. (1960). An analysis of fibre diameter and receptor characteristics of myelinated cutaneous afferent fibres in cats. *J. Physiol.* **153**, 99–112.

JÄNIG, W. (1971). Morphology of rapidly and slowly adapting mechanoreceptors in the hairless skin of the cat's hind foot. *Brain Res.* **28**, 217–232.

JÄNIG, W., SCHMIDT, R. F. & ZIMMERMANN, M. (1968). Two specific feedback pathways to the central afferent terminals of phasic and tonic mechanoreceptors. *Expl Brain Res.* **6**, 116–129.

JOHNSON, K. O. (1974). Reconstruction of population response to a vibratory stimulus in quickly adapting mechanoreceptive afferent fiber population innervating glabrous skin of the monkey. *J. Neurophysiol.* **37**, 48–72.

LEICHT, R., ROWE, M. J. & SCHMIDT, R. F. (1973). Cutaneous convergence on to the climbing fibre input to cerebellar Purkyně cells. *J. Physiol.* **228**, 601–618.

LYNN, B. (1971). The form and distribution of the receptive fields of Pacinian corpuscles found in and around the cat's large foot pad. *J. Physiol.* **217**, 755–771.

MAFFEI, L. (1968). Inhibitory and facilitatory interactions in retinal receptive fields. *Vision Res.* **8**, 1187–1194.

MERZENICH, M. M. & HARRINGTON, T. (1969). The sense of flutter-vibration evoked by stimulation of mechanosensitive afferents innervating the hairy skin of monkeys. *Expl Brain Res.* **9**, 236–260.

PERL, E. R., WHITLOCK, D. G. & GENTRY, J. R. (1962). Cutaneous projection to second-order neurons of the dorsal column system. *J. Neurophysiol.* **25**, 337–358.

PETIT, D. & BURGESS, P. R. (1968). Dorsal column projection of receptors in cat hairy skin supplied by myelinated fibres. *J. Neurophysiol.* **31**, 849–855.

RATLIFF, F., KNIGHT, B. W., TOYODA, J. & HARTLINE, H. K. (1967). Enhancement of flicker by lateral inhibition. *Science, N.Y.* **158**, 392–393.

RODIECK, R. & STONE, J. (1965). Analysis of receptive fields of cat retinal ganglion cells. *J. Neurophysiol.* **28**, 833–849.

ROWE, M. J. & CARMODY, J. J. (1970). Afferent inhibition over the response range of secondary trigeminal neurones. *Brain Res.* **18**, 371–374.

SATO, M. (1961). Response of Pacinian corpuscles to sinusoidal vibration. *J. Physiol.* **159**, 391–409.

SCHMIDT, R. F., SENGES, J. & ZIMMERMANN, M. (1967). Presynaptic depolarization of cutaneous mechanoreceptor afferents after mechanical skin stimulation. *Expl Brain Res.* **3**, 234–247.

TALBOT, W. H., DARIAN-SMITH, I., KORNHUBER, H. H. & MOUNTCASTLE, V. B. (1968). The sense of flutter-vibration: comparison of the human capacity with response patterns of mechanoreceptive afferents from the monkey hand. *J. Neurophysiol.* **31**, 301–334.

WALL, P. D. (1960). Two transmission systems for the skin senses. In *Sensory Communication*, ed. ROSENBLITH, W. A. New York: Wiley.