

UCSF

UC San Francisco Electronic Theses and Dissertations

Title

Modifications of Sml cortical neurons during a developing carrageenin-induced hyperalgesic inflammation

Permalink

<https://escholarship.org/uc/item/03x8s51v>

Author

Christian, Kirsten Kay

Publication Date

1990

Peer reviewed|Thesis/dissertation

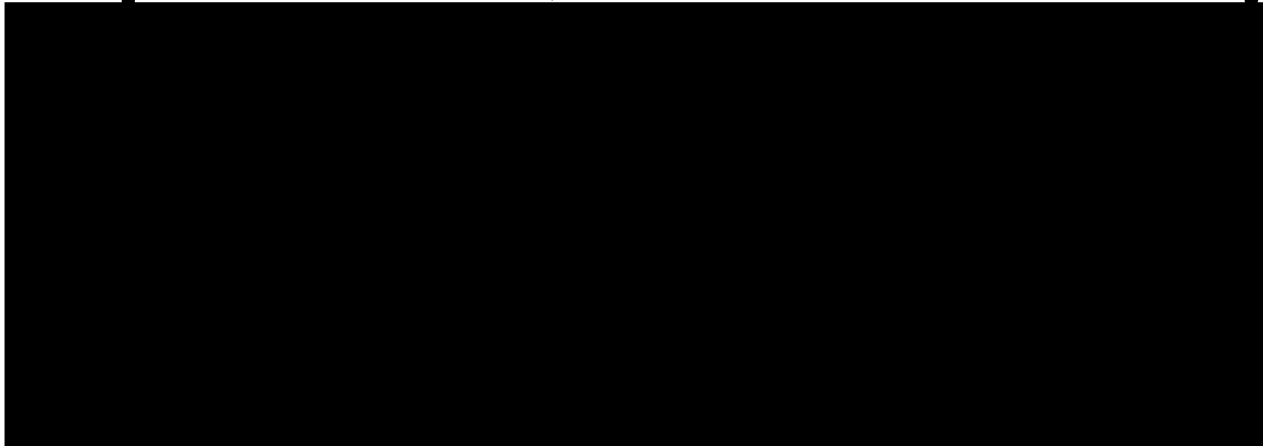
**MODIFICATIONS OF
SmI CORTICAL NEURONS
DURING A DEVELOPING
CARRAGEENIN-INDUCED
HYPERALGESIC INFLAMMATION**

**Kirsten Kay Christian
B.A. Pomona College, Claremont, CA**

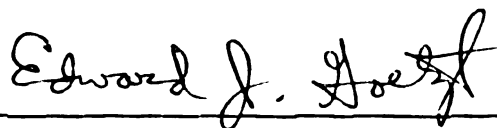
THESIS

**Submitted in partial satisfaction
of the requirements for the
degree of
M.D. with Thesis
at**

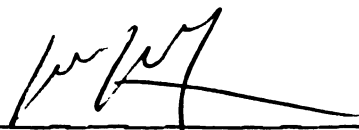
The University of California, San Francisco.



Thesis approval form for Kirsten K. Christian

A handwritten signature in cursive script, reading "Edward J. Goetzl", written above a horizontal line.

Dr. Edward Goetzl

A handwritten signature in cursive script, reading "Giselle Guilbaud", written above a horizontal line.

Dr. Giselle Guilbaud

A handwritten signature in cursive script, reading "H.J. Ralston, III", written above a horizontal line.

Dr. H.J. Ralston, III

Abstract:

The present electrophysiological study was designed to analyze the responsiveness of neurons in the first somatosensory cortex in rats during the development of a hyperalgesic carrageenin-induced inflammation. Neurons were characterized as being driven by noxious pinch (group 1), non-noxious light touch (group 2), or non-noxious articular movement (group 3). Responses and spontaneous activity of 22 (out of a total of 165) cells in these three categories were recorded up to 150 minutes after intraplantar injection of carrageenin. Group 1 cells showed a statistically significant increase in post-carrageenin responses and a moderate, but statistically insignificant increase in spontaneous activity. Group 2 cells, in contrast, displayed a very significantly increased spontaneous activity ($p < 0.001$ between 60 and 150 minutes after carrageenin). This increase contrasts sharply with the evolution of spontaneous activity in the ventrobasal nucleus of the thalamus after carrageenin. Group 2 cell responses displayed a general increase, but this increase was statistically significant only for cells with expanded receptive fields. Finally, group 3 cells showed no significant modifications in responses or spontaneous activity. The local anesthetic xylocaine depressed both responses and spontaneous activity when injected into the peripheral receptive field at approximately 60 minutes after carrageenin injection. This depressive effect indicates that neural rather than humoral mechanisms are responsible for the post-carrageenin modifications seen in the present study. The involvement of central mechanisms in the observed alterations of SmI cortical discharge during inflammation is discussed and mechanisms such as disinhibition of inhibitory synapses or recruitment of previously inactive synapses are proposed.

Table of Contents

Preliminary Pages	i-iv
<i>Title Page</i>	<i>i</i>
<i>Abstract</i>	<i>ii</i>
<i>Table of Contents</i>	<i>iii</i>
<i>List of Figures</i>	<i>iv</i>
Introduction	1
Methods	7
Results	11
Discussion	16
Figures	29-41
Bibliography	42

List of Figures

<u>Figure #</u>	<u>Illustrating</u>	<u>Page #</u>
1	ECoG Tracings	29
2	Dye Spot / Laminar Distribution	30
3	Laminar Distribution	31
4	Responses / Spontaneous Activity of Pinch and Light Touch Cells	32
5	Responses of One Noxious Neuron	33
6	Responses of One Neuron Driven by “ Light Touch”	34
7	Expanded Receptive Fields	35
8	Comparison of Responses with and without Expanded RFs	36
9	Comparison of Spontaneous Activity with and without Expanded RFs	37
10	Spontaneous Hyperactivity	38
11	Responses of Cells Driven by Non-Nociceptive Articular Movement	39
12	Comparison of Post-Carrageenin Spontaneous Activity in the Cortex and the VB	40
13	Spikes as seen in Oscilloscope	41

Introduction:

The study of pain is a rapidly expanding branch of the neurosciences. The literature using anatomical, pharmacological, behavioral and electrophysiological approaches to elucidate pain mechanisms and pathways is substantial and growing (1,2,3). Nevertheless, any study of pain encounters several obstacles. Possibly the most obvious problem is the ethical impossibility of conducting certain types of studies. Moreover, there are psychological factors for which no model exists. In addition, pain is not a uniform phenomenon, but rather a sensory and emotional experience manifested in many forms. Therefore, a model for the study of pain is both necessary and imperfect. The choice of an appropriate model is crucial.

Before considering the various models and techniques which constitute the history of pain research it seems wise to first distinguish between the notion of pain and that of nociception. The word "pain" is a purely psychological term defined as an unpleasant sensory and emotional experience associated with actual or potential tissue damage or described in terms of such damage (4). The word "nociception" was first used by Sherrington in 1906 to describe stimuli capable of damaging tissue (5). In Nicomachean Ethics, Aristotle was possibly the first to consider pain as a state or feeling of unpleasantness as opposed to a feeling of pleasure. Pain was not considered a "sense" by Aristotle who nevertheless proposed five senses describing qualities of external objects. The theory of the five senses inspired Johannes Muller in 1840 to propose five kinds of specific nerve "energies" or, in present terminology, sensory nerves corresponding to each sense. It was the development of modern electrophysiology which disproved Muller's theory by showing that electrical discharge of nerve fibers differs only in magnitude, frequency, and velocity (6), but his idea of sensory classification is reflected in modern classification of nociceptors into mechanical, thermal, and polymodal (7). Thus, the concept of pain, a purely psychological term has led to the concept of a nociceptor, a purely physiological term (4).

Initial investigations into the nature of pain often involved the study of animals in which lesions

similar to those seen in humans suffering from chronic pain had been introduced. More recently, various models for inflammatory pain have been developed. The most heavily studied model has been that of Freund's adjuvant-induced polyarthritis in rats. In this model a suspension of killed *Mycobacterium butyricum* is injected into the base of the tail inducing an immunological response similar to rheumatoid arthritis two weeks following the injection. This model has been studied using a variety of techniques including behavioral, biochemical, pharmacological and electrophysiological approaches. In particular, electrophysiological studies found that somatosensory neurons recorded in various parts of the nervous system were profoundly altered when compared to similar neurons in normal rats (8,9,10,11,12,13,14,15). Though this model has been quite successful for studying chronic pain mechanisms, Freund's adjuvant cannot be used to study acute inflammatory pain because several weeks are necessary for the development of the rheumatoid-like inflammation.

More recently local injection of the mucopolysaccharide carrageenin has been used as a model for acute inflammatory pain (12,16,17,18,19,20,21,22,23,24,25,26). This study represents the continuation of a series of studies which have used the carrageenin model to characterize signalling of nociceptive messages from the periphery to the CNS (16,19,20,21). Signalling of nociceptive messages follows an anatomical pathway from cutaneous nociceptors to the cortex. An external noxious stimulus in a normal intact system should cause the peripheral nociceptors to fire relative to the intensity and form of the stimulus (27,28). Two types of peripheral afferent fibers, A delta and C, carry noxious information (29). A delta fibers respond mainly to noxious mechanical stimuli whereas C fibers are polymodal and respond to mechanical, thermal or chemical stimuli (7,30). These fibers enter the spinal cord in the lateral division of the dorsal root and eventually form part of Lissauer's tract. A delta and C fibers then synapse upon neurons in laminae I, II, and V of the dorsal horn. Second order neurons transmit the noxious information through the anterolateral system. The anterolateral quadrant of the cord contains both descending and ascending tracts. The main ascending tracts are the spinothalamic, the spinoreticular and the spinotectal tracts (31). Second order neurons of these tracts synapse in the ventrobasal (VB) nucleus of the thalamus among other thalamic targets. Neurons in the ventrobasal complex project to the SI and SII regions of the cerebral cortex (32). It can therefore be expected that these cortical areas contain nociceptive cells. However, the role of the cerebral cortex in nociception

has been discussed and disputed since the beginning of the century.

Responses of somatosensory cortical neurons to mechanical skin stimulation and to movement of the joints have been well established (33,34,35,36). However, the role of the primary somatosensory cortex in processing of nociceptive information is still unresolved. In the beginning of the century arguments were generally against the participation of the cortex in nociception. In 1920 Head (37) studied humans with thalamic pain syndromes and concluded that the somatosensory cortex did not contain representations for noxious input. Nevertheless, he admitted that the cortex might play a role in localizing nociceptive stimuli. In 1968 Penfield and Rasmussen supported Head's conclusions after conducting experiments in which electrical stimulation of human somatosensory cortex rarely caused reports of pain (38). In 1969 White and Sweet reported that ablation of the somatosensory cortex seldom succeeded in reducing intractable pain (39). Nonetheless, numerous clinical and physiological reports contradict the above conclusions (40).

As early as 1915 Dejerine and Mouzon reported a syndrome in which cortical injury caused a loss of reaction to certain noxious stimuli in humans, whereas tactile sensation remained unchanged (41). In 1951 Marshall studied humans with cortical wounds and found that eleven of 18 patients showed loss in cutaneous pain appreciation (42). Furthermore, in 1952 Lewin and Philips reported that stimulation of the somatosensory cortex in humans suffering from intractable pain can produce verbalization of local pain sensations (43). Physiological evidence supports somatosensory cortical participation in nociceptive processing. Numerous studies of tooth pulp nerve fibers have been undertaken since these fibers represent the only pure afferent input transmitting exclusively nociceptive information. In 1957 Melzack demonstrated the existence of a thalamically-relayed afferent projection from the tooth pulp to the somatosensory cortex in the cat (44), and, in 1973, Shigenaga reported similar findings in the rat (45). Using comparable methods of tooth pulp stimulation in the cat, Vyklicky proposed that the somatosensory cortex differentially modifies the perception of noxious and other sensory stimuli (46). In 1975 Chatrian studied human cortical modifications during tooth pulp stimulation and suggested that the somatosensory cortex of man participates in the central processing of pain (47). More recently Kenshalo and Isensee conducted a series of electrophysiological experiments in monkeys in which

somatosensory cortical responses to noxious stimuli were recorded (48). They concluded that neurons with small receptive fields (RF) provide information about the localization, the intensity, and the temporal attributes of a noxious stimulus while cortical neurons with large RFs can signal only the occurrence and intensity of a noxious stimulation. Behavioral experiments in cats also support the forementioned clinical and physiological findings. For example, Durinyan et al found that stimulation of the second somatosensory cortex in the cat produced an "attention reaction" possibly reflecting a painful sensation (49). This body of clinical, physiological and behavioral evidence supports the role of the somatosensory cortex in nociception.

The forementioned description of a nociceptive pathway is but a bare outline. One must also consider the descending control systems. Signalling of nociceptive messages in the CNS is modulated by descending inhibitory systems which produce antinociception. Thus, higher centers of nervous control can modify sensory functions. For example, stimulation of the ventrobasal complex of the thalamus has been shown to have inhibitory effects on nociceptive signals projecting rostrally from dorsal horn neurons responding to peripheral noxious stimuli. Clinically this is reflected in the fact that intractable pain that doesn't respond to peripheral deafferentation has been successfully treated using ventrobasal thalamic stimulation (7). The ventrobasal complex projects heavily to the cortex and it therefore seems reasonable to anticipate similar findings in the cortex. Indeed, Zhang et al have demonstrated that the activity of STT neurons can be modulated by electrical stimulation of the sensory cortex, (50) and Owens et al have reported that electrical stimulation of sensory cortex increases the noxious threshold of spinothalamic tract cells (51).

The physiological interaction between carrageenin and the living organism can help clarify the resulting edema and hyperalgesia. The first three hours after intraplantar injection of carrageenin an edematous response (26) as well as hyperalgesia (52) are exhibited. This inflammatory development can be divided into two successive stages. The first stage consists of a nonphagocytic inflammatory response including all events occurring in the dermal and epidermal tissue up to 60 min after injection. This stage is followed by a phagocytic inflammatory response at about 60 min after injection of carrageenin. After 2 to 10 min an edema develops due to serotonin released by injured mast cells. Serotonin and histamines play a role in the hyperalgesia caused by carrageenin. For histamines, it was shown (24) that

the anti-histamine Thiazinamium, which acts only upon peripheral receptors, blocked the hyperalgesia but not the edema due to carrageenin. The development of edema from 10 to 50 minutes is postulated to result from osmotic forces as the edema during this interval is a function of the concentration of carrageenin (26).

At about 60 min after injection the phagocytic inflammatory response begins as a result of carrageenin cytotoxicity to the dermal endothelium. Neutrophils phagocytize carrageenin thereby destroying their phagocytic vacuoles and eventually release lysosomal enzymes into the dermal tissue. This results in release of prostaglandin intermediates increasing endothelial cell permeability and thereby increasing sensitivity of local nerve endings (26). After the initial phase during which histamine and serotonin are produced, kinins such as bradykinin are released at about 90 minutes. Bradykinin has been shown to produce algisia in peripheral fibers, the spinal cord, and supra-spinal regions (53,54,55,56). After approximately two hours prostaglandins are released. The action of carrageenin continues for several days, but for our study the above description of the first few hours is sufficient. Thus injection of carrageenin has two major effects, that of edema and that of hyperalgesia. The questions addressed by this study were based more heavily upon the hyperalgesic than the edematous effects of carrageenin during the acute inflammatory period.

The principle techniques used to study the forementioned models are behavioral, anatomical, and electrophysiological. The electrophysiological techniques employed in this study stem from DuBois-Reymond, who, in 1841, demonstrated the resting and the action potential by stimulating and recording neuronal activity with a galvanometer. In 1922 Erlanger and Gasser invented the cathode-ray oscilloscope. In 1939 Hodgkin and Huxley used longitudinal microelectrodes to measure membrane potentials, and in 1940 the field was described in depth by H. Schafer in Elektrophysiologie (quoted in 57). Both extra and intra-cellular electrode techniques were developed and experiments were carried out in various areas of the central nervous system to discover a variety of cellular discharges such as the excitatory and inhibitory postsynaptic potentials found in Betz cells of the cerebral cortex by Phillips in 1956 (58). Macroelectrodes were gradually replaced by microelectrodes until it became possible to record the activity of a single neuron with an extracellular microelectrode as done in this study (57).

Following sub-cutaneous, intra-muscular, or intra-articular carrageenin injection peripheral

sensitization is manifested in several ways. Experimenters have found a decrease in the activation threshold of fine afferent fibers for both thermal and mechanical noxious stimuli as well as a significant spontaneous activity in numerous cutaneous afferent fibers of both muscular and articular origin (12,59). In the CNS at the level of the dorsal horn of the spinal cord and the ventrobasal complex of the thalamus researchers have found a progressive increase in neuronal responses produced by nociceptive stimuli. In addition, behavioral and electrophysiological studies (12,16,22,60,61) have often shown the appearance of responses on the contralateral side of the recording site non-existent before carrageenin. These receptive fields showed an increasingly more intense response for the same stimulation. For example, in this study we at times observed an increase in neuronal responses for both the injected and the non-injected paw. This last observation implies the interaction of central mechanisms in the hyperalgesia caused by carrageenin.

This study was inspired by the forementioned investigations concerning the transmission of nociceptive messages in peripheral nerves, the dorsal horn of the spinal cord and especially in the ventrobasal complex of the thalamus using the carrageenin model. In addition, studies concerning characterization of the normal rat cortex especially with regard to integration of nociceptive messages from the periphery (62,63) motivated the study of the cortex under pathological conditions. The SmI somatosensory cortex has been studied much less heavily than the other portions of the pathway. In 1958 and 1971 Woolsey and Welker (64,65), respectively, showed that the rat SmI cortex is organized in a very precise somatotopic manner, receiving input from the contralateral half of the body. Rat SmI cortical neurons responding to stimulation of cutaneous peripheral receptive fields have been characterized by numerous groups (66,67,68,69,70). More recently, in a systematic study, Lamour et. al. (62,63) described several categories of SmI cortical neurons driven by distinct modes of stimulation. The three most prevalent modes of stimulation were 1) light non-nociceptive (brushing etc) 2) deep non-nociceptive (often articular) and 3) nociceptive (pinch). The present study characterized units using these same stimulation modes. Three principal findings resulted from the Lamour study. First, a relatively substantial number of cortical neurons receive noxious inputs. Second, such noxious input is somatotopically organized and, third, neurons driven by noxious inputs are able to encode stimulus parameters (63). In a second paper Lamour et. al. examined the laminar and columnar organization

of noxious and non-noxious inputs. In this paper they suggested that the somatotopic organization of noxious and non-noxious receptive fields are somehow independent and that noxious receptive fields have an independent somatotopic organization. (62).

This study is based on ideas and information from these previous studies, but no electrophysiological study has as yet addressed the question of cortical characterization using the carrageenin model for a developing inflammation.

Methods:

Surgical preparation :

The surgical preparation used in this study was identical to that used in previous studies performed both at the thalamic and the cortical level (62,63,71). During the surgical procedure rats were anesthetized using halothane at 2.5% in a mixture of one third O₂ and two thirds N₂O. A tracheotomy was performed to facilitate artificial ventilation using a Palmer pump, and the rats were paralyzed with an intra-peritoneal infusion of pancuronium (Pavulon, 10mg/kg initially and then in small doses over time to reach 25-30mg/kg i.p.) to minimize muscular movement and to maximize the probability of maintaining recording of the cell as long as possible. The frequency and volume of the pump were set in order to maintain a normal acid-base equilibrium as described by Freminet (72). End-expiratory CO₂ (4.5-5.5%) and cardiac rhythm were constantly registered. The animals were fixed in a stereotaxic apparatus to avoid any movement and the sutures of the bregma were identified using moderate magnification. The bregma was later used as a reference point for electrode recording. After the skin was reflected, a dental burr was used to drill two small longitudinal holes for the silver ball electrodes of the ECoG. In order to ensure that the cortex was not damaged by heat from the drilling procedure, drilling periods were interrupted by short intervals during which ringer was sprayed onto the drilling site. The ECoG was thus recorded from the dura mater. At 2.5% halothane anesthesia the ECoG was composed of slow, high voltage activity (1.5-2 Hz) accompanied by periods of depressed activity and

slow spikes.

The surgical procedure consisted of a craniotomy removing the minimal area covering the SmI hindlimb representation according to the coordinates of Hall and Lindholm (73). Following this procedure the exposed dura mater was covered with warm saline and surgical sponge to minimize edema and dehydration. The anesthesia was lowered to 0.5% and the ECoG was monitored to surveil the animal's recovery to a lightly anesthetized state (generally lasting between 45 and 60 minutes). The ECoG representing this state consisted of theta monomorphic waves of 4-5 Hz and 100-120 μ V associated with sparse spindles of 12-14 Hz. The ECoG was carefully monitored throughout the experiment. At various periods during the experiment the tail was pinched and the ECoG was monitored for any signs of arousal (see figure 1). When the ECoG showed no signs of arousal and was characterized by the more rapid, less depressed, activity of the lightly anesthetized state described above, the dura mater covering the SmI representation of the hind limb was removed. This was done under microscopic control to ensure the least possible damage to the exposed cortex.

Recording procedures :

Extracellular unit potentials were recorded with glass microelectrodes filled with a mixture of 2% Pontamine sky blue in 1M NaCl. The tips were then carefully brushed on lense paper to yield a tip diameter of about 1 μ m as judged by the impedance of 10 to 15 megohms. The site of penetration of the microelectrode was 2.5 to 2.7 mm lateral and -0.5 to -0.7 mm posterior to the bregma in accordance with the coordinates of Hall and Lindholm (73) for the hindpaw representation of SmI. The variance in recording location resulted from a variety of difficulties. Often arteries covered the ideal penetration site. Because of the extreme fragility of microelectrodes as well as risk of hemorrhage it was not possible to penetrate the vessel, and the recording site was thus diverted.

A minimal current was passed through the microelectrode during one or two seconds in order to reestablish the original impedance if abnormally increased during the penetration. When a single unit was isolated its activity was amplified and displayed on an oscilloscope. The frequency of discharge was displayed on a rectilinear strip chart recorder. For the 80 rats used an average of three penetrations

were made per rat.

Characterisation of receptive fields :

As the microelectrode penetrated the cortex continuous non- noxious stimulation of the hindpaw was used as a search stimulus. The unit's peripheral receptive field was then characterized. The stimulus was classified as either noxious or non-noxious. Few cortical cells were found to have convergent properties (see results). A stimulus was considered noxious when it was capable of giving a noxious sensation to the skin of the investigator. The non- noxious stimuli were further classified as either "light touch cells" which were stimulated by either brushing, stroking or gentle tapping or as "articular cells" which responded to flexion or extension of the articulations in the hindpaw. The noxious stimulation was a strong pinch often accompanied by a flexed or extended position of the articulations. The receptive field was then delimited and noted in order to determine possible RF expansions after carrageenin injection. Finally, temporal characteristics of the responses such as phasic, tonic or "on-off" responses were noted.

Once the unit was characterized and a minimum of 3 control stimulations were recorded the hindpaw was injected with 0.2 ml of a 1% solution of carrageenin in saline (Satia Laboratory Paris). The site of injection was generally in the center of the plantar surface such that the resulting inflammation necessarily involved the RF. For those neurons responding to light touch and showing a quite distal receptive field, the site of injection was slightly more distal at the base of the digit containing the RF. Stimuli were never applied earlier than 5 minutes after carrageenin administration so as to eliminate confusion of responses to stimuli and discharge due to irritation of the injecting process. In general the receptive field was stimulated every 5 to 10 minutes depending on the type of stimulation. If the stimulation was noxious, the receptive field was stimulated less often in order to avoid tissue damage. Individual stimulations lasted 15 seconds. Vascularization of the peripheral cutaneous receptive field was monitored by observing the color of the extremities and their recovery to a pre-stimulus state just after stimulation. Between stimulations the injected and non-injected hindpaws

were stroked and brushed to search for expansions in the receptive fields, but this search process was limited to a minimal contact time. Cellular activity during these search periods was not considered to be spontaneous, and was thus not considered in the statistical analysis for spontaneous activity.

If cells displayed a tendency toward an increased spontaneous activity then 0.05ml of the local anesthetic, Xylocaine, (lidocaine-epinephrine hydrochlorides, respectively 1 and 0.0025% in saline) was administered into the plantar surface of the inflamed paw. Xylocaine was administered after the cell had been observed for at least 60 minutes following carrageenin injection. Xylocaine's anesthetic effect was judged by a depression in responses to stimulation of the receptive field every three minutes, generally until 25 min after xylocaine administration. This procedure was carried out in order to determine whether the increased spontaneous activity was of peripheral or central origin.

Histological identification of recording site :

In order to identify the cortical layer in which the recorded unit resided, the final recording site of each fruitful penetration was marked by ejecting dye iontophoretically from the microelectrode (20-50 uA for 30 minutes). Finally the animal was sacrificed with an overdose of sodium pentobarbital and perfused through the aorta with saline followed by a solution of 10% formaldehyde in saline. The brain was then removed and frozen. 100 um sections of the brain were cut on a freezing microtome and then stained with safranin. A camera lucida was used to reconstruct the cortical layer in which the cell resided and to specify the depth and laterality of the recording site (see figure 2). From most superficial to deepest, cortical layers were numbered I, II-III, IV, Va, Vb, VIa, VIb. The presence of neuronal aggregates in layer IV in the histological reconstructions was considered as evidence for the correct positioning of the electrode within the SmI (74). However, as elicited by Dykes and Lamour (75,76), several sources of uncertainty result from this localisation method. The actual location of the cell was based upon an interpolated distance and had an uncertainty that was determined by the size of the dye spot (generally 100-150 um diameter) and the ambiguity of the location of the electrode relative to cells studied. The ambiguity of the cellular location arises from the fact that extracellular recordings allow an action potential to be recorded for a distance of at least 100 um on each side of the point of

maximum amplitude. This produces an uncertainty of plus or minus 100 μm and there is no clear proof that the electrode was recording from the cell body rather than a process such as the apical dendrite some distance from the actual location of the cell body (75). Nevertheless, the forementioned technique was considered adequately precise for localization of cells within a cortical layer.

Data analysis :

In order to facilitate statistical analysis, data for responses were grouped together into three periods of 5-30 minutes, 30-60 minutes and 60-150 minutes after carrageenin injection. These periods were chosen because they reflect hyperalgesic development and evolution as demonstrated by data from previous behavioral experiments (22). For each cell, mean values for responses during these periods were calculated. These mean values were then expressed individually as a percent of the control value of their respective cells, and used to calculate a global average for each period. These results were treated using a non-parametric test of variance (Kruskal-Wallis), and further analyzed by the Mann-Whitney test. Data for spontaneous activity were processed in a similar manner, but cellular discharge quantified into 60 second intervals was used in lieu of response data.

Results :

A total of 165 neurons (124 penetrations) were recorded in 66 animals during this study. Only data from experiments in which cells were recorded for a minimum of 40 minutes after carrageenin administration ($n=22$) were used for the statistical analysis. Of these 22 cells, $n=8$ responded to noxious stimuli (hindpaw pinch) and $n=14$ responded exclusively to non-noxious stimuli ($n=8$ light touch and $n=6$ articular movement). At 60 minutes after injection the medio-plantar circumference of the injected and the non-injected paws was 33.4 ± 0.6 and 22.4 ± 0.7 respectively showing a significant difference ($p<0.001$).

maximum amplitude. This produces an uncertainty of plus or minus 100 μm and there is no clear proof that the electrode was recording from the cell body rather than a process such as the apical dendrite some distance from the actual location of the cell body (75). Nevertheless, the forementioned technique was considered adequately precise for localization of cells within a cortical layer.

Data analysis :

In order to facilitate statistical analysis, data for responses were grouped together into three periods of 5-30 minutes, 30-60 minutes and 60-150 minutes after carrageenin injection. These periods were chosen because they reflect hyperalgesic development and evolution as demonstrated by data from previous behavioral experiments (22). For each cell, mean values for responses during these periods were calculated. These mean values were then expressed individually as a percent of the control value of their respective cells, and used to calculate a global average for each period. These results were treated using a non-parametric test of variance (Kruskal-Wallis), and further analyzed by the Mann-Whitney test. Data for spontaneous activity were processed in a similar manner, but cellular discharge quantified into 60 second intervals was used in lieu of response data.

Results :

A total of 165 neurons (124 penetrations) were recorded in 66 animals during this study. Only data from experiments in which cells were recorded for a minimum of 40 minutes after carrageenin administration ($n=22$) were used for the statistical analysis. Of these 22 cells, $n=8$ responded to noxious stimuli (hindpaw pinch) and $n=14$ responded exclusively to non-noxious stimuli ($n=8$ light touch and $n=6$ articular movement). At 60 minutes after injection the medio-plantar circumference of the injected and the non-injected paws was 33.4 ± 0.6 and 22.4 ± 0.7 respectively showing a significant difference ($p<0.001$).

Placement of neurons in cortical layers

A total of 67 cells (45 rats) were histologically reconstructed (see figure 3). Cells responding to non- nociceptive stimuli (n=31 light touch, n=9 articular movement) were found in layers II to VI. Cells driven by noxious stimuli were localized only in the deepest layers, Va to VIb. Regardless of the stimulation mode, the greatest number of cells were always found in layer Vb.

Neurons driven by noxious stimuli

Pre-carrageenin responses-

Among the 8 neurons used, 7 were driven exclusively by noxious hindpaw pinching and 1 was also responsive to light touch. All control responses were well-isolated and showed no or very little post-discharge. Of these 8 cells, 3 showed receptive fields only on the hindpaw contralateral to the recording site (mean amplitude of responses= 151 ± 63 spikes and mean duration= 13.7 ± 1.9 sec). Five showed bilateral receptive fields (mean amplitude and duration of responses for hindpaws contralateral and ipsilateral to the recording site were 170.3 ± 53 spikes per 20.0 ± 4.9 sec and 97.0 ± 17 spikes per 17.0 ± 1.3 sec respectively; comparison of two samples insignificant, $p > 0.05$ by Wilcoxon signed rank test). Sensitivity to noxious thermal stimulation (determined using hindpaw immersion in a 50° C water bath) was found for all tested cells (5 of 8 cells).

Post-carrageenin responses-

Mean responses up to 150 minutes after carrageenin injection are shown in figure 4. For the injected hindpaw, response intensity increased gradually over the three testing intervals reaching $221 \pm 21\%$ of the control value ($p \leq 0.001$) during the last period (60-150 minutes). Response duration also

increased gradually ($163 \pm 29\%$ of control value for final period), but in a statistically insignificant manner. In fact, only 1 of the 8 cells developed a clear post-discharge (i.e. duration greater than 30 sec). Carrageenin administration increased response intensity only for noxious and not light tactile stimuli of the forementioned unit exhibiting a wide dynamic range.

One of the 3 unilaterally responsive units developed a bilateral receptive field 40 minutes after carrageenin injection.

For the 5 units showing initially bilateral receptive fields, post-carrageenin response intensities for the non-injected hindpaw stimulation did not differ significantly from pre-carrageenin values, but showed a general tendency toward increased responses (see figure 5).

Three units with bilateral receptive fields responded to noxious stimulation by inhibition of their background activity. This inhibition occurred either during ($n=2$) or immediately after ($n=1$) the stimulation. Inhibitions appeared no earlier than 10 minutes after carrageenin injection and lasted up to 1 minute.

Spontaneous activity-

All unitary background activity during which there was no external stimulation was considered to be spontaneous. The average spontaneous cellular discharge before carrageenin was 6.0 ± 3.2 Hz for $n=8$ neurons. Between 30 and 60 minutes after carrageenin administration spontaneous activity reached a maximum of 363% of the control value, but this increase was not statistically significant ($p>0.05$). Nevertheless, as early as 30 minutes after carrageenin injection, three cells displayed intense periods of spontaneous hyperactivity (average frequency = 14 Hz) lasting from 5 to 30 minutes. For one of these cells xylocaine was administered, as described in the methods, 100 minutes after carrageenin during a period of spontaneous hyperactivity. Cellular discharge diminished to zero at 8 minutes and gradually recovered to pre-xylocaine levels at 20 minutes.

Neurons driven by light touch

Pre-carrageenin responses-

A total of $n=8$ units driven exclusively by light touch applied to the contralateral hindpaw were used. Receptive fields included one or two digital pads ($n=2$), one distal phalanx ($n=4$), or the entire finger ($n=2$). Control responses rhythmically followed the stimulation frequency and remained stable without fading for the 15 second stimulation period. Furthermore, no post-discharge was observed. Average response intensity was 135.5 ± 12.8 spikes.

Post-carrageenin responses-

Mean responses up to 150 minutes after carrageenin injection are shown in figure 4. Response intensity increased systematically over the three testing intervals reaching $243 \pm 83\%$ of the control value during the last period. However, in contrast to the results from noxious stimuli, this increase was not statistically significant (see figure 6). As soon as 15 minutes after carrageenin a distinct expansion of the receptive field was observed for $n=5$ units. The original outline of receptive fields appeared to enlarge, spreading to neighboring zones (see figure 7). Interestingly, when the forementioned $n=5$ units were isolated and reanalyzed, they showed a statistically significant increase in responses, in sharp contrast to the stability of the responses of the three cells without expanded RF (see figure 8). It was also noted that the largest expansions in RF were observed in rats whose injected hindpaw showed the largest medio-plantar edema at 60 minutes. Initial RFs were included in the edematous zone in 4/5 cells with expanded RFs and in 1/3 cells without expanded RFs.

Spontaneous activity-

For the eight cells studied, the average spontaneous discharge before carrageenin was 1.12 ± 0.28

Hz. After carrageenin administration, discharge increased in a statistically significant manner (figure 4). Maximum spontaneous activity was found between 60 and 150 minutes ($611.8 \pm 176\%$ of control values, $p \leq 0.001$). When units displaying expansions in their RFs were separated from those displaying no expansions, the evolution of spontaneous discharge was clearly distinct. When no RF expansion existed, spontaneous activity increased only slightly and in a statistically insignificant manner. In contrast, the statistically significant increase in spontaneous activity, seen when all eight cells are grouped together, persists for the five cells exhibiting expanded RFs (figure 9). The largest increases in spontaneous activity were significantly associated with the degree of RF expansion ($p \leq 0.001$). Though all cells displayed an increased spontaneous activity, the largest increase in spontaneous activity was observed in units in which the initial receptive field was included in the edematous zone. One cell, (see figure 10) discarded from previous statistical analysis, manifested paroxystic firing which appeared suddenly at 32 after carrageenin and lasted until xylocaine administration at 60 minutes. Immediately after xylocaine, cellular firing was abolished and reappeared at 34 minutes (10 minutes after recovery of evoked responses).

Neurons driven by articular movement

Pre-carrageenin responses-

A total of $n=6$ cells driven by articular movement of the ankle ($n=3$) or phalanges ($n=3$) were used. Control responses were quite phasic and often displayed an "on-off" character corresponding to the first and last stimulations of the hindpaw. Control responses were extremely variable ranging from 11.7 to 342.5 spikes per 15 seconds with an average of 102 ± 50 spikes.

Post-carrageenin responses-

Post-carrageenin responses for the 6 "articular" cells used are given in figure 11. Only 2 cells were recorded more than 60 minutes after carrageenin. These 2 cells had the smallest control values and

exhibited large increases in their responses (450% and 2800%). The 4 remaining cells, which were followed up to 60 minutes, maintained relatively constant responses. One of these 4 cells displayed an elevated control discharge relative to the other cells (342.5 spikes per 15 sec) and exhibited increasingly longer post-discharges (duration=1 minute for control, and 5 minutes half an hour post-carrageenin).

Spontaneous activity-

The average spontaneous cellular discharge before carrageenin was 1.62 ± 0.8 Hz for $n=6$ neurons. This unitary background activity did not evolve in a homogeneous manner and could not be correlated with modifications in responses. Three cells maintained their pre-carrageenin discharge levels, one increased moderately (230% of control value), and two increased dramatically (4000% and 7500% of respective control values). Spontaneous discharge of these last two cells manifested itself in strikingly different patterns. One cell showed a constantly increasing activity which reached a maximum discharge at the end of the recording period. Spontaneous activity of the other cell consisted of intense periods of hyperactivity lasting up to 10 minutes, as described in previous units.

Discussion :

The above results describe modifications of SmI cortical neurons during a developing acute inflammation. Comparison of cortical neurons with thalamic, spinal and peripheral units under the same acute inflammatory conditions can clarify the role of cortical neurons in pain sensations. Furthermore, mono- and polyarthritic rats provide a model for chronic inflammation and as such may illustrate more profound modifications which might be seen if carrageenin's inflammatory effects were prolonged. The use of these inflammatory models clarifies and supports a cortical role in nociception.

The goal of this discussion is to analyze the role played by the first somatosensory cortex in processing peripheral information due to an acute inflammation. In an attempt to simplify the

presentation of previous findings relevant to the present study the discussion is categorized according to stimulation mode used to activate neurons. Because peripheral afferent fibers project via the spinal cord to the VB and thus to the cortex, discussion of each stimulation mode shall follow this anatomical route to the cortex. Observations along the same anatomical path in the mono- and polyarthritic rat will then be discussed in order to compare the acute findings of the present study with results from chronic inflammation. Finally, some of the problems posed by drawing conclusions about alert states from anesthetized animals will be discussed. Nevertheless, the rats used in this study were "normal" prior to carrageenin administration. Thus, it is logical to begin with a comparison of control observations with findings in the normal rat.

Comparison of Controls With Normal Rats

Characteristics of cortical cells responding to both noxious and non-noxious stimuli prior to carrageenin administration were relatively similar to previous studies in normal rats conducted under the same anesthetic and electrophysiological conditions (62,63). For example, Lamour et al found that cells responding to non-noxious stimuli were generally characterized by small receptive fields whereas noxious stimuli were elicited from larger areas of the ankle and hindpaw. Cells responding to articular movement were less common (7.3%) than cells responding to light touch or pinch. Histologically, this study's results are similar to those of Lamour et al. (62,63) who reported that neurons driven by non-noxious cutaneous stimuli were found in layers II to V and were rare in layer VI while nociceptive neurons were almost exclusively found in layer V and VI. Lamour et. al. also claimed that receptive field size was a function of the laminar position of the neurons such that receptive fields were generally small in layer IV, larger in layer II-III and V and even larger in layer VI. Indeed, the largest receptive fields for cells responding to non-noxious stimuli in this study were situated in layer VI and all neurons in layer IV responding to light tactile stimuli had relatively smaller receptive fields. It is also interesting to note that our results are in accordance with Lamour et. al. in that no layer IV unit was driven by noxious stimuli. This is curious because the VB complex, containing a large number of neurons responding to noxious stimuli, relays information to the cortex via thalamo- cortical axons which

terminate heavily in layer IV.

Post-Carrageenin Modifications of Neurons Responding to Nociceptive Stimuli

Much as in previous studies in different parts of the nervous system our results confirmed an increased cortical responsiveness to noxious stimulation subsequent to administration of inflammatory and algescic substances. The effect of carrageenin on mechano-heat sensitive C-fibers in rats was investigated by Kocher et. al. using subcutaneously injected carrageenin in the area innervated by the saphenous nerve (23). They found an enhanced responsiveness and sensitization to heat stimulation. In addition, C-fibers receiving information from the inflamed area exhibited spontaneous activity and increased mechanical thresholds. In the present cortical study no change was seen in the mechanical threshold. None of the units located outside of the inflamed area displayed the forementioned sensitization.

Relatively few authors have directly characterized the changes that occur in nociceptive dorsal horn neurons during inflammation. Existing studies (54) have demonstrated that properties of responses, spontaneous discharge and receptive fields of dorsal horn neurons can change after injection of algescic substances. In an electrophysiological study Haley et. al. used a bradykinin B2 receptor antagonist to reduce cellular discharge caused by formalin injection into the receptive field of nociceptive rat dorsal horn cells (55). This study provides electrophysiological evidence for the role of bradykinin, a product of the carrageenin exudate, in nociception. Recently, modifications in cat dorsal horn neurons responding to nociceptive muscular stimulation subsequent to injection of bradykinin in their peripheral receptive fields were investigated by Hoheisel and Mense (77). After bradykinin they found expanded receptive fields, a lowering of mechanical threshold and the appearance of new RFs. In addition, the proportion of cells with both deep and cutaneous input and of those having background activity rose. Dorsal horn neurons thus seem rather similar to peripheral fibers in modifications due to various algescic substances.

The ventrobasal nucleus of the thalamus receives noxious information directly from the spinothalamic tract as well as indirectly via the spinoreticulothalamic tract (7,32). Participation of the

VB in nociception during inflammatory pain has been investigated using carrageenin (12,16,19,20,21). Guilbaud et. al. examined modifications of ventrobasal thalamic cells at different stages of carrageenin-produced inflammation (20). Results in the VB were similar to the present findings in the cortex for neurons driven by noxious stimuli. Thalamic responses were significantly increased to 215% of control values at one hour after carrageenin as compared to 221% in the cortex. In addition, spontaneous activity increased and expansions of the RF were noted for many of these nociceptive cells.

Post-Carrageenin Modifications of Cells Responding to Nonnociceptive Cutaneous Stimuli

Our results for light tactile stimuli differ substantially from studies in lower regions of the nervous system. Not only did we find increased responses to such stimuli, but a significantly increased spontaneous discharge not otherwise observed in the CNS or the PNS. Though thick myelinated fibers do not convey nociceptive information, they do transmit sensory stimuli to the SmI cortex (78). Modifications in these fast conducting fibers can therefore clarify our findings in the SmI cortex. Most studies investigating thick myelinated fibers involve the introduction of algogenic substances at the periphery. Fjallbrant and Iggo (79) studied the specificity of chemical excitants for different fiber types using close arterial injection of bradykinin and serotonin. They found that 5-HT and BKN are not specific for fibers and excite both nonnociceptive and nociceptive fiber types. Beck and Handwerker (53) carried out similar studies in order to elucidate the role of different fibers in the initiation of pain. They found that bradykinin and serotonin, products of carrageenin inflammation, excite fast fibers as well as non-myelinated nociceptive fibers. Indeed, Beck and Handwerker found that the differences between myelinated and unmyelinated fibers were not significant. Nonetheless, one must note that rapidly adapting myelinated fibers were excited by neither serotonin nor bradykinin. Only slowly adapting myelinated fibers were excited in about 30 to 50% of the cells tested. One can therefore conclude that signalling of light tactile stimuli transmitted via rapidly adapting myelinated fibers should not be modified by the algescic substances introduced by carrageenin administration.

Relatively few studies of the modifications of nonnociceptive dorsal horn cells exist. However, in 1972 Besson et. al. published a study of modifications of dorsal horn cell activity after intra-arterial

injection of bradykinin in the spinal cat (54). The cells of lamina IV were nonnociceptive, activated by tactile stimulation of low intensity from a small peripheral field. Only 16% of the lamina IV cells studied showed variations in their activity. The finding that these cells were not modified by bradykinin injection demonstrates that spinal and cortical processing of light touch differ.

Responsiveness of cells responding to nonnociceptive information, projected from the spinal cord to the thalamus, show the same lack of modification as the dorsal horn. In numerous studies, ventrobasal neurons driven by non-noxious stimuli such as light touch never showed an increase in responses nor any significant changes in the size of their receptive fields (12,16,19,20,21). Our cortical results differ from these findings in the ventrobasal thalamus in that we observed a marked increase in responses to light touch as well as a significant rise in spontaneous discharge. Furthermore, more than half of the cells studied which responded to light touch displayed an expanded receptive field after carrageenin injection. These observations strongly suggest that cortical processing of non-nociceptive somatic stimuli differs from signalling of this information via peripheral fibers and the thalamus. Thus our cortical results could demonstrate a cortical role in tactile allodynia (a state in which light tactile stimulus is perceived as painful).

In order to compare spontaneous activity in the VB and the cortex, data from previous experiments (19,20) with identical methods as the present study but located within the VB were used to calculate VB spontaneous activity. The eight experiments exploited involved only cells driven by light tactile stimuli. Exactly the same statistical analysis was used for this data set in order to ensure that the only variable was the location of cells recorded in the CNS. Spontaneous activity in the VB did not show a statistically significant increase and, in fact, increased only slightly. This is in sharp contrast to results of the present study showing that spontaneous discharge of cortical cells increases quite significantly as early as the first half hour after carrageenin administration. Using a parametric test (ANOVA), data from the VB were compared with data from the cortex for the same periods post-carrageenin. The two samples were highly significantly different ($p=0.008$)(see figure 12). The possibility that this increased spontaneous discharge results from cellular irritation is refuted by suppression and later recovery of both responses and spontaneous discharge after local anesthetic administration. This contrast between cortical and thalamic post-carrageenin modifications indicates that peripheral information is processed

differently in cortical cells than in the VB. This could be explained by the disinhibition of inhibitory cortical controls or, as proposed in the dorsal horn for noxious stimuli, by the induction of previously non-functional synapses (54).

Post-Carrageenin Modifications in Cells Responding to Nonnociceptive Articular Stimuli

Few authors talk about post-carrageenin modifications in cells responding to nonnociceptive articular stimuli. In general, however, our findings of no significant increase in responses agree with studies in other parts of the nervous system. Peripheral afferent fibers responding to articular movement have been examined under acute inflammatory conditions and findings demonstrate enhanced sensitivity. For example, peripheral afferent fibers with mechanosensitive receptive fields on medial and anteromedial regions of the cat knee joint were excited by small doses of bradykinin, a product of the carrageenin exudate (56). Kanaka et. al. concluded from these findings that all fine articular afferent units with local mechanosensitive receptive fields are also sensitive to bradykinin and possibly to a wide variety of other endogenous chemical substances. In another experiment using the cat knee joint Schaible and Schmidt investigated the effects of a developing arthritis including the time course of modifications on low and high-threshold units and on afferents that were non-receptive in the normal joint (60). They concluded that an acute experimental arthritis yields enhanced mechanosensitivity in nonnociceptive, nociceptive and initially unresponsive articular afferents. One explanation for these findings is that there exists a "special class of nociceptive afferents requiring manifest tissue damage for suprathreshold excitation (60)." As suggested by Schaible and Schmidt, recruitment of these special nociceptive afferents might influence central processing of nociceptive discharge from inflamed joints. Our results for articular movement are difficult to compare to these peripheral findings because articular movement was always nonnociceptive and cells were recorded for much shorter periods. Nevertheless, 1 of the 6 articular cells presented in our study showed the same pattern seen in 3 of the 16 similar cells illustrated in Schaible and Schmidt. This pattern consisted of strong initial responses in the control period and subsequently unchanged reactions. Otherwise, only one third of our "articular" cells demonstrated enhanced sensitivity of peripheral afferent nerve fibers during acute inflammation.

Nonetheless, our results for cortical cells responding to noxious hindpaw pinch show enhanced responses similar to those seen in peripheral fibers for comparable stimuli. The above theory concerning a "special class of nociceptive afferents" can most pertinently be applied to this group of cells.

Spinal neurons responding to nonnoxious articular movement during a developing inflammation have been studied by few groups. However, in 1987 Schaible et. al. published a study of ascending tract cells in the spinal cat during acute carrageenin and koalin-induced inflammation of the knee joint (61). Their findings differ greatly from our results in the cortex. Of 9 cells responding to flexion of the knee joint prior to inflammation 6 showed increased responses and spontaneous activity. Interestingly, 4 cells initially showing little or no response to flexion developed large responses within 1 or 2 hours after inflammation of the knee joint. It should be noted that Schaible et. al. injected algogenic substances directly into the knee joint thereby insuring inflammation of the joint tissue. In our study intra-plantar injection of carrageenin causes only indirect articular exposure. Thus, joints in our study may have been less affected by carrageenin than was the case for Schaible et. al. Another possibility is ,of course, that somatic information about articular movement is processed differently in the spinal cord than in the cortex.

Nonnociceptive neurons in the VB of the thalamus, however, have been examined during a developing carrageenin inflammation. Results for non-noxious articular movement are generally similar to results for light tactile stimuli. Thus, plantar injection of carrageenin did not produce an increase in responses or spontaneous discharge. It appears, therefore, that cortical and thalamic response characteristics of cells responding to nonnociceptive articular stimuli are similar. Certainly, one must consider the small number of cells that this study was able to consider and the fact that the two cells showing significantly increased responses in the cortex were those for which recording was maintained more than an hour. Nevertheless, we cannot presume that unchanged articular responses for cells recorded less than an hour would have increased if recording had been maintained. Large generalized increases in responses for cells driven by pinching and light touch were observed less than 60 minutes after carrageenin administration.

The question addressed above regarding the evolution of cells recorded for extended periods after carrageenin injection can logically be followed by the question of chronic inflammation. Mono- and

polyarthritic rats provide a model for a chronic inflammatory state.

Nervous Modifications in Poly- and Monoarthritic Rats

Modifications in peripheral afferent fibers due to chronic arthritis are more extreme than those during acute inflammation. Guilbaud et. al. compared a sample of 34 normal and 19 arthritic rats and found that arthritic joint capsule receptors had lower than normal thresholds to mechanical stimulation and were more easily fatigued on repeated stimulation (59). Furthermore, peripheral afferent fibers in arthritic rats demonstrated a heightened capacity to sustain a continuous background discharge (25% of cells showed increased background activity) and to respond to small joint movements. In comparison, 50% of the cortical cells responding to articular movement in the present study showed a similar increase in background activity. They concluded that these results were due in part to altered properties of the joint capsule receptors.

Dorsal horn cells of the polyarthritic spinal rat have been studied by Menetrey and Besson in an electrophysiological study (15). They found that cells in lamina I, containing neurons that respond exclusively or maximally to noxious stimulation (7), and lamina II of the dorsal horn, could be divided into two groups. The first group were those driven by non-oedematous skin showing properties similar to those observed in healthy rats. The second group consisted of cells driven by edematous skin. This second group of cells showed strikingly different electrophysiological properties including a relatively high level of background activity with frequent bursting patterns and sudden dramatic increases as well as an exaggerated reactivity to light mechanical stimuli. In another dorsal horn study Calvino et. al. made similar observations of increased spontaneous activity and responsiveness (9). They also noted that in some cases additional ipsilateral receptive fields developed. These studies demonstrate that cells responding to noxious stimuli in the dorsal horn display post-inflammatory modifications similar to those seen in nociceptive SmI cortical cells. Our results illustrate the same increased responses and spontaneous discharge as well as the appearance of new receptive fields and one case of an ipsilaterally expanded receptive field.

A recent study by Hylden et. al. investigated the contribution of dorsal horn mechanisms to the

above expansions in receptive fields in rats with chronic arthritis (13). They proposed two possible centrally mediated mechanisms which might contribute to these expansions; "an increase in synaptic efficacy of excitatory inputs or a release of inhibitory inputs in the dorsal horn." The former idea is reminiscent of Schmidt and Schaible's theory about a "special class of nociceptive afferents." Hylden pointed out that such an unmasking of previously existing but inoperative synapses has already been produced by electrical stimulation of ineffective inputs, noxious thermal stimuli, and noxious pinch (see 13). In order to explain the increased responses seen in the dorsal horn Hylden proposed that "increased spatial summation from neurons with enlarged receptive fields may contribute to the appearance of hyperalgesia. Expanded receptive fields would lead to a greater number of neurons activated by a given stimulus; the resulting increase in discharge may ultimately be perceived as more intense pain (13)." This theory could possibly explain our results, in that cells displaying expanded receptive fields showed significantly greater increases in both their responses and their spontaneous activity than cells with no expansions in RF. It should, however, be noted that the relatively deep chloralose anesthesia used by Hylden et. al. favors expanded receptive fields (80).

Thalamic neurons of polyarthritic rats display profound changes in responses to somatic stimuli (8). Compared with normal rats, polyarthritic rats show few neurons activated by intense mechanical stimulation whereas many cells respond to gentle stimulation of the joints and surrounding inflamed areas. In the VB of the arthritic rat more than 50% of the somatosensory neurons are driven by mild stimuli. Furthermore, in one study 50% of the neurons studied displayed unusual patterns of responses consisting of very long post-effects and paroxysmal discharges (10). This study suggested that the long-lasting afterdischarges could be a result of activation and sensitization of the cutaneous thin afferent fibers by algogenic substances present in the skin of the inflamed paw. Indeed, forementioned studies (15) in the dorsal horn of polyarthritic rats observed this same sensitization phenomenon.

More recently Guilbaud et. al. published findings further supporting the role of central phenomena in the forementioned changes seen in ventrobasal thalamic neurons of arthritic rats (11). In addition to previously mentioned results demonstrating the predominance of VB thalamic responses to joint movement, Guilbaud et. al. also found that a local injection of lidocaine in one hind paw strongly depressed the VB thalamic neuronal responses to mild stimulation of both ankles. Along with this

electrophysiological study a behavioral study was conducted whereby the vocalization threshold to paw pressure after a unilateral anesthetic block was examined. This behavioral study demonstrated bilateral hypoalgesia thereby confirming the electrophysiological findings. The results of this study demonstrated not only that the responsiveness of peripheral receptors but also that of central neuronal responses are modified by chronic arthritis (20,22).

Sml cortical neurons have as yet never been studied using the carrageenin model for acute inflammatory pain. Nevertheless, Lamour et. al. have examined these neurons using the polyarthritic rat (81). Their results resembled studies in the VB in terms of the lack of neurons driven by light cutaneous and intense mechanical stimulation found. However, the Sml cortex of arthritic rats differed from the VB in three ways. First, long afterdischarges largely outlasting the period of stimulation were not regularly observed. Second, paroxysmal discharges were never observed, and third, neurons driven by brushing the skin were rarely observed (6 out of 117 neurons) in the cortex, whereas they represented 16% of the neurons observed in the VB. These findings contrast equally with the results of the present study. The total absence of paroxysmal discharge in the arthritic rat's Sml cortex as compared to the significant increase in such discharge during acute inflammation of the peripheral field could possibly suggest that central processes of inhibition, somehow interrupted during the acute inflammatory phase, are subsequently amplified as a result of chronic inflammatory pain. This disinhibition could be a preliminary step in the dramatic sensory reorganization toward cells responding to joint movement seen in arthritic rats.

Another form of Sml cortical reorganization is seen in the somatosensory map of partially deafferented mammals. Dykes and Lamour have shown that a major deafferentation of the rat hindlimb causes certain neurons from the reorganized cortex to have an elevated spontaneous activity (75,76). They concluded that cells from deafferented cortex have fewer excitatory inputs and are under a weaker degree of inhibitory control than normal neurons. The increased responses and spontaneous activity of deafferented animals has also been seen in animals with peripheral neuropathies (82 and unpublished data). Although comparison of injury and pain in the form of deafferentation with that of acute to chronic inflammation associates two very different phenomena, central accommodation might have some common points. Is it possible that selective disinhibition of neurons driven by damaged receptive fields

is a universal nervous response to injury? Obviously, further investigation is necessary before such proposals can be discussed in depth.

Problems Involving Anesthesia

As in past studies (62,63), neuronal activity was recorded in rats under a stable inhalation anesthesia of 0.5% halothane in a mixture of 1/3 O₂ and 2/3 N₂O. The present study confirms that this level of anesthesia does not abolish cortical responses to noxious stimuli. Nevertheless, one must consider possible ramifications of the anesthetic such as inhibitory and excitatory effects as well as changes in receptive field size (83,84).

When the above halothane anesthesia is increased to 2 or 2.5% responses to nociceptive stimuli progressively disappear (80). This fact may explain the inability of early researchers to find nociceptive cortical neurons (6). In contrast, responses to light tactile and other nonnociceptive stimuli are not diminished with halothane. Indeed, responses to light tactile stimuli are increased under deeper levels of chloralose anesthesia (60 mg/kg iv) (33). As suggested by Lamour et. al., even the moderate anesthesia used could increase the threshold of cortical responses to peripheral stimuli thereby suppressing nonnociceptive pathways and inducing an apparent specificity which would not exist in unanesthetized animals (63). Lamour et. al. refuted this idea by pointing out that such an induced specificity can not explain the clear differences in laminar distribution of different categories of neurons found in their study. As described in the results the laminar distribution of the present study is similar to that of the Lamour study (62).

Receptive fields covering large parts of the body have been described under different levels of anesthesia. Andersson (85) suggested that this may be due to cortical arousal. However, the possibility that changes in receptive fields seen in the present study is due to anesthetic level is quite unlikely because of the lack of ECoG signs of such arousal during the experiments. It must be noted that the ECoG was continuously monitored and experiments were discontinued if ECoG signs suggested deterioration or arousal.

Finally, administration of the local anesthetic xylocaine decreased both evoked and spontaneous

discharge. This depressive effect does not seem to result from diffusion into the circulation to produce a systemic effect. Studies of the effects of i.v. lidocaine administration on dorsal horn neurons show that substantially greater doses are needed to produce a general anesthetic effect (86). Furthermore, increased spontaneous discharge does not seem to result from cellular irritation as shown by the depression and recovery of such activity resulting from local anesthetic. Finally, spikes of cells showing increased spontaneous activity did not have the appearance typical of damaged cells (figure 13).

Conclusion

The first major conclusion of the present study is a confirmation that the SmI cortex participates in nociception. Our findings also demonstrate that SmI cortical processing of peripheral noxious information resembles that of the ventrobasal thalamus, whereas SmI cortical processing of peripheral nonnociceptive light touch during a developing inflammation differs from that of lower parts of the nervous system. Finally, the significant increase in spontaneous activity of cortical cells driven by light touch, not observed in the ventrobasal thalamus, can be explained by disinhibition of previously inhibitory synapses or recruitment of previously non-functional synapses.

It is interesting to compare the expansion of receptive fields seen in the present study with similar expansions seen in previous studies. Notably, cortical neurons can become responsive to cutaneous inputs which are outside their normal skin fields if these fields are deafferented. In general, reports have dealt with chronic deafferentation, but there is increasing evidence that such expansions in receptive fields can occur within a few hours after nerve block or injury (87,88,89). As pointed out by Wall, these findings suggest that rapid alterations in cortical organization can occur when normal inputs are disrupted (90). A central mechanism such as the activation of previously undetected or inactive connections could explain these expanded receptive fields. The mechanism or mechanisms behind expansion of receptive fields, as well as increased spontaneous activity and responses, is certainly of neural rather than humoral origin as shown by the suppression of evoked and spontaneous discharge after local anesthesia of the receptive field (21,91).

In conclusion, modifications in the first somatosensory cortex during a developing carrageenin-induced inflammation are complex and, in some ways, unique in the nervous system. We have shown that an acute inflammation results in rapid cortical changes, at times reminiscent of alterations seen in chronic inflammation and even deafferentation. One must therefore ask if variations in cortical activity after peripheral injuries of very different severity might be due to the same mechanisms. Studies of cortical modifications on a subacute basis one or two days after carrageenin injection could answer this question. Likewise, development of techniques allowing cortical cells to be recorded for longer periods during the evolution of a peripheral inflammation or lesion would facilitate a better understanding of subsequent cortical modifications.

2/3 N₂O 1/3 O₂

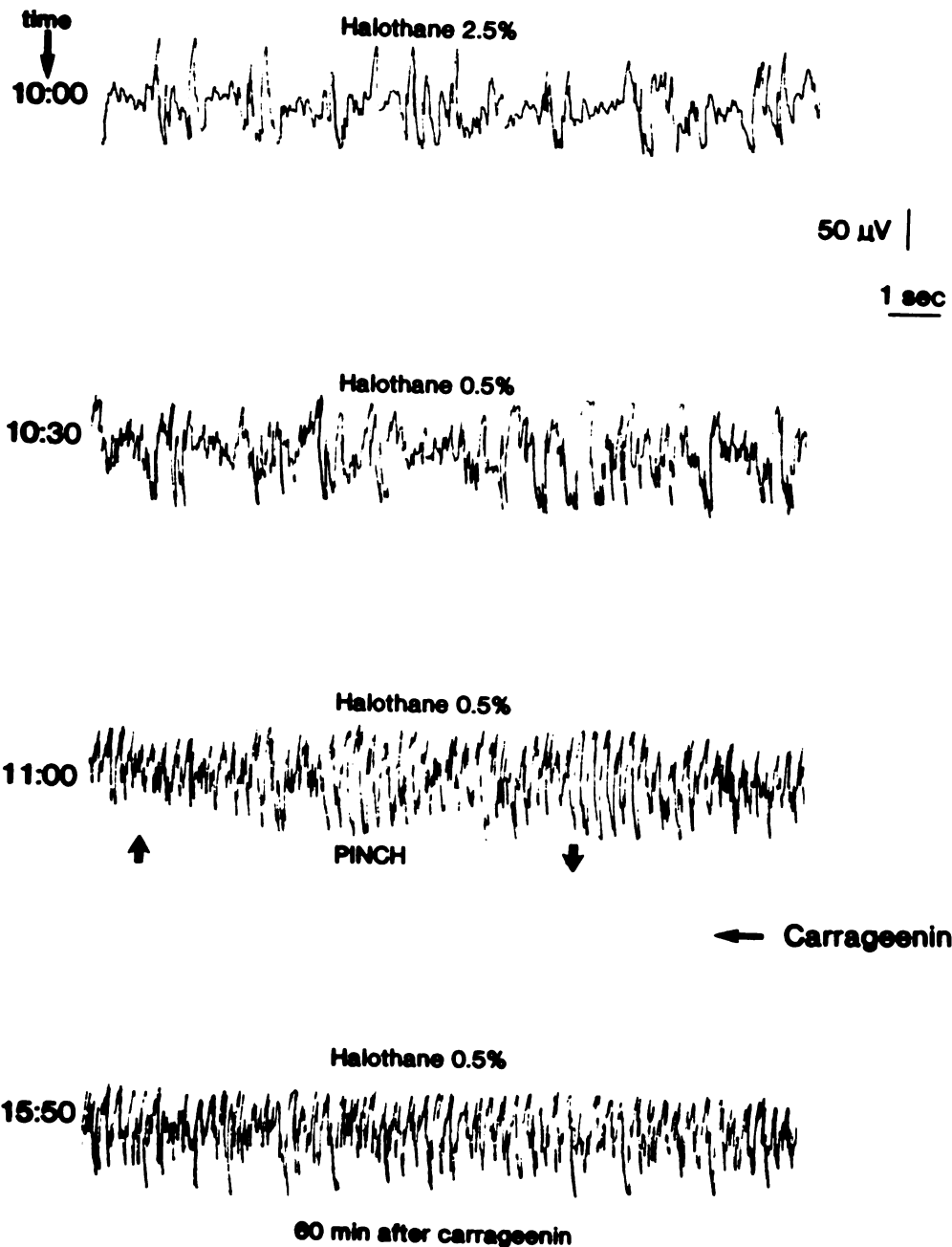


Figure 1) Electrocorticogram tracings are shown for different time periods during surgical preparation and post-carrageenin cellular recording. The pinch at 11:00 indicates the tail pinch used to determine whether the animal shows signs of arousal.

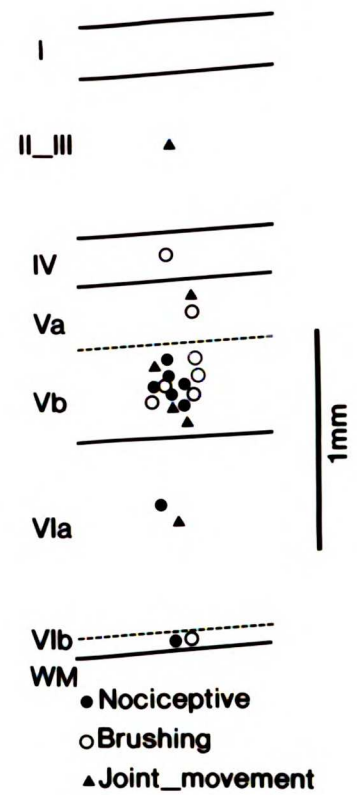
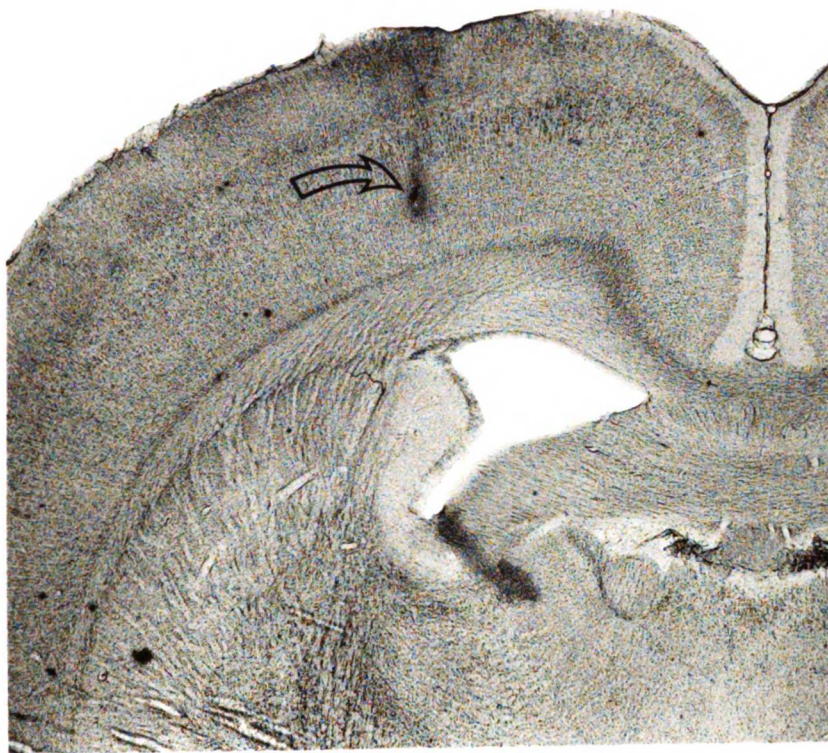


Figure 2) A photograph of the cortex showing the dye spot used to identify the cortical layer of the recorded cell. The laminar distribution of the 22 neurons used for statistical analysis is also shown.

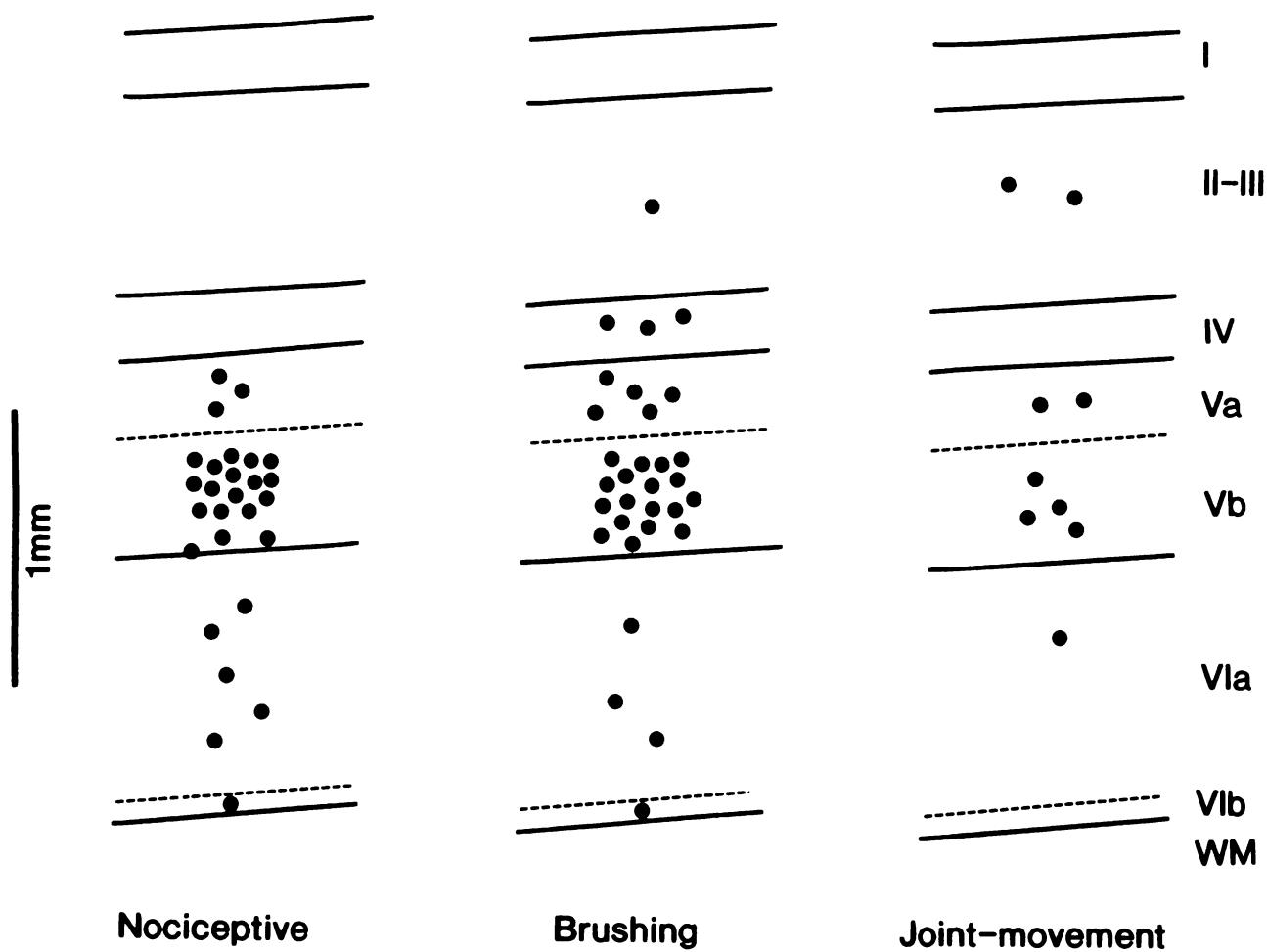
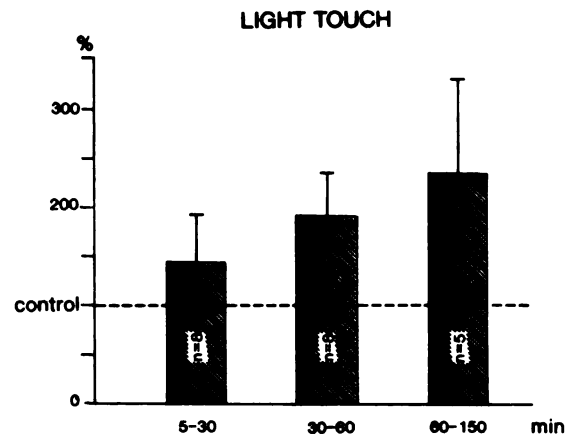
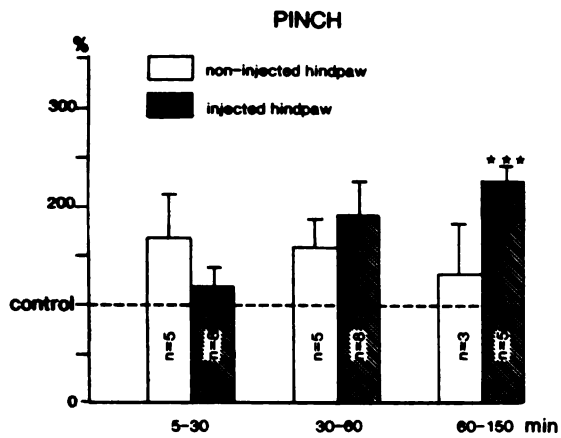


Figure 3) A laminar distribution of all cells recorded during the study. Notice the small number of cells driven by joint movement.

RESPONSES



SPONTANEOUS ACTIVITY

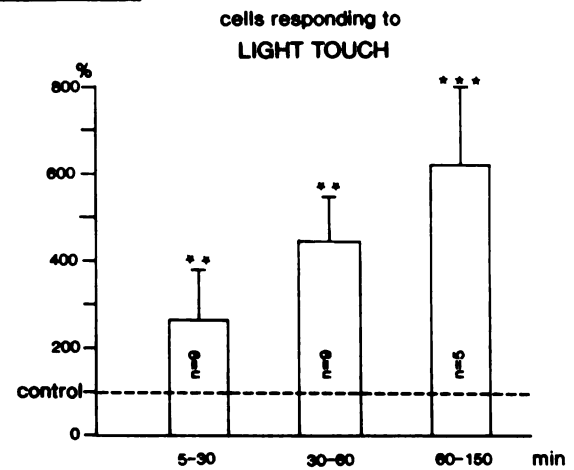
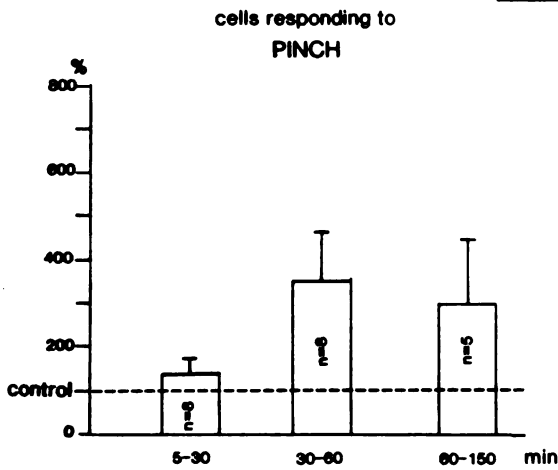


Figure 4) Results are given for responses and spontaneous activity of cells driven by noxious pinch and light nonnociceptive touch of the hindpaw subsequent to intraplantar carrageenin injection. Notice that all responses increased over the recording period whereas only nonnociceptive cells showed a clear tendency toward increased activity. Statistical significance is indicated by stars. *** indicates $p < 0.001$, ** indicates $p < 0.01$ and * indicates $p < 0.05$, ANOVA test.

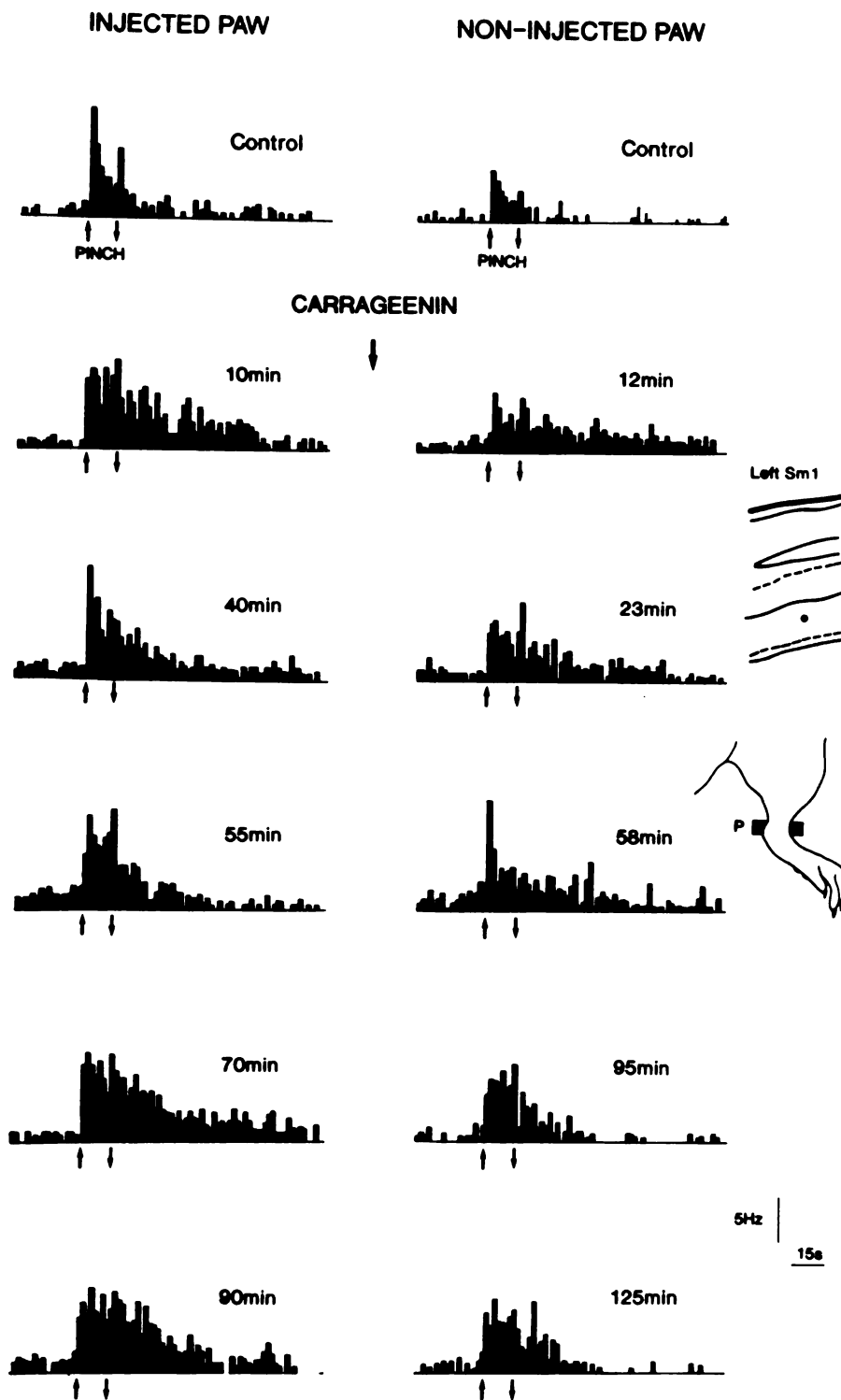


Figure 5) Responses of one cortical neuron driven bilaterally by noxious pinch of the hindpaw. Notice the increase in responses for both the injected and the non-injected paw after carrageenin administration. Spikes are counted in 2 second epochs.

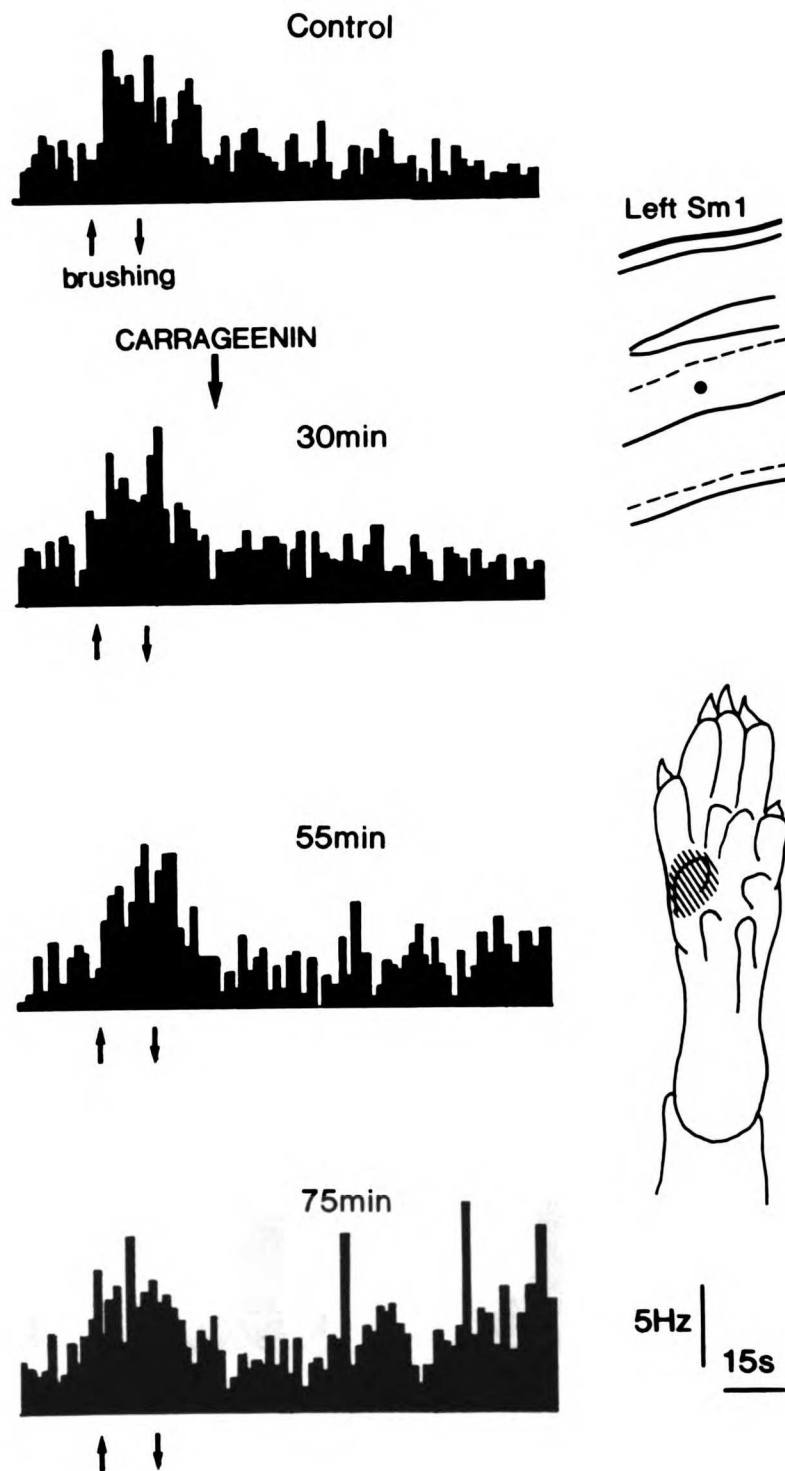


Figure 6) An example of a cortical cell responding to light touch. Notice that this cell did not show increased responses, but did show significantly increased spontaneous activity. Most cells driven by light touch showed both increased responses and spontaneous activity, but the lack of significance of this increase in responses accompanied by a high statistical significance of the increase in spontaneous activity seen in our results is well-illustrated by this cell.

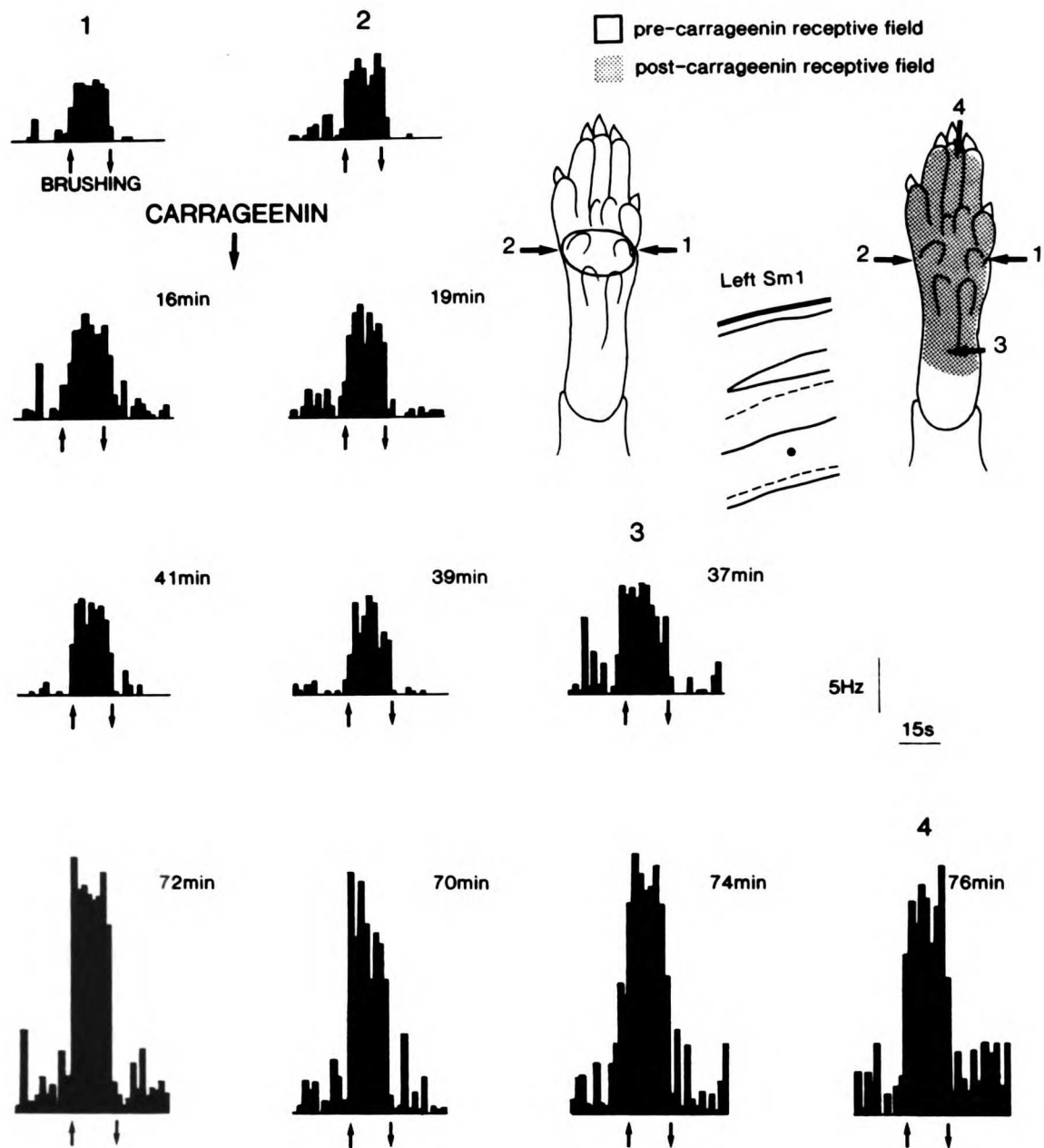


Figure 7) A cortical cell displaying marked expansions in its receptive field. Receptive fields and sites of stimulation are displayed in the two schematic drawings of the injected hindpaw. Note that prior to carrageenin injection the cell responded to stimuli of two different plantar pads (sites 1 and 2 in the schematic drawing of the paw prior to carrageenin). 37 minutes after carrageenin, the cell responded to stimulation of site 3. 76 minutes after carrageenin, stimulation of site 4 produced responses. Notice also the increased magnitude of responses and background activity with time.

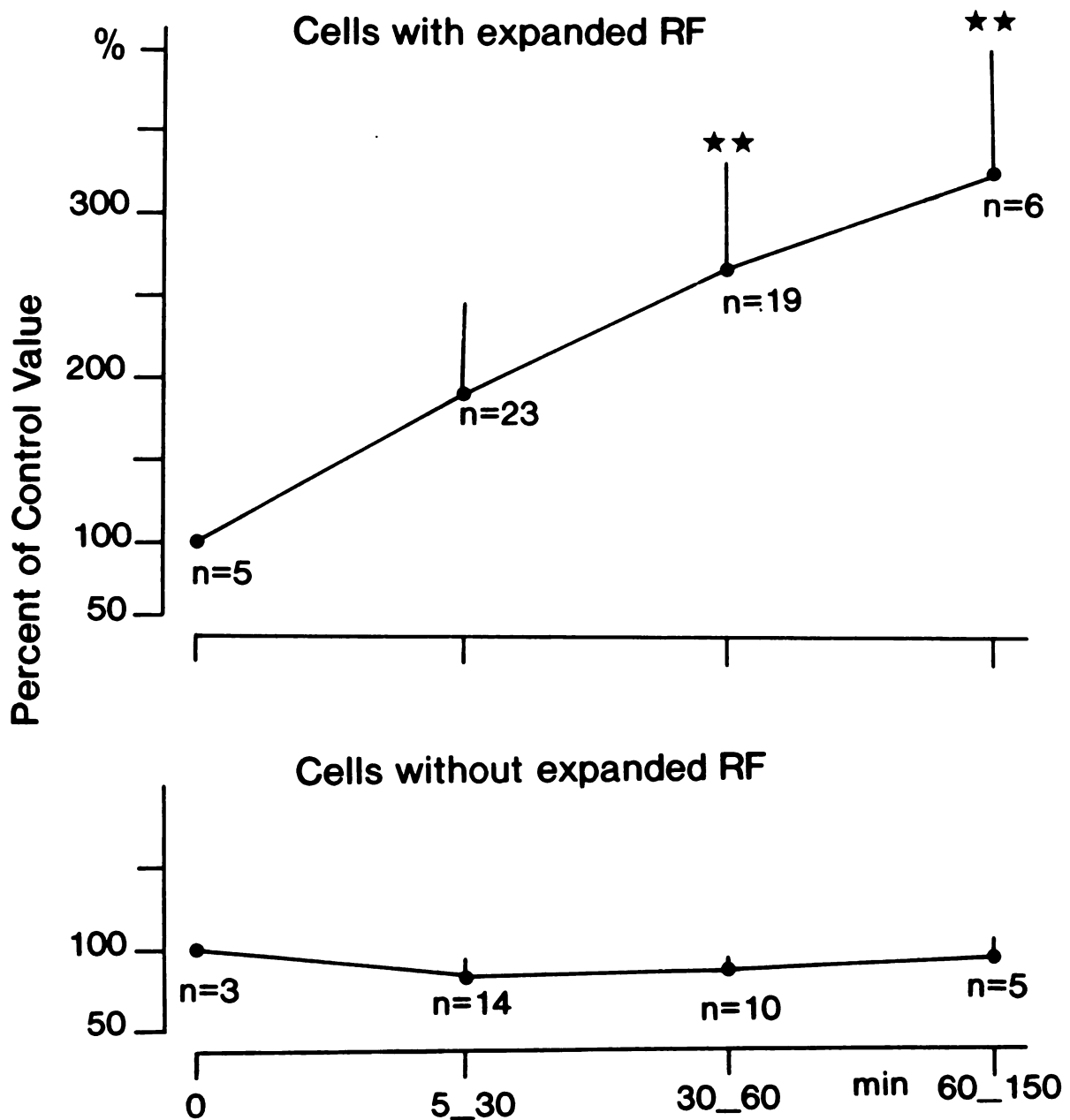


Figure 8) Post-carrageenin responses measured as a percent of the control value (100%) are shown for cells responding to light touch with and without expanded receptive fields. Notice that responses for cells with expanded RF increased significantly (** indicates $p < 0.01$ by ANOVA tests) whereas responses for cells without expanded receptive fields remained similar to control values.

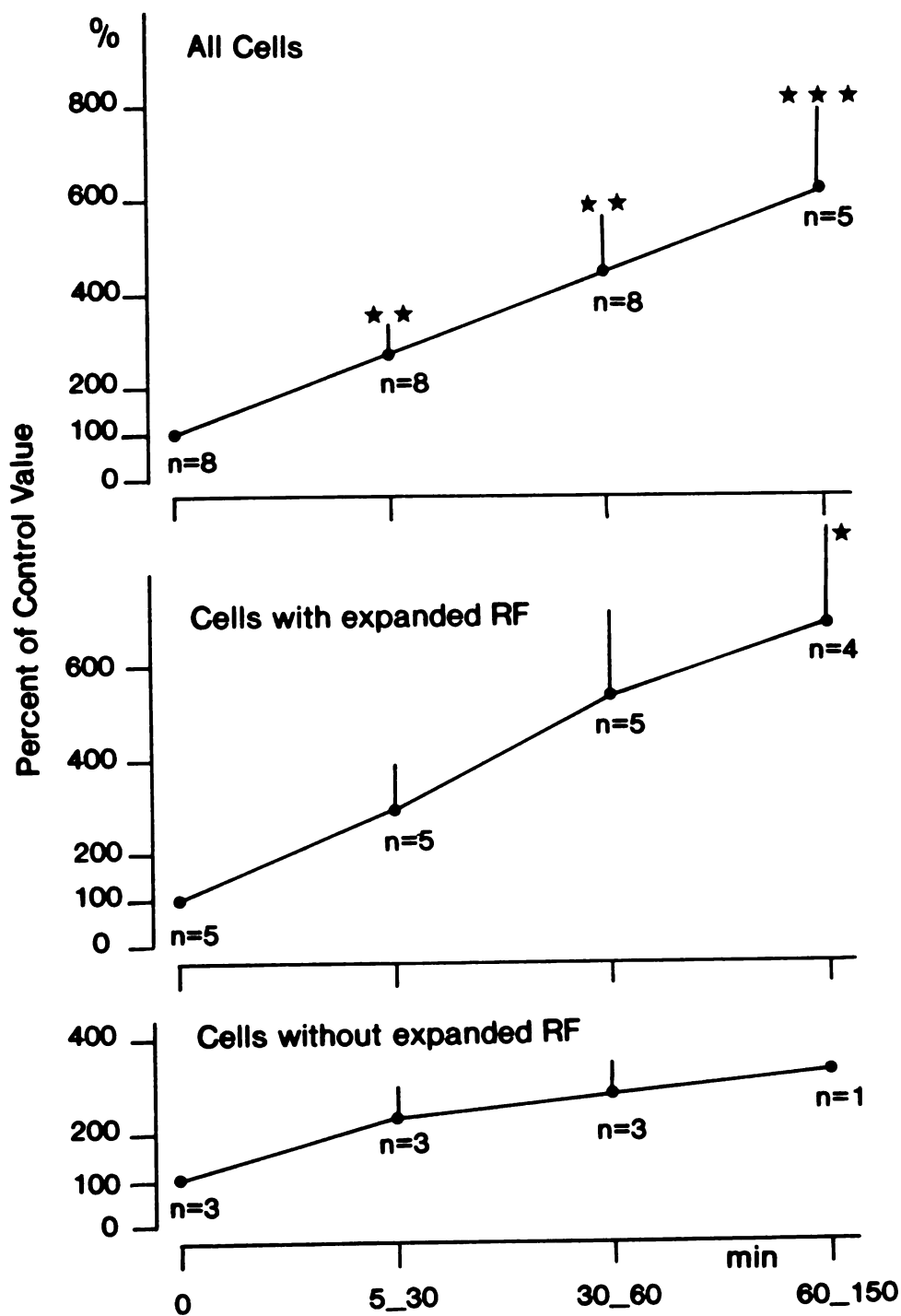


Figure 9) Post-carrageenin spontaneous activity of cells responding to light touch is shown for all cells used in the statistical analysis. This group of neurons is then divided into those with and those without expanded receptive fields. Note that cells show a tendency toward increased spontaneous activity regardless of RF expansion, but the degree and statistical significance of this increase varies according to RF expansion. (***) indicates $p < 0.001$, ** indicates $p < 0.01$, and * indicates $p < 0.05$, ANOVA tests)

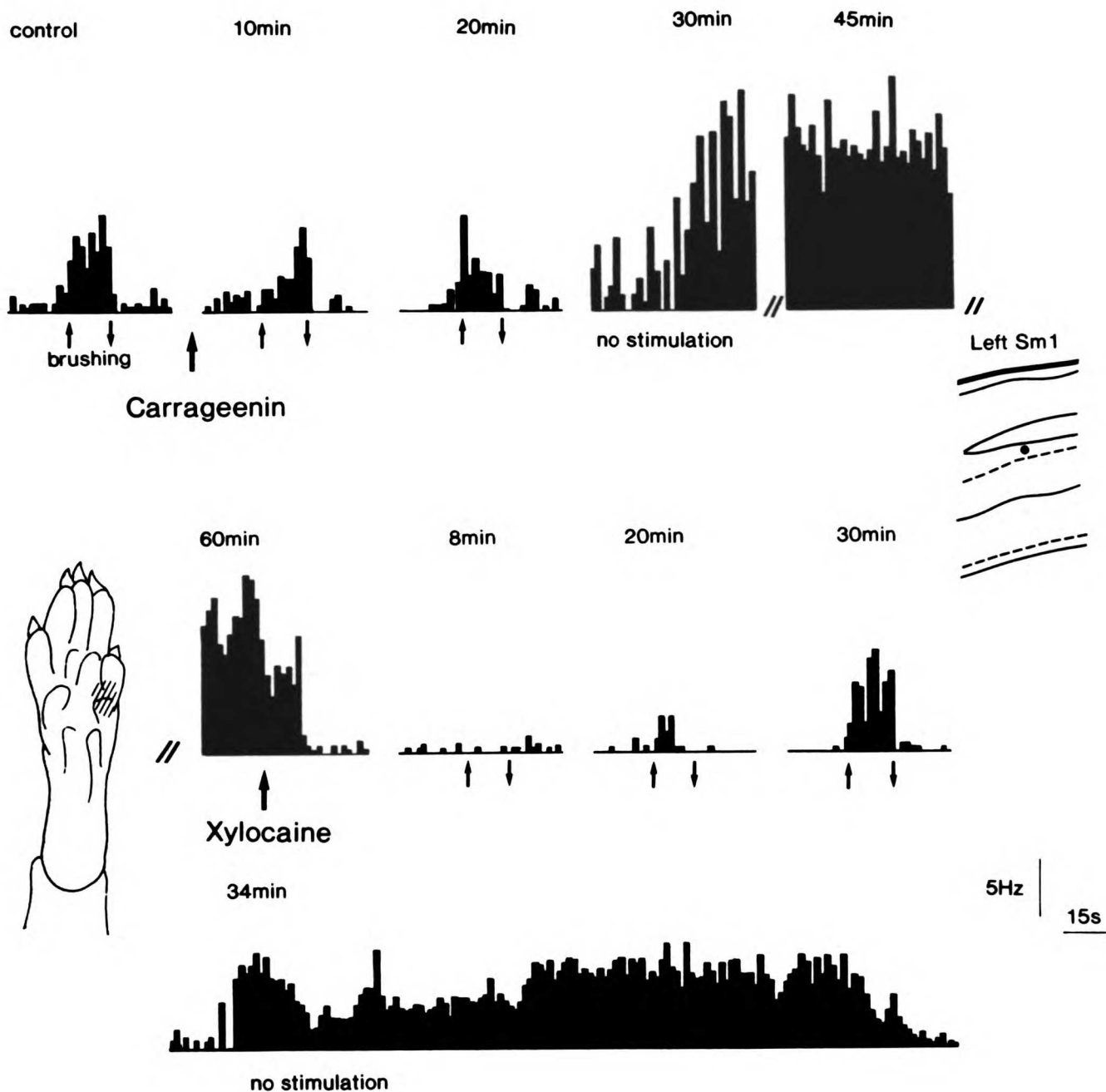


Figure 10) Modifications seen in one cortical cell responding to light touch during a recording session. Notice the sudden burst of spontaneous hyperactivity seen 30 minutes after carrageenin and lasting until subsequent administration of xylocaine at 60 minutes. Responses and background activity are depressed for 20 minutes and gradually return until a similar, but less intense, burst of hyperactivity begins at 34 minutes after local anesthetic.

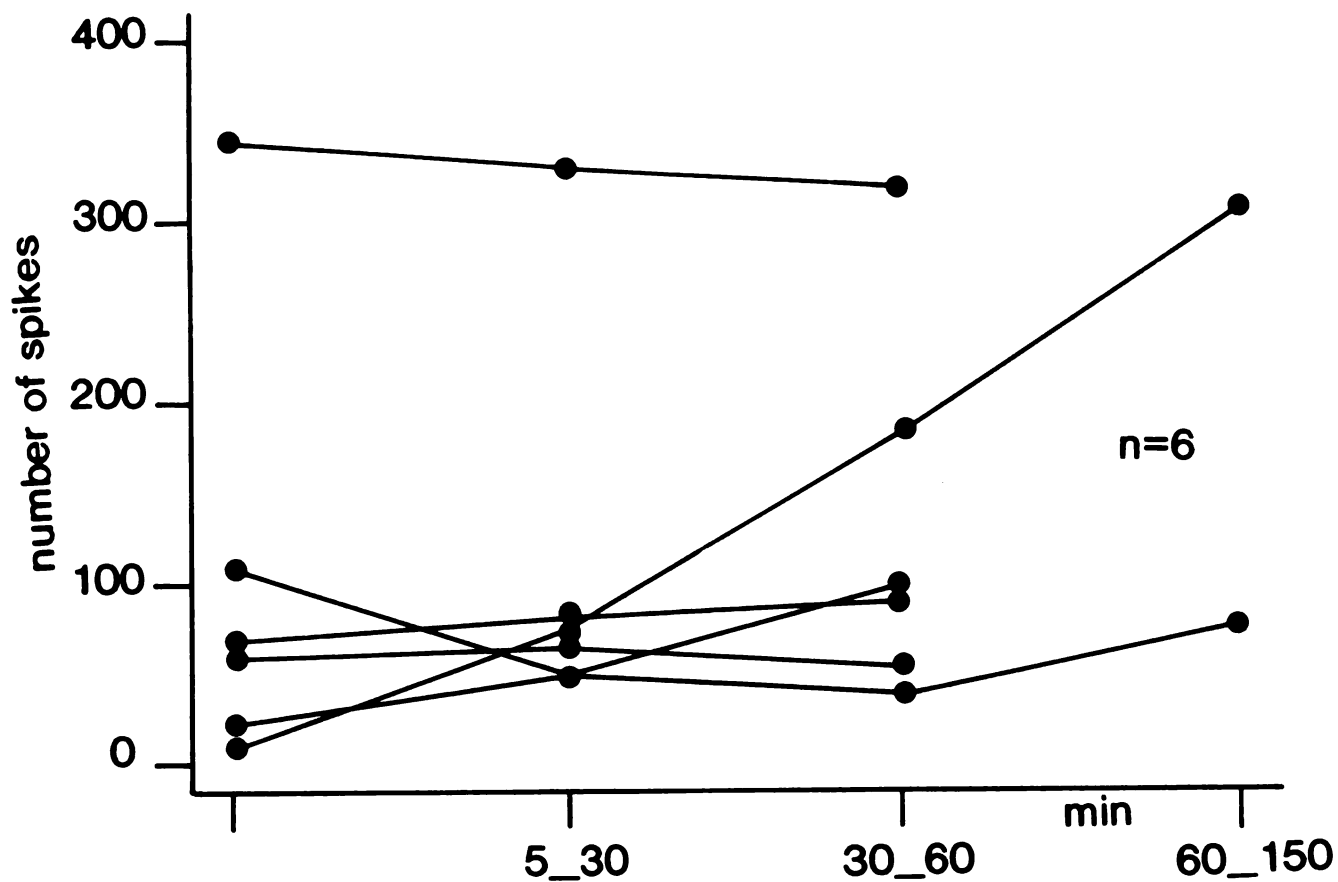


Figure 11) Post-carrageenin responses of cells driven by nonnociceptive articular movement are illustrated in number of spikes per time interval. Note that only two neurons displayed increased responses while four remained stable at control values. Note, however, that the two cells showing increased responses were also the two cells recorded for the longest time period.

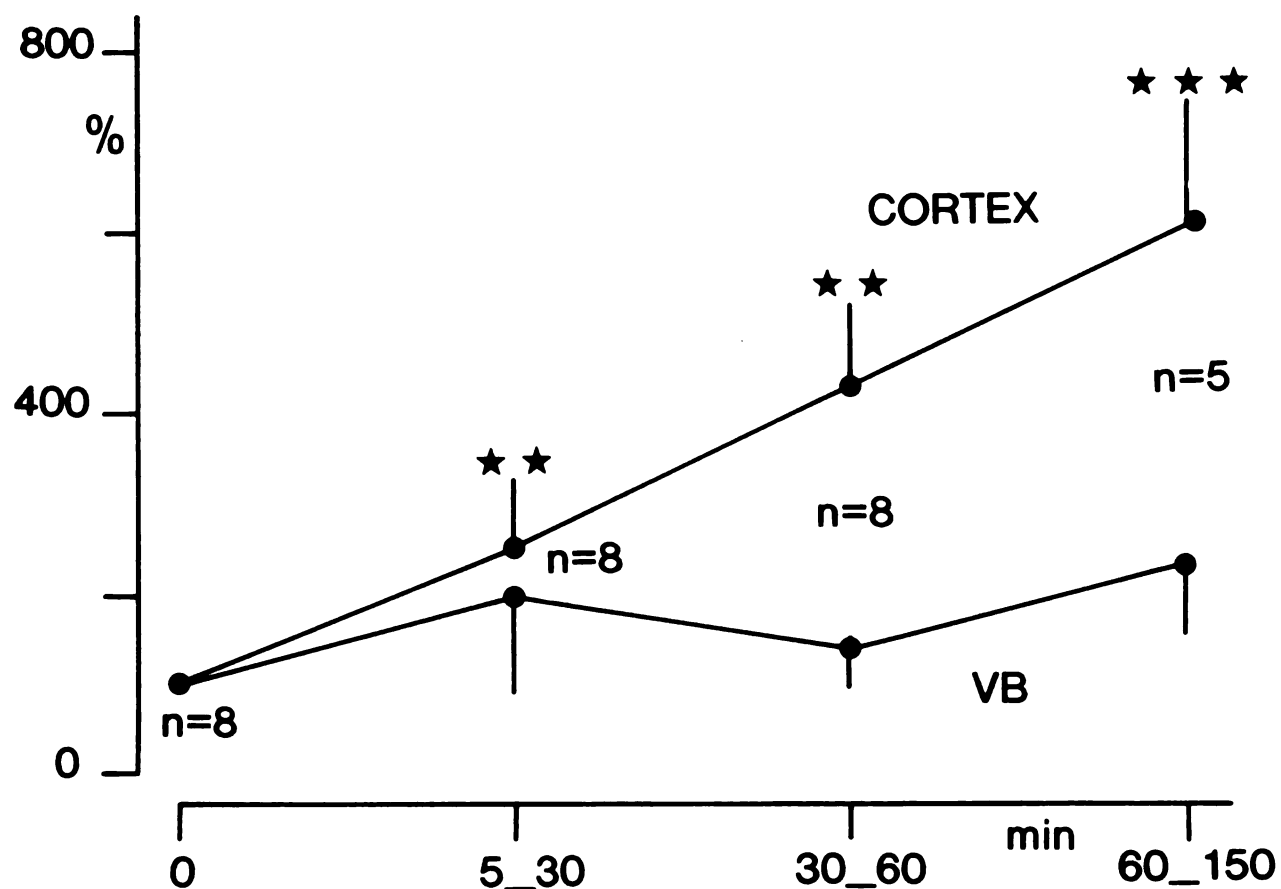


Figure 12) Post-carrageenin spontaneous activity of $n=8$ cells driven by light touch in the cortex is compared to such activity of $n=8$ cells in the ventrobasal complex of the thalamus under identical anesthesia for the same time periods. Note that cortical activity displays a statistically significant increase while thalamic activity does not differ statistically from pre-carrageenin values. (***) indicates $p<0.001$ and ** indicates $p<0.01$ by ANOVA tests)

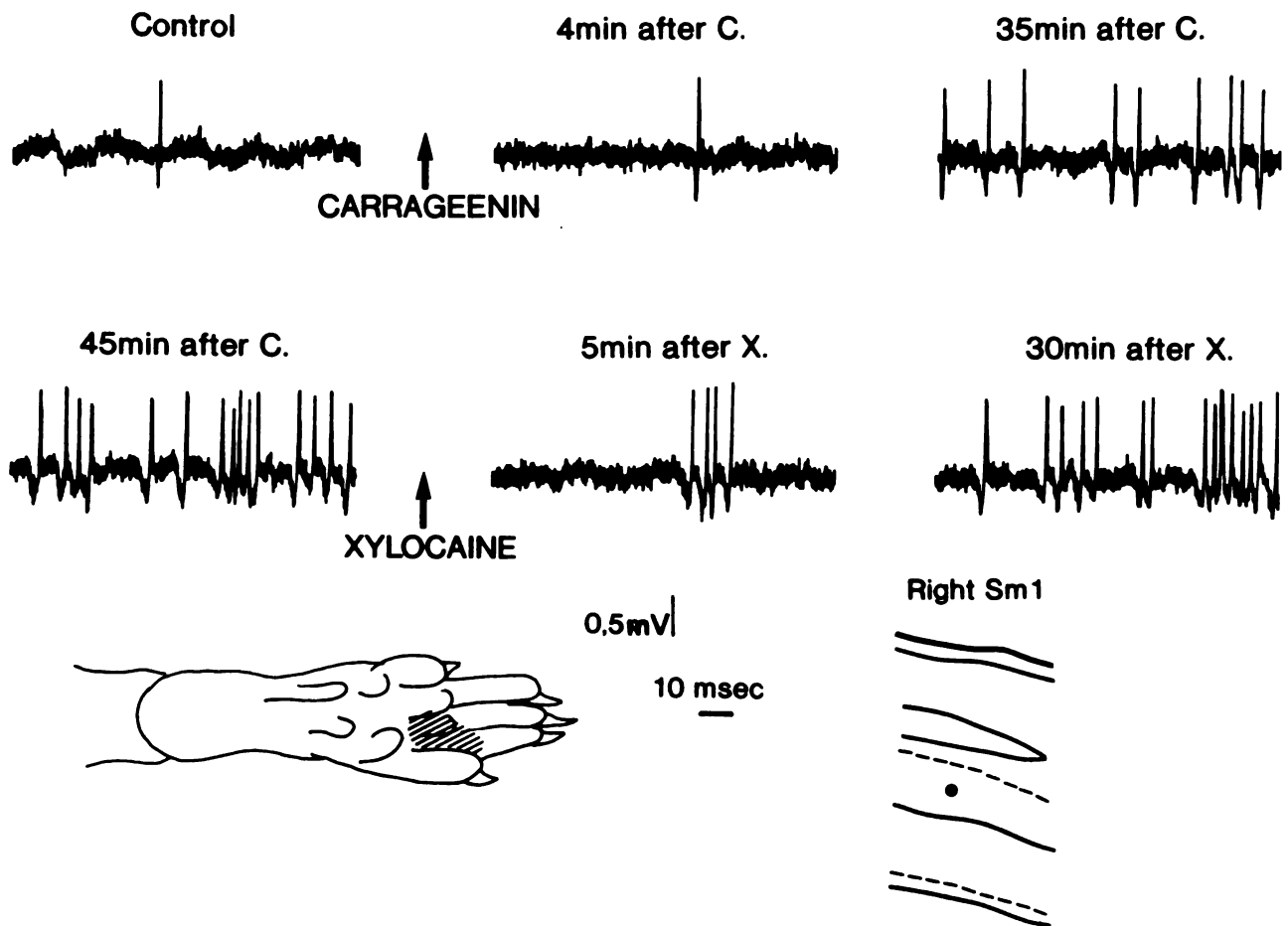


Figure 13) An illustration of spikes as seen in the oscilloscope. This cell responded to light touch though all illustrated spikes represent background activity not triggered by stimulation of the receptive field. Note the increased spontaneous activity until xylocaine was locally injected resulting in diminution and final recovery of cellular discharge 30 minutes after local anesthetic. Spikes do not have the typical appearance of damaged cells.

Bibliography

- 1) Bonica, J.J., Causalgia and Other Reflex Sympathetic Dystrophies. In Advances in Pain Research and Therapy, Vol. 3, edited by J.J. Bonica et al., Raven Press, New York, 1979.
- 2) Noordenbos, W., Sensory Findings in Painful Traumatic Nerve Lesions. In Advances in Pain Research and Therapy, Vol. 3, edited by J.J. Bonica et al., Raven Press, New York, 1979.
- 3) Willis, W.D., The Pain System, vol. 8, Karger, New York, 1985.
- 4) Wall, P.D., and McMahon, S.B., The relationship of perceived pain to afferent nerve impulses. Trends Neurosci. 6: 254-255, 1986.
- 5) Sherrington, C.S., The Integrative Action of the Nervous System, Yale Univ. Press, New Haven, CT, 1906.
- 6) Hardy, J.D., Wolff, H.G., and Goodell, H., Pain Sensations and Reactions, Hafner Publishing Company, N.Y., 1967.
- 7) Besson, J.M., and Chaouch, A., Peripheral and Spinal Mechanisms of Nociception. Physiol. Rev. 67: 67-186, 1987.
- 8) Besson, J.M., Guilbaud, G., The Arthritic Rat as a Model of Clinical Pain? Proceedings of the International Symposium on The Arthritic Rat as a Model of Clinical Pain?, Elsevier Science Publishers, N.Y., 1988.
- 9) Calvino, B., Villanueva, L., Le Bars, D., Dorsal horn (convergent) neurones in the intact anaesthetized arthritic rat. I. Segmental excitatory influences. Pain 28: 81-98, 1987.
- 10) Gautron, M., Guilbaud, G., Somatic responses of ventrobasal thalamic neurones in polyarthritic rats. Brain Research 237: 459-471, 1982.
- 11) Guilbaud, G., Benoist, J.M., Eschali r, A., et. al., Evidence for peripheral serotonergic mechanisms in the early sensitization after carrageenin-induced inflammation: electrophysiological studies in the ventrobasal complex of the rat thalamus using a potent specific antagonist of peripheral 5-HT receptors. Brain Res. 502: 187-197, 1989.
- 12) Guilbaud, G., Kayser, V., and Lombard, M.C., Les differents modeles experimentaux de la douleur chronique. Interet et critiques. in La Douleur et son Traitement, Arnette, Paris, 1988.
- 13) Hylden, J., Nahin, R., Traub, R., et. al., Expansion of receptive fields of spinal lamina I projection neurons in rats with unilateral adjuvant-induced inflammation: the contribution of dorsal horn mechanisms. Pain 37: 229-243, 1989.
- 14) Levine, J.D., Clark, R., Devor, M., et. al., Intraneuronal substance P contributes to the severity of experimental arthritis. Science 226: 547-549, 1984.

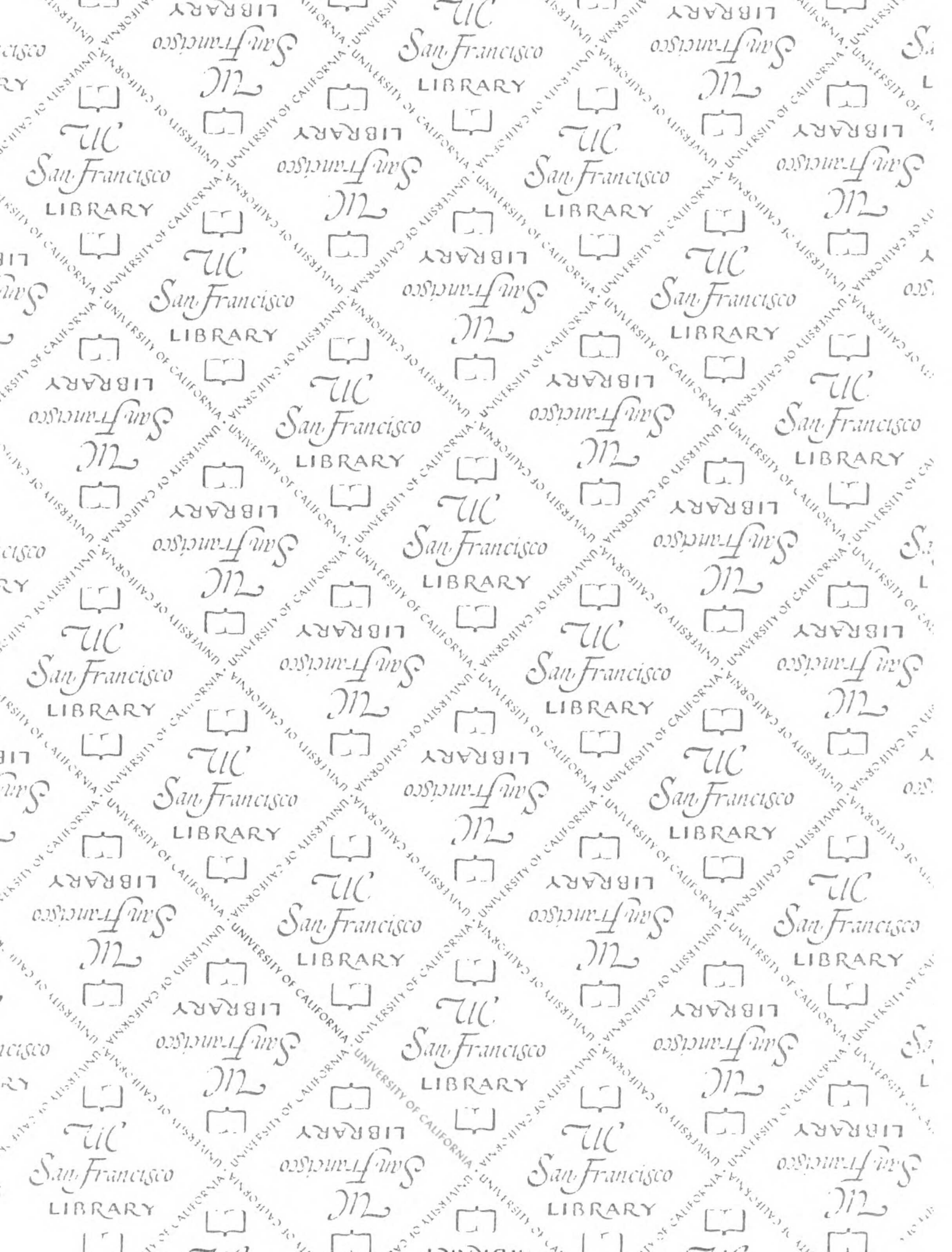
- 15) Men  tre  y, D., Besson, J.M., Electrophysiological characteristics of dorsal horn cells in rats with cutaneous inflammation resulting from chronic arthritis. *Pain* 13: 343-364, 1982.
- 16) Benoist, J.M., Kayser, V., Gautron, M. et. al., Changes in responses of ventrobasal thalamic neurons during carrageenin- induced inflammation in the rat. *Adv. in Pain Res. and Ther.* 9:295-303, 1985.
- 17) Di Rosa, M., Giroud, J.P., Willoughby, D.A., Studies of the mediators of the acute inflammatory response induced in rats in different sites by carrageenin and turpentine. *J. Path.* 104: 15-29, 1971.
- 18) Guilbaud, G., Benoist, J.M., Eschali  r, A., et. al., Evidence for central phenomena participating in the changes of responses of ventrobasal thalamic neurons in arthritic rats. *Brain Research* 484: 383-388, 1989.
- 19) Guilbaud, G., Benoist, J.M., Neil, A., et. al., Neuronal response thresholds to and encoding of thermal stimuli during carrageenin-hyperalgesic-inflammation in the ventro-basal thalamus of the rat. *Brain Res.* 66: 421-431, 1987.
- 20) Guilbaud, G., Kayser, V., Benoist, J.M., et. al., Modifications in the Responsiveness of Rat Ventrobasal Thalamic Neurons at Different Stages of Carrageenin-Produced Inflammation. *Brain Res.* 385: 86-98, 1986.
- 21) Guilbaud, G., Neil, A., Benoist, J.M., et. al., Thresholds and encoding of neuronal responses to mechanical stimuli in the ventro-basal thalamus during carrageenin-induced hyperalgesic inflammation in the rat. *Exp. Brain Res.* 68: 311-318, 1987.
- 22) Kayser, V., and Guilbaud, G., Local and remote modifications of nociceptive sensitivity during carrageenin-induced inflammation in the rat. *Pain* 28: 99-107, 1987.
- 23) Kocher, L., Anton, F., Reeh, P. et. al., The effect of carrageenin-induced inflammation on the sensitivity of unmyelinated skin nociceptors in the rat. *Pain* 29: 363-373, 1987.
- 24) Neil, A., Benoist, J.M., Kayser, V., et. al., Initial nociceptive sensitization in carrageenin-induced rat paw inflammation is dependent on amine autocoid mechanisms: electrophysiological and behavioral evidence obtained with a quaternary antihistamine, thiazinamium. *Exp. Brain Res.* 65: 343-351, 1987.
- 25) Roch-Arveiller, M., Dunn, C., Giroud, J.P., Comparaison des propri  t  s inflammatoires des fractions lamda et chi de carragenine. *J. Pharmacol.* 8,4,: 461-476, 1977.
- 26) Vinegar, R., Truax, J.F., Selph, J.L., Johnston, R., Venable, A.L., McKenzie, K.K., Pathway to carrageenin-induced inflammation in the hind limb of the rat. *Federation Proc.* 46: 118-126, 1987.
- 27) Burgess, P.R., and Perl, E.R., Cutaneous mechanoreceptors and nociceptors. In: Handbook of Sensory Physiology. Somatosensory System. edited by A. Iggo. Berlin: Springer-Verlag, Vol. 2, p. 29-78, 1973.
- 28) Iggo, A., Andres, K.H., Morphology of cutaneous receptors. *Ann. Rev. Neurosci.* 5: 1-31, 1982.

- 29) Fleischer, E., Handwerker, H.O., and Joukhador, S., Unmyelinated nociceptive units in two skin areas of the rat. *Brain Research* 267: 81-92, 1983.
- 30) Lynn, B., Carpenter, S., Primary afferent units from the hairy skin of the rat hind limb. *Brain Res.* 238: 29-43, 1982.
- 31) Kandel, E.R., and Schwarz, J.H. (eds), Principles of Neural Science, 2nd edn. Elsevier, 1985.
- 32) Nothias, F., Peschanski, M., and Besson, J.M., Somatotopic reciprocal connections between the somatosensory cortex and the thalamic PO nucleus in the rat. *Brain Res.* 447: 169-174, 1988.33) Armstrong-James, M., George, M.J., Bilateral receptive fields of cells in rat Sm1 cortex. *Exp. Brain Res.* 70: 155-165, 1988.
- 34) Chapin, J.K., Woodward, D.J., Somatic sensory transmission to the cortex during movement: Phasic modulation over the locomotor step cycle. *Exp. Neurol.* 78: 670-684, 1982.
- 35) Chapin, J.K., Woodward, D.J., Somatic sensory transmission to the cortex during movement: Gating of single cell responses to touch. *Exp. Neurol.* 78: 654-689, 1982.
- 36) Donoghue, J.P., Kerman, K.L., and Ebner, F.F., Evidence for two organisational plans within the somatic sensory-motor cortex of the rat. *J. Comp. Neurol.* 183: 647-664, 1979.
- 37) Head, H., Studies in Neurology. London: Oxford Univ. Press, vol 2, 1920.
- 38) Penfield, W., Rasmussen, T., The cerebral cortex of man. New York: Hafner, 1968.
- 39) White, J.C., Sweet, W.H., Pain and the neurosurgeon: A forty year experience. Springfield, IL: Thomas, 1969.
- 40) Berkley, K.J., Parmer, R., Somatosensory cortical involvement in responses to noxious stimulation in the cat. *Exp. Brain Res.* 20: 363-374, 1974.
- 41) Dejerine, J., Mouzon, J., Un nouveau type de syndrome sensitif cortical observé dans un cas de monoplegie corticale dissociée. *Rev Neurol* 28: 1265-1273, 1915.
- 42) Marshall, J., Sensory disturbances in cortical wounds with special reference to pain. *J Neurol Neurosurg Psychiatry* 14: 187-204, 1951.
- 43) Lewin, W., Philips, C.G., Observations on partial removal of the postcentral gyrus for pain. *J Neurol Neurosurg Psychiatry* 15: 143-147, 1952.
- 44) Melzack, R., Haugen, F.P., Responses evoked at the cortex by tooth stimulation. *Am. J. Physiol.* 190: 570-574, 1957.
- 45) Shigenaga, Y., Matano, S., Kusuyama, M., et al., Cortical neurons responding to electrical stimulations of the rat's incisor pulp. *Brain Research* 67: 153-156, 1974.

- 46) Vyklicky, L., Keller, O., Brozek, G., et al., Cortical potentials evoked by stimulation of tooth pulp afferents in the cat. *Brain Research* 41: 211-213, 1972.
- 47) Chatrian, G., Canfield, R., Knauss, T.A., et al., Cerebral responses to electrical tooth pulp stimulation in man. *Neurology* 25: 745-757, 1975.
- 48) Kenshalo, D.R., Isensee, O., Responses of primate SI cortical neurons to noxious stimuli. *J. Neurophysiology* 50: 1479-1496, 1983.
- 49) Durinyan, R.A., Reshetnyak, V.K., Rabin, A.G., Effects of stimulation of somatic cortical areas in unrestrained cats. *Zh. vyssh. nerv. Deyat. Pavlova* 22: 109-115, 1972.
- 50) Zhang, D., Owens, C.M., Willis, W.D., The responses of STT neurons to electrical stimulation of cortex and cerebral peduncle. *Neuroscience Abstracts* 15: 151, 1989.
- 51) Owens, C., Zhang, D., Willis, W.D., Electrical stimulation of sensory cortex increases the noxious threshold of spinothalamic tract cells. *Neuroscience Abstracts* 15: 151, 1989.
- 52) Lynn, B., Cutaneous Hyperalgesia. *Brit. Med. Bull.* 331: 103-102, 1977.
- 53) Beck, P., Handwerker, H., Bradykinin and serotonin effects on various types of cutaneous nerve fibres. *Pflugers Arch.* 347: 209-222, 1974.
- 54) Besson, J.M., Conseiller, C., Hamann, K.F., et al., Modifications of dorsal horn cell activities in the spinal cord, after intra-arterial injection of bradykinin. *J. Physiol.* 221: 189-205, 1972.
- 55) Haley, J.E., Dickenson, A.H., Schachter, M., Electrophysiological evidence for a role of bradykinin in chemical nociception in the rat. *Neuroscience Letters* 97: 198-202, 1989.
- 56) Kanaka, R., Schaible, H., Schmidt, R.F., Activation of fine articular afferent units by bradykinin. *Brain Research* 327: 81-90, 1985.
- 57) Bures, J., Petron, M., Zachar, J., Electrophysiological Methods in Biological Research, Academic Press, New York, N.Y., 1967.
- 58) Phillips, C., Intracellular records from Betz cells in the cat. *Quart J Exp Physiol* 41: 58-69, 1956.
- 59) Guilbaud, G., Iggo, A., Tegnér, R., Sensory receptors in ankle joint capsules of normal and arthritic rats. *Exp Brain Res* 58: 29-40, 1985.
- 60) Schaible, H., Schmidt, R.F., Time Course of Mechanosensitivity Changes in Articular Afferents During a Developing Experimental Arthritis. *Jour. of Neurophysio.* 60: 2180-2195, 1988.
- 61) Schaible, H., Schmidt, R.F., Willis, W.D., Enhancement of the responses of ascending tract cells in the cat spinal cord by acute inflammation of the knee joint. *Exp Brain Res.* 66: 489-499, 1987.

- 62) Lamour, Y., Guilbaud, G., and Willer, J.C., Rat Somatosensory (SmI) Cortex: II. Laminar and Columnar Organization of Noxious and Non-noxious inputs. *Exp. Brain Res.* 49: 46-54, 1983.
- 63) Lamour, Y., Willer, J.C., and Guilbaud, G., Rat Somatosensory (SmI) Cortex: I. Characteristics of Neuronal Responses to Noxious Stimulation and Comparison with Responses to Non-noxious Stimulation. *Exp. Brain Res.* 49: 35-45, 1983.
- 64) Welker, C., Microelectrode delineation of fine grain somatotopic organization of SmI cerebral neocortex in albino rat. *Brain Res* 26: 256-275, 1971.
- 65) Woolsey, C.N., Organization of somatic sensory and motor areas of the cerebral cortex. In: Harlow, H., Woolsey, C.N., (eds.) Biological and Biochemical bases of behavior. University of Wisconsin Press, Madison, pp 63-81, 1958.
- 66) Armstrong-James, M., The functional status and columnar organisation of single cells responding to cutaneous stimulation in neonatal rat somatosensory cortex S1. *J. Physiol.* 246: 501-538, 1975.
- 67) Axelrad, H., Verley, R., Farkas, E., Responses evoked in mouse and rat SI cortex by vibrissa stimulation. *Neurosci Lett* 3:265-274, 1976.
- 68) Chapin, J.K., Woodward, D.J., Modulation of sensory responsiveness of single somatosensory cortical cells during movement and arousal behaviours. *Exp. Neurol.* 72: 164-178, 1981.
- 69) Ito, M., Some quantitative aspects of vibrissa-driven neuronal responses in rat neocortex. *J Neurophysiol* 46: 705-715, 1981.
- 70) Simons, D.J., Response properties of vibrissa units in rat S1 somatosensory neocortex. *J. Neurophysiol.* 41: 798-820, 1978.
- 71) Guilbaud, G., Peschanski, M., Gautron, et. al., Neurons Responding to Noxious Stimulation in VB Complex and Caudal Adjacent Regions in the Thalamus of the Rat. *Pain* 8: 303-318, 1980.
- 72) Freminet, A., Bursaux, E. et Poyart, C., Mesure de la vitesse de renouvellement du lactate chez le rat par perfusion de 14 CU (L) lactate. *Pflugers Arch. ges. Physiol.* 334: 293-302, 1972.
- 73) Hall, R.D., Lindholm, E.P., Organisation of motor and Somatosensory neocortex in the albino rat. *Brain Res.* 66: 23-38, 1974.
- 74) Welker, C., Woolsey, T.A., Structure of layer IV in the somatosensory neocortex of the rat: Description and comparison with the mouse. *J. Comp. Neur.* 158: 437-454, 1974.
- 75) Dykes, R.W., and Lamour, Y., An electrophysiological laminar analysis of single somatosensory neurons in partially deafferented rat hindlimb granular cortex subsequent to transection of the sciatic nerve. *Brain Res.* 449: 1-17, 1988.
- 76) Dykes, R.W., and Lamour, Y., An electrophysiological study of single somatosensory neurons in rat granular cortex serving the limbs: A laminar analysis. *Jour. of Neurophysiol.* 60: 703-724, 1988.

- 77) Hoheisel, U., Mense, S., Long-term changes in discharge behaviour of cat dorsal horn neurons following noxious stimulation of deep tissues. *Pain* 36: 239-247, 1989.
- 78) Wiesenfeld-Hallin, Z., Hallin, R.G., Persson, A., Do large diameter cutaneous afferents have a role in the transmission of nociceptive messages?. *Brain Research* 311: 375-379, 1984.
- 79) Fjallbrant, N., Iggo, A., The effect of histamine, 5-hydroxytryptamine and acetylcholine on cutaneous afferent fibers. *J Physiol Lond* 156: 578-590, 1961.
- 80) Guilbaud, G., Quelques données sur les bases neurophysiologique de l'effet analgesique de la morphine et des anesthésiques généraux. *Circulation et métabolisme du cerveau* 3: 5-16, 1986.
- 81) Lamour, Y., Guilbaud, G., Willer, J.C., Altered properties and laminar distribution of neuronal responses to peripheral stimulation in the SmI cortex of the arthritic rat. *Brain Research* 273: 183-187, 1983.
- 82) Wall, P.D., Waxman, S., and Basbaum, A.I., Ongoing activity in peripheral nerve: Injury Discharge. *Exp. Neurol.* 45: 576-589, 1974.
- 83) Armstrong-James, M., George, M.J., Influence of anesthesia on spontaneous activity and receptive field size of single units in rat SmI neocortex. *Exp. Neurology* 99: 369-387, 1988.
- 84) Chapin, J.K., Lin, C., Mapping the Body Representation in the SI cortex of Anesthetized and awake rats. *J. Comp. Neurol.* 229: 199-213, 1984.
- 85) Andersson, S. A., Cortical responses after spinal lesions and to tooth pulp stimulation. *Neurosci Res Program Bull* 16: 125-132, 1978.
- 86) Dohi, S., Kitahata, L., Toyooka, H., et. Al., An analgesic action of intravenously administered lidocaine on dorsal horn neurons responding to noxious thermal stimulation. *Anesthesiology* 51: 123-126, 1979.
- 87) Kaas, J.H., Merzenich, M.M., Killackey, H.P., The reorganization of somatosensory cortex following peripheral nerve damage in adult and developing mammals. *Ann. Rev. Neurosci.* 6:325-356, 1983.
- 88) Merzenich, M.M., Kaas, J.H., Wall, J.T., et al., Progression of change following median nerve section in the cortical representation of the hand in area 3b and 1 in adult owl and squirrel monkeys. *Neuroscience* 10: 639-665, 1983.
- 89) Metzler, J., Marks, P.S., Functional changes in cat somatic sensory-motor cortex during short-term reversible epidural blocks. *Brain Res.* 177: 379-383, 1979.
- 90) Wall, J.T., Cusick, C.G., Cutaneous Responsiveness in Primary Somatosensory (S-I) Hindpaw Cortex Before and After Partial Hindpaw Deafferentation in Adult Rats. *The Jour. of Neuroscience* 4: 1499-1515, 1984.
- 91) Levine, J.D., Dardick, S.J., Basbaum, A.I., et. al., Reflex Neurogenic Inflammation I. Contribution of the peripheral nervous system to spatially remote inflammatory responses that follow injury. *Jour. of Neuroscience* 5: 1380-1386, 1985.



FOR REFERENCE

NOT TO BE TAKEN FROM THE ROOM

CAT. NO. 23 012

PRINTED
IN U.S.A.

