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A

OPTOELECTRONIC MONITORING OF DISCRIMINITAVE WHISKING IN THE HEAD FIXED RAT

by

MICHAEL A. HARVEY

A dissertation submitted to the Graduate Faculty in Psychology
in partial fulfillment of the requirements for the degree of Doctor of Philosophy, The City
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Chapter 1

Introduction

A major goal of neuroscience has been to understand how the nervous system processes information about the external world, and uses this information to control movement. It has become popular to categorize the way in which information is acquired into passive and active processes. This distinction is at least partially due to the vast amounts of physiological research that relies upon the experimenter passively applying stimuli to an animal. One of the major suggestions of this division is that there is some fundamental difference in the processing of the two types of stimulation, with the assumption being that passive stimulation is somehow relieved of the integrative functions necessary for evaluation of input derived from the active movement of a receptor organ. In the real world, however, very few if any processes are truly passive in nature. Almost all sensory input must be evaluated in context of the state of the organism regardless of visible movement.

Movements of a receptor organ that are specific to certain stimulus properties should reflect the way in which these different properties are encoded, and identified by the sensory system in use, such that the particular motor pattern used for the observation would be optimized to activate the appropriate components of that sensory system When a stimulus is encountered that has a particular relevance to an organism it may be subjected to a more detailed examination, presumably resulting in a richer sensory representation of that feature. To this end animals have developed specific behavioral strategies in order to maximize the efficacy of their sensory explorations and observations. An analysis of such directed movements should reveal certain basic principles by which the nervous system operates in the acquisition and decoding of that sensory information.

Examples of movements directed at the acquisition of sensory information can be found in all sensory modalities. Sniffing, a snakes tongue flicking, and the antaenuation of insects are all examples of movements directed at the acquisition of olfactory information. In the visual system saccadic eye movements, the rocking and peering of locusts and the bobbing behavior of pigeons as they approach the ground for a landing serve to enhance perception of visual stimuli. Head turns and movements of the pina allow for many species to track and focus auditory information The most prominent of all sensory explorations, however, surely belong to the somatosensory system where objects are actively palpated and manipulated as animals decipher there particular characteristics.

Movement of the sensory organ across an object surface is critical in the somatosensory system where maximum resolution is attained by a lateral motion across an object surface, this differs from the visual system in which maximum resolution is achieved by the fixation of the fovea on an object, (Johnson, & Lamb, 1979). The differences in tactual discrimination that exists between a fixed touch of an object and the lateral scanning of an object have been attributed to the differential activation of subsets of afferent fibers under these two conditions. A fixed touch excites the slowly adapting fibers, providing the system with only the topographic pattern, and intensity dimensions of a surface. When the sensory organ, (e.g. finger or hand), is moved the slowly adapting fibers will provide temporal information, with additional input arising from the movement dependent responses of the rapidly adapting and pacinian afferents, (Ibid). Humans, when asked to make haptic identifications of different object properties, i.e. hardness, volume, or shape, typically use hand movements specific for each task (Lederman and Klatzky, 1996). This specificity might reflect the encoding of the various dimensions of the stimulus, including intensity (the overall number of spikes), temporal patterning (when the spikes occur) or isomorphic patterning (where the spikes occur on the receptor sheet). Well-organized patterns of movement could potentially maximize the resolution of any one of these properties; with a particular strategy manifesting itself once a relevant stimulus dimension has been established. For instance in a discrimination where the periodicity of the stimulus is critical, it may be beneficial for an animal to move its receptor at a constant velocity, thereby maintaining the temporal integrity of the stimulus, (Sinclair, & Burton, 1991).

The rodent vibrissa system One of the most highly conserved of mammalian somatosensory organs is the sensory hair. While almost all hairs are capable of transmitting somatosensory information the facial tactile hairs possess further specializations allowing them greater sensitivity and in some cases mobility. These facial hairs, or vibrissae, have larger follicles that are embedded in a blood sinus and receive a much richer innervation than a normal body hair. The mammalian facial vibrissae can be either mobile or immobile, with the former being seen in species such as canids, higher primates, and cetaceans. The mobile hairs may be further divided into sporadically moving, seals, sea lions, cats, etc. and rapid whisking, rodents and other small nocturnal animals. It is the fast, repetitive, and long lasting whisker movements that distinguish the whisking animals from others whose facial vibrissae tend to move slowly and whose movements terminate rather rapidly. It is unclear what purpose whisking subserves, but there are strong implications that it maybe a mechanism which allows the animal to enhance the resolution of tactile stimuli.

The rodent vibrissal system is particularly attractive the examination of sensorimotor systems, since each vibrissa is both an effector and a sensory organ, functioning as an element in a receptive array scanned across object surfaces. Input generated during the scan is used to control movements of the head, limbs and the vibrissae themselves. The vibrissae of the rat are arranged about its head in what Wineski (1983) described as a planar sensory field. The field is dynamic, as it is continuously changing its spatial relationship to the animal, and to the sensory world. Whisking is defined as the protraction and retraction of the vibrissae, with one such cycle being defined as one whisk, (Whelker, 1962). Protractions of the vibrissae are generated by the contraction of intrinsic mystacial pad muscles, which form a sling surrounding pairs of adjacent follicles of the same row. (Dorfl, 1982; Carvell et al. 1991). The subsequent retraction of the vibrissae is thought to be due to the relaxation of the intrinsic muscles, the elasticity of which return the whisker to its resting position. The mystacial pad into which the vibrissae are imbedded can be moved by contraction of the extrinsic pad musculature, with such contractions leading to a change in position of the entire array of whiskers. The amplitude of whisker protraction can be very small, a few millimeters, or quite large, up to several centimeters, (Carvell & Simons, 1990).

Anatomy and physiology of the vibrissa system

The central representation of the vibrissae is somatotopically patterned at all levels of the trigeminal neuraxes, with the cortical barrels, thalamic barreloids, and brainstem barrelettes being the visible consequence of this somatotopy. In each case it is the clustering of the afferent projections, be they thalamocortical, lemniscal, or peripheral that form these structures. The individual clusters, in all cases, are isomorphic with the patterning of the vibrissae on the animal's snout, and are considered to be uniquely activated by their corresponding vibrissa.

Each vibrissa is innervated independently by up to 100 myelinated fibers, which project to the trigeminal ganglion. Trigeminal ganglion afferents, in turn send collaterals to the principal nucleus of the fifth trigeminal nerve (PrV), and the spinal nucleus of the fifth trigeminal nerve (SpV). In PrV, and in portions of SpV individual vibrissae are somatotopically represented by dense clusters of afferents terminals, (Rice & Muenger, 1986). Aggregates of these clusters are called barrelettes. Projections from PrV and SpV ascend via the lemniscal and paralemniscal tracts respectively, to the ventral posterior medial nucleus (VPM), and the rostral portion of the posterior complex (POM). The proportion of trigeminal cells that terminate in POM and VPM are divided unequally between cells arising from PrV, and SpV. In POM there is dominance by paralemniscal afferents, and in VPM by lemniscal afferents, (Chiaia et al., 1991). The targets of SpV and PrV within VPM differ as well, the lemniscal projections targeting a tight focus of cells in the barreloid region, while paralemniscal projections terminate in the extrabarreloid septa, and show patchy anterograde labeling indicating that their axons quite small in diameter, (Williams et al., 1994).

Cells in the barreloid region of VPM show center surround response characteristics, with a given cell being driven primarily by single vibrissae, and to a lesser degree by 2 or 3 adjacent vibrissae generally in the same row, (Welker, 1971; Waite, 1973). These receptive fields are extremely sensitive to the level of anesthesia used during recording, with the contribution of the RF surround shrinking with increasing depth of anesthesia. POM cells, however, are driven equally well by any one of several adjacent vibrissae, with the best stimulus being the consecutive stimulation of vibrissae in a row, (Lee et al., 1994)

The receptive field properties of VPM cells can be altered by eliminating the input which arises from PrV neurons in the trigeminal brainstem nuclear complex, resulting in these VPM neurons adopting multiple vibrissae receptive fields, comparable to neurons in POM, (Diamond, et al., 1992). Lesions of TRN also produce VPM cells with expanded RF's, which are similar to the expanded RF's seen in PrV lesions. Subsequent lesion of the spinal subnucleus, interpolaris (SpVi), restores the RF's to their original size, indicating that in a normal animal responses from surround cells may be enhanced by SpV afference, with inhibition of this activity generated by TRN. The relief of this inhibition, through lesion of TRN expands the surround, and the subsequent lesion of SpVi shrinks it again, (Lee et al. 1994). The presence of single whisker receptive fields in VPM is indicative of a high-resolution system, which would allow an animal to differentiate inputs received by several vibrissae simultaneously.

Both POM and VPM project to somatosensory cortex (SI) to a region that is characterized in some species of rodents by characteristic assemblages of neurons called "barrels". Cortical barrels are roughly circular structures comprised of cell dense sides, and cell poor hollows, (Woolsey & Van Der Loos, 1970). The relative cell density of the hollows differs markedly among species. In rat SI that has been stained for nissel substance, the barrels are manifested as solid, round patches. Therefore rats are said to have solid barrel architecture. Thus far there has been no good explanation for the existence of a barrel field, which was initially thought to be a specialization brought on by the presence of mystacial vibrissa. However comparative studies have revealed several species that possess vibrissa, but do not show barrel architecture. Similarly there has been no correlation found between animals that actively whisk, and those that have barrels. It is possible that the barrel field is simply an exaggeration of a design present in all species, (Woolsey, et al., 1974).

Specific Aims

The observing behavior of an animal during the solution of a sensory task provides a unique window onto the neural mechanisms by which that task is solved since different observing responses result in different patterns of neural activity that are critically related to the animals perception. Observation of the development of task specific patterns of whisking depends upon our ability to make high-resolution measurements of vibrissal position over time, and our ability to control precisely the receptor sheet to which the stimulation is applied. Similarly in order to make accurate assessments of the correlation between a successful trial, and the presence of a specific whisking strategy, we must be able to resolve the types of whisks made on a per trial basis. This work was aimed specifically at the development of behavioral preparations, which allow for detailed examinations of whisker movements during the acquisition of tactile discrimination tasks, restrict access to the tactile discriminanda to the mystacial vibrissae on one side of the animals head (allows for within animal controls), eliminate confounds due to head and body movements, separate the observing and conditioned response, and are compatible with electrophysiological methodologies. To meet these requirements we have utilized a head fixed preparation, and optoelectronic transduction systems which allowed us to isolate and monitor a single vibrissae in real time, with high spatial-temporal resolution while permitting precise control over the stimuli. One aim of this Dissertation is to examine the development of whisking strategies under different stimulus conditions, so as to correlate the presence of a particular strategy, with the solution of a particular tactile task. A second is to begin to characterize the relation between "barrel cortex" and whisking in the rat. We do this by examining the effects of "barrel-cortex" ablations upon the rat's *unconditioned* whisking behavior. Taken together, the two experiments lay the foundation for systematic studies of the contribution of the "barrel cortex" to the rodent's discriminative whisking behavior.

Chapter 2

Tactile Detection and Discrimination

Materials and Methods

Subjects

The data presented in this report were obtained 8 male and female rats of the Long-Evans and Sprague-Dawley strains, ranging in age from three months to a year. [A number of additional animals were used but were discarded, either because they lost their head mounts or failed to acquire and/or maintain stable performance on the operant ever-pressing task]. Animals were housed individually under a 12:12 reversed light-dark cycle. During testing, subjects were water-deprived and maintained at 80-85% of free-feeding weight, by a 23.5 hr. water deprivation schedule with food available ad libitum.

Surgery

Subjects were anesthetized with Ketamine/Xylazine (100 mg/kg, i.p./5.5 mg/kg, i.m). With the head held in a stereotaxic device a midline incision was made in the scalp beginning at 1 cm anterior to bregma. The periosteum was retracted and pushed down to the two sides of the skull. Six self-tapping stainless steel screws (Small Parts #Q-TX0-2) were used as anchors for a dental cement platform. Screws were inserted to a depth of 1-½ turns. Four screws were placed on the top of the skull approximately 3 mm lateral to the midline and 5 mm rostral or caudal to the bregma. The remaining two screws were inserted into the two sides of the skull. A mounting screw (Small Parts #Q-TSB-632-12) was embedded in the central portion of the dental cement platform above bregma. The incision was closed with a single suture at its caudal portion to minimize irritation of the incision edges.

Apparatus

Training and testing were carried out in a soundproof chamber (Industrial Acoustics: interior dimensions 80 x 60 x 60 cm). The chamber contained a gravity-driven water delivery system, a white-noise generator, a tone generator (Radio Shack #273-074A, 2.5 kHz), a stepping motor (Alpha Products) optoelectronic monitoring devices ((PAS 11L, laser micrometer, Hama Laboratories, Palo Alto, CA. 94306) and a CCD camera. Illumination for closed-circuit video observations was provided by an LED emitting infrared illumination at a wavelength (950 nm) beyond the visible spectrum of the rat (Rosenberger and Ernest, 1970; Messing RB, 1972). The water delivery system, stepping motor and optoelectronic devices were connected to a personal computer (486 PC) via a standard A/D card (Alpha,). Specially written QuickBasic programs were used to control stimulus presentation, reinforcer (water) delivery and data storage. Figure 1 illustrates the testing situation.

Optoelectronic transduction of individual vibrissa movements

Whisker movements were monitored in the rostro-caudal plane using an optoelectronic system (laser-emitter and detector) which has been described in detail elsewhere (Bermejo, et al. 1998). Briefly, interruption of the emitted beam by the shadow of a whisker produces a voltage shift in a subset of shaded sensors (CCDs). Whisker movement trajectories produce successive displacements in the position of that voltage shift, which are linearly related to whisker position. A comparator circuit identifies the

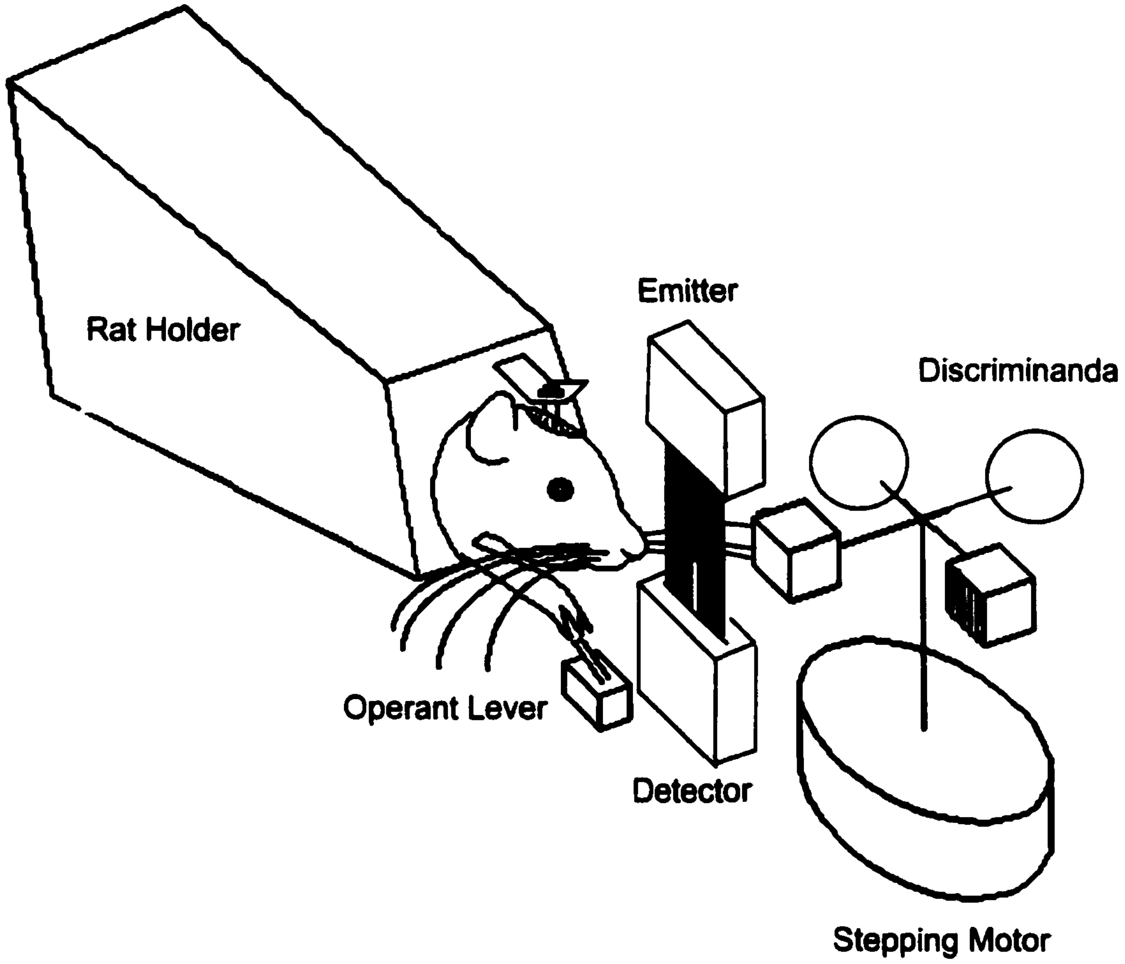

Figure 1 Monitoring Of Individual Vibrissae Movements During A Tactile Detection Task
Illustration of Experimental setup showing the rat inside of a V shaped restraining device. The animal's head is fixed to a metal bar protruding from the top of the restrainer, and his paw rests on a microswitch used to measure lever presses. A turntable rotates discriminanda into the path of the vibrissae. The visibility of one whisker with respect to the others in increased by the attatchment of a light, foam marker. As the marked whisker interrupts the beam, a voltage shift in the detector is registered on a PC.

successive positions of voltages above a preset threshold and outputs the data to a microprocessor for computation and display of the trajectory. (The device has a resolution of 1.4 ms; 26 mμ). To monitor an individual whisker trajectory with the other whiskers present light 3-4-mg foam marker is glued along the side of the C1 whisker. This increases its "visibility" with respect to surrounding vibrissae without significantly affecting whisking kinematics (Bermejo, et al., op.cit). In this study, only movements of the left C-1 whisker were monitored, but all whiskers were present.

Calibration

In order to transform data on sensor locations into a record of angular whisker positions, a calibration procedure is carried out for each animal at the start of each recording session. The whisker is manually positioned at 90° from the horizontal; i.e., perpendicular to the animal's snout. The detector is placed so that the whisker shaft intercepts the CCD array at its mid point, and at a fixed distance (ca. 10 mm) from the whisker base. The position of the CCD (e.g., 1-2496) which is intersected by the shadow of the whisker at its initial (i.e., 90°) position is recorded. The angular displacement of the vibrissa may then be calculated using the formula ArcTan (θ) = Opposite/Adjacent, where the opposite is the distance moved along the CCD array and the adjacent is some known distance. The procedure is illustrated schematically in Figure 2. Note: Since the whisker moves in an arc, the point on the vibrissa shaft that interrupts the beam at the beginning of the trajectory will not be identical with the point interrupting the beam at its end. This could result in an overestimation of the distance traveled by the whisker. To compensate for this we oriented the CCD array in parallel with the animal's face. Since the kinematic

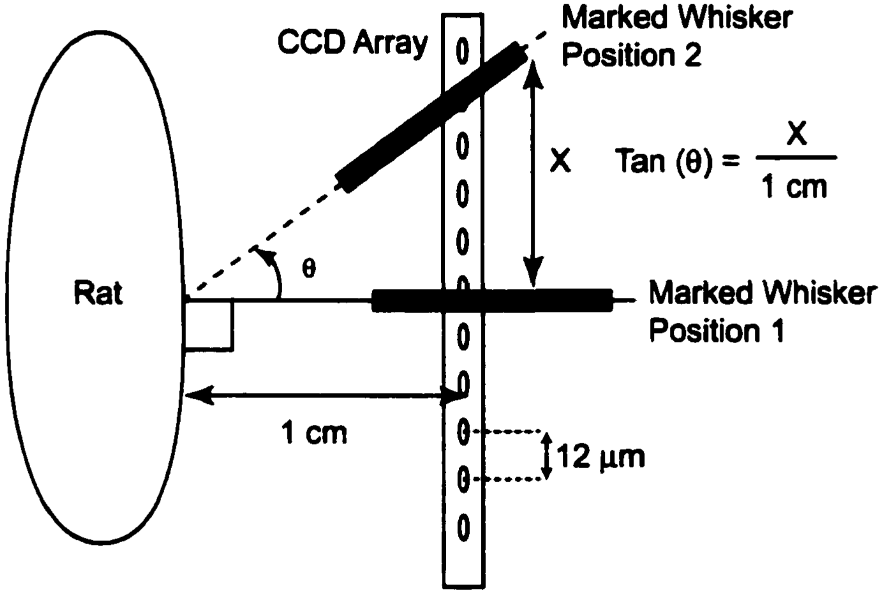

Figure 2 Calculation of Whisking Amplitudes
Whisking amplitudes are calculated by first calibrating the system such that the the CCD array is 1 Cm from the from the base of the whisker when that whisker is 90° to the animals face. The tangent of the triangle can then be used to calculate the angular displacement.

properties of the trajectories measured using this procedure were comparable to those reported using videographic methods (see below: RESULTS), we believe that minor errors introduced by the procedure did not significantly bias the results.

Behavioral Testing

For testing, the animal's body movements are constrained and its head fixed in position using a V shaped device made of plexiglass and containing a metal bracket to which the head mounting screw is fixed (Bermejo, et al. 1996). A microswitch attached to the platform of the restraining device served as an operant lever. During testing, the restraining device was fixed in place in the testing chamber and the stepping motor was used to for stimulus presentation.

An operant lever pressing response served as the *indicator response.* Head-fixed animals were initially hand-shaped to press the lever for reinforcement, (water, in 40 μl aliquots) delivered initially on a Continuous Reinforcement Schedule. Subsequently, acquisition of the lever pressing response was achieved by gradually increasing the ratio of lever pressing responses/reinforcement (Fixed Ratio10). Animals stable on the FR schedule were switched to a Variable Interval schedule, in which reinforcement was delivered for the first response following a predetermined time interval. When stable, moderate response rates were obtained on a VI 20' schedule, subjects were trained either to detect the presence of a 2 x 2 x.5 cm acrylic cube faced with No. 8 sandpaper, or to discriminate between the same acrylic cube and an acrylic sphere, 1cm in diameter. The two discriminanda in this task differed in size, surface texture and shape, In both tasks the stepping motor was used to rotate tactile discriminanda into position parallel to the vibrissae on one side of the snout. The stimuli were positioned such that whisker contact was possible only when the whiskers were protracted, but they were within reach of even low-amplitude whisking movements.

Detection or discrimination training was conducted in 40-minute daily sessions using a two-component (reinforcement/extinction) schedule in which the presence or absence of a reinforcement (delivered on a VI 30' schedule) was associated with a specific stimulus condition (S+/S-). A variable interval schedule was chosen to provide a stable, slow rate of responding, as well as to keep the animal from using the water as a discriminant. Individual components were created in a quasi-random fashion with no component being shorter than the maximum VI. S+ and S- components were then matched and presented randomly, with 20 components for each condition. So an individual session consisted of 40 components, 20 S+ and 20 S-. Trials could end without reinforcement; however since there was never a component shorter than the maximum VI, reinforcement was available in all S+ trials.

These conditions effectively transformed the operant VI schedule into a successive (Go/NoGo) discrimination paradigm. Stimuli were presented in a pre-arranged, quasi-random order, for discrete intervals of variable length. There was no constraint on the number of consecutive S+ or S- trials. However there were always an equal number of components for S+ and S-, namely 20. Trial order and duration were randomized, with the shortest trial exceeding the longest possible Variable Interval. Within trials, the VI schedule determined the occurrence of reinforcements. Trials could end without reinforcement. However, since there was never a component shorter than the maximum VI, reinforcement was available in all S+ trials.

In the detection task, subjects were reinforced for lever presses in the presence of the cube (S+) and extinguished in its absence (S-). In the discrimination paradigm subjects were reinforced for lever pressing in the presence of the sphere (S+) and extinguished in the presence of the cube (S-). For both tasks, the ratio of S+/S- responses was calculated at the end of each test session. Under these conditions, it is possible that animals could press at a low rate until delivery of a reinforcement, and then emit a burst of responses, artificially elevating the percentage of correct responses. To control for this possibility, we also recorded the latency from the time of stimulus presentation to the first lever pressing response. Criterion performance on both tasks was defined as 80% correct responses for 3 successive sessions. Subjects who achieved criterion were either transferred to a different task served in one of several control procedures.

Control Procedures

Several features of the experimental setup were designed to insure that solution of the detection and discrimination tasks were mediated by tactile (whisker) inputs, rather than by other sensory cues. Visual cues were eliminated by running the animal in a darkened chamber, illuminated (for videographic purposes) by an infrared LED at a wavelength outside the rat's spectral range. To control for cues from rotation of the stepping motor which presented the stimuli, the motor had four stopping positions. The four stimulus positions were arranged such that a 90 degrees rotation of the stepping motor could take the animal into either S+ or S- component, regardless of the current position of the motor. This was done so that the animal could not use differences in the duration of motor operation as an auditory cue. In addition, two animals that had achieved criterion were tested with the stepping motor programmed normally but the discriminanda absent. In three Detection subjects that had met criteria using the left whisker array, several training sessions were carried out in which the discriminanda were presented to the whiskers on the right side of the face to test for transfer. After reacquiring criterion, these animals were placed on a texture discrimination task that required them to discriminate between rough (Norton #47750/60 grit: Course) and smooth (Norton # 44710/-220 grit: Very Fine) surfaces.

Data Collection and Analysis

The basic epoch of data selected for analysis was defined by the presentation of the stimulus at the start of each individual trial. Data on lever pressing was saved throughout the extent of each trial. For trials longer than 24 seconds, data on vibrissa movements was saved for the initial and final 12 seconds of the trial—including whisks made during the reinforcement period. The top portion of Figure 3 presents a record of whisking and lever pressing responses recorded during a single trial on the discrimination task. For analysis, the transformed whisking data for each test session were plotted in angular coordinates and displayed on a computer monitor as a plot of whisker position against time. A specially written, cursor-driven program was used to display individual data epochs. Individual whisks were selected for analysis on the basis of their general shape (i.e., a protraction followed by a retraction). Once selected, a specially written algorithm identified critical points defining the start, peak and end of the whisk—the peak being defined simply as the maximum forward position attained prior to retraction—and calculated the values for specific kinematic parameters (e.g., whisk duration, peak

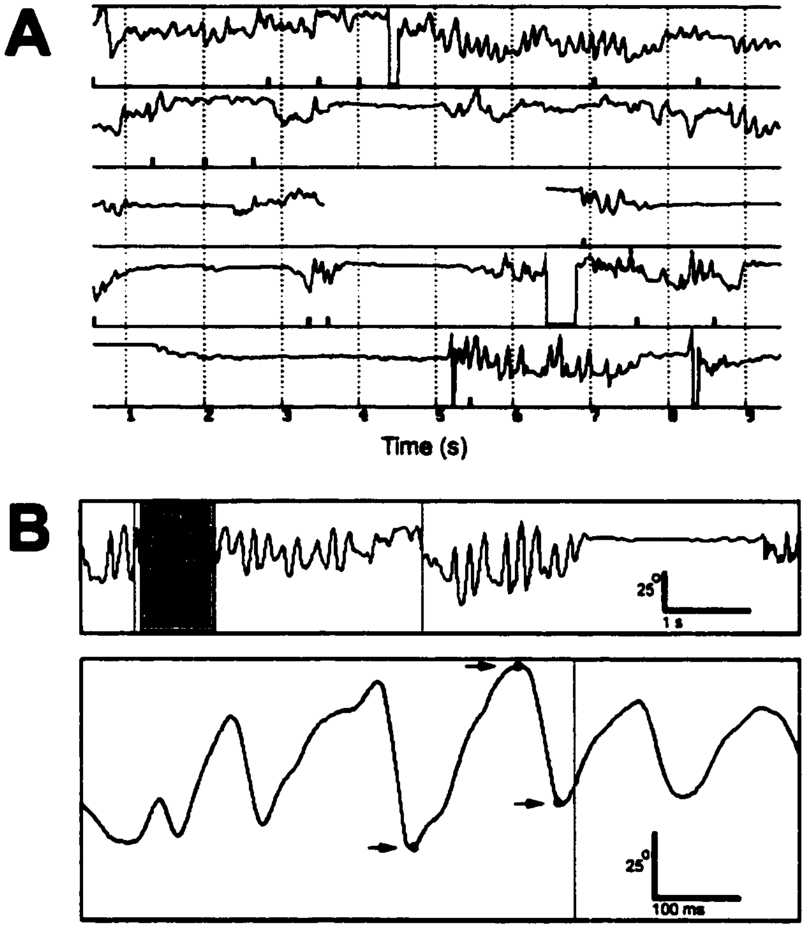

Figure 3 Optoelectronic Monitoring and Data Analysis of Real Time Whisking

A. A trace of whisking from a single trial in an animal performing a tactile discrimination task. Hash marks below the trace indicate lever presses, and the break in the middle indicates a period of unrecorded time during the trial. B In the top portion of this figure is a low resolution viewof a 7 second epoch of whisking with lever pressing. The grey box is the area selected for analysis and is displayed below in a higher resolution. Arrows point to the start, peak,and end points of a whisk selected for kinematic analysis. Grey vertical bars reflect lever presses in both the upper and lower portions of the figure.

amplitude, velocity) as well as the number of whisking responses. The bottom portion of Figure 3 illustrates the application of this program to the selection and analysis of a subset of whisking data on numbers of lever presses, latency to first lever press, number of whisking movements and derived kinematic values were saved and exported to a standard spread sheet program, for further computations, graphic presentation and statistical analysis. The values for the kinematic parameters were measured both for total trials (S+/S-) and (separately) for S+ and S- trials. Time series data were analyzed using a Fast Fourier analysis [FFT] of 45 s blocks of whisking. The results presented in this report reflect events over the course of task acquisition for individual animals. They are based upon an analysis of approximately 50,000 whisking responses (ca. 5,000,000 individual data points) obtained from eight animals.

Results

Detection: Task acquisition

Five of the seven animals reached criterion performance on the detection task within 30 days. Good performance on the task was indicated both by the differential rate of responding on the S+ component of the schedule and by differences in the latency to the first response on a given trial. Figure 4 presents cumulative records of lever pressing on a Variable Interval task (A) prior to the introduction of the discrimination paradigm and (B) after achieving criterion performance on the detection task. Note that lever pressing appears to be under good stimulus control, with little or no lever pressing on the S- trials, and an immediate increase in responding at the onset of the S+ trial.

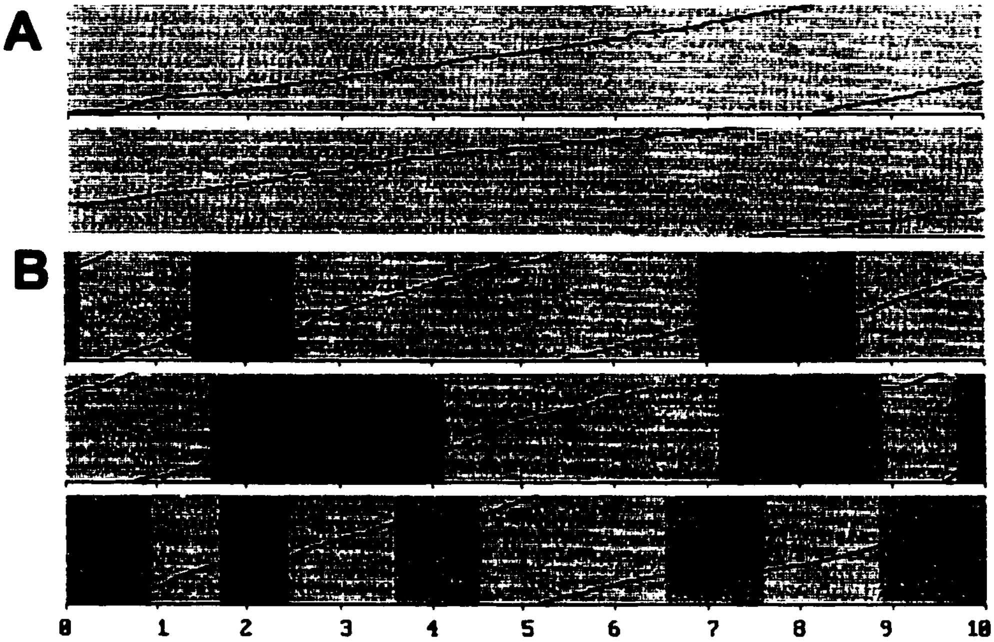

Figure 4 Cumulative Records of Lever Pressing Before and After Learning of a Tactile Detection Task: Rat 704
A. Cumulative record of lever pressing during a VI20 schedule of reinforcement. B. Cumulative responses after the animals has reached criterion performance on a tactile detection task. Dark areas represent S- trials and the lightly shaded areas represent S+ trials. In both A and B cumulative records reset after 70 responses.

The left hand portion of Figure 5 presents learning curves for three representative animals on the detection task. Two of the animals (704, 705 achieved criterion performance; the third (706), though obviously responding at a higher rate on the S+ trials, did not. The right hand portion of the figure plots, for these animals, the mean latency to the first lever press in each schedule component. The two successful animals show a marked divergence in latencies on the S+ and S- components which is associated with performance changes during the course of acquisition, but which is not seen in the unsuccessful animal.

Insert Figure 5 (Left and Right) here

Detection: Whisking behavior

To assess changes in whisking behavior accompanying acquisition of the detection task, data on whisk duration, mean protraction amplitude and peak velocity, and number of whisks were analyzed for each of the three rats whose learning curves are presented in Figure 5. For all measures, the analysis was based upon the first, middle and last three sessions. Only one of these measures (number of whisks) varied significantly over the course of training. The animals achieving criterion performance consistently emitted substantially (30-40%) more whisks on the S- trials over the course of task acquisition (Figure 6). A Fourier analysis of whisking across sessions based upon 45 sec. of whisking data from each session, failed to reveal any significant trends in whisking frequency.

To relate whisking movement parameters to performance on the indicator (lever pressing) response we constructed a trial-by-trial analysis of the last session of training for all three animals. Correlations were calculated separately for S+ and S- trials. Once

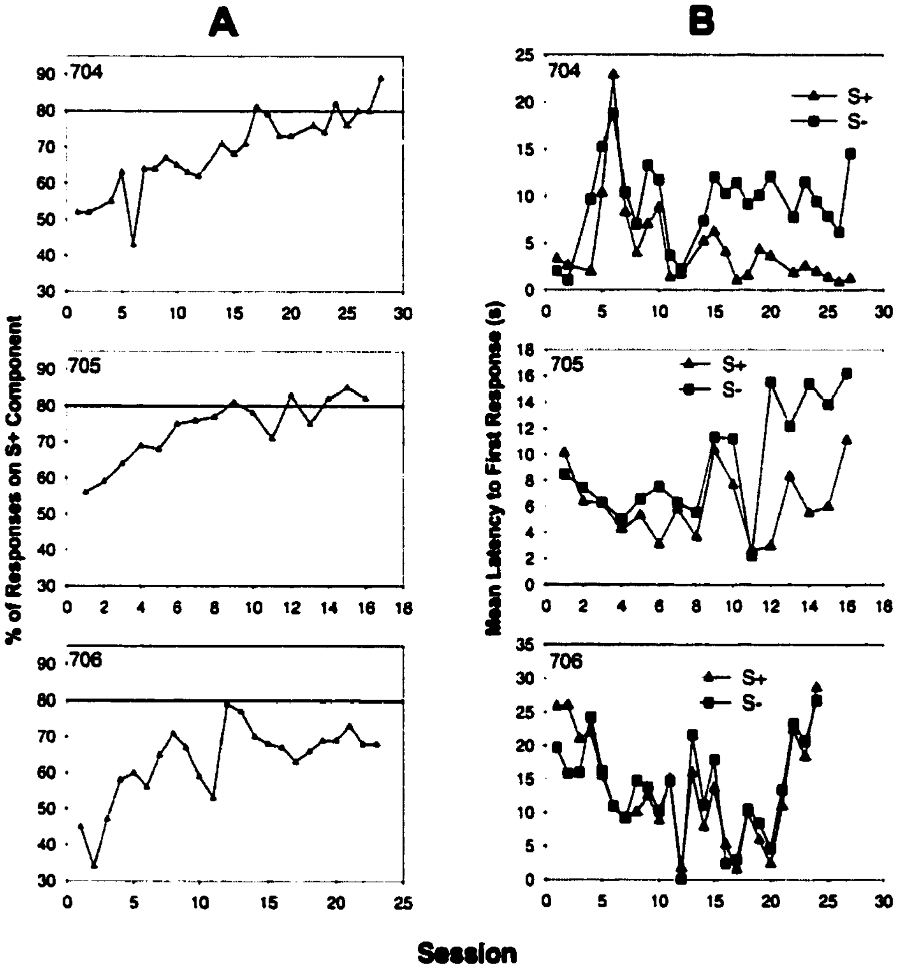

Figure 5 Acquisition of a Tactile Detection Task
Learning curves for three animals are shown in A. The Horizontal line at 80 percent correct responses indicates criterion performance. The latency to first lever press is shown in B for the same three animals. In both A and B the top two panelsshow animals which achieved criterion performance and the bottom pannel shows an animal which failed to learn the task.

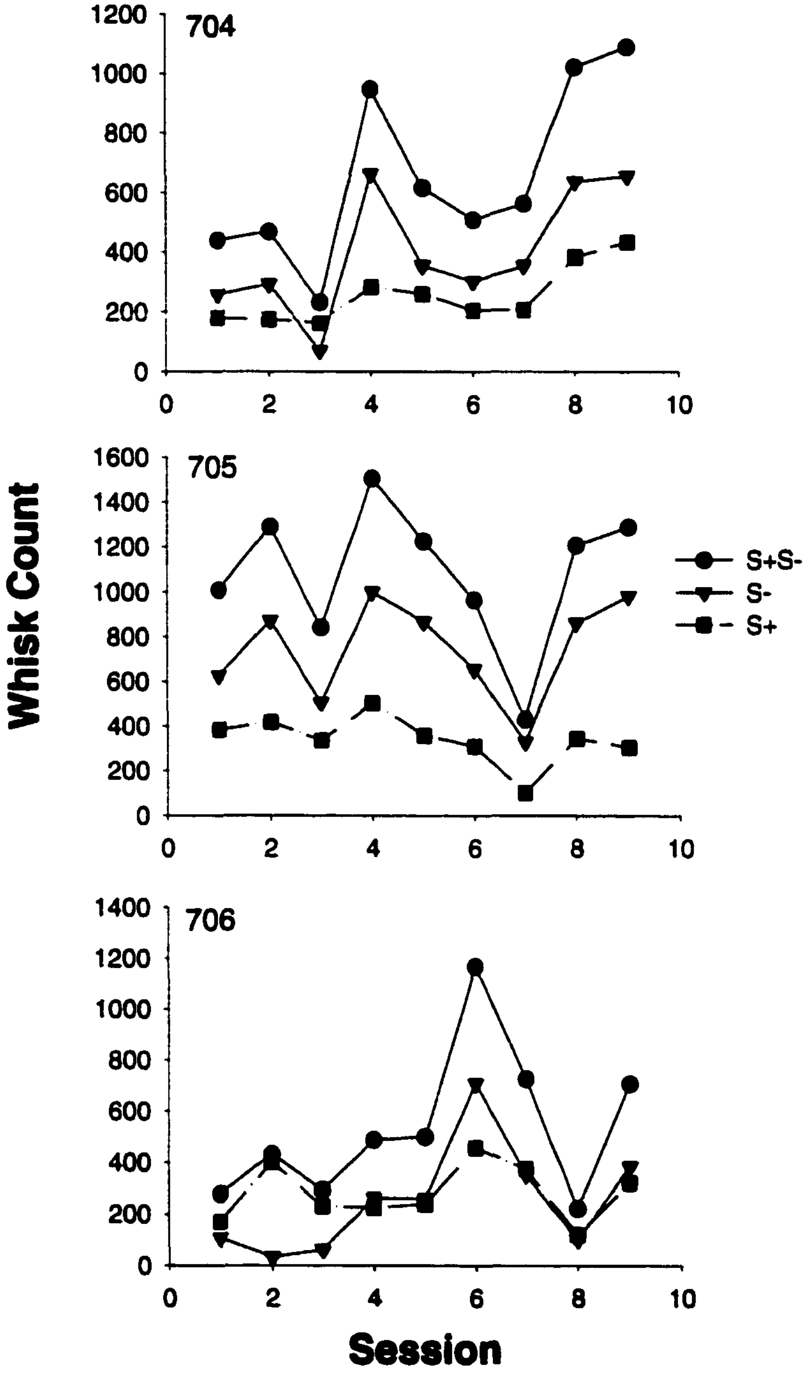

Figure 6 WhiskCount vs. Session During Tactile Detection

Over all numbers of whisks for the first, middle, and last three sessions of training are plotted for three animals. Counts are over all trials, (S+S-), and also within each condition.

again, only the amount of whisking was significantly correlated with a performance variable (lever pressing). For Rat 704, these correlations were significant (p: < .01) only on S+ trials. For rat 705 they were significant for both S+ trials (p < .004) and S- trials (p < .001). Lever pressing rates for the last session of Rat 706 were so low that meaningful correlations could not be calculated.

Discrimination: Task acquisition

Of four animals which entered the study, one lost its head mount in the middle of training, two achieved criterion (801 and 825), and one (821) failed to achieve criterion after two months of training. Performance data on the discrimination task is presented for these three animals in Figure 7. Note the striking difference in acquisition time between 801 and 825, and that the performance of 821, though highly variable, remains consistently above chance level for the last month of testing. As with the detection animals, acquisition of the discrimination was associated with a decreased in the latency to the first level press on S+ trials for the two successful animals. .

Discrimination: Whisking behavior

Acquisition of the object discrimination task was associated with changes in several whisking movement parameters. As in the detection task, the successful animals emitted more whisks (ca. 30%) on S- trials during the final third of the sessions. All three rats showed a decrease in protraction amplitude from the first to the final sessions, and the difference was highly significant for the two rats that achieved criterion [801, 825: $p < .001$; 821: $p < .05$). Both successful animals showed a decreased mean whisking velocity over the course of acquisition, which was due, primarily to a reduction of about 20% on

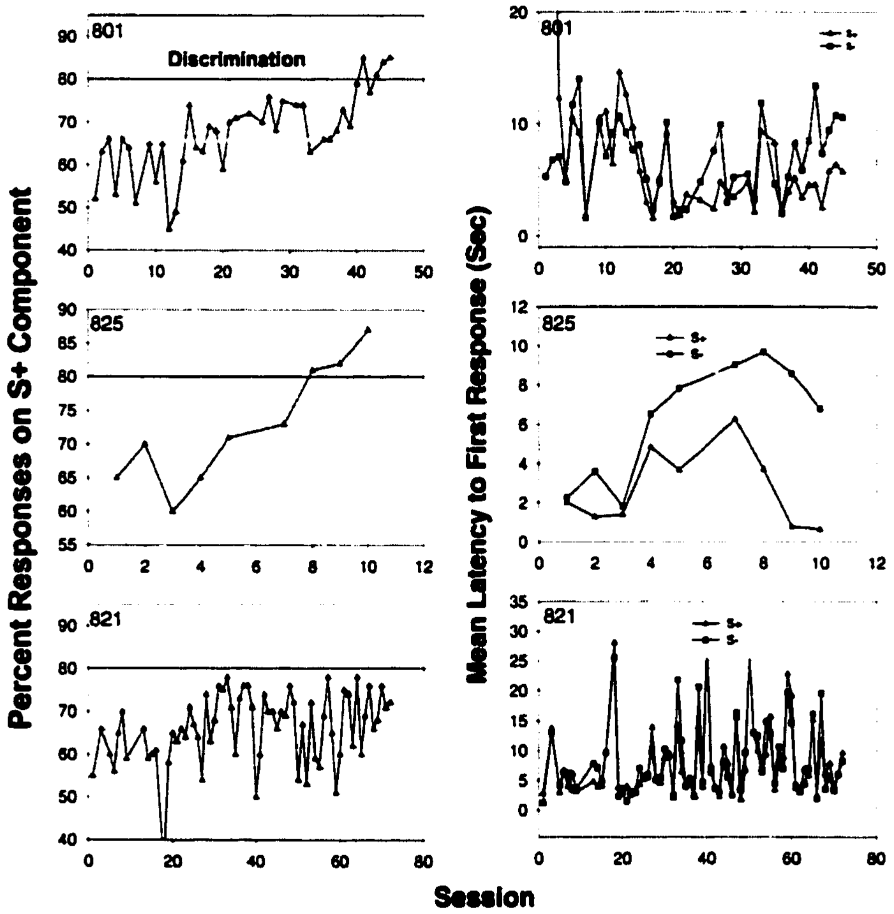

Figure 7 Acquisition of a Tactile Discrimination Task
Learning curves for three animals are shown in A. The Horizontal line at 80 percent correct responses indicates criterion performance. The latency to first lever press is shown in B for the same three animals. In both A and B the top two panelsshow animals which achieved criterion performance and the bottom pannel shows an animal which failed to learn the task.

the S+ trials. Moreover, both animals that achieved criterion showed a significant decrease in whisk duration from the first to the final training session (801: $p < .001$; 825: $p < .001$). The reduction in whisk duration seen in the two successful rats was consistent with a shift towards higher frequencies in the whisking power spectra of those animals (Figure 8) but not in those of the unsuccessful animal.

A trial-by-trial analysis of the correlations among whisking movement parameters and lever pressing rates was carried out for S+ and S- trials during the last test session in all three subjects. For rat 801, correlations were positive and significant on the S+ trials for the number of whisks ($p < .007$), for whisk durations ($p < .0003$) and whisk amplitudes ($p < .05$). For rat 825, they are significant only for number of whisks ($p < .0009$). For rat 821, which did not achieve criterion, there are significant negative correlations (on S+ trials) between the rate of lever pressing and the duration ($p < .004$) and amplitude ($p < .008$) of whisking movements.

Figure 9 summarizes the relation between whisking movement parameters (kinematics) and performance over the course of task acquisition for one detection (Det) and one object discrimination (OD) animal, each of which had attained criterion performance on the task. The figure plots the session means of each of the kinematic variables (duration, amplitude, velocity, number of whisks) as well as a regression line. Significance levels for each kinematic variable were obtained using an ANOVA. Note that performance on the detection task is significantly correlated only with the number of whisks emitted, while performance on the detection task is correlated both with shorter whisk durations and smaller protraction amplitudes.

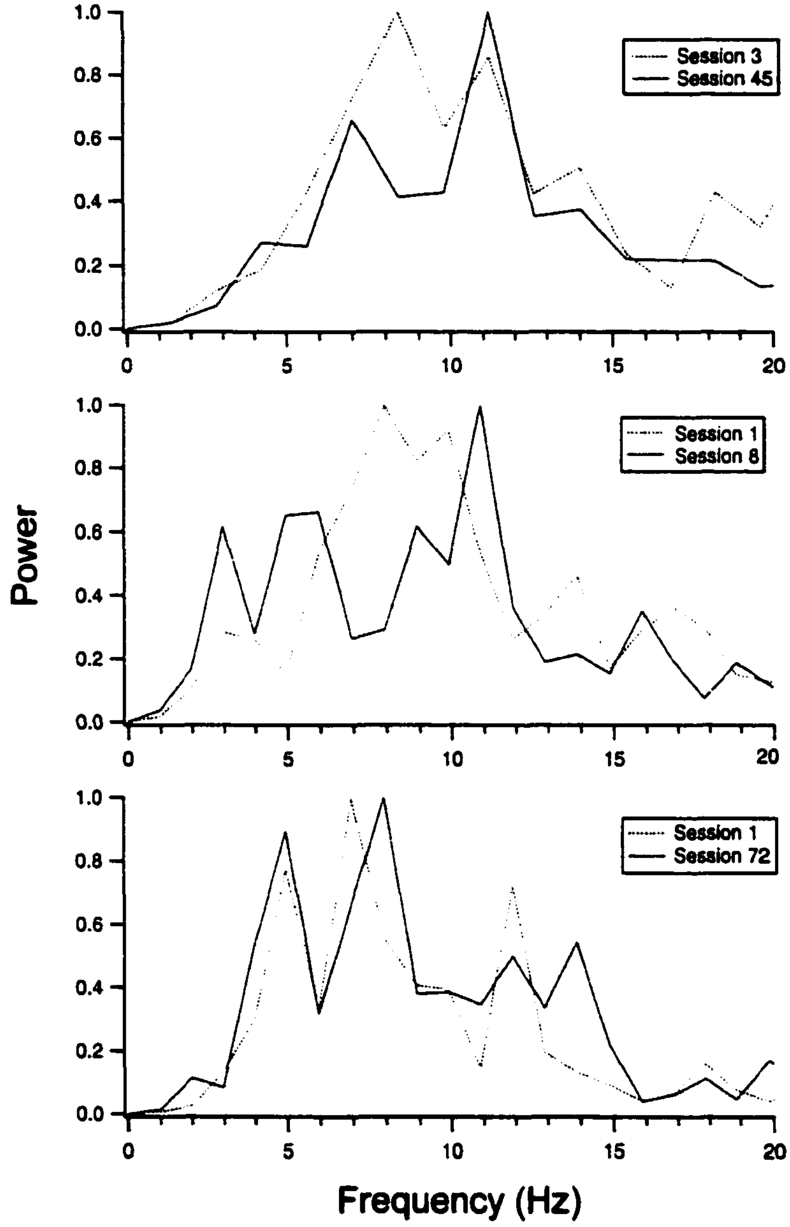

Figure 8 Power Spectra of Whisking Frequencies During Tactile Discrimination The top two panels show power spectra for the first and last sessions of training forin two animals that reached criterion performance on a tactile discrimination task. The bottom panel shows the same data for an animal which failed to acquire the task.

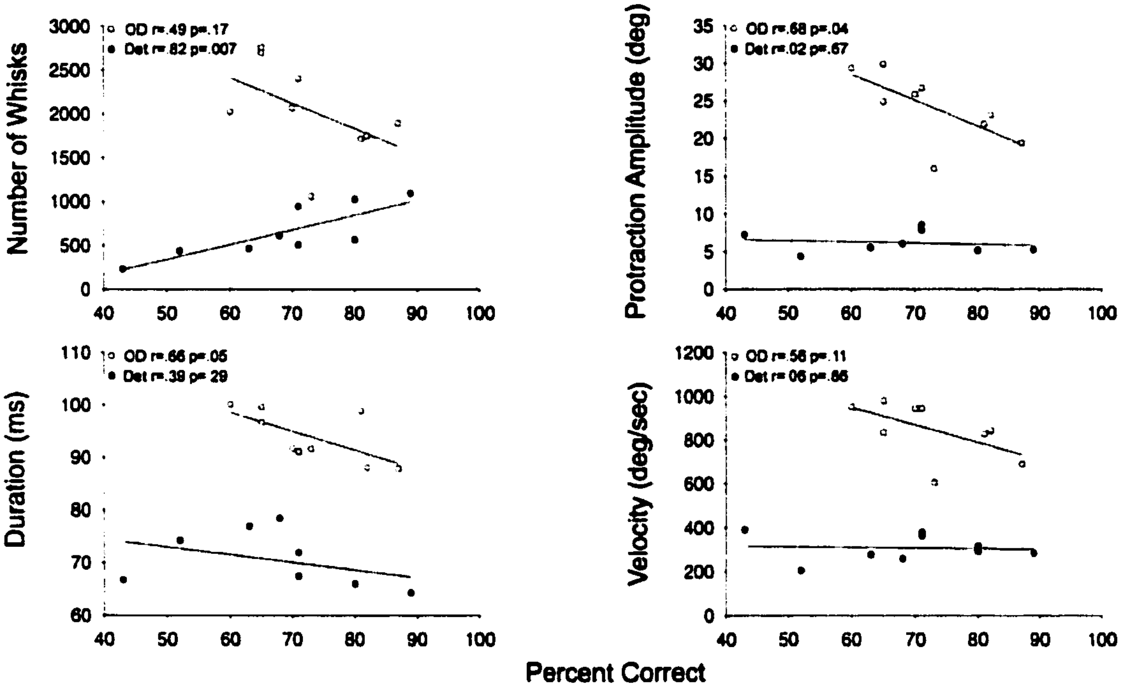

Figure 9 Relationships Between Whisk Properties and Perfromance in Tactile Detection and Object Discrimination

Scatter plots show significant relationships between whisk duration and protraction amplitude in the object (OD) discrimination task. The only significant correlation in the tactile detection (Det) task is that between number of whisks and performance.

Control procedures

In two detection animals that had attained criterion performance, the stimulus was removed from the stepping motor prior to the session, and the trials then proceeded normally These animals continued to make lever presses during the first few trials and then gradually stopped responding. In three additional animals which had achieved criterion, stimulus presentation was shifted from the whiskers on the left to those on the right. This transfer was followed by an initial decrease in performance, but all three animals regained criterion performance within 2 sessions. These animals were then shifted switched to a texture (rough/smooth) discrimination. A precipitate drop in differential responding followed transfer to the new task, and performance remained at this level for several weeks. Data for two of these "transfer" animals are presented in Figure 10.

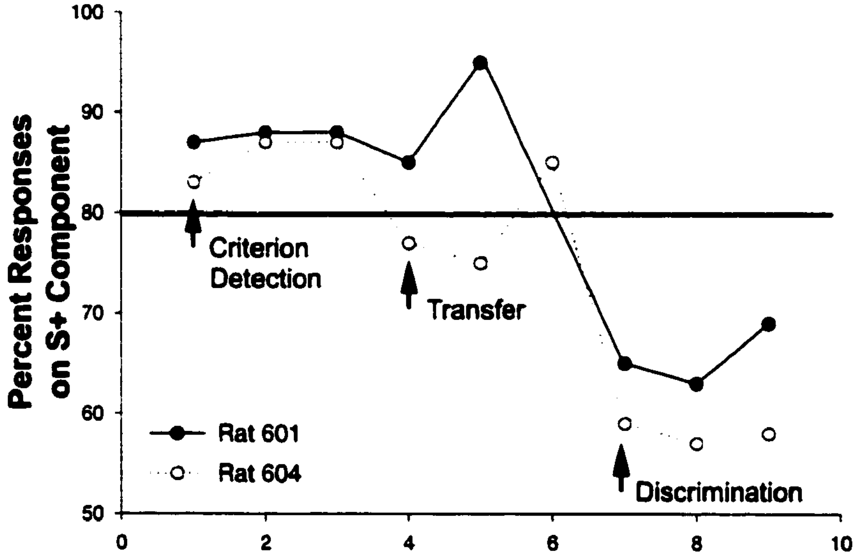

Figure 10 Transfer and Transition to Discrimination After attaining criterion performance on tactile detection the side of the face to which the stimulus is presented is transferred from right to left. Following reacquisition of criterion, animals are switched to a rough/smoothe discrimination. The horizontal line at 80% correct responses indicates criterion performance.

Chapter 3

Effects of Cortical Barrel Field Ablation Upon Passive Whisking

The rat's mystacial vibrissae function as sensorimotor elements in an "active touch" system. At rest and during periods of exploration, mystacial vibrissae exhibit a rhythmic pattern of alternating protraction and retraction movements--whisking, (Welker, 1964). These movements generate patterns of somatosensory input that are used to guide discriminative behaviors, and, recursively, to control patterned movements of the vibrissae themselves (Carvell and Simons, 1990). The representation of the mystacial pad occupies a substantial portion of the rat's cortical somatosensory area, and the vibrissae are represented within that region by discrete neuronal aggregates ---"barrels"—(Woolsey and Van der Loos, 1970), whose spatial organization replicates the pattern of individual whiskers on the rat's snout. Decortication, or removal of substantial portions of rostral cortical regions does not abolish the "basic" (7-9Hz) whisking rhythm, which is assumed to reflect the operation of a brainstem Central Pattern Generator.

Ablation of the cortical "barrel field" has no effect upon tactile detection or discrimination tasks involving *passive touch*, but disrupts performance on *active touch* tasks such as gap detection, (Hutson and Masterton, 1986), and texture discrimination, (Guic-Robles, et al, 1992). These observations suggest that such disruptions reflect lesion effects upon the sensorimotor modulation of vibrissa movement patterns. However, in neither of the lesion studies was whisking behavior directly measured. Moreover, previous cortical ablation studies did not assess effects upon whisking kinematics. In the present study we provide such data by comparing movement patterns recorded from mystacial vibrissae, ipsilateral and contralateral to the ablated cortical "barrel" field.

General Methods

Three Long-Evans rats were anesthetized with 50 mg / kg of Nembutal. The anesthetic state of the animal was monitored and supplements (5 mg / kg) were given when necessary. Small holes were drilled in the skull for self-tapping screws. A craniotomy was made over S1 cortex. Carbon fiber electrodes were advanced into the cortex and multiunit recordings were used to determine the receptive field of neurons. Whiskers were stimulated with a hand held probe. The vibrissal S1 cortex was mapped and the boundaries of barrel cortex were delineated, using coordinates measured from Bregma. Landmarks like the blood vessel pattern evident on the surface of the brain were sketched and recording sites were aligned to these landmarks. Using the information gathered from the recording, and blood vessel pattern information, subpial aspiration of the defined region was carried out, the cavity filled with gelfoam and the scalp replaced over the ablated area. A dental cement crown with an embedded mounting screw was constructed to allow head fixation.

Behavioral Testing

For about a month postoperatively, subjects were used in an experiment in which they were reinforced with water for lever pressing on a Variable Interval schedule, but no discrimination training was given. Approximately eight months postoperatively, the movement trajectories of a pair of bilaterally homologous whiskers (C-1 Right, Left) were monitored during three successive 30 min. daily test sessions. Data on whisking patterns in normal animals were obtained from two additional head-fixed animals.

Subjects were maintained on a 23.5 h water deprivation schedule, with food available *ad lib*. The rat's body was restrained in a V-shaped acrylic enclosure, and its head fixed to a metal bracket using bolts embedded in its dental cement crown. During testing water was delivered at random intervals on a non-contingent schedule. Note: In both normal and ablated subjects, *all whiskers on both sides of the face were intact*

Results

Histological analysis

Eight months after surgery, animals were sacrificed with an overdose of Nembutal, perfused with heparinized 0.1 M (Phosphate buffer) followed by 4% paraformaldehyde. The brain was removed, post-fixed over night and sectioned. Coronal sections were stained for Cytochrome c-oxidase (Wong-Riley, 1979) and the extent of the lesion was determined. Figure 11 presents coronal sections at the level of the lesion in each of the three animals. In all three, the ablations were restricted to the posterior-medial barrel field. The medio-lateral extent of the ablation was 3-5 mm; its rostro-caudal extent was 6 mm. The depth of the ablation varied a bit from animal to animal, sometimes extending into the internal capsule. The brain on the ablated side is distorted. Nevertheless the ablation includes all of S1 barrel cortex in all three animals, covering 3-5 mm mediolaterally and nearly 6mm rostro-caudally. There is some interanimal variability in the depth of the ablation, which extends into the internal capsule in two of the three animals.

V7 (Animal 1)

V 9 (Animal 2)

V11 (Animal 3)

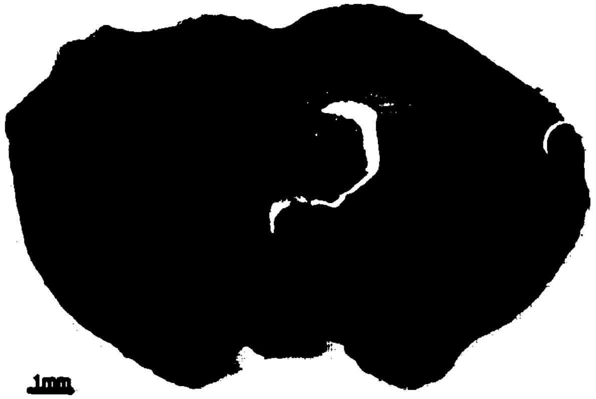

Figure 11 Lesion Reconstructions for V07, V09, and V11

Coronal sections at the center of PMBSF lesions in three animals. In all cases there was a complete ablation of the barrel field. Bars represent 1 mm.

Kinematic analysis

Figure 12 presents records of the movements of the Right and Left C-1 vibrissa in a normal head-fixed animal during part of a single test trial. Although there are some variations in whisking amplitude over the course of the record, and movements of the two whiskers are sometimes slightly out of phase, whisking movements on the two sides have similar amplitudes and are emitted at similar frequencies. Figure 13 presents comparable data from a rat with a confirmed ablation of the cortical barrel field. In the experimental animal, whisking amplitudes appear to be larger for the (Right) whisker, contralateral to the ablated cortex. This conclusion is supported by the data presented in Figure 14, which plots the Mean whisking amplitude for the Right and Left C-1 whisker in a normal, Control and the three Experimental animals. For all three, the amplitude of whisking movements made by the vibrissa contralateral to the ablation is significantly greater, but no such differences are seen in normal animals. In Figure 15 frequency distributions of whisking amplitudes are shown for the R and L whiskers in the three experimental animals. Amplitude differences are most noticeable at the large end of the distribution. Figures 16-19 present functions relating whisking amplitude to two kinematic variables—velocity and rise time for the three experimental animals and a normal control. In the Experimental group, as in normal animals, the scaling of protraction amplitude involves primarily control of velocity, rather than rise time.

To examine ablation effects upon whisking frequency, a Fast Fourier analysis was applied to the movement data for the L and R whiskers in each of the experimental animals as well as in a normal control, (Fig 20-23). In all animals, the power spectra show similar frequency peaks for both whiskers. Figure 24 compares cross correlation plots for the R and L whiskers in a lesioned and a normal rat. In both animals, there is a high degree of coherence in the whisking rhythms on the two sides of the face.

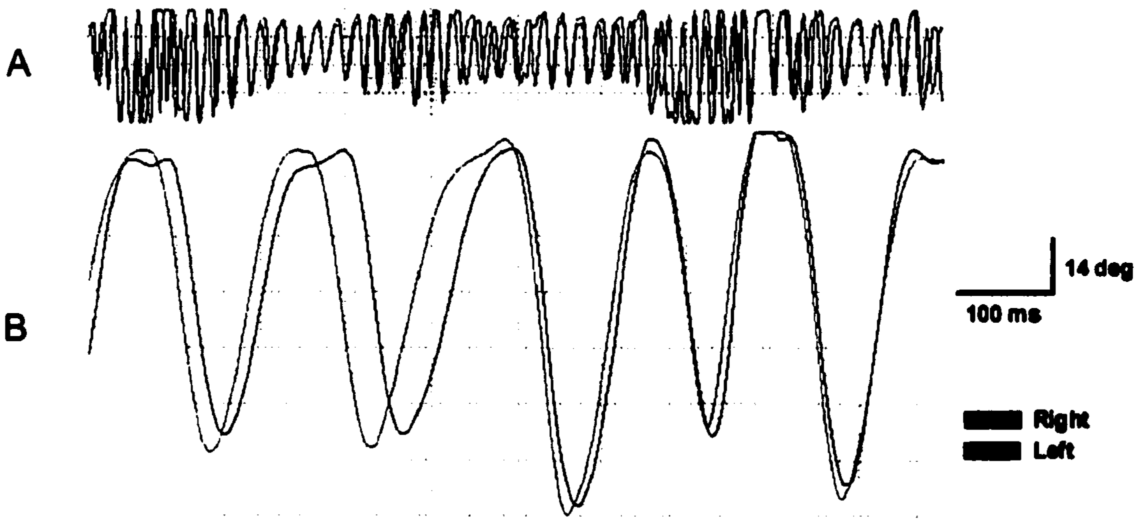

Figure 12 Movements of the Left and Right Whiskers in a Normal Animal
A. Low resolution record of bilateral whisking in the left and right C1 vibrissae in a normal control animal. Shaded portion is shown in higer resolution in B. High concordance between amplitude, velocity, and frequency within homologous pairs of vibrissae in the normal animal.

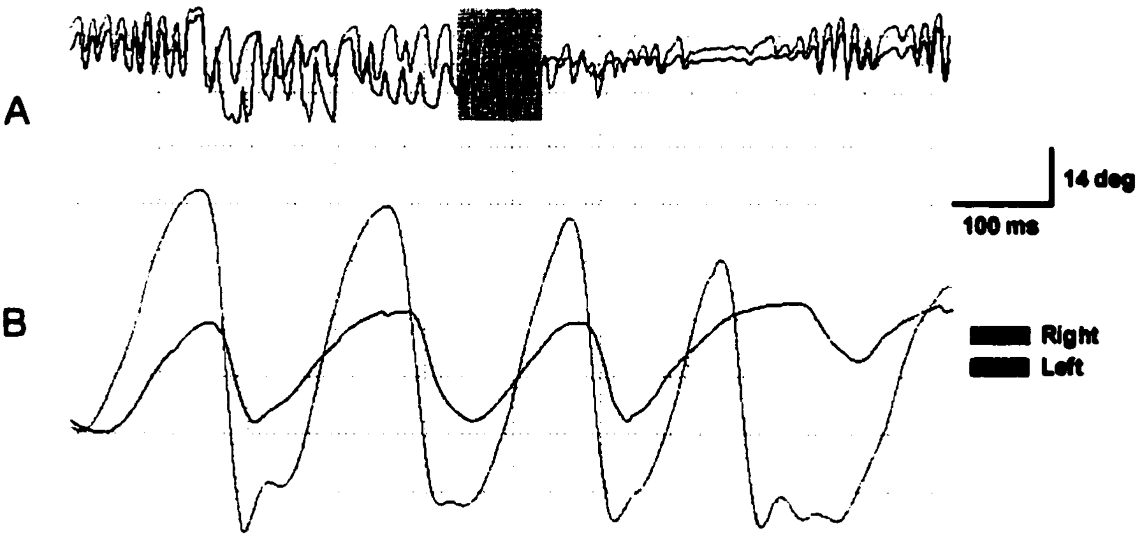

Figure 13 Movements of the Left and Right Vibrissae in an Animal With a Left PMBSF Lesion

A. Low resolution record of bilateral whisking in the left and right C1 vibrissae in a barrel field lesion animal. Shaded portion is shown in higher detail in B. A striking difference in amplitude between homologous whiskers on the left and right sides.

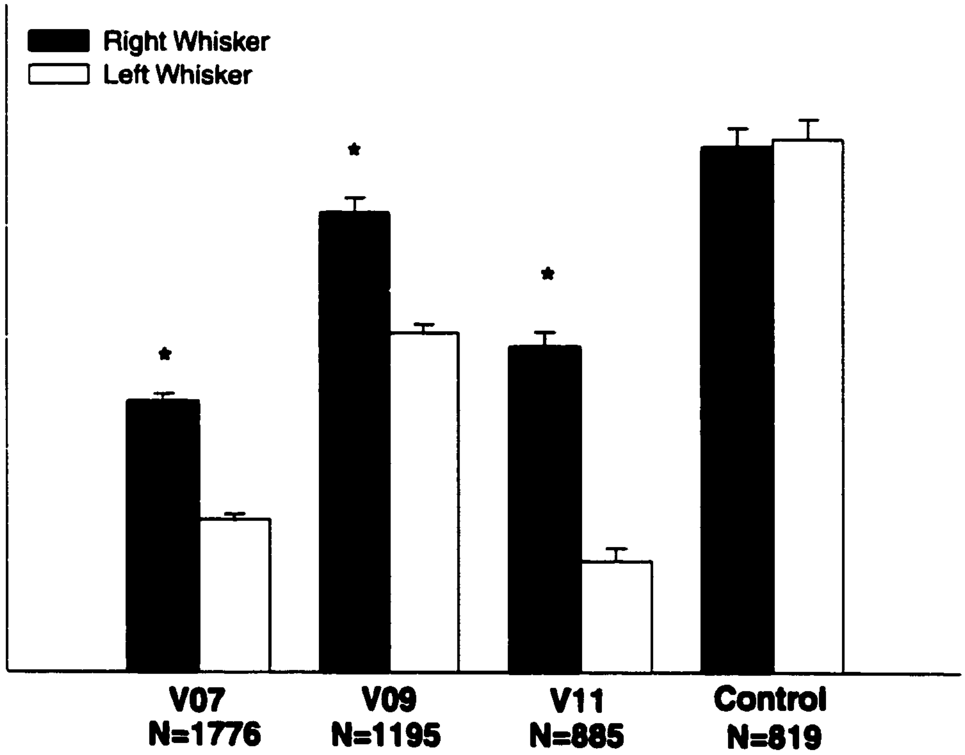

Figure 14 Mean Whisker Protractions (Left vs. Right)
Mean whisking amplitudes for the left and right whiskers of three animals with left PMBSF lesions and one normal control.

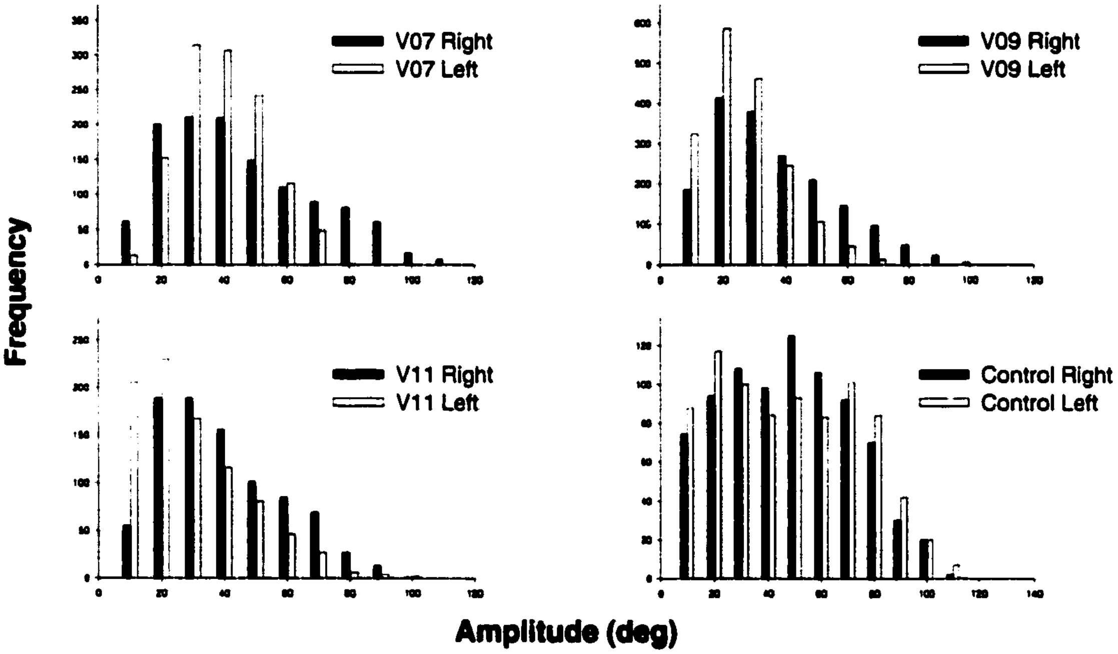

Figure 15 Frequency Distribution of Protraction Amplitudes for Left and Right Vibrissae Panels A-C show the relative frequencies of different protraction amplitudes for the left and right vibrissae of three rats with lesions to the left PMBSF. Panel D shows the same data for a normal control.

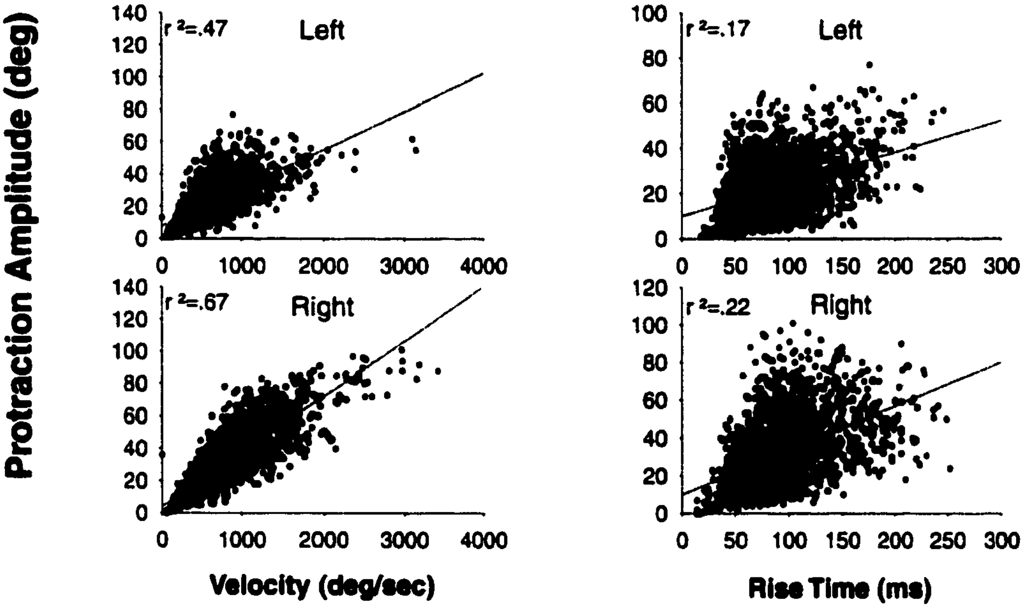

Figure 16 Whisking Kinematics of Left vs. Right Vibrissae: Rat V07 Peak velocity (left) and rise time (right) vs protraction amplitude in right and left C1 vibrissae in an animal sustaining a lesion to left PMBSF.

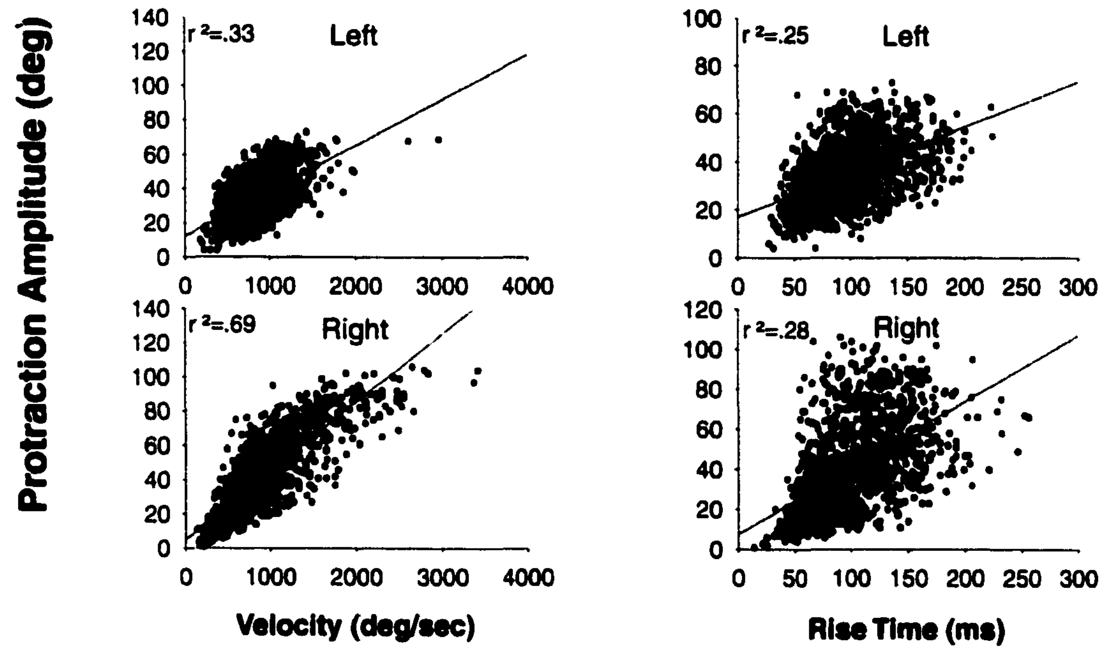

Figure 17 Whisking Kinematics of Left vs. Right Vibrissae: Rat V09 Peak velocity (left) and rise time (right) vs protraction amplitude in right and left C1 vibrissae in an animal sustaining a lesion to left PMBSF.

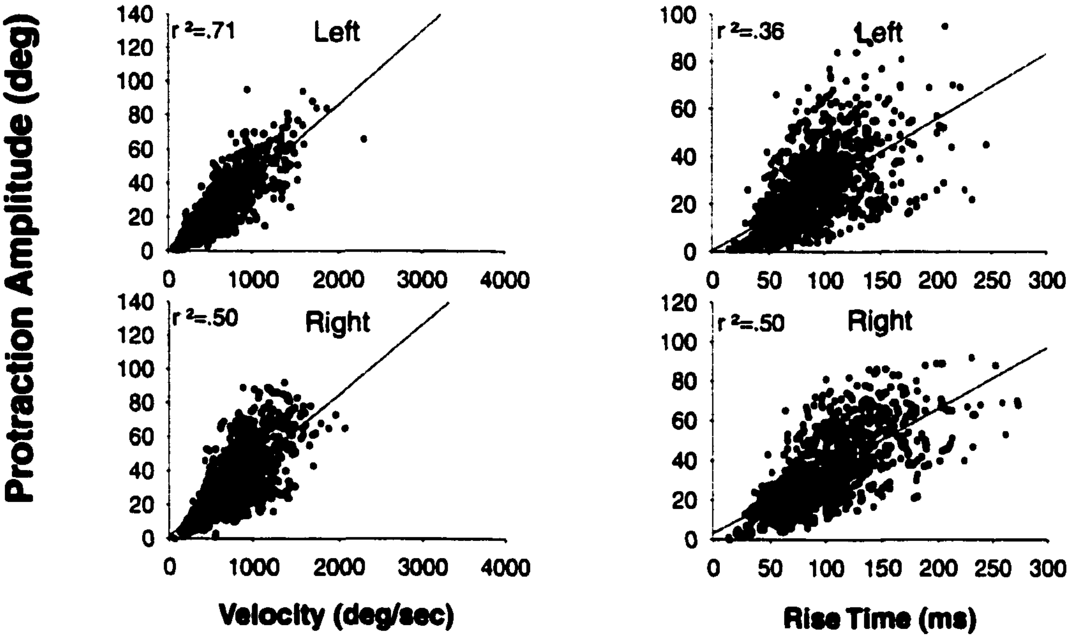

Figure 18 Whisking Kinematics of Left vs. Right Vibrissae: Rat V11 Peak velocity (left) and rise time (right) vs protraction amplitude in right and left C1 vibrissae in an animal sustaining a lesion to left PMBSF.

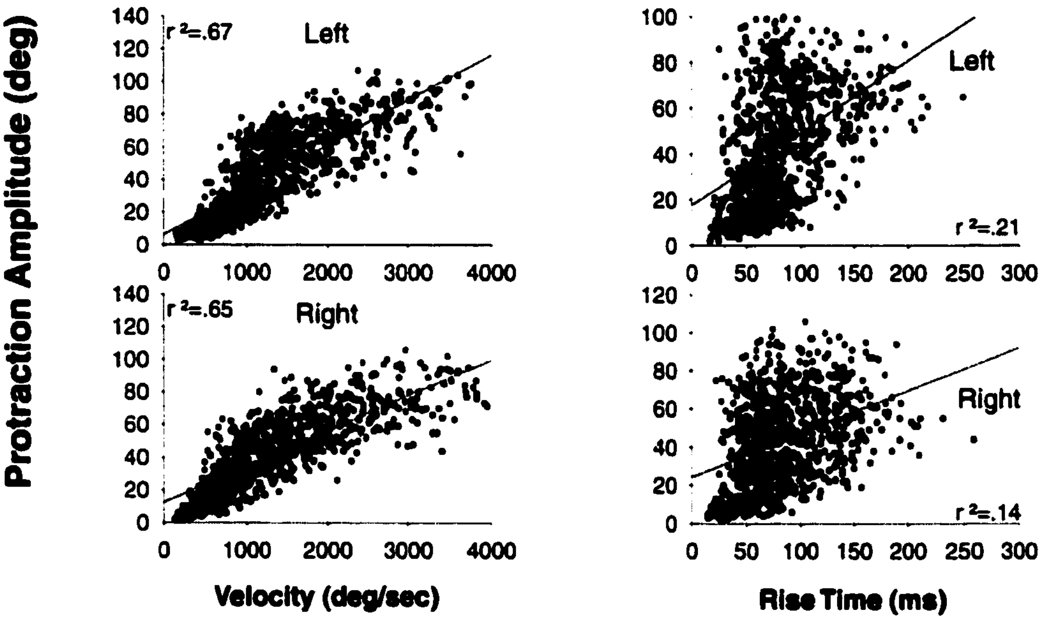

Figure 19 Whisking Kinematics of Left vs. Right Vibrissae:Normal Control

Peak velocity (left) and rise time (right) vs protraction amplitude in right and left C1 vibrissae in a normal control animal.

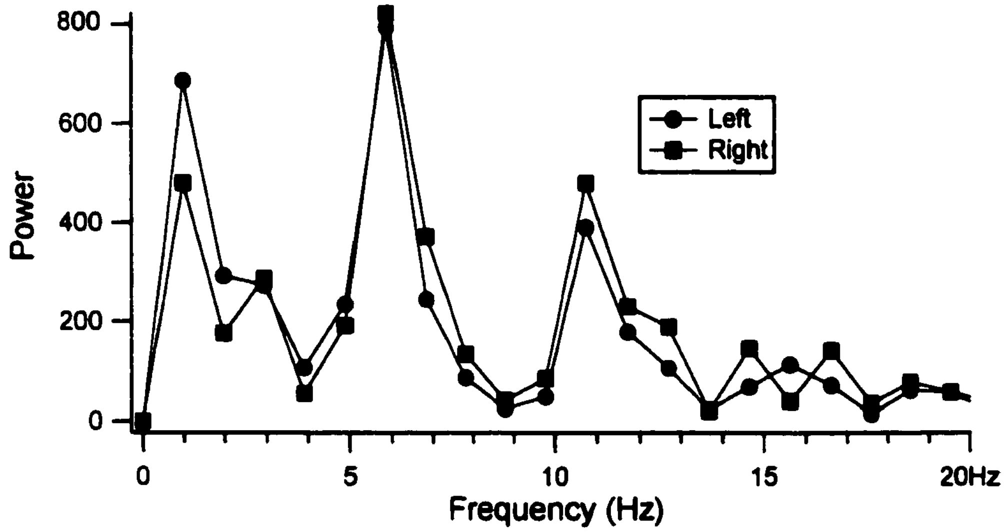

Figure 20 FFT Right vs. Left Whisker: Rat V07
Power spectra for left and right C1 Vibrissae in an animal with a lesion of left PMBSF.

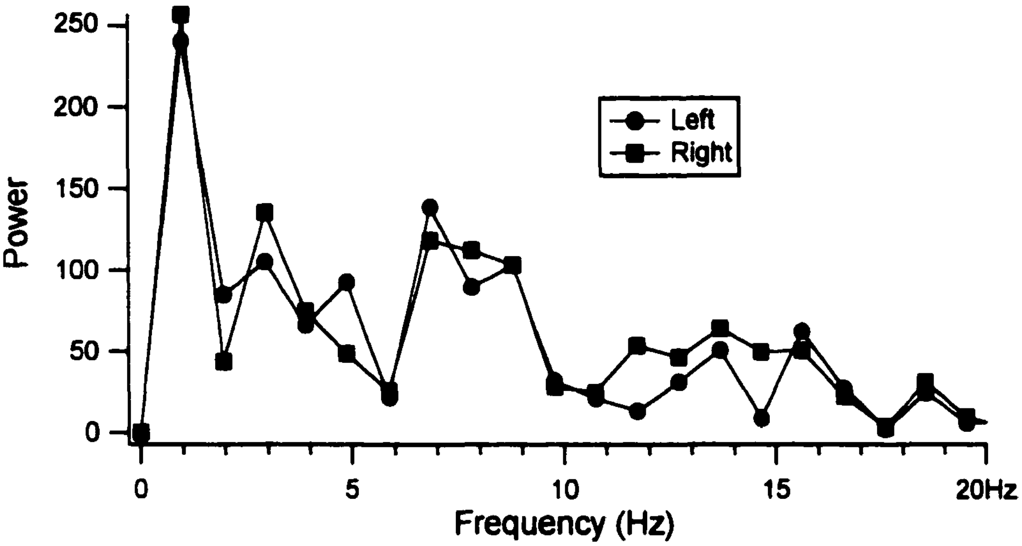

Figure 21 FFT Left vs. Right Whisker: Rat V09
Power spectra for left and right C1 vibrissae in an animal with a lesion to left PMBSF.

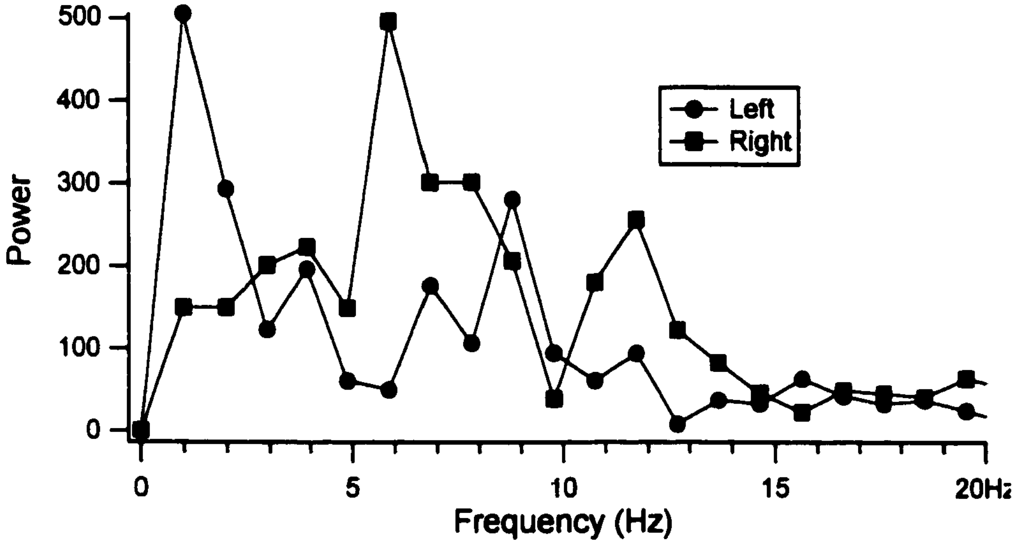

Figure 22 FFT Left vs. Right Whisker: Rat V11
Power spectra for left and right C1 vibrissae in an animal with a lesion of left PMBSF

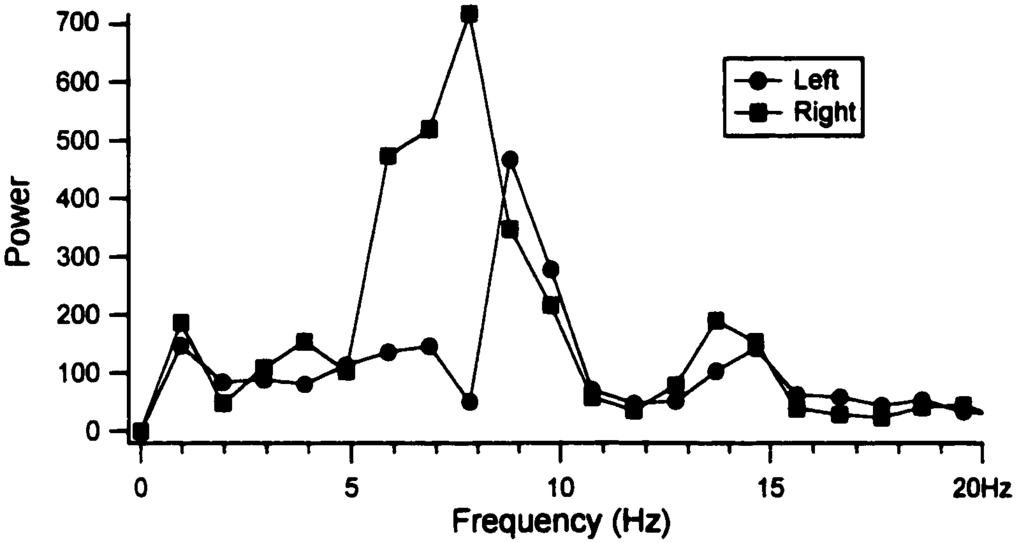

Figure 23 FFT Left vs. Right Whisker: Normal Control Power spectra for left and right C1 vibrissae in a normal control animal.

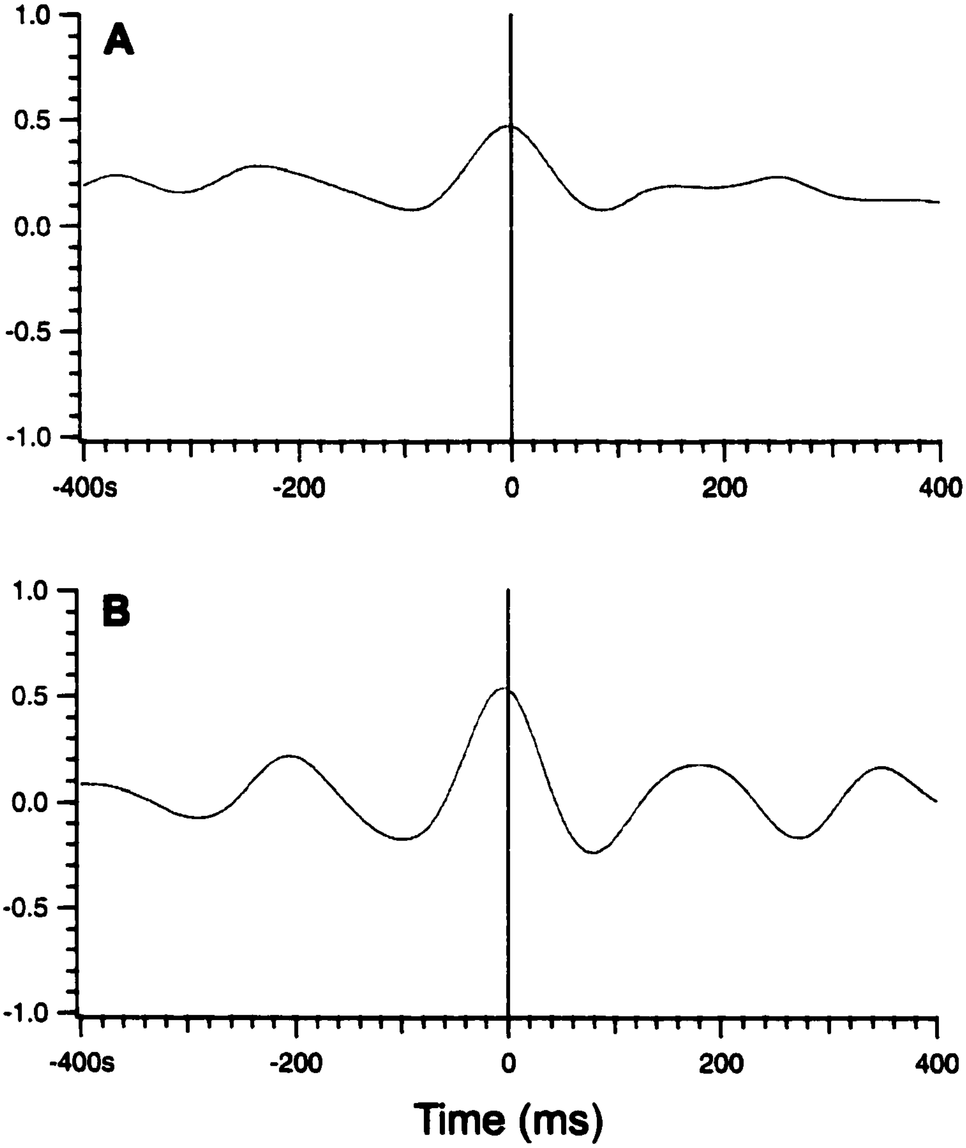

Figure 24 Concordance Between Left and Right Whisker Movements
Cross correlations reveal a high degree of synchrony in the movements of the left and right C1 vibrissae in both the ablated (A) and normal (B) animals.

Chapter 4

Discussion

Discriminative Whisking in the Head Fixed Rat

We have shown that (a) rats can solve tactile object detection and discrimination tasks using only the large, motile mystacial vibrissae (*macro*vibrissae)and (b) without engaging in head movements. We have confirmed and extended the previous findings (a) that acquisition of discrimination tasks is accompanied by changes in whisking behavior patterns, and (b) that successful performance on the different tasks is associated with changes in different whisking movement parameters (Carvell and Simons, 1995). Our data were obtained in head-fixed, immobilized subjects using a methodology that permits us to track the movements of an individual whisker with high spatio-temporal resolution and in 'real' time. Our findings are thus of both substantive and methodological interest.

Methodological Considerations

The present methodology is quite different from current procedures for the study of discriminative whisking behavior, with respect to training paradigm, stimulus control, response measurement and data analysis. Immobilization and head fixation eliminate confounds related to head movement, and facilitate precise control of stimulus presentation (e.g., to *either* the right or left whiskers, or *exclusively* to the macrovibrissae). They eliminate the need for visual occluders, and, with the use of an operant 'indicator' response, minimize the need for the repeated experimenter/animal interactions typical of most rodent tactile discrimination paradigms. Head fixation is a prerequisite for the use of an optoelectronic monitoring system, which provides far higher spatio-temporal resolution (1.4 ms; 26mμ) than is available with all but the most sophisticated video motion-analysis systems. Moreover, optoelectronic monitoring facilitates rapid and efficient (computer-assisted) acquisition and analysis of whisking data, making it possible to collect data on thousands of whisks (tens of thousands of data points) on large numbers of individual trials, over the long series of training sessions associated with tactile discrimination learning—including both negative and positive trials. [The optoelectronic system does not provide information on whisker/object interactions (e.g. whisker bending) or contact onset and offset (but see Bermejo and Zeigler, 2000)]. The operant discrimination paradigm facilitates dissociation of the rat's "observing responses" (discriminative whisking), from its "indicator" response (lever pressing). It allows us to measure each response class independently and to analyze the process by which the indicator response comes under the stimulus control of information acquired by whisking behavior.

In principle, these methods represent a considerable advance over current procedures, but only if they generate data consistent with those obtained by more conventional procedures. Put succinctly, do the current procedures (including head fixation, marking of a single whisker, and monitoring of the whisking trajectory only in the rostro-caudal plane) disrupt normal patterns of discriminative whisking or bias data collection in a systematic manner? First, with respect to the marker, we have measured whisking movements of a single vibrissa with and without the marker and found no significant differences in movement kinematics (Bermejo et al. 1998). Second, Carvell and Simons

(1990) found that, during discriminative whisking, vibrissae on the same side of the face move in synchrony. We have confirmed this observation videographically using marked C-1 whiskers on the two sides of the animal. Third, while the whisking trajectory has both a rostro-caudal and a dorso-ventral component, our measurement procedure is based on the widely held assumption that, to a significant extent, individual whisker movements may be described "solely in terms of (variations) in a single angle as a function of time" (Kleinfeld, 1999). Thus, high resolution monitoring of the movements of a single whisker in the rostro-caudal plane during discriminative whisking should reflect important kinematic parameters of the whisker array. Our data indicate that the relation between discriminative whisking patterns and performance observed in head-fixed animals are (a) systematic and (b) similar to those obtained from unconstrained rats using videographic recording (see below). [Of course, the technique does not distinguish between whisker displacements attributable to the action of follicular *(intrinsic)* muscles and those reflecting changes in the movement baseline produced by action of the mystacial pad *(extrinsic)* muscles (Dorfl, 1982)].

Moreover, our results strongly suggest that solution of the detection and discrimination tasks was mediated by tactile (whisker) inputs, rather than by extraneous stimuli fortuitously associated with reinforcement. Perhaps the most convincing evidence is the contrast between the rapid reacquisition of criterion performance following presentation of the stimuli to the "naiive" whiskers, and the precipitate drop in performance following the shift from the detection to the texture discrimination task (Fig. 10). In the latter task, subjects continued to press the lever but responded non-differentially, i.e., during both S+ and S- trials. Their persistence in responding in both conditions is consistent with their prior training, since (on the detection task) lever presses were reinforced in the presence of a discriminandum and extinguished in its absence. In the texture discrimination a discriminandum is present on both S+ and S- trials.. The absence of differential responding in this situation [with all other testing conditions constant] is strong evidence that the lever pressing of these animals was controlled by stimulus-reinforcer contingencies associated with vibrissa-mediated tactile inputs from the stimulus objects.

One final methodological consideration is worth noting. In previous studies of tactile localization and discrimination, the rats were initially allowed to palpate the discriminanda with their snouts and the discriminanda were then gradually moved until the indicator response (gap-jumping) was presumably controlled only by the whiskers (Hutson and Masterton, 1986; Carvell and Simons, 1995). Thus the final discrimination performance involves *both* initial transfer of stimulus control from snout afferents to vibrissa, and subsequent vibrissa-mediated acquisition. The present testing arrangements allow us to track the development of a whisking" strategy", mediated solely by the *macro*vibrissae, over the course of task acquisition.

Acquisition and performance on the detection and discrimination tasks

Unlike the "species-typical" jumping/reaching responses of previous studies, the lever pressing response had to be acquired and maintained by the rat prior to the start of discrimination training. Acquisition and maintenance of this "indicator response" was difficult for many animals, probably due to the physical constraints associated with immobilization and head fixation, and males seemed to adapt better to the testing situation than females. In addition to the need for handling and a period of adaptation to the test situation, the size, force requirements and position of the lever relative to the animal were critical variables. We found that initial acquisition of the lever press was facilitated by relatively severe (transient) water deprivation and the use of a successive approximation (shaping) procedure in the initial session(s). A small microswitch, with minimal force requirements, placed horizontally, in front of and parallel with the paw, was effective, but the use of different animal-response configurations and deprivation-reinforcement arrangements might facilitate performance.

Most animals which acquired the response and entered the discrimination phase, performed satisfactorily over prolonged periods of testing. While there was considerable individual variability in the time course of acquisition for both tasks, improvement in performance, on both, was signaled by an *increased* latency of lever pressing on S- schedule components, and a *decreased* latency on S+ trials. Since subjects had initially been trained to press the lever at a steady but moderate rate, these variations in latency during discriminative behavior provide additional evidence for modulation of response rate by the discriminanda.

Whisking kinematics during detection and discrimination

The two tasks make quite different demands on the vibrissa as an "active touch" system. In the detection task, the animal must simply confirm the presence of an object within the "planar sensory field" formed by the whiskers (Wineski, 1983). This may not require modulation of the basic whisking movement pattern. In the discrimination task, however, the animal has to distinguish the stimulus properties of one object from those of another.

Identifying salient features that distinguish the two discriminanda may require the animal to actively engage the object and process the inputs that characterize those features. Differences between the whisking patterns associated with successful performance on the two tasks may thus reflect differing task demands.

Five whisking movement parameters were examined over the course of task acquisition: whisking frequency, the amplitude, velocity and duration of individual whisks, and the amount of whisking. For both the detection and discrimination animals, whisking frequencies during the initial sessions were typical of the basic (6-9 Hz) "exploratory" whisking pattern. The power spectra of the *detection* animals did not change over the course of learning in either the successful or the unsuccessful animals. However, the two successful object discrimination animals showed a shift to higher frequencies over the course of training, which was not seen in the unsuccessful rat. A significant association between bandwidth and successful discrimination performance has been reported in previous studies of discriminative whisking Carvell and Simons, 1995, 1996). Confirmation of that relationship for an object discrimination task in the head-fixed animal both extends the generality of the finding and suggests that our testing paradigms do not disrupt "normal" patterns of discriminative whisking.

Successful performance in the detection animals is associated with variations in the amount of whisking, manifested as a significant difference between the number of whisks emitted on S+ and S- trials during the last three training sessions. Interestingly, both successful animals show consistently more whisking during the S- trials, but no such difference is seen in the unsuccessful subject. The increased whisking on the S- trials may reflect the utility of repeated sampling which would increase the likelihood of detection and decrease the probability of incorrect responses. The fact that none of the other kinematic variables, including frequency, were correlated with performance suggests that acquisition of the detection task involves modulation of the *amount* but not the type of whisking.

Successful performance on the object discrimination task was associated not only with higher whisking frequencies, but with decreased mean durations of individual whisking movements, changes in protraction amplitude and increased whisking on S- trials during the last three sessions—a period when discrimination performance was at or approaching criterion levels. While discrimination performance did not seem to be associated with the absolute amplitudes of emitted whisks, the relative *change* in mean amplitude over the course of training was much greater for the two animals that met criterion than for the third, which did not (7-10° vs. 1°). These data strongly suggest that acquisition of the object detection task requires modulation of *both* the amount and the type of whisking. The only kinematic variable that did not appear to correlate significantly with task performance was velocity, although the two rats that achieved criterion performance showed a trend towards decreasing velocity over the course of training.

Because our primary focus was methodological, we did not examine the effects of systematically varying stimulus dimensions on the development of whisking movement patterns. Nevertheless, despite substantial differences in training paradigm, data recording and analysis we have confirmed the previous finding that successful discriminative behavior is associated with the presence of systematic variations in whisking movement patterns which may function as whisking 'strategies' (Carvell and Simons, 1995)

Functional characterization of whisking behavior: Differential contributions of micro- and macrovibrissae.

Brecht et al (1997) has noted that the different whisker types that make up the vibrissa array have often been treated as functionally equivalent. Using an ingenious testing paradigm, involving either localization of or discrimination between small palatable and unpalatable cookies differing in shape, these investigators showed that the effects of removing either the *micro*-or *macro* vibrissae varied with task type. *Macro*vibrissae removal disrupted the localization, but not the recognition task. *Macro*vibrissae removal disrupted the object recognition but not the localization task. Based upon these findings and differences in the topography and spatial density of the two classes of sensory hair, Brecht et al (1997) concluded that the *macro*vibrissae function "as a distance detector array;...the *micro*vibrissae...as a high resolution tactile sensor" (1997, p. 97).

That the *macro*vibrissae contribute to spatial perception has been repeatedly demonstrated (Vincent, 1912; Schiffman, Lore, Passaficieme, and Neeb, 1970; Hutson and Masterton, 1986; Harris, Petersen and Diamond, 1999). The contribution of the *micro*vibrissae to high-resolution discriminative behavior has been less convincingly documented. Brecht notes, "The object recognition deficit introduced by shaving is very transient—one day in most animals (Brecht, *pers. comm.*, 1998). Dissociation of the contributions of *micro*vibrissae and snout skin afferents is also difficult. On the other hand, Carvell and Simons (1990, 1995) have shown that the large motile *macro*vibrissae can support high-resolution texture discrimination. Furthermore, the present study demonstrates that object detection and discrimination may be accomplished using only the *macro*vibrissae. The discriminanda used in the object discrimination task differed substantially along several stimulus dimensions (size, shape, texture) so that its solution did not require high resolution encoding of any of these properties.

Identification of the "function" of a biological structure such as a sensory organ will depend upon the range and type of behavioral tasks used to 'interrogate' that structure. Characterization of vibrissal function is complicated by substantial differences in task demands and testing conditions in the available studies (distal vs. proximal stimuli; high vs. low resolution requirements). The use of a gap-crossing paradigm and distal stimuli obviously engages the *macro*vibrissae but does not exclude a contribution by inputs generated by lateral head movements across the *micro*vibrissae. [Note: Carvell and Simons (1995) report that, during acquisition of the texture discrimination, the small rostral whiskers remain protracted and in contact with the discriminanda during active palpation with the large caudal whiskers (Carvell and Simons, 1990). Use of an elevated stage or open field for an object recognition task involving proximal stimuli (Brecht, et al 1997, *pers. comm.*) may produce a bias towards the use of the *micro*vibrissae. The paradigm used in the present study insures exclusive use of the *macro*vibrissae in a low resolution discrimination task. We have no reports of performance on a high resolution discrimination tasks under conditions which insure exclusive use of *either* the macro-or microvibrissae. The currently available data suggests that both classes of vibrissa can support a variety functions depending upon behavioral context.

Conclusions

Under laboratory conditions, discriminative behaviors may be conceptualized as involving two distinct response classes: (1) responses upon which differential reinforcement is directly contingent (e.g. lever pressing, gap jumping, button pushing, speech) and which serve as indicator responses for the experimenter; (2) observing responses which, while not differentially reinforced, provide, for the animal, (a) exposure to the relevant stimuli and (b) encoding of critical stimulus properties. In audition and vision, observing responses may either be internal or indirectly inferred from head or eye movements. For the human somatosensory system it is possible to identify hand movement patterns which serve as an interface between the stimulus and the indicator response and whose topography is highly correlated with the stimulus properties to be encoded (Lederman and Klatzsky, 1987; Morley et al, 1983). Similar patterns may be observed in the whisking behavior of rats. In their seminal papers on discriminative whisking in rodents, Carvell and Simons (1995, 1996) recognized that stimulus-related differences in the motor pattern of whisking might provide clues as to the way in which different stimulus properties are encoded by the relevant sensory system.

The present report uses novel methodologies to extend and clarify previous studies of the sensorimotor control of discriminative whisking in rats. Those methodologies allows us to track discriminative whisking movements with high resolution under well-controlled conditions. They make it possible to use each animal as its own control in lesion studies by comparing the behavior of homologous whiskers connected to either intact or lesioned central whisker projections at different levels of the neuraxis. They are compatible with long-term studies of correlated behavioral and neural activity over the course of task acquisition. Whisking" behavior offers an unique neurobehavioral "window" onto the processes by which the brain encodes information about the external world and uses it to generate movements. The methods used in the present study may facilitate analysis of those processes.

Unilateral Barrel Field Ablation Changes the Relationship of Whisking Amplitudes between Left and Right Vibrissae

Unilateral ablation of the cortical "barrel field" does not abolish the rat's rhythmic whisking pattern, disrupt mechanisms of amplitude scaling, or impair the bilateral coordination of whisking. It does produce significant increases in the amplitude of whisking responses emitted by vibrissae contralateral to the ablated area. [Although preoperative data for these animals was not obtained, amplitudes for the whisker ipsilateral to the ablation fall within the range reported for normal animals (Carvell and Simons, 1990; Bermejo, et al. 1998).

These observations are consistent with current models (e.g. Kleinfeld, 1999) which assume that the "basic" whisking rhythm is produced by a brainstem Central Pattern Generator (CPG). Convincing behavioral evidence for a whisking CPG, based upon differentiation studies, has recently been reported (Gao, Bermejo and Zeigler, 1999). In such models, CPG activity is modulated by descending outputs from vibrissal motor cortex, bilaterally coordinated by callosal connections (Carvell, Miller, and Simons, 1996). Because input from the "barrel-field" makes a substantial contribution to the vibrissal motor cortex, its removal might be expected to decrease motor cortex output to structures along the descending pathway, *unilaterally*, producing an imbalance in the modulatory input to the CPG. If we assume that the CPG normally operates in a "free-running" mode, cortical influences upon the CPG are likely to be primarily inhibitory. Unilateral ablation of the "barrel field" would produce a disinhibition of activity in the contralateral CPG and an increase in its excitability. Note that despite the observed increase in whisking amplitude, modal whisking frequencies on the side contralateral to the ablation are essentially unchanged. This finding suggests that the shift in whisking amplitude produced by the lesion is being monitored and whisking rhythm regulated by controlling whisking velocity. Re-afference generated by whisker movements has been reported at both ganglion (Zucker and Welker, 1969) and thalamic levels (Brown and Waite, 1974).

Whatever the adequacy of this account, the present results may clarify previous findings that "barrel-field" lesions disrupt discriminative behaviors dependent upon "active touch". During vibrissa-mediated tactile discrimination, the rat progressively refines its whisking pattern by modulating vibrissa movement parameters, including whisking amplitude (Carvell and Simons, 1995). Thus, in addition to sensory processing, active touch involves a significant degree of sensorimotor control of the vibrissa. Our data suggest that the effects of barrel cortex ablation upon tactile discrimination may reflect impaired sensorimotor control of whisking movement parameters.

Future Directions

This paradigm provides a useful tool for future investigations of sensorimotor function. An intriguing question that can now be answered directly is to what extent does an animals inability to effectively modulate its whisking contribute to the deficits seen in discriminative performance in animals with cortical lesions. Similarly how do the various brainstem and thalamic structures contribute to the animals ability to appropriately pattern its whisking to different sensory tasks. These questions can be addressed both through lesion studies as well as through electrophysiological methods using the head fixed preparation. The ability confine the animal's whisker to defined physical location allows us to ask questions about sensory coding in an active touch system, where we can better control what is impinging on the sensory organ at any given time.

Further this preparation will allow us to combine behavioral studies with fine electrophysiological recordings in awake, and undrugged animals that should help to clarify not only some long-standing controversies in the literature, but also may hopefully cast some light some of the basic mechanisms by which sensory information is acquired, encoded, and ultimately applied to the regulation of fine motor tasks.

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