Proc. Natl. Acad. Sci. USA
Vol. 93, pp. 4942–4947, May 1996
Neurobiology

Quantitative long-term imaging of the functional representation of a whisker in rat barrel cortex

(sensory representation/vibrissae/intrinsic signals/optical imaging)

S. A. MASINO AND R. D. FROSTIG

Department of Psychobiology and the Center for the Neurobiology of Learning and Memory, University of California, Irvine, CA 92717-4550

Communicated by James L. McGaugh, University of California, Irvine, CA, December 20, 1995 (received for review July 28, 1995)

ABSTRACT **In this study, we implement chronic optical imaging of intrinsic signals in rat barrel cortex and repeatedly quantify the functional representation of a single whisker over time. The success of chronic imaging for more than 1 month enabled an evaluation of the normal dynamic range of this sensory representation. In individual animals for a period of several weeks, we found that: (*i*) the average spatial extent of the quantified functional representation of whisker C2 is surprisingly large—1.71 mm^2 (area at half-height); (*ii*) the location of the functional representation is consistent; and (*iii*) there are ongoing but nonsystematic changes in spatiotemporal characteristics such as the size, shape, and response amplitude of the functional representation. These results support a modified description of the functional organization of barrel cortex, where although a precisely located module corresponds to a specific whisker, this module is dynamic, large, and overlaps considerably with the modules of many other whiskers.**

Repeating functional modules are a prominent organizing theme of sensory cortex (1, 2). Detailed evaluation and quantification of ongoing spatiotemporal characteristics of these modules underlie a comprehensive evaluation of cortical function. Although many features of the functional organization of sensory cortex have been revealed, several characteristics in both the spatial and temporal domains remain relatively inaccessible. For example, the opportunity to assess sensory response areas with high spatial resolution, particularly for more than one experimental day, has been practically unavailable. The techniques providing high spatial resolution are either inherently invasive to the neuronal tissue (closely spaced microelectrode recordings) or a terminal procedure (2-deoxyglucose mapping). Consequently, there is little information quantifying functional modules, particularly their dynamics over time in individual animals. Given the emergent view of sensory cortex as dynamically regulated throughout life (3–6), an understanding of the ongoing stability versus plasticity of cortical modules is fundamental to understanding cortical processing.

*I*ntrinsic *s*ignal optical *i*maging (ISI) provides a high-resolution (≈50 μm) visualization of functional activity over large spatial areas of the brain surface. Although ISI provides limited temporal resolution, its spatial resolution lends itself to visualizing and subsequently quantifying functional modules (for reviews, see refs. 7–9). In addition, ISI records activity-dependent changes in light reflectance and requires no invasion to the cortical tissue. This noninvasive imaging allows repeated sampling without preset limits on experimental duration. Thus, ISI achieves high spatial resolution simultaneously with the potential for extending the temporal domain beyond 1 day. Successful chronic optical imaging through an optical chamber with the underlying skull and dura removed has recently been reported in kitten visual cortex (10).

In the present study, we introduce a chronic ISI preparation to investigate the dynamics of cortical sensory processing over time. Because of its unique structural (11, 12) and functional (refs. 13 and 14; for recent reviews, see refs. 15 and 16) features, the cortical representation of a whisker in barrel cortex presents a tractable model for quantifying a functional module and its dynamics. Successful qualitative visualization and characterization of whisker representations using ISI in rat barrel cortex have been accomplished (17–19), including visualization through a thinned skull (17, 19). In this report, we further develop the thinned skull preparation into a chronic imaging preparation and characterize a specific whisker representation quantitatively and over time.

Using chronic, quantitative ISI, we found that stimulation of a single whisker consistently activates a surprisingly large area of barrel cortex. In addition, spatial aspects, such as the location of the functional representation, and temporal aspects, such as the time course of the stimulus-related intrinsic signal response, are consistent. However, over time we found ongoing but unsystematic changes both in the shape and the areal extent of the whisker representation, as well as the amplitude of the intrinsic signal response. Thus, these findings indicate that the functional module in barrel cortex activated by a single whisker is both large and dynamic over time.

MATERIALS AND METHODS

Subjects. Repeated imaging was performed on six adult male Sprague–Dawley rats (350–700 g). For the duration of the experimental period, all animals were individually housed with water and food ad lib and a 12 h light/12 h dark cycle. Animals were removed from the housing environment only on experimental days.

Chronic Preparation. Before any data collection, all rats underwent an initial surgical procedure. Rats were anesthetized with Nembutal (50 mg/kg i.p.) and a surgical level of anesthesia was maintained with supplemental doses. Using a sterile procedure, a 6 × 6 mm area of skull overlying the left somatosensory cortex was progressively thinned with a drill bit to a final thickness of 100–200 μm. A thin layer of clear epoxy was applied over the thinned area of skull to prevent skull and tissue regrowth, and a wall of dental cement was built around the outer edges of the thinned area. The skin was sutured around the edges of the dental cement wall, leaving the thinned skull exposed but protected from scratching and grooming behaviors. Topical (Neosporin) and systemic (ampicillin, 100 mg/kg) antibiotics were administered, and each rat recovered without incident. This resulted in a chronic preparation that required no additional surgical procedure on subsequent imaging days. In two animals, the first imaging sessions were performed on the same day as the initial chronic surgery. In all other cases, to allow adequate recovery time, the interval

The publication costs of this article were defrayed in part by page charge payment. This article must therefore be hereby marked "*advertisement*" in accordance with 18 U.S.C. §1734 solely to indicate this fact.

Abbreviation: ISI, intrinsic signal optical imaging.

between the initial surgery and the first imaging session was no less than 5 days and was extended in one case to 50 days.

Data Collection. Each rat was imaged repeatedly (2–4 times each) with successive imaging days at least 1 week apart. On each imaging day, the rat was anesthetized with Nembutal (50 mg/kg i.p.) and placed in a stereotax. An appropriate level of anesthesia (weak eyeblink reflex, no tail or paw reflex) was maintained during imaging with a continuous Nembutal infusion. Before imaging, the epoxy layer was removed and the skull rethinned slightly with the drill bit. In each case the skull was thinned to where the major cortical vasculature was visible when the thinned skull was coated with silicone oil. Measurements of the thicknesses of many other thinned skulls in acute experiments, as well as the thickness after the final chronic imaging experiment, determined this thickness to be 100 ± 20 μm. An additional wall of petroleum jelly was built around the dental cement wall, filled with silicone oil, and sealed with a glass coverslip. The thinned area overlying the somatosensory cortex was positioned under a Photometrics (Tucson, AZ) model CH230 slow-scan charge-coupled device camera and illuminated with a stabilized 630-nm light source. After each day of imaging, the oil and petroleum jelly were removed, a thin layer of epoxy was applied, and this epoxy layer was left intact until the next imaging day.

A detailed description of data collection is provided in Masino *et al.* (17). Briefly, light reflectance values from the somatosensory cortex were continuously collected with the camera for nine consecutive 500-msec frames that included data before, during, and after whisker stimulation. The reflectance values were collected over a 6.8 × 5.1 mm area. Each trial of whisker stimulation consisted of a computer-controlled 0.5-mm rostrocaudal deflection of whisker C2 at a distance of 15 mm from the snout. This deflection amplitude did not cause any detectable movement of any other whiskers as verified by inspection under a microscope. After two frames of baseline data (1 sec), the stimulation was delivered at 5 Hz for 1 sec during each trial. Between each trial there was a fixed interstimulus interval of 15 sec. Each data session consisted of multiple presentations of identical stimulus trials randomly interlaced with control trials (no stimulation delivered; 20% of the trials were controls). After imaging, the animal recovered and remained in the home cage for a minimum of 1 week between successive imaging sessions.

Data Analysis. A detailed description of all aspects of data analysis regarding ISI in barrel cortex is provided (20) and a brief summary is included here. As described (17), to visualize the functional representation of a whisker, the data frames collected 500-2500 msec after the start of whisker stimulation were divided by data frames collected without stimulation within the same trial. We have found that it is beneficial to divide by frames collected within the same trial to avoid incorporating slow, global changes in overall light reflectance that are larger (by as much as an order of magnitude) than the stimulus-induced changes. In the current experiments, each image is created by applying the above division to an average of 128 trials. After division, the data array is mapped to gray scale values, and a coherent black area identifies the location of the functional representation of the whisker.

Spatial analysis: Areal quantification. A computer algorithm locates the peak value of the data array within the functional representation of the whisker. Considering the difference between this peak value and the median value of the data array to be 100% (the median serves as an image-specific reference value), borders can be drawn that include any predefined percentile of this difference. Because the difference between the peak and the median is normalized to 100%, general changes in excitability should not affect the quantified size of the functional representation.

In the present study, the area enclosed within the 50% border (area at half-height) is used as the quantification criteria. Before quantifying the areal extent of the functional representation, Gaussian filtering reduces high-frequency noise inherent in the divided data. To preserve the dynamic range of the data array, filtering and quantification are applied before mapping the data array onto gray scale values for visualization as an image.

As the filtering process increases the size of the area at half-height, we applied Gaussian filters of different dimensions (half-widths 3, 5, 7, 9) to each data array to determine the extent of such an increase. As expected, there is a linear increase in the quantified area at half-height when applying Gaussian filters of increasing dimensions. The intercept of the regression line approximates the areal extent in the absence of any filtering for each individual case. Unless otherwise noted, the areal values reported herein represent those obtained using this regression-based correction procedure.

Each areal quantification in this report was performed using an average of 128 trials. Each trial collected 4.5 sec of data containing 1 sec of whisker stimulation. As many as 5 of these 128-trial data sessions were collected during a single day of imaging. All data sessions collected within a day were averaged to determine the average areal extent of the whisker representation for that experimental day.

Amplitude and temporal analysis. Ongoing variabilities in both the amplitude and the temporal profile of the intrinsic signal response were investigated. The amplitude reported here is the peak fractional change obtained when averaging the response within the area at half-height. The average amplitude was calculated for each 128-trial data session as well as for each experimental day. In addition, the temporal profile (time-to-peak) of the intrinsic signal response was compared across successive imaging days.

RESULTS

The size and functional dynamics of a single whisker representation in rat barrel cortex were quantified repeatedly in individual animals using ISI. In the present study, successful chronic imaging through a thinned but intact skull was performed more than 2 months after the initial surgical preparation. Over the time period examined thus far, image quality and the spatiotemporal characteristics of the intrinsic signal response were maintained. However, the areal extent of the functional representation and the amplitude of the intrinsic signals exhibited dynamic changes over time.

The average areal extent of the functional representation of whisker C2 quantified over time is 1.71 mm^2. Overall averages for individual rats varied between 1.465 and 2.19 mm^2. These large values for the area at half-height emphasize that a large area of barrel cortex responds to stimulation of a single whisker. The functional area at half-height is more than 10× the size of the corresponding anatomical representation of whisker C2 in layer IV, the "barrel," as assessed histochemically using either succinic dehydrogenase (21) or cytochrome oxidase staining (22).

Without a deliberate manipulation to sensory processing, there was variability in the areal extent of the functional whisker representation. The variation within individual rats and the differences between rats over multiple experimental days are shown in Fig. 1. The average difference from the mean value within a rat ranged from 0.36% (CP4) to 48.2% (CP5). At this point, with only a small number of imaging days from each individual rat, it is unclear whether the amount of variability in a whisker representation is a property specific to individual rats or represents the ongoing variability in the rat population as a whole.

Clear images of the functional representation of whisker C2 were regularly obtained during repeated imaging sessions. Representative images from 4 successive weeks of imaging (chronologically ordered A–D) in an individual animal (CP1)

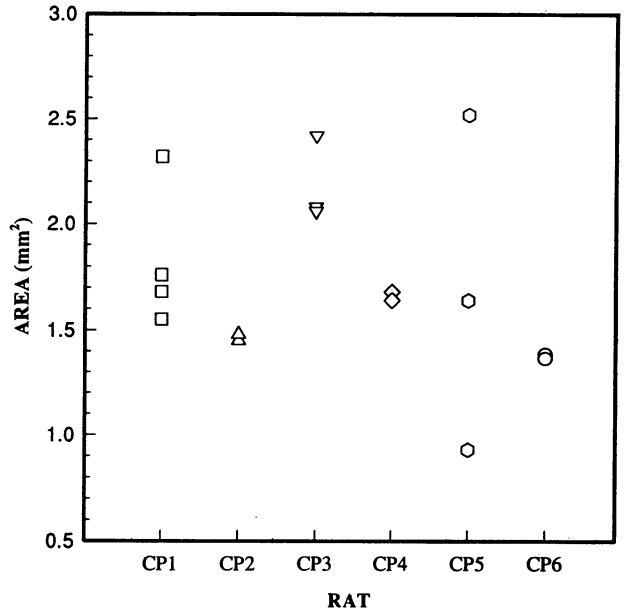

FIG. 1. Areal measures quantified for each individual rat. Each point is the overall average of the area at half-height obtained during 1 experimental day in each of the six subjects (CP1–CP6). Individual rats varied both in the average size of the functional representation of whisker C2 and in the functional range demonstrated over 2–4 experimental days.

are shown in Fig. 2. In each case, the white border outlines the area at half-height of the functional representation of whisker C2 in the left somatosensory cortex. Note that there are ongoing changes in the size and the shape of the whisker representation.

In agreement with the precise topography of barrel cortex, the coordinates of the location of the whisker representation were consistent over time. The quantification borders surrounding the functional representation of whisker C2 obtained during each of the 4 successive weeks illustrated in Fig. 2 are superimposed on an image of the thinned skull (Fig. 3*A*) and on an image of the functional representation (Fig. 3*B*). This composite verifies the consistent location of the whisker representation over time. Even though there is some variability in the border quantifying the area at half-height, the majority of the response area is centered on the same cortical area. In this case, at least 60% of the functional area of any single day is common to all imaging days. In general, the common area shared by the functional areas quantified during successive imaging days ranged from 48% (CP6) to 95% (CP2).

In addition to evaluating the position and shared area over time, this chronic imaging preparation enabled quantification of functional dynamics within the whisker representation of six individual animals. Like the topographic location of the representation, the time course of the intrinsic signal response was consistent. The peak intrinsic signal response always occurred during the 0.5-sec frame collected 1.5–2.0 sec after stimulus onset. However, there were changes in both the area at half-height and the amplitude of the intrinsic signal response over time. Fig. 4 illustrates these ongoing but unsystematic changes in both measures over time. Because of the small sample size and an unequal number of imaging days from individual animals, statistical analyses of this data set were limited. However, when comparing the area and the amplitude on the first imaging day versus an imaging day 2 weeks later using a paired t test, there was no evident trend in either measure ($n = 4$; area, $P = 0.62$; amplitude, $P = 0.49$). The average area at half-height and intrinsic signal amplitude obtained on each imaging day as well as an overall average for each individual rat is summarized in Table 1. Here we also indicate the time interval between imaging sessions and the interval between the initial surgical preparation and the first imaging session.

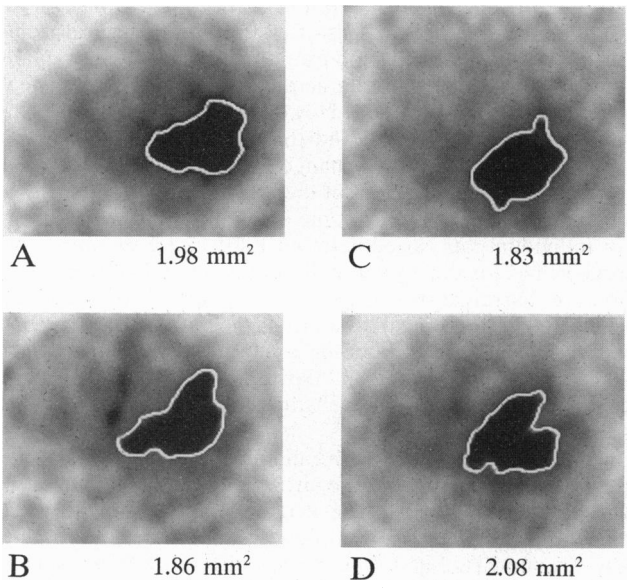

FIG. 2. Representative images obtained over 4 consecutive weeks from an individual animal (CP1). Each image is an average of 32 trials of whisker stimulation of 5 Hz at 1 sec, and the border on each image surrounds the area at half-height. These images are examples of the areal quantification before the regression-based correction. Note that there are changes in the size and shape of the representation over time. Images are in chronological order from *A* to *D*. Although there are ongoing changes, over 4 weeks there is no systematic trend. Furthermore, over this experimental period there was no deterioration of image quality. Each image shown is of a 5.6 × 4.6 mm cortical area.

DISCUSSION

This study describes the advent of chronic, quantitative ISI in rat barrel cortex and its ability to assess the size and ongoing functional dynamics of the representation of a specific whisker. ISI provides the opportunity for cortically noninvasive, high-resolution sampling of a sensory representation without preset limitations on experimental duration. In the present report, individual rats were imaged as often as once a week. Over the time period examined thus far, distinct images of the functional representation of a single whisker were consistently obtained, and features of this representation were repeatedly quantified.

The average size of the whisker representation quantified herein is 1.71 mm² (area at half-height; whisker C2). This areal extent is substantially larger than the areal extent of the

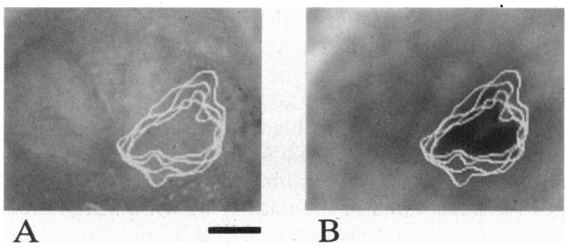

FIG. 3. Consistent location of the functional representation over time. The quantification borders obtained over 4 weeks (shown in Fig. 2) are superimposed on *(A)* an image of the thinned skull showing the major underlying vasculature and *(B)* on an image of the functional activity. Note that the functional representation remains reliably centered on the same area. In this example, the common area shared among all imaging days is more than 60% of the quantified area at half-height common to any individual day. Each image is 5.0 × 4.0 mm cortical area. (Scale bar = 1 mm.)

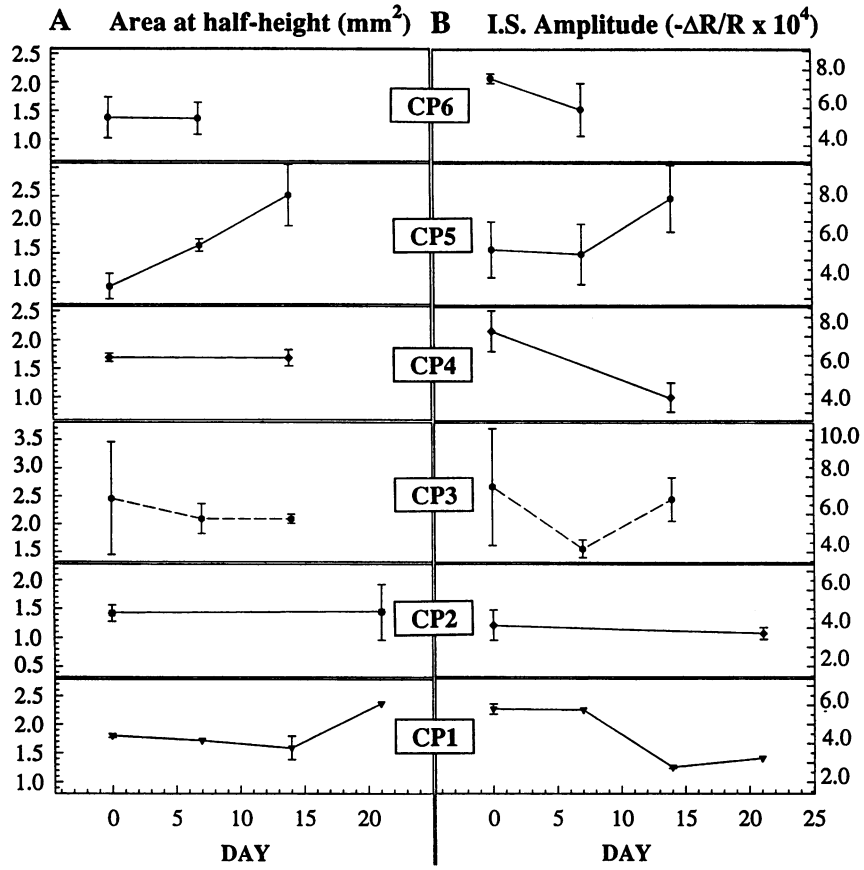

FIG. 4. A comparison of the ongoing changes in the *(A)* area versus the changes in the *(B)* intrinsic signal amplitude. Successive imaging days for each rat are represented including the standard error for the imaging sessions collected during each day. Only a single imaging session was collected on two experimental days (CP1, days 7 and 21), and thus these points lack an error bar. Area is represented in mm^2, and intrinsic signal amplitude is calculated as the peak fractional change in light reflectance (R).

functional representation of a whisker as quantified with 2-deoxyglucose mapping (up to approximately 0.5 mm^2; refs. 23 and 24). Although this area seems large, particularly when considering that only the half-height of the intrinsic signal response area is quantified, both structural and functional evidence supports the potential for a widespread functional area corresponding to a single whisker. For example, injections of anterograde and retrograde tracers into a whisker representation labeled widespread horizontal connections within the upper cortical layers extending to a diameter of ≈2 mm (25). The range of these anatomical connections could account for the large functional area described here, and thus mediate responses to a single whisker throughout the majority of barrel cortex.

In general, electrophysiological studies have not addressed specifically the spatial extent of the cortical area activated by

Table 1. Summary of average area and intrinsic signal amplitude for each day of imaging

Rat	Days after initial surgery	Area (mm^2)		IS amplitude ($-\Delta R/R \times 10^4$)	
		Each day	Average	Each day	Average
CP1	17	1.76	1.83	5.80	4.40
	24	1.68		5.77	
	31	1.55		2.78	
	38	2.32		3.25	
CP2	0	1.45	1.465	4.19	3.88
	21	1.48		3.56	
CP3	5	2.42	2.19	6.99	5.67
	12	2.07		3.70	
	19	2.07		6.32	
CP4	0	1.68	1.66	7.30	5.60
	14	1.64		3.90	
CP5	6	0.93	1.70	4.94	5.75
	13	1.64		4.71	
	20	2.52		7.61	
CP6	50	1.38	1.375	7.61	6.80
	57	1.37		5.98	

Chronological listing of area and intrinsic signal amplitude for each imaging day and the overall average for each subject (CP1–CP6). IS, intrinsic signal.

stimulation of a single whisker. However, some electophysiology experiments provide direct support for our present findings. For example, when systematically recording responses to deflection of a single whisker at several cortical sites, significant responses are recorded in adjacent whisker representations, particularly when long-latency responses are considered (>10 msec; refs. 26 and 27). Furthermore, overlapping and dynamic responses have also been found in the medical division of the ventral posterior nucleus, the thalamic nucleus that connects reciprocally with barrel cortex (28).

Previous recordings of either voltage-sensitive dyes (29, 30) or intrinsic signals (30) in rat barrel cortex also revealed an extensive area responding to stimulation of a single whisker. However, these qualitative studies recorded signal traces of either the dye-based or the intrinsic signal response, and did not visualize or quantify the functional representation of a whisker. To extend our findings, whiskers other than C2 also demonstrate a large functional representation in rat barrel cortex using quantitative ISI (31). Moreover, when relating optical responses and single-unit responses, detailed single-unit recordings after imaging confirmed that the spatial extent of the stimulus-related intrinsic signal response area and the stimulus-related neuronal response area correspond closely (32). Thus, the large area quantified with ISI reflects underlying neuronal activity potentially mediated by horizontal intracortical connections.

In agreement with the precise topography of barrel cortex and corroborated by the correspondence between the imaged representation and the anatomical representation, the spatial location was consistent over time. Previous experiments verified that the functional representation determined with ISI is centered over the anatomical representation of that whisker in layer IV (17). Accordingly, there was considerable overlap of the functional representation of whisker C2 quantified in the same animal on successive imaging days. Even though deflecting an individual whisker activates a large area of barrel cortex, this whisker dominates a reliably located functional area.

Based on these findings, we propose that the traditional view of the functional whisker representation in rat barrel cortex, i.e., a relatively narrow column corresponding to each layer IV barrel (23, 33), is practically applicable only during the first few milliseconds after whisker stimulation (26). We suggest that a description that emphasizes a large area of cortical activation and substantial functional overlap between whisker representations is more accurate. Similar to the present results in which a large cortical area responds to a restricted portion of the sensory periphery (a single whisker), ISI recently revealed that a large area of visual cortex responds to a point stimulus in the visual field (34). This analogous finding in the visual cortex of cats generalizes our results beyond the specific case of rat barrel cortex.

Although the normal range of a functional sensory representation in somatosensory cortex within an animal is seldom evaluated, individual differences in sensory representations have been explored and can be quite dramatic. The predominant source for such differences is most often attributed to unique experiences in the individual animal's life thus far (35, 36). Subtle changes most likely occur throughout life, and are significant after major alterations in sensory stimulation or experience such as during lactation (37) or learning (38, 39).

Individual differences have previously been found in the quantified areal extent of a functional whisker representation in rat barrel cortex. For example, 2-deoxyglucose mapping of the functional representation of whisker C3 in several adult rats also shows that the quantified area (calculated here from the published dimensions of the width and height) exhibited by individual rats may vary by >23% around the mean value for the small group examined (40). Similarly, the diameter of the functional representation of whisker C3 within each cortical layer shows variability across individual animals as assessed by 2-deoxyglucose uptake (23). In agreement with these results, rats showed marked variability in their individual representations of whisker C2 when quantified using ISI.

As expected in the absence of any manipulation and as a verification of the continued integrity of the chronic preparation, the ongoing variability in the whisker representation is unsystematic. Systematic functional reorganization of a cortical area, known as cortical plasticity, has been verified after a variety of experimental manipulations (3–6). Presumably, a manipulation to the system would induce a systematic change in the functional whisker representation. Indeed, using quantitative ISI we have collected data after a sensory manipulation suggesting this to be the case. Unilateral plucking while sparing one whisker throughout development results in a significant and systematic difference between the functional size of the spared whisker as compared with its contralateral control (41).

By sampling repeatedly over time, this investigation evaluated the normal dynamic range in the functional representation of a single whisker. Few previous experiments have performed a high-resolution functional mapping of ongoing dynamics. Results related to the present report are included in Jenkins *et al.* (38) when owl monkeys maintained passive (rather than active) contact of a digit with a static (rather than rotating) disc. Unlike the present experiment, which specifically did not include any manipulation, the passive stimulation group did receive altered sensory input. However, animals sampled before and after passive stimulation did not show the significant changes in their somatosensory cortex that were evident in other monkeys after active disk contact. In addition, there were smaller relative changes in the areal extent of the digit representation than those found here in the whisker representation. However, with only two animals in this passive stimulation group, it is impossible to make detailed comparisons.

Although all of the sources of the variability seen in the present experiments remain to be elucidated, we believe the main source to be ongoing dynamics in cortical function. At this point, we cannot rule out the possibility that subtle alterations in anesthesia level or other physiological parameters contribute to the ongoing variability. However, it is unlikely that these are the main sources of the variability considering that the depth of anesthesia and other physiological parameters (e.g., heart rate, temperature) were carefully monitored. Small fluctuations in the depth of anesthesia within an experiment would presumably be averaged out and would not significantly contribute to the observed differences across days. In acute experiments specifically testing different anesthetic states, Stryker *et al.* (42) concluded that the ongoing variability was larger than the effect of anesthesia or experimental error. Similar sources of variability are most likely producing the long-term changes reported here. In addition, in the present experiment the quantification algorithm accounts for overall levels of activity by normalizing the difference between the peak and the median for each data session (see *Materials and Methods*). Thus, the dynamics imaged here are thought to be ongoing changes in the relative amount of cortex responding to a specific whisker rather than overall changes in the level of cortical excitability.

To summarize, we quantified the size and ongoing dynamics of a whisker representation in rat barrel cortex using ISI. We found that stimulation of an individual whisker reliably activates a large area of barrel cortex, indicating substantial functional overlap between whisker representations. In light of these findings, we propose that a modified description of barrel cortex, one that considers that a single whisker activates a large and dynamic response area, reflects more accurately the functional properties of this area of sensory cortex.

We acknowledge the contributions of Kim Tran, Lloyd Mangnall, and Mandi Anvaripour to the imaging experiments and data analysis.

Also, we thank Cynthia Chen-Bee for a critical review of the manuscript. This work was supported by National Institutes of Health Grant MH-50362, National Institute of Aging Grant A605142–09, National Science Foundation Grant IBN-9507936 (R.D.F.), and National Institutes of Health Training Grant 5 T32 MH14599–17 (S.A.M.).

1. Mountcastle, V. B. (1979) in *The Neurosciences: Fourth Study Program*, eds. Schmidt, F. O. & Worden, F. G. (MIT Press, Cambridge, MA), pp. 21–42.
2. Hubel, D. H. & Wiesel, T. N. (1977) *Proc. R. Soc. London B* **198,** 1–59.
3. Merzenich, M. M., Recanzone, G. H., Jenkins, W. M. & Nudo, R. (1990) in *Natural and Artificial Parallel Computation*, eds. Arbib, M. A. & Robinson, J. A. (MIT Press, Cambridge, MA), pp. 177–210.
4. Gilbert, C. (1995) in *The Cognitive Neurosciences*, ed. Gazzaniga, M. (MIT Press, Cambridge, MA), pp. 73–90.
5. Kaas, J. (1995) in *The Cognitive Neurosciences*, ed. Gazzaniga, M. (MIT Press, Cambridge, MA), pp. 51–72.
6. Weinberger, N. W. (1995) *Annu. Rev. Neurosci.* **18,** 129–158.
7. Grinvald, A., Bonhoeffer, T., Malonek, D., Shoham, D., Bartfeld, E., Arieli, A., Hildesheim, R. & Ratzlaff, E. (1991) in *Memory: Organization and Locus of Change,* eds. Squire, L. R., Weinberger, N. M., Lynch, G. & McGaugh, J. L. (Oxford Univ. Press, New York), pp. 49–85.
8. Frostig, R. D. (1994) in *Cerebral Cortex,* eds. Peters, A. & Rockland, K. S. (Plenum, New York), Vol. 10, pp. 331–358.
9. Frostig, R. D., Masino, S. A., Kwon, M. K. & Chen, C. (1995) *Int. J. Imag. Syst. Technol.* **6,** 216–224.
10. Kim, D. S. & Bonhoeffer, T. (1994) *Nature (London)* **370,** 370–372.
11. Woolsey, T. A. & Van der Loos, H. (1970) *Brain Res.* **17,** 205–242.
12. Killackey, H. P. (1973) *Brain Res.* **51,** 326–331.
13. Welker, C. (1976) *J. Comp. Neurol.* **166,** 173–190.
14. Simons, D. J. (1978) *J. Neurophysiol.* **41,** 798–820.
15. Armstrong-James, M. (1995) in *Cerebral Cortex*, eds. Jones, E. & Diamond, I. (Plenum, New York), Vol. 11, pp. 333–374.
16. Simons, D. J. (1995) in *Cerebral Cortex*, eds. Jones, E. & Diamond, I. (Plenum, New York), Vol. 11, pp. 263–298.
17. Masino, S. A., Kwon, M. C., Dory, Y. & Frostig, R. D. (1993) *Proc. Natl. Acad. Sci. USA* **90,** 9998–10002.
18. Narayan, S. M., Santori, E. M. & Toga, A. W. (1994) *Cerebr. Cortex* **4,** 195–204.
19. Peterson, B. E. & Goldreich, D. (1994) *J. Neurosci. Methods* **54,** 39–47.
20. Chen-Bee, C., Kwon, M. C., Masino, S. A. & Frostig, R. D. (1996) *J. Neurosci. Methods*, in press.
21. Killackey, H. P., Belford, G., Ryugo, R. & Ryugo, D. K. (1976) *Brain Res.* **104,** 309–315.
22. Land, P. W. & Simons, D. J. (1985) *J. Comp. Neurol.* **238,** 225–235.
23. Kossut, M., Hand, P. J., Greenberg, J. & Hand, C. L. (1988) *J. Neurophysiol.* **60,** 829–852.
24. Levin, B. E., Craik, R. L. & Hand, P. J. (1988) *Brain Res.* **443,** 261–271.
25. Hoeflinger, B. F., Bennett-Clarke, C. A., Chiaia, N. L., Killackey, H. P. & Rhoades, R. W. (1995) *J. Comp. Neurol.* **354,** 551–563.
26. Armstrong-James, M. & Fox, K. (1987) *J. Comp. Neurol.* **263,** 265–281.
27. Armstrong-James, M., Fox, K. & Das-Gupta, A. (1992) *J. Neurophysiol.* **68,** 1345–1358.
28. Nicolelis, M. A. L., Lin, R. C. S., Woodward, D. J. & Chapin, J. K. (1993) *Proc. Natl. Acad. Sci. U S A* **90,** 2212–2216.
29. Orbach, H. S., Cohen, L. B. & Grinvald, A. (1985) *J. Neurosci.* **5,** 1886–1895.
30. Grinvald, A., Lieke, E., Frostig, R. D., Gilbert, C. D. & Wiesel, T. N. (1986) *Nature (London)* **324,** 361–364.
31. Chen-Bee, C. H. & Frostig, R. D. (1996) *J. Neurophysiol.*, in press.
32. Frostig, R.D., Masino, S.A. & Kwon, M.C. (1994) *Soc. Neurosci. Abstr.* **20,** 1384.
33. Welker, C. (1976) *J. Comp. Neurol.* **166,** 173–190.
34. Das, A. & Gilbert, C. D. (1995) *Nature (London)* **375,** 780–784.
35. Merzenich, M. M. (1985) in *Comparative Neurobiology: Modes of Communication in the Nervous System,* eds. Cohen, M. J. & Strumwasser, F. (Wiley, New York), pp. 138–157.
36. Merzenich, M. M., Nelson, R. J., Kaas, J. H., Stryker, M. P., Jenkins, W. M., Zook, J. M., Cynader, M. S. & Schoppmann, A. (1987) *J. Comp. Neurol.* **258,** 281–296.
37. Xerri, C., Stern, J. M. & Merzenich, M. M. (1994) *J. Neurosci.* **14,** 1710–1721.
38. Jenkins, W. M., Merzenich, M. M., Ochs, M. T., Allard, T. & Guic-Robles, E. (1990) *J. Neurophysiol.* **63,** 82–104.
39. Recanzone, G. H., Merzenich, M. M., Jenkins, W. M., Grajski, K. A. & Dinse, H. R. (1992) *J. Neurophysiol.* **67,** 1031–1056.
40. Code, R. A., Eslin, D. E. & Juliano, S. L. (1992) *Exp. Brain Res.* **88,** 341–344.
41. Masino, S. A. & Frostig, R. D. (1995) *Soc. Neurosci. Abstr.* **21,** 123.
42. Stryker, M. P., Jenkins, W. M. & Merzenich, M. M. (1987) *J. Comp. Neurol.* **258,** 297–303.