the past 3 years of experimentation, the 100 percent susceptibility of all inoculated animals, the reduction of the incubation period, the duration of the disease, and the nature and distribution of the lesions as seen with the light and electron microscope (*6*).

ELIAS E. MANUELIDIS
Departments of Pathology and *Neurology, Yale University School of Medicine, New Haven, Connecticut 06510*

References and Notes

1. C. J. Gibbs, Jr., D. C. Gajdusek, D. M. Asher, M. P. Alpers, E. Beck, P. M. Daniel, W. B. Matthews, *Science* **161**, 388 (1968).
2. C. J. Gibbs, Jr., and D. C. Gajdusek, *ibid.* **165**, 1023 (1969); D. C. Gajdusek and C. J. Gibbs, Jr., *Nature (Lond.)* **230**, 588 (1971); C. J. Gibbs, Jr., and D. C. Gajdusek, *Science* **182**, 67 (1973).
3. B. Brownell, M. J. Campbell, L. W. Greenham, Annual Meeting, American Association of Neuropathologists, 51st, New York, 30 May to 1 June 1975, abstract.
4. The neurological disease of this 54-year-old female patient became apparent 1 month before she was admitted to the hospital. She showed forgetfulness, fatigue, difficulty with her balance, and had a short episode of blurring of vision. In addition she had been rather lethargic and slept much of the time. On admission to the Hartford Hospital, 3 months prior to death, the patient was found to walk cautiously with broadened gait. Coordination, sensation, motor power, and reflexes were intact. A pneumoencephalogram showed a minimal dilation of the ventricular system and subarachnoidal space. Ten days later the patient exhibited gradually increased difficulty with her mentation and myoclonic seizures of her extremities, that were more severe on the right side. The clinical course progressively worsened and the patient was transferred to the Yale–New Haven Hospital. Here she exhibited marked mental deterioration, was bedridden, mute, and incontinent. She assumed a flexion posture of the upper extremities and her hands were tightly gripped; the legs showed a flaccid tone. There was a 3+ jaw jerk and ankle clonus. The electroencephalograph recordings showed marked decrease in activity and finally became flat except for myoclonic bursts of activity. A cerebral biopsy was made 10 days before the patient developed pneumonia and died.
5. P. W. Lampert, D. C. Gajdusek, C. J. Gibbs, Jr., *Am. J. Pathol.* **68**, 626 (1972).
6. E. E. Manuelidis, in preparation.
7. The technical assistance of P. Johnson is gratefully acknowledged. Supported by grants CA-15044-02 and NS-12674-01.

19 May 1975; revised 6 August 1975

Commissural and Cortico-cortical "Columns" in the Somatic Sensory Cortex of Primates

Abstract. *Anatomical experiments demonstrate that commissural and cortico-cortical fibers arising and terminating in the somatic sensory cortex of monkeys terminate in layers I through IV in a mosaic of precisely ordered vertical bands. The cells of origin of these fibers, found predominantly in layer III, are also arranged in vertical aggregations.*

The sensory areas of the cerebral cortex have a pronounced vertical cellular organization that can be demonstrated anatomically (*1*) and electrophysiologically (*2*). In some areas (*3*) this verticality seems to be related to a precise ordering of the thalamic afferent fibers entering the area. In the visual cortex, for example, bundles of thalamic afferents are aligned in register so as to form a series of bands approximately 500 μm wide, each of which is related to one eye. In the somatic sensory cortex, individual columns of similar dimensions are related to a single modality (*3*).

In our study of the somatic sensory cortex in rhesus (*Macaca mulata*) and squirrel (*Saimiri sciureus*) monkeys, commissural and cortico-cortical fibers have also been found to terminate in discrete vertical groupings. The cells of origin of the commissural fibers and, to a lesser extent, those of cortico-cortical fibers have been found to be arranged in distinct clusters.

Commissural and cortico-cortical fibers of eight monkeys were demonstrated with autoradiography (*4*); this method depends upon the axoplasmic transport of isotopically labeled proteins from the cell body to the terminals of its axon. The tritium-labeled amino acids proline and leucine were injected singly or multiply into the first somatic sensory area (SI). Single injections containing 5 μc of radioactivity in 0.1 μl of solution caused heavy labeling (*4*) of cells over an area of approximately 1 to 2 mm^2. After survival periods ranging from 12 hours to 6 days, the brains were prepared for autoradiography (*4*). Multiple, large injections containing up to 100 μc of activity coalesce so as to heavily label virtually all of SI. Transported label is accumulated in the opposite SI and SII, though only in the face, head, trunk and proximal limb representations (*5*). Within these parts of the body representation, the cortex is not uniformly labeled. Instead, the label resolves itself into a number of blocklike formations (Fig. 1C); in each of these, a few loosely packed grains extend from the white matter up to layer IV. Here a band of intense terminal labeling expands in width to as much as 600 to 1000 μm within the confines of layer IV. Superficial to layer IV, the grain concentration becomes reduced and the band narrows, but it continues to layer I. A second band of moderately intense terminal labeling is commonly situated in layer II and the upper half of layer III. After a short postoperative time (12 to 24 hours), when most of the transported label is located in axon terminals (*4*), the labeling in layers V and VI is indistinct; this suggests that the majority of the commissural fibers terminate in the granular and supragranular layers.

In SII, the bands of terminals have the same laminar distribution, but they tend to be slightly wider. In both SI and SII, the interval between adjacent bands varies according to the part of the representation examined, but in the face area, which receives many bands (Fig. 1C), the interval is usually between 500 and 1000 μm wide. A single small injection of isotope in SI in an area of about 1 mm^2 leads to labeling of only one or two columnlike bands of commissural fibers in the opposite SI and SII.

Since the discontinuous distribution of the commissural fibers demonstrated autoradiographically might result from the unequal labeling of the different parts of SI (even with a large number of injections of isotope), three additional experiments were performed. The whole of SI was ablated, and a survival period of 5 days was allowed. The brains were then prepared by the Wiitanen modification of the Nauta method for staining degenerating axons and their terminals (*6*). The total complement of commissural fibers was thereby labeled. Orthographic reconstructions (Fig. 2, A and B) show the size and relative distribution of the bands of commissural fiber terminals as determined from measurements of the widest parts of the foci of terminal degeneration (which, as in the case of the autoradiographic experiments, occurs in layer IV).

Though SI contains four cytoarchitectonic fields (areas 3a, 3b, 1, and 2, recognized by counterstaining the Nauta preparations with a cellular stain), the distribution of the commissural bands is not by area. In the face and trunk representations, all four areas contain labeled bundles arranged in an anteroposterior sequence across SI. Apart from these, a mediolaterally oriented row extends along the representation of axial portions of the body at the junction of areas 1 and 2 (Fig. 2A).

In autoradiographs and counterstained Nauta preparations, it is often possible to see (Fig. 1, B and C) that each commissural fiber bundle is associated with an aggregation of five or more of the large pyramidal cells typical of the deep one-half to two-thirds of layer III (layer III B) (*7*). In intervening portions of the cortex lying between the commissural fiber bundles and in parts of SI and SII related to the distal aspects of the limbs, the large pyramidal cells, though still present, often appear fewer in number and tend to be confined to the deepest part of layer III B.

Five correlative experiments used the uptake by axon terminals and the retrograde axonal transport of the enzyme horseradish peroxidase (*8*) to demonstrate the cells of origin of the commissural fibers. The large pyramidal cells of layer III B are the only cells in SI to give rise to

commissural fibers (Fig. 1A). After multiple, very large injections (up to 1000 μg) of the enzyme in SI of one side, the cells in layer III B in the face, head, trunk, and proximal limb regions of the contralateral SI are labeled in discrete clusters 500 to 1000 μm wide (Fig. 1A) (*9*). Although there remains the possibility that the whole of the ipsilateral SI was not equally labeled, it is, nevertheless, possible to make a map comparable to that generated from the distribution of the bundles of commissural axons (Fig. 2C). This suggests that commissural axons may arise from and terminate upon exactly homotopic groups of pyramidal cells. The bilaminar pattern of termination described might reflect terminations of the commissural axons on the pronounced tufts of dendritic branches generated in layers III A and IV, respectively, by the primary apical and basal dendrites of these nerve cells.

Discrete vertical bundles of ipsilateral cortico-cortical fibers have also been demonstrated in the somatic sensory area and in certain related cortical areas. Multiple, but discontinuous, injections of labeled amino acids in SI label a dense band of terminals in layer IV of the ipsilateral SII, but at intervals, a band of terminal labeling between 500 and 800 μm wide ascends through the supervening layers to the deep part of layer I (Fig. 1D). Each ascending band is separated from its neighbors by a gap of approximately 500 μm. Each is considered to represent the terminals of a bundle of cortico-cortical axons emanating from cells at the center of each injection mass. Each bundle demonstrated anatomically is a topographic one and cannot, therefore, be considered a functional unit in the sense of the electrophysiological "column" (*2*), but we believe it to be a reflection of the basic vertical pattern of organization upon which such columns must depend. That is, more than one functional column of cortical cells may be contained within each ascending bundle of cortico-cortical axons.

After single small injections of isotope in areas 3a and 3b of SI, single labeled bundles are found in SII and in areas 1, 2, and 5. Each again consists of a band about 500 μm wide in layers I through IV. The cortico-cortical bands are, thus, similar to the vertical arrays of commissural fibers; despite the obvious uncertainty of obtaining injections of comparable size in different experiments, the relative consistency in the dimensions of the bands is striking.

Experiments with the retrograde, horseradish peroxidase tracing method show that most cortico-cortical axons arise from pyramidal cells of all sizes in layers III A and III B, while fewer arise in layers II and V and virtually none in layer VI. After in-

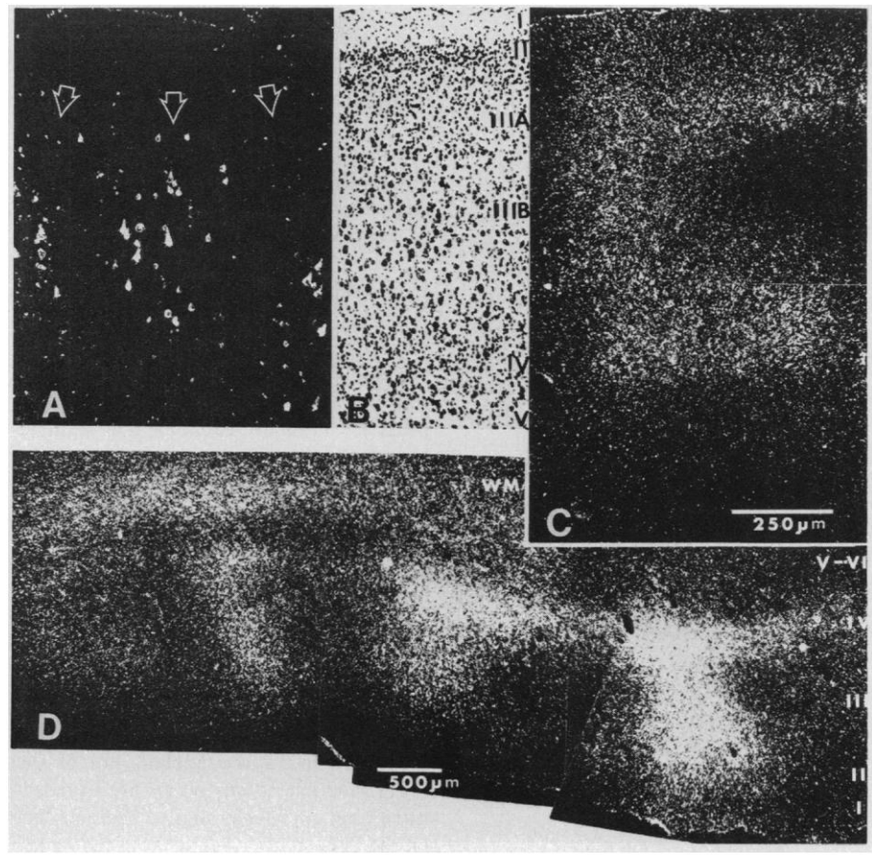
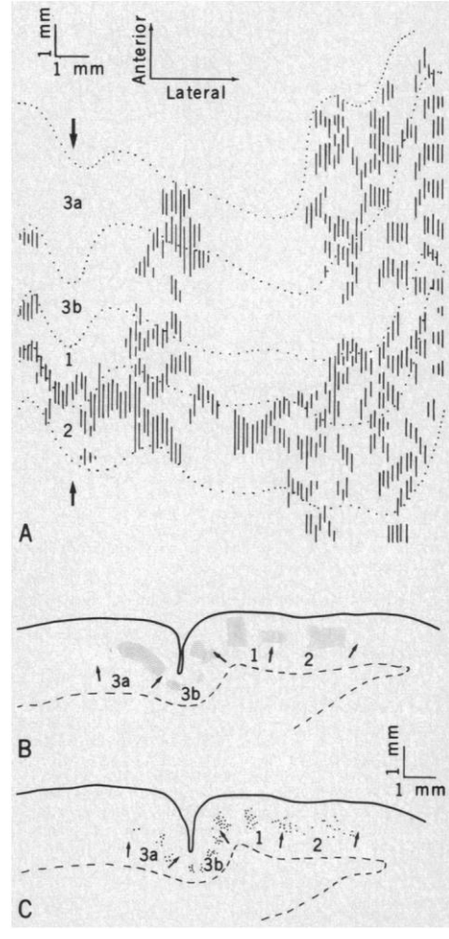


Fig. 1 (top). (A to C) Three photomicrographs at the same magnification. (A) A dark-field preparation, showing three vertical groupings of pyramidal cells in layer II B labeled by retrograde transport of horseradish peroxidase; the injection labeled an area of the opposite SI considerably larger than that shown here. (B and C) Respectively bright- and dark-field photomicrographs of the same part of SI showing the laminar distribution of a vertical band of commissural axon terminals labeled autoradiographically. (D) Dark-field photomicrograph showing several vertical arrays of labeled cortico-cortical axon terminals in SII of a squirrel monkey following multiple, small injections of isotopically labeled amino acids in the ipsilateral SI; *WM*, labeled axons in white matter. This figure is inverted with respect to (B) and (C) because SII lies in the upper bank of the lateral sulcus. Fig. 2 (right). (A) Expanded surface view of the four architectonic fields (3a, 3b, 1, and 2) of the SI cortex of a squirrel monkey showing columnar distribution of degenerating commissural axon terminals in a brain in which the contralateral SI was removed 5 days previously. Each vertical line represents a band of terminal degeneration from a single parasagittal section. Unshaded areas receive no commissural terminals. The largest of these are the hand and foot representations. Arrows indicate medial border of hemisphere where SI extends from lateral onto medial surface. (B) Projection drawing of a single parasagittal section through the trunk representation showing the patches of terminal degeneration in layers II to IV. (C) Projection drawing of a comparable parasagittal section from a brain in which commissurally projecting cells (each indicated by a dot) were labeled by the retrograde transport of horseradish peroxidase. The cells lie in layer III B and are arranged in clusters similar to the terminal bundles of commissural fibers indicated in (B).



jections of the enzyme in SI, retrogradely labeled cells in SII and in the motor cortex (area 4) appear in narrow vertical groupings, which may be single or multiple, depending on the size of the injections.

These observations provide further evidence that the sensory cortex can be considered as a mosaic of functional units based on afferent input. In the two types of afferent systems examined here, the columnar pattern probably reflects a precise topographic ordering of cells and their axons. The width of the columnlike bands of axons and terminals, and their distribution, when taken in conjunction with available physiological data (*2*), make it unlikely that each band represents a single somesthetic modality; however, they probably indicate the anatomical organization that underlies the cortical representation of both place and modality. The investigation also shows, with previous studies on the thalamic afferents (*7*), that the granular and supragranular layers are the sites of termination of all three major afferent systems. This points up an interesting duality in the organization of the cortex, for evidence is accruing that the efferent fibers to subcortical centers such as the thalamus, midbrain, pons, and dorsal column nuclei arise only in layers V and VI (*10*).

E. G. JONES
H. BURTON, R. PORTER
Department of Anatomy, Washington University School of Medicine, St. Louis, Missouri 63110

References and Notes

1. R. Lorente de Nó, *Physiology of the Nervous System* (Oxford Univ. Press, London, 1938), pp. 288–313.
2. M. Abeles and M. N. Goldstein, Jr., *J. Neurophysiol.* **33**, 172 (1970); D. H. Hubel and T. N. Wiesel, *J. Physiol. (Lond.)* **160**, 106 (1962); V. B. Mountcastle and T. P. S. Powell, *Bull. Johns Hopkins Hosp.* **105**, 173 (1959).
→ D. H. Hubel and T. N. Wiesel, *J. Comp. Neurol.* **146**, 421 (1972); H. P. Killackey, *Brain Res.* **51**, 326 (19 → T. N. Wiesel, D. H. Hubel, D. M. K. Lam, *ibid.* **79**, 273 (1974).
→ W. M. Cowan, D. I. Gottlieb, A. E. Hendrickson, J. L. Price, T. A. Woolsey, *Brain Res.* **37**, 21 (1972).
5. E. G. Jones and T. P. S. Powell, *Brain* **92**, 717 (1969); D. N. Pandya and L. A. Vignolo, *Brain Res.* **15**, 49 (1969).
6. J. T. Wiitanen, *Brain Res.* **14**, 546 (1969).
7. E. G. Jones, *J. Comp. Neurol.* **160**, 167 (1975).
8. J. H. LaVail, K. R. Winston, A. Tish, *Brain Res.* **58**, 470 (1973).
9. S. Jacobson and J. Q. Trojanowsky [*ibid.* **74**, 149 (1974)] observed retrograde labeling of pyramidal cells in both layers III and V of the rhesus monkey frontal cortex following injections of horseradish peroxidase on the opposite side.
10. C. G. Gilbert and J. P. Kelly, paper presented at the fourth annual meeting of the Society for Neuroscience, St. Louis, 20 to 23 October 1974; R. D. Lund, J. S. Lund, A. H. Bunt, A. E. Hendrickson, A. F. Fuchs, *ibid.*; S. P. Wise, *Brain Res.* **90**, 139 (1975).
11. Supported by NIH grants NS-10526 and NS-09809. R.P. was on leave from the Department of Physiology, Monash University, Clayton, Victoria, Australia, and was supported by a Fulbright-Hays scholarship and by Washington University. We thank B. McClure and M. W. Rhoades for technical and photographic assistance.

3 April 1975; revised 5 May 1975

Delayed-Type Hypersensitivity to Sheep Red Blood Cells: Inhibition of Sensitization by Interferon

Abstract. *Interferon, when given or induced 24 hours before contact of mice with sheep red blood cells, prevented sensitization, and no delayed-type hypersensitivity reaction could be elicited 4 days later, after challenge with the antigen, as shown by the absence of footpad swelling in treated animals.*

Interferon, originally discovered because of its antiviral properties, has more recently been shown to affect the immune system (*1*). Our group has been interested mainly in the effects of interferon on cell-mediated immune reactions, and we have previously reported that, in mice sensitized to picryl chloride or sheep red blood cells (SRBC), expression of delayed-type hypersensitivity (DTH) is significantly inhibited if the animals are treated with interferon on the day before or the day of challenge with the antigen (*2*). We believe that this phenomenon contributes to the frequently described inhibition of DTH which occurs as a result of virus infection (*3*).

In view of this very pronounced action of interferon on the efferent arc of the DTH reaction, it was important to determine whether interferon also had an effect on the afferent arc, that is, the actual sensitization. This was examined with two different approaches: induction of interferon with Newcastle disease virus (NDV) in congenic high and low interferon producing mice, and administration of exogenous interferon.

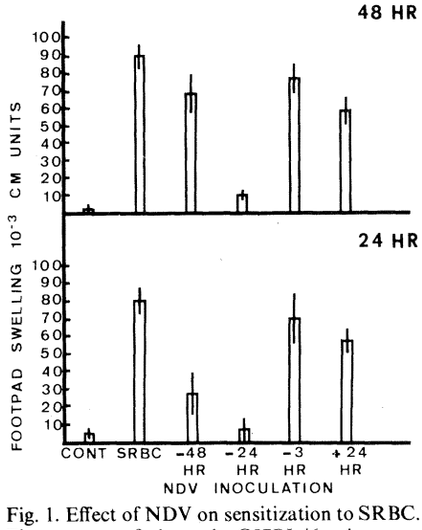


Fig. 1. Effect of NDV on sensitization to SRBC. Five groups of six male C57BL/6 mice were sensitized against SRBC. Three of these had been inoculated intravenously with 10^7 PFU of NDV, respectively 48, 24, or 3 hours before sensitization. The fourth group received NDV 24 hours after SRBC and the fifth group, which served as a sensitized control, received SRBC without any NDV injection. All mice, 4 days after sensitization, plus an additional group of unsensitized animals, were injected with 10^8 SRBC into the left footpad. Average footpad swelling (± S.E.) is graphically represented for each group 48 hours and 24 hours after sensitization. All values were virtually back to normal after 72 hours. Abbreviations: cont, unsensitized controls; SRBC, sensitized controls (SRBC without NDV injection).

The induction of DTH to SRBC in mice was carried out by the procedure of Lagrange *et al.* (*4*), in which mice are sensitized with an intravenous inoculation of 10^6 SRBC suspended in phosphate-buffered saline (PBS) and 4 days later, challenged by an inoculation into the left footpad of 10^8 SRBC suspended in 40 μl of PBS. Footpad swelling is measured with dial-gauge calipers 24, 48, and 72 hours later, and is expressed as the difference between the left (challenged) and the right (unchallenged) foot. Histological examinations made during previous experiments had shown that DTH reaction typically occurred 24 hours after footpad inoculation (*2*).

An experiment was carried out to determine whether interferon had any effect on sensitization to SRBC, and, if so, to determine the optimal conditions of timing. Different groups of six C57BL/6 mice received one intravenous inoculation of 10^7 plaque-forming units (PFU) of NDV, each group at a different time with regard to the time of sensitization, which was the same for all animals. The amount of virus inoculated was calculated to induce about 200,000 units (*5*) of circulating interferon at peak levels, that is, about 9 hours after virus injection. Footpad swelling, measured 24 and 48 hours after challenge, is illustrated in Fig. 1. All groups reacted to footpad challenge except the one that had received NDV 24 hours before sensitization, whose members behaved like nonsensitized animals. The fact that NDV had no effect when given on the day of or the day after sensitization was important, since it ruled out the possibility that the inhibition of footpad swelling in mice inoculated on the day preceding sensitization might have been due to an effect on the reactive or efferent phase.

That NDV acted through interferon induction was confirmed in congenic high and low interferon producers. Two strains of mice, genetically identical as far as all identifiable loci are concerned, but differing by their alleles at the If-1 locus, were used. One strain carries the If-1^h