A Potential Noninvasive Neurobiological Test for Alzheimer's Disease

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Currently Alzheimer's disease, which affects more than 20 million people worldwide, can only be definitively diagnosed by histological examination of brain tissue obtained at autopsy or biopsy. There is a great need for an early, noninvasive, sensitive, and easily administered diagnostic test of Alzheimer's disease. Here it is reported that patients diagnosed with probable Alzheimer's disease by standard clinical criteria exhibited a marked hypersensitivity in their pupil dilation response to a cholinergic antagonist, tropicamide, placed in their eyes. It was possible to distinguish 18 of 19 individuals (95%) either clinically diagnosed with Alzheimer's disease or classified as suspect Alzheimer's individuals by neuropsychological screening from 30 of 32 normal elderly controls (94%).

Alzheimer's disease is a neurodegenerative disorder of the central nervous system that affects almost 1 in 10 individuals who survive beyond the age of 65. With advancing age, the prevalence of the disease increases to an estimated 19% for individuals 75 to 85 years old and greater than 45% for individuals over 85 (*1*). The disease is characterized by progressive memory loss and the decline of other higher cognitive functions such as attention (*2–5*). This cognitive decline is presumably the consequence of the synaptic loss and extensive neuronal cell death that occur in regions of the brain involved in cognition and memory (*6, 7*). Two characteristic neuropathological lesions have been found in these regions: intracellular neurofibrillary tangles, composed of abnormally phosphorylated cytoskeletal proteins; and complex protein deposits, called amyloid, consisting primarily of a ~40–amino acid peptide, Aβ (*5, 7–9*). In addition, the amyloid deposits contain an inflammation-associated antiprotease (α-antichymotrypsin), the enzyme butyryl cholinesterase (*10*), and a protein involved in lipid transport (apolipoprotein E) (*11, 12*). The presence of amyloid deposits and neurofibrillary tangles are required for the definitive diagnosis of the disease but can only be detected by microscopic examination of brain tissue (usually at autopsy). In the absence of such histopathological analysis, a provisional diagnosis of probable or possible Alzheimer's disease can be rendered only after extensive neurological and neuropsychological testing.

Currently Alzheimer's disease may be incorrectly diagnosed clinically in as many as 25 to 40% of cases in nonresearch diagnostic settings (*1, 13*). Neuropsychological tests commonly used in the diagnosis of Alzheimer's disease are relatively insensitive to cognitive changes in the very early stages of the disease and are often not specific (*14*). It would be very useful to have a test for Alzheimer's disease that could be administered during life and would allow for early and positive diagnosis. Such a test would make it easier to identify appropriate individuals for future pharmacological treatments that would be aimed at stemming the progression of the disease.

The potential test for Alzheimer's disease reported here derives from a consideration of certain similarities between patients with Down syndrome and patients with probable Alzheimer's disease. Individuals with Down syndrome who live beyond the age of 30 develop the same brain lesions that characterize Alzheimer's disease and in most cases exhibit a related dementia (*8, 15–20*). This similarity encouraged us to search for physiological characteristics associated with Down syndrome that might also be present in Alzheimer's disease (*21, 22*). It has been shown that individuals with Down syndrome exhibit a hypersensitivity to compounds that act as antagonists of acetylcholine neurotransmission (*22–24*). This hypersensitivity can be detected by measuring changes in heart rate or pupil size in response to these agents (*22–24*). In this report we show that patients with clinically diagnosed Alzheimer's disease are hypersensitive to the pupil-dilating effect of the acetylcholine receptor antagonist tropicamide.

We tested 58 individuals for their pupil response to a very dilute solution of tropicamide. These subjects were divided into five experimental groups (two groups of patients and three control groups of community-dwelling elderly individuals). One patient group consisted of 14 individuals who had been previously diagnosed with probable Alzheimer's disease on the basis of standard clinical criteria (*25, 26*). The other patient group was a pilot sample of four patients diagnosed with one of the following non-Alzheimer's type dementias: Korsakoff's syndrome, multi-infarct dementia, and dementia with an extrapyramidal syndrome. The remaining 40 elderly individuals were assigned to one of three control groups according to neuropsychological screening criteria defined before the initiation of the study. Normal controls consisted of 32 individuals who performed at or above age norms on a battery of neuropsychological tests that assessed intellectual capacity, attention, memory, and language (*26*). Five individuals whose performance yielded abnormalities in memory and discrepancies between estimated life-long intelligence quotient and current performance in cognitive tests were classified as "suspect" Alzheimer's individuals. Three individuals who exhibited abnormal performance on cognitive tests but had no salient memory deficit were classified as "cognitively abnormal" elderly for this study.

Before being tested, subjects were seated in a comfortable semidarkened room 1.5 m in front of a television screen and given sufficient time (2 to 3 min) for their eyes to accommodate to the dim illumination. After resting pupil diameter (baseline) measurements were recorded for 1 min from each eye, a single drop of a very dilute solution of tropicamide was administered to one eye (arbitrarily chosen) and a drop of a control solution (sterile water) to the other eye. The researcher administering the drops was blind to which solution was being applied to which eye. Pupil diameter data were obtained from each eye for 30-s samples at scheduled times over the course of 1 hour.

A comparison of the pupil dilation response of patients with clinically diagnosed Alzheimer's disease and experimental controls to the acetylcholine receptor antagonist tropicamide is shown in Fig. 1. As expected, the treated pupils of the normal controls (Fig. 1A, lower curve) showed a minimal increase in pupil diameter over the course of the hour. In contrast, the patients clinically diagnosed with Alzheimer's disease displayed a pronounced response to the pupil-dilating effect of tropicamide (Fig. 1A, upper curve). A comparison of the response of patients with clinically diagnosed Alzheimer's disease, the suspect Alzheimer's individuals, the cognitively abnor-

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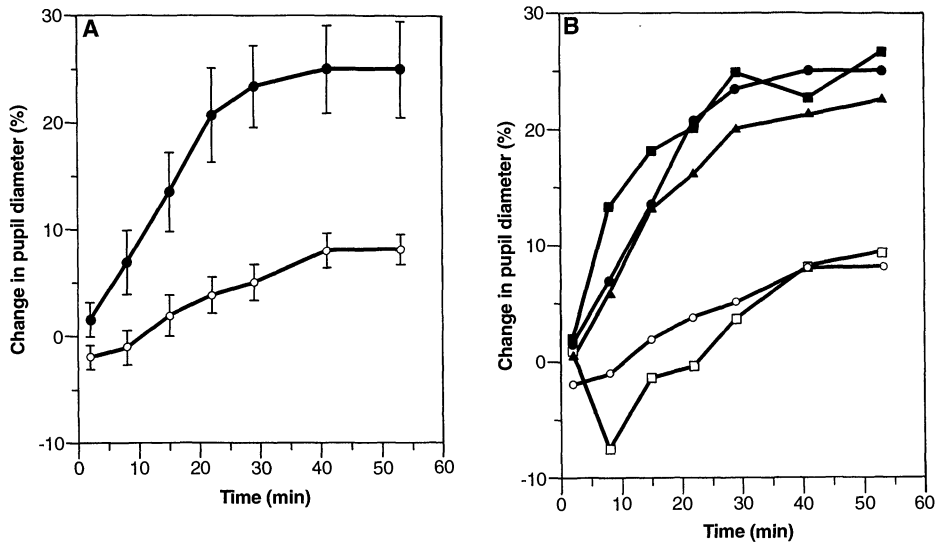

Fig. 1. Pupil dilation response to the acetylcholine receptor antagonist tropicamide in patients with probable Alzheimer's disease and experimental control groups. The pupil dilation (mydriasis) of patients with probable Alzheimer's disease and experimental control groups was elicited by the topical application of tropicamide in one eye and a solution of sterile water applied to the other eye. Tropicamide, a synthetic analog of atropine, is commonly used in ophthalmology to dilate the pupil and allow examination of the fundus. Solutions of tropicamide at 0.5 to 1.0% are normally used to dilate the pupil maximally in 20 to 40 min. The concentration of tropicamide used in this study (0.01%) was chosen so as to cause minimal dilation of normal eyes. After application of tropicamide, pupil diameter was measured seven times over the course of 1 hour for 30 s at 2, 8, 15, 22, 29, 41, and 51 min. Pupil diameter was measured with a video-based pupil center-to-corneal reflection system capable of measuring eye position and pupil diameter (Applied Science Laboratories, Bedford, Massachusetts). The sampling rate was 60 Hz, yielding 1800 samples per 30-s measurement cycle. Between measurement intervals, subjects were shown segments from the videotape *Fantasia* to reduce anxiety, fatigue, and boredom from sitting in a semi-darkened room. Each time point shown in (**A**) represents the mean percentage change in pupil diameter over resting pupil diameter (baseline) measurement in the treated eye of patients and normal controls. A Kruskal-Wallis pairwise multisample test was used to determine the significance of the differential tropicamide sensitivity of the Alzheimer's (●) and control groups (○). Overall, the results indicated that at minute 29 there was a 23.4% (SEM 3.8%) change in the pupil diameter of patients with probable Alzheimer's disease compared with a 5% (SEM 1.7%) change for normal subjects (P = 0.009). The five curves representing all subjects fall broadly into two groups. As shown in (**B**), the percentage change in pupil diameter of the treated eye over baseline of the suspect Alzheimer's subjects (▲) and the cognitively abnormal subjects (■) closely parallels that of the patients with probable Alzheimer's disease (●), whereas the patients with non-Alzheimer's type dementia (□) exhibit a pattern like that of normal controls (○).

mal subjects, the patients with non-Alzheimer's type dementia, and the normal controls is shown in Fig. 1B. Both the suspect Alzheimer's disease individuals and the cognitively abnormal subjects show an almost identical pattern of pupillary response to that of patients with clinically diagnosed Alzheimer's disease. In contrast, the response of the group of patients diagnosed with non-Alzheimer's type dementia was similar to the performance of the normal controls.

The complete set of data for the minute 29 sampling point (the point of maximal separation of clinically diagnosed Alzheimer's patients and normal elderly control subjects) is presented in Fig. 2A. Each symbol represents the percent change in pupil size over baseline of a single individual. The means and the ±95% confidence intervals of the means for patients with probable Alzheimer's disease and the normal controls are plotted in Fig. 2B. There was a clear separation between these groups beginning

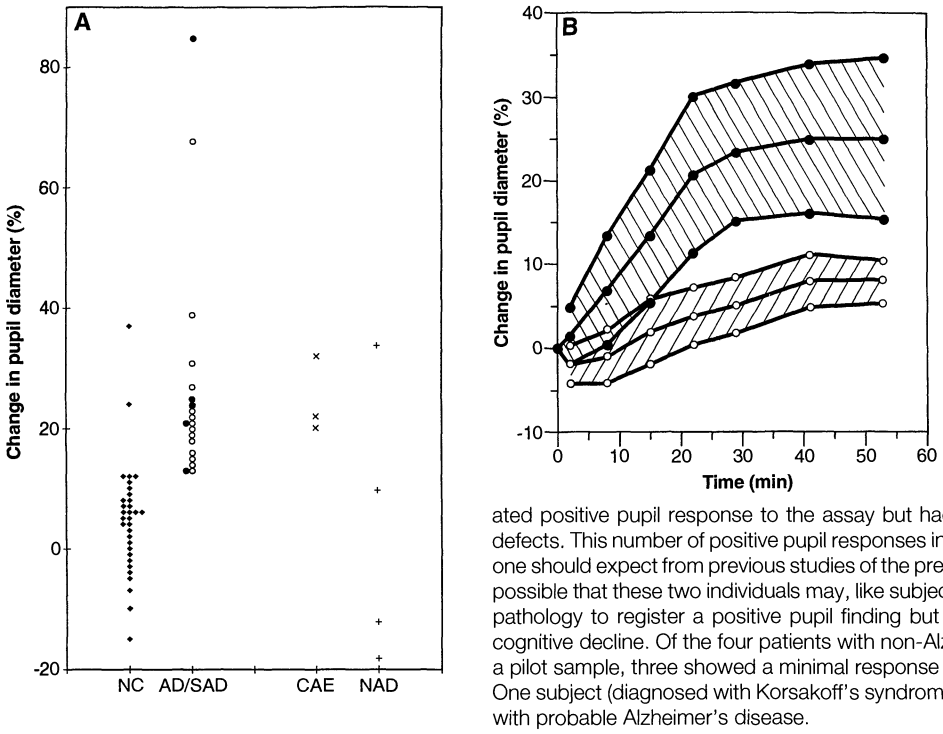

Fig. 2. Individual pupil dilation scores (percent change over baseline) at minute 29 for patients with probable Alzheimer's disease and all control subjects (**A**), and mean percentage change in pupil dilation with ±95% confidence intervals for patients with probable Alzheimer's disease and normal elderly controls (**B**). A minimum overlap in the pupil dilation scores between groups and between individuals in different groups was obtained by designating 13% change in pupil diameter at minute 29 of the assay as a cutoff point. Of the 40 elderly subjects from the community that were tested [normal elderly controls (NC), suspect Alzheimer's dementia individuals (SAD), and cognitively abnormal elderly subjects (CAE)], nine showed a positive response to the assay that was ≥13% at minute 29, of which seven were either in the suspect Alzheimer's disease group or the cognitively abnormal elderly group. Thus, only 2 of 32 normal elderly controls exhibited an exaggerated positive pupil response to the assay but had no other clinically notable cognitive or neurological defects. This number of positive pupil responses in our "normal" sample is within the order of magnitude one should expect from previous studies of the prevalence of this disease in the community. It is therefore possible that these two individuals may, like subject SG discussed in the text, have sufficient Alzheimer's pathology to register a positive pupil finding but do not yet exhibit clinically discernible symptoms of cognitive decline. Of the four patients with non-Alzheimer's type dementia (NAD) who were included as a pilot sample, three showed a minimal response to the pupil assay and reacted as the normal sample. One subject (diagnosed with Korsakoff's syndrome) exhibited a pupil response similar to that of patients with probable Alzheimer's disease.

at minute 15. This distinct separation between the groups was maintained at minute 29 after instillation for the ±99% confidence intervals (*27*).

These graphs indicate that, with few exceptions, both the patients with a diagnosis of probable Alzheimer's disease and the subjects we have classified as "suspect" Alzheimer's individuals could be distinguished from the normal controls on the basis of their hypersensitivity to tropicamide. Furthermore, the fact that the response of patients in our pilot sample with non-Alzheimer's–type dementias is similar to that of normal controls suggests that the pupil dilation assay may be specific for Alzheimer's pathology, although more studies will be necessary to demonstrate specificity and to determine the mechanism of the response (*28*).

When we combine the data from the 14 patients with probable Alzheimer's disease and the 5 subjects we have classified as suspect Alzheimer's individuals, 18 of 19 subjects exhibited a positive response to the pupil dilation assay. This 95% concordance between the clinical or suspected diagnosis and the results of the pupil assay is consistent with the finding in our dementia clinic that 95% of patients whom we clinically diagnose with probable Alzheimer's disease and who are subsequently brought to autopsy have pathologically confirmed Alzheimer's disease (*29*).

Several findings from this study suggest that the tropicamide pupil dilation test may be able to identify individuals with Alzheimer's disease before the onset of clinical symptoms of dementia. First, patients with a clinical diagnosis of Alzheimer's disease who exhibited an exaggerated mydriatic response included the most mildly demented individuals as measured by the Information-Concentration-Memory subtest of the Blessed Dementia Rating Scale, and we found no correlation between patients' dementia scores and a positive pupil result. The lack of such a correlation suggests that the pupil assay may be sensitive to the earliest stages of the disease. Secondly, almost all of the elderly individuals living in the community whom we tested and who showed a positive pupil response also exhibited neuropsychological deficits and most were found to have a salient memory impairment consistent with Alzheimer's disease.

Of particular interest is the case of patient SG. This elderly subject living in the community initially exhibited a positive pupil response to tropicamide but showed no obvious cognitive deficits and only a self report of mild difficulty with some daily living activities. He was retested 9 months later and continued to show a positive pupil response. During this interval he exhibited a substantial decline (from 0 to 6) on the Information-Concentration-Memory subtest of the Blessed Dementia Rating Scale and developed clear memory deficits. These results indicate that the pupil dilation assay was sensitive enough to detect an abnormal response in an elderly community-dwelling individual who subsequently developed symptoms consistent with a diagnosis of probable Alzheimer's disease.

If these early results are supported by further testing, the tropicamide pupil dilation test might be able to identify Alzheimer's patients early in the disease process, when they could most benefit from therapies currently in clinical trials designed to slow the progression of the disease. Unlike other biochemical and physiological tests now being developed, the pupil dilation response is safe, relatively noninvasive, sensitive, and easy to quantitate with already available, automated instrumentation.

REFERENCES AND NOTES

1. D. A. Evans, *J. Am. Med. Assoc.* **262**, 2551 (1989).
2. L. F. M. Scinto *et al.*, *Arch. Neurol.* **51**, 682 (1994).
3. L. L. Heston, A. R. Mastri, V. E. Anderson, J. White, *Arch. Gen. Psychiatry* **38**, 1084 (1981).
4. B. A. Yankner and M. M. Mesulam, *N. Engl. J. Med.* **325**, 1849 (1991).
5. R. D. Terry and R. Katzman, in *The Neurology of Aging*, R. Katzman and R. D. Terry, Eds. (Davis, Philadelphia, 1983), pp. 51–84.
6. J. P. Blass, *Neurology* **43** (suppl. 4), 825 (1993).
7. D. L. Price, *Annu. Rev. Neurosci.* **9**, 489 (1986).
8. G. G. Glenner and C. W. Wong, *Biochem. Biophys. Res. Commun.* **122**, 885 (1984).
9. B. Muller-Hill and K. Beyreuther, *Annu. Rev. Biochem.* **58**, 287 (1989).
10. M. M. Mesulam and C. Geula, *Ann. Neurol.*, in press.
11. Y. Namba, M. Tomonaga, H. Kawasaki, E. Otomo, K. Ikeda, *Brain Res.* **541**, 163 (1991).
12. T. Wisniewski and B. Frangione, *Neurosci. Lett.* **135**, 235 (1992).
13. W. A. Rocca *et al.*, *Ann. Neurol.* **19**, 415 (1986).
14. R. C. Peterson *et al.*, *Neurology* **44**, 867 (1994).
15. H. Potter, *Am. J. Hum. Genet.* **48**, 1192 (1991).
16. O. Bugiani *et al.*, *Neurosci. Lett.* **119**, 56 (1990).
17. J. T. Coyle, M. L. Oster-Granite, J. D. Gearhart, *Brain Res. Bull.* **16**, 773 (1986).
18. J. Hardy, A. Goate, M. Owen, N. Rossor, *Lancet* **ii**, 877 (1989).
19. M. I. Olson and C. M. Shaw, *Brain* **92**, 147 (1969).
20. H. M. Wisniewski, A. Rabe, K. E. Wisniewski, in *Molecular Neuropathology of Aging*, P. Davies and C. Finch, Eds. (Banbury Report, Cold Spring Harbor Laboratory, Cold Spring Harbor, New York, 1988), pp. 399–413.
21. H. Potter, L. N. Geller, D. Dressler, in *Genes in Medical Diagnosis and Therapy*, E. Kurstak, Ed. (Dekker, New York, in press).
22. B. Sacks and S. Smith, *J. Neurol. Neurosurg. Psychiatry* **52**, 1294 (1989).
23. W. S. Harris and R. M. Goodman, *N. Engl. J. Med.* **279**, 407 (1968).
24. J. M. Berg, M. W. G. Brandon, B. H. Kirman, *Lancet* **ii**, 441 (1959).
25. G. McKhann *et al.*, *Neurology* **34**, 939 (1984).
26. The 14 male and female patients with a diagnosis of probable Alzheimer's disease, mean age 74 ± 7 years, were drawn from the Massachusetts Alzheimer's Disease Research Center, Boston, MA. Patients classified as having probable Alzheimer's disease (AD) met strict National Institute of Neurological, Communicative Disorders and Stroke, and Alzheimer's Disease and Related Disorders Association (NINCDS-ADRDA) diagnostic criteria for probable Alzheimer's disease and performed significantly worse than the 32 cognitively normal elderly controls (NC) on the Information-Memory-Concentration subtest of the Blessed Dementia Rating Scale, a standard clinical measure of disease severity (AD = 17 ± 7, range = 4–27; NC = 0.7 ± 0.85, range = 0–3; $P < 0.01$). The scores on the Blessed Dementia Rating Scale range from 0 to 37 with 37 representing the most severe impairment. The NINCDS-ADRDA criteria for a diagnosis of probable Alzheimer's disease require that (i) dementia be established by clinical examination and be documented by a measure such as the Blessed Dementia Rating Scale, (ii) the patient exhibits deficits in two or more areas of cognition, (iii) there is a progressive worsening of memory and other cognitive functions, (iv) the patient has no disturbance of consciousness, (v) there is an onset between the ages of 40 and 90, and (vi) there is an absence of systemic or brain diseases that in and of themselves could account for progressive deficits in memory and cognition. Initially, 43 elderly individuals, either spouses of patients or healthy volunteers recruited through advertisement in the metropolitan Boston area, were enrolled in this study to participate as controls. Three individuals did not meet screening criteria because of significant ocular pathology and therefore were not studied. Of the remaining 40 individuals who participated in the study [mean age 72 ± 6 years.(no significant difference from the patient groups)], 32 were considered cognitively normal on the basis of neuropsychological screening. A pilot sample of four patients with a diagnosis of non-Alzheimer's type dementia were also included for study, mean age 66 ± 6 years. Two of these patients had a primary diagnosis of Korsakoff's syndrome, one was diagnosed as having multi-infarct dementia, and one a dementia with an extrapyramidal syndrome. All subjects completed an informed consent agreement. With the exception of the control patient with the extrapyramidal syndrome (parkinsonian-like), all subjects had unremarkable findings on a neuro-ophthalmological examination evaluating saccades, smooth pursuit, visual fields to confrontation, and partial field optokinetic nystagmus. No individuals were accepted into the study who had glaucoma or iridectomies or if they were found to have a narrow anterior chamber predisposing them to closed angle glaucoma in response to tropicamide. Three potential normal controls volunteering for this study were not tested because of iridectomies in one or both eyes. No potential subjects were rejected on the basis of a narrow anterior chamber. None of the normal controls had diseases of the central nervous system by history. Medication use in patients and all control subjects was comparable. No subject was taking medications with known interaction effects with tropicamide. No patients were taking any experimental acetylcholinesterase inhibitors (such as Cognex) that could have interfered with the assay. All subjects had a brief bedside neurological examination. Subjects were then screened with a neuropsychological battery of standard measures that assessed attention (Digit Span), memory (Wechsler Logical Memory Subtest and Visual Reproduction Subtest), naming (Boston Naming Test), and general intellectual ability (National Adult Reading Test) to determine if they were cognitively normal. The screening was carried out by an individual who was blind to any findings from the pupil assay. On the basis of their performance on the neuropsychological battery of tests, the subjects were assigned to one of three groups as described in the text.
27. L. F. M. Scinto *et al.*, data not shown.
28. Further research will be necessary to identify the precise site and mechanism of the defect in the pupil dilation response we have observed. The control of pupil diameter represents a balance between the cholinergic and adrenergic innervation of the iris muscle and is influenced directly and indirectly by central and autonomic nervous system inputs. Any of these systems could be affected in Alzheimer's disease and lead to a hypersensitivity to tropicamide. Possibilities include: (i) reduced in-

nervation of the target muscle through neuronal cell death, axon retraction, or reduced release, increased degradation, or increased re-uptake of neurotransmitter; (ii) altered amounts or function of a neurotransmitter receptor; or (iii) the impairment of central equilibrating mechanisms.
29. B. H. Price *et al.*, *Arch. Neurol.* **50**, 931 (1993).
30. The research reported in this article was made possible in part by grants from the Alzheimer's Association to L.F.M.S. and K.R.D., seed funds to H.P. and L.F.M.S., and the provision of laboratory space by the Beth Israel Hospital. H.P. and D.D. were supported in part by grants from NIH, the Alzheimer's Association, and the Freudenberger family. We thank Applied Science Laboratories for engineering support and for the loan of laboratory equipment. We are grateful to J. Guinessey for neuropsychological testing of subjects.

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Coding of Visual Space by Premotor Neurons

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In primates, the premotor cortex is involved in the sensory guidance of movement. Many neurons in ventral premotor cortex respond to visual stimuli in the space adjacent to the hand or arm. These visual receptive fields were found to move when the arm moved but not when the eye moved; that is, they are in arm-centered, not retinocentric, coordinates. Thus, they provide a representation of space near the body that may be useful for the visual control of reaching.

Premotor cortex is involved in the preparation and guidance of movement (*1*). In monkeys, many premotor neurons are active when the animal moves. In ventral premotor cortex, neurons also respond to visual stimuli and may play a role in the visual guidance of movement. Most of these visual neurons also respond to tactile stimuli; they have tactile receptive fields (RFs) on the face or arms, and corresponding visual RFs extend outward from the tactile fields into the space surrounding the body (Fig. 1) (*2*, *3*). The tactile RFs are somatotopically organized (*4*), and therefore the corresponding visual RFs provide a map of the visual space near the body (*5*). Although the visual RFs are large, each one giving only crude information about spatial location, a population of these cells could specify the location of targets for limb and body movements.

In most other regions of the brain, visual RFs are retinocentric. That is, when the eyes move, the visual RFs move with them, thereby remaining at the same retinal site. Such cells form a spatial coordinate system that can measure the position of a stimulus with respect to the eye. However, some investigators have suggested that a more stable coordinate system attached to the head or trunk might better serve visuospatial function (*6*). We studied the visual responses in ventral premotor cortex (ventral area 6) to determine how they encode the space near the body. Are the RFs of these cells retinocentric, or are they expressed in a coordinate system attached to the head, trunk, or some other part of the body? We concentrated on studying the bimodal cells with tactile RFs on the arm and tested the effect of varying the angle of gaze and the position of the arm on their visual responses. We found that most of these cells code space in arm-centered coordinates.

Single neuron responses in ventral premotor cortex (Fig. 1A) (*7*) were studied in two tame male *Macaca fascicularis* (6.0 and 7.0 kg). For one monkey, weekly recording sessions were conducted while the animal was anesthetized with nitrous oxide and oxygen and immobilized with pancuronium bromide. For the second monkey, daily recording sessions were conducted while the animal was unanesthetized and trained to fixate. The animal's head was fixed in place, and the arm contralateral to the recording electrode was restrained. Eye position was monitored with a scleral search coil (*8*).

We plotted somatosensory RFs by manipulating the joints and stroking the skin. Visual RFs were plotted with objects presented on a wand. To distinguish a visual response from a tactile response, we also tested the cells with the animal's eyes covered. Visual responses were tested quantitatively with stimuli presented by a motorized track.

In the anesthetized preparation, 141 neurons were studied, of which 42% (n = 59) were somatosensory, 1% (n = 2) were visual, 27% (n = 38) were bimodal visual-somatosensory, and 30% (n = 42) were unresponsive to our stimuli. In the awake preparation, 211 neurons were studied, of which 36% (n = 75) were somatosensory, motor, or both (9); 8% (n = 17) were visual; 31% (n = 65) were bimodal; and 25% (n = 54) were unresponsive. Of the visual and bimodal cells, only nine showed any response during overt movements of the animal.

A typical example of a bimodal cell studied in the anesthetized preparation is shown in Fig. 1B. When a visual stimulus was moved within 10 cm of the tactile RF on the face, the cell responded. By approaching the face from various angles, we measured the extent of the visual RF in three dimensions. Figure 1C shows another cell studied under the same condition. It had a tactile RF on the contralateral arm. When the arm was moved toward the ipsilateral side, the visual RF was dragged across the midline and into the ipsilateral field of view, even though the eyes remained fixed; that is, the visual RF was not retinocentric; rather, it was arm-centered.

In the awake preparation, we studied the effects of changing the position of both the animal's arm and gaze. Figure 2

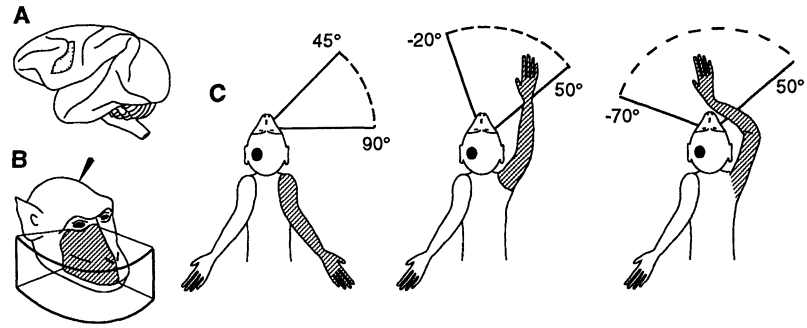

Fig. 1. (**A**) Ventral premotor cortex (shaded). (**B** and **C**) Two examples of RFs of bimodal, visual-tactile neurons studied in the anesthetized preparation. In (B), the tactile RF (stippled) and the visual RF (boxed) correspond in location. The arrowhead indicates the hemisphere recorded from. In (C), the lateral borders of the visual RF are shown by solid lines. As indicated by the dashed line, the RF extended more than 1 m from the animal. The black dot on the head indicates the hemisphere recorded from. When the arm was out of view (left), the visual RF extended from 90° to 45° contralateral. When the arm was moved forward (center), the visual RF moved to the front of the animal. When the arm was bent toward the ipsilateral side (right), the visual RF moved with it.

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