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Complex–unoriented cells in a subregion of primate area 18

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In primates, both the primary and secondary visual cortical areas can be subdivided histologically by staining for the mitochondrial enzyme cytochrome oxidase[1–3]. In the primary visual cortex (area 17, the first cortical receiving area for visual information) these histological differences correspond to functional subdivisions[1–6], cytochrome-dark regions being concerned with information about colour and cytochrome-light regions concerned with form. Here we report that the second visual area, area 18, which receives its main cortical input from area 17 (refs 7, 8), similarly has functional subdivisions that correspond to the cytochrome oxidase staining pattern. In area 18 the segregation between form and colour is maintained, reinforcing our notion that form and colour information follow parallel pathways. The specific differences between cells in areas 17 and 18 suggest that a possible step in hierarchical information processing is spatial generalization, analogous to the difference between simple and complex cells.

In area 17 the cytochrome oxidase staining pattern consists of small dark-staining polka-dots (blobs) on a lighter-staining background[1,2]. Cells in the blobs are not orientation-selective and at least half are colour-coded, whereas most cells in the inter-blob regions are orientation-selective with only a small number colour-coded[6]. In area 18 the dark-staining regions are much coarser and form stripes, ~0.5–1 mm wide, which run perpendicular to the 17/18 border and extend the full width of area 18 to its anterior border (Fig. 1; refs 3–5). In some macaques and in most squirrel monkeys, the dark stripes are generally of two types, alternating thin and thicker, slightly lighter stripes. Although the thick stripes are not always easy to distinguish from the thin stripes by cytochrome oxidase staining alone, they differ anatomically in that they have a different laminar distribution of thalamic inputs[4]. Thus, area 18 seems to consist of at least three types of subregions: thin stripes, thick stripes and, in between, much lighter interstripes.

We have recorded from 1,019 cells in area 18 of 11 young adult female *Macaque fascicularis* monkeys and 7 adult female *Saimiri sciurius* (squirrel) monkeys. The recording apparatus was as described previously[6]. We made electrode penetrations perpendicular to the cytochrome oxidase stripes as nearly parallel to the surface as possible.

It was clear that area 18, like area 17, is physiologically heterogeneous but with a much coarser periodicity. Figure 2 shows the results from one macaque monkey, in which we recorded from 9 mm of area 18; almost all of the penetration appears in this single section. The figure indicates that we passed through four cytochrome oxidase-dark regions. Two of these stripes (the second and fourth) consisted exclusively of cells that entirely lacked orientation selectivity, whereas almost all the cells in the other two stripes and in the interstripe regions were orientation-selective. We found such long stretches of

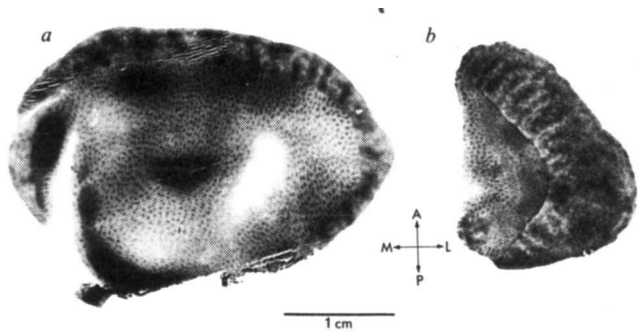

Fig. 1 Sections of monkey visual cortex stained for cytochrome oxidase. *a*, Section from a macaque monkey. Most of the section is in area 17 except for a 0.5-cm-wide strip of area 18 at the top. *b*, Section from a squirrel monkey. Area 17 is to the left with the fine polka-dot pattern and area 18 is to the right with a coarser-striped pattern. The alternation of thick and thin stripes is clearest near the top of the squirrel monkey section and is not apparent in the macaque.

unoriented cells in both macaques and squirrel monkeys (211 cells), always in cytochrome oxidase-dark regions. In the macaques it was not usually possible to say whether the dark stripes containing unoriented cells were thick or thin, but they usually alternated with stripes containing oriented cells, as in Fig. 2. In the squirrel monkeys the unoriented cells were clearly in the thin stripes. All these results are consistent with our earlier observation that the blobs in area 17 and thin stripes in area 18 are interconnected.

In macaques, about two-thirds of the unoriented cells showed colour opponency; in the squirrel monkey the proportion of colour-coded cells was much smaller. Of the colour-coded cells, a few had colour opponent centres and no antagonistic surrounds (for example, red-on centre, green-off centre, Type II; ref. 6), but most had some form of double-opponent colour coding (for example, red-on centre, red-off surround, green-off centre, green-on surround). Similar to blob cells in area 17, the unoriented cells in area 18 (whether or not they are colour-coded) responded more strongly as spot size was increased up to an optimum size beyond which the responses declined. What was different about many of these cells, compared with the blob cells in area 17, is that they responded to a spot of optimum size regardless of where it was positioned, over an area that was itself many times larger than the optimum spot. Thus, in these cells a spot large enough to cover the entire activating region was usually ineffective.

An example of one such colour-coded cell is shown in Fig. 3. It gave on responses to small red spots, off responses to small blue or green spots and no responses to white or to yellow spots (580 nm). In response to an optimal spot, 0.5° in diameter, the cell gives on responses to red and off to blue or green throughout an activating region 3.5° in diameter, maximum near the centre and falling off gradually with distance; increasing the spot beyond 0.5° resulted in a fall-off in responses (Fig. 3*b*, *c*). When we shone a small red and a small blue spot simultaneously in the same position (giving a yellow spot), the cell gave little or no response. We did not try spatially to separate red and blue spots with this cell (but in other similar cells, simulating with a small red spot and a small blue spot side-by-side within the activating region gave mixed on/off responses). This cell was located in layer 5 of a cytochrome oxidase-dark stripe in area 18 of a macaque.

We also observed cells, similar but not colour-coded, some giving on and others off responses regardless of wavelength over an activating region larger than the optimum spot size. Baizer *et al.*[9] have described 'spot cells' in area 18 that are similarly unoriented, not colour-coded and respond best to a small spot anywhere in a large receptive field. We term all of these cells, whether or not they are colour coded, 'complex-unoriented', because they differ from ordinary centre-surround cells in a manner analogous to the difference between oriented simple and complex cells[10]. (The distinction lies in the permissible

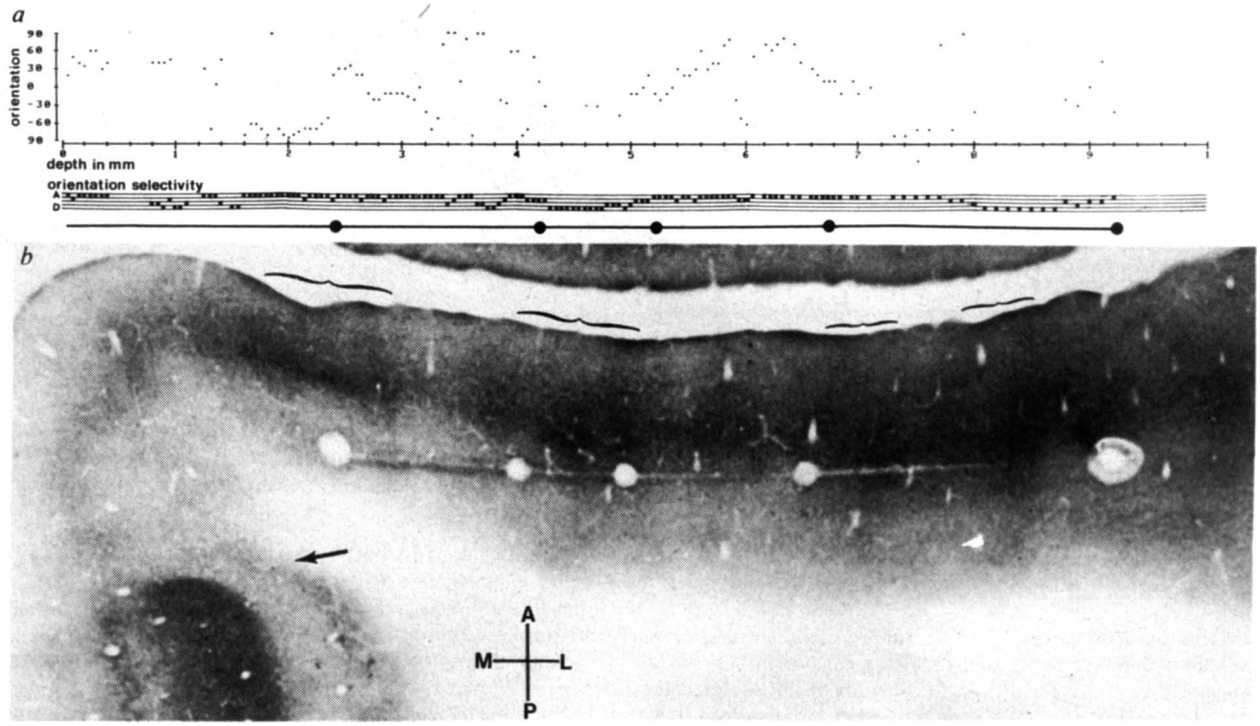

Fig. 2 *a*, *b*, Reconstruction of an electrode track in area 18 of macaque. *b*, Transverse section through area 18 just anterior and parallel to the area 17/18 border. Part of the border is visible at the left edge of the section (arrow). The space shown just above area 18 is the lunate sulcus; the midline is on the left. The electrode track, most of which can be seen together with five electrolyte lesions, runs from left to right. The section was stained for cytochrome oxidase[22] and the cytochrome-dark regions are indicated by brackets. *a*, From above downwards, preferred orientation (0°, vertical); orientation selectivity (A, well oriented, D, unoriented); and the location of the electrolyte lesions. Optimum orientation and orientation selectivity were judged subjectively by listening to the response of the cell to slits of light generated by a hand-held slide projector. Observations were made every 0·05 mm. Note the correspondence between two of the cytochrome-dark regions (between second and third lesions and just before the final lesion) and the absence of orientation selectivity. (At these points no orientation could be plotted.) The entire track after the first 0.5 mm was below layer 4.

variation in position; see ref. 10.) In both cases, although the size of the region of the visual field in which the higher level cell can be activated is larger (in our complex-unoriented cells, from two to several times the diameter), the optimum stimulus size for a given eccentricity remains the same, as if an 'or-gate' operation had been performed on many cells with similar receptive fields scattered over an area larger than each separate receptive field. Thus, although resulting in a loss of precision of localization, complexification need not lead to a loss of resolution or acuity, because at a given eccentricity the average optimum stimulus size, such as line width or spot diameter, shows no such increase. The increase in receptive-field size reported in several pre-striate areas[11–14] is presumably, at least in part, the result of such transformations. The occurrence of similar operations in different systems suggests that similar circuits are used, which would doubtless simplify the problem of building the cortex. Despite the frequency with which this operation seems to occur in the visual system, its role in information processing is still a matter of conjecture. (See page 285 of this issue.)

Some of the unoriented cells we found in area 18 were not complex and seemed the same as cells that we have observed in area 17. We are now trying to determine whether the different orders of unoriented cells in area 18 are segregated into different layers, as any sequence of unoriented cells tended to contain either ordinary or complex unoriented cells. In area 17, however, we have not seen complex-unoriented cells. The transformation of colour information in going from area 17 to 18 might be predicted to involve the acquisition of orientation selectivity, but this seems not to be so.

In the squirrel monkey it is clear that the regions in area 18 containing complex-unoriented cells are the same regions, the thin stripes that we previously found anatomically to be connected to the blobs in area 17. These blobs contain unoriented, often colour-coded cells[6]. In macaque monkeys the histological distinction between two types of cytochrome-dark stripes is often less clear, but, as shown in Fig. 2, only some of the stripes (usually alternate ones) are also characterized by colour-coded unoriented cells.

We have also studied the characteristics of the cells in the other stripes: cells in both the interstripes and the thick stripes show orientation selectivity and are not colour-coded. The light-staining interstripes in area 18 are reciprocally connected with the inter-blob regions of area 17, which contain oriented non-colour-coded cells[6]; the connections of the thick stripes are not yet known, but they clearly receive input from area 17 (see Fig. 30 of ref. 6). The most striking characteristic of cells in the interstripe regions is that over half of them are end-stopped[11], responding optimally to slits of a particular length and less well to longer slits. This suggests that the system that in area 17 carries information about the orientation of edges might next acquire information about their degree of curvature. In the thick stripes the cells seem to be concerned primarily with binocular disparity: many fail to respond to either eye alone but do respond if the eyes are aligned with a particular disparity, corresponding either to near, far or peaking at zero disparity. DeYoe and Van Essen[15], by injecting two different retrograde tracers into the pre-striate visual areas MT and V4 of macaques, have found that in area 18 alternating cytochrome-dark stripes project to MT and to V4. The results of Shipp and Zeki[16] in the accompanying paper confirm these observations. Given the high proportion of colour-coded cells reported in V4 (ref. 14), the V4-projecting stripes would be expected to be the ones to receive input from the blobs in area 17 and contain unoriented, often colour-coded cells: in a squirrel monkey these would be the thin stripes. Our finding that the thick stripes contain orientation-selective cells is consistent with their involvement with some system other than the colour system; they could correspond to the MT-projecting stripes described by DeYoe and Van Essen[15]. The segregation in area 17 of two systems, one apparently concerned with form

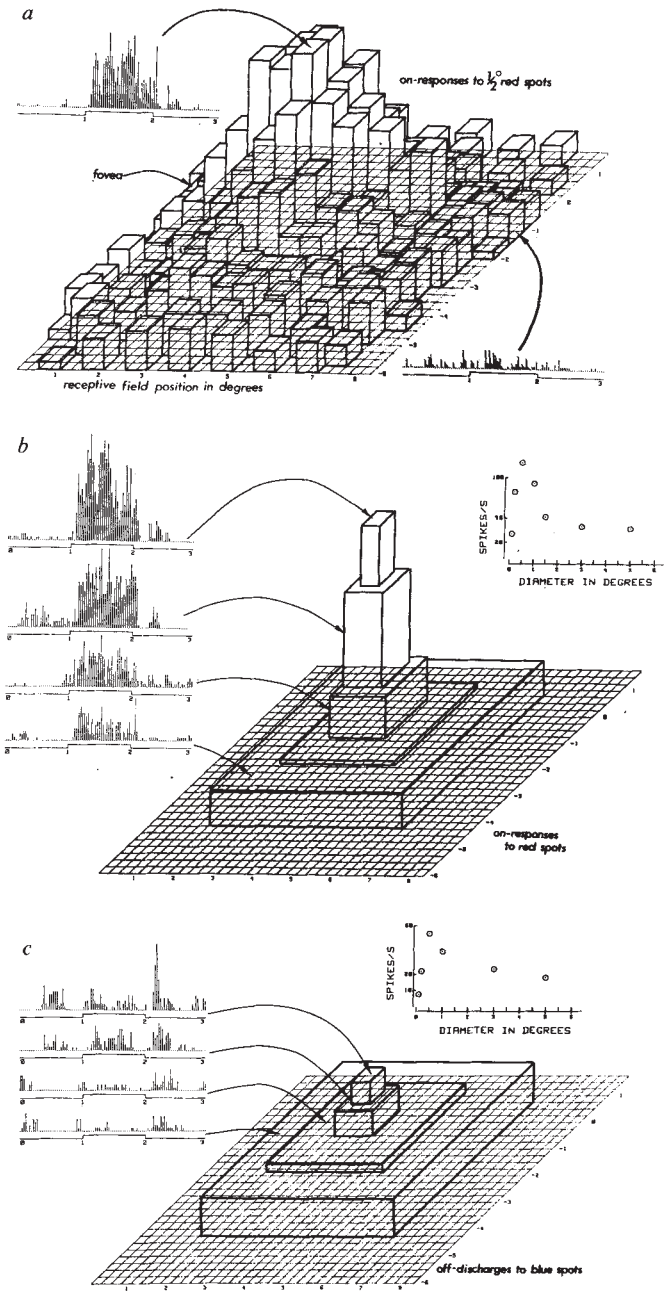

Fig. 3 Responses of a complex double-opponent cell in area 18 of a macaque monkey to red and blue spots of light, varying size and position of spot. The grid represents the visual field, marked off in degrees, with the fovea at (0, 0). The red and blue spots were produced by Kodak Wratten filters (29 and 47B) placed in front of a slide projector. *a*, Three-dimensional plot illustrating responses to red spots of optimal size, 0.5° in diameter. The plot is made over a region 8×8°, centred 5° lateral and 2.5° below the fovea. Responses were obtained over an area occupying 3×3°, outside which the bars represent spontaneous background activity. Bars represent means of five responses to stimuli of 1 s duration. The histogram for the area giving the peak response, shown in the inset on the upper left, represents 85 impulses during this 'on' period. The lower right inset shows one of the histograms for a region giving little or no response above the background firing rate. *b*, Diagram of on-responses to red light spots of various sizes. Each histogram is a mean of five stimuli. The three-dimensional graph illustrates the responses to the optimum size and to larger stimuli; the inset on the left shows the histograms obtained for four of these five spots. The upper right inset shows the entire curve of response versus spot size. Note that the 3.5 and 5° stimuli evoked responses that were barely visible above background firing levels, even though these covered the area over which 0.5° spots gave vigorous responses. *c*, Same cell, off-discharges to blue spots of different sizes.

and the other with colour, evidently persists into area 18 and beyond. That a similar segregation occurs in humans is apparent from reports of subjects with local cortical damage who experience loss of colour vision but retain normal form, movement and depth perception[17–21].

We thank Debra Broihier and Janet Robbins for histology and Marc Peloquin for photography. This work was supported by NIH grant EY 00605-25 and grants from E. I. du Pont de Nemours and Co., the Monsanto Co. and NSF.

Received 12 December 1984; accepted 25 March 1985.

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Induction of regulatory T-lymphocyte responses by liposomes carrying major histocompatibility complex molecules and foreign antigen

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Regulatory (helper and suppressor) T lymphocytes become activated only when foreign antigen is presented to them on the surface of antigen-presenting cells (APC), together with class II major histocompatibility complex (MHC) molecules (heterodimers of polypeptides of 28,000 and 35,000 relative molecular mass)[1–4]. Once activated by a certain foreign antigen—MHC combination, T cells react to the same antigen only in combination with the same MHC molecule, a phenomenon termed MHC restriction of T-cell recognition (reviewed in refs 1, 5). Studies of the mechanisms involved in antigen presentation and MHC restriction have been hampered mainly by the virtual impossibility of inducing T-cell responses in the absence of APC. We describe here the production of synthetic lipid vesicles with inserted class II MHC molecules and a protein antigen coupled covalently to the lipid. These liposomes are shown to stimulate cloned helper T cells and T-cell hybridomas in an antigen-specific, MHC-restricted manner in the absence of APC. Thus, the recognition of foreign antigen together with class II MHC molecules seems to be the only signal required for the activation of antigen-primed regulatory T cells. Furthermore, 'processing' of antigen by APC is not essential for its recognition by T cells.

To determine the minimal requirements of T-cell activation, we constructed liposomes carrying a foreign protein antigen, or class II MHC molecules, or both, and tested whether these liposomes could activate antigen-specific class II-restricted T cells in the absence of APC. Class II MHC molecules were isolated from lipopolysaccharide-stimulated spleen cells. The membrane proteins were solubilized with 50 mM β-octyl gly-