

CONTINUITY OF ORIENTATION COLUMNS BETWEEN SUPERFICIAL AND DEEP LAMINAE OF THE CAT PRIMARY VISUAL CORTEX

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SUMMARY

1. Recent reports of a marked and consistent dislocation between orientation columns in the superficial and deep layers of cat striate cortex (Bauer, 1982, 1983) directly contradict the traditional view of the system (Hubel & Wiesel, 1962). This has considerable implications for our current understanding of cortical organization, and in order to clarify the issue we have carried out experiments to test the continuity of the columnar system with depth, in central regions of area 17.

2. In twenty-four penetrations, eighteen of which were placed as perpendicular as possible to the surface of the cortex, orientation preference was assessed at regular intervals both qualitatively and using a randomly interleaved quantitative technique. The distribution of preferred orientations was analysed with reference to a detailed histological reconstruction of the electrode track, including the location of laminar boundaries and the course of radial tiers of cells and capillaries.

3. From a further series of eighteen near-perpendicular penetrations, the change in average orientation between one superficial and one deep layer recording site was compared with the deviation of the track from perpendicular to the surface and hence parallel to the orientation columns.

4. In penetrations perpendicular to the surface of the cortex, orientation preference showed little variation between superficial and deep laminae. In oblique penetrations, preferred orientation generally changed according to a single, smooth trend. Those irregularities that were encountered were confined to oblique penetrations, and were distributed throughout the cortical laminae.

5. In conclusion, our evidence does not support the presence of a systematic discontinuity with depth within the orientation columnar system. It is therefore entirely consistent with earlier evidence on the subject.

INTRODUCTION

Neurones in the visual cortex show a high degree of selectivity for a number of stimulus parameters, one particularly notable example being the angle or orientation of a contour passing over the receptive field (Hubel & Wiesel, 1959, 1962, 1968; Henry, Dreher & Bishop, 1974; Gilbert, 1977; Hammond & Andrews, 1978). Not only are the majority of cells clearly tuned to this aspect of their visual input, but the

distribution of preferred orientations is non-random (Hubel & Wiesel, 1962, 1963, 1965, 1968, 1974; Albus, 1975). Neurones sharing the same or similar specificity are aggregated into a system of 'columns' lying perpendicular to the surface of the cortex, which are themselves arranged in an orderly progression throughout the tangential extent of the area. It is hardly surprising that the orientation column has been accepted as one of the characteristic features of the functional organization of the visual cortex, nor that it has provided a basis for speculation about the generation of orientation selectivity and other aspects of receptive field structure. It is therefore of some importance that recent work has called into question a fundamental characteristic of the columnar concept.

Following their original work on the subject, Hubel & Wiesel (1962, 1963, 1965, 1968, 1974) stressed that electrodes penetrating perpendicular to the surface of the cortex encountered cells of similar specificity throughout the oriented layers. This led to the view that orientation columns, in common with ocular dominance columns (Hubel & Wiesel, 1963, 1968), extend throughout the depth of the cortex from layer II to layer VI. Their interpretation was apparently substantiated by data obtained utilizing [¹⁴C]-2-deoxyglucose to visualize the patterns of high metabolic activity generated in the visual cortex, during the presentation of a stimulus composed of a single orientation (Hubel, Wiesel & Stryker, 1978; Albus, 1979; Lang & Henn, 1980; Schoppmann & Stryker, 1981; Singer, 1981; Albus & Sieber, 1984). Furthermore, although electrophysiological studies by other authors have revealed a degree of random variation in the distribution of orientation selectivity at the columnar level (Albus, 1975; Lee, Albus, Heggelund, Hulme & Creutzfeld, 1977), they did not until recently challenge the conclusion that cells with a common orientation bias are broadly aggregated into units extending throughout the cortical laminae.

However, these findings have now been questioned by reports that there is a marked difference in orientation preference above and below the non-oriented layer IVc in the monkey cortex, which cannot be accounted for by lateral movement of the electrode (Bauer, Dow & Vautin, 1980; Kruger & Bach, 1982; Bauer, Dow, Snyder & Vautin, 1983; Dow & Bauer, 1984). Similarly in the cat area 17, it is claimed that a clear discontinuity in the orientation sequence is almost invariably found at the junction between layers IV and Va (Bauer, 1982, 1983). The magnitude of this shift in the cat, although variable, is said to be frequently as high as 90 deg. The authors argue for a marked and consistent dislocation between the columnar organisation of the superficial and deep laminae, such that overlying regions have orthogonal rather than corresponding orientation preference (see Fig. 1). This of course implies an important and fundamental error in the observations made both by Hubel & Wiesel, and by subsequent investigators in the field (Albus, 1975; Lee *et al.* 1977). However, it is fair to say that none of these studies demonstrated truly unequivocal evidence for the continuity of the columns between superficial and deep layers; the arrangement was either inferred or lacked good histological control.

The work of Bauer and colleagues has thus highlighted the need for a more detailed and quantitatively confirmed study of the pattern of orientation preference in area 17, with an emphasis on its distribution as a function of cortical depth. The experiments reported here attempt to fulfil this need. A specific investigation has been made of the vertical continuity of orientation columns, giving particular

attention to histological verification of electrode angle, and the location of any large shifts in orientation preference. The results confirm that overlying regions of the cat primary visual cortex share a common specificity, and that a marked and consistent dislocation between the superficial and deep laminae is difficult to support. A preliminary report of this work has been made (Murphy & Sillito, 1984).

METHODS

A total of twenty-one cats were used in this series of experiments. They were anaesthetized with 70% N₂O and 30% O₂, supplemented with 0.1–0.4% halothane, paralysed with gallamine triethiodide (10 mg/kg. h), and artificially ventilated at a rate and depth that maintained end-tidal CO₂ levels at 3.8–4.2%. All wound margins were treated with subcutaneous injections of procaine hydrochloride, and the ear bars of the stereotaxic apparatus were coated with antiseptic lignocaine hydrochloride gel. Electroencephalogram and heart rate were monitored continually in order to confirm the state of anaesthesia. A bilateral cervical sympathectomy was performed. The eyes were treated with atropine methonitrate and phenylephrine hydrochloride, protected with plastic contact lenses, and brought to focus upon a semi-opaque tangent screen at a distance of 1.14 m. Further details concerning the preparation, care, and maintenance of the animals have been given elsewhere (Sillito & Kemp, 1983).

928 recordings, 755 of which were from fully isolated and in most cases fully classified single units, were made in area 17 of the cortex, using glass micropipettes filled with 2% (w/v) Pontamine Sky Blue in 0.5 M-sodium acetate. Recordings were made on the tip of the lateral gyrus between 2 and 4 mm posterior to stereotaxic zero. All were within 10 deg, and the majority within 5 deg, of the area centralis. Receptive field properties were assessed initially using hand-manipulated stimuli projected from the rear of the tangent screen. Orientation preference and tuning were then confirmed quantitatively in a representative sample of cases. Care was taken to use the longest bar to which a given cell would respond reliably, as orientation selectivity may decrease with bar length (Henry *et al.* 1974). In the earliest experiments, an automatically controlled optical projection system and custom-built averager were used to generate peristimulus time histograms for each of the tested orientations individually (see Sillito, 1976, 1979). However, the bulk of the quantitative data were obtained using a computer system based upon the Computer Automation ALPHA LSI-2 and a modified C.E.D. 502 interface, driving a Joyce Electronics Ltd. C.R.T. display. This had the great advantage of producing orientation multihistograms with random interleaving of all stimulus presentations. These data are presented here primarily in the form of polar diagrams showing the relative magnitude of the response to each orientation tested, usually the supposed optimum and 15, 30, and 60 deg on either side, averaged over five or more trial repeats (see Fig. 2 for further details). The system is believed to give a very accurate representation of the orientation tuning of the cells. Comparison of these data with the qualitatively determined optima showed that the initial assessment was rarely more than 10 deg in error, and usually better. Hence the purely qualitative data are also believed to be acceptably accurate.

A total of forty-two separate penetrations were made, for which two basic strategies were adopted.

'Group 1' (twenty-four penetrations)

In eighteen cases the electrode was placed at a location and angle judged to be perpendicular to the cortical surface, in order to record from neurones located in or near a single cortical 'column'. Due to the highly curved nature of the cat cerebral cortex, however, very small positional errors led to substantial deviations from the intended course. Consequently, only seven penetrations were virtually perpendicular, while eleven were oblique in a principally medial to lateral direction. A further six penetrations were deliberately oblique, but with a caudal to rostral slope. In either case, neurones were sampled from throughout the track in order to assess the trends in orientation change seen with depth and location. A minimum of eight and a maximum of thirty-one recordings were made per penetration, the most common number being thirteen.

'Group 2' (eighteen penetrations)

The electrodes were again placed as close to the perpendicular as possible, and only those which were approximately so were included in the analysis. In contrast to the previous experiments, however, closely spaced groups of three to five quantitatively assessed cells were recorded from each of two locations; one above and one below the layer IV/V border region. This allowed a rapid comparison of the difference between superficial and deep layer orientation preferences, and the horizontal distance between recording sites.

At the termination of each penetration, the course of the electrode and at least three key locations were marked with Pontamine Sky Blue. Discrete spots were made by passing 4–8 μ A (negative) current through the recording barrel for 6–8 min, while the same current passed during withdrawal of the electrode at a rate of 2–4 μ m/s left a fine trace along the entire length of the track. The accuracy of this method is of course limited by the degree to which the electrode shunts the tissue as it moves. This did not in fact appear to be a problem with the fine electrodes used in this study. Nevertheless, the marked locations were always verified during the analysis of the histology, both by checking the distances of the blue spots from one another and from the surface, and by comparison with the distinctive changes in receptive field type encountered over the course of the penetration (Gilbert, 1977). Where a major discrepancy was detected the data were discarded.

The brain was removed from the animal prior to fixation, and blocks containing the primary visual cortex were immersed in 10% (w/v) formol saline (frequently renewed) for a period of two weeks. The fixed tissue was impregnated with a solution of 1% gum arabic and 30% sucrose, frozen sectioned to a thickness of 50 μ m, mounted and stained with neutral red. In all cases except those few in which the electrode was inserted with a caudal to rostral slope, the blocks were cut in the coronal plane perpendicular to the cortical surface. The remainder were sectioned sagittally. Relevant sections were drawn using a Wild M8 zoom microscope with drawing attachment. Each electrode track was then reconstructed from serial sections, in order to determine the angle of penetration and the location of the major laminar boundaries (after O'Leary, 1941). Hence changes in orientation preference could be related with certainty to the cytoarchitecture of the cortex. This information was considered essential to the final analysis of the data; so much so that any penetration for which a full histological record was not available was also discarded.

For blocks that were sectioned coronally, the deviation of the electrode from perpendicular was assessed in the plane of section by comparison with the courses of the radial tiers of cells, and in the orthogonal direction by comparing the course of the electrode from section to section with those of nearby radial blood vessels. Since the number of sections involved was usually very small, an accurate three-dimensional reconstruction was easily obtained in this way. The position of each neurone was then projected along a radial line to the surface of the cortex, and the total deviation of the track in both coronal and sagittal planes translated into distance moved with respect to the surface. The intention was to provide a form of measurement that would not be affected by the compression of the tissues in the deeper layers, and which would therefore reflect, in a simple linear fashion, the accelerating rate at which an oblique penetration cuts through the columnar system in those layers. This method gave consistent results and made the detection of unusual shifts in orientation preference considerably easier.

RESULTS

The controversy central to this study is summarized in Fig. 1. This shows a schematic representation of a section through area 17, including the outline, the main laminar boundaries, and the course of a few of the radial tiers of cells. A pair of hypothetical electrode tracks are also shown, one perpendicular to the cortical surface and one slightly oblique. At regular intervals throughout the course of each track are marked the sites of recorded cells, for which two sets of orientation preferences are shown in the form of bars drawn at the corresponding angle. Those relating to the perpendicular penetration are shown to the left of the Figure, and

those relating to the oblique penetration to the right. The first column in either set represents the type of data that might be expected if orientation columns are continuous throughout the cortical laminae. Thus in the vertical penetration the orientation preferences recorded are very similar throughout, while in the oblique penetration they change according to a single over-all trend. The second column

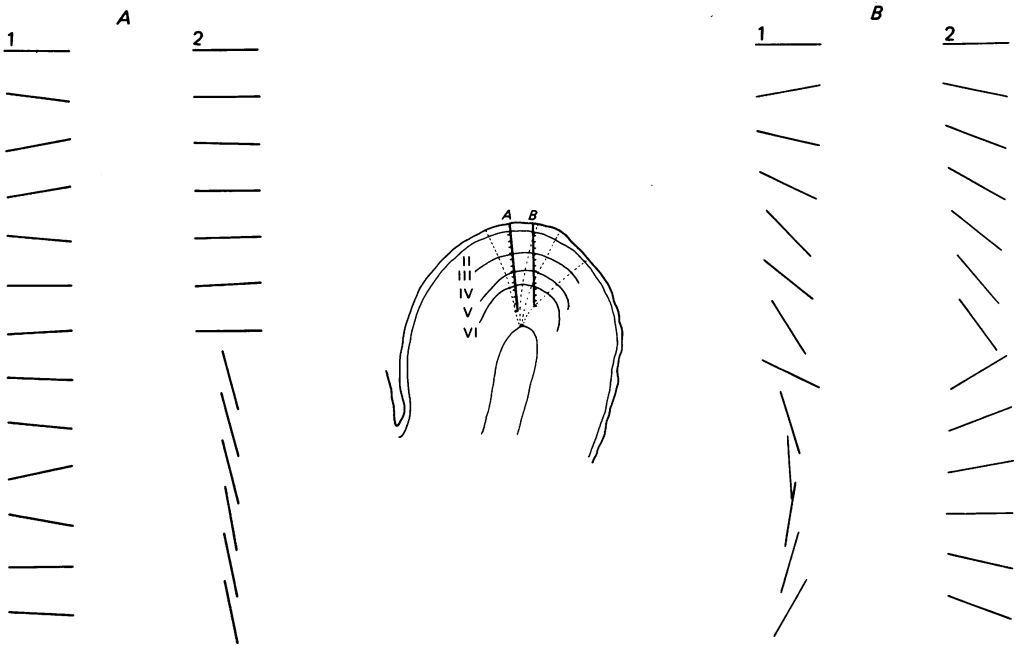


Fig. 1. Hypothetical results of two penetrations passing through layers II-VI of the feline visual cortex. The central Figure shows a schematic reconstruction of one perpendicular (A) and one oblique (B) penetration with respect to a coronal section through area 17, including the major laminar boundaries and a few of the radial tiers of cells and capillaries (dotted lines). For each track two alternative sets of orientation preferences are shown, illustrating the trends described by (1) Hubel & Wiesel (1962, 1974) modified according to Albus (1975), and (2) Bauer (1982, 1983).

illustrates the type of data reported by Bauer (1982, 1983). They differ from those described above in that there is a marked dislocation in the region of the IV/V border, such that the orientations recorded in the deeper layers are approximately orthogonal to those which might otherwise have been expected. The 'group 1' experiments were designed with the intention of locating such discontinuities, if present, and confirming their laminar distribution.

General findings

Of the total of twenty-four 'group 1' penetrations, for which neurones were recorded at regular intervals along the track, seven were found to run parallel, or very nearly so, to the radial tiers of cells and capillaries in both coronal and sagittal planes. They were therefore believed to be virtually confined within a single, anatomically defined cortical column. In these penetrations, orientation preference showed no

substantive change between superficial and deep layers, although there was some minor variation in bias seen from cell to cell.

In those penetrations that were to some extent oblique, orientation preference was most commonly found to change from cell to cell according to a single, over-all trend. The rate of change tended to fall within or below the ranges reported by Hubel &

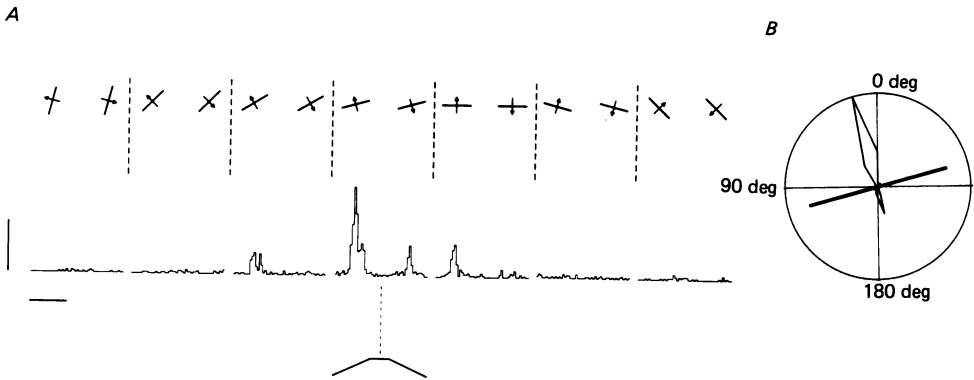


Fig. 2. Quantitative assessment of orientation tuning for a single cell. *A*, peristimulus time histograms showing the response of the cell to five randomized stimulus presentations at each of seven trial orientations; vertical ± 15 , 30, and 60 deg. Each includes both forward and backward motion of the stimulus, as indicated by the ramp symbol beneath the central Figure and the orientation and direction symbols above. Calibration vertical 100 spikes/s, horizontal 2 s. *B*, normalized orientation tuning curve for the same cell, in the form of a polar diagram. The axes correspond to the orientation of the stimulus as seen from the cat's eye view. The magnitude of the response to each test orientation is marked at the orthogonal to this angle, in the direction corresponding to the motion of the stimulus. The heavy bar shows the optimum orientation chosen on the basis of these data.

Wiesel (1962, 1963, 1974) and Albus (1975), with a mean value of 108 deg/mm of tangential distance moved with respect to the surface (range 40–200 deg/mm, s.e. of the mean ± 9 deg). This difference we believe stems from a combination of the predominantly coronal course, both of the majority of the penetrations and of the columns in this area of cortex (Singer, 1981; Albus & Sieber, 1984), and the method used to estimate the tangential movement of the electrode in the coronal plane (see Methods). Indeed, the drift rate appeared to correlate to some extent with the direction of the deviation of the electrode from the perpendicular. Where the slope was predominantly in the caudal to rostral direction, the average drift rate was 120 deg/mm of tangential distance, whereas in those cases where the error was exclusively in the coronal plane this value was 95 deg/mm. The difference was not, however, statistically significant. Regardless of the explanation the results were consistent within themselves, and the difference was always taken into account in the analysis of irregularities in the expected trends.

In common with the findings of Albus (1975) and Lee *et al.* (1977), a significant random element was seen in the precise distribution of optimum orientations. This scatter was variable, but fell within the range of ± 5 –15 deg of a best-fit line for the

data for all but the few cells discussed below. These exceptions were neurones for which the optimum orientation deviated by more than twice the normal range of variation (that is, by more than 30 deg) and up to 90 deg from an otherwise orderly trend. Twenty such cells were recorded, two in the superficial layers (which differed by 45 and 60 deg from that expected), twelve in layer IV (mean value 55 ± 5 deg from

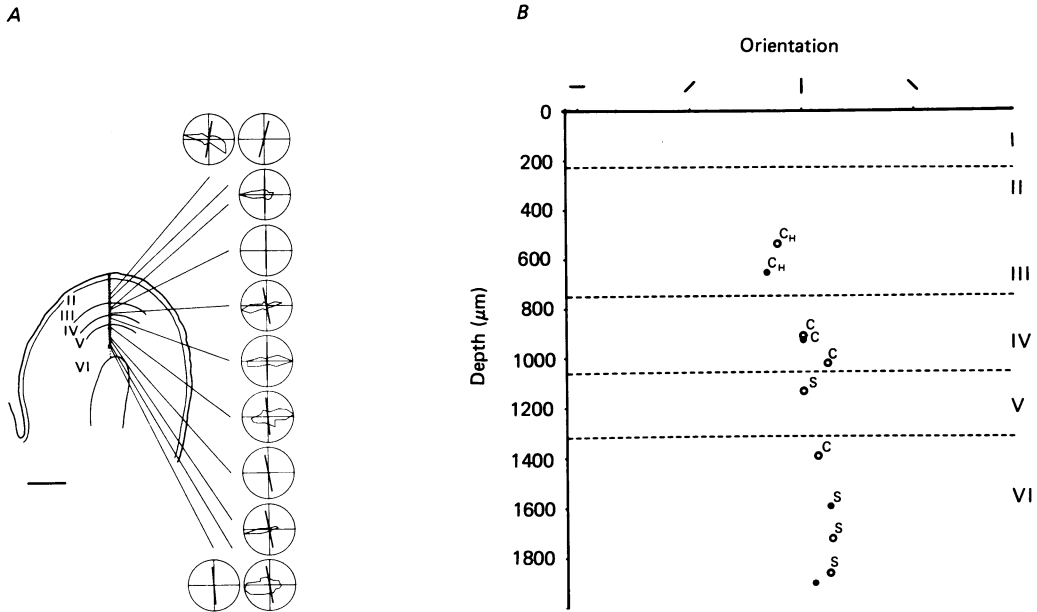


Fig. 3. Results of a 'group 1' perpendicular penetration. *A* illustrates the course of the electrode track, in the same form as for Fig. 2. Scale bar 1 mm. To the right are shown the orientation preferences of eleven cells recorded at the locations indicated. Quantitative data are represented by polar diagrams of the entire tuning curve; qualitative data by a bar drawn across a set of polar axes at the optimum orientation. *B*, a graph of orientation preference with respect to the depth of the recording below the surface of the cortex, and to the locations of the major laminar boundaries. Open circles indicate quantitative and filled circles qualitative data. Cells classified where possible as complex (C) or simple (S), with (H) or without end-stopping. CS denotes special complex in following Figures.

that expected), and six in the deeper layers (mean value 50 ± 4 deg from that expected). Hence, although they were encountered in all laminae, the majority were seen in layers IV–VI. This is reflected in the difference in average scatter for recordings in layers II/III (9 ± 1 deg to either side of the best-fit curve), IV (18 ± 3 deg), and V/VI (11 ± 1 deg). One important point to note is that in three penetrations a cluster of two or three such cells in layers IV and V gave the appearance of a 'double shift' (Bauer, 1983) in the middle layers.

Apart from these instances, step-wise shifts in orientation preference were rare. Their frequency and distribution, along with those of other encountered irregularities, will be analysed in detail in the following sections, with particular reference to the possibility of a dislocation in the columnar system between the superficial and deep layers.

Continuity at the IV/V border in 'group 1' experiments

The first and most important point is that such a discontinuity was never encountered in a penetration that ran truly perpendicular to the cortical laminae, and in which all the recordings were therefore made from within a narrow column of tissue. For example, Fig. 3 shows the data from a penetration that was parallel

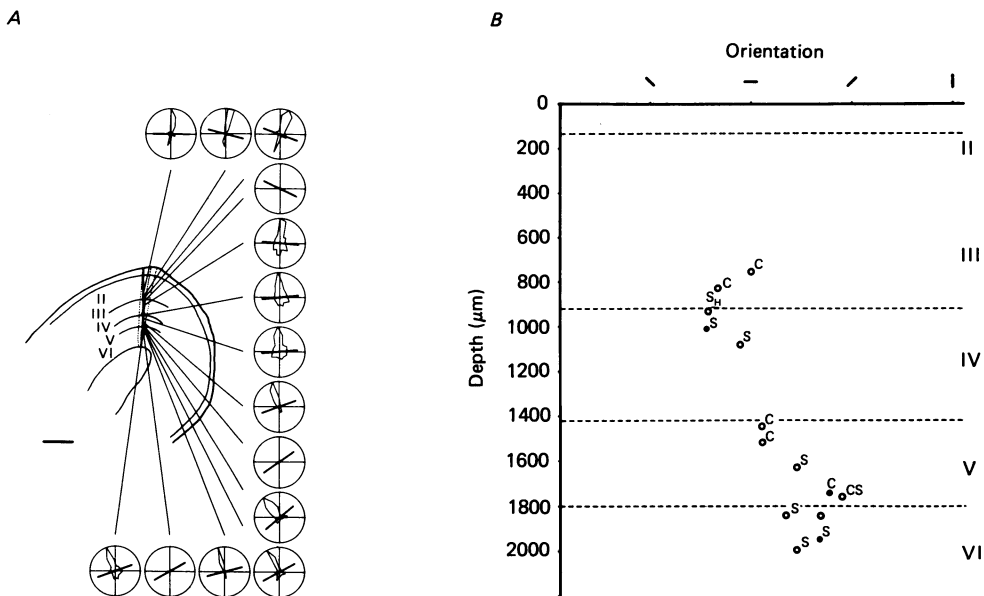


Fig. 4. Results of a near perpendicular penetration from 'group 1'. Format as for Fig. 3.

to the radial tiers of cells and capillaries, and was hence indistinguishable from perpendicular according to the present histological techniques. Fig. 3A includes the histological reconstruction of the track, and the orientation preferences of a number of cells recorded at specified points. These are given in the form of polar diagrams, showing the entire tuning curve in the case of quantitative data (see Fig. 2), but the optimum orientation only in the case of qualitative data. Fig. 3B shows these same results converted into the form of a graph, of preferred orientation *versus* distance from the surface. It is immediately clear that, although the optimum orientations varied over a range of 25 deg in total, there is little over-all change along the length of the penetration. There is no doubt that all the neurones in this vertical cylinder of tissue shared a closely related specificity, and certainly no evidence of a major shift between the superficial and deep layers. Such results argue very strongly in favour of continuity within individual columns.

Moreover, in the majority of the remaining penetrations the drift in orientation remained constant throughout the cortical layers, with no evidence of a sustained shift in preference that was greater than the inherent variability within the track (this was defined as greater than 30 deg in regions of average base-line scatter). For

example Figs. 4 and 5 show the results from two penetrations that ran at an angle to the perpendicular. In the first case the deviation was slight, and the drift in orientation preference away from the initially recorded values was therefore very small. It is quite clear that no major dislocation in the orientation system was encountered in the course of this penetration. Fig. 5 in contrast, represents the data

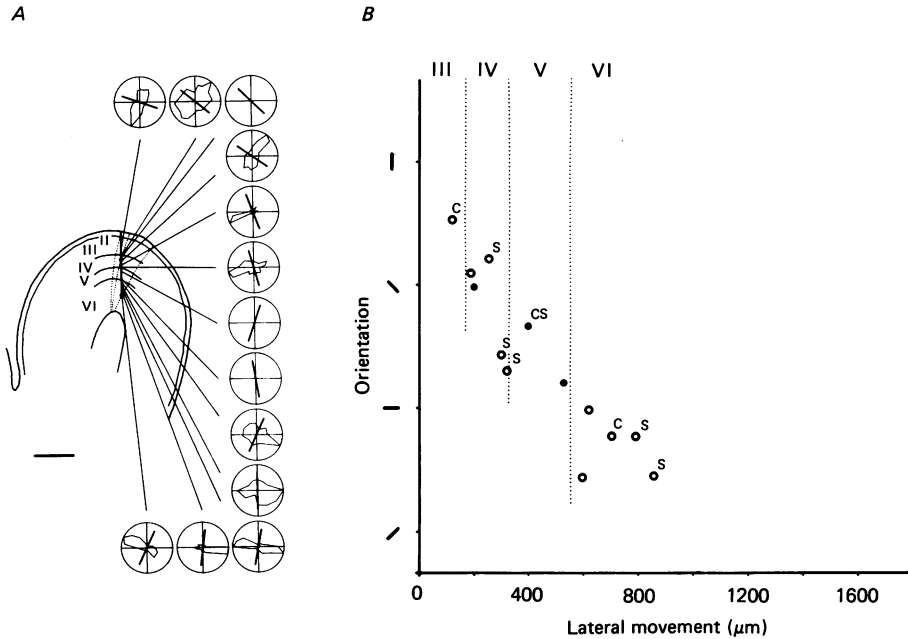


Fig. 5. Results of a markedly oblique penetration from 'group 1'. Format of *A* as for Fig. 3*A*. *B*, a graph of orientation preference with respect to the tangential distance of the recording sites from the entry point of the penetration, measured along the cortical surface. Also shown are the locations at which the electrode crossed the major laminar boundaries. All else as for Fig. 3*B*.

from a track that deviated significantly in the coronal plane. For this reason the graph (Fig. 5*B*) is presented as orientation *versus* tangential movement of the electrode, measured as distance along the surface of the cortex from the entry point of the penetration to the point radially above each recording site. As may be seen, the orientation preferences recorded in layer VI are approximately orthogonal to those initially encountered in the superficial layers. However, there is a lateral distance of 700 μm between these sites, and the total change is therefore well within the range expected from this and previous work (Hubel & Wiesel, 1962, 1974; Albus, 1975). Furthermore, although the individual values show a significant degree of scatter, the over-all trend is quite smooth and again shows no sign of a large discontinuity at the border between layers IV and V, or indeed in any other lamina.

Those irregularities that were observed in our data fell into three basic types. In three penetrations, the last few cells had orientation preferences that shifted progressively away from the previously recorded trend. In each case, however, these

cells were in the deeper parts of layer VI, where the electrode had moved at a marked angle through the radial tiers of cells and capillaries. Given the compression of the columnar system at this depth, an accurate estimate of the expected drift rate is impossible. These data cannot therefore be taken as evidence of a similar shift in the orientation columns themselves. On the other hand, three penetrations showed the

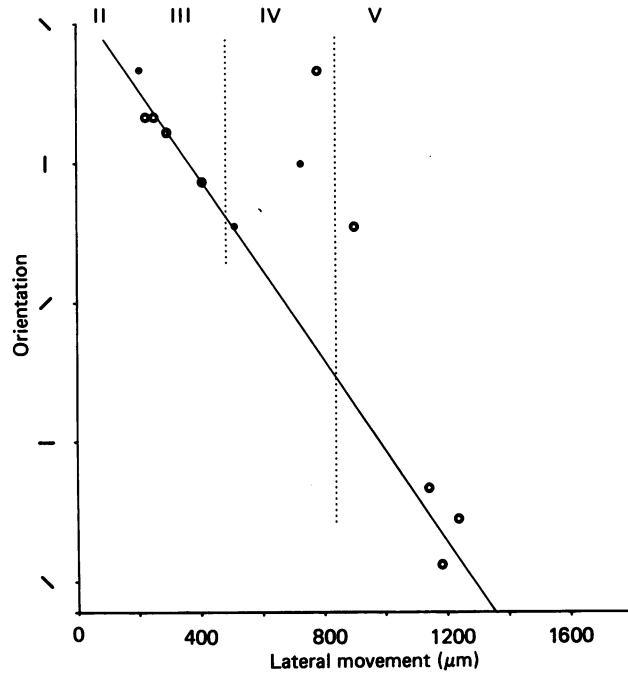


Fig. 6. Results for an oblique 'group 1' penetration, in which three cells were encountered in the vicinity of the IV/V border with orientation preferences that differed by up to 90 deg from an otherwise orderly trend. Format as for Fig. 5B, but cell type omitted for the sake of clarity.

apparent 'double shift' already described, of which an example is illustrated in Fig. 6. Whether these do indeed represent fractures in either the vertical or horizontal organisation of the columnar system, or simply the coincidence of several exceptional cells within an otherwise orderly system, is impossible to resolve. Certainly the orientation preferences recorded above and below these cells followed a single, common trend, suggesting that there was no significant disruption of the system as a whole. Finally, three single fractures were encountered. One is undoubtedly genuine, having a magnitude of 90 deg and occurring in mid-lower layer VI. The remaining two are ambiguous, in that they appear as a result of only one or two cells with orientations that deviate substantially from the expected trend. For example, in the penetration illustrated in Fig. 7 there appears by eye to be a fracture in the upper part of layer VI, yet a regression line drawn through the layer III-V data passes through the last point, leaving only the two intermediate cells with unexpected preferences.

Hence, it may be seen that where some form of discontinuity was observed it was rarely of the type reported by Bauer (1982, 1983); that is to say, a 45–90 deg step-wise shift in orientation preference separating regions with very much lower drift rates. Nevertheless, the distribution of the observed instances is shown in Fig. 8, which

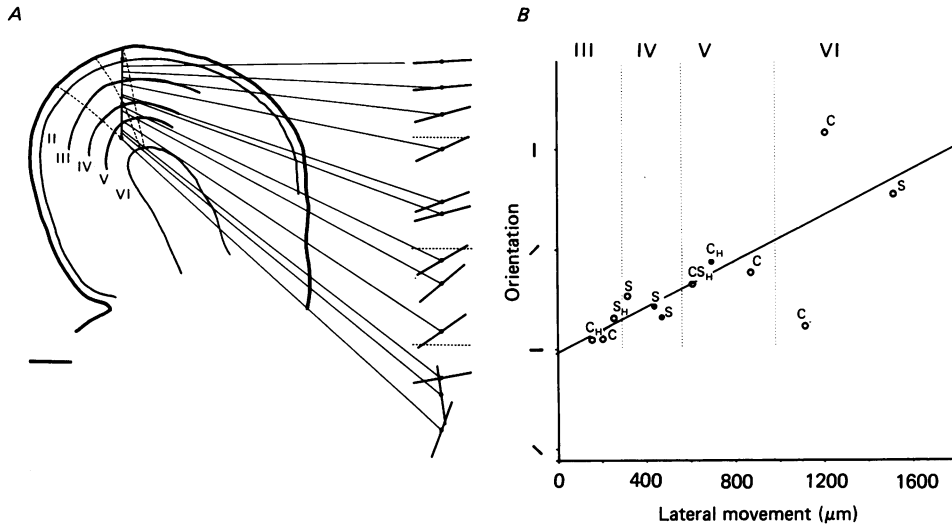


Fig. 7. Results of a markedly oblique penetration from 'group 1', in which a possible discontinuity was encountered. The format is identical to that of Fig. 5, with the exception that the optimum orientation only is indicated in *A*, for the sake of simplicity. Regression line calculated from the layer III–V data.

is a histogram summarizing the frequency and location of all the irregularities encountered in the 'group 1' data, compared with the number of occasions when no such evidence was found. The horizontal axis represents the layers of the visual cortex, with layers III and VI divided into upper and lower halves in order to give eight approximately equal sections. Below this axis are plotted those occasions when an electrode passed through and beyond each section, and recorded a sufficient number of cells to show without doubt that no discontinuity was encountered. Above it are those occasions when a discontinuity (defined as a sustained shift in orientation preference greater than the inherent variability within the track; that is greater than 30 deg in regions of average base-line scatter) did occur, or when the data could be interpreted in either way. A major fault in the columnar system, of the type described by Bauer (1982, 1983), would lead to a large peak in the upper section of the histogram in the division marked by the arrowhead, and a corresponding trough in the lower section. This is clearly not the case.

Continuity at the IV/V border in 'group 2' experiments

For the eighteen 'group 2' penetrations, the change in average orientation between the groups of cells recorded above and below the IV/V border was compared with the tangential distance between recording sites. Fig. 9 illustrates the results of this

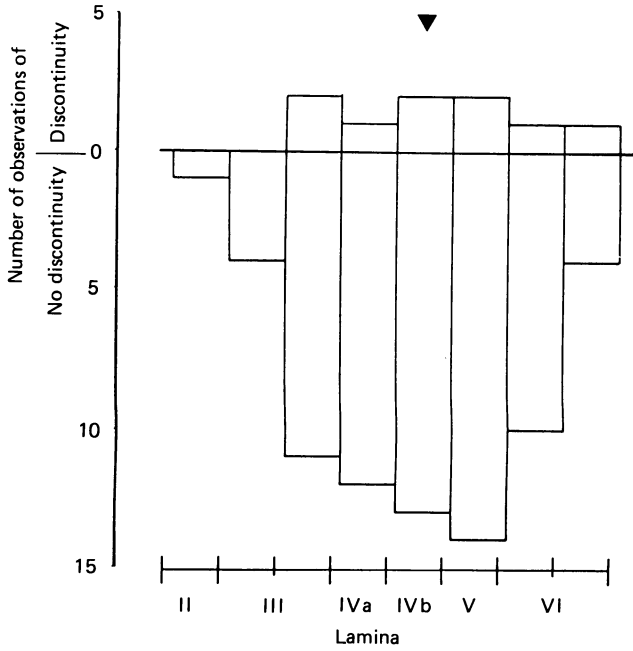


Fig. 8. Histogram illustrating all those occasions when an electrode passed through and beyond each cortical lamina, with (upper section) and without (lower section) any indication of an irregularity in the orientation trend. See text for further details.

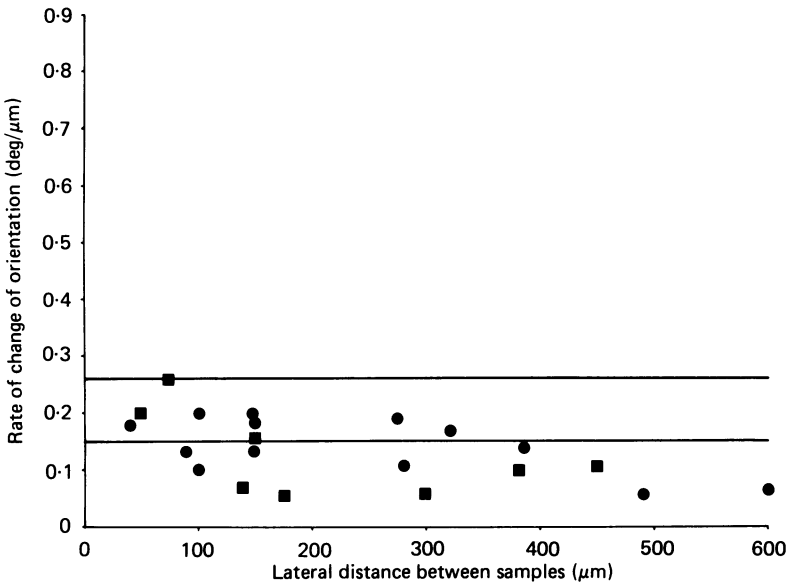


Fig. 9. Graph of the rate of change recorded between two sets of orientation preferences, one above and one below the IV/V border region in each of twenty-one near perpendicular penetrations, as compared with the tangential distance between them. Circles refer to 'group 1' and squares to 'group 2' experiments. The parallel horizontal lines indicate the limits set by previous authors for the expected drift rates in area 17 of the cat (Hubel & Wiesel, 1962, 1974; Albus, 1975).

analysis for the eight penetrations in which superficial and deep layer recordings were made within a horizontal distance which, when projected to the cortical surface, was less than 600 μm . This is the greatest distance within which an unambiguous (i.e. less than 90 deg) shift in orientation might be expected (Hubel & Wiesel, 1962, 1974; Albus, 1975). In every instance the values recorded were either within the limits expected from the work of other authors (Hubel & Wiesel, 1962, 1974; Albus, 1975), in which case it may be assumed that no discontinuity in the columnar system was encountered, or somewhat lower, the range of values being 50–250 deg/mm. This compares well with the results shown in the previous section for the 'group 1' penetrations, as does the fact that the lower drift rates tended to be associated with penetrations for which no caudal to rostral slope could be detected. In order to illustrate the overlap between these data more fully, Fig. 9 also shows the results of thirteen 'group 1' penetrations, for which suitable blocks of data were subjected to the same analysis. In none of the latter cases was there a major discontinuity between the relevant recording sites. In view of the consistency between these two sets of results, it is concluded that the 'group 2' experiments contain no evidence for a difference between deep and superficial layer orientation preferences.

DISCUSSION

The results of the 'group 1' penetrations from this series of experiments confirmed the broad pattern described by Hubel & Wiesel (1962, 1963, 1974), in that neighbouring and overlying cells were found to share similar orientation preferences, with a tendency for that preference to change in the tangential direction according to a gradual, over-all trend. Our results differed from those of Hubel & Wiesel only in the greater degree of random variability that was superimposed upon these trends. However, the normal level of scatter agreed well with that seen in the data of Albus (1975), while the appearance of single cells with widely deviant preference were also as reported previously (Albus, 1975; Lee *et al.* 1977).

No evidence was found to confirm the presence of a discontinuity in the orientation domain between the superficial and deep layers. This is illustrated most clearly in the results from the few truly perpendicular penetrations, in none of which was a significant change in orientation preference encountered at any cortical depth. It is self evident that these results could not have been obtained if the system were organized in the way described by Bauer (1982). This is further confirmed by the results of those oblique penetrations in which the orientation trends remained constant throughout the length of the track. Again it should have been obvious had the electrode passed through a major dislocation in the columnar system. Those irregularities that were encountered occurred only in oblique penetrations, and were distributed through the depth of the cortex rather than confined to any single layer. The logical explanation is that they represent fractures in the tangential organization of the system rather than with depth; that is to say, fractures between groups of columns rather than within individual columns. Such fractures have been reported in previous electrophysiological studies, both by Hubel & Wiesel (1962, 1963, 1965) and by Albus (1975). They are presumed to represent blind endings and other irregularities in the pattern that the columns project in tangential section, as

revealed by 2-deoxyglucose experiments (Hubel *et al.* 1978; Albus, 1979; Lang & Henn, 1980; Schoppmann & Stryker, 1981; Singer, 1981; Albus & Sieber, 1984). It is interesting to note that there is evidence from such work to suggest that, at the eccentricity from which we were recording, that pattern is far more fragmentary and complex than elsewhere (Singer, 1981; Albus & Sieber, 1984). One would therefore expect to encounter these discontinuities with comparative frequency, in penetrations that were to any extent oblique. This comment applies equally well to the double as to the single shifts.

In the final analysis, our results entirely support the traditional concept of orientation columns or sheets that are continuous throughout the cortical depth. They are therefore in direct contrast to those of Bauer (1982, 1983). This is surprising in light of the now extensive documentation of this phenomenon in the primate visual cortex (Bauer *et al.* 1980, 1983; Kruger & Bach, 1982; Dow & Bauer, 1984).

The failure of previous authors to find a discontinuity has been explained in terms of the specificity of the experimental method used (Bauer, 1982). Thus it has been claimed that other investigators failed to correlate large orientation shifts in their data with lamina, because they did not design their experiments or analyse their results with that possibility in mind. This is certainly true of Albus (1975) and Lee *et al.* (1977), neither of whom contributed decisive evidence in this matter. However, the argument is far less convincing when applied to the work of Hubel & Wiesel, who made an extensive and very specific investigation of all aspects of the geometry of the orientation domain (Hubel & Wiesel, 1962, 1963, 1974). Their experiments were designed to study the organization of the system in both the tangential and the radial plane, and the continuity of the columns was one factor that received specific attention. In fact, not only did they state that orientation remains constant throughout the cortical laminae, but they published illustrations in which this is seen to be the case (e.g. Hubel & Wiesel, 1962, Fig. 13; 1965, Fig. 2). Thus the presence of a discontinuity would imply a major error on their part. While this is by no means impossible, it does seem unlikely in light of the present results. The disparity between the conclusions of this study and that of Bauer is even more striking, since the specific aims and the basic experimental protocol were very similar in either case.

One possibility is that there is a population of cells, encountered occasionally in all cortical layers in this study but most commonly in layers IV–VI, with orientation preferences that differ markedly from that of the basic columnar unit. This would, to take just one example, be an expected feature of inhibitory interneurons associated with the generation of orientation selectivity in the surrounding cells (Sillito, 1975, 1979, 1984; Sillito, Kemp, Milson & Berardi, 1980). If this were so, then a column could perhaps appear continuous or discontinuous, according to the sampling characteristics of the micro-electrode used. The three 'group 1' penetrations in which a cluster of two or three such aberrant cells produced an apparent double shift could certainly be taken as evidence for this, since they resembled in some respects the data reported by Bauer (1983). However, although this idea is appealing, we have found no real evidence that it is so. In contrast to Bauer we found double shifts only in penetrations that were markedly oblique, and so consideration of these instances cannot be restricted to an interpretation based on the vertical movement of the electrode alone. As indicated earlier, disruptions in the tangential pattern of

the columns could as easily explain our observations. Furthermore, it cannot be ruled out that we occasionally recorded from the axonal or dendritic processes of cells in columns distant from the electrode, and that these recordings account for the apparent shifts.

However, it is pertinent to consider the fact that the data quoted by Bauer are not entirely unambiguous. Although the results from seventy-five penetrations have now been reported, only twenty-three of these, less than half of which showed a discontinuity at the IV/V border, were histologically reconstructed (Bauer, 1982, 1983). The locations of the remaining large orientation shifts were inferred from these examples, while the deviation from perpendicular of the unreconstructed tracks remains unknown. It is therefore possible that the majority of these penetrations were to some extent oblique, especially since those examples of reconstructed tracks published in Bauer (1982) appear to be quite markedly so. As such they would have encountered fractures in the tangential organization of the columnar system, as well as the normal drift in orientation preference. It is easy to see how the data from such penetrations could have been misinterpreted as evidence of a laminar discontinuity under these circumstances. The more recent report states that a dislocation is found as often in layer VI as at the IV/V border, and that there is a marked shift in receptive field location congruent with the shift in orientation (Bauer, 1983). Both facts tend to confirm the suspicion that these shifts are associated with tangential movement of the electrode.

In short, taking the present evidence into account, we can see no reason to support the view that there is a systematic dislocation in the representation of orientation preference between superficial and deep layers of the cat primary visual cortex. This view is consistent with the work of Hubel & Wiesel, with the results of 2-deoxyglucose experiments, and with many facets of the anatomical organization of the cortex (such as dendritic bundles) which favour some type of functional continuity through the depth of the cortex. Models of visual cortical processing based upon the presence of such a dislocation in the cat (Bauer, 1983) must therefore be regarded with caution.

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