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The pattern of ocular dominance columns in cat primary visual cortex: intra- and interindividual variability of column spacing and its dependence on genetic background

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Abstract

We present a comprehensive analysis of the intrinsic variability of the periodicity of ocular dominance columns in cat primary visual cortex (area 17) and its relationship to genetic background and visual experience. We characterized the intra-areal and interindividual variability of column spacing in a large set ($n=49$) of ocular dominance patterns adapting a recently developed technique for the two-dimensional analysis of orientation column patterns. Patterns were obtained from three different cat colonies (termed F, M and D), the cats having either normal visual experience or experimentally induced strabismus. Two-dimensional maps of local column spacing were calculated for every pattern. In individual cortices, local column spacings varied by >50% with the majority of column spacings ranging between 0.6 and 1.5 mm in different animals. In animals from colonies F and M ($n=29$), the mean column spacing ranged between 1.03 and 1.27 mm and exhibited no significant differences, either between the two breeds or between strabismic and normal animals. The mean spacing was moderately clustered in the left and right brain hemisphere of individual animals but not in littermates. In animals from colony D ($n=2$), average column spacing ranged between 0.73 and 0.95 mm, and was thus significantly different from the distribution of spacings in animals from breeds F and M, suggesting an influence of genetic factors on the layout of ocular dominance columns. Local column spacing exhibited a considerable systematic intra-areal variation, with largest spacings along the representation of the horizontal meridian and smallest spacings along the peripheral representation of the vertical meridian. The total variability of ocular dominance column spacing comprised 24% systematic intra-areal variation, 18% interindividual differences of mean column spacing and 58% nonsystematic intra-areal variability.

Introduction

The columnar organization of neuronal circuits in the cerebral cortex is a universal principle of neocortical architecture. In the visual cortex, for instance, the afferents from the left and the right eye terminate in alternating patches, called ocular dominance columns (Wiesel *et al.*, 1974; Shatz *et al.*, 1977; LeVay & Nelson, 1991). In a plane parallel to the cortical surface, neuronal selectivities vary so that left and right eye domains form highly organized two-dimensional (2D) patterns (Löwel & Singer, 1987; Anderson *et al.*, 1988; Löwel, 2002). The function of the system of ocular dominance columns is unclear at present. Studies of macaque monkey primary visual cortex demonstrated a substantial interindividual and intra-areal variability of spacing and size of ocular dominance columns (Horton & Hocking, 1996). In the squirrel monkey, ocular dominance columns vary so strongly that a significance of ocular dominance columns for visual perception seems rather unlikely in this species (Adams & Horton, 2003). These observations raise three important questions. (i) Do ocular dominance columns vary to a similar extent in the primary visual cortex of other species? (ii) What is the origin of this variability, i.e. what factors control column size and spacing during development? (iii) Is there a general systematic dependence of column spacing on retinotopic position in the map of visual space, such that different parts of the field of view are processed by columns of different sizes? In order to conclusively answer these questions it is necessary to assess column spacing both quantitatively and locally within area 17 and to quantitatively disentangle and compare the different components contributing to the total variability of column spacings. Previous quantitative analyses of ocular dominance patterns, however, estimated only the average spacing of the columns, neglecting possible intra-areal variability of the column layout, and thereby gave a rather incomplete picture of the intrinsic variability of column layouts.

In this study, we present a precise and reliable quantitative assessment of the local spacing of ocular dominance columns in cat primary visual cortex (area 17). In order to achieve such a characterization, we adapted a recently developed image analysis technique, based on the wavelet transform (Kaschube *et al.*, 2000, 2002), for the characterization of ocular dominance columns. Our analysis extracts a 2D map of local column spacings from patterns of ocular dominance columns, labelled with either 2-deoxyglucose or proline autoradiography (Löwel & Singer,

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1987, 1992, 1993a,b; Löwel, 1994, 2002; Rathjen & Löwel, 2000; Rathjen *et al.*, 2002). Based on these maps, we quantitatively characterized the intrinsic interindividual and intra-areal variability of column spacing and the systematic dependence of column spacing on the position of columns within area 17 (retinotopic location), and assessed the relative contribution of these factors to the total variability of column spacing. Furthermore, we examined the dependence of local column spacing on visual experience and on genetic background.

Our analyses provide important baseline information for studies investigating the mechanisms underlying ocular dominance column formation. One key question of this research is whether experimental modification of neuronal activity patterns can change column spacing: because the spacing of columns is not influenced by purely local modifications of column shapes from a predefined grid (such as the shrinkage or expansion of individual domains, as happens with monocular deprivation), the successful experimental induction of an abnormal spacing of ocular dominance columns by manipulation of neuronal activity patterns would demonstrate that the pattern predominantly forms by an activity-dependent mechanism and is not predetermined by a hidden proto-map (see Stryker, 1991; Löwel, 1994; Hensch & Stryker, 1996; Crowley & Katz, 2002). As pointed out by Horton & Hocking (1996), every experimental modification of column layouts has to be disentangled from a potentially large intrinsic, intra-areal and interindividual variability of columnar systems. A precise and reliable quantitative characterization of the intrinsic variability of ocular dominance patterns is therefore an indispensable prerequisite for any study trying to modify basic layout properties of ocular dominance columns by experimental manipulations of neuronal activity patterns. Some of the results have been reported in abstract form (Kaschube *et al.*, 2001a,b).

Materials and methods

Animals

We analysed patterns of ocular dominance columns in primary visual cortex (area 17) of 31 cats (49 hemispheres). Some of the ocular dominance maps of these animals have already been published (Löwel & Singer, 1987, 1993a,b; Löwel *et al.*, 1988; Löwel, 1994, 2002; Rathjen & Löwel, 2000; Rathjen *et al.*, 2002). Ocular dominance patterns were labelled with either 2-[^{14}C]-deoxyglucose (2-DG) autoradiography (Sokoloff *et al.*, 1977) after monocular stimulation of the animals or by [^{3}H]-proline autoradiography after injection of the labelled proline into one eye which labels the thalamocortical afferents of that eye in cortical layer IV (Grafstein, 1971; Wiesel *et al.*, 1974). All experimental procedures are described in detail in Löwel and Singer

TABLE 1. Dataset used for quantitative analysis of ocular dominance (OD) maps in cat area 17

Cat ID	Hemi-sphere	Label	Autoradio-graphy (*n*)	Weight (g)	Age (weeks)	Experience S/N	V+H
M1-L7	L+R	2-DG	5+3	820	7.5	S	–
M2-L7	R	2-DG	6	840	8.5	S	–
M3-L8	L	2-DG	7	980	11	S	–
M4-L8	L	2-DG	5	1050	12	S	–
M5-L9	L	2-DG	3	1100	14	N	–
M6-L9	L	2-DG	4	1100	12	S	–
M7	L+R	2-DG	5+6	1370	12	S	–
M8	L+R	2-DG	2+4	330	4	S	–
M9	L	2-DG	4	540	6	N	–
F1-L4	L+R	2-DG	4+4	850	9.5	N	L+R
F2-L4	L+R	2-DG	4+3	1080	10.5	N	L
F3	L+R	2-DG	4+4	490	5	S	R
F4-L1	L+R	2-DG	4+4	350	4	N	L+R
F5-L1	L+R	2-DG	4+4	340	4	S	L
F6-L2	L+R	2-DG	3+3	720	6	S	–
F7-L2	R	2-DG	3	550	6	N	–
F8	L+R	2-DG	3+2	320	5	N	–
F9-L3	L+R	2-DG	4+3	490	6	S	L+R
F10-L3	L+R	2-DG	2+4	460	6	N	–
F11	L	2-DG	2	460	5	N	–
F12	L+R	2-DG	4+3	430	4	S	L
F13	L+R	2-DG	4+2	1820	14	N	L
F14-L5	L+R	2-DG	4+4	840	8	S	L+R
F15-L5	L+R	2-DG	4+4	1510	13	S	L+R
F16	L	2-DG	4	940	10	S	L
F17	L	2-DG	4	1120	10	S	L
F18	L	2-DG	4	880	9	S	–
F19-L6	R	Proline	1	980	8.5	S	–
F20-L6	L	Proline	1	1000	9	S	–
F20-L6	L	2-DG	4	1000	9	S	R
D1-L10	L+R	Proline	1+1	1200	12	N	–
D2-L10	L+R	Proline	1+1	1200	12	N	–

We analysed 49 hemispheres from 31 animals from three different cat colonies. Animals belonging to the same colony are labelled accordingly: nine cats, M1–M9, (12 hemispheres) were born and raised in the animal house of the Leibniz-Institut für Neurobiologie in Magdeburg, Germany; 20 animals, F1–F20, (33 hemispheres) were born and raised in the animal house of the Max-Planck-Institut für Hirnforschung in Frankfurt am Main, Germany; two cats, D1 and D2, (four hemispheres) were bought from a commercial breeding company (Gaukler, Offenbach, Germany). Furthermore, the dataset included 10 litters (L1–L10) and contained 19 strabismic animals (seven from colony M, 12 from colony F). Hemisphere (L, left; R, right; L + R, both); Label, the labelling method; (*n*), the number of autoradiographs analysed; Wt (g), body weight; Age (wk), the age in weeks at the time of the experiment; S/N, the visual experience of the animal (S, strabismus vs. N, normal visual experience); V+H, the subset of hemispheres for which the cortical representation of the vertical and horizontal meridian could be identified (see Materials and methods). Results presented in Figs 7c, 9 and 10 are based on this subset.

(1987, 1993a,b) and Löwel (1994). Table 1 lists details of the dataset used in the present analysis. The animals were from three different cat colonies. 'Colony M' consisted of nine cats (12 hemispheres) born and raised in the animal house of the Leibniz-Institut für Neurobiologie in Magdeburg, Germany. 'Colony F' comprised 20 animals (33 hemispheres) born and raised in the animal house of the Max-Planck-Institut für Hirnforschung in Frankfurt am Main, Germany. Two additional cats (four hemispheres) were bought from a commercial animal breeding company (Gaukler, Offenbach, Germany); we refer to them as 'colony D'. Furthermore, the dataset contains patterns from 19 strabismic animals (seven animals, 10 hemispheres from colony F and 12 animals, 19 hemispheres from colony M; see Table 1). All animal experiments have been performed according to the German Law on the Protection of Animals and corresponding European Communities Council Directive of November 24 1986 (86/609/EEC).

Analysis method

Patterns of ocular dominance columns were analysed using methods adapted from our previous quantitative analyses of orientation column patterns in cat area 17 (Kaschube *et al.*, 2000, 2002). Compared to orientation columns, the labelling of ocular dominance columns was often weaker and more nonhomogeneous in signal contrast. Therefore, we introduced additional steps in the preprocessing of the data and calculated wavelet transforms with higher resolutions. All steps of the present analysis are described in detail in the following paragraphs. Basically, as sketched in Fig. 2, the spacing of columns was estimated by matching the 2D structure of the ocular dominance column pattern in a small circular region with an appropriate localized stereotype pattern, the wavelet. The wavelet itself was varied in spacing (the distance of modulations within the wavelet) in order to find the optimal overlap with the column structure at this location (compare Fig. 2c). The spacing of the wavelet leading to an optimal overlap was used as a measure of the local column spacing. One can think of this procedure as some kind of local power spectrum analysis, from which the typical spatial frequency of a signal is extracted. Finally, after having determined the local column spacing for each location in each cortex, hemisphere and population averages were extracted from these quantities to facilitate the comparison between column spacing in different hemispheres or at different cortical locations.

Image digitization

Proline and 2-DG autoradiographs were photographed using a digital camera (Nikon Coolpix 950 Digital, 1024 × 768 pixel resolution), and resampled by bicubic interpolation using Adobe Photoshop 5.5 to an effective spatial resolution of at least 8.4 pixels/mm cortex and 256 grey levels per pixel. For every autoradiograph this yielded a 2D array of grey values $I_0(\mathbf{x})$, where $\mathbf{x}$, a 2D vector, is the position within the area and I_0 its intensity of labelling. Figure 1 illustrates representative 2-DG and proline autoradiographs of the ocular dominance patterns in cat area 17.

Region of interest

For every autoradiograph a region of interest encompassing the pattern labelled in area 17 was defined by visual inspection. The 17/18 border was identified based on the larger column spacing and different pattern layout in area 18 than in area 17 (Löwel & Singer, 1987, 1993a,b; Löwel, 1994). The manually defined polygon encompassing the entire ocular dominance pattern within area 17 was stored together with every autoradiograph. Only the pattern within area 17 was used for subsequent quantitative analysis. The representation of the blind spot, monocular segment, regions with very low signal, and minor artefacts (scratches, folds, and air bubbles) were excluded from further analysis.

Preprocessing

All digitized patterns were preprocessed in order to remove overall variations in the intensity of labelling. To achieve this, first the local average labelling intensity

$$\bar{I}(\mathbf{x}) = \frac{\int_{A17} d^2y I_0(\mathbf{y}) K(\mathbf{y}-\mathbf{x})}{\left(\int_{A17} d^2y K(\mathbf{y}-\mathbf{x})\right)} \quad (1)$$

was calculated using the kernel $K(\mathbf{y}) = \frac{1}{2\pi\sigma_x^2}\exp(-\mathbf{y}^2/2\sigma_x^2)$ and subtracted from the pattern

$$I_1(\mathbf{x}) = I_0(\mathbf{x}) - \bar{I}(\mathbf{x}). \quad (2)$$

The spatial width σ_x of the kernel was chosen such that patterns with a wavelength <1.5 mm, the range of column sizes, were only weakly attenuated by the preprocessing. We used $\sigma_x = 0.43$ mm, for which the attenuation at scale 1.5 mm was 20%. The attenuation for wavelengths <1.5 mm decreased rapidly. The pattern was then centred to yield $\int_{A17} d^2y I(\mathbf{y}) = 0$ and thresholded to uniform contrast by setting $I(\mathbf{x}) = 1$ in regions with positive values and $I(\mathbf{x}) = -1$ in regions with negative values. Finally, grey values in artefact regions and in regions outside area 17 were set to zero. Figure 2 shows a representative example of a preprocessed ocular dominance pattern (the upper left pattern of Fig. 1). The applied high pass filtering removed overall variations in labelling intensity but left the column pattern unaffected in all autoradiographs. For most of the patterns this procedure was inevitable. In several cases in the present study the strength of labelling was fairly homogeneous across parts of area 17 (e.g. in the upper left, upper middle and lower middle examples in Fig. 1). We used these cases as control examples, because the column spacing in these regions could also be estimated using a weaker filtering. For these regions, filtering changed the measured value often by <0.02 mm. The impact of the filtering on the relative difference in spacing between maps is expected to be small because all maps were affected by the filtering to a similar extent. The results presented in this work remain valid with a smaller signal attenuation. However, spacing values then get more affected by labelling variations leading to less accurate estimates. After the described preprocessing procedure, the labelling of columns was of fairly equal quality across different hemispheres and across each single area 17, thus minimizing any bias which might have been caused by such a inhomogeneity.

Analysis of local column spacing $\Lambda(\mathbf{x})$

For each analysed pattern of ocular dominance columns we determined a 2D map representing the column spacing at each cortical location. We first calculated wavelet representations of a given pattern $\mathbf{I}(\mathbf{x})$ using wavelets which only covered a few hypercolumns but exhibited a strong periodicity. According to the wavelet's confined spatial extension, the estimation at a given location was unaffected by regions at least $2-3$ hypercolumns apart. The representations were obtained from

$$\hat{I}(\mathbf{x},\theta,l) = \int_{A17} d^2y I(\mathbf{y}) \psi_{\mathbf{x},\theta,l}(\mathbf{y}), \quad (3)$$

where $\mathbf{x},\theta,l$ are the position, orientation and scale of the wavelet $\psi_{\mathbf{x},\theta,l}$ and $\hat{I}(\mathbf{x},\theta,l)$ denotes the array of wavelet coefficients. We used complex-valued Morlet wavelets defined by a mother wavelet

$$\psi(\mathbf{x}) = \exp\left(-\frac{\mathbf{x}^2}{2}\right) e^{i\mathbf{k}_\psi \mathbf{x}} \quad (4)$$

and

$$\psi_{\mathbf{x},\theta,l}(\mathbf{y}) = l^{-1}\psi\left(\Omega^{-1}(\theta)\frac{\mathbf{y}-\mathbf{x}}{l}\right) \quad (5)$$

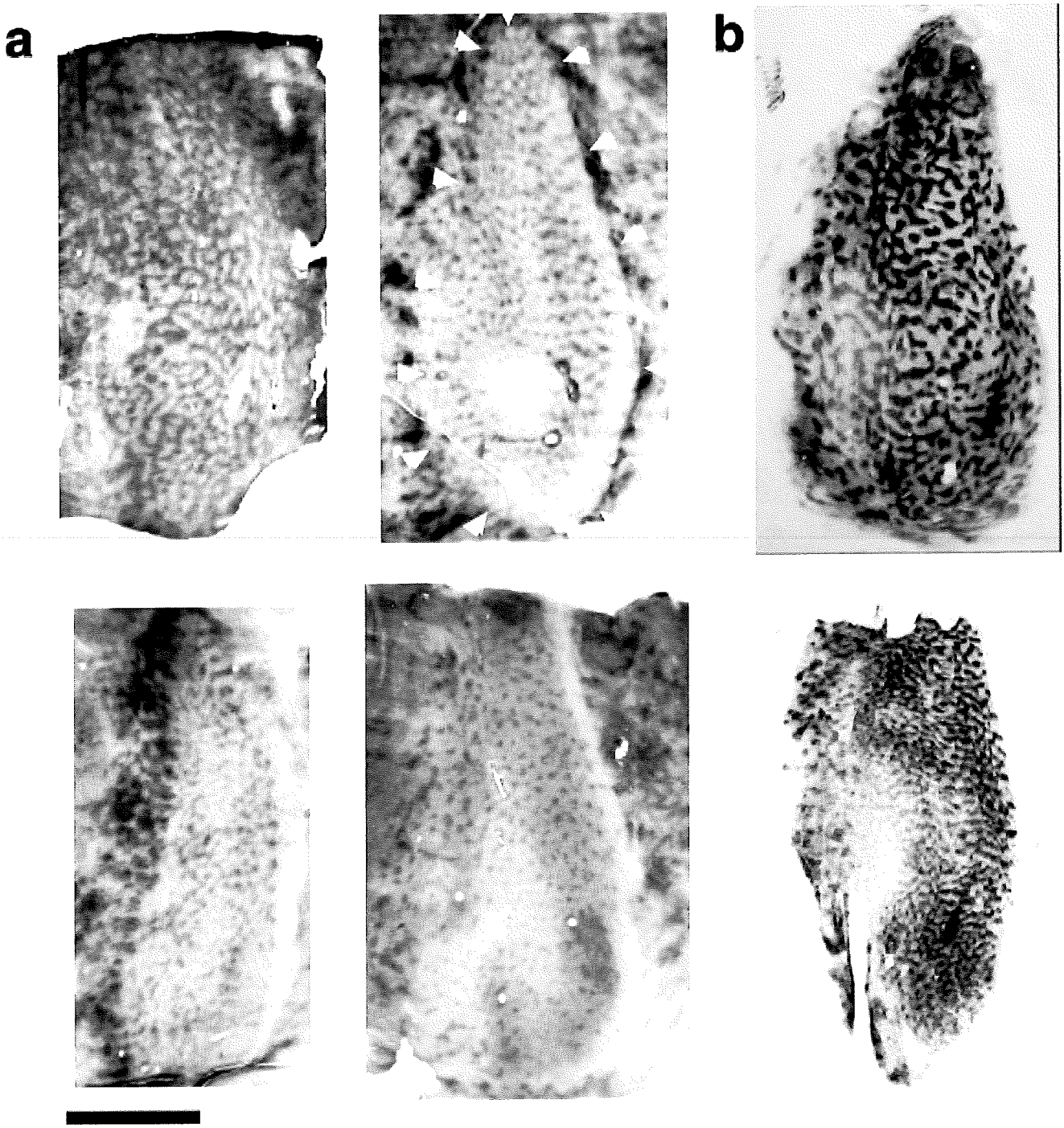

FIG. 1. Representative examples of ocular dominance (OD) column patterns in the primary visual cortex (area 17) of cats (from left to right, hemispheres in upper row: F14 left, F4 right, F20 left, ; in lower row: F3 right, F2 left, D2 right). (a and b) OD domains labelled by 2-[^{14}C]-deoxyglucose (2-DG) (a) and by [^{3}H]-proline (b) appear as dark grey to black patches on lighter grey to white background. OD patterns were visualized on cortical flat-mount sections and thus contain OD domains within the entire area 17. Area 17 is the drop-shaped region in the centre of the figures (marked by white arrowheads in the upper middle OD pattern), which is reliably discernible by its distinct pattern compared to surrounding cortical regions. All OD column patterns are displayed so that the 17/18 border appears left in all panels (to this end, the right hemisphere patterns were mirror-inverted) to aid comparison. Anterior is at the top, posterior is at the bottom of each figure. Scale bar, 10 mm.

with the rotation matrix

$$\Omega(\theta) = \begin{pmatrix} \cos(\theta) & -\sin(\theta) \\ \sin(\theta) & \cos(\theta) \end{pmatrix}.$$

The characteristic wavelength of a wavelet with scale l is $\Lambda_\psi l$ with $\Lambda_\psi = 2\pi/|\mathbf{k}_\psi|$. For large values of $|\mathbf{k}_\psi|$ the wavelets (eqn 4) exhibit a high resolution of spatial frequencies. For small values of $|\mathbf{k}_\psi|$ they are localized in the cortical coordinates enabling high spatial resolution. Therefore we used relatively large $|\mathbf{k}_\psi|$ wavelets [$\mathbf{k}_\psi = (7,0)$] to estimate local column spacing.

From these representations we estimated at each location the wavelet with optimal periodicity, i.e. the wavelet which best matched

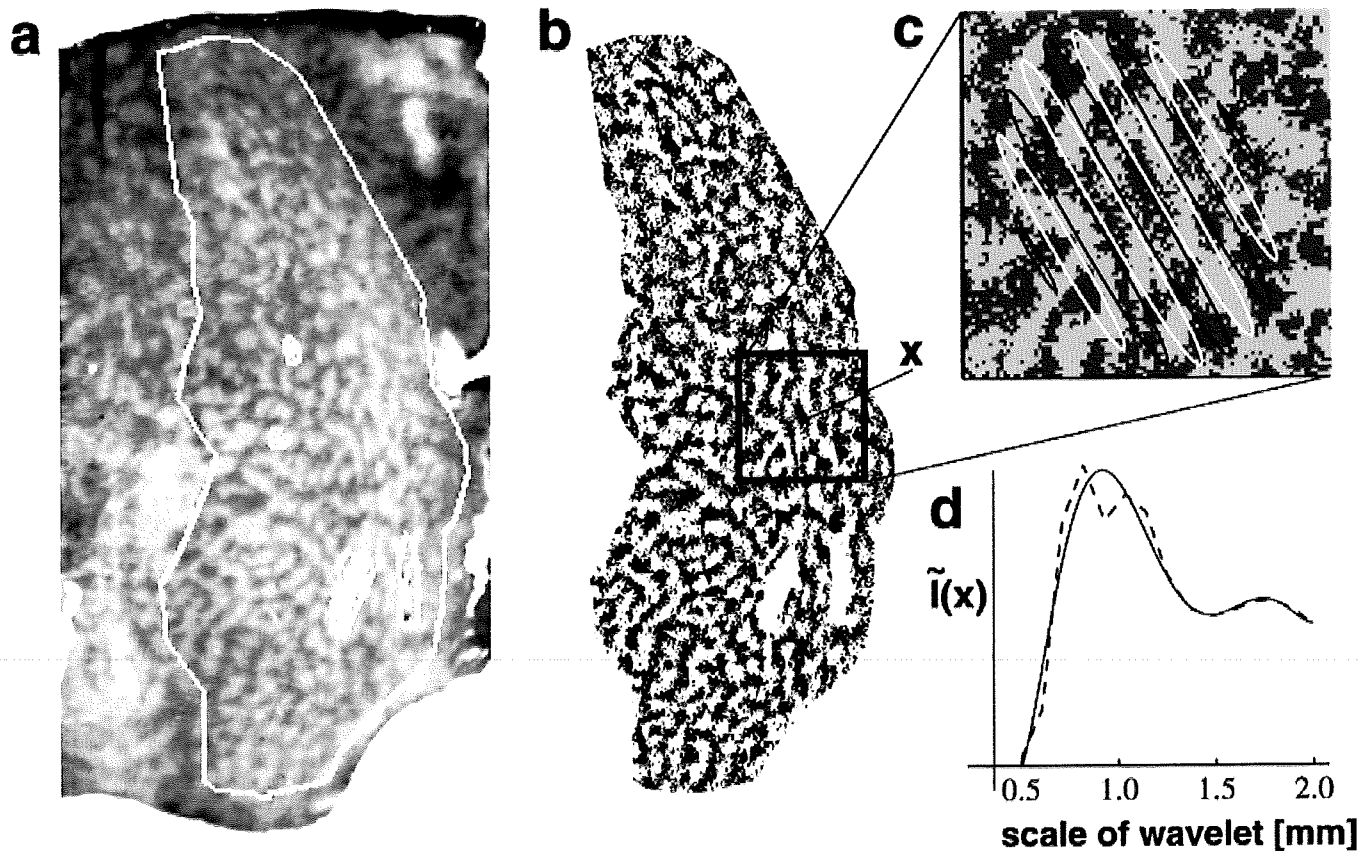

FIG. 2. Calculation of local column spacing based on wavelet analysis. (a) Typical 2-DG pattern of OD columns (the upper left pattern of Fig. 1). The region of interest (restricted to area 17, without artefacts) is marked by the white line. (b) After preprocessing (see Materials and methods), the OD pattern in the region of interest is contrast-enhanced locally (compare a with b). The representation of the blind spot, monocular segment and minor artifacts, delineated by white lines in (a), were excluded from further quantitative analyses. The region within the black box in (b) is shown enlarged in (c) to illustrate the calculation of column spacing: example of a wavelet $\psi_{\theta,\mathbf{x},l}$ of optimal scale centred at $\mathbf{x}$ superimposed on the OD pattern (background labelling illustrated in grey). Contour lines show the spatial structure of the real part of the complex-valued wavelet (Eqn 5). Positive regions are delineated by white lines, negative ones by dark lines. Note that the darkly labelled OD domains are mostly covered by the dark regions of the wavelet yielding a large absolute value of the wavelet coefficient. For nonoptimal scales the OD domains get more spread among both positive and negative regions of the wavelet leading to smaller values of the coefficients. Thus, the wavelet orientation averaged modulus $\bar{I}(\mathbf{x},l)=\int_0^{\pi}\frac{d\theta}{\pi}|\hat{I}(\mathbf{x},\theta,l)|$, shown in (d) (dashed line), exhibits a peak at a certain scale [estimated by a polynomial fit (solid line)] which we define as the local column spacing at position $\mathbf{x}$.

the column pattern around this location (Fig. 2). We first calculated the orientation averaged modulus

$$\bar{I}(\mathbf{x},l)=\int_0^{\pi}\frac{d\theta}{\pi}|\hat{I}(\mathbf{x},\theta,l)| \tag{6}$$

of the wavelet coefficients for every position $\mathbf{x}$ and then determined the scale

$$\tilde{l}(\mathbf{x})=argmax\left(\bar{I}(\mathbf{x},l)\right) \tag{7}$$

maximizing $I(\mathbf{x},l)$ with respect to l. The corresponding characteristic wavelength

$$\Lambda(\mathbf{x})=\tilde{l}(\mathbf{x})\Lambda_{\psi} \tag{8}$$

was used as an estimate for the local column spacing at the position $\mathbf{x}$ (Fig. 2). For every position (spatial grid size 0.12 mm) wavelet coefficients for 12 orientations $\theta_i\in\{0,\pi/12,\ldots,11\pi/12\}$ and 15 scales l_j [with $l_j\Lambda_{\psi}$ equally spaced at (0.5 mm, 2 mm)] were calculated. The scale maximizing $\bar{I}$ ($\mathbf{x}$, l) was then estimated as the maximum of a polynomial in l fitting the $\bar{I}$ ($\mathbf{x}$,l_j) for a given position $\mathbf{x}$ (least-squares fit). Figure 3 illustrates the 2D maps $\Lambda(\mathbf{x})$ of local column spacing for the ocular dominance maps depicted in Fig. 1. In all examples, spacing mostly changes smoothly with cortical location except for a few regions with rapid transitions.

Population averaged map $\Lambda_{pop}(\mathbf{x})$ of local column spacing

We superimposed and averaged 2D maps $\Lambda(\mathbf{x})$ of local column spacing obtained from different animals to define a population spacing map $\Lambda_{pop}(\mathbf{x})$ reflecting the systematic trend of local column spacing across area 17. For superposition, we first localized the representations of the vertical meridian (VM), the horizontal meridian (HM) and the area centralis on the autoradiographs and then aligned the 2D maps of local column spacing from different animals using these landmarks. The representation of the VM can be identified based on the larger column spacing and different pattern layout in area 18 compared to area 17 (Löwel & Singer, 1987; Löwel, 1994), the approximate location of the area centralis can be determined using the slightly weaker labelling of ocular dominance columns in this region than in more peripheral visual field representations (Löwel & Singer, 1987), and the HM crosses the vertical meridian at right angles (Tusa *et al.*, 1978; Löwel *et al.*, 1987). The alignment of spacing maps based on these topographic references matches corresponding locations from different hemispheres and reveals the values typical for a particular visual field representation.

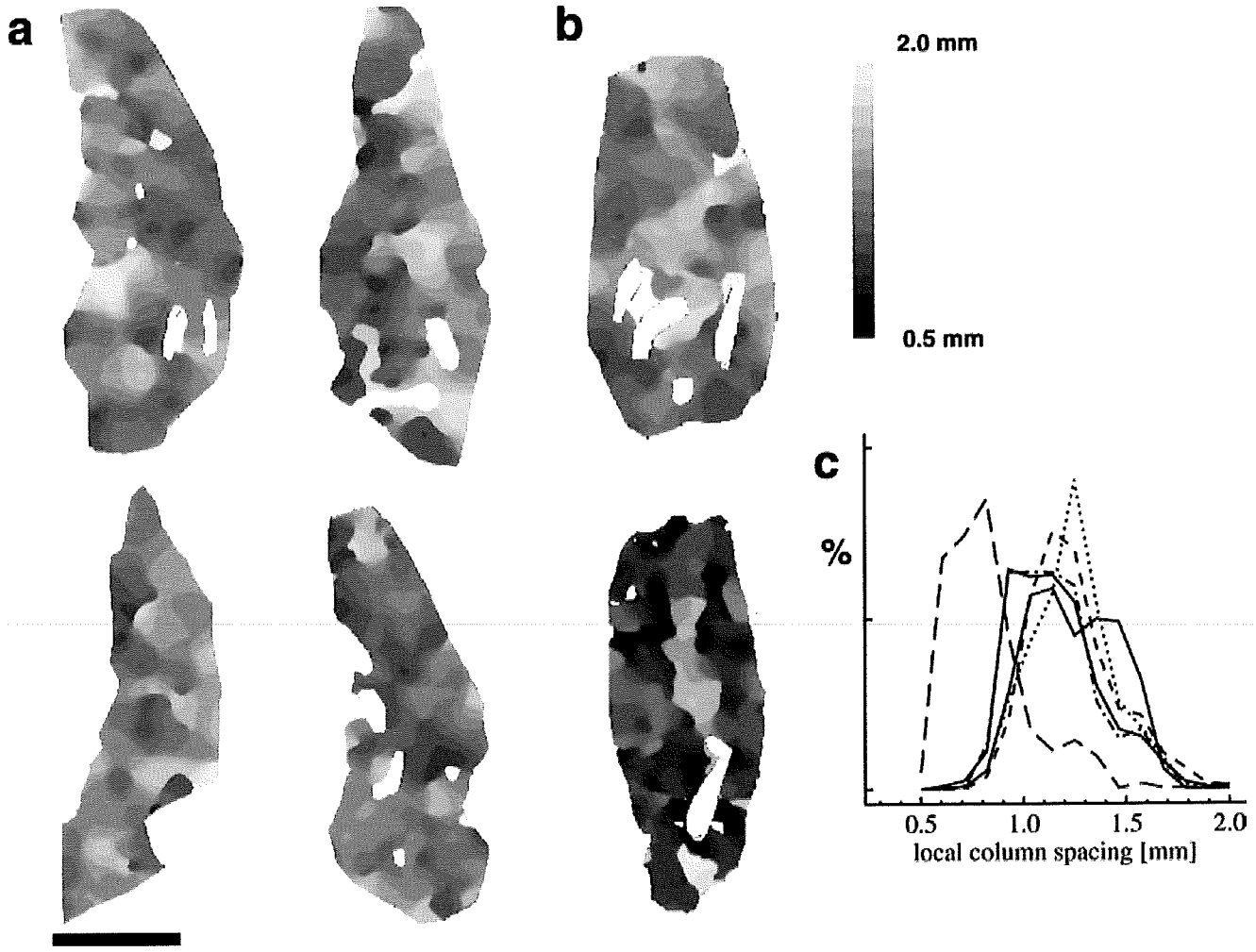

FIG. 3. 2D maps of local column spacing for the OD patterns displayed in Fig. 1, coded in grey scale. (a and b) Light grey regions exhibit a larger spacing then the average spacing of the population; dark grey regions exhibit smaller-than-average spacing (see inset). (c) Histograms of local column spacings for the six maps displayed in (a) and (b). In five of the maps (the three upper maps, the lower left map and the lower middle map), spacing values ranged primarily between 0.9 and 1.5 mm. The most frequent value varied between 1.1 and 1.3 mm. In the lower right map column spacing was considerably smaller: values range between 0.6 and 1.1 mm and the most frequent value was ≈0.8 mm (large dashed line). Scale bar, 10 mm.

In 18 hemispheres from 13 animals, the representation of the VM and the central visual field representation could be reliably estimated. Therefore, this subset of hemispheres (listed in Table 1) was used for computing the population column spacing map (Fig. 9), the local correlation of column spacing (Fig. 7) and the comparison of the interindividual and the intra-areal variability (Fig. 10).

Mean spacing Λ and spacing inhomogeneity σ_Λ

In order to characterize the spacing of ocular dominance columns in individual hemispheres conveniently, we defined two parameters capturing the main properties of the 2D map of local column spacing $\Lambda(\mathbf{x})$ (Figs 2 and 3). (i) The mean column spacing $\Lambda = \langle \Lambda(\mathbf{x}) \rangle_{\mathbf{x}}$, i.e. the average of all local column spacing values over the entire area 17. It measures whether a pattern predominantly contains large or small ocular dominance columns. (ii) The spacing inhomogeneity $\sigma_\Lambda = \sqrt{\langle (\Lambda(\mathbf{x}) - \Lambda)^2 \rangle_{\mathbf{x}}}$, the SD of local column spacing within area 17. The parameter σ_Λ measures the variability of spacing of ocular dominance columns within a single hemisphere, i.e. whether a pattern contains regions of different spacing or whether it is rather homogeneous in spacing.

Accuracy and reliability of parameter estimation

In order to estimate the parameters reliably and precisely, it is important to pool values obtained from single flat-mount sections at various cortical depths for each brain hemisphere. Values generally vary among different sections, due to the different noise level of labelling and due to the differences of the artefact-free region which can be analysed in each section. Parameters were estimated by averaging the values from up to seven different sections per hemisphere, and errors were computed as SEMs from these values when more than one section was available.

To estimate Λ and σ_Λ for a given hemisphere, there are two alternatives. We refer to them as method 1 and method 2. Method 1 consists of first calculating a parameter for each available flat-mount section and then averaging over different sections if more than one section is available. Method 1 can be carried out for every hemisphere. Method 2 consists of first aligning (see above) the available sections for each hemisphere to enable an averaging of local spacing values at every cortical location before calculating the average over different locations. Method 2 is more accurate, but can only be applied to a subset of the hemispheres where such an alignment is possible (listed in Table 1).

Error of local column spacing $\Lambda(\mathbf{x})$ and the population column spacing map $\Lambda_{pop}(\mathbf{x})$

In order to analyse the error of the local column spacing, $\Delta\Lambda(\mathbf{x})$, it is necessary to match corresponding locations between different sections from the same hemisphere. Therefore, the error estimation was based on the subset of data appropriate for method 2 (listed in Table 1). We only considered locations where we sampled from at least three different sections. Errors $\Delta\Lambda(\mathbf{x})$ were on average 0.060 mm, corresponding to 44% of the SD of local spacings $\Lambda(\mathbf{x})$ (see Table 2). Regions with relatively high contrast labelling in all analysed sections yielded very consistent values of spacing across sections and thus gave rise to small error measurements, whereas lower signal-to-noise ratios (in other regions) sometimes caused larger errors. The error of the population spacing map $\Lambda_{pop}(\mathbf{x})$ was calculated by error propagation from the error of the local spacing, which is $\Delta\Lambda_{pop}(\mathbf{x}) = \sqrt{\langle\Delta\Lambda(\mathbf{x})^2\rangle_{pop}}/\sqrt{N_{pop}}$, where the average is taken over the population of the N_{pop} hemispheres contributing to the population map $\Lambda_{pop}(\mathbf{x})$ (listed in Table 1). The mean of this error over all locations $\mathbf{x}$ was $\langle\Delta\Lambda_{pop}(\mathbf{x})\rangle_{\mathbf{x}} = 0.031$ mm.

Error of spacing parameters Λ and σ_Λ

The value of the mean spacing Λ and the spacing inhomogeneity σ_Λ are expected to be more reliable than typical local spacings $\Lambda(\mathbf{x})$, because they are averages over many local values. For method 1, errors of both parameters ranged between 0.001 and 0.066 mm. For the mean spacing Λ the error averaged over all hemispheres was 0.018 mm, corresponding to 34% of the SD of mean column spacings Λ from different hemispheres. For the spacing inhomogeneity σ_Λ the mean error was 0.012 mm, equal to 42% of the SD of values for σ_Λ in different hemispheres (Table 2). For method 2, the accuracy of the mean spacing Λ is estimated by error propagation from the error of the local spacing, $\Delta\Lambda(\mathbf{x})$, as $\sqrt{\langle\Delta\Lambda(\mathbf{x})^2\rangle_{\mathbf{x}}}/\sqrt{N_{ind}}$, where N_{ind} is the number of independent estimates of $\Lambda(\mathbf{x})$ within the map. N_{ind} depends on the nature of the noise causing the error. If the spatial correlation length of the noise is smaller than the wavelet with typical wavelength equal to Λ, N_{ind} is given by the area of the map ($\approx 250\,mm^2$) divided by the area of the wavelet with typical wavelength equal to Λ ($\approx 5\,mm^2$), which yields $N_{ind} \approx 50$. However, if the noise is correlated over a larger range than the wavelet, then N_{ind} might be considerably smaller. Hence, using method 2, the error for Λ ranged between 0.010 and 0.074 mm, or between 16 and 112% of the SD of the mean spacing Λ in the population.

The advantage of method 2 is more pronounced for calculating σ_Λ. The knowledge of the local error $\Delta\Lambda(\mathbf{x})$ allows compensation for the impact of this error on σ_Λ: inserting the local spacing with its error, $\Lambda(\mathbf{x}) + \Delta\Lambda(\mathbf{x})$, into the equation for the variance of local column spacing yields $s_\Lambda^2 = \sigma_\Lambda^2 + \langle\Delta\Lambda(\mathbf{x})^2\rangle_{\mathbf{x}} + \langle 2\Delta\Lambda(\mathbf{x}(\Lambda - \Lambda(\mathbf{x}))\rangle_{\mathbf{x}}$, of which the third term should be negligible, thus leading to $\sigma_\Lambda = \sqrt{s_\Lambda^2 - \langle\Delta\Lambda(\mathbf{x})^2\rangle_{\mathbf{x}}}$ as the proper estimate for the true value of σ_Λ. The values of the spacing inhomogeneities σ_Λ when calculated by method 2 are expected to be smaller and more accurate than when they are calculated by method 1. Because deviations from the mean spacing contribute quadratically to σ_Λ, first averaging over sections is safer because it reduces the occurrence of the (often artifactual) extreme spacing values strongly biasing σ_Λ towards large values.

Methods 1 and 2 were in very good agreement with each other in the case of the mean spacing Λ, but the results deviated more strongly in the case of the spacing inhomogeneity σ_Λ. The two values of Λ for a given area 17 differed typically only $\approx$0.010 mm from each other and were strongly correlated ($r = 0.99$, $P < 0.0001$). The spacing inhomogeneity σ_Λ calculated by method 2 was on average 0.08 mm smaller than its value obtained by method 1. The correlation between the two results was relatively small ($r = 0.28$) and not significant. The reason for this difference is that, by first averaging over sections, the local spacing $\Lambda(\mathbf{x})$ is estimated more precisely and its reduced error also reduces the error of the spacing inhomogeneity σ_Λ.

Influence of labelling methods

To check whether the estimation of the local column spacing $\Lambda(\mathbf{x})$, the mean column spacing Λ and the spacing inhomogeneity σ_Λ depends on whether column patterns were labelled with 2-DG or with proline, we additionally compared these parameters in one double-labelled hemisphere (animal F20). Parameter values obtained from a proline-labelled ocular dominance pattern were compared with the same parameters obtained from 2-DG-labelled sections at four different depths from the identical hemisphere of the same animal. The difference between the mean column spacing Λ for the two labellings was only 0.019 mm, corresponding to 35% of the interindividual variability (SD) of Λ. For the spacing inhomogeneity σ_Λ the two values differed by $\approx$0.027 mm from each other or 55% of the interindividual variability of σ_Λ. These differences between 2-DG- and proline-labelled patterns were within the range of differences which we normally obtained from analysing 2-DG patterns from different sections of the same hemisphere (see Table 2), indicating that neither parameter value is affected by the type of labelling of ocular dominance columns.

Dependence on the parameters of the wavelet analysis

The calculated values for the local column spacing $\Lambda(\mathbf{x})$, the mean column spacing Λ and the spacing inhomogeneity σ_Λ were insensitive to variation in the parameters of the image analysis method such as $\sigma_{\mathbf{x}}$ and $\mathbf{k}_\psi$, and the number of used wavelet orientations and scales.

Quantification and decomposition of variability

The variability of a given spacing parameter [e.g. Λ, $\Lambda_{pop}(\mathbf{x})$] was quantified by the SD of the values for that parameter corrected for the systematic error which is caused by the errors of these values. The interindividual variability σ_{div} of the mean column spacing Λ was calculated by $\sigma_{div} \approx \sqrt{s_{div}^2 - \langle\Delta\Lambda^2\rangle_{hemis}}$, where s_{div} is the SD of the values of the mean column spacing Λ for different animals and $\langle\Delta\Lambda^2\rangle_{hemis}$ is the squared error of Λ averaged over hemispheres from all animals. Its square root is $\sqrt{\langle\Delta\Lambda^2\rangle_{hemis}} = 0.022$ mm (by method 1). Analogously, the systematic intra-areal variability σ_{sys} of the local column spacing was calculated from the SD s_{sys} of values in the population map $\Lambda_{pop}(\mathbf{x})$ and their errors $\Delta\Lambda_{pop}(\mathbf{x})$ by $\sigma_{sys} \approx \sqrt{s_{sys}^2 - \langle\Delta\Lambda_{pop}(\mathbf{x})^2\rangle_{\mathbf{x}}}$. The square root of the spatially averaged squared error yielded $\sqrt{\langle\Delta\Lambda_{pop}(\mathbf{x})^2\rangle_{\mathbf{x}}} = 0.013$ mm. The total variability

TABLE 2. Accuracy of estimation

Parameter (P)	Range (mm)	s	ΔP (mm)	ΔP/s
Λ	1.03–1.27	0.053	0.018	34%
σ_Λ	0.10–0.27	0.028	0.012	42%
$\Lambda(\mathbf{x})$	0.75–1.65	0.138	0.060	44%
$\Lambda_{pop}(\mathbf{x})$	1.00–1.35	0.066	0.031	47%

Accuracy of estimation for the mean column spacing Λ, the spacing inhomogeneity σ_Λ, the local spacing $\Lambda(\mathbf{x})$ and the local population spacing $\Lambda_{pop}(\mathbf{x})$ using up to seven 2-DG autoradiographs obtained from flat-mount sections from various cortical depths. Values for Λ and σ_Λ are based on data from colony M and F, values for $\Lambda(\mathbf{x})$ and $\Lambda_{pop}(\mathbf{x})$ are based on the subset of data from colony F listed in Table 1. For all parameters, the table lists: Range, the total range of variation; s, the SD of the parameter values; ΔP, the averaged SEM of the parameter values; and ΔP/s, the relative error as a percentage of the SD.

σ_{tot}, i.e. the variability of ocular dominance column spacing across all animals, is given by $\sigma_{tot} = \sqrt{s^2_{tot} - \langle \Delta\Lambda(\mathbf{x})^2 \rangle_{tot}}$, where $\sqrt{\langle \Delta\Lambda(\mathbf{x})^2 \rangle_{tot}} = 0.087$ mm is the square root of the error of the local spacing squared and averaged over all locations in all hemispheres and s_{tot} is the SD of local spacing values $\Lambda(\mathbf{x})$ from all hemispheres. The total variability is through

$$\sigma^2_{tot} \approx \sigma^2_{div} + \sigma^2_{sys} + \sigma^2_{ran} \quad (9)$$

composed of the interindividual variability of the mean spacing, σ_{div}, the systematic intra-areal variability of the local spacing, σ_{sys}, and that part of intra-areal spacing variation, σ_{ran}, in addition to the systematic part. This decomposition provides an estimate for the relative magnitudes of the different contributions to the total variability of ocular dominance column spacing across the population.

Permutation tests

Because parameter values were not Gaussian-distributed, permutation tests were used to assess statistical significances of differences between groups of hemispheres (e.g. between colonies and between strabismic and normal animals), significances of correlations of parameter values, and significances of littermate clustering. The approach can be summarized as follows: to assess whether two populations exhibited significantly different parameter distributions, we calculated their averages and SDs and compared the differences of these averages and SDs to those obtained for populations composed of randomly chosen sets of animals from the combined population, called pseudo-populations. Statistically, there is a significant difference, if the hypothesis 'parameter values exhibit the same distribution for both populations' must be rejected, i.e. if the observed difference is too large, making it extremely unlikely to occur by chance. For a quantity Q (either population mean or population variance) with difference ΔQ between population 1 and 2, the P-value is given by

$$p = \langle \Theta(\Delta Q_p - \Delta Q) \rangle_p \quad (10)$$

where $\Delta Q_p = |\overline{Q}^1_{p(i)} - \overline{Q}^2_{p(i)}|$ is the difference between pseudo-population 1 and 2 generated by a suitable permutation p(i) of hemispheres, $\Theta(\cdot)$ is the Heaviside function and $\langle \cdot \rangle_p$ denotes the average over permutations. Similarly, to test significance of the correlation of parameter values in the left and right hemispheres, or of the correlation between parameters, actual correlations were compared to those gathered from randomly chosen assignments between hemispheres. For testing significance of littermate clustering, a permutation test was used according to Kaschube *et al.* (2002). In this test P-values were calculated using either arbitrary random permutations of hemispheres (P_{II}) or only such permutations which preserved left–right pairs of hemisphere indices (P_I). For all permutation tests, P-values were calculated using 10 000 randomly generated permutations of the data.

Results

Adapting a recently developed technique for the analysis of orientation column patterns (Kaschube *et al.*, 2000, 2002), we performed a detailed 2D quantification of ocular dominance column patterns in cat primary visual cortex (area 17). In particular, we focused on the local spacing of adjacent columns as a major layout property of the column patterns. Ocular dominance columns were labelled by either 2-[^{14}C]-deoxyglucose autoradiography (2-DG) after monocular stimulation of the animals (44 hemispheres from 28 animals; Löwel & Singer, 1993a,b; Löwel, 1994, 2002; Rathjen & Löwel, 2000; Rathjen *et al.*, 2002) or by [^{3}H]-proline autoradiography after injection of the labelled proline into one eye which labels the thalamocortical afferents of that eye in cortical layer IV (six hemispheres from four animals; Löwel & Singer, 1987; Löwel *et al.*, 1988; Löwel, 1994) (listed in Table 1). Double-labelling of ocular dominance columns with both techniques was performed in one animal (Löwel & Singer, 1993a,b). Figure 1 illustrates representative examples of ocular dominance column patterns in flat-mount sections (Freeman *et al.*, 1987; Löwel & Singer, 1987) of cat visual cortex. In all investigated hemispheres, and as described previously in a number of studies (Shatz *et al.*, 1977; Löwel & Singer, 1987; Anderson *et al.*, 1988; for a review see: Löwel, 2002), ocular dominance columns are arranged in complex repetitive patterns in a plane parallel to the cortical surface. However, columns differ considerably in their spacing in different regions of area 17 and in individual animals.

Maps of local column spacing

All results presented in this study are based on 2D maps of local spacing calculated from the patterns of ocular dominance columns observed in the analysed animals. These maps of local spacing $\Lambda(\mathbf{x})$ already revealed a strong variability in column spacing across area 17 and among different individuals, illustrated in Fig. 3a and b, showing the local column spacing $\Lambda(\mathbf{x})$ for the ocular dominance column maps depicted in Fig. 1. The maps demonstrate that local spacing values $\Lambda(\mathbf{x})$ vary considerably between different regions of area 17. For each map the histogram of values $\Lambda(\mathbf{x})$ within the map is depicted in Fig. 3c. To enable comparison, the six histograms are shown superimposed on each other. Five of the maps (the three upper maps, the lower left map and the lower middle map) exhibited column spacing values mostly between 0.9 and 1.5 mm. The most frequent value varied for different maps between 1.1 and 1.3 mm. The other map (the lower right map in Fig. 3) exhibited a column spacing which differed considerably. Values for this example ranged between 0.6 and 1.1 mm and the most frequent value was $\approx$0.8 mm. The degree of intra-areal and interindividual variability of spacing was similar in the remaining 43 hemispheres analysed. Typically, local spacing values varied >50% within an individual area 17 and up to 70% among different individuals.

Interindividual variability

To analyse the interindividual variability of ocular dominance columns, we assessed the interindividual variability of the mean column spacing Λ and the spacing inhomogeneity σ_Λ. Both parameters provided an appropriate characterization of the histograms of local column spacing in area 17 (Fig. 3c), because these typically exhibited only one pronounced peak of variable width and position. We observed that both the mean spacing Λ and the spacing inhomogeneity σ_Λ varied considerably between different animals. Figure 4 depicts the mean spacing Λ (Fig. 4a) and the spacing inhomogeneity σ_Λ (Fig. 4b) for all analysed hemispheres, namely the 2-DG maps of colonies M and F and the proline maps of colonies F and D. All values displayed in Fig. 4 were calculated using method 1 (i.e. by averaging over cortical locations before averaging over different brain sections; see Materials and methods). Over the entire dataset, mean spacings Λ varied between 0.73 and 1.27 mm with an average value of 1.12 mm. Spacing inhomogeneities σ_Λ varied between 0.10 and 0.27 mm with an average value of 0.18 mm. To quantify the interindividual variability of the mean spacing Λ, we calculated the SD s_{div} of mean column spacings Λ in different animals. We obtained an interindividual variability of $s_{div} = 0.108$ mm for all hemispheres of colonies M, F and D.

Inter-colony variability

Because our data were obtained from three different cat colonies, F, M and D, we asked whether spacing values were more similar in animals

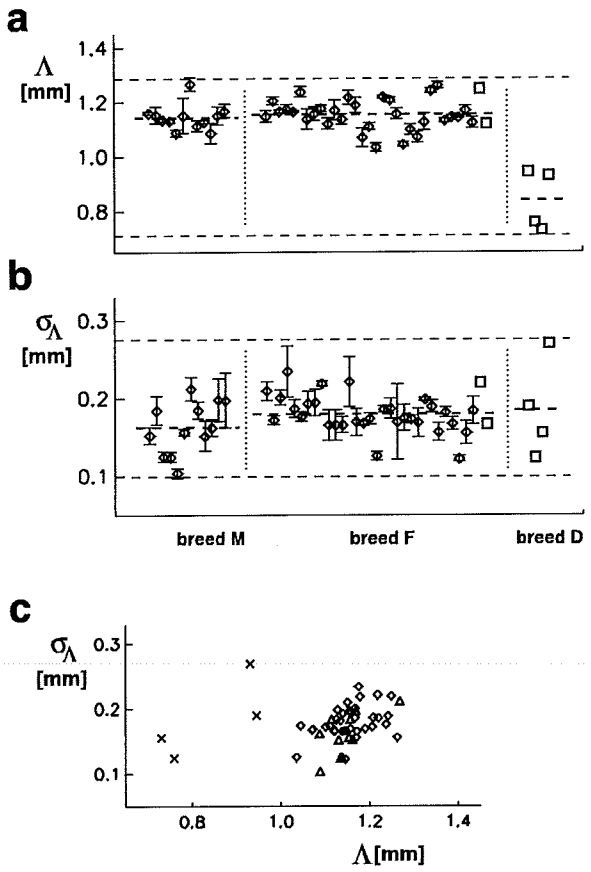

FIG. 4. (a and b) Interindividual variability and (c) relative independence of (a) mean column spacing Λ and (b) spacing inhomogeneity σ_Λ in 49 hemispheres from 31 animals. Mean spacing values from individual 2-DG-labelled hemispheres are indicated by diamonds arranged horizontally in (a) and (b). Values obtained from the six proline-labelled maps are indicated by boxes (displayed at the right). Error bars show SEM of the estimated parameter values (see Materials and methods; no error bars for cases where only a single section was available and for all proline reconstructions). The vertical dotted lines separate values from three different colonies of cats (left, colony M; middle, colony F; right, colony D; for further details see Materials and methods). The average value for each colony is marked by a horizontal dashed line. Note that both mean column spacing Λ and spacing inhomogeneity σ_Λ display significant interindividual and intercolony variability: mean column spacings Λ (a) varied between 0.73 and 1.27 mm, whereas for colony M and colony F they only ranged between 1.03 and 1.27 mm, and for colony D only between 0.73 and 0.95 mm. Differences in average values between colonies M and D and between colonies F and D are significant ($\Delta = 0.30$ mm, $P < 0.0001$; and $\Delta = 0.31$ mm, $P = 0.03$, respectively). Spacing inhomogeneities σ_Λ (b) varied between 0.10 and 0.27 mm in different animals. Average values did not significantly differ between different colonies ($P > 0.18$). (c) Scatter plot of the mean column spacing Λ and the spacing inhomogeneity σ_Λ. Values from individual hemispheres of colony M are indicated by triangles, values from colony F by diamonds and values from colony D by crosses. Parameters are moderately correlated (M, $r = 0.63$; F, $r = 0.38$) so that column spacings Λ and spacing inhomogeneities σ_Λ are not totally independent of each other; this explains a small fraction of the variability of σ_Λ (M, 39%; F, 14%).

from the same colony, i.e. in animals with a similar genetic background. Interestingly, single colonies indeed exhibited different average values of the mean spacing Λ and smaller interindividual variabilities s_{div}. Figure 4 shows that, in comparison to the total range of mean spacings Λ between 0.73 and 1.27 mm, values for both colonies M and F ranged solely between 1.03 and 1.27 mm, and for colony D they were confined between 0.73 and 0.95 mm. Thus, mean spacings Λ from colonies M and F were distributed without any overlap with the distribution of values from colony D. The average of mean spacings Λ from colony M was $\Lambda_M = 1.14$ mm which was very similar to the average value $\Lambda_F = 1.16$ mm of colony F, but both values deviated substantially from the average spacing observed in colony D of $\Lambda_D = 0.84$ mm. Thus, the average mean spacing Λ differed between colonies M and D by $d_{MD} = 0.30$ mm, and between colonies F and D by $d_{FD} = 0.31$ mm, a relative difference of >25%. Both differences were significant (d_{MD}, $P < 0.0001$; d_{FD}, $P < 0.03$; permutation test; see Materials and methods). Although the sample of colony D consisted of only two animals, these substantial differences clearly showed that column spacings in colony D statistically differed from those of colonies M and F. Indeed, the values observed in colony D were, compared to the distribution of spacings in colonies M and F, 6 × SD smaller. Thus, although these values did not provide a good estimate of the average column spacing in colony D, the data rejected the hypothesis that spacings from colony D are drawn from the same distribution as those of colonies M and F. We would like to stress that this conclusion holds in spite of the two animals from colony D being littermates. Furthermore, we would like to emphasize that littermate clustering does not explain the observed difference, because values from the litter of colony D simply do not occur in colonies M and F. The difference of $d_{FM} = 0.01$ mm between colony M and colony F, however, was not significant ($P = 0.51$). The spacing inhomogeneity σ_Λ did not differ in the three colonies. Values varied between 0.10 and 0.27 mm in each colony. Colony averages were $\sigma_\Lambda^M = 0.17$ mm, $\sigma_\Lambda^F = 0.18$ mm and $\sigma_\Lambda^D = 0.19$ mm. Differences between colonies were, with $d < 0.018$ mm, not significant ($P > 0.30$). Taken together, mean column spacings Λ and spacing inhomogeneities σ_Λ were both indistinguishable in colonies M and F, exhibiting distributions with almost identical average values and similar ranges, but for colony D mean spacing values Λ differed substantially. This indicates that spacing of ocular dominance columns can vary strongly between animals from different breeds. In order to obtain a statistically homogeneous subset of the data, we excluded colony D from subsequent quantitative analyses. For colonies M and F together, the error corrected (see Materials and methods) interindividual variability of the mean spacing Λ was $\sigma_{div} = 0.049$ mm.

Relationship between spacing parameters

To determine whether the mean spacing Λ and the spacing inhomogeneity σ_Λ measured independent aspects of the local column spacing map, we calculated their correlation. Correlations, depicted in Fig. 4c, were generally modest (method 1: individual colonies: M, $r = 0.63$; colony F, $r = 0.38$; colony M and F together, $r = 0.44$; method 2: $r = 0.45$, data not shown), so that column spacings and spacing inhomogeneities are not totally independent of each other. Λ explains a small fraction of the variability of σ_Λ (M, 39%; F, 14%; M + F, 20%; Method 2, 20%).

Influence of strabismus on spacing parameters

Because our data pool contained column patterns from both normal and strabismic animals, and because there is an ongoing debate as to whether strabismus influences column layouts (Goodhill, 1993; Löwel, 1994; Goodhill & Löwel, 1995; Jones *et al.*, 1996; Tieman & Tumosa, 1997; Sengpiel *et al.*, 1998; Wolf *et al.*, 2000; Rathjen *et al.*, 2002; Schmidt *et al.*, 2002), we checked whether some of the observed variability of spacing parameters was induced by strabismus. To this end, we compared the parameters in normally raised and strabismic animals from colonies M and F (Fig. 5). We found that the mean column spacings Λ did not exhibit any systematic trend towards an influence of strabismus: in colony M, averages of the mean spacing Λ for strabismic and control animals (calculated by method 1) were 1.14 and 1.16 mm, respectively. The small difference of $d = 0.018$ mm was

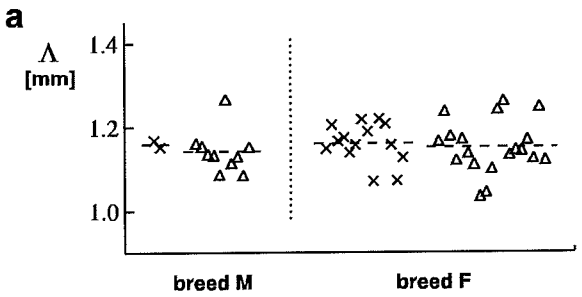

FIG. 5. The effect of strabismus on the mean spacing Λ of ocular dominance columns in cat area 17. Parameter values for animals from two different colonies (M, left; F, right) raised with different visual experience are compared. Values from individual hemispheres of normally raised cats are marked by crosses, arranged horizontally. Values from individual hemispheres of strabismic animals are represented by triangles. Average values are marked by the horizontal dashed lines. For both colonies M and F, the mean spacing Λ was not significantly different between squinting and normally raised animals, either the average value of Λ or the variability (SD) of Λ (colony M: average squinting, 1.14 mm, average control animals, 1.16 mm, *P*-value for difference, $P = 0.57$; colony F: average squinting, 1.15 mm, average control animals, 1.16 mm, $P = 0.68$; colonies F and M together: average squinting, 1.15 mm, average control, 1.16 mm, $P = 0.48$; for differences in SD, $P > 0.27$). Thus, no clear influence of strabismus on the mean column spacing Λ was observed.

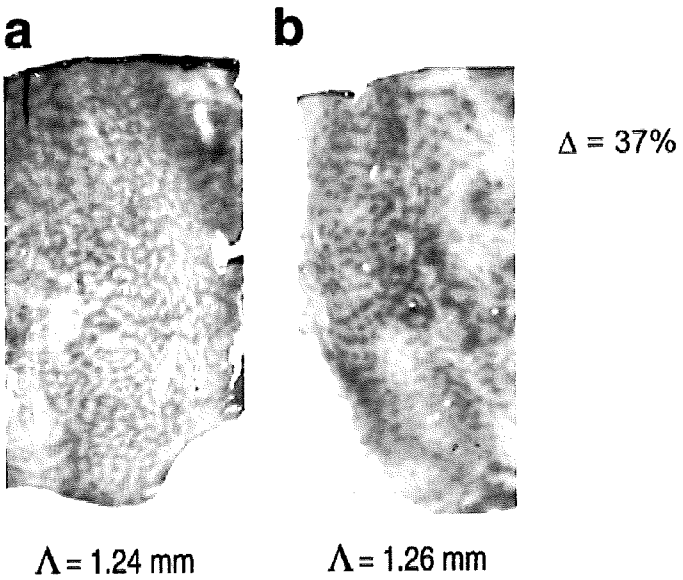

FIG. 6. Example of patterns of 2-DG-labelled ocular dominance columns in the two hemispheres of an individual animal. The patterns in both (a) the left and (b) the right hemisphere of cat F14 are displayed. Note that the spacing of adjacent ocular dominance columns looks rather similar in the two hemispheres ($\Delta = 37\%$). Note furthermore that the parameter mean column spacing Λ quantitatively captures this similarity: Λ in the left and right area 17 of cat F14 was 1.24 and 1.26 mm, respectively, corresponding to a difference relative to the SD of all values from colonies F and M of 37%. Thus, in the illustrated case, Λ differed by only 20 μm in the two hemispheres of an individual brain, although the mean column spacing may differ by up to 240 μm in hemispheres from different animals (of colonies M and F).

not significant ($P = 0.57$). In colony F, average mean spacings for strabismic and control animals were 1.15 and 1.16 mm, respectively. The difference of $d = 0.008$ mm was not significant ($P = 0.68$). For colonies M and F together, average values for strabismic and control animals were 1.15 and 1.16 mm, respectively. The difference of $d = 0.012$ mm was again not significant ($P = 0.48$). In addition to the average of the mean spacing Λ, the variability (SD) of the mean spacing was also indistinguishable for strabismic and control animals in all cases ($P > 0.27$). Further, in the case of the spacing inhomogeneity σ_Λ we did not observe any significant tendency for strabismic animals to exhibit values different from normal animals. *P*-values were generally >0.3. We conclude that our data did not indicate that strabismic and normal animals systematically differ in column spacing, thereby confirming a recent report by Rathjen *et al.* (2002).

Column spacing in the left and right hemisphere

Because the pronounced difference in column spacing found between animals from different colonies suggests a substantial influence of genetic background, we asked whether ocular dominance columns exhibit a similar spacing in the two hemispheres of an individual brain. Because the two hemispheres of a brain are genetically identical, genetically controlled features of visual cortical architecture are expected to be similar in the two brain hemispheres. An example of a left and right ocular dominance pattern is presented in Fig. 6 (animal F14). The mean spacing Λ differs in this example by 0.020 mm, 37% of the SD s_{div} of all values from colonies F and M. This example is representative for a general trend: the mean spacing Λ indeed differed less between the left and right hemisphere in the 16 complete brains from our dataset of colonies M and F than between two randomly chosen pairs of hemispheres. This is demonstrated in Fig. 7a displaying the mean spacing Λ for the left hemispheres vs. that for the right hemispheres (calculated by method 1). The difference between the two hemispheres was on average 0.043 mm and the correlation between the two hemispheres was significant with $r = 0.52$ ($P = 0.02$, permutation test). The results obtained by method 2 ($r = 0.83$, $P = 0.04$) support this finding (data not shown). In contrast, the spacing inhomogeneity σ_Λ did not exhibit a reduced variability within an individual brain. Figure 7b depicts the values of the spacing inhomogeneity σ_Λ for the left hemisphere vs. the values for the right hemisphere. The correlation between the two hemispheres was only weak and not significant ($r = 0.05$, $P = 0.42$). The correlation obtained by method 2 confirmed this picture ($r = 0.40$, $P = 0.29$). Neither of the spacing parameters showed any tendency towards lateralization, i.e. systematic differences between the left and right hemisphere.

Because the two brain hemispheres were correlated in their mean spacing Λ but not in their spacing inhomogeneity σ_Λ it is natural to ask whether the local spacing $\Lambda(\mathbf{x})$ is similar in two brain hemispheres, i.e. whether columns at corresponding cortical locations in the left and right hemispheres exhibit a similar spacing (locations with receptive field positions of similar altitude and opposed azimuth in the field of view). Therefore, we mirror-inverted the left area 17 and aligned the representations of the vertical and horizontal meridian of both areas 17. We then calculated the correlation of the local column spacing values $\Lambda(\mathbf{x})$ for all pairs of hemispheres: an especially convenient measurement for our purpose because it is unaffected by the mean column spacing Λ. The correlation coefficients for the real brains were then compared to the correlations obtained from pseudo brains consisting of two randomly assigned hemispheres. Figure 7c displays a histogram of correlation coefficients which were computed from all possible pseudo brains (solid line). Using the suitable subset of 18 hemispheres (listed in Table 1) we gathered a total of $18 \times (18-1)/2 = 153$ coefficients. The histogram is centred around 0.04. The coefficients for the real brains are marked by the boxes and crosses. Typically, the coefficients for the real brains were not larger than those for the pseudo brains, as would be expected if local column spacing values $\Lambda(\mathbf{x})$ were correlated in the left and right hemisphere. In fact, the real coefficients were spread around zero correlation with an average of −0.1 indicating that a real pair is not more strongly correlated than two arbitrary hemispheres. Thus, we observed no evidence for a local similarity of spacing of ocular

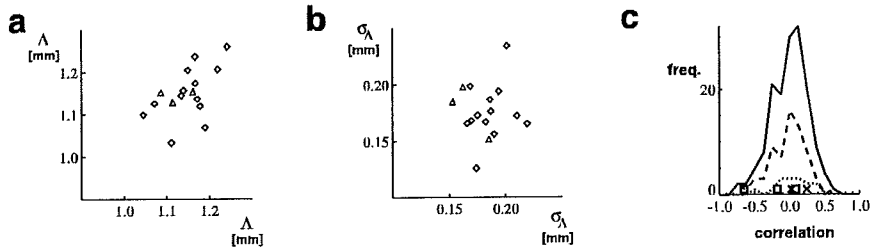

FIG. 7. Spacing of ocular dominance columns in left and right brain hemispheres. (a) Mean column spacing Λ for left hemispheres (abscissa) vs. mean spacing Λ for right hemispheres (ordinate) of all complete hemisphere pairs. Values from individual pairs of colony M are indicated by triangles, values from pairs of colony F by diamonds. Values were significantly correlated in the two hemispheres (for M and F together, $r=0.52$, $P=0.02$). (b) Spacing inhomogeneity σ_Λ for all complete hemisphere pairs. Values were not significantly correlated (for M and F together, $r=0.05$, $P=0.42$). (c) Correlation of the local column spacing $\Lambda(\mathbf{x})$ in left and right hemispheres. Correlation coefficients for five hemisphere pairs (see text) are marked by boxes (strabismic animals) and crosses (normal animals) along the ordinate. Their average correlation was −0.10. For significance estimation of these correlations, histograms of coefficients obtained from randomly assigned pseudo hemisphere pairs are depicted in the same graph. The solid histogram includes all available pseudo pairs (average correlation, 0.04), the dashed histogram includes all pairs of hemispheres from two strabismic animals (average, 0.01), and the dotted histogram includes all pairs from two normal animals (average, 0.07). Note that the correlation for a real hemisphere pair is typically not larger than for a pseudo brain. Note further that for both the real brains and the pseudo brains the correlation was larger for the normal cases than for the strabismic cases, though this tendency was not significant.

dominance columns at corresponding retinotopic locations in left and right brain hemispheres.

Column spacing in littermates

A genetic influence on the mean column spacing Λ is suggested from both the similarity of mean column spacing Λ in the left and right brain hemisphere (Fig. 7) and its statistical difference in individual colonies (Fig. 4). Moreover, various features of orientation columns in cat area 17, including the mean spacing, are genetically determined to a substantial extent (Kaschube *et al.*, 2002). Using an approach analogous to our previous study, we here tested whether mean column spacing Λ of ocular dominance columns was also more similar in littermates than in unrelated animals. Our data included nine litters whereby each litter consisted of two animals (see Table 1; in order to use a statistically homogeneous sample, L10 has been excluded from this analysis). Complete hemisphere pairs were available in four litters. Two litters consisted each of one animal with a complete hemisphere pair and one animal with only one hemisphere. In three litters each animal contributed only one hemisphere. An example of two ocular dominance patterns from a litter is presented in Fig. 8 (hemisphere F1 right and F2 left). The mean column spacing Λ differed in this example by 0.04 mm which was 75% of the SD s_{div} of mean spacings Λ from colonies M and F. In general, differences in the mean spacing Λ in littermates and in unrelated animals were comparable. For method 1 the average interindividual variability within litters was $s_{lit}=0.047$ mm (compared to $s_{div}=0.051$ mm for all hemispheres of colonies M and F), and values were not clustered in littermates ($P_{II}=0.69$, $P_I=0.83$, permutation test; see Materials and methods). Results based on method 2 were in line with this finding ($P_{II}=0.18$, $P_I=0.33$). For the spacing inhomogeneity σ_Λ similar results were obtained. For both methods 1 and 2 the spacing inhomogeneity was far from being significantly clustered in littermates (method 1, $P_{II}=0.60$, $P_I=0.92$; method 2, $P_{II}=0.60$, $P_I=0.83$). Furthermore, the spacing was not correlated locally in littermates. Correlation coefficients (r) for hemisphere pairs from littermates were on average $r=0.05$, which was not significantly different from the average correlation between two arbitrary spacing maps of $r=0.04$ (see Fig. 7). Taken together, we observed no evidence for any enhanced similarity of spacing of ocular dominance columns in littermates.

Systematic intra-areal variability

So far we arrived at a picture of ocular dominance column spacing to exhibit a substantial variability across area 17 and among different individuals. It is conceivable that the strong intra-areal variability common to all hemispheres reflects a systematic trend across area 17, e.g. columns in a particular visual field representation might exhibit a larger (or smaller) than average spacing and therefore spacings might not vary independently in different hemispheres. To distinguish the systematic part of the intra-areal variability from the nonsystematic, apparently random, part, we calculated the population spacing map $\Lambda_{pop}(\mathbf{x})$, a 2D map of local column spacing averaged over the population. To this end, individual maps of local column spacing were superimposed so that the representations of the vertical and horizontal meridian were aligned and spacings were averaged over the population at each cortical location (for details see Materials and methods). Figure 9 shows the population spacing map $\Lambda_{pop}(\mathbf{x})$ which demonstrates a considerable amount of systematic intracortical variation in column spacing. The map is grey-scale-coded as the maps of local spacing $\Lambda(\mathbf{x})$ in Fig. 3. The values of $\Lambda_{pop}(\mathbf{x})$ varied considerably in different cortical regions. Along the representation of the horizontal meridian, values were larger than the mean value of $\langle\Lambda_{pop}=\rangle\Lambda_{pop}(\mathbf{x})\rangle_x=1.16$ mm. Regions along the representation of the peripheral part of the vertical meridian exhibited a smaller than average spacing. To illustrate the variation across different cortical regions, values drawn along the representation of the horizontal and the anterior part of the vertical meridian are depicted in Figs 9b and c, respectively. The histogram of the population map $\Lambda_{pop}(\mathbf{x})$ in Fig. 9d illustrates the magnitude of systematic intra-areal variability. Values typically ranged between 1.00 and 1.35 mm. From the SD of $s_{sys}=0.066$ mm we obtained (by error correction; see Materials and methods) a systematic variability of $\sigma_{sys}=0.056$ mm. Correlations between the population spacing map $\Lambda_{pop}(\mathbf{x})$ and spacing maps from individual hemispheres were modest. The correlation ranged between $r=-0.18$ for the left hemisphere of animal F17 and $r=0.77$ for the left hemisphere of animal F5, and exhibited an average value of $r=0.39$. We conclude that a considerable part, though certainly not all, of the intra-areal variability of ocular dominance column spacing is explained by its systematic variation across the cortex.

The different components of the total variability

To identify the prevalent factor for the substantial variability of spacing of ocular dominance columns throughout the population (colony F), we decomposed the total variability, i.e. the variability of the local spacing $\Lambda(\mathbf{x})$ in all animals, into three mutually independent parts: (i) the interindividual variability σ_{div} of mean column spacings Λ; (ii) the

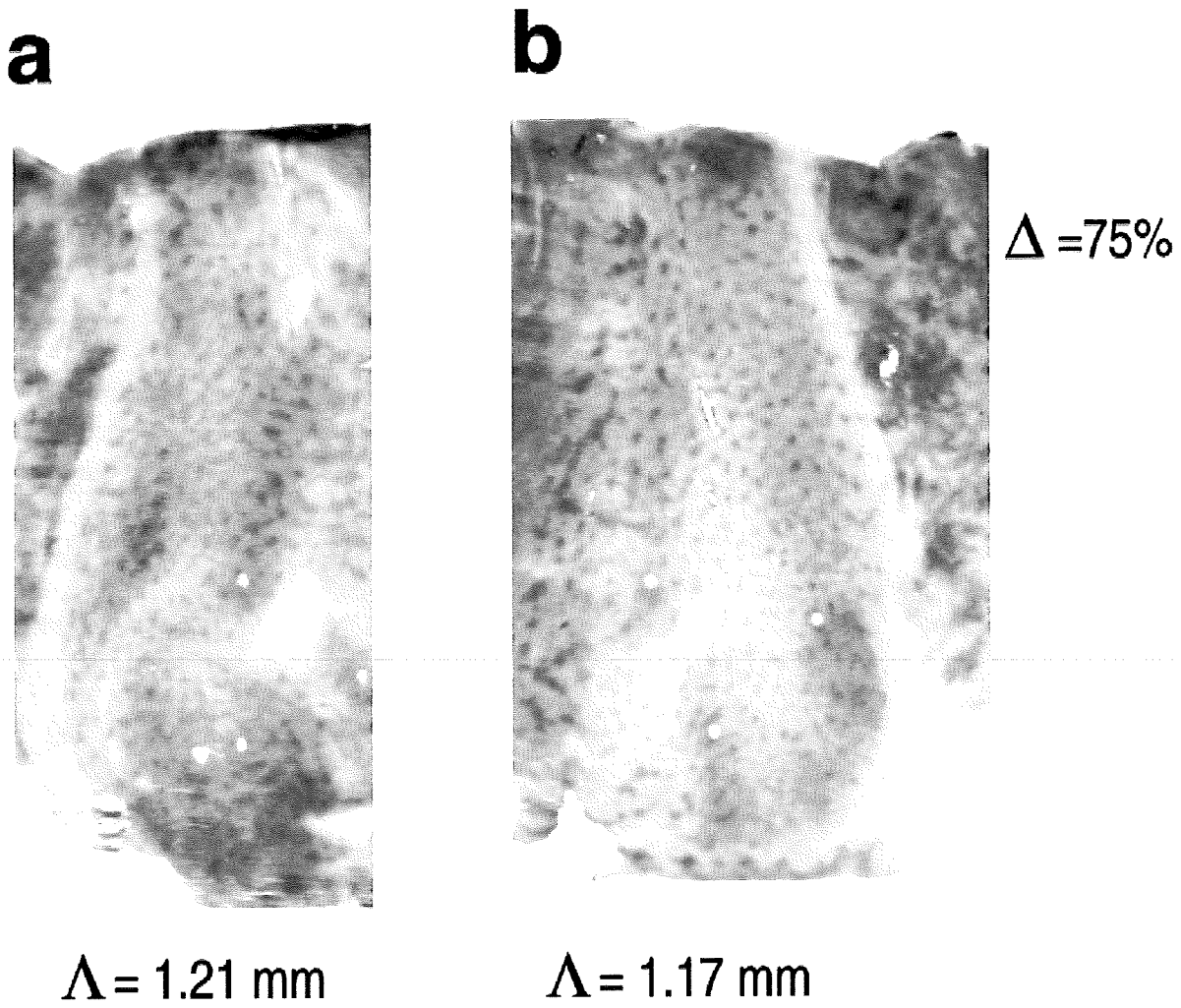

FIG. 8. Example of patterns of 2-DG-labelled ocular dominance columns in littermates. The ocular dominance column patterns from (a) the right hemisphere of cat F1 and (b) the left hemisphere of cat F2 (both from litter L4) are displayed. Note that compared to Fig. 6 the spacing of adjacent ocular dominance columns is less similar. Note furthermore that the parameter mean column spacing Λ quantitatively captures this relative dissimilarity: mean column spacing in the right area 17 of cat F1 was 1.21 mm and, in the left area 17 of cat F2 1.17 mm, a difference of 75% of the SD of all values from colonies M and F. In contrast to pairs of left and right hemispheres from individual animals (compare Figs 6 and 7) the variability of parameter values in littermates was indistinguishable from the variability observed in unrelated animals from the same colony.

systematic intra-areal variation σ_{sys} expressed by $\Lambda_{pop}(\mathbf{x})$; and (iii) an apparently random nonsystematic intra-areal variability σ_{ran} of local column spacing $\Lambda(\mathbf{x})$ (see Materials and methods). The relative magnitude of these different variability components is illustrated graphically in Fig. 10 showing the histogram of all local column spacing values $\Lambda(\mathbf{x})$ in the population (solid line), the histogram of local column spacings from a 'typical' single hemisphere (dotted line) and the histogram of the population spacing map $\Lambda_{pop}(\mathbf{x})$ (already displayed in Fig. 9). The interindividual variability of the mean column spacing contributes relatively little to the total variability because the histogram of all column spacings was only slightly wider than that of a 'typical' hemisphere. The SD of all column spacings was s_{tot} 0.14 mm and the SD for the typical hemisphere was $s_{typ} = 0.12$ mm. Thus the variability of column spacing in a single hemisphere was comparable to the variability observed in the entire population. The width of the histogram of the population spacing map $\Lambda_{pop}(\mathbf{x})$ suggests the systematic part of the intra-areal variability σ_{sys} to account partly for the total variability. The large gap to the histogram of the 'typical' hemisphere, however, indicates a dominance of the nonsystematic ('random') part of intra-areal variability. Quantitatively, the total variability, after error correction (see Materials and methods), yielded $\sigma_{tot} = 0.114$ mm. The interindividual variability of the mean column spacing Λ was $\sigma_{div} = 0.049$ mm and the systematic intra-areal variability of the local column spacing $\Lambda(\mathbf{x})$ was $\sigma_{sys} = 0.056$ mm. The nonsystematic variability resulted in $\sigma_{ran} = \sqrt{\sigma_{tot}^2 - \sigma_{div}^2 - \sigma_{sys}^2} = 0.086$ mm. Thus, the total variability of ocular dominance spacing comprised 18% interindividual variability of the mean column spacing, 24% systematic intra-areal variability, and 58% nonsystematic intra-areal variability. Hence, in a given hemisphere, the intra-areal variability consisted of 29% systematic and 71% nonsystematic variation. We conclude that the largest fraction of variability in ocular dominance column spacing in cat area 17 is attributable neither to systematic intracortical variation nor to interindividual differences in the mean spacing. The apparently random part of the intra-areal variability is dominating the total variability of ocular dominance spacing.

Discussion

Our study presents the first locally resolved high accuracy quantification of ocular dominance column spacing in primary visual cortex

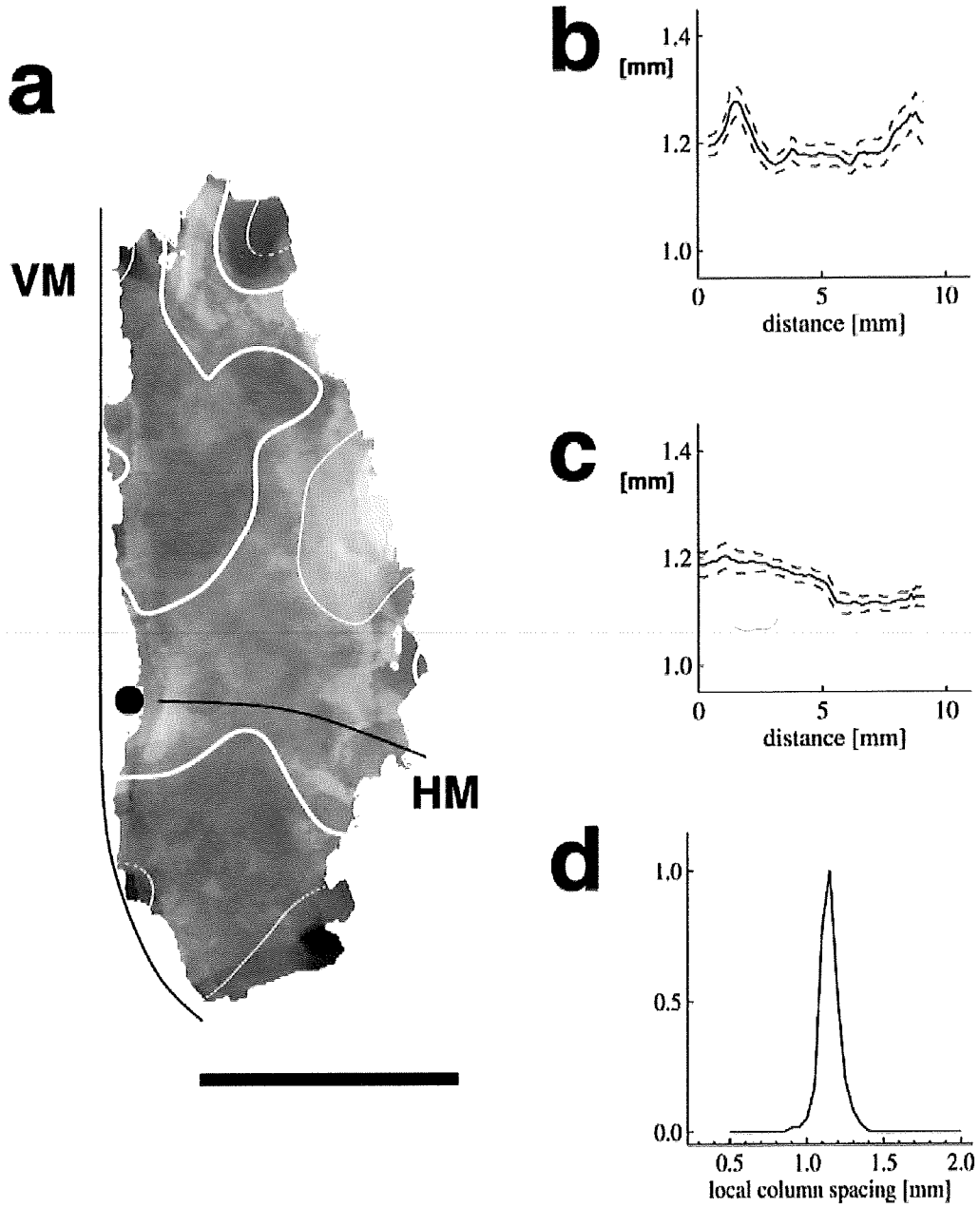

FIG. 9. The systematic intra-areal variation of ocular dominance spacing. (a) Population spacing map $\Lambda_{pop}(\mathbf{x})$, a superposition and average of all maps of local spacing $\Lambda(\mathbf{x})$ such that the representations of the horizontal and vertical meridian (HM and VM, respectively) were aligned in each map (see Materials and methods). The map is coded in grey scale: light grey regions exhibit larger-than-average spacing, dark grey regions exhibit smaller-than-average spacing. The black circle marks the central visual field representation. The black lines delineate the HM and VM. The white contour lines in the spacing map indicate levels of spacing: the thick white line indicates positions with the average value Λ_{pop} of the population spacing map $\Lambda_{pop}(\mathbf{x})$; the thinner white lines indicate positions with average value $\pm$SD of $\Lambda_{pop}(\mathbf{x})$. The contour lines were calculated from a smoothed version of the map. Anterior is at the top, posterior is at the bottom of the figure. Scale bar, 10 mm. (b) Values of the population map $\Lambda_{pop}(\mathbf{x})$ along the HM from the central representation towards the peripheral representation. Dashed lines indicate $\Lambda_{pop}(\mathbf{x}) \pm$ SEM for each location. Values are smoothed for illustration. (c) Values along the VM from the central representation towards the peripheral representation in the anterior region of area 17. (d) Histogram of spacings in the population map $\Lambda_{pop}(\mathbf{x})$. Values range between 1.00 and 1.35 mm. The mean value is $\Lambda_{pop} = 1.16$ mm. Note that there is a general tendency for ocular dominance columns to exhibit a larger-than-average spacing along the representation of the horizontal meridian, and smaller-than-average spacing along the representation of the peripheral part of the vertical meridian. The magnitude of this systematic variation is expressed by the SD of the population map $\Lambda_{pop}(\mathbf{x})$, $s_{sys} = 0.066$ mm.

(area 17) of a large number of animals. The patterns were obtained from cats raised with different visual experience (normal and strabismic) and from different genetic backgrounds (different cat colonies). In all animals, column spacings exhibited a pronounced variability across area 17. A fraction of the intra-areal variability (29%) is explained by systematic variation of column spacing across area 17: the column spacing was generally larger than average along the cortical representation of the horizontal meridian and smaller along the peripheral representations of the vertical meridian.The major part of variability in each hemisphere (71%) was an apparently random variation in column spacing across cortex. The mean column spacing in area 17 varied in different animals from cat colonies M and F between 1.03 and 1.27 mm, which was considerably smaller than the intra-areal variability of spacings (22% of the intra-areal variability).

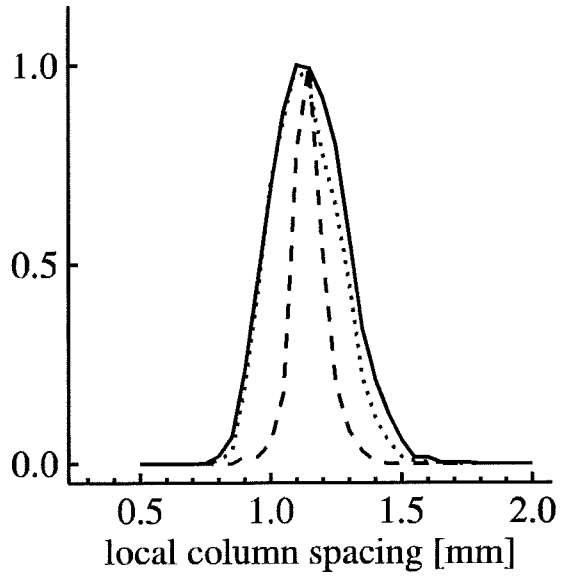

FIG. 10. The different components of the total variability of ocular dominance column spacing. Depicted are the histograms of three distributions (bin size, 0.05 mm): The solid line is the histogram of local spacing values $\Lambda(\mathbf{x})$ from all hemispheres expressing the total variability of column spacing. The distribution is centred around 1.16 mm and exhibits a variability (SD) of $s_{tot} = 0.138$ mm. The dotted line is the histogram of a 'typical' individual hemisphere. It was calculated by averaging the histograms of local column spacing from individual hemispheres, after normalizing each one to yield a mean value equal to the hemisphere average of mean spacings Λ of 1.15 mm, and to yield a SD of $s_{typ} = 0.12$ mm, equal to the average of all spacing inhomogeneities σ_Λ (calculated by method 2; see Materials and methods). The dashed line is the histogram of the population spacing map $\Lambda_{pop}(\mathbf{x})$ shown in Fig. 9 and represents the systematic variation of column spacing across area 17. It is centred around 1.16 mm and has a variability of $s_{sys} = 0.066$ mm. All histograms were calculated from the subset of data listed in Table 1 (see Materials and methods). All histograms were normalized to a maximum value equal to 1. Note that the total variability is surprisingly similar to the variability in a 'typical' map, indicating the prevalence of intra-areal variability over the interindividual variability of the mean spacing of ocular dominance columns. Note further that a considerable fraction of intra-areal variability is systematic such that column spacing does not vary independently in different hemispheres. The large gap between the histogram of the population map and the histogram of the 'typical' hemisphere, however, demonstrates a dominance of intra-areal variability which is non-systematic and different for each individual.

On average, the mean column spacing was 1.15 mm which was quite similar to the average of 1.17 mm obtained previously for the spacing of orientation columns (Kaschube *et al.*, 2002). On the other hand, the interindividual variability of the mean column spacing was only 60% of that observed for orientation columns (Kaschube *et al.*, 2002). In contrast to our previous results on orientation columns, a clustering of mean column spacings among littermates is not supported by our data. Nevertheless, a dependence of ocular dominance column spacing on the genetic background of the animals is suggested by the substantial difference in column spacing observed in cats from colony D (where mean column spacings ranged between 0.73 and 0.95 mm) and by the similarity of column spacings in the left and right hemispheres of individual brains. Strabismus did not, however, affect column spacings.

Previous attempts to characterize the spacing of ocular dominance columns (Obermayer & Blasdel, 1993; Löwel, 1994; Rathjen *et al.*, 2002; Schmidt *et al.*, 2002) were based on nonlocal image analysis techniques and were therefore restricted to an estimation of the mean spacing in the ocular dominance pattern in a particular hemisphere. In contrast, the use of localized filters (wavelets) in the present study enabled us for the first time to analyse the local spacing of ocular dominance columns in different regions of area 17. We estimated column spacing locally with an average error of 31 μm by using multiple autoradiographs obtained from different sections of visual cortical flat-mounts. Two spacing parameters were used to quantitatively characterize column spacing in area 17: mean column spacing Λ and spacing inhomogeneity σ_Λ, both estimated with an error of <20 μm (relative errors of <2 and 10%, respectively). Error measurements were similar to those obtained from the analyses of orientation columns in cat primary visual cortex using similar techniques (Kaschube *et al.*, 2002). All errors were small compared to the variabilities of spacing parameters.

Our results partially agree with observations in macaque primary visual cortex. Horton & Hocking (1996; see also LeVay *et al.*, 1985) have analysed the variability of spacing of ocular dominance columns in the primary visual cortex of six macaque monkeys. They reported that the average column spacing along the representation of the vertical meridian varies up to two-fold between different animals and that columns generally exhibited the largest spacing in the foveal representation. In the periphery, columns were wider along the vertical than along the horizontal meridian for a given eccentricity. These findings partially agree with our observations in cat primary visual cortex. Within a cat colony, mean column spacings varied by ≈30% between different animals. Among the largest column spacings were generally those observed in the central representation (Fig. 9). In contrast to the observations in macaque monkeys, however, with greater eccentricity, column spacings tended to be smaller along the vertical but not along the horizontal meridian.

Besides this systematic variation of ocular dominance column spacing within area 17, our study reveals a predominant part of intra-areal variability which cannot be explained by systematic variation. Neither was this component affected by the drastic change in visual input induced by strabismus nor did it exhibit any correlation in related animals or in the left and right brain hemispheres. This variablity in spacing might be caused by individually different patterns of activity during development (Weliky & Katz, 1999) or by constraints from other cortical maps such as retinotopy or orientation selectivity. Random influences through either of these mechanisms might explain the observed variability. However, a very sensitive dependence of ocular dominance column spacing on genetic factors would also be consistent with our findings. Genetic factors might directly control the termination patterns of axons from the eye-specific layers of the LGN, or determine cellular parameters such as the width of dendritic or axonal arborizations in the cortex (Gierer, 1988; Miller, 1995; Swindale, 1996; for a detailed discussion, see Kaschube *et al.*, 2002).

Data consistent with an influence of strabismus on the spacing of ocular dominance columns were reported originally by Löwel (1994) (see also Tieman & Tumosa, 1997). However, other studies (Jones *et al.*, 1996; Sengpiel *et al.*, 1998) and in particular the recent study by Rathjen *et al.* (2002) comparing littermates raised with and without strabismus did not support the original interpretation noting that it was difficult to disentangle the influence of litter membership and visual experience in the previous study (Löwel, 1994). The present study, which is based on a very large dataset (including the data of Löwel, 1994) and which uses a refined method for assessing the spacing of columns, confirms and extends the recent interpretation by revealing no indication for a change in column spacing induced by strabismus: for a given cat colony, both mean column spacing Λ and spacing inhomogeneity σ_Λ did not differ significantly between strabismic and normally raised animals. Also locally, the spacing of columns did not

show an influence of squint, though our sample was rather limited in this analysis. While strabismic animals exhibited a tendency towards larger column spacings in the central representation of area 17, this tendency was not statistically significant (data not shown). Because spacing values can differ substantially between different colonies, pooling of values from different colonies can lead to a nonhomogeneous sample resulting in improper significance values. The sample of hemispheres used in Löwel (1994) was relatively small and included the normally raised animals from colony D with their substantially smaller column spacing. This substantially increased the apparent difference between normal and strabismic animals.

Recent studies show that a pattern of ocular dominance columns is present functionally (Crair *et al.*, 1998; Rathjen & Löwel, 2000) and anatomically (Crair *et al.*, 2001; see also Crowley & Katz, 2000, 2002) at the time of eye opening in the second postnatal week. Theoretical considerations suggest that squint which is induced after ocular dominance columns have formed, as might have been the case in the study of Löwel (1994), would not result in a change in column spacing (Wolf *et al.*, 2000). In a framework of activity-dependent Hebbian mechanisms with dynamic regulation of synaptic connections, column spacing is shown to depend on the correlation of activity patterns in the two eyes before the segregation of ocular dominance columns. If the initial formation of ocular dominance columns happens before eye opening, the alternation of the correlation of retinal activity patterns induced by squint should have no effect on column spacing.

While the present analyses thus clarify that there is no significant difference in the spacing of adjacent ocular dominance columns in strabismic and normally raised cats, they additionally suggest an influence of genetic background on column spacing. Our analyses of column spacing in different cat colonies showed that column spacings in colony D significantly and strongly differed from those in colonies F and M. Furthermore, the observed correlation of the mean column spacing Λ in the left and right hemisphere of individual animals is also consistent with a genetic influence on column spacing. The absence of an enhanced similarity in column spacing between littermates does not necessarily contradict this conclusion. In fact, it is conceivable that the genetic makeup of an individual influences its column spacing such that even small genetic differences might cause large differences in spacings. In this case, even the reduced difference of genes in littermates might be sufficient to cause a variability in spacing almost equal to that within a colony. An alternative and certainly more parsimonious interpretation would be that genetic influences might be relatively weak, such that ocular dominance columns would be strongly affected by random events (see, e.g. Swindale, 1996; Wolf & Geisel, 1998; 2003) and therefore exhibit an equal amount of variability among littermates and among unrelated animals, as indicated by our data. In contrast to the present observations, a substantial clustering of mean column spacing was observed for the system of orientation columns in cat primary visual cortex using comparable methods and an equally large sample of 2-DG-labelled hemispheres (Kaschube *et al.*, 2002). However, the intrinsic interindividual variability of the mean spacing of orientation columns was almost 70% larger than that of ocular dominance columns. Thus, the intrinsic 'clustering' of spacing values might make it more difficult to detect littermate clustering for ocular dominance column spacing.

The substantial variability in ocular dominance column spacings in cat primary visual cortex revealed by our study raises the question of the reason for this variability. Local column spacing was found to vary by an almost equal extent in a given brain hemisphere as in an entire colony of cats and, despite the fact that the hemisphere mean of column spacings was relatively similar in different individuals of a colony, locally their column spacing did not covary. A functional significance for this substantial variability within and among individual hemispheres remains unclear at present. Sensory abilities such as stereopsis and acuity have repeatedly been linked to the processing of binocular responses (Daw, 1995) and it is conceivable that differences in column spacing reflect variable processing abilities (compare Chklovskii, 2000). These abilities are expected to vary among different individuals and depending on the eccentricity in the visual field. On the other hand, the substantial variability observed in the present study is also consistent with the hypothesis that ocular dominance columns are a functionally less relevant by-product of the development of other cortical features and thus susceptible to various incoherent influences (Adams & Horton, 2003). Further investigations which functionally compare visual performance and the layout of ocular dominance columns in individuals might help to clarify these issues. Our detailed characterization of the variability of ocular dominance spacing provides a necessary prerequisite for identifying covariances of visual performance and column layout.

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Abbreviations

2D, two-dimensional; 2-DG, 2-[^{14}C]-deoxyglucose; HM, horizontal meridian; Λ, mean column spacing; r, correlation coefficient; s, SD; σ, variability; σ_Λ, spacing inhomogeneity; VM, vertical meridian.

References

Adams, D.L. & Horton, J.C. (2003) Capricious expression of cortical columns in the primate brain. *Nature Neurosci.*, **6**, 113–114.

Anderson, P.A., Olavaria, J. & Van~Sluyters, R.C. (1988) The overall pattern of ocular dominance bands in cat visual cortex. *J. Neurosci.*, **8**, 2183–2200.

Chklovskii, D.B. (2000) Binocular disparity can explain the orientation of ocular dominance stripes in primate primary visual area (V1). *Vision Res.*, **40**, 1765–1773.

Crair, M.C., Gillespie, D.C. & Stryker, M.P. (1998) The role of visual experience in the development of columns in cat visual cortex. *Science*, **279**, 566–570.

Crair, M.C., Horton, J.C., Antonini, A. & Stryker, M.P. (2001) Emergence of ocular dominance columns in cat visual cortex by 2 weeks of age. *J. Comp. Neurol.*, **430**, 235–249.

Crowley, J.C. & Katz, L.C. (2000) Early emergence of ocular dominance columns. *Science*, **290**, 1321–1324.

Crowley, J.C. & Katz, L.C. (2002) Ocular dominance development revisited. *Curr. Opin. Neurobiol.*, **12**, 104–109.

Daw, N.W. (1995) *Visual Development*. Plenum Press, New York.

Freeman, B., Löwel, S. & Singer, W. (1987) Deoxyglucose mapping in the cat visual cortex following carotid artery injection and cortical flat-mounting. *J. Neurosci. Meth.*, **20**, 115–129.

Gierer, A. (1988) Spatial organization and genetic information in brain development. *Biol. Cybern.*, **59**, 13–32.

Goodhill, G.J. (1993) Topography and ocular dominance: a model exploring positive correlations. *Biol. Cybern.*, **69**, 109–118.

Goodhill, G.J. & Löwel, S. (1995) Theory meets experiment: correlated neural activity helps determine ocular dominance column periodicity. *Trends Neurosci.*, **18**, 437–439.

Grafstein, B. (1971) Transneuronal transfer of radioactivity in the central nervous system. *Science*, **172**, 177–179.

Hensch, T.K. & Stryker, M.P. (1996) Intracortical interactions regulate ocular dominance column segregation. *Soc. Neurosci. Abstr.*, **22**, 682.15.

Horton, J.C. & Hocking, D.R. (1996) Intrinsic variability of ocular dominance column periodicity in normal macaque monkeys. *J. Neurosci.*, **16**, 7228–7239.

Jones, D.G., Murphy, K.M. & Van Sluyters, R.C. (1996) Spacing of ocular dominace columns is not changed by monocular deprivation or strabismus. *Invest. Ophthalmol. Vis. Sci. Suppl.*, **37**, 1964.

Kaschube, M., Wolf, F., Geisel, T. & Löwel, S. (2000) Quantifying the variability of patterns of orientation columns in the visual cortex of cats. *Neurocomputing*, **32–33**, 415–423.

Kaschube, M., Wolf, F., Geisel, T. & Löwel, S. (2002) Genetic influence on quantitative features of neocortical architecture. *J. Neurosci.*, **22**, 7206–7217.

Kaschube, M., Wolf, F., Rathjen, S., Geisel, T. & Löwel, S. (2001a) 2-D analysis of ocular dominance patterns in cat primary visual cortex. *Soc. Neurosci. Abstr.*, **27**, 821.26.

Kaschube, M., Wolf, F., Geisel, T., Rathjen, S. & Löwel, S. (2001b) 2-D analysis of patterns of ocular dominance columns in cat primary visual cortex. In Elsner, N. & Kreutzberg, G.W. (eds), *Proceedings of the 4th Meeting of the German Neuroscience Society 2001*, Vol. I. {28th Göttingen Neurobiology Report 2001}. Thieme Verlag, Stuttgart, p. 546.

LeVay, S., Connolly, M., Houde, J. & Van Essen, D.C. (1985) The complete pattern of ocular dominance stripes in the striate cortex and visual field of the macaque monkey. *J. Neurosci.*, **5**, 486–501.

LeVay, S. & Nelson, S.B. (1991) Columnar organization of the visual cortex. In Cronly-Dillon, J.R. (ed.), *Vision and Visual Dysfunction.* Macmillan, Houndmill, pp. 266–315.

Löwel, S. (1994) Ocular dominance column development: Strabismus changes the spacing of adjacent columns in cat visual cortex. *J. Neurosci.*, **14**, 7451–7468.

Löwel, S. (2002) 2-Deoxyglucose architecture of cat primary visual cortex. In Payne, B. & Peters, A. (eds), *The Cat Primary Visual Cortex.* Academic Press, San Diego, pp. 167–193.

Löwel, S., Bischof, H.-J., Leutenecker, B. & Singer, W. (1988) Topographic relations between ocular dominance and orientation columns in the cat striate cortex. *Exp. Brain Res.*, **71**, 33–46.

Löwel, S., Freeman, B. & Singer, W. (1987) Topographic organization of the orientation column system in large flat-mounts of the cat visual cortex: a 2-deoxyglucose study. *J. Comp. Neurol.*, **255**, 401–415.

Löwel, S. & Singer, W. (1987) The pattern of ocular dominance columns in flat-mounts of the cat visual cortex. *Exp. Brain Res.*, **68**, 661–666.

Löwel, S. & Singer, W. (1992) Selection of intrinsic horizontal connections in the visual cortex by correlated neuronal activity. *Science*, **255**, 209–212.

Löwel, S. & Singer, W. (1993a) Monocularly induced 2-deoxyglucose patterns in the visual cortex and lateral geniculate nucleus of the cat. I. Anaesthetized and paralysed animals. *Eur. J. Neurosci.*, **5**, 846–856.

Löwel, S. & Singer, W. (1993b) Monocularly induced 2-deoxyglucose patterns in the visual cortex and lateral geniculate nucleus of the cat. II. Awake animals and strabismic animals. *Eur. J. Neurosci.*, **5**, 857–869.

Miller, K.D. (1995) Receptive fields and maps in the visual cortex: Models of ocular dominance and orientation columns. In Domany, E., van Hemmen, L.J. & Schulten, K. (eds), *Models of Neural Networks III* Springer-Verlag, New York, pp. 55–78.

Obermayer, K. & Blasdel, G.G. (1993) Geometry of orientation and ocular dominance columns in monkey striate cortex. *J. Neurosci.*, **13**, 4114–4129.

Rathjen, S. & Löwel, S. (2000) Early postnatal development of functional ocular dominance columns in cat primary visual cortex. *Neuroreport*, **11**, 2363–2367.

Rathjen, S., Schmidt, K.E. & Löwel, S. (2002) Two-dimensional analysis of the spacing of ocular dominance columns in normally raised and strabismic kittens. *Exp. Brain Res.*, **145**, 158–165.

Schmidt, K.E., Stephan, M., Singer, W. & Löwel, S. (2002) Spatial analysis of ocular dominance patterns in monocularly deprived cats. *Cereb. Cortex*, **12**, 783–796.

Sengpiel, F., Godecke, I., Stawinski, P., Hübener, M., Löwel, S. & Bonhoeffer, T. (1998) Intrinsic and environmental factors in the development of functional maps in cat visual cortex. *Neuropharmacology*, **37**, 607–621.

Shatz, C.J., Lindstrom, S. & Wiesel, T.N. (1977) The distribution of afferents representing the right and left eyes in the cat's visual cortex. *Brain Res.*, **131**, 103–116.

Sokoloff, L., Reivich, M., Kennedy, C., Des Rosiers, M.H., Patlak, C.S., Pettigrew, K.D., Sakurada, O. & Shinohara, M. (1977) The [^{14}C]deoxyglucose method for the measurement of local cerebral glucose utilization: theory, procedure, and normal values in the conscious and anesthetized albino rat. *J. Neurochem.*, **28**, 897–916.

Stryker, M.P. (1991) Activity-dependent reorganization of afferents in the developing mammalian visual system. In Lam, D.M. & Shatz, C.J. (eds), *Development of the Visual System.* MIT Press, Cambridge (MA), pp. 267–287.

Swindale, N.V. (1996) The development of topography in the visual cortex: a review of models. *Network*, **7**, 161–247.

Tieman, S.B. & Tumosa, N. (1997) Alternating monocular exposure increases the spacing of ocularity domains in area 17 of cats. *Visual Neurosci.*, **14**, 929–938.

Tusa, R.J., Palmer, L.A. & Rosenquist, A.C. (1978) The retinotopic organization of area 17 (striate cortex) in the cat. *J. Comp. Neurol.*, **177**, 213–235.

Weliky, M. & Katz, L.C. (1999) Correlational structure of spontaneous activity in the developing lateral geniculate nucleus in vivo. *Science*, **285**, 599–604.

Wiesel, T.N., Hubel, D.H. & Lam, D.M. (1974) Autoradiographic demonstration of ocular-dominance columns in the monkey striate cortex by means of transneuronal transport. *Brain Res.*, **79**, 273–279.

Wolf, F. & Geisel, T. (1998) Spontaneous pinwheel annihilation during visual development. *Nature*, **395**, 73–78.

Wolf, F. & Geisel, T. (2003) Universality in visual cortical pattern formation. *J. Physiol. (Paris)*, in press.

Wolf, F., Pawelzik, K., Scherf, O., Geisel, T. & Löwel, S. (2000) How can squint change the spacing of ocular dominance columns? *J. Physiol. (Paris)*, **94**, 525–537.