Changes during development of the optic disc projection relative to the area centralis position in the visual field were studied in the kitten. The determinations were based both on direct measurement and physiological location of these retinal landmark projections. The results showed that the relative distance between these retinal landmarks in visual space became shorter with age, indicating that the visual field coordinates change extensively with age. Since the area centralis cannot be seen in most young kittens, the mean distances we have determined for the landmarks in the visual field may provide a useful means for estimating the position of area centralis from the projected position of the optic disc. Our results also confirm the nonuniform growth of the retina suggested already from anatomical observations. Taking into account both eye growth and the changes in the visual field coordinates allowed some reinterpretation of changes in physiological properties of the visual cells known to occur during development, such as decreased size of the visual receptive fields, increased spatial resolution and increased responsiveness to high velocity visual stimuli.

In young kittens, as early as the day of eye opening, some cortical visual cells already show specific properties. To properly analyze these physiological properties at a given age and/or compare them during development, it is necessary to know the size of the receptive fields and their location in the visual field. The coordinate axes of the visual field must also be established precisely as a function of age, particularly as the growth of the eye changes the geometric relations between the retina and the visual field. In adult cats, these coordinates are now established because of the well known relative position of both optical landmarks: the area centralis and the optic disc. On the contrary, in kittens, these parameters have only been roughly estimated because of the embryonic corneal and vitreous vascularization which clouds the intraocular media and prohibits or makes it difficult ophthalmologically to identify and localize the area centralis. The media of the eye do not become clear until around 5–6 weeks of age. The optic disc, on the other hand, can be identified at the earliest ages.

In the present study, we have determined the projected angular position of the area centralis and the optic disc in visual field coordinates. Two methods have been used: first, its projection, when ophthalmoscopic recognition was possible; and second, electrophysiological recordings of receptive field properties of binocular cells located in the cortical projection zone of the area centralis. The results will allow, solely from the projections of the optic disc, a close estimation of the position of the area centralis and of the coordinate axes for the visual field. Particular attention is given to the fact that the retinal image of an object placed at a given distance from the eye increases because the intraocular focal length roughly doubles between birth and adulthood.

A preliminary account of the present results was reported elsewhere.

Materials and Methods

Animals

In this study, we used 75 kittens, born in our colony, reared normally or in the dark from birth, and seven adult cats. Animals of both sexes were used.

Retinal Landmarks Projection

The animals were anaesthetized with Saffan-Glaxo (1.2 ml/kg intramuscularly, i.e., 10.8 mg/kg of alfaxone and 3.6 mg/kg of alfadolone acetate). A catheter
was inserted into the radial vein to supply additional anaesthetic as required and a cannula was introduced into the trachea. The electrocardiogram was monitored, and the body temperature was maintained at 38°C through an electronically controlled heating pad. The animal was held with minimum trauma in a stereotaxic frame which assured the integrity of the visual field. The nictitating membranes were retracted with neosynephrine 5%. The pupillary alignment was checked before the animal was paralysed with flaxedil (gallamine triethiodide) and artificially ventilated. Contact scleral lenses were placed (using atropine 1%) to protect the corneae from drying. Before mydriasis, the pupillary alignment was again checked. The optic discs (od) of the retina were ophthalmoscopically projected (OD) on a tangent screen, placed at 57 cm of the eyes (Fig. 1). For animals whose ocular media was clear enough to estimate the area centralis position (ac) on the retina, it was back-projected onto the screen (AC). In almost all cases “ac” was easily identified in kittens from 5–6 weeks of age. However, over the years and a large number of young kittens studied, it was sometimes possible to identify the “ac” location on the retina even in kittens as young as 13 days and to back-project it onto the tangent screen. Both ac and od retinal landmarks were alternately projected five to ten times, because of small variations from one projection to another. The final reference position was taken as the geometrical center of the projections marks. The A and E distances (Fig. 1) of OD on the vertical and horizontal meridians (crossing in AC) were measured on the tangent screen and corrected for eccentricity so that they corresponded directly to the angular values α and ε with respect to the eye, as 1 cm = 1° at 57 cm.

Even when conditions were appropriate, the ophthalmoscopic inspection of the fundus of the eye in young kittens was difficult, as was the determination of “ac” because: (1) parts of the retina were masked by “clouds” in the optical media; (2) only a very small area could be seen at the same time; and (3) the zone of blood vessel convergence, the center of which appeared free of vascularisation, has a “crescent” shape in young kittens. It is horizontally elongated with a curve open to the inferior part of the retina. Using the fixed orientations of the main vessels for comparison during development it appeared that the lateral third of the “crescent” corresponds to “ac.”

Accuracy of the projection and determination of the landmarks position on the screen were better in the older kittens than in the younger ones as judged by the scatter of the measured values. Successive plots of the landmarks in the youngest kittens were often scattered as much as 5°, whereas in older kittens or adult, it was only around 1°.

Additional Preparation and Recording Procedure

Small openings were made bilaterally in the skull above the visual cortex and the dura was removed to expose the lateral gyrus. Throughout the recording session, anaesthesia and paralysis were maintained with continuous infusion of saffan (3.6 mg/kg/hr), flaxedil (10 to 15 mg/kg/hr), plasmagel and glucose mixture. Expired CO₂ was monitored and maintained at 4%. Extracellular recordings from single cells were performed with tungsten microelectrodes (1 to 2 MΩ at 1000 Hz). Penetrations were made vertically in the lateral gyrus in the caudal part of area 17 (P2–P5; L1–L2 Horsley Clarke coordinates) where the projection corresponding to the central part of the visual field is located or in area 18 (A5–P2). Along one such track, recordings were made approximately every 100 μm over distances of 1500 μm to 5000 μm depending on the laterality. When laterality was around 1.2 mm, tracks began orthogonal to the cortical surface and continued up to 5000 μm in the medial bank of the lateral gyrus, where it crossed layers obliquely. Two small electrolytic lesions (10 μA, 10 sec) were made, one at the end of each track and the other at an intermediate depth, for later reconstruction.

Visual Stimulation

An estimate of the characteristics of the cells was first obtained with a hand-held stimulator. Moving and stationary light stimuli (13 cd/m²) of different sizes and shapes (spots, bars, edges) were projected onto a faintly lighted screen (0.2 cd/m²) at a distance of 57 cm from the cat’s eyes. Luminances were kept constant throughout the experiments. Using back-projected visual stimuli (bright or black ones), the receptive field (RF) and the cell characteristics were then carefully analyzed. Since a particular emphasis was placed upon the RF position in the visual field, special care was taken to plot the field limits for each eye separately (see refs. 3 and 17 for detailed procedure). Briefly, the preferred orientation was established, for each eye; as the orientation of a bar eliciting the best response from the cell, the RF limits were plotted using the optimal stimulus parameters to determine the “minimum response field” and the size of the RF was the area of the field. After each experiment, all RFs mapped through visual stimulation of each eye were redrawn with respect to the corresponding optic disc.

Torsion

When the RFs of binocular cells with good responsiveness and their preferred orientation had been...
plotted with either eye, it was possible to check for the eye rotation, usually referred to as torsion. Torsion was assumed to exist if systematic differences were obtained for one preferred orientation of binocular cells when tested through each eye. The torsion was measured as the angular difference between the preferred orientations established for each eye. In practice, the drawings of the RFs recorded for one eye were turned until they fit the RFs recorded through the other eye. However, in most cases there was no torsion or a very limited one, which is in agreement with previous reports. When random small differences were observed from cell to cell, between preferred orientation tested through each eye, it was assumed they represented an orientation disparity.

Central Vertical Meridian, an $\alpha$ Determination

The central vertical meridian position was determined by shifting the RFs mapped through one eye to superpose them with those mapped through the other eye. Small random mislocations were considered as position disparity. The distance between the two projected ODs was measured, corrected for eccentricity, and $\alpha$ was taken as one-half of this distance.

The accuracy of the plotted RF was very good since the dispersion never differed from successive trials by more than 1°. Since, in the current study, only cells providing clear and vigorous responses were used, we assume that this accuracy was not age-dependent. The vertical meridian position is expected to have been very reliably established.

Determination of the AC Position and $\epsilon$ Values

This determination was not as easy as that of $\alpha$ and was not obtained in all the animals. To precisely locate the AC, many recording tracks in and around the AC projection zone in area 17 were needed. In this cortical region having the highest magnification factor, the tracks whose RFs were considered as corresponding to AC were those for which the shift of RF centers was minimum, the area covered by the RFs was the smallest and the overlap with those of the neighbouring tracks was the largest. The AC position was established in each experiment as the geometrical center of the area covered by the RFs of the tracks determined as defined above.

Histological Reconstruction

At completion of the experiment, the animals were perfused transcardiatically with a solution of 10% formaldehyde and the brain was removed. After fixation, 100 µm slices in the frontal plane were cut with a freezing microtome. Slices were then stained with cresyl-violet and electrode penetration tracks could be reconstructed using the electrolytic lesions. The position of the border between areas 17 and 18 was determined, using identical electrophysiological and morphological criteria, whatever the age of the animal.

The investigations on animals described herein conform to the ARVO Resolution on the Use of Animals in Research.

Results

Classically, in the paralyzed adult cat, the area centralis (AC) and the optic disc (OD) positions in the visual field can be determined for each eye by projection onto the screen, and the coordinates of the visual field established by vertical and horizontal meridians passing through the AC position (Fig. 1). Generally,
the AC position is deduced from that of OD, which is easier to project. Thus, according to data in the literature, the AC position is defined by an azimuth $\alpha$ of $13^\circ$6 to $15^\circ$7 and an elevation $\epsilon$ of $5^\circ$5 to $6^\circ$24.5.14 for the adult cat. The mean values of our measurements were $\alpha = 14^\circ$8 ($13^\circ$8 to $15^\circ$5) and $\epsilon = 5^\circ$8 ($5^\circ$7 to $6^\circ$).

In the youngest kittens, the projected OD positions from each eye were more separated on the screen than in the adult. This did not result from a divergent strabismus since the receptive fields of binocular cells were superimposed. A similar observation has been made by Olson and Freeman,23 who showed that in spite of an apparent divergent strabismus the visual axes are convergent even in the youngest animals. Moreover, our observations indicate that the angular distance (2A) between the OD projections decreases as a function of age (Fig. 1).

Values of angle $\alpha$, at all ages, have been obtained through either direct measurements of the back projected positions OD and AC (see Methods) in 22 kittens, or through OD projections with corrections in position by mean of binocular RF analysis in the visual cortex (see Methods) in 51 kittens. These values are reported in Figure 2A, with the regression curve from 2-week-old to adult cats. Values from normally or dark-reared kittens have not been differentiated since they appeared similar.

The calculated function of the regression curve is:

$$\alpha = 14^\circ$8 + 13^\circ$9 \times e^{-A/33.6} \quad (r = 0.78) \quad (1)$$

where A is the age of the animal in days. This function gives $\alpha$ values indicated weekly in Table 1.

These observations are in agreement with the results previously reported by Olson and Freeman18 that $\alpha$ decreases during postnatal development according to one exponential function: $\alpha = 15^\circ$7.
+ $16^\circ 1 \times e^{-A/24.4}$, reported in Figure 2A as a dashed curve. Both $\alpha$ curves are very similar.

$\epsilon$ has been experimentally determined using the same methods as those used for $\alpha$ measurements in 21 kittens with projected retinal landmarks and in 13 kittens with electrophysiological determinations; these are plotted in Figure 2B. A regression curve (continuous line) also shown in Figure 2B was calculated from the function:

$$
\epsilon = 5.08 + 9.09 \times e^{-A/29} \quad (r = 0.80)
$$

(2)

The $\epsilon$ values given on a weekly basis in Table 1 were also calculated using the above function.

Since the growth of the eye changes its focal length, it is expected to have the same influence in all directions. In this case, the elevation $\epsilon$ would be expected to vary proportionally with $\alpha$.

A simple method to estimate $\epsilon$ from one measured $\alpha$ would be to apply the ratio $\alpha/\epsilon$ of adult cats during development. From data of Vakkur et al, this ratio $(\alpha/\epsilon = 13^\circ 6/5^\circ 5$ or $15^\circ 7/6^\circ 2$) is equal to 2.47 or 2.53, respectively (mean 2.50). Hence a developmental curve for $\epsilon$ could be found from $\alpha/2.50$ for $\alpha$ at all ages. Such a curve is reported as the dashed one in Figure 2B.

The two curves show important differences between 2 and 6 weeks, indicating that more than postnodal distance lengthening is involved during eye growth.

In conclusion, it can be seen that in kittens, the angular distances $A$ and $E$ decrease in an exponential fashion until at least 10 weeks of age. The values given in Figure 2 and Table 1 allow location of the AC in the visual field from the projected OD position and establishing the visual field coordinates as a function of age.

In fact, since the exponents are not the same in the equation for $\alpha$ and $\epsilon$, it can be seen that the ratio $\alpha/\epsilon$ will vary exponentially with age. When the ratio of the two equations were calculated at different ages, it was found that the ratio converges to 2.5 at about 10 weeks from a minimum of 1.93 at day 1.

**Discussion**

In the present study, we have determined in the kitten the values of angles $\alpha$ and $\epsilon$, respectively azimuth and elevation of OD, as a function of age. Both angles decrease exponentially in kittens from 2 weeks of age to adulthood with different exponential constants especially between 2 and 6 weeks.

Previous reports which have attempted to determine the AC position in kitten either have considered it as identical with the adult, or have taken the orthogonal projection of the nose upon the tangent screen as the center of gaze. However, the former procedure neglected the effects of eye growth on $\alpha$ and $\epsilon$, and the latter did not take into account the fact that, generally, visual axes under paralysis are not correctly positioned. Consequently, in both cases, the error can be as much as $10^\circ$. Actually, the values we have established give the AC position from OD projection as a function of age more precisely than applying other methods in young kittens. However, in kittens older than 2 months, the most accurate method remains the careful projection of both OD and AC for each experimental animal.

During eye growth, two factors influence the $\alpha$ and $\epsilon$ changes: the postnodal distance (PND) lengthening and the retinal growth. PND lengthening would reduce the angular values of a retinal segment constant in size, and the retinal growth would increase the angular values of a retinal segment determined by equivalent points. As the observed changes of $\alpha$ and $\epsilon$ are decreases it is obvious that PND lengthening appears more important than retinal growth. But, as both phenomena are known to occur, the observed values of $\alpha$ and $\epsilon$ in young kittens are smaller than they would have been without any retinal growth.

Since the change in the focal length (PND) has the same influence in all directions, the discrepancy between the experimental $\epsilon$ curve and the deduced one from $\alpha$ values (Fig. 2B) indicates a nonuniform growth of the retina. The fact that experimental values were greater than the calculated ones shows that before 6 weeks, the retinal size corresponding to $E$ is closer to the adult one than the retinal size corresponding to $A$, ie, from 2 to 10 weeks the radial retinal growth following nasal axis ($\alpha$) is larger than that following inferior axis, at the eccentricity of OD ($\epsilon$).
This conclusion agrees with anatomical results by Mastronarde et al. In their Figure 6, which gives retinal growth in every axis from 3 weeks to adulthood, it appears that the radial growth in nasal axis is larger than the tangential one. At eccentricities between 2 and 3.5 mm from AC, which include the OD–AC distance, the ratio of radial on tangential growth is constant and can be estimated to 1.14. This allows one to determine an $a/e$ ratio in 3-week-old kittens by dividing the adult $a/e$ ratio (2.5) by the growth ratio (1.14). It gives 2.19, ie, a value very similar to the 2.18 obtained value from 3 weeks $a/e$ of Table 1. This similarity strongly supports the nonuniform retinal growth observations.

Neglecting the effects of the eye growth on the angular distances during development may lead to actual errors on angular measures due to mislocation of AC from the OD projection. Using the correct position of AC would be particularly important in comparative studies of the physiological properties of visual cells as a function of eccentricity, such as binocular integration, stimulus velocities, receptive field sizes, extent of overlap of the ipsilateral and contralateral hemifields along the central vertical meridian, or even the extent of the visual field. Below, we reconsider some interpretations for the development of the physiological characteristics of visual cells which have been shown to change with age.

Receptive Fields Size

It has been shown that the size of the receptive fields of visual cells decreases during development in the retina, in the lateral geniculate nucleus  and in the cortex. It is interesting to note that, by considering eye growth alone, a given retinal area which would “see” 20 deg. in space in a 6-week-old kitten, but will “see” only 9 deg. in the adult cat. So the extent of visual field from which a visual cell receives information gradually decreases as the optical components of the eye grow. This effect has already been suggested by Rusoff and Dubin and Rusoff to explain the decreasing size of ganglion cell receptive field.

However, eye growth is not the only factor to take into account in interpreting the decrease of the receptive fields size. The growth of the dendritic fields of the visual cells at every level of the visual system as well as modifications in neuronal connectivity should also be relevant. This has been discussed elsewhere to explain the evolution of the receptive field sizes of different types of cells recorded in area 18.

Spatial Resolution

The decrease in size of the visual receptive fields with age could also be a factor in explaining the improvement of spatial resolution during development observed at different levels of the visual system, either electrophysiologically or behaviorally. Since visual acuity is considered to depend upon the resolving power at the area centralis and as it is the first retinal region to mature, improvement of spatial resolution with age must be attributed to eye growth.

Stimulus Velocities

The effective stimulus velocities in activating visual cortical cells increase with age. Is velocity detection affected by the modifications of the optical components during development? For a given stimulus moving across a receptive field on the screen, the velocity of its image on the retina becomes faster as the PND lengthens. Conversely, for a given stimulus velocity on the retina, the larger the PND, the slower must the velocity of the stimulus be on the screen. Thus, in fact, the increasing length of the PND does not “explain” the observed improvement in stimulus velocity detection; on the contrary, it acts in the opposite way. Consequently, other mechanisms have to be considered, on the one hand, to account for the observed changes in velocity detection, and on the other hand, to counterbalance the eye growth effect.

Key words: kitten, optical landmarks, development, visual field coordinates

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