Postnatal development of disparity sensitivity in visual area 2 (V2) of macaque monkeys

I. Maruko, B. Zhang, X. Tao, J. Tong, E.L. Smith III, and Y.M. Chino

College of Optometry, University of Houston
Houston TX, 77204-2020

Abbreviated title: Disparity sensitivity in infant visual cortex

Figures: 12
Pages: 35
Words in text: 5851

Key words: visual area 2, primary visual cortex, development, disparity sensitivity, stereopsis, macaque monkeys

Acknowledgement: This work was supported by National Institutes of Health research and CORE Grants, EY-08128 (YMC), EY-03611 (ELS), and RR-07146. Corresponding author:

Yuzo Chino
College of Optometry
University of Houston
505 J. Davis Armistead Bldg.
Houston, TX 77204-2020
ychino@uh.edu
713-743-1955
713-743-2053 (fax)
Abstract

Macaque monkeys do not reliably discriminate binocular depth cues until about 8 weeks of age. The neural factors that limit the development of fine depth perception in primates are not known. In adults, binocular depth perception critically depends on detection of relative binocular disparities and the earliest site in the primate visual brain where a substantial proportion of neurons are capable of discriminating relative disparity is visual area 2 (V2). We have examined the disparity sensitivity of V2 neurons during the first 8 weeks of life in infant monkeys and compared the responses of V2 neurons to those of V1 neurons. We found that the magnitude of response modulation in V2 and V1 neurons as a function of interocular spatial phase disparity was adult like as early as 2 weeks of age. However, the optimal spatial frequency and binocular response rate of these disparity sensitive neurons were more than an octave lower in 2- and 4-week-old infants than in adults. Consequently, despite the lower variability of neuronal firing in V2 and V1 neurons of infant monkeys, the ability of these neurons to discriminate fine disparity differences was significantly reduced compared to adults. This reduction in disparity sensitivity of V2 and V1 neurons is likely to limit binocular depth perception during the first several weeks of a monkey’s life.
Introduction

In macaque monkeys, stereopsis is absent during the first 2-3 postnatal weeks and binocular disparities are not reliably detected until about 6-8 weeks of age (O'Dell and Boothe, 1997). Similarly human infants do not demonstrate stereopsis until about 4-6 months of age (Birch et al, 1983; 1993; Brown et al, 2007). The neural factors that constrain the early development of binocular depth perception have not been extensively investigated. The ability of the primary visual cortex (V1) neurons to process local disparity information is thought to be a fundamental requirement for stereopsis and is likely to put some limits on performance (Prince et al, 2000; Cumming and DeAngelis, 2001; Nienborg et al, 2004). Previously, we found that an adult-like proportion of V1 neurons is sensitive to interocular spatial phase disparity only 6 days after birth (Chino et al, 1997), indicating that the basic binocular connections in V1 required for stereoscopic vision are present near birth.

It is becoming increasingly clear, however, that in adult monkeys, V1 neurons alone could not directly support binocular depth perception (Cumming and Parker, 1999; Read et al, 2002). Instead, extrastriate neurons that are sensitive to relative binocular disparity are thought to more directly underlie fine depth perception (Roe et al, 2007; Parker, 2007 for reviews). Thus, an emerging view on binocular vision development is that immaturities in cortical neurons beyond V1, yet to be discovered, are likely to be involved in limiting stereopsis in neonates (O'Dell and Boothe, 1997; Chino et al, 2004; Kiorpes and Movshon,
2004). Consistent with this view, several lines of evidence suggest that the overall functional maturation of extrastriate visual areas appears to proceed at a slower pace relative to V1 (Distler et al, 1996; Batardiere et al, 2002; Kiorpes and Movshon, 2004; Zhang et al, 2005a; Zheng et al, 2007).

In adults, V2 is the earliest site beyond V1 where a substantial proportion of neurons are capable of detecting relative binocular disparity (Thomas et al, 2002). Also, a link between the disparity sensitivity of cortical neurons in individual monkeys and their ability to discriminate depth cues can be established by quantifying their choice-related activity in these neurons while the monkeys perform a discrimination task. Disparity sensitive V2 neurons, but not V1 neurons, have been shown to exhibit ‘choice-related’ activity (Nienborg and Cumming, 2006). However, it is not known whether V2 neurons of infant monkeys are sensitive to relative binocular disparity, or whether disparity selective V2 neurons in infants show choice-related activity. This is simply because at these early ages, it is not feasible to conduct microelectrode recording experiments in awake behaving monkeys. In this study, therefore, we estimated the ability of V2 and V1 neurons to discriminate fine disparity differences by analyzing their sensitivity to binocular phase disparities, their optimal spatial frequencies, and their response amplitudes and variabilities during the first several postnatal weeks (Nover et al, 2005; Yang and Maunsell, 2004; Nienborg and Cumming, 2004). We found that although the abilities of V2 neurons to combine binocular signals and to detect interocular spatial phase disparities were qualitatively adult like as early as 14 days after birth, the median
disparity threshold of V2 neurons was over 4 times higher in 2- and 4-week-old infants than in adults. We also found that relative changes in the median disparity threshold for V2 and V1 neurons between 2 and 8 weeks of age paralleled behavioral improvement in stereoacuity (O’Dell and Boothe, 1997).

**Materials and Methods**

Microelectrode recording experiments were conducted in anesthetized and paralyzed monkeys (*Macaca mulatta*). All experimental procedures conformed to the National Institute of Health guidelines for the use of animals in research and were approved by the University of Houston’s Institutional Animal Care and Use Committee.

**Subjects.** Five 2-week-old, four 4-week-old, three 8-week-old infant monkeys and four adult monkeys served as subjects. The weights of the infant monkeys varied between 480 g and 600 g at 2 weeks, between 500 g and 525 g at 4 weeks, and between 550 g and 750 g at 8 weeks of age. Some results on monocular response properties of cortical neurons from these monkeys have been previously reported (Zheng et al., 2007; Zhang et al., 2007).

**Preparation.** The surgical preparation and recording procedures have been described in detail elsewhere (Chino et al., 1997; Zhang et al., 2005; Zheng et al., 2007). Briefly, the monkeys were anesthetized initially with an intramuscular injection of ketamine hydrochloride (15-20 mg/kg) and acepromazine maleate (0.15-0.2 mg/kg). After all surgical procedures were completed, the animals were
paralyzed by an intravenous injection of vercuronium bromide (Norcuron; 0.1 mg/kg/hr) and artificially ventilated with a mixture of 59% N₂O, 39% O₂, and 2% CO₂. Anesthesia was maintained by the continuous infusion of a mixture of Propofol (4 mg/kg/hr) and Sufentanyl Citrate (0.05 µg/kg/hr). Core body temperature was kept at 37.6°C. Cycloplegia was produced by topical instillation of 1% atropine and the animals’ corneas were protected with rigid gas permeable, extended-wear contact lenses. Retinoscopy was used to determine the contact lens parameters required to focus the eyes on the stimulus screens. Additional spectacle lenses were also used if necessary.

Recording and Visual stimulation. Tungsten-in-glass microelectrodes (FHC, Inc., Bowdoinham, ME) were used to record and isolate the activity from individual cortical neurons. A typical penetration in V1 began several millimeters posterior to the lunate sulcus and about 1.5 cm from the midline and ended when the electrode tip entered the white matter. The tangential penetrations in V2 (the angle of deviations from perpendicular was about 20°) were typically started right behind the blood vessels running along the lunate sulcus and also about 1.5 cm from the midline. The penetration ended when the electrode tip exited V2. All receptive fields were located within 5.0° of the center of the projected fovea.

For each isolated neuron, the receptive field for each eye was mapped and its ocular dominance was initially determined using hand held stimuli (Hubel and Wiesel, 1962). Responses to drifting sine wave gratings (3.1 Hz, 80% contrast) were measured for a broad range of stimulus orientation and spatial frequency from which the preferred orientations, direction of stimulus drift, and spatial
frequencies were determined for each unit. The visual stimuli for these experiments were generated on a monochrome monitor (VRG) with ultra-short persistence (frame rate = 140 Hz; 800 x 600 pixels, screen size = 20° x 15° at 114 cm and mean luminance = 50 cd/m²). Recorded action potentials were digitized at 25 kHz and sampled at a rate of 140 Hz (7.14 msec bin widths) and compiled into peristimulus time histograms (PSTHs) that were equal in duration to, and synchronized with, the temporal cycle of the grating (TDT data acquisition system, TD, Inc, FL). Cells were classified as simple or complex on the basis of the temporal characteristics of their responses to a drifting sine wave grating of the optimal spatial frequency and orientation (Skottun et al., 1991).

**Measurements of orientation, spatial frequency and disparity tuning functions.**

**Orientation tuning.** The preferred orientation and orientation bandwidth for each receptive field were determined by fitting the orientation tuning functions with wrapped Gaussian functions (Fig 1a) (Swindale, 1998):

\[
G(\theta) = m_1 \sum_{n=\infty} \exp\{-((\theta-m_2+180n)^2/(2m_3^2))\}
\]

Where \( \theta \) = orientation, \( m_1 \) = the response rate, \( m_2 \) = the preferred orientation, and \( m_3 \) = the standard deviation of the Gaussian function.

**Spatial frequency tuning:** To determine each cell’s optimal spatial frequency and spatial resolution, the spatial frequency response data were fitted with Gaussian functions (Fig 1b) (DeAngelis et al., 1993):

\[
G(m_0) = m_1 \exp\{-(m_0-m_2)^2/(2m_3^2)\}
\]
Where \( m_0 = \) spatial frequency, \( m_1 = \) the response rate, \( m_2 = \) the optimal spatial frequency, and \( m_3 = \) the standard deviation of the Gaussian function. The spatial resolution for each unit was determined by locating the highest spatial frequency that evoked responses which were significantly higher than the cell’s average spontaneous firing rate (i.e., > ±2 SDs).

**Binocular interactions index (BII):** To determine the strength and the nature of binocular interactions, responses were collected for dichoptic sine wave gratings of the optimal spatial frequency and orientation as a function of the relative interocular spatial phase disparity of the grating pair (Fig 1c). The sensitivity to relative interocular spatial phase disparities was quantified using a *binocular interaction index* that was calculated from the sine function fit to the binocular phase tuning data (Ohzawa and Freeman, 1986a,b; Smith et al, 1997; Prince et al, 2002a). Binocular interaction index here is defined as

\[
\text{BII} = \frac{R_{\text{max}} - R_{\text{min}}}{R_{\text{max}} + R_{\text{min}}}
\]

Where \( R_{\text{max}} \) is the largest response on the tuning function, and \( R_{\text{min}} \) is the smallest response. These values were obtained from fit functions.

**Disparity discrimination index (DDI):** To take effects of variability in neuronal firing and firing rates of individual cells into account, we also calculated the DDI (Prince et al, 2002a; Uka and DeAngelis, 2003):

\[
\text{DDI} = \frac{(R_{\text{max}} - R_{\text{min}})}{[(R_{\text{max}} - R_{\text{min}}) + 2.\text{RMS}_{\text{error}}]}
\]
Where $R_{\text{max}}$ is the largest response on the tuning function, and $R_{\text{min}}$ is the smallest response. RMS $\text{error}$ is the square root of the residual variance around the mean across the entire tuning function.

To characterize whether binocular signal interactions were facilitatory or suppressive in nature, the peak binocular response / dominant monocular response ratios (Peak B/M ratios) were calculated for each unit and expressed in terms of relative strength (db), i.e., $10 \log \text{Peak B/M}$. Negative Peak B/M values signify binocular suppression and positive values indicate binocular facilitation. For the calculation of the BII and the DDI, and the Peak B/M ratio was performed on the square root of firing rate (Prince et al, 2002a; Uka and DeAngelis, 2003).

**Histology.** At the end of each penetration, small electrolytic lesions (5μA, 5 sec, electrode negative) were made at three points along the track for later reconstruction. Experiments were terminated by administering an overdose of sodium pentobarbital (100 mg/kg) and the animals were killed by perfusion through the heart with an aldehyde fixative. Frozen sections were stained for Nissl substance and cytochrome oxidase. The laminar distribution of individual units was estimated from the recording depths and the histologically identified electrode tracks. Our sampling frequency was generally uniform and similar in all subject groups.

**Results**
We recorded responses from a total of 788 neurons (158 simple & 361 complex in V2 and 78 simple & 191 complex in V1) and quantitatively analyzed the binocular and monocular response properties of 104 V2 and 78 V1 neurons in 2-week-old infants, 84 V2 and 48 V1 neurons in 4-week-old infants, 138 V2 and 63 V1 neurons in 8-week-old infants, and 193 V2 and 80 V1 neurons in adult monkeys. Since we did not find major differences between simple and complex cells or laminar differences in any of the response measures that we studied, we have combined data in these cell types from all cortical layers for the subsequent analyses. Statistical significance was tested for group differences with Kruskal-Wallis test for median values, unless specified otherwise.

**Binocular signal combination in V2 is adult-like at 2 weeks of age**

One of the most efficient and highly sensitive ways to assess the strength and characteristics of binocular signal combination in a visual cortical neuron is to measure its binocular interaction index (BII) (Ohzawa and Freeman, 1986a,b; Smith et al, 1997; Chino et al, 1997; Prince et al, 2002a). As we have previously reported for V1 neurons (Chino et al, 1997) and as can be seen in Figure 2, the response modulation of the representative V2 neuron from a 2-week-old infant as a function of interocular spatial phase disparity was as robust as those in older infants and adults; and the calculated BII values were also comparable at all ages. However, it is important to note that the mean binocular response rates (dotted lines) for these representative units from infants were substantially lower than those from adults.
The population analysis of disparity tuning functions revealed that in V2 the median BII values were 0.18, 0.16, and 0.19 in 2-, 4- and 8-week-old infants, respectively, and were not statistically different from the median BII value in adults (0.18)(p > 0.01)(Fig 3, left column). The median BII values in V1 also did not change over age (p > 0.01)(Fig 3, right column). Moreover, the proportion of ‘disparity sensitive’ units, defined as neurons that exhibited a statistically significant response modulation as a function of phase disparity (Prince et al, 2002a) was similar for all the infant groups and adults (p > 0.01, $\chi^2$ test)(filled bars in Fig 3). Note that the frequency distribution of the BII in V1 of our adult monkeys was similar to that previously reported for awake behaving adult monkeys (Prince et al, 2002a).

The binocular interaction index (BII) of a give neuron does not take the variability of firing into account, and tends to be negatively correlated with the mean firing rate (Prince et al, 2002a). Because V2 and V1 neurons in our infant macaques showed much lower firing rates than in adults (Chino et al, 1997; see Fig 6), BII values of 2- and 4-week-old infants might have been artificially elevated and might have not reflected the credible estimate of neuron’s sensitivity to binocular disparity at these early ages. To examine this possibility, we calculated the disparity discrimination index (DDI)(Prince et al, 2002a; Uka and DeAngelis, 2003). The frequency distribution of DDI values in our adult V1 (Fig 4) was very similar to that in awake adult monkeys (Prince et al, 2002a). More importantly, the distribution and the median DDI values in V2 and V1
neurons of all infant groups were not significantly different from those in adults (p > 0.01)(Fig 4).

The nature of binocular signal interactions (i.e., excitatory versus inhibitory) in V2 and V1 neurons were examined by taking the ratio of the peak binocular response over the dominant monocular response, and the resulting values were expressed in terms of relative strengths (db), i.e., 10 log peak B/M. Although the median B/M value (0.45 db) at 2 weeks of age was considerably lower than that in adults (0.76 db), the difference did not reach statistical significance (p > 0.01)(Fig 5). A similar difference was found for V1 neurons at 2 weeks of age (0.33 db in infants vs. 0.56 db in adults)(p > 0.01). The median ratios for 4- and 8-week-old-infants (0.83 db and 0.52 db in V2, 0.69 db and 0.39db in V1) were not different from the corresponding ratios in adults (p > 0.01). The overall distribution of Peak B/M ratios for V1 neurons in our anesthetized and paralyzed adults were similar to the distribution of binocular/monocular response ratios of V1 neurons found in awake behaving adult monkeys (Prince et al, 2002b).

**Firing rates, response variability and optimal spatial frequency are low in infants**

The ability of individual V1 neurons to discriminate fine disparity differences depends on a cell’s optimal spatial frequency, discharge rate, and response variability (Prince et al, 2002a; Nienborg et al, 2004; Nakatsuka et al, 2007; also see Nover et al, 2005; Yang and Maunsell, 2004 for a similar analysis). Thus we examined the maturation of these response properties. In V2, although the binocular/monocular ratios were adult like at 2 and 4 weeks of age, the median
binocular response rate at 2 weeks of age (8.61 spikes/sec) was less than half of
the adult value (19.93 spikes/sec) and was significantly lower at 4 weeks (8.33
spikes/sec) and 8 weeks of ages (13.41 spikes/sec) than in adults (p < 0.01)(Fig
6a). Similarly the mean binocular response rate of V1 neurons in all infant groups
was significantly lower than the response rate in adults (p < 0.01).

As anticipated from these low firing rates in infants, however, the response
variances of V1 and V2 neurons were also much lower in infants than in adults.
The median variance-to-mean ratio of V2 neurons at 2 weeks of age ($\sigma^2 / \mu$,
calculated for the duration of 322 msec for 20 epochs at each phase disparity)
was only one half (1.10) of the adult value (2.20)(p < 0.01)(Fig 6b). Similarly, the
median variance-to-mean ratios of V2 neurons at 4 (1.27) and 8 weeks of age
(1.13) were also significantly lower than that in adults (p < 0.01). The median
ratio for V1 neurons was 1.04 at 2 weeks, 1.06 at 4 weeks, and 0.94 at 8 weeks
of age, all of which were significantly lower than that in adults (1.59)(p < 0.01).
Thus, the firing pattern of V1 and V2 neurons in our infants was less noisy than in
adults. It is important to note that the differences in variance-to-mean ratios
between V2 and V1 were statistically significant at 8 weeks of age (p < 0.01) and
for adults (p < 0.01) whereas there were no significant differences at 2- and 4-
weeks of age (p > 0.01).

One of the most important variables affecting the disparity sensitivity of
individual cortical neurons is its spatial frequency tuning characteristics. The
optimal spatial frequency was determined for each neuron from its fitted spatial
frequency tuning function (Fig 1b). In V2, the median optimal spatial frequency at
2 (0.70 c/deg) and 4 (0.91 c/deg) and 8 weeks of age (1.37 c/deg) were significantly lower than that in adults (2.04 c/deg, p <0.01)(Fig 7a). Similar differences were found between infants and adults (2.04 c/deg) for V1 in 2- (1.0 c/deg, p < 0.01) and 4-week-old infants (1.02 c/deg, p < 0.01), but not in 8-week-old infants (1.60 c/deg, p > 0.01).

The median spatial resolution of V2 neurons at 2 weeks of age (3.49 c/d), determined by locating the highest spatial frequency that evoked responses significantly higher than the average spontaneous firing of the unit (> ± 2 SD), was less than half the adult median value (8.19 c/d)(p < 0.01)(Fig 7b). The median spatial resolutions at 4 and 8 weeks of age (3.65 c/d and 5.1 c/d, respectively) were also significantly lower compared to adults (p < 0.01). In contrast, 2- (4.00 c/deg) and 4-week-old infants (4.41 c/deg), but not 8-week-old infants (5.50 c/deg) had lower median spatial resolutions for V1 neurons compared to adults (7.50 c/deg)(p < 0.01). Our data on the developmental changes in the average spatial resolution of V1 neurons (open triangles) are similar to those previously reported (Kiorpes and Movshon et al, 2004). Although we did not directly measure the contrast threshold of V2 or V1 neurons for high spatial frequency gratings, the lower spatial resolutions found for infants using gratings of 80% contrast suggest that the contrast sensitivity of these neurons for higher spatial frequency is likely to be substantially lower than that in adults.

Normalized optimal disparity sensitivity is low at 2- and 4-weeks of age
Based on the response modulation as a function of spatial phase disparity, optimal spatial frequency and response variance of each neuron, we estimated its ability to discriminate small disparity differences, defined here as the neuronal disparity threshold (Fig 8). The effects of the optimal spatial frequency (2.0 c/deg) on disparity sensitivity for a representative disparity-tuned V2 neuron are shown in Fig 8a. The changes in the cell’s absolute firing rate per unit of angular disparity \( (\text{right ordinate, spikes/sec/arcmin}) \) were calculated for the phase disparity tuning function. Note that the highest sensitivity is found at the steepest slope of the disparity tuning function (indicated by an arrow and the thick line at spatial phase disparities around 180 degrees). The optimal disparity sensitivity for this unit therefore was 2.99 spikes/sec/arcmin.

In adults, the ability to detect small disparity differences is also influenced by the variability of neuronal firing (Yang and Maunsell, 2004; Nover et al, 2005; Nakatsuka et al, 2007). To take response variability into account, the relationship between the mean firing rate and response variance was initially determined by plotting a cell’s mean response rate as a function of its response variance. Figure 8b shows data for a representative V2 neuron. From the best linear fit for the data points, the response variance of this unit was estimated for the response rate that corresponded to the steepest portion of its phase disparity function (the dotted line). The optimal disparity sensitivity for this unit (2.99 spikes/sec/arc min) was then normalized with square root of response variance. The disparity threshold for this V2 neuron was, then, calculated by taking the inverse of the...
ratio of cell’s firing rate per unit of angular disparity / √response variance (2.13 arc min).

The population analysis for the optimal disparity sensitivities demonstrates the effects of the low optimal spatial frequencies and the low firing rates of V2 and V1 neurons in infants (Fig 9). Specifically the median optimal disparity sensitivity of V2 neurons at 2 weeks of age (0.14 spikes/sec/arcmin) was nearly 10 times lower than that in adults (1.21 spikes/sec/arcmin; p < 0.01). At 4 and 8 weeks of age, the median disparity sensitivity was still far lower than in adults (p < 0.01). In V1, the optimal disparity sensitivity at 2 and 4 weeks of age was significantly lower than in adults (p < 0.01), but at 8 weeks of age, the median optimal disparity sensitivity was indistinguishable from adults (p > 0.01), primarily because optimal spatial frequencies of V1 neurons had become adult-like (Fig 7a).

The median disparity threshold of V2 neurons in 2-week-old infants was more than 4 times higher than that in adults. In addition, for all of the other infant groups, the median disparity threshold of V2 neurons was significantly higher than that in adults (p < 0.01)(Fig 10). The median threshold values for V1 units were also significantly higher than in adults but only for 2- and 4-week-old infants (p < 0.001). Interestingly the median threshold of V2 neurons in 2-week-old infants was significantly higher than that in V1 neurons (p < 0.01). It is worthwhile to note that the adult median disparity threshold values (4.51 arcmin in V2 and 5.26 arcmin in V1) were very similar to the median ‘neurometric disparity threshold’ value of V1 neurons in awake behaving monkeys (estimated from Fig...
13 of Prince et al, 2000), which was on average at least 4 times higher than their ‘psychometric disparity thresholds’.

To obtain a clearer picture of which ‘limiting’ factor may have relatively larger impact on the neuronal disparity threshold, we compared the relative developmental change in neuronal ‘disparity-acuity’ (1/disparity threshold) with improvement in the BII/DDI, variance-to-mean ratio, spatial frequency (optimal and resolution), and the response rate of V1 and V2 neurons (Fig 11). The median values for these response measures at 2, 4, and 8 weeks of age were normalized to their respective values in adults. The BII or DDI values did not significantly change over age in V2 or V1. For V1 neurons, the improvement in disparity acuity was closely associated with their age-dependent changes in optimal spatial frequency, resolution, and response rate. In V2, however, the disparity acuity of neurons at 2 weeks of age was far more immature than any of these responses. Together, in V2, but not in V1, additional limiting factor(s) might have contributed to the lower disparity acuity at 2 weeks of age.

Discussion

The primary findings of this study are that the binocular responses of V2 neurons of 2- and 4-week-old infant monkeys modulate at near adult levels as a function of binocular spatial phase disparity, but that their relatively low binocular response amplitudes and their coarser spatial frequency tuning impose severe limits on their ability to discriminate fine disparity differences.
According to the only psychophysical study in the literature on the normal maturation of stereopsis in monkeys, stereoscopic vision could not be detected during the first 3 postnatal weeks even for random-dot stimuli having disparities as large as 30 arcmin (O'Dell and Boothe, 1997). By 4 weeks of age, however, nearly 80% of monkeys showed “evidence of stereopsis” and all eleven monkeys exhibited stereopsis by 8 weeks. The average stereo-acuity rapidly dropped from 21.7 arcmin at 3 weeks (n =6) down to about 3.3 arcmin by 8 weeks of age (n=11), although the stereoacuity at 8 weeks of age was several times worse than in adults (O'Dell and Boothe, 1997; Prince et al, 2000). Assuming that these represent an estimate of the infant’s optimal visual performance, what prevents the emergence of stereopsis during the first 2-3 weeks of life and what limits stereoacuity development during the first 8-10 weeks of life? The critical immaturities that have been previously proposed to explain reduced binocular performance in infant primates are 1) abnormal binocular alignment and vergence eye movements (e.g., O'Dell and Boothe, 1997), 2) the slow maturation of cortical binocular mechanisms (e.g., Birch, 1993; Held, 1993; Kiorpes and Movshon, 2004), and 3) the poor ‘visibility’ of binocular stimuli (e.g., Schor et al, 1984, Brown et al, 2007) due to coarse spatial frequency tuning and/or low contrast sensitivity (e.g., Chino et al, 1997; Zheng et al, 2007).

Alignment and vergence

Normal binocular vision requires high degrees of coordination between motor and sensory systems. In particular, stereopsis has been shown to depend on
precise interocular alignment (Harwerth et al, 1995). A critical variable to be considered for behavioral assessments of stereopsis in primate infants, then, would be potential immaturities in alignment and vergence eye movements that compromise or prevent fusion of binocular images. For the present experiment, this was not an issue because we employed anesthetized and paralyzed preparations and the binocular disparities for our experiments were optically controlled with a high degree of precision. However, according to the only data available in the literature on alignment and vergence in awake behaving infant monkeys (published only as an abstract), neonates and young infant monkeys were reported to exhibit unsteady fixation and interocular alignment (Boothe and Gong, 1992). Thus, O'Dell and Boothe (1997) concluded that immaturities in oculomotor mechanisms were the primary reason why they could not reliably detect stereopsis during the first several postnatal weeks.

Several lines of evidence suggest, however, that the role of errors in interocular alignment in limiting binocular vision development in monkeys may not be as substantial as these investigators suggested. For example, in human infants of equivalent ages, binocular alignment and/or vergence eye movements do not appear to significantly limit stereoscopic vision (Birch et al, 1983; Braddick, 1996, Hainline and Riddell, 1995; Brown et al, 2007). Consistent with this view, we have found in our infant monkeys that the fundamental sensory requirements for binocular fusion and alignment (e.g., relatively well developed binocular mechanisms and adult-like sensitivity to binocular disparity) are present in V1 and V2 as early as 6-14 days after birth, if not at birth (also see Chino et al,
1997). A recent study demonstrated that many infants between 7 and 24 weeks of age show ‘adult like’ latencies for accommodation and vergence responses during binocular viewing (Tondel and Candy, 2008). Equally important are the previous findings that fixating eye movements (saccades) of human infants are relatively mature near birth (Aslin, 1977), and as early as 2-3 months of age (roughly equivalent to 2-3 weeks in monkeys), adult like amplitude and velocities are attained in reactive saccades (Hainline et al, 1984; Matsuzawa and Shimojo, 1997; Richards and Holley, 1999; Garbutt et al, 2006). These results suggest that subcortical mechanisms supporting ‘motor’ components of fixation and alignment and the extraocular musculature are relatively well developed near birth provided that stimuli for fixation are ‘visible’ to infants.

Maturation of binocular mechanisms

The present results rule out a lack of disparity detectors in V2 (or V1) as the primary source of stereo-deficiencies in infants. We found that the binocular response modulation of neurons in these areas as a function of interocular spatial phase disparity (BII and DDI) was as robust as in adults. Moreover, the distributions of binocular/monocular response ratios of V2 and V1 neurons in our infant monkeys were very similar to those for adults and to the distribution of binocular/monocular response ratios found in V1 of awake, behaving adult monkeys (Prince et al, 2002b). Interestingly, these investigators also found that those units in which the peak binocular responses were substantially greater than the maximum monocular responses were classified as tuned excitatory (“TE”) or
far cells ("FA") while those with binocular to monocular ratios around 1.0 or slightly less than 1.0 were classified as either near or tuned inhibitory cells, respectively. In this respect, it is tempting to speculate that cortical circuitry underlying conventional disparity cell types (i.e., “near”, “far”, “tuned-excitatory”, and “tuned-inhibitory”) may be largely adult like in V2 and V1 at 2 weeks of age.

Although qualitatively adult-like binocular mechanisms exist in the visual cortices of neonates, it is not known whether disparity sensitive V2 neurons in our infants are sensitive to relative disparity. As mentioned earlier, this is because microelectrode recording experiments in awake, behaving monkeys are not currently plausible at these ages. However, there is indirect evidence from our separate studies (e.g., Zhang et al, 2005) that relative disparity sensitivity might be very weak or absent all together in V2 near birth. In order to have robust sensitivity to relative binocular disparity, extrastriate neurons must be able to compare and ‘integrate’ signals over relatively large areas, e.g., units with RF centers and surrounds that are tuned to different disparities (Nienborg and Cumming, 2006; Umeda et al, 2007; Roe et al, 2007; Parker, 2007). In this respect we have previously found that the RF center/surround organization of V2 neurons is exceedingly immature at 2 weeks of age, for example, there are little or no measurable RF surrounds. There is considerable improvement by 4 weeks of age, and the RF centers and surrounds become largely adult like by 8 weeks of age (Zhang et al, 2005), the age at which infant monkeys typically exhibit good stereoacuities (O’Dell and Boothe, 1997). Thus, immaturities in the relative disparity detectors of V2 could be a major factor limiting stereopsis development.
Effects of spatial frequency tuning, firing rate and contrast sensitivity

Even if disparity tuned V2 neurons in our infant monkeys were sensitive to relative disparity, their performance in stereo-tasks could still be limited by the reduced ability of these neurons to detect fine disparity differences. We found that the median disparity threshold of V2 neurons was about 4-5 times worse in 2-week-old infants than in adults (Fig 10). The primary reason for the poor discrimination performance of V2 neurons was their low optimal spatial frequencies (Fig 5a) and their low response rates at these ages (Fig 6a).

How might the normalized optimal disparity sensitivity of V2 neurons in our infant monkeys affect their binocular depth perception? Figure 12 compares the relative time courses for the improvement in behavioral stereoacuity (adopted from O'Dell and Boothe, 1997) with the disparity thresholds of V2 and V1 neurons obtained in this study (Fig 10). The best neuronal threshold value (~ 5 arcmin in our adults, see Fig 10) on the right scale was aligned with the optimal value in the left scale for behaviorally measured stereoacuity in adults (< 0.5 arcmin, Harwerth et al, 1995; O'Dell and Boothe, 1997) to facilitate the comparison. The different data sets were fitted with exponential functions that characterized the relative changes in disparity sensitivity between the maximum value at birth and the minimum value for adults. Relative changes in the median disparity threshold for V2 neurons (and somewhat less in V1 neurons) closely paralleled the behavioral improvement in stereoacuity between 2 and 8 weeks of age. Comparing time constants ($T_c$) is a simple and efficient way to determine
whether the relative changes in perception and cortical physiology are different. The $Tc$ for perceptual changes was 2.92 weeks, which was similar to the $Tc$ for V2 disparity sensitivity (2.60 weeks). The $Tc$ for changes in disparity sensitivity of V1 neurons was slightly longer (4.02 weeks) because at 2 weeks of age disparity sensitivity of V1 neurons was far better than in V2 (Fig 10), i.e., the V1 changes were more moderate than that in V2, not because the functional maturation of disparity mechanisms was delayed in V1. Considered together, the spatial frequency tuning, firing rate and response variability of V2 and V1 neurons in the developing visual brain are likely to play a critical role in limiting binocular vision development because the basic cortical mechanisms in V2 and V1 for binocular combination are adult like as early as 2 weeks of age.

Similar conclusions were reached in previous psychophysical studies in human infants. For example the development of stereopsis was shown to largely depend on the maturation of spatial frequency tuning and contrast sensitivity (Schor, 1985). A more recent study with human infants demonstrated that the most critical factor limiting stereopsis in infants was their poor visual performance due to ‘insensitivities’ to stimulus contrast (Brown et al, 2007). Common to both studies, then, is that binocular vision development is severely constrained by the ‘visibility’ of stimuli. Our recent studies also found that in comparison to adults the contrast sensitivity of infant V2 neurons is substantially lower until 8 weeks of age (Zheng et al, 2007; Zhang et al, 2005).

Conclusions
During early postnatal development, the sensitivity of binocular disparity mechanisms in early cortical processing (V1/V2) is constrained by the relatively coarse spatial frequency tuning, the lower response rate, the low contrast sensitivities and/or the immaturities in the receptive-field center-surround organization of cortical neurons. One or more of these immaturities in turn are likely to limit the development of binocular depth perception. Alternatively, immature cortical mechanisms beyond V2, yet to be discovered, could be another limiting factor. However, the present study unambiguously demonstrates that regardless of the maturational state of extrastriate visual areas beyond V2, immaturities in the response properties of V2 and V1 neurons described in this study are likely to impose substantial constraints on binocular performance of infant primates.

References


Maruko et al


**Figure legends**

**Figure 1.** Methods used to measure the disparity sensitivity of a V2 neuron.  

a. Determination of the preferred orientation from its orientation tuning function fitted with a wrapped Gaussian function.  

b. Determination of optimal spatial frequency and spatial resolution of the unit after fitting its spatial frequency response function with a Gaussian function.  

c. Disparity tuning function of the V2 neuron. Binocular response amplitude was plotted as a function of interocular
spatial phase disparity. BII, *binocular interaction index*. DDI, *disparity discrimination index*, Peak B/M, *peak binocular response amplitude/monocular response amplitude*. Note that the calculation of these values was performed after taking the square root of firing rates. Dotted line indicates the mean binocular response amplitude. Filled triangle, dominant monocular amplitude. Open triangle, non-dominant monocular amplitude. Asterisk, spontaneous activity.

**Figure 2.** Examples of binocular spatial phase tuning functions of V2 (a-d) and V1 neurons (e-f) of 2-, 4-, 8-week-old infants and adults. Plotting conventions are same as in Fig 1c. Note that the disparity tuning function of the V2 neuron at 2 weeks of age was very similar to that for the adult unit except its low mean binocular response rate.

**Figure 3.** Population data for disparity tuning functions in infants and adults. *Open histograms*, the distribution of BII values for all V2 and V1 neurons in infants and adults. *Filled histograms*, the distribution of V2 and V1 neurons that had statistically significant disparity tuning, i.e., disparity sensitive units (one-way ANOVA, p < 0.05, Prince et al, 2002a). Mean (±se) and median values are indicated by triangles and circles respectively. Note that the calculation of the BII values was performed after taking the square root of firing rates.

**Figure 4.** Developmental changes in disparity tuning functions.
Open histograms, the distribution of DDI values for V2 and V1 neurons in infants and adults. Filled histograms, the distribution of DDI values for V2 and V1 neurons that had statistically significant disparity tuning (one-way ANOVA, p < 0.05, Prince et al, 2002a). Mean (±se) and median values are indicated by triangles and circles respectively. Note that the calculation of the DDI values was performed after taking the square root of firing rates.

Figure 5. Frequency histograms illustrating the distribution of peak binocular response over monocular response ratios of V2 and V1 neurons in infants and adults. Mean (±se) and median values are indicated by triangles. The dotted lines indicate the border between excitatory (> 0.0 db) and inhibitory (< 0.0 db) binocular interactions. Note that the calculation of the Peak Binocular/Monocular ratios was performed after taking the square root of firing rates.

Figure 6. Developmental changes in neuronal firing patterns of V2 and V1 neurons. a. The average (±se)(open circles) and median (filled squares) mean binocular response rate of V2 and V1 neurons in infants and adults. Rectangular boxes indicate the range of quartile values. b. The distribution of response variance-to-mean ratios of V2 and V1 neurons in infants and adults. Plotting conventions are same as in a. Note the lower firing rates and variance-to-mean ratios of infant’s units.
Figure 7. Developmental changes in spatial frequency tuning. The average (±se)(open circles) and median (filled squares) optimal spatial frequency of V2 and V1 neurons in infants and adults. Rectangular boxes indicate the range of quartile values. b. The distribution of spatial resolution of V2 and V1 neurons in infants and adults. Plotting conventions are same as in a. Note the lower optimal spatial frequency and resolution of infant’s units.

Figure 8. Procedures for estimating the disparity threshold for a representative V2 neuron. a. Disparity tuning function (solid curve) of a V2 neuron from an adult monkey from which the optimal disparity sensitivity of the cell (dotted curve) was determined by taking its optimal spatial frequency (2.0 c/d) into account. The optimal disparity sensitivity at the steepest portion of the tuning function (2.99 spikes/sec/ arc min at the thick line) is indicated with a large arrow. b. Mean binocular responses (data points) were plotted on log-log coordinate as a function of response variances for the same unit as in a. Each data point indicates one spatial phase disparity including spontaneous firing (the lowest data point). The solid line is the best linear fit. The response variance was estimated from the fit function for the steepest portion of the phase tuning function (dotted lines). The optimal disparity sensitivity was then normalized with its variance of responses to obtain its disparity threshold.
Figure 9. The distribution of optimal disparity sensitivity (spikes/sec/arc min) of V2 and V1 neurons in infants and adults. Mean (±se) and median values are indicated by triangles.

Figure 10. Developmental changes in disparity threshold of V2 and V1 neurons. Mean (±se) and median values are indicated by triangles.

Figure 11. Comparisons of relative developmental changes in neuronal ‘disparity-acuity’ (1/disparity threshold) with improvement in BII/DDI, variance-to-mean ratio, spatial frequency (optimal and resolution), response rate. The median values for these response measures at 2, 4, and 8 weeks of age were normalized to their respective adult values. BII, binocular interaction index, DDI, disparity discrimination index, VMR, variance-to-mean ratio, SR, spatial resolution, SF, optimal spatial frequency, RR, response rate, DA, disparity acuity.

Figure 12. Comparisons of developmental changes between behaviorally measured stereoacuity of infant monkeys (O'Dell and Boothe, 1997) and the disparity threshold (‘neuronal disparity acuity’) of V2 (solid line) and V1 (dotted line) neurons for our infant monkeys. Note that the relative time course of improvement in the disparity threshold of V2 and V1 neurons parallels improvement in behaviorally measured stereoacuity. Data points were fitted with an exponential function, y = (R_{max}-R_{min})\exp(-x/Tc) + R_{min}, where R_{max} is the
maximum value at birth and $R_{\text{min}}$ is the minimum value in adults, $x$ is time in weeks, and $T_c$ is time constant.
<table>
<thead>
<tr>
<th>Orientation (deg)</th>
<th>Spatial Frequency (c/deg)</th>
<th>Spatial Phase Disparity (deg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Response (spikes/sec)</td>
<td>BII= 0.56</td>
<td>DDI= 0.76</td>
</tr>
</tbody>
</table>
Peak Binocular / Monocular (db)

Proportion of units (%)

V2

- 2 weeks
- 4 weeks
- 8 weeks
- adult

V1

- 2 weeks
- 4 weeks
- 8 weeks
- adult

Peak Binocular / Monocular (db)
A

V2

Response (spikes/sec)

V1

Age (weeks)

B

V2

Variance to Mean Ratio

V1

Age (weeks)
Spatial Phase Disparity (deg)  
Response (spikes/sec)  

Disparity Sensitivity (spikes/sec/arc min)  
Optimal SF=2 c/deg  
Optimal Disparity Sensitivity = 2.99

Mean Response (spikes/sec)  
Variance (spikes/sec)^2  
Neuronal Threshold = 2.13 arc min

A

B
Optimal Disparity Sensitivity
(spikes/sec/arc min)

V2

2 week

n=104
0.14
0.13 ± 1.15

4 week

n=84
0.32
0.24 ± 1.19

8 week

n=138
0.65
0.48 ± 1.15

Adult

n=193
1.21
1.16 ± 1.09

n=193
0.1

V1

n=78
0.27
0.20 ± 1.17

n=48
0.28
0.24 ± 1.19

n=83
0.75
0.60 ± 1.20

n=80
0.65
0.89 ± 1.18

Proportion of units (%)

0
0.01
0.1
1
10
0.1
1
10
Optimal Disparity Sensitivity
(spikes/sec/arc min)
Disparity Threshold (arc min)

Proportion of units (%)

V2

V1

2 week

4 week

8 week

adult

n=104

n=78

n=84

n=48

n=138

n=63

n=193

n=80

18.88 ± 1.14

11.91 ± 1.14

10.45 ± 1.16

11.19 ± 1.16

8.43 ± 1.12

5.92 ± 1.17

4.51 ± 1.08

5.26 ± 1.15

5.75 ± 1.15

5.26 ± 1.15

5.49 ± 1.08

10.02 ± 1.16

10.96 ± 1.16

6.90 ± 1.17

8.43 ± 1.17

10.45 ± 1.16

11.19 ± 1.16

11.91 ± 1.14

18.88 ± 1.14
$Y = (R_{\text{max}} - R_{\text{min}})^*e^{(-x / T_c)} + R_{\text{min}}$

- Perception: $T_c = 2.92$ (week)
- V2: $T_c = 2.60$ (week)
- V1: $T_c = 4.02$ (week)