4. DISCUSSION

Differentiation of bipolar cells in the rabbit retina

Development of bipolar cells in the vertebrate retina has been well documented in chicks (Quesada et al., 1981), in ferrets (Miller et al., 1999), and recently in mouse (Morgan et al., 2006). The present study adds another characterization of bipolar cell maturation in the rabbit retina (Fig.6). However, development of rod bipolar cells in the rabbit retina was first reported by Casini et al. (1996). Using protein kinase C to label rod bipolar cells, they showed that the first immunoreactivity of rod bipolar cells was not detected until P6. By using dye injection and gene gun labeling, we could follow development of bipolar cells as early as P0. Although occasionally recognizable bipolar cells can be found in some early mature rabbits at birth, most often morphologically identified bipolar cells are not detected until P1-2. This is in consistent with the ferret study that detection of immunoreactivity is later than direct labeling by dye injection (Miller et al., 1999). Earlier studies indicated that axonal arbors of bipolar cells diffusely ramify in the IPL initially, and then become narrowly stratified throughout developmental stages. Recent studies seem to support that axonal terminals of developing bipolar cells are preferentially determined in either ON or OFF layers of the IPL. Our results apparently showed that bipolar cells have preferred ON or OFF arborizations once they are recognizable as bipolar cells.

Previous electron-microscopic studies have shown that ribbon synapses appear first in the OFF layer, and later in the ON layer, of the IPL during development of the human and monkey (Hendrickson, 1996), cat (Crooks and Morrison, 1989), rabbit (McArdle et al., 1977), and mouse retinas (Olney, 1968a, 1968b). However, the study of ferret bipolar cell development did not find any temporal asymmetry between ON and OFF layer stratification (Miller et al., 1999). In contrast, Sherry et at. (2003) have reported that expression of vesicular glutamate transporter 1 in the mouse retina reveals temporal ordering in development of ON vs. OFF circuits. In the present study, we also observed this temporal asymmetry between ON and OFF layers, in which OFF bipolar cells apparently develop earlier than ON bipolar cells.

Disruption of glutamate-mediated interactions during development prevents the pattern of ganglion cell dendritic stratification (Bodnarenko and Chalupa, 1993; Bodnarenko et al., 1995; Bodnarenko et al., 1999). It is possible that bipolar cells facilitate the remodeling of ganglion cell dendrites by providing spatially localized signals. Our results that morphologically identified bipolar cells can be detected as early as P1-2 suggest the potential roles of bipolar cells on affecting ganglion cell development.

Effect of visual deprivation on bipolar cell maturation

In the present study, we have shown that light deprivation causes a delay for

development of bipolar cells. Interneuron maturation effects by dark-rearing were observed for the first time (Zhang et al., 2005). Bipolar cells have been considered the latest neuron cells in the retinal development, and thus are thought to be influenced by other early-developed retinal neurons. The delay phenomenon of bipolar cells seen in this study indicates that light may promote the maturation of the bipolar cells. APB treatments of developing cat retina cause the ON/OFF segregation of the dendrites of the ganglion cells in IPL delays but not permanently arrest it (Bodnarenko et al., 1995). Under the dark-rearing environment, continuous release of the glutamate by depolarization of the photoreceptors corresponds to the treatment of mGluR6 agonist APB. This result implies that disruption of glutamate releasing by APB or light has a similar effect in the normal circuitry formation.

How the light deprivation caused the delay of the bipolar cells maturation found in the present study is uncertain. It has been demonstrated that light deprivation would affect the IPL circuitry during the developmental stages in turtles (Sernagor and Grzywacz, 1996; Kay et al., 2004), and mice (Tian and Copenhagen, 2003). Besides, ON/OFF segregation of ganglion cells would be affected by interruption of inner retina circuitry (Vistamehr and Tian, 2004). Recent results from our lab (Chan and Chiao, unpublished data) also showed that light deprivation did not disturb the main functions of the DSGC (direct selectivity ganglion cell), but affect receptive field

properties mediated by the inner retina circuitry. It is possible that the fine tuning of the amacrine cells' dendrites may be refined by the axonal terminals of developing bipolar cells. Therefore, our findings may be interpreted as light deprivation make the photoreceptors release glutamate constantly, and bipolar cells delay for refining the amacrine cells, consequently this results in alternations of certain properties of retinal circuitry.