Results

S-cone and M-cone distribution across the rabbit retina

According the previous reports (Sezl et al., 1994; Juliusson et al., 1994; Famiglietti and Sharpe, 1995), the rabbit retina was shown having unequal S cone distribution across the whole retina. Because of using different S cones labeling from previous studies, the cone distribution across the rabbit retina needs to be rechecked, especially the density of the S cone and dual opsin expressing cones.

Retina was divided into four regions: dorsal side, visual streak, ventral side, and blue streak (Fig. 6). Five images of each region (145 × 145μm) were analyzed (Fig. 7). In general, most retinas exhibit a dominance of M cones versus S cones (Table 1): 5 to 1 in the dorsal side, 11 to 1 in the visual streak, and 3 to 1 in the ventral side. In contrast, the ventral most 5% to 6% of the whole retinal area (blue streak) showed a high density of blue cones (11,000/mm^2), where there was little M cone opsin expression. This pattern is identical with previous reports (Sezl et al., 1994; Juliusson et al., 1994; Famiglietti and Sharpe, 1995). But in terms of total cone density, visual streak in our study is apparently lower than in the previous report. For dual opsin expressing cone population, there is a gradient from zero in the dorsal side retina to a high ratio (about 18% of total cone population) in the blue streak (Table 1). This result is also consistent with the report by Sezk et al. (1994).

The cone connection of “common” bipolar cell types

From Kouyama and Marshak (1992) study in primate, the blue cone bipolar cells were showed having similar morphology in different location in the retina. Therefore, in seeking potential blue cone bipolar cells, I only used the dorsal side retina because of the sparse distribution of S cone and the lowest dual opsin expressing cone population. In doing so, I can avoid the problems of chanced specific S cone connection and non-specific S cone connection with opsin co-expressing cones.

I followed the bipolar cell categorization identified in MacNeil et al. (2004). All injected bipolar cells were divided into 12 cone bipolar cell types and 1 rod bipolar cell type (Fig.8 Top). The axonal stratification of the bipolar cell in the IPL was not used as a strict criterion for determining the type of bipolar cells because microinjection in our experiment tends to compress the retina which makes the IPL shortened at the injection side. Therefore, cells were considered to be a single type if all three conditions were met: 1) the dendritic and axon terminal arbors exhibit similar branching patterns and distributions of varicosities; 2) the axonal fields have similar diameters; 3) the dendritic fields have similar diameters. Despite similar cell appearances, the sizes of dendritic and axonal fields tend to have a little difference by their locations in the retina. Therefore, the bipolar cells were categorized by their morphology first when they had similar morphological pattern but a little difference in cell size.

Images were analyzed from 82 “common” bipolar cell types (axonal field diameter is smaller

than 50μm) and 17 wide-field bipolar cells (axonal field diameter larger than 50μm). I obtained at least 2-10 bipolar cells for each type identified in MacNeil et al. (2004). All 12 distinct functional types of cone bipolar cells receive inputs from all cones (regardless M cones or S cones) in their dendritic fields (Fig. 8 Bottom). These results are consistent with previous reports (Boycott and Wassle, 1991; Bloomfield and Dacheux, 2001; and MacNeil et al., 2004).

The cone connection of wide-field bipolar cells

17 wide-field bipolar cells were subdivided into 4 populations (Fig. 9): Blue cones contact only bipolar cells (BB), BB-like bipolar cells (BL), Wide-field bipolar cells whose axonal terminals stratified in the sublamina a (WA), and Wide-field bipolar cells whose axonal terminals stratified in the sublamina b (WB). There were two BB cells with about 60 and 80μm in axonal field diameter and smaller dendritic field diameter (about 40 and 50μm) (Fig. 9A Top). The BB cells have only 4-5 primary dendrites varied in length and no branch. All dendrites contact S-cone pedicles exclusively at their terminal claw structures (Fig. 9A Bottom). The axons of BB cells were stratified in sublamina 1 of the IPL and ended with some varicosities. Thus, they are considered to be OFF cone bipolar cells. Five of BL cells identified have similar morphology as BB (Fig. 9B). The major difference between BB and BL was S cone connectivity. BL cells contact only 1-2 S cones in their 4 or 5 primary dendrites (Fig. 9B Bottom). Furthermore, the axon strata of BL cells tend to have broadly distribution from 10 to 35% of the IPL in some cases. WA cells have similar axon strata

distribution with BB-like bipolar cells but differ in their morphological appearances (Fig. 9C). WA cells have many branches in their dendrites and received all cone inputs in their dendritic fields (Fig. 9C Bottom). The axons of the WA cells have less varicosity in terminal but more loops in process (Fig. 9C Top). The WB cells have similar morphological feathers as WA cells in both dendrite and axon processing but different axonal stratification (Fig. 9D). The axon strata of WB cells were from 50 to 55% of the IPL. The properties of wide-field bipolar cells were summary in Table 2.

The cone connection of type C like horizontal cell

Famiglietti (1990) reported a new type of wide field horizontal cell (named type C), and he suggested that this type C horizontal cells could presumably link to S cones specifically in rabbit retina. The type C horizontal cell exhibits a notable feature: multiple, thick, branching and wide-ranging processes, extending well beyond the dendritic tree. The long dendritic arbors give rise to occasional claw structure to cone pedicle and terminate in a single small knob.

Some horizontal cells I injected have similar features like type C horizontal cell described in Famiglietti (1990). However, the extended long arbors were hard to fill dye completely. It was difficult to determine whether the cell are type C horizontal cell or not with my incomplete injections. Therefore, those cells were named type C like horizontal cell in this study (Fig. 10). The type C like horizontal cells have no cone selectivity as A-type and B-type horizontal cells reported previously (Hack and Peichl, 1999). However, the extended arbors of the type C like horizontal

cells were uncertain about the cone selectivity because of poor image quality.