Dimensional Differences among the Groups of Retinal Ganglion Cells According to the Retinal Zones in Chicks

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ABSTRACT. The populations of retinal ganglion cell (RGC) groups (Groups I, II, III, IV) were similar each other between the central and intermediate zones, but the population in the peripheral zone were clearly different from those in the central and intermediate zones due to increase of Group III and IV cells and decrease of Group I cells. The dimensions of somal area and dendritic field of Group I cells increased very gradually toward the peripheral zone, but those of other three Groups grew steeply in the peripheral zone. The correlation index between somal area and dendritic field of RGCs showed high coefficient in the central (r=0.73) and intermediate (r=0.77) zones, but lowered clearly in the peripheral zone (r=0.64) due to increase of Group III cells, which showed nonlinear relation between somal area and dendritic field.

KEY WORDS: chick retina, dimension, ganglion cell.

The density of the retinal ganglion cells (RGCs) is different in various animals according to the retinal regions such as the central area, dorsal area, and peripheral area. The characteristic pattern of the cell density is closely related to visual behavior of the animals [9]. However, the chick central and dorsal areas are not so clear as found in mammals or some birds [5]. On the other hand, it has been known that RGCs are classified into several types according to somal size and dendritic arborization from fish to mammals, including fish [8, 10], amphibian [15], reptile [11], bird [16], and mammal [4]. Recently, we divided chick RGCs into six groups based on the somal area, dendritic field and dendritic arborization patterns as follows [13]. Group Ic: small somal area, narrow dendritic field, and simple arborization, Group Is: small somal area, narrow dendritic field, and complex arborization, Group IIs: medium-sized somal area, middle-sized dendritic field, and simple arborization, Group IIIs: medium-sized somal area, wide dendritic field, and complex arborization, Group IVc: large somal area, wide dendritic field, and complex arborization [6, 13]. The differences in these morphological factors of RGCs bring about various effects on the visual function. However, it has hardly been reported about relationship between the RGC groups and the retinal regions. In this short communication, we attempted to show the relationship between the morphological classification of chick RGCs and the retinal regions.

Retinal ganglion cells were labeled with fluorescent dyes, Dil (1,1'-dioctadecyl-3,3',3'-tetramethylindocarbocyanine perchlorate; Molecular Probes, Eugene, OR) or dilithium Lucifer Yellow (Polysciences, U.S.A.). Twenty chucks of the White Leghorns were anesthetized by overdose of sodium pentobarbital (45 µg/g body weight). The eyeballs were dissected out from the orbit by cutting the optic nerve very close to the optic chiasm, and some small crystals of Dil were implanted onto a section of the optic nerve stump. The eyeball with optic nerve was incubated at 37°C in 1% paraformaldehyde in 0.1 M phosphate buffer (pH 7.4) for 4 weeks. The RGCs labeled with Dil were examined using a G-filter set (excitation 545 nm, absorption 590 nm, Olympus, Japan) under a fluorescence microscope. In the intracellular injection, the method was the same as this described previously [6, 13]. Briefly, individual RGCs labeled with Fast Blue (Sigma) beforehand were evenly chosen in the same fluorescent microscopic view as far as possible and penetrated with a glass micropipette filled with a 3% Lucifer Yellow in distilled water. Lucifer Yellow was released at a negative current of 5–10 nA square waves from 30 seconds to 2 min through a UV-filter set (excitation 365 nm, absorption 420 nm) (BX 50WI, Olympus, Japan).

Images of 282 labeled RGCs were photomicrographed using a fluorescence microscope (Olympus, Japan) through a UV-filter set (for LY-injected RGC) or a G-filter set (for Dil-labelled RGC). The image data were entered into an image analyzing system (Luzex III, Nireco, Tokyo, Japan) via a CCD-TV camera, and were reported as numerical data in terms of the somal areas and dendritic field. These numerical data were statistically processed with the aid of a computer software package (Excel ver 8.0, Microsoft).

We divided the retina into three zones concentrically, central, intermediate, peripheral zones to examine the changes in the morphological properties of RGCs based on data on density of chick RGCs [5] (Fig. 1D). Almost all of the RGC groups were found in the central (Fig. 1A) and intermediate (Fig. 1B) zones. Group I cells occupied the largest population of RGCs in the central (70.7%, 55 of 75 cells) and intermediate (65.5%, 56 of 85 cells) zones. On the other hand, Group III (about 5%) and Group IV (1–4%) showed very small populations in both zones (a single...
Group IV cell was observed in the central area (6). In the peripheral zone (Fig. 1C), however, the cell population of Group I decreased to about 42.60% (52 of 122 cells), and other groups considerably increased, especially Group III (16.4%, 20 of 122 cells) and Group IV (8.2%, 10 of 122 cells). The constitution of the RGC groups in the peripheral zone was clearly different from those in the central and intermediate zones.

Figure 2 showed the comparison of the sizes of somal area and dendritic field between different RGC groups in three retinal zones. The somal area and dendritic field increased in size toward the peripheral zone in every group. Although the somal size of Group I cells did not show significant difference (P>0.05) between the intermediate and peripheral zones, those of other cell groups became larger obviously within a 5% confidence limitation (P<0.05) in the intermediate and peripheral zones (Fig. 2A). In particular, increasing rate of somal area was obviously high in the peripheral zone. The same trend was also found in the dendritic field (Fig. 2B). Two kinds of these morphological quantities indicate that Group I cells have the tendency to become larger very gradually toward periphery, but other group cells become larger steadily toward periphery, particularly they grow steeply in the peripheral zone.

The correlation index between the somal area and dendritic field of RGCs showed a high value of coefficient of

![Fig. 1. Three circular graphs showing constitution of RGC population in the central zone (A), intermediate zone (B), and peripheral zone (C). Each zone is indicated on the retinal map in D. Note that the RGC population resembles between the central and intermediate zones each other, but they are clearly different in the peripheral zone. Abbreviations: CA: central area, CZ: central zone, DA: dorsal area, IZ: intermediate zone, N: nasal retina, PZ: peripheral zone, T: temporal retina.](image1)

![Fig. 2. Comparisons of the sizes of somal areas (A) and dendritic fields (B) in each group. Group IV cells are not found in the central zone. Note that both sizes become larger toward the peripheral zone in all groups, particularly, Groups III-IV cells become clearly larger in the peripheral zone.](image2)
correlation in the central (r=0.73) and intermediate (r=0.77) zones (Fig. 3A, B), but it lowered clearly in the peripheral zone (r=0.64) (Fig. 3C). It is considered from these data that the high correlation index in the central and intermediate zones is due to high population (70%) of Group I cells with morphological uniformity, and the lower correlation index in the peripheral zone is caused by increase of groups III and IV cells that have wide dendritic field and middle-sized and large somata.

Group I cells correspond very well to the brisk-sustained RGCs judging from their cellular dimension [2]. These RGCs occupy the highest population in the central zone, and shows high morphological uniformity as described above. These facts suggest strongly that Group I cells, particularly, Group Ic cells are very similar to reptile G2-cells [1] and also correspond to X-cells defined physiologically. On the other hand, Group IV cells correspond dimensionally to brisk-transient-linear cells [3]. In addition, Group IV cells were found most frequently in the periphery zone. These data made us presume the correspondence of Group IV cells to Y-cells. In contrast to the Groups I and IV, physiological properties of Groups II and III have not understood sufficiently. However, Group II and III cells display some similar features in terms of the somal area and dendritic field of the subtype of W cells that show various shapes. For instance, Group II cells are similar to the complex RGCs [3, 7] and Group III cells seem to be ε-cells [12, 14] in mammals. These findings suggest strongly that the chick central and intermediate zones have many RGCs exhibiting similar properties in both of morphology and physiology. Consequently, chick retina could possess a wide zone like the central area in mammals.

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