Spatial Properties of Goldfish Ganglion Cells

JOSEPH BILOTTA and ISRAEL ABRAMOV

From the Visual Research Laboratory, Department of Psychology, Brooklyn College of City University of New York, Brooklyn, New York 11210

ABSTRACT We systematically classified goldfish ganglion cells according to their spatial summation properties using the same techniques and criteria used in cat and monkey research. Results show that goldfish ganglion cells can be classified as X-, Y-, or W-like based on their responses to contrast-reversal gratings. Like cat X cells, goldfish X-like cells display linear spatial summation. Goldfish Y-like cells, like cat Y cells, respond with frequency doubling at all spatial positions when the contrast-reversal grating consists of high spatial frequencies. There is also a third class of neurons, which is neither X- nor Y-like; many of these cells' properties are similar to those of the "not-X" cells found in the eel retina.

Spatial filtering characteristics were obtained for each cell by drifting sinusoidal gratings of various spatial frequencies and contrasts across the receptive field of the cell at a constant temporal rate. The spatial tuning curves of the cell depend on the temporal parameters of the stimulus; at high drift rates, the tuning curves lose their low spatial frequency attenuation. To explore this phenomenon, temporal contrast response functions were derived from the cells' responses to a spatially uniform field whose luminance varied sinusoidally in time. These functions were obtained for the center, the surround, and the entire receptive field. The results suggest that differences in the cells' spatial filtering across stimulus drift rate are due to changes in the interaction of the center and surround mechanisms; at low temporal frequencies, the center and surround responses are out-of-phase and mutually antagonistic, but at higher temporal rates their responses are in-phase and their interaction actually enhances the cell's responsiveness.

INTRODUCTION

When studying specific retinal functions, the choice of a species has often been dictated by convenience as much as by the questions being asked. For example, the responses of the goldfish retina to color have been carefully analyzed because of the general similarity of goldfish color vision to that of humans and because of the convenience of the goldfish retina as a physiological preparation. Although the goldfish is often used as a model of vertebrate color vision, the spatial properties of its retina have been somewhat ignored. Much of the work on spatial processing has been carried out on the cat, whose ganglion cells can be divided into three classes (X, Y, and...
W) according to the linearity of their spatial summation (see Enroth-Cugell and Robson, 1984). There have been only a few attempts to conduct similar investigations on goldfish ganglion cells, and the results have often been completely contradictory. For example, some reports claim that all goldfish ganglion cells are linear (Spekreijse and van den Berg, 1971), while others report that the majority are non-linear (Levine and Shefner, 1979).

The present work attempts to resolve these discrepancies as well as to “bring the goldfish up to date” by quantitatively assessing the spatial properties of goldfish ganglion cells using the same criteria and techniques used on cat neurons. Only through this kind of assessment can the spatial summation of goldfish ganglion cells be unequivocally related to spatial summation in other vertebrate retinas. Also, examining the spatial summation properties of the different classes of goldfish ganglion cells may provide valuable information regarding the neuronal mechanisms underlying the organization of the receptive fields; this is similar to the analysis of ganglion cells in the cat retina done by Hochstein and Shapley (1976a, b). From a more general perspective, if this classification scheme exists in evolutionarily diverse species, such as cat and goldfish, then these properties must constitute a basic principle by which visual systems process information and not just a species-specific attribute.

Another aspect of a ganglion cell’s spatial processing is its spatial filtering characteristics. This was examined by measuring each cell’s response to sinusoidal gratings, of various spatial frequencies and contrasts, drifting across the receptive field at a constant temporal rate. Since the general receptive field properties of goldfish ganglion cells are comparable to those of cat ganglion cells, goldfish cells should behave like those of the cat and other species. For example, since the center and surround arrangement of goldfish ganglion cells is similar to other species, they should possess the same spatio-temporal interactions (e.g., Derrington and Lennie, 1982; Enroth-Cugell et al., 1983).

METHODS

Preparation and Recording

Subjects were common goldfish (Carassius auratus) measuring 10–15 cm in length; they were maintained in an aquarium with a water temperature of 21°C, and a 12 h light/dark cycle. 2–4 h before surgery, the fish was placed in a dark-adaptation tank. This facilitated the retraction of the pigment epithelium from the cones, making it easier to separate the retina from the pigment epithelium with minimal damage to the cones. After dark-adaptation, the animal was killed by decapitation, and the eye was enucleated and hemisected. The retina was removed from the eyecup and placed, receptor side up, on a glass plate within the isolation chamber. The isolation chamber was maintained at a constant 17°C throughout the experiments. A steady flow of moist 100% oxygen was passed through the isolation chamber at a constant rate of 75 ml/min (see Abramov and Levine, 1972, for more details).

Extracellular recordings from single ganglion cells were made with glass-insulated platinum-iridium microelectrodes (Wolbarsht and Wagner, 1963) lowered into the retina, from the receptor side; a platinum-iridium indifferent electrode was placed on the edge of the retina. Ganglion cell responses were amplified, monitored with an oscilloscope and audio speaker, and were recorded by a computer for analysis.
Optical Stimulator

The optical system consisted of four independent beams joined by mixing cubes and projected onto the ganglion cell side of the retina. Three of the beams were used to present monochromatic spots and annuli to the receptive field of the cell. (For a complete description of the three spectral beams, see Abramov and Levine, 1972.) These stimuli were used to determine the spectral characteristics of the cell.

The fourth beam was used to produce spatial and temporal stimuli. It consisted of a high resolution oscilloscope (CRT) (model 606, P51 phosphor; Tektronix, Beaverton, OR) which displayed the output of an electronic visual stimulator (Milkman et al., 1978). This stimulator was designed to produce sinusoidal gratings whose spatial frequency (cycles/millimeter on the retina), orientation, contrast and temporal frequency were independently variable. The visual stimulator was also capable of producing a grating whose contrast was reversed as a sinusoidal function of time (i.e., contrast-reversal gratings). The spatial position (spatial phase) of this grating on the retina could also be manipulated. Each pattern was modulated around a mean luminance to maintain a constant adaptational state. The contrast of the stimulus was defined as: (maximum luminance − minimum luminance)/(maximum luminance + minimum luminance). Since the function relating the CRT’s light intensity to Z-input voltage was not linear, a linearizing circuit was added to correct this distortion (Harris and Abramov, 1983). The CRT’s image entered the optical system’s beam splitter and was projected onto the ganglion cell side of the retina by a high quality camera lens. The image was restricted to a 7.5 mm circular field on the retina and produced a retinal illuminance of 0.2 lm/m². The stimulus generator was also capable of restricting the stimulus pattern to a central window or to its surround while the remaining portion of the display was maintained at the same mean luminance.

Procedures

Once a cell was isolated, the CRT display was turned on and a few minutes were allowed for the retina to adapt to the CRT’s illumination. The contrast, spatial frequency and temporal frequency of the contrast-reversal gratings were selected by the experimenter to optimize the responses of the cell. These gratings were then presented at various spatial phases of the cell’s receptive field to establish whether a null point could be found; that is, a position at which the cell responded as if there were no modulated stimulus. To aid the experimenter, an on-line Fourier analysis of the cell’s responses to each stimulus was displayed by the computer.

To determine the cell’s spatial filtering characteristics, sinusoidal gratings of various spatial frequencies, were drifted across the cell’s receptive field at a constant temporal rate (cycles per second crossing the field). Each spatial frequency was presented at several contrasts. Temporal contrast response functions were determined by presenting a contrast-reversal grating of “zero” spatial frequency (i.e., a spatially uniform field whose intensity was sinusoidally modulated in time). The stimulus duration was varied to maintain the same number of stimulus presentations across temporal frequencies. These stimuli were also presented at various contrasts.

When possible, the various parameters of the different types of stimuli were varied. For example, contrast-reversal gratings were presented at several temporal and spatial frequencies to determine their influence on the spatial summation properties of the cell; and drifting gratings were presented at different drift rates to examine the cell’s spatio-temporal interactions.

For some cells, the spectral characteristics were also examined; monochromatic stimuli of 450, 510, and 700 nm (all wavelengths were equated for equal quantal content) were presented for 1 s to the center (0.63 mm diam spot of light) and surround (annulus with inner
and outer diameters of 1.95 and 6.31 mm, respectively) portions of the cell's receptive field. These values were sufficient to stimulate either the center or surround mechanism while minimizing the influence of the other mechanism (Daw, 1968). These three wavelengths were presented at several intensities covering the response range from threshold to saturation in five approximately equal logarithmic steps. The maximum intensity corresponded to $2.75 \times 10^{12}$ quanta/cm$^2$/s. Because the cis peak of the long-wavelength cones (L-cones) is located at ~420 nm, these three wavelengths represent isoabsorption points for the L-cones (Harosi, 1976). Therefore, if only L-cones were present, then the responses at these three wavelengths would be identical. This allowed the experimenter to identify quickly the spectral characteristics of the cell. Cells were classified as spectrally opponent if there was a change in response sign across the three wavelengths (e.g., ON-excitation at one wavelength and OFF-excitation at another). If there was no change in response sign across the wavelengths, then the cell was classified as spectrally nonopponent (see Abramov, 1972). Each cell was also classified as either red-ON, red-OFF, or ON-OFF based on the response to a 700 nm spot presented to the cell's receptive field center (see Mackintosh et al., 1987, for details).

**RESULTS**

**Spatial Summation**

Spatial summation was tested by presenting a contrast-reversal grating at different spatial phases across the receptive field of the ganglion cell. The grating's contrast was reversed according to a specified sinusoidal temporal function. The temporal function was divided into discrete time bins and the cell's responses to each reversal cycle were superimposed to provide the average number of spikes per time bin. These values were converted into spike rates and the response amplitudes of the first ten harmonics were derived from a discrete Fourier transform.

**X-like cells.** For a cell to be classified as X-like, three criteria had to be met: (a) there must have been a spatial position at which there was no response to the contrast-reversal grating (i.e., a null point); (b) when the grating was positioned away from the null point, the response was modulated at the same frequency ($f$) as the contrast-reversal; and (c) the amplitude at $f$ was a sinusoidal function of spatial phase.

27 (21%) of a total of 126 ganglion cells successfully isolated were classified as X-like. Fig. 1 illustrates the presence of a null point and the dependence of the response on spatial phase for one of these cells. The spatial frequency of the grating was 1.52 cy/mm, the contrast was 6%, and the contrast-reversal rate was 4 Hz. The abscissa represents one reversal cycle of the stimulus grating (250 ms). The stimulus cycle was divided into 30 discrete time bins (8.3 ms each), and the response reflects the average rate per time bin. Fig. 1a shows the averaged response of the cell to the stimulus positioned at the midpoint of the receptive field. At this position, there was no response to the grating. As the grating was positioned away from this null point, in either direction, the cell responded at the same temporal frequency modulation as the stimulus' cycle reversal; the response amplitude also increased as the grating was positioned farther away from the null. Note that the temporal phase of the responses (with respect to the stimulus) on one side of the null point was 180 degrees out-of-phase with the responses on the other side of the null point.

Fig. 2 shows the relative amplitudes of the fundamental and second harmonic
components of the response as a function of spatial phase in the same cell as above. The amplitude of the fundamental component was a sinusoidal function of the spatial phase of the stimulus grating. Since the responses on either side of the null point were $180^\circ$ out-of-phase, one side was arbitrarily designated as a positive response and the other side as a negative response. The curve represents the best fit sinusoid.

For X-like cells, the presence of a null point was tested at various spatial frequencies, temporal frequencies, and contrasts; these parameters did not affect the existence of a null point in these cells. The ratio of the second harmonic to the fundamental component ($2f/f$) was a good indicator of the dominant component in the response (Hochstein and Shapley, 1976a). This ratio was always less than one for X-like cells, indicating that the response was dominated by the fundamental component.

**Y-like cells.** A cell was classified as Y-like if a null point could not be found and the cell responded to a contrast-reversal grating at double the temporal frequency...
Figure 2. Relative amplitudes of the first two Fourier components of an X-like cell's responses to a contrast-reversal grating as a function of spatial phase of the stimulus. The cell (C30A) and the stimulus parameters are the same as in Fig. 1. Circles refer to the amplitude of the fundamental component of the response; triangles represent the response amplitude of the second harmonic component. The curve is the best fit sinusoid to the data.

Figure 3. Averaged responses of a Y-like cell (C32A) to two contrast-reversal gratings at various spatial positions on the receptive field. For both spatial frequencies, the contrast-reversal rate was 4 Hz and maximum contrast was at 25%. The values above each graph refer to the position of the grating, in degrees, on the receptive field; zero degrees represents the best estimate of the midposition of the receptive field. The two spatial frequencies were 3.05 cycles/mm (a, c, and e) and 0.38 cycles/mm (b, d, and f). The number of stimulus presentations per graph was 37. The bottom illustrations show one complete contrast-reversal cycle of the stimulus. Spectral classification: red-on center, spectrally opponent.
at all spatial phases. These criteria had to be satisfied at high, but not necessarily at low spatial frequencies. At low spatial frequencies, the responses of the small non-linear subunits found in Y cells are overshadowed by the contributions of the larger center and surround mechanisms (Hochstein and Shapley, 1976a).

53 (42%) of 126 ganglion cells isolated were classified as Y-like. Figs. 3 and 4 show a typical Y-like cell’s response to a contrast-reversal grating at two different spatial frequencies. The gratings were presented at 25% contrast with a reversal rate of 4 Hz. At high spatial frequencies (e.g., 3.05 cy/mm), the cell responded at twice the stimulus temporal frequency at all spatial phases (Figs. 3, a, c, e, and 4 b). However, at low spatial frequencies (e.g., 0.38 cy/mm) a null point was found (Fig. 3 d). This cell had a zero spontaneous rate so there were no spikes recorded at the null point.

(Not all Y-like cells had a zero spontaneous rate; there was no apparent relationship between the cell’s spatial summation class and its spontaneous rate.)

Fig. 4 shows the relative amplitudes of the fundamental and second harmonic components of the response, from the same cell as in Fig. 3, as a function of spatial phase at a high spatial frequency (Fig. 4 b), and the fundamental component amplitude as a function of spatial phase at a low spatial frequency (Fig. 4 a). Positive responses were again arbitrarily assigned to one side of the null position and negative responses to the other side. The curve represents the best fit sinusoid. At high spatial frequencies, the second harmonic component dominated the response. Note that the second harmonic amplitude did not depend on the spatial phase of the
stimulus. Also, there was no phase shift in the second harmonic component with spatial phase; hence all responses were arbitrarily designated as positive. The fundamental component, at high spatial frequencies was very weak; however, it was still a sinusoidal function of spatial phase.

At low spatial frequencies, Y-like cells displayed X-like characteristics (e.g., Fig. 4 a). The fundamental component dominated the response, a null point could be found, and the amplitude of the fundamental component was a sinusoidal function of spatial phase. The second harmonic component of the responses of this cell is not shown since it was somewhat distorted due to the fact that this cell had a spontaneous rate of zero.

The $2f/f$ ratio varied as a function of the spatial frequency of the grating in Y-like cells. At low spatial frequencies, the ratio was less than one. However, as spatial frequency increased so did the $2f/f$ ratio. At high spatial frequencies, this ratio typically reached values of two to three or even higher. Clearly, the mechanisms responsible for the nonlinearities in Y-like cells were responsive primarily to high spatial frequencies. These findings were robust across various temporal frequencies and contrasts, including low contrasts.

W-like cells. Cells that did not satisfy the criteria for X- or Y-like cells were classified as W-like. 46 (37%) out of 126 cells were classified as W-like. Fig. 5 illustrates a W-like cell’s response to a contrast-reversal grating of 1.52 cy/mm. The stimulus was presented at 6% contrast, with a reversal rate of 4 Hz. The responses of this and other W-like cells to a contrast-reversal grating contained both fundamental and second harmonic components. At spatial phases away from the “null” position the fundamental component dominated (Fig. 5, a and c). However, as the spatial phase of the grating approached a point midway between those extremes, a doubling of the response occurred, indicating domination by the second harmonic component (Fig. 5 b). Fig. 5 d shows that the fundamental component was, again, a sinusoidal function of spatial phase; it also shows a relatively strong second harmonic component, compared with X-like cells, across all spatial phases. Once again, positive and negative responses were arbitrarily assigned and the curve represents the best fit sinusoid.

The $2f/f$ ratio of W-like cells was never as high as in Y-like cells. Also, the general response characteristics of W-like cells were independent of spatial frequency. This implies that the mechanisms responsible for the nonlinearities in W-like cells are not the same as in Y-like cells, since Y-like cells behave linearly at low spatial frequencies and W-like cells do not.

Spatial and Temporal Tuning

To examine the spatial filtering characteristics of each cell, sinusoidal gratings were drifted across the receptive field at a constant temporal frequency. For most cells, the sensitivity at each spatial frequency was derived by interpolation on the response vs. contrast curve to find the contrast necessary for a constant response amplitude. However, since most cells responded linearly up to moderate contrasts, responses within the linear range could also be used directly as a measure of sensitivity. To illustrate the linear response range of an X-like cell as a function of stimulus contrast, Fig. 6 shows the amplitude of the fundamental component of the response.
plotted against contrast for each of several spatial frequencies. The values at zero contrast represent the amplitude of the fundamental component to a stimulus with no modulation around mean luminance; this was a measure of the cell’s noise level. Virtually all cells examined, regardless of type, responded linearly up to at least 13% contrast. However, to be strictly correct, functions derived directly from the response measure will be designated as log relative “response” rather than “sensitivity” on the ordinate.

**FIGURE 5.** Averaged responses of a W-like cell (C27A) to a contrast-reversal grating at various positions on the receptive field. The stimulus consisted of a contrast-reversal grating of 1.52 cycle/mm, at 6% contrast, modulated at a rate of 4 Hz. The values above each graph (a, b, and c) refer to the position of the grating, in degrees, on the receptive field; zero degrees represents the best estimate of the midposition of the receptive field. The number of stimulus presentations per graph was 37. The bottom illustration shows one complete contrast-reversal cycle of the stimulus. d shows the relative amplitudes of the Fourier components of the cell’s responses to a contrast-reversal grating as a function of spatial phase. Stimulus parameters were the same as in a, b, and c. Circles refer to the amplitude of the fundamental component; triangles represent the amplitude of the second harmonic component. The curve is the best fit sinusoid to the data. Spectral classification: ON-OFF, spectrally opponent.

*X-like cells.* Since X-like cells display linear spatial summation and the response is dominated by the fundamental component, the amplitude of the fundamental component was used as the response measure. Fig. 7 shows a typical spatial contrast sensitivity function from an X-like cell (the cell is the same as in Fig. 6). The stimulus grating drifted across the receptive field at a rate of 4 Hz. The shape of the function was similar to those obtained in other species: there was a sharp high frequency drop, as well as the low frequency attenuation believed to be due to the mutual antagonism of the center and surround portions of the cell’s receptive field.
FIGURE 6. Response vs. contrast curves of an X-like cell (C23A) at several spatial frequencies. The response measure was the amplitude of the fundamental component. The stimuli were sinusoidal gratings of (a) 0.19, (b) 0.38, (c) 0.76, and (d) 1.52 cy/mm, drifting at 4 Hz. The dotted lines represent the amplitude of the fundamental component when a grating of zero contrast was presented; they are measures of the cell's noise level. Spectral classification: red-ON center, spectrally opponent.

The spatial contrast sensitivity functions obtained from single neurons in the goldfish are qualitatively similar to psychophysical functions from this species (Northmore and Dvorak, 1979). The peak of the psychophysical function is at ~0.3 cy/deg and the acuity limit, or the highest frequency detectable at 100% contrast, is between 1 and 2 cy/deg. To compare the physiological and psychophysical functions, cycles per millimeter on the retina were converted to cycles per degree, using values from a schematic eye of the goldfish (Charman and Tucker, 1973). The appropriate conversion value is ~19 deg/mm (see Bilotta, 1987, for more details).

FIGURE 7. Spatial contrast sensitivity function of the same X-like cell (C23A) shown in Fig. 6. The response measure was the amplitude of the fundamental component. The stimuli were gratings drifting at 4 Hz; the number of stimulus presentations per point was 30. Sensitivity was obtained by interpolation on the response vs. contrast curves to find the contrast necessary for a constant response amplitude.
Extrapolating from the cell's function in Fig. 7, the acuity limit is between 12 and 20 cy/mm. This converts to between 0.6 and 1.0 cy/deg, which is close to the behavioral limit. However, the peak of the cell's function is at ~0.8 cy/mm, which converts to 0.04 cy/deg, well below the behavioral peak.

Fig. 8 shows the spatial contrast response curves of several X-like cells obtained at a drift rate of 4 Hz. At high spatial frequencies, these cells were similar in response; however, two of the cells (Fig. 8, b and d) did not appear to possess any low frequency attenuation. It is worth noting that these two cells also did not possess a surround mechanism; that is, they were spatially nonopponent. This supports the notion that the low frequency attenuation found in neurons is a result of an antagonistic interaction between the center and surround portions of the receptive field.

To examine the spatio-temporal interactions in goldfish ganglion cells, spatial contrast response functions were obtained at different drift rates. The results for one X-like cell are shown in Fig. 9. Functions were obtained for gratings of 13%
contrast drifting at rates of 1, 4, and 8 Hz; all values were normalized with respect to one maximum value. The shape of the spatial tuning curves depended on the drift rate of the stimulus, but only at low spatial frequencies. At high spatial frequencies, the three functions were very similar; the curves deviated only at low spatial frequencies. At the lower drift rates of 1 and 4 Hz, there was a decrease in response to low spatial frequencies. However, at 8 Hz there was less low frequency attenuation.

In cat, the differences in the spatial contrast sensitivity functions across drift rates appear to reflect changes in the temporal interactions between the center and surround mechanisms (Hochstein and Shapley, 1976a, b). To examine the center and surround interactions in more detail, temporal contrast response functions were obtained from the entire receptive field as well as from the center and surround areas separately. Fig. 10 shows typical results. The response measure was the amplitude of the fundamental component; all points were normalized with respect to one maximum value. The full-field stimulus consisted of a spatially uniform circular field (7.5 mm diam) whose intensity varied sinusoidally in time to a maximum of 25% contrast. The “center” function was derived with the temporally modulated stimulus restricted to a 1 mm by 1 mm square while the remaining portion of the field was maintained at the same mean luminance as the center; for the “surround” function, the modulated stimulus was restricted to the surrounding portion of the field with the center square maintained at mean luminance.

It should be pointed out that because the center and surround mechanisms overlap spatially, it is impossible to stimulate the entire area of one without stimulating the other; any spot that stimulates the center mechanism must inevitably stimulate the middle portion of the surround mechanism. The values used here were simply intended to maximize the influence of one area while minimizing the input from the other. As can be seen, at low temporal frequencies, the center and full-field values are similar while the surround values are much lower. But, at these low frequencies the center is somewhat more responsive than the full field, due to the antagonism between the center and surround mechanisms when the entire field is stimulated. However, as temporal frequency increases, the response of the surround mechanism increases, and actually peaks at a higher temporal frequency (8 Hz) than the
center mechanism (2–4 Hz). Thus, the surround is quite responsive at higher temporal frequencies, and in fact, appears to be as responsive as the center at high frequencies. Also, at a temporal frequency of 8 Hz, the cell is more responsive to the full-field stimulus than either the center or surround portions separately; this suggests that, under these conditions, not only is there no center and surround antagonism, but that these mechanisms must be synergistic.

Some of the differences among the above functions can be explained by comparing the differences in phase lag between the center and surround responses. In Fig. 10, each point represents the temporal phase difference, with respect to the stimulus, between the cell’s responses when the middle portion of the field (“center” mechanism) is modulated and when the outer portion of the field (“surround” mechanism) is modulated. In other words, each point is the phase lag of the center mechanism’s response to the stimulus minus the phase lag of the surround mechanism’s response. At low temporal frequencies, the phase lag difference between the center and surround mechanisms is about 180°; thus, they are mutually antagonistic. However, as the temporal frequency of the stimulus increases, the phase lags of the

![Figure 10](image-url)
center and surround mechanisms also change but at different rates. At 8 Hz the center and surround responses are roughly 360° apart and are now in-phase and actually synergistic; their interaction now increases the cell's responsiveness.

**Y-like cells.** Since the spatial summation of Y-like cells is nonlinear, the amplitude of the fundamental component is not always appropriate as a response measure. Although the fundamental component provided a means to compare the linear components of the responses of Y-like and X-like cells, it did not completely describe the response pattern of Y-like cells. To illustrate the presence of a nonlinear response component in Y-like cells, Fig. 11 compares a Y-like cell's spatial contrast sensitivity function based on the fundamental component of the response with one based on the maximum response minus the minimum response (i.e., the peak-to-peak amplitude of the response). The drift rate of the grating was 4 Hz.

The cell's sensitivity values determined from the fundamental component and the maximum-minimum response were similar at low to moderate spatial frequencies, but not at high spatial frequencies. Based on the fundamental component of the response the cell was relatively insensitive to gratings higher than 3.05 cy/mm, whereas the maximum-minimum response shows that the cell was still responsive at these values. The differences no doubt reflect the presence of small nonlinear subunits similar to those found in cat Y cells (Hochstein and Shapley, 1976b). But, since the function for the fundamental component is similar to those shown earlier for X-like cells, it appears that the fundamental components of both X- and Y-like cells are the result of comparable interactions of the responses of the center and surround mechanisms of the receptive field. It is also interesting to note that the spatial resolution, based on the linear response component, of goldfish X- and Y-like cells is similar; this is not the case with cat ganglion cells.

Like X-like cells, the Y-cells' spatial tuning depended on the drift rate of the stim-
ulus grating. Fig. 12 compares the spatial contrast sensitivity functions of a Y-like cell at different stimulus drift rates. The response measure was the amplitude of the fundamental component and all values were normalized with respect to one maximum value. Although the 1- and 4-Hz functions differ in terms of absolute sensitivity (i.e., the cell is more sensitive overall to the temporal rate of 4 Hz) the shape of the two functions is similar (they superimpose on one another). However, there were dramatic changes in the shape of the function at the 16-Hz drift rate, primarily at low spatial frequencies. Increasing the stimulus drift rate decreased the degree of low frequency attenuation in the function.

To examine the temporal properties of the Y-like cells' receptive field mechanisms, temporal contrast response functions were determined for the center, surround, and full field. Fig. 13 shows typical results. As in X-like cells, the center and surround mechanisms are mutually antagonistic at low temporal frequencies; that is, the cell is less responsive to full-field stimulation than to stimulation of the center mechanism alone, and the responses of the center and surround mechanisms are 180° out-of-phase. However, at high temporal frequencies, the center and surround mechanisms appear synergistic; the full-field response values are higher than either the center and surround values. Also, the phase lags of the center and surround mechanisms coincide at the temporal frequencies at which the full-field response is larger (i.e., 16 and 32 Hz).

W-like cells. Unlike X- and Y-like cells, which appeared to be similar in most respects except for the presence of the small nonlinear subunits in Y-like cells,
W-like cells were a class apart. There was so much variability across W-like cells that it is difficult to make general statements about them. For the most part, spatial contrast sensitivity functions could be obtained from W-like cells and many of them had functions that were similar to X-like cells. That is, there were no indications that any W-like cell possessed the small nonlinear subunits found in Y-like cells. The nonlin-
ear responses of W-like cells must be due to some other aspects of the cell's mechanisms. Fig. 14 shows the spatial contrast sensitivity function of a W-like cell. The response measure was maximum-minimum and the stimulus drift rate was 2 Hz. Note that at high spatial frequencies there was no indication of nonlinear subunits like those found in Y-like cells. If present, these would certainly be apparent with the maximum-minimum response (compare with the Y-like cell in Fig. 11). Some, but not all, W-like cells possessed the low frequency attenuation found in cells with an antagonistic surround (as in Fig. 14). As with X- and Y-like cells, the shape of the function for W-like cells with an antagonistic mechanism depended on the drift rate of the stimulus grating.

**DISCUSSION**

**Spatial Summation**

This work has shown that goldfish ganglion cells can be classified as X-, Y-, and W-like based on their spatial summation properties. Using the same criteria as for cat X cells, goldfish X-like cells display linear spatial summation. Goldfish Y-like cells, for the most part, are similar to cat Y cells in their response characteristics; both cat Y cells and goldfish Y-like cells respond to high spatial frequency, contrast-reversal gratings at twice the temporal modulation frequency at all spatial positions. The nonlinearities of goldfish Y-like cells are most likely due to small, nonlinear subunits, as in cat Y cells (Hochstein and Shapley, 1976b); this hypothesis is supported by the fact that the nonlinearity is most apparent at high spatial frequencies where such small subunits are most responsive. Aside from this nonlinearity at high spatial frequencies, the fundamental component of the Y-like cell's response behaves as in the X-like cell, suggesting that the organization of X- and Y-like cell receptive fields is similar, except that Y-like cells possess nonlinear subunits. These findings agree with what is known about cat ganglion cells (Enroth-Cugell and Robson, 1966; Hochstein and Shapley, 1976a, b).

On the other hand, cat Y cells' and goldfish Y-like cells' responses are somewhat different when the contrast-reversal grating consists of low spatial frequencies. For goldfish Y-like cells, a spatial null point can be found if the grating is of low spatial frequency. Cat Y cells, however, display a slight doubling response at the midposition of the receptive field; but, when the grating is shifted away from that position, the response is dominated by the fundamental component, as in goldfish Y-like cells (see Hochstein and Shapley, 1976a). Thus, although the nonlinear behavior of the cells in both species can be best explained by the presence of small nonlinear subunits, there are qualitative differences in the subunits' properties. For the goldfish, at the null position of the center and surround mechanisms at low spatial frequencies, the subunit responses appear to be absent. For cat Y cells, although a "null" position can be found at low spatial frequencies for the center and surround mechanisms, the rectifying subunits are still responsive, producing a small response at double the temporal modulation of the stimulus.

Victor and Shapley (1979) have suggested that the subunits in cat Y cells result from the direct input of bipolar cells. It is possible that differences between the species' subunits may be a reflection of the strength of the surround mechanisms of
their bipolar cells. The surround mechanism's antagonism to the center in goldfish bipolar cells (Kaneko, 1970) is much stronger than that found in the cat (Nelson and Kolb, 1983). Thus, at low spatial frequencies, stimulating both the centers and surrounds of the goldfish's subunits could result in no net response. The surrounds of these subunits respond less to high spatial frequencies, leaving predominately the subunits' center responses which would be like those from cat Y cells.

The relationship between goldfish W-like cells and cat W cells is more difficult to determine. Since it is relatively difficult to isolate and maintain stable responses from cat W cells, their spatial properties have not been studied in as much detail as those of X and Y cells. Also, because of the response variability across W cells in the cat, it is difficult to specify common characteristics, and this category most likely contains a variety of subclasses and characteristics (see Rodieck, 1979). The same appears to be true for the goldfish W-like cells. The reason for the variability among goldfish W-like cells may be the same as for similar cells found in the eel retina (Gordon and Shapley, 1978; Shapley and Gordon, 1978). Many goldfish W-like cells displayed response characteristics similar to the "not-X" cells found in the eel. To account for the responses of these "not-X" cells, Gordon and Shapley proposed a receptive field organization that is different from the center/surround organization of X and Y cells. The receptive fields of these cells consist of two slightly overlapping Gaussian distributions in which one area responds with ON-excitation, the other with OFF-excitation. The degree of overlap of the areas varies from cell to cell, thus accounting for the inconsistency within this classification. It is also interesting to note that some eel "not-X" cells displayed spatial contrast sensitivity functions with low frequency attenuation. Gordon and Shapley (1978) attributed this finding to the possibility of a "silent surround" (see Barlow, 1953). Some of the W-like cells in the goldfish also displayed attenuation at low spatial frequencies. It is quite likely that many goldfish W-like cells and the eel "not-X" cells are similar in their receptive field organization.

The fact that goldfish ganglion cells can be classified by their spatial summation properties confirms the results of Levine and Shefner (1979) and Levine (1982) who found that goldfish ganglion cells could be divided into X-like and not-X-like classes based on their response to a pinwheel of light that was rotated to various positions within the center of the receptive field. The present work has elaborated on their findings by examining the nature of the nonlinearity of the not-X-like cells and by examining the linearity of the entire receptive field. These not-X-like cells can be subdivided into Y- and W-like based on their response properties. Despite the differences in stimuli across the studies, the proportions of the various cell types in both studies are similar. Levine and colleagues found approximately two-thirds (64%) of their cells to be not-X-like. In this study, combining Y- and W-like cells into one "nonlinear" category, 79% of the cells were nonlinear. However, these percentages are much different than those found in the cat retina. Cat X cells constitute ~45% of the ganglion cell population, while Y cells make up ~4–7% of the population (see Rodieck, 1979).

On the other hand, the present findings are in disagreement with the findings of Spekreijse and van den Berg (1971) who found that all goldfish ganglion cells possessed linear spatial summation. The discrepancies between the present study (and
the work of Levine and colleagues) and Spekreijse and van den Berg (1971) probably are related to the type of stimuli used. Spekreijse and van den Berg (1971) presented a checkerboard pattern in which a square was sinusoidally modulated out-of-phase with an adjacent square; they were able to adjust the phase and contrast of each pattern to create a “null” response from the ganglion cell. Our results show that the spatial frequency of the stimulus is crucial in the determination of the cell’s spatial summation properties. For example, although a large checkerboard contains a large number of spatial frequencies, it has most of its power at low spatial frequencies. In this study, it was found that a stimulus consisting of low spatial frequencies was not sufficient to activate the small, nonlinear subunits, and under these conditions, the cell would behave linearly. It is quite possible that the stimulus used in the Spekreijse and van den Berg (1971) study was insufficient to examine the nonlinear subunits found in Y-like cells and thus, these cells would be classified as linear. It is possible, to a first approximation, to compare their stimuli with those used in the present study since they provide some of their checkerboard sizes. In all of their figures in which cells display linear summation, the square widths are 0.35 mm. It has been shown that the fundamental component of a checkerboard lies along its diagonal at 1.41 times the pattern’s spatial frequency (see Kelly, 1976). Assuming one square represents one-half cycle and performing the necessary calculations, one obtains a value of 2.02 cy/mm. In the present study (e.g., Fig. 11), to clearly identify a Y-like cell, a grating of 3.05 cy/mm was necessary (although occasionally a cell would show hints of nonlinearity to a grating of 1.52 cy/mm). In one example where Spekreijse and van den Berg used counterphase-modulated, monochromatic, 0.23 mm squares, they could not null the cell’s response. A 0.23 mm square converts to 3.07 cy/mm, a spatial frequency that would most likely activate a Y-like cell’s nonlinear subunits. But, this explanation does not account for the absence of W-like cells in their findings. Perhaps it was due to the difficulty in identifying these cells. Using contrast-reversal gratings, we found W-like cells very difficult to classify, since they possessed X-like properties at some spatial positions but not others.

In summary, it appears that the differences among the studies of spatial summation of goldfish ganglion cells are due to the nature of the stimuli and the criteria used for determining linearity. One purpose of this study was to examine spatial summation processing in goldfish ganglion cells using the same stimuli and criteria as used in work in other species, primarily cat. By using these techniques, we found that spatial summation processing in goldfish ganglion cells is similar to that of cat neurons. Regarding the relationship between a cell’s spatial summation class and its spectral properties, there was none. That is, an X-like cell could be a red-ON or red-OFF center cell and spectrally opponent or nonopponent. This relationship between the cell’s spatial and spectral properties will be examined in detail in a later paper.

Spatial Tuning

The spatial filtering characteristics of goldfish ganglion cells are at least qualitatively similar to those of neurons in cat and monkey (Kaplan and Shapley, 1982). In most cases, the spatial filtering appears to be bandpass in that the neuron is most sensitive
to middle spatial frequencies and less sensitive to higher and lower frequencies. At low spatial frequencies (and low temporal frequencies) the cell's response is presumed to be the result of mutual antagonism between the center and surround portions of the receptive field. However, we have shown that the center and surround, under certain conditions, may be synergistic. This is demonstrated by examining the relative phase lags of the responses of center and surround mechanisms to a temporally modulated stimulus. At low temporal frequencies, the center and surround mechanisms are out-of-phase; thus, stimulating both areas produces a mutual antagonism. However, as the temporal frequency of the stimulus is increased, the phase lags of both the center and surround mechanisms also increase, but at different rates. It is possible, and was always found to be the case in this study, that there could be a temporal frequency at which the center and surround responses are in-phase, resulting in an increase in response when the entire receptive field is stimulated.

The phenomenon of center and surround interactions changing with temporal frequency has been demonstrated in *Limulus* (Ratliff et al., 1969), in cat lateral geniculate nucleus (LGN) and ganglion cells (Kaplan et al., 1979), and monkey ganglion cells (Gouras and Zrenner, 1979). This also explains the variation in the shape of spatial tuning curves at different stimulus drift rates (Enroth-Cugell et al., 1983). At high spatial frequencies, there is little or no change in the function with stimulus drift rate (at these frequencies there is only the response of the center—there is no interaction of center and surround). The most dramatic change in the shape of the function occurs at low spatial frequencies (stimuli to which both center and surround mechanisms are responsive), where increasing the temporal frequency rate of the stimulus produces less attenuation. Similar spatio-temporal interactions have been found in cat ganglion cells (Derrington and Lennie, 1982; Enroth-Cugell et al., 1983; Frishman et al., 1987) as well as macaque LGN neurons (Derrington and Lennie, 1984).

Although the spatial contrast sensitivity functions obtained from single neurons are similar in shape across the different species, there are some differences. The most important difference is in the range of spatial frequencies to which the neuron is sensitive. Compared with the cat and monkey functions, the goldfish function is shifted to much lower spatial frequencies, implying that the goldfish has poor acuity and is unable to detect fine detail in its environment. This is not surprising since the goldfish typically lives in murky water.

The high spatial frequency limit of the goldfish ganglion cells agrees with the psychophysically determined function of the goldfish (Northmore and Dvorak, 1979). However, there is a large discrepancy between the peak sensitivities of the psychophysical and physiological measures. The peak of the physiological curve is shifted to much lower spatial frequencies than the behavioral function. It is not entirely clear why there is this discrepancy between the two measures. One possibility is that the differences between the functions are a result of the stimulus used to derive the spatial contrast sensitivity functions. The psychophysical functions were determined with static, sinusoidal gratings while the ganglion cells' functions were derived from responses to drifting sinusoidal gratings.

Another apparent difference between the goldfish and other animals is in the
similarity of the acuity limits of X- and Y-like cells. In cat ganglion cells at any given eccentricity (Linsenmeier et al., 1982), the differences in acuity limit between X and Y cells are much larger than in the goldfish. The spatial resolution of a ganglion cell depends on the size of its receptive field center (Cleland et al., 1979). It is hypothesized that in the area centralis of the cat (where neurons have the smallest receptive fields), direct input from one bipolar cell constitutes the receptive field center of a ganglion X cell. Moving away from the area centralis, the receptive field centers become larger and therefore must receive more than one bipolar cell input, resulting in poorer spatial resolution. However, the goldfish retina possesses no area centralis and is roughly uniform throughout (Schellart, 1973). Thus, each goldfish ganglion cell probably receives a similar number of direct bipolar cell inputs to its receptive field center. The spatial resolution of each ganglion cell is too poor to be the result of a single bipolar cell input to its center. Therefore, goldfish X-like cells are probably most similar to the X cells found in the cat peripheral retina which must also have multiple bipolar cell input to their center mechanism. What is interesting is that, unlike the cat Y cells, the goldfish Y-like cells appear to have the same size center as the X-like cells. Thus, although the spatial resolution of the linear components of goldfish X- and Y-like cells is similar, the nonlinear subunits of Y-like cells, which most likely consist of single bipolar cell input (see above), are capable of resolving finer gratings than X-like cells.

This work is based on a dissertation submitted by J. Bilotta in partial fulfillment of the requirements for a Ph.D. degree from the City University of New York, New York. The authors would like to thank Drs. James Gordon, Elizabeth A. Lemerise, and Maureen K. Powers, and Paul J. DeMarco for their comments and suggestions.

This work was supported by grants from the National Institutes of Health Eye Institute (EY-01697), and from the PSC-BHE Research Award Program of the City University of New York (11188, 665192, and 666344). J. Bilotta was supported by National Institutes of Health Eye Institute grant EY-06088 during the final preparation of this manuscript.

Original version received 19 October 1987 and accepted version received 10 November 1988.

REFERENCES

Abramov, I. 1972. Retinal mechanisms of colour vision. In Handbook of Sensory Physiology: Volume VII. Physiology of Photoreceptor Organs. M. G. F. Fuortes, editor. Springer-Verlag, Berlin 567–607 pp.

Abramov, I., and M. W. Levine. 1972. The effects of carbon dioxide on the excised goldfish retina. Vision Research. 12:1881–1895.

Barlow, H. B. 1953. Summation and inhibition in the frog’s retina. Journal of Physiology. 119:69–88.

Bilotta, J. 1987. Spatial and spectral properties of the goldfish retina. Ph.D. thesis. City University of New York, New York. 306 pp.

Charman, W. N., and J. Tucker. 1973. The optical system of the goldfish eye. Vision Research. 13:1–8.

Geland, B. G., T. H. Harding, and V. Tulunay-Keeseey. 1979. Visual resolution and receptive field size: examination of two kinds of cat retinal ganglion cell. Science. 205:1015–1017.
Daw, N. W. 1968. Colour-coded ganglion cells in the goldfish retina: extension of their receptive fields by means of new stimuli. Journal of Physiology. 197:567–592.

Derrington, A. M., and P. Lennie. 1982. The influence of temporal frequency and adaptation level on receptive field organization of retinal ganglion cells in cat. Journal of Physiology. 333:343–366.

Derrington, A. M., and P. Lennie. 1984. Spatial and temporal contrast sensitivities of neurones in lateral geniculate nucleus of macaque. Journal of Physiology. 357:219–240.

Enroth-Cugell, C., and J. G. Robson. 1966. The contrast sensitivity of retinal ganglion cells of the cat. Journal of Physiology. 187:517–552.

Enroth-Cugell, C., and J. G. Robson. 1984. Functional characteristics and diversity of cat retinal ganglion cells: basic characteristics and quantitative description. Investigative Ophthalmology and Visual Science. 25:250–267.

Enroth-Cugell, C., J. G. Robson, D. E. Schweitzer-Tong, and A. B. Watson. 1983. Spatio-temporal interactions in cat retinal ganglion cells showing linear spatial summation. Journal of Physiology. 341:279–307.

Frischman, L. J., A. W. Freeman, J. B. Troy, D. E. Schweitzer-Tong, and C. Enroth-Cugell. 1987. Spatiotemporal frequency responses of cat retinal ganglion cells. Journal of General Physiology. 89:599–628.

Gordon, J., and R. M. Shapley. 1978. Contrast sensitivity and spatial summation in frog and eel retinal ganglion cells. In Visual Psychophysics and Physiology. J. C. Armington, J. Krauskopf, and B. R. Wooten, editors. Academic Press, New York. 315–329.

Gouras, P., and E. Zrenner. 1979. Enhancement of luminance flicker by color-opponent mechanisms. Science. 205:587–589.

Harosi, F. I. 1976. Spectral relations of cone pigments in goldfish. Journal of General Physiology. 68:65–80.

Harris, C. M., and I. Abramov. 1983. Linearizing the z-axis of an oscilloscope display. Behavior Research Methods and Instrumentation. 15:662.

Hochstein, S., and R. M. Shapley. 1976a. Quantitative analysis of retinal ganglion cell classifications. Journal of Physiology. 262:237–264.

Hochstein, S., and R. M. Shapley. 1976b. Linear and nonlinear spatial subunits in Y cat retinal ganglion cells. Journal of Physiology. 262:265–284.

Kaplan, E., and R. M. Shapley. 1982. X and Y cells in the lateral geniculate nucleus of macaque monkeys. Journal of Physiology. 300:125–143.

Kelly, D. H. 1976. Pattern detection and the two-dimensional Fourier transform: flickering checkerboards and chromatic mechanisms. Vision Research. 16:277–287.

Levine, M. W. 1982. Retinal processing of intrinsic and extrinsic noise. Journal of Neurophysiology. 48:992–1010.

Levine, M. W., and J. M. Shefner. 1979. X-like and not X-like cells in goldfish retina. Vision Research. 19:95–97.

Linsenmeier, R. A., L. J. Frishman, H. G. Jakiela, and C. Enroth-Cugell, 1982. Receptive field properties of X and Y cells in the cat retina derived from contrast sensitivity measurements. Vision Research. 22:1173–1183.

Mackintosh, R. M., J. Bilotta, and I. Abramov. 1987. Contributions of short-wavelength cones to goldfish ganglion cells. Journal of Comparative Physiology A. 161:85–94.
Milkman, N., R. Shapley, and G. Schick. 1978. A microcomputer-based visual stimulator. *Behavior Research Methods and Instrumentation*. 10:539–545.

Nelson, R., and H. Kolb. 1983. Synaptic patterns and response properties of bipolar and ganglion cells in the cat retina. *Vision Research*. 23:1183–1195.

Northmore, D. P. M., and C. A. Dvorak. 1979. Contrast sensitivity and acuity of the goldfish. *Vision Research*. 19:255–261.

Ratliff, F., B. W. Knight, and N. Graham. 1969. On tuning and amplification by lateral inhibition. *Proceedings of the National Academy of Sciences*. 62:733–740.

Rodieck, R. W. 1979. Visual pathways. *Annual Review of Neuroscience*. 2:193–225.

Schellart, N. A. M. 1973. Dynamics and statistics of photopic ganglion cell responses in isolated goldfish retina. Ph.D. thesis. University of Amsterdam, Netherlands.

Shapley, R. M., and J. Gordon. 1978. The eel retina: ganglion cell classes and spatial mechanisms. *Journal of General Physiology*. 71:139–155.

Spekreijse, H., and T. J. T. P. van den Berg. 1971. Interaction between colour and spatial coded processes converging to retinal ganglion cells in goldfish. *Journal of Physiology*. 215:679–692.

Victor, J. D., and R. M. Shapley. 1979. The nonlinear pathway of Y ganglion cells in the cat retina. *Journal of General Physiology*. 74:671–689.

Wolbarsht, M. L., and H. G. Wagner. 1963. Glass-insulated platinum microelectrodes: design and fabrication. In *Medical Electronics*. H. Bostem, editor. University of Liege Press, Liege. 510–515.