J. Physiol. (1986), **375**, *pp.* 121–135
With 6 text-figures
Printed in Great Britain

INTERACTION BETWEEN THE SOMA AND THE AXON TERMINAL OF RETINAL HORIZONTAL CELLS IN *CYPRINUS CARPIO*

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(*Received 19 June 1985*)

SUMMARY

1. Intracellular recordings were made from the monophasic horizontal cells of the carp retina which are known to respond with a sustained hyperpolarization to all visible monochromatic light. The receptive field of each subcellular structure, the soma and the axon terminal, was determined using a long narrow slit of light.

2. Somata and axon terminals showed receptive fields that encompassed almost the entire retina. This observation suggests that each aggregate of the subcellular parts forms a synctial structure. However, with increasing distance from the slit, the response peak decayed more steeply in somata than in axon terminals.

3. The spatial decline of the peak consisted of two exponential functions in somata, while a single exponential function in axon terminals.

4. The length constant of the axon terminal was similar to the larger length constant revealed in the soma. This finding suggests an electrical communication at work between the soma and the axon terminal.

5. A quantitative account was made in light of a discrete resistive network model which consists of a pair of syncytia coupled through connecting axons; one represents the contiguous layer of somata and the other the contiguous layer of axon terminals. Relevant response properties computed from the model analysis were in satisfactory agreement with experimental data.

6. It was concluded that the soma and the axon terminal of the horizontal cell are electrically connected in the cyprinid retina.

INTRODUCTION

The horizontal cell of the vertebrate retina consists of a soma and an axon terminal which are connected by a slender axon (Boycott & Kolb, 1973; Mitarai, Asano & Miyake, 1974; Stell, 1975; Nelson, Lützov, Kolb & Gouras, 1975; Leeper, 1978*a*; Gallego, 1982). Each soma and axon terminal is electrically coupled with its neighbours (Yamada & Ishikawa, 1965).

In a number of animal species, such as reptiles and mammals, the soma and the

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axon terminal are believed to be electrically isolated and function independently. Both morphological and physiological studies have shown that these two structures contact different sets of photoreceptors (Kolb, 1974; Nelson *et al.* 1975; Leeper, 1978*b*; Wässle, Boycott & Peichl, 1978; Leeper & Copenhagen, 1979). Furthermore, cable-theoretic analysis of the thin and long axon revealed little possibility of electrotonic signal spread from one end to the other (Nelson *et al.* 1975; Ohtsuka, 1983). Contrary to these animals, axon terminals of teleost fish horizontal cells have no synaptic contacts with photoreceptors (Stell, 1975). Despite the lack of photoreceptor inputs, axon terminals of fish horizontal cells show light-evoked responses which are nearly identical in shape and amplitude to those recorded in somata (Mitarai *et al.* 1974; Weiler & Zettler, 1979). They differ only in the amount of spatial summation (Kaneko, 1970; Marmarelis & Naka, 1972). The most likely explanation is that in fish, in contrast to other animal species, the soma and the axon terminal are connected electrically by the axon. However, it is not well understood how this transmission takes place without much decrement. To interpret the spread of electrical signals, Weiler & Zettler (1979) proposed a hypothesis that the axon and the axon terminal have a voltage-dependent amplifying property which compensates for the decrement of signals during tonic spread.

More recently, it was found that the axon of the carp horizontal cell has abundant gap junctions with axon terminals of other horizontal cells immediately after the axon leaves the soma (Kouyama, Watanabe & Shimatani, 1984). This finding provides a possibility that the electrical signal generated in the soma can spread and converge to axon terminals without passing through the entire length of the axon. To evaluate this possibility, the spatial properties of the carp horizontal cell were measured and analysed in terms of a resistive network model. The results strongly suggest that these two structures have an electrical connexion.

METHODS

Preparation

Experiments were performed on the isolated retina of the carp, *Cyprinus carpio*. After the eye was excised from pithed light-adapted fish, it was opened at the equator and the frontal half was removed. The whole eyecup was placed in a chamber vitreous side down. The sclera was separated from the choroid, and the choroid-pigment epithelium complex was detached from the retina. The isolated retina, about 13 mm in diameter, was spread flat on the bottom of the chamber, receptor side up, and was washed of debris by physiological saline: (in mM) NaCl, 116; KCl, 2; $CaCl_2$, 2; $MgCl_2$, 1; $NaHCO_3$, 10; dextrose, 20 and pH 7·6. After the saline was drained off, the chamber was supplied with a moist gas mixture of 95 % O_2 and 5 % CO_2. The experiments were performed from 30 min to 2½ h after the isolation. During the experiment, the retina was maintained under a photopic condition by a steady white background illumination of 2·0 μW cm^{-2}, which was equivalent to 620 nm monochromatic light of $2{\cdot}0 \times 10^{12}$ photons cm^{-2} s^{-1} in producing an equal amount of hyperpolarization in monophasic horizontal cells.

Recording and cell identification

Intracellular recordings were made from monophasic horizontal cells with 3 M-K-acetate-filled glass micro-electrodes (resistance 80–150 MΩ). The monophasic horizontal cells were characterized by hyperpolarizing responses to monochromatic light flashes of all visible wave-lengths (Mitarai *et al.* 1974). No rod-driven horizontal cells were recorded in these photopic retinae. Since somata of the established three (mono-, bi- and triphasic) types of cone horizontal cells and axon terminals

are arranged in ordered sublayers in the inner nuclear layer, the recording sites can be identified from the electrode depth of a single penetration and the spectral response (Mitarai *et al.* 1974). Thus, in a single penetration from the receptor side, the electrode penetrates the soma of a monophasic cell first, the soma of either a biphasic or a triphasic cell next, and finally the axon terminals (of several types in series). Therefore, the most scleral monophasic unit recorded was identified to be the soma, and those recorded more vitreal to the bi- or triphasic cells were identified as the axon terminal. Forty-two somata and nineteen axon terminals (twelve pairs of these were recorded in single penetrations, see Table 1) were analysed.

Signals were stored on FM magnetic tapes (TEAC R-410, frequency range 0–2 kHz ± 1 dB at a tape speed of 15·24 cm s^{-1}) and the reproduced signals were later sampled at 1 kHz by a 12-bit analog to digital converter after passing through a low-pass filter (cut-off frequency of 60 Hz). Response amplitudes were measured as the voltage difference between the response peak and the voltage at the stimulus onset.

Light stimulus

A dual-beam optical stimulator equipped with a 500 W xenon arc lamp was used in the present study. Monochromatic light was obtained by narrow band width interference filters after the infra-red radiation was blocked by a heat filter. The intensity of the light stimulus was adjusted by neutral density filters. The maximum intensity of the monochromatic light was $3{\cdot}9 \times 10^{14}$ photons cm^{-2} s^{-1} in all wave-lengths and that of white light was 150 μW cm^{-2}. A narrow slit of light (0·3 × 4·0 mm on the retina) was projected on to the retina. The stimulus was moved by changing the position of the projection lens set 11·2 cm above the retinal surface.

Data analysis

Horizontal cells of the same response types are considered to form a single sheet of syncytial cells (Teranishi, Kato & Negishi, 1982; Kaneko & Stuart, 1984). If the coupling is uniform and resistive, the spatial distribution of the response to a spot of light is equivalent to the potential spread from a point source, as predicted by a Bessel function (Naka & Rushton, 1967; Lamb, 1976). In the case of a slit of light with infinite length, however, the current spread in the syncytium can only be at right angles to the slit and the response distribution is given by a single exponential function along the transverse axis of the slit. In this case, it is relatively easy to estimate the tightness of coupling by the length constant. Practically, this condition was simulated by Lamb (1976) using a long slit of light (0·08 × 2·33 mm). In the present study, a slit of light (approximately 1·7 times longer than that used by Lamb) was used to analyse the spatial properties of the horizontal cell response. The slit was initially centred on the impaled cell under the microscope and then displaced along the transverse axis in 0·1 mm steps. 10 ms flashes were given as stimuli at 2 s intervals. After the slit travelled to a point 2·0 mm from the recording site, it was moved back in the opposite sequence of steps to confirm the stability of recordings. The slit was moved to both sides to examine the symmetry of the receptive field. In most experiments, dim ($3{\cdot}9 \times 10^{12}$ photons cm^{-2} s^{-1}) 620 nm monochromatic light was used. The centred slit evoked hyperpolarizing responses of 4–6 mV in somata and 2–3 mV in axon terminals. Diffuse illumination at this intensity evoked hyperpolarizing responses of 10–15 mV which were one-fourth to one-third of the saturating response. The response amplitude was kept small to minimize changes in voltage-dependent membrane conductances (Byzov & Shura-Bura, 1983; Tachibana, 1983).

RESULTS

Horizontal cell responses to a slit placed at various distances

A flash of the slit evoked hyperpolarizing responses both in the soma and the axon terminal. As illustrated in Fig. 1, the response amplitude decreased as the distance between the slit and the recording site was increased. The two structures of the horizontal cell showed different spatial properties. When the response amplitudes obtained in a single electrode track were compared for the soma and the axon terminal, the response amplitude of the soma was larger than that of the axon

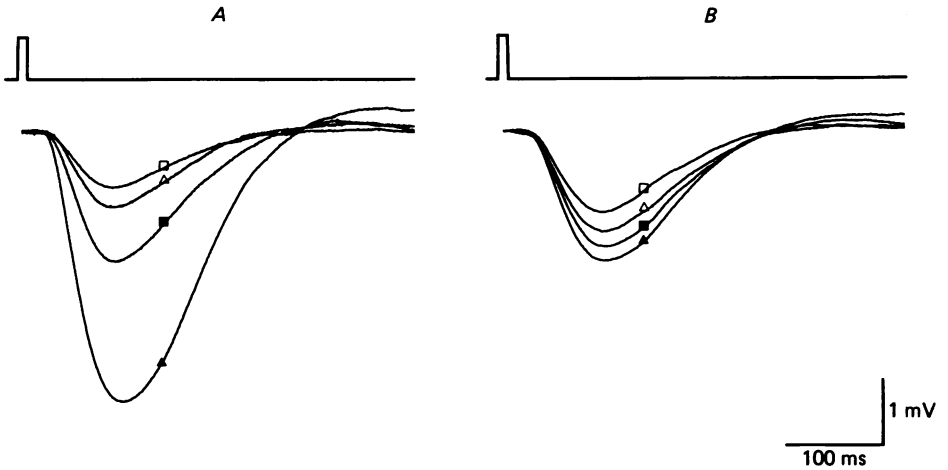

Fig. 1. Responses recorded from the horizontal cell soma (*A*) and from the axon terminal (*B*) to a slit placed at various distances from the recording site. *A* and *B* were recorded sequentially during a single penetration. A light slit (0·3 mm × 4·0 mm, 620 nm monochromatic light of $3{\cdot}9 \times 10^{12}$ photons cm^{-2} s^{-1}) was first centred on the impaled cell (0 mm; filled triangles) and flashed (duration 10 ms, indicated above the responses), and then moved transversely by 0·2 (filled squares), 0·4 (open triangles) and 0·6 mm (open squares) as indicated on each response trace and flashed again.

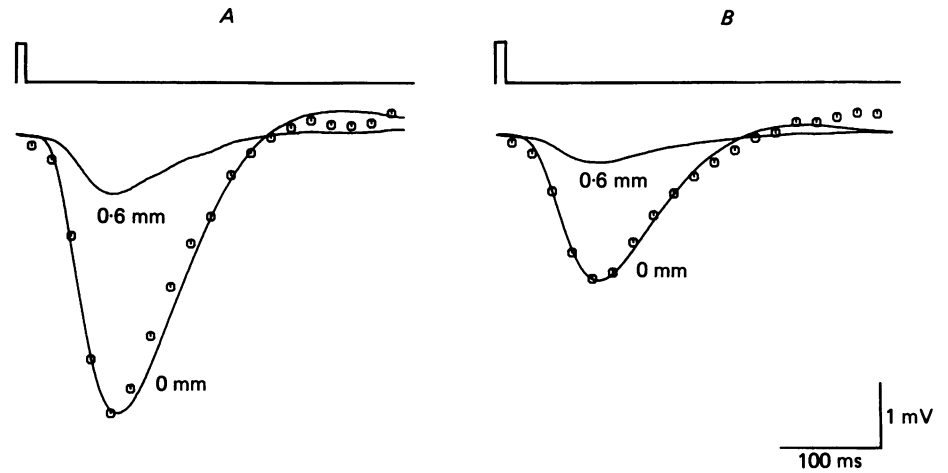

Fig. 2. Responses recorded from the horizontal cell soma (*A*) and from the axon terminal (*B*) to a slit placed at the centre and at 0·6 mm from the recording site. The open circles represent the responses to the 0·6 mm displaced stimulus but amplified by 4·6 (soma) and 5·1 (axon terminal) so that the peak amplitude becomes equal to that of the response to the centred slit.

terminal at the centre (0 mm) and at 0·2 mm. However, at > 0·4 mm, this relation was reversed; the response amplitude of the axon terminal exceeded that of the soma.

Centred slits and laterally displaced slits evoked responses of nearly identical wave forms (Fig. 2). This was observed in the soma as well as in the axon terminal. In this Figure, the response to the stimulus laterally displaced by 0·6 mm was amplified

(illustrated by open circles) so that the response peak became equal to that of the response evoked by the centred stimulus. The similarity of the response wave form was particularly strong during the rising phase (from the response initiation to the peak) of each response. A small deviation seen during the recovery phase may be due to an activation of some non-linear components, but it was not analysed further in the present study since the difference was small. The response wave form of the soma and the axon terminal was also very similar. These observations provide a basis to analyse the horizontal cell syncytia as resistive networks.

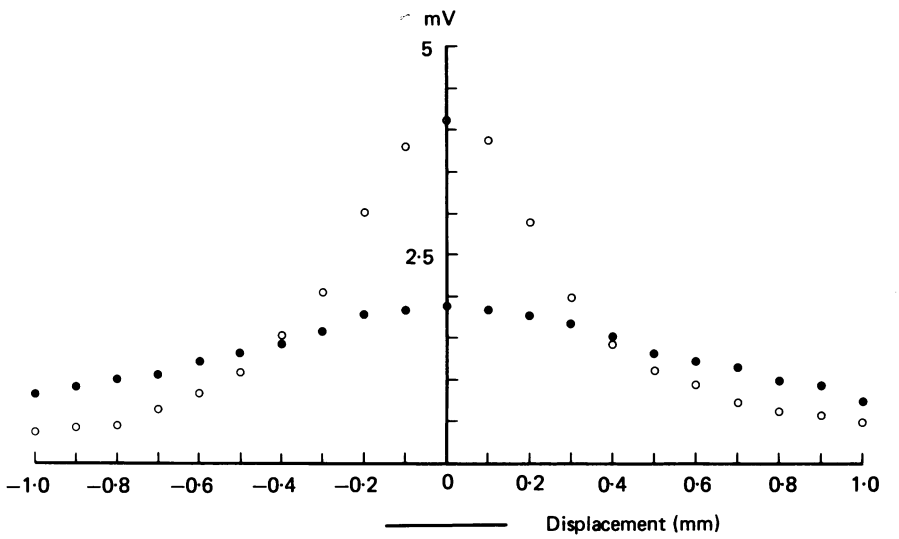

Fig. 3. The peak response amplitude of the soma (open circles) and the axon terminal (filled circles) as a function of distance from the centre of the slit. The slit was moved in 0·1 mm steps. The horizontal bar below the abscissa indicates the slit width.

Response peak as a function of distance from the slit

The decline of the peak response amplitude showed a clear difference between the soma and the axon terminal (Fig. 3). The response amplitude of the soma decreased steeply with distance, reaching 1/e of the peak value evoked by the centred stimulus at approximately 0·4 mm. The decay was almost symmetrical in both directions. On the other hand, the response amplitude of the axon terminal decayed less steeply than that of the soma. Approximately 1·0 mm was needed for response to decay to 1/e of the response to the centred slit. The decay was also symmetrical. These observations suggest that axon terminals couple more tightly than somata.

Analysis by a one-dimensional network

The length constant of the syncytium was measured on the assumption that current flows only in a transverse direction from the slit. In Fig. 4, the response peaks were plotted as a function of distance from the edge of the slit in semilogarithmic ordinates. The decline of the response of the soma (Fig. 4*A*) could not be fitted by a line with a single slope. Obviously, the slope consists of two components, a steep one near the

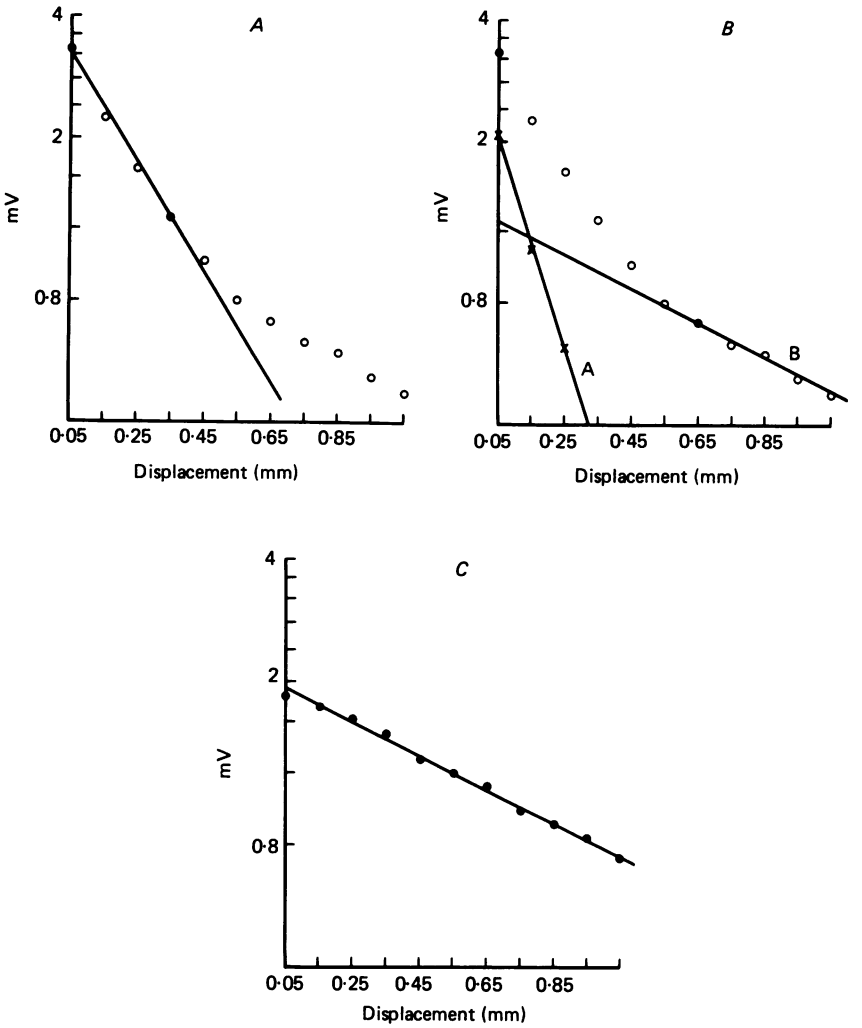

Fig. 4. Comparison between the spatial decline of the peak response amplitude with a single exponential decay. The peak response amplitudes were plotted as a function of distance from the edge of the slit in semilogarithmic ordinates. Responses of the soma and of the axon terminal were obtained in a single penetration. The straight lines were drawn by eye. *A*, the decline of the response peak of the soma could not be fitted by a single straight line. *B*, the decline slope of the response of the soma could be fitted by two straight lines with different gradients. First, line B was drawn by using data points from 0·55 mm to 1·05 mm. Line A was drawn to fit to the difference between the data points and line B (crosses). *C*, the decline of the axon terminal was fitted by a single straight line.

slit and a flat one far from the slit (Fig. 4*B*). To estimate the flat component, a straight line (line B) was fitted by eye to data points in the far region from the slit (0·55 mm ~ 1·05 mm). The steep component was obtained from the differences between the data points and the extrapolated line B in the near region from the slit. The calculated steep component (crosses) could be fitted by another straight line (line A). This procedure was applied to twelve pairs of cell responses recorded during single electrode penetrations (Table 1).

TABLE 1. Comparison of length constants between the soma and the axon terminal (a.t.) and ratio of response amplitude to diffuse illumination. Each of twelve pairs of cell responses was recorded in a single penetration. Length constants of the soma and the axon terminal were obtained by fitting a straight line by eye to data points plotted in semilogarithmic ordinates. Response amplitudes to diffuse illumination (10 mm in diameter) of dim ($3{\cdot}9 \times 10^{12}$ photons cm^{-2} s^{-1}) 620 nm monochromatic light and their ratio are also described. (N.e.: not examined)

	Length constants (mm)				
	Soma			Response amplitude to diffuse light	Ratio (%)
Pair No.	Line A	Line B	A.t.	(a.t. *vs.* soma (mV))	(a.t./soma)
1	0·19	0·99	0·83	12·7/11·2	113
2	0·18	0·81	0·94	N.e.	—
3	0·13	1·15	1·16	12·5/14·5	86
4	0·16	0·71	0·92	N.e.	—
5	0·18	0·91	1·13	N.e.	—
6	0·26	0·83	0·97	13·0/13·5	96
7	0·21	0·87	1·24	15·7/15·3	103
8	0·13	1·01	1·06	13·8/14·8	93
9	0·12	0·78	1·23	N.e.	—
10	0·16	0·82	1·21	N.e.	—
11	0·13	0·72	0·79	N.e.	—
12	0·17	1·13	1·10	12·7/10·6	120
Mean ± S.D.	0·17 ± 0·04	0·89 ± 0·14	1·05 ± 0·15	13·4 ± 1·1/13·3 ± 1·8	102 ± 12

The response amplitude recorded from the axon terminal showed a single exponential decay (Fig. 4*C*, Table 1). The length constants of twelve pairs of cells were (mean ± S.D.): 0·17 ± 0·04 mm (near region) and 0·89 ± 0·14 mm (far region) for the soma and 1·05 ± 0·15 mm for the axon terminal. The length constant obtained for the axon terminal was close to that obtained for the soma in the far region from the slit. Student's *t* test provided no reason to reject the null hypothesis that they were the same (at the 2% level of significance). This evidence, together with the observation that the response amplitude of the axon terminal exceeded that of the soma in the far region from the slit, suggests that the soma and the axon terminal are electrically connected.

A new model: coupled two-layer network including somata and axon terminals

A discrete resistive network model was designed to interpret the above observation. The model consists of a pair of inter-connected two-dimensional resistive networks, but for simplicity of representation one-dimensional networks are illustrated (Fig. 5); one represents the syncytium of the soma and the other represents that of the axon terminal. A square grid arrangement with mean cell spacing D was assumed for the spatial distribution of the soma and the axon terminal for simplicity. From this assumption, each of these networks can be treated as a one-dimensional network because a slit stimulus was used. The network of the soma was expressed by a total membrane conductance of each cell (g_m) and a coupling conductance between two adjacent cells (g_s). The syncytium of the axon terminal was expressed similarly by a total membrane conductance (h_m) and a coupling conductance between two adjacent cells (h_s) with the same mean cell spacing as that of the soma. Although the axon terminal is a long fusiform process which probably has no preferential orientation

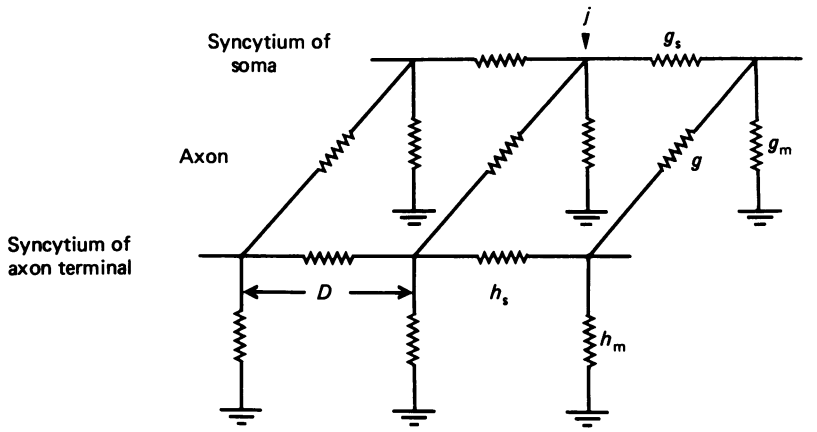

Fig. 5. A resistive network model for the carp horizontal cell (interpretation in text).

(Stell, 1975), the one-dimensional network with the same cell spacing as the soma may represent adequately the properties of the syncytium made of axon terminals. Since it has been demonstrated morphologically that a carp horizontal cell has a single, non-bifurcating axon terminal (Stell, 1975), the number of axon terminals in a unit area must be the same as that of the soma. The two networks were connected at each node by a conductance g which represents the conductance of the axon that connects the axon terminal to the soma. However, g does not necessarily represent the conductance of an axon along the entire length (see Discussion). The leakage conductance of the connecting axon was omitted for simplicity.

The response amplitudes at each node of the network are expressed by equations:

$$S_{n-1}-[(g_m+g)/g_s+2].S_n+S_{n+1} = -A_n.g/g_s-j_n/g_s, \tag{1}$$

$$A_{n-1}-[(h_m+g)/h_s+2].A_n+A_{n+1} = -S_n.g/h_s, \tag{2}$$

Here, S_n and A_n represent the response amplitudes of the soma and the axon terminal at the nth node. j_n signifies the driving current induced by the illumination. Since the potential distribution is symmetrical for both sides of the slit, only positive values for n are examined in this section. Outside the slit, j_n is considered to be zero and the relevant response amplitudes are given from eqns. (1) and (2) as

$$S_n = a_1.\exp(-n.d_1)+a_2.\exp(-n.d_2), \tag{3}$$

$$A_n = b_1.\exp(-n.d_1)+b_2.\exp(-n.d_2). \tag{4}$$

Here d_1 and d_2 are functions of g_m, g_s, h_m, h_s and g (see Appendix 1). And a_1, a_2, b_1 and b_2 are constants which are determined by a boundary condition (see Appendix 2).

If the coupling conductances g_s or h_s are large enough for $(g/g_s).(g/h_s)$ to be eliminated, the solutions (3) and (4) can be expressed (see Appendix 1) by

$$S_n = a_1.\exp(-n.D/\lambda_1)+ a_2.\exp(-n.D/\lambda_2), \tag{5}$$

$$A_n = b_1.\exp(-n.D/\lambda_1)+ b_2.\exp(-n.D/\lambda_2). \tag{6}$$

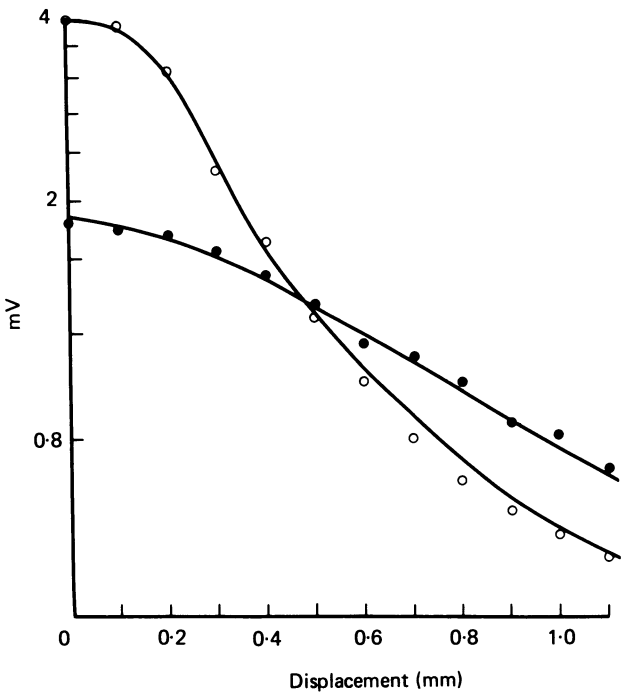

Fig. 6. Comparison between the model and the experiment. The simulation was made on the response of the soma and of the axon terminal which were obtained in a single penetration (Table 1, cell 8). The response amplitudes of the soma (open circles) and the axon terminal (filled circles) were plotted as a function of distance from the centre of the slit in semilogarithmic ordinates. The continuous curves were computed from the model. $(g_m+g)/g_s$ and $(h_m+g)/h_s$ were estimated from the length constant of the line A of the soma and that of the axon terminal in Table 1 by eqns. (7) and (8) in the text. The ratios, $g_m/g_s : g/g_s$ and $h_m/h_s : g/h_s$, were adjusted until an adequate fit was obtained. In this case $g_m/g_s = 0{\cdot}026$, $g/g_s = 0{\cdot}06$, $h_m/h_s = 0{\cdot}001$, $g/h_s = 0{\cdot}01$. Note that the adjusted parameters satisfy $g/g_s . g/h_s \ll 1$, as was required in the prediction of the length constants λ_1 and λ_2 (see Appendix 1). A horizontal bar under the abscissa shows the width of the illuminated region.

Here, the length constants λ_1 and λ_2 reflect the tightness of the electrical coupling: λ_1 for the syncytium of the soma and λ_2 for that of the axon terminal. They are related to the ratio $\gamma_1 \equiv (g_m+g)/g_s$ and $\gamma_2 \equiv (h_m+g)/h_s$ (similar to eqns. (14*a*) and (14*b*) of Lamb & Simon, 1976) through

$$\gamma_1 = 2(\cosh D/\lambda_1 - 1), \tag{7}$$

$$\gamma_2 = 2(\cosh D/\lambda_2 - 1). \tag{8}$$

Under the condition that $\lambda_1, \lambda_2 \gg D$, γ_1 and γ_2 are expressed by

$$\gamma_1^{-1} = g_s/(g_m+g) \simeq (\lambda_1/D)^2, \tag{9}$$

$$\gamma_2^{-1} = h_s/(h_m+g) \simeq (\lambda_2/D)^2. \tag{10}$$

The model predicts the response to decay with two exponential components. Spatial properties of the response in the soma and in the axon terminal can be

interpreted from this model as follows. The electrical signal is evoked in somata which lie under the slit. The signal spreads laterally to the neighbouring somata through g_s with the small length constant λ_1 and also spreads to the axon terminal through g. For reasons to be discussed in reference to morphology (see Discussion), the conductance of the axon, g, is probably large enough to permit electrical communication between the syncytium of the soma and that of the axon terminal. Thus, at the centre of the slit, one can record a reasonably large response amplitude in the axon terminal. Since the axon terminal is thought to be coupled more tightly to its neighbours than the soma (represented by a large length constant λ_2), decrement of the response amplitude might be smaller in axon terminals than in somata. Therefore, a larger response amplitude is expected in axon terminals than in somata at a region far from the slit, where the electrical signal possibly spreads back from the axon terminal to the soma. As a consequence, the response of the soma might reflect that of the axon terminal, particularly at a distant region from the slit, and the spatial decrement of the response amplitude becomes similar between the soma and the axon terminal. Current spreading in the syncytium of the soma with length constant λ_1 would also affect the response of the axon terminal. But the influence is likely to be experimentally indiscernible because the junctional conductance between axon terminals is high (see Appendix 2).

To evaluate these explanations, the computed and experimental response distributions were compared in Fig. 6. The computation was carried out by solving eqns. (1) and (2) in a matrix form with the aid of the Gauss–Jordan method. The cell spacing was chosen as 50 μm (Kaneko & Stuart, 1984). An adequate fit was obtained both in the soma and the axon terminal. This result suggests that the two syncytia have an electrical connexion and that the responses of the soma and the axon terminal interact with each other during the course of the lateral signal spread.

From the model, a ratio of the response amplitude of the axon terminal to that of the soma can be obtained by $g/(g+h_m)$. The adjusted parameters predict that diffuse illumination would produce responses in axon terminals whose amplitudes are approximately 90 % as large as those in somata. In fact, the response amplitudes of axon terminals evoked by diffuse illumination with dim light were 102 ± 12 % (mean $\pm$ S.D., $n = 6$) as large as those of somata (Table 1). Variation could be attributed to the variable amount of damage caused by the penetration of the electrode.

DISCUSSION

The present study has demonstrated a difference of the receptive field characteristics in the soma and the axon terminal of fish horizontal cells. Both receptive fields were uniform in structure, but they were larger in the axon terminal than in the soma, in agreement with earlier reports (Kaneko, 1970; Marmarelis & Naka, 1972; Teranishi *et al.* 1982). The spatial decline of the response amplitude of the soma measured by a displaced slit of light consisted of two exponential components, while that of the axon terminal showed a single exponential component. Furthermore, the larger length constant of the soma had a value very close to that of the axon terminal. The present results strongly suggest that, unlike horizontal cells of other animal species,

the voltage responses evoked in the syncytial network of somata can conduct to the terminal network, and that the potential changes in the two separate syncytia are influenced by each other. These spatial properties were described quantitatively by a resistive network model expressing the two electrically connected syncytial layers of somata and axon terminals.

The idea of electrotonic spread from the soma to the axon terminal became very likely due to a recent morphological study of the fish retina. Kouyama *et al.* (1984) examined horseradish-peroxidase-injected axon terminals in an electron microscope and found that each axon terminal has numerous gap junctions with neighbouring axons and axon terminals almost continuously along its whole length. This morphological observation suggests that the axon makes electrical contacts with axon terminals of other horizontal cells immediately after it leaves the soma. Therefore, it is likely that the effective length of the axons connecting the terminal syncytium to the soma syncytium is much shorter than the actual length (approximately 300 μm) of the axon. This observation supports a bidirectional electrical communication between the soma and the axon terminal, as proposed in the present study. In the present experiment, all axon terminals showed a maximum response to the centred slit. If the response recorded from an axon terminal spreads from its own soma travelling all the way along the axon, the maximal amplitude would have been recorded when the slit was displaced into the direction of soma location. This observation also suggests that the effective length of the axon is much shorter than its morphological length.

It might be argued that one of the two length constants obtained for soma is attributable to scattered light. However, this possibility can be ruled out because the terminal syncytium showed only a single component and because it occurred at distances over which light scattering is considered to be of minor importance. It might also be argued that the finite length of the slit deformed one-dimensional potential decay, particularly when the slit was displaced by a long distance. In horizontal cells of the turtle retina, however, reliability of the one-dimensional analysis has been demonstrated by Lamb (1976) using a slit of a finite length. In his experiment, the slit (0·08 × 2·33 mm) was displaced up to 0·8 mm (several times the length constant), but the response still followed a single exponential decay. Since the length of the slit used in the present study was 1·7 times as long as that used by Lamb, the error due to the finite length of the slit might be much smaller.

The following conditions were given to parameters in eqns. (1) and (2) to obtain good agreement with the experimental results.

1. A larger membrane conductance (a total membrane conductance of each cell) to the soma than to the axon terminal, i.e. $g_m > h_m$. Since the potential decay was measured in the unilluminated region, the adjusted parameters reflect a total conductance value in the dark. In the unilluminated region, the conductance of the soma would be high by an addition of the synaptic conductance which is activated by the transmitter released from photoreceptors (Byzov & Trifonov, 1968; Kaneko & Shimazaki, 1975), provided that the non-synaptic conductance of the soma is nearly identical to that of the axon terminal membrane.

2. A larger coupling conductance value to the junction between the axon terminal than between the soma, i.e. $h_s > g_s$. This condition is relevant to apparent single

exponential decay of the response of the axon terminal (see Appendix 2) and possibly fulfilled since axon terminals have abundant gap junctions (Kouyama *et al.* 1984).

3. A 10 times larger value to the connecting conductance (between the soma and the axon terminal) than to the membrane conductance of the axon terminal, i.e. $g/h_m \simeq 10$. In a previous study, Tachibana (1983) obtained a conductance value of 10^{-9} S in the enzyme-treated isolated horizontal cell (soma) of the cyprinid fish at a membrane potential of around -30 mV. The transmitter-mediated conductance was not activated in isolated cells. If the membrane properties of the axon terminal are the same as the non-synaptic membrane of the soma, the conductance of the axon terminal is likely to be of the same order of magnitude. From these observations, the connecting conductance g was roughly estimated to be 10^{-8} S. If the axon is 0·5 μm in diameter and filled with axoplasm having a specific conductance of 2×10^{-2} S cm^{-1}, this value corresponds to 50 μm in the case of an infinite membrane resistance. This length might represent the effective length between the soma and the axon terminal.

A regenerative propagation of signals from the soma to the axon terminal has been proposed by Weiler & Zettler (1979). The present study suggests that signal propagation seems possible without a regenerative process, but does not eliminate its possibility. A different experiment, which is in progress, of examining membrane properties of axons and axon terminals of isolated horizontal cells may elucidate the presence of such a process.

APPENDIX 1

We will derive the solutions of the following set of difference equations which are obtained from eqns. (1) and (2) in the text:

$$S_{n-1}-(\alpha_1+\alpha_2+2).S_n+S_{n+1}=-\alpha_2.A_n-E_n, \tag{A 1.1}$$

$$A_{n-1}-(\beta_1+\beta_2+2).A_n+A_{n+1}=-\beta_2.S_n, \tag{A 1.2}$$

where $\alpha_1 \equiv g_m/g_s$, $\alpha_2 \equiv g/g_s$, $\beta_1 \equiv h_m/h_s$, $\beta_2 \equiv g/h_s$,

and

$$E_n = j_n/g_s = E(-k \leqslant n \leqslant k; \text{ inside the slit of width } 2k), \tag{A 1.3}$$

$$= 0(n < -k, k < n; \text{ outside the slit}). \tag{A 1.4}$$

First, the voltage distribution for (A 1.4) is obtained. By assuming solutions of form $S_n = S_0.e^{n\delta}$, and $A_n = A_0.e^{n\delta}$ where S_0, A_0 and δ are constant, we obtain the following equations from (A 1.1) and (A 1.2)

$$S_0.e^{\delta}-S_0.(\alpha_1+\alpha_2+2)+S_0.e^{-\delta}=-\alpha_2.A_0, \tag{A 1.5}$$

$$A_0.e^{\delta}-A_0.(\beta_1+\beta_2+2)+A_0.e^{-\delta}=-\beta_2.S_0. \tag{A 1.6}$$

On combining (A 1.5) and (A 1.6), it is found that e^{δ} must satisfy the following fourth-order equation:

$$(e^{\delta}-(\alpha_1+\alpha_2+2)+e^{-\delta}).(e^{\delta}-(\beta_1+\beta_2+2)+e^{-\delta})-\alpha_2.\beta_2=0. \tag{A 1.7}$$

Obviously, (A 1.7) has four solutions for e^{δ}. If e^{δ} satisfies (A 1.7), so does $e^{-\delta}$. Therefore the boundary conditions that $S_n, A_n \to 0$ as $n \to \pm\infty$ and a symmetric distribution of

the voltage, i.e. $S_n = S_{-n}$, give the solutions expressed by

$$S_n = a_1.\exp(-|n|.d_1)+a_2.\exp(-|n|.d_2), \tag{A 1.8}$$

$$A_n = b_1.\exp(-|n|.d_1)+b_2.\exp(-|n|.d_2), \tag{A 1.9}$$

where a_1, a_2, b_1, b_2, d_1 and d_2 are constants ($d_1, d_2 > 0$).

Next, the voltage distribution for (A 1.3) is obtained. In this case the solutions can be expressed by

$$S'_n = a'_1.\exp(n.d_1)+a'_2.\exp(n.d_2)+a'_1.\exp(-n.d_1)+a'_2.\exp(-n.d_2)+S^*, \tag{A 1.10}$$

$$A'_n = b'_1.\exp(n.d_1)+b'_2.\exp(n.d_2)+b'_1.\exp(-n.d_1)+b'_2.\exp(-n.d_2)+A^*. \tag{A 1.11}$$

Here, a'_1, a'_2, b'_1 and b'_2 are constants. S^* and A^* are specific solutions, which correspond to the voltage evoked by diffuse illumination, expressed by

$$S^* = (g+h_m).E/(g.g_m+g.h_m+g_m.h_m), \tag{A 1.12}$$

$$A^* = g.E/(g.g_m+g.h_m+g_m.h_m). \tag{A 1.13}$$

The following approximation gives the simplified solutions. When the coupling conductance g_s or h_s is larger relative to the connecting conductance g (i.e. $g_s \gg g$, or $h_s \gg g$), then the term $\alpha_2.\beta_2 (= g/g_s.g/h_s)$ can be ignored. Therefore, e^{δ} is obtained from (A 1.7) as the solution of quadratic equations:

$$e^{\delta}-(\alpha_1+\alpha_2+2)+e^{-\delta} = 0, \tag{A 1.14}$$

$$e^{\delta}-(\beta_1+\beta_2+2)+e^{-\delta} = 0. \tag{A 1.15}$$

Thus the required solutions are

$$\exp(-d_1) = (\gamma_1+2)/2-\sqrt{(\gamma_1^2+4.\gamma_1/2)}, \tag{A 1.16}$$

$$\exp(-d_2) = (\gamma_2+2)/2-\sqrt{(\gamma_2^2+4.\gamma_2/2)}, \tag{A 1.17}$$

where $\gamma_1 \equiv \alpha_1+\alpha_2 = (g_m+g)/g_s$, $\gamma_2 \equiv \beta_1+\beta_2 = (h_m+g)/h_s$. If the length constant λ_1 and λ_2 are defined (see eqn. (14*a*) of Lamb & Simon, 1976) by

$$\gamma_1 = 2(\cosh D/\lambda_1-1), \tag{A 1.18}$$

$$\gamma_2 = 2(\cosh D/\lambda_2-1), \tag{A 1.19}$$

then the solutions (A 1.8), (A 1.9) and (A 1.10), (A 1.11) can be expressed by

$$S_n = a_1.\exp(-|n|.D/\lambda_1)+a_2.\exp(-|n|.D/\lambda_2), \tag{A 1.20}$$

$$A_n = b_1.\exp(-|n|.D/\lambda_1)+b_2.\exp(-|n|.D/\lambda_2), \tag{A 1.21}$$

and

$$S'_n = a'_1.\exp(n.D/\lambda_1)+a'_2.\exp(n.D/\lambda_2) +a'_1.\exp(-n.D/\lambda_1)+a'_2.\exp(-n.D/\lambda_2)+S^*, \tag{A 1.22}$$

$$A'_n = b'_1.\exp(n.D/\lambda_1)+b'_2.\exp(n.D/\lambda_2) +b'_1.\exp(-n.D/\lambda_1)+b'_2.\exp(-n.D/\lambda_2)+A^*. \tag{A 1.23}$$

These solutions provide an intuitive interpretation for the lateral spread of the electrical potential in terms of length constant and mean cell spacing.

APPENDIX 2

The apparent single exponential decay of the response of the axon terminal is shown by examining the constants a_i, b_i, a_i' and b_i' $(i = 1, 2)$. By substituting (A 1.8) and (A 1.9) in (A 1.1) and comparing the coefficients of $\exp(-|n|.d_1)$ and $\exp(-|n|.d_2)$, we obtain

$$b_1 = -a_1.(e^{d_1} - (\alpha_1 + \alpha_2 + 2) + e^{-d_1}).\alpha_2^{-1}, \tag{A 2.1}$$

$$b_2 = -a_2.(e^{d_2} - (\alpha_1 + \alpha_2 + 2) + e^{-d_2}).\alpha_2^{-1}. \tag{A 2.2}$$

On combining (A 1.10), (A 1.11) and (A 1.1), we also find

$$b_1' = -a_1'.(e^{d_1} - (\alpha_1 + \alpha_2 + 2) + e^{-d_1}).\alpha_2^{-1}, \tag{A 2.3}$$

$$b_2' = -a_2'.(e^{d_2} - (\alpha_1 + \alpha_2 + 2) + e^{-d_2}).\alpha_2^{-1}. \tag{A 2.4}$$

In addition to these four equations, the following boundary conditions are required to give expressions for a_i, b_i, a_i' and b_i' $(i = 1, 2)$ in terms of the slit width of $2k$ and parameters involved in the model.

$$S_k = S_k', \tag{A 2.5}$$

$$S_{k+1} = S_{k+1}', \tag{A 2.6}$$

$$A_k = A_k', \tag{A 2.7}$$

$$A_{k+1} = A_{k+1}'. \tag{A 2.8}$$

The spatial distribution of the voltage can be obtained by solving this set of equations. However, the apparent single exponential decay can be predicted from eqns. (A 2.1) and (A 2.2). We assume that the approximation, $\alpha_2.\beta_2 \simeq 0$ in Appendix 1, is satisfied due to $\beta_2 \simeq 0$. This assumption leads to (A 1.16) and (A 1.17) and we obtain

$$e^{d_1} - (\alpha_1 + \alpha_2 + 2) + e^{-d_1} = 0, \tag{A 2.9}$$

$$e^{d_2} - (\beta_1 + \beta_2 + 2) + e^{-d_2} = 0. \tag{A 2.10}$$

By applying (A 2.9) to (A 2.1), it is shown that $b_1 \simeq 0$ in the case of $\alpha_2 \neq 0$, while b_2 has a finite value.

The author thanks Professor Genyo Mitarai for his continual encouragement and numerous suggestions throughout this work, and Professor Akimichi Kaneko for his valuable comments and discussions on the manuscript. Thanks are also due to Drs S. Yasui, T. Ohtsuka, M. Tachibana, M. Tauchi and S. Usui for their comments on the manuscript. This research is part of a dissertation, submitted in partial fulfilment of the requirements for a Ph.D. degree from Nagoya University.

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