Light Adaptation of Discrete Waves in the *Limulus* Photoreceptor

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**ABSTRACT** Light adaptation affects discrete waves in two ways. It reduces their average size and decreases the probability that a photon incident at the cornea causes a discrete wave. There is no effect of light adaptation on the latency of discrete waves, or on their time-course.

**INTRODUCTION**

The photoreceptor of the lateral eye of *Limulus* responds to a flash of light by depolarizing. The amount of depolarization depends on the energy of the stimulus and on the quantity of light absorbed by the photoreceptor in the minutes preceding the stimulus presentation (Benolken, 1962; Fuortes and Hodgkin, 1964). A fixed light stimulus is less effective if the photoreceptor was recently exposed to light than if it was not, and this effect is called light adaptation. (For a review of the effects of light adaptation on the *Limulus* eye see Wolbarsht and Yeandle, 1967.)

The depolarizing response of the photoreceptor is composed of many discrete depolarizations called discrete waves. Each discrete wave results from a single photon absorption (Wolbarsht and Yeandle, 1967; Srebro and Yeandle, 1970). This paper is concerned with the effect of light adaptation on the stochastic properties of discrete waves. Two studies with similar objectives have already been published (Adolph, 1964; Dodge et al., 1968). In these studies the fluctuating level of depolarization that resulted from steady illumination was analyzed using the theory of shot noise, and the stochastic properties of discrete waves were deduced. However, the shot noise model provides no information about the timing of discrete waves (latency). Our approach is different and does provide this information. We used low energy flashes of light which resulted in the absorption of a small and variable number of photons on each presentation, and we observed the effects of light adaptation on the responses. The properties of responses were related to the stochastic properties of discrete waves.
THEORY

In this section we discuss the stochastic properties of discrete waves, and show how they relate to the stochastic properties of responses to low energy flashes of light.

When a light flash containing $E$ photons is incident at the surface of the ommatidium, only a fraction of that light is absorbed, and a fraction of the light absorbed is effective in producing discrete waves. We define the product of these fractions as the quantum efficiency of discrete wave production and denote its value by $p$. The average number of light-induced discrete waves per flash is $pE$. We use the phrase "effective absorption" to label a photon absorption that produces a discrete wave.

A photon effectively absorbed at time $t = 0$ will produce a discrete wave at time $t$ following the absorption. The latency, $t$, is a random variable. We define the probability density function, $r(t)$, so that

$$r(t)At = \text{probability (a discrete wave during $\Delta t$ at $t/ \text{one effective absorption at } t = 0$)}$$

where $\Delta t$ is a small increment of time.

Let $S(t)$ be a normalized function denoting the time-course of a discrete wave, and let $A$ be a random variable denoting the discrete wave amplitude. Discrete waves may be represented by $A \cdot S(t)$.

The latency of the first discrete wave to follow a flash of light has the probability density function $L(t)$ given by

$$L(t) = (\lambda r(t) + \mu) \exp (-\lambda \int_0^t r(x)dx - \mu t)$$

where $t$ is measured from the onset of the light flash, $\mu$ is the rate at which spontaneous discrete waves occur, $\lambda = pE$, and $r(t)$ has already been defined. Equation 1 has been derived and its predictions tested by Srebro and Yeandle (1970). We define $\bar{L}$ as the average value of the latency over a time interval, $T$, just sufficient to include all light-induced discrete waves. It can be shown that

$$\mu = -(1/T) \log_e (q_D) \quad (1 a)$$

and

$$\lambda = -\log_e (q_L/q_D) \quad (1 b)$$

where $q_L$ is the probability that no discrete wave occurs in the interval $(0, T)$.
and \( q_0 \) is the probability that no spontaneous discrete wave occurs in a period of duration \( T \) \( \) (Srebro and Yeandle, 1970).

Let a flash of light be presented at time \( t = 0 \) and let \( v(t) \) be the amount of depolarization produced at time \( t \). Let the flash be presented many times and denote by \( \overline{v(t)} \) and \( \sigma_n^2(t) \) the average depolarization and its variance at a specified time, \( t \). In the following development we show that the fractional variation of the depolarization at a specified time following a flash, \( \sigma_n(t)/\overline{v(t)} \), does not change when the amplitudes of discrete waves are changed by a simple scaling operation. This conclusion is similar to the well-known result that scaling a random variable does not alter its fractional variation. But the depolarization at a specified time following a flash is a function of several random variables and the proof is more complicated.

The number of photons effectively absorbed during a flash of light varies from trial to trial, and follows the Poisson distribution.

\[
p(n) = \frac{\lambda^n}{n!} \exp(-\lambda)
\]

where \( p(n) \) is the probability that exactly \( n \) effective absorptions occur on any one trial. Let \( \overline{v_n(t)} \) and \( \sigma_n^2(t) \) be the mean and variance of the depolarization at a specified time, \( t \), for those trials which result in exactly \( n \) effective absorptions. Then

\[
\overline{v(t)} = \sum_{n=0}^{\infty} p(n) \overline{v_n(t)}, \quad (2)
\]

and

\[
\sigma_n^2(t) = \sum_{n=0}^{\infty} p(n) \sigma_n^2(t). \quad (3)
\]

If more than one photon is effectively absorbed, and if each behaves independently, then

\[
\overline{v_n(t)} = n\overline{v(t)} \quad (4)
\]

and

\[
\sigma_n^2(t) = n\sigma_1^2(t). \quad (5)
\]

For the case \( n = 1 \) consider an increment of time \( \Delta t' \) at \( t' < t \). The probability that the one discrete wave begins during \( \Delta t' \) is

\[
r(t')\Delta t'
\]
and the depolarization caused by that discrete wave at \( t \) is

\[ A \cdot S(t - t') \]

with probability \( p(A) \). Then, as \( \Delta t' \to 0 \),

\[
\overline{v(t)} = \int_{0}^{t} \int_{A} A p(A) r(t') S(t - t') \, dA \, dt'
\]

\[
\overline{v(t)} = \bar{A} \int_{0}^{t} r(t') S(t - t') \, dt'
\]

and

\[
\overline{v(t)} = \bar{A} \overline{G(t)}, \tag{6}
\]

where \( G(t) \) is the value of the integral. The second moment of \( v(t) \), denoted by \( E_2(v(t)) \), is given by

\[
E_2(v(t)) = \int_{0}^{t} \int_{A} A^2 p(A) S^2(t - t') r(t') \, dA \, dt'
\]

\[
E_2(v(t)) = E_2(A) \int_{0}^{t} S^2(t - t') r(t') \, dt'
\]

\[
E_2(v(t)) = E_2(A) H(t)
\]

where \( E_2(A) \) is the second moment of \( A \) and \( H(t) \) is the value of the integral. From the definition of the second moment,

\[
\sigma^2(t) = (\sigma^2 + \bar{A}^2) H(t) - \bar{A} \overline{G^2(t)} \tag{7}
\]

where \( \sigma^2 \) is the variance of the discrete wave amplitude, \( A \). From equations 2, 4, and 6 it follows that

\[
\overline{V(t)} = \lambda \overline{A} \overline{G(t)} \tag{8}
\]

and from equations 3, 5, and 7 it follows that

\[
\sigma^2(t) = \lambda [\sigma_A^2 H(t) + \bar{A}^2 (H(t) - \overline{G^2(t)})]
\]

which may be written

\[
\sigma^2(t) = \lambda (\sigma_A^2 H(t) + \bar{A}^2 D(t)) \tag{9}
\]

where

\[
D(t) = H(t) - \overline{G^2(t)}.
\]
The fractional variation, \( f(t) \), is given by \( \sigma^2_s(t)/\mu(t) \) and from equations 8 and 9

\[
f(t) = \sqrt{\left[ \sigma^2_s H(t) + \bar{A} D(t) \right]/\Lambda^2 G^2(t)}. \tag{10}
\]

Under the operation of scaling all discrete wave amplitudes by any factor, the ratio \( \sigma^2_s/\bar{A} \) remains constant. From equation 10 it follows that \( f(t) \) is also invariant under the operation.

From equation 8 it is seen that the average depolarization at a fixed time following a fixed flash could be reduced by four possible processes: a reduction in the average number of effectively absorbed photons per flash, \( \lambda \), a reduction in the amplitudes of the discrete waves that compose the response, a shortening of the time-course of a discrete wave, \( S(t) \), or a lengthening of the probability density function associated with the times of occurrences of individual discrete waves, \( r(t) \). All the processes named, with the single exception of a reduction in the amplitudes of discrete waves, also result in an increase in the fractional variation.

The maximum value of a response to a flash of light is denoted by \( V_{\text{max}} \) and defined as the largest value of the resulting depolarization during an interval of time \( T \) following a flash of light where \( T \) is sufficiently long to include all light-induced depolarization. The value of \( V_{\text{max}} \) is different for each response, and its distribution depends on the properties of discrete waves. The relationship of the probability density function of \( V_{\text{max}} \) to \( S(t) \), \( r(t) \), \( \lambda \), and \( A \) is identical to the problem solved by Souček for endplate potentials (Souček, 1971). The problem can be simplified by noting that for each presentation of light flash there will result either no discrete wave, one discrete wave, or more than one discrete wave. If there is no discrete wave, \( V_{\text{max}} \) will be near the resting membrane potential, and we choose to call this value 0. If there is one discrete wave \( V_{\text{max}} \) is equivalent to \( A \). If there is more than one discrete wave the distribution of \( V_{\text{max}} \) depends on both the number of discrete waves and the degree of their temporal overlap. Generally, \( V_{\text{max}} \) has a broad distribution with multiple maxima, but in some particularly fortunate cases the discrete waves are sufficiently separated in time and the distribution of \( V_{\text{max}} \) for a low energy flash reflects the distribution of \( A \) in a simple way (for example see Fig. 5).

**METHODS**

A microelectrode was inserted into the ommatidium of the excised lateral eye using standard techniques. A small quantity of tetrodotoxin was added to the bathing fluid to stop the occurrence of action potentials, and the cell was kept in the dark for approximately 45 min. The temperature of the cell was adjusted to make the rate of spontaneous discrete waves low. (The temperature used was usually near 11°C.) A
A 20 msec flash of light that delivered $E_t$ photons was selected so that it evoked at least one light-induced discrete wave in a large majority of its presentations (the threshold flash). Another 20 msec flash which delivered $E_a$ photons per presentation was selected (the adapting flash). A sequence of 400 flashes was presented at 5-sec intervals, and consisted of the alternate presentations of the adapting and threshold flashes. In each group of 10 flashes in the sequence, one of the threshold flashes was inhibited, and we called this a false flash. The entire sequence was called a run. After each presentation of the threshold flash, equally spaced samples of $v(t)$ were taken, at 100 samples/sec, starting at the onset of the flash and ending after 2 sec. The samples were stored on incremental magnetic tape. The latency of the first discrete wave to follow a threshold flash was determined by measuring the time required for the resulting depolarization to just exceed a predetermined fixed level. The fixed level of depolarization was selected by the experimenter so that the automatic detection of discrete waves was in accord with his visual impression of their occurrence. If no discrete wave occurred in the 2 sec interval following the flash, no latency measurement was recorded, and the trial was marked as a "no response" trial.

In separate runs, the ratio $E_a/E_t$ was systematically varied from 1 to 125. Every run in which $E_a/E_t$ was greater than one was preceded by a run in which $E_a/E_t$ was exactly one and followed by ½ hr of darkness, and another run for which $E_a/E_t$ was one. A group of nine runs constituted an experiment. During any experiment the temperature of the cell, the quantal content of the threshold flash, and the critical depolarization used to detect the occurrence of a discrete wave remained constant. False flash results were processed exactly as threshold flash trials, but were treated separately. We report results for 12 different retinula cells.

RESULTS

Fig. 1 shows the responses to 18 consecutive threshold flashes for a run in which $E_a/E_t$ was one. The following measurements were made, and tabulated separately for threshold and false flashes, from records similar to those shown in Fig. 1:

1. the latency distribution for the first discrete wave,
2. the fraction of trials for which there was no response,
3. the distribution of the maximum depolarization of responses.

The average latency of the first discrete wave, $\overline{L}$, was calculated from the tabulated data. The average number of spontaneous discrete waves per second, $\mu$, and the average number of light-induced discrete waves per trial, $\lambda$, were estimated using equations 1a and 1b. In addition the computer was programmed to operate as a calculator of average transients and provide the time-course of the average depolarization, $\overline{\sigma(t)}$, due to all the threshold flash presentations in a run. The time-resolved variance $\sigma^2(t)$ was calculated at the same time.

Each run lasted approximately 33 min and we checked the stability of the
run by plotting the values of $V_{max}$ in the serial order of their occurrences during the run. There was no significant drift in $V_{max}$. There was often a small hyperpolarizing drift of the membrane potential during the course of a run.

Fig. 2 shows the latency histograms for a typical experiment. There is no systematic change in the latency distribution for values of $E_a/E_t$ from 1 to 28. The histogram associated with $E_a/E_t$ equal to 125, however, has a peak at a slightly later time than those associated with $E_a/E_t$ equal to one.

![Figure 2. Latency histograms for a typical experiment.](image)

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Fig. 3 shows the average depolarization, $v(t)$, as a function of time for the same runs as shown in Fig. 2. There is a systematic decrease in the magnitude of $v(t)$ as $E_a/E_t$ increases, but there is no appreciable alteration of its time-course.

In order to investigate the change in fractional variation with light adaptation we selected a time, $t_{max}$, near the maximum depolarization due to the threshold flash in each experiment. At this time the average depolarization and its variance were relatively large and accurate estimates of both could be made. Denoting the depolarization at $t_{max}$ by $V$ and the fractional variation at $t_{max}$ by $F$, each run could be summarized by five measurements, namely, $V$, $F$, $\lambda$, $\mu$, and $\lambda$. Let $X$ stand for any one of the five measurements. Let $X_a$ stand for $X$ in a run in which $E_a/E_t$ was greater than one, and let $X_b$ stand for the average value of the same $X$ for the two runs, with $E_a/E_t = 1$, that preceded and followed it. The per cent change in each of the five measurements was tabulated and averaged over the 12 cells studied.
FIGURE 2. Distributions of the latency of the first discrete wave following threshold flashes. The abscissa gives time in seconds. Time 0 corresponds to the onset of the light flash. The ordinate gives the number of trials for which the latency was that shown on the abscissa. The histogram bin size is 10 msec; all the records are from a single cell. The number to the right of each histogram gives the ratio of the adapting flash energy to the threshold flash energy. Retinula cell, 12.0°C.
Fig. 4 shows the per cent change of the five measurements as a function of $E_a/E_t$. Curve 1 shows that $V$, the maximum average depolarization, decreases as $E_a/E_t$ increases. There is no obvious threshold for this effect. For example, an adapting flash with energy 8.7 times the threshold flash was sufficient to reduce $V$ by 21%. In contrast to the behavior of $V$, the average latency of the first discrete wave, $L$, is virtually unaffected by adapting flashes with energy as high as 28 times the threshold flash. There is a small increase in $L$ when $E_a/E_t$ is 125 (Curve 2). Curve 3 shows that there is a progressive increase in the fractional variation of the depolarization, $F$, as $E_a/E_t$ is increased. Curves 4 and 5 show that $\lambda$, the average number of light-induced discrete waves per threshold flash, and $\mu$, the average rate of spontaneous discrete waves per second, are reduced as $E_a/E_t$ is increased. The estimate, $\lambda$, is less affected by light adaptation than is $V$. For example, at $E_a/E_t = 28$, $V$ is reduced by 43% while $\lambda$ is reduced by 22%, and this suggests that the sizes of discrete waves may also decrease as $E_a/E_t$ increases. Also, $\mu$ is less affected by light adaptation than is $\lambda$. At $E_a/E_t = 125$, $\lambda$ is reduced by 62% while $\mu$ is reduced by 37%.

Fig. 5 shows several histograms for $V_{max}$, the maximum depolarization of responses, for several runs of a single experiment. Each row in the figure represents a single run, and columns 1 and 2 represent, respectively, the threshold and false flash data. Each histogram has two peaks. The peak near $V_{max} = 0$ represents trials for which no discrete wave was recorded. The second peak occurs at different values of $V_{max}$ in each row. In the first run (top) the second peak is near $V_{max} = 2.3$ mv, and in the last run (bottom) the second peak is
near 1.4 mv. The relative number of trials associated with the first peak increases as $E_a/E_t$ increases, and the location of the second peak shifts to lower values as $E_a/E_t$ is increased.

Although all the histograms of $V_{\text{max}}$ showed changes similar to those shown in Fig. 5, there was considerable variation from cell to cell in their detail. In most experiments, the second peak was much broader than in Fig. 5 and in others there were two peaks for $V_{\text{max}} > 0$. We examined the responses trial by trial and found that the variation in the $V_{\text{max}}$ histograms reflected a large variation in the degree of overlap of discrete waves.

Fig. 6 shows responses that consist of isolated discrete waves for two different runs in the same experiment. By an isolated discrete wave we mean one which was not overlapped in time by another discrete wave. The figure was con-

![Graph](image-url)

**Figure 4.** Per cent change for five parameters of threshold flash responses. The abscissa gives the ratio of the adapting flash energy to the threshold flash energy. The ordinates give per cent change (see text for details of calculation). Each point is the average value for 12 cells. Curve 1, per cent change of average depolarization. Curve 2, per cent change of the average latency. Curve 3, per cent change of fractional variation. Curve 4, per cent change of average number of light-induced discrete waves per flash. Curve 5, per cent change of average number of spontaneous discrete waves per second.
structed by selecting the first 9 or 10 trials of the run for which an isolated discrete wave resulted. Column A shows isolated discrete waves for a run in which $E_a/E_i$ was one, while column B shows isolated discrete waves for a run in which $E_a/E_i$ was 125. The gain corresponding to the traces of column A is 0.4 times the gain corresponding to the traces of column B. The discrete waves are reduced in size by light adaptation, but there is no systematic change in their time-course.

A small and variable hyperpolarization often followed the depolarizing phase of a discrete wave. We did not investigate this phenomenon in detail, but we think that it was somewhat greater during light adaptation.

![Figure 5: Distribution of maximum response depolarizations.](image)

The abscissa gives maximum depolarization in millivolts. The ordinates give the number of trials for which maximum depolarization was that shown on the abscissa. The ordinate scale at the left refers to all histograms. The histogram bin size was approximately 0.15 mV. Column 1, threshold flashes; column 2, false flashes. All the histograms are from a single experiment. The number to the right of each row gives the ratio of the adapting flash energy to the threshold flash energy. Retinula cell, 11.0°C.
CONCLUSIONS

Our results suggest the following conclusions.

(a) The probability density function associated with the times of occurrence of individual discrete waves is not affected by light adaptation. This is supported by the findings that the latency distribution for the first discrete wave is not affected by light adaptation for ratios of adapting-to-threshold flash energy as large as 28, and that the time-course of the average depolarization is not affected by light adaptation. (The slight increase in the average latency for adapting flash-to-threshold flash energy ratio 125 can be accounted for by a decrease in the quantum efficiency of discrete wave production under these conditions. See item d below.)

(b) The discrete wave time-course is not affected by light adaptation. This is supported by the result that the time-course of the average depolarization is not affected by light adaptation, as well as by our observations of isolated discrete waves which reveal no systematic change in their time-course.

(c) The sizes of discrete waves are reduced by light adaptation. This is supported by our observations of isolated discrete waves. It is also supported by the results that the peak of the distribution of the maximum response depolarization shifts progressively to lower values as the ratio of adapting-to-threshold flash energy increases, and that the reduction in the maximum
average depolarization is only partly accounted for by a reduction in the average number of light-induced discrete waves per flash.

\(d\) The quantum efficiency of discrete wave production is decreased by light adaptation. This is supported by the following results. (i) The fractional variation of the depolarization increases as the ratio of adapting-to-threshold flash energy increases. (ii) The estimate of the average number of light-induced discrete waves decreases as the ratio of adapting to threshold flash energy increases. (iii) The distribution of the maximum depolarization of responses shows a progressive increase in the relative number of trials that result in no discrete wave. In view of conclusions (a) and (b) above, the increase in fractional variation is particularly important since it rules out the interpretation that the observed decrease in the quantum efficiency of discrete wave production was the result of the reduction in discrete wave size and therefore merely reflected a failure to detect some of them.

In summary, we have found that light adaptation has two major effects on discrete waves. It reduces the size of the discrete waves and it reduces the quantum efficiency of their production.

**DISCUSSION**

In this section we consider the possible mechanisms by which light adaptation may affect discrete waves.

**Quantum Efficiency**

The reduction of quantum efficiency could be due to either (1) a decrease in the average number of photons absorbed per flash or (2) a reduction in the probability that an absorbed photon produces a discrete wave. The first possibility could occur if there were a significant loss of visual pigment during a run. A calculation shows that this is not likely. Consider a run in which the threshold flash delivers, on the average, one effectively absorbed photon per presentation and the adapting flash delivers 10 times this number. From Fig. 4 we would expect a 20% reduction in the size of the response and a small decrease in the quantum efficiency. About 2160 effective photons are absorbed during the run. Suppose that no visual pigment regenerates, and that the probability that a photon absorbed by the visual pigment produces a discrete wave is 0.25 (Millecchia and Mauro, 1969); then, at most, 8640 visual pigment molecules are lost during the run. But there are about \(10^4\) visual pigment molecules per ommatidium (Hubbard and Wald, 1960), and, therefore, only a negligible change in optical density. The same argument holds even for the highest adapting flash energy used in these experiments.

The second possibility would be plausible only if the photoreceptor contained compartments of visual pigment molecules so constituted that an absorption in one visual pigment molecule disabled the others. But a significant
reduction in quantum efficiency occurs when the adapting flash delivers only 10 effectively absorbed photons. The compartments would therefore have to be of the order of the size of a retinula cell. However, Fuortes and Yeandle (1964) showed that the frequency of discrete waves is a linear function of the light intensity for steady low energy stimuli, and thus the compartment model is not a likely explanation.

A possible mechanism of the decrease in quantum efficiency is the migration of screening pigment in the ommatidium. Screening pigment migration occurs upon exposure to light in a wide variety of arthropods (Mazokhin-Porshnyakov, 1969). The finding that the rate at which spontaneous discrete waves occur is less affected by light adaptation than is the quantum efficiency of discrete wave production also suggests that screening pigment migration may occur.

Our conclusion that the quantum efficiency of discrete wave production is reduced by light adaptation is different from that reported by Dodge et al. (1968). These authors used steady illumination, kept their cells at a higher temperature than we did, and used considerably higher light intensities than we did. It is not clear which of these factors is critical to the difference in our results.

**Discrete Wave Size**

There are several ways in which the sizes of discrete waves could be reduced by light adaptation. (1) The driving force of the process, that is the sodium electromotive force (EMF), may be reduced. (2) The load resistance of the cell (the resistance through which the current associated with a discrete wave is dissipated) may decrease. (3) The external calcium ion concentration may increase.

The first possibility follows from the fact that sodium ions carry most of the current associated with the response of the cell to light (Millecchia and Mauro, 1969). But a simple calculation shows that the first possibility is unlikely. Consider the experimental situation presented in the discussion of quantum efficiency. Approximately 2160 discrete waves occur during the run. A discrete wave probably has a peak current of no more than $10^{-9}$ amp (Millecchia and Mauro, 1969) and lasts 0.2 sec. Each discrete wave transfers $2 \times 10^{-10}$ coul of charge, or approximately $3 \times 10^8$ sodium ions, across the cell membrane. At most, approximately $10^{-9}$ mmole of sodium enters the ommatidium during the run. The ommatidium consists of approximately 10 retinula cells, each roughly a cylinder $10^{-2}$ cm long and $2 \times 10^{-3}$ cm in diameter, with a volume of $3 \times 10^{-7}$ cc. Therefore, the concentration of sodium is raised by less than 3 mmoles/liter, hardly enough to reduce the sodium EMF. If the increase in the intracellular concentration of sodium was restricted to a cellular compartment, such as a single microtubule, a substantial lowering of the sodium EMF could...
result for that compartment. But if this were the mechanism of the discrete
wave size reduction it would also require that photons be absorbed by only a
small fraction of the available microtubules, and this is not a reasonable as-
sumption. A similar calculation shows that if the influx of sodium ions reduced
the concentration in a completely restricted extracellular space, that space
would be a zone less than approximately 0.1 μ surrounding each retinula cell
(smaller if the restriction was incomplete). Furthermore, a restricted space of
this kind implies the existence of a standing electrical potential and a volume
change associated with light adaptation. It is therefore unlikely that a change
in the sodium EMF causes the reduction in discrete wave size.

With regard to the second possibility we know of no evidence that the load
resistance is reduced by light adaptation. In fact, Lisman and Brown (1971)
found that light adaptation increases the load resistance in the ventral photo-
receptor.

The third possibility is suggested by the results of Millecchia and Mauro
(1969). They found that the discrete waves of the ventral photoreceptor
were larger in calcium-free seawater than in normal seawater. If the third
possibility were correct, calcium ions would have to move from the intra-
cellular to the extracellular space surrounding the cell during light adapta-
tion. In addition, there would probably have to be some restriction to the free
diffusion of calcium ions in the extracellular space surrounding the cell. How-
ever, this restriction would not be as stringent as for sodium ions because pre-
sumably a small increase in the calcium ion concentration would be sufficient
to mediate the discrete wave size reduction.

Our conclusions that light adaptation reduces the average size of discrete
waves, and that there is no threshold for the reduction, are in agreement
with those of Dodge et al. (1968).

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