

Macaque ganglion cells, light adaptation, and the Westheimer paradigm

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Abstract

Retinal adaptation mechanisms are considered relative to the Westheimer paradigm. Responses to a probe presented upon pedestals were obtained from macaque ganglion cells. On-center magnocellular (MC) cell responses decreased to a plateau as pedestal diameter increased, consistent with operation of a local adaptation pool. Off-center cells also demonstrated a vigorous response with small pedestals, but as pedestal size increased, responsivity decreased and then partially recovered as pedestals encroached upon the surround. The response trough was due to a profound suppression of maintained activity. Comparison with psychophysical data suggests a multiple physiological substrate for the Westheimer paradigm, involving an interaction between adaptation pools, changes in maintained firing due to center-surround mechanisms and a cortical component.

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1. Introduction

Crawford (1940) originally described how the threshold for a small spot stimulus changed with the size of a surround pedestal; as pedestal diameter increased, threshold rose to a peak and then decreased. Westheimer (1965, 1967) suggested this phenomenon was due to the interaction of excitatory and inhibitory regions of ganglion cell receptive fields. The increase in threshold with pedestal size was thought to result from an increase in adaptation (excitation) brought about by spatial summation within the receptive field center, and the subsequent decrease was attributed to a decrease in adaptation through lateral inhibition from the surround.

Psychophysical studies of the Westheimer paradigm have not resolved if the effect is solely due to local retinal

adaptation or if cortical mechanisms are also involved (see Westheimer, 2004; for review). Several studies have indicated that local contour or edge effects contribute to the Westheimer curve, in which case a center-surround model may be inadequate (Fry & Bartley, 1935; Lennie & MacLeod, 1973; Wyatt, 1972). Although Westheimer found no dichoptic transfer of the effect, later workers (Yu & Essock, 1996a, 1996b; Yu & Levi, 1997) did show weak binocular transfer and demonstrated an alteration of the classical Westheimer curve in amblyopes (Yu & Levi, 1997), which may suggest a cortical component. In addition, Makous (1997) considered the Westheimer paradigm in terms of the spatial Fourier spectra of the stimuli, and interpreted the effect in terms of spatial filters of cortical origin.

Westheimer did not provide a precise definition for the concept of adaptation, but two interpretations are possible. Adaptation may refer to the excitation level within a receptive field; with a high excitation level, a probe stimulus may be less visible. If this is the case,

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responses of on- and off-center cells should be modulated in opposite directions as a bright pedestal surrounding the probe stimulus is increased in size. Alternatively, light adaptation per se may be occurring. Such adaptation may take place within local adaptation pools; adaptation pools close to the size of the X-cell receptive field center have been suggested for cat ganglion cells (Cleland & Freeman, 1988; Enroth-Cugell & Shapley, 1973; Levick, Cleland, & Dubin, 1972). This explanation would predict effects for on- and off-center cells of the same polarity, but not explain the secondary threshold decrease at larger pedestal sizes.

Few physiological studies have looked at the Westheimer paradigm directly. Essock, Lehmkuhle, Frascella, and Enoch (1985) measured responses of cat lateral geniculate nucleus neurons to a temporally modulated sinusoidal probe on different pedestals. On- and off-center X cells showed a biphasic relationship between response and pedestal diameter as in the Westheimer curve, while Y cells showed only a monophasic decrease in responsivity as pedestal size increased. These authors also recognized that on- and off-center cells should show different behavior based on a simple Westheimer model.

Our goal in these experiments was to use a protocol based on the Westheimer paradigm to study local light adaptation in the primate retina and to determine the contribution of retinal components to the effect. We then compared ganglion cell responses to psychophysical contrast thresholds with the sinusoidal probe used in our physiological measurements. We also determined psychophysical Westheimer curves with a variety of temporal waveforms in an attempt to separate the roles of the on- and off-center pathways.

The electrophysiological experiments showed data consistent with adaptation pools comparable to center size in both magnocellular (MC) on- and off-center cells. However, interactions of responsivity with maintained firing rate caused a marked difference in behavior in on- and off-pathways, although we were unable to identify this asymmetry psychophysically. The physiological substrate for the Westheimer paradigm most likely consists of a retinal spatial summation mechanism, involving an adaptation pool nearly the size of a receptive field center, which causes the initial threshold increase. The secondary decrease may be partly associated with changes in retinal maintained activity, but may also reflect the operation of cortical edge detection mechanisms.

2. Methods

2.1. Electrophysiology

Cell responses were recorded extracellularly from the retinas of four adult anesthetized macaque monkeys

(*M. radiata* and *M. fascicularis*). Animals were initially sedated with an intramuscular injection of ketamine (10 mg/kg). Anesthesia was continued with 10 mg/kg thiopental and further maintained with isoflurane (0.2–2%) in a 70%:30% nitrous oxide-oxygen mixture. Local anesthetic was applied to points of surgical intervention. Electroencephalogram, electrocardiogram, heart rate, temperature, and exhaled CO₂ level were monitored continuously to ensure animal health and adequate depth of anesthesia. Muscle relaxation was maintained by a constant infusion of gallamine triethiodide (5 mg/kg/h) with accompanying dextrose Ringer's solution (6 ml/kg/h). Body temperature was kept close to 37.5°C by an electric heating pad. End-tidal CO₂ was brought close to 4% by adjusting the rate of respiration. Urine samples were analyzed for the presence of glucose and protein. All procedures were approved by an on-campus Institutional Animal Care and Use Committee and conformed to the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research.

A tungsten-in-glass recording microelectrode was introduced to the retina through a scleral hole using established techniques (Crook, Lange-Malecki, Lee, & Valberg, 1988). Cells were recorded in the parafoveal retina. Subsequent to isolating a ganglion cell's activity, we ascertained the cell type and plotted receptive field location using a hand-held stimulus projector focused on a tangent screen 226 cm from the eye. MC-pathway cells were identified by their transient responses to all colors, and their high contrast responsivity to achromatic stimuli. Parvocellular (PC) pathway cells were identified by their sustained responses to colored stimuli.

Ganglion cells' spike trains were recorded and stored in peri-stimulus time histograms representing two cycles at 64 bins per cycle. Response amplitude, in the form of first-harmonic amplitude, was derived from the histograms by Fourier analysis.

2.1.1. Stimuli

Visual stimuli were generated through a graphics controller (Visual Stimulus Generator series 3; Cambridge Research Systems, Cambridge, UK) and presented on a 21-in. CRT monitor (CPD-520; Sony, Tokyo, Japan) 226 cm from the eye. Care was taken to ensure proper centering of the stimulus on the receptive field, and centering was controlled by testing a 3 × 3 matrix of test probe locations.

The stimuli consisted of a circular probe 4 min of arc in diameter surrounded by a concentric pedestal of variable diameter ranging from 4' to 192' (Fig. 1a). The central probe was modulated sinusoidally at 4.34 Hz at either 50% or 100% contrast with mean luminance level of 20 cd/m², or at 100% contrast with mean luminance level of 50 cd/m². Maintained activity with the probe unmodulated was also measured as a function of pedestal size. The luminance of the steady pedestal

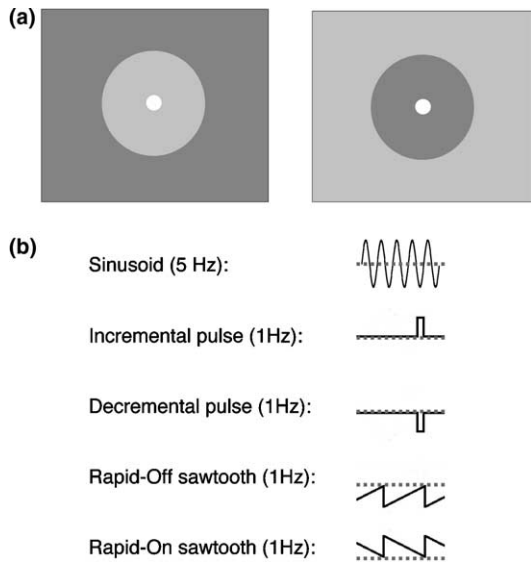


Fig. 1. (a) Left: Spatial luminance profile of Westheimer protocol. Large circular area represents a pedestal that is variable in size. Small central circle represents a probe that undergoes temporal modulation. Right: Inverted contrast protocol. (b) Temporal luminance profiles of central probe. Each luminance profile corresponds to a psychophysical condition. Sinusoidal modulation was at 5 Hz. The incremental pulse was 10 ms. The decremental pulse was likewise 10 ms. The rapid-on condition consisted of a step increase in luminance from pedestal luminance toward maximum luminance followed by a ramp back to pedestal luminance. The rapid-off condition consisted of a step luminance decrease from pedestal luminance toward zero luminance followed by a ramp back to pedestal luminance.

was 20 cd/m^2 and that of the background was 1.31 cd/m^2 . For some cells, a decremental pedestal was also used. It was obtained by switching the luminance of the pedestal and surround (Fig. 1a).

2.2. Psychophysics

2.2.1. Stimuli

Visual stimuli were generated by a graphics controller (Visual Stimulus Generator series 5; Cambridge Research Systems, Cambridge, UK) and displayed on a 21-in. CRT monitor (CPD-520; Sony, Tokyo, Japan) 1.72 m away from the observers, with a 120 Hz refresh rate and a spatial resolution of 1024×764 pixels.

Stimuli similar to those in the physiological measurements were used in the psychophysical experiments. When presented foveally, the central probe was $4'$ in diameter and the pedestal size varied between $4.5'$ and $28'$ in diameter. When presented at 5° extrafoveally, the central probe was $8'$ in diameter and the pedestal size varied between $8'$ and $192'$ in diameter.

The luminance of the central probe was modulated with 5 Hz sinusoidal modulation with mean luminance of 20.5 cd/m^2 , a periodic incremental or decremental pulse (10 ms every 1 s), or a rapid-on or rapid-off sawtooth of 1 Hz (Fig. 1b). Luminance for the incremental

pulse ranged from that of the pedestal (20.5 cd/m^2) to a maximum luminance of 96.3 cd/m^2 . The decremental pedestal was also tested with the sinusoidal probe.

2.2.2. Procedure

Thresholds were measured using the method of adjustment. Observers adjusted the contrast of the probe so that it was just visible. Results are expressed in Michelson contrast for sinusoidal and sawtooth waveforms. Weber contrast is used for pulses. Each session included 13 pedestal sizes and 10 repetitions at each pedestal size. The pedestal size and the initial contrast setting of the probe were randomized from trial to trial.

2.2.3. Observers

An author (JK) and two observers (HT and HP) naïve to the purpose of the experiment participated. The naïve observers were recruited from the State University of New York College of Optometry. Observers ranged in age between 22 and 25 years. All subjects had normal color vision tested with the Farnsworth-Munsell 100 hue test. JK and HT are emmetropic, while HP is myopic and wore corrective lenses. Prior to testing, informed, written consent was obtained from the subjects according to a protocol conforming to the Declaration of Helsinki and approved by the State University of New York, State College of Optometry Institutional Review Board.

3. Results

3.1. Physiological measurements

Responses to the stimulus as a function of pedestal size were obtained from 40 MC cells and 15 PC cells. In addition, several other measurements were made on subsets of cells. These included the protocol with inverted contrast polarity and area-summation estimates with discs of increasing diameter.

3.1.1. Responses of Magnocellular cells

The responses of a typical MC on-center cell to the highest probe contrast are shown in Fig. 2a. Some spike histograms are presented with a plot of response vs. pedestal diameter. The solid line represents a fit of a function described below Eq. (1). With the smallest pedestal diameter, there is a vigorous response to the probe, as is evident in the corresponding histograms. Upon increasing pedestal diameter, the response gradually decreases and eventually plateaus without any secondary rise. Every on-center cell behaved much like the one in Fig. 2a. The plot in Fig. 2b shows the cell's maintained activity at different pedestal sizes with the central probe unmodulated. Maintained firing increases with pedestal diameter and then decreases.

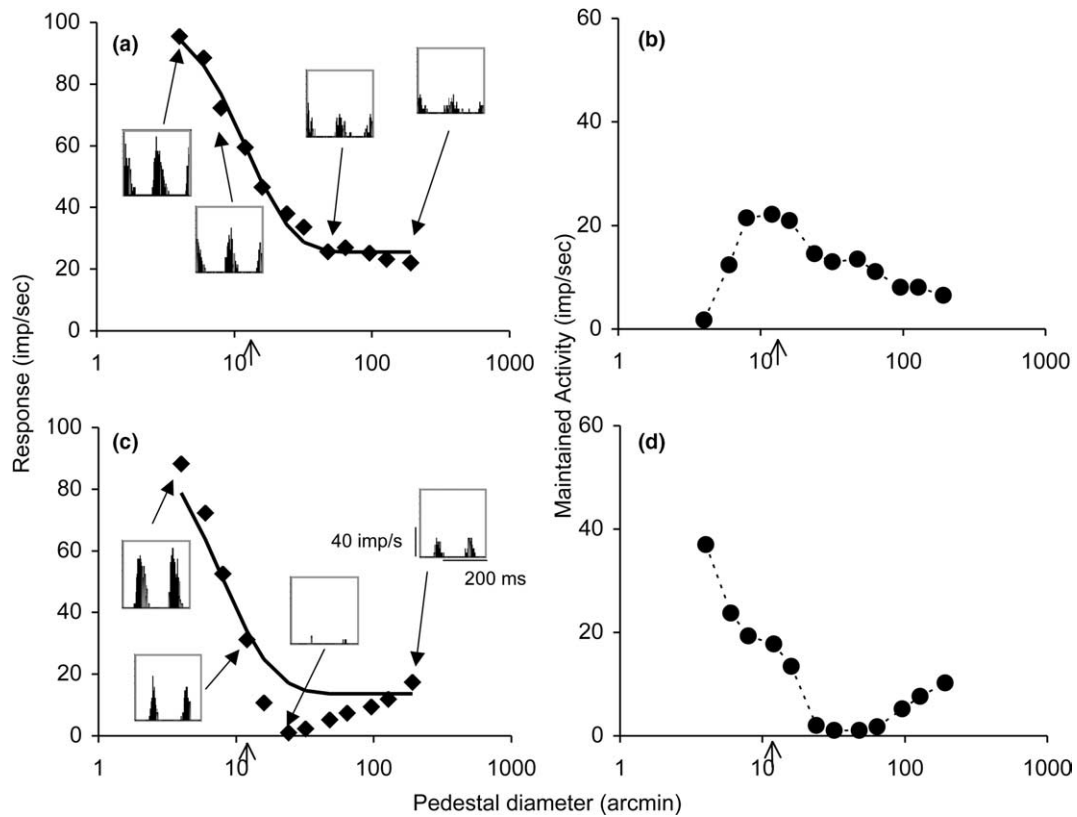


Fig. 2. (a) MC on-center cell (ER73U29) response to Westheimer protocol (100% contrast) with 4.3Hz central modulation. Cell response, in the form of first-harmonic amplitude, was derived from PSTH histograms by Fourier analysis. Solid line represents a model fit to cell response. Fit parameters: $r_c = 8.150$; $c = 0.0072$; baseline = 103.087. Histogram represent two cycles at 64 bins per cycle, corresponding to responses at pedestal sizes indicated by arrows. (b) MC on-center cell maintained activity under Westheimer protocol with steady central probe. (c) MC off-center cell (ER73U13) response to Westheimer protocol (100% contrast) with 4.3Hz central modulation. Solid line represents a model fit to cell response. Fit parameters: $r_c = 7$; $c = 0.02$; baseline = 98.17. Histograms as in (a). (d) MC off-center cell maintained activity with steady central probe (Thin arrows denote receptive field center size.).

Results from a typical MC off-center cell are shown in Fig. 2c. Responsivity was initially high and at first decreased, as in the MC on-center cell. Subsequently, there was a deep dip in responsivity over intermediate pedestal sizes. This was characteristic of MC off-center cells. At larger pedestal sizes, responsivity recovered to a level lower than the responsivity at the smallest pedestal size. The magnitude of response suppression over intermediate pedestal sizes varied from cell to cell. The maintained activity for the MC off-center cells (Fig. 2d) varied in a way opposite to the on-center cell (Fig. 2b). Maintained activity levels of off-center cells were similar at large and small pedestal sizes but a profound inhibition of maintained activity at intermediate sizes occurred. This corresponded to when a pedestal just filled the center (Fig. 2d), as judged from results from area-summation experiments described below. The arrows for each cell show center diameters (95% of Gaussian) so derived. The dip in responsivity in off-center cells we thus attribute to an 'iceberg' effect related to the depression of maintained activity.

Since MC cell responses tend to saturate with increasing contrast (Benardete, Kaplan, & Knight,

1992; Lee, Pokorny, Smith, Martin, & Valberg, 1990), it is possible that responsivity curves as in Fig. 2 could be distorted by response saturation. Three different contrast values were used in the central probe stimulus for all cells. However, all three contrast conditions yielded similar curve shapes, although when responsivity was low the data became noisy. This is illustrated for an off-center MC cell in Fig. 3. Results were also analysed by fitting the response data obtained under the three contrasts with the Naka and Rushton (1966) function. This yielded similar curves to those in Fig. 2a and c, although with more variability. Data obtained using the highest contrast were thus used in the analysis.

MC on- and off-center cells showed a similar decrease in responsivity at the largest compared to the smallest pedestal size. Average small:large pedestal responsivity ratios for all on- and off-center MC cells tested were 3.58 ($n = 20$; SD = 1.15) and 4.01 ($n = 20$; SD = 1.76), respectively. The difference in ratios between on- and off-center cells was not statistically significant ($P = 0.221$). This would be consistent with the operation of an adaptation pool in both cell types.

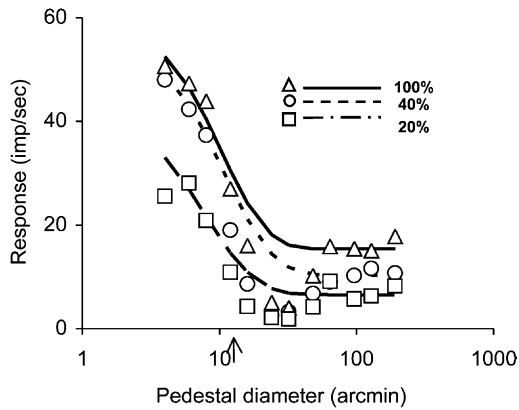


Fig. 3. MC off-center cell responses to central probe modulation at three contrast levels. Lines represent model fits to cell response at different contrasts. Thin arrow denotes receptive field center size.

Data from on-center cells were fitted with a function equivalent to a feed-forward gain control mechanism to estimate the diameter of such an adaptation pool:

$$R(r_{ped}) = \frac{R_m}{1 + c \left(\int_{r_{probe}}^{r_{ped}} F(\sigma_{pool}) dr \right)} \quad (1)$$

where $R(r_{ped})$ is response as a function of pedestal radius, r_{ped} , R_m is the maximum response amplitude (with the smallest pedestal), c is a scalar, and $\int_{r_{probe}}^{r_{ped}} F(\sigma_{pool}) dr$ is the integral of the Gaussian representing an adaptation pool of radius σ_{pool} over the pedestal area. The function gave good fits to response profiles of MC on-center cells, as seen by the solid curve in Fig. 2a. However, for off-center cells, response profiles could not be fit properly due to the substantial drop in response over intermediate pedestal sizes. To attempt to capture some features of the cell response, we fixed the adaptation pool radius at the mean value for on-center cells. The function then successfully captured the initial drop and final recovery level in response at small and large pedestal size, respectively (Fig. 2c). Mean adaptation pool Gaussian radii for on-center cells was 7.36'. This value was close to the range reported for radii of MC-cells' receptive field centers in the parafovea (reviewed in Lee, Kremers, & Yeh, 1998).

3.1.2. Area summation experiments

In order to examine in more detail the relation between the diameter of an adaptation pool and receptive field center size, in a subset of ganglion cells we measured area summation functions directly. A standard area summation protocol (Barlow, FitzHugh, & Kuffler, 1957; Cleland & Enroth-Cugell, 1968; Peichl & Wässle, 1979) was performed. Receptive field center sizes were then compared to adaptation pool sizes derived from analyzing cell responses to the Westheimer protocol.

Responses to 4Hz modulation were recorded at 3–4 contrast levels at different disc sizes. At each disc size, re-

sponse amplitude as a function of contrast was fitted with a Naka and Rushton (1966) function to give an estimate of contrast gain. An example of contrast gain as a function of disc area is shown in Fig. 4 for an on- and an off-center cell. Data have been fitted with a Difference-of-Gaussians (DOG) model. Data and fitted curves from the same cells obtained with the Westheimer protocol are also shown in Fig. 4. Fit parameters are given in the figure legends. For on-center cells tested under both the Westheimer and area summation protocols, adaptation pool radii (mean 6.77'; $N = 10$) obtained using the Westheimer protocol were similar to center radii (mean 5.32'; $N = 10$) obtained using the area-summation method. This direct comparison was not possible for off-center cells. However, for the off-center cell of Fig. 2c, receptive field center radius was used as a spatial parameter in the fit of Eq. (1). The physiological data are consistent with operation of an adaptation pool comparable in size to the receptive field center.

3.1.3. Response and detectability

Response amplitude may not necessarily be a reliable measure of detectability of neuronal signals if there are maintained firing rate changes as in Fig. 2a and b, since this could affect signal-to-noise ratio. An analysis of response variability was thus carried out. Fourier analysis was performed on each of the 64 cycles from our post-stimulus time histograms. The distances (D_i) between the amplitudes of the fundamental Fourier components of the responses to individual cycles of the periodic stimulus and their geometric mean in the complex plane were calculated. Where n is the number of cycles, we defined noise as (Croner, Purpura, & Kaplan, 1993):

$$\text{Noise} = \sqrt{\frac{\sum D_i^2}{n - 1}} \quad (2)$$

Fig. 5 shows noise and response amplitudes, as well as signal-to-noise ratio (first-harmonic amplitude/noise amplitude), of a typical MC on-center cell and an MC off-center cell to pedestals of varying diameter.

For the on-center cell, the level of noise remained similar at all pedestal sizes and the signal-to-noise ratio curve is of similar shape to that of response amplitude alone. For the off-center cell, noise was similar at small and large pedestal sizes, but decreased at the point where the receptive field center was covered by a pedestal. One may interpret the reduction of noise at intermediate pedestal size in off-center cells to be a consequence of the cessation of maintained activity; when no impulses occur, noise is undefined.

These results suggest that, if psychophysical detectability is solely a function of retinal signal-to-noise ratio as for the on-center cell in Fig. 5c, retinal noise does not change with maintained firing so as to affect detectability.

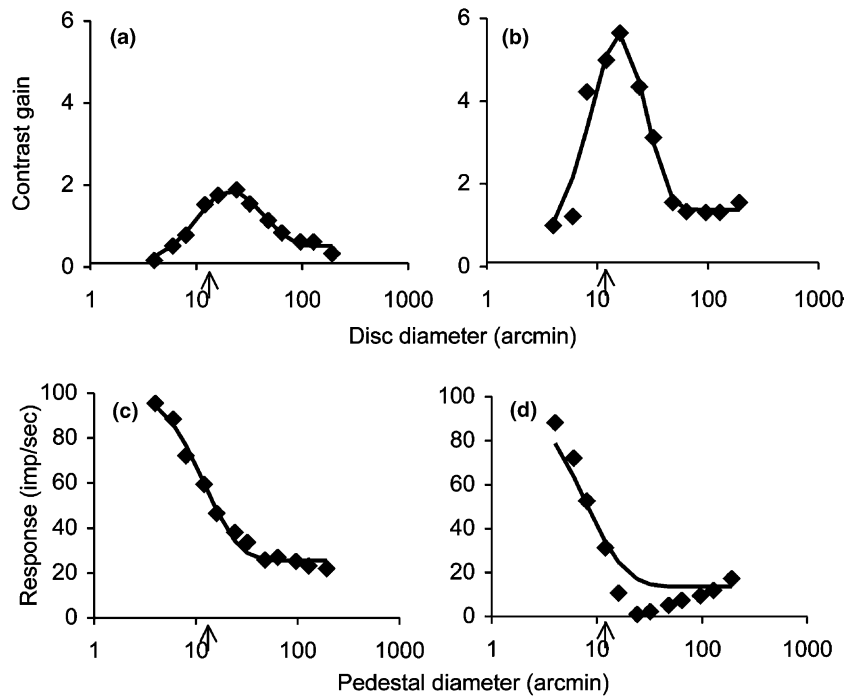


Fig. 4. (a) MC on- and (b) MC off-center cell response to area summation protocol. Solid lines represent DOG fits to response amplitudes. On-center cell fit parameters: $r_c = 3.94$; $c = 0.023$; $r_s = 16.94$; $s = 0.00096$. Off-center cell fit parameters: $r_c = 3.58$; $c = 0.106$; $r_s = 9.25$; $s = 0.0135$. Corresponding cell response to Westheimer protocol (100% contrast) for same (c) on- and (d) off-center cells, respectively (Thin arrows denote receptive field center size.).

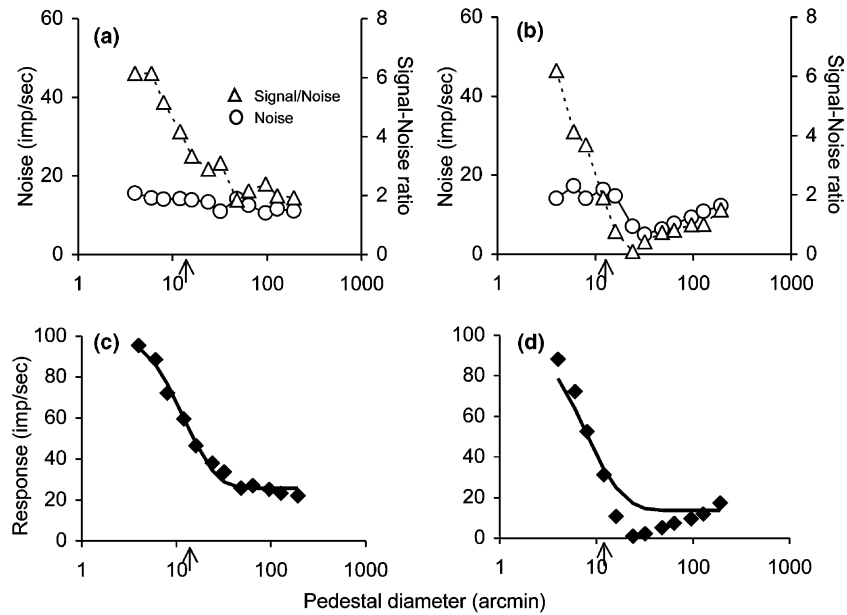


Fig. 5. (a) MC on-center cell noise (circles) as calculated using method in text. Signal/noise ratio (triangles/dotted line) with Westheimer protocol (100% contrast). (b) MC off-center cell noise (circles). Signal/noise ratio (triangles/dotted line) under Westheimer protocol (100% contrast). Corresponding cell response to Westheimer protocol (100% contrast) for same (c) on- and (d) off-center cells, respectively. (Thin arrows denote receptive field center size.).

3.1.4. Parvocellular cells

Responses to a luminance probe at small pedestal sizes of some PC on-center cells were vigorous (Fig. 6a; +L–M on-center cell). As pedestal size increased,

the probe response became greatly attenuated. The same response profile was observed for PC off-center cells (Fig. 6c), but responsivity of off-center cells to the small probe was much lower than that seen in on-center cells

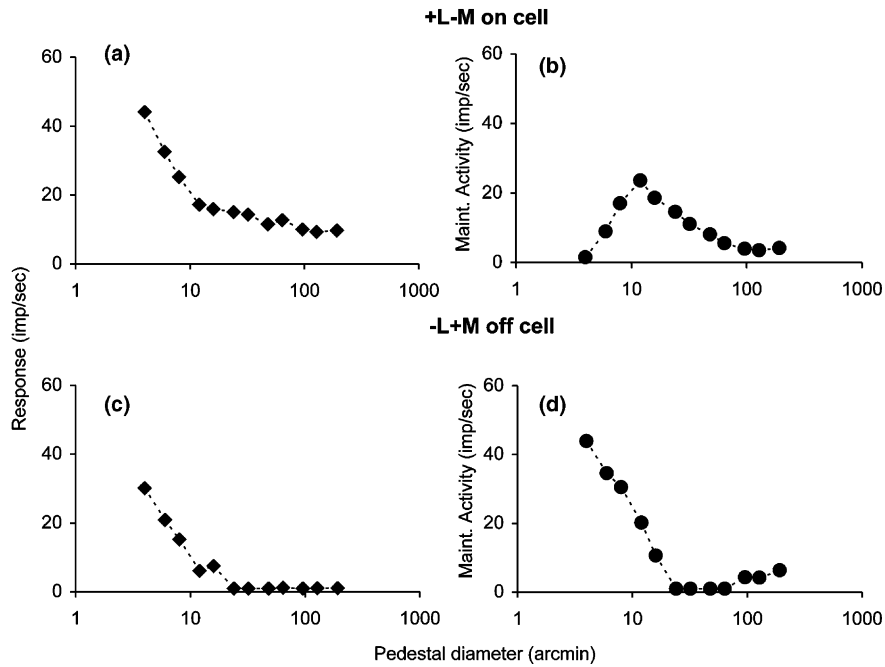


Fig. 6. (a) PC red on-center cell (ER70U8) response to Westheimer protocol (100% contrast). (b) PC red on-center cell maintained activity under Westheimer protocol with steady central probe. (c) PC red off-center cell (ER70U16) response to Westheimer protocol (100% contrast). (d) PC red off-center cell maintained activity under Westheimer protocol with steady central probe.

at intermediate and large pedestal sizes. Under large pedestal conditions, mean response amplitude for PC on- and off-center cells was 14.57 ($N = 12$) and 4.07 ($N = 3$) impulses/sec, respectively. For comparison, mean values for MC cells were 22.14 ($N = 20$) and 19.39 ($N = 20$) impulses/sec, for on- and off-center cells, respectively.

Maintained activity (Fig. 6b and d) also varied in a manner similar to that in MC cells. Although results from PC cells resembled those from MC cells, there was considerable inter-cell variability in responsivity.

3.1.5. Inversion of contrast polarity

We tested the adequacy of the adaptation pool model using an analog of the original Westheimer paradigm in the form of an inverted contrast polarity stimulus. These experiments involved dark pedestals with the same sinusoidal central probe stimulus, as shown in Fig. 1. Such an inversion of the original Westheimer paradigm was also tested by Sinai, Essock, and McCarley (1999), whose main intention was to attempt to isolate the off-pathway psychophysically.

On- and off-center MC-cells showed similar responses to the inverted contrast protocol. Responsivity of typical MC on- and off-center cells was low and changed little at small to intermediate pedestal sizes, as seen in the examples in Fig. 7, which also shows the same cells' data from the normal protocol. Responsivity then increased toward a plateau at larger pedestal sizes. At no point was maintained firing completely abolished for either

cell type, and there was no comparable deep dip in responsivity as seen with the standard protocol. Thus, the similar behavior of on- and off-center cells when maintained activity is present supports the 'iceberg' explanation for the trough in off-center cell responsivity with the standard Westheimer protocol.

We fitted data from on- and off-center cells with Eq. (3), a modification of Eq. (1).

$$R(r_{ped}) = \frac{R_m}{1 + c \int_{r_{ped}}^{r_{\infty}} F(\sigma_{pool}) dr} \tag{3}$$

where $R(r_{ped})$ is response, R_m is the maximum response amplitude, c is a scalar and $\int_{r_{ped}}^{r_{\infty}} F(\sigma_{pool}) dr$ is the integral of the Gaussian representing that part of the adaptation pool covered by the light background. The function gave good fits to response profiles of both MC on- and off-center cells, as seen by the solid curves in Fig. 7a and c, indicating that the adaptation pool model provided a reasonable description of the data.

On comparing large:small pedestal response ratios obtained from the inverted contrast protocol to small:large pedestal ratios obtained from the Westheimer protocol for both on and off cells, there was no statistically significant difference (on-center cells— $P = 0.11$; off-center cells— $P = 0.23$). However, mean radii values obtained for on- and off-center cells were 15.64 and 10.87 ($P = 0.018$), respectively. A statistically significant difference in radii was observed between data obtained using the Westheimer protocol and the inverted contrast

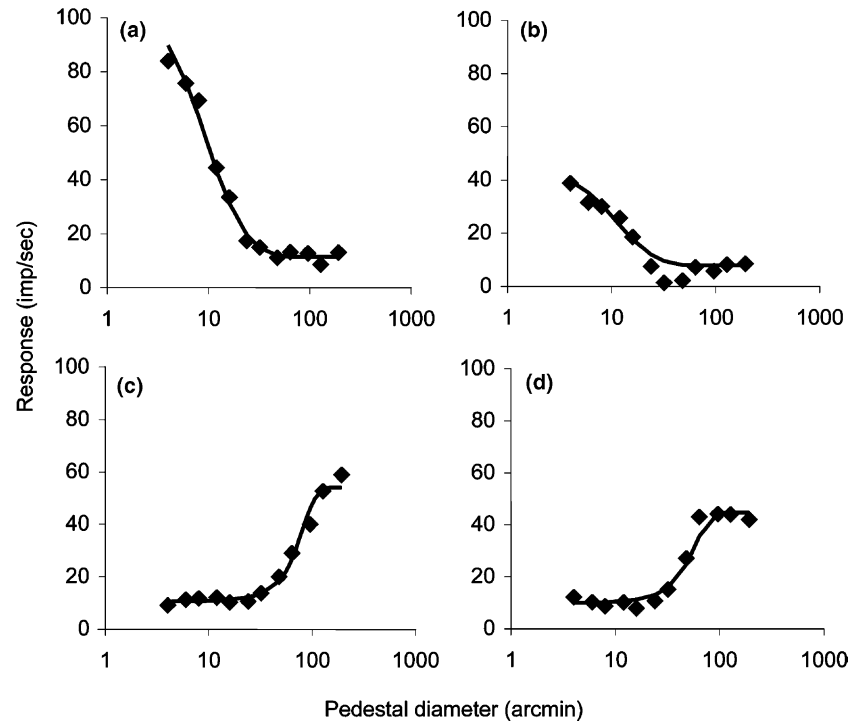


Fig. 7. Comparison between cell response to inverted contrast protocol and Westheimer protocol. (a) MC on-center cell (ER71U1) response to Westheimer protocol (100% contrast). Model (line) fit parameters: $r_c = 9.734$; $c = 0.0137$; baseline = 105.116. (b) MC off-center cell (ER71U7) response to Westheimer protocol (100% contrast). Model (line) fit parameters: $r_c = 9$; $c = 0.009$; baseline = 44. (c) MC on-center cell response to inverted contrast protocol (100% contrast) with central sinusoidal modulation (4.3Hz). Model (line) fit parameters: $r_c = 19.09$; $c = 0.00035$; baseline = 10.52. (d) MC off-center cell response to inverted contrast protocol (100% contrast) with central sinusoidal modulation (4.3Hz). Model (line) fit parameters: $r_c = 14$; $c = 0.00063$; baseline = 10.

protocol for on- ($P = 0.002$) and off-center ($P = 0.011$) cells. This may either be due to light scatter with the dark pedestal protocol or to saturation effects within the adaptation pool.

Psychophysical measurements using inverted contrast stimuli similar to those used in the physiology were performed under foveal fixation. Contrast threshold to central sinusoidal luminance modulation was high at small and intermediate pedestal sizes, but was significantly reduced at large pedestal sizes. The shape of the threshold curves resembled the physiological results.

3.2. Psychophysical measurements

3.2.1. Sine and pulse stimuli

An incremental pulse stimulus has usually been used in the Westheimer paradigm whereas the physiological experiments here used a sinusoidal probe. We therefore measured thresholds for sine and pulse probes and a comparison for two observers is shown in Fig. 8. Curve shapes were similar for the two conditions; this was also the case for a third observer (not shown). We also tested incremental and decremental pulses to try and isolate the MC on- and off-pathways. In view of the differences

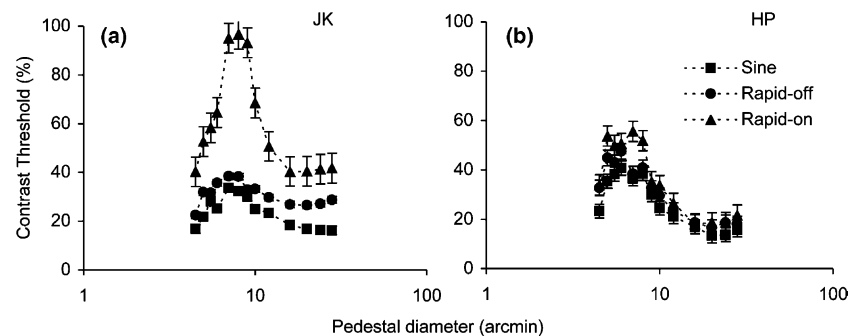


Fig. 8. Psychophysical Westheimer curves obtained with sine, incremental pulse, and decremental pulse probe stimuli for observers JK (a) and HP (b) under foveal fixation.

observed between these pathways physiologically, a difference in curve shape might then occur. However, the curve shapes were again similar, which might suggest that incremental and decremental pulses might not discriminate between on- and off-pathways with our stimulus configuration.

For all three observers, thresholds for the incremental pulse condition were lowest, those for the decremental pulses were the highest, and those for the sinusoidal stimulus condition were intermediate but curves were otherwise very similar in shape for each observer. As in Westheimer’s original data, thresholds peaked at pedestal sizes of approximately 6–7’ in diameter. Results were similar in a control for stray light using a higher background luminance (3 cd/m², rather than 1.31 cd/m²).

3.2.2. Sawtooth stimuli

Sawtooth stimuli may be more effective at isolating on- or off-pathways than pulses (Bowen, Pokorny, & Smith, 1989; Kremers, Lee, Pokorny, & Smith, 1993). We therefore used rapid-on and rapid-off sawtooth modulation; if rapid-off sawtooth stimuli isolate the off-pathway, one might expect a dramatic increase in threshold at intermediate pedestal sizes.

There was very little difference in the shape of the Westheimer curve for each condition (Fig. 9), however, there were differences between absolute threshold levels for the different waveforms, and differences between observers. Bowen et al. (1989) found that some observers showed lower thresholds to rapid-off than rapid-on sawteeth. Differences shown in Fig. 8 were (if present) in the opposite direction.

3.2.3. Parafoveal stimuli

Parafoveal (5°) experiments were performed since the electrophysiological experiments involved ganglion cells located at parafoveal eccentricities (5°–10°). Curves for two observers peaked at ~20’ pedestal diameter (Fig. 10). Such values are similar to the pedestal areas at which MC cell responses plateau for on-center cells and dip for off-center cells.

3.2.4. Small–large pedestal ratios of contrast threshold

Ganglion cells’ responses at small pedestal sizes differed from those at large pedestal sizes by a factor of ~4. In contrast, psychophysical thresholds with large and small pedestals were similar; ratios are summarized in Table 1. Values are generally close to unity, An

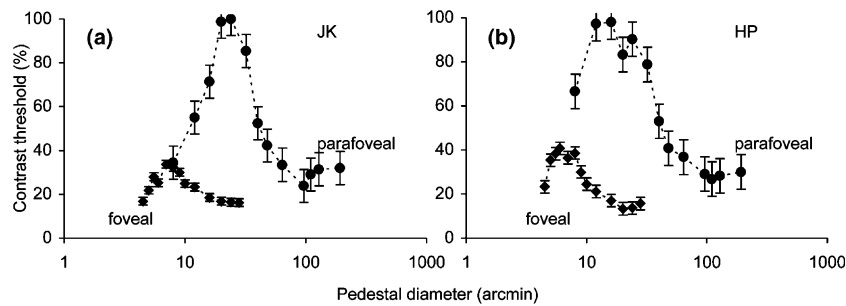


Fig. 9. Psychophysical Westheimer curves obtained with sine (square), rapid-on sawtooth and rapid-off sawtooth probe stimuli for observers JK (a) and HP (b) with foveal fixation.

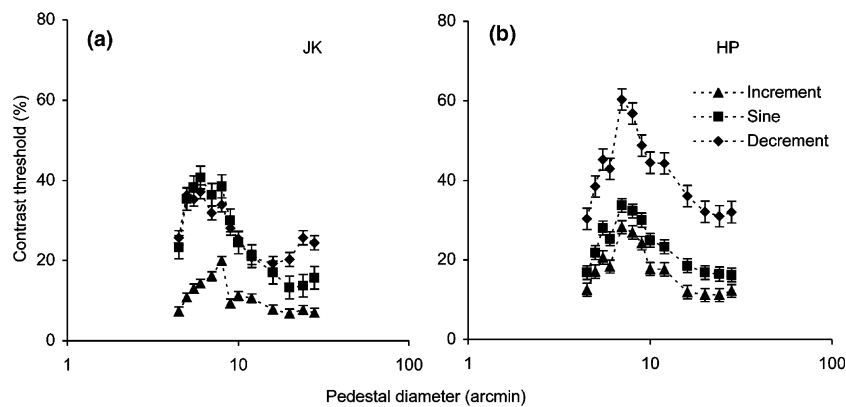


Fig. 10. Psychophysical Westheimer curves obtained from foveal and parafoveal fixation on the central test probe with sine probe for observers JK (a) and HP (b).

Table 1
Small:large pedestal ratios for three subjects under five conditions

	Sine	Rapid-off	Rapid-on	Increment	Decrement
JK	0.96	1.28	1.04	0.98	1.05
HP	0.67	0.55	0.64	0.96	0.95
HT	1.09	1.1	2.24	1.61	n/a
Mean	0.91	0.99	1.31	1.18	1

analysis of variance (ANOVA) yielded an F value of 0.353 indicating no significant between-group difference among the different conditions.

These psychophysical data do not parallel ganglion cell responsivity. Although the initial increase in psychophysical threshold with pedestal size may have a direct retinal correlate, the secondary decrease in psychophysical threshold may not have a direct retinal substrate.

4. Discussion

4.1. Physiological measurements

Westheimer interpreted his original findings in terms of receptive field center-surround organization at the ganglion cell level. The initial increase in threshold was a consequence of the pooling of adaptation signals within an on-cell center and the subsequent decrease a result of inhibitory signals from the surround antagonizing the signals from the center. However, this interpretation is physiologically incomplete (Lennie & MacLeod, 1973) since it leaves the roles of on- and off-pathways unclear and there is no evidence for an antagonistic surround to adaptation pools in the retina. For example, Cleland and Freeman (1988) found that adaptation pools of cat ganglion cells were similar in size to X-cell receptive field centers, but there was no indication of a surround mechanism. Cleland and Enroth-Cugell (1968) had also shown a lack of surround antagonism in their Westheimer-like experiments under scotopic conditions. However, Essock et al. (1985) tested a protocol intended to directly parallel the Westheimer stimulus and found X cells showed a biphasic change in responsivity (a decrease and then an increase) as a function of pedestal diameter, which might be interpreted as a surround effect; Y cells showed a simple responsivity decrease, as in on-center MC cells. The authors did not note any difference between on- and off-center cells. They did not specifically address interactions of responsivity with maintained activity, which may account for the discrepancy with other work.

In any event, data presented here suggest the presence in macaque MC cells of an adaptation pool close in size to the field center, without any antagonistic surround. Adaptation pools were found to be similar in size to MC-cell centers both by comparison with reports in the literature (Lee et al., 1998) and by comparison with

direct measurements. On- and off-center MC cells showed a decline in responsivity from smallest to largest pedestal size despite their opposite center-surround organization, as expected if an adaptation pool (i.e., gain control) mechanism operated on both cell types. However, maintained activity showed opposite behavior for on- and off-center cells, as expected if the changes in maintained activity derived from center-surround mechanisms. For MC off-center cells, maintained activity recovered as pedestals encroached upon the excitatory surround and we suggest that responsivity recovered as a result of resumption of maintained activity.

Cleland and Freeman (1988) suggested that, in the cat, adaptation pools might be located in the photoreceptor layer, with gap junctions mediating spatial summation. However, there is psychophysical evidence (MacLeod & He, 1993; MacLeod, Williams, & Makous, 1992) that adaptation pools in primate outer retina are local, of the dimensions of single cones, and physiological results are consistent with this view (Lee, Dacey, Smith, & Pokorny, 1999); the primate differs from non-mammalian vertebrates in this respect (e.g., Cadenas, Reifsnider, & Tranchina, 1994). MC cells show a greater degree of adaptation than is present in outer retina and show a much greater change in temporal response with luminance level than outer retinal neurons (Lee et al., 1990; Smith, Pokorny, Lee, & Dacey, 2001), which suggests that inner retinal adaptation mechanisms, i.e., adaptation pools, are present for this pathway. It is unclear how this might be managed anatomically. On- and off-center pathways diverge immediately after the cones and ramify in different strata of the inner plexiform layer, yet they presumably share the same light adaptation signal.

The degree of adaptation and temporal response of PC cells do not appear to be substantially modified compared to outer retinal neurons (Lee et al., 1990; Smith et al., 2001). Although there was inter-cell variability, responsivity of PC on-center cells with small pedestals could be vigorous but at large pedestal sizes responses to the probe were weak. The difference in MC and PC luminance responsivity observed with the small probes used here was not as great as observed with large fields (Lee, Martin, & Valberg, 1989) or gratings (Derrington & Lennie, 1984). Local stimulation of a PC cell center by the small probe on a dark background may explain this difference. A gain control arising from a PC cell non-classical surround may attenuate responsivity, since increasing an annular surround size reduces the gain of a PC on-center cell to a central spot (Benardete & Kaplan, 1997; Valberg, Lee, Tigwell, & Creutzfeldt, 1985).

4.2. Psychophysical measurements

The shape of the Westheimer function was found to be similar under all conditions tested. There were

differences between probe waveforms in the size of the initial rise in threshold, but these were not consistent between observers. Sinai et al. (1999) performed psychophysical experiments aimed at isolating the off-pathway using decrement-on-dark stimuli and their results were comparable to ours.

Waveforms which might have segregated on- and off-pathways did not appear to do so in our psychophysical measurements. With large field stimuli, rapid-off and rapid-on sawtooth stimuli may isolate the off- and on-pathways, respectively (Kremers et al., 1993). With large stimuli, contrast at threshold is low and the salient element of the sawtooth is the rapid transition. However, with the small probes used here contrast at threshold was high and observers reported the probe was visible during the slow phase of the sawtooth. This may have led to less effective isolation of the on- and off-pathways. With pulse probes, it is less clear why selective activation was not achieved.

4.3. *The physiological substrate of the Westheimer function*

It is likely that the initial increase in psychophysical threshold with pedestal size has a physiological substrate in the decrease in responsivity observed physiologically. The increase in threshold is of similar amplitude to the decrease in responsivity, and the increase peaks at a diameter which corresponds to MC-cell center size, as originally noted by Oehler (1985). However, the physiological substrate appears to lie in an adaptation pool rather than the center per se.

The secondary decrease in threshold may not have a substrate entirely in the retina. MC off-center responsivity was deeply depressed at intermediate pedestal sizes before partial recovery. If a central detection mechanism relied on activity in both on and off pathways, the recovery in off cells might contribute to the secondary threshold decrease. However, MC on- and off-center cell responsivity at large pedestal sizes was lower than at small pedestal sizes by a factor of ~ 4 . The nearly identical psychophysical thresholds with small and large pedestals (Table 1) support further, possibly cortical, contributions to the Westheimer paradigm. If a central contrast detector mediates threshold, when pedestal area is small there may be masking by the edge of the pedestal (Wyatt, 1972). As pedestal diameter increases, the pedestal edge falls outside the profile of the detector, decreasing masking and contributing to the secondary threshold decrease.

One difficulty in linking the physiology and psychophysics is the presence of fixational eye movements in the awake observer, since transients from this source (Hayhoe & Smith, 1989; Teller, Andrews, & Barlow, 1966; Tulunay-Keesey & Jones, 1977; Tulunay-Keesey

& Vassilev, 1974) may contribute to the Westheimer function. Nevertheless, persistence of sensitization (the secondary threshold decrease) with stabilized images suggests that transients are not the only mechanism involved. Also, image fading makes the stabilized image approach somewhat ambiguous, as does the possibility of residual eye movements (Barlow & Sakitt, 1973; Teller et al., 1966). Finally, eye movements would be expected to have a much smaller effect at higher eccentricities due to increased receptive field size but, apart from spatial scaling with retinal eccentricity (Oehler, 1985), the Westheimer curve retains a similar shape.

Another uncertainty in linking the physiology and psychophysics is the role of maintained activity when detecting a superimposed perturbation in firing. We show in Fig. 4 that changes in maintained activity do not affect response variability, i.e., detectability, of ganglion cell responses per se. However, it is possible to conceive of cortical detectors which might be affected by maintained firing levels. On the other hand, the LGN may partially filter out maintained firing changes (Kaplan & Shapley, 1984; Lee, Virsu, & Creutzfeldt, 1983).

Westheimer (2004) has recently reviewed center-surround antagonism in spatial vision in the context of his paradigm. He concludes that the effect is predominantly retinal, but that some cortical masking effects are also present. Our results would be consistent with this view, although the physiological substrate in the retina may lie in adaptation mechanisms rather than center-surround interaction. However, Westheimer proposes that the changes in the Westheimer function in amblyopia may be due to a retinal deficit. There is substantial physiological evidence from the cat that amblyopia leaves ganglion cell resolution unaffected (Cleland, Crewther, Crewther, & Mitchell, 1982; Crewther, Crewther, & Cleland, 1985); this may suggest a somewhat greater role for cortical processing than he implies.

5. Conclusions

There is extensive evidence that the magnocellular pathway is responsible for detecting achromatic stimuli such as pulses and the sinusoidal probe used here (Lee et al., 1989; Lee, Pokorny, Smith, & Kremers, 1994; Lee et al., 1990) and it is plausible that this pathway underlies detection in the Westheimer paradigm. The results presented suggest that retinal and cortical mechanisms both contribute to the Westheimer result. Were it known how on- and off-pathways combine in cortical contrast detectors, it would be possible to parse the Westheimer paradigm's substrate more precisely.

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