Macaque ganglion cell responses to probe stimuli on modulated backgrounds

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In the natural environment, visual targets have to be detected and identified on changing backgrounds. Here, responses of parasol (magnocellular) ganglion cells to probes on modulated backgrounds are described. At low frequency, the adaptation level of the background influences the probe response, but with increasing frequency there is a strong interaction with the response to the background per se, so that on- and off-center cell responses are modulated in different phases. Interactions with the background response include both thresholding effects (when the cell’s firing is suppressed and no pulse response occurs) and saturation effects (when the background response is vigorous the pulse generates few additional spikes). At 30 Hz, the effect of the pulse is largely a suppression or phase shift of the background response. The data are relevant to the probed-sinewave paradigm, in which pulse detection thresholds are modulated with pulse phase relative to a sinusoidal background. The physiological substrates of the psychophysical results with the probed-sinewave paradigm appear complex, with on- and off-center cells likely to contribute to detection at different pulse phases.

Keywords: ganglion cell, magnocellular, parasol, light adaptation, temporal processing


Introduction

In the natural environment, visual targets must be identified and detected on changing backgrounds. In outer retina, horizontal cell responses to sinusoidal (Lee, Dacey, Smith, & Pokorny, 1999) or pulse probes (Lee, Dacey, Smith, & Pokorny, 2003) added to a sinewave background were studied to investigate mechanisms of adaptation; it was found that adaptation was rapid and cone-specific, although falling short of Weber’s law (Smith, Pokorny, Lee, & Dacey, 2001). Adaptation in ganglion cells has been measured after step changes in illumination in cat (Enroth-Cugell & Shapley, 1973) and macaque (Yeh & Lee, 1996). Gain controls are complete within a few tens of milliseconds or less. However, sinewave backgrounds have not been used at the ganglion cell level. From a psychophysical perspective, sensitivity to flashes presented upon modulated backgrounds has received periodic attention since Boynton, Sturr, and Ikeda (1961) first made such measurements. Although these authors used square-wave modulation, most subsequent workers have used sinusoidal backgrounds and the protocol is often termed the probed-sinewave paradigm. The original motivation for such studies was to investigate the properties of light adaptation; the amplitude and phase lag between the modulation of psychophysical threshold and the modulation of background might indicate the degree and time course of adaptation.

In the probed-sinewave paradigm, psychophysical thresholds of a test probe vary as a function of the phase of the test probe relative to the background, but the threshold-versus-phase curve is frequently not sinusoidal in shape and may be phase shifted; there is an overall elevation of threshold compared to pulses presented on a steady background (reviewed in Hood, Graham, von Wiegand, & Chase, 1997; Wolfson & Graham, 2006). Hood et al. (1997) considered several models of adaptation in relation to the probed-sinewave paradigm and found all of them to be more or less inadequate. Dichoptic presentation experiments have shown that the threshold elevations are almost entirely retinal rather than central in origin (Wolfson & Graham, 2001b).

Physiological substrates and mechanisms were often not addressed in the psychophysical studies. One of these is the presence of both on- and off-center ganglion cells, which might mediate detection (but see Hood & Graham, 1998). On- and off-center cells respond in counterphase
and their responses rectify; during inhibition of firing, a pulse response may not break through the suppression of cell activity, so that pathways could mediate detection at different phases. Another mechanism often omitted is contrast gain control (but see Snippe, Poot, & van Hateren, 2000), which leads to response saturation. Background modulation contrast used in the psychophysical studies has usually been high enough to activate contrast gain controls, and so responses to pulses upon a vigorous background response are likely to be suppressed by response saturation. Contrast gain control is marked in parasol ganglion cells of the magnocellular (MC) pathway (Benardete, Kaplan, & Knight, 1992; Yeh, Lee, & Kremers, 1995) of the primate, which is thought to be responsible for psychophysical detection of luminance modulation (Lee, Pokorny, Smith, Martin, & Valberg, 1990) and pulses (Lee, Pokorny, Smith, & Kremers, 1994).

This paper describes ganglion cell responses to probes on modulated backgrounds. The first aim was to replicate at the ganglion cell level previous work in outer retina (Lee et al., 1999, 2003) and the second aim to investigate the effects of modulated backgrounds on probe stimuli with reference to the probed-sinewave paradigm. Probes were added to sinewave-modulated backgrounds and cell responses measured. At low background modulation frequencies, responses of both on- and off-center MC cells to the background were weak, and probe responses were at least partly determined by background illuminance level. At higher frequencies (e.g., 9.8 Hz), cell responses to the background were vigorous, and the interaction between pulse response and background response became dominant. At 30 Hz, responses to the background were vigorous and in saturation, and responses to the pulsed probe consisted of a suppression or phase disturbance within the ongoing background response. This suggests that detection of targets on modulated backgrounds has a complex physiological substrate.

**Methods**

Cell activity was recorded from the retinas of anesthetized macaques (M. fascicularis). The animals were initially sedated with an intramuscular injection of ketamine (10 mg/kg) followed by thiopental (10 mg/kg). Anesthesia was maintained with inhaled isoflurane (0.2–2%) in a 70:30 N2O–O2 mixture. Local anesthetic was applied to points of surgical intervention. EEG and ECG 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 i.v.) with accompanying dextrose Ringer solution (6 ml/kg/h). Body temperature was kept close to 37.5°C. End tidal CO2 was kept close to 4% by adjusting the rate of respiration. All procedures were approved by the State University of New York Animal Care Committee and conform to the Association for Research in Vision and Ophthalmology guidelines for ethical care of animals.

Neuronal activity was recorded directly from retinal ganglion cells by a tungsten-in-glass electrode inserted through a cannula entering the eye behind the limbus. The details of the preparation can be found elsewhere (Crook, Lange-Malecki, Lee, & Valberg, 1988). We recorded responses of cells between 4° and 12° eccentricity. Cell identification was achieved through standard tests (Lee, Martin, & Valberg, 1989). These included achromatic contrast sensitivity and responses to lights of different chromaticity. Additional tests, e.g., measuring responses to heterochromatically modulated lights (Smith, Lee, Pokorny, Martin, & Valberg, 1992), were employed in cases when identification was difficult. PC cells could generally be identified by their tonic responses and spectral opponency, and MC cells by their phasic responses and lack of spectral opponency. For each cell, the locus of the receptive field center was determined and the stimulus was centered on this point.

Visual stimuli were generated using a light-emitting-diode (LED)-based Maxwellian-view system (Lee et al., 1990). The LEDs were driven by modulation of a high-frequency pulse train, which provided a high degree of linearity. Only two LED sources (modulated in phase) were used in these experiments. In some measurements, they had peak wavelengths of 554 nm and 638 nm, and in others peak wavelengths of 518 and 660 nm. The two sets of LEDs had mean chromaticities with dominant wavelengths of 595 and 578 nm, respectively. The chromaticity of each LED and their relative luminance were calculated from each LED’s spectrum and the Smith and Pokorny cone fundamentals (Smith & Pokorny, 1975). Retinal illuminance was measured (Westheimer, 1966) to be 1500 td. Background modulation and the superimposed pulse or sinewave probes were coextensive and presented as 4-degree fields with a dark surround.

In the set of experiments with sinusoidal probes, a 19.6-Hz sinusoidal probe was superimposed on a sinusoidal background modulated at 0.61, 1.22, or 2.44 Hz. This resembles the protocol used earlier in outer retina experiments (Lee et al., 1999, 2003). Various background and probe contrasts were tested; details are given in the figure legends. In the set of experiments with pulsed probes, a 6.4-ms pulse was superimposed on a sinusoidal background modulated at 1.22, 4.88, 9.76, or 30 Hz, and 33% contrast. The probes were presented at 8 background phases. Three incremental (500, 1000, and 2000 td) and two decremental (–500 and –1000 td) pulse amplitudes were tested. For 1.22- and 4.8-Hz backgrounds, a test pulse was presented on every background cycle. For the 9.8-Hz background, a test pulse was presented every third cycle and at 30 Hz every fifth cycle. Cell responses were recorded for either 20 or 40 probe presentations depending on the background frequency. In addition, control measurements
were carried out at background modulation contrasts other than 33%. Responses to test probes (both sinusoidal and pulsed) were also recorded on steady backgrounds that corresponded to the background luminance levels at the instants when the probes were presented. Times of spike occurrence were recorded to an accuracy of 0.1 ms and averaged histograms of spike trains were simultaneously accumulated.

**Results**

This analysis concentrates on cells of the MC pathway. Responses of a sample of PC cells to the achromatic background modulation and the brief (6.4 ms) luminance pulses were also measured, but PC cell responses to test stimuli were a factor of 6–10 less vigorous than those of MC cells. Data shown here were obtained from a sample of 27 on-center MC cells and 32 off-center MC cells, although few cells were tested on all the protocols. We first describe the responses of cells with sinusoidal test probes to provide a link to outer retinal data. We then describe the response of cells with pulse probes. Last, we consider the detectability of pulses by central mechanisms.

**Cell responses to sinusoidal or pulse probes on a background**

**Sinusoidal probes**

Sinusoidal probes and backgrounds are a convenient physiological stimulus, since responses are readily amenable to linear analysis. The stimulus protocol is shown in Figure 1A. A test probe (19.5 Hz) is added to a sinusoidal background (0.61 Hz) to give the waveforms shown; stimulus modulation contrasts are described in the legend. Figure 1B shows the response of an H1 horizontal cell to this protocol, as described elsewhere (Lee et al., 1999); the top trace shows the response to the combined waveform, and the middle trace to the background alone; the traces have been vertically displaced for clarity. The probe response in the bottom trace is derived by subtraction of background response from combined responses; it is modulated by the background, and its amplitude can be described by a divisive adaptation mechanism with delay (Lee et al., 1999).

Figure 1C shows responses of on- and off-center MC cells to this protocol, again with a background modulation
of 0.61 Hz and a test probe of 19.5 Hz. The responses to the background alone (middle traces) differ in shape for the on- and off-center cells and are not precisely in counterphase. This is typical of MC cells at low frequencies, probably due to an interaction between on and off cell responses with ongoing adaptation mechanisms. At higher frequencies, on and off cells respond in counterphase (Lee et al., 1990; see examples in later section). The response to the combined stimulus (upper traces) shows the modulation due to the test probe; there is a difference in modulation profile over the background cycle for the two cells, with a larger test response when a background response is present. This is illustrated in the lower panels, where the background histogram has been subtracted from the “background + test.” This is most obvious for the off-center cell; when its firing is suppressed by the high luminance of the background in the first half of the histogram, there is little response.

We attempted to describe MC cell responses with a modification of the divisive model used for horizontal cells. Figure 2 shows this approach for the on- (Figure 2A) and off-center cells (Figure 2B) of Figure 1. The traces in the top panels show the responses to the combined “background + test” waveforms and the background alone, replotted from Figure 1. The combined waveform response \( R(\omega t) \) was fitted with

\[
R(\omega t) = \frac{R_0}{B\left(C_{bg}\sin(\varphi_{bg} + \varphi_{delay}) + 1\right) + 1 + R_{bg}},
\]

where the sinusoidal test probe, of frequency \( \omega \) with a response of phase \( \varphi_{test} \), is represented by the second sine term. The first fractional term represents a gain control in which \( R_0 \) is a scaling parameter, \( B \) determines the degree of adaptation caused by the background, \( C_{bg} \) is background contrast, \( \varphi_{bg} \) is background phase, and \( \varphi_{delay} \) is a phase delay representing the time course of adaptation. \( R_{bg} \) is the measured background response waveform. Free parameters are \( B, \varphi_{delay}, R_0, \) and \( \varphi_{test} \). The middle panels in Figures 2A and 2B show the background response used.

Figure 2. Qualitative description of responses of cells of Figure 1 ((A) on-center, (B) off-center) based on a divisive adaptation model. (Top traces) Histograms from Figure 1, smoothed and superimposed. (Middle traces) Smoothed background response redrawn from the top panel, and putative test response modulated as described in text. (Lower panels) Background response again replotted, together with sum of measured background response and test model response, followed by rectification. The sum curves were fitted to the measured background + test curves (upper panels) using a least-squares criterion and give a reasonable fit.
in the fit (given negative values for the off-center cell when its firing is inhibited) and the modulated test response. The sum of modulated test response and the background was rectified and was fitted to the data using a least-squares criterion.

The lower panels show the fitted curves for the combined response and the replotted curves for the background response alone. There is a good correspondence between the actual combined response data (top panel) and the fitted curves (bottom panel). This simple approach indicates that an early rapid adaptational gain control added to the background response provides a reasonable first approximation to cell behavior at these low frequencies, when background responses are not vigorous.

We tested background frequencies up to 2.44 Hz, above which the limited number of test cycles per background cycle made interpretation difficult. To analyze the data, we used the same approach as in the horizontal cell analysis. We subtracted the response to the background alone from the combined response, as in the lower panels in Figure 1C. Each cycle of the test response was then Fourier analyzed and the 1st harmonic amplitude plotted against background phase. The results are shown in Figure 3 for 0-, 0.61-, 1.22-, and 2.44-Hz background frequencies; 0 Hz refers to the steady-state condition, where responses were measured at different luminance levels corresponding to the instantaneous background luminance level at each phase. Data shown in Figure 3 are averaged from 3 to 8 MC cells for each curve. At 0 Hz, the test response modulates with adaptation level and follows a similar course for the on- and off-center cells. The difference between maximum and minimum responses is approximately that expected from Weber’s law. At 0.61 Hz, the phase difference between on- and off-center cells seen in Figures 1 and 2 becomes apparent, and by 2.44 Hz the response modulation of on- and off-center cells is almost in counterphase. We tested three different test amplitudes and two background contrasts for each cell and found a broadly similar pattern of results except for changes in response amplitude.

These data suggest that modulation of responsivity of on and off pathways by the background is determined by adaptation level at very low background frequencies but moves into counterphase as frequency increases to 2.44 Hz.

Use of sinusoidal test probes is impractical at still higher background frequencies. Pulse probes do not suffer from this limitation, and in the next section, we describe responses to incremental and decremental pulses.

### Pulsed probes

We measured responses of MC cells to pulses superimposed on sinusoidal backgrounds of five different frequencies (0 to 30 Hz) at 8 different background phases. In preliminary experiments, we explored several background contrasts with both incremental and decremental pulses. We used a 33% background contrast, which permitted use of decremental pulses; for each background condition, 3 incremental or 2 decremental pulse amplitudes were tested. We first give a qualitative description of cell responses.

Figure 4 shows responses of on- and off-center MC cells to a pulse probe (500 td) presented at difference phases of a sinusoidal background (33% contrast). Figures 4A–4C show responses with different background frequencies, and each column represents responses at a given background phase, while the 5th column shows responses to the background alone.

Figure 4A shows responses to 0-Hz background, i.e., when pulses were delivered at different steady luminance levels corresponding to the instantaneous background luminance level at the phase of the sinusoidal modulation. The on-center cell responds with a short latency, transient burst of impulses. Excitatory responses of off-center cells have a longer latency and duration; they correspond to the second lobe of the biphasic impulse response function.

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**Figure 3.** Effects of increasing temporal frequency on amplitude of test responses. First harmonic test-response amplitudes (mean of 4 on-center, 4 off-center cells) are plotted as a function of the phase of the test cycle within the background (background phase). Four frequencies are shown. 0 Hz corresponds to the steady-state condition, i.e., test probes presented at different mean luminances. Responsivity of on- and off-center cells modulates in phase. At the three higher frequencies, the responsivity of on- and off-center cells pulls apart in phase as frequency increases. Retinal illuminance of 1500 td, response average of 20–40 background cycles per condition. Background contrast: 80%, test contrast: 20%.
expected of these transient cells with a light pulse. The shape and duration of the impulse response expected at the illuminance level used (Lee et al., 1994) have been drawn into each panel. Modulation of pulse response amplitude varies with the background illuminance level, becoming larger at the lower illuminance level. The change roughly corresponds to Weber’s law; response amplitude at 90 compared to 270 deg differs by about a factor of two, which is expected at the 33% contrast level.

With 1.22- and 4.88-Hz backgrounds (Figures 4B and 4C), there is an interaction between the pulse response and the background response, although clear peaks can still be distinguished with reference to the impulse response inserts. When the pulse response superimposes upon the excitatory response to the background, a peak is seen, but this is less vigorous when the pulse response corresponds to a response trough (e.g., 4.88 Hz, on-center cell, phase 180 deg). Thus, there is a phase shift between the effect of the background on on and off pathways. The ordinate scale has been changed at 4.88 Hz to accommodate the more vigorous background responses at this frequency.

Figure 5 shows data from the two higher frequencies tested (±1000 td pulse amplitude). At 9.76 Hz, responses to the background are vigorous. If the pulse response occurs at a phase at which a response to the background is present, there is an incremental peak. If the timing is such that it falls in a period where cell firing is inhibited because of the background, little or no response is elicited. Impulse responses have again been drawn in for reference.

We also tested a 30-Hz background; this was the frequency employed by Boynton et al. (1961). MC cells respond vigorously at this frequency at the retinal illuminance used. The impulse response inserts provide a
reference as to where a response should be expected. Where the added pulse is at a phase when an increase in the background response peak might be anticipated, little enhancement is apparent, presumably due to response saturation. However, responses to the background can be reduced, abolished, or phase shifted by the probe. This appears to occur when a negative lobe of the impulse response corresponds to the background peak.

Responses to decremental probes showed a similar pattern (not shown), except that pulse responses were inverted in polarity.

It should be stressed that these experiments involve repetitive delivery of the pulse probes on the continuous background. Inspection of the histograms in Figures 4 and 5 suggests that the response to the background had recovered to a steady state before the next pulse was delivered; note that for the 9.8- and 30-Hz background modulations, pulses were delivered every third and fifth background cycles, respectively. In the following analysis, we will treat the responses to the pulses as independent events with the assumption that the effects of one pulse had dissipated before the next test.

These results are consistent with those using sinusoidal probes, i.e., as frequency increases, it is interaction with the background response that is the major determinant of the resultant response.

**Interaction between probe and background responses**

The results presented suggest that at least three factors play a role in shaping responses on modulated backgrounds. Adaptation changes cell responsivity as a function of luminance level. This is obvious at low background frequencies when MC cell responses are weak, in the limit at 0 Hz (the steady-state condition). The second factor is rectification; when a cell’s firing is inhibited, responses to an added pulse cannot generate a peak. Since on- and off-center cells respond in counterphase, this effect results in counterphase modulation of their responsivities. Lastly, if responses to the background modulation are vigorous and the cell response is in saturation (for example, a background modulated at 30 Hz), an added pulse generates little excitatory response, but responses to the background may be reduced or phase shifted. We implemented a simple model (results not shown) in which a pulse response was added to a modulated background, and responses modified by rectification and a saturating non-linearity. The general features of the responses in Figures 4 and 5 could be reproduced.

With 30-Hz backgrounds, additional complications are present. One of these is physical distortion of the background wave by the superimposed pulse. It has usually
been assumed that the energy in the pulse is small compared with that in the background modulation. However, as frequency increases, the pulse occupies a significant fraction of the background waveform. This may significantly distort the Fourier spectrum of that cycle where the pulse occurs. To provide an estimate of the amount of distortion expected, Fourier analysis was performed on the “background + pulse” waveforms with pulses presented at various phases relative to the background (33% contrast). The higher harmonic components introduced by the pulses have been neglected. The amplitude of the first harmonic is plotted as a function of pulse phase in the top panel of Figure 6A. There is a modulation of the background contrast, which ranges from ca. 22% to ca. 44% around the 33% baseline. There is also some distortion of phase (Figure 6A, lower panel).

A further complication with a 30-Hz background is that cells’ impulse responses are of comparable duration to the background period. Figure 6B sketches this effect for on- (middle panels) and off-center (lower panels) cell responses to a pulse presented at 2 different phases (left and right columns) of a sinusoidal background. The top traces show stimulus waveforms for two pulse phases; background and pulse are plotted separately. In the panels below are impulse responses of a duration and waveform expected from MC cells at the retinal illuminance used (Lee et al., 1994) and the expected rectified background responses with appropriate phase (Lee et al., 1990). Since the time course of the impulse response is similar to the cycle period, for 90 deg pulse phase the pulse and background responses might be expected to augment one another for both on- and off-center cells, whereas for the 180 deg phase a cancellation is expected for both cells. This is illustrated in the linear sum curves below. Response saturation has not been taken into account in this sketch; it is intended as a qualitative illustration of timing interactions when the duration of the impulse response becomes comparable to the cycle length.

We stress these last two factors, since at lower frequencies pulse responses of on- and off-center cells may modulate out of phase through the background cycle, whereas at 30 Hz the effects shown in Figures 7A and 7B will cause their responses to modulate in phase.

Detectability of pulse responses

Psychophysical approaches to pulse detection in the probed-sinewave paradigm often assume some kind of peak detector (e.g., Hood et al., 1997). However, the data in Figures 4 and 5 suggest that at high temporal frequencies response peaks are less apparent, and that an elision of response or phase shift is the most obvious effect of the pulse. We therefore undertook a neurometric analysis based on the structure of responses. We do not suggest that central detection mechanisms operate in this way; our aim was to perform an analysis that makes optimal use of the neural signal.

Figure 7A shows the impulse response of an MC cell at mean retinal illuminance used in these experiments (Lee et al., 1994). It consists a biphasic response with a total duration of 40–50 ms and a latency of 10–15 ms. To capture this response, we Fourier analyzed each impulse train in a window encompassing the response. The window started with a 15-ms latency and had a width of 40 ms (25 Hz). Figure 7A shows the window fitted onto the impulse response. We assumed that the presence of the modulating background does not substantially alter the time course of the impulse response.

If response to a pulse is superimposed onto a background response, an interaction as in Figure 7B would be expected. In the 40-ms window, a 1st harmonic Fourier component is present due to the background alone, dependent on phase. By comparing the 1st harmonic amplitude and phase for “background + pulse” with those for background alone, we could estimate the effect of the added pulse. This estimation neglects higher harmonic distortions in the cell response but gave a satisfactory first approximation for quantifying the effect of the pulse response on the background. For the different pulse phases on the background, the analysis window was set so that it aligned with the beginning of the stimulus pulse. We verified that minor variation of latency and cycle duration of the Fourier analysis did not substantially affect the analysis.

Figures 7C and 7D display averaged 1st harmonic amplitudes of on- and off-center cells at two frequencies (1.22 and 30 Hz). Responses for the 8 phases tested have been plotted on vector coordinates, and results for each phase pair (background + pulse and background alone) connected by line segments. Data are averaged over 3–5 cells per point. Results were similar for all pulse amplitudes (the intermediate pulse amplitude is shown here). Seven other cells for which the full range conditions were not tested showed similar results.

For the 1.22-Hz data, there is little 25-Hz response to the background alone and the data points are near the origin, i.e., the cell response to the 1.22-Hz background alone has little energy at the frequency that corresponds to the 40-ms analysis duration. With a test, the response vectors of on- and off-center cells (the “background alone” to “pulse + background” line segments) are radial, but in counterphase, with respect to the origin. This corresponds to the response to the pulse being a sharp peak superimposed on the ongoing background response (as was apparent from the histograms in Figure 4), with inverted test response polarity for on- and off-center cells. The response vectors of both on- and off-center cells to the superimposed pulse vary in length with background phase.

For the 30-Hz background modulation, the width of the 40-ms analysis window is similar to the background period. The 1st harmonic response to the background
alone is vigorous, and it rotates around the origin with the pulse phases (which determined the start of the analysis period). The points for the “background + pulse” conditions have been connected by solid lines, and “background alone” by dashed lines. Response vectors for the on- and off-center cells are approximately in counterphase (see color coding). The vectors connecting the point pairs for different pulse phases are generally not radial in direction, indicating that the added pulse changes the phase of the background response. In addition, the most prominent effect of the pulse is a reduction of 1st harmonic amplitude; points for the “background + pulse” condition often lie closer to the origin than with background alone. The reduction is most marked at 180–270 deg for both on- and off-center cells. This differs from behavior at lower frequencies, where modulation of responsivity of on- and off-center cells tended to be in counterphase.

For the 0-Hz steady-state condition (not shown), the results of this analysis, when plotted as in Figure 7, are straightforward since no background response is present. Response amplitude was dependent on background level. At 4.88 and 9.8 Hz (not shown), features intermediate between the 1.22- and 30-Hz data in Figures 7C and 7D were observed.

We assessed detectability of pulses added to the background assuming that the length of the line segments connecting the “background + pulse” and “background alone” points in Figure 7 represent the available signal to a central detector. This resembles an ideal “single-cell observer” approach (Lee, Sun, & Zucchini, 2007). This is unlikely to be completely realistic, and possible limitations are taken up in the Discussion section.

For each frequency and pulse phase, vector lengths for the three pulse amplitudes were fitted with either a linear relationship, or a Naka–Rushton function, or a linear relation with a threshold (the threshold represents conditions when the inhibition from the background response prevent the low pulse to break through). We estimated cycle-to-cycle variability in the response to “pulse + background” and to background alone for the different pulse phases from individual spike trains. As a measure of response variability, noise (effectively a standard deviation of response vectors) was calculated as in Croner,
Purpura, and Kaplan (1993) and averaged over the different pulse phases. For all background frequencies, noise was similar for “pulse + background” versus background alone (within shaded regions) gives an estimate of the effect of the added pulse on the background waveform. (C) First harmonic responses plotted in vector coordinates for pulse responses on a 1.22-Hz background. The color-coded symbols indicated data for the different pulse phases. For the on- and off-center cells, response vectors are roughly in opposite phases, because the impulse responses are inverted. Response vectors linking “background alone” and “background + pulse” are radial, indicating that the 1st harmonic response to background alone is small, and the analysis is capturing the response peak. (D) Similar analysis for 30-Hz background. Since background period is now comparable to the analysis window, there is a substantial 1st harmonic component to background alone (symbols connected by dashed lines). Responses to “background + pulse” (symbols connected by solid lines) show tangential (i.e., phase shifts) as well as radial components. There is little excitatory response to the pulse; the most prominent feature of responses is a decrease in background response peak (solid lines lie within dashed lines). Examination of the color coding shows that it is the same pulse phases (blue, yellow) that generate the largest decrement in response for both on- and off-center cells; 1000 td pulse data used, average of 3–6 cells per point.

Figure 7. Neurometric analysis of responses. (A) Sketch of sine window used for Fourier analysis relative to anticipated cell impulse response. Fourier analysis of spike trains was performed for a 40-ms window beginning 15 ms after the start of the test pulse; the window width roughly matches the duration of the impulse response. (B) A comparison of response amplitude and phase for “background + pulse” versus background alone gives an estimate of the effect of the added pulse on the background waveform. (C) First harmonic responses plotted in vector coordinates for pulse responses on a 1.22-Hz background. The color-coded symbols indicated data for the different pulse phases. For the on- and off-center cells, response vectors are roughly in opposite phases, because the impulse responses are inverted. Response vectors linking “background alone” and “background + pulse” are radial, indicating that the 1st harmonic response to background alone is small, and the analysis is capturing the response peak. (D) Similar analysis for 30-Hz background. Since background period is now comparable to the analysis window, there is a substantial 1st harmonic component to background alone (symbols connected by dashed lines). Responses to “background + pulse” (symbols connected by solid lines) show tangential (i.e., phase shifts) as well as radial components. There is little excitatory response to the pulse; the most prominent feature of responses is a decrease in background response peak (solid lines lie within dashed lines). Examination of the color coding shows that it is the same pulse phases (blue, yellow) that generate the largest decrement in response for both on- and off-center cells; 1000 td pulse data used, average of 3–6 cells per point.

A similar analysis was performed on responses to decremental pulses. Cell threshold curves (not shown) at lower frequencies resembled those in Figure 8, i.e., modulation of threshold for on- and off-center cells was in counterphase. However, at 30 Hz, an analysis as in Figure 6 predicts that effects of contrast on the background and interactions of background and pulse response should invert and undergo a 180 deg phase shift. The pattern of responses plotted as in Figure 7D was inverted (not shown). Cell threshold curves for decremental pulses
at 30 Hz are plotted in Figure 8F. The modulation in cell threshold has now undergone a 180 deg phase shift for both on- and off-center cells relative to the incremental pulse condition. These data suggest that at low frequencies, modulation of responsivity of on- and off-center cells occurs in phase since adaptation mechanisms are dominant. As frequency increases, thresholds for on- and off-center cells pull apart and become out of phase. However, at 30-Hz modulation of on- and off-center pathways come back into phase again. These physiological results indicate that detection of targets on modulated backgrounds has a complex physiological substrate and is difficult to attribute to a single adaptation process.

Discussion

It is shown here that the physiological response to test probes upon a modulated background has a complex substrate. Light adaptation per se would appear to play only a minor role in modulating the responses to pulse or sinewave probes except at the lowest temporal frequencies (ca. 1 Hz or less). At higher frequencies, interaction between the probe response and the background response itself appears to be the main determinant of probe responsivity, even though outer retinal data suggest that adaptation mechanisms are able to follow to these frequencies (Lee et al., 2003).

We have focused on responses of MC cells since available evidence suggests that this pathway is responsible for detection of luminance modulation. Responses of PC cells to brief luminance pulses on luminance-modulated backgrounds were weak and difficult to analyze. We also measured responses of PC cells to chromatic probes on luminance- and chromatic-modulated backgrounds. This protocol has received little psychophysical attention, and physiological and psychophysical results will be reported elsewhere.

Two interactions between background and test probe are demonstrated here. First, probe responses are suppressed when the probe is presented at such a background phase that a cell’s firing is inhibited. Second, when a cell is responding very vigorously to the background, especially at higher frequencies (9.8–30 Hz), the background saturates the cell response and little additional response...
can be evoked by an added pulse. Because of these effects, probe responsivity tends to modulate in counterphase for on- and off-center cells, except at high frequencies (30 Hz in our data) when the duration of a background cycle period is so short that further interactions tend to bring modulation of on- and off-center cell responsivities back into phase rather than in counterphase, as was shown in Figures 7 and 8.

We performed a neurometric analysis based on a Fourier analysis of the period within the background response when the pulse response occurred. We chose a period of 40 ms based on the duration of the impulse response function of MC cells. One critical assumption in the analysis is that this time course is not changed by the background. Judging from inspection of the histograms, superimposed pulse responses and recovery of the background response appeared to be consistent with this time window. In addition, we tested durations of the analysis window from 30 to 60 ms and did not find any substantial change in the results.

These data are relevant to psychophysical results using the probed-sinewave paradigm. It has been noted (Wolfson & Graham, 2006) that although the general pattern of psychophysical findings appears to be maintained between studies, there is significant variability, especially in the mid-frequency range. For example, some studies report two peaks in threshold through the background cycle as pulse phase is varied. The dual-peak effect may be due to an envelope of on and off channels, both of which can mediate detection. In any event, in view of the presence of complex interactions in the underlying physiology, the balance between different physiological effects in psychophysical measurements is likely to depend on several factors, for example, background contrast, background frequency, and pulse duration. In addition, retinal illumination level will change the temporal dynamics and the duration of the impulse response, which may also modify interactions. Lastly, the spatial structure of the stimulus might also play a role. In some reports, background and probe were coextensive, in others the probe diameter was smaller than the background, with or without a sharp boundary between them.

The analysis of Hood and Graham (1998) explicitly modeled the effect of on and off pathways. At low temporal frequencies (ca. 1 Hz), the modeled ganglion cell responses show rectification and they proposed that psychophysical thresholds follow the envelope of sensitivities of on and off pathways (their Figure 4). The 1.22-Hz analysis presented here partially resembles their prediction. They also considered the 30-Hz background condition. However, their analysis does not correspond to our results here. The main reason for the discrepancy is probably that their model did not include response saturation. On the whole, however, the data presented here are consistent with psychophysical detection derived from an envelope of on- and off-pathway sensitivities.

Our analysis predicts that modulation of psychophysical thresholds should be similar for incremental and decremental pulses except at 30 Hz. Wolfson and Graham (2001a) compared psychophysical thresholds to incremental and decremental probes at a variety of background frequencies and found only minor differences in the relation between psychophysical threshold and pulse phase on the background. However, we show here that both incremental and decremental probes may be detectable by both on and off pathways (as these authors discuss) with similar threshold/pulse phase relationships. Thus, the similarity of the incremental and decremental pulse curves in Wolfson and Graham is consistent with our results.

However, these authors also asked observers to identify pulses as increments and decrements and found that identification was possible at threshold under most conditions. Nevertheless, our informal observations indicated that incremental and decremental pulses presented upon modulated backgrounds, although perhaps distinguishable, are not obvious as increments and decrements, except at the lower temporal frequencies. This issue thus remains unresolved, although one factor is that Wolfson and Graham used small probes on a large background (and thus spatial context was present) whereas we used coextensive probes and backgrounds.

The data in Figure 8 are based on a neurometric analysis. How far central mechanisms make optimal use of ganglion cell signals is uncertain. The neurometric analysis depends on both changes in response amplitude and phase being used by a detection mechanism. This assumption may not be satisfactory. More realistically, three possible central detector mechanisms might be considered. The first possibility is a straightforward pulse detector looking for peaks on the ongoing background response. We tested such an algorithm, but it did not work well at 9.8 and 30 Hz (not shown). The second possibility is a mechanism that detects vector length (with a phase component, as in the Figure 8 analysis), but psychophysical ability to discriminate temporal phase decreases rapidly above 10 Hz (White, Sun, Swanson, & Lee, 2002). A third possibility would be a mechanism that detects changes in response amplitude of the background irrespective of phase. The underlying physiological substrate is so complex that distinguishing between these possibilities is difficult.

Detection of targets upon modulated backgrounds is likely to occur in the natural environment, and how this is achieved is pertinent to recent analysis of physiological and psychophysical responses to natural scenes. The probed-sinewave paradigm provides a direct test of performance in this context, although its original goal, to investigate mechanisms of adaptation, may not have been achieved. We are currently exploring psychophysical correlates of the results presented here, such as the use of rapid-on and rapid-off sawteeth to better separate on- and
off-center pathways. One further feature, especially of natural scenes, is the presence of spatial context. Psychophysical and physiological variants of the probed-sinewave protocol in which more spatial context is provided may be valuable for interpretation of visual detection under changing conditions.

Conclusions

MC ganglion cell responses to probed-sinewave paradigm proved to be much more complex than those of horizontal cells. Light adaptation per se would appear to play only a minor role in modulating the responses to pulse or sinewave probes except at the lowest temporal frequencies (ca. 1 Hz or less). At higher frequencies, interaction between the probe response and the background response itself appears to be the main determinant of probe responsivity.

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References


