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Retinal Ganglion Cell Adaptation to Small Luminance Fluctuations

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Freeman DK, Graña G, Passaglia CL. Retinal ganglion cell adaptation to small luminance fluctuations. *J Neurophysiol* 104: 704–712, 2010. First published June 10, 2010; doi:10.1152/jn.00767.2009. To accommodate the wide input range over which the visual system operates within the narrow output range of spiking neurons, the retina adjusts its sensitivity to the mean light level so that retinal ganglion cells can faithfully signal contrast, or relative deviations from the mean luminance. Given the large operating range of the visual system, the majority of work on luminance adaptation has involved logarithmic changes in light level. We report that luminance gain controls are recruited for remarkably small fluctuations in luminance as well. Using spike recordings from the rat optic tract, we show that ganglion cell responses to a brief flash of light are modulated in amplitude by local background fluctuations as little as 15% contrast. The time scale of the gain control is rapid (<125 ms), at least for ON cells. The retinal locus of adaptation precedes the ganglion cell spike generator because response gain changes of ON cells were uncorrelated with firing rate. The mechanism seems to reside within the inner retinal network and not in the photoreceptors, because the adaptation profiles of ON and OFF cells differed markedly. The response gain changes follow Weber's law, suggesting that network mechanisms of luminance adaptation described in previous work modulates retinal ganglion cell sensitivity, not just when we move between different lighting environments, but also as our eyes scan a visual scene. Finally, we show that response amplitude is uniformly reduced for flashes on a modulated background that has spatial contrast, indicating that another gain control that integrates luminance signals nonlinearly over space operates within the receptive field center of rat ganglion cells.

INTRODUCTION

A ubiquitous property of biological systems is adaptation, and as a consequence, much research is devoted to elucidating the biophysical mechanisms underlying the phenomenon and understanding its role in normal system function. A well-documented example is luminance adaptation (also known as light adaptation), which increases or decreases the gain and dynamics of retinal neurons to maintain visual sensitivity and avoid response saturation as mean illumination level changes (Shapley and Enroth-Cugell 1984; Walraven et al. 1990). The mechanisms responsible for this sensitivity adjustment exist both within the photoreceptor cells (Dowling 1967; Perlman and Norman 1998; Pugh et al. 1999; Schneeweis and Schnapf 1999) and the retinal network (Cicerone and Green 1980; Dunn et al. 2007; Green and Powers 1982; Rushton and Westheimer 1962). Another mechanism of sensitivity adjustment at work in the retina is contrast adaptation. It modulates response gain and dynamics based on the variance of luminance in the scene (i.e., contrast), regardless of the mean luminance level (Baccus and Meister 2002; Chander and Chichilnisky 2001; Kim and Rieke

2001; Manookin and Demb 2006; Shapley and Victor 1978; Smirnakis et al. 1997). The mechanism has processes that operate both within and outside the classical receptive field of ganglion cells.

Although the evidence for luminance and contrast adaptation is ample and compelling, the extent to which the two mechanisms are physiologically distinct is not entirely clear. Luminance adaptation is generally described as a logarithmic process, which normalizes local variations in incident light intensity by the ambient level so that the retinal output faithfully represents the contrast of objects viewed in different lighting environments (Troy and Enroth-Cugell 1993). The process is often modeled using a quasi-linear approach, in which the retinal response to a stimulus is specified by a spatiotemporal filter that is parameterized by the steady-state retinal illuminance (Purpura et al. 1990; Smith et al. 2008). It is known, however, that retinal neurons integrate luminance adaptive signals over a region of space about the size of their receptive field center (Cleland and Enroth-Cugell 1968). Given the small size of the luminance adaptation pool, it has been suggested that the process might be recruited under constant illumination conditions because head and eye movements across the visual scene would produce local fluctuations in mean light intensity over the retinal surface (Dunn and Rieke 2006). In support of the idea, the range of mean luminances within patches of natural scenes about the size of ganglion cell receptive field centers was found to differ by a factor of 2 within regions of sky and a factor of 10 within foliage backlit by sky (Frazor and Geisler 2006). These ranges are substantial and, based on ganglion cell and geniculate cell responses to luminance steps of similar amplitude (Mante et al. 2005; Wark et al. 2009; Yeh et al. 1996), they have the potential to recruit light adaptive mechanisms.

The aim here was to evaluate the hypothesis with in vivo recordings from retinal ganglion cells and directly assess how much luminance variation is needed to produce measurable changes in response gain. We show that rat ganglion cells adapt to relatively small fluctuations in local light intensity (>15% contrast), which is remarkable for a system that operates over several orders of magnitude. The sensitivity is comparable to that reported for temporal contrast adaptation in the retina (Baccus and Meister 2002; Chander and Chichilnisky 2001; Kim and Rieke 2001; Manookin and Demb 2006), raising the possibility that the phenomena may be the same or closely related. We find that the luminance adaptive mechanism is distinct though from spatial mechanisms of contrast adaptation. Because both modulate ganglion cell responsiveness, the two gain controls must act in concert under normal viewing conditions to dynamically shape the retinal output. In addition, we find differences in the adaptation profiles of ON and OFF cells, implying that the gain changes occur downstream of the photoreceptors, after the retinal signal is split into ON and

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OFF pathways. This inner retinal component of luminance adaptation is thus distinct from outer retinal components that are also quite sensitive (Lankheet et al. 1993; Lee et al. 2003). Together, the findings show that receptoral and postreceptoral mechanisms of luminance gain control play an active role in the dynamic encoding of visual scenes.

METHODS

Physiological preparation

The experimental procedures have been described in detail elsewhere (Freeman et al. 2008). Briefly, adult Brown Norway rats were anesthetized with an intraperitoneal injection of ketamine hydrochloride (49 mg/kg) and xylazine (1 mg/kg) that was supplemented intravenously at 28.6 and 0.6 mg/kg/h, respectively, infused at a rate of 1.6 ml/h. The pupil was dilated with 1% atropine sulfate, and a clear contact lens was placed over the eye to protect the cornea from drying. The animal was positioned in a stereotaxic apparatus, and a 5-mm hole was drilled in the skull over bregma. To prevent eye movements, which would cause the location of the ganglion cell receptive field to drift during the experiment, the animal was paralyzed with an intravenous injection of 8 mg gallamine triethiodide supplemented by a 24.3-mg/kg/h infusion. The animal was mechanically ventilated with room air at a rate that maintained end-tidal CO_2 at a normal level. All procedures were approved by the Boston University Animal Care and Use Committee.

Recording and visual stimulation

Custom-made tungsten-in-glass microelectrodes were used to record extracellularly from rat retinal ganglion cell axons (Levick 1972). The electrode was advanced into the brain through a protective guide tube located 0.5 mm anterior to bregma for optic chiasm recording or 1.5 mm lateral to bregma for optic tract recording. A video monitor with 100-Hz frame rate (Multiscan 17se, Sony, 40.4 × 30.2 cm) was placed 16 cm from the eye. The mean luminance was 26 cd/m², which is in the photopic range for rat (Cicerone and Green 1980). Custom software controlled the monitor output using Matlab, LabView, and the Psychophysics Toolbox (Brainard 1997). The monitor was gamma corrected to linearize the input-output relationship governing monitor intensity. Upon isolation of a single unit spike train, the receptive field was located and centered on the monitor. Spike times were recorded with a digital spike discriminator at 10 kHz (Acquisition Processor Module, FHC, Bowdoin, ME).

Cells were classified using previously described methods (Freeman et al. 2008). Briefly, ON- and OFF-center cells were determined from their response to handheld wands. Cells having a center-surround receptive field structure and brisk response were identified as X- and Y-like (Cleland and Levick 1974), whereas those with more complex receptive field structure and a sluggish response were considered W-like (Troy and Shou 2002). The latter were encountered too infrequently for inclusion. X- and Y-like cells were differentiated using a sinusoidal grating of high spatial frequency that reversed sinusoidally in contrast at 2 Hz. Unlike X cells, Y cells produce a strong second harmonic (F2) response for gratings that are too fine to elicit a modulated response (F1) from the center mechanism (Hochstein and Shapley 1976). Brisk cells that had a significant F2 response (F2 > F1) were therefore considered Y-like cells and those that did not were considered X-like cells. Data for both cell types were pooled after the results showed no clear differences between them.

The location of the receptive field center on the monitor was determined using a 100% contrast bipartite field oriented vertically and then horizontally. When the contrast border of the stimulus is centered on the receptive field, alternating contrast produces no F1 response in X-like cells and a pure F2 response in Y-like cells. The size of the receptive field center was estimated from the largest spot

diameter that gave an F1 response in the linear range of 5–10 imp/s (ips) to sinusoidal input of 2 Hz.

Probed-sinewave paradigm

The luminance of a spot overlaying the receptive field center was modulated sinusoidally in time at a given contrast (0–30%) and frequency (0, 0.25, 1, 4 Hz), where contrast is defined as

$$C_S = \frac{L_{max} - L_{min}}{L_{max} + L_{min}} \times 100\%$$

where L_{max} is the maximum luminance and L_{min} is the minimum luminance of the spot. The effect of the sinusoidal modulation on ganglion cells sensitivity was probed using a 100-ms flash presented at various temporal phases (0, 45, 90, 135, 180, 225, 270, and 315°), where 90° is the brightest phase and 270° is the darkest. Each flash was presented 8–12 times in random order and consecutive flashes were spaced ≥6 s apart. The contrast of the probe was defined as

$$C_P = \frac{L_{probe} - L_{mean}}{L_{mean}} \times 100\%$$

where L_{mean} is the mean luminance of the monitor. Probe contrast was adjusted to give a response that is less than half-maximal and within the linear range of the cell. ON cells were presented with bright flashes and OFF cells with dark flashes. The peak firing rate of probe responses was measured after subtracting the cell's response to sinusoidal modulation with no flash. Probe responses on a modulated background were normalized by the control response to a probe on a constant gray background. The control flash was presented 10 times in 3-s intervals before and after the probed-sinewave stimulus to confirm that ganglion cell sensitivity did not drift. The control response was measured as the peak firing rate minus the resting rate. Firing rate was calculated with a 40-ms bin size and smoothed with a 60-ms moving average window. For 0-Hz modulation, spot luminance was held for several seconds at the intensity level specified by each phase of the sinusoidal modulation, and sensitivity was probed at each phase in random order.

Probed split-sine paradigm

To assess the spatial integration of adaptive signals within the receptive field center, the spot was split in half vertically, and each half was countermodulated. The modulation waveform was a 0.25-Hz sinewave of 25% contrast. Again, ganglion cell sensitivity was probed by flashing a 100-ms spot before, during, and after background modulation. The flash stimulated both halves of the receptive field center equally. Probe response amplitude was measured with respect to the background rate, which equaled the resting rate in this case because the counter-phase modulation centered on the receptive field produces no F1 response.

RESULTS

The threshold for luminance adaptation was studied in rat retinal ganglion cells with a probed-sinewave stimulus paradigm, in which the luminance of a small spot matched in size to the receptive field center of recorded cells was sinusoidally modulated in time against a uniform gray background. The mean background luminance was the same for all experiments and within the photopic range of the animal. For various modulation phases, the spot was flashed brighter for ON cells or darker for OFF cells to probe for response adaptation. The flash interval was always >3 s, because initial experiments showed this gave independent probe responses. The stimulus paradigm has several attractive features. First, it has frequently been used in psychophysical studies, which have shown that a phase-

dependent modulation of flash sensitivity is indicative of luminance adaptation and a phase-independent reduction in sensitivity of contrast adaptation (Hood et al. 1997; Snippe et al. 2000; Wolfson and Graham 2006). This facilitates interpretation of our results, because the distinction between the processes blurs when small luminance fluctuations are involved. It also facilitates comparisons of our results to humans. Second, the probed-sinewave paradigm avoids onset and offset transients that can obscure responses to the probe stimulus if background luminance were stepped up and down. Third, it confines the adaptive signal to the receptive field center, isolating the actions of local luminance gain controls from the potentially confounding effects of contrast gain controls in the extraclassical receptive field of retinal ganglion cells (Enroth-Cugell and Jakiela 1980; Passaglia et al. 2001, 2009).

Adaptation to local luminance fluctuations

Figure 1 shows the effect of local luminance fluctuations on the sensitivity of a typical ON cell. The modulation waveform in this case was a 0.25-Hz sinewave of 35% contrast. The baseline sensitivity of the cell ($R_{control}$) was measured before and after the stimulus paradigm by flashing the spot against the unmodulated gray background (Fig. 1A). Flash contrast was fixed at 50% or less relative to this background so that probe responses were in the linear range of the cell based on measured contrast response functions (Fig. 1B). Data were excluded from analysis if $R_{control}$ showed drift. Probed-sinewave responses (R_{sine}) were quantified by subtracting the response to background modulation without

flashes (Fig. 1C). The ratio of the two measurements ($R_{sine}/R_{control}$) defined the change in response gain of the cell, which depended on the phase of flash presentation with respect to the local background modulation (Fig. 1D). The flash response grew by ~30% when spot intensity decreased and shrank by roughly the same amount when spot intensity increased. This nonlinear behavior is indicative of a luminance adaptive process, which is known to counteract increases in light intensity with decreases in response gain and vice versa. The data show that the process is sensitive enough to modulate ganglion cell responsiveness even when local luminance fluctuates by a fraction of a log unit and scene luminance is basically constant.

It may be noted that flash contrast is defined here relative to the ambient light intensity. If flash contrast were instead defined relative to the instantaneous local luminance, it would not be constant. It too would vary sinusoidally in time. This raises an alternative interpretation of the results, which is that flash responses got smaller when spot intensity increased and larger when intensity decreased because flash contrast varied, and ganglion cells are known to act as contrast detectors. This interpretation does not, however, detract from the conclusion that spot modulation caused light adaptation. The system must still adapt to local luminance for flashes containing the same number of photons to produce responses of different size.

Sensitivity of luminance adaptation to stimulus contrast

The contrast of the sinewave was altered to determine the threshold amount of background modulation needed to measur-
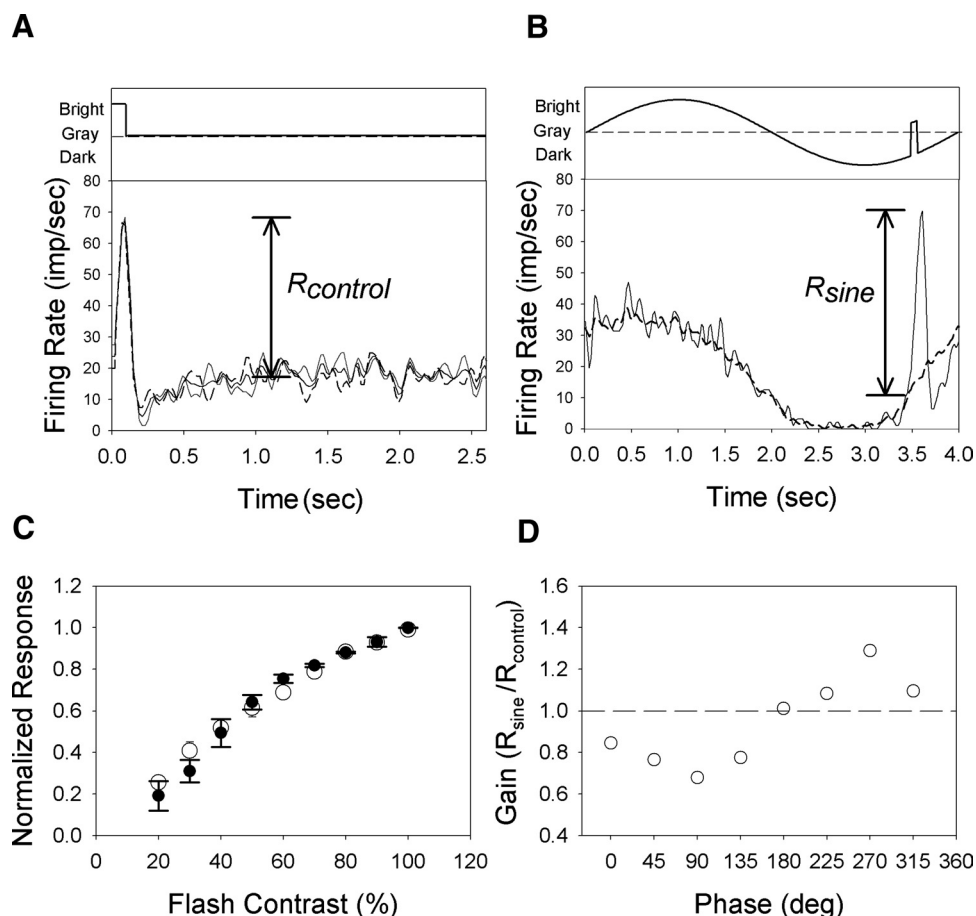

FIG. 1. A: the response of an ON cell to a 100-ms flash (averaged over 10 presentations) on a uniform gray background presented both before (gray line) and after (dashed line) the probed sinwave stimulus. The average of these 2 responses (solid line) is used to estimate the amplitude of the control flash response ($R_{control}$). B: the response to a sinusoidally oscillating spot of 35% contrast and 0.25-Hz frequency (dashed line) and the response to the same sinusoid with a 100-ms flash presented at a phase of 315° (3.5 s) (solid line). Flash response amplitude is quantified as R_{sine}. Above each response in A and B is an illustration of the stimulus. C: average response to 100 ms flashes of varying contrasts for ON cells (white circles, $n = 6$) and OFF cells (dark circles, $n = 6$). Response is normalized to the response at 100% contrast. D: response gain, or sensitivity, of the ganglion cell as estimated by the amplitude of the flash response presented at various phases of the sinusoid relative to the control ($R_{sine}/R_{control}$).

ably activate the luminance gain control, and the results were averaged across all ON cells ($n = 7$ for 5% contrast, $n = 8$ for 15 and 25%, $n = 9$ for 35%). No change in response gain was detectable at 5% contrast, but the gain varied significantly above and below the baseline level for contrasts of 15% and higher (Fig. 2). These contrasts are within the threshold range for evoking a response from rat ganglion cells (Fig. 1B). Across the cell population, flash response amplitude was consistently reduced for phases that the spot was brighter than gray (45–135°) and enhanced for phases that the spot was darker than gray (225–315°). For the highest contrasts, the response enhancement was underestimated for certain dark phases as the modulated rate of some ON cells fell to zero (Fig. 4). That such low contrasts not only affected response gain but increased the gain for some phases makes it unlikely that static nonlinearities like spike rate clipping or saturation were involved. The data also provide little evidence for contrast adaptation. Such a process would suppress probe responses at all phases (Hood et al. 1997; Wu et al. 1997), but adaptation profiles appear fairly symmetric about the unity gain line at low contrast and are not centered markedly below.

The probed-sinewave experiment was also carried out for a population of OFF cells using a range of modulation contrasts ($n = 8$ for 5%, $n = 6$ for 15 and 25%, $n = 7$ for 35%). For these cells, the probe flash was darker than background so that the flash response was also an increase in spike rate. Because OFF cells have a lower maintained discharge than ON cells (Freeman et al. 2008), the background modulation often caused clipping of the probe response at zero firing rate for some modulation phases. The response clipping makes a decrease in gain hard to interpret. Nevertheless, an increase in responsivity

above baseline was apparent for several flash phases at modulation contrasts of 15% or more (Fig. 3), indicating that OFF cells adapt to relatively small fluctuations in luminance as well.

The adaptation profiles of ON and OFF cells show a marked phase difference. Response enhancement was maximal in ON cells when the spot was darkest (~270°), whereas it was maximal in OFF cells when the spot transitioned from light to dark (~180°). If the gain control mechanism responsible for these changes resides in photoreceptors, the adaptation profiles of ON and OFF ganglion cells should be mirror opposites because the photoreceptor output would pass to both ON and OFF bipolar cells. The gain control thus seems distinct from outer retinal adaptation described in cone and horizontal cells (Lee et al. 2003; Schneeweiss and Schnapf 1999) and lies within the inner retinal circuitry. A likely site is the signal transfer from bipolar cells to ganglion cells. Postreceptor adaptation was recently localized to this junction in primate (Dunn et al. 2007), although it was not the dominant site for ganglion cells at photopic light levels. Based on our results, separate gain controls must exist for the ON and OFF pathways to have different adaptation profiles.

Independence of response adaptation from response rate

The effect of local background modulation on response gain was not coupled to its effect on mean firing rate. Figure 4 plots the rate modulation profile of ON and OFF cells for different contrasts. The profile was constructed by dividing the sinewave modulated rate with the resting rate and averaging across cells. ON cells tended to fire maximally at 45°, whereas OFF cells tended to fire maximally at 180°. The fact that ON and OFF cell responses at 0.25 Hz are not 180° apart is indicative of filtering differences between
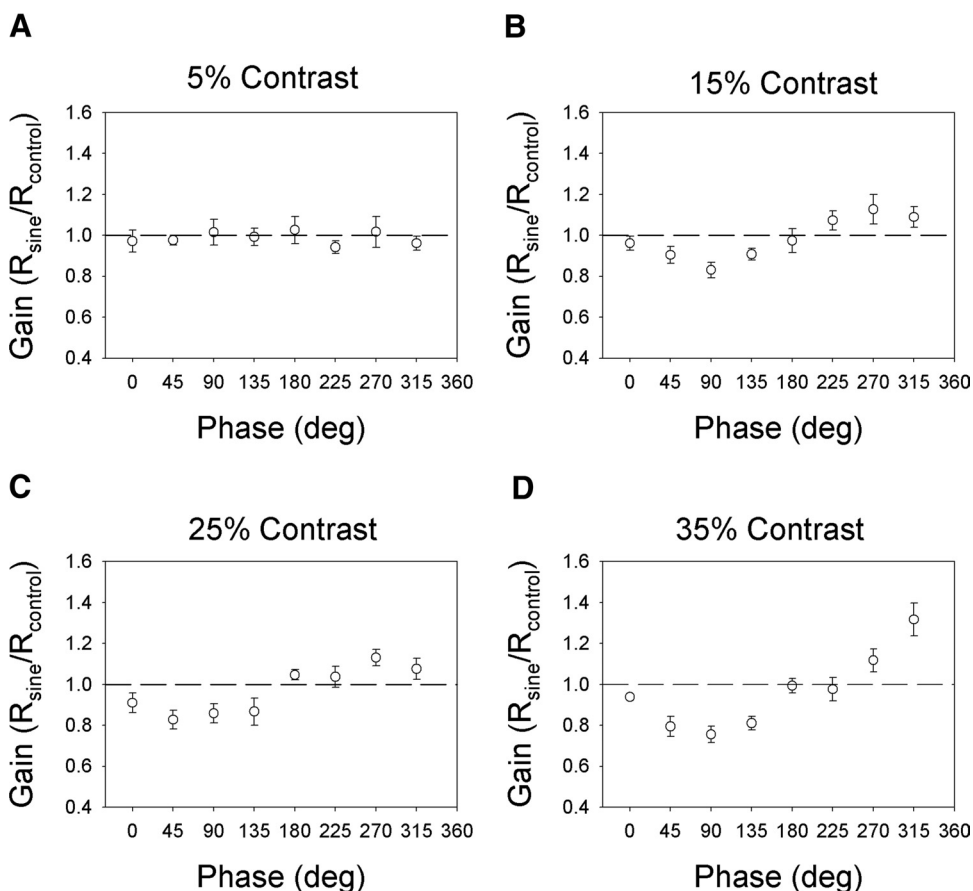

FIG. 2. A–D: results of the probed sinewave paradigm averaged over all ON cells for a sinusoidal frequency of 0.25 Hz and varying contrasts. The sensitivity oscillates above and below the baseline level, indicative of luminance adaptation. Best fit sinusoids of 0.25 Hz show the depth of modulation. Error bars indicate SE.
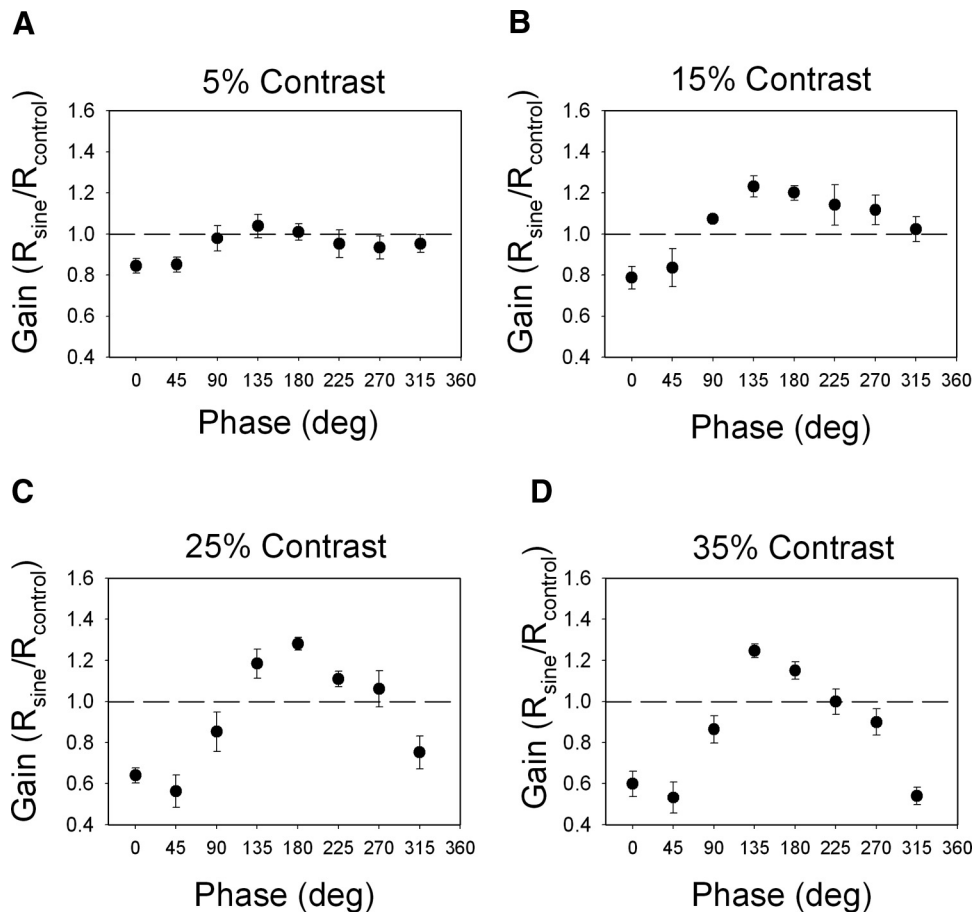
A

B

C

D

FIG. 3. *A–D*: results of the probed sinewave paradigm averaged over all OFF cells for a sinusoidal frequency of 0.25 Hz and varying contrasts. Although the reductions in sensitivity may be exaggerated by response clipping, the increases in sensitivity above baseline must be caused by luminance adaptation. Error bars indicate SE.

the ON and OFF pathways, which has been reported previously (Chichilnisky and Kalmar 2002). The rate modulation profile of ON cells was phase advanced by ~225° with respect to their probe adaptation profile (cf. Figs. 2 and 4*A*). This means that, when the modulated rate equaled the resting rate, the response gain was significantly reduced for some phases (135°) and elevated for others (315°). The response reduction at resting spike rates is additional evidence against the gain changes being a product of response compression. The phase difference between the two profiles also means that the effect was not a property of the ganglion cell spike generator, like spike frequency adaptation (O'Brien et al. 2002). This further limits the adaptive process to a site between bipolar cell and ganglion cell dendrites, at least for

the ON pathway. The location is less certain for OFF cells. Although changes in response gain and firing rate were correlated, response clipping in this pathway could have masked a small phase difference between the probe adaptation and rate modulation profiles.

Temporal dynamics of luminance adaptation

To determine whether the observed sensitivity changes require the luminance to be modulated dynamically in time, spot luminance was held constant for several seconds at the levels specified by different phases of a 25%-contrast sinewave, and response gain was probed at each phase in
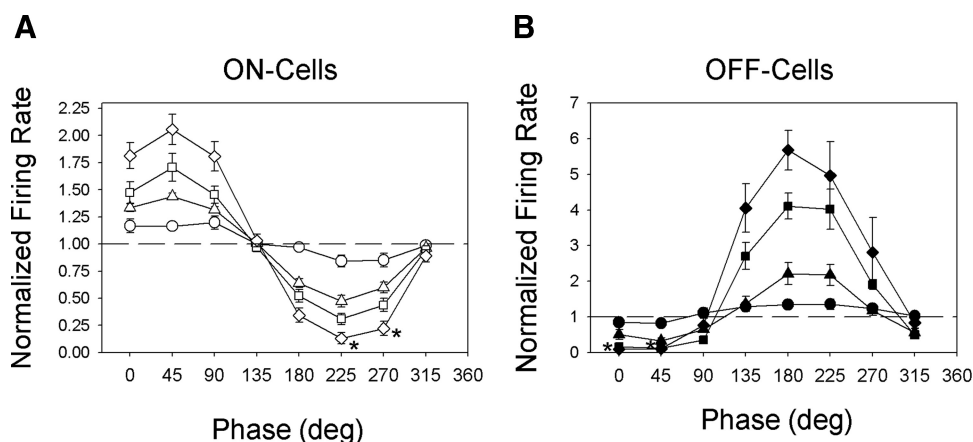
A

B

FIG. 4. The modulation depth for ON cells (*A*) and OFF cells (*B*) was measured in response to sinusoidally varying luminance at 0.25 Hz at various contrasts. The normalized response was quantified as the fractional deviation from the mean rate where a value of 1 represents no change in firing rate. Average results over all cells are shown for contrasts of 5 (circle), 15 (triangle), 25 (square), and 35% (diamond). Asterisks show the points in which the firing rate was occasionally clipped at 0 imp/s, causing an underestimate in gain.

A

0Hz

B

1Hz: ON-Cells

C

1Hz: OFF-Cells

D

4Hz: High MFR
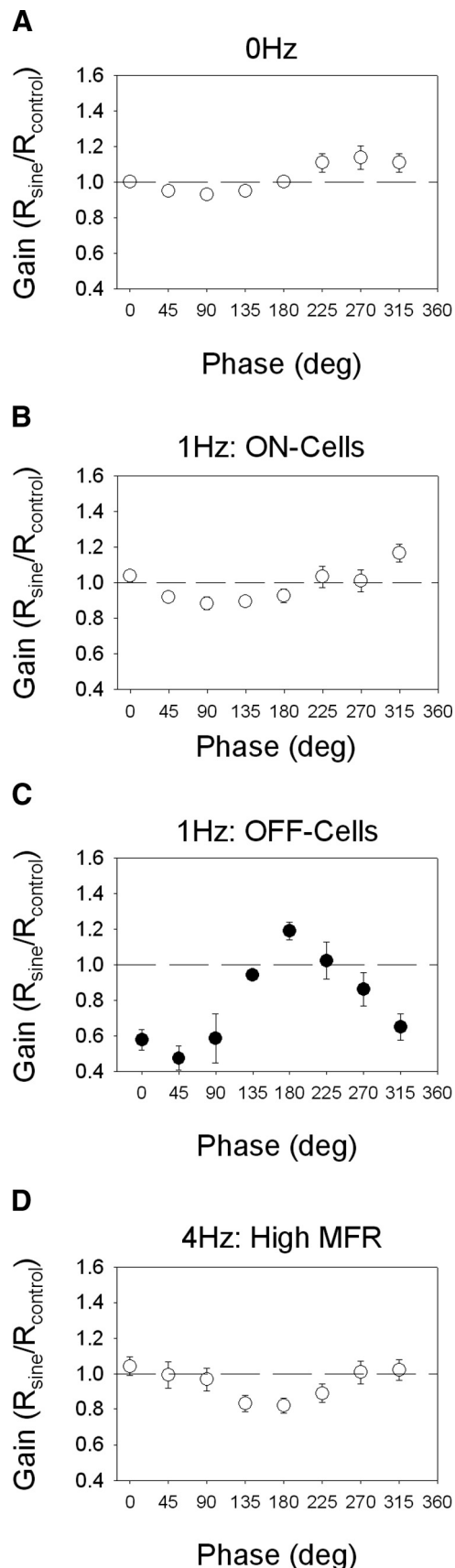
random order. This gives the steady-state gain at each phase of the probed-sinewave experiment, and the results are shown in Fig. 5A ($n = 5$ ON cells). Ganglion cell sensitivity was reduced for an increase in standing contrast (45–135°) and enhanced for a decrease in standing contrast (225–315°), consistent with luminance adaptation. Thus the gain changes reported here do not require dynamic changes in luminance.

Luminance adaptation can span multiple timescales ranging from tens of milliseconds to several minutes (Walraven et al. 1990). We sought to determine the time scale of the luminance adaptation observed at low contrasts. However, estimating the speed of this mechanism in rats is complicated by the fact that the flash response can outlast the modulation period at high temporal frequencies. In addition, response clipping and phase locking are more pronounced at fast modulation rates (>1 Hz). For stimulation at 1 Hz, ON and OFF cells both showed measurable luminance adaptation to 15% background modulation, as evidenced by the elevation in response gain above the baseline level for some probe phases (Fig. 5B). For a subset of ON cells that had a high maintained discharge rate (>30 imp/s), it was possible to probe faster speeds because the modulated response did not show distortion. For stimulation at 4 Hz, the sensitivity profile varied sinusoidally with phase as well (Fig. 5C). Together, the results show that luminance adaptation acts on a timescale of <500 ms in OFF cells and <125 ms in ON cells. There is also suggestion of a phase-independent reduction of gain in the 1- and 4-Hz data. The shift was small (phase-averaged gain: 0.95 ± 0.09 for 4 Hz), but it could indicate the presence of temporal contrast adaptation at higher modulation frequencies.

Weber's law behavior at low stimulus contrast

Retinal sensitivity has been shown to decrease in inverse proportion to light level over the photopic range, allowing ganglion cells to signal contrast instead of absolute luminance (Shapley and Enroth-Cugell 1984; Troy and Enroth-Cugell 1993). This behavior is referred to as Weber's law and can be described by the equation

$$G_0 = \frac{\alpha}{L_0}$$

where G_0 is the baseline gain, L_0 is background light level, and α is a constant of proportionality. The relationship implies that increasing light level by a factor of 10, for example, would cause retinal sensitivity to decrease by a factor of 10. What has not been considered are the gain changes predicted by Weber's law for stimuli of low-to-moderate contrast and the degree to which retinal ganglion cells achieve them.

Consider a ganglion cell viewing a sinusoidally modulated spot on a steady background. If the luminance of the spot at a

FIG. 5. The time scale of adaptation was estimated by varying the temporal frequency of the probed-sinewave stimulus. A: response gain of ON cells ($n = 5$) for 0-Hz stimulation (i.e., constant luminance). Spot responses were acquired at each phase after several seconds of adaptation to the luminance level of a 25% contrast sinewave at the specified phases. B–C: response gain of ON ($n = 13$) and OFF ($n = 3$) cells for 1-Hz stimulation at 15% contrast. D: response gain of a subset of ON cells ($n = 9$) with a high maintained discharge rate (>30 imp/s) for 4-Hz stimulation at 15% contrast.
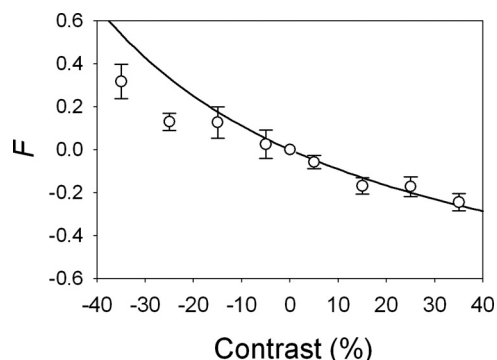
FIG. 6.   The fractional change in ganglion cell sensitivity, F, for the 0.25 Hz probed-sinewave stimulus (circles) as a function of background contrast. The solid line shows the change in response gain predicted by Weber's law. Error bars give SE.

given point in time is L_1, the contrast of the spot relative to the background is

$$C = \frac{L_1 - L_0}{L_0}$$

Assuming that luminance adaptation is as fast as the luminance modulation, the fractional change in response gain F predicted by Weber's law is

$$F = \frac{G_1 - G_0}{G_0} = \frac{L_0}{L_1} - 1 = \frac{-C}{1 + C}$$

where G_1 is the response gain at a luminance of L_1. This value is the same for any value of α and for any background light level. Interestingly, the predicted gain changes are not symmetric about the baseline level (Fig. 6, line). For example, at 25% contrast, the gain gets 33% larger when the spot darkens but only 20% smaller when the spot brightens.

To evaluate whether ganglion cell adaptation to small luminance fluctuations maintains Weber's law behavior, the maximum and minimum change in response gain measured for the 0.25-Hz modulation at each contrast was compared with Weber's law (Fig. 6). Only ON cell data were considered because of the marked clipping of OFF cell responses even at low contrasts. The data follow theoretical predictions for positive contrasts and deviate slightly at negative contrast, which may be caused in part by response clipping. Hence, although low contrast stimuli are usually thought to produce linear responses, they do activate luminance gain controls, and the

amount of activation is consistent with Weber's law. At higher contrasts, the nonlinear response distortions associated with luminance adaptation become apparent, and gain changes may be asymmetric for stimulus increments and decrements.

Adaptation to spatial contrast

Although the probed-sinewave data show that small fluctuations in light intensity over the receptive field of ganglion cells can cause luminance adaptation, the data provide little indication for an involvement of contrast adaptation because probe sensitivity profiles were not shifted much below baseline. This means that either the rat retina does not have contrast adaptive mechanisms or that these mechanisms require spatial intensity gradients within the receptive field to activate. We tested for spatial contrast adaptation by hemisecting the spot vertically and modulating the luminance of each half of the spot in counterphase. Because ganglion cells integrate signals linearly over their receptive field center, excitatory input from the bright and dark halves cancel, and the hemisected spot produces no modulated response to the background. At various phases of the counterphase modulation, both halves of the spot were flashed together to probe for response adaptation.

Figure 7 shows the results for a group of ON ($n = 13$) and OFF ($n = 8$) cells using 0.25-Hz counterphase modulation at 25% contrast. Interestingly, response gain was suppressed by ~40% for both cell types at all flash phases. This could reflect an asymmetry in luminance adaptation, except that the suppression for ON cells was larger than that for the probed-sinewave stimulus (Fig. 2), even though the adaptation field was split in two and adaptive signals from each field should partially cancel. Instead, the phase-independent reduction of response gain is evidence for a gain control mechanism within the receptive field center that integrates luminance signals nonlinearly and is thereby sensitive to spatial contrast. This is consistent with a study of contrast adaptation that used spatial contrast noise as the stimulus (Brown and Masland 2001). The striking dissimilarity of the sinewave and split-sinewave data implies that the spatial contrast and luminance gain controls are separate mechanisms mediated by distinct retinal circuits.

DISCUSSION

Using a probed-sinewave stimulus, retinal ganglion cells in rats were shown to change their response gain for local luminance fluctuations as low as 15% contrast. Gain increases and
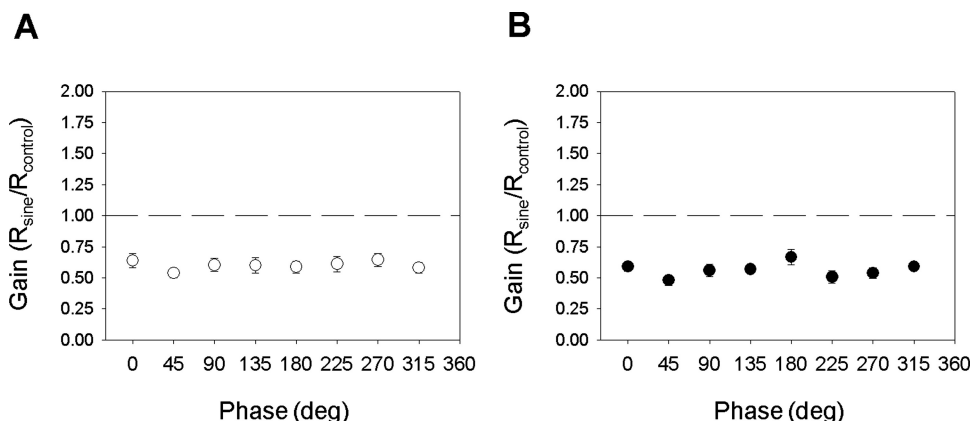

FIG. 7.   Average response gain for ON cells (A, $n = 13$) and OFF cells (B, $n = 8$) as a function of modulation phase for the probed split-sine paradigm. For this paradigm, the spot was hemisected and the two halves were counterphase modulated sinusoidally in time. For a well-centered spot, the resting discharge of the cell is unaltered by the background modulation. Bars above the figure indicate the bright and dark phases for each hemifield. Probe response amplitude was measured before/after ($R_{control}$) and during (R_{sine}) counterphase modulation of 0.25 Hz and 25% contrast. Error bars give SE.

decreases were both measured when local luminance, respectively, varied below and above the ambient level, indicating that the gain changes were mediated by a luminance adaptive process and not the result of contrast adaptation or response compression. The process was active at low contrasts in both ON and OFF ganglion cells but differed in temporal phase between the two cell types. The phase difference places the site of adaptation downstream of the photoreceptors and in the retinal network. The ganglion cell spike generator can be excluded as a possible site for the ON pathway as the gain changes were independent of firing rate. A similar conclusion could not be made for the OFF pathway because of response clipping. These findings are not based on any particular model of luminance gain control and are thereby free of modeling assumptions.

Relationship to physiological studies of retinal adaptation

The mammalian retina contains several mechanisms of luminance gain control, including photopigment bleaching, rod-cone signaling, receptor adaptation, and network adaptation. Although the gain changes described here could perhaps reflect a novel high-sensitivity mechanism, they are predicted by Weber's law and more likely the product of network adaptation. The existence of such a mechanism has been shown (Dunn et al. 2007; Green and Powers 1982; Ichinose and Lukasiewicz 2007), but its threshold of activation has not been established because the state of retinal adaptation is varied in most studies by changing light level over orders of magnitude (Cleland and Enroth-Cugell 1968, 1970; Enroth-Cugell and Shapley 1973a,b; Enroth-Cugell et al. 1975). Within a given state, ganglion cell responses are considered to be approximately linear for subsaturating stimuli. We show here that network mechanisms of adaptation are engaged even by relatively small luminance fluctuations and that they are more than sensitive enough to modulate ganglion cell responses over the 2- to 10-fold range of local luminance levels typically found in a natural scene (Frazor and Geisler 2006). Their effects on response gain are apparent for fluctuations of ≥1 Hz for OFF cells and 4 Hz for ON cells. This suggests that the adaptive mechanism examined here acts rapidly (<125 ms) and are thus fast enough to adjust retinal gain between saccadic eye movements (~200 ms) (Saito and Fukada 1986). In short, our results experimentally validate the hypothesis that luminance gain controls in the retina operate not just as we move from one lighting environment to the next but also as we scan a visual scene under constant ambient illumination (Dunn and Rieke 2006).

The findings raise the possibility that luminance gain controls could have been active in some studies of contrast adaptation. One argument advanced against this possibility is that such studies generally use white noise stimuli of constant mean intensity and no discernible change in cone or horizontal cell response gain was measured based on noise analysis (Baccus and Meister 2002). Nevertheless, it was recently shown that intensity changes that are insufficient to adapt photoreceptors can still induce luminance adaptation downstream in the retinal network (Dunn et al. 2007). It might also be argued that luminance adaptation acts too slowly to factor in contrast adaptation studies because a white noise stimulus varies rapidly in time. However, we show that network mechanisms of luminance gain control are fast. At the typical frame rates of white noise presentation, it can be expected that the mean input over the integration time of ganglion cells would fluctuate in time because the stimulus contains energy at all temporal frequencies. Given the remarkable sensitivity of the luminance gain control described here, if gain changes induced by luminance increments and decrements are asymmetric in strength (Fig. 6) or dynamics (Snippe et al. 2004), the mechanism could produce an adaptive effect like that reported of stimulus contrast, particularly in cases where the receptive field center is uniformly illuminated. How substantial the putative contributions might be is uncertain.

Our results provide weak evidence for temporal contrast adaptation within the receptive field of ganglion cells. Such a mechanism would cause phase-independent response suppression, but a downward shift in response gain was seen only for 4-Hz modulation and it was fairly weak (~5%). This finding is seemingly at odds with several contrast adaptation studies (Chander and Chichilnisky 2001; Baccus and Meister 2002; Beaudoin et al. 2007; Kim and Rieke 2001). In some cases, the disparity might be explained by differences in stimulus configuration, because many of these studies illuminated the entire retina, whereas the input was confined here to the receptive field center. Indeed, our results provide strong support for spatial contrast adaptation (Fig. 7) and are consistent with the few studies that introduced spatial contrast into the receptive field (Brown and Masland 2001; Mante et al. 2005). We did not study the lateral extent of this spatial contrast adaptation mechanism, so it may correspond to the contrast gain control that is known to operate within and beyond the receptive field of ganglion cells in other mammals (Enroth-Cugell and Jakiela 1980; Passaglia et al. 2001, 2009; Shapley and Victor 1978). Consistent with this idea, the bipartite spot experiment implies the involvement of nonlinear subunits like those that comprise the retinal contrast gain control. Also, spatial contrast adaptation acts on both X and Y cells, as does the contrast gain control.

Relationship to psychophysical studies of luminance adaptation

The probed-sinewave stimulus has been used extensively in psychophysics to study the dynamics of light adaptation. For modulated backgrounds of high contrast (>50%), human visual sensitivity to a spot of light flashed at different phases was shown to vary above and below the unmodulated baseline at photopic light levels (Hood et al. 1997; Wolfson and Graham 2006), indicative of luminance adaptation. In addition, for modulation frequencies >0.4 Hz, the sensitivity profiles are often offset to an overall higher threshold level (Hood et al. 1997; Snippe et al. 2000). This phase-independent reduction of sensitivity has been attributed to contrast adaptation (Snippe et al. 2000, 2004). Only two studies have used modulated backgrounds of <50% contrast, and their findings also show sinusoidal oscillations of threshold with an overall threshold elevation at higher frequencies (Shickman 1970; Wolfson and Graham 2001).

Our physiological results confirm the psychophysical evidence that low-contrast stimuli can activate a local mechanism of luminance adaptation in the mammalian retina. However, our results do not provide support for a retinal origin to the perceptual effects of temporal contrast adaptation because ganglion cell sensitivity profiles showed a much smaller dc-offset from baseline compared with the psychophysical results. One reason for this may be a difference in stimulus configuration. In most psychophysical experiments, the modulated background is several times larger than the spot used to

measure visual sensitivity, whereas in our experiments, the background modulation was constrained to the same spot that probed the receptive field center sensitivity of ganglion cells. This means that retinal mechanisms that operate outside the classical receptive field center and surround, such as contrast gain control (Enroth-Cugell and Jakiela 1980; Passaglia et al. 2001, 2009; Shapley and Victor 1978), could have factored prominently in the psychophysical data but not ours. Hence, probed-sinewave experiments using low contrast background modulations that are co-spatial with the probe are needed to compare with our results more directly.

REFERENCES

Baccus SA, Meister M. Fast and slow contrast adaptation in retinal circuitry. *Neuron* 36: 909–919, 2002.

Benardete EA, Kaplan E, Knight BW. Contrast gain control in the primate retina: P cells are not X-like, some M cells are. *Vis Neurosci* 8: 483–486, 1992.

Brainard DH. The psychophysics toolbox. *Spat Vis* 10: 433–436, 1997.

Brown SP, Masland RH. Spatial scale and cellular substrate of contrast adaptation by retinal ganglion cells. *Nat Neurosci* 4: 44–51, 2001.

Chander D, Chichilnisky EJ. Adaptation to temporal contrast in primate and salamander retina. *J Neurosci* 21: 9904–9916, 2001.

Chichilnisky EJ, Kalmar RS. Functional asymmetries in ON and OFF ganglion cells of the primate retina. *J Neurosci* 22: 2737–2747, 2002.

Cicerone CM, Green DG. Light adaptation within the receptive centre of rat retinal ganglion cells. *J Physiol* 301: 517–534, 1980.

Cleland BG, Enroth-Cugell C. Quantitative aspects of sensitivity and summation in the cat retina. *J Physiol* 198: 17–38, 1968.

Cleland BG, Enroth-Cugell C. Quantitative aspects of gain and latency in the cat retina. *J Physiol* 206: 73–91, 1970.

Cleland BG, Levick WR. Brisk and sluggish concentrically organized ganglion cells in the cat's retina. *J Physiol* 240: 421–456, 1974.

Dowling JE. The site of visual adaptation. *Science* 155: 273–279, 1967.

Dunn FA, Lankheet MJ, Rieke F. Light adaptation in cone vision involves switching between receptor and post-receptor sites. *Nature* 449: 603–607, 2007.

Dunn FA, Rieke R. The impact of photoreceptor noise on retinal gain controls. *Curr Opin Neurobiol* 16: 363–370, 2006.

Enroth-Cugell C, Jakiela HG. Suppression of cat retinal ganglion cell responses by moving patterns. *J Physiol* 302: 49–72, 1980.

Enroth-Cugell C, Lennie P, Shapley RM. Surround contribution in light adaptation in cat retinal ganglion cells. *J Physiol* 247: 579–588, 1975.

Enroth-Cugell C, Shapley RM. Adaptation and dynamics of cat retinal ganglion cells. *J Physiol* 233: 271–309, 1973a.

Enroth-Cugell C, Shapley RM. Flux, not retinal illumination, is what cat retinal ganglion cells really care about. *J Physiol* 233: 311–326, 1973b.

Frazer RA, Geisler WS. Local luminance and contrast in natural images. *Vision Res* 46: 1585–1598, 2006.

Freeman DK, Heine WF, Passaglia CL. The maintained discharge of rat retinal ganglion cells. *Vis Neurosci* 25: 535–548, 2008.

Green DG, Powers MK. The mechanisms of light adaptation in rat retina. *Vision Res* 22: 209–216, 1982.

Hochstein S, Shapley RM. Linear and nonlinear spatial subunits in Y cat retinal ganglion cells. *J Physiol* 262: 265–284, 1976.

Hood DC, Graham N, von Wiegand TE, Chase VM. Probed-sinewave paradigm: a test of models of light-adaptation dynamics. *Vision Res* 37: 1177–1191, 1997.

Ichinose T, Lukasiewicz PD. Ambient light regulates sodium channel activity to dynamically control retinal signaling. *J Neurosci* 27: 4756–4764, 2007.

Kim KJ, Rieke F. Temporal contrast adaptation in the input and output signals of salamander retinal ganglion cells. *J Neurosci* 21: 287–299, 2001.

Lankheet MJ, Van Wezel RJ, Prickaerts JH, van de Grind WA. The dynamics of light adaptation in cat horizontal cell responses. *Vision Res* 33: 1153–1171, 1993.

Lee BB, Dacey DM, Smith VC, Pokorny J. Dynamics of sensitivity regulation in primate outer retina: the horizontal cell network. *J Vis* 3: 513–526, 2003.

Levick WR. Another tungsten microelectrode. *Med Biol Eng* 10: 510–515, 1972.

Manookin MB, Demb JB. Presynaptic mechanism for slow contrast adaptation in mammalian retinal ganglion cells. *Neuron* 50: 453–464, 2006.

Mante V, Frazor RA, Bonin V, Geisler WS, Carandini M. Independence of luminance and contrast in natural scenes and in the early visual system. *Nat Neurosci* 8: 1690–1697, 2005.

O'Brien BJ, Isayama T, Richardson R, Berson DM. Intrinsic physiological properties of cat retinal ganglion cells. *J Physiol* 538: 787–802, 2002.

Passaglia CL, Enroth-Cugell C, Troy JB. Effects of remote stimulation on the mean firing rate of cat retinal ganglion cells. *J Neurosci* 21: 5794–5803, 2001.

Passaglia CL, Freeman DF, Troy JB. Effects of remote stimulation on the modulated activity of cat retinal ganglion cells. *J Neurosci* 29: 2467–2476, 2009.

Perlman I, Norman RA. Light adaptation and sensitivity controlling mechanisms in vertebrate photoreceptors. *Prog Retin Eye Res* 17: 523–563, 1998.

Pugh EN, Nikonov S, Lamb TD. Molecular mechanisms of vertebrate photoreceptor light adaptation. *Curr Opin Neurobiol* 9: 410–418, 1999.

Pupura K, Tranchina D, Kaplan E, Shapley RM. Light adaptation in the primate retina: analysis of changes in gain and dynamics of monkey retinal ganglion cells. *Vis Neurosci* 4: 75–93, 1990.

Rushton WAH, Westheimer G. The effect upon the rod threshold of bleaching neighboring rods. *J Physiol* 164: 318–329, 1962.

Saito H, Fukada Y. Gain control mechanisms in X- and Y-type retinal ganglion cells of the cat. *Vis Res* 26: 391–408, 1986.

Schneeweis DM, Schnapf JL. The photovoltage of macaque cone photoreceptors: adaptation, noise, and kinetics. *J Neurosci* 19: 1203–1216, 1999.

Shapley RM, Enroth-Cugell C. Visual adaptation and retinal gain controls. *Prog Retin Eye Res* 3: 263–346, 1984.

Shapley RM, Victor JD. The effect of contrast on the transfer properties of cat retinal ganglion cells. *J Physiol* 285: 275–298, 1978.

Shickman GM. Visual masking by low-frequency modulated light. *J Opt Soc Am* 60: 107–117, 1970.

Smirnakis SM, Berry MJ, Warland DK, Bialek W, Meister M. Adaptation of retinal processing to image contrast and spatial scale. *Nature* 386: 69–73, 1997.

Smith VC, Pokorny J, Lee BB, Dacey DM. Sequential processing in vision: the interaction of sensitivity regulation and temporal dynamics. *Vision Res* 48: 2649–2656, 2008.

Snippe HP, Poot L, van Hateren JH. A temporal model for early vision that explains detection thresholds for light pulses on flickering backgrounds. *Vis Neurosci* 17: 449–462, 2000.

Snippe HP, Poot L, van Hateren JH. Asymmetric dynamics of adaptation after onset and offset of flicker. *J Vis* 4: 1–12, 2004.

Troy JB, Enroth-Cugell C. X and Y ganglion cells inform the cat's brain about contrast in the retinal image. *Exp Brain Res* 93: 383–390, 1993.

Troy JB, Shou T. The receptive fields of cat retinal ganglion cells in physiological and pathological states: where we are after a half century of research. *Prog Retin Eye Res* 21: 263–302, 2002.

Victor JD. The dynamics of the cat retinal X cell centre. *J Physiol* 386: 219–246, 1987.

Walraven J, Enroth-Cugell C, Hood DC, Macleod DI, Schnapf JL. The control of visual sensitivity: receptoral and postreceptoral processes. In: *Visual Perception: The Neurophysiological Foundations*, edited by Spillmann L, Werner JS. San Diego: Academic Press, 1990, p. 53–101.

Wark B, Fairhall A, Rieke F. Timescales of inference in visual adaptation. *Neuron* 61: 750–761, 2009.

Wolfson SS, Graham N. Comparing increment and decrement probes in the probed-sinewave paradigm. *Vis Res* 41: 1119–1131, 2001.

Wolfson SS, Graham N. Forty-four years of studying light adaptation using the probed-sinewave paradigm. *J Vis* 6: 1026–1046, 2006.

Wu A, Burns SA, Elsner AE, Eskew RT, He J. Rapid sensitivity changes on flickering backgrounds: tests of models of light adaptation. *J Opt Soc Am A* 14: 2367–2378, 1997.

Yeh T, Lee BB, Kremers J. The time course of adaptation in macaque retinal ganglion cells. *Vis Res* 36: 913–931, 1996.