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AMPLITUDE AND PHASE OF RESPONSES OF MACAQUE RETINAL GANGLION CELLS TO FLICKERING STIMULI

BY B. B. LEE*, P. R. MARTIN* AND A. VALBERG†

*From the *Department of Neurobiology, Max Planck Institute for Biophysical Chemistry, D-3400 Göttingen, FRG and †Section of Biophysics, Department of Physics, University of Oslo, Oslo, Norway*

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SUMMARY

1. We have measured responses of macaque retinal ganglion cells to a uniform flickering field, with variation in luminance, chromaticity or both (heterochromatic flicker).

2. With heterochromatic flicker, as the luminance ratio of the flicker components was varied, phasic ganglion cell activity went through a minimum and an abrupt phase change close to equal luminance. Tonic ganglion cell responses underwent a gradual phase change without any minimum close to equal luminance. For red on-centre cells, when wavelengths above 570 nm were altered with white, a progressive phase advance occurred as luminance ratio (L_λ/L_W) was increased. With wavelengths below 570 nm a progressive phase lag occurred. For green on-centre cells, the opposite pattern was found. For all tonic cells, the higher the temporal frequency, the more rapidly did such phase changes occur. A simple model incorporating a centre–surround delay of 3–8 ms could quantitatively account for these changes.

3. With luminance flicker of different dominant wavelengths, amplitudes and phase of responses of phasic ganglion cells were independent of wavelength at all frequencies. The amplitude and phase of the responses of tonic ganglion cells was very dependent on wavelength, as well as on flicker frequency. Their characteristics hardly ever resembled results from phasic cells.

4. For achromatic flicker, response phase of tonic cells at or above 10 Hz was variable, probably due to the centre–surround delay. Such variability was not seen among phasic cells.

5. An interesting implication of these results is that the ability of tonic ganglion cells to unambiguously signal rapid chromatic or spatial change is limited.

INTRODUCTION

In the previous paper (Lee, Martin & Valberg, 1989*a*), we have described the sensitivity of different ganglion cell types of the macaque to luminance and chromatic flicker, and provided evidence that phasic (M-pathway) and tonic (P-pathway) cells respectively are the physiological substrate for detection of these two sorts of flicker by the human observer. It is often assumed such stimuli are

detected through different channels (Kelly & van Norren, 1977), for which these cell types would thus form the physiological basis.

Early studies of retinal ganglion cells had revealed that tonic cells were predominantly colour-opponent, whereas phasic cells were non-opponent with a spectral sensitivity (measured by increment thresholds) very similar to the human photopic luminosity, or V_λ function (de Monasterio & Gouras, 1975; de Monasterio, 1978). A correspondence between these cell types and psychological channels did not find acceptance, however. One reason was the failure to find evidence for phasic cells near the fovea. This assumption was based on the difficulty in recording from such cells there, and on the weak short-latency antidromic potential recorded in the foveal region in comparison with that recorded more peripherally (Gouras, 1969).

When tonic cells were stimulated with luminance flicker of different wavelengths, Gouras & Zrenner (1979) noted that tonic cells became more broad band in their spectral responsiveness, and attributed this effect to a centre–surround latency difference. The latency difference they postulated was about 15–20 ms, which is large in comparison with that suggested for cat ganglion cells (Enroth-Cugell, Robson, Schweizer-Tong & Watson, 1983). It was proposed (e.g. Zrenner, 1983) that such cells could do 'double duty' and serve as both chromatic and achromatic channels depending on temporal frequency.

More recent evidence has suggested the phasic, M-pathway could indeed play an important role in visual performance. Firstly, Perry & Silveira (1988) localized putative phasic, M-pathway cells histologically, and found them numerous around the foveal pit but buried deep in the layers of ganglion cells there. This could account for both the weak antidromic potential and the difficulty in finding such cells with an electrode. Secondly, phasic cells underlie certain psychophysical tasks. Heterochromatic flicker is the alternation of lights of differing chromaticity. A human subject who adjusts the relative intensity of two such flickering lights is able to minimize or abolish the sensation of flicker. It was mostly from such data that the human photopic luminosity function was established. We have shown elsewhere that phasic ganglion cells possess all the properties necessary for a physiological substrate for this task (Lee, Martin & Valberg, 1988).

In the previous paper (Lee *et al.* 1989*a*), we presented further evidence that phasic cells play a role in visual performance. Here, we examine ganglion cell responses in more detail, especially in relation to a latency difference between centre and surround of tonic cells. These cells, when stimulated with an alternation of two different wavelengths at, say, 10 Hz, showed a gradual change in response phase as the relative intensity of the lights was altered. We show that this can be attributed to a delay of the surround relative to the centre of a few (3–8) milliseconds. We also demonstrate the effect of this delay on the responses of tonic cells to other stimuli.

METHODS

Recordings from ganglion cells were obtained from the retinae of the anaesthetized macaque monkey. A detailed description of anaesthetic regime, preparation and visual stimulation is given in the accompanying paper (Lee *et al.* 1989*a*).

To make clear the relationship between the flickering stimulus and cell response, we have sketched the composition of heterochromatic flicker in Fig. 1. The stimulus is made up of

achromatic (Wh) and chromatic (λ) components, which are modulated in counterphase. The two components are shown added together in panel *A*, and separately in panel *B*. In *B*, a response waveform is also shown, with a phase lag relative to the white light of ϕ radians. We have adopted the convention that a phase lag is represented by a *positive* value of ϕ. Note that this differs from that adopted by Enroth-Cugell *et al.* (1983). For a stimulus with a temporal waveform sin ωt, at an angular velocity of ω the response (r) of a cell is given by

$$r(t) = 2a\ (\sin\ (\omega t - \phi) + 1),$$

where a is response amplitude. The angular velocity is related to frequency, f, by $\omega = 2\pi f$. From Fourier analyses of cell responses we obtain phase relative to the maximum firing rate of the cell, and this measure, ϕ', is given by $\phi' = \phi + \pi/2$.

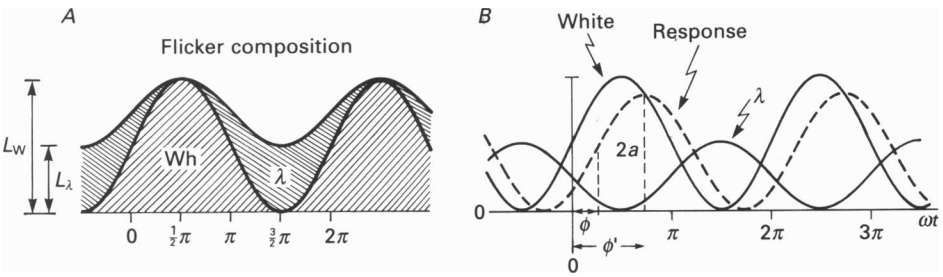

Fig. 1. Heterochromatic flicker is the alternation of two lights of differing chromaticity. In *A*, L_W and L_λ indicate luminance, the components being added together. They have been separated in *B*. Response waveform (dashed line) lags the white component by ϕ rad.

RESULTS

We classified cells as phasic or tonic on the basis of their response to chromatic spots, and from their achromatic contrast sensitivities, as described in the accompanying paper (Lee *et al.* 1989*a*). The results discussed here were obtained from a sample of eighty-five tonic and fifty-one phasic ganglion cells.

Responses to heterochromatic flicker

Typical responses of a phasic and a tonic cell to heterochromatic flicker are shown in Fig. 2. Responses to two cycles of flicker are shown in each histogram; the flicker composition is sketched on the left. For each cell, a wavelength of 506 nm was alternated with a white reference at 10 Hz. The numbers next to each histogram are the 506 nm/white luminance ratios (L_λ/L_W). For the phasic cell, as a luminance ratio of one is approached, the cell becomes less responsive, until close to a ratio of one (corresponding to flicker minimization by human subjects) only a small, frequency-doubled response remained. Such frequency-doubled responses are discussed elsewhere (Lee, Martin & Valberg, 1989*b*). At a ratio greater than one, the cell's response grows more vigorous, and an abrupt change in response phase has occurred.

The green on-centre cell shows qualitatively different behaviour. As luminance

ratio changes, there is no minimum in the response but a gradual phase shift. These differences in the behaviour of the two cells are illustrated in Fig. 2*B*, where response amplitude and phase are plotted as a function of luminance ratio. The sharp minimum in the phasic cell's response and the abrupt phase shift contrast with the vigorous response and slow phase change of the green on-centre cell. A more extensive description of such results is given by Lee *et al.* (1988).

With heterochromatic flicker at temporal frequencies of 4 Hz or above, gradual phase changes were seen in all tonic cells as luminance ratio was altered. These changes were dependent on the wavelength used and temporal frequency, as well as on stimulus field size. It is convenient to visualize these changes in polar co-ordinates, where the amplitude and phase of the response is represented as a vector, and such graphs can be seen in Fig. 3. A response phase of $\pi/2$ implies that a cell's firing is in phase with the intensity modulation of the white component of the heterochromatic flicker (Fig. 1).

In the example of Fig. 3*A*, a green on-centre cell was stimulated with heterochromatic flicker at 10 Hz, with different monochromatic wavelengths being alternated with a white reference light. Responses were recorded at different relative intensities of the flickering lights, starting with the white component about 10 times more luminous than the chromatic component, and ending with the chromatic component about 6 times more luminous than the white component. These two end-points are indicated by W and λ respectively. The luminance ratio closest to one is indicated by the filled symbol in each curve, and the arrows indicate increasing luminance ratio. The curves do not start at the same point for even at low luminance ratios the chromatic flicker component influenced response amplitude and phase.

It is immediately obvious from Fig. 3*A* that with wavelengths below 570 nm a clockwise rotation of the response vector takes place as luminance ratio is increased (corresponding to a progressive phase advance), while above 570 nm, an anti-clockwise rotation occurs (corresponding to a progressive phase lag). At 570 nm, response phase is almost independent of luminance ratio, and the curve passes close to the origin, indicating a minimum at equal luminance. White and 570 nm lie close to a tritanopic confusion line, so that at equal luminance a lack of response of a cell which lacks S-cone input is to be expected. Examples of original response histograms may be found in Lee *et al.* (1988).

Figure 3*B* and *C* illustrates a similar result from another green on-centre cell, in this case stimulated at different temporal frequencies with 506 nm and 622 nm alternated with the white reference. As with the cell of Fig. 3*A*, for 506 nm a progressive phase advance takes place as luminance ratio increases, while at 622 nm a progressive phase lag occurs. The magnitude of these phase changes is related to temporal frequency, so that at 1 Hz response phase is almost constant, while at 40 Hz the biggest phase shift takes place.

Figure 4 illustrates the behaviour of a typical red on-centre cell. We tested five different wavelengths at 1 and 10 Hz. At 1 Hz, little phase change occurs. At 10 Hz, the opposite pattern to that displayed by the green on-centre cell appears, with a progressive phase *advance* with wavelengths above 570 nm, and a phase *lag* below 570 nm. Again, at 570 nm response phase is almost independent of luminance ratio.

The results shown in Figs 3 and 4 were typical for all the green on-centre (fifteen

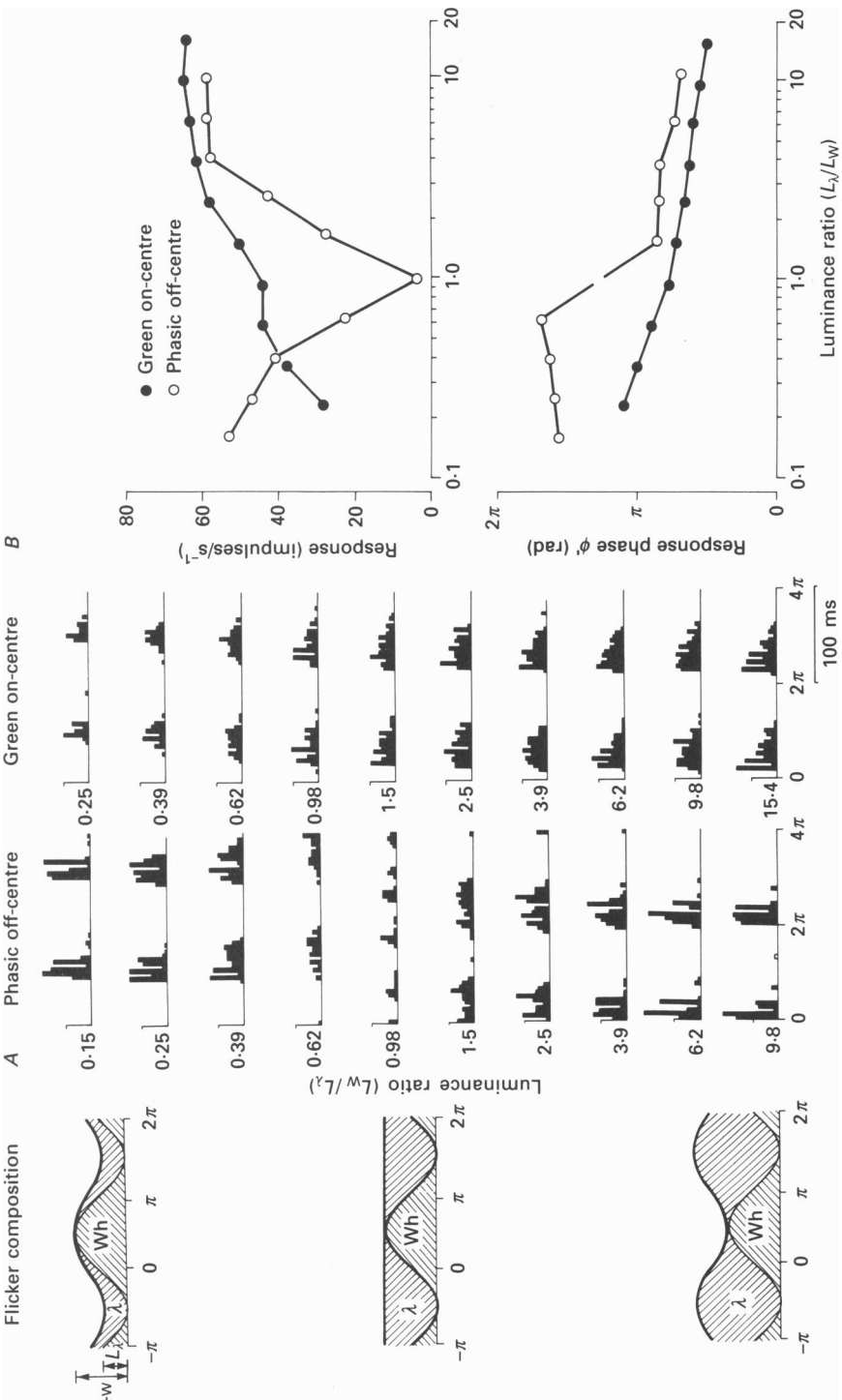

Fig. 2. *A*, response histograms of a phasic off-centre cell and a green on-centre cell to heterochromatic flicker (10 Hz, 4 deg field, mean luminance 50 cd m^{-2}), the composition of which is illustrated on the left. The modulation of luminance (L) of each light is shown; the upper line shows the total luminance of the mixture. White was alternated with 506 nm. Each histogram represents response to two cycles of flicker, beginning at the mean intensity of the white component. Luminance ratio (L_λ/L_w) is indicated for each histogram. The modulated response of the phasic cell goes through a minimum close to a luminance ratio of one, at which point an abrupt change in phase is seen. For the green on-centre cell there is a gradual change in response phase without any indication of a minimum. Six seconds of activity were averaged for each histogram. Bin width was 4 ms. Calibration bars indicate a firing rate of 40 impulses s^{-1}. In *B*, amplitude of the fundamental response component and response phase are plotted against luminance ratio for the two cells.

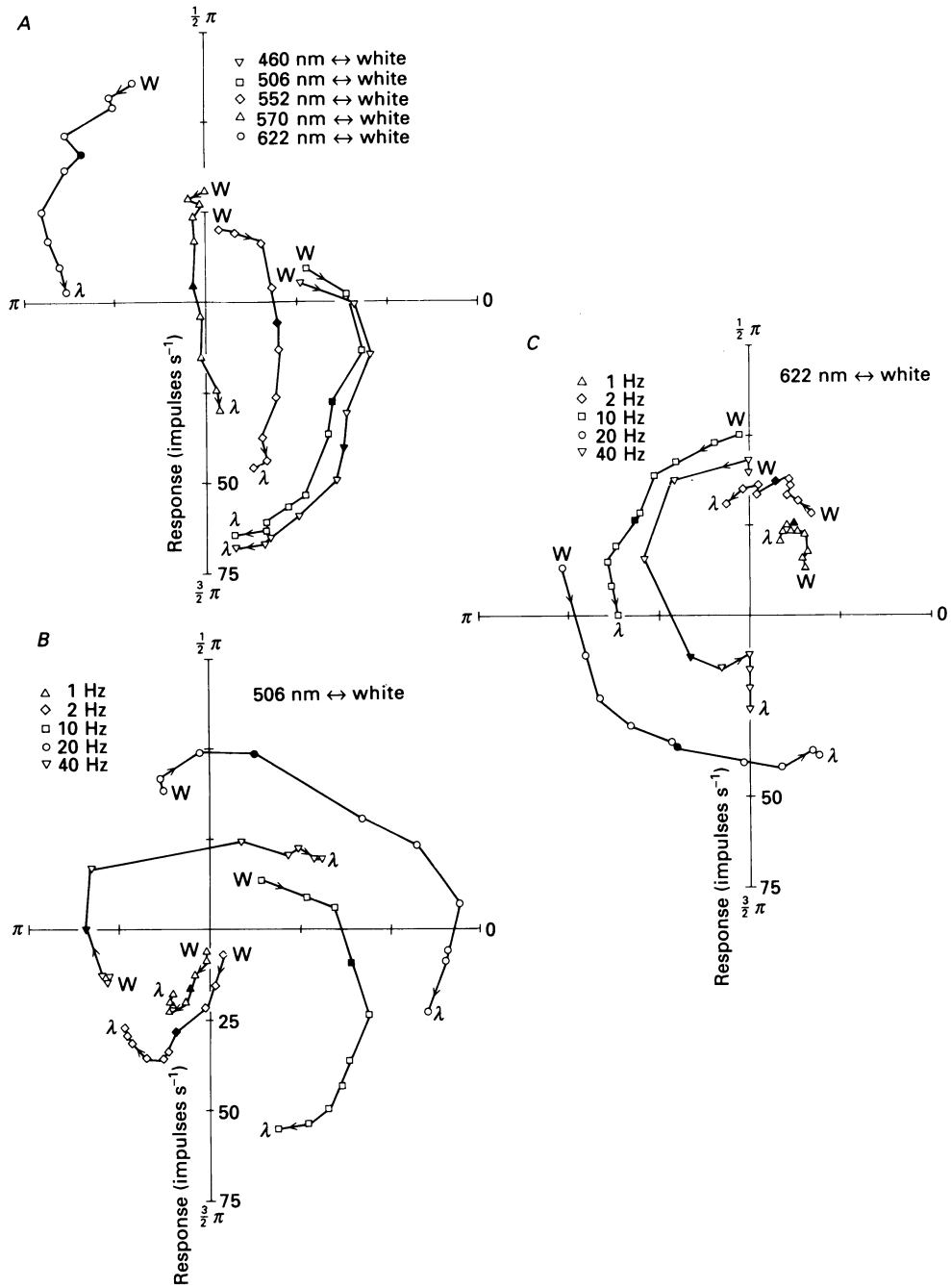

Fig. 3. For legend see opposite

cells) and red on-centre (thirteen cells) investigated with this protocol. Red and green off-centre cells tested (thirteen cells) also showed changes in response phase similar to those in Figs 3 or 4.

This dependence of response phase on luminance ratio, and the interaction with temporal frequency, suggests a delay between opponent cone mechanisms. It is intuitively obvious that if two sinusoidal signals, with a phase lag between them, are added, then the sum will also be a sinusoid the phase of which will be dependent on their relative amplitudes. It is not directly obvious what delay is required to account for the change observed, and why wavelengths above and below 570 nm cause phase shifts in opposite directions. We therefore analysed cell response in a quantitative manner.

Response phase to heterochromatic flicker

We have employed a linear model, although the range of intensities used was probably sufficient for non-linearites in the cone signals to occur (e.g. Lee, Valberg, Tigwell & Tryti, 1987). In the following, we have assumed that a straightforward *transport* delay is present, so that the phase delay is proportionally related to frequency, and to angular velocity, ω.

The response to a sinusoidal alternation of two lights will be a function of the activation of the opponent cone mechanisms. Taking as an example a red on-centre cell, a response vector, N, can be decomposed into N_{real} and N_{imag}. Using an analysis similar to that of Enroth-Cugell *et al.* (1983), we obtain:

$$N_{\text{real}} = A_{\text{r}} R\cos \omega \text{d} - A_{\text{g}} G\cos \omega(d + d_\delta), \tag{1}$$

and

$$N_{\text{imag}} = A_{\text{r}} R\sin \omega \text{d} - A_{\text{g}} G\sin \omega(d + d_\delta), \tag{2}$$

where A_{r} and A_{g} are the weightings of the M– and L–cone mechanisms, ω is angular velocity of the flicker ($2\pi f$), d is a time delay common to centre and surround, due, for example, to latency, and d_δ is an additional delay present only in the surround mechanism. R and G represent the modulation generated in each cone mechanism by a given flicker condition, calculated from

$$R = L_{\text{W}} - L_\lambda R_\lambda, \tag{3}$$

and

$$G = L_{\text{W}} - L_\lambda G_\lambda, \tag{4}$$

Fig. 3. Polar diagrams of the response and response phase of two green on-centre cells to heterochromatic flicker. Response amplitude is represented radially, and response phase as the vector angle. A phase of 0 deg is referenced to the sine of the white flicker component (see Fig. 1). Luminance ratio was changed in 0·2 log unit steps, and the ratio closest to one is indicated by the filled symbol. The ends of the curves associated with low (white component larger) and high (chromatic component larger) ratios are marked with W and λ respectively. Increasing ratio is indicated by the arrows on the curves. *A*, different wavelengths were alternated with white at 10 Hz. For wavelengths below 570 nm an anticlockwise rotation of the response vector occurs, indicating a gradual phase advance (cf. Fig. 8*A*) For 570 nm, the curve passes close to the origin, indicating that the cell's responses went through a minimum. For wavelengths above 570 nm, a clockwise rotation occurs, indicating an increasing phase lag. *B* and *C*, plots similar to those in *A*, for another green on-centre cell for one short and one long wavelength at different temporal frequencies.

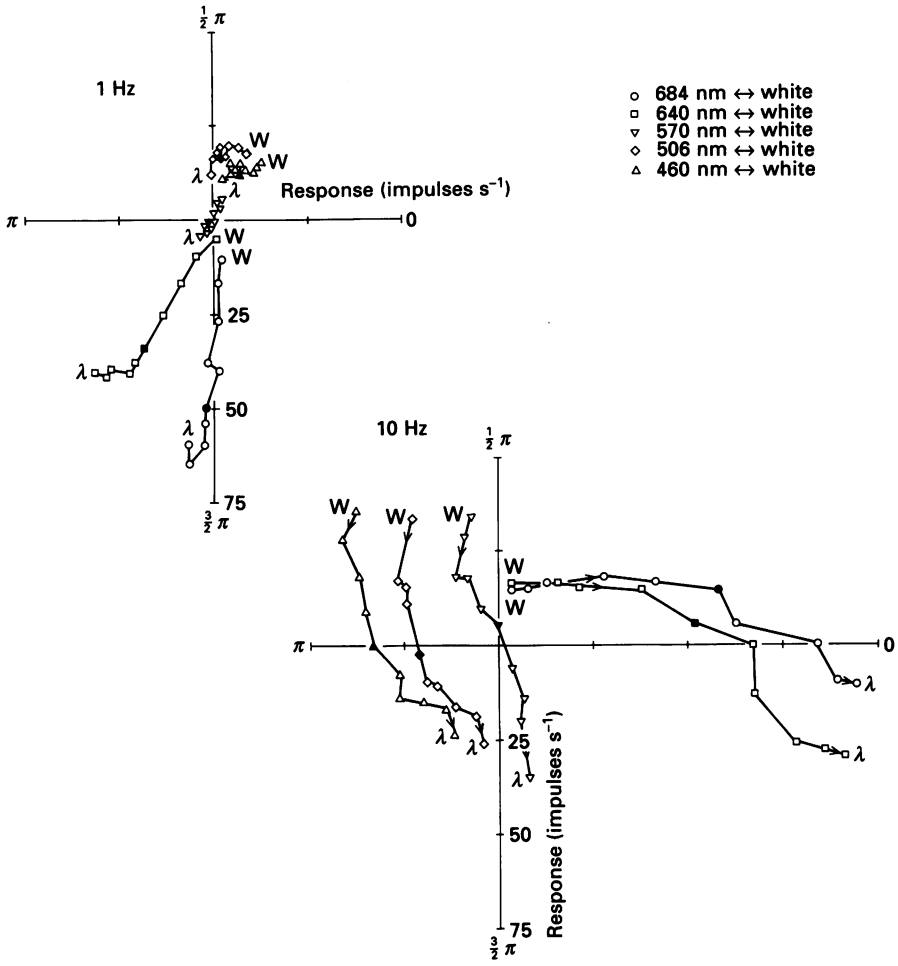

Fig. 4. Responses of a red on-centre cell presented in the same way as in Fig. 3. Different wavelengths were alternated with white at 1 and 10 Hz. The response phase changes which occur are in the opposite direction in comparison to the green on-centre cell in Fig. 3.

where L_W and L_λ are the amplitudes of the white and chromatic components of the flicker (in candela m^{-2}) and R_λ and G_λ are the excitations generated in the two cone mechanisms by the chromatic component, relative to those generated by the white component of the same luminance. A description of the method used to calculate cone excitations is given in the accompanying paper (Lee *et al.* 1989*a*). For wavelengths below about 570 nm, $R_\lambda < 1 < G_\lambda$ while for wavelengths longer than 570 nm $R_\lambda > 1 > G_\lambda$. White and 570 nm lie close to a tritanopic confusion line, so that for 570 nm $R_\lambda \approx G_\lambda \approx 1$.

The response phase ϕ' will be given by

$$\phi' = \tan^{-1}(N_{imag}/N_{real}), \tag{5}$$

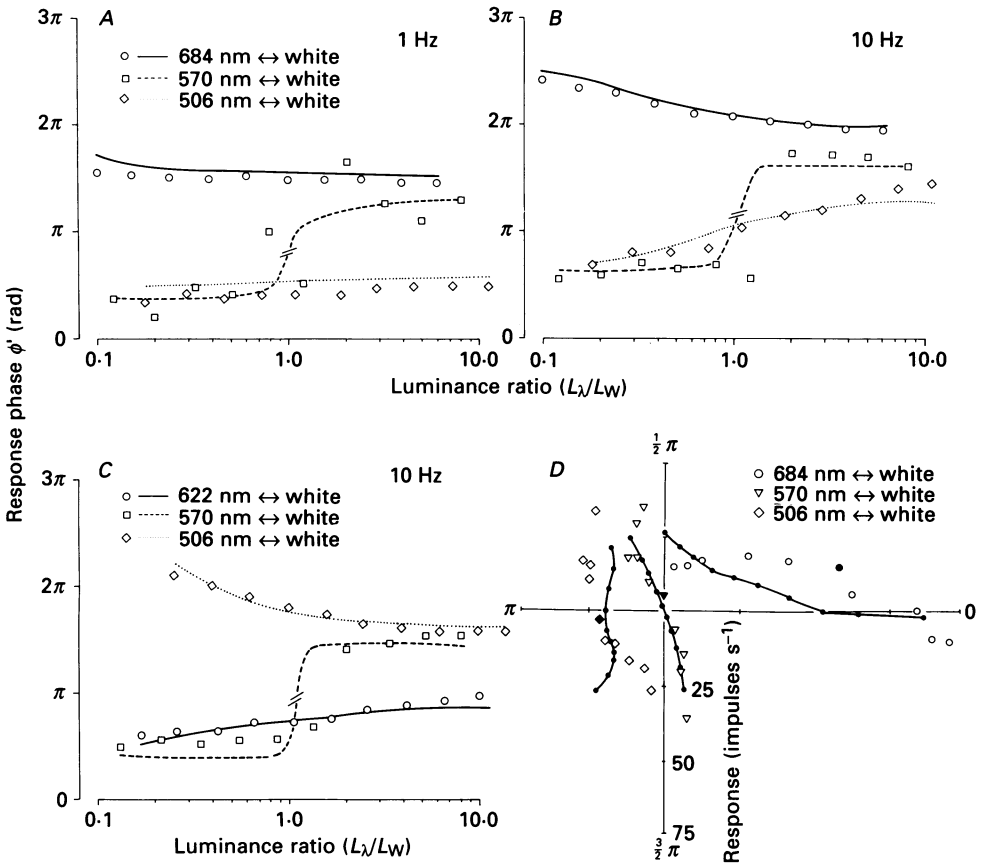

Fig. 5. Simulation of changes in response phase on the basis of a phase delay between centre and surround. In *A* and *B*, data of the red on-centre cell in Fig. 4 were used. Only three wavelengths are shown, although all wavelengths at 1 and 10 Hz were used for fitting the curves. The fit to the phase data is satisfactory. In *C*, results of similar fitting procedure was carried out for the cell of Fig. 3*A* (three out of seven wavelengths shown). For both cells, response phase can be accounted for satisfactorily. In *D*, response amplitude and phase are compared on polar co-ordinates for the same data as in *B*. Each curve is associated with the points lying closest to it. Prediction of response amplitude is barely adequate.

and response amplitude

$$N = (N_{\text{imag}}^2 + N_{\text{real}}^2)^{1/2}. \tag{6}$$

For response phase, using the expressions (1) and (2) above, we can derive

$$\phi' = \omega d - \tan^{-1}(\sin \omega d_\delta / (A_r R / A_g G - \cos \omega d_\delta)). \tag{7}$$

It can be seen that as luminance ratio is increased a progressive advance in response phase will occur when a wavelength longer than 570 nm is used, for R decreases more rapidly than G (as a consequence of eqns (3) and (4)). Conversely, for wavelengths below 570 nm a progressive phase lag occurs as G decreases more rapidly than R. This is the behaviour exhibited by the red on-centre cell in Fig. 4, and by other

red on-centre cells so that results were consistent with the M-cone mechanism (feeding the surround) being delayed relative to the L-cone mechansism (feeding the centre).

In the case of a green on-centre cell, the same analysis may be applied. If for such cells the L-cone mechanism (feeding the surround) is delayed with respect to the M-cone mechanism (feeding the centre), the reverse behaviour would be expected, as seen in the green on-centre cell of Fig. 3, and in other such neurones.

In the eqn (7) above, response phase is a function of three free parameters, d, d_δ and A_r/A_g. For eight cells we attempted to adjust these parameters to provide a best fit to response phase. A pseudo-Gauss–Newton non-linear regression procedure was used (BMDP statistics package).

A simulation (using seven wavelengths) of response phase for the red on-centre cell of Fig. 4 is shown for three wavelengths (506, 570 and 684 nm) in Fig. 5*A* and *B*. Optimal values for the parameters were for d, 26 ms, for d_δ, 6·7 ms, and for A_r/A_g, 1·06. The phase behaviour of the cell is well reproduced at both temporal frequencies, 96% of the variance in phase being accounted for by the simulation.

Results from a simulation of response phase of the green on-centre cell in Fig. 3*A* are shown in Fig. 5*C*, again for three wavelengths. The simulation can account for response phase reasonably well. Optimal values for the parameters were for d, 15·0 ms, for d_δ, 4·3 ms and for A_g/A_r, 1·2, with 92% of the variance of phase being accounted for by the simulation.

Response phase for the green on-centre cell in Fig. 3*B* and *C* could also be simulated satisfactorily, as was also the case for the other six cells analysed. Values of d ranged from 14 to 30 ms, and of d_δ from 3 to 8 ms.

We also attempted to account for response amplitude using the values of d, d_δ, and A_r/A_g derived from eqn (7). A threshold non-linearity was introduced, in that instantaneous firing rate of the cell was not allowed to go below zero. Such rectification is apparent in the responses shown in Fig. 2.

Simulation of response amplitude was less satisfactory than for phase. In Fig. 5*D* results are shown for the red on-centre cell for 10 Hz. The points are redrawn from Fig. 4, and the curves associated with each set of symbols are derived from the simulation. The shape of the calculated curves does not adequately reproduce the experimental data.

There are a number of reasons why, for response amplitude, the simple approach used here may be unsatisfactory. For example, it may be necessary to take into account the non-linearity in cone signals (Valberg, Lee & Tryti, 1987). However, a more significant factor may have been the change in time-averaged mean chromaticity and luminance at different ratios and wavelengths. This could have caused a change in the weightings, A_r and A_g. Such a change would be expected to affect response amplitude much more than response phase, for which only the ratio A_r/A_g is a determining factor. For most opponent cells, this ratio of cone weightings is close to one (Derrington, Krauskopf & Lennie, 1984; Lee *et al.* 1987), and a small change would not substantially affect the response phase simulation; the ratio R/G in eqn (7) changed 60-fold over the intensity range employed.

We conclude that phase of tonic cell response to heterochromatic flicker can be accounted for by assuming a centre–surround latency difference of 3–8 ms. We have

recently been able to confirm these values using a more direct measurement technique (Smith, Lee, Pokorny, Martin & Valberg, 1989). In subsequent sections we discuss the effect of this delay on responses to other stimuli.

Heterochromatic flicker responses of cells with S-cone input

Cells with S-cone input responded well when spectral mixtures lying along a tritanopic confusion line were alternated. Five blue on-centre cells were tested with heterochromatic flicker using various wavelengths and frequencies, as was one cell with inhibitory S-cone and excitatory M-cone input. This latter cell class is rare in the lateral geniculate nucleus (Valberg, Lee & Tigwell, 1986) though forming a distinctive population. All cells responded well to 570 nm–white flicker. As with red and green on-centre cells, response phase varied with luminance ratio. However, the changes observed for different wavelengths varied among different blue on-centre cells. Although we have not attempted to model these cells' responses, it seems likely that the changes observed may also be attributed to a difference in phase delay between opponent cone mechanisms.

Responses to luminance flicker of different wavelengths

Changes in the spectral responsiveness of tonic cells as flicker frequency is increased were first noted by Gouras & Zrenner (1979), and were attributed to a centre–surround latency difference. They suggested a centre–surround delay of 15 ms. We have shown above that a delay of 3–8 ms is more plausible. It was also reported that on average the spectral responsiveness of tonic cells approximates the V_λ function at 33 Hz, for opponent M- and L-cone signals should add at this frequency with a 15 ms centre–surround delay. However, a latency difference of 3–8 ms would not generate a synchronization of opponent cone signals until 62·5 Hz at least. Few tonic cells responded at such temporal frequencies.

We recorded responses to luminance flicker of different wavelengths from all major classes of ganglion cells, twenty-four tonic cells receiving $+M-L$ or $+L-M$ input, six receiving S-cone input and six phasic cells. Spectral tuning of phasic ganglion cells is illustrated in Fig. 6 for an on- and an off-centre phasic cell. Although amplitude is temporal frequency dependent, at a given frequency almost all wavelengths of luminance flicker evoke a similar response. This is as expected of a cell type with V_λ sensitivity (de Monasterio, 1978; Crook, Lee, Tigwell & Valberg, 1987; Lee *et al.* 1988). Also, phase is independent of flicker frequency, except at 646 nm. With this wavelength, a decrease in amplitude and phase change at higher temporal frequencies is seen. Almost all phasic cells showed this effect.

For tonic cells, variation in spectral responsiveness with temporal frequency was substantial, and is shown in Fig. 7 for a green on-centre and a red on-centre cell. At low frequencies (1 or 2 Hz), cells show the expected spectral responsiveness. The green on-centre cell is an illustrative example. Responses peak at two spectral loci, with little response at an intermediate wavelength, the so-called cross-over point. At this point, a sharp phase reversal occurs, indicating that an excitatory response was present below about 570 nm and an inhibitory response above that value. Increasing flicker frequency causes a change in spectral responsiveness. It becomes more broadband, and response phase now changes gradually with wavelength. For the red

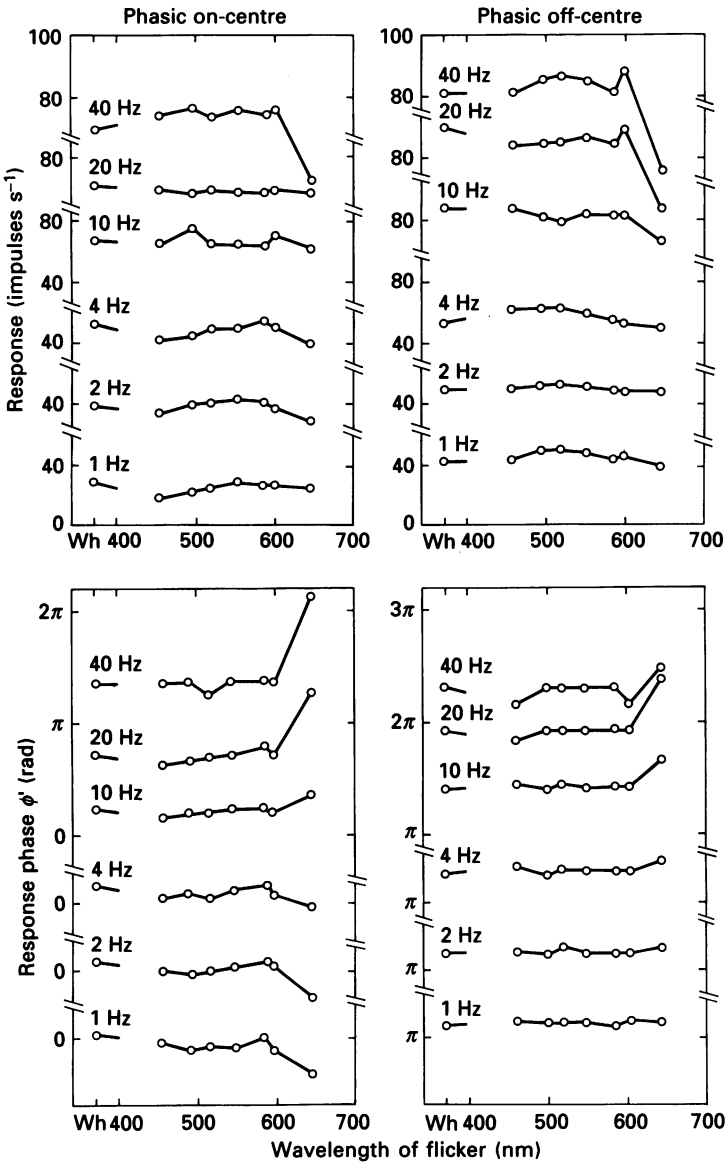

Fig. 6. Response amplitude and phase for luminance flicker with different dominant wavelengths and for white (Wh), for two phasic cells at different temporal frequencies. Except with a wavelength of 646 nm, response amplitude and phase are independent of the colour of the flickering light. Four degree fields were used.

on-centre cell, spectral responsiveness does not change so markedly as frequency is increased, but response phase becomes very wavelength-dependent. Although the cells of Fig. 7 are to some extent representative, great intercell variability was found for all cell types, both in spectral responsiveness and response phase. For eleven cells, we tested different contrast levels, with similar results.

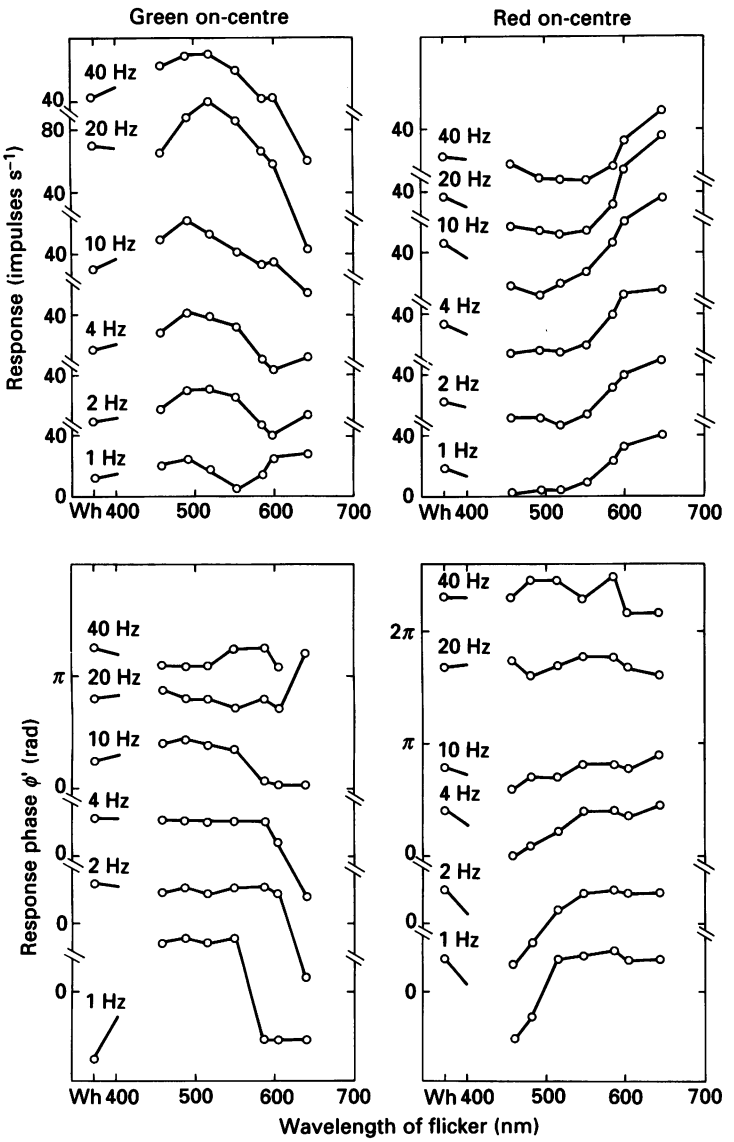

Fig. 7. Response amplitude and phase for a red and a green on-centre cell for luminance flicker (90% contrast) at different dominant wavelengths and temporal frequencies. Spectral responsiveness is frequency dependent. Four degree fields were used.

The changes shown in Fig. 7 are more complicated than expected from the description of Gouras & Zrenner (1979). These authors implied that opponent cells could acquire V_λ spectral responsiveness at high flicker frequencies. We found it to be very rare for a tonic cell to even approximate such a spectral responsiveness as the phasic cells in Fig. 6, though there was a tendency for them to become more broadband.

Figure 8*A* shows a schematic illustration of what may occur. For an opponent cell,

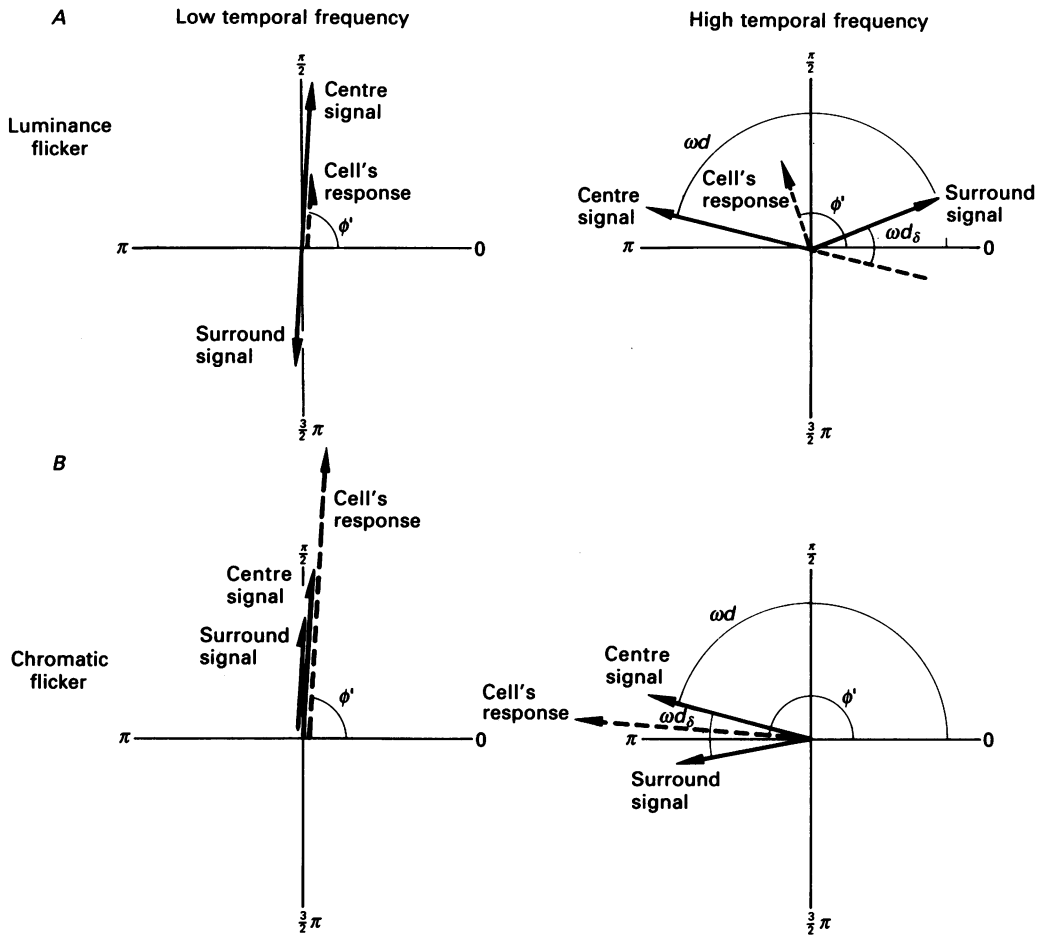

Fig. 8. *A*, a schematic illustration of the effect a phase delay on response and phase of tonic cells. For luminance flicker, at low temporal frequency, centre and surround are directly in antiphase, and cell response is dependent on the difference in activation of centre and surround, without response phase (ϕ') being affected. At high temporal frequency, the centre signal is delayed by ωd, and the surround signal by an additional ωd_δ radians. Cell response and phase now become dependent not only on the relative activation of centre and surround, but also on ωd_δ and thus on temporal frequency. Response phase can vary over $\pi - \omega d_\delta$. For chromatic flicker, centre and surround signals reinforce one another at low temporal frequencies. Changing temporal frequency separates their signals by ωd_δ radians, but this will not cause such a big change in response phase as with luminance flicker.

centre and surround have different spectral sensitivities. With luminance flicker, at low temporal frequencies, centre and surround exert effects almost in counterphase (π rad), and a cell's response is related to the difference between centre and surround signals. With luminance flicker of different wavelengths, the relative lengths of the centre and surround vectors change, and the resultant response changes sharply in phase when the centre–surround difference changes sign.

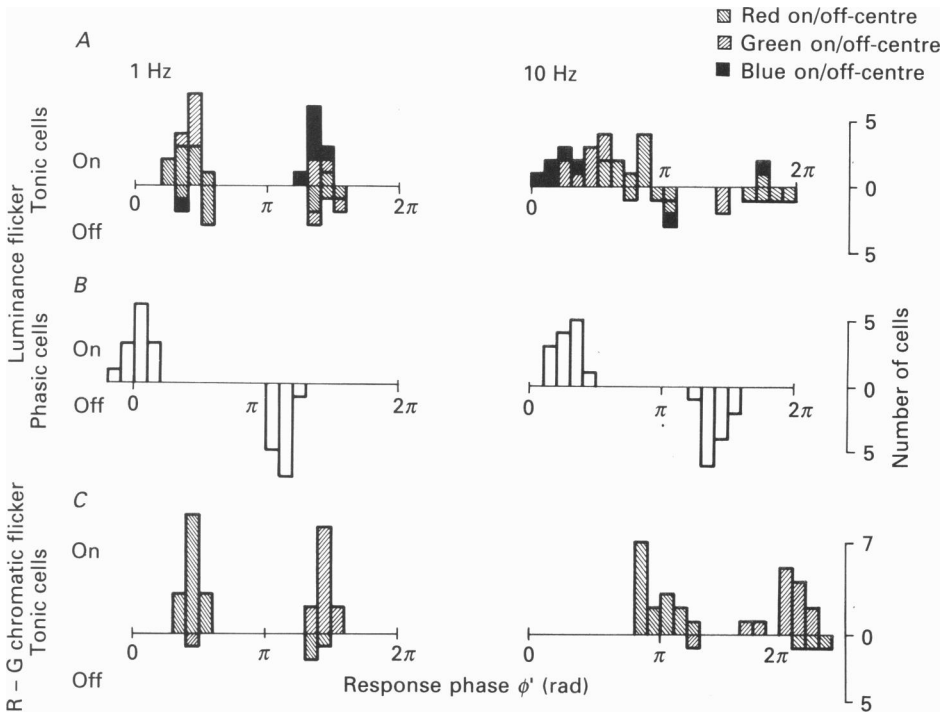

Fig. 9. *A*, response phase of tonic cells becomes variable as frequency of luminance flicker is increased. This is shown for achromatic luminance flicker. *B*, with phasic cells, this was not the case. *C*, with tonic cells and red–green (R–G) chromatic flicker (when the centre and surround vectors lie only ωd_δ apart), less variability was present at 10 Hz, as might be predicted from Fig. 8.

As temporal frequency is increased, a phase delay rotates both centre and surround vectors. If the delay of the centre vector is ωd, and the delay of the surround vector is greater than that of the centre vector by ωd_δ, the two vectors make an angle of $\pi - \omega d_\delta$. The cell's response has a phase of ϕ'. With different wavelengths, the relative magnitude of centre and surround vectors will change, altering both response magnitude and response phase. For a particular cell, the changes which occur will depend on both the relative strengths of the opponent mechanisms, and on the value of the phase delay, ωd_δ, of the surround relative to the centre. Thus, a substantial intercell variability is possible.

In Fig. 8*B* we have sketched the same kind of analysis for chromatic flicker. Signals of centre and surround now act synergistically. Centre and surround vectors will only be separated by ωd_δ radians, and thus less variability in response phase is to be expected.

If this explanation is valid, response phase among a population of tonic cells, stimulated with, for example, white luminance flicker, should become variable as flicker frequency is increased.

In Fig. 9*A* and *B*, we compare response phase of tonic ganglion cells to achromatic and chromatic flicker at 1 and 10 Hz. At 1 Hz, response phase (ϕ') of tonic cells is

close to $\pi/2$ or $3\pi/2$. This indicates that the modulation of cells' firing rate closely follows the luminance modulation of the stimulus. Generally, those cells classified as red or green on-centre are excited by an increase in achromatic luminance, and those classified as red or green off-centre by a decrease in luminance. At 10 Hz with luminance flicker, variability in response phase of tonic cells is substantial, as can be seen from the distribution of the cells along the phase axis.

With chromatic flicker, variability in response phase should be more restricted. This is shown in Fig. 9*C* to be the case. Cells with +M−L (green on-centre and red off-centre) and +L−M (red on-centre and green off-centre) cone inputs responded out of phase with one another, as would be expected.

We also show a similar analysis for phasic cells in Fig. 9. At 10 Hz, response phase was much less variable than with tonic cells. At 1 Hz, response phase is earlier than for tonic cells by about $1/2\pi$, indicating cells respond when rate of luminance change is maximal. The difference in time course of response of phasic and tonic cells (Dreher, Fukuda & Rodieck, 1976; Schiller & Malpeli, 1978) may account for the difference in response phase between the two cell types.

DISCUSSION

For a non-opponent ganglion cell, the only way to separate contributions to the response from centre and surround is to manipulate the spatial configuration of a stimulus. For X-cells of the cat, response phase and amplitude are dependent on the spatial frequency of a grating pattern, and this can be modelled in terms of centre and surround mechanisms between which there is a phase delay (Enroth-Cugell *et al.* 1983; Frishman, Freeman, Troy, Schweitzer-Tong & Enroth-Cugell, 1987). For tonic, wavelength-opponent retinal ganglion cells of the monkey, another possibility for differential stimulation of centre and surround is present, namely variation in spectral composition of the stimulus.

In the results presented here, we have shown that several distinctive features of the responses of wavelength-opponent cells probably arise from a delay between the antagonistic cone mechanisms which feed centre and surround. Firstly, under most conditions of heterochromatic flicker, tonic cells showed gradual changes in response phase as the relative luminance of flicker components was altered, and this was dependent on the temporal frequency and on the wavelength of the flicker components. On the basis of a simple model, we could show that the direction and magnitude of these phase changes could be explained by a delay of 3–8 ms between centre and surround. This is roughly the range found by Enroth-Cugell *et al.* (1983) for X-cells of the cat.

Secondly, the spectral responsiveness of tonic cells to luminance flicker with lights of different wavelengths depends on flicker frequency. Response phase with different wavelengths is also shown to be very dependent on frequency. These effects can also be explained on the basis of a phase delay, as was first postulated by Gouras & Zrenner (1979), who first noted this effect. However, the averaged curve they show, resembling the V_λ function, may mask substantial intercell variability, for individual opponent cells hardly ever conformed to the luminosity function in spectral responsiveness. Given the variability in cone weightings, and variability in phase delay, the very heterogenous behaviour found is not surprising.

Thirdly, phase of response to achromatic flicker at 10 Hz varied from cell to cell, but less so in the case of chromatic flicker. This may be understood in terms of the response vectors for the opponent mechanisms. If they are approximately in counterphase, as is the case for achromatic flicker, minor rotation of one vector relative to the other (due to a phase delay) will result in an increase in response and a marked change in response phase as temporal frequency is increased. For chromatic flicker, with the vectors at a similar angle, response amplitude and phase will be less sensitive to the presence of a small phase delay angle between them.

Two sources of such a phase delay could be either a *transport delay* or a *low-pass filter* (Enroth-Cugell *et al.* 1983). We assumed that a transport delay was present, that is, the phase delay is proportional to temporal frequency. This assumption is satisfactory for X-cells of the cat (Enroth-Cugell *et al.* 1983). On the other hand, Derrington & Lennie (1984) found a simple transport delay model did not work well for sensitivity of wavelength-opponent cells to achromatic gratings of different spatial and temporal frequencies. Since we did not test extensively with spots of different sizes, we do not have comparable data.

A further consequence of such a centre–surround phase delay will be a marked increase in sensitivity to luminance flicker as temporal frequency is increased, in contrast to chromatic flicker, for which sensitivity changes little (Lee *et al.* 1989*a*).

Consequences for psychophysics

In the accompanying paper (Lee *et al.* 1989*a*), we show that psychophysical detection of luminance flicker is consistent with detection by the phasic cell system whereas detection of chromatic flicker at low temporal frequencies is attributable to detection based on the activity of opponent cells. Thus, the different channels thought to mediate flicker detection psychophysically (Kelly & van Norren, 1977) may have physiological substrates in the phasic and tonic cell systems.

An alternative viewpoint is that summation between tonic cells could mediate achromatic flicker detection. This is unlikely, for tonic cells respond vigorously to chromatic flicker at 10 Hz, but this signal does not seem available for detection. It is difficult to understand how summation should be possible in one situation while in the other, tonic cells' signals are discarded. From the results presented here, summation is rendered even less likely. The phase variability observed with achromatic flicker would tend to substantially degrade any signal generated by simple combination of tonic cell activity.

As argued in the accompanying paper (Lee *et al.* 1989*a*), the significantly greater sensitivity at high temporal frequencies to chromatic flicker of tonic cells in comparison with a human observer implies that high-frequency components in these cells' signals are not utilized for detection. This 'filtering' of the signal presumably permits the observer to carry out a task such as flicker photometry on the basis of phasic cell activity alone.

We show here that opponent cell responses do not bear a straightforward relation to the spectral composition of a stimulus at high temporal frequencies; as far as chromaticity is concerned, they are a red herring. Also, there is evidence that phase information is critical for reconstruction of the visual image (Julesz & Schumer, 1981). From our results, response phase of opponent cells is likely to be dependent on both the chromatic composition in an image and the velocity with which it moves

across the retina. Thus, spatial information would be readily confounded by these other variables. We suggest that, since high-frequency components of tonic cell activity do not deliver an unambiguous chromatic or phase signal, they are discarded.

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