

# An Examination of Physiological Mechanisms Underlying the Frequency-Doubling Illusion

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**PURPOSE.** The frequency-doubling illusion is an apparent doubling of spatial frequency when a sinusoidal grating is modulated rapidly in temporal counterphase. It has been proposed that the illusion arises from a spatially nonlinear ganglion cell class. The current study reexamines this possibility and investigates other mechanisms that may underlie the illusion.

**METHODS.** Responses of macaque magnocellular (MC) retinal ganglion cells were recorded to counterphase-modulated sinusoidal gratings of various spatial frequencies, and linearity of spatial summation was assessed. Human psychophysical thresholds were measured for a variety of phase discrimination and matching tasks.

**RESULTS.** Consistent with lateral geniculate recordings reported by other authors, no evidence was found of a separate nonlinear ( $M_y$ ) MC cell class. The small, spatially nonlinear responses found were least at the low spatial frequencies used in clinical testing. Further analysis showed that no spatially modulated signal can be expected from the nonlinear response of a ganglion cell; the nonlinearity of spatial summation gives a doubled response in time but not across space. Psychophysical performance was consistent with an inability to distinguish the temporal phase of counterphase-modulated gratings when the illusion occurs. From 4 to 40 Hz, the zero-crossings of the modulated sinusoidal grating provided a spatial cue and were matched to comparison patterns at twice the stimulus spatial frequency.

**CONCLUSIONS.** These results are inconsistent with the hypothesis that spatially nonlinear ( $M_y$ ) retinal ganglion cells are the physiological substrate of the frequency-doubling illusion. A cortical loss of temporal phase discrimination may be the principle cause of the illusion. (*Invest Ophthalmol Vis Sci*. 2002;43:3590-3599)

The frequency-doubling illusion was first described by Kelly.<sup>1</sup> An achromatic grating made to flicker in counterphase is perceived to be twice its spatial frequency, provided the grating is of low spatial frequency and the counterphase temporal modulation is rapid (20–30 Hz). Recently, a method

for clinical testing for glaucoma was developed based on an interpretation of the physiological basis of this illusion.<sup>2</sup> It has been proposed that the physiological substrate of the percept is “Y-like,” spatially nonlinear, magnocellular (MC) retinal ganglion cells in the retina.<sup>3</sup>

The terms “X” and “Y” cell were originally coined to describe cat ganglion cells<sup>4–10</sup>; a characteristic feature of cat Y-cells is nonlinear spatial summation. This nonlinearity is thought to arise because of subunits within the field structure that rectify the visual signal.<sup>6</sup> In studies in Old-World primates such as macaque, at the level of the lateral geniculate nucleus (LGN), only a small fraction of cells show some degree of nonlinear spatial summation.<sup>11–14</sup> These cells were thought to be a subset of cells of the MC pathway and hence were termed Y-like MC ( $M_y$ ) cells, with the remainder of spatially linear cells being termed X-like.<sup>13</sup> It should be noted that spatial nonlinearity is not related to the temporal nonlinear summation of cone photoreceptor signals seen with chromatic stimuli, which is a property of all MC cells.<sup>15</sup>

Tests of linearity of spatial summation commonly involve a counterphase-modulated grating stimulus. An illustration of this stimulus and the response of a highly linear MC cell compared with that of a more nonlinear MC cell is shown in Figure 1. Averaged responses to the stimulus at four grating positions are shown. For the spatially linear cell, there is a null position of the grating, where there is no response. The first harmonic response of the less-linear cell also shows a null position, but the second harmonic response (a temporally frequency-doubled response) shows no variation at different grating locations. Figure 1 also shows that the temporal phase of the linear cell's response is quite different to gratings 180° spatially out of phase, whereas the temporally frequency-doubled response of the less-linear cell does not vary with spatial location. This inability of less-linear cells to distinguish between the onset and offset of light in terms of spatial phase led to a proposal that these cells may form the retinal substrate of the frequency-doubling illusion.<sup>3</sup> It has been argued that if these cells were large and sparse across the retina, with limited redundancy, then retinal ganglion cell damage in the early stages of glaucoma could produce reduced sensitivity for the frequency-doubling illusion.<sup>3</sup>

This study was an examination of the physiological feasibility of the hypothesis that a subclass of MC cells underlies the percept of the frequency-doubling illusion. We reviewed physiological properties of the MC pathway in the context of spatial nonlinearity, and considered whether this might be a substrate for the illusion. We present findings suggesting that spatially nonlinear ganglion cells are unlikely to be the substrate for the illusion. We then consider alternative, cortical origins of this percept.

## METHODS

### Physiological Experiments

We recorded *in vivo* from the retinas of six adult anesthetized macaque monkeys (*Macaca fascicularis*). The animals were initially sedated with an intramuscular injection of ketamine (10 mg/kg). Anesthesia was maintained with inhaled isoflurane (0.2%–2%) in a 70:30 N<sub>2</sub>O-O<sub>2</sub>

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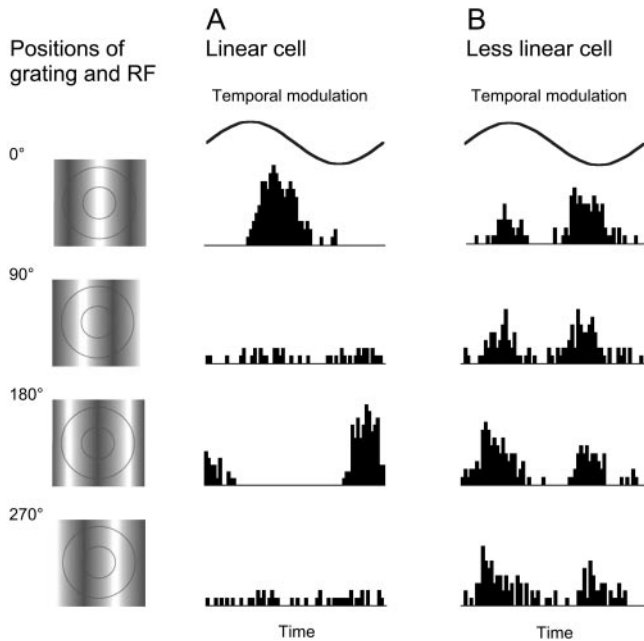
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**FIGURE 1.** Typical responses of a spatially linear MC cell (A) and a less-linear MC cell (B). *Left:* relative position of the grating and the cell's receptive field (RF). The histograms show the cells' responses to one cycle of temporal modulation (temporal frequency, 17.4 Hz; spatial frequency, 1.6 cyc/deg; binwidth, 0.9 msec; 64 bins/cycle). When a grating was centered on the receptive field (spatial phase 0° or 180°), the linear cell (A) responded vigorously, with a 180° temporal phase shift in response between the spatial 0° and 180° conditions. In the 90° and 270° conditions, there was little response, because summation is linear. From the less-linear cell (B), there was a nonlinear response (two peaks per cycle of temporal modulation) at all grating spatial phases, as well as a linear response component.

mixture. Local anesthetic was applied to points of surgical intervention. Electroencephalogram (EEGs) and electrocardiogram (ECGs) 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 per hour, intravenously) with accompanying dextrose Ringer's solution (5 mL/h). Body temperature was kept close to 37.5°C. End-tidal CO<sub>2</sub> was brought close to 4% by adjusting the rate of respiration. All procedures were approved by the State of Lower Saxony Animal Welfare Committee and conformed to European Union guidelines for ethical care of animals and to the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research.

A tungsten-in-glass recording microelectrode was introduced to the retina through a scleral hole by established techniques. The details of the preparation can be found elsewhere.<sup>16</sup> Cells were recorded both in the central and peripheral retina. The location of each cell's receptive field was mapped onto a tangent screen 114 cm from the eye. Cells were initially characterized on the basis of both their antidromic latency after electrical stimulation of the optic chiasm and their responses to luminance and chromatic stimuli. Spatial dimensions of cells' receptive fields were also measured by using drifting gratings of different spatial frequencies.

Visual stimuli were generated through a graphics controller (Visual Stimulus Generator [VSG] series 3; Cambridge Research Systems, Cambridge, UK) and presented on a CRT monitor (Barco, Kortrijk, Belgium) 114 cm from the eye (frame rate 195 Hz, mean luminance 40 cd/m<sup>2</sup>). The stimuli were temporally counterphase-modulated sinusoidal gratings of various spatial frequencies. The temporal frequency was 3.68 or 17.44 Hz. The spatial frequency was 0.2, 0.4, 0.8, 1.6, 3.2, or 6.4 cyc/deg. The spatial phase of the grating was advanced from 0° to 360° across the receptive field in 45° increments.

## Psychophysical Experiments

The same visual display system was used as that used in the physiological experiment, but with a viewing distance of 0.48 m. At this distance the visible area of the display subtended 40° by 26° of visual angle. Mean luminance of stimulus and background was 40 cd/m<sup>2</sup>, and chromaticity was (0.45, 0.47) in International Commission on Illumination (CIE) *x, y* coordinates.

Psychophysical experiments included measurements of phase-discrimination threshold, detection threshold, and zero-crossing matching. In all experiments, the observer viewed the targets monocularly.

In some experiments, we used rectified gratings generated with a graphics controller (VSG 2/5; Cambridge Research Systems) and presented on a 21-in. monitor (Trinitron; Sony, Tokyo, Japan) at a viewing distance of 148 cm. Other conditions were very similar to the main experimental set.

**Phase Discrimination and Detection.** The stimuli consisted of a pair of temporally counterphase-modulated sinusoidal gratings. The two horizontal gratings (each 19° wide, 25° high) were presented simultaneously side by side with a 30-minute gap. In further experiments, sawtooth or square-wave gratings, or a rectified sine wave grating was used. Phase discrimination and detection contrast thresholds were measured with a two-alternative, forced-choice procedure involving randomly interleaved dual staircases.

For the discrimination task, each trial included two 300-ms presentations with a 200-ms interstimulus interval. One presentation contained a pair of gratings that were temporally in phase, and the other a pair of gratings 180° out of phase (for counterphase-modulated gratings, 180° temporally out of phase is equivalent to 180° spatially out of phase). The observer's task was to indicate which of the two presentations contained the in-phase gratings.

For the detection task, similar stimuli were used, except that only one of the two presentations contained a pair of in-phase gratings, and the other presentation was blank. The observer's task was to indicate which of the two presentations contained the gratings.

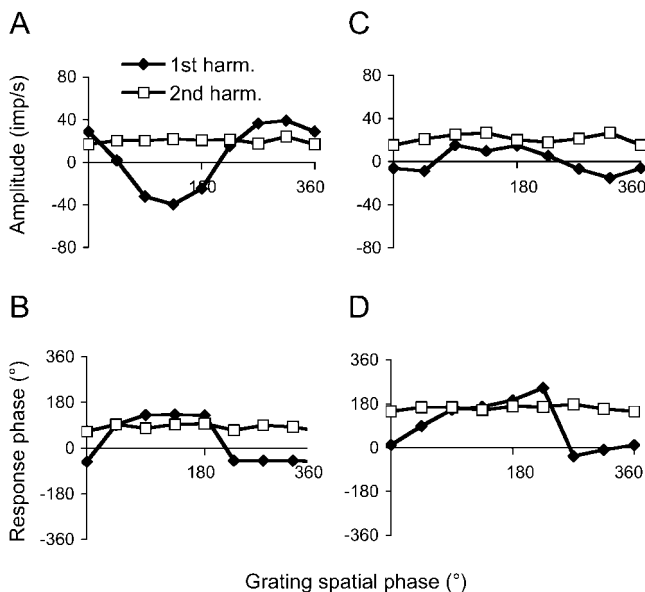
Contrast thresholds (71% correct responses) were computed as the means of 20 reversals of four staircases for both tasks. A complete data set was obtained with foveal fixation (spatial frequency: 0.2 and 1.0 cyc/deg, temporal frequency: 1, 2, 4, 8, 11, 16, 20, 30, and 40 Hz). For comparison, a partial data set was collected at 5°, 10°, and 20° eccentricity (spatial frequency: 0.2 cyc/deg, temporal frequency: 4–40 Hz).

In further experiments, discrimination thresholds were measured between pairs of in-phase gratings and pairs of temporally or spatially 90° phase-shifted gratings at 0.2 cyc/deg and 20 Hz.

**Zero-Crossing Matching.** The stimuli consisted of a pair of horizontal gratings simultaneously presented side by side with a 30-minute gap. One grating, the test, was a counterphase-modulated sinusoidal or sawtooth grating (spatial frequency: 0.2 cyc/deg, temporal frequency: 4–20 Hz). The other grating, the comparison pattern, was a static square-wave grating with a restricted duty cycle (10%) so that it appeared on the screen as a series of equally spaced, thin, bright lines on a dark background. The test and comparison gratings were of equal contrast. The observer's task was to adjust the spatial frequency and phase of the comparison grating until the bright bars aligned with the regions of minimal flicker (the zero-crossing regions) of the test grating. Initial training for all observers was with a 20-Hz test grating, because the zero-crossings were most prominent at this and higher temporal frequencies.

Matching spatial frequency and phase were measured with the method of adjustment. The temporal frequency and spatial phase of the reference gratings were randomized from trial to trial. Each matching result was an average of 10 settings. Data were collected with foveal fixation.

**Observers.** Three observers participated in the experiments. Observers AJRW and HS were authors, and JK was a naïve observer. All observers had normal color vision, as assessed with a Neitz anomaloscope, Ishihara pseudoisochromatic plates and Farnsworth-Munsell 100-Hue (Munsell Color Services, New Windsor, NY). Observer HS is



**FIGURE 2.** Amplitude and phase of the first and second harmonic components of MC cells' responses to counterphase-modulated sinusoidal gratings (17.4 Hz, 0.2 cyc/deg). (A, B) Results for a spatially linear MC cell (nonlinearity index = 0.59). (C, D) Equivalent data for a spatially less-linear cell (nonlinearity index = 1.73). When first harmonic response phases undergo an approximate 180° shift, response amplitude is made negative. Second harmonic amplitude and phase are independent of spatial phase. Stimulus parameters as described in Figure 1.

myopic and wore corrective lenses. Observers AJRW and JK are emmetropic. Observers provided informed written consent according to a protocol conforming to the Declaration of Helsinki and approved by the State University of New York, State College of Optometry Institutional Review Board.

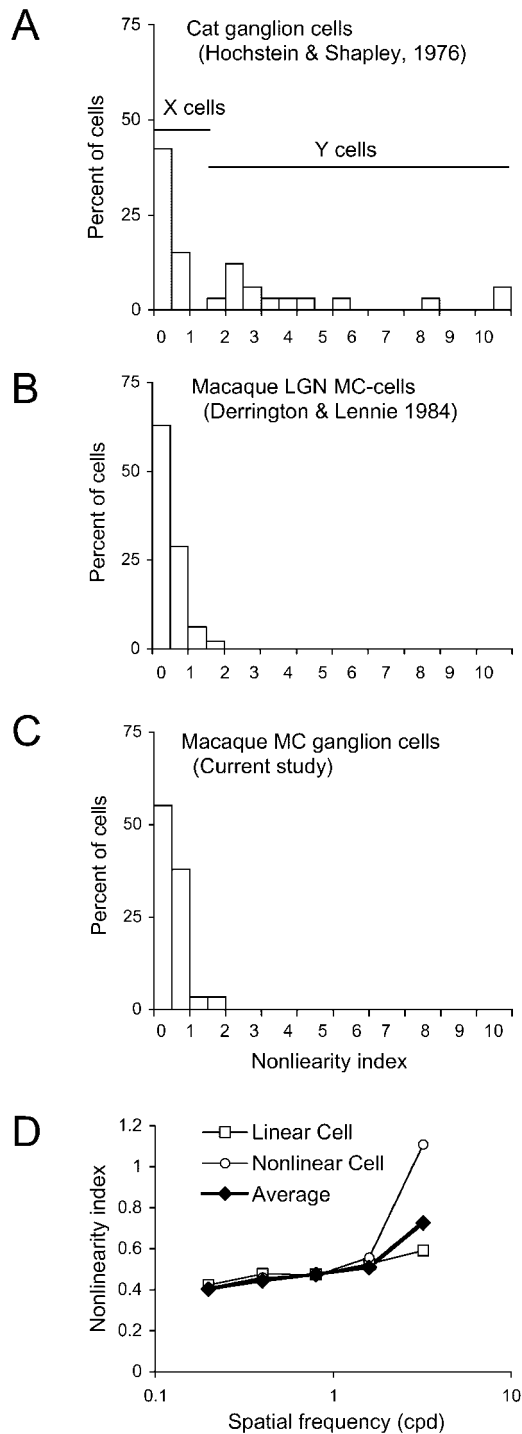
## RESULTS

### Evaluation of Spatial Nonlinearity in Macaque MC Ganglion Cells

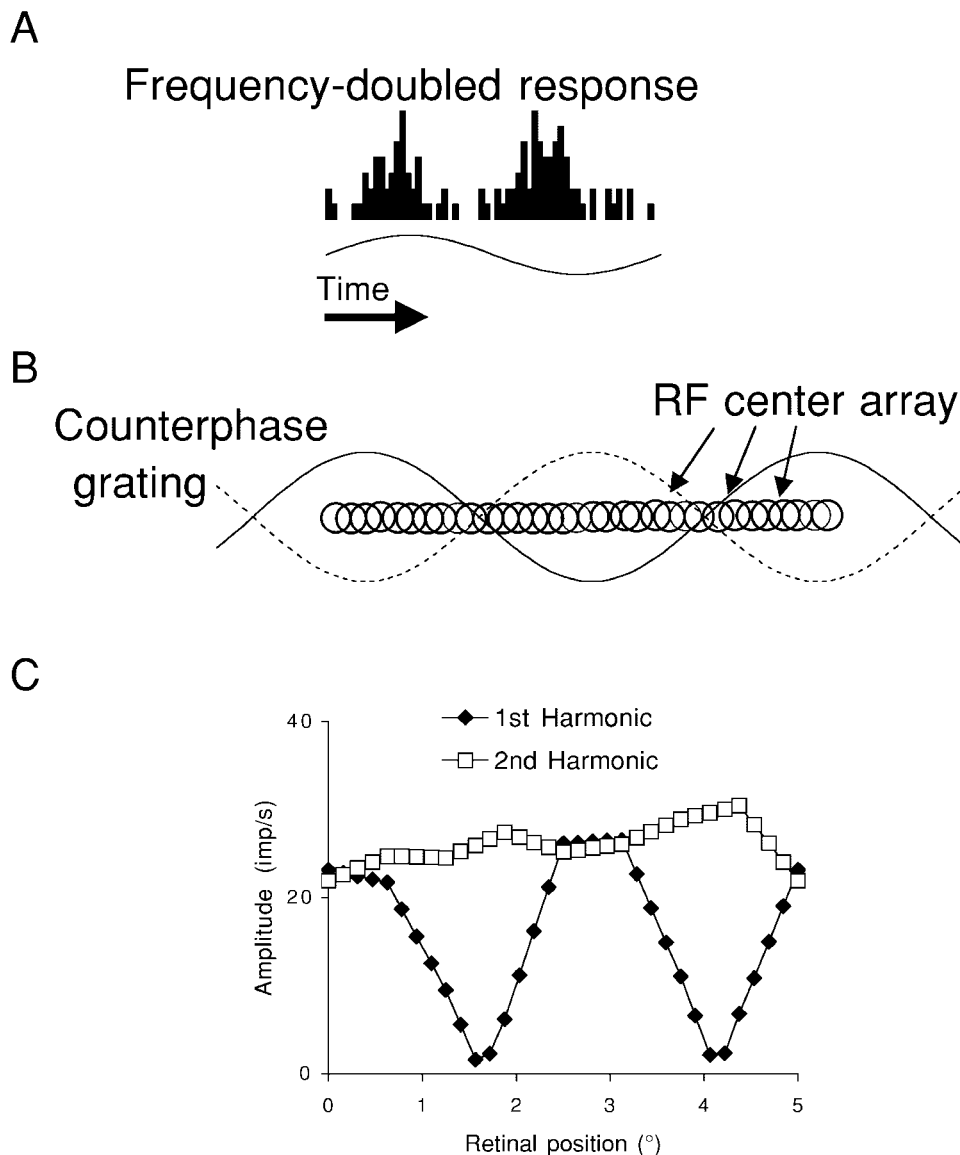
Previous tests of spatial linearity in the macaque visual pathway have largely been restricted to LGN recordings.<sup>11-14</sup> For comparative purposes, we tested retinal ganglion cells. Recordings were obtained from 36 MC cells from both fovea (11 cells) and peripheral (25 cells, 20–50°) retina, because a critical region for clinical use of frequency-doubling technology (FDT) is in the peripheral visual field.<sup>2,17</sup> Cells were classified as MC cells on the basis of their response properties to handheld stimuli (high contrast sensitivity, no color selectivity, phasic responses). Antidromic latency measurements ( $4.74 \pm 0.72$  ms [SD],  $n = 16$ ) confirmed this identification.

All cells were tested with counterphase luminance-modulated sinusoidal gratings at a spatial frequency twice that which gave the peak response. Typical cells from our sample are shown in Figure 1 and further examples are shown in Figure 2. In the histograms (Fig. 1), the amplitude of the first harmonic depended on the grating location, whereas the amplitude and phase of the second harmonic response appeared independent of grating position. In Figure 2 are plots of response amplitude and phase of a linear cell (Figs. 2A, 2B) and less-linear cell (Figs. 2C, 2D) which quantify these results. Amplitudes are made negative when the first harmonic phase changes by 180°.

A measure of spatial nonlinearity is the nonlinearity index, as originally defined in cat<sup>5</sup> and adapted for use in macaque



**FIGURE 3.** (A) Spatial nonlinearity indices of cat retinal ganglion cells ( $n = 33$ ). Data adapted from Hochstein S, Shapley RM. Linear and nonlinear spatial subunits in Y cat retinal ganglion cells. *J Physiol.* 1976;262:265–284. (B) Spatial nonlinearity indices of MC cells from macaque LGN ( $n = 97$ ). Data adapted from Derrington AM, Lennie P. Spatial and temporal contrast sensitivities of neurones in lateral geniculate nucleus of macaque. *J Physiol.* 1984;357:219–240. (C) Spatial nonlinearity indices of retinal MC cells ( $n = 36$ ). Macaque ganglion cells show a bimodal distribution for X- and Y-cells. Cat ganglion cells showed a unimodal distribution. (D) Nonlinearity index of macaque MC cells as a function of grating spatial frequency, averaged across cells ( $n = 11$ ), as well as for a highly linear cell and a less-linear cell.



**FIGURE 4.** (A) Spatial nonlinearity caused a frequency doubling in time. (B) A single row in an array of parafoveal MC cells (either on- or off-center) based on literature estimates. Each *disk* represents the center of a receptive field. *Solid* and *dashed sinusoids*: profiles of a counterphase-modulated 0.2-cyc/deg grating. (C) Response amplitudes expected for cells in the ganglion cell array as a function of spatial position. The points for individual cells were interpolated from actual measurements for a less-linear MC cell. Note that the amplitude of the first harmonic had high spatial modulation, whereas that of the second harmonic did not. Thus, no spatially modulated percept can derive from second harmonic responses.

LGN.<sup>12</sup> It is defined as the mean second harmonic response divided by the peak first harmonic response. There was no difference in nonlinearity indices at the two retinal eccentricities ( $t = 1.58$ ,  $P > 0.10$ ), and data have been combined. Figure 3 shows original cat data,<sup>5</sup> a published distribution for LGN MC cells,<sup>12</sup> and the distribution of nonlinearity indices for our MC cells. The cat data show the bimodal distribution of X- and Y-cells in this species, with Y-cells showing indices greater than 2. Macaque retinal and LGN distributions were similar and were unimodal, with few cells having indices greater than 1. The distributions remained unimodal when recalculated with the indices scaled logarithmically. The data in Figure 3 are in agreement with the previous studies in the macaque LGN,<sup>11-14</sup> in which investigators were unable to identify a separate MC cell Y-like class. It appears unlikely that electrode sampling bias could have led to  $M_y$  cells being missed in both our results from ganglion cells and previous data sets from LGN recordings. Factors influencing electrode sampling would presumably be unrelated at the two locations.

In tests for spatial nonlinearity, it is common to use spatial frequencies above the optimal spatial frequency for a cell. This decreases the first harmonic response and allows the second harmonic response to become more apparent. However, in

FDT, a spatial frequency of 0.25 cyc/deg is usually used.<sup>2,17</sup> Based on estimated center size,<sup>16</sup> this is below the peak spatial frequency of MC cells at eccentricities less than 30°. We therefore tested the dependence of the nonlinearity index on spatial frequency for the parafoveal cell sample ( $n = 11$ ) at spatial frequencies from 0.2 to 6.4 cyc/deg. At each spatial frequency, a nonlinearity index was calculated whenever either the first or second harmonic average response was greater than 10 impulses/sec. Figure 3D shows examples of the nonlinearity index as a function of spatial frequency. At less than 1 cyc/deg, the nonlinearity index was small and similar for all cells but increased at higher spatial frequencies in some cases. At the low spatial frequencies used in clinical tests,<sup>2,17</sup> linear response components are dominant in the parafovea.

### Spatial Nonlinearity as a Basis for the Frequency-Doubling Percept

In the previous section, we show that physiological evidence for a separate  $M_y$  class is weak. We then considered whether the frequency-doubling percept could be derived from such a nonlinear response, per se. Figure 4A shows a further example of a temporally frequency-doubled response from a less-linear

MC cell. The spatial nonlinearity caused a frequency-doubled response in time. There were two response peaks per temporal cycle of the stimulus. We then considered whether this implies a frequency-doubled response in space from a ganglion cell array.

Each ganglion cell type tiles the retina in a semiregular array, with separate arrays for on- and off-center MC cells and for other cell types.<sup>18</sup> The plots in Figure 2 represent the responses of a single cell relative to the location of a counterphase-modulated grating. The plot can be reformulated to represent the responses of an array of ganglion cells (of a single type with similar properties) to a single counterphase-modulated grating, as sketched in Figure 4B. The density of cells and center size shown are approximately what might be expected of MC cells at 5° eccentricity.<sup>18</sup> The grating spatial frequency is 0.2 cyc/deg. Amplitudes of the first and second harmonics were interpolated from the response of a single cell to predict the responses from the array (Fig. 4C). The amplitude of the first harmonic components would be modulated across the ganglion cell array. However, the second harmonic response, although frequency doubled in time (Fig. 4A), is not modulated in space. Thus, based on response amplitude, there would not appear to be a spatial cue in the second harmonic response to account for the doubled percept.

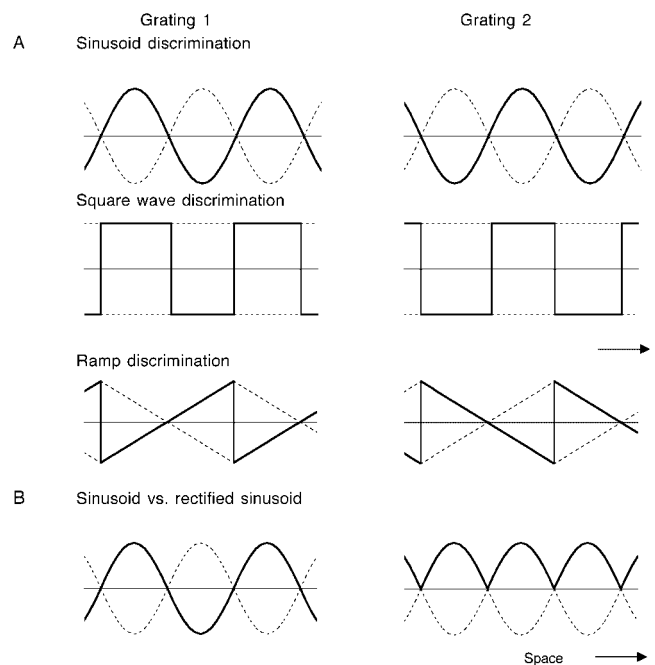
We considered whether the response phase of the second harmonic response could in any way act as a spatial cue. Response phase of first and second harmonic responses of two cells with differing nonlinearity indices were plotted as a function of grating position at 0.2 cyc/deg in Figure 2. Response phase of the first harmonic varies with grating spatial phase, regardless of whether the cell has a high or low nonlinearity index. Phase of the second harmonic is independent of grating phase for both cells shown. To confirm this result, we compared the phases of the second harmonic at the two peaks of the first harmonic amplitude. At 0.2 cyc/deg, mean phase difference of the second harmonic at these two peaks was  $32.7^\circ \pm 14.4^\circ$  (SD;  $n = 11$ ), which was not statistically significant (Watson and Williams test,  $P > 0.05^{19}$ ). The same analysis was repeated on the same cells at the highest spatial frequency that gave a response (average first harmonic or second harmonic response greater than 10 impulses/sec). This yielded a similar result. The possibility that the second harmonic phase provides spatial information can thus be rejected.

We suggest that the notion that the frequency-doubling illusion can arise from a nonlinear spatial response is based on a confounding of the nature of the nonlinearity: Although it is a nonlinearity of spatial summation, it causes a temporal rather than a spatial doubling of frequency.

### The Frequency-Doubling Illusion and Loss of Phase Information

Figure 4 showed responses of an array of ganglion cells as a function of spatial position. Observation of the frequency-doubling illusion suggests bars of intense flicker separated by regions of minimal flicker. These minima appear at approximately twice the spatial frequency of the test stimulus and correspond to the zero-crossings of the sinusoidal grating. A plausible explanation of the regions of minimum flicker would be minimal first harmonic responses of ganglion cells. On either side of these regions, ganglion cells respond vigorously. On- and off-center cells respond in counterphase. The doubled nature of the illusion implies that discrimination of the half-cycle temporal phase difference of neighboring flickering bars is difficult. We tested this by measuring the discrimination threshold of counterphase-modulated gratings at several spatial and temporal frequencies.

Two aligned gratings were presented side by side, each modulated in counterphase. In a two-alternative, forced-choice



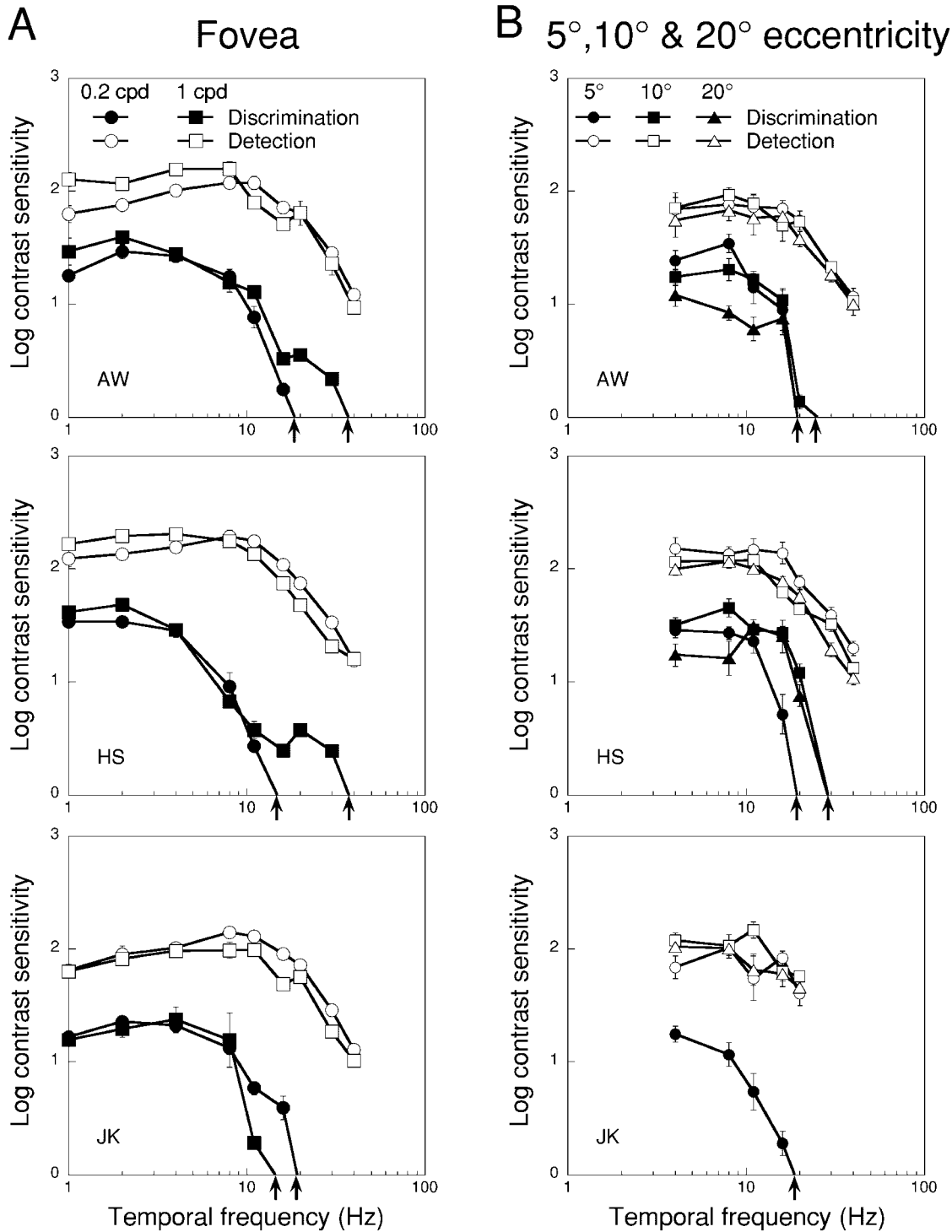
**FIGURE 5.** (A) Spatial waveforms in a discrimination task. Sinusoids, square waves, and ramps were counterphase modulated at different temporal frequencies, and observers were instructed to discriminate between the two gratings shown. (B) In a further discrimination task, a counterphase-modulated rectified sinusoid was compared with the standard condition. Note that for the rectified sinusoid there is always a residual response in cells near the zero-crossings. Under conditions for which the residual response was expected to be small, the rectified and standard sinusoidal gratings were indistinguishable.

procedure, observers had to make the discrimination illustrated in Figure 5A. Contrast thresholds for the discrimination were measured at 0.2 and 1 cyc/deg.

Discrimination and detection thresholds are shown in Figure 6. Figure 6A shows foveal data for the three observers. At the lowest temporal frequencies, the grating phase could be reliably distinguished if contrast was approximately 0.5 log unit above detection threshold. However, grating phase discrimination rapidly degraded beyond 8 Hz and became impossible beyond approximately 20 Hz (contrast sensitivity became less than one, indicated by arrows). Detection sensitivity is preserved well beyond 20 Hz. Similar results were found for both the 0.2-cyc/deg grating and the 1-cyc/deg grating. Similar results were also found when the task was repeated with the 0.2-cyc/deg grating in more peripheral visual field locations (Fig. 6B).

Loss in temporal phase discrimination should also occur for other counterphase-modulated waveforms. The phase-discrimination task was repeated for square and sawtooth spatial waveforms at 0.2 cyc/deg at 20 Hz, as illustrated in Figure 5A. For both the square and sawtooth waveforms, none of the three observers could discriminate the phases at 100% contrast. As with sinusoids, the gratings themselves were detected at 20 Hz with a low contrast detection threshold (square wave: 0.96% contrast; sawtooth: 1.55% contrast). Thus, the lack of phase discriminability is not restricted to a counterphase-modulated sinusoid.

If a loss of temporal phase discrimination is indeed responsible for the frequency-doubling illusion, under certain conditions an observer should be unable to distinguish between a counterphase-modulated sinusoid and reversal of a rectified



**FIGURE 6.** (A) Psychophysically derived foveal sensitivity functions for the grating phase-discrimination task at a range of temporal frequencies for the three observers identified at lower left. Where log contrast sensitivity became less than zero (indicated by arrows), observers were unable to perform the task. Individuals' detection sensitivities for the same conditions are shown on the same axes. (B) Data for the same task at 5°, 10°, and 20° eccentricities.

sinusoid at high stimulus temporal frequencies, as illustrated in Figure 5B. A linear ganglion cell's response amplitude for this waveform would go through a minimum near the minimum of the rectified sinusoid but not null out completely. The residual response would be dependent on receptive field center size and contrast. We calculated that in central retina ( $\pm 10^\circ$ ) a grating of 0.1 cyc/deg at 30% contrast would evoke a negligible

response in an MC cell positioned at the crossover (equivalent to <1% contrast for an optimally positioned sinusoid). Under these conditions, five observers compared the contrast-reversed rectified sine wave paired with the standard grating reversed stimulus. At low temporal frequencies, the gratings were clearly distinguishable. At 20 Hz, they appeared very similar.

### Possible Mechanisms for Loss of Phase Discrimination

These results show that observers are unable to distinguish the temporal phase of counterphase-modulated gratings at and above 20 Hz, although the modulation is easily detectable. A possible cause for this loss of phase sensitivity would be variation in response phase at the ganglion cell level. To analyze this further, we examined variation of response phase on a cycle-by-cycle basis for each cell. For peak responses (i.e., Fig. 1 spatial phases  $0^\circ$  and  $180^\circ$ ), the standard deviation of phase from cycle to cycle was  $32.3^\circ$  ( $n = 11$ ). Phase difference for the peak responses (at 0.2 cyc/deg) was  $176.1^\circ \pm 2.7^\circ$ ,  $n = 11$ . For a single sweep, based on responses of a single ganglion cell at each counterphase location, a central mechanism would reliably ( $P < 0.001$ ) distinguish the phase of counterphase modulation.

Thus, ganglion cell data do not support the hypothesis that the psychophysical loss in temporal phase sensitivity is due to phase variation among members of the ganglion cell array. An alternative hypothesis is that there is a loss of temporal phase sensitivity at a cortical site.

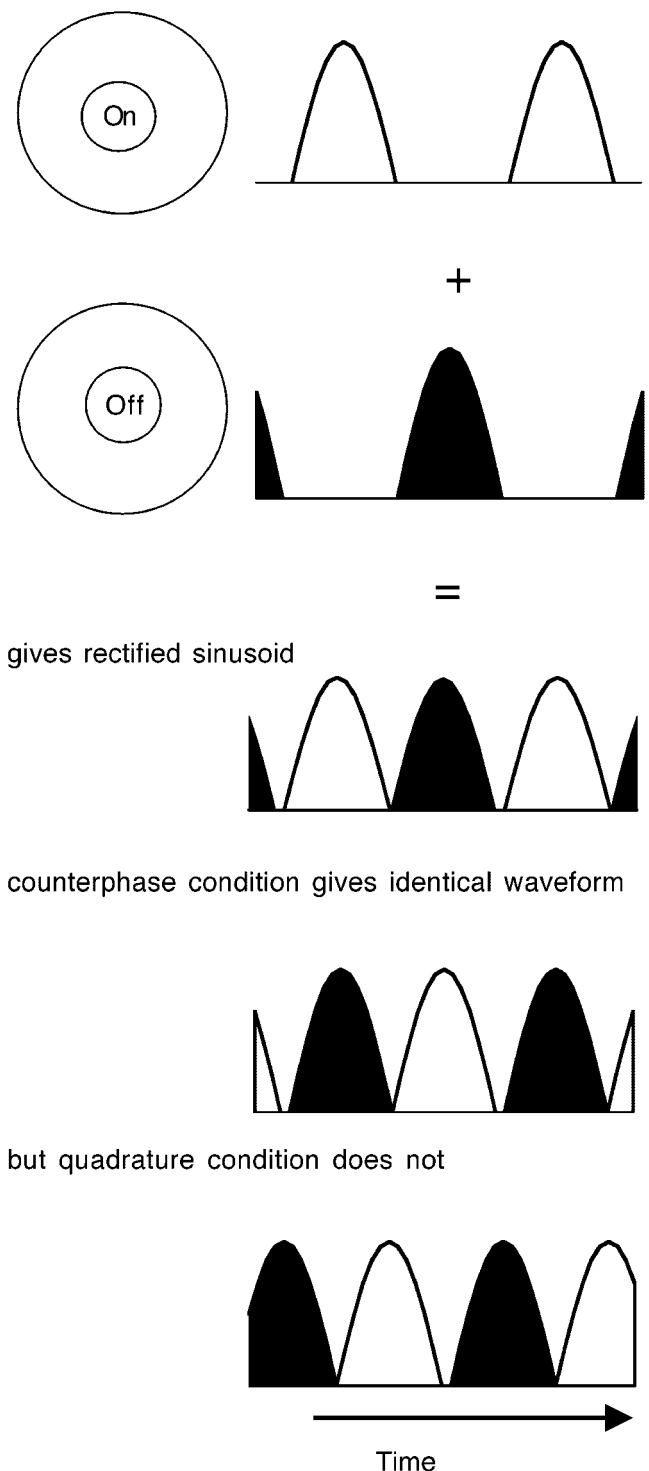
Other studies have suggested temporal rectification as the origin of the frequency-doubling illusion.<sup>3,20,21</sup> One mechanism by which this might occur is by summation at high temporal frequencies of on- and off-center cell signals in the cortical mechanism responsible for detecting high-frequency flicker. In the retina, two MC cell arrays are present, one each for on- and off-center ganglion cells. Summation at a cortical site of responses of these two cell types would be equivalent to full-wave rectification of the stimulus. This model would account for the psychophysical discrimination results described so far.

The hypothesis is shown in more detail in Figure 7. Signals from on- and off-center cells combine to give a full-wave rectified response. This would be indistinguishable from the counterphase condition. However the quadrature ( $90^\circ$ ) condition should be discriminable. To test this, observers were asked to perform the discrimination task for a pair of counterphase gratings with the temporal phase difference set at  $90^\circ$  instead of  $180^\circ$ . The two gratings should appear different if a straight-forward temporal rectification is taking place. None of the three observers could make the discrimination at 100% contrast, which suggests that the temporal rectification hypothesis does not account for the loss of phase discriminability. We propose that this evidence shows that the frequency-doubling illusion is due to a loss of phase sensitivity at a cortical locus. The reason for this loss remains uncertain.

### Spatial Cues from the Ganglion Cell Array

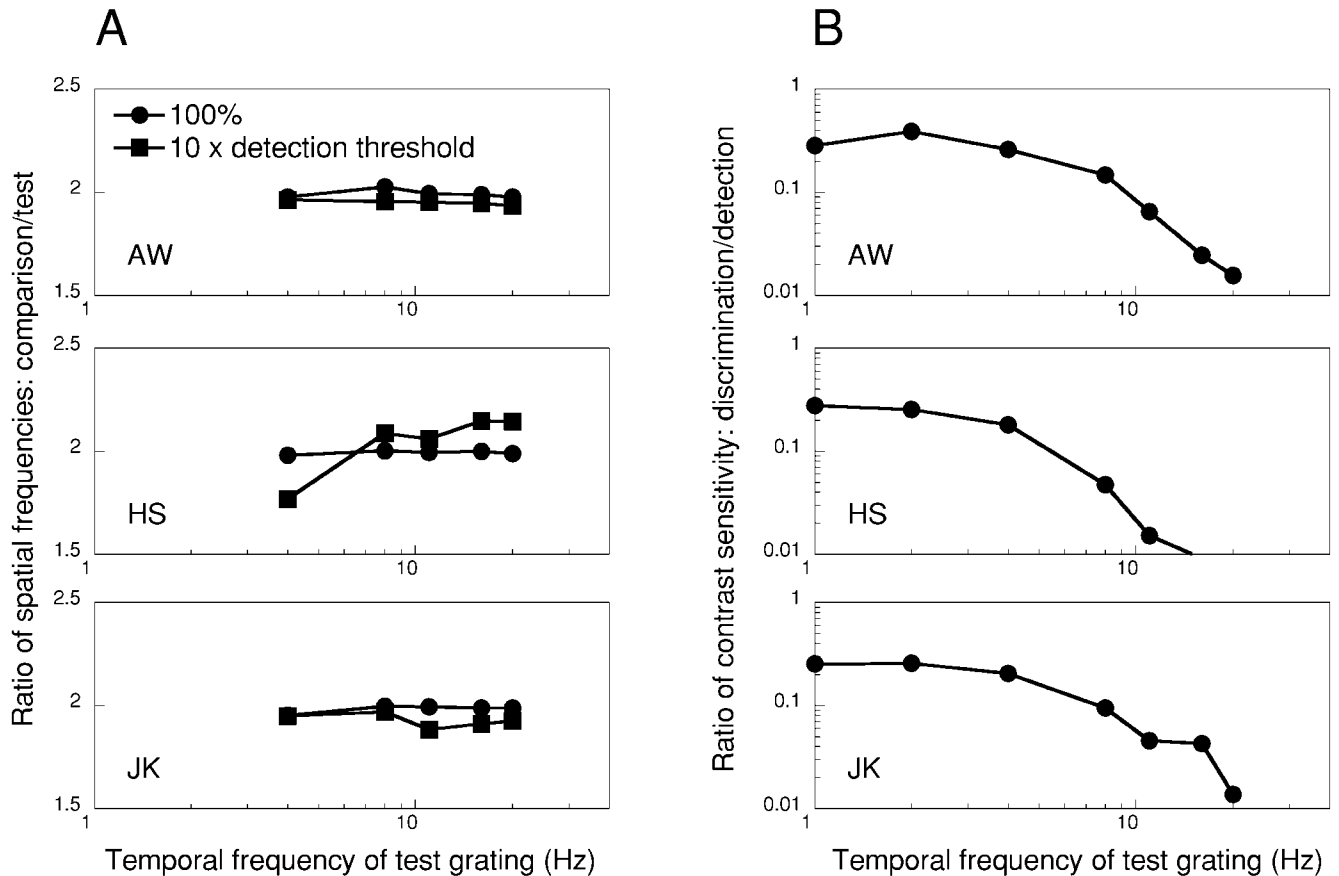
Although there is a loss of temporal phase information at high stimulus temporal frequencies, spatial cues are present at these frequencies. The most obvious spatial cue was the zero-crossings (regions of minimal flicker). To characterize this phenomenon quantitatively, subjects were asked to match these regions of minimal flicker to rectangular bars of adjustable distance and position at a variety of temporal frequencies. This test was performed at 100% contrast and a lower contrast (10 times detection threshold) for each observer. The test sinusoidal grating was 0.2 cyc/deg. The spatial frequency of the square-wave bars matched to the regions of minimal flicker was close to twice the spatial frequency of the test stimulus (Fig. 8). The spatial position of the bars that the observers set approximated the position of the zero-crossings of the sinusoidal grating (mean phase difference at 20 Hz,  $0.6^\circ \pm 5.5^\circ$  [SD],  $n = 3$ ). Even at lower temporal frequencies (4 Hz), at which it is possible to distinguish counterphase-modulated grating phase, it was still possible to match the zero-crossings (Fig. 8). This is

### Central summation of on and off-center signals -



**FIGURE 7.** Cortical rectification mechanism by which phase information might be lost. A combination of on- and off-center cell signals would yield a full-wave rectified signal indistinguishable from the temporal counterphase condition. However, a temporal quadrature stimulus should be distinguishable, which was not the case.

consistent with the spatial cue provided by the zero-crossings' acting as a reference for the perceived spatial frequency. For comparison, the ratio of detection to discrimination thresholds



**FIGURE 8.** (A) Spatial frequency of static bars matched to the regions of minimal flicker of a counterphase sinusoidal grating for the three observers identified at *lower left*. The task was performed at 100% contrast, and a contrast that was 10 times that of the counterphase grating detection threshold for each observer. The match occurred to twice the actual grating frequency, independent of temporal frequency down to 4 Hz, although within this frequency range phase discriminability changed drastically (B).

(Fig. 8B) is shown to change radically over this frequency range.

If the spatial information provided by the zero-crossings at high temporal frequencies defines perceived spatial frequency, a grating with fewer zero-crossings should not result in the frequency-doubling illusion, as suggested by Tyler.<sup>20</sup> A sawtooth waveform has one zero-crossing, and so the perception of frequency doubling should not occur. To characterize the spatial information seen at high temporal frequencies with this waveform, the zero-matching task was performed at 0.2 cyc/deg with 20-Hz flicker. The spatial frequency of the matched rectangular bars was approximately the same as the spatial frequency of the test grating (matched spatial frequency =  $0.2 \pm 0.004$  cyc/deg [SEM]).

To test the hypothesis that at high temporal frequencies, spatial information is preserved in the zero-crossings of counterphase-modulated sinusoidal gratings, the phase-discrimination task was repeated for two sinusoidal gratings. This time, the gratings were temporally in phase but spatially in quadrature—that is, offset by  $90^\circ$ . The hypothesis was that for 20-Hz counterphase gratings, if only spatial information were preserved in the zero-crossings then a spatial offset of the gratings would always be obvious. This was the case. The discrimination threshold was comparable to the detection threshold (discrimination threshold: 7.91% contrast, 0.7 log unit above detection threshold,  $n = 3$ ). This suggests that the absolute response amplitude to the counterphase flicker is available for

spatial discrimination; it is only temporal phase information that is unavailable.

## DISCUSSION

One of the critical assumptions made in the design of FDT was the presence of a significant subgroup of highly spatially nonlinear MC cells in primate retina.<sup>3,22</sup> However, the original description of spatially nonlinear MC cells<sup>13</sup> did not provide a distribution of nonlinearity indices. Results from prior and current physiological experiments are not consistent with the presence of a separate group of spatially nonlinear My ganglion cells. As with previous findings in the macaque LGN,<sup>11-14</sup> the distribution of MC cell nonlinearity indices is unimodal. Spatial nonlinearity is not found to the extent described in cat Y cells, in which the nonlinearity index can reach 10, under appropriate stimulus conditions.<sup>5</sup> Recent studies of retrogradely labeled macaque ganglion cells projecting to macaque LGN have also failed to isolate such a cell class (Dacey DM, personal communication, April 2001).

Second, if such a class of cells were to exist, it is unclear why they should dominate the percept of the counterphase grating while responses of other cells (e.g., linear MC cells) are perceptually ignored.

Last, we argue that any temporally frequency-doubled response of putative  $M_y$  cells must be spatially invariant and thus cannot be a substrate for the illusion. Other mechanisms for

the frequency-doubling illusion must be considered. Our findings are consistent with a cortical loss of temporal phase-discrimination ability. In a study of primary visual cortex, the temporal response was slower than in the retina or LGN,<sup>23</sup> but the investigators did not comment on the precision of the cells' response phase. The locus of the cortical loss of phase information thus remains uncertain.

A psychophysical examination of the temporal and spatial parameters of the frequency-doubling illusion, based on phase-discrimination ability, substantially conformed to the original description made by Kelly.<sup>1</sup> The rapid elevation in grating phase-discrimination threshold (Fig. 5) quantifies an abrupt onset of the frequency-doubling illusion over a restricted temporal frequency range, as in previous descriptions.<sup>1,24,25</sup> We suggest that the location of zero-crossings, separated by regions of perceived flicker, provides the spatial cue for the illusion when phase discrimination is not possible. By matching the zero-crossings to bars, it was possible to show that not only were they at twice the grating spatial frequency, but the bars did not appear to change in perceived spatial frequency or phase at temporal frequencies from 4 to 20 Hz. Over this same region of temporal frequencies, temporal phase discrimination changed from good (within 0.5 log unit of detection threshold) to poor (nonmeasurable, >2 log units above detection threshold). Investigators in several studies have reported that in this temporal frequency range the perceived spatial frequency of counterphased gratings is intermediate—between one and two times the fundamental.<sup>24–26</sup> Because we did not find evidence that the apparent spacing of the zero-crossings changes with temporal frequency, we suggest that the finding of intermediate apparent spatial frequencies may be due to the observer's being forced to reconcile conflicting cues. Parker,<sup>27</sup> based on an adaptation paradigm, also suggested that the illusion arises in processing by central mechanisms, rather than in the retina.

There has been a report<sup>28</sup> of a frequency-doubling illusion with chromatic gratings. This would be a puzzling finding if nonlinear ganglion cells gave rise to the percept, because PC-cells, for example, show linear summation properties.<sup>12,13</sup> However, loss of phase discrimination at a cortical locus might also occur with chromatic mechanisms and give rise to a similar illusion.

Cortical mechanisms can detect flicker up to at least 40 to 50 Hz.<sup>23</sup> However, in the discrimination tasks reported in the current study, phase information appeared to be lost at much lower frequencies. In contrast, with rapidly moving targets in a Vernier task, the temporal precision achieved by the observers is in the millisecond range, with rapidly drifting gratings. Mechler and Victor<sup>29</sup> recently showed that, in a Vernier task, it is the spatiotemporal context that enables such precise temporal assessment; temporal information alone is not sufficient. The current results fit with this suggestion.

The current commercially available clinical test uses a decrease in contrast sensitivity to detect visual field loss.<sup>2,17</sup> The clinical test does not depend on perception of the frequency-doubling illusion, per se.<sup>17,30</sup> Recent evidence suggests that the mechanisms underlying the illusion resemble to some extent those for detection of full-field flicker, which appears to be accomplished through the MC pathway.<sup>30</sup> Thus, the test is most likely a probe of contrast sensitivity of the MC pathway.

In summary, the present study builds on previous physiological studies in primate LGN to show that, as in LGN, spatial nonlinearity is found to a very limited extent in the primate retina. Unlike Y-cells in the cat, which form a physiologically and anatomically separate population of ganglion cells, spatially nonlinear MC cells in primate are the extremes of a

unimodal distribution of spatial nonlinearity. Furthermore, we have shown that spatially nonlinear cells are unlikely to be the origin of the frequency-doubling illusion. Instead, we postulate an alternate model for the illusion, which may occur because of a reduced cortical sensitivity to the temporal phase of achromatic counterphased gratings. This has important implications for the design of future clinical screening tests for glaucoma.

## References

1. Kelly DH. Frequency doubling in visual responses. *J Opt Soc Am*. 1966;56:1628–1633.
2. Maddess T, Goldberg I, Dobinson J, Wine S, Welsh AH, James AC. Testing for glaucoma with the spatial frequency doubling illusion. *Vision Res*. 1999;39:4258–4273.
3. Maddess T, Henry GH. Performance of nonlinear visual units in ocular hypertension and glaucoma. *Clin Vis Sci*. 1992;7:371–383.
4. Enroth-Cugell C, Robson JG. The contrast sensitivity of retinal ganglion cells of the cat. *J Physiol*. 1966;187:517–552.
5. Hochstein S, Shapley RM. Quantitative analysis of retinal ganglion cell classifications. *J Physiol*. 1976;262:237–264.
6. Hochstein S, Shapley RM. Linear and nonlinear spatial subunits in Y cat retinal ganglion cells. *J Physiol*. 1976;262:265–284.
7. Shapley R, So YT. Is there an effect of monocular deprivation on the proportions of X and Y cells in the cat lateral geniculate nucleus? *Exp Brain Res*. 1980;39:41–48.
8. So YT, Shapley R. Spatial properties of X and Y cells in the lateral geniculate nucleus of the cat and conduction velocities of their inputs. *Exp Brain Res*. 1979;36:533–550.
9. Sur M, Sherman SM. Linear and nonlinear W-cells in C-laminae of the cat's lateral geniculate nucleus. *J Neurophysiol*. 1982;47:869–884.
10. Victor JD, Shapley RM. Receptive field mechanisms of Cat X and Y retinal ganglion cells. *J Gen Physiol*. 1979;74:275–298.
11. Blakemore C, Vital-Durand F. Organization and post-natal development of the monkey's lateral geniculate nucleus. *J Physiol*. 1986;380:453–492.
12. Derrington AM, Lennie P. Spatial and temporal contrast sensitivities of neurones in lateral geniculate nucleus of macaque. *J Physiol*. 1984;357:219–240.
13. Kaplan E, Shapley RM. X and Y cells in the lateral geniculate nucleus of the macaque monkeys. *J Physiol*. 1982;330:125–143.
14. Levitt JB, Schumer RA, Sherman SM, Spear PD, Movshon JA. Visual response properties of neurons in the LGN of normally reared and visually deprived macaque monkeys. *J Neurophysiol*. 2001;85:2111–2129.
15. Lee BB, Martin PR, Valberg A. Nonlinear summation of M- and L-cone inputs to phasic retinal ganglion cells of the macaque. *J Neurosci*. 1989;9:1433–1442.
16. Crook JM, Lange-Malecki B, Lee BB, Valberg A. Visual resolution of macaque retinal ganglion cells. *J Physiol*. 1988;396:205–224.
17. Johnson CA, Samuels SJ. Screening for glaucomatous visual field loss with frequency-doubling perimetry. *Invest Ophthalmol Vis Sci*. 1997;38:413–425.
18. Silveira LCL, Perry VH. The topography of magnocellular projecting ganglion cells (M-ganglion cells) in the primate retina. *Neuroscience*. 1991;40:217–237.
19. Zar JH. *Biostatistical Analysis*. Englewood Cliffs, NJ: Prentice Hall; 1999.
20. Tyler CW. Observations on spatial-frequency doubling. *Perception*. 1974;3:81–86.
21. Kelly DH. Nonlinear visual responses to flickering sinusoidal gratings. *J Opt Soc Am A*. 1981;71:1051–1055.
22. Maddess T, Hemmi JM, James AC. Evidence for spatial aliasing effects in the Y-like cells of the magnocellular visual pathway. *Vision Res*. 1998;38:1843–1859.
23. Hawken MJ, Shapley RM, Grosf DH. Temporal-frequency selectivity in monkey visual cortex. *Vis Neurosci*. 1996;13:477–492.

24. Parker A. The effects of temporal modulation on the perceived spatial structure of sine-wave gratings. *Perception*. 1983;12:663-682.
25. Richards W, Felton TB. Spatial frequency doubling: retinal or central? *Vision Res*. 1973;13:2129-2137.
26. Maddess T, Kulikowski JJ. Apparent fineness of stationary compound gratings. *Vision Res*. 1999;39:3404-3416.
27. Parker A. Shifts in perceived periodicity induced by temporal modulation and their influence on the spatial frequency tuning of two aftereffects. *Vision Res*. 1981;21:1739-1747.
28. Dobkins KR, Bosworth CF, Sample PA. Form: detection (F:D) contrast threshold ratios are elevated for luminance, with respect to chromatic stimuli. *Invest Ophthalmol Vis Sci*. 1999;40(4):S355. Abstract nr 1886.
29. Mechler F, Victor JD. Comparison of thresholds for high-speed drifting vernier and a matched temporal phase-discrimination task. *Vision Res*. 2000;40:1839-1855.
30. Anderson AJ, Johnson CA. Mechanisms isolated by frequency-doubling technology perimetry. *Invest Ophthalmol Vis Sci*. 2002;43:398-401.