Visible flicker from invisible patterns

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USING a laser interferometer we can create grating patterns of high optical contrast (interference fringes) directly on the retina[1–3]. With coarse fringe patterns, the alternating light and dark bars of the pattern can be seen, but the bars of the finest fringes are not subjectively resolved. We report here that when we rapidly modulate the contrast of a fine fringe pattern (keeping overall luminance constant), observers experience flicker, even if the fringes are too finely spaced to be perceived as a grating. For this flicker to be seen, the pattern needs to be resolvable by the photoreceptors themselves, but not necessarily by later stages of visual processing. It can be explained if, in man, signals associated with individual cone receptors do not depend linearly on light intensity, but instead are scaled by a fast sensitivity-regulating or light-adaptation mechanism. Contrast-modulation flicker is not demonstrable in rod vision; rod vision therefore lacks such a local adaptation process.

Subjects in our experiments viewed an interference fringe of time-varying contrast (Fig. 1, top panels). Generally, the spatial frequency of the fringe was so high that the stripes could not be resolved by the subject and the field looked uniform (except for laser speckle). The contrast fluctuated sinusoidally between a peak value (generally unity) and zero. Our subjects saw flicker, as if the overall luminance of the field was changing, even though the total amount of light in the test field remained constant during the modulation (Fig. 1, lower left panels). This contrast-modulation flicker arises not simply from detection of the local changes of intensity that occur within the test field, but from a reduction in the perceived overall brightness of the field when the contrast is increased: we could cancel, or null the flicker by modulating the overall physical luminance of the field in synchrony with the modulation of contrast (Fig. 1, lower right). At a suitably adjusted amplitude of this nulling luminance modulation, the perceived flicker was minimized and the field appeared almost steady as well as uniform.

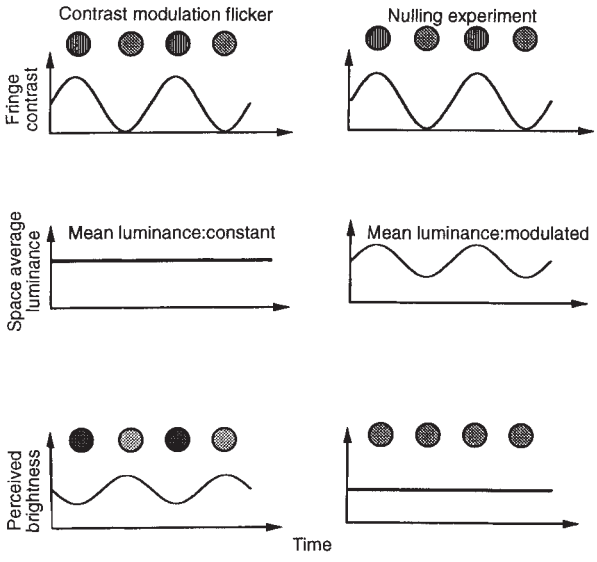

FIG. 1 The stimulus used to generate contrast-modulation flicker (left panel), and the stimulus used to null or cancel it (right panel). Fringe contrast (top left) varied sinusoidally between a peak value (generally unity) and zero. If luminance was constant during the modulation of contrast (middle left), flicker could be seen. The observer then adjusted the amplitude and phase of the nulling luminance modulation (middle right) in order to minimize the perceived flicker. It proved helpful to superimpose on the depicted stimulus a fixed 'pedestal' luminance modulation. In alternating 1-s intervals, distinguished by computer-generated beeps, this pedestal was arranged to alternately cancel or reinforce the contrast-modulation flicker. Any failure to null the contrast-modulation flicker revealed itself as a difference in the resultant flicker amplitude between these two intervals. The test field was a disc subtending 50′ of visual angle, illuminated with 2,000 trolands (td) of red (633 nm) light from a He–Ne laser and enclosed in a uniform equiluminous surround. It was centrally fixated, and so fell within the central fovea where the photoreceptors are small enough to preserve high optical contrast for fringes as fine as 100 cycles per degree, far above the resolution limit[2].

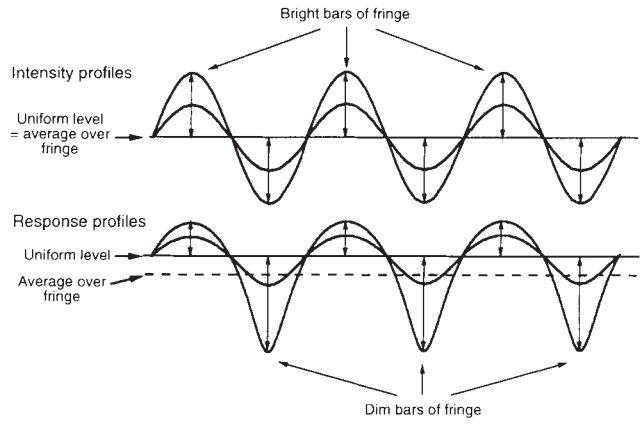

FIG. 2 Top, each curve is a cross-section of the light intensity profile over the test field, at some moment during the modulation of fringe contrast. Horizontal line is when fringe contrast is zero (uniform field); other curves show fringe at intermediate or at peak contrast. Bottom, corresponding response profiles if the relation between response and intensity is compressively nonlinear. The average response with the fringe at peak contrast falls below the response to the uniform field.

Contrast-modulation flicker can be explained if each cone photoreceptor's response depends in a nonlinear way on light intensity[4], and if later neurons sum these nonlinear signals over their receptive fields to determine the perceived overall luminance of the unresolved fringe. When the interference pattern, having reached peak contrast, is then succeeded by the physically uniform phase of the test field, the luminance of the light bars goes down by as much as the luminance of the dark bars goes up. This keeps the overall space-average luminance constant (Fig. 2, upper curves) and, if each photoreceptor's signal were a fixed linear function of local intensity, the average of the receptor signals would remain constant as well. But if each cone receptor's response is a compressively nonlinear function of intensity (or equivalently, if individual cone signals are adaptively scaled by a sensitivity-regulating process), then the response profiles will be asymmetrically distorted as shown by the lower curves in Fig. 2, and the decrement in response in the previously bright bars will be less than the increment in the previously dark bars. So the space-averaged response will be greater during the uniform phase (Fig. 2, continuous horizontal line) than it was for the fringe (dashed line).

In our nulling experiment, this difference is compensated by a visually equivalent increase in overall physical luminance when the fringe is near peak contrast. The nulling luminance variation was more than a factor of two between maximum and minimum under some conditions, but it depended strongly on fringe peak contrast: halving the fringe contrast reduced the equivalent luminance excursion about fourfold. This roughly quadratic dependence on fringe contrast is theoretically expected if the cone output is a smoothly compressive nonlinear function of intensity, with a large quadratic term in its power series expansion.

The responsible nonlinear process must be at a stage within the visual system where the fringe can be resolved. If it takes place within the photoreceptor cells, contrast-modulation flicker should require only that the fringe be present with high contrast at the photoreceptor level. Whether it is resolved perceptually, or by later neural stages of the visual system, would then be irrelevant: the flicker amplitude should depend entirely on the fringe contrast as seen by the cones themselves, which is limited mainly by the size of the individual cone receptors. Figure 3 supports this prediction. The finer the fringe, the less the flicker (Fig. 3*a*); but the equivalent amplitude decreases only very slowly as the fringe spatial frequency increases past the visual resolution limit (here just over 60 cycles per degree of visual angle). Contrast-modulation flicker is measurable even above 100 cycles per degree. Evidently the responsible nonlinear elements resolve optical detail much better than does the entire visual system of the human observer.

Figure 3*b* shows just how little spatial integration of light intensity at the input to the nonlinear elements is implied by the results of Fig. 3*a*. The gaussian curve is the point spread function corresponding to the curve of Fig. 3*a*. Evidently the nonlinearity is preceded by spatial integration of light intensity over a region spanning only 18 seconds of arc, or 1.5 μm on the retina (full width at half height). This is equal to the smallest histological estimate of the diameter of the light-collecting inner segment of the central foveal cones[5,6], and provides a functional estimate of the cone's light-collecting area that is in rough agreement with other recent evidence[2]. This leaves no margin for any neural spatial integration. The nonlinear process is therefore local and early: either it is inside the cones themselves, or otherwise it operates independently on the signals from each cone, before further spatial integration obliterates the representation of the finest fringes at later neural stages and in perception.

We have investigated the dynamic aspect of this early visual nonlinearity by varying the modulation frequency (the frequency at which the fringe appears and disappears). The equivalent (nulling) amplitude remains substantial up to 30 Hz (Fig. 4,

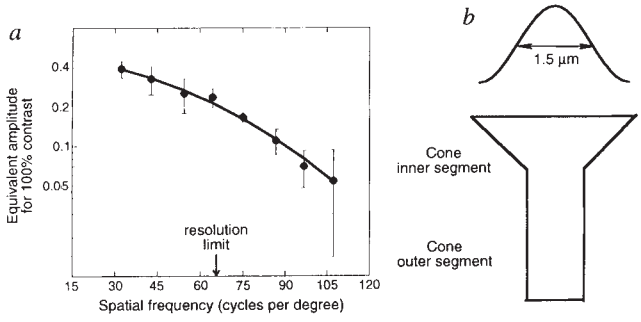

FIG. 3 *a*, Filled circles, contrast-modulation flicker as a function of fringe spatial frequency for test fields fluctuating sinusoidally at 15 Hz between unity contrast and zero contrast. The equivalent (nulling) luminance modulation approaches 40% (a sinusoidal modulation of overall luminance between 1,200 and 2,800 trolands) at low fringe frequencies, and decreases only slowly with increasing frequency, remaining measurable at 100 cycles per degree of visual angle. Error bars are the 95% confidence intervals, based on variation among the mean settings from four different sessions. Curve is best-fitting gaussian. Two other subjects (D.I.A.M. and one other subject naive as to the purpose of the experiment) were tested; they yielded similar results. The indicated visual resolution limit is the highest fringe frequency at which a two-alternative forced-choice discrimination between horizontal and vertical fringes of unity contrast could be made with 75% reliability. *b*, point spread function for light intensity at the input to the nonlinear element, compared with the dimensions of a central foveal cone photoreceptor[6]. Effective contrast at the input to the nonlinear elements was assumed proportional to the square root of the equivalent amplitude from *a*; the corresponding radially symmetrical point spread function shown here is gaussian in spatial coordinates, and is given by the Fourier transform of the function relating contrast to spatial frequency.

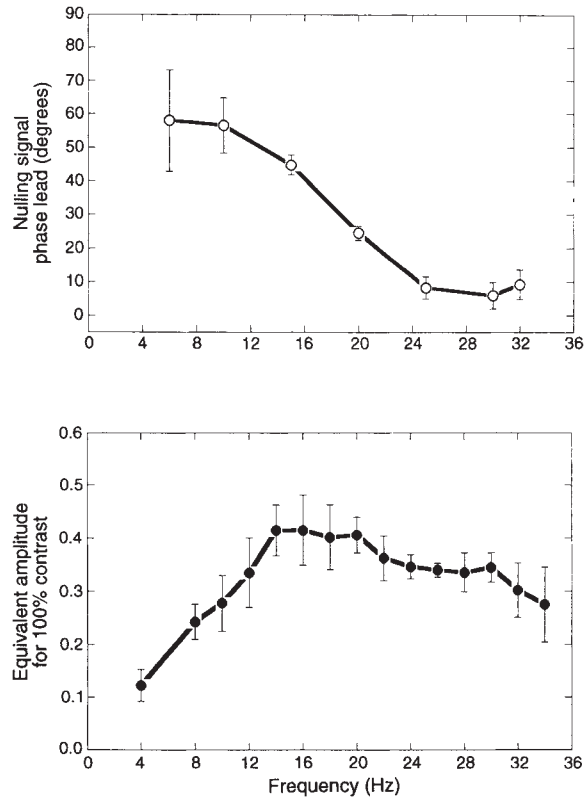

FIG. 4 Open circles, phase of luminance modulation optimally effective in perceptually cancelling contrast-modulation flicker, for a range of temporal frequencies. Spatial frequency 30 cycles per degree. Positive phase values mean the luminance peak had to precede the contrast peak. Error bars are the 95% confidence intervals, based on variation among the mean settings from four different sessions. Results for the two other subjects were similar. Filled circles, amplitude of luminance modulation required to cancel contrast-modulation flicker at different modulation frequencies.

filled circles). At the lower frequencies, the luminance modulation for an optimal null at high peak contrast has to be considerably advanced in phase relative to the modulation of contrast (empty circles, Fig. 4). This excludes instantaneous compression (with a fixed relationship between input intensity and output, as assumed in Fig. 2) as a model for the nonlinear process, because that model predicts no phase mismatch[4]; it also excludes models in which time-average light exposure speeds the visual response as well as reducing its sensitivity[7,8], because these predict a nulling signal lag, not a lead[9]. But the phase lead is expected if a relatively fast sensitivity-regulating process scales sensitivity roughly in inverse proportion to the recent input level for each cone. Its disappearance at 25–30 Hz is expected if the adaptation process fails to follow the input closely at these high frequencies[9].

Our results suggest that a major component of human photopic (cone) visual adaptation to light is very fast and strictly local. A strictly local process could reside either within the cones themselves or in later neural structures (such as the cone-bipolar synapse) that are fed by single cones. We find (in preparation) that contrast-modulation flicker is visible down to low photopic light levels, below 1,000 quanta per cone per second. This intensity range is comparable with the range of adaptation in monkey gross receptor potentials (isolated with aspartate in the monkey foveal electroretinogram[4]), but extends well below the range reported for cone outer segment photocurrent[10]. The situation for rod vision is quite different: contrast-modulation flicker in the rod system (measured 15 degrees extrafoveally with 515 nm laser light on a deep red steady background) is absent or minimal. This supports other evidence[2,11–14] that in human

vision the cone system, but not the rod system, has substantial receptor-specific sensitivity regulation. □

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Proliferation of oligodendrocyte precursor cells depends on electrical activity in axons

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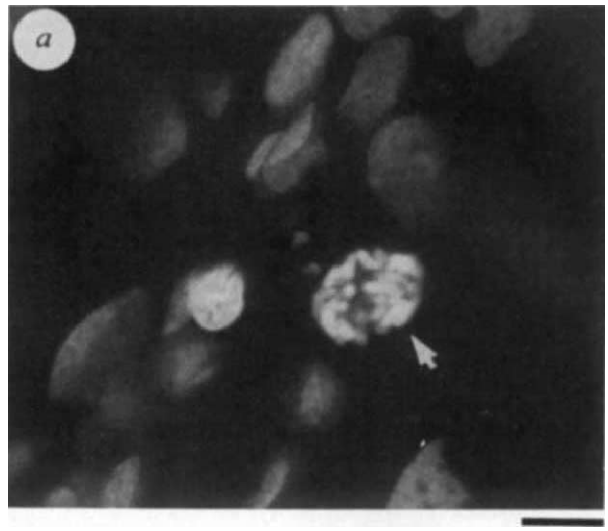
OLIGODENDROCYTES myelinate axons in the vertebrate central nervous system. It would, therefore, make sense if axons played a part in controlling the number of oligodendrocytes that develop in a myelinated tract. Although oligodendrocytes themselves normally do not divide, the precursor cells that give rise to them do. Here we show that the proliferation of oligodendrocyte precursor cells in the developing rat optic nerve depends on electrical activity in neighbouring axons, and that this activity-dependence can be circumvented by experimentally increasing the concentration of platelet-derived growth factor, which is present in the optic nerve and stimulates these cells to proliferate in culture. These findings suggest that axonal electrical activity normally controls the production and/or release of the growth factors that are responsible for proliferation of oligodendrocyte precursor cells and thereby helps to control the number of oligodendrocytes that develop in the region.

To examine the role of axons in the proliferation of oligodendrocyte precursors, we transected one of the optic nerves in postnatal day 8 (P8) rats. After 4 days, the average number of mitotic figures per section was 90% lower in transected nerves than in the uncut control nerves (Fig. 1*a*, *b*). To test the possibility that the axons normally display a glial mitogen on their surface (as has been suggested for axons in the peripheral nervous system (PNS)[1]), which is lost when the axons degenerate and are phagocytosed after transection, we studied the effect of



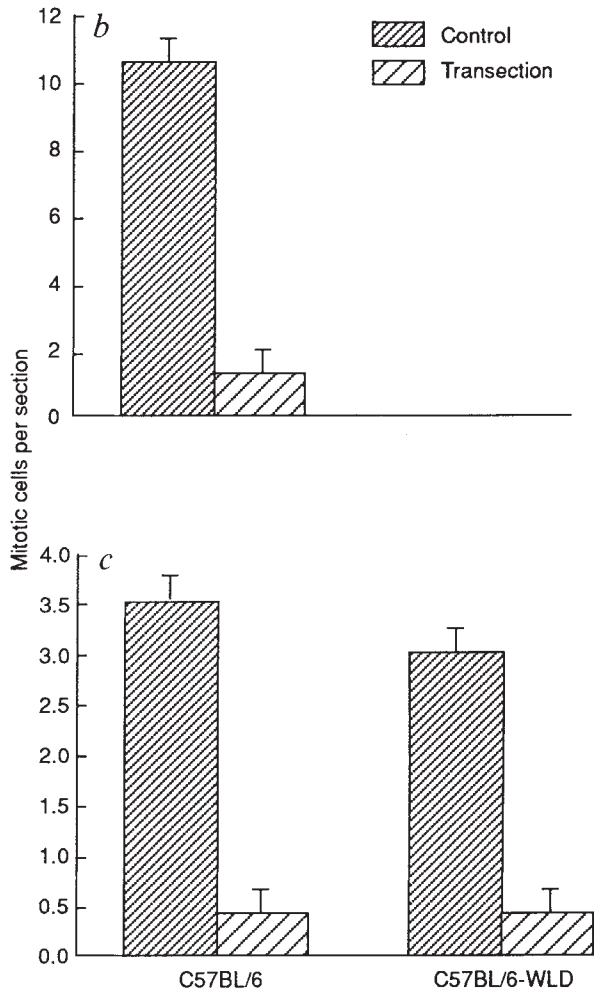


FIG. 1 The effect of nerve transection on cell proliferation in the developing optic nerve in rats (*b*) and in C57B1/6 and C57B1/6-WLD mice (*c*). A typical mitotic figure in a P12 optic nerve section stained with the nuclear counterstain, propidium iodide, is shown in *a*.
METHODS. P8 (*a*, *b*) rats or P12 (*c*) mice were anaesthetized with ether and the right optic nerve was cut just behind the eyeball. Four days later, the rats were reanaesthetized with ether and perfused with 4% paraformaldehyde, as previously described[7]. The optic nerves were incubated in 4% paraformaldehyde at 4 °C overnight, transferred to 30% sucrose in PBS until equilibrated, frozen in OCT compound (Miles) and cut into 8 μm longitudinal sections, which extended the entire length of the nerve, with a Bright cryostat. The sections were collected onto gelatinized glass microscope slides, air dried, post-fixed in 70% ethanol for 10 min at −20 °C, and stained with propidium iodide (4 μg ml^{-1}) in minimal Eagle's medium buffered with HEPES (MEM/HEPES) and containing DNase-free RNase A (100 μg ml^{-1}) for 30 min at 37 °C[7]. The slides were washed three times in PBS and mounted in Citifluor. The number of mitotic figures per section was determined by averaging the number of mitotic figures counted in five optic nerve sections per animal. In this and the following figures, the results obtained in four animals are shown as means ± s.e.m. The section shown in *a* was examined with a MRC-600 laser-scanning confocal imaging system in conjunction with a Nikon Optophot microscope. Scale bar, 8 μm.