13.1 g of ethanol per kilogram of body weight, with a standard deviation of 1.29. The percentage of the total caloric intake which was ingested as ethanol was 44.8 percent. The mean body weight of the group was 308.2 g, which was not significantly different from the free-feeding starting weight of 315.9 g.

Three to four weeks from the end of the experiment, we selected four animals for observation during alcohol withdrawal. They were removed from the experimental cages at 7 a.m., and were placed in individual observation cages with water, but no alcohol, available. Food rations, in amounts equivalent to those defined by the experiment, were given at the appropriate times. Within 3 to 4 hours after the last feeding period (5 to 6 a.m.), when most of their last draughts of ethanol presumably occurred, the animals became hyperactive. A shaking of keys near the top of the cage for 1 to 2 seconds resulted in a tonic-clonic convulsion in rat No. 8. For the next hour, tremors, spasticity, and clonic head movements occurred, and finally, a second seizure ended in death. When keys were shaken (2 to 5 seconds) for the first time after 9½ hours of withdrawal, a clonic running episode was produced in rat No. 2, followed shortly by death from a tonic-clonic seizure. Rat No. 7 showed all the preconvulsive symptoms, but keys shaken (up to 20 seconds) after 15 hours of withdrawal had no effect. Rat No. 1 was similar, but no attempt was made to trigger a convulsion by shaking keys. Attempts to produce convulsions in normal Holtzman rats by prolonged key shaking were unsuccessful, and no preconvulsive, hyperactive behavior was observed.

We are unaware of any previous report demonstrating physical dependence on ethanol in the rat as indicated by withdrawal convulsions, although this has been obtained in other species, as well as in man (*9*). We could also find no previous report of the development of physical dependence by self-administration in animals other than man when ethanol in water was available as the sole drinking solution.

Other methods for the production of ethanol dependence in animals have involved administration by intravenous and intragastric routes, by liquid diets, and by inhalation (*9, 10*). While useful, these methods have certain disadvantages; they are more removed from a model of alcoholism involving oral self-administration of an aqueous ethanol solution than is the method reported here. In animals consuming liquid diets in which 35 to 36 percent of the calories are derived from alcohol, the concentrations of ethanol in the blood appear to be rather low (*10*) unless the animals suffer considerable concomitant weight loss with presumed changes in their capacity to metabolize ethanol (*6*). The present method maintains high concentrations in the blood without significant body weight loss.

The percentage of total caloric intake derived from ethanol is low in most experiments utilizing ethanol in the drinking water. One study reports a caloric intake from ethanol as high as 30 percent (*11*). Attempts to incorporate 45 percent of the calories as ethanol into a liquid diet resulted in death of the animals (*12*). The 44.8 percent caloric intake in the present experiment compares favorably with this picture, and with the amount selected by human alcoholics (*13*).

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Chromatic Specificity of the Visual Evoked Response

Abstract. *The temporal alternation of red and green stripes in a structured field produces successive contrast, which can elicit cortical potentials recorded from the scalp. Amplitudes of the major frequency components of the potentials correspond to the relative intensities of red and green producing the contrast. The amplitude variations are color-specific, since total luminance and structure are held constant.*

In studies of cortical processing of color in man, the stimulus procedure used to elicit the visual evoked response (VER) has been the spatial and temporal alternation of two colors presented in a striped or checkerboard field (*1, 2*). The major advantage of the alternation procedure is that chromaticity can be varied independently of luminance, which remains constant. An assumption has been that contribution of the two colors to the response is equal or constant. For example, red and green alternated 180° out of phase (that is, the colors exchanging places at a given frequency) produce a presumably comparable response to each alternation regardless of direction of color change. Thus, the response is a difference measure between the two colors, and a response to a given color per se is not available.

Our purpose here is to demonstrate that with temporal alternation alone (successive contrast) in a structured field, responses to red-to-green alternations are different from those to green-to-red alternations. This previously unreported effect (*2, 3*) represents a fundamental difference between the red and the green processing channels. This difference is apparent because their respective contributions to the response are not equal, and thus a degree of chromatic specificity is obtained. We also show that VER differences between the red and green channels are independent of luminance.

Color stimuli were produced by narrow-band interference filters (Baird-Atomic) in a Maxwellian view system. For monocular stimulation, two channels presented a field of 20° to the subject's right eye, with central fixation aided by a bite bar. Two identical channels for the left eye were available for binocular stimulation. Within the field, subjects with normal color vision saw

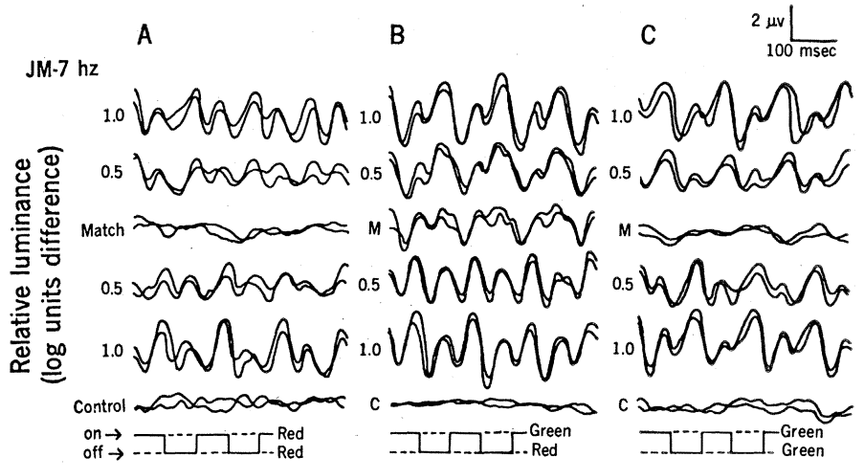

Fig. 1. Monocular VER's to alternating chromatic stimuli. Two separate trials of 120 summations each are superimposed for each trace. For control trials, light sources were occluded; *M*, match point. (A) Both stimuli in the alternation are 630 nm. (B) The first stimulus is 520 nm, and the second is 630 nm. Green is relatively brighter in traces above the match, and red is relatively brighter in traces below. (C) Both stimuli are 520 nm.

stationary vertical black stripes subtending 30′ of visual angle, with an equal number of interspersed 30′ stripes of red [peak wavelength (max λ), 630 ± 2.5 nm bandwidth at 50 percent]. Each 71.43 msec the red stripes were instantaneously replaced by green (max λ, 520 ± 2.5 nm bandwidth at 50 percent) stripes. The alternating stimuli were produced by out-of-phase 7-hz square-wave modulation of two glow modulator tubes through Ronchi rulings aligned in each channel. The alternations produced no real or apparent movement.

The successive contrast produced by heterochromatic alternation of the red and green typically gives the asymmetrical VER wave train shown in Fig. 1B at the match point. The red-to-green alternations differ from the green-to-red alternations, and we obtained similar asymmetries again and again despite varying our matching procedure and careful photocell monitoring to preclude gaps or overlaps in the alternations.

To more clearly establish the effect as chromatic and to rule out luminance or other artifacts, we unbalanced the match in four steps of 0.25 log unit in each direction, by counterrotating neutral density filters, for a total luminance difference between the stimuli of 1.0 log unit (+0.5 log unit to one color and −0.5 from the other); overall luminance was kept constant. Unbalancing that favored the green (Fig. 1B, top) produced VER trains different in shape from those produced when red was favored (Fig. 1B, bottom).

The procedures used for red-green alternation were then repeated with red-red alternation and green-green alternation. With monochromatic alternation, no VER should be elicited at the match point if matching is adequate, because there is no chromatic interaction. There is little or no VER when matched red stimuli (Fig. 1A) or matched green stimuli (Fig. 1C) are alternated (*4*). The unbalancing, or luminance per se, appears to affect only the magnitude of the patently different response trains produced by red alternations (Fig. 1A) or green alternations (Fig. 1C). Further, the VER responses obtained with red and green monochromatic alternations are comparable to those produced by unbalancing the heterochromatic alternations to favor red or green, respectively.

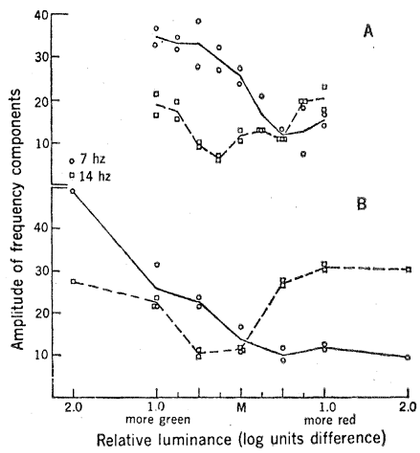

Fig. 2. Amplitudes of major frequency components given by Fourier analysis. (A) Data are given for session with psychophysical match as midpoint on abscissa. (B) Data are given for subsequent session with minimum response as midpoint.

Visual inspection suggested that the VER wave shape differences resulted from variations in the response frequency components. Fourier analysis by the fast Fourier transform was performed on the digitized VER's to give the amplitude and phase of the frequency components of the VER. As expected from the visual inspection, the frequencies at 7 and 14 hz were the most significant components, and varied systematically from one stimulus condition to another. Figure 2A gives amplitudes of the 7- and 14-hz components for the analog VER's in Fig. 1B. With the green stimulus much brighter than the red, the 7-hz amplitude is greater than the 14-hz amplitude, and this difference reverses as the red is made brighter than the green. In an attempt to see whether the reversal point could be obtained closer to the match, the session was repeated with a "minimum response" criterion (*5*) for the VER and with the total density difference extended to 2.0 log units. Activity of the two major components was essentially replicated (Fig. 2B), but the reversal point occurred at the electrophysiological match.

The same dominant frequency components (7 hz with chromatic contrast favoring green and 14 hz with red favored) were found with flicker square-wave "on-off" stimulation when the red or green channel was occluded. Thus, the frequency specificity is not restricted to steady-state stimulation and appears to be a fundamental difference in cortical processing of red and green. Additional support for this conclusion was found when we stimulated binocularly and dichoptically. Binocular red stimulation gave a predominant 14-hz component, and binocular green gave a predominant 7-hz component, while for dichoptic stimulation with the red and green 180° out of phase, the 7- and 14-hz components were similar in amplitude.

The most probable interpretation is that the red and green processing channels differ in temporal characteristics (different "on" or "off" responses, or differences in both), although direct examination is not possible with these data (*6*). It is known that photopic spectral sensitivity varies with stimulus duration (*7*), and that the phase of one color must be shifted to eliminate residual flicker in heterochromatic sinusoidal alternation (*8*). Our Fourier analyses did show large phase shifts

and reversals of the major components in the data, but direct manipulation of stimulus phase is necessary for adequate interpretation. It has often been suggested, in accounting for phenomena such as Benham's Top, that the red system is somehow "faster" than the green system.

This suggestion could account for the chromatic effect reported here. Consider the case where a red stripe alone is presented as a square wave at 7 hz. The "faster" red response could allow the system to respond maximally to the beginning and ending of each red presentation, and indeed in this condition we get a train of 14 waves comparable in amplitude, and consequently a large 14-hz component. Conversely, when a green stripe alone is presented, the "slower" green response is not yet completed when the stimulus ends; this could result in a smaller response to the following absence of green, which in turn would give the obtained large 7-hz component.

Such an interpretation is tentative beyond the obvious reasons, since it is restricted to the stimulation frequency used. Psychophysical research offering support for a temporal interpretation also suggests that the effect would vary with the frequency of stimulation (*9*). It has also been reported that with nonalternating stimuli, amplitude of the fundamental frequency component of the VER varies with wavelength of the stimulus and of the surround field (*10*).

Regardless of the ultimate efficacy of our (or any) temporal interpretation, the use of temporal chromatic alternation (successive contrast) allows the study of a given chromatic VER response almost per se. This strong conclusion is based on the large and consistent differences found between red-to-green and green-to-red interactions, and is presumed to reflect different red and green processing systems with different properties.

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3. Riggs and Sternheim (*2*) stated (p. 637), "Furthermore, there are changes of phase and shape in addition to the measured changes of amplitude, of waves resulting from various wavelength pairs." These changes were not shown or elaborated upon further, but they probably were not similar to the effect we report here (L. A. Riggs, personal communication). In our method we used only temporal modulation of the stimuli (successive contrast) rather than the temporal and spatial modulation (successive and simultaneous contrast) used by others (*1*). In fact, we assume that our use of only successive contrast allowed the effect to be seen so clearly and consistently. Riggs and Sternheim (*2*) also stated that they obtained more consistent VER's by placing stationary black stripes in front of the boundaries between the alternating color stripes. This, in effect, reduced simultaneous contrast.
4. The data presented in Figs. 1 and 2 are from one subject and were replicated in additional sessions. The other subject's data were similar in all significant aspects.
5. Different methods can yield different heterochromatic "matches"; for example, see R. M. Boynton and P. K. Kaiser [*Science* **161**, 366 (1968)]. In addition, heterochromatic flicker photometry is difficult for the subject at the relatively low rate of 7 → J. B. Siegfried, D. I. Tepas, H. G. Sperling, and R. H. Hiss [*ibid.* **149**, 321 (1965)] found the VER minima at the heterochromatic match point, but D. Regan [*Vision Res.* **10**, 163 (1970)] subsequently found the VER minima displaced off the match point. Our results correspond more closely to Regan's, perhaps because we also examined the VER frequency components rather than amplitude of the gross waveform. We could shift the point where the two fundamental VER frequencies were approximately equal (for example, Fig. 2, A and B) and obtain even smaller VER's with monochromatic alternation (for example, *M* condition in Fig. 1, A and C) by varying luminance of either color channel slightly (< 0.1 log unit). This extreme VER sensitivity to both luminous and chromatic contrast suggests that a "minimum response" criterion would be appropriate. Average retinal illuminance of the stripes was 56 trolands, measured by the method described by G. Westheimer [*ibid.* **6**, 669 (1966)].
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Pumps or No Pumps

The article by Culliton (*1*) would lead one to believe that active transport occurs in all living cells and that the membrane is the rate-limiting factor for the entrance of ions and other solutes into the living cells. The definition of active transport is met when it is demonstrated that an ion is moved against an electrochemical gradient through a semipermeable membrane. This phenomenon has been clearly demonstrated to occur across cellular material such as the frog skin; and the ions and water are in solution on both sides of the "membrane." The movement of substances from within a given cell against an electrochemical gradient is postulated to occur because of an energy-requiring, membrane situated pump. These ideas are fundamental to the biologists' view of the living cell and are an integral part of a general theory which is termed the membrane or ionic theory.

The pump concept was first postulated explicitly by Dean in 1941 to account for the apparent movement of sodium against an electrochemical gradient. The creation of the pump concept is a natural consequence of a fundamental, yet unproved, postulation that the proteins, ions, and water are in free solution within the cell. In fact, there is no definitive experiment published, to my knowledge, that shows sodium or any other ion to move from a free solution of low concentration within a cell to a free solution of high concentration outside the cell. Part of the confusion that exists is that a rigorous demonstration of active transport is lacking; and the failure to demonstrate this phenomenon, even in extruded squid axons, has been replaced by criteria other than net movement of ions against an electrochemical gradient. For example, ouabain sensitivity and the presence of sodium-potassium activated adenosine triphosphatase have been substituted for active transport.

Furthermore, according to Ling, the energy requirements of the sodium pump alone are excessive. Ling's calculations on the energy requirements of the sodium pump have gone unchallenged—perhaps they were overlooked (*2*). Consider the following: (i) Assume first that ions and water are in a free solution, that the pumps are 100 percent efficient, and that Ling's calculations are incorrect. (I must add that I think the available evidence demonstrates that all these assumptions are incorrect, but I present them so that I may ask a question which perhaps will be answered by the scientific commu-