

THE MAMMALIAN COLOUR MODULATORS

RAGNAR GRANIT

*The Nobel Institute for Neurophysiology, Karolinska Institutet,
Stockholm, Sweden*

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BY DETERMINING the colour thresholds of isolated elements in the mammalian eye before and during polarization of the retina Gernandt (1) first showed that the on/off-elements were selectively influenced in different wave-lengths by the polarizing current. The wave-lengths selected were relatively few, the strength of the current constant (1.0 mA), and much of the experimental work was devoted to examining the scope and value of the new method.

I have now carried out a systematic analysis of the spectral properties of 28 elements, measured at every 0.020μ in the spectrum. In order to make it possible to select the strength of the polarizing current properly, the rheobase was first determined.

PROCEDURE

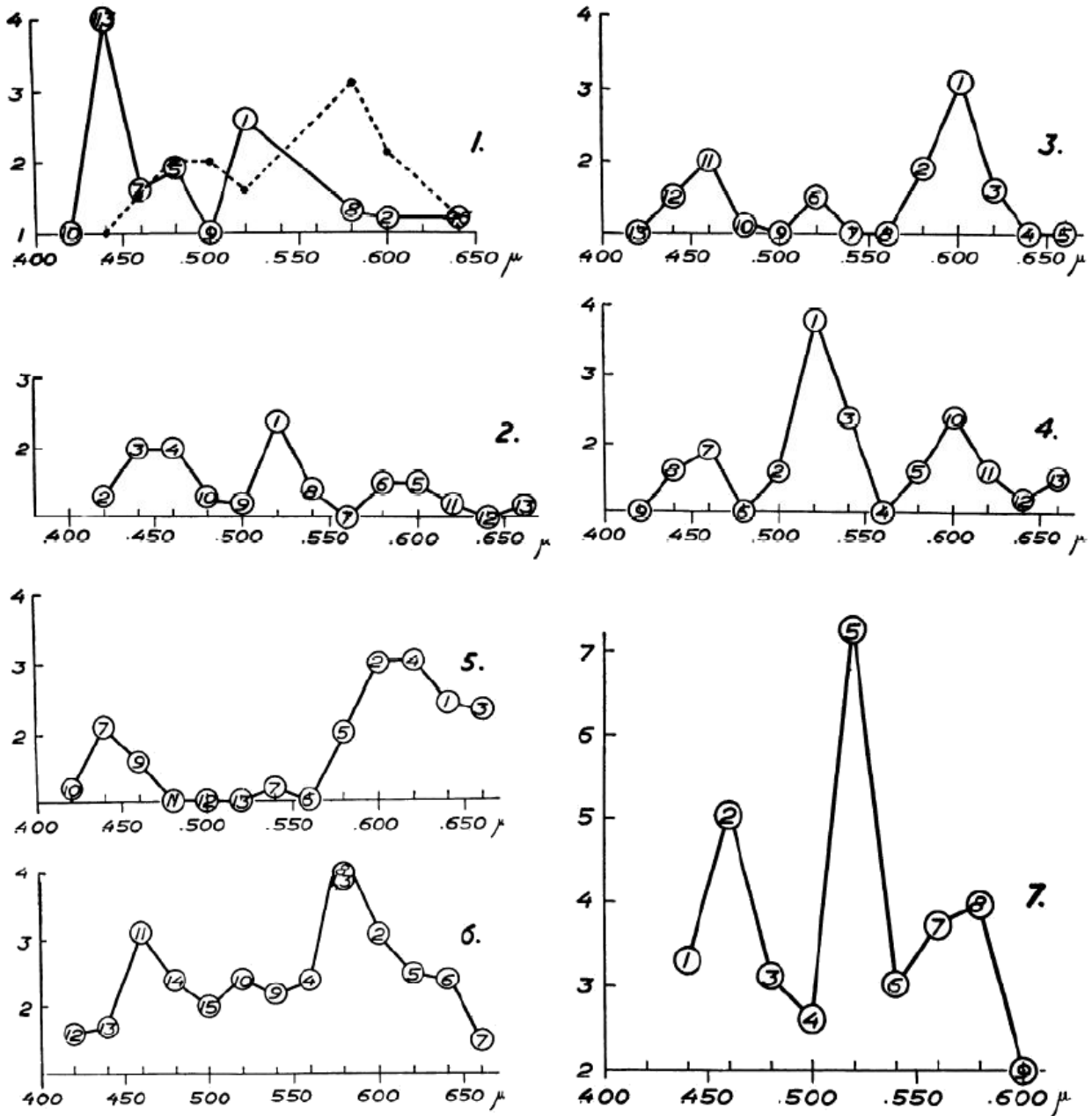
The principal effects of polarizing current and the technique itself has been described in another paper in the same issue of this journal (5). The electric current, passed through the retina of the dark-adapted decerebrated cat, facilitates or inhibits the light threshold (5). Depressions of almost any order of magnitude can easily be obtained in certain types of on/off-elements, modest facilitations are common, large facilitations less common. The effects have been shown to be a function of current strength and type of element (5).

In the present work very strong effects on the light threshold have been avoided because a gradual change then superimposes itself upon the effects to be measured, and, in this particular case, it has been desirable to keep the retina constant so as to be able to compare threshold changes in different parts of the spectrum of our Wright colorimeter (8).

In each wave-length the normal light threshold of the well-isolated spike was first determined, then the threshold during polarization of suitable strength. Dividing the one by the other gives a "polarization factor." This factor has been plotted for some elements as a function of wave-length in Figures 1-7. A factor of 8 (times the normal threshold) was considered very high. In 26 elements facilitations were measured, in 2, depressions. There were 26 on/off-elements and 2 pure off-elements.

RESULTS

Material. The results are based on 566 measurements, half of which consist of thresholds before, the other half of thresholds during, polarization. The wave-lengths were taken in haphazard order, as shown by Figures 1-7. It was generally attempted to measure 13 wave-lengths for each element but often there was insufficient energy at the ends of the spectrum and occasionally an element was lost in the midst of an experiment. Most missing values were at the extreme ends of the spectrum. Generally the off-components of an on/off-element were found easier to measure than the on-components. The reason for this is that they, as a rule, are preceded by definite inhibition of the spontaneous rhythm during the period of illumination (for



FIGS. 1-7. Polarization factors as described in text plotted against wave-lengths for seven separate elements. The numbers within the circles show order of taking the observations. 1, Anodal facilitation at 5 rheobases. On-component drawn in full, off-component dotted. 2, Anodal facilitation of off-component at 14 rheobases. 3, Cathodal inhibition of pure off-element at 4 rheobases (highly inhibitable element). 4, Anodal facilitation of off-component at 10 rheobases. 5, Exceptional element. Cathodal, in spite of which the off-component illustrated is facilitated by anode at 14 rheobases. 6, Cathodal inhibition of off-component at 4 rheobases (highly inhibitable element). 7, Anodal facilitation of pure off-element. Strong current, but by mistake polarization threshold not determined. Only nine observations obtained.

3 sec.) and so stand out better against the background activity caused by the polarizing current than the on-components. In this material there were 11 on-components and 17 off-components.

Polarization factors. Figures 1-7 show samples of individual elements.

For one of them (Fig. 1) both on- and off-components have been measured. The two components are seen to have had their maxima in different regions of the spectrum. Similar spectral differences between the on- and the off-component have also been noted by Gernandt (1). There is little to add to the figures, which are self-explanatory. The spectral location, shape and magnitude of the effects should be observed. It is a striking fact that the broad visual purple distribution of sensitivity with maximum around 0.500μ does not play any rôle in these curves.

Analysis of results. To begin with, let us for the sake of argument regard the 283 polarization factors as experimental errors in the determination of the thresholds. It might be added, however, that each polarization factor actually is established on the basis of a very large number of observations around the threshold (6). If the variations in the thresholds are due to experimental errors, then what is the probability, in each wave-length, for this error to exceed a given value? The following seven probabilities were computed: that the error exceeds 1.4 times the normal thresholds, that it exceeds 1.9 times, 2.4, 2.9, 3.4, 3.9, 4.4 and 4.9 times the normal threshold. Some of those probabilities are plotted as ordinates against wave-length in Figure 8.

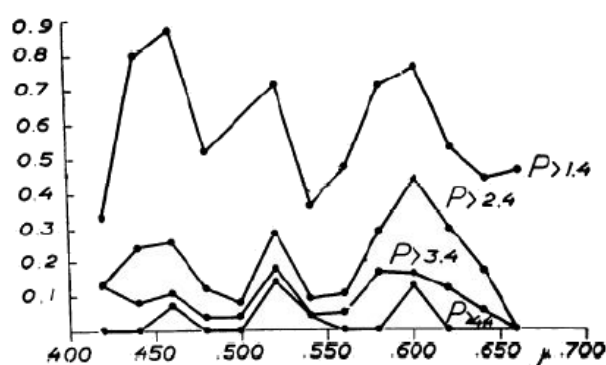


FIG. 8. The probabilities that the polarization factors exceed the values marked against the curves have been plotted against wave-lengths. See text.

It is clear from the figure that a very large number of the thresholds, taken during polarization, have been changed more than 1.4 times by the current. Such low probabilities tend to bring out broad, overlapping zones. By prescribing higher values for the probabilities, the regions within which effects occur become narrower.

In order to obtain some idea of both the top and the base of the curve the seven probabilities have all been averaged. In Figure 9 these

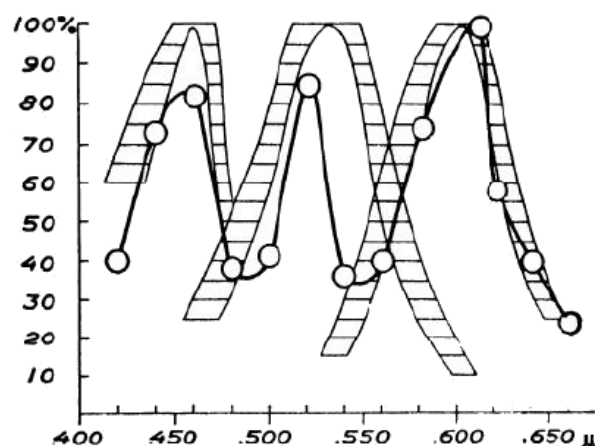


FIG. 9. Curve through readings: The seven probabilities, given in the text, have been averaged and plotted against wave-length with the maximum adjusted to 100. This curve is to be compared with the averaged modulators (3) as obtained by selective adaptation (lines enclosing hatched areas).

averages have been plotted in per cent of maximum. The other curve illustrates the averaged colour modulators in cat's eye, determined by the method of selective adaptation (3). Considering the very different methods applied in the two types of experiment as well as the fact that the off-components dominated in this work whereas the work on selective adaptation was carried out with on-components alone, there is good agreement between the two curves.

Since the individual curves varied a great deal, I chose to deal with them in the manner set forth above. But in addition the results were given to Dr. L. Goldberg (of the Caroline Institute) for statistical analysis. The average curve of Figure 10, surrounded by areas illustrating the mean variations was obtained from him. It shows that the average polarization factors for the 28 receptors also convincingly reproduce the three regions of predilection, seen in Figures 1-9.

DISCUSSION

Nature of polarization effect on colour reception. In view of Figure 10 and of the direct measurements of facilitations and inhibitions in the previous

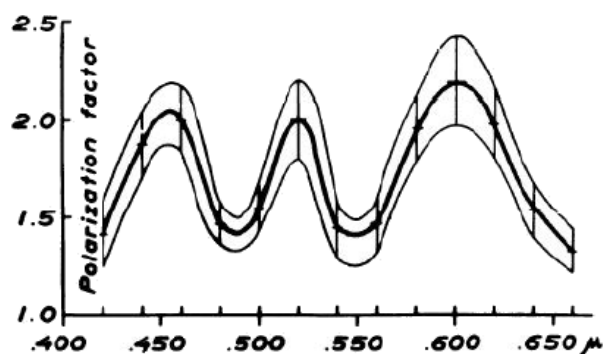


FIG. 10. The averaged polarization factors (thick line) and their mean variations plotted against wave-length.

paper (5), there would seem to be no need for regarding the results as errors of measurement although it was convenient to present them in that fashion. The work on selective adaptation (3) showed that the on/off-elements are inhomogeneous from the point of view of colour. Extremely definite results demonstrating the same fact have recently been obtained by Gernandt (2). When on/off-elements are light-adapted with red, green or blue light sometimes one, sometimes an-

other of the three colours succeeds in removing a large fraction of the on- or the off-component. Whether the on- or the off-component is affected is largely dependent upon the off/on-ratio in combination with specific colour sensitivity. It is therefore certain that the on/off-elements consist of structures of different colour sensitivity. Let us imagine, for instance, that a given element consisted of 1 green- and 20 red-sensitive structures. Now, since there is a definite effect of polarization on the light threshold (5), this must change the polarization factor of the element in that particular region of the spectrum for which it is specifically sensitive, in this case chiefly in the red region. It is therefore clear that the polarization method of analysis must yield information about the design of the mechanism of colour vision.

If it is natural, on the basis of the facts set forth above, to expect colour changes related to the spectral composition of the on/off-elements it is by no means primarily evident why the colour bands obtained with the polari-

zation factors should be as narrow as the individual curves of Figures 1-7. They share this property as well as the regions of predilection with the modulator curves (3, 4). In view of the results in the analysis of the effect of polarizing currents of different strength on the light thresholds (5), it has now become possible to explain this fact in this particular type of experiment.

It was shown in that work that the light thresholds of the pure on-elements, which are the simplest ones from the point of view of design and are lacking inhibition, also were practically insensitive to inhibition and but moderately sensitive to facilitation by polarizing currents up to exceedingly high strengths. These elements represent the receptor properties, less modified by synaptic factors than any other types of element. Thus the pure receptor responses are relatively insensitive to polarizing currents. The large facilitations and inhibitions of the light thresholds were only found in certain on/off-elements which for such purposes must possess specific structures, absent in the on-elements. Many on/off-elements are lacking these structures also. In the experiments on colour reception we have, accordingly, chiefly been playing upon specific facilitatory and inhibitory structures, and not been dealing with effects of polarization upon the receptors themselves. This can be shown in another way. In the fully dark-adapted eye, used in these experiments, the spectral sensitivity of the elements is wholly dominated by the broad visual purple curve with its maximum around 0.500μ (3, 4). Facilitation over the receptors ought to have reproduced this curve slightly modulated by other sensitivity curves, but there is, in fact, not a sign of it. On the contrary, the maximum effect of polarization is in the red end of the spectrum where the photosensitivity of visual purple is very low. Thus the narrow curves of Figures 1-7 have been obtained because of the fact that facilitation and inhibition of the light thresholds in on/off-elements depends upon specific structures for facilitation and inhibition in those elements.

Where, in the retina, are those specific structures situated? Undoubtedly somewhere in the synapses. The enormous range of variation of the off/on-ratio, a range of at least 100,000 (6), and the fact that the on- and off-components of the same on/off-element may have very different colour thresholds (2) show that these elements are composite structures to which units of different colour and other properties have contributed. These units can only have been joined to the same "final common path," the one from which the micro-electrode picks up the discharge, by the internuncial neurones of the retina. Accordingly, these experiments demonstrate that modulators obtained by polarization may vary in form, location and relative magnitude because of specific influences of facilitation and inhibition distributed over specific structures, almost certainly dependent on internuncial channels.

This leads to the final conclusion that the agreement in form and spectral locus of the modulator curves obtained (i) directly (4), (ii) by selective adaptation (3), and (iii) by polarization in this work signifies that the modulators need not represent primary colour-sensitive substances but can be, and in this particular work *are*, transformations of the spectral absorption

curves of these substances by internuncial facilitation and inhibition. In particular, inhibition can be assumed to have reduced the ordinates along the regions where primary curves overlap. In this way inhibition can be responsible for the narrowness of the curves and their variations in the three spectral regions within which modulators tend to appear. Possibly facilitation also contributes to this effect but the details of the mechanism must now be analyzed by experiments of a different type.

The primary curves. It is difficult to deduce the form of the three primaries from the modulators but it seems likely that the fundamental sensation curves, given in a previous paper in this journal (3), cannot be far from the truth. These curves were based on the three spectral regions of predilection within which modulators have been found. The average curve of the polarization experiments, given in Figure 10, must also be a fairly simple derivative of the primary curves. All these results suggest that it would be very difficult to synthesize the colour curves of the receptors with less than three variables. The number of variables may, however, be greater. For instance, by all three methods the modulators in the red region of the spectrum fall into groups around $0.600\text{--}0.620\mu$ (red modulators) and around $0.600\text{--}0.580\mu$ (yellow modulators). This is clearly shown also by the Figures 1-7.

Evidence for the existence of modulators in the human eye within two of the spectral regions of predilection, red and green, has recently been obtained by Thomson (7) who used small areas and low levels of intensity.

SUMMARY

The colour thresholds have been determined for single spikes isolated in the cat's retina with the micro-electrode technique before and during polarization of the retina.

The threshold is selectively altered in different regions of the spectrum by polarization. By dividing the threshold values *before* with those obtained *during* polarization a factor is obtained, called the "polarization factor," which illustrates the effects of polarization.

The polarization factors, as functions of wave-length, reproduce the colour modulators of the cat's eye.

The large effects of polarization on the light threshold have been shown to be due to specific structures for inhibition and facilitation (5) and not to receptor effects. Hence the modulators obtained by plotting polarization factors against wave-length should be modified by such effects and need not represent primary colour sensitivities.

The modulators obtained by polarization agree in the main with those obtained by other methods. Hence modulator curves can be transformations of primary colour curves by processes of facilitation and inhibition.

A recent extremely interesting report by Morton and his co-workers (BALL, COLLINS, and STUBBS, *Nature*, 1948, 161: 424) summarizes their attempts to influence the chromophore of visual purple, Wald's retinene, shown by Morton and Goodwin to be vitamin A aldehyde, by various chemical agents. A number of photosensitive substances have been obtained which reproduce the spectral regions of the modulators. The colour bands are even narrower than those obtained by myself from the optic nerve fibres by direct recording (see 4).

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