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DARK-ADAPTATION AND THE REGENERATION OF RHODOPSIN

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Ever since Kohlrausch (1922) measured dark-adaptation by plotting the logarithm of the threshold against the time in the dark, it has been clear that the recovery of visual sensitivity occurs in two stages. For, in general, the curve plotted exhibits two quasi-independent branches which meet at a pretty sharp kink. As Kohlrausch demonstrated, the early and faster process is associated with day vision: the later process with night vision. An enormous amount of subsequent work has substantiated this interpretation, but we are on much less secure grounds if we conclude (as is often done) that the first branch is concerned with cones only, the second with rods only and that there is no rod–cone interaction.

The 'cone branch' is obtainable from the rod-free fovea; it has the spectral sensitivity of daylight vision, monochromatic test flashes are usually seen as coloured, acuity is good, and a large Stiles–Crawford effect is found. This is strong evidence that cones are involved, but weak evidence that rods are not involved. To be sure, they are not involved at the central fovea, but the cone branch there is not quite the same shape as that found at the parafovea, where rods are anatomically present (Hecht, Haig & Wald, 1935; Sloan, 1950).

The 'rod branch' is obtainable from all parts of the retina where rods are present and is absent from the only places (fovea and blind spot) where they are not. This branch exhibits the spectral sensitivity of twilight vision which (after correction for absorption by the ocular media) corresponds very closely to the absorption spectrum of rhodopsin (Crescitelli & Dartnall, 1953), a photosensitive pigment which can be seen contained in the living rods (Boll, 1876). Test flashes appear colourless whatever their wave-length, acuity is poor and the Stiles–Crawford effect is small or absent.

This is strong proof that rods are involved, but evidence is not so clear whether cones contribute or not. If rhodopsin is the only pigment which absorbs quanta at threshold throughout the second branch, then after any

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fixed bleach, the curves obtained with test flashes of various wave-lengths should all coincide when suitably displaced parallel to the axis of log. *I*. Such coincidence, however, cannot be obtained from some curves which appear reliable. Kohlrausch (1931) and Wolf & Zigler (1955) found that adaptation with a blue test proceeded faster and more extensively than with an orange test—as might be expected if cones made some contribution to the excitability of the second branch.

On the other hand Sloan (1958), who alternated the colour of the test flash (blue/white) throughout the course of each run (instead of making separate runs for each colour tested), found no change in relative threshold. Sloan's is clearly the surer method of detecting any change in relative sensitivity. We have employed it, using alternately blue and yellow or green and yellow tests and looking for any variation in the differential sensitivity during the course of rod adaptation. We confirm Sloan in finding an exact equality throughout the second branch of the curve and conclude that, in the conditions of our experiments, cones enter no more into any part of this branch than they do at the final stage of complete dark-adaptation.

Actually the rod and cone curves do not meet quite sharply at a kink. Almost all published results show a rounded transition lasting a minute or so, as might be expected from the *probability summation* of two independent inputs (Stiles, 1949, p. 223). But the creeping transition which Wald (1960) has recently presented, lasting from the seventh to the seventeenth minute of dark-adaptation, has neither been seen in his earlier publication (Hecht *et al.* 1935; Wald & Clark, 1937), nor in those of other workers; nor was it found in our own experiments, where a very similar technique established a fairly abrupt transition from cones to rods.

It has been widely believed that the fall in threshold exhibited by the rod branch of the dark-adaptation curve represents in some way the regeneration of rhodopsin. Such a relation between threshold and pigment was put forward by Hecht at first to explain the recovery in sensitivity of the marine animals *Mya* and *Ciona*. The analogy with human dark-adaptation led to these ideas being carried over to a domain where exact measurements made them finally unacceptable, and Hecht's (1937) well-known review has only one thing quantitative to say about dark adaptation and the photochemical theory. It is, that the instantaneous threshold in the dark at the end of light-adaptation—the first point upon the dark-adaptation curve—is related to the light level to which the eye had been adapted by a formula which was derived from photochemical considerations and fitted the experiments very well. But the considerations which predicted the first point were quite unable to explain the rest of the curve (as Hecht was well aware), and actually the first point itself was only explicable by assuming that a retinal illumination of 0·03 td will bleach half the rhodopsin away. But this illumination corresponds to the incidence of less

than 1 quantum per rod per second and the direct measurements of Campbell & Rushton (1955) have shown that actually a light one million times stronger is needed. A more detailed discussion of the achievements and difficulties of the photochemical theory has been given elsewhere (Rushton, 1959).

In a determination of the dark-adaptation curve, the actual value of threshold intensity will obviously depend upon the size, duration, wave-length, etc., of the test flash; but if the curve represents simply the regeneration of rhodopsin, its *shape* should remain unaffected by changing these test parameters, since they will produce nothing more than a vertical shift. The observed results, however, turn out to be more complex. For, though change of wave-length may simply result in the vertical shift of the curve (as discussed above) an increase in the area or duration of the flash does increase somewhat the extent of dark-adaptation and prolong the recovery time (Craik & Vernon, 1941; Arden & Weale, 1954). Other factors, therefore, besides the regeneration of rhodopsin must contribute to the increase of excitability during dark-adaptation—presumably a reorganization of nerve connexions, as Lythgoe (1940) pointed out long ago, and as electrophysiology has since confirmed (Barlow, Fitzhugh & Kuffler, 1957*a*, *b*).

The difficulty in establishing securely the photochemical theory has been to measure in the same eye both the visual threshold and the rhodopsin level. For, before the development of retinal densitometry, it was not possible to estimate the visual pigments in man, and in animals not only does the quantitative extraction of rhodopsin need great care and patience, but the concomitant determination of the visual threshold by electrophysiology or behaviour constitutes a difficult and totally different problem. Such measurements, moreover, as were undertaken (Granit, Munsterhjelm & Zewi, 1939; Peskin, 1942) did not suggest any simple or indeed unique relation between rhodopsin content and e.r.g. in the frog. And there was very little evidence indeed to suggest that the increased sensitivity in dark-adaptation was related to the regeneration of rhodopsin, otherwise than through the fact that each represented a return to an equilibrium which had been disturbed by bleaching.

This was the position when retinal densitometry threw some new light upon the relation by permitting the visual pigment level to be measured in the living eye during bleaching and regeneration. It soon appeared that human rhodopsin is more or less completely regenerated by 40 min after the end of a full bleach (Rushton, Campbell, Hagins & Brindley, 1955), whereas human cone pigments are back in about 8 min (Rushton, 1958). Both these times correspond closely to the time needed for return to full visual sensitivity in the dark.

This coincidence strongly suggests that there is some intimate relation between threshold and pigment concentration, for while the pigment is regenerating the threshold is falling, and at about the stage where regeneration stops the improvement in sensitivity also ceases. But is the relation between threshold and pigment unique, and if so what is it? That is the question this paper sets out to answer.

It is known from the work of Hecht and others (Müller, 1931; Hecht, Haig & Chase, 1937; Haig, 1941; Mote, Riopelle & Meyer, 1950) that rod dark-adaptation curves following substantial light-adaptations are all very similar in shape, but are displaced laterally along the time axis to various extents. The greater the bleaching, the more the curve is displaced to the right and the later the recovery at every stage. On the other hand, a more profound rhodopsin bleach will also take longer to reach each fixed stage of pigment regeneration. Are the curves of pigment regeneration also related simply by lateral displacement? And is the time shift the same both for the recovery of threshold and for the regeneration of rhodopsin?

In the experiments to be described the rods were light-adapted to various extents and the subsequent regeneration in the dark was followed by retinal densitometry. The rod thresholds were defined by a single point, namely, the kink in the curve—the moment when the rods first became more sensitive than the cones (always after more than 8 min of dark-adaptation). Thus, after each light-adaptation it was possible in a single run to measure both the curve of pigment regeneration and the moment of cone–rod transition simply by interrupting the former for the short interval required to determine the latter.

It will be shown that no matter how much pigment had to be regenerated ($> 25\%$) the kink always occurred at the moment when 92% had been resynthesized. And in the Discussion it will be argued that this in conjunction with certain well-established facts defines the relation between threshold and rhodopsin content.

METHODS

Dark-adaptation. Records were traced upon Gunkel & Bornschein's (1957) modification of standard equipment. The subject fixated a small dim red light, and observed the flash of a 1° circular patch situated at 12° from the fovea, continually flashing on for $\frac{1}{2}$ sec, off for $\frac{1}{2}$ sec. The log. brightness was controlled by a photometric wedge driven by a motor, and records were traced upon a rotating drum. The direction of the motor was reversed by a switch operated by the subject in such a manner that when the flash was seen, the motor made it progressively dimmer until finally it was not seen, whereupon the subject reversed the switch and waited until he perceived the flash again. A saw-tooth tracing was thus obtained which continually oscillated about the true threshold.

Now in dark-adaptation measurements the moment of transition from first to second branch is usually detected by the kink in the curves. This kink is apt to show very clearly in the curves which authors draw through their points, but one sometimes feels doubts

when faced with one's own experimental results. Which irregularities are erroneous and should be smoothed out; which is the transition point, which perhaps could be emphasized a little? The use of test flashes with alternating colours has permitted us to locate the transition point securely.

In front of the test flash was interposed either a yellow interference filter ($\lambda = 580$ mμ) or a green one (520 mμ) with a neutral density 0·68 fixed to it, making the combination indistinguishable from the yellow in scotopic vision. In photopic vision, however, the yellow threshold was more than half a log. unit lower. These two filters were alternately interposed, being changed every minute throughout dark-adaptation. The result is as shown in Fig. 1. Throughout the first branch of the curve the threshold changed with each change in filter, and the tracing resembles a battlement. Upon the second branch *per contra* the two lights are equivalent and the battlements vanish. The moment of transition could be appreciated by the subject as the point when an exchange of filters suddenly produced practically no change in the colour or the brightness of the test flash.

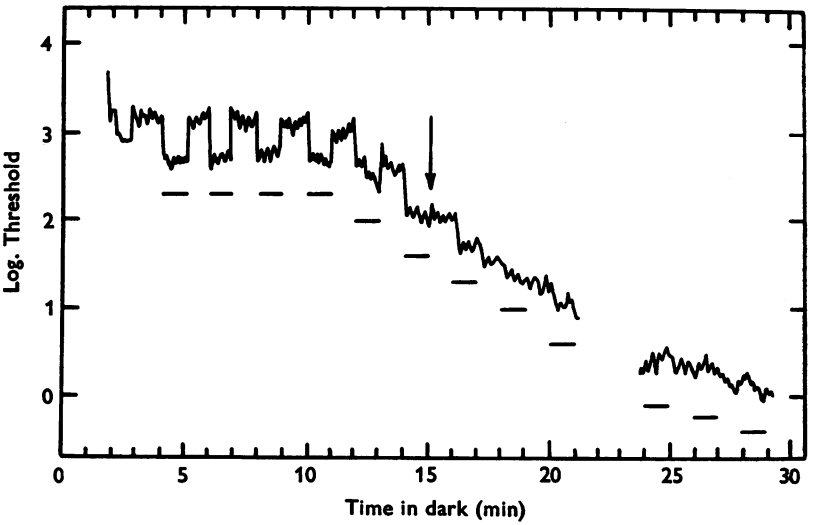

Fig. 1. Dark-adaptation following total bleach. Irregular line is the saw-tooth tracing recorded (with pause at 21–24 min). Test flashes were yellow during the 1 min periods indicated by the short horizontal lines. During the alternate minutes they were green of equal scotopic value. Arrow indicates cone–rod transition for yellow test light.

Measurement of rhodopsin. The retinal densitometer had the simple form indicated in Fig. 2. The light source A was a vertical coiled filament of a 6 V car headlamp. From it two beams diverged and were united at the polarizing beam-splitter B, upon which the filament was imaged by lenses L_1, L_2 and the two images were adjusted to coincide. Polaroids P_1, P_2 completed the orthogonal polarization of the two united beams, which were finally reflected into the eye by the tiny mirror M. The lens L_4 focused the filament upon the cornea and at the same time brought sharply into focus upon the retina the image I_1 of the stop S_1, which was uniformly illuminated by coinciding with the image of lenses L_1, L_2 formed by L_3. The light reflected from the eye fell upon L_5 and an image of I_1 was formed at I_2 in the plane of the iris stop S_2, which was shut down so as to be entirely filled by I_2. The light passing through S_2 (and that only) fell upon the cathode of an E.M.I. photomultiplier tube $P.C.$, the output of which was measured.

Principle of measurement. The deep red filter F_1 transmitted a light which was not appreciably absorbed by rhodopsin, so the reflexion of this light into *P.C.* was unaffected by rhodopsin density and hence could be used as a reference standard. The filter F_2 transmitted light of wave-length 516 mμ, which is strongly absorbed by rhodopsin; thus the amount of this light reflected into *P.C.* was less the greater the density of the rhodopsin through which it had twice passed. The polaroid P was mounted in a ball race and rotated at a rate of 40/sec. As it rotated it cut off alternately the red and the green light. Thus, if the rotation rate were n radians/sec, the instantaneous output of the photocell would be

$$r\sin^2 nt + g\cos^2 nt,$$

where r and g are the maximal outputs from red and green lights alone and t is time (sec).

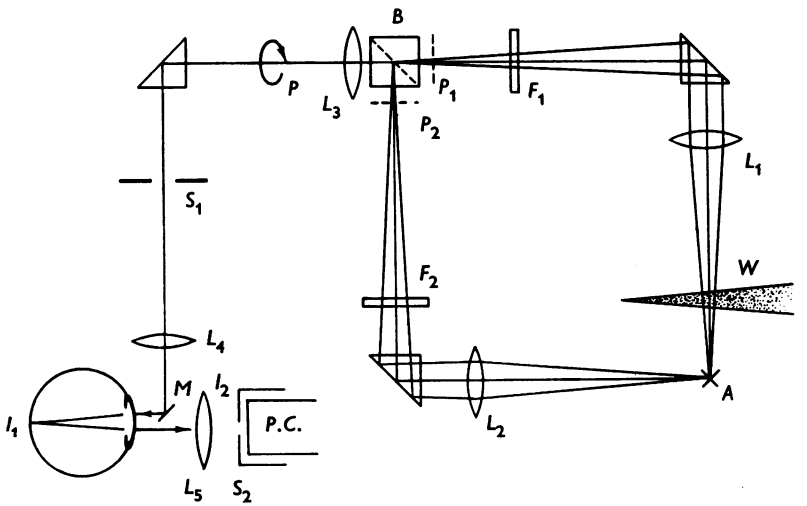

Fig. 2. Diagram of simple form of retinal densitometer. For details see text.

By shifting the photometric wedge W, the value of r could be made equal to g, whereupon the output becomes

$$g\,(\sin^2 nt + \cos^2 nt) \equiv g$$

so the a.c. output becomes zero.

It will now be plain how rhodopsin can be measured. The a.c. output is detected and adjusted to zero by shifting W. Any increase in rhodopsin density will affect g but not r, and so the a.c. output will no longer be zero. When balance has again been restored the wedge W must have introduced into the red path a density increment equal to that introduced by rhodopsin into the green path.

The signal/noise ratio is very much improved by the use of a phase-sensitive rectifier. An additional steady plane-polarized light (not shown in Fig. 2) is passed through the rotating polaroid P and focused upon a second photomultiplier cell whose output is *multiplied* by the output from the eye. The integrated product is passed through a sensitive galvanometer which is brought to zero deflexion by suitable setting of the wedge W. Now when t is great

$$\frac{2}{t}\int_0^t \sin nt \sin pt\, dt \to 0,$$

except when $p = n$, when the expression becomes unity. In this way the integrals of all noisy components in the signal average out to zero except such as happen to coincide in frequency with n, the signal itself, and are more or less in phase.

Procedure

The subject's pupil was dilated with cyclogyl (cyclopentolate HCl 1·0 %: Merck Index) and he was aligned in the densitometer, being kept in position by biting upon a dental impression and steadying his head with a moulded forehead rest. He fixated upon a small red light (not shown in Fig. 2) so placed that the 3° region of retina measured for rhodopsin coincided with the 1° test flash detected during dark-adaptation. This adjustment was secured by removing the filters F_1, F_2 (Fig. 2) so that a bright white light fell upon the retina while the red light was being fixated. Then immediately the subject moved to the dark-adaptation apparatus and adjusted the fixation light there so that the test flash fell upon the after-image still prominent from the white densitometer exposure.

The pre-adaptation bleaching light was a very bright white light presented in Maxwellian view for 30 sec. The subject fixated just outside the edge of the Maxwellian lens at a marked spot, so the fixation spot was not greatly light-adapted, and the regions to be measured lay at the centre of a 20° bleached area.

Campbell & Rushton (1955) showed that the Bunsen–Roscoe law ($It = k$) holds for the bleaching of rhodopsin with exposures up to 45 sec, and Rushton (1956) found that half the rhodopsin was bleached by a retinal illumination of 6·8 $\log_{10}$ troland sec, delivered within 45 sec. The fraction bleached by other illuminations is given by Fig. 6 of that paper, in which the bleaching light without any filter interposed was 7·8 log. troland sec. Alternatively the result may be computed from the formula

$$\log_{10} \log_{10} \frac{1}{p} = \log_{10} It - 7{\cdot}3, \qquad (1)$$

where p is the fraction of rhodopsin remaining unbleached by exposure to a light It (scotopic) troland sec. In this way it is possible to bleach a predetermined fraction $1-p$ of the rhodopsin in any retinal region.

An important detail in the technique was for the subject to acquire the skill to quit the densitometer and later to resume an identical position there, so that measurements of rhodopsin (in equilibrium) were repeatable. With this skill it was not necessary to remain clamped and immovable throughout the long time course of rhodopsin regeneration, and moreover measurements of dark-adaptation could be interpolated.

The actual course of an experimental run was as follows. After a preliminary bleach the subject remained half an hour in the dark, sometimes tracing a curve such as Fig. 1. Then one or two preliminary measurements were made of the wedge settings for the fully regenerated rhodopsin density. The eye was then exposed for 30 sec to the Maxwellian bleaching light at a predetermined intensity level. The subject immediately resumed position in the densitometer and readings were taken of wedge settings, frequently at first, less so as recovery slowed, with results such as those shown in Fig. 3. When the intervals became as long as 2 min the subject left the equipment and rested in the dark between readings. The eyes were always kept shut except during the 7 sec or so during which wedge settings were being made.

For the first run the rhodopsin was bleached completely, so that the change in wedge setting which corresponded to full bleaching was known from the outset. The setting corresponding to 85 % regeneration was therefore easily computed. When in each run regeneration was seen to reach this point, the subject left the densitometer and took up position at the dark-adaptation equipment, and at once began to record his threshold, employing the alternate yellow and green flashes matched for scotopic equality.

Without exception, at the time when regeneration was 85 % complete, the yellow flash appeared much brighter than the green, showing that cones were still the more sensitive system. Within 5 min, however, conditions changed, and the two lights appeared identical both in brightness and colour. The subject could signal the moment of this change and it

corresponded to the kink in the recorded curve, and to the change from castellation to smoothness. Dark-adaptation measurements were continued for 2 or 3 min to establish the rod curve more securely, and then the subject returned to the densitometer to complete the regeneration curve. In conclusion, the fully dark-adapted threshold was recorded upon the tracing.

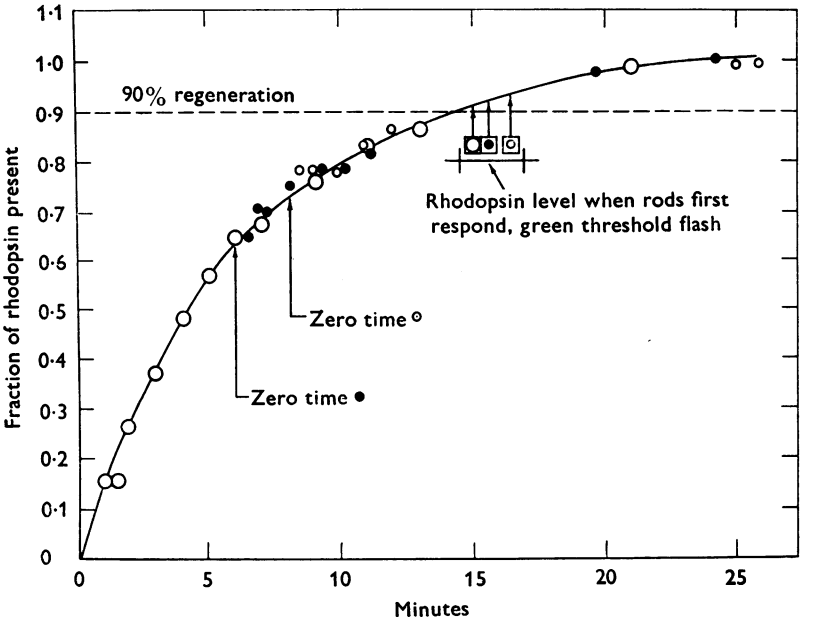

Fig. 3. Regeneration of rhodopsin in the dark. The curve is an exponential of time constant 6 min. All black circles displaced to the right so that zero lies at point indicated. Small white circles similarly displaced but further. For each run the cone–rod transition of the dark-adaptation curve lay where shown by the arrow. ○ 25 %; ● 35 % bleach; ◯ total bleach.

RESULTS

The results of one experiment are shown in Fig. 3, where the large white circles show the fraction of rhodopsin regenerated at various times after a total bleach. The curve is an exponential of half-decay time 4 min. The moment in dark-adaptation when the rods first responded to the green flash is shown by the arrow and corresponds to a regeneration of 92 % rhodopsin after an interval of 15 min in the dark.

The black circles represent a repetition of the experiment with a weaker pre-adapting light which bleached 35 % of the rhodopsin. These results have been plotted in Fig. 3, displaced 6 min to the right so that zero time falls at the point shown. This is the displacement required to bring the first point into coincidence with the exponential curve, and it is seen that all the subsequent points lie also upon the same curve. The moment when

rods first responded to the green flash is shown by the second arrow, and though this is now only 10 min from zero time it still corresponds to about 92 % rhodopsin regeneration.

The small white circles represent a third run, now with a 25 % bleach. The method of plotting is analogous to that used for the black circles and rods still first appear at about 92 % rhodopsin regeneration.

DISCUSSION

The important fact which appears from the experiment of Fig. 3 is that, despite very different extents of bleaching and subsequent periods in the dark, the threshold for rods is found to be the same (namely just below the dark-adapted cone threshold and 1·9 log units above absolute rod threshold), when rhodopsin regeneration has reached a fixed value of about 92 %. Nor is this fixed correspondence between threshold and regeneration likely to be confined to that point alone.

Most investigators (Müller, 1931; Hecht *et al.* 1937; Haig, 1941; Mote *et al.* 1950) who have made a careful study of the effect of various *substantial* bleachings upon the subsequent dark-adaptation have shown that the various rod curves are superposable by lateral displacement. And we have seen that the regeneration curves of rhodopsin appear also to be superposable by lateral displacement. If this is accepted it must follow that the displacements of Fig. 3 which cause all the pigment curves to coincide will also bring into coincidence the threshold points, not only at the first moment of their appearance which we have tested but throughout.

It must be admitted, however, that measurements of the final 8 % of rhodopsin regeneration are not accurate enough to establish the exact superposability of this tail end of the curves. Our confidence in it rests upon the following argument.

We have seen from Fig. 3 that the over-all regeneration of rhodopsin follows pretty accurately the relation

$$y = y_0 e^{-t/6}, \tag{2}$$

where y is the fraction of opsin still uncombined at time t (min), and y_0 is y at zero time. But from the chemistry of rhodopsin regeneration (Wald & Brown, 1956) we know that

$$\begin{array}{ccccc} \text{retinene} & + & \text{opsin} & \rightarrow & \text{rhodopsin} \\ x & & y & & (1-y). \end{array}$$

Thus if x is the concentration of retinene, the rate of regeneration of rhodopsin is given by

$$-\frac{dy}{dt} = kxy,$$

where k is the velocity constant. But differentiating (2) gives

$$-\frac{dy}{dt} = \frac{1}{6}y_0 e^{-t/6} = y/6.$$

$$\therefore \quad x = 1/6k \tag{3}$$

Thus the exponential regeneration of rhodopsin implies a constant value for x, the concentration of 11-*cis* retinene, as has already been shown in the case of cone pigments (Rushton, 1958). Now the retinene is formed either by oxidation from the 11-*cis* vitamin A store or by isomerization of the all-*trans* retinene liberated upon bleaching, and it is removed by combining with opsin to form rhodopsin. The exponential curves show that when removal is relatively fast the retinene replacement maintains the concentration x at a very fixed level. Thus it would be surprising if this level could no longer be maintained in the later stages of regeneration when removal is only slight. But if the level here does remain fixed, the curves must remain strictly exponential, and we are justified in our belief in their precise superposability. From this we conclude that there is a fixed relation between threshold and the fraction of opsin still uncombined, and we may now ask what that relation is.

Each member of the series of dark-adaptation curves obtained by Haig (1941) after various bleaching exposures was shown to fall upon a mathematical curve of the form

$$\log I/I_D = ae^{-t/b}, \tag{4}$$

where I_D is the dark-adapted threshold and a, b are constants. It is true that the equation quoted by Haig looks much more complex than (4) but it may be reduced to this form by a suitable change of constants. Haig found the time constant b to increase with increased bleaching, but for bleaches greater than 25 %, b reached an upper limit of about 6 min. Now 6 min is the time constant for rhodopsin regeneration, so combining equations (4) and (2) we obtain

$$\log I/I_D = ae^{-t/6} = ay/y_0. \tag{5}$$

Thus, if threshold is expressed in units of the dark-adapted value I_D, we derive the required relation—log. threshold is proportional to the fraction of opsin still uncombined. This is the relation which Wald, Brown & Smith (1955) have deduced (for small bleaches) from the 'compartment hypothesis' of Wald (1954). It receives experimental support from the impressive paper of Dowling & Wald (1960) which has just appeared, wherein several important changes in structure and function of the rat's retina are studied in relation to the removal of rhodopsin. One aspect of this work concerns the change in log. threshold which accompanies the bleaching

and regeneration of rhodopsin. Albino rats had all their rhodopsin bleached away by exposure to bright light. Then, during the subsequent stay in the dark, the e.r.g. was determined at different times in response to flashes of various strengths. The rhodopsin present at each state was found by killing a rat and extracting its rhodopsin, one pair of retinas yielding a solution from which a measurement could be made with good precision. Experimental details are not given, but this exacting work appears to have been well performed, and demonstrates good linearity between the rhodopsin content and the log. threshold for the first appearance of the e.r.g. This investigation therefore established experimentally for the rat the linear log. threshold relation which we confirm in man at a very different level of visual activity.

Dowling & Wald (1960) then went further and extended the relation to the condition of night blindness which follows deprivation of vitamin A from the diet. Young rats were reared upon food lacking vitamin A but containing vitamin A acid. They grew well and appeared normal in health except for the rods, which at first lost rhodopsin and later lost their structure and degenerated. During the period of 7–10 weeks, throughout which rhodopsin progressively fails but structural change is not yet severe, the e.r.g. at maximum is nearly normal in size, though it needs a much stronger flash to elicit it. The relation between the e.r.g. log. 'threshold' and the fraction of rhodopsin present, as compared to normal, at each stage is again linear. And the line is the same one which fitted the relation described above. Thus removal of rhodopsin either by bleaching or by vitamin lack raises the 'threshold' in identical fashion.

Now the 'threshold' for an e.r.g. is some 1000 times higher than that for rod vision, so one should hesitate to jump from relations about e.r.g. in the rat to perceptual thresholds in man, though Karpe & Tansley's (1948) work goes some way to support this. The present results, however, do show rather directly that the same logarithmic relation applies.

In this paper the evidence that log. threshold depends immediately upon rhodopsin content is fairly straightforward, but the grounds for believing the relation to be linear are more indirect. We are strengthened in this belief, however, not only from Dowling & Wald's results upon the rat, but also from some observations upon a 'rod monochromat' where the same relation could be demonstrated upon a far more extended scale. This is described in the papers immediately following.

SUMMARY

1. Dark-adaptation curves were traced by using a test flash which alternated in colour each minute between yellow and green. The coloured filters were adjusted for scotopic equality, thus they were indistinguishable on the rod branch of the curve (Fig. 1) but exhibited a battlement tracing upon the cone branch. In this way one knew at each minute whether cones or rods were involved.

2. The regeneration of rhodopsin was measured by reflexion densitometry in the normal eye following light adaptations which bleached various fractions of rhodopsin from 25 to 100%. Regeneration always returned along an exponential curve of 4 min half-return time.

3. During each run of pigment measurement the procedure was interrupted for a few minutes to permit some dark-adaptation measurements to be made. These always included the cone–rod transition, which occurred at about 2 log. units above the full dark threshold.

4. At the moment of the transition the rhodopsin had always regenerated about 90% independently of the amount bleached initially.

I should like to thank Dr R. D. Gunkel for the loan of his dark-adaptation equipment and for valuable assistance in the course of these experiments, and I am grateful also to all those who offered themselves as subjects in this work. Above all am I indebted to Dr M. G. F. Fuortes, who proved to be the most steady and consistent subject I have ever had. From his eye are taken the records shown in this paper: to his skill is due such accuracy as has been achieved.

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