THE RELATION BETWEEN THE WAVE-LENGTH OF LIGHT AND ITS EFFECT ON THE PHOTOSENSORY PROCESS.*

BY SELIG HECHT.

(From the Physiological Laboratory, College of Medicine, Creighton University, Omaha.)

(Received for publication, November 11, 1920.)

I.

1. In a series of publications I have described several phases of the mechanism which underlies the sensitivity of certain animals (Mya and Ciona) to light (Hecht, a, b, c, d). The basis of this mechanism is essentially a process involving two intimately connected chemical reactions. The first of these is a reversible photochemical reaction

\[ S \rightleftharpoons P + A \]

in which \( S \) is the sensitive substance, \( P \) the principal product of decomposition, and \( A \) its accessory. The second reaction is an ordinary chemical reaction

\[ L \rightarrow T \]

which is catalyzed by the products of reaction of the photochemical reaction. The thermolabile material \( T \) is the end-product of the sensory process, and a definite quantity of it is required for the initiation of an impulse which results in the response of the animal to illumination.

The properties of these different substances and reactions are derived from quantitative studies of the responses of animals under conditions of experimentation calculated to bring them out. As the characteristics of the reactions and their components become known

* These experiments were carried out with the aid of a grant from the Elizabeth Thompson Science Fund, to the trustees of which I wish here to express my thanks.

375

The Journal of General Physiology
in greater detail, their identification becomes increasingly possible. Heretofore attention has been centered mainly on the kinetic and dynamic relations of the sensitive system. It is the purpose of the present observations to furnish the basis for the description of a physical property of the photosensitive substance $S$.

2. It is axiomatic to say that in a photochemical transformation only the light which is absorbed is active in producing chemical change. Although the converse,—that all the light which is absorbed gives rise to chemical action,—is not proved, it is well known that the absorption spectrum of a chemical system is intimately connected with its photochemical behavior (Sheppard, 1911, p. 140). Thus, if in a given photochemical system we know the relative effectiveness of a series of lights of different wave-length, it is possible to draw certain conclusions with regard to the absorption spectrum of the sensitive substance in the range of wave-lengths which have been investigated (Lasareff, 1907). Experiments were therefore undertaken to determine the effectiveness of light of different wave-length on the photosensory responses of $Mya$ arenaria.

3. The method used is in its essentials as follows. The reaction time of $Mya$ to light varies inversely as the logarithm of the intensity (Hecht, 1920–21, e). The relation between these two variables may be found for light of any quality. When using monochromatic illumination, the curve expressing the relation between the intensity ($I$) and the reaction time ($t$) may be called an $I$-isochrome, in analogy to the curve at constant temperature called an $I$-isotherm. The experiments consist in mapping out the $I$-isochromes for the photic response of $Mya$ to lights of different composition. The relations among the isochromes will give the relative effectiveness of the different lights.

The experiments were performed in the Marine Biological Laboratory at Woods Hole, Massachusetts, during the summer of 1920.

II.

1. Monochromatic light is secured by means of Wratten Light Filters Nos. 70 to 76 inclusive. These are made by the Eastman Kodak Company, and consist of specially stained gelatin films cemented between glass plates. The absorption spectra of these light filters
FIG. 1. Transmission spectra of the Wratten Light Filters used to secure monochromatic illumination.
have been carefully measured.\footnote{The measurements are given on p. 61 of a booklet—Wratten Light Filters—published by The Eastman Kodak Company.} They are presented graphically in Fig. 1. The white spaces are the transmission areas for the different screens in terms of the per cent transmission of the incident light.

It is apparent that each screen transmits a rather narrow portion of the visible spectrum. In the significant part of the spectrum ($\lambda = 400$ to 600 $\mu\text{m}$) the screens transmit bands about 40 $\mu\text{m}$ wide; most of the light transmitted is confined to an even narrower band.

2. With these screens I used a 400 watt, concentrated-filament Mazda lamp, running on an ordinary lighting circuit of 115 volts. This source of light may be considered as a point source, and different relative intensities may be obtained by placing animals at different distances from the light, the intensities being computed on the inverse square law.

For ordinary sources of light the energy content of the spectrum varies with the wave-length. It is therefore important to know the exact values of the relative energy distribution of the light in order that the energy transmitted by the filters be known. The results of the determination of the energy distribution for the 400 watt lamp which I used in these experiments are shown in Fig. 2, which gives the relative energy content of the different wave-lengths of the visible spectrum.

From Fig. 2 we know the energy incident on the filters. Fig. 1 in turn tells us the portion of this incident energy which is transmitted by each of the filters. It is thus a simple computation to find out the energy content of the light transmitted by each filter in conjunction with the lamp. The transmission values in Fig. 1 and the energy values in Fig. 2 are both determined for bands 10 $\mu\text{m}$ wide. The total energy transmitted by any filter is therefore the sum of the energy transmitted by its constituent 10 $\mu\text{m}$ bands. These totals are given in Table I.

The Mazda lamp plus a screen may thus be considered as a source of energy which radiates a narrowly defined portion of the spectrum, and whose intensity is proportional to the energy content of the transmitted light. For convenience I have used for the relative intensity the same figures as those given in Table I for the transmitted energy.
Fig. 2. Energy distribution in the spectrum of the 400 watt lamp used in conjunction with the light filters.

TABLE I.
Relative Energy Transmitted by Wratten Light Filters Plus a 400 Watt Lamp.

<table>
<thead>
<tr>
<th>Filter No.</th>
<th>Energy</th>
</tr>
</thead>
<tbody>
<tr>
<td>70</td>
<td>366.8</td>
</tr>
<tr>
<td>71</td>
<td>285.3</td>
</tr>
<tr>
<td>72</td>
<td>34.8</td>
</tr>
<tr>
<td>73</td>
<td>88.6</td>
</tr>
<tr>
<td>74</td>
<td>36.8</td>
</tr>
<tr>
<td>75</td>
<td>106.9</td>
</tr>
<tr>
<td>76</td>
<td>38.1</td>
</tr>
</tbody>
</table>
The distribution of the energy transmitted by each screen is given in Fig. 3. For uniformity the total transmission is given the value of 100. It will be seen that the point of maximum energy transmission is occasionally shifted toward the red end as compared with the per cent transmission given in Fig. 1. This is, of course, because the energy content of the spectrum increases steadily toward the red end, as shown in Fig. 2.

3. In Figs. 1 and 3 the transmission of the filters is given only for the visible spectrum. All the filters transmit infra-red rays. It may, however, be said at once that rays beyond $\lambda = 680 \, \mu\text{m}$ are ineffective as stimulating agents for *M. ya*. I have determined this by subjecting animals to very intense illumination from which all the visible rays have been filtered out by means of Wratten Light Filter No. 87. This screen transmits the rays beyond $\lambda = 680 \, \mu\text{m}$, and none below that wave-length. In no case did the animals respond to these infra-red rays. As a source of error, therefore, infra-red rays may be left out of consideration.

The screens do not transmit ultra-violet light. In addition, the light from the incandescent filament has to pass through several centimeters of glass before reaching the animal. Therefore, the ultra-violet rays are screened out in this way as well, and do not enter as a source of error.

The filters are quite photostable. But in order to avoid any possible bleaching effect, a shutter is placed between the light source and the filter, so that the filter is exposed to light only during the few seconds required to expose an animal and to measure its reaction time. Moreover, the light is turned off between exposures in order that as little heat be formed as possible. This is a necessary precaution, because a 400 watt lamp produces a tremendous amount of heat, which might otherwise pass even through the shutter and affect the gelatin between the glass plates of the filter.

From what has been said it is evident that these Wratten Light Filters furnish a simple method of procuring practically monochromatic light of known intensities, many times greater than can ordinarily be produced with the aid of a dispersion spectrum.

4. Further than the use of the screens, the experiments involve no new principle. A filter is placed in the path of the beam of light im-
Fig. 3. Energy distribution of the light transmitted by the filters when used with the 400 watt lamp. The total energy for each filter is placed arbitrarily at 100. Cf. Table I.
mediately in contact with the shutter (for a description of the apparatus used, see Hecht, 1919-20, c). Several animals which have been thoroughly dark-adapted are then used. The reaction time of each animal is measured with a stop-watch. After a rest of 15 minutes in the dark, the animals are again tested, but at a distance from the light nearer or farther than before, depending on the magnitude of the previous reaction time. After another period in the dark, the reaction time is measured at still another intensity, and the process repeated until enough responses have been measured to cover a range between 2.0 and 4.0 seconds in the reaction time.

The curve giving the relation between the reaction time and the intensity is the II-isochrome for this filter. The entire procedure is then repeated with another filter, until all the filters have been tested. In this way a series of II-isochromes are mapped out for the seven portions of the visible spectrum shown in Fig. 3.

III.

1. Several preliminary experiments agreed in showing that the light coming through Filter 75 is the most effective portion of the spectrum. A final set of experiments was then arranged using nine dark-adapted animals. The results are given in Fig. 4. Each point is the average of nine determinations of the reaction time, one for each animal. The curves are the II-isochromes drawn smoothly through the points. An exception is the curve for Filter 76, which for obvious reasons is drawn parallel to the other six curves.

The range of intensities required to produce similar responses is very large; from 2 to 16,000 units. To present them in a single figure is manifestly impractical. Moreover, it has been shown that the reaction time is inversely proportional to the logarithm of the intensity. Consequently, the abscissae of Fig. 4 are the logarithms of the intensities and not the intensities themselves. The use of such a logarithmic plot makes all the II-curves parallel to one another, and renders their comparison more simple than would otherwise be the case.

The II-isochromes of Fig. 4 show at once that the most effective portion of the spectrum is the light transmitted by Filter 75. This corresponds to a band whose maximum is at 490 \( \mu \mu \), in the blue-green.
It requires the least amount of energy to produce a given photosensory effect.

2. It will be of significance to determine quantitatively the relative effectiveness of the different parts of the spectrum. The method used in this connection deserves a little consideration. It is required to find the relative effectiveness of a series of reagents on a given system. The common procedure of comparing the different effects produced by the same concentration of the reagents, though giving qualitative results, is utterly fallacious for quantitative purposes, except in rare instances such as when the effect produced is a linear function of the concentration. A quantitatively correct evaluation requires the comparison of the concentrations of the different reagents which will produce the same effect.

I need not labor the point, because Brooks (1920) has presented it in some detail in its bearing on the theoretical interpretation of hemolysis data. Applied to our immediate problem, it follows that the important thing in working with spectral light is not to have a spectrum of equal energy distribution with which to determine the relative effects of different parts of it, but to have a spectrum of variable, known energy distribution, with which to determine the...
energy required to produce the same sensory effect. The value of the latter method will become apparent when the attempt is made in the next section to interpret the experimental findings.

In the present instance the same sensory effect is represented by responses produced in the same reaction time. It is therefore required to measure the different intensities necessary to produce the same reaction time. This can be done by reading from the $I_t$-isochromes of Fig. 4 the intensities corresponding to definite values of the reaction time.

I have done this for three values; for 2.5, 3.0, and 3.5 seconds. In each instance the intensity for Filter 75 is placed at unity, and the relative intensities of the others computed in terms of it. The three values so obtained for each portion of the spectrum are given in Table II. In treating them further, I have used the average of the three figures for each filter.

### Table II.

Relative Intensities Required to Produce the Same Reaction Time.

<table>
<thead>
<tr>
<th>Reaction time (sec.)</th>
<th>Intensity for filter No.</th>
<th>Filter 70</th>
<th>Filter 71</th>
<th>Filter 72</th>
<th>Filter 73</th>
<th>Filter 74</th>
<th>Filter 75</th>
<th>Filter 76</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.5</td>
<td>4,030</td>
<td>452</td>
<td>66.9</td>
<td>22.8</td>
<td>13.2</td>
<td>1.0</td>
<td>5.1</td>
<td></td>
</tr>
<tr>
<td>3.0</td>
<td>3,650</td>
<td>392</td>
<td>65.4</td>
<td>22.0</td>
<td>12.9</td>
<td>1.0</td>
<td>6.5</td>
<td></td>
</tr>
<tr>
<td>3.5</td>
<td>3,240</td>
<td>312</td>
<td>63.0</td>
<td>20.7</td>
<td>12.6</td>
<td>1.0</td>
<td>5.3</td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td>3,640</td>
<td>385</td>
<td>65.1</td>
<td>21.8</td>
<td>12.9</td>
<td>1.0</td>
<td>5.6</td>
<td></td>
</tr>
</tbody>
</table>

3. The relative effectiveness of the different lights may be represented by the reciprocals of the relative intensities. Fig. 5 shows these values in graphic form. For convenience, and for another reason which will presently become apparent, the reciprocal of the maximum is made equal to 100, the other values being computed accordingly and employed as ordinates. The wave-length is used as abscissa. Each point has been plotted as a rectangle, whose center is the point of maximum energy transmission as given in Fig. 3, and whose horizontal dimension represents the points between which 75 per cent of the transmitted energy is confined. A smoothed curve through the points shows strikingly that the most effective portion
Fig. 5. Relative effectiveness of the spectrum in sensory stimulation. The curve also represents the absorption spectrum of the photosensitive substance of Mya. In the latter case the ordinates represent per cent of absorption by the sensitive substance.
of the spectrum for the sensory stimulation of $M_{ya}$ lies between 490 and 510 $\mu \text{m}$. The effectiveness of the spectrum drops rapidly on both sides of this maximum, the parts above 610 $\mu \text{m}$ and below 420 $\mu \text{m}$ being practically ineffective as stimulating agents.

IV.

1. On the basis of certain considerations it is possible to place a physical interpretation on the results as presented in Fig. 5 and Table II. Lasareff (1907) has shown that for dyes that are bleached rapidly by light, the photochemical effect is directly proportional to the energy absorbed per interval $\Delta \lambda$, independent of the value of $\lambda$ and of the position of the absorption maximum. These results have been confirmed on other photochemical reactions (Weigert, 1911, p. 90); and a similar state of affairs has been shown to be true for the bleaching of visual purple (Trendelenburg, 1911).

This means that in order to produce a given photochemical effect, sufficient light of any wave-length $\lambda_n$ must be delivered, such that the amount absorbed will be the same at all values of $\lambda_n$. The relative effectiveness of different parts of the spectrum therefore depends solely on the absorption spectrum of the sensitive substance. The greater the proportion of light of a given wave-length that is absorbed, the less of that light is necessary to produce a given effect.

2. If we apply these findings to the present data, it will become apparent that Fig. 5 represents the absorption spectrum of the photosensitive substance $S$ in the reaction

$$S \rightarrow P + A$$

which is responsible for the photic sensitivity of $M_{ya}$.

Let us assume that in the position of maximum effectiveness ($\lambda_{\text{max}} = 500 \mu \text{m}$) there is complete absorption of the incident energy. At any other value of $\lambda_n$ the same amount of energy will be required to be absorbed in order to produce the same effect. But the amount of incident energy is greater than this, in inverse proportion to the magnitude of absorption at that wave-length. We know the incident energy from Figs. 4 and 5 and Table II. The absorbed energy is equal to the amount required at the maximum effectiveness ($\lambda =
500 μμ. At any other wave-length λ, then, the per cent of absorption will be given by the ratio of the energy at λ_{max} divided by the incident energy at λ_n.

The absorption at λ_{max} will then be 100 per cent, and at any other value of λ_n it will be equal to the reciprocal of the incident energy. Therefore Fig. 5 represents the absorption spectrum of the photosensitive substance S, the ordinates now being per cent of absorption. Absorption spectra are not infrequently given in this way (Henri, 1919, p. 42, 65). I shall therefore not give the absorption spectrum in terms of the absorption index. The latter can be calculated according to certain assumptions as to the thickness of the absorbing layer etc. (Cf. Bovie, 1918–19), in themselves, however, only of speculative interest.

3. Because of its interpretation as the absorption spectrum of S, the shape of the curve in Fig. 5 becomes of significance. Many known substances possess absorption bands in position and extent similar to that shown here (Cf. Uhler and Wood, 1907, Fig. 17 and those following). As a rule such curves are symmetrical with regard to the point of maximum absorption. This, however, is by no means universal, because many substances show skew absorption spectra similar to that in Fig. 5; for example, uranine, as studied by Uhler and Wood (1907, Fig. 15). In such cases it is usual to assume that there are really two, or more, vibrators in the molecule, their combined effect being given by the total curve as found. If we suppose that in the photosensitive substance S there are present two vibrators, one whose period corresponds to 500 μμ and the other to about 570 μμ, each giving a symmetrical resonance curve, the compound curve of Fig. 5 would be their resultant.

Leaving aside these speculative matters, it may be noted that the appearance of the absorption curve, though not particularly distinctive, is sufficiently so to serve as a corroboration of the identity of the photosensitive substance S in future experimentation.

v.

In recent years there has appeared a number of careful measurements of the most effective portion of the spectrum for the stimulation of different organisms (Laurens and Hooker, 1920; Loeb and
Wasteneys, 1916; Mast, 1917). Although for many animals this point lies in the yellow-green, and for many plants in the blue-green, there is an increasing number of organisms which do not conform to this classification. *Mya arenaria* clearly belongs in this non-conforming category, because it is an animal for which the maximum effectiveness is in the blue-green.

The particular significance of the above attempts at classification is due to Hess (1910). As the result of experiments with a number of animals, Hess has come to the conclusion that "the curves of the relative stimulating values of different homogeneous lights coincide, approximately or exactly, with the luminosity curve of the totally color blind human eye" (Hess, 1910, p. 362). From which he proceeds to draw certain conclusions with regard to the nature of the responses of animals to light. Loeb and Wasteneys (1916) have shown that these conclusions not only fail to agree with the facts of other investigators, but also that the process of arriving at them involves a decided logical fallacy. If after this any additional evidence were needed, the results with *Mya* certainly show that Hess' original statement is not generally true. The curve of effectiveness of the spectrum for *Mya* possesses nothing in common with the luminosity curve for the color blind or the dark-adapted human eye.

It may be added that Hess' method of experimentation is open to criticisms which invalidate his conclusions. Working with certain clams whose photic sensitivity is similar to that of *Mya*, he employs criteria of the effectiveness of light which cannot be considered quantitative. He notes the length of the extended siphon, or the extent of its retraction as the animal is moved along the projected spectrum. My experience with *Mya* has been that these are variable characteristics, even when care is taken to insure uniformity otherwise. In Hess' experiments no values are given for the energy content of the spectrum; no time is allowed for the recovery of an animal in the dark between stimulations; no measurements are made of the time of exposure; no record is made of the reaction time. In fact, few measurements of any kind are given. Even the "curves" on which the comparison with the human eye rests fail to appear in the article (Hess, 1910). Thus, even if Hess' logic were correct, his experimental findings are open to grave doubt; and the conclusions drawn from them are therefore doubly invalid.
The results with spectral lights therefore sum up to this. With *Mya* the effectiveness of the spectrum depends exclusively on the absorption spectrum of the photosensitive substance in the sense organ. The different positions of the maximum effectiveness for various organisms merely show that the sensitive substances concerned are different entities, each possessing its own absorption spectrum.

**Summary.**

1. Following the description of a simple method of securing high intensities of monochromatic illumination, it is shown that the most effective portion of the spectrum for the stimulation of *Mya* is near \( \lambda = 500 \mu \mu \).

2. The quantitative data secured is interpreted in terms of certain photochemical findings, and as a result the absorption spectrum of the photosensitive substance of *Mya* is tentatively mapped out.

Through the courtesy of Doctor M. Luckiesch of the Nela Research Laboratory, Mr. W. E. Forsythe and Mr. F. E. Cady determined the energy distribution for the 400 watt lamp which I used in these experiments. I take pleasure in thanking them for their help in this connection.

**Bibliography.**


Hecht, S., (a) Sensory equilibrium and dark adaptation in *Mya arenaria*, *J. Gen. Physiol.*, 1918–19, i, 545; (b) The nature of the latent period in the photic response of *Mya arenaria*, 657; (c) The effect of temperature on the latent period in the photic response of *Mya arenaria*, 667; (d) Intensity and the process of photoreception, 1919–20, ii, 337; (e) Time and intensity in photosensory stimulation, 1920–21, iii, 367.


Sheppard, S. E., Photochemistry, New York, 1914.


Weigert, F., Die chemischen Wirkungen des Lichts, Stuttgart, 1911.