ON THE REACTION TO LIGHT OF *MYXINE GLUTINOSA* L.

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That the effectively eyeless hag, *Myxine glutinosa*, is responsive to light was reported by Gustafson (1935). Our own interest was aroused by the behaviour of some hags (kindly sent to us by Dr Gustafson) which were being photographed. A tank containing a number of them was brightly illuminated for a few seconds only. Some 20 sec. later the animals, which had previously lain motionless in the semi-darkness, started, rather suddenly, to swim. The long delay between stimulus and response seemed to merit study while the definiteness of the response promised an important help to observation. The present paper reports the behaviour of hags under different conditions of illumination and a preliminary investigation of the sensory physiology of their responses.

METHODS

In two successive years over thirty animals were received from Sweden and from each batch twenty, as uniform in size as possible and with the largest and smallest individuals excluded, were selected for our work. Each of these animals was kept in a separate rectangular tank 24 in. long, 12 in. wide and 12 in. deep under about 3 in. of sea water. No aeration was provided, and the glass floors of the tank were left bare since *Myxine* given mud or gravel will always disappear into it. The tanks were kept in a room from which daylight was excluded and whose temperature stayed between 11 and 14° C.

For the convenience of the observers, and to permit records to be made of the behaviour of animals while in 'darkness' the room was lit by two 15W. red-painted bulbs throughout the period of the observations. Occasionally dim red light from a hand torch was used to supplement the general illumination. The background illumination did not reach 10 ca.m. (candle metres) at the floor of any of the tanks. We shall hereafter refer to animals as being in the dark when their only illumination was from these sources.

To test their responses the animals were illuminated, one at a time, by means of electric light bulbs mounted in a hood which could be rested on top of the tanks. While lit in this way an animal could be watched through the side of its tank. This method does not provide a uniform light intensity over the floor of the tank and to this is due one of the uncontrolled variables in our observations. Another is

a consequence of the different positions adopted by animals when at rest during stimulation by light. They usually lay on their sides, but sometimes lay on their bellies, or even, rarely, on their backs, and the aspect presented to the source of light varied accordingly.

When in the dark under these conditions our hags normally lay quite still. That 'spontaneous' movements do occur was, however, very evident and so before any animal was subjected to a light treatment it was watched for 1 min. in the dark. If during that time it made any visible movement it was deserted for at least 15 min. and then watched for a further period of 1 min. In no case was it necessary to defer an observation on an animal more than twice.

After treatment with light animals were usually allowed a minimum of 30 min. in the dark to recover, but for certain experiments this period was reduced to 15 min.

OBSERVATIONS

(1) *The activity of animals in the dark*

We have two measures of the frequency of 'spontaneous', i.e. non-induced, movement in our animals. In the first place we made a record of each occasion upon which an observation had to be deferred because of movement during the 1 min. pre-treatment period. In the second place we watched each of one batch of animals continuously for 1 hr. in the dark and recorded every spell of activity shown.

Thus on 679 occasions we approached one or other of a group of twenty animals for the purpose of testing its responses to light. Of that number forty-two approaches were deferred because the animal moved during its 1 min. period in the dark, or was moving when first approached. Of the forty-two deferred approaches three represent observations twice deferred. There seem to be real differences between the levels of spontaneous activity of the animals when measured in this way, but there is no obvious correlation between this and the differences in the responses to light which also appeared.

When twenty animals were watched continuously for an hour in the dark ten of them remained still for the whole of that period. Of the other ten, four engaged in a single burst of swimming, five in two separate bursts, and one in three separate bursts. In other words, swimming occurred 17 times in 20 'animal-hours' of observation.

Such movements in animals under observation could, of course, be mistaken for responses to light stimulation. Their effect will then be to produce mean reaction times lower than the correct figures or to indicate a response where none is evoked. This source of error would clearly be serious where the reaction time approached the mean interval between spontaneous movement. As, however, we were always concerned with reaction times of less than 5 min. (and usually less than 1 min.) we can safely ignore this source of error, although we believe that the very few of our measurements of reaction times that are strikingly less than the mean of their kind may be accounted for in this way.

(2) *Response of* Myxine *to general illumination*

The illumination of *Myxine* elicits general locomotory activity which begins sometimes with a local response at the head or tail but more often as a general stirring of the whole animal. These movements are always sharp and follow a period, never less than a few seconds, during which the animal remains still. Active locomotion may take some time to develop after these first stirrings. The hag in the dark usually lies on its side. On illumination it gets on to its belly and then, after intermittent stirring assumes a swimming posture by throwing its body into waves. Soon afterwards the full locomotory response emerges. Once this happens the animal may swim for several minutes, making periodic attempts to burrow. We shall not concern ourselves further with these later phases in the

Table 1. *Reaction times of twenty animals to general illumination at* 344 *ca.m.*

Animal	Time to first response (sec.)					Mean
A	30	34	37	24	22	29·4
B	14	17	14	12	20	15·4
C	6	7	14	9	11	9·4
D	36	22	27	19	17	24·2
E	24	47	56	23	24	34·8
F	20	21	32	14	18	21·0
G	15	12	22	16	20	17·0
H	20	18	13	18	26	19·0
I	15	17	18	21	15	17·2
J	32	33	32	27	23	29·4
K	23	24	34	20	29	26·0
L	8	16	17	21	15	15·4
M	29	19	29	13	27	23·4
N	13	16	14	17	21	16·2
O	33	22	23	18	16	22·4
P	8	14	19	9	18	13·6
Q	13	14	14	13	13	13·4
R	12	13	11	14	11	12·2
S	20	12	13	29	11	17·0
T	28	21	33	43	49	34·8
Total	399	399	472	380	406	—
Mean	19·95	19·95	23·6	19·0	20·3	20·56

behaviour of an animal aroused by light. But the sharpness of the first response makes it possible to use this in measuring reaction-time, and most of our work has been concerned with the measurement of this reaction time under various conditions.

Reaction times of a group of twenty selected animals illuminated 5 times by a 40 W. lamp (mean intensity 344 ca.m. at the floor of the tank) are given in Table 1. From the mean reaction time of about 20 sec. we see that *Myxine* is slow to respond even to fairly intense illumination. Some corresponding figures from other animals at intensities of this order are: ammocoetes, 2 sec. (Young, 1935; Steven, 1950); tadpoles of *Rana clamitans*, 2 sec. (Obreshkove, 1921); *Ciona*, 5–10 sec. (Hecht, 1918); *Mya*, 1·3–1·5 sec. (Hecht, 1919*a*). Only *Proteus* (Hawes, 1945) has a comparably slow response.

Table 1 also shows how much the reaction time can vary under our experimental conditions, even when the light source remains the same. This variation may be

due in part, and perhaps in the main, to differences of intensity in different parts of the tank and differences in the lighting of an animal according to the way it lies. However, the means of the separate series of the entire group do not depart far from the general mean for the five series taken together.

It is also clear from Table 1 that there is a big variation in reaction time from one animal to another, some are clearly 'fast' and others 'slow'. Thus animals E and T gave mean reaction times 3–4 times as long as C or R. For this reason, in certain work to be described later, we confined observations to a few, and in some cases to single, animals whose performance had been established as relatively consistent.

Intensity/time data

It was to be expected that the response of *Myxine* to light would vary with the intensity of illumination, and tests showed that though the character of the response is the same over the whole range of intensities that give responses at all, the reaction time varies inversely with intensity.

Our data on the effects of varying intensity on the reaction time fall into three groups: (1) results on twenty selected animals whose reaction times at a single intensity were given in Table 1; on these we only obtained three readings for each animal at each of four intensities; (2) results on five animals with eight readings at each of five intensities; (3) results on one animal with twenty-five readings at each of seven intensities. The results of each of these are given graphically in Fig. 1 *A–C*, and the numerical data for Fig. 1 *C* are given in Table 2. The picture is the expected one. At high intensities the reaction time approaches a minimum and, in this region, big increases in intensity have little effect upon it. At lower intensities the reaction time is a matter of minutes and it becomes difficult to be certain that one is not dealing with a spontaneous movement.

Strict adherence to the Bunsen–Roscoe Law is not to be expected, since we know from Hecht's work (1918, 1919*a*) on *Ciona* and *Mya* that in them reaction time in a response to light is made up of two components, of which one alone varies with intensity in the Bunsen–Roscoe manner. Thus if we extract from Table 2 the data on the three lowest intensities we find that the products of reaction time and intensity are 162, 146 and 131 approximately, and not nearly as constant as the Bunsen–Roscoe Law demands.

Sensitization time and latent period

As in the response to light in other animals, it is not necessary to illuminate *Myxine* for the full period of the reaction time to get a normal response. It is clear, therefore, that the reaction time is composed of the two components of Hecht mentioned above, sensitization-time—the period of exposure to light necessary for response in minimum time; and latent period—during which the animal need not be illuminated. We tested the reaction times of our groups of twenty animals with illumination of limited duration, and the results are set out in Table 3. One can reduce the period of illumination to 5 sec. at this intensity (344 ca.m.) without any reduction in the reaction time. If further reduced to 1 sec. most of the animals still

respond but only after a much longer time. Even with a flash of approximately 0·1 sec. duration, ten out of the twenty animals responded, usually from 2 to 5 min. later.

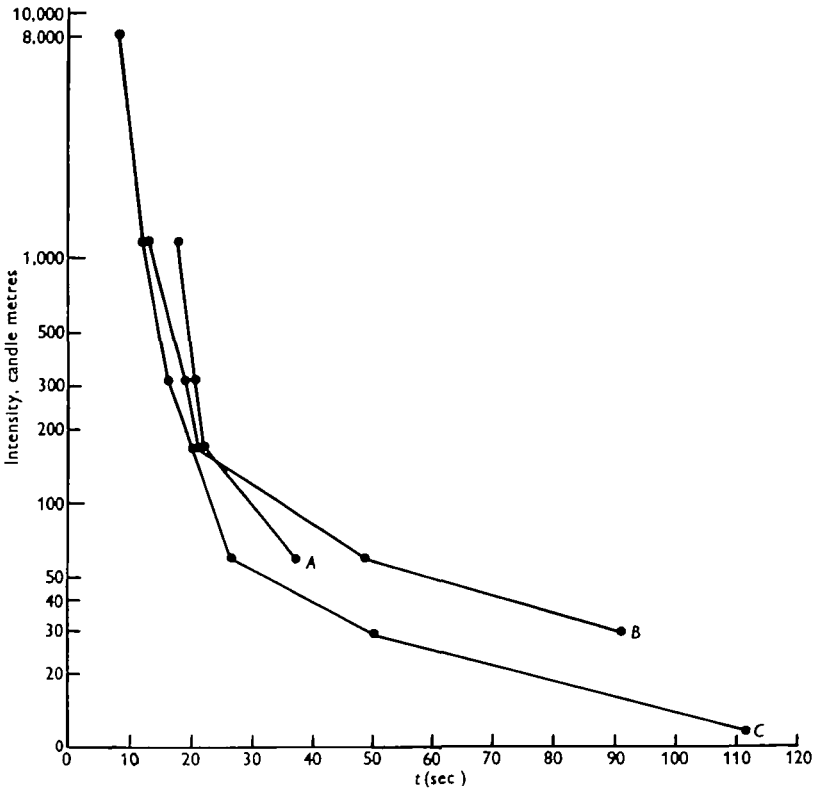

Fig. 1. The relation between reaction time and intensity. *A*, means of twenty animals and three readings at each intensity; *B*, means of five animals and eight readings at each intensity; *C*, means of twenty-five readings at each intensity on a single animal.

Table 2. *Reaction times of animal F to general illumination at different intensities. Means of twenty-five observations at each intensity with standard deviations*

Intensity (ca.m.)	Mean reaction time (sec.)
8650	8·8 ± 1·60
1258	12·3 ± 1·65
344	16·3 ± 2·18
184	20·5 ± 2·06
65·6	26·5 ± 4·06
31·2	50·4 ± 10·4
12·6	111·7 ± 30·0

It was important to determine how much of the reaction time was taken up with sensitization and how much with latent period over a wide range of intensities, and especially in the long reaction times with low intensities. We did this simply by

finding the shortest periods of illumination that would still give reaction times of minimal value at several intensities.

Our most complete data on sensitization are based on three animals only, and for one of these sensitization was determined at a number of different intensities. The first two of these animals yielded the results given in Table 4. Inspection of these results indicated a sensitization time of 5 sec. in both cases, with latent periods of 21 and 22 sec. Here the latent period is by far the longer of the two components, and most of the long reaction time is due, not to the photochemical process, but to the time taken by the products of the photochemical reaction to initiate the response. It is a little unfortunate that these two animals had almost identical mean reaction times so that they cannot tell us whether differences between animals are to be

Table 3. *Reaction times of twenty animals to general illumination at* 344 *ca.m. with varying exposure periods*

Duration of illumination (sec.) ...	15	10	5	1	Flash 0·1 sec. approx.
Mean reaction time (sec.)	31·25	31·55	31·84	72·06	177·22
No. of animals responding	20	20	19	16	9

Table 4. *Reaction times (sec.) of animals D and N to general illumination at* 344 *ca.m. with varying exposure times (means of* 20 *tests)*

Duration of illumination	Until roused	10	9	8	7	6	5	4	3	2	1
RT Animal D	26·0	24·2	—	—	—	—	22·9*	36·5	40·1	45·6	56·5
RT Animal N	26·8	24·4	26·1	27·6	25·4	28·5	29·6*	55·8	—	—	—

* Probable sensitization period.

attributed to differences in sensitization period or latent period. Our third animal, whose performance at different intensities is examined below, can contribute something on this point. At the intensity (344 ca.m.) used in determining sensitization in the other animals it had a reaction time of 16 sec. and a sensitization period of 6 sec. This result suggests that sensitization time is roughly constant from animal to animal and that it is the latent period that is responsible for the big and consistent differences between animals that were noted in an earlier section.

Table 5 consists of sensitization data from animal F at five different intensities. Five readings of reaction time were taken for each exposure at each intensity. The simplest way of expressing sensitization time from the figures is to take it as the shortest of the exposure times giving a reaction time not appreciably longer than the reaction time when the light remains on until the animal is roused. At 344 ca.m. this would be 6 sec., though it is not until one reduces the exposure time to 3 sec. that all the reaction times in a series exceed the minimal figure. Perhaps the halfway position of 4·5 sec. between the exposure time when delayed responses first

appear (6 sec.) and that when all responses are delayed (3 sec.) would give a closer approximation to the true sensitization period. However, the sensitization periods given in the table for the five different levels of intensity are those obtained by the method of inspecting the means and picking out the last one that still gives near-minimal reaction times.

The figures show that while the highest intensities of all failed to speed up sensitization period as much as one might have expected, the effect is very clear at moderate and low intensities. The latent period varies much less, but it is not constant and on this we shall comment in the discussion. Thus *Myxine*'s comparatively slow reaction at higher intensities is mainly taken up by the latent period. But the exceptionally slow reactions at the lower intensities are mainly due to the time required for sensitization.

Table 5. *Reaction times, sensitization periods and latent periods of animal F at five intensities. Reaction times are means of five readings*

Intensity (cm).													Sensitization period	Latent period
1258	Exposure time	UR*	5	4	3	2	.	.	.	.	.	.	5	9·8
	Reaction time (sec.)	14·8	16·2	20·8	26·0	†	.	.	.	.	.	.		
344	Exposure time	UR*	10	9	8	7	6	5	4	3	2	.	6	9·8
	Reaction time (sec.)	15·8	15·6	15·8	15·6	15·8	16·2	18·6	20·2	25·6	†	.		
184	Exposure time	UR*	15	14	13	12	11	10	.	.	.	.	14	11·4
	Reaction time (sec.)	25·4	24·6	24·2	31·6	32·0	37·2	†	.	.	.	.		
65·6	Exposure time	UR*	28	27	26	25	24	23	22	21	20	19	26	11·0
	Reaction time (sec.)	37·0	35·8	41·4	41·6	57·8	46·8	51·8	59·6	78·0	58·2	†		
31·2	Exposure time	UR*	44	42	40	38	36	34	.	.	.	.	42	16·2
	Reaction time	58·2	64·0	52·4	76·8	81·2	†	†	.	.	.	.		

* Until roused.
† Less than five responses obtained.

At the same time we have an example of the prolongation of the reaction time by illuminating for periods far below the sensitization period. Indeed it is difficult to get down to an exposure time at any intensity when the reaction fails altogether. When Hecht encountered this effect in *Mya* (1919*b*) and in *Ciona* (1926) he overlooked, in our opinion, certain of its implications. He simply regarded it as the lengthening of the latent period. For reasons that we shall go into later we find it unsatisfactory to apply the term latent period to the very different conditions that exist when exposure time falls far short of the sensitization period. What is noteworthy in this effect is the essential unity of the sensory process in spite of its two stages. Thus one can get the same result in terms of reaction time from a very brief exposure at a high intensity as one gets from prolonged exposure at a lower one.

Other factors affecting reaction time

A number of factors besides intensity influence reaction time. As might be expected, temperature has an effect. Thus an animal that reacted to light at 344 ca.m. after 36 sec. at 12·2° C., reacted after 62 sec. at 6·7° C. Another which reacted at 14 sec. at 11·7° C. did so after 32 sec. at 4·0° C. Such a sensitivity to temperature again indicates the large proportion of the reaction time that is taken up by non-photochemical reactions.

Exposure to light also has the effect of slowing down the responses for some time afterwards, and if the illumination is continued for a long time, e.g. half an hour, the animal can be made temporarily altogether insensitive. The return of sensitivity after such treatment is slow, as we can see from the following experiment on ten hags. These were exposed to continuous illumination at 344 ca.m. for 30 min. Then after 2 min. in the dark only one animal responded to a period of illumination lasting 2 min. After 5 more minutes in the dark, 2 animals responded, after 10 further minutes nine animals responded; after 15 more minutes again only nine

Table 6. *Reaction times of animals F and N (means of five readings) before and after illumination at* 344 *ca.m. for* 1 *min.*

	Animal F (sec.)	Animal N (sec.)
Before illumination	13·7	28·7
1–3 min. after illumination	25·0	47·6*
3–6 min. after illumination	17·2	30·7
6–9 min. after illumination	15·6	26·9
9–12 min. after illumination	14·2	29·1

* Mean of three readings only.

responded and it was only after another 30 min. in the dark that all ten had recovered their sensitivity to light. The mean reaction times of the animals responding after 10, 15 and 30 min. in the dark were 117, 67 and 54 sec. respectively, compared with 18·1 sec. at the beginning. Clearly a long time in the dark is required for return to normal sensitivity after such treatment.

The effect of shorter periods of illumination on reaction time was also investigated. The animals were illuminated for 1 min. and then the reaction times to further exposures after certain times had elapsed were measured. As readings could not be taken until the animals had come to rest, it was not always possible to do tests within 3 min. of the first exposure. The results from two of these animals are given in Table 6. They show that it can be assumed that reaction time is back to normal, in other words, that dark adaptation is complete, within a few minutes after exposures of the kind used in our experiments, and that an interval of 15 min. between successive readings was long enough to permit comparable results to be obtained.

Surprisingly, the reaction to light is delayed if the animal is already active when the illumination begins. Thus if a hag is roused by sharp tactile stimulation and

then illuminated while it is still swimming or in a swimming posture, the response to light follows much later than in the resting animal. Ten such tests were carried out with one of our animals and gave the following results: mean reaction time of ten controls at 344 ca.m. illuminated at rest—20·3 sec., mean reaction time of same ten animals when illuminated while response to tactile stimulation still in progress—40·2 sec. It may seem strange that one can still determine the reaction time to light under these conditions. In fact the activity induced by a tactile stimulus was usually sufficiently short-lived for the hagfish to relax completely after the light was turned on. Then after lying still for a time, the typical response to light developed. Evidently the reaction time to a light stimulus can be lengthened by the central nervous system. This is a warning against believing this kind of behaviour to be automatic and invariable.

Response to local illumination

By using Perspex rods and a 36W. light source it was possible to explore the surface of the animal to find out where the light sense is located. As a preliminary, twenty animals were examined with a rod 3·8 × 0·8 cm. in cross-section. Fig. 2*a* shows the areas illuminated by this rod, the head, the branchial apertures, the cloaca and the tail. All responded to local illumination on the head, the tail and the cloaca, but at the branchial apertures only ten responses were obtained. The intensity on these illuminated patches was 790 ca.m., and the reaction times somewhat longer than with general illumination of this intensity. It seemed that *Myxine* had regions of high sensitivity to light anteriorly and posteriorly, and areas of lesser sensitivity in the mid-body region.

With rods which illuminate smaller patches it was possible to delimit the light-sensitive areas more precisely. A rod 2 × 0·8 cm. in cross-section gave the following picture on the same group of twenty animals:

Anterior head	20 responses
Posterior head	10 ,,
Branchial apertures	2 ,,
Midway branchial apertures and tail	2 ,,
Cloaca	19 ,,
Tail	7 ,,

Using a third rod of dimensions 1·05 × 0·8 cm. in cross-section two hags were explored along the whole of their lengths. The results are shown in Fig. 2 on the outline drawings of these animals. Moving the rod along the body gave twenty-six bands 0·8 cm. wide in animal D and twenty-four in animal F. Each band was illuminated 5 times. Except where there are definable landmarks, e.g. branchial apertures, tail tip, cloaca and mouth, the location of the bands varied a little from one series of readings to another, but it so happens that it was these most easily marked regions that proved to be most important. The number of responses to the five tests is indicated in each band on the drawing and the mean reaction time below.

These results show that *Myxine*'s light sense is located mainly near the anterior end and in the cloacal-caudal region. Probably *Myxine* has light-sensitive end-organs over most of its body, which are dense at certain places in the head and in the cloacal and tail region, but sparse elsewhere. Presumably, if local illumination is to rouse the animal a minimum number of these end-organs must be excited. Thus light from the smaller rods fails to rouse if directed anywhere between points just posterior to the mouth and anterior to the cloaca. Even in the more sensitive areas, the reaction to light fails if only very tiny areas are illuminated. Thus when

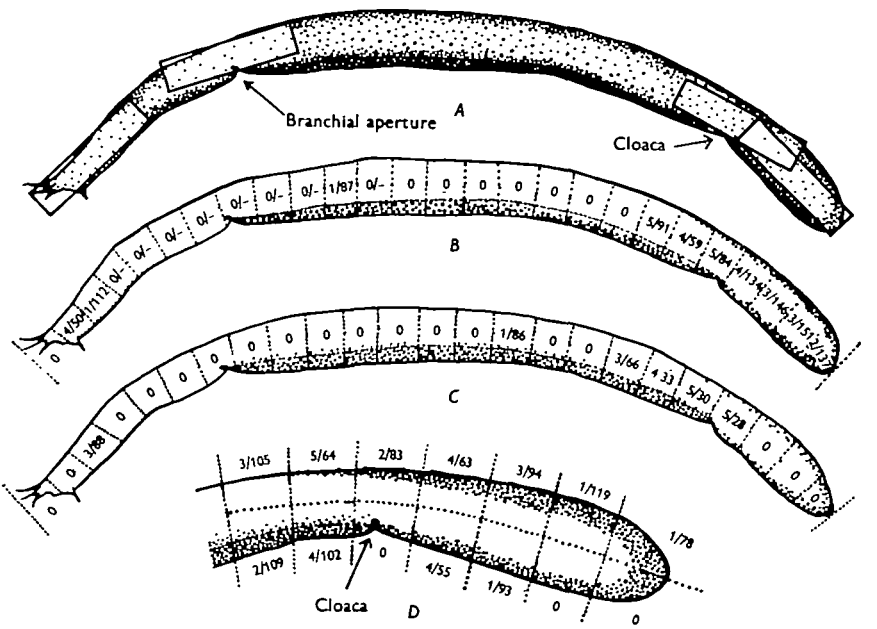

Fig. 2. Local illumination of *Myxine*. *A*, areas illuminated with rod 3·8 × 0·8 cm. cross-section; *B*, areas illuminated on animal D with rod 1·05 × 0·8 cm. cross-section; *C*, areas illuminated on animal *F* with rod 1·05 × 0·8 cm. cross-section; *D*, areas illuminated on posterior region of animal D. In *B*–*D* and *D* the figures refer to the number of responses obtained in five trials and to the mean reaction time in seconds where responses occurred.

the skin was explored with a rod 0·4 × 0·4 cm. in cross-section, no response occurred even when this was directed on to the sensitive places in the head and near the cloaca.

The region of the cloaca and the tail is the most extensive area of high sensitivity to light. In the ammocoete the main light-sensitive organs occur in the tail (Parker, 1905; Young, 1935; Steven, 1950), but in *Myxine* the immediate vicinity of the cloaca is the most sensitive place of all. Tests using a smaller rod showed that the lips of the cloaca itself are not light sensitive. This point is illustrated by Fig. 2*D* in which the results of separate dorsal and ventral illumination of the last 7 bands into which this animal (D) may be divided are shown on an outline drawing.

The light-sensitive area in the head is more restricted in extent, apparently not more than 0·8 cm. wide in either of the two animals D and F. While these, and most

other animals, gave no sign of sensitivity to light at the extreme anterior end, we did encounter one remarkable animal which always responded to local illumination here within 4–8 sec.

The highly sensitive region in the head is well in front of the vestigial eyes. While it might almost be assumed on general anatomical grounds, e.g. the poor optic nerves, that the eyes are not involved in the response, it seemed wise to test the matter. The eyes of two hagfish were destroyed under urethane anaesthesia. Both continued to respond to local illumination on the head. This, and the location of the light-sensitive region in the head, shows that the light sense here is non-optic.

It is generally true that our hagfish reacted more quickly to general than to local illumination. But perhaps more important than the quantitative differences in the reaction time are certain qualitative differences in the kinds of reactions resulting from the two kinds of illumination. While it is true that many animals gave the typical response after local illumination at the posterior end, in many other cases they made only local responses, a flexing of the tail or some other movement that did not involve the rest of the body. These local movements were a common feature of the response; and they always took the same form in those animals which showed them.

The role of the nervous system in the response of Myxine *to light*

It has been implied in the earlier sections that the response to light takes its origin from specifically light-sensitive end-organs situated in the skin. The possibility that changes in light intensity are acting directly on the central nervous system cannot however be ruled out, especially in view of Young's (1935) evidence for certain responses of ammocoetes to light—and relatively slow ones at that—being due to the light acting directly on the spinal cord. Indeed the fact that *Myxine* is most sensitive to light at the head and near the tail, is consistent with direct excitation of this kind since these are the places where the central nervous system is nearest the body surface.

Two kinds of experiment were carried out to test this matter. In one, a length of brain or spinal cord was exposed and its sensitivity to direct illumination was tested. In the other, the skin was removed over several centimetres in front of the cloaca and the sensitivity to light within this skin-free area compared with that of the light-sensitive region just behind it. In these tests, the animals were, as usual, immersed in sea water.

In the first experiments the brain and the anterior end of the spinal cord were exposed and then illuminated locally with our Perspex rods in the usual way. If we are dealing with direct effects of light on the spinal cord and brain, it would be fair to assume that the removal of the skin and other tissues covering these parts of the central nervous system would increase the effective intensity of the incident light and so reduce the reaction time. In the event the two animals tested in this way had reaction times to illumination on the operated region that were more than doubled by the operation.

Table 7 shows the results of the experiments in which the skin was removed from

an area just in front of the cloaca known to be highly sensitive to light before the operation. The figures show quite clearly that the light sense goes with the skin. We are satisfied that the response to light in *Myxine* cannot be attributed to direct stimulation of any part of the central nervous system.

The chief neurological problem raised by this demonstration of photo-sensitivity in the skin at the posterior end lies in establishing the sensory pathways from the photo-receptors to the central nervous system. Added interest is attached to this problem in *Myxine* by the fact that the light receptors in the tail of *Lampetra* have

Table 7. *Responses of* Myxine *to local illumination before and after removal of skin anterior to cloaca*

Before operation						After operation					
Region for skin removal			Region to be left intact			Skin-free region			Skin intact		
No. of trials	No. of responses	Mean *RT* (sec.)	No. of trials	No. of responses	Mean *RT* (sec.)	No. of trials	No. of responses	Mean *RT* (sec.)	No. of trials	No. of responses	Mean *RT* (sec.)
24	24	25·3	23	23	41·7	29	4	155·2	31	26	56·1

Table 8. *Responses of* Myxine *to local illumination after transection of the spinal cord*

Head illumination			Cloacal-caudal illumination		
No. of trials	No. responses from head	No. responses from tail	No. of trials	No. responses from head	No. responses from tail
36	36	0	50	2	46

Table 9. *Responses of* Myxine *to general illumination after transection of the spinal cord*

No. of trials	No. responses from head	No. responses from tail	Mean *RT* head (sec.)	Mean *RT* tail (sec.)
33	33	32	42·4	35·1

been shown to be innervated through the lateral-line nerves to the tail (Young, 1935). This is shown in the ammocoete by the fact that the head alone responds when the tail is illuminated after transection of the spinal cord. This response disappears when the lateral line nerves are severed.

The spinal cord was transected under anaesthesia in two groups of five hagfish in successive years. The results appear in Table 8. It can be seen at a glance that the situation in *Myxine* is quite unlike that in the ammocoete. In spinal *Myxine*, if the head is illuminated, only that part of the animal anterior to the cut responds; posteriorly the animal remains quite passive. If the cloacal-caudal region is illuminated, only that part of the animal posterior to the cut responds. The situation is revealed equally clearly when spinal animals are exposed to general illumination.

Then, as Table 9 shows, independent responses occur in the two halves of the animal. In the spinal ammocoete, general illumination only causes responses of the head.

Thus the spinal cord in *Myxine* provides the pathway along which impulses from the photo-receptors near the cloaca travel anteriorly to the brain. But even when the posterior part of the body is isolated by spinal section, local responses are still possible through local reflex arcs which take slightly different forms in different individuals. There is none of this in the ammocoete. This evidence that the spinal nerves conduct the excitation from the photo-receptors to the central nervous system reveals an arrangement hitherto not known with certainty in a vertebrate. For while it is true that other cases of sensitivity to light in the skin of vertebrates (blinded tadpoles, *Proteus*, blind fish, etc.) may well be due to innervation through

Table 10. *Responses of* Myxine *to local illumination before and after severance of spinal nerves anterior to cloaca*

Before operation						After operation					
Region of operation			Region to be left intact			Operated region			Intact region		
No. of trials	No. of responses	Mean *RT* (sec.)	No. of trials	No. of responses	Mean *RT* (sec.)	No. of trials	No. of responses	Mean *RT* (sec.)	No. of trials	No. of responses	Mean *RT* (sec.)
15	15	24·8	15	15	37·2	21	2	78·0	21	18	70·7

the spinal nerves, this has not been established. In an additional experiment we exposed a length of spinal cord and stripped it of all its connexions with surrounding tissues severing ventral as well as dorsal roots. The operated region was usually taken just in front of the cloaca, or even anterior to that if such a region proved to be sensitive to light in preliminary tests. After the operation the sensitivity of the operated region was compared with the intact cloacal-caudal region just behind. The results obtained from four animals are given in Table 10. The skin of a region operated upon in this way loses its sensitivity, but there is a good deal of spontaneous activity after such drastic operations and this, in our opinion, explains the inconsistencies in the results. This is additional evidence that the spinal nerves serve the photo-receptors of the cloacal-caudal region.

Nothing has been done, because of the technical difficulties involved, to trace out the innervation of the much more restricted light-sensitive region in the head. However, in some of our animals it must be said that the head is the more sensitive of the two regions. Several of them had to be rejected for some of the experiments which required high sensitivity to light in the posterior region because only the head responded to the local illumination used in these tests.

It would seem that photoreception in *Myxine* is a general skin sense subject to individual variation in sensitivity and distribution, its connexions with the central nervous system are provided by the sensory roots of the spinal nerves in the posterior region, and probably by analogous sensory fibres in one of the cranial nerves in the head.

DISCUSSION

Myxine provides an example of a non-visual reaction to light which apparently conforms to the general principles derived by Hecht from work on more rapidly responding invertebrates. These are that the reaction time is composed of a photo-

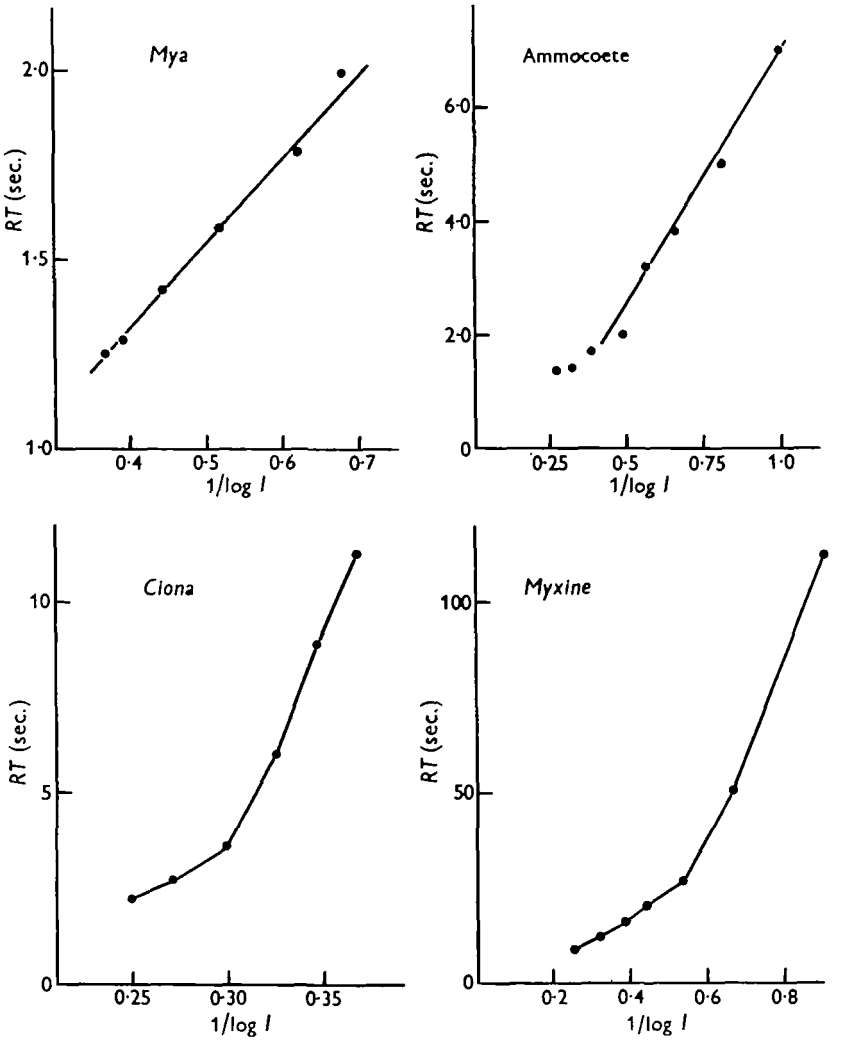

Fig. 3. The relation between the reaction time and the reciprocal of the logarithm of the intensity in *Mya*, ammocoete, *Ciona* and *Myxine*. The curves for *Mya* and ammocoete are redrawn from Hecht (1921), fig. 2, and Steven (1950), fig. 4, respectively. The curve for *Ciona* is constructed from Hecht (1918), table 2. The curve for *Myxine* is constructed from the data of Table 2 of this paper.

chemical phase, whose intensity/time relations are roughly in accordance with the Bunsen–Roscoe Law, and a non-photochemical phase appearing as a latent period, taken up with certain secondary processes. In considering our results with *Myxine*,

however, we have found it necessary to modify the conception of the latent period originally proposed by Hecht.

Hecht's latent period is not a quantity that can be measured directly. It is a difference, a quantity obtained by subtracting a period of exposure to light from a total reaction time. We have seen that in *Myxine*, as in *Ciona* and *Mya* on which Hecht worked (1918, 1919*a–c*), there is a certain critical exposure time, the sensitization period, beyond which further exposure does not shorten the reaction time at any given intensity. At this exposure the reaction time is minimal, and according to Hecht (1918) the latent period obtained by subtracting the sensitization period from the reaction time is constant in *Ciona* over a considerable range of intensities. We were a little surprised, therefore, when our own values for the latent period in *Myxine*, with full sensitization, were not constant over our range of intensities. They went from 15·2 sec. at the lowest intensity (31 ca.m.) to 9·8 sec. at the highest (1258 ca.m.). With even more intense illumination (8650 ca.m.) the reaction time, of which the latent period is a part only, fell to a mean value of 8·8 sec. Our determinations of the latent period were only rough approximations, yet the progressive and consistent shortening of this difference between critical exposure time and reaction time needs explanation.

Further consideration suggested, however, that a constant latent period with varying intensity is only to be expected when sensitization is almost instantaneous as in *Mya*. Indeed this follows from Hecht's own conception of the nature of the photosensory process, as a coupled photo-chemical reaction in which, to quote his (1919*b*) summary:

> The events which happen in the sense organ of *Mya* when it is stimulated by light may, according to our findings, be expressed as follows. The photosensitive substance (*S*), originally formed from its two precursors (*P* and *A*—Precursor and Accessory), is changed back into them under the influence of light, both reactions being given by the expression $S \underset{\text{dark}}{\overset{\text{light}}{\longleftrightarrow}} P-A$. This happens during the exposure to light or during the sensitization period when the exposure is prolonged. One or both of the freshly formed precursor substances then immediately serves to catalyse the transformation of an innocuous material (*L*) into a stimulating substance (*T*). This occurs during the latent period. When a sufficient amount of the stimulating substance (*T*) has been accumulated, it acts on the nervous connections to the sense organs and initiates the retraction of the siphon. The entire sensory process may therefore be summed up in the following two reactions:
>
> $$S \rightarrow P+A; \quad L//P+A// \rightarrow T,$$
>
> in which the symbol $//P+A//$ signifies catalysis by one or both of the precursor substances.

The weakness of this conception lies in its assumption that the latent period and the reaction $L \rightarrow T$ are co-extensive in time. In practice this reaction will surely not wait until the completion of the photochemical reactions $S \rightarrow P+A$ before itself beginning, and it seems to us better to regard these two reactions not as consecutive but as contemporary or, at least, overlapping. The latent period is then not the entire duration of the reaction producing T but simply the time required, over and above the sensitization period, to produce T.

If this is so then changes in intensity will affect not only the sensitization phase but through it the latent period also. Hence one may expect the latent period to

change with intensity in a way which while unimportant where sensitization is very brief (e.g. in *Mya*) is marked where this is relatively long (e.g. in *Ciona* and *Myxine*).

A further complication, which we do not profess to explain, must be the cause of a feature of the intensity/time relationship in *Ciona*, the ammocoete and *Myxine*, which has not before received attention. For if the sensitization process obeys the form:

$$\text{Photochemical effect} = k \times \text{duration of stimulus} \times \log \text{intensity},$$

where k is a constant, then the curve of reaction time against the reciprocal of the log intensity should be a straight line cutting the time axis at a value corresponding to the latent period where this is a constant. In the case of *Mya* the data fit this expectation well, but inspection of Steven's (1950) curve for the ammocoete shows that while the relation is linear for low intensities it is impossible to derive a real value for the latent period from this part of it. Hecht's figures for *Mya* and *Ciona*, Steven's for the ammocoete and our own for *Myxine* are plotted in Fig. 3. In *Ciona* and *Myxine* the upper (low intensity) part of the curve is, or may well be regarded as, straight; but if produced cuts the time axis at a negative value. The lower (high intensity) part departs quite markedly from the linear. Steven's data are quite consistent with a similar situation in the ammocoete, although he did not work with intensities sufficiently high to bring the animals well into the non-linear part of their reactions. If, then, Hecht's picture of the reaction time only provides an adequate description of events where the sensitization period is short compared with the total reaction time, our own modification is still unable to explain the actual situation in *Ciona*, the ammocoete and *Myxine* where sensitization is relatively long. We do not think that the data at our disposal are sufficiently accurate and detailed to make it worth while fitting a deduced curve to them and we must reluctantly leave this matter unexplained.

We may here comment briefly on the biological significance of *Myxine*'s light sense. Parker (1909) believed that dermal photo-reception was found only in those fish that were predominantly fresh water in habitat, and he was driven by this to revise his earlier views on the evolutionary origin of the vertebrate eye. Here it is only necessary to point out that *Myxine*, by robbing Parker's premise of its general validity, must weaken his argument from it.

From the comparative point of view it is of some interest to discover that though each of the groups of living cyclostomes is light sensitive at the tail end of the body, the innervation of the sense organs is different in each case. This makes it possible that the light sense was evolved independently in lampreys and hags. The alternative is that one mode of innervation replaced the other in one of the two groups, but we think it more likely that the two senses are of independent origin and testify to the ability of the vertebrate skin to evolve photoreceptors and the peripheral nervous system to innervate them in situations in which they meet the needs of the animal. Possibly the light sense in *Myxine* developed in animals that lacked a lateralis system in the tail and trunk and hence could only be innervated by spinal nerves, while in lampreys a fully functional lateralis system permitted an alternative

nervous pathway. It would certainly be of great interest to know whether lateral line nerves do serve dermal photoreceptors in any other aquatic vertebrates.

There are obvious differences, however, in the relative importance of the light sense to the lampreys and hags. The light sense of the lamprey tail has an important and well understood role to play in the life of the ammocoete at least, but it is possible to doubt the value of *Myxine*'s sense both because it is so feeble and because many *Myxine* are caught in water so deep that it must there be inoperative. We are not, however, inclined to dismiss the light sense as a functionless evolutionary survival in *Myxine*, as Hawes felt bound to do in the rather different case of *Proteus*. The matter is discussed fully by Steven (1955) on the basis of his findings on the spectral sensitivity of the light sense, but for our part we feel that the very localization of the sense points to a continuing role of light perception in the life of those hags inhabiting, or visiting, relatively shallow waters. Its effect would seem to be, if we argue from behaviour in the laboratory, to keep the hag buried during daylight hours and in particular to ensure that the tail as well as the head is withdrawn beneath the surface of the sea-bed. It may also be expected to deter the movement of hags from deeper to shallower waters, but discussion of such points only emphasizes our ignorance of *Myxine*'s normal mode of life.

SUMMARY

1. *Myxine glutinosa* responds to illumination by active locomotory movements.

2. The response to light occurs some time after the onset of illumination. This time can be resolved, after the method of Hecht, into a sensitization period and a latent period.

3. Analysis of the relation of sensitization period and latent period to intensity of illumination and other factors shows that photoreception in *Myxine* is essentially similar to that of a number of other animals, including the ammocoete, but suggests that the secondary reactions initiated by the production of photolytes during sensitization occur during both sensitization and latent periods and not during the latent period alone.

4. The photoreceptors of *Myxine* are located in the skin and are present only, or mostly, at the anterior end of the head and in the region of the cloaca. Nervous impulses travel from the posterior photoreceptors through spinal nerves to the spinal cord.

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REFERENCES

GUSTAFSON, G. (1935). On the biology of *Myxine glutinosa* L. *Ark. Zool.* **28**A (2), 1–8.

HAWES, R. S. (1945). On the eyes and reaction to light of *Proteus anguinus*. *Quart. J. Micr. Sci.* **86**, 1–53.

HECHT, S. (1918). The photic sensitivity of *Ciona intestinalis*. *J. Gen. Physiol.* **1**, 147–66.

HECHT, S. (1919*a*). Sensory equilibrium and dark adaptation in *Mya arenaria*. *J. Gen. Physiol.* **1**, 545–58.

HECHT, S. (1919*b*). The nature of the latent period in the photic response of *Mya arenaria*. *J. Gen. Physiol.* **1**, 657–66.

HECHT, S. (1919*c*). The effect of temperature on the latent period in the photic response of *Mya arenaria*. *J. Gen. Physiol.* **1**, 667–85.

HECHT, S. (1921). Time and intensity in photosensory stimulation. *J. Gen. Physiol.* **3**, 367–73.

HECHT, S. (1926). The effect of exposure period and temperature on the photosensory response in *Ciona*. *J. Gen. Physiol.* **8**, 291–301.

OBRESHKOVE, V. (1921). The photic reactions of tadpoles in relation to the Bunsen-Roscoe Law. *J. Exp. Zool.* **34**, 235–79.

PARKER, G. H. (1905). The stimulation of the integumentary nerves of fishes by light. *Amer. J. Physiol.* **14**, 413–19.

PARKER, G. H. (1909). The integumentary nerves of fishes as photoreceptors and their significance for the origin of the vertebrate eyes. *Amer. J. Physiol.* **25**, 77–80.

STEVEN, D. M. (1950). Some properties of the photoreceptors of the brook lamprey. *J. Exp. Biol.* **27**, 350–64.

STEVEN, D. M. (1955). Experiments on the light sense of the hag, *Myxine glutinosa* L. *J. Exp. Biol.* **32**, 22–38.

YOUNG, J. Z. (1935). The photoreceptors of lampreys. I. Light-sensitive fibres in the lateral line nerves. *J. Exp. Biol.* **12**, 229–38.