THE EFFECTS OF SPATIAL SUMMATION IN THE RETINA ON THE EXCITATION OF THE FIBERS OF THE OPTIC NERVE

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In a previous paper (Hartline, 1940) it was shown that a ganglion cell in the peripheral retina of the vertebrate eye is excited by activity in many convergent pathways, from sensory elements distributed over a receptive field covering approximately a square millimeter of retinal area. Illumination of any portion of the receptive field of a retinal ganglion cell will accordingly produce a discharge of impulses in its axon, the strength of the response to illumination of a fixed retinal area usually being greater the higher the intensity of the stimulating light. The present paper will show that the discharge of impulses in a single optic nerve fiber also depends upon the size of the illuminated area. The excitation of a ganglion cell is therefore controlled by the number of active pathways which converge upon it, as well as by the degree of activity in the individual pathways.

Spatial summation in the vertebrate retina has previously been demonstrated by Adrian and Matthews (1927–1928). They showed that the latency of the discharge of impulses in the whole optic nerve of the cat was shorter the larger the area of the retina illuminated, and the latency of the response to four spots of light was shorter than the shortest latency obtained with any of the spots singly. This summation was enhanced by the application of strychnine, indicating that it depended upon the nervous interconnections within the retina. The study of the activity in single optic nerve fibers has now furnished more direct evidence for the convergence of excitatory effects within the retina; the present paper is concerned with the extension of this study to an analysis of spatial summation, in terms of the activity of the individual units of the retina.

METHOD. The method for studying the activity of single optic nerve fibers in the retinas of cold-blooded vertebrates, and for determining the location and extent of their receptive fields has been described in previous papers (Hartline, 1938, 1940). In the present experiments the eyes from

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large frogs (R. catesbiana) were used. None of the receptive fields of the fibers studied lay within or near the area acuta of the retina; the properties here reported are those of the peripheral retina.

The apparatus for illuminating the retina has likewise been described previously. It provided a beam of light which could be directed upon any part of the exposed retina, more than large enough to cover the region under investigation. The illumination was restricted to any desired area within this beam by means of diaphragms, with apertures of suitable size and shape imaged on the retina. Sharpness of focus was assured, in every experiment, by direct observation of the patterns of light on the retina by means of a dissecting microscope (×32). The diaphragms were readily interchangeable, and slipped into place against mechanical stops in a holder. The accuracy and reproducibility of their alignment in the beam was checked by exposing photographic plates in the place of the retina. Fine adjustments on the diaphragm holder enabled it to be shifted slightly, within the limits of the beam, so that the patterns of illumination could be accurately centered upon the receptive field of the fiber under observation.

RESULTS. In figure 1 are shown oscillograms of the amplified action potentials in a single optic nerve fiber, obtained in response to illumination of the retina with patches of light of various sizes. The areas illuminated, which were circular in shape, had been carefully centered upon the most sensitive portion of the fiber's receptive field and fell well within its limits. The larger the area of the stimulus patch, the shorter was the latency of response, and, for moderate degrees of stimulation, the higher was the frequency and the greater the number of impulses in the discharge. The fiber used in this experiment was one responding with a burst of impulses at the onset of illumination, and again when the light was turned off (no discharge during steady illumination). Fibers giving other types of response (cf. Hartline, 1938) show a similar dependence of the discharge upon the area illuminated.

Varying the area of the retina illuminated by a fixed intensity thus affects the response in a single optic nerve fiber: this effect, moreover, is exactly similar to that obtained by varying the intensity of illumination upon a fixed retinal area (cf. Hartline, 1938). To permit a comparison, two series of records are shown in figure 1, obtained at two different intensities of illumination. The responses in the right hand column were obtained with an intensity ten times that used in the left. It is to be noted that responses at the higher intensity are comparable to those obtained with areas approximately ten times larger, at the lower intensity. Only the total luminous flux falling upon the retina (area X intensity) is of significance in determining the response of the ganglion cell. This rule has been found to hold, except for very strong stimulation, to within the
limits of accuracy of this method. It applies only to illumination falling well within the receptive field of the fiber under observation.

A simple demonstration of this reciprocal relation between the area and intensity necessary to produce a constant effect in an optic nerve fiber is furnished by the determination of the threshold intensity, $I_{\text{thresh}}$, for various areas, $A$, of retinal illumination, plotted in figure 2. The line through the experimental points has a slope of $-1$, representing, on this double logarithmic plot, the relation

$$A \cdot I_{\text{thresh}} = \text{constant.}$$
This relation was demonstrated by Adrian and Matthews (1927–1928) in the optic discharge of the eel's eye; the present experiments show it to be a property of the individual retinal ganglion cells. Its limitation to retinal distances less than 1 mm., as reported by Adrian and Matthews, is due to the fact that the diameter of the receptive field of a ganglion cell is, on the average, of this order of magnitude.

Measurements of the reciprocal of the latent period and of the initial frequency of the discharge of impulses (in the same fiber whose responses are shown in fig. 1) are plotted, in figure 3, as functions of the area of illumination, for various levels of intensity. For moderate degrees of stimulation, these measures of the response increase steadily and approximately linearly with the logarithm of the area illuminated. Curves ob-

![Graph showing relation between area of retina illuminated (A) and threshold intensity, I\text{thres}, for stimulation of a single optic nerve fiber. For each arrow, upper point gives lowest intensity which elicited one or two impulses; lower point gives highest intensity which failed to elicit any response (determinations made to nearest 0.3 or 0.4 log unit). Where duplicate determinations coincided, arrows are drawn heavier. Line drawn through points has slope of \(-1\). (Log I = 0 equivalent to 3.10^6 meter candles; area in mm.².)](http://ajplegacy.physiology.org/)

ained at different levels of intensity are separated, parallel to the axis of abscissae, by amounts roughly equal to the logarithms of the ratios of their intensities, in accordance with the reciprocity relation discussed above.

Figure 3 shows that the responses increase with increasing area only up to a certain point. Beyond this point the responses actually decrease with increasing size of stimulus area, although these areas are well within the limits of the receptive field of the fiber. It is furthermore to be noted that the higher the intensity the smaller is the area at which this decrease begins. This effect may also be seen in the right hand column of figure 1, where the response to the largest area contains fewer impulses than the response to the area one-fourth as large. A similar depressing effect on the response has been reported, when the intensity of retinal illumination
on a fixed retinal area is increased above an optimal value (Hartline, 1938). It is as though the ganglion cell can be "overloaded," and the fact that this can be accomplished by increasing the area of the retina illuminated, as well as by increasing the intensity of the light, serves to emphasize the principle that the final response of the ganglion cell is determined by the sum total of activity reaching it over many convergent pathways.

It has been pointed out previously that in these experiments the sensitivity to light of any point on the retina must be defined with respect to the particular optic nerve fiber under observation. The sensitivity, thus defined, is not uniform over the receptive field of a fiber; the outlying portions are less effective in producing responses than is the central region (Hartline, 1940). It is reasonable to suppose that the outlying portions of the receptive field also contribute less to the total summed excitation of the ganglion cell. To test this point, and to study the relative contributions from the component portions of an illuminated area under different conditions, the following series of experiments have been performed.

A square area, large enough to cover nearly all of the receptive field of a fiber under observation, was subdivided into 25 small squares by means of diaphragms with appropriate apertures. Each of these small areas could be illuminated separately and the response to it compared with the response to illumination of the entire area, or of areas comprising several of the small subdivisions.

The requirements for threshold excitation of a fiber responding at "on" and "off" (only the "on" response recorded) are given in figure 4. The minimal intensity necessary to produce a response was determined for each
small square illuminated alone, and also for areas covered by 4, 9 and 25 of these small squares, as indicated in the figure. The reciprocals of these threshold intensities are entered in the respective squares, so that the greater the number in a particular square the more effective was that area in producing excitation of the ganglion cell. It is to be seen that the region of maximum sensitivity of the receptive field of this fiber was covered by eight of the nine central squares; the 16 border subdivisions

\[
\begin{array}{cccc}
0.1 & 0.1 & 0.1 & 0.05 \\
0.5 & 0.1 & 0.1 & 0.1 \\
0.5 & 0.1 & 0.5 & 0.25 \\
0.5 & 0.1 & 0.1 & 0.1 \\
0.25 & 0.25 & 0.5 & 0.25 & 0.25
\end{array}
\]

Fig. 4. Chart of the relative effectiveness, in stimulating a single optic nerve fiber, of different portions of the fiber's receptive field. "Effectiveness" of a region of the retina defined as reciprocal of threshold intensity for that region. Upper left: numerical values of effectiveness of 25 subdivisions of large square area tested individually. (Comparative scale of retinal distance given above.) Threshold intensity of the most effective subdivisions set equal to 1 (equivalent to 8.10^-3 meter candles) Lower left: effectiveness of area covering 4 of the central subdivisions (heaviest outline in upper left). Lower right: effectiveness of area covering the 9 central subdivisions (heavy outline in upper left). Upper right: effectiveness of entire large square. Fiber gave 'on' and 'off' bursts. 'Threshold' taken as the lowest intensity (within 0.3 or 0.4 log unit) which would reliably produce an 'on' burst of one or two impulses.
were all considerably less effective. When the larger area covered by four of the central squares was illuminated, the threshold intensity was one-fifth that of any of its subdivisions alone; when the still larger area covered by the nine central squares was exposed the threshold was still lower—only one-tenth of the threshold of the most sensitive subdivision. Thus, for the central portion of the receptive field, large illuminated areas were more effective in exciting the ganglion cell than any of their subdivisions. However, when the entire area covered by the 25 small squares was illuminated, the threshold intensity was not measurably lower than the threshold of the central region covered by only nine squares. Adding the 16 border subdivisions did not appreciably increase the effectiveness of the illumination, in this experiment. To judge from other experiments, the outlying portions of the receptive field do contribute somewhat to the total effect, and this might have been observed in the present experiment, had the thresholds been determined more closely. Nevertheless, the inclusion of less sensitive regions of the receptive field contributes correspondingly little to the summed effect; illumination of areas entirely outside the receptive field contributes nothing at all to the excitation of the ganglion cell.

Spatial summation in the vertebrate retina is thus limited to the receptive field of the retinal ganglion cell, and its effects are most readily observable in the more sensitive central portion of that field. A series of experiments has been performed, designed to analyze the contributions from component subdivisions of an illuminated area, which in every case lay well within the receptive field of the fiber under observation.

The experiment of figure 4, just cited, furnishes evidence of the summation of subliminal excitation. Thus illumination of any single square at an intensity \(1/I = 10\) failed to produce a response, yet this illumination must have produced some degree of activity in the pathways converging upon the ganglion cell, for when the nine central squares were illuminated together, at this intensity, impulses were discharged in the optic nerve fiber. Another example is furnished by an experiment on a fiber responding only to the cessation of illumination. At a suitable intensity, illumination of any one of four small squares singly produced no responses, but when all four were illuminated together "off" responses were regularly elicited, consisting of at least 7 impulses, at frequencies of 45 to 60 per second. Evidently, weak light can produce effects in the individual subdivisions of an area which are subliminal when they act alone, but which sum to reach the threshold of the ganglion cell when all act together. Since the activity in the retinal pathways presumably involves nerve impulses, we must conclude that more than one impulse must reach the retinal ganglion cell in order to excite a response in its axon.

The experiment of figure 1 shows that spatial summation not only affects
the threshold intensity to which a ganglion cell will respond, but also determines the magnitude of response at intensities above threshold. By testing different subdivisions of an area separately it can be shown, first, that the responses to illumination of a given area may be augmented by subliminal excitation from adjacent regions of the receptive field, and second, that illumination strong enough to elicit responses from each single subdivision of an area produces still greater excitation when the total area is exposed.

In an experiment (fig. 5) on a fiber responding only to cessation of illumination, only two of the central squares, out of the 25, would elicit a response (one impulse) when illuminated singly. However, when the area covered by the nine central squares was exposed, at this same intensity,
responses of 6 to 10 impulses, at average frequencies of 10 to 20 per second, were elicited. And when the 16 border subdivisions were added, the response increased to 18 impulses, at 53 per second, although none of these border squares alone could produce any response at this intensity. While

![Diagram A](image1)

![Diagram B](image2)

**Fig. 6.** a. Chart of the responses of a single optic nerve fiber (responding at "on" and at "off") to illumination of different portions of its receptive field, at a fixed intensity (0.3 meter candles). Left: frequencies of discharge (1st 6 impulses) of "on" and "off" bursts (upper and lower numbers, respectively) for each of 9 small squares tested individually (scale of distance given above). Right: frequencies of discharge of the "on" and "off" bursts (upper and lower pairs of numbers, respectively) in response to illumination of entire area covered by the 9 small squares. Upper member of each pair of numbers gives value obtained before testing the small squares, lower member the value afterwards. b. Chart of the frequencies of maintained discharge (13th to 15th second of continuous illumination) of single optic nerve fiber, in response to illumination of each of 9 small squares (left) compared with response to illumination of entire area covered by these squares (right). Scale given above. Intensity 300 meter candles.

It has been shown that border subdivisions contribute less to the summed effect of the illumination than do the more central ones, this experiment shows that their contribution nevertheless may be quite appreciable. This is especially true at low levels of excitation, where a slight increase in the stimulus usually causes a considerable increase in the response.
At an intensity moderately above threshold, the response to illumination of a large area is greater than the greatest response to illumination of any subdivision of this area at this same intensity. Illumination of nine small squares individually at an intensity above threshold resulted in the responses tabulated in figure 6a. When the entire area covered by these nine squares was exposed, at this same intensity, the frequency of the discharge was greater than in the responses of even the most effective subdivision illuminated alone. With fibers of this kind, responding to a change in illumination, both the "on" and the "off" bursts show the effects of spatial summation. A similar result, with a fiber whose discharge was maintained during steady illumination, is shown in figures 6b and 7. The frequency of the steady discharge resulting from illumination of each of the subdivisions singly is given in the respective square in figure 6b. When the entire area was illuminated, the frequency of the resulting discharge exceeded the highest frequency obtained from any of the small squares alone. Figure 7 shows the records of the responses to illuminating the entire area and to illuminating its most effective subdivision at the same intensity.

As noted previously, excitation above an optimal limit results in diminished responses in an optic nerve fiber. Thus it can happen that the response to the total area is actually less than that to any of its component subdivisions. The fiber, cited above, whose "off" responses illustrated the summation of subliminal effects from four subdivisions of an area, gave the following responses when tested at an intensity 100 times higher. The individual squares, illuminated singly, gave "off" bursts having
initial frequencies of 265, 230, 205 and 195 impulses per second. In response to illuminating the whole area covered by these four squares, at the same intensity, the initial frequency of impulses in the burst was only 175. That this diminished response was due to the excessively high total excitation was shown by reducing the intensity of the light to $\frac{1}{4}$ its previous value; illumination of the whole area then gave a response whose initial frequency was 240 impulses per second. Summation of excitation due to activity in convergent pathways takes place over the entire range of the response of the retinal ganglion cell.

Spatial summation can take place, of course, only where there is convergence of the effects of stimulation. In the more simple eye of *Limulus*, there is no convergence, and the response in a given optic nerve fiber depends only upon the illumination of the sensory cell giving rise to that fiber. Illumination of adjacent areas of the eye has no effect upon this response (Graham, 1932). But where there is convergence there need not be summation; the response in the final common path might be determined solely by the most strongly excited component. This is not so in the vertebrate retina, as was originally evident from the studies of Adrian and Matthews. The present experimental study furnishes direct evidence that the excitation of a single retinal ganglion cell is determined by the summated effects of activity in the pathways converging upon it.

**SUMMARY**

A study has been made of the action potentials of single optic nerve fibers of the frog's retina, in response to illuminating areas of the retina of various sizes. In these experiments the fibers used were from the peripheral retina, where many receptor elements are connected with each retinal ganglion cell.

The discharge of impulses in a single optic nerve fiber is stronger the larger the area of the retina illuminated, within the limits of the fiber's receptive field. Except for very strong illumination, the responses have a shorter latency and a higher frequency the greater the number of receptors illuminated. The threshold intensity is also lower the larger the area of the stimulating patch of light.

Varying the area of the retina illuminated by a fixed intensity affects the discharge of impulses in a single optic nerve fiber in the same way as varying the intensity of illumination of a fixed area. For threshold excitation and for levels of response above threshold, only the total quantity of light (A·I) determines the response, provided the illumination is confined to the central portion of the fiber's receptive field.

Excitation of a retinal ganglion cell above an optimal limit results in diminished responses in its optic nerve fiber: this effect can be produced by increasing either the intensity or the area of the retinal illumination.
The discharge of impulses in response to illumination of a given area within the receptive field of an optic nerve fiber has been compared with the responses to illumination of subdivisions of this same area. 1. Illumination of the less effective subdivisions in the margins of the receptive field contributes correspondingly little to the summed effect upon the ganglion cell. Illumination of areas entirely outside the receptive field has no effect upon the discharge of impulses. 2. Subliminal effects from the subdivisions of an area can sum to reach the threshold of the ganglion cell when all the subdivisions are illuminated together. From this it is concluded that more than one nerve impulse must reach the retinal ganglion cell, over the pathways converging upon it, in order to excite a discharge in its optic nerve fiber. 3. The discharge of impulses in response to illumination of a given area is stronger than the strongest response from any subdivision of this area, illuminated at the same intensity. This is true provided the ganglion cell is not stimulated too strongly; at very high levels of excitation the response to illumination of the entire area is diminished.

An optic nerve fiber is the final common path for nervous activity originating in many receptor elements of the retina; excitation due to the activity in the retinal pathways converging upon a single ganglion cell summates to determine the response in its optic nerve fiber.

REFERENCES

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