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Lateral Inhibition in an Insect Eye*

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Summary. Intracellular light-evoked potentials were measured from both visual cells and secondary neurons (monopolar neurons type I) in the eye of *Calliphora* at varying angles of light-incidence. From these measurements and from the characteristic curves we obtained a relationship between the effective light intensity and the angle of incidence of the light stimulus for both cell types. These curves must be identical for the two cell types in the absence of a lateral information processing as a theoretical reflection shows. From the experimental results that the curve (effective light intensity versus light angle of incidence) of the monopolar neurons was considerably narrower as that of the visual cells, it was concluded that a lateral inhibition in the first optic ganglion of the fly retina exists. Although the information coding in the secondary neurons of the fly retina was completely different (graded potentials) from that of corresponding neurons in the *Limulus* eye (spikes), it appeared that the same principles of information processing existed in both instances.

Introduction

The concept of "lateral inhibition" was used to describe the mutual influence of receptive units in *Limulus* eye (Hartline, Wagner and Ratliff, 1956). Every receptive unit possessed an eccentric cell (secondary neuron) whose excitation was dependent not only on the stimulus intensity which excited the visual cells of this unit, but also on the excitation of the neighboring eccentric cells.

Electrophysiological investigations of single cells of the vertebrate retina provided results pointing to the existence of lateral inhibition also in the vertebrate eye. For example, potentials of varying form and polarity have been recorded from the bipolar neurons and cones of a vertebrate retina depending upon whether the retina was stimulated with a light spot or with a light annulus (Werblin and Dowling, 1969; Kaneko, 1970; Baylor, Fuortes and O'Bryan, 1971).

Likewise, as with the described cells of the vertebrate eye, both the primary receptors and the secondary neurons of the insect retina showed no spike activity in our experiments. Therefore, in these retinae the in-

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formation coding in the secondary neurons was of a completely different type as that in the corresponding cells of the *Limulus* eye. We could show, however, from a comparison of the visual fields of receptors and neurons that the principle of lateral inhibition as found in the *Limulus* eye also existed in the first optic ganglion of an insect retina.

Methods

Light-evoked potential responses were recorded in the first optic ganglion (Lamina ganglionaris) of the fly *Calliphora erythrocephala* using microglass capillaries. The recording technique and the method of cell identification have been reported by Zettler and Järvilehto (1971) and Järvilehto (1971). Two measurement series were recorded from every investigated cell, whereby either the light intensity or the angle of incidence of the light stimulus was varied.

The measurement of the potentials as a function of the light intensity was accomplished using a punctiform light source (2 mm diameter and 100 mm distant) positioned so that a maximal potential could be elicited. The intensity was varied using grey filters (Zeiss).

The relationship of the response to the angle of incidence could be measured by turning the light source step by step along the equator of the eye, always at the same distance from the eye. The stimulus intensity was thereby kept constant. At every point a square wave stimulus of 250 msec duration was applied.

Identification of the investigated cell was carried out by marking the cell with Procion-Yellow M-4R (Fig. 1). The recording site could be identified through histological localization of the electrode tip (Zettler and Järvilehto, 1970).

Results

The potentials, examined in this investigation originated from the axons of the visual cells 1–6 or from the axons of the postsynaptic monopolar neurons of type I (Fig. 1). Both in the axons of the visual cells (Järvilehto and Zettler, 1970; Ioannides and Walcott, 1971; Alawi and Pak, 1971) and in the axons of monopolar neurons (Autrum, Zettler and Järvilehto, 1970; Zettler and Järvilehto, 1971) the stimulus response consisted not of spikes, but rather gradiated de- or hyperpolarizing potentials respectively. The potentials of both cell types were sensitive to the stimulus intensity (Järvilehto and Zettler, 1971) as well as to the light angle of incidence. For this reason, the potentials strongly depended on the position of the light source. For every cell one could find an exact position from which a maximal response could be elicited. A variation of even a few degrees caused a considerable decrease in the response.

The relationship of the responses to the angle of incidence of the light stimulus was investigated in four visual cells of type 1–6. The depicted visual field shown in Fig. 4a was obtained from these measurements. A detailed description of these potentials can be avoided here as their angular sensitivity has already been amply described (Washizu,

Burkhardt and Streck, 1964; Vowles, 1966; Scholes, 1969; Mote, 1970; Zettler and Järvilehto, 1970).

The relationship of the hyperpolarizing potentials of a monopolar neuron to the light angle of incidence is shown in Fig. 2d. The original recordings in Fig. 2 show the potential gradiation, caused both by the light intensity and by the light angle of incidence. The light intensity is diagrammed in relative logarithmic units. The maximal intensity occurs at about 10^4 Lux. The variation of the angle of incidence was carried out at an intensity of 9×10^{-4} relative units. The angle 0° is the position of the light source at which the maximal response was obtained.

Discussion

The light-evoked potentials which one records from a single visual cell in the fly eye have various amplitudes depending upon which direction the light stimulus strikes the eye (Washizu, Burkhardt and Streck, 1964). The basis for this angular dependence of the responses lies in that the light intensity impinging upon specific visual cells varies when the direction of incidence of the light changes. The light intensity impinging upon specific visual cells was called the "effective light intensity". In addition to a direct measurement (Kuiper, 1962) the relationship of the effective light intensity to the angle of incidence can also be obtained indirectly through electrophysiological studies (Washizu, Burkhardt and Streck, 1964). When the light-sensitivity is known (characteristic curve), then one can ascertain from this curve the effective light intensity corresponding to every potential which has been measured at a certain light angle of incidence. Thus one obtains curves describing the effective light intensity as a function of the light angle of incidence (Fig. 4a). The efficiency of the light stimulus is defined as 100% when the position of the light source elicits a maximal response (0°). If direction of incidence of the light stimulus differs from this angle, then in spite of constant intensity of the light source the efficiency of the light decreases.

The same considerations can be applied to the potentials of the monopolar neurons. They also show a distinct dependence on the angle of incidence of the light stimulus. If one measures this relationship (Fig. 2c), then one can again conclude from the characteristic curve (Fig. 2a) the relationship of the effective light intensity to the angle of incidence (Fig. 4b). The visual fields of the two cell types (Visual cell type 1–6 and monopolar neuron type I) are depicted side by side in Fig. 4 for comparison. Both curves were obtained from potential measurements under the same physiological conditions.

To make statements about a probable lateral information processing from these comparison one must also consider the morphological situation.


a

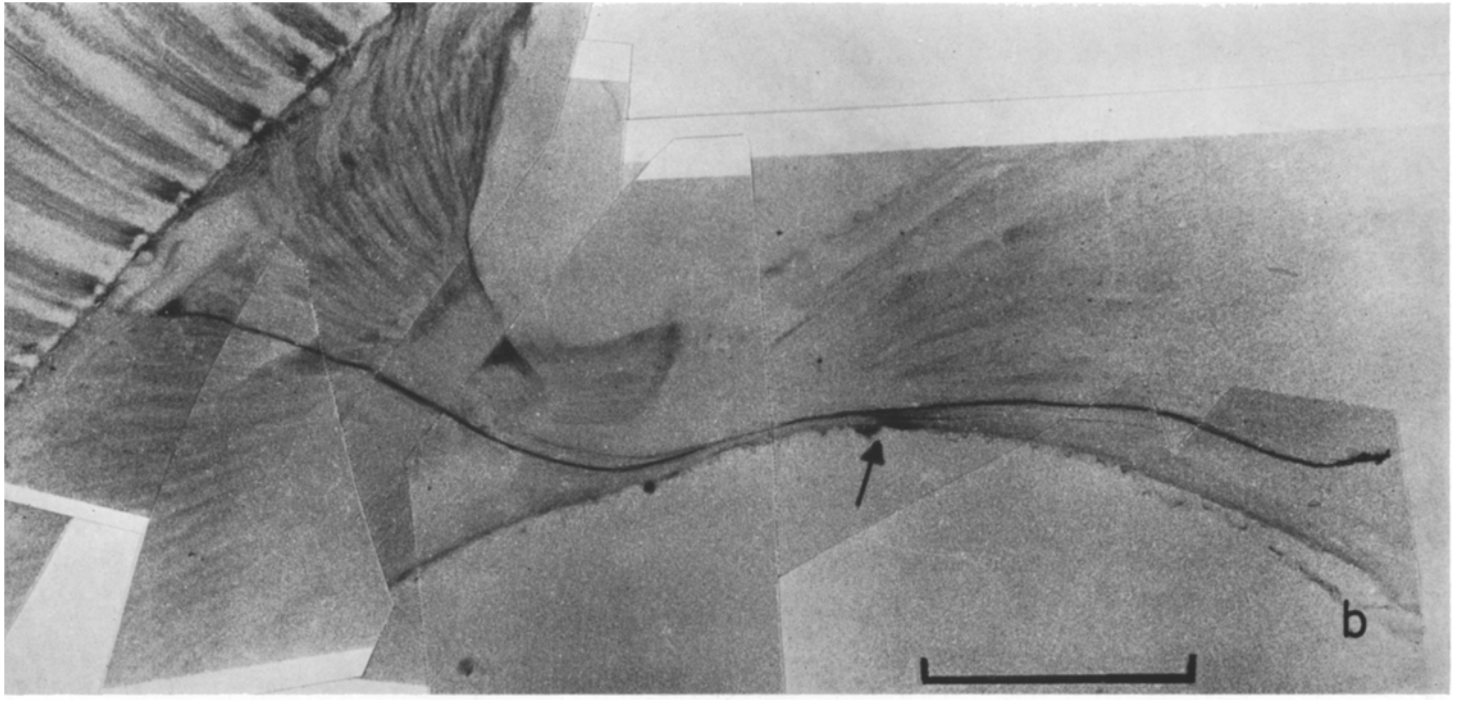

Fig. 1 a and b. *Calliphora erythrocephala*. Cellular identification through intracellular injection of Procion-Yellow M-4R. Photomontage from a black-white negative picture. The recording site is marked with an arrow. a Visual cell of type 1–6. Scale 50 μm. b Monopolar neuron of type I. Scale 100 μm

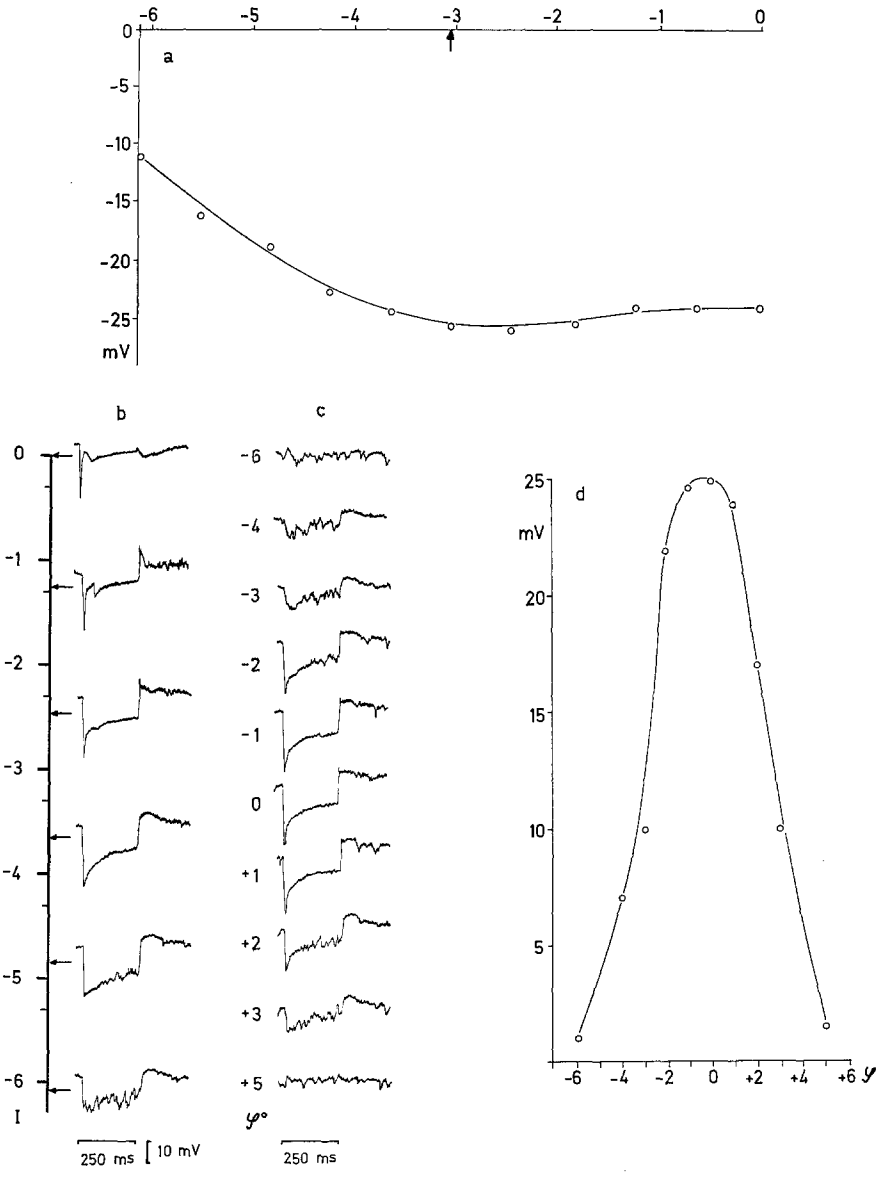

Fig. 2a–d. *Calliphora*. Intracellular potentials from the axon of a monopolar neuron. a Characteristic curve of the amplitude of the on-effect. The relative units of the intensity are diagrammed in a logarithmic scale. b Potentials at different intensities and constant direction of incidence of the light stimulus ($\varphi = 0°$). c Potentials at different directions of incidence and constant intensity ($I = 9 \times 10^{-4}$) of the light stimulus. d Amplitude of the on-effect as a function of the angle of incidence of the light

Fig. 3 depicts the morphological connections between the visual cells and the monopolar neurons. In a neuro-ommatidium (cartridge) of the Lamina ganglionaris each of the six visual cells are in synaptic contact with both monopolar type I neurons (Trujillo-Cenóz and Melamed, 1963; Trujillo-Cenóz, 1965; Boschek, 1971). These six visual cells come from six different ommatidia in the external retina (Cajal and Sánchez, 1915; Trujillo-Cenóz, 1966; Braitenberg, 1967), but are considered as a single receptive unit because they all "see" the same point in the invironment (Autrum and Wiedemann, 1962; Kirschfeld, 1967).

To our specific stimulus situation this means that from a given position of the light source which maximally stimulated one visual cell, the other five visual cells of the same cartridge were also maximally stimulated. This means that the visual field of the whole cartridge-unit is identical with that of a single visual cell of this unit. One monopolar neuron, therefore, will be excited by six different visual cells which possess one and the same visual field.

Assumming that a monopolar neuron A (Fig. 3) maintains synaptic contact only with the six visual cells of cartridge A, it can be expected that its visual field is identical with the visual fields of the six visual cells belonging to cartridge A and hence with the visual field of each of the individual six visual cells.

This proposition can be based on the following: the visual field of a visual cell will be defined as the curve for the effective light intensity:

$$I = f_R(\varphi) \quad \text{(the subscript } R \text{ refers to the receptor).}$$

This function is supported from the measured curves: response versus light angle of incidence

$$U_R = g(\varphi),$$

and the characteristic curve

$$U_R = h(I).$$

From these equations I becomes a function of φ:

$$I = h^{-1}(g_{(\varphi)})$$

where h^{-1} is the reciprocal function of h. Therefore,

$$f_R(\varphi) = h^{-1}(g_{(\varphi)}).$$

Analogously, the visual field of a monopolar neuron is given by

$$I = f_N(\varphi),$$

where the subscript N refers to the visual field of a neuron.

The above stated proposition means: $f_N(\varphi) = f_R(\varphi)$, under the assumption that no lateral exchange with neighboring elements exists. When one considers the receptive unit A (Fig. 3), this assumption means

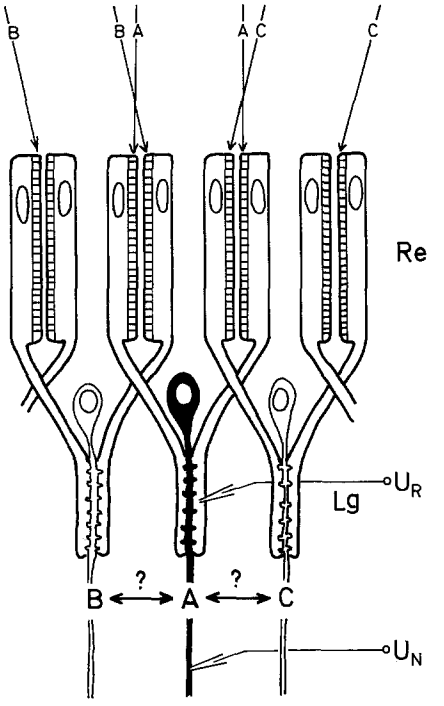

Fig. 3. *Calliphora.* Scheme of the synaptic connections of the primary receptors (type 1–6) and monopolar neurons (type I). A, B and C are receptive units. All receptors which belong to the same laminar unit have the same maximal effective light angle of incidence. *Re* retinula layer (retina externa), *Lg* lamina ganglionaris (first optic ganglion), U_R potential in the axon of a visual cell, U_N potential in the axon of a monopolar neuron

that the potential of the neuron at A is only a function of the potential of the visual cells at A and not resulting from the excitation of neighboring elements B and C.

Therefore,

$$U_N = k\,(u_R)^1.$$

It is under this assumption that the above proposition must be proven.

U_R is a function of φ as well as a function of I, so the assumption can be split into two equations:

$$U_N = k\,(g_{(\varphi)}),$$

1 It has been shown (Järvilehto and Zettler, 1971) that the potential amplitude U_R could not be the generator potential for the amplitude U_N that is, one cannot cause the other. It does not mean, however, that no functional relationship can exist between the two values (both values stand in functional relation to the light intensity, therefore a functional relationship between them must also exist).

and

$$U_N = k\,(h_{(I)}).$$

By rearranging the two equations and explicitly describing I, one obtains the relationship of the effective light intensity I to the angle φ for the monopolar neuron

$$I = h^{-1}\,(g_{(\varphi)}).$$

This is the same relationship as holds for the receptor and thus the proposition is proven.

It is obvious that by the existence of a lateral mutual influence the assumption no longer holds true and that therefore the curve for the effective light intensity of a monopolar neuron $f_{N(\varphi)}$ must be different from that of a receptor $f_{R(\varphi)}$.

The morphology of the lamina provides sufficient ground for the supposition that the monopolar neuron A also maintains synaptic connections with laterally extended laminar cells in addition to those with the six mentioned visual cells. Such laterally branched cells are: horizontal cells, amacrine cells, tangential cells and the monopolar neurons of type L_4 (Cajal and Sánchez, 1915; Strausfeld, 1970; Strausfeld and Braitenberg, 1970; Strausfeld, 1971). Possible mutual interaction between the cartridge A and its neighboring elements B and C is symbolized in Fig. 3 by double-headed arrows.

If such a mutual lateral influence between the individual cartridges occurs, one can no longer expect, as has been shown, that the visual field of a monopolar neuron A is identical with the visual field of the visual cells belonging to cartridge A. The effective light intensity which causes the neuronal potential is in this instant no longer the effective light intensity only, which causes the visual cell potentials of cartridge A, but in addition to it, also the intensity which effects the visual cell potentials of the cartridges B, C and others. If the influence of the neighboring elements B and C on the neuron A is excitatory, the effectivity of light will be increased and one must expect a wider range of the visual field for neuron A. In the case of an inhibitory influence, the effectivity of light will be decreased and thus the visual field of a monopolar neuron must be narrower than that of an individual visual cell.

The curves depicted in Fig. 4 for the two cell types differ considerably. The visual field of the monopolar neurons is considerably restricted compared to that of the visual cell. The maximal effective light intensity (at 0°) was defined as 100% for every investigated cell. The angles of the visual field by which the effective light intensity for visual cells was decreased to 50, 10 or 5 percent, were 4.5°, 9° and 11° respectively.

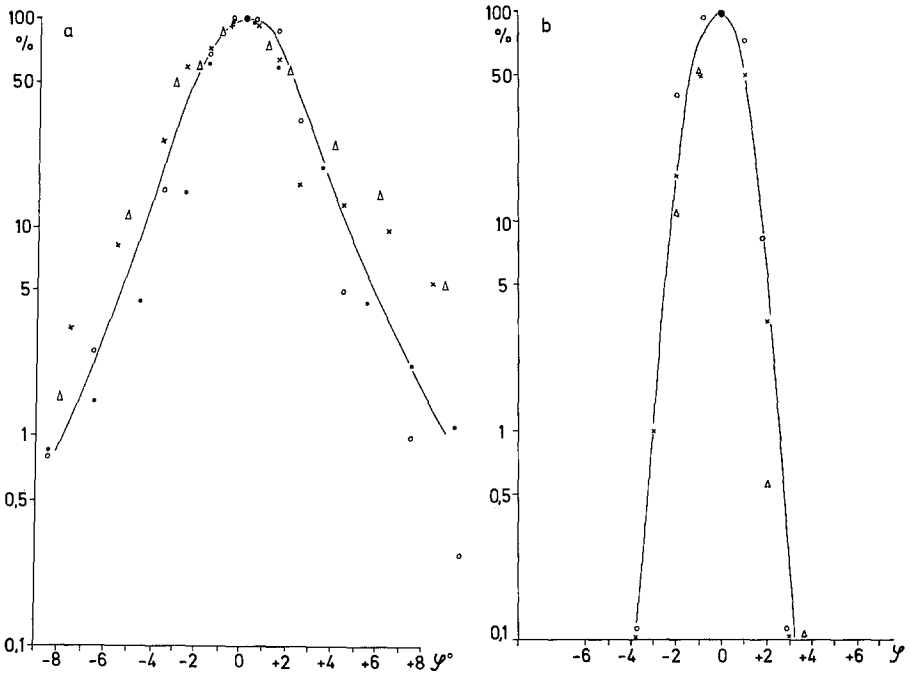

Fig. 4a and b. *Calliphora*. Effective light intensity as a function of the light angle of incidence. The greatest effectiveness ($\varphi = 0°$) is defined as 100%. a Mean value curve from 4 visual cells. b Mean value curve from 3 monopolar neurons

The corresponding angles for the investigated monopolar neurons under the same conditions were 2°, 2.5° and 3°.

On the basis of the above accomplished deliberations one must conclude from these findings that a lateral inhibitory processing occurs in the first optic ganglion. The question, however, which path this processing takes, that is, which cells are involved, is not to be answered at present.

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