

RNA CONTENT IN NEURON-NEUROGLIA SYSTEMS OF THE RETINA
AND VISUAL CORTEX FOLLOWING LIGHT DEPRIVATION AND PHOTIC
STIMULATION

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Dual-beam cytospectrophotometric investigation of sections stained with galloxyanin and chrome alum showed that keeping adult rats in complete darkness for 30 days led to a decrease in the RNA content in the neuroglia of the ganglion-cell layer of the retina but no changes in the corresponding neuron. No changes were found either in neurons or in neuroglia of layers II-III of the visual cortex. At the end of light deprivation, stimulation by a steady light or by flashes for 2 h had no effect on the RNA content in the neurons of either of these parts of the visual system, whereas in control rats it evoked definite accumulation of RNA in the neurons. Qualitative changes in the metabolism of intracellular RNA in the nerve tissue of adult animals under the influence of light deprivation are emphasized. Differences in the biochemical properties of the neuron-neuroglia system depending on its location in the visual system are discussed.

KEY WORDS: *Neuron; neuroglia; retina; cerebral cortex; RNA; light deprivation.*

The writers showed previously [5] that prolonged hypofunction of the visual system in adult animals leads to marked changes in the RNA content in neurons of different parts of that system. In recent years much evidence has been accumulated [6] to show that the neuron is only part of a complex hypercellular community, a combined functional and metabolic system of neuron and neuroglia, the properties of which largely determine the character of function of nerve tissue.

The object of this investigation was to compare biochemical changes evoked by light deprivation and by photic stimulation in neuron-neuroglia systems of the sensor and peripheral structures of the visual system in adult rats.

EXPERIMENTAL METHOD

Deprivation was produced in adult male rats weighing 180-220 g by keeping the animals in complete darkness for 30 days. At the end of the experiments the animals were quickly decapitated in dim red light, which does not significantly affect the albino rat retina [11]. At the end of deprivation some of the animals were exposed for 2 h to illumination by a constant light with an intensity of 40 lx or to flashes with a frequency of 2 Hz and the same intensity of illumination. Animals kept for 30 days under standard animal house conditions and with natural illumination (not over 6 lx in intensity) served as the control.

Immediately after decapitation the retina and visual cortex were excised, fixed by Carnoy's method, and embedded in paraffin wax. Sections 7 μ thick were stained with galloxyanin and chrome alum to detect nucleic acids. The nucleic acid content (per cell) was determined by dual-beam cytospectrophotometry 550 and 465 nm, with allowance for the volumes of the cells calculated from linear measurements of the cells made with an ocular microm-

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TABLE 1. Nucleic Acid Concentration and Total Content and Cell Volume for Neurons and Neuroglia of Ganglion-Cell Layer of Rat Retina after Light Deprivation and Photic Stimulation ($M \pm m$)

| Experimental conditions | Neurons | | | Neuroglia | | |
|---------------------------|---|----------------------|--|---|----------------------|--|
| | nucleic acid concentrations, logarithmic extinction units | cell volume, μ^3 | nucleic acid content calculated per cell, conventional units | nucleic acid concentrations, logarithmic extinction units | cell volume, μ^3 | nucleic acid content calculated per cell, conventional units |
| Control | $0,394 \pm 0,015$ | 157 ± 3 | $61,9 \pm 1,6$ | $0,665 \pm 0,016$ | $52,4 \pm 1,3$ | $34,7 \pm 0,7$ |
| Constant light | $0,506 \pm 0,014^*$ | $181 \pm 14^*$ | $92,1 \pm 1,9^*$ | $0,599 \pm 0,051$ | $34,6 \pm 0,5^*$ | $20,6 \pm 0,4^*$ |
| Flashes | $0,482 \pm 0,009^*$ | $204 \pm 8^*$ | $98,4 \pm 3,3^*$ | $0,554 \pm 0,038^*$ | $47,5 \pm 1,6$ | $26,3 \pm 3,0^*$ |
| Darkness | $0,370 \pm 0,002$ | 153 ± 5 | $56,8 \pm 4,2$ | $0,576 \pm 0,023^*$ | $41,0 \pm 0,9^*$ | $23,6 \pm 0,7^*$ |
| Darkness + constant light | $0,420 \pm 0,016$ | 158 ± 11 | $66,4 \pm 2,0$ | $0,571 \pm 0,012^*$ | $42,5 \pm 1,1^*$ | $24,9 \pm 0,9^*$ |
| Darkness + flashes | $0,461 \pm 0,016^*$ | 147 ± 6 | $67,8 \pm 2,0$ | $0,566 \pm 0,013^*$ | $48,8 \pm 1,7$ | $27,6 \pm 0,8^*$ |

*Here and in Table 2 differences from control statistically significant at the level $P < 0.05$.

TABLE 2. Nucleic Acid Concentration and Total Content and Cell Volume for Neurons and Neuroglia of Layers II-III of Visual Cortex of Rats after Light Deprivation and Photic Stimulation ($M \pm m$)

| Experimental conditions | Neurons | | | Neuroglia | | |
|---------------------------|---|----------------------|--|---|----------------------|--|
| | nucleic acid concentrations, logarithmic extinction units | cell volume, μ^3 | nucleic acid content calculated per cell, conventional units | nucleic acid concentrations, logarithmic extinction units | cell volume, μ^3 | nucleic acid content calculated per cell, conventional units |
| Control | $0,294 \pm 0,012$ | 322 ± 13 | $94,5 \pm 2,2$ | $0,397 \pm 0,006$ | $62,6 \pm 1,0$ | $24,1 \pm 0,1$ |
| Constant light | $0,356 \pm 0,013^*$ | 323 ± 4 | $115,0 \pm 4,1^*$ | $0,275 \pm 0,024^*$ | $70,3 \pm 1,9$ | $19,0 \pm 0,7^*$ |
| Flashes | $0,317 \pm 0,016$ | $388 \pm 6^*$ | $122,9 \pm 4,3^*$ | $0,340 \pm 0,028$ | $77,6 \pm 1,9^*$ | $26,0 \pm 1,1$ |
| Darkness | $0,337 \pm 0,017^*$ | 318 ± 11 | $107,0 \pm 1,6^*$ | $0,401 \pm 0,017$ | $61,1 \pm 2,0$ | $25,9 \pm 1,7$ |
| Darkness + constant light | $0,271 \pm 0,009$ | 323 ± 9 | $87,4 \pm 2,9$ | $0,387 \pm 0,004$ | $68,3 \pm 1,8$ | $26,5 \pm 1,0$ |
| Darkness + flashes | $0,347 \pm 0,044^*$ | $242 \pm 7^*$ | $84,2 \pm 2,3$ | $0,381 \pm 0,013$ | $70,2 \pm 0,7$ | $26,8 \pm 0,7$ |

eter. The experimental details, apparatus, technique of the cytophotometric measurements, and all mathematical calculations were described previously [5].

Each series of experiments was carried out on six rats; in each animal 60 to 100 neurons in each location and the same number of neuroglial cells from each animal were examined photometrically. All calculations and statistical analysis of the results, by Student's method, were carried out with the Minsk-22 digital computer.

EXPERIMENTAL RESULTS

As Tables 1 and 2 show, light deprivation and photic stimulation in some cases altered both the concentration of nucleic acids and the volume of the cells. In the adult organism the nuclear DNA level in cells of the nervous system is stable [2, 6]; for that reason changes discovered in the combined RNA + DNA, especially after exposure for 2 h, were entirely attributable to changes in the RNA content.

Photic stimulation caused a definite increase in the RNA content both in the ganglion cells of the retina (Fig. 1) and, to a rather lesser degree, in the neurons of layers II-III of the visual cortex (Fig. 2), in agreement with data in the literature [1, 2]. It is interesting to note that this accumulation of RNA in the neurons in nearly every case (except the effect of flashes on the visual cortex) was accompanied by a decrease in the RNA content in cells of the perineuronal neuroglia (Figs. 1 and 2).

Light deprivation itself had little effect on the RNA level. Distinct changes (a fall in the RNA content) were observed only in the glial cells of the retina (Fig. 1). However, the action of photic stimulation on rats after 30 days in darkness was somewhat different from its action on intact animals. Neither constant light nor flashes led to the accumulation of RNA in the retinal or visual cortical neurons. The glial cells, however, preserved

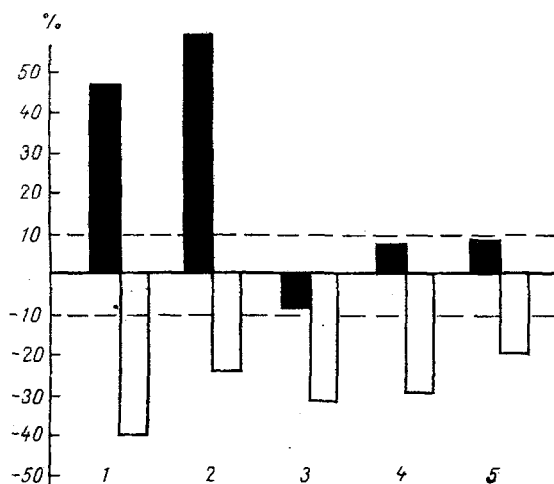


Fig. 1

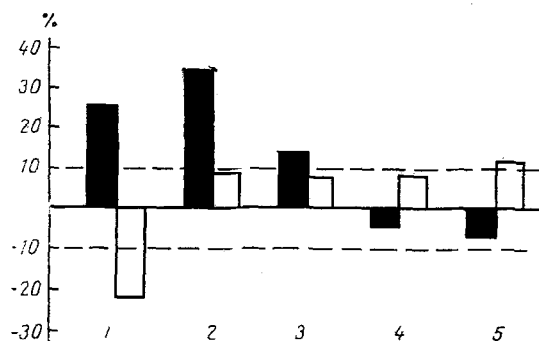


Fig. 2

Fig. 1. Changes in RNA content in retinal neurons and neuroglia of rats after light deprivation and photic stimulation. Black columns represent neurons, unshaded columns neuroglia. Broken lines mark 95% confidence limits. 1) Stimulation of intact animals by constant light; 2) stimulation of intact animals by flashes; 3) light deprivation; 4) light deprivation followed by stimulation with constant light; 5) light deprivation followed by stimulation by flashes. Ordinate, deviations of RNA content (calculated per cell) in per cent of corresponding control value.

Fig. 2. Changes in RNA content in visual cortical neurons and neuroglia of rats after light deprivation and photic stimulation. Legend as in Fig. 1.

their original response to illumination, but only in the retina (Fig. 1). In the glial cells of the visual cortex constant light, which evoked a decrease in the RNA content in intact rats, had no significant effect on animals subjected to light deprivation (Fig. 2). Several workers have demonstrated [3, 4, 8-10] quantitative changes in the metabolism of the nervous system, including in RNA metabolism [1, 8], during postnatal maturation of the visual system under conditions of prolonged light deprivation. The results of the present experiments indicate that in adult animals hypofunction of the visual system for 30 days alters the properties of the cell structure of this system. Although the relationship between RNA breakdown and synthesis, which determines the level of intracellular RNA, showed little change under the influence of deprivation, this factor had a marked effect on the ability of visual neurons to respond by biochemical changes to photic stimulation. In the writers' view, this indicates qualitative changes in nucleic acid metabolism in the structures of the visual system during a long period of hypofunction.

In neurons of the central (cortex) and peripheral (retina) structures of the visual system the action of deprivation was practically identical. In the glial cells, on the other hand, the effect of deprivation on the metabolic response to illumination was clearly defined in the visual cortex but absent in the retina. Presumably the specific features of metabolism in concrete neuron-neuroglia systems may depend in some cases also on specific differences between the neuroglial cells.

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EFFECT OF MUSCULAR EXERTION ON INVERTASE ACTIVITY OF THE SMALL INTESTINE

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Invertase activity of the proximal, middle, and distal parts of the small intestine after muscular exertion lasting 2, 4, and 10 h (forced swimming in water at $35 \pm 1^\circ\text{C}$) was studied in acute experiments on male albino rats. After swimming for 2 h the invertase activity in the first two parts of the intestine was sharply reduced and it returned to its initial level after 48-72 h. This decrease was much less marked in the distal portion. Swimming for both 4 and 10 h led to a slight increase in enzyme activity in all three parts of the intestine 24 h after the beginning of the experiments, followed by a decrease in the first two parts and a marked increase in the distal portion 48 h after the beginning of the experiment. It is suggested that these changes are brought about through the hypothalamic-pituitary-adrenal system in accordance with the principle of the general nonspecific adaptation syndrome.

KEY WORDS: *Contact digestion; invertase; muscular exertion.*

Intensive muscular exertion leads to considerable changes in metabolism [10], the hormonal status [12], and the motor-evacuatory, secretory, and absorptive activity of the gastrointestinal tract [2, 3, 7, 9, 11]. However, the state of the mechanisms of contact digestion, which plays an important role in the digestion and absorption of the main components of the diet [6], has received extremely inadequate study under these conditions, with the result that difficulties exist in the solution of certain problems connected with the physiology of nutrition during muscular activity.

In the present investigation, in which an intestinal enzyme participating in the final stages of carbohydrate hydrolysis (invertase — E.C. 3.21.36) was used as the example, the effect of muscular exertion of varied duration on enzyme activity in different parts of the small intestine was studied.

EXPERIMENTAL METHOD

Experiments were carried out on 99 noninbred male albino rats weighing 100-120 g and kept on a standard diet. The animals were divided into three experimental (30 rats in each group) and one control (9 rats) group. The animals of the experimental groups were forced to swim in water at $35 \pm 1^\circ\text{C}$ for 2, 4, and 10 h, respectively, and then decapitated immediately or 4, 24, 48, and 72 h after swimming. The rats of the control group were kept under similar conditions but were not subjected to any special procedure. To determine enzyme activity homogenized preparations are obtained from the everted proximal, middle, and distal portions of the small intestine. These preparations were incubated in 1% sucrose at 37°C for 10 min. Enzyme activity was expressed in micromoles glucose formed per minute per gram wet weight of tissue [4]. Activity of the homogenate reflected the total reserves of enzymes, indirectly reflecting the rate of protein synthesis; surface activity of the intact portions of intestine characterized that fraction of the enzymes that was included in the composition of the membrane surface of the microvilli [6].

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