the labeling density in cells with dystrophic changes and in cells with no such changes. As has already been pointed out [1], dark neurons have a higher level of RNA synthesis than pale neurons. Meanwhile dark neurons with dystrophic changes had a higher intensity of RNA synthesis, although not statistically significantly, than dark neurons with no dystrophic changes. Similar data also were obtained when pale neurons were compared. This is shown by examination of the autoradiographs (Fig. 1-3), which revealed maximal labeling in the nucleus and nucleolus of damaged neurons. In other words, nerve cells with marked dystrophic changes were characterized by a high level of RNA synthesis.

As was observed previously for hepatocytes [3], a reparative process such as RNA synthesis is recorded in neurons simultaneously with destructive changes. However, unlike in hepatocytes, it does not develop in different cells, but in the same cells, due to the intracellular type of regeneration which is a feature of neurons.

LITERATURE CITED

1. E. Ya. Sanovich, A. A. Pal'tsyn, and V. P. Tumanov, Byull. Éksp. Biol. Med., No. 4, 498 (1981).
2. D. S. Sarkisov, A. A. Pal'tsyn, and B. V. Vtyurin, Adaptive Restructuring of Biorhythms [in Russian], Moscow (1975).
3. D. S. Sarkisov, A. A. Pal'tsyn, and B. V. Vtyurin, Electron-Microscopic Autoradiography of Cells [in Russian], Moscow (1980).
4. D. Brown and J. Gurdon, Proc. Natl. Acad. Sci. USA, 51, 139 (1964).
5. M. Buongiorno-Nardelli and F. Amaldi, Nature, 225, 946 (1970).
6. R. P. Perry, Exp. Cell Res., 29, 400 (1963).
7. R. P. Perry, J. Cell Biol., 91, 28 (1981).
8. F. M. Ritossa and S. Spiegelman, Proc. Natl. Acad. Sci. USA, 53, 737 (1965).

ACTION OF CORTICAL EXTRACTS FROM THE LEFT AND RIGHT CEREBRAL HEMISPHERES ON RESTORATION OF CONDITIONED REFLEX ACTIVITY IN RATS AFTER UNILATERAL EXTIRPATION OF THE FRONTAL CORTEX

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Proof has recently been obtained of neurochemical asymmetry of the brain of man and animals [9] and of considerable differences in the changes taking place in neurotransmitter concentrations after injury to the left or right cerebral hemispheres [12, 13]. Unilateral injury to the cerebellum [1], vestibular nuclei [6], or cerebral cortex [2] of animals and the creation of unilateral generators of pathologically enhanced excitation (GPEE) in the brain [5, 6] or spinal cord [8] have been shown to lead to the appearance of peptide regulators in the brain tissue with a lateralized influence on muscle tone. The disparity between the biochemical consequences of injury to the hemispheres suggests that, in principle, they can be selectively corrected and, consequently, partial recovery of disturbed functions may be possible with the aid of endogenous factors specific for each hemisphere.

To test this hypothesis, the investigation described below was undertaken to study the effect of low-molecular-weight components of cortical extracts of the left and right hemi-

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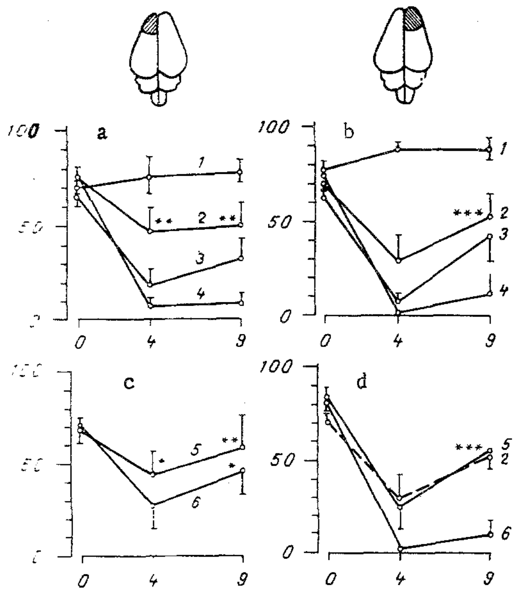

Fig. 1. Effect of cortical extracts from left and right cerebral hemispheres on restoration of AACR after unilateral extirpation of the frontal cortex. Above — diagram showing injury to cerebral cortex in recipients; a and c) changes in AACR after extirpation of left frontal cortex, b and d) of right cortex. Ordinate, coefficient of preservation of AACR (in %). Abscissa, time after operation (in days). a and c) State of AACR: 1) in animals after mock operation, 2) in rats after left-sided (a) and right-sided (b) frontal lobectomy and treated with LE and RE from healthy animals, respectively; 3) the same as in 2, but receiving LE and RE treated with Pronase; 4) after injection of physiological saline (control); c and d) state of AACR: 2) the same graph as in b (for comparison with size); 5) receiving RE taken nine days after left-sided lobectomy on donors; 6) receiving extracts of the contralateral hemisphere of normal animals. $^{*}p < 0.05$, $^{**}p < 0.02$, $^{***}p < 0.01$ Compared with control.

spheres of rats on the time course of recovery of active avoidance conditioned reflexes (AACR) disturbed by unilateral extirpation of the frontal cortex.

EXPERIMENTAL METHOD

Experiments were carried out on noninbred male albino rats (81 animals weighing 180-200 g). A defensive AACR was formed in the animals in a shuttle box. Flashes were the conditioned stimulus, and the unconditioned stimulus was electrical stimulation of the limbs through the floor of the box, switched on 5 sec after the beginning of the conditioned stimulus. Intervals between combinations measured 30 sec. The criterion of conditioning was 10 correct avoidance responses in succession. Not more than 50 combinations were used in one experiment. Integrity of the reflexes formed was tested after seven days. The coefficient of preservation of AACR was calculated by the equation:

$$K = \frac{H_1 - H_2}{H_1} \cdot 100,$$

where H_1 denotes the number of combinations with which the conditioned reflex was formed and H_2 the number of combinations at which it was reproduced seven days after training. Right- or left-sided lobectomy was performed on the trained animals, under pentobarbital anesthesia (50 mg/kg), by extirpation of the cerebral cortex (the frontal and postfrontal region as described by Svetukhina [10]). Animals undergoing the mock operation, namely trepanning of the skull without injury to the meninges and cerebral cortex, under the same conditions, served as the control.

The animals were divided into three experimental groups: 1) undergoing a mock operation, 2) with left- or right-sided extirpation of the frontal cortex, receiving an intraperitoneal injection of 0.5 ml of sterilized physiological saline daily after the operation, and 3) with left- or right-sided extirpation of the frontal cortex, receiving intraperitoneal injections of 0.5 ml of lyophilized cortical extract (1 mg/kg) from the left or right cerebral hemisphere (LE and RE), daily after the operation. Group 1 consisted of three animals, groups 2 and 3 of 5-11 animals each. The method of preparing the extract was described previously [7]. To obtain the extracts, the cortex of each cerebral hemisphere of normal animals and of rats undergoing left-sided lobectomy nine days previously was used. This time was chosen in accordance with our previous investigations [3] which showed that restoration of motor food conditioned reflexes takes place on the 9th day after bilateral lobectomy to the extent of 30-40%, and this is accompanied by marked changes in concentrations of monoamines (serotonin, noradrenalin, and dopamine) and of their metabolites in various brain structures [3, 4].

In some of the experiments RE and LE were subjected to the action of a combination of proteolytic enzymes (Pronase) [6, 7]. To inactivate the enzymes later the solutions were boiled for 10 min and then used for injection into the animals.

Changes in muscle tone of the hind limbs were estimated 1 h after injection of LE or RE in all the animals of group 3 by the passive extension test [6]. The state of the AACR was studied in the animals of all three groups on the 4th and 9th days after the operation. To assess the significance of differences between the samples Student's t test was used. The significance of the difference between the level of preservation of AACR in the animals of group 3 relative to that of group 2 (control) was evaluated.

EXPERIMENTAL RESULTS

Unilateral lobectomy led to deep inhibition of AACR, which was not fully restored nine days after the operation (Fig. 1: a, 4 and b, 4). Physiological saline was injected intraperitoneally into all the animals of this group daily after the operation. A particularly marked disturbance of conditioned-reflex activity of the rats was observed after right-sided lobectomy: on the 4th day the AACR was absent in all animals without exception. Inhibition of AACR was due to the brain damage itself, because no disturbance of the reflexes was found in rats after a mock operation (Fig. 1: a, 1 and b, 1).

Injection of cortical extracts from the ipsilateral damaged cerebral hemispheres of normal animals from the 1st day after the operation (Fig. 1: a, 2 and b, 2) led to significant improvement of restoration of AACR compared with control rats receiving physiological saline (Fig. 1: a, 4 and b, 4). In the case of injection of LE the difference was statistically significant by the 4th day and highly significant by the 9th day (Fig. 1: a, 2).

A significant effect of restoration of AACR on the 9th day also was obtained after injection of RE from animals undergoing right-sided lobectomy (Fig. 1b, 2).

To determine the chemical nature of the active principle of the brain extracts which stimulated recovery of AACR after unilateral lobectomy, proteolyzed solutions of LE and RE were used. Proteolysis greatly reduced the effectiveness of action of LE and RE on restoration of AACR (compare a, 3 and b, 3 with a, 2 and b, 2 in Fig. 1). Experiments with proteolyzed RE and LE are evidence of a possible peptide nature of their active factors.

The effect of cortical extracts from the contralateral hemisphere on changes in AACR induced by unilateral lobectomy varied. Whereas injection of RE improved the restoration of AACR in rats with left-sided lobectomy (Fig. 1c, 6) LE had no effect on restoration of AACR in animals undergoing right-sided lobectomy (Fig. 1d, 6).

The next step was to study changes in activity of extracts isolated from the cortex of the right hemisphere of animals undergoing left-sided lobectomy nine days previously. RE of the cortex of these animals was found to be more effective than RE obtained from healthy rats in its action on restoration of AACR after left-sided lobectomy (compare c, 6 and c, 5 in Fig. 1). By the 4th day the level of restoration of AACR was significantly higher than in the control, and on the 9th day the significance of the difference was higher than in the case of injection of RE from normal animals. However, no stimulating effect of RE from animals undergoing leftsided lobectomy on AACR in rats with damage to the cortex on the right side was observed (compare d, 2 and d, 5 in Fig. 1). Consequently, the increased effectiveness of the stimulating action of RE from donor animals after left-sided lobectomy was manifested only when administered to recipient rats with left-sided cortical damage.

The data described above are proof of differences in the effectiveness of extracts obtained from the cortex of the right or left cerebral hemispheres of rats in relation to their action on restoration of AACR disturbed by unilateral lobectomy. The results of the investigation are evidence that the stimulating action of the extracts on the process of compensatory recovery depends on the side of the lesion. For instance, whereas RE of healthy animals improves restoration of AACR disturbed by unilateral lobectomy. The results of the investigation are evidence that the stimulating action of the extracts on the process of compensatory recovery depends on the side of the lesion. For instance, whereas RE of healthy animals improves restoration of AACR in rats after both right- and left-sided lobectomy, LE activates conditioned-reflex activity only of rats undergoing left-sided lobectomy.

Unilateral injury to the cerebral cortex is known to change the direction of the animals' movements in a T-maze [11], and this is bound to have some effect on the results of an investigation in which the correct choice of side is the basis of the animal's behavior. It must also be recalled that LE and RE can induce lateralized changes in muscle tone in both normal and lobectomized animals. In the present investigation these circumstances were monitored. First, a bilateral avoidance method was used, in which there was no preference for any one direction. Second, preservation of AACR began to be investigated when the action of LE and RE on muscle tone was long after completion.

Thus the results of this investigation, indicating lateralization of the effect of LE and RE on restoration of AACR when disturbed by unilateral lobectomy, point to the possibility that endogenous peptides specific for each cerebral hemisphere can exert an oriented effect on restorations of CNS functions.

LITERATURE CITED

1. G. A. Vartanyan and Yu. V. Balabanov, Gagra Colloquia [in Russian], No. 7, Tbilisi (1979), pp. 279-287.
2. G. A. Vartanyan and B. I. Klement'ev, Fiziol. Cheloveka, 9, No. 1, 122 (1983).
3. T. V. Grekhova, G. A. Romanova, and A. N. Sovetov, Patol. Fiziol., No. 4, 57 (1985).
4. T. V. Grekhova, V. S. Kudrin, I. I. Miroshnichenko, and G. A. Romanova, Byull. Éksp. Biol. Med. (in press).
5. G. N. Kryzhanovskii, Determinant Structures in Pathology of the Nervous System [in Russian], Moscow (1980).
6. G. N. Kryzhanovskii, V. K. Lutsenko, and M. Yu. Karganov, Byull. Éksp. Biol. Med., No. 10, 404 (1981).
7. G. N. Kryzhanovskii, V. K. Lutsenko, and M. Yu. Karganov, Byull. Éksp. Biol. Med., No. 1, 14 (1982).
8. G. N. Kryzhanovskii, V. K. Lutsenko, and M. Yu. Karganov, Neuropeptides: their Role in Pathology [in Russian], Tomsk (1985), pp. 144-145.
9. V. K. Lutsenko and M. Yu. Karganov, Neirokhimiya, 4, No. 2, 197 (1985).
10. V. M. Svetukhina, Arkh. Anat., 42, No. 2, 31 (1962).
11. E. I. Synkov, Mechanism of Brain Plasticity in Functional and Pathological Situations [in Russian], Vol. 2, Makhachkala (1982), pp. 130-131.
12. T. Morau, K. L. Kubos, P. R. Sanberg, and R. G. Robinson, Behav. Neurosci., 98, 742 (1984).
13. R.-G. Robinson and J. T. Coyle, Brain Res., 188, 63 (1980).