function of a system [1,4]. Such a mechanism may in fact underlie the action of many low-intensity factors, particularly physical ones, determining their manifest influence on biological systems.

The specific nature of resonance structures of the body may be very different: mechanisms of electron and ion transport underlie virtually all the processes of production and utilization of energy, metabolism regulation, and different intricate physiological functions. The most important of these are regulatory rhythms of the brain, local processes of organic self-regulation, as well as intracellular rhythmic processes of intercoordinated activity of organelles and, above all, processes involving membrane structures which determine electrical and chemical gradients, ion transport, etc.

REFERENCES

1. I. I. Blekhman, *Synchronization in Nature and Technology* [in Russian], Moscow (1981).
2. V. I. Dontsov and T. A. Ismailov, *Pat. Fiziol.*, № 5, 65-66 (1984).
3. A. A. Podkolzin and E. P. Markin, *Izv. Akad. Nauk SSSR*, **54**, №10, 1961-1963 (1990).
4. E. N. Chirkov and Yu. N. Babaev, *Electromagnetic Nature of Immunity and Cell Differentiation* [in Russian], Preprint, Akad. Nauk Ukr. SSR, Institute of Cybernetics, № 62, (1987), pp. 1-18.
5. A. J. Cunningham, *Nature*, **207**, 1106-1107 (1965).
6. E. Ettlenne, P. Hoeng, and R. Frankel, *Effect of Magnetic Fields on Ca Transport in Isolated Muscle Microsomes: NSWC/URSI Spring Meet.*, Seattle, Washington (1979), p. 442.
7. J. Walleczek and R. P. Liburdi, *FEBS Lett.*, **271**, № 1-2, 157-160 (1990).

Comparative Study of Transport of Exogenous Radioactive Histone Administered by Various Routes

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Accumulation of radioactive label in olfactory bulbs is found to outstrip the label content in the blood of animals to which histone is administered intranasally. The concentration of the label in the blood is highest after intraperitoneal injection, but shortly afterwards radioactive histone is detected in the brain as well.

Key Words: *radioactive histone; various routes of administration; accumulation in the blood and brain*

The intranasal route of drug delivery is becoming more and more popular of late, particularly when it concerns a certain class of pharmacological compounds. This applies first of all to peptides, among which many substances have been found to be useful for the treatment of many serious diseases, including diseases of the central nervous system (CNS). The possibility of such dosage forms and their sufficient efficacy necessitate a detailed study of this route of drug administration in order to better validate it and use it on a wider scale, with other drugs of peptide and nonpeptide nature as well.

In the present research we compared the penetration of peptides or small proteins into the CNS after intranasal and intraperitoneal adminis-

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tration thereof. Iodine-labeled histone, a polycationic protein of calf thymus characterized by high transport properties vis-a-vis the blood-brain barrier [1], was used.

MATERIALS AND METHODS

Sulfate of histone from calf thymus (Reachim, Latvia, Biolar Research and Manufacturing Conglomerate) was labeled with iodine monochloride after modified McFarline's method [6] in experiments with intranasal and intraperitoneal administration. The Specific activity of the labeled preparation was 5 μcCi/mg. For historadioautographic studies iodination of histone and bovine serum albumin (Sigma, USA) was carried out by the standard method with chloramine [3]. The specific activity of the labeled preparations was 60 μcCi/mg for total histone and 40 μcCi/mg for bovine serum albumin.

Experimental studies of various routes of administration of radioactive histone were carried out with 60 laboratory mice weighing 25-30 g. Labeled histone was administered intranasally in normal saline in a dose of 0.35 mg per animal. The same amount of histone was used for intraperitoneal injection. The animals were decapitated 15, 30, and 60 min after histone administration, blood was collected, and various portions of the brain (olfactory bulbs, cerebral hemispheres) were taken for examination. Activity of ^{125}I γ-radiation was measured with a Gamma-12 device. Radioactivity level was assessed per 100 mg dry mass of tested tissue, with a histone solution of the concentration used in the experiment taken as the reference.

The penetration of labeled histone and albumin in various portions of the brain was historadiographically assessed in rats weighing 175-200 g. The agents were injected into the right common carotid artery of anesthetized rats in a dose of 8-12 mg of protein per animal. Perfusion of the vascular bed with a mixture of Ringer's solution and cattle serum was carried out 10-12 min after the infusion was started. Various portions of the brain were then removed and fixed in 10% neutral formalin. Penetration of ^{125}I histone in various structures of the brain was assessed by the semiquantitative visual historadiographic method. For control ^{125}I-labeled albumin was used, which is known never to cross the blood-brain barrier. Samples of the motor cortex, olfactory bulbs, hippocampus, hypothalamus, and cerebellum after fixation were embedded in paraffin by the standard method. An emulsion (Ilford-L4, England) was layered onto the slices. After exposure and development of the samples the slices were stained with hematoxylin eosin. The distribution of radioactivity "tracks" was examined under an Opton microscope.

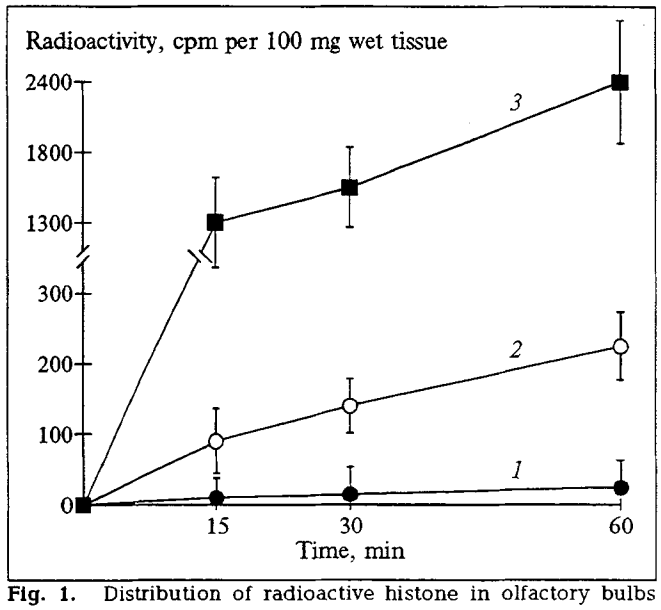

Fig. 1. Distribution of radioactive histone in olfactory bulbs (*1*), cerebral hemispheres (*2*), and blood (*3*) in various periods after intraperitoneal injection.

RESULTS

Histone was detected in high concentrations in the blood and all the tested brain structures 15 min after intraperitoneal injection (Fig. 1), radioactivity accumulation in the blood being more rapid than in the nervous system structures. The radioactivity level was higher in the cortical parts than in the

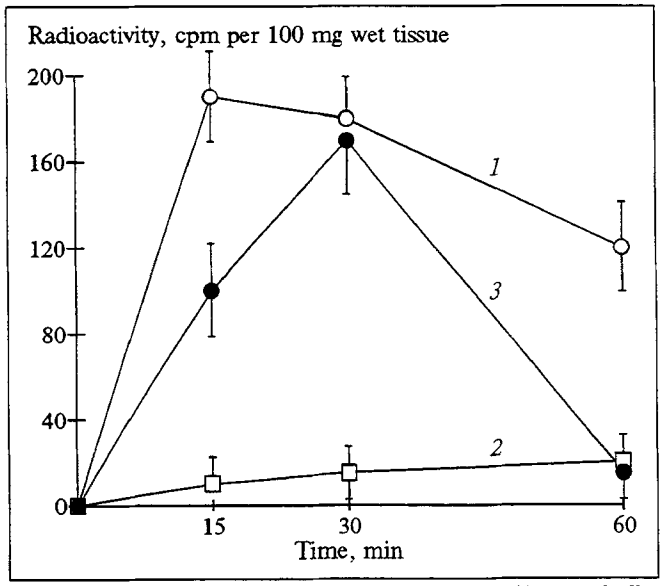

Fig. 2. Distribution of radioactive histone in olfactory bulbs (*1*), cerebral hemispheres (*2*), and blood (*3*) in various periods after intranasal administration.

olfactory bulbs and was still increasing one hour after administration in both preparations, as well as in the blood. Quite a different pattern of radioactivity distribution in the blood and brain preparations was observed after intranasal administration of ^{125}I histone (Fig. 2). During the first 15 min the content of radioactive histone in the olfactory bulb was almost twice as high as in the blood and 40 times higher than in the cerebral cortex. After 30 min its content was similar in the bulbs and blood, and after one hour it started decreasing.

On the other hand, reduction of the ^{125}I histone content in the blood was approximately 5 times more rapid than in the olfactory bulbs. The histone content in the cerebral cortex detected in all periods was much lower than in the blood and olfactory bulbs. The results of the experiments with intranasal administration of histone indicate its independent transport into the olfactory bulbs and blood. Its entry into other parts of the brain, however, may be secondary and mediated by transport via the circulation, provided that labeled histone molecules can cross the blood-brain barrier. Historadiographic studies on rats confirmed such a capacity of histone molecules. Ten to twelve minutes after injection of ^{125}I histone into the blood and perfusion, radioactive label was detected in all the tested structures of the brain. Analysis of historadiograms showed an uneven distribution of ^{125}I histone in various parts of the brain. Its highest levels were revealed in the hippocampus and hypothalamus. The intensity of histone entry in these brain portions was virtually the same. In the hypothalamic area the greatest accumulation of the label was observed in extracellular formations, that is, in the neuropil, in contrast to cellular structures. In the hippocampal zone ^{125}I histone was found mostly at sites of accumulation of neuronal cellular bodies and, to a lesser degree, in the neuropil. Penetration of labeled histone into the sensorimotor cortex was lower than into the above-named brain portions. The content of ^{125}I histone in the cerebellum was negligible, although it was detected in all its cellular layers. Histone penetration in olfactory bulb tissue was negligible as well. In addition, intensive impregnation of the vascular plexus of the third ventricle with labeled histone was observed, with "label" localized along the endothelial lining and "tracks" observed both in the plasma and basal membrane zones. Iodine-labeled serum albumin used for control did not penetrate into the corresponding brain parts. The incidence of "tracks" over nervous tissue of the brain did not reliably differ from the baseline one outside the histological preparation. Hence, the historadiographic study revealed penetration of exogenous histone from blood plasma to nervous tissue of the brain. The pattern of label distribution in such parts of the brain as the sensorimotor cortex and olfactory bulbs in historadiographic experiments with rats fully coincided with the results of experiments with intraperitoneal injection of ^{125}I histone to mice (Fig. 1). It is possible that labeled histone binds, first of all, to the anion sites of fenestrated capillaries of the brain, such as the vascular plexus, and penetrates into cerebral ventricles, from which it later enters the parenchyma of various parts of the brain. Such a type of histone entry in the brain is observed both after its direct administration into the blood and after intraperitoneal injection, this suggesting that histone can cross the blood-brain barrier and pass through cell membranes; this picture can hardly be distorted at the expense of ^{125}I histone degradation products in experiments of the chosen duration [2]. The different pattern of radioactive label distribution after intranasal administration of histone permits us propose two independent channels of its entry in various tissues of an animal. Labeled histone entry into the blood may be mediated by the capillary net of the respiratory epithelium of the nasal cavity, which is very well developed. Our results demonstrate that the maximal increase of the content of radioactive histone in the blood is observed by the 30th min, after which it gradually decreases. A similar pattern of entry into the blood is observed for intranasal administration of calcitonin in experiments on human volunteers [8]. Another portion of radioactive histone may be directly transported to the olfactory bulbs either by axoplasmic transport via fibers of olfactory neurons, or via the lymphatic system. Unfortunately, there are no published data on the contribution of the lymphatic system, but there are reports about the presence in the olfactory system of rapid axon transport realized in both directions [9]. The velocity of movement along the sciatic nerve of mammals for rapid axoplasmic transport is about 17 mm/h [7]. Since the distance which the transported molecules have to overcome from the surface of the nasal cavity along the olfactory nerve bundles to the olfactory bulbs is very short in mice, just a few millimeters, a 15-min exposure of radioactive histone in the nasal cavity is sufficient for its transport and accumulation in the olfactory bulbs. The data of other research with wheat agglutinin conjugated with horseradish peroxidase [4,5] demonstrating the presence in the olfactory system of transsynaptic transport of high-molecu-

lar compounds may be one more piece of evidence of the possibility of axonal transport of radioactive histone in the olfactory system structures.

REFERENCES

1. D. Al-Sufi, O. A. Goryukhina, G. P. Dem'yanenko, and Yu. V. Bobryshev, *Vestnik Leningr. Gos. Univ.*, Ser.3, Issue 3, № 17, 129-132 (1991).
2. O. A. Goryukhina and I. P. Ashmarin, *Vopr. Med. Khim.*, **20**, № 6, 652-655 (1974).
3. L. A. Osterman, *Study of Biological Macromolecules by Electrofocusing, Immunoelectrophoresis, and Radionuclide Methods* [in Russian], Moscow (1983), pp. 236-239.
4. H. K. Baker and R. F. Spenser, *Exp. Brain. Res.*, **63**, 3461-3473 (1986).
5. S. K. Itaya, *Brain Res.*, **409**, № 2, 205-214 (1987).
6. A. S. McFarline, *Nature*, **182**, 53 (1985).
7. S. Ochs, *J. Physiol. (Z.)*, **227**, 627-645 (1972).
8. A. M. Pontizoli, A. Aiberetto, E. Calderara, *et al.*, in: *Delivery Systems for Peptide Drugs*, London (1986), Vol. 125, № 7, pp. 249-253.
9. G. Weiss and K. Buchner, in: *Molecular Neurobiology of the Olfactory System*, London (1988), № 7, pp. 217-233.

PHARMACOLOGY

Drug Correction of Impaired Strophanthin Tolerance during Simulated Cardiac Decompensation

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The risk of glycoside intoxication under conditions of simulated acute and subacute cardiac insufficiency may be reduced by ajmaline, anaprilin, and combinations of each of these drugs with pentamine or nitroglycerin.

Key Words: *correction of strophanthin toxicity, cardiac insufficiency*

Administration of cardiac glycosides representing the main group of cardiotonic agents for emergency use in acute cardiac failure caused by coronary heart disease, for example, sometimes does not result in adequate improvement of myocardial contractility and recovery of impaired hemodynamics. The therapeutic effect of glycoside cardiotonics is limited by a sharply increased sensitivity to their toxic action [3,10,11]. Since management of heart failure envisages that, in addition to cardiac glycosides, quite a number of other drugs be used, indicated individually for each specific clinical situation, we thought it interesting to test on models of acute and subacute cardiac insufficiency the effects of some drug combinations most frequently used in clinical cardiology on tolerance for the toxic effect of strophanthin.

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MATERIALS AND METHODS

Experiments were carried out with 385 Wistar rats weighing 160-170 g narcotized with sodium thio-

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