THE ROLE OF DERMORPHIN IN THE REGULATION OF THE PROCESSES
OF HIBERNATION IN MAMMALS

T. N. Soilertinskaya, É. N. Nuridinov, and M. F. Obukhova

Data are presented in this paper regarding the influence of the opioid peptide, dermorphin, and its conjugate on the conditioned reflex activity of sousliks. The administration of dermorphin (0.1 µg, subcutaneously) is accompanied by a fall in body temperature, slowing of the baseline vegetative indices, and the sinking of the animal into a sleep-like state. A substantial disturbance of conditioned reflex activity was identified in the late period (delayed effects). Immunization of the hibernating sousliks with a conjugate of dermorphin leads to a gradual awakening of the animals from hibernation, to the normalization of conditioned reflex activity, and to the appearance of the motoric component of a conditioned defense reaction. The question of a possible specific role of dermorphin in the mechanisms of hibernation is discussed.

The problem of the neurochemical regulation of the processes of higher nervous activity and the functional state of hibernating animals is a topical and little-studied subject in ecological physiology. According to the data of foreign authors, an increase in the number of opioid neuropeptides in the blood plasma [22] is observed in the process of hibernation. In the opinion of S. M. Parin and E. V. Golanov [12], the opioid peptidergic system occupies an important place in the physiological mechanisms of adaptation and in increasing resistance to noxious factors. Margulis, Goldman, and Fink [18] advanced the position, based on investigations involving the administration of naloxone, an antagonist of opiate receptors, to hibernating mammals, according to which hibernation is an opioid-dependent state, and that endogenous opioids may be specifically involved in the mechanisms of its regulation. There are now data in the literature pointing to the fact that a number of peptides (bombesin, thyroliberin, opioids, etc.) are regulators of the physiological functions of hibernating mammals [17, 24]. The detailed study of the physiological effects of the neokyotorphin-peptide, isolated from the brain of the souslik, on hibernating and nonhibernating animals, was made in a paper of D. A. Ignat'ev, et al. [13]. It was found that a stimulating effect of this peptide on cardiac activity, level of oxygen consumption, and the emergence of the animal into the normothermic state is observed when it is administered to hibernating sousliks. However, few investigations are known of the role of the opioid neuropeptides in the regulation of the higher nervous functions, the most delicate indicator of the functional state of the organism of hibernating animals. Of these, from our point of view, dermorphin is of special interest. Isolated in 1980 from the skin of a frog, it is a heptapeptide possessing the very prolonged opioid activity [9]. There has been information in recent years in the foreign literature regarding the discovery of dermorphin-like peptides in the brain structures of mammals [20, 21].

Its marked analgesic effect is a characteristic feature of dermorphin. This fact has served as the basis of attempts to use this neuropeptide in clinical practice for the elimination of painful symptoms of varied etiology. There are few data regarding the influence of dermorphin on behavioral activity and memory processes in mammals. Castellano and Povono [16] observed disturbances of passive behavior after it was administered to mice. The changes observed were dose-dependent in character. Broccardo [15] observed the phenomenon of catalepsy when large doses of dermorphin were administered to rats. According to the data of E. Yu. Baturina, et al. [2], the administration of dermorphin to rats is accompanied by a marked analgesic effect and a change in motoric activity. In the opinion of T. G. Emel'tyanova, et al. [3], dermorphin may be regarded as a thermoregulatory peptide influencing both central and peripheral thermoregulatory mechanisms. It was found in comparative physiological investigations of the laboratory of A. I. Karamyan that the administration of dermorphin to lizards (monitors) and hedgehogs leads to their sinking into a sleep-like state, to a fall in body temperature,
and to a marked analgesic effect. A significant disruption of conditioned reflex activity was observed in the hedgehogs.
Changes in behavioral activity following the administration of dermorphin were most prolonged in lizards [7, 8, 10]. It seemed to be of interest to investigate the influence of dermorphin on the acquired and inborn forms of nervous activity in another representative of the hibernating mammals, the rodents (the Aral yellow souslik). In the light of the promise of the new approach developed by I. P. Ashmarin [1], consisting in the possibility of prolonged alteration of the physiological and biochemical status of the organism though the active immunization of animals with conjugates of endogenous regulatory peptides, it seemed of interest to study the effect, in addition to dermorphin itself, of its conjugate with a carrier antibody in the hibernation period.

We have not found such studies in the literature.

METHODS

The experiments were carried out in 16 Central Asian Aral yellow sousliks on the basis of the unified motoric-defense methodology of V. M. Bekhterev-Protopopov. To study the features of the higher nervous activity of the souslik, the animal was placed in a specially designed mount so as to fix the trunk while leaving the head and extremities free. The electrogram was picked up by means of plate electrodes in the thoracic lead. The active electrode was placed in the region of the heart, and the indifferent electrode was placed on the lower extremity. The pneumogram was recorded by means of a special angle sensor.

The electrodermal stimulation was applied to the right lower extremity by means of a special cuff. The duration of the electrodermal stimulation was 200 msec, twice the threshold of the motoric reaction of the animal.

The body temperature was measured in the rectum with an infant’s thermometer. All the indices of the conditioned reflex activity were recorded on an ink-writing encephalograph produced by Sanei (Japan). A sound of 500 Hz, 50 dB above the threshold of human hearing, was used as the conditional stimuli for the sousliks. The time of the isolated action of the conditional signal was 6 sec. There were four series of experiments. In the first series, conditioned reflexes to the acoustic stimuli were developed in the intact sousliks in the active state; in the second series, the possibility of developing and maintaining conditioned reactions in the period of hibernation was studied; in the third series, the changes in higher nervous activity in the awake animals following the administration of dermorphin were studied. In the fourth series, the changes in the functional state of the organism and in conditioned reflex activity in the hibernating sousliks were investigated against the background of immunization with a conjugate of dermorphin. The dermorphin preparation (Serva) was injected subcutaneously in physiological solution in a dose of 0.1 mg/kg 10 min immediately before the experiment. The dermorphin conjugate with bovine serum albumin, obtained using glutaraldehyde, as described in reference[s] [1, 11], was injected subcutaneously in a dose of 0.15 mg/kg with Freund’s complete adjuvant, twice at an interval of 10 days. The statistical analysis of the data was done in blocks, averaging the data of three experiments. The significance of the differences was established on the basis of the character of the distribution using the Student’s and Fisher’s tests.

INVESTIGATION RESULTS

Features of the Higher Nervous activity of Awake and Hibernating Sousliks. The experiments on the formation of conditioned reflexes in sousliks were carried out in several stages. First the animals were accustomed to the experimental conditions, then the orienting reaction was extinguished, and only then was the development of the conditioned reflexes commenced. The study of the sequence of the inclusion of various functional systems in the conditioned reflex activity of the brain has shown that it proceeds in a stepwise fashion. The initial changes were recorded in the vegetative indices. Thus, the rate of respiratory movements increased by 54.0 ± 5.7% on the average. The heart rate increased (by 2.5 ± 1.9% on the average) in the first experiments (at the stage of generalization of the conditioned reactions). A pronounced tendency toward their decrease (by 1.5 ± 0.12% on the average) as compared with the baseline was found in the stage of concentration of the conditioned reactions. The motoric component of the conditioned defense reaction appeared later (after 25–30 combinations), was less pronounced, and was consolidated with difficulty. It only reached the 50–65% level at the stage of the consolidated conditioned reactions, and was variable, both with respect to its manifestation and with respect to its latent period. The latent period ranged on the average from 2 to 4 sec. The formation of extinctive inhibition was wave-like in character. The motoric component of the conditioned defense reaction extinguished first (four