7. D. E. Rotter and J. M. Rowlands, J. Pharmacol., **1**, 13 (1981).
8. J. A. Scott, B. A. Khaw, J. T. Fallon, et al., J. Mol. Cell Cardiol., **18**, 1243 (1986).
9. J. Wolf and A. B. Jones, Proc. Nat. Acad. Sci. USA, **65**, 454 (1979).

β-LIPOTROPIN AND β-ENDORPHIN IN MECHANISMS OF COMPENSATION OF FUNCTIONS AFTER DESTRUCTION OF THE LATERAL HYPOTHALAMUS

F. I. Dzhafarov

UDC 612.81+612.391:612.015.348

KEY WORDS: β-lipotropin; β-endorphin; lateral hypothalamus

Investigations have demonstrated the physiological role of opioids and other oligopeptides in the central mechanisms of hunger and thirst [3, 7, 12, 14]. Of all the neuropeptides, with their broad spectrum of action, one which is particularly interesting is β-lipotropin (β-LPT), which is widely distributed in various structures of the CNS and, in particular, in the hypothalamus [1, 13]. There is also evidence that β-LPT and its derivative β-endorphin can change the character of hunger-motivated food-related instrumental activity in rabbits and drinking behavior in rats [4-10].

Accordingly, in the investigation described below the basic parameters of biological motivations and related physiological functions were studied after destruction of the lateral hypothalamus in order to establish the possible involvement of β-LPT and its derivative, β-endorphin, in the mechanisms of compensation of these functions under the conditions specified, a problem which still remains virtually unstudied.

EXPERIMENTAL METHOD

Experiments were carried out on 45 noninbred male rats weighing 200-250 g. The animals were divided into three groups, with 15 in each group. Group 1 (control) consisted of animals in which the lateral hypothalamus was destroyed and physiological saline injected into the lateral ventricles in a volume of 3-5 μl; in the animals of group 2 the lateral hypothalamus was destroyed and intraventricular microinjections of β-LPT given; in the rats of group 3, after destruction of the lateral hypothalamus, β-endorphin was injected into the lateral ventricles. The lateral hypothalamus of the animals was destroyed by electrical coagulation under general (pentobarbital) anesthesia, using stereotaxic coordinates of an atlas of the rat brain [11]: P = 1.7-2.0 mm, L = 1.5-1.7 mm, H = 8.0-8.5 mm. For electrolytic destruction an anodal current of 50 mA was passed for 5 sec. To inject the substances cannulas 0.84 mm in diameter were inserted into the lateral ventricles in accordance with stereotaxic coordinates: AP = 1.5-1.7 mm, L = 2 mm, H = 4-4.5 mm [17]. These peptides were injected through the implanted cannulas in concentrations of $91.5 \cdot 10^{-6}$ μmoles/μl (for β-LPT) and $269 \cdot 10^{-6}$ μmole/μl (for β-endorphin) in a volume of 3-5 μl (in physiological saline) by means of a microinjector.

β-LPT and β-endorphin were obtained from the Laboratory of Protein Hormones, Research Institute of Experimental Endocrinology and Hormone Chemistry, Academy of Medical Sciences of the USSR, and were isolated from bovine pituitary glands [5].

*Deceased.

P. K. Anokhin Research Institute of Normal Physiology, Academy of Medical Sciences of the USSR, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR A. V. Val'dman.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 111, No. 6, pp. 629-632, June, 1991. Original article submitted February, 1990.

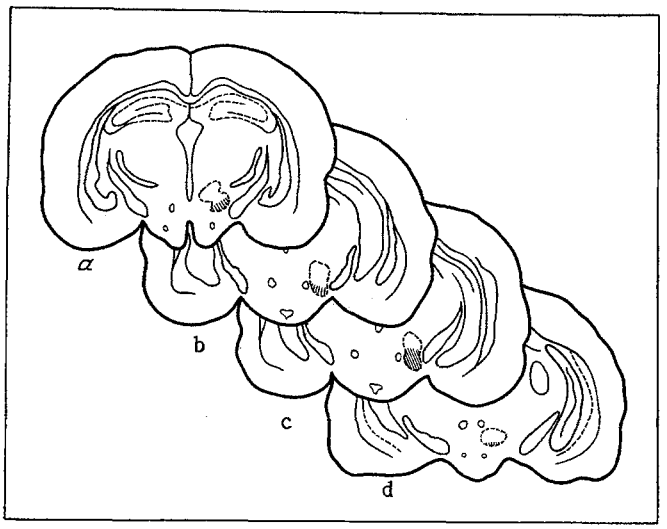

Fig. 1. Reconstruction of brain sections showing location of zone of coagulation of lateral hypothalamus in rat No. 97. a) Section passes through hypothalamus at level of dorsomedial and ventromedial hypothalamic nuclei. P = 2.5; L = 0.5-2.0; H = 4.5-5.5. b) Section passes through level of mammillary body. Zone of destruction involves lateral region of hypothalamus (its caudal part), and also medial lemniscus (above and laterally). P = 2.7, L = 0.5-2.0, H = 5.0-6.0. c) Level close to level B, but somewhat more caudally. Destruction observed even more ventrally than at previous levels and involves lateral region of hypothalamus more fully. P = 3.2, L = 0.5-1.5, H = 5.5-6.5. d) Level of section lies in region of substantia nigra and caudal part of lateral hypothalamus. P = 3.5, L = 0.5-1.0, H = 6.0-7.0.

The rats were kept in individual cages equipped with hanging containers of food and water. The experiments were conducted in three stages: I) adaptation of the animals to individual cages (10-12 days), II) background observation (10 days), III) experimental procedure (15 days). The daily quantity of food and water consumed, the diuresis, body weight and temperature, and manifestations of orienting-investigative activity (rearings, horizontal movements), comfort behavior (grooming, cleaning, licking), and spontaneous drowsiness and sleep were recorded at intervals in all the animals.

After the end of the experiments the animals were killed, the brain removed, histological sections were cut, and the location of the zone of destruction in the region of the lateral hypothalamus was then determined by a rapid photographic method, after which reconstruction was carried out on brain sections stained by Nissl's method, and morphometric analysis was undertaken. Reconstruction of brain sections showing the site of destruction of the lateral hypothalamus is illustrated in Fig. 1.

Values of the physiological parameters studied were subjected to statistical analysis by traditional methods of biometrics and Student's t test.

EXPERIMENTAL RESULTS

In the animals of group 1 (control) two of the 15 rats died as a result of destruction of the lateral hypothalamus on the 7th-8th days. They showed progressive loss of body weight, reduction of food and water consumption, and lowering of the diuresis and body temperature. The remaining 13 rats showed general tendencies of these parameters, which were most marked on the 9th-12th day after destruction of the lateral hypothalamus. Data for this group are pooled in Table 1. It must be emphasized that toward the 9th-12th days after destruction of the lateral hypothalamus, compared with the background values a decrease was observed in body weight (on average by 19.6%), food intake (on average by 30%), and water intake (by 26.8%). The diuresis also was reduced (on average by 26%). The body temperature showed a tendency to

TABLE 1. Time Course of Physiological Parameters after Destruction of Lateral Hypothalamus in Rats (average for group, n = 13)

Parameter	Background 10 days be before destruction	Days after destruction					
		1	3	6	9	12	15
Body weight, g	242,9±6,09***	222,0±6,51*** (91,4)	211,7±7,95*** (87,2)	200,7±9,48*** (82,6)	197,8±10,29*** (81,4)	207,3±7,86*** (85,3)	215,7±7,78*** (88,8)
Quantity of food, g	9,0±0,26***	1,2±0,33** (13,3)	4,2±0,90*** (46,7)	6,8±1,33*** (75,6)	6,3±1,09** (70,0)	7,3±0,94*** (81,1)	6,8±0,58** (75,6)
Volume of water, ml	11,2±0,76***	2,7±0,70** (24,1)	5,4±1,16*** (48,2)	8,3±1,27*** (74,1)	8,2±1,70** (73,2)	7,4±0,96*** (66,1)	6,4±1,40** (57,1)
24-hourly diuresis, ml	7,3±0,60***	6,7±0,54*** (91,8)	5,2±0,72*** (71,2)	5,0±0,92*** (68,4)	5,4±1,39** (74,0)	6,6±1,38*** (90,4)	6,8±1,30*** (93,2)
Body temperature, °C	37,2±0.10***	37,9±0,29*** (100,9)	37,8±0,16*** (101,6)	37,4±0,21*** (100,5)	37,4±0,29*** (100,5)	37,7±0,42*** (101,3)	37,4±0,37*** (100,5)
State of immobility, min	44,5±1,32***	30,5±4,44*** (68,5)	40,7±3,17*** (91,5)	37,1±4,24*** (83,4)	39,5±3,77*** (88,8)	35,8±2,88*** (80,5)	36,1±1,98*** (81,1)
Comfort-associated behavior, min	8,7±0,68***	18,5±4,04** (212,6)	13,7±2,12*** (157,5)	12,3±2,10*** (141,4)	12,9±2,20*** (148,3)	5,4±0,88*** (177,0)	16,4±1,69*** (188,5)
Orienting-investigative activity, min	0,9±0,21**	0,8±0,33** (88,9)	0,2±0,06** (22,2)	0,2±0,08* (22,2)	0,5±0,13** (55,6)	0,5±0,12** (56,6)	0,6±0.18** (66,7)

Legend. Here and in Tables 2 and 3, percentage of change compared with background value, taken as 100%, shown between parentheses; significance of difference compared with background values: *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

TABLE 2. Time Course of Physiological Parameters in Rats Receiving β-LPT after Destruction of Lateral Hypothalamus (average for group, n = 15)

Parameter	Background 10 days before destruction	Days after destruction and injection of β -LPT					
		1	3	6	9	12	15
Body weight, g	206,6±4,48***	176,6±4,67*** (85,5)	180,5±4,69*** (87,4)	187,5±4,70*** (90,8)	190,1±4,09*** (92,0)	204,4±5,08*** (98,9)	227,3±4,94*** (110,0)
Quantity of food, g	7,9±0,24***	3,9±0,14** (49,4)	5,7±0,51*** (72,2)	8,0±0,54*** (101,3)	8,6±0,95** (108,9)	7,9±0,23*** (100)	8,3±0,58** (105,1)
Volume of water, ml	20,6±0,57***	6,6±0,18** (32)	11,1±1,42*** (53,9)	17,4±5,27** (84,5)	20,3±1,58** (98,5)	20,6±0,33*** (100)	20,3±1,26*** (98,5)
24-hourly diuresis, ml	17,0±1,06***	4,2±0,68*** (24,7)	7,4±1,01*** (43,5)	11,7±1,49*** (68,8)	16,5±1,46*** (97,1)	16,4±0,87*** (96,5)	16,2±1,49*** (95,3)
Body temperature, °C	37,0±0,05***	37,0±0,11*** (100,8)	37,3±0,12*** (100)	37,0±0,08*** (100(	37,0±0,08*** (100)	37,0±0,05*** (100)	37,0±0,05*** (100)
State of immobility, min	42,5±1,0***	41,6±2,12*** (97,9)	36,1±2,05*** (84.9)	37,4±3,04*** (88)	34,4±1,33*** (80,9)	38,4±1,25*** (90,4)	33,5±0,32*** (78,8)
Comfort-associated behavior,	11,3±0,56***	8,4±0,79*** (74,3)	11,9±0,77*** (105,3)	13,1±1,07*** (115,9)	10,3±0,85*** (91,2)	9,8±0,58*** (86,7)	10,3±0,86*** (91,2)
Orienting-investigative activity, min	0,9±0,20**	1,3±0,15*** (144,4)	1,1±0,26** (122,2)	1,6±0,38** (177,8)	1,5±0,38** (166,7)	1,1±0,25** (122,2)	1,6±0.40** (177,8)

rise. Unlike the other parameters tested, the volume of water consumed by the rats of this group continued to fall until the 15th day of observation. These animals also showed an increase in the frequency of comfort-associated forms of behavior, reduction of orienting-investigative activity, and shortening of the period of sleep and drowsiness (state of immobility, Table 1). An example of the individual trend of the parameters after destruction of the lateral hypothalamus is given in Fig. 2.

Immediately after destruction of the lateral hypothalamus the rats of group 2 were given intraventricular microinjections of β-LPT. By contrast with the animals of group 1, development of the syndrome of destruction of the lateral hypothalamus had specific differences in the animals of this group. For instance, starting with the 3rd day of destruction of the lateral hypothalamus and subsequent injection of β-LPT, most rats showed a tendency for all the autonomic parameters (except body temperature) recorded to be restored. By the 15th day, these parameters were virtually restored to the background values. Moreover, the body weight of all animals exceeded (on average by 10%) the background values by this time (Table 2). The most characteristic feature of the action of β-LPT after destruction of the lateral hypothalamus was increased orienting-investigative activity, accompanied by inhibition of comfort-associated behavior and lengthening of the period of wakefulness. An individual example of the trend of these parameters is given in Fig. 3.

In rats of group 3, after electrical coagulation of the lateral hypothalamus intraventricular microinjections of β-endorphin were given. The action of β-endorphin in these animals had specific differences. Unlike in animals of the control group and group 2, food consumption and body weight of the rats of group 3 were restored almost to their background levels after 12-15 days, starting with the 1st day after injection of β-endorphin the volume of diuresis rose sharply, and by the 15th day of observation it was 34.5% higher than in the background period, but the volume of water

TABLE 3. Time Course of Physiological Parameters in Rats Receiving β-Endorphin after Destruction of Lateral Hypothalamus (average for group, n = 15)

Parameter	Background 10 days before destruction	Days after destruction and injection of β-endorphin					
		1	3	6	9	12	15
Body weight, g	222,7±6,30***	196,41±5,18*** (88,2)	197,7±5,71*** (88,8)	202,0±5,16*** (90,7)	212,5±5,27*** (95,4)	217,8±5,31*** (97,8)	225,7±6,10*** (101,3)
Quantity of food, g	10,6±0,54***	1,9±0,80* (17,9)	7,1±1,32*** (67,)	9,5±0,89*** (89,6)	10,3±0,75*** (97,2)	10,2±0,58*** (96,2)	10,3±0,59*** (97,3)
Volume of water, ml	10,6±0,72***	1,8±0,51** (17)	5,8±0,87*** (54,7)	8,7±0,70*** (82,1)	10,0±0,38*** (94,3)	10,0±0,48*** (94,3)	0,6±0,61*** (90,6)
24-hourly diuresis, ml	5,5±0,28***	6,7±1,03*** (121,8)	7,6±0,29*** (138,2)	6,0±0,72*** (109,1)	6,8±0,61*** (123,6)	7,3±0,61*** (132,7)	7,4±0,88*** (134,5)
Body temperature, °C	37,1±0,07***	38,6±0,22*** (104,1)	38,0±0,09*** (102,5)	37,5±0,26*** (101,1)	37,7±0,20*** (101,6)	37,8±0,16*** (101,9)	37,8±0,27*** (101,9)
State of immobility, min	35,1±2,08***	38,3±3,63*** (109,1)	41,0±3,35*** (116,8)	38,5±2,29*** (109,7)	34,5±1,51*** (98,3)	37,9±2,78*** (108)	41,5±0,81*** (118,2)
Comfort-associated behavior, min	12,9±0,49***	13,2±2,36*** (102,3)	12,1±1,45*** (93,8)	14,6±2,34*** (113,2)	14,5±1,29*** (112,4)	14,4±1,71*** (111,6)	11,2±0,53*** (86,8)
Orienting-investigative activity, min	0,6±0,09***	0,9±0,40* (150)	0,6±0,19** (100)	0,41±0,11** (66,7)	0,3±0,14* (50)	0,4±0,17* (66,7)	0,3±0,14* (50)

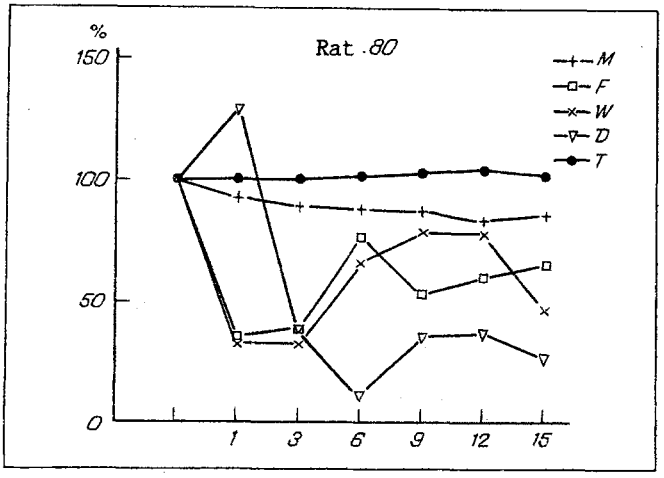

Fig. 2. Time course of changes in food-getting and associated autonomic parameters after destruction of lateral hypothalamus. M) Body weight; F) quantity of food; W) volume of water; D) 24-hourly diuresis; T) body temperature. Abscissa, days of investigation; ordinate, percentage of change relative to background (taken as 100%).

consumed remained low at this time. Parallel with these changes the number of manifestations of comfort-associated behavior and the number of episodes of spontaneous drowsiness and sleep increased in these animals, the level of wakefulness diminished, and orienting-investigative activity was suppressed (Table 3).

The results thus showed that destruction of the lateral hypothalamus in the animals is accompanied by the formation of a specific central-peripheral syndrome, including disturbances of food-getting and thinking behavior, and also of mechanisms of body weight, of a constant body temperature, and the level of diuresis, in the animals. Intraventricular microinjections of endogenous β-LPT and β-endorphin led on the whole to compensation of the functions disturbed by destruction of the lateral hypothalamus. The character of the compensatory effects of β-LPT and β-endorphin differed in many respects. The idea of "chemical" compensation of the disturbed brain functions after intracerebral intervention was put forward by Zilov [2] and Kotov [8], and involvement of oligopeptides in the mechanisms of compensation in chemical blockade of protein synthesis in the brain was examined by Sudakov [9]. Destruction of the lateral hypothalamus in the present experiments evidently led to a disturbance of synthesis of β-LPT and β-endorphin, although specific opiate receptors were present in other brain structures and in the body as a whole, their exogenous influence being adequate for realization of the basic adaptive functions [13, 15], including compensatory.

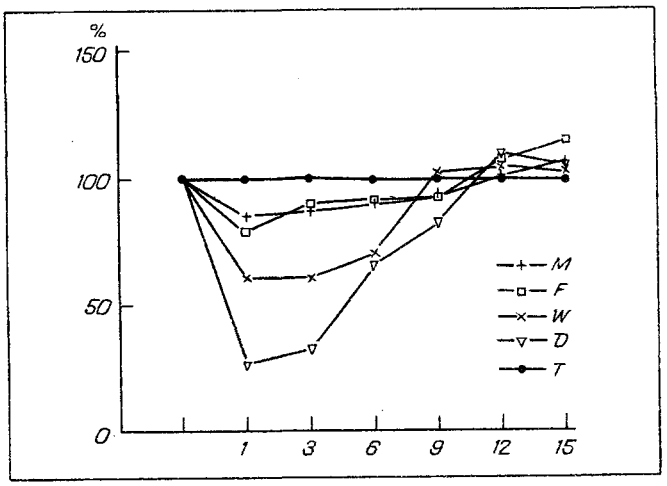

Fig. 3. Time course of change in food-getting and associated autonomic parameters in response to injection of β-LPT after destruction of lateral hypothalamus. Legend as to Fig. 2.

The results thus suggest the systemic character of involvement of regulatory peptides in the mechanisms responsible for adaptive activity of the organism as a whole.

LITERATURE CITED

1. I. P. Ashmarin, Progress in Science and Technology. Series: Physiology of Man and Animals [in Russian], Vol. 34, All-Union Institute of Scientific and Technical Information (Moscow), p. 184.
2. V. G. Zilov, Vestn. Akad. Med. Nauk SSSR, No. 2 (1982).
3. P. K. Klimov, Peptides and the Digestive System [in Russian], Leningrad (1983).
4. A. V. Kotov, V. F. Martynov, S. M. Tolpygo, and L. F. Kelesheva, Byull. Éksp. Biol. Med., **97**, No. 3, 265 (1984).
5. Yu. A. Pankov and G. P. Elizarova, Probl. Éndokrinol., No. 5, 91 (1971).
6. P. F. Rokitskii, Biological Statistics [in Russian], Minsk (1964).
7. K. V. Sudakov, Zh. Vyssh. Nerv. Deyat., No. 1, 78 (1987).
8. K. V. Sudakov and A. V. Kotov, Zh. Vyssh. Nerv. Deyat., No. 6, 1022 (1985).
9. K. V. Sudakov, Oligopeptides in Mechanisms of Biological Motivations and Their Compensation during Inhibition of Protein Synthesis [in Russian], Irkutsk (1987), p. 127.
10. S. M. Tolpygo, Yu. S. Komarov, A. V. Kotov, et al., Byull. Éksp. Biol. Med., No. 12, 643 (1981).
11. M. J. Pellegrino et al., Stereotaxic Atlas of the Rat Brain, New York (1979).
12. S. H. Snyder, Science, **209**, 976 (1980).
13. E. Olson, K. D. Olson, and A. J. Kastin, Peptides, **6**, 769 (1985).
14. E. James, J. Ladina, A. Willam, et al., Peptides (1980-1981).
15. L. F. Agnati, K. Fuxe, M. Loli, et al., Acta Physiol. Scand., **128**, 201 (1986).