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EFFECT OF ENKEPHALINS ON FUNCTION OF CALCIUM-REGULATING ENDOCRINE GLANDS IN SHOCK

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One of the distinctive features of dysfunction of the endocrine system in various critical states is disturbance of the activity of calcium-regulating endocrine glands [2, 4, 12, 15]. The writers showed previously that increased activity of the C-cells of the thyroid gland and biphasic changes in parathyroid gland function are observed in traumatic, cardiogenic, and burn shock. The hypocalcemia thus arising greatly aggravates the course of the pathological process and is an unfavorable prognostic sign in shoch [4, 5, 10, 11]. These disturbances are evidently based not only on specific factors (disturbances of the peripheral circulation, tissue hypoxia, toxemia), but also on nonspecific mechanisms (a powerful post-aggressive response to extremal stimulation). It can be postulated that measures aimed at preventing stress will lead to improvement of the activity of the parathyroid glands and of the C cells of the thyroid gland in shock. Data have been obtained to show a marked anti-stress effect of enkephalins and endorphins, which are endogenous opioid peptides [7, 8],

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TABLE 1. Concentrations of PTH, CT, and Calcium (CA) in Blood Plasma of Rats With Traumatic and Hemorrhagic Shock, Receiving and Not Receiving Enkephalin

Experimental conditions	PTH, units/ml	CT, pg/ml	Ca, mmoles/liter
Control	3,74±0,22	109,2±11,5	2,49±0,04
Traumatic shock			
3 h	4,89±0,25*	208,9±24,4*	2,68±0,05*
18 h	5,31±0,37*	188,9±19,1*	2,58±0,05
48 h	2,89±0,20*	127,1±12,4	2,19±0,04*
Traumatic shock + enkephalin			
3 h	4,23±0,31	140,1±14,1	2,51±0,06
18 h	4,75±0,18*	163,9±13,8*	2,59±0,05
48 h	3,52±0,20	124,2±14,8	2,38±0,04
Hemorrhagic shock			
3 h	4,68±0,23*	151,7±26,4	2,64±0,04*
18 h	2,92±0,18*	157,2±10,1*	2,17±0,05*
48 h	2,71±0,19*	92,9±14,2	2,04±0,04*
Hemorrhagic shock + enkephalin			
3 h	4,19±0,27	140,2±16,1	2,53±0,05
18 h	3,91±0,28	137,9±18,5	2,42±0,04
48 h	3,34±0,25	120,0±14,4	2,37±0,05

Legend. *$p<0.05$ compared with control.

and also that disturbances of the calcium balance can be corrected by injection of enkephalins [6]. This may be indirect evidence of an influence of enkephalins on function of the calcium-regulating organs.

In this investigation the effect of a stable analog of Leu-enkephalin on function of the calcium-regulating gland were studied in traumatic and hemorrhagic shock.

EXPERIMENTAL METHOD

Experiments were carried out on male rats weighing 180-220 g. Hemorrhagic shock was induced by bleeding from the right common carotid artery in a volume equivalent to 3% of the animal's body weight. Traumatic shock was induced by application of a specially designed vise to the animal's hind limbs for 6 h. Intact animals served as the control. The rats were decapitated. All experiments were conducted under superficial ether anesthesia. The stable analog of Leu-enkephalin, D-Ala^2-Leu^5-Arg^6-enkephalin (synthesized in the Laboratory of Peptide Synthesis, All-Union Cardiologic Scientific Center, Academy of Medical Sciences of the USSR; Director, Dr. Med. Sci. M. I. Titov), was injected intraperitoneally in a single dose of 500 µg/kg body weight. The concentration of parathormone (PTH) and calcitonin (CT) in the blood plasma was determined by radioimmunoassay, using kits from Fleurus (Belgium) and from Byk-Mallinckrodt (West Germany) respectively. Radioactivity was counted on a Tracor Gammaspectrometer (USA). The plasma calcium concentration was determined spectrophotometrically with kits from Bio-La-Test (Czechoslovakia). Optical density was recorded on a Specord M-40 spectrophotometer (East Germany). The numerical results were subjected to statistical analysis.

EXPERIMENTAL RESULTS

In both hemorrhagic and traumatic shock the blood PTH concentration was found to be statistically significantly increased 3 h after the beginning of the experiment (Table 1). However, after 18 h the blood PTH level in animals with blood loss showed a statistically significant decrease, whereas in animals with traumatic shock it still remained higher than in the control. After 48 h the blood PTH concentration in rats of both groups was statistically significantly lower than in the control.

The plasma CT concentration in rats with hemorrhagic shock showed no significant change throughout the period of the experiment, and not until 18 h was it statistically significantly higher than in the control. In animals with traumatic shock the blood CT concentration rose considerably after 3 and 18 h, but fell to the control values 48 h after the beginning of the experiment.

The trend of the blood calcium concentration was proportional to changes in the PTH concentration, in agreement with data obtained by the writers previously [5]. The free calcium concentration rose in rats with hemorrhagic shock after 3 h and in animals with traumatic shock after 3 and 18 h. Rats with blood loss developed hypocalcemia 18 and 48 h, and those with crushing or the soft tissues 48 h respectively after the beginning of the experiment.

Injection of the stable analog of Leu-enkephalin into rats with hemorrhagic and traumatic shock prevented the blood PTH level from rising 3 h after the beginning of the experiment. After 18 h the blood PTH level in animals with traumatic shock remained significantly higher than in the control, although it was lower than the corresponding parameter in animals not receiving the enkephalin. In rats with hemorrhagic shock the blood PTH concentration at this period did not differ significantly from the control. After 48 h the blood PTH concentration was within limits of the control values in both groups of animals.

The blood CT concentration in rats with hemorrhagic shock receiving enkephalin did not change significantly throughout the period of the experiment. In animals with traumatic shock and receiving enkephalin the blood CT concentration was statistically significantly ($P < 0.05$) lower than its level in untreated rats, although it remained a little higher than in the control. The plasma calcium concentration in animals of both groups showed no statistically significant changes throughout the period of the experiment.

As these results show, biphasic changes in the blood PTH concentration took place in animals with hemorrhagic and traumatic shock; an initial period of increase was followed by a decrease in the blood hormone concentration. The first phase of the changes, namely an increase in functional activity of the parathyroid glands in shock, is a manifestation of a response to stress [5] and is connected with the stimulating action of catecholamines on the glands [15]. The second phase, a decrease in parathyroid gland activity — arises as a result of "collapse" of the defensive mechanisms and is an extremely unfavorable factor in various critical states [5, 10]. The reason for the lowering of the blood PTH concentration is evidently exhaustion of the glands as a result of preceding hyperfunction under the conditions of hypoxia and toxemia present in shock [5]. These last two factors may perhaps play a particularly important role in the pathogenesis of the disturbances mentioned above, for in various extremal states which are not shock producing, namely immobilization, emotional stress, etc., activity of the parathyroid glands increases considerably, but they do not develop exhaustion. An increased PTH concentration under these circumstances is recorded throughout the period of action of the stressor [1, 15].

The degree of lowering of the PTH and calcium levels in the blood is proportional to the severity of the course of shock [5, 11]. This may evidently explain the more rapid onset of hypofunction of the glands in hemorrhagic shock, which was accompanied by a higher mortality than traumatic shock (68 and 45% respectively).

The increase in activity of the C-cells of the thyroid gland under extremal conditions is nonspecific in nature [1, 13, 14]. The writers have shown that elevation of the blood CT level in postaggressive states is less important in the genesis of hypocalcemia than parathyroid deficiency, but nevertheless it may aggravate the disturbances of calcium metabolism in shock [5].

Injection of the Leu-enkephalin analog into animals with traumatic shock prevented the rise of the PTH level in the initial period of the postaggressive response. This effect is evidently linked with the antistress action of enkephalins and their ability to inhibit the peripheral effects of catecholamines [9]. Injection of the stable analog of Leu-enkephalin into animals with hemorrhagic and traumatic shock also prevented the fall in the blood PTH concentration in the later stages after trauma. Evidently by preventing the initial hyperactivity of the parathyroid glands in shock, the opioid peptides themselves protected them against subsequent exhaustion.

Rapid inhibition of parathyroid gland function in hemorrhagic shock in the present experiments could be largely attributable to the presence of marked tissue hypoxia rather than to nonspecific factors. This suggests that an important role in the normalizing action of opioids in this case is played by their direct antihypoxic action [3].

Enkephalins also normalized to a greater or lesser degree the functions of the thyroid C-cells in traumatic and hemorrhagic shock. Elevation of the CT level in extremal states is basically adaptive in its role and is aimed at limiting the stressor reaction and protecting

the adrenals against exhaustion [1]. However, in various critical states of the organism, release of CT aggravates disturbances of calcium metabolism and worsens the course of the pathological process [5, 12]. Consequently limitation of CT release in shock under the influence of enkephalins can also be regarded as a favorable factor, more especially because opioid peptides also modulate the activity of the endocrine system in extremal states and prevent its exhaustion [7].

Leu-enkephalin analogs thus promote normalization of the function of the calcium-regulating endocrine glands and calcium metabolism during shock.

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