mechanisms in the striatum is one of the factors underpinning the activation, stabilization, and maintenance of the activity of the GPEE in this structure, this promoting the development of parkinsonism.

REFERENCES

1. G. I. Kryzhanovskii, *Determinant Structures in Nervous System Pathology* [in Russian], Moscow (1980).
2. G. N. Kryzhanovskii, R. F. Makul'kin, A. A. Shandra, *et al., Byull. Eksp. Biol. Med.*, **103**, № 6, 650-653 (1987).
3. D. M. Araujo, P. A. Lapchak, Y. Robitaille, et al., J. Neurochem., **50**, № 10, 1914-1923 (1988).
4. T. Arendt, V. Bigl, A. Tennstedt, and A. Arendt, *Neuroscience*, **14**, № 1, 1-14 (1985).
5. T. Bartfai, G. Fisone, and U. Langel, *Trends Pharmacol. Sci.*, 13, № 8, 312-317 (1992).
6. V. Chan-Palay, *J. Comp. Neurol.*, **273**, № 4, 543-557 (1988).
7. J. N. Crawely and G. L. Wenk, *Trends Neurosci.*, **12**, № 8, 278-282 (1989).
8. F. Javoy-Agid and Y. Agid, *An Introduction to Neurotransmission in Health and Disease*, Oxford (1990), pp. 206-220.
9. J. Lehman and B. Scatton, *Brain Res.*, **252**, № 1, 77-89 (1982).
10. M. Myslobodsky, O. Kofman, and M. Mintz, *Epilepsia*, **27**, № 5, 559-568 (1981).
11. S. O. Ogren and A. Pramanik, *Neurosci. Lett.*, **128**, № 3, 253-256 (1991).
12. C. Paxinos and G. Watson, *The Rat Brain in Stereotaxic Coordinates*, Sydney (1982).
13. L. Turski, T. Klockgether, W. A. Turski, *et al., Brain Res.*, **512**, № 1, 125-131 (1990).
14. Z. Wiesenfeld-Hallin, X. J. Xu, M. J. Villar, and T. Hokfelt, *Neurosci. Lett.*, **109**, № 1-2, 217-221 (1990).
15. W. Zieglgansberger and T. R. Tolle, in: *International Society of Pathophysiology: Constituent Congress: Proceedings*, Moscow (1991), p. 29.

The Effect of Synthetic Analogs of Enkephalins on the Development of Traumatic Cerebral Edema

V. E. Novikov and V. V. Yasnetsov

UDC 616.831-005.98-092.9-02:615.31:547.943:547.95]-07

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 116, № 8, pp. 125-127, August, 1993
Original article submitted March 18, 1993

Key Words: *cerebral edema; synthetic analogs of enkephalins*

Cerebral edema (CE) is the most grave complication after cranio-cerebral trauma. According to current views [6], pathophysiological and biochemical changes in the brain after trauma should be regarded as a response of the organism to severe stress. In view of this, timely correction of the stress reaction and preventing compensatory changes from becoming pathological are of utmost importance for prophylaxis and therapy of CE.

Special attention has been paid lately to the discussion of the role of endogenous opioid peptides as regulators of the biochemical processes and functions of the organism under extreme conditions. It is noted that opioid peptides attenuate the stress reaction and protect the organism from stress injury [1,3]. Earlier, we demonstrated the involvement of the endogenous opioidergic system in CE formation [5]. The present study was undertaken to study the effect of synthetic analogs of enkephalins on the development of traumatic CE.

Department of Pharmacology, "Gidrobios" Research Center, Russian Ministry of Public Health, Moscow. (Presented by P. V. Sergeev, Member of the Russian Academy of Medical Sciences)

MATERIALS AND METHODS

The experiments were performed on 179 nonpedigree albino rats of both sexes weighing 160-220 g. The animals were subjected to a trauma, and the development of CE was assessed from the content of total water and the density of the cerebral

0007-4888/93/0008-0907$12.50

tissue 1-4 days after brain injury, as described earlier [4].

The following analogs of enkephalins were used in the study: dalargin (Tyr-D-Ala-Gly-Phe-Leu-Arg), DAGPLAN (Tyr-D-Ala-Gly-Phe-Leu-Arg-NH-Et), DAGMPLAN (Tyr-D-Ala-Gly-MePhe-Leu-Arg-NH-Et), DAGPLG (Tyr-D-Ala-Gly-Phe-Leu-Glu), DAGO (Tyr-D-Ala-Gly-MePhe-Gly-ol), and DSLET (Tyr-DSer-Gly-Phe-Leu-Thr). All peptides were synthesized at the Cardiology Research Center, Russian Academy of Medical Sciences. The peptides were dissolved in isotonic NaCl solution and administered intraperitoneally to the animals in a dose range of 1 μg/kg - 1 mg/kg according to the scheme: the first injection was performed 15 min prior to trauma, followed by daily injections during the experiment, and the last injection was given 30 min prior to decapitation. The control animals received injections of isotonic NaCl solution according to the same scheme. In some cases naloxone (1 mg/kg), administered 15 min prior to injection of the peptides, was used as a blocker of opioid receptors. The data obtained were processed statistically using Student's t test [2].

RESULTS

Cerebral edema developed 24 hours after trauma, as was evidenced by a statistically reliable increase in the total water content in the brain and a decrease in the density of the cerebral tissue (Table 1). Four days after trauma the signs of CE were exacerbated (Table 2).

It turned out that among six peptides tested, three peptides (DAGMPLAN, DAGO, and DSLET)

TABLE 1. Effect of Synthetic Analogs of Enkephalins on the Development of Traumatic Cerebral Edema 24 h After Trauma ($M \pm m$)

Traumatic influence and peptides n=6-10	Total water, %	Density of brain tissue, g/cm^3
Intact animals	77.75±0.14	1.0412±0.0002
Trauma (control)	78.86±0.38*	1.0385±0.0003*
Trauma + dalargin		
10 μg/kg	78.43±0.22*	1.0393±0.0003*
100 μg/kg	79.37±0.15*	1.0385±0.0002*
1 mg/kg	78.20±0.35*	1.0389±0.0004*
Trauma + DAGPLAN		
1 μg/kg	78.64±0.49*	1.0391±0.0004*
10 μg/kg	79.10±0.42*	1.0388±0.0002*
100 μg/kg	78.83±0.23*	1.0391±0.0002*
1 mg/kg	79.32±0.45*	1.0387±0.0003*
Trauma + DAGMPLAN		
10 μg/kg	78.33±0.37	1.0390±0.0004*
100 μg/kg	78.01±0.25**	1.0403±0.0002**
1 mg/kg	79.17±0.43*	1.0387±0.0003*
Trauma + DAGPLG		
10 μg/kg	78.57±0.41*	1.0390±0.0004*
100 μg/kg	79.22±0.41*	1.0388±0.0004*
1 mg/kg	79.64±0.44*	1.0385±0.0002*
Trauma + DAGO		
1 μg/kg	79.51±0.33*	1.0385±0.0002*
10 μg/kg	78.65±0.14*	1.0391±0.0002*
100 μg/kg	77.73±0.27**	1.0401±0.0003**,*
1 mg/kg	77.91±0.18**	1.0400±0.0004**,*
Trauma + DSLET		
10 μg/kg	78.77±0.12*	1.0393±0.0002*
100 μg/kg	78.03±0.39	1.0402±0.0003**,*
1 mg/kg	78.15±0.24	1.0395±0.0002**,*
Trauma + naloxone		
1 mg/kg	79.09±0.19*	1.0391±0.0002*
Naloxone (1 mg/kg) + DAGO		
(100 μg/kg)	79.70±0.20	1.0388±0.0003
Naloxone (1 mg/kg) + DSLET		
(100 μg/kg)	79.11±0.28	1.0391±0.0002

Note. Here and in Table 2 one asterisk denotes significance of differences (p<0.05) in comparison with intact rats, two asterisks – in comparison with control.

TABLE 2. Effect of Synthetic Analogs of Enkephalins on the Development of Traumatic Cerebral Edema 4 Days after Trauma ($M \pm m$)

Traumatic influence and peptides n=6-10	Total water, %	Density of brain tissue, g/cm^3
Intact animals	77.75±0.14	1.0412±0.0002
Trauma (control)	79.69±0.25	1.0384±0.0002*
Trauma + DAGO (100 μg/kg)	79.03±0.29*	1.0399±0.0003**
Trauma + DSLET (100 μg/kg)	78.07±0.36**	1.0406±0.0002**
Trauma + DAGMPLAN (100 μg/kg)	79.64±0.31*	1.0384±0.0002*

exerted an antiedemic activity, while the others (dalargin, DAGPLG, and DAGPLAN) remained neutral (Table 1). For example, DAGO and DSLET in doses of 100 μg/kg and 1 mg/kg significantly reduced the total water content in the brain and increased tissue density ($p<0.05$). However, in lower doses (1 and 10 μg/kg) they proved ineffective (Table 1). In contrast to the above-indicated substances, DAGMPLAN manifested antiedemic properties only in a dose of 100 μg/kg, while in doses of 10 μg/kg or 1 mg/kg it did not produce any positive effect. We also failed to reveal an antiedemic activity of dalargin and its analogs (DAGPLG and DAGPLAN) in whole dose range tested (1 μg/kg - 1 mg/kg) (Table 1). Naloxone (1 mg/kg) did not inhibit the development of CE 24 h after trauma but completely blocked the positive effect of DAGO and DSLET (Table 1).

In the next series of experiments (Table 2) we used only those peptides that possessed antiedemic activity at early stages and studied their effect on CE development 4 days after trauma, i.e., at the late stage of the process.

As was established, DAGO (100 μg/kg) did not prevent the accumulation of water in the brain but reliably increased the tissue density. DAGMPLAN (100 μg/kg) did not affect either the content of total water or the density of the cerebral tissue 4 days after injury. Unlike these two peptides, DSLET (100 μg/kg) statistically reliably ($p<0.05$) inhibited the development of CE in this period; in other words, it displayed its antiedemic properties at the late stage of the process.

Thus, DSLET attenuates the compensatory edemic changes in the brain 24 h and 4 days after trauma, while DAGO and DAGPLAN produce a positive effect only at the early stage of CE development (one day after trauma).

It is most likely that the antiedemic effect of these peptides stems from their stimulation of the opioid receptors of the brain. This is confirmed by the finding that naloxone blocks their protective effect. As is known [7-9], DSLET predominantly stimulates delta receptors (in the brain tissue its affinity to Δ-receptors is 20-24-fold and 1000-fold higher than that to μ- and κ-receptors, respectively), while DAGO is a selective agonist of μ-receptors (in the brain its affinity to m-receptors is 166-187-fold higher than that to Δ-receptors and there is practically no κ-agonist activity). Altogether, the data obtained provide evidence that different types of opioid receptors, in particular, μ- and Δ-receptors may be involved in the pathogenesis of CE.

The authors express their gratitude to Dr. Zhanna Bespalova, head of the Laboratory of Peptide Synthesis, Cardiology Research Center, for kindly supplying the synthetic analogs of enkephalins.

REFERENCES

1. E. O. Bragin and V. V. Yasnetsov, in: *Advances in Science and Technology, Series Human and Animal Physiology* [in Russian], Vol. 47, Moscow (1991), pp. 1-223.
2. V. A. Kokunin, *Ukr. Biokhim. Zh.*, **6**, 776-791 (1975).
3. F. Z. Meerson and M. G. Pshennikova, *Adaptation to Stress Situations and Physical Loads* [in Russian], Moscow (1988).
4. V. E. Novikov, Dep. VINITI, № 2662-B90 (1990).
5. V. E. Novikov, V. V. Yasnetsov, and A. N. Sharov, *Byull. Eksp. Biol. Med.*, **114**, № 8, 163-165 (1992).
6. G. F. Rybakov, *Cranio-Cerebral Trauma* [in Russian], Vologda (1992).
7. J. S. Heyman, J. L. Vaught, R. B. Raffa, and F. Porreca, *TIPS*, **9**, 134-138 (1988).
8. H. W. Kosterlitz, *Pol. J. Pharmacol. Pharm.*, **39**, 571-576 (1987).
9. G. A. Tejwani and S. H. Hanissian, *Neuropharmacology*, **29**, 445-452 (1990).