REFERENCES

1. E. N. Barkova, E. V. Chesnokov, and A. L. Korkin, *Byull. Eksp. Biol. Med.*, **111**, № 7, 96 (1991).
2. Yu. A. Vladimirov, *Priroda*, № 3, 36 (1987).
3. V. B. Gavrilov and M. I. Meshkorudnaya, *Lab. Delo*, № 3, 33 (1983).
4. I. D. Stal'naya and T. G. Garishvili, *Modern Methods in Biochemistry* [in Russian], Moscow (1977), p. 66.
5. Yu. S. Sukhanov, *Lab. Delo*, № 6, 17 (1982).
6. G. Bjorneboe, A. Bjorneboe, B. Hagen, *et al.*, *Biochim. Biophys. Acta*, **918**, 236 (1987).
7. J. Buttriss and A. Diploch, *Biochim. Biophys. Acta*, **962**, 81 (1988).
8. A. Cederbaum, E. Dicker, E. Rubin, and G. Cohen, *Biochem. Biophys. Res. Commun.*, **78**, 1254 (1977).
9. W. Cohn, F. Loechleiter, and F. Weber, *J. Lipid Res.*, **29**, 1359 (1988).
10. J. Gutteridge, D. Rowley, and E. Griffiths, *Clin. Sci.*, **68**, № 4, 463 (1985).
11. B. Halliwell and J. Gutteridge, *TIBS*, **11**, 372 (1986).
12. R. Litov, D. Irwing, G. Downey, and A. Tappel, *Lipids*, **13**, 305 (1978).
13. S. Rall, K. Weisgraber, R. Maley, and T. Innerarity, *J. Lipid Res.*, **25**, 1277 (1984).
14. B. Martinec, *Clin. Chem.*, **10**, 1078 (1964).
15. E. Toranzo, A. Marzi, and G. Castro, *Toxicology*, **19**, 77 (1981).

Effect of Blood Serum of Patients Undergoing Intravenous Laser Therapy on the Parameters of Synaptic Transmission

D. I. Azbel', N. V. Egorushkina, I. Yu. Kuznetsova, A. S. Ratushnyak, S. M. Shergin, A. M. Shurgaya, and M. B. Shtark

UDC 615.849.19.015.4:616.153

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 116, № 8, pp. 149-151, August, 1993
Original article submitted February 23, 1993

Key Words: *blood serum; intravenous laser therapy; hippocampus; p-spike*

Currently, biological objects (systems) able to respond to an array of external factors are being widely used in order to study the combined effect of different enzymes and biologically active substances (BAS) [1]. Surviving hippocampal sections may be counted among such systems. They are highly sensitive to BAS and demonstrate comparatively simple reactions which are easily registered [3]. These properties make it possible to use this model for testing the blood serum of patients who have undergone intravenous laser therapy (IVLT). The mechanisms of the therapeutic effect of laser are still to be elucidated, but it has been established that the effects of laser on the blood are many and varied [2], their individual analysis involving very costly and very complicated procedures. Therefore, hippocampal sections used for testing the activity of such a complex system as the blood serum are one of the most appropriate models for experiment.

In the present study we investigated the effect of blood serum of patients with ischemic heart disease, exposed to low-energy laser radiation, on the parameters of synaptic transmission in neurons of the rat hippocampus.

MATERIALS AND METHODS

Fourteen patients (living in Yakutia) aged 36 to 57 years with different types of ischemic heart disease were examined. Six patients had suffered myocardial infarction in the past. The course of laser therapy comprised 6 sessions, each of 30 min

Laboratory of Clinical Diagnostics and Treatment, Institute of General Human Pathology and Ecology; Laboratory of Central Regulation and Control Mechanisms, Institute of Medical and Biological Cybernetics, Siberian Department of the Russian Academy of Medical Sciences, Novosibirsk

0007-4888/93/0008-0939$12.50

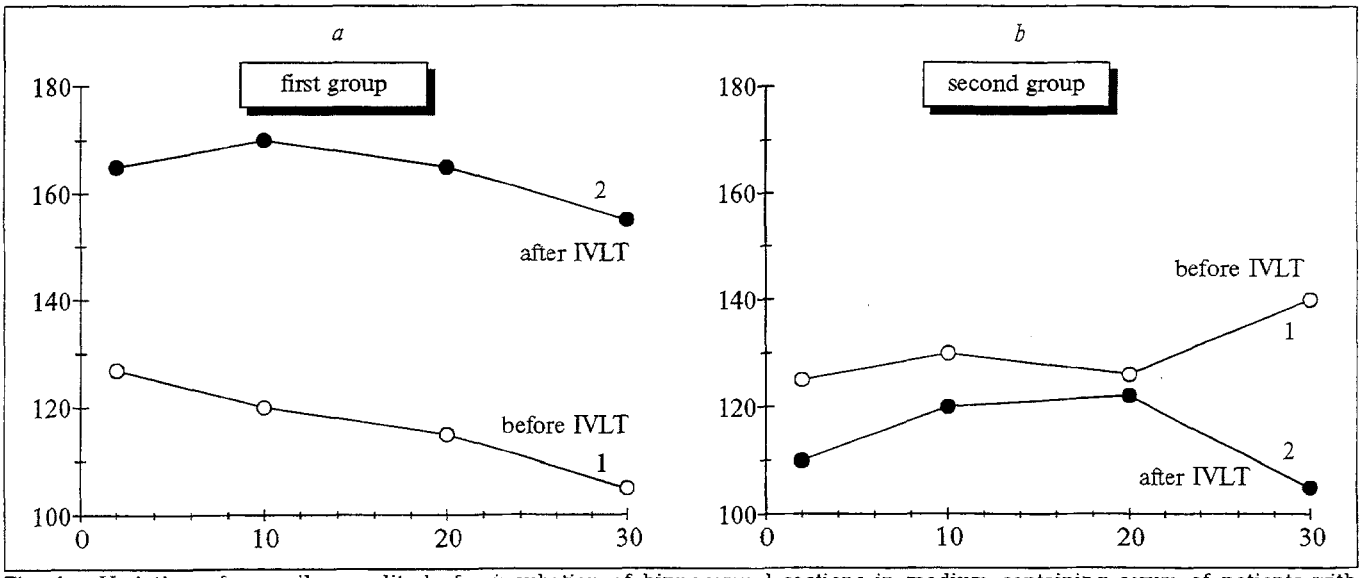

Fig. 1. Variation of p – spike amplitude for incubation of hippocampal sections in medium containing serum of patients with initially low (*a*) and initially high (*b*) neuronal activity of serum.

duration. He-Ne laser with a power of 1 mW/cm^2 was used in the treatment. The light conductor was inserted in the cubital vein. Blood for testing was taken twice: before the first session of IVLT (control) and after the last session (experiment). Wistar rats weighing 150-180 g were used in the experiments. The experiments were performed on hippocampal cross-sections of a routine preparation [4], 400-500 μ thick. After isolation, the sections were incubated under conditions of a constant flow of fresh Yamamoto medium at a constant temperature for one hour (for adaptation and to remove the harmful products of cell decay). One of the sections was then transferred to the chamber for testing, which was preliminarily filled with Yamamoto medium. Stimulating electrodes were placed on the moss fibers. Electrical stimulation was performed with rectangular pulses (100 V, frequency 0.067 Hz, pulse duration 1 msec). The electrical activity of pyramidal neurons was led off from the CA3 area of the hippocampus with the aid of extracellular glass electrodes filled with 2.5 mM NaCl (tip diameter 3-5 μ, resistance 2-5 MOhm). The amplitude and the latency of the population spike (p-spike) of the pyramidal neurons were recorded to assess the efficacy of synaptic transmission and of the electrical pulse conductance of the nerve fibers, respectively. The p-spike amplitude was determined as the sum of positive and negative deviations from the baseline. The latency was calcu-

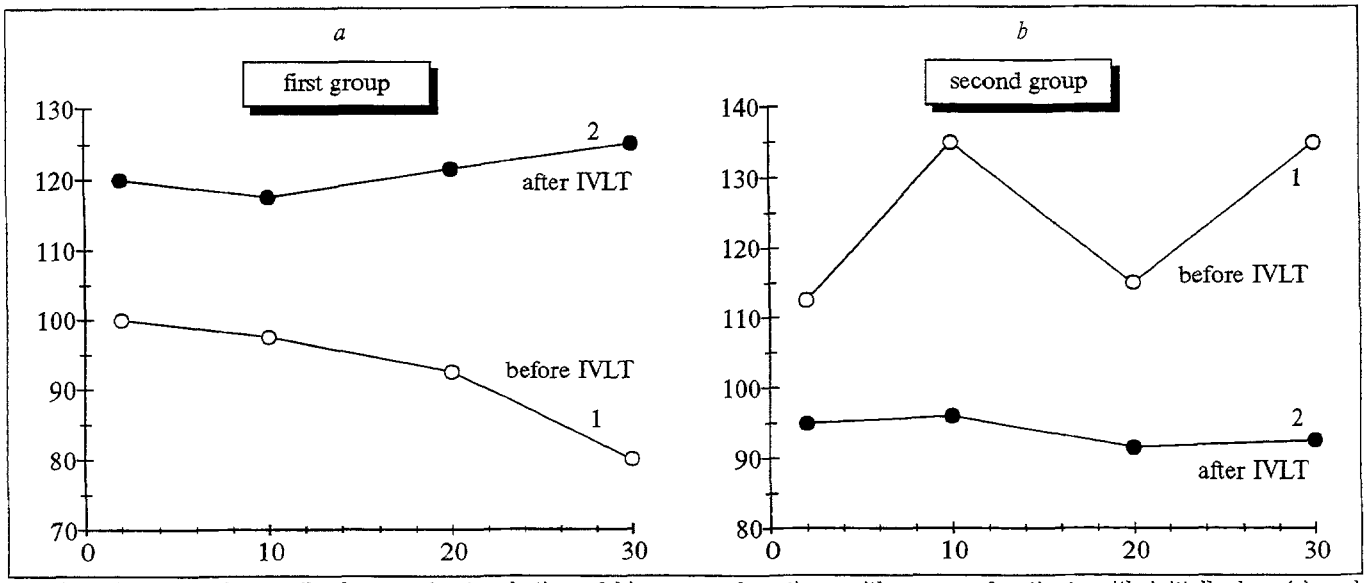

Fig. 2. Variation of p – spike latency for incubation of hippocampal sections with serum of patients with initially low (*a*) and initially high (*b*) neuronal activity of serum.

lated as the interval between the beginning of stimulation and the peak of the spike. The parameters were computer-averaged and recorded. The amplitude and the latency of the p-spike during incubation in Yamamoto medium served as the reference (baseline value) taken as 100% in each experiment. After that, for testing the sera of patients before and after laser therapy, the Yamamoto medium in the chamber was partially replaced by the serum (the ion composition and pH of the artificial incubation medium and of the serum were virtually the same). Then, 2, 10, 20, and 30 min after serum application, the amplitude and the latency of the p-spike were measured and expressed as percentage of the amplitude and latency of the cell response before serum application. Curves of the cell response amplitude vs. time and of the p-spike latency vs. time were plotted using the values of p-spike amplitude and latency expressed in arbitrary units for each fixed time interval.

RESULTS

Two groups of patients were distinguished in the analysis of the level of amplitude of the p-spike for the addition of control serum (before IVLT). The first group comprised patients whose serum, when added to the medium for incubation of hippocampal sections, virtually did not changed the amplitude of the p-spike as compared to the initial (without serum) value taken as 100% (Fig. 1, *a*, *1*). This group consisted of 6 patients. The second group comprised patients whose serum markedly increased the amplitude after a 30-min incubation with a hippocampal section. The serum derived from this group initially exhibited an increased neuronal activity (Fig. 1, *b*, *1*).

After IVLT, the serum of 5 patients (out of 6, comprising the group with a low neuronal activity of the serum) markedly increased the p-spike amplitude (Fig. 1, *a*, *2*), this value dropping below the initial level for the serum of just one patient.

The sera of 6 patients (out of 7 comprising the group with an initially increased neuronal activity of the serum) markedly reduced the p-spike amplitude (Fig. 1, *b*, *2*). One may assume that experimental serum of patients for whom the control p-spike amplitude was close to 100% raised the amplitude of the p-spike, while the serum of patients for whom the control p-spike amplitude exceeded 100% reduced this value; in other words, normalization of this parameter occurred.

The second parameter in question - latency - is less specific; it is largely a function of the variability of the method. Hence, it is more difficult to interpret the results. Nevertheless, two groups of control sera (before IVLT) were distinguished. The first group comprised those patients, the addition of whose serum markedly reduced the latency as compared to the baseline level (without serum) taken as 100%. This group was made up of 3 patients (Fig. 2, *a*, *1*). The second group comprised patients whose serum either failed to alter the velocity of pulse conductance or slowed it down as compared to the baseline level (Fig. 2, *b*, *2*).

After laser therapy, the p-spike latency markedly increased after application of the serum for all the patients of the first group (Fig. 2, *a*, *1*). At the same time, the serum of 6 patients (out of 8 comprising the second group) reduced the latency of the p-spike (Fig. 2, *b*, *2*). Thus, the p-spike latency increased when experimental serum was used for which an initially low value of this parameter was observed, and it decreased when serum was used for which an initially high latency was observed; in other words, normalization of the latency of p-spike was noted.

However, an unequivocal interpretation of the results obtained is difficult, because the sample of patients examined is small and nonuniform. In addition to the main disease, almost all the patients suffered from different associated disorders. Against the background of laser therapy, all the patients were receiving drugs which might (directly as well as indirectly) have affected the parameters being recorded. However, the above-noted tendency toward normalization of the parameters characterizing synaptic transmission and conductance of the nerve fibers in the hippocampal sections, which changed under the influence of the serum of patients before and after IVLT, allows to assume that these parameters make it possible to record objectively the state of the patient during the course of treatment.

REFERENCES

1. S. P. Bidei, in: *Immobilized Cells and Enzymes. Use of Immobilized Mammalian Cells for Quantitative and Qualitative Assay of Hormones* [in Russian], Moscow (1988), p. 177.
2. V. B. Matyushichev and V. V. Titov, in: *Mechanisms of the Biostimulating Effect of Low-Intensity Laser Radiation* [in Russian], Vest. Leningrad Gos. Univ., Ser. Biologiya (1986), pp. 1-30.
3. N. A. Otmakhov and A. G. Bragin, *Brain Res.*, **506**, № 2, 173-183 (1990).
4. C. Yamamoto, K. Matsumoto and M. Takagi, *Exp. Brain Res.*, **38**, 469-472 (1980).