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Changes in Blood Serum Neuroreactivity in Coronary Parients

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Key Words: neuroreactivity; blood serum; membrane potential

Isolated mollusk neurons have been used as a test object to assess blood serum neuroreactivity. Such neurons are devoid of a glial sheath, this making their membrane readily accessible to serum substances. Coronary patients' blood sera have been tested over a course of intravenous laser therapy. Such therapy is widely practiced in the treatment of cardiology patients, but the mechanisms of the therapeutic effect of light are not clear and there are no definite criteria for assessment of the therapeutic effect [2,3].

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MATERIALS AND METHODS

Blood was collected before therapy and after a course of intravenous laser therapy (IVLT) consisting of six sessions [4]. Blood sera were frozen and kept before testing at -80°C. They were defrosted directly before the experiments. Sera of 13 patients were tested. Neurons of Lumnaea stagnalis aged 0.5 to 2 years were used. After 40 min incubation of nervous tissue in 0.3% enzyme solution (pronase, Serva, Germany) the neurons (without glial cells) were removed from the peripharyngeal ganglia and placed on the glass bottom plate of the chamber. Physiologycal saline of the following composition was used: 55 mM NaCl, 1.6 mM KCl, 4 mM CaCl₂, 1.5 mM MgCl₂, 5 mM Tris, pH 7.8. The

serum ionic composition was matched to the used saline by mixing the sera with a compensating solution in a 1:2.6 ratio, the compensating solution consisting of 3.5 mM CaCl, and 1.24 mM MgCl₂. The volume of the chamber in which the neurons were placed was 100 µl. Sera were added to the chamber in a dose of 20 µl. One patient's serum was tested in one chamber. Electrophysiological experiments were carried out 12-18 h after neuron isolation. Intracellular electrodes were used to record biopotentials. The membrane potential (MP) and spontaneous and evoked responses were recorded and processed with computers and KAMAK equipment. Membrane potentials were defined as the difference between the electrode potential values outside and inside the cell.

RESULTS

A change of the MP was a common tendency in the neuronal reaction to application of the test sera. The degree and time course of these changes were individual.

The least pronounced reversible changes in neuronal MP were caused by the sera of three patients. After a drop in the MP which developed within the first 0.5-1.5 min, normalization occurred within 2 to 5 min.

Some patients' sera caused hyperpolarization to develop during the first 1-5 min, followed in all cases by depolarization. Neuronal hyperpolarization indicates an increased intracellular concentration of potassium ions.

A membrane potential drop, or cell depolarization, caused by interaction of the membrane with serum components, indicates the presence in the serum of substances disrupting the activity of the ion-transporting systems and impairing the membrane. During the first 20 to 30 min the damaging effect of the sera manifests itself in cell depolarization. Replacement of the solution containing the serum with physiological saline did not lead to recovery of the initial value of the MP. The damaging effect was assessed as a percent ratio of the maximal MP depolarization to the value of theis parameter in solution before serum application.

Comparison of the damage caused by control (before IVLT) and experimental sera permitted the patients to be divided into three groups (Fig. 1).

Group 1 consisted of patients whose sera after experimental IVLT (E) depolarized neurons to a lesser degree than the sera before therapeutic exposure (C). The damaging effect of sera on the membrane was noticeably reduced (by 43±10.3%) after

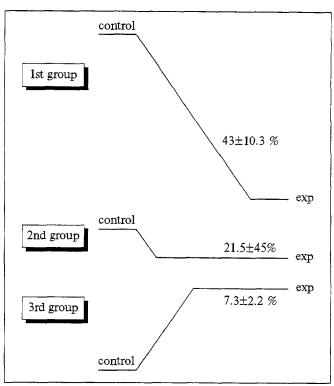


Fig. 1. Changes in effects of control (before laser exposure, Control) and experimental (after six sessions of laser exposure, Exp) sera on membrane potential of isolated nerve cells in three groups of patients (percent neuroreactivity changes).

laser therapy. This group consisted of 6(46.15%) patients.

Group 2 were patients whose sera after laser exposure had a less pronounced injurious effect on the membrane (4 patients, 30.7%), but the differences between the experimental and control samples were less marked $(7.3\pm2.2\%)$.

Group 3 consisted of patients whose sera after laser exposure had a more injurious effect (by $21.5\pm4.5\%$).

In health a nerve cell maintains on its surface membrane a potential difference from -45 to -70 mV. Tension is created because of an irregular distribution of ions (mainly potassium) on both sides of the cell membrane [1].

The marked drop of the MP gives suggests that the sera caused disorders in the ion-transport cellular systems responsible for potassium and sodium (and other) ion distribution against their concentration gradient.

The functioning of these systems is maintained by a metabolic process, namely the ATP-dependent mechanism of sodium and potassium ion transport or by the Na-K pump which maintains low sodium and high potassium concentrations inside the cell, actively transporting sodium from and potassium into the cell. The drop of the MP (in some cases all the way to zero) confirms the hypothesis

about serum-induced disorders in normal ion ditribution. Another ion-transporting system of the cell, consisting of chemo- and potential-regulated channels, may be disturbed as well. Ions cross the membrane via these ion-selective channels along their concentration gradient. The state of ion channels is known to be regulated by many factors, among which are some substances acting on the external surface of the membrane.

The above facts suggest that the de- and hyperpolarization changes detected in the first few minutes may reflect the effects of substances in the serum on regulatory subunits of the ion channel. Intravenous laser therapy in the majority of cases reduces the activity of these factors present in patients' sera which probably disturb the normal functioning of the ion-transporting systems of the neuronal membrane.

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The Costimulating Effect of Chorionic Gonadotropin on Lymphokine-Activated Splenocytes. New Aspects of the Immunomodulating Effect of the Pregnancy Hormone

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Chorionic gonadotropin (CG) is known to possess a well expressed immunomodulating activity. As a rule, the effects of the hormone are of a dose-dependent nature and can change direction depending on the differentiation level of immunocytes and the presence of female steroid hormones [1,3,4]. Since CG is a pregnancy hormone, investigation of its immunomodulating effects is of great importance for understanding the mechanisms of immunological tolerance formation in the maternal organism vis-a-vis the semiallogenic fetus. The CG secretion by neoplastic cells [10] and the hor-

Department of Biochemistry, Perm Medical Institute (Presented by A. D. Ado, Member of the Russian Academy of Medical Sciences) mone's ability to intensify carcinogenesis and metastasis [6] give grounds for regarding this hormone as a factor promoting development of the neoplastic process, which always goes along with immune mechanism disorders.

The object of the present study was to examine the effect of CG on the processes of antigen-independent differentiation of immunocompetent cells under the influence of differentiative signals varying in level and quality.

MATERIALS AND METHODS

The experiments were performed on female CBA mice weighing 20-22 g. Chorionic gonadotropin