

## 1. Muscle Physiology

**CD<sup>2+</sup> AND NI<sup>2+</sup>-INDUCED CONDUCTIVITY CHANGES OF FAST AND SLOW INACTIVATING Ca CHANNELS IN THE MUSCLE MEMBRANE.** M. Henček, D. Zacharová, Institute of Molecular Physiology and Genetics, Slovak Academy of Sciences, Bratislava.

The aim of the present work was to compare the effects of the Ca<sup>2+</sup> antagonists, Ni<sup>2+</sup> and Cd<sup>2+</sup>, on fast and slow inactivating Ca channels present in the crayfish muscle membrane (4). Time courses of the conductivity of both Ca channels could be studied separately using extended software based on the Hodgkin-Huxley equation for the estimation of conductivity (2,3). Ca ionic channels were recorded from internally perfused muscle segments under conditions of the vaseline-gap voltage clamp. Similarly as in other structures (1), Cd<sup>2+</sup> depressed the conductivity of the slow Ca channel more than that of the fast one, whereas Ni<sup>2+</sup> had a stronger effect on the fast Ca channel. Cd ions proved more effective than Ni ions in inhibiting both Ca channel types.

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**MECHANISMS OF INTRACELLULAR pH REGULATION IN VASCULAR SMOOTH MUSCLE.** H. Nguyen Duong, Department of General Physiology, University of Ulm, Ulm, Germany.

Intracellular pH (pH<sub>i</sub>), an important determinant of several cell functions, e.g. metabolism, contractility and ion channel conductivity, underlies strict and precise regulation. The aim of this study was to characterize the mechanisms responsible for this control in vascular smooth muscles, which basically occur on a short-term basis by intracellular buffering and on a long-term basis by ionic exchange across the cell membrane. One method of studying these mechanisms is to perturb pH<sub>i</sub> by exposure to weak acids or bases at constant external pH (pH<sub>o</sub>) and to analyze the resulting contractility changes. In isolated porcine coronary arteries (PCA) the pattern of contractile responses to alkaline or acid loading coincides exactly with expected pH<sub>i</sub> changes as they have been measured in many other cells and tissues, an induced acidification correlating with contraction and alkalization with relaxation. This indicates clearly that pCa<sub>i</sub> and pH<sub>i</sub> in PCA vary directly with one other. The results obtained are in agreement with the assumption of a dual regulation system involving both the activation of a Na<sup>+</sup>/H<sup>+</sup>-exchanger for the correction of a shift to acidic pH-values and the activation of an Cl<sup>-</sup>/HCO<sub>3</sub><sup>-</sup>-exchanger for the correction of a shift to alkaline values.

**VOLTAGE DEPENDENCE OF SLOW INWARD CURRENT RELATED TO CONTRACTION (I<sub>slc</sub>) IN DOG VENTRICULAR MUSCLE.** J. Šimurda, M. Šimurdová, P. Bravený, J. Šumbera, Faculty of Medicine, Masaryk University, Brno.

The voltage dependence of the total slow inward current (I<sub>sl</sub>) and isometric contraction were measured in thin ventricular trabeculae at a low and a high level of contractility achieved by two standard conditioning voltage-clamp protocols. The aim was to separate the contraction related component I<sub>slc</sub> from the calcium current I<sub>Ca</sub>. The experiments were performed at 30 °C in the presence of TTX (20 μM). The analyses of responses to 300 ms depolarizing pulses showed that I<sub>slc</sub> and contraction had the same threshold voltage which was lower than that of I<sub>Ca</sub>. At increased contractility, the net I<sub>slc</sub> could be recorded in a narrow voltage range between both thresholds. In another series of experiments I<sub>slc</sub> was measured as a tail current following short (10 ms) standard depolarizing pulses at voltages negative to threshold of I<sub>Ca</sub>. In the voltage range between -90 and -35 mV the experimental current-voltage relations of I<sub>slc</sub> match fairly well with the theoretical curves derived from a quantitative model based on the hypothesis that I<sub>slc</sub> is identical with the current of the electrogenic Na-Ca exchange process.

**THE CHANGES IN CYTOSOLIC FREE CALCIUM CONCENTRATION IN ISOLATED NERVE TERMINALS UNDER DIFFERENT DEPOLARIZING CONDITIONS.** *R. Bottlíková, J. Orlický,* Institute of Molecular Physiology and Genetics, Slovak Academy of Sciences, Bratislava.

By using an intracellular fluorescence probe Fura-2AM the changes of the cytosolic free calcium concentration in synaptosomes were monitored in order to search for the possible role of  $\text{Na}^+$ ,  $\text{Ca}^{2+}$ ,  $\text{K}^+$ -channels and  $\text{Na}/\text{Ca}$  exchanges in the regulation of intrasynaptosomal calcium.  $\text{K}^+$ -depolarization ( $10\text{-}60\text{ mmol.l}^{-1}$ ) increased the intrasynaptosomal free  $[\text{Ca}^{2+}]_i$  by  $100\text{-}200\text{ nmol}$  through the  $\text{Ca}^{2+}$ -channel and/or  $\text{Na}/\text{Ca}$  exchanger in  $[\text{Na}^+]_i$  rich synaptosomes. Veratridine ( $4\text{-}45\text{ }\mu\text{mol.l}^{-1}$ ),  $\text{Na}^+$ -channel opener, caused an increase in  $[\text{Ca}^{2+}]_i$  by  $60\text{-}300\text{ nmol}$  in a dose-dependent manner on the extrasynaptosomal  $[\text{Na}^+]_o$  concentration.  $\text{K}^+$ -channel inhibitor, 4-aminopyridine ( $0.01\text{-}10\text{ mmol.l}^{-1}$ ), elevated  $[\text{Ca}^{2+}]_i$  by  $15\text{-}120\text{ nmol}$ . Subsequent  $\text{K}^+$ -depolarization can induce  $\text{Ca}^{2+}$ -channels to fire synchronously. These results suggest that 4-aminopyridine may mimic more closely the physiological depolarization by repetitive stimulation of the nerve terminal than elevated KCl.

**INFLUENCE OF HIGH EXTRACELLULAR LEVELS OF CALCIUM ON THE ENERGY METABOLISM OF MUSCLE TISSUE.** *A. Breier<sup>1</sup>, Z. Sulová<sup>2</sup>, T. Stankovičová<sup>3</sup>, D. Hagarová<sup>1</sup>,* <sup>1</sup>Institute of Molecular Physiology and Genetics, <sup>2</sup>Institute of Chemistry and <sup>3</sup>Institute for Hear Research, Slovak Academy of Sciences, Bratislava.

Tissue of the mouse diaphragm was incubated in Liley's solution with a content of 2, 4, 6 and 8  $\text{mmol.l}^{-1}$  calcium. The energy metabolism of muscle tissue was studied on the basis of estimation of tissue respiration parameters, glucose incorporation as well as the tissue content of ATP, lactate and pyruvate. It can be concluded from the results that 1) at the initial time, the high level of calcium accelerated transport of glucose through sarcolemma but after 20 min of incubation the transport rate was considerably inhibited, 2) extracellular calcium level exciting 6  $\text{mmol.l}^{-1}$  also had an inhibitory effect on the respiration of muscle tissue, 3) 30 min of incubation of the tissue in an increased concentration of calcium in the bath lead to a decreased content of ATP and pyruvate and an increased content of lactate.

**EXPRESSION OF BETA-CARDIAC AND SLOW TWITCH MYOSIN HEAVY CHAINS IN RAT INTRAFUSAL FIBRES.** *T. Soukup, F. Pedrosa<sup>1</sup>, L.E. Thornell<sup>1</sup>,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague and <sup>1</sup>Department of Anatomy, University of Umea, Sweden.

Intrafusal fibres express different myosin heavy chains (MHC) often with regional variation along the length of the fibres. We have compared the staining of monoclonal antibodies (mAb) against beta-cardiac MHC (Sera-Lab, England) and mAb 9812 against slow-twitch MHC (gift of Dr. G.K. Dhoot, London), on fresh cryosections of the extensor digitorum longus muscle from 3-week-old rats.

Both mAbs stained extrafusal type 1 fibres; in the spindles, bag<sub>1</sub> fibres were stained in their polar zones and bag<sub>2</sub> fibres along their entire length except for a short region around the equatorial zone. Unexpectedly, mAb 9812 stained more of the juxtaequatorial zone of the bag<sub>2</sub> fibre than mAb against beta-cardiac MHC. However, beta-cardiac MHC and skeletal slow twitch MHC are thought to be encoded by the same gene. Thus the staining pattern with both mAbs was almost identical and the small difference in staining of bag<sub>2</sub> fibres might be explained by quantitative difference in the number of available epitopes.

**INCREASED ACTIVITY OF SARCOLEMAL ( $\text{Na}^+ - \text{K}^+$ )-ATPASE AFTER *IN VIVO* ADMINISTRATION OF 7-OXO-PGI<sub>2</sub>.** A. Džurba, A. Ziegelhöffer, T. Ravingerová, N. Tribulová, Institute of Heart Research, Slovak Academy of Sciences, Bratislava.

The influence of a stable prostacycline derivative, 7-oxo-PGI<sub>2</sub>, on the specific activity of ( $\text{Na}^+ - \text{K}^+$ )-ATPase was investigated in isolated rat heart sarcolemma. When administered in a single dose of 50  $\mu\text{g}\cdot\text{kg}^{-1}$  i.m. 24 or 48 hours prior to excision of the heart, the drug induced a significant ( $p < 0.001$ ) increase in specific activity of ( $\text{Na}^+ - \text{K}^+$ )-ATPase. Kinetic analysis of activation by ATP of the particulate enzyme revealed decreased  $K_m$  values ( $p < 0.01$ ). Both effects of PGI<sub>2</sub> on ( $\text{Na}^+ - \text{K}^+$ )-ATPase could be antagonized considerably by simultaneous administration of an arbitrarily chosen dose (1  $\text{mg}\cdot\text{kg}^{-1}$  i.m.) of a protein synthesis inhibitor, cycloheximide ( $p < 0.05$ ). The results obtained may be explained in two ways: a) by synthesis of a mediator which affects the ( $\text{Na}^+ - \text{K}^+$ )-ATPase by increasing its turnover number or b) by *de novo* synthesis of ( $\text{Na}^+ - \text{K}^+$ )-ATPase differing from the original one in increased affinity for the substrate.

**THE EFFECT OF 7-OXO-PGI<sub>2</sub> ON THE MUSCLE MEMBRANE ( $\text{Na}^+ - \text{K}^+$ ) PUMP.** T. Stankovičová, H. Zemková<sup>1</sup>, A. Ziegelhöffer, F. Vyskočil, Institute for Heart Research, Slovak Academy of Sciences, Bratislava and <sup>1</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The effect of 7-oxoprostacycline (PGI) on membrane sodium pump *in situ* was analysed after preliminary findings indicating that sarcolemmal ( $\text{Na}^+ - \text{K}^+$ )-ATPase is modulated by PGI.

The diaphragms of female mice pretreated for 30–34 h with 50  $\mu\text{g}/\text{kg}$  PGI i.p. were used for measuring the resting membrane potential (RMP) with microelectrodes. RMP of muscles from pretreated mice was significantly higher ( $-83.7 \pm 5.7$  mV, mean  $\pm$  S.D.) than in control animals ( $-70.8 \pm 7.7$  mV) and this hyperpolarization was abolished by ouabain.

In acute experiments where  $10^{-7}$   $\text{mol}\cdot\text{L}^{-1}$  PGI (i.e. in a concentration comparable to systemic administration) was applied into the bath with Na-loaded muscles, the increase of RMP due to the electrogenic pump was about 30 mV and reached about -100 mV. This increase was abolished by addition of ouabain as in previous experiments.

The data indicate that PGI has a facilitating effect on this part of RMP which is produced by the electrogenic ( $\text{Na}^+ - \text{K}^+$ ) pump.

**PREVENTIVE EFFECT OF 7-OXO PROSTACYCLIN ON ISCHAEMIC AND REPERFUSION INJURY OF THE RAT HEART.** T. Ravingerová, A. Džurba, A. Ziegelhöffer, V. Trégerová, Institute for Heart Research, Slovak Academy of Sciences, Bratislava.

Late (48 h after i.m. administration) protective antiischaemic action of 7-oxo prostacyclin (7-oxo PGI<sub>2</sub>) was studied on the isolated, perfused rat heart (Langendorff preparation) during 30 min global ischaemia followed by 30 min reperfusion. Functional parameters of the heart (heart rate, contractility and coronary flow), as well as those of energy metabolism (ATP, ADP, AMP, ADN, LAC, PYR) were investigated. A protective effect of 7-oxo PGI<sub>2</sub> was manifested by a better maintenance of heart function during reperfusion, by a better preservation of myocardial ATP content in the course of ischaemia, as well as by a normalization of the enhanced lactate accumulation, caused by ischaemia. The protective antiischaemic effect of 7-oxo PGI<sub>2</sub> is supposed to be related to a stimulation of the sarcolemmal ( $\text{Na}^+ - \text{K}^+$ )-ATPase specific activity with a consequent stabilization of cell membranes, thus preventing the alteration of sodium and calcium homeostasis.

**THE PROTECTIVE EFFECT OF 7-OXO-PROSTACYCLIN ON RAT HEART IN POSTISCHAEMIC AND  $Ca^{2+}$  PARADOX CONDITIONS. A MORPHOLOGICAL AND BIOCHEMICAL STUDY.** *N. Tribulová, J. Slezák, T. Ravingerová, L. Okruhlicová, A. Ziegelhöffer, A. Džurba.* Institute for Heart Research, Slovak Academy of Sciences, Bratislava.

On the model of isolated perfused rat heart, both postischaemic reperfusion and  $Ca^{2+}$  paradox injury resulted in structural and metabolic derangement of myocardial cells. This was manifested histochemically by depressed activity of glycogen phosphorylase and oxidoreductases in the ischaemic heart. The activity of these enzymes as well as that of ATP-ases and alkaline phosphatase was more depressed in the heart injured during the  $Ca^{2+}$  paradox. These changes were accompanied by irreversible fine structural alterations of myocytes.

The stable  $PGI_2$  analogue, 7-oxo-prostacyclin (50  $\mu g/kg$  i.m.) 48 hours after administration exerted a cardioprotective effect. It was manifested by better preservation of the ultrastructure of myocytes.

We assume that the prostacyclin-induced protective effect may be explained by influencing the processes at the cardiac membrane level resulting in a decreased  $Ca^{2+}$  overload.

**SEXUAL DIFFERENCES IN PROLIFERATION OF RAT VASCULAR SMOOTH MUSCLE CELLS (SMC) *IN VITRO*.** *L. Bačáková, M. Baudyšová, V. Mareš.* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The SMC proliferation plays a role in the development of vascular diseases. In an earlier study we showed sexual differences in growth of rat aortic SMC in culture. The male SMC had a shorter doubling time (41 vs. 46 h in females) and reached a higher maximum population density (170 000 vs. 80 000 cells/cm<sup>2</sup>). Here we report the parameters of cell kinetics obtained in cultured aortic SMC derived from young male and female rats (150 g, SPF). The cells were cultured in a modified Eagle's minimum essential medium with 10 % of foetal calf serum and labelled with <sup>3</sup>H thymidine (1  $\mu Ci/ml$ ). We found that the male SMC have a higher labelling index (27 vs 19 %), a higher mitotic index (6 vs. 3 %), a shorter cell cycle (19 vs. 27 h) and a higher growth fraction (82 vs. 57 %). In the 2nd and 3rd passages, the male SMC contain more hyperploid SMC (6 %) than the female SMC (2 %). Similar differences, although smaller, were also found in conventionally raised rats. We assume that the sexual differences in the proliferation of vascular SMC are due more to genetic factors than to epigenetics.

## 2. Physiology of the Blood

**BONE MARROW REACTION TO DAMAGE CAUSED BY ACUTE SUBLETHAL IRRADIATION.** *E. Nečas, V. Znojil.* Institute of Pathological Physiology, First Faculty of Medicine, Charles University, Prague.

The bone marrow of experimental mice was damaged by acute total body irradiation with doses of 1.5 Gy or 2.6 Gy. Changes in total cellularity of the femoral bone marrow, in the incidence of "haematopoietic stem cells" of the CFU-S and GM-CFC types, as well as in the proliferation rate of "CFU-S stem cells", were examined. The total bone marrow cellularity recovered several days before the recovery in the number of either type of "stem cells". Of these, the more mature GM-CFC recovered more rapidly than the less mature CFU-S. The recovery pattern differed when different irradiation doses were used. Its analysis favours the hypothesis that the cell production rate is primarily regulated by the stem cell populations rather than by a constant stem cell number. The results confirm that normal bone marrow cellularity can be obtained even when the incidence of stem cells is reduced to about 10 % of normal.

REACTION OF MURINE HAEMOPOIETIC TISSUE TO THE DAMAGE CAUSED BY CYCLOPHOSHAMIDE. *L. Šefc, E. Nečas*, Institute of Pathological Physiology, First Faculty of Medicine, Charles University, Prague.

Cyclophosphamide (135 mg/kg b.w.) caused a prolonged perturbation in the regulation of haemopoietic stem cells. Both CFU-S and CFU-GM reached a transient peak after an initial decrease in the bone marrow. Thereafter, a secondary decline followed with nadir day 7 after CY. At this time, the CFU-S ceased to divide (6 % CFU-S in S-phase vs 30 % in control mice).

The kinetics of stem cell recovery in the spleen was quite different from that in the bone marrow. The initial damage was much more pronounced in the spleen. At the time when only a few stem cells were present in the bone marrow, there was a ten-fold increase in their number in the spleen as compared with normal values. The contribution of changes in the microenvironment and/or stem cell migration is discussed.

ONTOGENETIC CHANGES IN HAEMOPOIETIC STEM CELL COMPARTMENTS OF RATS. *A. Bartoníčková, A. Vacek*, Institute of Biophysics, Czechoslovak Academy of Sciences, Brno.

The haemopoietic activity of blood forming tissues exhibited, during the life of rats, a decrease in the pool of compartments of polypotential haemopoietic stem cells (CFUs) in the spleen and its elevation in the bone marrow.

Methods of *in vitro* cultivation of fibroblast progenitors (CFUf) of haemopoietic tissues, constituting a substantial part of the haemopoietic inductive microenvironment (HIM) were elaborated and changes of CFUs and CFUf in the bone marrow and spleen of rats of different ages were compared.

At the age between 3rd and 12th months of life the concentration of CFUf in the bone marrow of rats increased 2.2 times, while in the spleen there was a six-fold decrease. The decrease was accompanied by a drop of the proliferative activity of both tissues, after sublethal gamma-ray irradiation of 12 week-old rats, an elevation of the CFUs and CFUf pools was observed as well as a restoration of the ability of the spleen to support macroscopic spleen colonies after bone marrow transplantation. The results document the ontogenetic changes in the compartment of CFUf, i.e. HIM, and its role in the ontogenetic changes in the CFUs compartment of the spleen is discussed.

CHANGES IN THE PHAGOCYtic ACTIVITY OF CELLS AFTER DEXTRAN SULPHATE ADMINISTRATION. *J. Kautská, A. Vacek, A. Lojek*, Institute of Biophysics, Czechoslovak Academy of Sciences, Brno.

Using the method of luminol enhanced chemiluminescence on an LKB Wallace 1251 apparatus, the phagocytic activity of splenic and femoral bone marrow cells was studied in the resting state and after activation with starch. The measurement was carried out after a short-term influence of i.p. dextran sulphate (DS) administration (0.5, one two or three hours) and a long-term (one, three or five days) action of dextran sulphate. Significant stimulation of the phagocytic activity of femoral bone marrow cells against the control group was found within one hour after dextran sulphate administration, while the metabolic activity of spleen cells was significantly depressed at this period. A significant decrease of the phagocytic activity of splenic cells and femoral bone marrow cells against control groups was also found on days 1 and 5 after dextran sulphate administration.

**THE EFFECT OF ANTIBIOTICS ON THE CHEMILUMINESCENCE OF HUMAN POLYMORPHONUCLEAR LEUKOCYTES.** *D. Kubičková, L. Beneš, A. Lojek, D. Jandová*, Institute of Biophysics, Czechoslovak Academy of Sciences, Brno.

Modern therapy requires the use of antibiotics without suppressive effects on the immune system, and this need is most apparent in patients treated with immunosuppressive agents, or those suffering from various other immunodeficiencies. The metabolic activity of human phagocytic cells was studied after their exposure to several antibiotics used as an antibiotic shield during heart operations (oxacillin, cefalothin and cefotaxime). The functional activity of phagocytes was investigated using luminol- and lucigenin-dependent chemiluminescence (CL). The particles to be phagocytosed were those of rice starch and opsonized *E. coli*. Cephalosporins (cefalothin and cefotaxime) in therapeutic concentrations did not affect the CL reaction of phagocytes significantly, in some cases the antibiotics exhibited a mild stimulatory effect. Oxacillin in therapeutic concentrations had a stimulatory influence on the CL response of phagocytes.

**CHEMILUMINESCENCE ANALYSIS OF THE METABOLIC ACTIVITY OF HUMAN PHAGOCYTES AND ITS USE FOR TESTING THE ACTIVITY OF ANTIOXIDANTS.** *A. Lojek, D. Jandová, D. Kubičková*, Institute of Biophysics, Czechoslovak Academy of Sciences, Brno.

Phagocytes in the whole blood of healthy donors and isolated polymorphonuclear leukocytes were studied for their metabolic activity. Phagocytic cells were activated by opsonized and non-opsonized microbes and particles (starch, zymosan, *E. coli*). The kinetics of the phagocytic reaction was monitored using a method of chemiluminescence - lucigenin was used for detection of the superoxide anion, and luminol for that of hydrogen peroxide.)

Starch and opsonized particles were found to be the best activators of oxygen radical production by phagocytes *in vitro*. Other experiments revealed a significant decrease in lucigenin and luminol oxidation on the addition of ascorbic acid (down to 20 µg/ml), thus proving its antioxidant, but at the same time protective effects on cells and tissues, which might otherwise become damaged when exposed to these two types of reactive oxygen metabolites.

**INFLUENCE OF CALCIUM CHANNEL BLOCKERS TO VINCRISTINE-RESISTANT MOUSE LEUKEMIA CELL LINE L1210/VCR.** *L. Poleková<sup>1</sup>, M. Barančík<sup>2</sup>, A. Breier<sup>1</sup>*, <sup>1</sup>Institute of Molecular Physiology and Genetics and <sup>2</sup>Institute for Heart Research, Slovak Academy of Sciences, Bratislava.

The vincristine-resistant mouse tumor cell line L1210/VCR was established in our laboratory from the mouse leukemia cell line L1210 (a gift from the Institute of Oncology in Bratislava). These resistant mutant cells were prepared by stepwise selection during subculturing in increasing concentrations of vincristine.

Calcium channel blockers verapamil, galopamil, nifedipine were tested for their ability to overcome multidrug resistance. The L1210/VCR cells were used in this study were 30 to 100 times more resistant to vincristine. We studied the ability of calcium channel blockers to reverse the multidrug resistance in doses 25<sup>-1</sup> mmol.l<sup>-1</sup>.

### 3. Methods

OPTICALLY-COUPLED ANALOG AMPLIFIER. *J. Mihálik*, Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

An amplifier has been constructed for the needs of an electrophysiological laboratory. It consists of two parts characterized by isolation of the input reference potential from the common output potential. The input analog signal enters into the differential input of the amplifier stage with a high input impedance and unit gain. An opto-coupler serves for signal transmission to the input of the common amplifier stage with a gain 100 or 500. Transmitted signal stability is achieved by using a current source and a pair of opto-couple units. They represent the resistance and capacitance barrier between both amplifier stages. It is also possible to set the reference level to zero and it can also filter the DC signal. It can be used for amplitudes up to 100 mV p-p in the frequency range from 0 to 10 Hz.

A COMPUTER GRAPHIC METHOD OF PLOTTING THE SPATIAL DENSITY OF PARTICLES. *J. Křekule<sup>1</sup>, M. Martone<sup>2</sup>, S.J. Young<sup>2</sup>, P.M. Groves<sup>2</sup>*, <sup>1</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague and <sup>2</sup>Department of Psychiatry, Medical Center, UCSD, La Jolla, USA.

Visualization of the density of particles (e.g. neurones) within a biological tissue helps in the study of their spatial allocation or even relations to other particles (e.g. patches of a neurotransmitter). Instead of the usual evaluation of the density in every point of a given space, the density contribution of each particle is evaluated according to the method proposed. The density is modelled by gray values stored in the video RAM of the display. All contributions are added to obtain the overall density map. By applying basic image processing routines, i.e. segmentation and edge detection, a set of equidensity lines is plotted. The plotting is simplified by overflowing arithmetic when summing individual density contributions. The method is illustrated by mapping neuronal density in serial sections of nucleus caudatum of the cat (1).

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PERSONAL COMPUTER SOFTWARE FOR EVALUATION OF SPIROERGOMETRIC FUNCTIONAL DIAGNOSIS. *V. Soulek<sup>1</sup>, M. Stork<sup>2</sup>*, <sup>1</sup>University Hospital, Hradec Králové and <sup>2</sup>Institute for Mechanical and Electrical Engineering, Plzeň.

We describe a program SPIRO for IBM PC which has been developed at the Institute for Mechanical and Electrical Engineering in Plzeň in cooperation with the University Hospital in Hradec Králové. The program address evaluate functional diagnostic tests in sport medicine. These tests are accomplished by using either a bicycle or tread-mill ergometer. To cope with any methods of functional diagnosis the program allows the following 4 operational set-ups: 1) standard load in steps of 50 W up to the maximum value, 2) any presentable load (sub- or maximal) for evaluation of the ventilation threshold, 3) a standard method of evaluation W 170 including the evaluation of blood pressure and estimation of the  $VO_{2max}$ , 4) spirometry without evaluation of indexes at the maximal value for determination of the  $O_2$  deficit. The input data are loaded from a diskette or a hard disk. The program is controlled by a keyboard or through a command file. Program SPIRO also provides editing of input data, print-outs of results and sorting of data sets according to conditions defined by the user. The program has 190KB. It has been written in Turbo Basic with time critical routines programmed in the assembler.

SOFTWARE FOR RECORDING OF ELECTROPHYSIOLOGICAL EXPERIMENTS AND FURTHER POSSIBILITIES OF IBM PC-ASSISTED DATA PROCESSING. *I. Stavrovský, M. Karhánek, J. Pavelková*, Institute of Molecular Physiology and Genetics, Slovak Academy of Sciences, Bratislava.

The software allows experimental data recording via a converter, graphical representation and storage in data files. The program employs a 12-bit ADDA card with a voltage range of 0-9 V and a minimum sampling interval of 130  $\mu$ s, although it also can work with other types of converters. Simultaneous sampling from a maximum of 5 channels and external synchronization are possible. Discrete or analog displaying modes are possible. The individual records can be displayed independently or simultaneously. The scale type can be selected. All sampling and display parameters can be set in advance and stored in a parameter file. The program also allows record averaging, measurements using two cursors, and screen hardcopying. The software has been written in C, and it is menu-operated. An extension to this software was developed to convert data files into ASCII files so that they can be imported by any commercial or independently developed software allowing analysis of experimental records.

COMPUTER-ASSISTED MEASUREMENT OF RADIOACTIVITY FLUXES. *J. Hochmann, J. Hochmannová, V. Šajter, D. Zacharová*, Institute of Molecular Physiology and Genetics, Slovak Academy of Sciences, Bratislava.

The method uses a radioactivity detector to record pulses from an experimental object. The pulses are processed in a shaping circuit and fed to a pulse amplifier. They leave the amplifier via an interface and enter a pulse counter to be stored in the computer memory. The time interval of recording, background interval and time measurement all are computer controlled. The values measured are displayed on the working space of a screen. The software developed serves mathematical computing of constants, subtraction of partial backgrounds, as well as mechanical printout of the results. The major contribution of the method lies in a substantial improvement of measurement flexibility and evaluation.

#### 4. Cardiac Physiology

EFFECT OF LOW CONCENTRATION OF CREATINEPHOSPHATE (CP) ON PROTEIN COMPOSITION OF THE RAT MYOCARDIUM. *V. Pelouch, M. Milerová<sup>1</sup>, B. Ošťádal*, Institute of Physiology, Czechoslovak Academy of Sciences and <sup>1</sup>Kardiocentrum, Motol Hospital, Prague.

Current concepts of the role of CP in cardiac contraction is controversial: CP may play a role a) as a carrier from mitochondria to myofibrils or b) as an energy buffer for ATP. The aim of our study was to estimate the effect of CP on protein profiling of the developing cardiac muscle. Chronic treatment (5 weeks, starting from 15th day) with GPA (guanidinopropionate-1 % in drinking water) induced low stable CP levels in the myocardium. GPA-induced retardation of body growth was connected with increased relative weight of the myocardium. The concentration of collagenous proteins in both ventricles was significantly elevated due to higher synthesis of the soluble collagenous fraction (predominantly collagen III). On the other hand, the concentration of non-collagenous fractions (contractile and sarcoplasmic proteins) was markedly decreased; this effect was more pronounced in the right ventricle. Our results thus point to the important role of CP in synthesis of individual myocardial protein fractions during the early postnatal period.

**THYROID CONTROL OF POSTNATAL DEVELOPMENT OF CARDIAC CONTRACTILE FUNCTION IN THE RAT.** F. Kolář, E.K. Seppet<sup>1</sup>, R. Vetter<sup>2</sup>, J. Procházka, J. Grünemel, B. Ošťádal, Institute of Physiology, Czechoslovak Academy of Sciences, Prague, <sup>1</sup>Tartu University, Tartu, Estonia and <sup>2</sup>Institute of Cardiovascular Research, Berlin-Buch, FRG.

Hyper- and hypothyroidism were induced in newborn rats by daily s.c. injections of L-triiodothyronine ( $10 \mu\text{g} \cdot 100 \text{g}^{-1}$  b.w.) and by 0.05 % 6-n-propyl-2-thiouracil in drinking water to mothers, respectively, both during 3 weeks. Hyperthyroid animals exhibited increased cardiac contractility, lower sensitivity to the negative inotropic effect of verapamil, and higher inhibition of the relaxation rate by ryanodine as compared with euthyroid rats; hypothyroidism resulted in opposite changes. The oxalate-supported  $\text{Ca}^{2+}$  uptake in ventricular homogenates was increased by 117.6 % in hyper- and decreased by 63.4 % in hypothyroid hearts. These data indicate that the normal postnatal increase of the cardiac sarcoplasmic reticular function is markedly affected by the changes in thyroid status. The contribution of transsarcolemmal  $\text{Ca}^{2+}$  fluxes to the regulation of contractile function seems to be, therefore, lesser in hyper- and greater in hypothyroid rats, when compared to euthyroid animals of the same age.

**THE NECESSARY NUMBER OF SECTIONS FOR EVALUATING THE INFARCT SIZE IN THE DOG.** M. Dostál, S. Doležel, V. Pavlíček, S. Bartáková, I. Kocur, Institute of Pathological Physiology, Masaryk University, Brno.

The LAD coronary artery was ligated and the infarct size in frozen heart ventricles was examined morphometrically after three hours by manually dividing the ventricles into five transverse slices. Furthermore, five equidistant macrosections were cut from each slice and stained by the PAS technique which makes it possible to estimate the infarct size according to the loss of glycogen. The standard error of the mean indicates the precision of the method by this calculation. For this reason, we used a gradually increasing number of macrosections for the calculation: only one macrosection of each slice was taken for calculation at the first step and 5 sections of each of the 5 slices were used at the last fifth step. It was observed that the S.E.M. decreased when the number of macrosections was greater. The character of the decrease of S.E.M. was hyperbolic. The asymptotic part of the hyperbole was reached by using 15-20 macrosections. Thus, the use of a larger number of macrosections could not appreciably contribute to a greater precision of the technique and, *vice versa*, insufficient precision was obtained if less than 15 sections were assessed.

**ELECTROCARDIOLOGIC FEATURES OF RIGHT VENTRICULAR DILATATION.** I. Ruttkay-Nedecký, E. Vanžurová<sup>1</sup>, K. Kandlíková<sup>1</sup>, L. Stanová<sup>1</sup>, L. Valentíková<sup>1</sup>, M. Malacký, R. Osvald, Institute of Normal and Pathological Physiology, Slovak Academy of Sciences and <sup>1</sup>Institute for Cardiovascular Diseases, Bratislava.

The atrial septal defect is a natural model of right ventricular volume overload resulting in dilatation of the ventricle. Its electrocardiological features were identified as those undergoing regression after surgical repair of the defect in a group of 55 children and adolescents aged 3-24 years (median 9). Postoperative studies were obtained 1-9 years after the intervention. The regression of right ventricular dilatation resulted in a significant increase of the spatial magnitude of the vector of left ventricular activation (40 ms after QRS onset). This is attributed to the normalization of the left ventricular end-diastolic volume and decrease of the magnitude of vectors indicating activation of the basal parts of the right ventricle (50-70 ms QRS). Furthermore, it is accompanied by orientational changes, due to the regression of right ventricular dilatation. The dipolar electrocardiotopographic representation improved the comprehensive nonparametric treatment of the observed data.

VCG IMAGE OF VARIATION OF THE LOCALIZATION OF SPECIFIC CONDUCTING SYSTEM TERMINATIONS IN THE LEFT VENTRICLE. *V. Szathmáry, I. Ruttkay-Nedecký, R. Osvald*, Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

A realistic computer model of the propagation of ventricular activation was used to study the effect of varying the position of specific conducting system terminations in the left ventricle and septum, representing the places of initial activation, on the resulting spatial heart vectors. Four differently localized foci of initial activation, each of them represented by one model element, were considered: in the central part of the left septal surface, posteriorly about one third of the distance from the apex to the base, paraseptally anteriorly about half of the ventricular height and in the upper part of the anterior free wall. During the model experiments, the positions of the initial elements were shifted  $\pm 5$  mm in the vertical and lateral direction either separately or in different mutual combinations.

Small variations of the initial activation site in the basal parts of the left ventricle led to significantly smaller changes of the vectorcardiographic loop than variations of the same extent if the initial activation was located more apically.

THE FIRST DERIVATION OF THE VECTORCARDIOGRAPHIC QRS LOOP DURING HYPERVENTILATION AND HYPOXIC VENTILATION. *Š. Kujaník, L. Baldovský<sup>1</sup>*, Department of Physiology, Faculty of Medicine, Šafárik University and <sup>1</sup>Department of Functional Diagnostics, Faculty Hospital with Policlinic, Košice.

The influence of normal ventilation at rest (control), hyperventilation (HV) and hypoxic ventilation (HXV) on the magnitude and direction of the positive (PM) and negative maxima (NM) of the first derivation of the Frank lead ECG was investigated in 27 young healthy women. PM and NM were measured after HV lasting 75 s, during the third and fifth minutes and during maximal alterations of HXV in the intermediate respiratory position. Significant alterations were found only in the frontal plane after HXV - a decrease in PM magnitude during the 3rd and 5th min and HXV<sub>max</sub> changes of the PM angle and HXV<sub>max</sub> and a decrease in the NM magnitude during the 3rd min, 5 min and HXV<sub>max</sub>. The intrinsicoid deflection was not significantly altered. It is being suggested that the decrease in PM and NM is caused by a lowering of the maximal speed of the QRS loop record and changes in the PM direction by slowing of ventricular depolarization around the 30th millisecond.

CHANGES OF THE SPECIFIC MYOCARDIAL POTENTIAL DURING THE DEVELOPMENT OF EXPERIMENTAL LEFT VENTRICULAR HYPERTROPHY. *L. Bachárová<sup>1</sup>, M. Bemadič<sup>2</sup>*, <sup>1</sup>Research Institute of Medical Bionics and <sup>2</sup>Institute of Physiology, Comenius University, Bratislava.

Changes in the cardioelectric field were studied during adaptation of the heart to an experimentally induced volume overload in rabbits. Frank orthogonal electrocardiograms were recorded and related to the LV mass. A new parameter - the specific potential - was calculated as a ratio of maximum spatial vector magnitude (QRS<sub>max</sub>) and left ventricular mass. After a transient decrease of QRS<sub>max</sub> in the early postoperative period it increased in the stage of developed hypertrophy. Values of the specific potential were decreased during the whole period of observation, they did not reach the initial values even at the stage of developed hypertrophy, when the maximal increase of QRS<sub>max</sub> values was observed. Changes in the cardioelectric field during LVH development are not proportional to LV mass increase. The suggested new parameter - the specific potential - makes it possible to quantify this relationship.

## 5. Physiology of Blood Circulation

**DYNAMIC VASCULAR COMPLIANCE (DVC) AND ITS USE IN NONINVASIVE MEASUREMENT OF BLOOD PRESSURE.** *J. Peňáz*, Department of Physiology, Faculty of Medicine, Masaryk University, Brno.

When blood pressure is measured in the finger using volume clamp method (1), the condition of zero transmural pressure must be fulfilled. One of its criteria is the DVC of the artery under the cuff which can be estimated in the following way. A relatively rapid vibration (50 Hz, 1-2 kPa) is superimposed on the cuff pressure and vascular volume oscillations thus produced are measured by a photoelectric system. The volume oscillations are selectively amplified and rectified to obtain a continuous curve representing the DVC. The shape of the DVC pulse curve depends on the transmural pressure (TP): at positive TP (cuff pressure below arterial pressure), it resembles the inverted pressure pulse, at slightly negative TP, it shows a marked positive systolic peak. The principle was used for continuous control of the setpoint in a new noninvasive blood pressure monitor(2).

1. *Peňáz J.*: Cs. Patent Nr. 133205, 1969.

2. *Peňáz J.*: U.S. Patent Nr. 4869261, 1989.

**BLOOD PRESSURE AND ITS REACTIVE CHANGES IN PREMATURE NEONATES.** *O. Andrásyová, E. Kellerová, E. Árendášová<sup>1</sup>, D. Gabrielová<sup>1</sup>*, Institute of Normal and Pathological Physiology, Slovak Academy of Sciences and <sup>1</sup>Faculty of Medicine, Comenius University, Bratislava.

The aim of this study was to investigate the resting blood pressure (BP) and heart rate (HR) and their reactive changes in 126 premature newborns in a stabilized health state, no longer under intensive care. Gestational age 25-37 weeks, average postnatal age 29 days, birthweight  $2017 \pm 393$  g, actual body weight  $2336 \pm 433$  g. The resting values of BP  $64 \pm 11/39 \pm 9$  mm Hg are practically identical with those in term-neonates in the first week of life, except for HR which was significantly higher in prematures  $143 \pm 19$  beats per min as compared with HR in term-neonates 115 beats. Systolic BP increases in correlation with postnatal age. The reactive increments of BP and HR to crying and prone body position are evident but significantly smaller in comparison to term-neonates. In the head-up position the BP changes are individually remarkably variable and insignificant if averaged, in contrast to full-term newborns, in whom the BP increases by 17-19 % and HR by 8 % on the average (1). It seems that maturity is one of the factors which could explain the different opinions on the existence of BP reactions caused by head-up position in neonates.

1. *Kellerová E, Andrásyová D.*: Bratisl. lek. Listy 91: 241-246, 1990.

**DISTRIBUTION OF BLOOD PRESSURE VALUES IN PRESCHOOL CHILDREN.** *V. Regecová, E. Kellerová*, Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

This is a part of an epidemiological study "Blood pressure variability in relation to age, development, familiar and environmental factors", performed in a sample of 1500 kindergarten children in Bratislava. From the analysis of the distribution of blood pressure (BP) values in the horizontal position in children up to 5 years old follow the limits for the upper and lower quartiles for systolic BP 90-105 mm Hg, diastolic 55-60 mm Hg and for heart rate (HR) 80-104 beats.min<sup>-1</sup>. In the ortho-position the limits shifted to 95-115 mm Hg systolic, 60-70 mm Hg diastolic BP and 90-120 beats per min for HR. Only in 5 % of subjects the BP values were above 120/70 mm Hg, HR above 118 per min in the supine position and 125/75 mm Hg and 126 beats per min in the ortho-position. In older children, the BP distribution changes and the upper limits rise by 5-15 mm Hg. The orthostatic values increase by 3-10 % of the horizontal ones on the average. This increment decreases with age. With the exception of boys 5.5-6.5 years, in whom the orthostatic BP was significantly higher, no sex-related differences were found in the mean values or in the distribution of BP values.

**MECHANISMS PARTICIPATING IN THE INHIBITION OF SYMPATHETIC CONSTRICTION OF SAPHENOUS ARTERY AFTER I.A. INJECTION OF ACETYLCHOLINE.** *M. Gerová, E. Barta<sup>1</sup>, J. Gero,* Institute of Normal and Pathological Physiology and <sup>1</sup>Institute for Heart Research, Slovak Academy of Sciences, Bratislava.

A significant decline of sympathetic constriction was found in the saphenous artery (SA) dilated by i.a. administered acetylcholine (ACh). The decline was due to a direct stimulation of EDRF release. However, interfering inhibition of noradrenaline (NOR) release from nerve terminals by ACh could also be involved. In 8 dogs, dilation of SA was induced by nitroglycerin which is irrelevant for nerve terminals, and the magnitude of sympathetic constriction was compared in controls and after nitroglycerin dilation of SA. No significant difference between the above two responses was found. It can thus be concluded that two mechanisms participate in the decline of sympathetic constriction of SA after i.a. ACh administration: a) besides EDRF release b) inhibition of NOR release from nerve terminals.

**THE INCIDENCE OF SPONTANEOUS LESIONS IN THE FEMORAL ARTERY OF THE DOG.** *A. Klasová, V. Smieško,* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

During morphological study, the occurrence of spontaneous lesions of the intima were observed in the femoral artery of mongrel dogs. At the light microscopical level, these regions are characterized by focal interruptions of lamina elastica interna. Scanning electron microscopical examinations revealed the presence of a heterogenic cell population covering the luminal surface of the damaged areas. One type of cells has a smooth surface and the other is characterized by numerous surface protrusions. Further investigation is needed for explaining the cause of this lesion formation. Some association with the development of atherosclerosis is being proposed.

**REMOVAL OF ENDOTHELIUM IN RABBIT EAR ARTERY BY AIR BUBBLES UNDER CONTROL CONDITIONS.** *F. Kristek, R. Töröková,* Department of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

Technique for selective removal of endothelium according to Ralevic *et al.* (1) has been applied to *in vitro* isolated ear artery (CEA) under control of physiological parameters. Segments of CEA were perfused and superfused in a bath. The length, longitudinal tension, perfusion pressure and diameter of the artery were monitored throughout the whole procedure. The protocol for endothelial removing was as follows: 1) high flow (about 15 ml/min) of Krebs solution for 5 min, 2) intermittent perfusion by air bubbles (3 s) and by high flow (3 s) – for a period of 6 min, 3) and high flow for 5 min. The biophysical parameters monitored were in the physiological range during the whole procedure. The absence of the endothelium was diagnosed in the electron microscope. The integrity of smooth muscle was verified by transmural stimulation.

*Relevic V., Kristek F., Hudlicka O., Burnstock G.: Circ. Res. 64: 1190–1196, 1989.*

**IMPAIRMENT OF ENDOTHELIUM-DEPENDENT RELAXATION IN THE RABBIT AORTA AFTER COLD STORAGE.** *J. Török, F. Kristek, M. Mokrášová*, Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

The purpose of the present investigation was to determine the effects of cold storage on endothelium-dependent relaxation (EDR) elicited by acetylcholine (ACh) in rings of rabbit thoracic aorta. In preparations which were precontracted to a stable plateau by  $10^{-6}$  mol/l noradrenaline, ACh elicited relaxation in fresh aortas if the endothelium was intact. Relaxation produced by ACh was gradually decreased after 3-7 days cold storage. A dose-dependent relaxation curve obtained by sodium nitroprusside in fresh aortas was not different from those obtained after 3-7 days cold storage. Electron microscopy of the tissue after 3 days in cold storage showed partial damage of endothelial cells (EC). The number of damaged EC was increased after 7 days. Some of them were damaged to such an extent that part of the internal elastic lamina was deprived of endothelial cell lining. These findings demonstrate that the impairment of EDR is probably due to the gradual destruction of EC.

**TURNOVER OF NUCLEIC ACIDS IN THE CORONARY WALL AND MYOCARDIUM DURING A SHORTLASTING PRESSURE OVERLOAD.** *O. Pecháňová, M. Gerová, E. Barta<sup>1</sup>, J. Kožík*, Institute of Normal and Pathological Physiology and <sup>1</sup>Institute for Heart Research, Slovak Academy of Sciences, Bratislava.

An increase in RNA synthesis was revealed in the coronary wall after it had been exposed to changes that accompany a shortlasting volume overload of the heart. Furthermore, RNA synthesis in the coronary wall was determined after shortlasting pressure overload (4 hours) of both the heart and coronary artery. Blood pressure of 180-200 mm Hg was induced by stepwise constriction of the abdominal aorta above the renal arteries and lasting 3-4 hours. Control experiments were performed without aortal constriction. The spectrophotometric method was used for analysis of nucleic acids, separately in the RIA and RC wall. The RNA concentration increased by 11.6 % and 1.0 % respectively in pressure overload animals in comparison to that of the controls. In the myocardium, the RNA concentration increased by 12.7 %. However, no changes were found in the concentration of DNA.

**CEREBRAL BLOOD FLOW IN RATS AFTER EXPOSURE TO STAGNANT ANOXIA (POSITIVE RADIAL ACCELERATION 10g).** *S. Trojan, J. Kapitola*, Institute of Physiology and Laboratory for Endocrinology and Metabolism, Faculty of Medicine, Charles University, Prague.

Cerebral blood flow was studied in 41 adult rats of our own breed of the average weight for males  $277 \pm 15$  g and for females  $254 \pm 14$  g. Animals were exposed to positive radial acceleration of 10g for a period of one minute shorter than the lethal dose, i.e. for 2 minutes. Twenty hours later the measurements using <sup>85</sup>Sr microspheres according to Kapitola *et al.* (1) were performed in anaesthetized animals. The cardiac output was calculated from the capacity of microparticles in the blood (femoral artery), the blood flow in the tissue was determined from the cardiac output and the capacity of microparticles in % of the dose. The blood flow was determined in the cortex, subcortical areas, the cerebellum and in the medulla oblongata. The measurements demonstrated a tendency to decrease MV both in male and female rats. An opposite trend was exhibited by the caption of <sup>85</sup>Sr microspheres which was slightly increased after anoxia ( $P < 0.05$ ) in all the examined regions of the brain. The hyperaemia which we found may not be an advantage for the brain recovering from hypoxia, because of the more intensive toxic influence of the arising superoxide radical O<sub>2</sub>.

1. Kapitola J. *et al.*: Physiol. Bohemoslov. 36: 155-158, 1987.

## UTILIZATION OF AUTOOXYGENATION DURING A CARDIOPULMONARY BYPASS.

*I. Gabauer, J. Styk, J. Slezák, D. Panca, Institute for Heart Research, Slovak Academy of Sciences, Bratislava.*

The cardiopulmonary bypass has been used in cardiac surgery for more than 35 years. In spite of technical improvements, oxygenators are still traumatic to the blood. The lungs are the best and the least traumatic "oxygenator". A double pump system without an oxygenator was used in 20 dogs. The cardiopulmonary setup consisted of two separate systems bypassing the right and left sides of the heart for one hour. The results have confirmed the full reliability of the autooxygenation during CPB. All evaluated haemodynamic parameters varied in the normal range. Arterial O<sub>2</sub> saturation achieved 100 %, haemolysis was less severe than when oxygenators were used and the loss of thrombocytes was minimal. We believe that this method may further improve the results of aorta-coronary bypass surgery besides the cost reduction obtained by excluding an oxygenator.

A HAEMODYNAMIC EFFECT OF SUBSTANTIA NIGRA STIMULATION UNDER CONDITIONS OF HAEMORRHAGIC HYPOVOLAEMIA. *I. Bračoková, L. Angyán<sup>1</sup>, M. Murín, I. Ivančo, Department of Physiology, Medical Faculty, Šafárik University, Košice and <sup>1</sup>Pécs, Hungary.*

Stimulation of the substantia nigra (SN) evokes a somatomotor as well as a circulatory response. With the aim to differentiate whether the haemodynamic response is primary or secondary to the somatomotor response, the haemodynamic effect was analyzed in 5 conscious cats and in 10 anaesthetized cats under normal and hypovolaemic conditions. It was found that the increase of blood pressure and the acceleration of heart rate were evoked by the stimulation of SN both in conscious and anaesthetized animals, but the threshold intensity in the anaesthetized animals was higher. The quantitative characteristics of the haemodynamics response were directly dependent on the intensity of the stimulation. After haemorrhage, repeated stimulation with the same intensity as in the normal animals evoked a significantly higher response. These results suggested that SN can play role in cardiovascular integration as well as in adaptation changes of the organism.

## 6. Endocrinology

INTERACTION OF DEXAMETHASONE WITH RAT LIVER NUCLEI. *M. Alexandrová, D. Maščuchová, Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.*

Glucocorticoids combine with a high-affinity cytoplasmic receptor to form steroid-receptor complexes which are translocated to the nucleus, the site of hormone action. We studied the effect of adrenalectomy (ADX) and dexamethasone (Dex) injection on the interaction of <sup>3</sup>H-Dex with rat liver nuclei. ADX rats received an injection of Dex and were killed 2 h or 24 h later. The liver cytosol was isolated and incubated with increasing concentrations of <sup>3</sup>H-Dex in the presence or absence of Dex at 0 °C for 2 h, then isolated nuclei were added and incubated at 25 °C for 1 h. The number of <sup>3</sup>H-Dex binding sites was estimated by Scatchard analysis. ADX alone caused a 50 % decrease of <sup>3</sup>H-Dex binding to nuclei in comparison with intact animals, K<sub>d</sub> decreased significantly. Additional lowering of <sup>3</sup>H-Dex binding was observed 2 h after Dex injection and even 24 h later <sup>3</sup>H-Dex binding was not yet fully recovered. K<sub>d</sub> values returned to those of intact rats. We conclude that the distribution of glucocorticoid binding sites between cytosol and nuclei depends on the concentration of available hormone.

**THE PROPERTIES OF LIVER INSULIN RECEPTOR TYROSINE KINASE IN HEREDITARY HYPERTRIGLYCERIDAEMIC RATS (HTG).** *M. Ficková, P. Hubert<sup>1</sup>, I. Klimeš, M. Hromadová, E. Sebková, P. Bohov, J. Cemel<sup>1</sup>, L. Macho,* Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava and <sup>1</sup>INSERM U338, Strasbourg, France.

The aim of this study was to investigate the activity of insulin receptor (IR) tyrosin kinase (TK) when the lipid composition of the liver plasma membrane (PM) had been changed. HTG and control Wistar rats were fed a standard diet with P/S ratio = 1 (polyunsaturated : saturated fatty acids) or a modified diet with P/S = 3 (source-fish oil, group FO) for 3 weeks *ad libitum*. Both diets were isocaloric. HTG animals displayed significantly elevated values of total serum triglycerides (TG) together with their content in VLDL and LDL lipoprotein fractions. The FO diet significantly reduced total TG, VLDL-TG and LDL-TG. In PM, the FO diet significantly increased n-3 PUFA content, with a concomitantly decrease of n-6 PUFA. In PM of HTG rats, the elevated content of total and esterified cholesterol was reduced by the FO diet. PM fluidity was not affected by the changes of membrane lipid composition. IR autophosphorylation was not influenced by the diet, or by the type of animals. Exogenous TK activity of liver IR was independent of the PM lipid composition. The changes in the lipid composition of liver PM of HTG rats thus did not influence the insulin receptor tyrosine kinase activity.

**ADENYLATE CYCLASE ACTIVITY CHANGES IN RAT HYPOPHYSIS LOBES AFTER MILD PROTEOLYTIC DIGESTION WITH CHYMOTRYPSIN.** *V. Klenarová, P. Šída,* Institute of Pathologic<sup>1</sup> Physiology, First Medical Faculty, Charles University, Prague.

The role of the adenylate cyclase (AC) complex in individual lobes of the rat hypophysis is still not clear. In this study we used mild proteolytic digestion with chymotrypsin (CHY) to disclose possible adrenergic effects which are not detectable under regular assay conditions. AC activity was estimated by a procedure using <sup>32</sup>P-alpha-ATP as substrate. The addition of GTP to AC assay appeared to be essential for full expression of the isoproterenol (ISO) effects. CHY (10 µg/ml) increased AC activity independently of the stimulating agent used; in the intermediate lobe, CHY increased the effects of ISO substantially, while those of forskolin were enhanced only slightly. In the neurohypophysis, the addition of CHY did not evoke any adrenergic effects. It can be concluded that mild proteolytic digestion of AC preparations did not prove to be useful for a closer characterization of AC in the rat hypophysis.

**CENTRAL REGULATION OF PROLACTIN (PRL) SECRETION DURING HAEMORRHAGIC STRESS IN RATS.** *J. Jurčovičová, G.B. Makara<sup>1</sup>, M. Dobráková, Z. Opršalová,* Institute of Experimental Endocrinology, Bratislava and <sup>1</sup>Institute of Experimental Medicine, Hungarian Academy of Sciences, Budapest, Hungary.

Haemorrhage (HEM) corresponding to 25 % of blood volume is accompanied by a fall of blood pressure by about 5.2 kPa and leads to a pronounced release of PRL in nonanaesthetized male rats. When studying the mechanism of this phenomenon we focused our attention to structures of the central nervous system involved in the PRL response to other stimuli. Lesions of the paraventricular nucleus (PVN) or posterior isolation of the mediobasal hypothalamus (MBH) both performed one week before the experiments, lowered the PRL secretion due to HEM. Posterior pituitary denervation (one week before the experiment) eliminated the HEM-induced PRL release. These results suggest the following conclusions: the structures located posteriorly to the MBH are involved in the transfer of signals triggering PRL release due to HEM. PVN and the neural lobe represent important regions for PRL releasing activity under HEM.

DOES THE PAINFUL STIMULATION AFFECT THE PROLACTINE LEVEL? R. Rokyta, S. Hrabětová, J. Jurčovičová<sup>1</sup>, J. Hrabě<sup>2</sup>, H. Šťastná, Department of Physiology, Third Medical Faculty, <sup>2</sup>Department of Mathematics, Faculty of Science, Charles University, Prague and <sup>1</sup>Institute of Endocrinology, Slovak Academy of Sciences, Bratislava.

Prolactine may affect nociceptive perception. Experiments were performed in 6 groups of rats (10 animals each): 1-1 controls, 1-2 rats with the administration of 0.9 % NaCl (0.5 ml i.m. 4-times, 1 per week), 1-3 rats with neopholline administration (applied in the same manner), 2-1 nociceptively stimulated rats only (Rokyta et al. 1985), 2-2 nociceptively stimulated rats with the administration of 0.9 % NaCl, 2-3 nociceptively stimulated rats with neopholline administration. The level of prolactine and pain sensitivity (tail-flick method) were measured one week after the last injection. The data were processed by analysis of variance and factor analysis. Painful stimulation had no significant effect on the prolactine level, while in the 2-3 group it had an opposite effect on nociceptive perception than in the other groups studied, i.e. it decreased it. After neopholline administration the weight of the hypophysis and prolactine level were increased while the weight increment was decreased.

Rokyta R., Kubík V., Korpancová J.: *Ontogenesis of the brain* 4: 199-203, 1985.

EFFECTS OF LH-RH AND OF ITS CHEMICAL ANALOGUES ON OXYTOCIN, VASOPRESSIN AND ESTRADIOL SECRETION BY BOVINE GRANULOSA CELLS *IN VITRO*. S.V. Nikolaiev, A.V. Sirotkin<sup>1</sup>, J. Nitray<sup>1</sup>, S.V. Burov, Chemical Research Institute, Leningrad State University, USSR and <sup>1</sup>Research Institute of Animal Production, Nitra.

Oxytocin, arginine-8-vasopressin and estradiol-17beta production by bovine granulosa cell culture were analysed without, or in the presence of LH-RH, its agonist (cycle-/Pro<sup>1</sup>, O Phe<sup>6</sup>/LR-RH, des<sup>1-3</sup>/D-Ala<sup>6</sup>/LH-RH), or antagonist (/D Phe<sup>2</sup>, D Phe<sup>6</sup>/LH-RH, /D Phe<sup>2</sup>, D Phe (NH<sub>2</sub>)<sup>6</sup>/Lh-RH cycle /Pro<sup>1</sup>, D Phe<sup>2</sup>, D Phe<sup>6</sup> /Lh-RH). It was found that all the preparations used stimulated oxytocin and estradiol secretion. Vasopressin release was also significantly increased after all Lh-RH antagonists (but not after LH-RH or its agonist) treatments. The present observations have demonstrated the direct influence of LH-RH and its analogues on both steroid and nonpeptide hormone production by bovine granulosa cells. Independent mechanisms of LH-RH and the action of its analogues on pituitary and ovarian oxytocin and estradiol are proposed. On the other hand, the chemical, but not the functional resemblance of LH-RH receptors in the hypophysis and vasopressin-producing granulosa cells can not be excluded.

PRODUCTION OF PROLACTIN (PRL) OR PROLACTIN LIKE ACTIVITY (PLA) BY OVARIAN GRANULOSA CELLS OF COWS *IN VITRO*. A.V. Sirotkin, A. Skokanová, V.P. Polítov<sup>1</sup>, G.V. Martinčenko<sup>1</sup>, J. Nitray, J. Jurčovičová<sup>2</sup>, Research Institute of Animal Production, Nitra, <sup>1</sup>Research Institute of Reproduction and Genetics of Farm Animals, Leningrad, USSR and <sup>2</sup>Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.

In ovarian granulosa cells m-RNA for PRL has been found (1) indicating that PRL is synthesized by the ovaries. We therefore measured the production of PRL (PLA) by ovarian granulosa cells of cows *in vitro* with or without addition of peptide hormones. Basal PRL values in the incubation medium varied between 1.2 - 1.6 ng 10<sup>6</sup> cells per day. FSH and growth hormone (GH) (10-10<sup>4</sup> ng ml<sup>-1</sup>) stimulated PRL (PLA) production in a dose-related pattern. Oxytocine in low doses inhibited and in high doses stimulated PRL (PLA) accumulation. Arginine-vasopressin, arginine-vasotocin and LHRH were without effects. These results speak for the PRL (PLA) production by the ovaries which can be affected by FSH, GH and oxytocine.

I. Einspanier E., Pitzel L., Wuttke W., Hagendorf G., Preuss K.D., Kornalinou E., Scheit K.H.: *Febbs Lett.* 204: 34-37, 1986.

**INHIBITORY EFFECT OF OESTROGEN ON BONE BLOOD FLOW IN RATS.** *J. Kapitola, J. Kubíčková<sup>1</sup>, Laboratory for Endocrinology and Metabolism and <sup>1</sup>Third Medical Clinic, Faculty of Medicine, Charles University, Prague.*

Local circulatory changes in the bones of rats after the administration of oestrogens were studied by means of <sup>85</sup>Sr-labelled microsphere method. After four weeks administration of oestradiol benzoate (EB, Agofollin Depot, CSFR) the blood flow through the tibia of female rats decreases to 57.1 % of the controls; the reaction involved is local (no change in cardiac output, decrease in <sup>85</sup>Sr-microsphere uptake). The decline is more frequent in the trabecular tissue; it can also be demonstrated in the kidneys, but not in the heart, liver, intestine, muscles, skin and fat. EB exerts his action also in male rats. Daily injections of oestradiol dipropionate (Agofollon) already reduce tibial blood flow during the first days, the decrease being significant by the 9th day. EB reduces the incorporation of <sup>45</sup>Ca and <sup>3</sup>H-proline as well as the uptake of <sup>85</sup>Sr in the bone. The physiological or pathological importance of the presented findings remains to be elucidated.

**BRAIN AND ATRIAL NATRIURETIC PEPTIDE BINDING SITES IN THE RAT ADRENAL GLAND.** *T. Torda, J.M. Saavedra<sup>1</sup>, Institute of Experimental Endocrinology, Bratislava and <sup>1</sup>LCS, NIMH, Bethesda, USA.*

We determined the interaction of porcine brain natriuretic peptide (BNP) with rat atrial natriuretic peptide (ANP) receptors in the adrenal glands of rats. Both [<sup>125</sup>I]-peptides bound to the zona glomerulosa, zona fasciculata and adrenal medulla. BNP inhibited ANP binding to its receptors in all adrenal areas. In zona glomerulosa, BNP competes with ANP for the same binding sites, with lower affinity, demonstrated by autoradiography and binding to soluble receptors. [<sup>125</sup>I]-BNP had a lower number of maximal binding sites and lower apparent affinity than [<sup>125</sup>I]-ANP. Dose response activation of guanylate cyclase exhibited a lower affinity of BNP when compared to ANP. Changes in amino acid sequences are most probably responsible for the differences in binding kinetics between ANP and BNP.

**THE EFFECT OF ACUTE AND CHRONIC ADRENALECTOMY (ADX) ON THE SECRETION OF ATRIAL NATRIURETIC PEPTIDE (ANP) AND NATRIURESIS DUE TO SALINE INFUSION.** *Š. Lichardus, J. Ponc, D. Jeřová, F. Földes, Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.*

The adrenal steroids regulate ANP gene expression, and represent the bulk of digoxin-like substances. Some have been claimed to play a role of another natriuretic hormone, inhibitor of the sodium pump. Both acute and chronic ADX should thus impair natriuresis and chronic ADX should also impair ANP release. It was, however, found in rats under Nembutal anaesthesia that natriuresis in both acute and chronic ADX did not statistically differ from that in the controls (it rose in controls from  $0.23 \pm 0.9$  to  $7.8 \pm 1.0$   $\mu\text{mol}/\text{min}$  and was matched during peak natriuresis in acute ADX rats by plasma ANP of  $114.9 \pm 15.4$   $\text{pg}/\text{ml}$  but in chronic ADX rats only by  $30.9 \pm 9.3$   $\text{pg}/\text{ml}$ ). It is concluded that adrenal steroids are indeed important for ANP release but their role in natriuresis due to saline loading is doubtful. Thus the natriuretic hormone-inhibitor of the sodium pump does not seem to be produced by the adrenals.

**ENDOGENOUS INHIBITOR OF  $\text{Na}^+ - \text{K}^+ - \text{ATPASE}$  : WHOLE BLOOD AND ERYTHROCYTE ACTIVITY.** N. Michajlovskij, B. Lichardus, P. Bakoš, Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.

As early as in 1970, Lichardus *et al* (1) could show that partially purified extracts of whole bovine blood had strong antinatriferic activity on frog skin (inhibiting  $\text{Na}^+ - \text{K}^+ - \text{ATPase}$  activity). Blood sample extracts were prepared using rapid deproteinization with trichloroacetic acid (TCA), centrifugation, and subsequent ether extraction of the TCA from the supernatant. The results of the present work 1. provided support for a strong antinatriferic effect of rat and human volunteer blood extracts prepared in the same manner; 2. demonstrated that erythrocytes are the major carriers of antinatriferic activity of the whole blood, whereas heparinized plasma remains almost without any effect. It can be suggested that the erythrocyte  $\text{Na}^+ - \text{K}^+ - \text{ATPase}$  functions as a receptor of the circulating endogenous inhibitor, and TCA releases it from the enzyme. The inhibitor is ether-insoluble.

I. Lichardus B., Pliska V., Uhrin V., Barth T.: In: Regulation of Body Fluid Volumes by the Kidney, J.H. CORT, B. LICHARDUS (eds), Karger, Basel, 1970, pp. 114-121.

## 7. Neuroontogenesis

**CORTICAL AFTERDISCHARGE THRESHOLDS IN DEVELOPING RATS,** V. Makal<sup>1</sup>, P. Mareš<sup>1,2</sup>, M. Malá<sup>1</sup>, M. Miňová<sup>1</sup>, H. Kubová<sup>2</sup>, <sup>1</sup>Department of Pathological Physiology, Third Medical Faculty, Charles University and <sup>2</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Rhythmic cortical stimulation induced afterdischarges (ADs) and minimal clonic seizures in adult rats. An identical motor pattern was observed during the stimulation (1). An ontogenetic study was performed on rats 12, 18, 25 and 90 days old to establish the threshold intensities necessary for eliciting clonic movements during stimulation, cortical ADs accompanied by clonic seizures and the transition to the second "limbic" type of ADs. Stimulation series of 8-Hz frequency and 15-s duration consisted of bipolar pulses of 1-ms duration with an intensity from 200  $\mu\text{A}$  to 15 mA. Each age group comprised at least 20 rats. Clonic movements accompanying stimulation were induced by lower current intensity than ADs in each rat in all age groups. The threshold intensity necessary for evoking ADs was lowest in 18-day-old and highest in adult rats. The transition from spike-and-wave AD to "limbic" AD was observed only in adult animals.

I. Kubová H., Makal V., Miňová M., Vaňková S., Mareš P.: Arch. Int. Pharmacodyn. 307: 49-59, 1990.

**ANTICONVULSANT ACTION OF LAMOTRIGINE DURING ONTOGENESIS IN RATS.** L. Staňková, P. Mareš, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The mechanism of action of a new anticonvulsant drug Lamotrigine (LMT, Wellcome<sup>®</sup>) probably depends on an inhibition of glutamate release. Motor seizures induced by metrazol were used to obtain basic data on the action of LMT during ontogenetic development. LMT was injected intraperitoneally in doses of 5, 10 and 20 mg/kg to rats aged 7, 12, 18, 25 and 90 days. Pentamethylenetetrazol (PTZ) was injected s.c. 30 min after LMT in a dose of 100 mg/kg in all age groups except the 18-day-old rats where a 90-mg/kg dose was given. LMT was unable to affect the incidence, pattern and latency of minimal (i.e. predominantly clonic) metrazol seizures. On the contrary, LMT selectively abolished the tonic phase of major, i.e. generalized tonic-clonic seizures. Doses of 10 and 20 mg/kg were effective in all the age groups. The clonic phase as well as the latency of clonic seizures remained unaffected by LMT. The profile of action of LMT is similar to that of phenytoin; this action did not change during ontogenesis in rats.

**ONTOGENETIC DEVELOPMENT OF KAINATE-INDUCED AUTOMATISMS.** *L. Velišek, L. Boháčenková, M. Čapková, P. Mareš*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Kainic acid (KA) applied systematically (2-14 mg/kg IP) induces automatisms. We tested the action of alpha-2-adrenomimetic clonidine and 5-HT receptor antagonist ritanserin against KA-induced automatisms in rats aged 7, 12, 18, 25 and 90 days. Whilst scratching was evoked by KA during the whole developmental period studied, a substantial number of wet dog shakes (WDS) was observed after day 18 of age. Clonidine (0.25 mg/kg IP) decreased the incidence of scratching in all age groups whereas the incidence of WDS was reduced only in 18- and 90-day-old animals. Ritanserin (20 mg/kg IP) influenced neither scratching nor the incidence of WDS substantially; moreover, the incidence of tonic-clonic seizures was increased in 7-day-old pups. It appears that the automatism induced by KA might be suppressed by alpha-2-adrenomimetic drugs.

**ONTOGENY OF LEARNING AND DECISION MAKING IN PUPS OF THE LABORATORY RAT.** *S. Fraňková*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Ontogeny of learning was studied in a situation which enabled the pup to select one out of two equivalent solutions. The method employed was the homing response (1). Three six-sided boxes were arranged so that the central box (C) was connected on the right (R) and left (L) sides with a box of the same size by small openings. Three pups were placed into R, other three into L. The tested pup learned to find the way "home" from box C. It was free to select between the R and L box. Learning was compared with that in the situation, where only one box was adjacent to C. On days 16, 21 and 27, the running speed to the "home" box, preference for R or L, and the alternations between R and L runs were recorded. For all age groups learning with the necessity of decision was more difficult when compared with that without decision making. Ontogenetic changes appeared in the running times, in the decreased preference for one side, in the increased number of alternations and oscillations between R and L.

*J. Fraňková S.: Act. Nerv. Super. 1991 (in press).*

**IMPACT OF ETHANOL-DRINKING FOR SEVERAL GENERATIONS ON BEHAVIOUR AND LEARNING DEVELOPMENT.** *Z. Chaloupka, J. Mysliveček*, Institute of Pathophysiology, Charles University, Medical Faculty, Plzeň.

Impact of ethanol drinking throughout several generations at two concentrations (5 % or 10 %) was studied in Wistar rats. Behavioural changes in various generations were not uniform. In rats drinking 5 % ethanol (corresponding to 12° beer), deviations of maternal behaviour were found in the 2nd generation. The 3rd generation is characterized by increased activity ( $p < 0.05$ ) without any significant change in the relative amounts of individual activity types. In pole-jumping avoidance, alcohol drinking rats tended to learn the criterion of 5 consecutive avoidances more slowly, and the reappearance of the first avoidance after extinguishing was significantly delayed ( $p < 0.01$ ). In rats consuming 10 % ethanol (approximately corresponding to wine), learning was already significantly impaired in the first generation.

**STRAIN DIFFERENCES IN CORTICAL BIOELECTRIC ACTIVITY AFTER PRENATAL ADMINISTRATION OF DIAZEPAM.** *J. Hassmannová<sup>1</sup>, J. Mysliveček<sup>2</sup>, R. Rokyta<sup>1</sup>*, <sup>1</sup>Department of Physiology, Third Medical Faculty, Charles University, and <sup>2</sup>Institute of Hygiene and Epidemiology, Prague.

We have shown previously that prenatal administration of diazepam in amounts corresponding to therapeutical dosage, influences learning and memory development in laboratory rats (1,2). We compared the changes of spontaneous cortical bioelectric activity in adult albino Wistar and pigmented Long Evans rats whose dams received 3 injections of diazepam (H, Seduxen 1 mg/kg i.m.) during the third week of gestation. Electroencephalograms of visual (occipital) and auditory (temporal) areas were recorded with stainless-steel needle electrodes; average amplitudes and percentage contribution of individual waves were evaluated. Most pronounced changes after diazepam were found in both strains in the occipital cortex. Whereas the EEG spectrum in albinos was shifted towards faster spectrum components, slow waves prevailed in the pigmented rats. Thus the comparison of EEG frequency spectrum after diazepam exhibited a mirror image in the two strains.

1. *Hassmannová J., Mysliveček J.: Homeostasis 33: 77-88, 1991.*

2. *Mysliveček J., Josifko M., Hassmannová J.: Physiol. bohemoslov. 38: 382, 1989.*

**HOW DOES KETAMINE AFFECT DEVELOPING EMBRYONIC MOTILITY?** *J. Sedláček*, Institute of Physiology, Faculty of Medicine, Charles University, Prague.

Considering that ketamine is believed to act primarily through the NMDA receptor complex, it may be used for neuropharmacological studies involving analysis of the EAA role in the development of embryonic spontaneous motility. 1. Ketamine depressed embryonic motility both after acute (12.5 mg/kg e.w.) and chronic application (average dose  $6.37 \pm 0.72$  mg/kg e.w./24 h, continuous application from day 4 to day 8, 12 or 16 of incubation). 2. The development of resting motility was depressed by 23.1-36.0 % of the control value. This effect appeared during the first 4 days of chronic ketamine application. Extreme changes were found in the activating effect of strychnine (deficit to 48.5 % of control value). 3. It is concluded from these results that ketamine interferes with development of the intrinsic rhythm of spontaneous activity of developing spinal motor generator and with the developing mechanism of GABA-ergic inhibition.

**ANTICONVULSANT ACTION OF OXCARBAZEPINE AND HYDROXYCARBAMAZEPINE DURING ONTOGENESIS IN RATS.** *H. Kubová, P. Mareš*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Oxcarbazepine (OXCZ) is a derivative of carbamazepine devoid of some of its side effects; it is metabolized to hydroxycarbamazepine (HYCBZ), a drug possessing its own anticonvulsant activity. Due to a gift from Ciba Geigy we had the possibility of examining the effects of these two drugs against motor seizures induced by metrazol (pentamethylenetetrazol, PTZ) in developing rats. Animals 7, 12, 18, 25 and 90 days received an i.p. injection of either drug (doses of 15, 30 and 60 mg/kg). Thirty minutes later, PTZ was administered s.c. in a dose of 100 mg/kg with the exception of 18-day-old rats where the 90-mg/kg dose was used. Neither drug influenced minimal (i.e. predominantly clonic) seizures, whereas in major generalized tonic-clonic seizures the tonic phase was selectively suppressed by both drugs in a dose-dependent manner. No significant differences were observed during ontogenesis. The two drugs exhibited the same profile of action as phenytoin and carbamazepine; HYCBZ has an efficacy of its own and may thus be responsible for a part of the anticonvulsant action of OXCZ in human patients.

## 8. Human Neurophysiology

**HYPERVENTILATION DYSPNOEA.** V. Šmejkal, P. Charamza<sup>1</sup>, Institute of Pathophysiology, Second Medical Faculty and <sup>1</sup>Department of Statistics, School of Physics and Mathematics, Charles University, Prague.

Dyspnoea produced in healthy volunteers by resistance breathing and/or by rebreathing depends on the magnitude of the resistance and the drive. We wanted to compare this dyspnoea with hyperventilation dyspnoea. Ventilation was voluntarily increased in 11 medical students. Tidal volume ( $V_T$ ), respiratory frequency, minute ventilation,  $P_{ETCO_2}$  and time were measured. Dyspnoea was assessed by the Borg visual analogue scale (1) on which 0 = no and 10 = maximum breathlessness. The correlation of dyspnoea can be expressed by a regression equation: degree of dyspnoea =  $0.89 V_T - 1.35 P_{ETCO_2} + k$  ( $k$  = individual constant in the range from +3.9 to +8.5). The correlation coefficient  $r = 0.95$ . In contrast to airway-obstruction and hypercapnia, during hyperventilation the higher degrees of dyspnoea correlate with lower  $P_{ETCO_2}$ .

*I. Borg G.: Scand. J. Rehab. Med. 2-3: 92-98, 1970.*

**DIFFERENCES IN SACCADIC EYE-MOVEMENT RELATED POTENTIALS OVER THE OCCIPITAL AND OTHER BRAIN AREAS.** F. Jagla<sup>1</sup>, G. Dimitrov<sup>2</sup>, V. Zikmund<sup>1</sup>, <sup>1</sup>Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava and <sup>2</sup>Institute of Physiology Bulgarian Academy of Sciences, Sofia.

In a previous study the differences in saccadic eye-movement related potentials were described over the right and left occipital areas (1). In the present report, a lower amplitude and shorter latency of premotion positivity, and a longer latency of the lambda response were found over the parietal as compared to the occipital area. The positive potential over the motor cortex and the positive wave with a very slow increase and decrease over the frontal eye fields were markedly expressed during the execution of saccades. The amplitude of the lambda response over the left parietal and of the positive wave over the left frontal areas were higher as compared to the corresponding areas on the right side.

*I. Jagla F., Zikmund V.: Act. nerv. super. 30: 287-289, 1988.*

**VISUAL-MOTOR COORDINATION AND LEARNING DISABILITIES IN MINIMAL BRAIN DISORDERED (MBD) AND HEALTHY SCHOOL BOYS.** P. Cakirpaloglu, T. Radil, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

A computer based method was used for analyzing visual-motor coordination of 120 MBD and 181 healthy boys. Three different types of visual stimuli were generated on the display: meaningless geometrical patterns, numbers and letters. The task for the subjects was to reproduce certain combinations of symbols by pushing the corresponding keys on a computer keyboard with one of the four fingers of the right hand (except the thumb). The program evaluated the average reaction time (RT) for sets with 1, 2 and 3 symbols, mean RT for each finger, the total RT, and the number of errors. The teachers had estimated (using school grades) the success of boys in reading, writing and calculation. The results pointed to difficulties in creating and using specific symbolic entities such as letters and numbers in 7 to 11-year-old MBD boys. This correlated with the general learning disability reflected in school grades.

**DIFFERENCES IN THE LATENCIES OF AUDITORY BRAIN STEM RESPONSES RECORDED SIMULTANEOUSLY FROM SYMMETRICAL AREAS OF THE SCALP IN HEALTHY SUBJECTS.** *J. Petřek*, Institute of Physiology, Faculty of Medicine, Palacký University, Olomouc.

Using analyser HEAD/TNS (1) auditory brain stem responses (ABR) to condensation and rarefaction clicks (60dB SL, 12/s) applied to the right or left ear were recorded simultaneously from symmetrical scalp sites (T<sub>3</sub>, T<sub>4</sub>) of 28 healthy subjects (13 males, 15 females). Linked earlobes served as the reference electrode to provide identical conditions for simultaneous recording of ABRs from T<sub>3</sub> and T<sub>4</sub>. Usually two averages of 1600 consecutive individual responses were obtained from each ear and for each test stimulation. Evaluation of latencies of ABR waves P I, P III and P V has shown that in symmetrical sites of the scalp their average latency was always longer in ABR records of the T<sub>4</sub> electrode than in records of the T<sub>3</sub> electrode, irrespective of which ear was stimulated.

*J. Petřek J., Chytil J., Vašků S.:* Acta Univ. Palacki, Olomouc (Olomouc), Fac. Med. 130, 1991 (in press).

**MODULATION OF THRESHOLDS FOR ELICITING EARLY EXTRALEMNISCAL AUDITORY RESPONSES (EEARs) BY CONDITIONING.** *R. Jirsa, T. Radil*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

EEARs (onset latencies 4-20 ms) evoked by tone - clicks were recorded from different brain stem nuclei and from the dentate gyrus. Thresholds for eliciting EEARs were higher by about 50 dB in comparison with those recorded from the inferior colliculus (IC). Acquisition of a shuttle-box conditioned avoidance reaction to the same stimulus lowered the thresholds for eliciting EEARs by about 15 dB. The stimulus threshold for eliciting the above avoidance reaction was approximately the same as that for eliciting the auditory response in IC. These stimuli, however, were insufficient for evoking the EEARs. Our results suggest that diffuse extralemniscal brain stem excitation is not a necessary condition for active avoidance to auditory signals.

**STANCE ON A TILTED PLATFORM.** *T. Radil, Z. Bohdanecký, J. Vorlíček*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Stabilographic recordings have been performed in humans standing on a platform either horizontal or tilted 7° or 14°; the subjects being oriented in all 4 possible directions with respect to the direction of the tilt and having their eyes either closed (C) or open (O). The instrument used (1) provided graphical output of integral numerical values corresponding to the extent of excursions from the central position of the projection of body mass upon the platform along the anteroposterior (AP) and lateral (L) dimension for the 30 s recording periods. Statistical processing proved that the values are always higher for O than C, higher for 14° than 7° and for the L position with respect to the tilt in comparison with AP. This approach might be adopted for clinical purposes.

*J. Hlavačka F. et al.:* Physiol. bohemoslov. 29: 441-442, 1980.

**BIPOLAR MONOAUURAL GALVANIZATION OF THE LABYRINTH IN MAN.** *V. Mihalik, M. Križková*, Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

Monopolar monoaural or bipolar binaural galvanization techniques are mostly used in research of the vestibular system. The bipolar monoaural galvanization technique has not been reported in the available literature. We employed this technique and have obtained the following results: 1. Bipolar monoaural galvanization evokes directionally nonspecific deviations of the gravity centre of the body (BGC). Terminal to the starting positions (the static phases) of BGC replacement does not show any direction preference. From this we infer that this type of galvanization does not cause intralabyrinth asymmetry, but probably changes the interlabyrinth afferentation differences of functionally reversely directed parts of the maculae. 2. Direction of the dynamic phase of BGC replacement (10 % - 90 % of deviation amplitude) is also laterally unspecific but directionally lies more closely to the interlabyrinth line.

**GALVANIC VESTIBULAR STIMULATION IN AVIATION MEDICINE.** *M. Sázal*, Institute of Aviation Medicine, Prague.

We previously reported our experience of using galvanic vestibular stimulation in pilots on a flight simulator (1,2,3). In the present report, 27 pilots were examined by bipolar, binaural stimulation (sinusoidal  $\pm 1$  mA, direct 2 mA/40 s) during simulated straight-and-level flight. The deviations of the control stick (right hand) and the aircraft in the frontal and sagittal plane were measured before and during these stimulations. Data were analysed by computer including the fast Fourier transformation. The influence of stimulation on the pilot's limb movement of the control stick was evident. The most marked effect of sinusoidal current was found at 0.2 Hz frequency in the frontal plane of movement, when the power spectrum density increased. The alteration of movement in the sagittal plane was not so apparent. The possibility of using galvanic vestibular stimulation for aviation medical assessment is suggested.

1. Sázal M., Hanousek J.: Voj. zdrav. Listy 58: 29 - 33, 1989.

2. Sázal M., Novotný B.: Physiol. bohemoslov. 38: 539, 1989.

3. Sázal M.: Čs. fyziol. (in press.).

**VESTIBULAR-PROPRIOCEPTIVE INTERACTION DURING SPACE POINTING MOVEMENTS IN MAN.** *F. Hlavačka, Th. Mergner<sup>1</sup>, Ch. Siebold<sup>1</sup>, G. Schweigart<sup>1</sup>*, Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava and <sup>1</sup>Abteilung Neurologie, University of Freiburg.

Goal directed motor behaviour depends on the complex interplay of several sensory systems. We studied the excursions of the left arm when subjects tried to keep it stationary in space during various combinations of vestibular and proprioceptive stimuli (angular range  $\pm 8^\circ$ ). The gain and phase of resulting compensatory movements were analysed in the range from 0.025 to 0.4 Hz. The compensatory arm movements were close to "ideal" at the higher stimulus frequencies in all the tested combinations. At the low frequencies, good compensation only occurred during counterphase (functionally synergistic) combinations when the vestibular and proprioceptive stimuli were of a similar magnitude. Otherwise, the results mainly reflected the vestibular input alone. The results were being explained by a non-linear model.

FEEDING LEADS TO SYNCHRONIZATION OF INFANT'S EEG. *K. Paul, J. Díttrichová*, Institute for Care of Mother and Child, Prague.

Emotional stimuli can significantly influence the EEG activity in infants. We found a large increase of synchronization in the EEG even during feeding. Twenty healthy full-term infants were examined during feeding at the age of 2, 10, 18 and 26 weeks. The EEG activity from leads C<sub>3</sub>P<sub>3</sub> and C<sub>4</sub>P<sub>4</sub> was quantified and the "theta index" and "mean amplitude" were statistically evaluated. Both these parameters were significantly higher ( $p < 0.01$ ) during feeding than before and after feeding at the age of 18 and 26 weeks. Synchronization was not present in the younger age groups. Individual infants differed in the intensity of synchronization and these differences were stable on repeated examinations (Kendal's coefficient of concordance  $W = 0.588$   $p < 0.01$ ). Adaptation to the novelty of the experimental situation was followed by increased synchronization. The differences between the 1st and the 3rd day were significant: the "theta index" -  $p < 0.01$ ; the "average amplitude" -  $p < 0.02$ .

PHARMACOLOGICAL VERIFICATION OF A MODEL OF HUMAN ABSENCE SEIZURES. *R. Vondříčková, L. Velišek, M. Pohl, H. Kubová, P. Mareš*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Rhythmic metrazol activity (RMA), i.e. the spike-and-wave rhythm induced by low doses of pentamethylenetetrazole (PTZ), represents an adequate model of human absence seizures (1). The action of antiabsence drugs was tested in this model. Adult rats were implanted with cortical electrodes and after one week of rest the recordings started. RMA was elicited by an i.p. injection of PTZ in a dose of 40 or 35 mg/kg. EEG activity was recorded for 30 min after the injection. The latency of the first RMA and the first generalized RMA were measured. The number, total and average duration of RMA episodes were counted between the 10th and 15th as well as between the 20th and 25th minute after PTZ administration. Rats were pretreated i.p. with ethosuximide (31, 25, 62.5, or 125 mg/kg), clonazepam (0.02 or 0.1 mg/kg) or valproate (200, 300 or 400 mg/kg) 10 min before PTZ. All three drugs exhibited a dose-dependent effect on the incidence and duration of RMA, the results with high doses were statistically significant.

*J. Schickerová R., Mareš P., Trojan S.:* *Activ. nerv. super.* 31: 16-20, 1989.

AUTO AND HETEROHYPNOSIS INFLUENCE UPON CUTANEOUS PAIN THRESHOLD. *P. Hájek, B. Jakoubek<sup>1</sup>, T. Radil<sup>1</sup>*, Medical Polyclinic, Litoměřice and <sup>1</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

A gradual increase in cutaneous pain threshold (c.p.t.) was found previously in healthy subjects and patients with atopic eczema during repeated hypnotic sessions. Repeated sessions without specific suggestions were ineffective. It follows that the effect observed was not caused by mere hypnosis but by specific suggestions during it. Repeated autohypnosis induced the same increase of c.p.t. as heterohypnosis. Autohypnosis was performed regularly after the end of the therapeutic sessions. It maintained the elevated level of c.p.t. or even increased it. The analgesis effect outlasted the hypnotic sessions by several months. It could, however, be suddenly reduced by appropriate hypnotic suggestion. It seems probable that the endogenous analgesic systems (irrespective of whether they are of opioid or nonopioid nature) are being engaged in the effects described by some hitherto unknown mechanism.

## 9. Experimental Neurophysiology

**HIERARCHIC ORGANIZATION OF THE VOCABULARY OF NEURONAL BURSTING ACTIVITY.** *D. Svorad*, Institute of Molecular Physiology and Genetics, Slovak Academy of Sciences, Bratislava.

Chaos in the behaviour of neuronal bursting activity (NBA) shows statistically defined principles in its organization. This study was aimed to prove the hypothesis that NBA is a hierarchic system (S). a) Construction of a NBA S. NBA is an object which can be considered to be a whole and simultaneously something consisting of many connected building parts (interimpulse intervals), i.e. NBA is a S. b) Definition of a NBA S. A NBA S can be defined by its vocabulary. Its organization is governed by several principles, e.g. Zipf law, Yule-Herdan-Mandelbrot law, etc. (4,1). c) Identification of hierarchy (3) in a NBA S. A NBA hierarchy can be described by the Waring-Herdan distribution (2).  
1. *Egghe L., Rousseau R.: J. Inf. Sci. 12: 193-197, 1986.*

2. *Herdan G.: The advance theory of language as choice and chance. Springer, Berlin, 1966.*

3. *Kennard C., Swash M. (eds): Hierarchies in Neurology. A Reappraisal of a Jacksonian Concept. Springer, Berlin, 1989.*

4. *Svorad D.: Activ. nerv. super. 26: 163-165, 1984.*

**FUNCTIONAL RECOVERY AFTER TRANSPLANTATION OF PRECULTURED GLIAL CELLS AND B-FGF TREATMENT.** *V. Valoušková, P. Kessler<sup>1</sup>, C.W. Cotman<sup>1</sup>*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague and <sup>1</sup>Department of Psychobiology, UCI, Irvine, USA.

Bilateral lesions of the sensory-motor cortex of adult rats produce a spatial deficit in the Morris water maze task. A unilateral homotopic transplant of embryonic tissue can ameliorate the behavioural impairment of this task. In the present study we compared the effects of embryonic neural transplants (ED 14), cultured astrocytes and purified neurotrophic factor (b-FGF) on the functional recovery in order to determinate the requirements of the injured system for behavioural recovery. The rats were tested in the Morris water maze for 14 days, 2, 4, 6 and 8 months after surgery. Transplants of purified astrocytes were shown to accelerate the long-term recovery in the spatial task in a similar manner as solid foetal cortical grafts. The mechanism of influence of b/FGF which slow down the recovery of spatial deficit was different from that of cultured glial cells.

**POSITIVE EFFECT OF TACRINE ON THE SYNTHESIS OF ACETYLCHOLINE IN BRAIN SLICES.** *S. Tuček, V. Doležal*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

We reported earlier that tacrine (TA, tetrahydroaminoacridine) increased the synthesis of acetylcholine (ACh) in cortical prisms incubated under "resting" conditions (3 mmol/l K<sup>+</sup>). We have now investigated the increased synthesis in more detail. While the synthesis of unlabelled (total) ACh was enhanced by TA during incubations at 3 mmol/l K<sup>+</sup>, the synthesis of (<sup>14</sup>C)ACh from (<sup>14</sup>C)choline added into the medium was considerably diminished. Hemicholinium-3 (HC-3), a specific inhibitor of the high affinity uptake of choline, 10 μmol/l inhibited the synthesis of ACh by more than 80 %, but the synthesis was augmented by 140 % when HC-3 (10 μmol/l) and TA (100 μmol/l) were present simultaneously). The effect of TA was not associated with its inhibitory action on cholinesterase. Apparently, TA stimulates the synthesis of ACh by increasing the utilization of choline from intracellular sources, making the synthesis of ACh less dependent on the supply of choline from the extracellular space.

**IMMUNOSTAINING FOR GLIAL FIBRILLARY ACID PROTEIN (GFAP) IN RAT SPINAL CORD DURING POSTNATAL DEVELOPMENT AND FOLLOWING X-IRRADIATION.** Z. Šimonová, E. Syková, I. Hájek, Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague.

GFAP is the major protein of intermediate filaments in differentiated mature astrocytes and in activated astrocytes during nervous tissue injury. Antibodies against GFAP were used to identify mature astrocytes, and activated astrocytes after X-irradiation, in 2 to 20-day-old rats. The lumbar region of the spinal cord in 1-day-old rats was x-irradiated. We found that the size of astrocytes in control rats and immunostaining increased gradually from 2 days to 10 days of age. In irradiated rats, astrogliosis and more dense immunostaining for GFAP than in control rats were already observed at 7 days of age. Our findings show that astrocytes in the rat spinal cord mature during the first 10 days postnatally. X-irradiation produces astrocyte proliferation and hypertrophy. Furthermore, our experiments with ion-selective microelectrodes revealed that the immature astrocytes, as well as activated astrocytes, are not able to ensure  $K^+$  and pH homeostasis in the spinal cord.

**EXTRACELLULAR SPACE VOLUME INCREASE IN THE SPINAL CORD OF THE RAT WITH EXPERIMENTAL AUTOIMMUNE ENCEPHALOMYELITIS.** J. Svoboda, E. Syková, P. Orkand<sup>1</sup>, C. Bernard<sup>2</sup>, Institute of Experimental Medicine, Prague, <sup>1</sup>Institute of Neurobiology, Puerto Rico and <sup>2</sup>Department of Psychology, La Trobe University, Melbourne.

Experimental encephalomyelitis (EAE) is an autoimmune disease widely used as an experimental model of multiple sclerosis (MS). Acute EAE in Lewis rats has been characterized by a breakdown of the blood-brain barrier, oedema, perivascular infiltration of inflammatory cells and astrocyte hypertrophy and proliferation – signs which, besides demyelination, may contribute to EAE manifestation. We studied the absolute changes in extracellular space volume (ECSV) in the spinal cord of rats with EAE and compared them with tissue inflammation and astrocyte immunostaining for GFAP. In early stages of the disease, i.e. 12-16 days after injection of myelin basic protein, ECSV increased reversibly from 20-25 % (control rats) to 40-50 % (EAE rats). The increase was found in the intermediate region and in the ventral horns concomitantly with inflammation and an increase in GFAP immunostaining. We suggest that the expansion of the ECSV may contribute to the clinical signs of EAE and may be important in MS.

**EXTRACELLULAR  $K^+$  AND pH CHANGES FOLLOWING X-IRRADIATION OF LUMBOSACRAL REGION OF THE NEONATAL RAT SPINAL CORD.** E. Syková, P. Jendelová, I. Hájek, Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague.

It has been suggested that glial cells buffer the activity-related extracellular  $K^+$  ( $[K^+]_e$ ) increase and extracellular pH ( $pH_e$ ) shifts. Since gliogenesis occurs during the first 10 days postnatally, the lumbosacral region of 1-day-old pups was X-irradiated (4000 r) to block gliogenesis. The stimulation-evoked  $[K^+]_e$  and  $pH_e$  changes were measured in the isolated spinal cord of control and irradiated pups using ion-selective microelectrodes. The enhanced  $[K^+]_e$  changes and the alkaline shifts were found in control 2 to 6-day-old pups, while in 10 to 14-day-old pups the  $[K^+]_e$  ceiling level decreased and the acid shift became dominant as is the case in adult rats. In irradiated pups the enhanced  $[K^+]_e$  changes and the alkaline shifts persisted even in 10 to 14-day-old pups. The alkaline shifts were enhanced by amiloride and STS, blocked by  $Mg^{2+}$  but not by  $Ba^{2+}$ . We conclude that glial cells play an important role in  $K^+$  and pH homeostasis in the neonatal rat spinal cord.

**DOPAMINERGIC D-1 RECEPTORS ARE NOT INVOLVED IN PRESYNAPTIC REGULATION OF ACETYLCHOLINE RELEASE FROM RABBIT STRIATUM.** *V. Doležal, C. Allgaier<sup>1</sup>, R. Jackisch<sup>1</sup>, G. Herting<sup>1</sup>*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague and <sup>1</sup>Department of Pharmacology, University of Freiburg, FRG.

In experiments on superfused slices of rabbit striatum, antagonists of D-1 (SCH 23390) and D-2 (domperidone) receptors had no influence on the release of acetylcholine (ACh) evoked by single electrical pulses and the D-1 agonist SKF 38393 was also without effect. D-2 agonist quinpirole decreased the release of ACh dose-dependently. Its inhibitory effect on the release of ACh was lesser in the presence of SCH 23390 but was not changed by D-1 agonist SKF 38393, which did not, however, influence the diminished potency of quinpirole in the presence of SCH 23390. The results confirm that the presynaptic dopaminergic inhibition of ACh release is mediated by D-2 receptors and indicate that SCH 23390, generally regarded as a specific D-1 antagonist, also binds to D-2 receptors at low ( $1 \mu\text{mol/l}$ ) concentrations.

**DIRECT ELECTRICAL STIMULATION OF THE NUCLEUS RETICULARIS GIGANTOCELLULARIS RESETS THE LICKING FREQUENCY GENERATOR.** *D. Megjrian, G. Brožek<sup>1</sup>, J. Bureš<sup>1</sup>*, University of Calgary, Faculty of Medicine, Alberta, Canada and <sup>1</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The nucleus reticularis gigantocellularis was unilaterally stimulated during a period of continuous licking (after 8 regular consecutive licks) of water in freely moving thirsty rats implanted with bipolar stimulating electrodes (coordinates AP 10, L 1, V 10 according to Fířková and Mařála). This stimulation (10 rectangular pulses, 0.1 ms, 100 Hz) of adequate intensity (25-50  $\mu\text{A}$ ) caused a phase shift of licks following stimulus delivery, but it did not change the licking frequency. The current, necessary for the licking pacemaker reset was lowest, when the stimulation was delivered in the middle of the inter-lick interval and highest during the tongue protrusion period. The stimulation of higher electrical intensity ( $> 100 \mu\text{A}$ ) suppressed spontaneous licking for several seconds.

**PENETRATION AND RETURN OF SPREADING DEPRESSION WAVES BETWEEN NEOCORTEX AND CAUDATE NUCLEUS.** *L.V. Vinogradova, V.I. Koroleva, J. Bureš<sup>1</sup>*, Institute of Higher Nervous Activity and Neurophysiology, USSR Academy of Sciences, Moscow and <sup>1</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Spreading depression (SD) waves elicited from the parietooccipital cortex of anaesthetized rats penetrated in 40 % of experiments through the temporal lobe structures (amygdala) into the caudate nucleus. Almost 70 % of these SD waves did not terminate in the caudate but returned to the cortex. Longer cortico/caudate than caudate-cortical conduction times suggest that SD enters and leaves the caudate by two different routes. SD waves elicited by KCl microinjection into the caudate reached the frontal and parieto-occipital electrodes with latencies indicating that the transit point is 5 mm closer to the rostral than to the caudal electrode. The region best satisfying this condition corresponds to the rostral claustrum. The directionally biased SD conduction through the transit zone provides a re-entry path for cortico-caudate-cortical SD propagation.

**VOLTAMMETRIC MONITORING OF POTASSIUM-STIMULATED CATECHOLAMINE CHANGES IN STRIATAL SYNAPTOSOMAL PREPARATIONS AND IN THE CORPUS STRIATUM IN RATS.** J. Pavlídek<sup>1</sup>, J. Orlický<sup>2</sup>, K. Murgas<sup>3</sup>, <sup>1</sup>Institute of Normal and Pathological Physiology, <sup>2</sup>Institute of Molecular Physiology and Genetics and <sup>3</sup>Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.

The voltametric technique was used to compare the effects of K<sup>+</sup>-induced depolarization on catecholamine (CA) levels in synaptosomal preparations of the corpus striatum *in vitro* with those in the corpus striatum of anaesthetized rats *in vivo*. *In vitro*, a 30 s lasting increase of CA-oxidation current could be observed after the addition of KCl to dopamine loaded synaptosomes into the medium with an elevated K<sup>+</sup> concentration (90 mmol.l<sup>-1</sup>) and a decreased concentrations of Na<sup>+</sup> (75 mmol.l<sup>-1</sup>) and Ca<sup>2+</sup> (0.75 mmol.l<sup>-1</sup>). In *in vivo* experiments, a microinjection of KCl (3 µl of 0.5 mol.l<sup>-1</sup> KCl) resulted in an increase of the CA-oxidation current (to severalfold of the control values) followed by a decrease or even total disappearance, with a gradual return to control values. K<sup>+</sup>-depolarization under conditions of depletion of extracellular calcium by EGTA confirmed the key role of calcium in the release of CA transmitters as well as in processes regulating the uptake and metabolism of these transmitters.

**INFLUENCE OF ANTIEPILEPTIC DRUGS CARBAMAZEPINE AND PHENYTOIN ON SPONTANEOUS UNIT ACTIVITY OF CEREBELLAR CORTEX NEURONES.** J. Buřitová, S. Hrabětová, J. Hrabě<sup>1</sup>, V. Pavlík<sup>2</sup>, P. Mareš<sup>2</sup>, Department of Physiology, Third Medical Faculty, <sup>1</sup>Department of Mathematics, Faculty of Science, Charles University and <sup>2</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Unit activity was measured in cerebellar cortex neurones in 37 males Wistar rats. Carbamazepin (CBZ, 25 mg/kg i.p.) was administered to 13 animals, phenytoin (PTH, 60 mg/kg i.p.) to 14 animals and 10 animals received the solvents of both drugs (i.e. propylenglykol : ethanol : water = 5 : 2 : 3). The activity of 50 neurones was recorded before and 73 neurones after the injection. The activity of 30 neurones was recorded both before and after administration of the drugs. Statistical processing was employed to assess the distribution of unit activity frequencies and the changes induced by the above mentioned drugs. It was shown that both CBZ and PHT significantly reduce the unit activity frequency, but the PHT effect is less expressed. A correlation between the frequency changes and the original uninfluenced frequencies was also studied.

**OPIOID RECEPTORS IN THE RAT SPINAL CORD AFTER LONGLASTING DEAFFERENTATION.** I. Hájek, J. Buřitová<sup>1</sup>, N. Kříž<sup>1</sup>, Institute of Experimental Medicine, Czechoslovak Academy of Sciences and <sup>1</sup>Department of Physiology, Third Medical Faculty, Charles University, Prague.

*In vitro* binding of specific opioid ligands to respective sites in the membrane fraction and the contribution of individual receptor classes (mu, delta, kappa) was studied in rats after longlasting (about 2 years) deafferentation of spinal dorsal roots in the cervical (Cm<sub>2,8</sub>) or thoracic (Th<sub>1,4</sub>) regions. This procedure leads to autotomy or scratching of the skin on the operated side. The total number of receptors in the cervical and thoracic spinal cord increased to more than twofold values in comparison with intact controls of the same age. This increase mainly concerned a rise in the cervical region in the number of free receptors, while both free and saturated receptors were increased in the thoracic region. On the deafferented side, a decrease in receptor selectivity, was found especially in the delta and kappa types. The increase in number of opioid receptors and their decreased saturation by endogenous ligands might be responsible for the development of longlasting pain syndromes after deafferentation.

**PERIPHERAL RESTRICTION OF THE PREFERRED FOREPAW IS MORE EFFECTIVE THAN ITS CENTRAL BLOCKADE DURING REVERSAL OF 'HANDEDNESS' IN RATS.** *E.I. Mikhayeva, J. Bures<sup>1</sup>*, Institute of Higher Nervous Activity and Neurophysiology, USSR Academy of Sciences, Moscow and <sup>1</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Reversal of forepaw preference was studied in rats under conditions of peripheral (bracelet) or central (inactivation of contralateral motor cortex and caudate nucleus by intracerebral injection of tetrodotoxin) blockade of the preferred limb. A persisting change of the forepaw preferred when reaching for food into a tubular feeder was achieved on the average after 91 (n=27) and 211 (n=26) reaches, respectively. A forced change of "handedness" during the tubular feeder task was accompanied by a similar change of preference during the shelf task in only 17 % rats. It is concluded that relatively independent lateralization of the two tasks reflects a differential contribution of the corticospinal, rubrospinal and ventromedial motor systems. Reversal training proceeds more rapidly with peripheral blockade which establishes not only the new preference but also inhibits the original one.

**IPSILATERAL INTERACTION OF CORTICAL AND SUBCORTICAL CENTRES IN THE ACQUISITION OF CONDITIONED TASTE AVERSION IN RATS.** *M. Gallo, J. Bures<sup>1</sup>*, Department of Psychology, University of Granada and <sup>1</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The possibility of lateralizing the neural circuits mediating conditioned taste aversion (CTA) has been examined by combination of functional hemidecortication by cortical spreading depression (CSD) and unilateral tetrodotoxin (TTX) injection into the parabrachial nucleus (PB). Rats drinking saccharin (CS) during CSD in the right hemisphere and receiving unilateral PB TTX (10 ng) shortly after an i.p. injection of LiCl (US) formed CTA when CSD and TTX were applied to the same hemisphere but not when applied to different hemispheres. Rats with an intact brain drinking saccharin and receiving unilateral TTX overlapping with LiCl administration learned a weak CTA the retrieval of which was disrupted by either ipsilateral or contralateral CSD during retention testing. It is concluded that CTA acquisition requires cooperation of ipsilateral cortical and subcortical centres but that the formation of an unilateral subcortical CTA engram does not warrant lateralization of the retrieval process.

**PARTIAL DISRUPTION OF PASSIVE AVOIDANCE LEARNING BY POSTACQUISITION INJECTION OF TETRODOTOXIN INTO THE PARABRACHIAL NUCLEI OF RATS.** *G. Tassoni, C. Bucherelli, J. Bures<sup>1</sup>*, Institute of Physiology, University of Florence and <sup>1</sup>Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The recent discovery that post-trial functional blockade of the parabrachial nuclei by intracerebral injection of 10 ng tetrodotoxin (TTX) disrupts the acquisition of conditioned taste aversion (CTA) has prompted attempts to ascertain the role of this structure in other types of inhibitory learning. Rats trained in a step-through avoidance task and receiving bilateral parabrachial TTX (2x10 ng) immediately after the acquisition trial displayed significantly weakened avoidance of the dark compartment two days later. The amnesic effect was significant when the acquisition - TTX delay had been prolonged to 24 but not to 48 hours. CTA is disrupted by similar TTX dosage up to 4 days after acquisition. It is concluded that the reversible blockade of parabrachial nuclei and of the adjacent reticular formation disrupts passive avoidance to a lesser degree and for a shorter time than CTA.

## 10. Metabolism

### CHANGES OF SERUM CHOLESTEROL AND LOW DENSITY LIPOPROTEIN METABOLISM IN GUINEA-PIGS WITH NUTRITIONALLY INDUCED VITAMIN C HYPOSATURATION.

*M. Jurčovičová, P. Bobek, E. Ginter, Research Institute of Human Nutrition, Bratislava.*

The study was performed on guinea-pigs after 15 weeks of chronic vitamin C deficiency. The control group, saturated with vitamin C, received a diet supplemented with 0.5 % of vitamin C and the experimental, vitamin C deficient group was fed with the same diet with only a maintenance dose of this vitamin to ensure normal growth. The negative influence of vitamin C deficiency on the increase of serum cholesterol levels was also manifested by its elevation in all the isolated lipoprotein fractions, mainly in atherogenic low density lipoproteins (LDL). All the calculated values of atherogenic indexes were significantly increased due to vitamin C deficiency. Kinetic studies *in vivo* with radionuclide  $^{125}\text{I}$  labelled native and modified homologous LDL showed that elevated serum cholesterol concentrations in vitamin C deficient animals are a consequence of decreased receptor mediated catabolism of atherogenic LDL.

### FISH OIL DECREASES SERUM LIPIDS AND MODIFIES THE LIPOPROTEIN PROFILE IN HEREDITARY HYPERTRIGLYCERIDAEMIC (HTG) RATS.

*M. Hromadová, E. Šeböková, I. Klímeš, M. Ficková, A. Vrána<sup>1</sup>, Institute of Experimental Endocrinology, Slovak Academy of Sciences and <sup>1</sup>Institute of Clinical and Experimental Medicine, Prague.*

The aim of the present work was to extend our knowledge on the hypolipemic effect of fish oil (FO) employing a model representing hypertriglyceridemia (HTG) conditioned genetically or evoked by a high saccharose diet (HS) in a line of rats with hereditary hypertriglyceridemia. In animals with HTG the increased TG in the serum and in the VLDL and LDL fractions of lipoprotein remained essentially unchanged after feeding the HS diet. However, supplementation of that diet with FO for 21 days significantly decreased serum ( $p < 0.05$ ) and VLDL - TG ( $p < 0.025$ ) levels. No differences in serum cholesterol or its distribution into lipoproteins were found between HTG and control animals. The administration of a diet with FO depressed total, LDL and HDL cholesterol ( $p < 0.05$ ). It may thus be concluded that 1. Changes in the lipoprotein profile conditioned by a genetic disorder of lipid metabolism were uninfluenced by the HS diet and 2. The hypolipidemic effect of FO manifests itself by lowering serum lipids in HTG rats and modifies the lipoprotein profile.

### FISH OIL INDUCES CHANGES IN LIPID COMPOSITION AND LIVER LDL RECEPTOR ACTIVITY IN HEREDITARY HYPERTRIGLYCERIDAEMIC (HTG) RATS.

*S. Šeböková, M. Hromadová, I. Klímeš, P. Bohov<sup>1</sup>, M. Hermann<sup>2</sup>, M. Ficková, Institute of Experimental Endocrinology and <sup>1</sup>Institute of Experimental Pharmacology, Slovak Academy of Sciences, Bratislava and <sup>2</sup>Institute of Medical Chemistry, Vienna University, Vienna, Austria.*

To elucidate further the mechanism of hypocholesterolemic action of n-3 PUFA under conditions of endogenous and/or exogenous HTG, the effects of fish oil (FO) on the cholesterol (CH) content, FA composition of serum and liver lipids and liver LDL receptor activity were investigated. Male hereditary HTG rats were fed a high sucrose (HS) diet (63 cal%) or basal chow alone or in combination with FO (30 wt% n-3 FA) for 21 days. The parameters were compared to control (C) normo-TG rats fed basal chow. In HTG rats fed the basal as well as HS diet, dietary FO decreased serum total, free and esterified CH content. The CH lowering effect of FO, also found in the livers of rats fed either HS ( $p < 0.05$ ) or the basal diet ( $p < 0.025$ ), was caused by a decrease of CH-esters only. Furthermore, FO raised the LDL binding capacity of liver membranes in both groups (basal: 1.8 fold,  $p < 0.001$ ; HS: 1.6 fold,  $p < 0.005$ ). In summary, 1. the hypo-CH action; of FO is equal in both the HTG as well as in the C rats, 2. and is not accompanied by an accumulation of CH in the liver, and 3. it may be in part mediated *via* an increased hepatic LDL receptor activity.

**FISH OIL IMPROVES THE SUCROSE DIET INDUCED DECREASE OF RAT MUSCLE GLYCOGEN SYNTHASE RESPONSE TO INSULIN ONLY WHEN COMBINED WITH ACIPIMOX (OLBETAM).** I. Klimeš, A. Minchenko<sup>1</sup>, E. Šebáková, M. Hromádová, P. Bohov<sup>2</sup>, A. Vrána<sup>3</sup>, Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava, <sup>1</sup>Institute of Endocrinology, Kiev, Ukraine, <sup>2</sup>Institute of Experimental Pharmacology, Slovak Academy of Sciences, Bratislava and <sup>3</sup>Institute of Clinical and Experimental Medicine, Prague.

To assess whether inhibition of FFA oxidation would not help to fully normalize the impaired insulin (I) action (A) by a sucrose diet (S), we studied the effect of acipimox (ACI) treatment of S fed rats on I stimulated glycogen synthase (GS) activity. S feeding raised fasting serum VLDL-TG, which could be equally suppressed by ACI, fish oil (FO) and/or FO+ACI. In S fed rats without and/or with additional treatment with FO or ACI, glycaemia remained unchanged due to an increase of serum I. Except for the FO+ACI group, fasting I and stimulation of GS activity in muscles by *I in vitro* showed a S-induced resistance to IA which was not normalized by FO and ACI alone. Finally, GS correlated negatively with the muscle TG content. Thus, 1. S-induced impairment of IA on the muscle GS is not abolished by suppression of TG production or; FFA oxidation alone, it requires both. 2. The effect of FO+ACI on GS provides new evidence for linking fat production and oxidation with non-oxidative glucose metabolism.

**OXYGEN-FREE RADICALS IN THE BLOOD OF PATIENTS WITH HYPERLIPOPROTEINAEMIA.** V. Soška, A. Zecumeister<sup>1</sup>, J. Siegelová<sup>2</sup>, A. Lojek<sup>3</sup>, Department of Clinical Biochemistry, Faculty Hospital, <sup>1</sup>Department of Anatomy and <sup>2</sup>Third Department of Medicine, Medical Faculty, and <sup>3</sup>Institute of Biophysics, Brno.

Oxygen-free radicals (OFR) are supposed to be a risk factor of atherosclerosis, as plasma lipids can undergo oxidation thus producing lipoperoxides, which are more atherogenic. We examined the production of OFR in 24 patients with hyperlipoproteinaemia (HLP) and the results were compared with 12 healthy subjects (HS). The method of luminol-dependent chemiluminescence was used (luminometer LKB 1251). Spontaneous production of OFR and stimulated production evoked by the stimulation of phagocytosis were measured in both groups. A significant decrease ( $p < 0.01$ ) of OFR was found in HLP. This finding can be evoked by the decrease of phagocytar activity of phagocytes, which are the main producers of OFR in the blood. It can be caused by damaging phagocytes due to the accumulation of LDL particles in HLP *via* acetyl-LDL receptors.

**UTILIZATION OF PROTEINS OF DIFFERENT QUALITY UNDER CONDITIONS OF HIGH FAT DIETS IN ANIMAL EXPERIMENT.** M. Krajčovičová-Kudláčková, Research Institute of Human Nutrition, Bratislava.

A diet with a optimum composition of a qualitatively different source of proteins (milk casein, wheat gluten) and subsequently high fat diets, HF 25 and HF 37.5 (25 %, 37.5 % fats) was given to 75-day-old male rats. The net protein ratio (NPR), the net protein utilization (NPU) and the process of gluconeogenesis in the liver (activity of PEPCK) was assessed. Under optimal nutritional conditions, the values NPR, NPU were significantly reduced for gluten (low quality protein, E/N=0.30), as compared with casein (E/N=0.79). The negative effect of the high fat diet (HF 37.5, optimal fat content 15 %) on NPU was more marked for gluten-reduction by 52 % (for casein by 31 %). NPR was also induced on the HF 25 diet for gluten (by 23 %); the inferior protein quality of gluten influenced the extent of proteosynthesis for the rapidly growing organism which is fully ensured by the superior quality of casein (a slightly unbalanced fat diet). The process of gluconeogenesis is more marked in the case of gluten (a greater supply of glucoplastic amino acids) and in correlation to NPU, NPR, this suggests the breakdown of body proteins.

**REDUCTION OF FAT STORES AND PLASMA TRIGLYCERIDES IN GENETICALLY HYPERTENSIVE NON-OBESE FEMALE RATS OF THE KOLETSKY TYPE BY BROMOCRIPTINE AND LISURIDE.** V. Golda, Institute of Experimental Neurosurgery, Hradec Králové.

Insulin is considered as a powerful lipogenic factor in the presence of prolactin (1). Insulin loses this effect when prolactin is reduced by a dopaminergic agonist such as bromocriptine (1). We verified latter statement in genetically hypertensive non-obese plasma prolactin and plasma insulin levels in the nonfasted state (unpublished data). We administered bromocriptine (10 + 10 mg/kg b.w.) and lisuride (0.4 + 0.4 mg/kg b.w.) twice a day (at 06.00 h and at 14.00 h), i.p. for 11 days. Neither drug influenced the final body weight, pellet and water intake. Both drugs caused a decrease in plasma triglycerides and in retroperitoneal fat pads; plasma cholesterol, lipoproteins and HDL were not significantly influenced.

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**MECHANISM OF THE INHIBITORY EFFECT OF VANADATE ON SUCCINYL-COA SYNTHETASE.** J. Křivánek, L. Nováková, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

We have recently shown that vanadate inhibits both the phosphorylation of the alpha-subunit of succinyl-CoA synthetase (SCS) and the activity of this enzyme (1,2). In this communication we report on some further aspects of the vanadate inhibition of SCS solubilized from the rat brain by Lubrol. Inorganic phosphate ( $P_i$ ), which has an analogous structure to vanadate, inhibited both phosphorylation and activity of SCS but to a much lesser extent than vanadate. Fifty per cent inhibition of phosphorylation was achieved at  $10^{-3}$  mol.l<sup>-1</sup> as compared to  $5.10^{-5}$  mol.l<sup>-1</sup> vanadate. The respective values for inhibition of SCS activity were  $3.10^{-3}$  mol.l<sup>-1</sup> and  $1.10^{-5}$  mol.l<sup>-1</sup>. In contrast to  $P_i$ , however, the vanadate effect was dependent on the concentration of succinate. In concentrations below 25 mmol.l<sup>-1</sup> no effect of  $1.10^{-5}$  mol.l<sup>-1</sup> vanadate could be observed. It appears that the effect of vanadate is more complex and cannot be accounted for merely by the product inhibition as in the case of  $P_i$ .

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**OXIDATION OF PALMITOYL-CARNITINE, PROPIONOYL-CARNITINE AND PYRUVATE BY LIVER MITOCHONDRIA AT EARLY STAGES OF LIVER REGENERATION.** V. Bláha, J. Šimek, Department of Physiology, Faculty of Medicine, Charles University, Hradec Králové.

The aim of this study was to evaluate the utilization of carbohydrates and lipids in the rat liver, regenerating after partial hepatectomy (PH). Laparotomy or PH (removal of 65-70 % of the liver tissue) was performed in rats. The animals were sacrificed by decapitation 3, 6, 18, 21, 24 and 48 h after surgery. The oxidation of palmitoylcarnitine, propionylcarnitine (lipid substrates) and pyruvate (final catabolite of glycolysis) was evaluated by measuring oxygen consumption of isolated liver mitochondria. 1. The oxygen consumption in the presence of palmitoylcarnitine decreased 3 h after surgery, with a return to initial values 6 h after PH. Another significant drop was noted 18 and 24 h after surgery. 2. After propionylcarnitine administration the mitochondrial oxygen consumption did not decrease below initial values. On the contrary, we recorded a significant increase 6 and 24 h after surgery as compared with non-operated controls. 3. The oxygen consumption decreased 3 h after PH in the presence of pyruvate. Six hours after surgery it increased to control values, then again decreased until 48 h after PH. The data obtained showed the importance of lipids as substrates at early stages of liver regeneration. During the first 48 h after PH increased the oxidation of lipid substrates in the regenerating rat liver. The oxygen consumption by pyruvate utilization decreased.

**EFFECT OF PROPYLTHIOURACIL ON LIVER REGENERATION AFTER PARTIAL HEPATECTOMY IN RATS.** Z. Červinková, J. Šimek, I. Hubáčková, Department of Physiology, Faculty of Medicine, Charles University, Hradec Králové.

In our experiments, the effect of propylthiouracil (PTU) on growth activity of the intact liver and liver regenerating after partial hepatectomy (PH) was studied. PTU (Propycil fy Kali Chemie, West Germany) was dissolved in drinking water ( $1.0 \text{ g.l}^{-1}$ ) and this solution was the only source of liquids for the experimental animals from 3 days before PH till the end of the experiment. In rats treated with PTU, a statistically significant decrease of liver DNA synthesis and mitotic activity of hepatocytes was found after PH at all the intervals studied. Part of this effect can be attributed to the spontaneous, lower intake of food by rats treated with PTU, as shown by comparison with pair-fed rats. The inhibitory effect of PTU on DNA synthesis in intact and regenerating liver was also observed in thyroidectomized rats even when thyroid hormones were administered. Our results demonstrate that the inhibitory effect of PTU on growth activity of the liver is mediated above all by a direct action of PTU on the liver tissue.

**INFLUENCE OF PENTOBARBITAL AND KETAMIN ON SUBSTRATE UTILIZATION DURING SHORT-TIME ANAESTHESIA IN THE RAT.** Z. Wilhelm, Department of Physiology, Medical Faculty, Masaryk University, Brno.

The aim of this study was to assess whether the opposite effects of two anaesthetics on respiration, inhibition by pentobarbital (P) and stimulation by ketamin (K), reflect different utilization of carbohydrates, fats and proteins and whether they are influenced by starvation. The utilization of substrates was determined during the first 30 min of anaesthesia assessed by  $\text{O}_2$  consumption,  $\text{CO}_2$  production and urinary nitrogen excretion. The rats were divided into 3 groups, each comprising 30 animals: I. with free access to food, II. fasting for 1 day, III. fasting for 3 days. Ten animals in each group received an i.p. injection of P (50 mg/kg), K (130 mg/kg) and 0.9 % NaCl as control, respectively. No significant differences were found in the metabolism of carbohydrates, fats or proteins of rats with a normal food supply. During fasting both anaesthetics potentiate the decrease of carbohydrate utilization but the effects on fat metabolism are opposite: K stimulates and P inhibits the utilization of fats. The differences in urinary nitrogen excretion due to fasting were insignificant.

**THE SUPPRESSIVE EFFECT OF TRYPTOPHAN ON FOOD INTAKE IN BROILERS DEPENDS UPON THE TIMING OF ITS ADMINISTRATION.** E. Baranyiová, Department of Physiology, University of Veterinary Sciences, Brno.

The effect of l-tryptophan (l-trp) on food intake (FI) was studied in Hybro broilers. Birds of 4 groups, 12 individuals each, were given single i.p.  $100 \text{ mg.kg}^{-1}$  doses of l-trp (Sigma, USA) at 6.30 h (on days 3, 8, 22 and 26) or at 9.30 h (on days 2, 6, 10, 20 and 24). Four groups were given saline. The suppressive effect of l-trp on FI after administration at 6.30 h was only noted on day 3 ( $P < 0.02$ ). However, in birds older than 2 days, FI was lower as against controls within 30 min of l-trp injection at 9.30 h: in broilers aged 6 ( $P < 0.01$ ), 10 ( $P < 0.01$ ), 20 ( $P < 0.05$ ) and 24 ( $P < 0.01$ ) days. These data show that l-trp suppresses feeding in birds older than 2 days. This effect was seen after the 6.30 h administration only in 3-day-old birds in which the FI pattern (1) (10-15 % of daily consumption eaten during the first morning hour (6 to 7h)) had not yet developed. In older birds, l-trp decreased the FI only when it was given after the large morning consumption, i.e. at 9.30 h. These data reveal that diurnal rhythms and FI pattern of any species must be considered when substances affecting their food intake are investigated.

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THE TIME COURSE OF THYMIDINE RELEASE FROM DEOXYRIBONUCLEIC ACID INTO MOUSE PLASMA AFTER WHOLE BODY GAMMA IRRADIATION. *J. Boháčěk, B. Hošek, J. Šituková*, Institute of Biophysics, Czechoslovak Academy of Sciences, Brno.

The thymidine levels in the plasma of irradiated mice were estimated as indicators of radiation damage at various intervals after irradiation. Hybrid mice (CBAXB1)F<sub>1</sub> were exposed to whole body irradiation with doses ranging from 25 to 400 cGy. Samples of blood taken from individual mice 2-6 h after the irradiation were centrifuged, deproteinized and injected into a chromatographic column. The levels of thymidine were found to be dose-dependent. For doses of up to 100 cGy the highest levels of thymidine were found around the 4th hour after the irradiation, while with doses of over 100 cGy the maximum of thymidine excretion was shifted to the 5th hour interval after the irradiation. The results confirmed the validity of thymidine as a biochemical indicator of radiation damage in mice.

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ADAPTATION OF TISSUES TO THE RADIOPROTECTIVE EFFECT OF HYPOXIA. *A. Váček, T. Tačev*, Institute of Biophysics, Czechoslovak Academy of Sciences and Research Institute of Clinical and Experimental Oncology, Brno.

The inhalation of 10 % oxygen decreases the oxygen tension in murine tissues by more than 50 % of the initial values and during irradiation with gamma rays it results in a radioprotective effect of hypoxia against the damage of tissues by ionizing radiation. The aim of the present study was to determine the relation of the intensity of radioprotection of mice on the duration of the exposure of the animals to hypoxia (10 % O<sub>2</sub>). It was observed that when the tissue hypoxia lasted more than 45 min before irradiation, its radioprotective effect decreased, despite the fact that the drop of pO<sub>2</sub> in the tissues was not changed. The decrease of the radioprotective effect of prolonged hypoxia was observed in GIT, haemopoietic tissues, protection of the skin against depilation and lethal effects of irradiation. The radioprotective effect of hypoxia in mice is short-termed, and a drop of its efficiency as a result of adaptation of tissues to hypoxia is discussed.

INTERACTION OF ANTRAQUINONES WITH Na<sup>+</sup>/K<sup>+</sup>-ATPase. *V. Boháčková, P. Dočolomanský, D. Hagarová, A. Breier*, Institute of Molecular Physiology and Genetics, Slovak Academy of Sciences, Bratislava.

Antraquinones such as Cibacron Blue F3GA (CB) and Remazol Brilliant Blue R (RB) were able to interact with the ATP-binding moiety of Na<sup>+</sup>/K<sup>+</sup>-ATPase (1). In this paper the interaction between 8 derivatives of anthraquinones and the Na<sup>+</sup>/K<sup>+</sup>-ATPase from the dog kidney were studied. All applied anthraquinones were able to inhibit the enzyme, but they were different in their inhibitory potency. The inhibitory action of these substances strongly increased with the decrease of the value of their apparent acidobasic dissociation constants. The presence of the triazine moiety in the substance molecule was the second important factor promoting its inhibitory action. However, exchange of the triazine moiety for a polar aliphatic chain in RB secured the inhibitory effect which was comparable with the effect of the highly potent CB.

*L. Ďurišová V., Vrbanová A., Ziegelhöffer A., Breier A.: Gen. Physiol. Biophys. 9: 519 - 528, 1990.*

**MORPHOMETRICAL MEASUREMENTS OF TUBULAR CELLS OF THE CHICK EMBRYONIC KIDNEY.** *L. Kubínová, Z. Zemanová, L. Antalíková<sup>1</sup>, I. Krekule*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague and <sup>1</sup>Research Institute of Animal Production, Prague-Uhřetíněves.

The nature of the process of tubular cystic dilation in the chick embryonic kidneys is still unknown. However, it can be elucidated by the measurement of morphological characteristics of tubular cells. In the present study, we introduced procedures for the estimation of mean tubular cell volume and mean cell dimensions in transverse sections of the tubule. The mean cell volume was estimated by the stereological method, so-called dissector (1), i.e. from measurements of two parallel sections of the kidney. The mean cell thickness and width were measured by a digitizer. The reliability of the presented methods and possibilities of the measurement of other tubular characteristics are discussed.

*I. Sterio D.C.: J. Microsc. 134: 127-136, 1984.*

**PATTERN OF RNA SYNTHESIS IN BOVINE OOCYTES AND CUMULUS CELLS DURING *IN VITRO* MATURATION.** *L.V. Kozikova, A.V. Sirotkin, J. Pivko*, International Laboratory, Research Institute of Animal Production, Nitra.

The pattern of RNA synthesis in individual bovine oocyte-cumulus complexes (OOC) were isolated from ovarian antral follicles (1-8 mm) and analysed after 1, 6, 12, 18 and 24 h of incubation in a medium TC-199 supplemented by 10 % fetal calf serum. OCC were classified in 3 classes based on the state of the cumulus layers: compact multilayered cumulus (1st class), friable multilayered cumulus (2nd class) and irregularly expanded or absent cumulus cells layer (3rd class). Total RNA synthesis was assessed by autoradiography, using <sup>3</sup>H-uridin (specific activity 740 G Bq/mmol, IRPAR, Czechoslovakia) and photoemulsion Ilford K-5, OCC were prepared by Tarkowsky's method. The great heterogeneity of the oocyte population was shown according to their capacity of synthesize RNA at the start of maturation: the content of silver grains per oocyte varied from 3 000 to 0. The incorporation rate of uridine during this time was significantly greater in oocytes in classes 1 and 2 than that in the cells of 3rd class. After 18-24 hours incubation (at metaphase II) this process disappeared in oocytes of all classes. Very intensive RNA synthesis was noted in cumulus cells. No differences in <sup>3</sup>H-uridine incorporation by cumulus cells of various OCC classes and stages of maturation were observed. The interrelationship between the pattern of RNA synthesis in oocytes and cumulus cells in the course of their maturation were discussed.

## 11. Physiology of Respiration and Sports

**pO<sub>2</sub> CHANGES IN THE SPINAL CORD BEFORE, DURING AND AFTER SPINAL CORD COMPRESSION IN DOGS.** *A. Horňáková, T. Badonič, J. Maršala*, Institute of Neurobiology, Slovak Academy of Sciences, Košice.

pO<sub>2</sub> values were measured in the lumbar spinal cord of dogs using membrane oxygen electrode during 5 min spinal cord compression followed by 30 min recirculation. Spinal cord compression was performed by a 200 g weight. The partial oxygen pressure monitored in the spinal cord was 5.8 kPa before compression, 0.2 kPa during the compression and 6.9 kPa immediately after compression. After 30 min the values of pO<sub>2</sub> were stabilized at the physiological level (5.8 kPa). The morphological features of neurones were also studied. Some of them revealed signs of argyrophilia and a light space in the pericellular region. The neuronal shape was also altered.

**BREATHING OF AWAKE RATS WITH COMPLETE DIAPHRAGM PARALYSIS.** *J. Nacházel, F. Paleček*, Institute of Pathophysiology, Second Medical School, Charles University, Prague.

Bilateral phrenicotomy (PX) in anaesthetized rats results in bradypnoea and hypoventilation (2). The aim of this study was to ascertain the effect of PX on ventilation in awake rats. Seven male Wistar rats were subjected to PX, 5 control rats to a sham operation. Tidal volume (VT) and breathing frequency (f) were measured in a body plethysmograph for unanaesthetized animals (1) during a control period and 7, 12 and 15 days after the operation. No changes of ventilation were observed in the control rats throughout the experiment. In rats with diaphragm paralysis f increased by 46 % and VT decreased by 35 %. Minute ventilation did not differ from that in the control rats. We conclude that the awake rats with complete diaphragm paralysis are, in contrast to anaesthetized rats, tachypnoeic.

1. *Bartlett D., Tenney S.M.*: Resp. Physiol. 10: 384-395, 1970.

2. *Nacházel J., Paleček F.*: Physiol. Bohemoslov. 39: 435-442, 1990.

**INSUFFICIENT BREATHING OF ANAESTHETIZED DOGS FROM NON-RELAXATION POSITIONS OF THE THORAX: INFLUENCE ON CARDIAC OUTPUT AND BLOOD PRESSURE.** *J. Hájek, O. Slezáková, A. Kurtanský, I. Šipinková, K. Kosák, A. Matášeje*, Department of Physiology, Faculty of Medicine, Comenius University, Bratislava.

Cardiac output and blood pressure were studied in 9 mongrel dogs during breathing from shifted positions of the thorax (maintained by pressure changes in a tank respirator) combined with a 50 % increase of the dead space. Ten controls breathed from the thorax equilibrium position. The Fick method was used for calculating the cardiac output. In the inspiratory position, the cardiac output and systolic pressure decreased by 606 ml and 6.88 kPa, respectively ( $P < 0.05$ ); an increase in dead space lowered the output again without altering the blood pressure. In the expiratory position, the two variables increased by 823 ml and 2.54 kPa, respectively ( $P < 0.05$ ); added dead space decreased cardiac output while blood pressure continued to rise. In the expiratory position, the increased dead space thus interferes with the parallelism of changes in blood pressure and cardiac output.

**LATHYRISM INHIBITS HYPOXIC PULMONARY HYPERTENSION IN RATS,** *J. Herges, J. Kawikova, V. Hampel*, Department of Pathophysiology, Second Medical School, Charles University, Prague.

In animals exposed to hypoxia, the new collagen fibres deposited in the pulmonary vascular wall and vessels are less elastic. A lathyrogenic agent, beta-aminopropionitrile (APN) inhibits the formation of crosslinks in collagen resulting in low stress resistance of connective tissue. Four groups of young male rats were used: C - treated with saline, normoxia, L - 20 mg/kg b.w. APN daily, 5 weeks, normoxia, CH - saline, 5 weeks, last 2 weeks exposed to hypoxia ( $F_iO_2 = 0.1$ ) and LH - APN as group L, hypoxia as group CH. Pulmonary arterial mean blood pressure in intact, anaesthetized rats was lower in group LH ( $23 \pm 5$ ) (torr  $\pm$  S.D.) than in CH ( $33 \pm 5$ ,  $p < 0.05$ ) and did not differ from the normoxic groups (C =  $17 \pm 2$ , L =  $18 \pm 2$ ). In the group LH the  $\Delta P/\Delta Q$  relationship, measured in isolated perfused lungs, had a lower intercept with the pressure axis than in the group CH. We conclude that collagen deposition in the pulmonary vessels plays a role in the development of pulmonary hypertension.

**PULMONARY GAS EXCHANGE DURING THERMAL POLYPNOEA IN THE DOG.** I. Šipinková, G. Hahn<sup>1</sup>, M. Meyer<sup>1</sup>, J. Piiper<sup>1</sup>, Department of Physiology, Faculty of Medicine, Comenius University Bratislava and <sup>1</sup>Department of Physiology, Max Planck Institute for Experimental Medicine, Göttingen, FRG.

Pulmonary gas exchange was studied in 7 conscious dogs (mean body weight 32 kg) provided with a chronic tracheostomy and an exteriorized carotid loop. Thermal polypnoea was elicited by increased ambient temperature (mean 27.5 °C, relative humidity 65 %). PO<sub>2</sub> and PCO<sub>2</sub> were determined in the arterial blood and the gas phase utilizing a special system for analysis of respiratory gas composition at high breathing frequencies. The following parameters were measured during thermal polypnoea (mean ± S.D.): breathing frequency 313 ± 19.min<sup>-1</sup>, tidal volume 167 ± 21 ml BTPS, effective alveolar ventilation 5.5 ± 1.3 l BTPS.min<sup>-1</sup>, series dead space ventilation 40.0 ± 1.0 l BTPS.min<sup>-1</sup>, PaO<sub>2</sub> 106.2 ± 5.9 Torr, PaCO<sub>2</sub> 27.2 ± 3.9 Torr, pH 7.45, O<sub>2</sub> uptake 193 ± 37 ml STPD.min<sup>-1</sup>, CO<sub>2</sub> output 168 ± 33 ml STPD.min<sup>-1</sup>. Pulmonary gas exchange during thermal polypnoea is characterized by decreased gas exchange efficiency due to increased parallel and series dead space ventilation and incomplete gas mixing in the lungs.

**CONTINUOUS ANALYSIS OF RESPIRATORY GAS COMPOSITION DURING HIGH FREQUENCY VENTILATION (HFV).** G. Hahn, M. Meyer, I. Šipinková<sup>1</sup>, Department of Physiology, Max Planck Institute for Experimental Medicine, Göttingen, FRG and <sup>1</sup>Department of Physiology, Faculty of Medicine, Comenius University, Bratislava.

Gas exchange analysis during HFV is limited by the response characteristics of conventional gas analyzers. The presented novel technique combined with respiratory mass spectrometry and precise flow measurement allows continuous monitoring of respiratory gas composition at breathing frequencies up to 30 Hz. This method is based on discontinuous collection of minute gas samples at any selected point of the respiratory cycle combined with a sample-hold device. In the phase-locked mode of operation, gas sampling is synchronous with breathing frequency and suitable for e.g. end-tidal gas concentration monitoring. In the scanning mode, samples are trapped from the respired gas asynchronously with the breathing frequency providing thus a method for the measurement of gas concentration profile during the whole respiratory cycle. The system may be applied in strictly regular breathing as encountered during mechanical HFV or in quasi-regular breathing observed e.g. in spontaneous thermal polypnoea.

**ADJUSTMENT OF HYPOCAPNIA DURING HIGH FREQUENCY JET VENTILATION.** A. Čalkovská, K. Javorka, Department of Physiology, Faculty of Medicine, Comenius University, Martin.

The possibility of adjusting hypocapnia by external addition of CO<sub>2</sub> during 5-hour hyperventilation induced a high frequency jet ventilator was studied in 15 anaesthetized and paralyzed rabbits. After a 30 min period of hyperventilation, the inflow (40-120 ml x min<sup>-1</sup>) CO<sub>2</sub> into the nozzle of the generator was started. The controls (7 rabbits) were ventilated with the same ventilatory pattern without addition of CO<sub>2</sub>. Specimens of arterial and mixed venous blood were obtained at 30 min intervals. Hyperventilation evoked hypocapnia in both subgroups (decrease of PaCO<sub>2</sub> from 3.8 ± 0.2 to 1.7 ± 0.2 kPa and 2.1 ± 0.2 kPa respectively), an increase of pH<sub>a</sub> and PaO<sub>2</sub> and decrease of right-left pulmonary shunts. The external addition of CO<sub>2</sub> during the unchanged ventilatory mode normalized PaCO<sub>2</sub> and pH<sub>a</sub>. Hyperventilation decreased HCO<sub>3</sub><sup>-</sup> and increased Cl<sup>-</sup> in the blood. The changes were eliminated by the addition of CO<sub>2</sub>.

**LACTATE ACCUMULATION DURING WORK PERFORMANCE LASTING THREE SECONDS.** *V. Klíner, E. Vinšová, J. Vřtvička*, First Medical Police Sport Centre and Second Laboratory of Clinical Biochemistry and Physiology of Top Sportsmen, Prague.

A group of 4 boys and 4 girls (aged  $16 \pm 1$  year) was tested. They ran at their maximal speed two sets  $4 \times 30$  m (flying start). The interval between the sets was 8 min, between separate runs 4 min. The average duration of each run was  $3.43 \pm 0.25$  s and there was no significant statistical difference between individual runs and between single sets. Average concentrations of blood lactate (La) and base excess (BE) (from the ear lobe) were  $1.5 \pm 0.3$  mmol/l and  $-1.2 \pm 0.7$  mmol/l before the start. Average concentrations of La and BE were  $9.4 \pm 1.1$  mmol/l and  $-7.0 \pm 1.9$  mmol/l after the run in the first set and  $11.6 \pm 1.8$  mmol/l and  $-10.5 \pm 2.4$  mmol/l in the second set. The obtained data showed that even at 3.5-second work performance increases blood lactate levels significantly. Our observations lead to the conclusion that it is always possible to find such a duration of a workload and of such intensity, which cannot be proved to be "anaerobic-alactic" work.

**CHANGES IN PERIMETRY AT MAXIMAL LOAD IN ICE HOCKEY PLAYERS.** *J. Heller, J. Pešek, R. Dlouhá, V. Bunc, J. Novotný*, Research Institute of Physical Culture, Charles University, Prague.

In order to evaluate alterations of the visual field due to a maximal exercise load, a group of first league ice hockey players ( $n=26$ , age  $23 \pm 4$  years,  $VO_{2max}$   $55 \pm 5$  mL $\cdot$ min $^{-1}$  $\cdot$ kg $^{-1}$ ) and a group of young ice hockey players ( $n=17$ , age  $11 \pm 1$  years,  $VO_{2max}$   $58 \pm 4$  mL $\cdot$ min $^{-1}$  $\cdot$ kg $^{-1}$ ) were examined by bicycle ergometry combined with perimetry, the latter group repeatedly after a 4 months training period. The lateral boundaries of the visual field (horizontal visual angle, HVA) were estimated at rest and within 15 s after finishing the maximal exercise. The resting HVA in league players ( $149 \pm 12^\circ$ ) was comparable to the values found in young players both in the 1st ( $144 \pm 13^\circ$ ) and in the 2nd ( $151 \pm 9^\circ$ ) measurement. At maximal load a decrease in HVA was observed in 60 %, an increase in 20 % and no changes in 20 % of both league and young players. The alterations of HVA were related neither to the degree of exertion at maximal load nor to the functional capacity of the players. However, repeated measurements in young players were significantly correlated both for resting HVA ( $r=0.68$ ,  $p<0.003$ ) and for HVA at maximal load ( $r=0.55$ ,  $p<0.02$ ).

**SOMATIC AND FUNCTIONAL CHARACTERISTICS OF KUWAIT TENNIS PLAYERS.** *A. Scholtzová, V. Štulrajter, A. Zrubák, L. Ramacsay*, Faculty of Physical Education and Sport, Comenius University, Bratislava

Measurements of somatotypes in Kuwait tennis players disclosed their pertinence to the ectomorphical zone with an adequate share of mesomorphy at the age of 13.2 years and to the endo and/or ecto mesomorphical zone at the age of 18.3 years. The reaction time (RT) in Kuwait players to central ( $255 \pm 26$  ms) and temporal ( $282 \pm 27$  ms) impulses were longer than in Czechoslovak players ( $214 \pm 46$  ms and  $230 \pm 51$  ms respectively). The disjunctive RT to two ( $272 \pm 31$  ms) and four ( $455 \pm 89$  ms) impulses was better in Kuwait players than in Czechoslovak ones ( $320 \pm 45$  ms and  $617 \pm 74$  ms respectively). Better results of motor tests in the standing jump ( $229 \pm 19$  cm) and in the deep forward bend ( $7.8 \pm 7.4$  cm) were achieved by Kuwait players than by the average Slovak population ( $178 \pm 20$  cm and  $3.9 \pm 5.5$  cm respectively). On the other hand, worse results were recorded in arm bends ( $2.0 \pm 1.5$ ) than in the average population ( $4.8 \pm 3.0$ ). We suggested that the RT and arm strength in Kuwait players should be improved by means of special exercises.

VENTILATORY RESPONSE DURING INCREMENTAL EXERCISE TESTS IN FEMALE ATHLETES OF DIFFERENT AGES. V. Bunc, J. Heller, Research Institute of Physical Education, Charles University, Prague.

The effect of progressively increasing work rate ( $20 \text{ W}\cdot\text{min}^{-1}$ ) up to exhaustion on the same course of  $\text{VO}_2$  and  $\dot{V}$  has been studied in groups of female biathlon athletes of different ages (I mean age =  $14.8 \pm 0.8$  years, mean  $\text{VO}_{2\text{max}} = 2.92 \pm 0.37 \text{ L}\cdot\text{min}^{-1}$ , II  $17.1 \pm 0.4$  years,  $3.62 \pm 0.28$  and III  $20.4 \pm 1.0$  years,  $4.00 \pm 0.42$ ). While  $\text{VO}_2$  increased linearly with the duration and work rate (mean slope in I  $0.22 \pm 0.04 \text{ L}\cdot\text{min}^{-2}$ , in II  $0.21 \pm 0.04$  and in III  $0.25 \pm 0.05$ ),  $\dot{V}$  changed its slope in two ranges. The first  $\dot{V}$  change of slope occurred at work loads below anaerobic threshold (AT) - (in group I  $14.93 \pm 42 \text{ L}\cdot\text{min}^{-2}$ , in II  $13.54 \pm 2.91$  and in III  $16.43 \pm 3.72$ ). The second changes of slope occurred in the range above AT (in I  $18.23 \pm 4.36 \text{ L}\cdot\text{min}^{-2}$ , II  $17.94 \pm 3.11$  and in III  $18.72 \pm 4.11$ ). The differences in above variables between all the three groups were not significant. The 1st slope of  $\dot{V}$  course might be the consequence of the different readjustment of  $\text{VO}_2$  on-response and hence of early lactate changes in different subjects. The 2nd slope seems to be dependent on the anaerobic capacity of the subjects investigated which was practically the same in our groups.

INDIVIDUAL VARIABILITY IN ENERGY AND OTHER FOOD COMPONENT INTAKE BEFORE, DURING AND AFTER A 100 KM RUN. J. Pařízková, J. Novák, Institute of Sports Medicine, Faculty of Physical Education, Charles University, Prague.

Previous studies of dietary intake during extreme work loads showed *inter alia* a marked energy deficit as related to its output. When following a number of parameters including dietary intake by the inventory method before, during and after a 100 km run, attention was focused on individual differences in the intake of energy, proteins, fats, carbohydrates, minerals and vitamins as related to the performance, and to the individual changes in body weight and composition. The results showed marked interindividual variations, which did not influence the performance markedly; these findings indicate obvious differences in the utilization of endogenous energy resources necessary for the performance.

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