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I. Neuromuscular Physiology

SATELLITE CELL ACTIVITY DURING MUSCLE GROWTH IN THE CRAYFISH. *M. Novotová, K. Rýdlová, B. Uhrik,* Institute of Molecular Physiology, Slovak Academy of Sciences, Bratislava.

Crayfish muscle fibres represent a suitable model for studying the process of muscle tissue formation during the moulting period. In this study, ultrastructural changes of m.extensor carpopoditi of *Astacus fluviatilis* were examined in the electron microscope. The areas of growth were observed within the material of external lamina both at the perifery of muscle fibres and in the lumen of sarcolemmal invaginations. They were characterized by the presence of myoblasts originating from activated satellite cells. The myoblast cytoplasm contained microtubules, vesicles, mitochondria and endoplasmic reticulum. Cytoplasmic outgrowths of myoblasts were delineated by patches of dense material and were approaching the fibre and fusing with the muscle cell membrane. Our results showed that satellite cells are of crucial importance not only in regeneration (1) but also in muscle growth during moulting.

1. Novotová M., Rýdlová K.: International symposium "Motoneuronal plasticity", p. 79, Bonn, 1990.

THE EFFECT OF NOCICEPTIVE STIMULATION INDUCING REFLEX MUSCLE ATROPHY IN RATS. *H. Urbancová, P. Hník, R. Vejsada,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Musle atrophy was induced in the soleus, gastrocnemius, tibialis ant. and extensor digitorum longus by unilateral fracture of the paw in adult rats under anaesthesia. Muscle wasting in all four muscles was comparable with denervation atrophy 3 and 7 days after sciatic nerve section. In four groups of animals dorsal roots L_4-L_6 were sectioned prior to metatarsal bone fracture (20 animals) or 2 h, 1 h and 2 min after the fracture (30 animals). It was shown that when deafferentation preceded the nociceptive stimulus, no loss of muscle weight occurred after 3 and 7 days. This may serve as evidence that this type of atrophy is indeed reflex in origin. However, the nociceptive stimulus does not have to act throughout the whole 7-day period, since even deafferentation performed 2 min after the fracture did not prevent muscle wasting. The nociceptive stimulus thus acts as a "trigger" for initiating muscle atrophy.

ON THE MECHANISMS OF SPONTANEOUS RELEASE OF ACETYLCHOLINE FROM THE RAT DIAPHRAGM DURING INCUBATION IN VITRO. *V. Doležal, S. Tuček,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Acetylcholine (ACh) is liberated from skeletal muscles during resting incubation *via* quantal and non-quantal release from nerve terminals and release from muscle fibres. The spontaneous non-quantal release is believed to account for an overwhelming proportion of ACh that is liberated from the nerve terminals. We have now found that the release of ACh is diminished by about 40 % in the presence of tetrodotoxin, or in the absence of Ca^{2+}. The inhibitory effect of vesamicol on the release is lost under such conditions. We suggest that tetrodotoxin and Ca^{2+} deprivation eliminate the release which is due to spontaneous impulse activity arising in nerve terminals after the inhibition of cholinesterases and that the proportion of quantal release on the total release is higher than has formerly been believed.

CHOLESTEROL SYNTHESIS IN RABBIT SKELETAL MUSCLES AFTER DENERVATION. *V. Mézešová, A. Drgová, G. Bezáková, D. Dobrota,* Department of Biochemistry, Jesenius Medical Faculty, Comenius University, Martin.

Denervation of fast rabbit skeletal muscles was followed by a 30 % increase in the cholesterol content of sarcotubular system membranes (STS) and a concomitant increase of the relative content of sphingomyelins (1). Changes in the content of cholesterol and phospholipids, as integral parts of the membranes, may affect the biological functions connected with membranes. We tried to explain the mechanism, which leads to the accumulation of cholesterol in membranes. One possible mechansim which is followed by an increase in the cholesterol content may be due to its enhanced synthesis. The study of cholesterol synthesis from ^{14}C mevalonic acid in fast muscle slices of the rabbits has shown that the rate of cholesterol synthesis in the denervated muscles is the same as in innervated muscles. The activity of the key enzyme of cholesterol synthesis, HMG-CoA reductase, was not changed in STS after denervation. The present results indicate that the increase of the cholesterol content in STS membranes after denervation is not due to increased activity of HMG-CoA reductase.

1. Lehotský J., Drgová A., Dobrota D., Mézešová V.: Gen. Physiol. Biophys. 10: 175-188, 1991.

THE EFFECT OF MURAMYLDIPEPTIDE (MDP) ON THE SEROTONERGIC SYSTEM IN ISOLATED ORGANS. *J. Slánský, O. Kadlec, K. Mašek,* Institute of Pharmacology, Czechoslovak Academy of Sciences, Prague.

A study has been made of the interaction of MDP with the serotonergic nervous system (1,2). MDP in a low concentration (50 $nmol.l^{-1}$) selectively enhanced contractions evoked by 5-HT in rat stomach strips and vas deferens, while contractions evoked either by acetylcholine, noradrenaline of KCl were not influenced. MDP sensitized preparations to 5-HT stimulation at the postsynaptic level. In the carp stomach strip, a high concentration of 5-HT (10 $\mu mol.l^{-1}$) evoked contractions that were further potentiated by MDP. A low concentration of 5-HT (0.1 $\mu mol.l^{-1}$) decreased the neurogenic twitches evoked by electric field stimulation; this was interpreted as presynaptic autoinhibition of 5-HT release. Such presynaptic autoinhibition of twitches was significantly more potentiated by MDP than the postsynaptic effect of 5-HT. MDP can be considered as a physiological antagonist of the 5-HT system.

1. *Mašek K., Kadlec O.:* The Lancet, Vol. 1, 1277, 1983.
2. *Horák P., Mašek K.:* Čs. fyziol. 38: 144, 1989.

IMMUNOHISTOCHEMICAL REACTIONS OF DEEFFERENTED RAT MUSCLE SPINDLES WITH SUPERNUMERARY INTRAFUSAL FIBRES. *T. Soukup, F. Pedrosa-Domellöf[1], G. Zachařová, L-E. Thornell[1],* Institute of Physiology, Czechoslovak Academy of Sciences, Prague and [1]Department of Anatomy, University of Umea, Umea, Sweden.

We studied the neoformation of supernumerary intrafusal fibres in the rat hind limb muscle spindles 6 weeks after neonatal deefferentation. Supernumerary fibres had smaller diameters, their myofibrillar ATPase, myosin heavy chain (MHC) and M-band protein composition generally corresponded to bag_2, bag_1 and chain fibres described in spindles 3 weeks after neonatal deefferentation (1). In 6-week-old rats, the number of chain fibres increased x 2.5 and of bag fibres x 1.68 in comparison with the average 2 bag and 2 chain fibres in control spindles. The differentiation of chain myotubes was probably more frequent because secondary intrafusal myotubes are intrinsically determined to coexpress fast twitch and neonatal MHCs. The presence of additional bag-like fibres suggests that their satellite myotubes are able to express the slow tonic MHC under the influence of acquired sensory innervation. The possibility that different types of satellite cells related to different intrafusal fibre types exist in muscle spindles deserves further attention.

1. *Soukup T., Pedrosa F., Thornell L.-E.:* Histochemistry 94: 245-256, 1990.

Supported by a grant from the Czechoslovak Academy of Sciences, No. 51145.

IMMUNOCYTOCHEMISTRY OF HUMAN INTRAFUSAL FIBRES. *L.-E. Thornell*[1], *T. Soukup, F. Pedrosa-Domellöf*[1], *M. Krupková, G. Zachařová,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague and [1]Department of Anatomy, University of Umea, Umea, Sweden.

We studied the expression of myosin heavy chain (MHC) and M-band isoproteins along the length of intrafusal fibres in human lumbrical muscles on serial cryostat sections. Bag_1 fibres were strongly stained with monoclonal (m) antibodies (Abs) against slow tonic (STO) MHC along their entire length, with slow twitch (STW) and M-protein mAbs in their polar zones; they did not stain with fast twitch (FTW) mAb and antineonatal (NN) MHC Ab. The presence of M-protein in the extracapsular polar zones correlates with our electronmicroscopical observations demonstrating the presence of an M-line in this region of bag_1 fibres. Bag_2 fibres were strongly stained along their whole length, with STO and STW mAbs; their staining with FTW, NN and M-band Abs was moderate to high. Chain fibres were strongly and uniformly stained with the FTW, NN and M-protein Abs. Thus, human intrafusal fibres coexpress several MHC isoforms, each with a characteristic distribution along the whole fibre length, similarly as was described in other species.
Supported by a grant from the Czechoslovak Academy of Sciences, No. 51145.

REGENERATION OF MUSCLE SPINDLES AFTER NEONATAL NERVE CRUSH IS NOT ENHANCED BY MARCAINE TREATMENT. *T. Svoboda, T. Soukup, R. Vejsada, I. Jirmanová, T. Valentová, L.-E. Thornell*[1], Institute of Physiology, Czechoslovak Academy of Sciences, Prague and [1]Department of Anatomy, University of Umea, Umea, Sweden.

Neonatal crush of the sciatic nerve leads to irreversible elimination of muscle spindles even if muscles are reinnervated during the 2nd postnatal week; only a few atypical spindles can be found (1). In our experiments we used a myotoxic drug, Bupivacaine (Marcaine), to initiate muscle degeneration and regeneration and to test whether the presence of regenerating myoblasts and myotubes available for sensory induction at the time when nerve fibres grow into the muscle cannot enhance the regeneration of muscle spindles. We found that in rat hind limb muscles only a limited number of atypical spindles regenerated within the preserved capsules. They contained one or more intrafusal fibres with atypical histochemical and immunocytochemical characteristics. The factors responsible for the failure of afferent innervation to induce spindle formation during early postnatal regeneration require further analysis.
1. *Zelená J., Hník, P.:* Physiol. Bohemoslov. 9: 373-381, 1960.
Supported by a grant from the Czechoslovak Academy of Sciences, No. 51145.

CRITICAL PERIOD IN THE DEVELOPMENT OF TACTILE LAMELLAR CORPUSCLES OF THE RAT. *J. Zelená, I. Jirmanová, T. Nitatori*[1], *C. Ide*[1], Institute of Physiology, Czechoslovak Academy of Sciences, Prague and [1]Department of Anatomy, Iwate Medical University, Japan.

The development of lamellar corpuscles was studied in rat toe pads under normal conditions and after crushing the sciatic nerve in 1- to 15-day-old animals. Rat lamellar corpuscles differentiate postnatally. By postnatal day 2, tiny axon terminals can be found in dermal papillae but, first, thin lamellae are formed around the terminals 8-12 days after birth. The corpuscles are structurally mature in 20-day-old rats. No lamellar corpuscles develop in the toe pads after crushing the sciatic nerve in newborn rats, and only occasional corpuscles regenerate after nerve crush at 5 days of age. After nerve crush in 10-day-old rats, lamellar corpuscles regenerate by the first month after the operation but they remain underdeveloped: their number and size are smaller than normal even 1 year after injury, and their terminals are encircled only by 1-3 lamellar layers. After nerve crush in 15-day-old rats, the corpuscles recover upon reinnervation and their size and lamellation become almost normal.

THE SOLANDT EFFECT IN MAMMALIAN SKELETAL MUSCLE. *J.A. Mejsnar, A.E. Chinet*[1], Charles University, Prague and [1]University Medical Center, Geneva.

The cranial gracilis muscle from cold – acclimated rats, placed *in vitro*, was simultaneously superfused with a standard Krebs-Ringer bicarbonate-buffered solution tonometered at pO_2 of 3.4 kPa and perfused with a same solution tonometered at pO_2 of 92 kPa. The effect of increased concentration of K^+ (20.9 mM) in the solutions was tested in experiments with this preparation. The increased concentration of K^+ in the superfusion solution had no effect on oxygen consumption. The increased concentration of K^+ in the perfusion solution enhanced the O_2 – consumption from 104.8 ± 0.2 (S.E.M.) to 126.2 ± 0.4 $nmol.g^{-1}.min^{-1}$ (7 experiments), i.e. by 21 %. The effect of extracellular K^+ is blocked by 75 % with 10 mM 2,3 butanedione monoxime (which blocks Ca^{2+} release from the sarcoplasmic reticulum). These results thus confirm the Solandt effect in a mammalian skeletal muscle.

II. Cell Physiology

GAMMA-GLUTAMYL TRANSPEPTIDASE AS A MARKER OF QUINOLINIC ACID-INDUCED DAMAGE IN THE DEVELOPING RAT BRAIN. *F. Šťastný, R. Haugvicová, V. Lisý,* Instituite of Physiology, Czechoslovak Academy of Sciences, Prague.

Quinolinic acid (QUIN), a tryptophan metabolite acting as a glutamate agonist at the NMDA receptor, caused neuronal loss 48 h after its intracerebroventricular injection followed by a massive bilateral neurodegeneration in the hippocampal formation of 30-day-old rats. In contrast, QUIN was almost ineffective in 12-day-old animals. For better localization of the damage and understanding of its dynamics we used changes in the activity of gamma-glutamyl transpeptidase (GGT) in various fields of the hippocampal formation in 12-, 30- and 50-day-old rats. The excitotoxicity of QUIN is, partially, involved in triggering the initial hippocampal GGT increase, but the subsequent decreases of this enzyme activity are related to degeneration of pyramidal and dentate granule neurones. These changes may be initiated by stimulation and later by chemical damage of neurones in the entorhinal cortex. This hypothesis is supported by fluctuation of GGT activity in the hippocampal formation of younger and older rats with bilateral lesions of the perforant path fibres.

ANALYSIS OF SARCOLEMMAL BETA-RECEPTOR FRACTION FROM THE DOG HEART. *L. Okruhlicová, T. Rada, M. Barančík, J. Šmisterová*[1], Institute for Heart Research, Slovak Academy of Sciences and [1]Institute of Experimental Pharmacology, Slovak Academy of Sciences, Bratislava.

The sarcolemmal fraction from the dog myocardium was analysed using radioligand, cytochemical and immunological methods. The receptor density B_{max} of the membrane fraction was determined using ^{3}H-dihydroalprenolol. The functional state of beta-receptors was demonstrated by isoproterenol stimulation of adenylate cyclase activity. Polyclonal antibodies against beta-receptors were prepared by immunization of rabbits (1) with a sarcolemmal fraction. The proteins of this fraction were electrophoretically transferred to a nitrocellulose membrane and using the method of immunoblotting incubated with immune sera. The binding of antibodies with proteins, which have the molecular weight 65 000 Da, was detected. We assume that binding of the antibodies with the beta-receptor, which has also a molecular weight of 65 kDa. Specific methods are needed for further specific detection.
1. *Rada* T., Okruhlicová L., Slezák J.: Brat. lek. Listy 92: 3-4, 138-141, 1991.

EFFECT OF PHORBOL ESTER ON I_{Ca} IN RAT AND GUINEA-PIG CARDIAC MYOCYTES. *L. Lacinova, M. Morad*[1], Institute of Molecular Physiology and Genetics, Slovak Academy of Sciences, Bratislava and [1]Department of Physiology, Medical School, University of Pensylvania, Philadelphia, USA.

The negative effect of phorbol esters on I_{Ca} in neuronal cells was recently reported (1). We investigated in more detail the effect of externally applied phorbol-12,13-diacetate (PDA) on cardiac myocytes. PDA was found to suppress I_{ca} in both rat atrial and ventricular myocytes with shallow dose-dependence in the concentration interval 10-500 nM. The effect of PDA on Ca^{2+} current in guinea-pig ventricular myocytes was more complex with a bell-shaped dose-dependence within the same concentratioin interval. 100 nM PDA increased I_{Ca} which was usually transient. This dual effect of PDA may be due to its diffusion into the cell and subsequent interaction with the protein kinase C system.

1. *Hockberger P., Toselli M., Swandula D., Lux D.H.:* Nature 338: 340-342, 1989.

EFFECT OF ALCURONIUM ON THE BINDING OF MUSCARINIC LIGANDS IN CHICK HEART AND RABBIT LUNG. *J. Jakubík, S. Tuček,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

It has been discovered in earlier work from our laboratory that the neuromuscular blocking drug acuronium has a positive allosteric effect on the binding of N-(^{3}H)metylscopolamine to muscarinic receptors in rat heart atria, ileal smooth muscle and the cerebellum. The positive effects of alcuronium appeared to be specific for the genetically defined m_2 receptor subtype, while the binding to the m_1 and m_3 sybtypes was not affected. Muscarinic receptors in the chick heart differ from those in the rat heart and probably belong to the m_4 subtype, similarly as the receptors in peripheral parts of the rabbit lung. We have now found that alcuronium also increases the binding of N-(^{3}H)methylscopolamine to these m_4 receptors; it has no positive effect on the binding of (^{3}H)quinuclidinyl benzilate.

ACTIVATION OF THE REACTION OF THE PLASMA-MEMBRANE CA^{2+}-PUMP BY NEGATIVELY CHARGED PHOSPHOLIPIDS. *J. Lehotský, L. Raeymaekers*[1], *R. Casteels*[1], Department of Biochemistry, Jesenius Medical Faculty, Comenius University, Martin, Czechoslovakia and [1]Laboratory of Physiology, KUL Leuven, Belgium.

The (Ca^{2+} + Mg^{2+})-ATPase of the plasma membrane is activated by negatively charged phospholipids. The mechanism of this activation was investigated by studying the effect of phospholipids on the steady-state phosphointermediate level and the p-nitrophenylphosphatase activity, an enzymatic reaction catalyzed by the Ca^{2+}-ATPase but differing from the ATPase reaction in several characteristic. Both paramaters were differentially affected by different acidic phospholipids. The level of phophoprotein intermediate was not affected by phosphatidylserine (PS) (30 % of total phospholipid), but it was increased by 60 % by phosphatidylinositol-4-phosphate (PIP). PS increased the p-nitrophenylphosphatase activity, whereas PIP had no significant effect. The results indicate that at least two reaction steps are involved in the activation. PIP mainly affects a reaction steps which accelerates the formation of the phosphointermediate, whereas the action of PS would affect two steps, one upstream and one downstream of the phosphointermediate.

LONGLASTING INCREASE OF RESPONSES TO REPEATED APPLICATIONS OF L-HOMOCYSTEIC ACID IN MOUSE CULTURED HIPPOCAMPAL NEURONES. *L. Vyklický Sr., V. Vlachová,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The concentration-response relationship (c-rr) of whole cell currents induced by L-homocysteic acid (L-HC) was studied in mouse embryonic hippocampal neurones in culture (n=56). In most neurones two phases in the c-rr could be distinguished. The first was characterized by responses to 3-300 μM L-HC which were desensitized in a concentration-dependent manner and were antagonized by 2-amino-5-phosphonovaleric acid (APV) indicating that NMDA receptors had been activated. The second phase was characterized by sustained responses which were insensitive to APV, indicating activation of non-NMDA receptors. In eight neurones, two phases in c-rr were not observed at the beginning of the recording. A 3-4 fold increase of the membrane current to L-HC (30-300 μM) developed after repeated L-HC applications. This increase was blocked by APV. It is suggested that the long-term increase of the membrane current reflects relief from desensitization of the NMDA receptors in which Ca^{2+} play an important but not an exclusive part.

PLASMA MEMBRANE INDEPENDENT POOL OF THE GS (ALPHA) PROTEIN IN THE LOW-DENSITY MEMBRANE FRACTION OF S49 LYMPHOMA CELLS. *P. Svoboda, P. Kvapil*[1], *P.A. Inse*[2], *L. Ransnas*[1], Institute of Physiology, Czechoslovak Academy of Sciences, Prague, [1]Department of Clinical Chemistry, University of Göteborg, Göteborg, and [2]Department of Pharmacology, UCSD, San Diego.

We report that the compartmentation of the stimulatory GTP-binding protein Gs exists in S49 lymphoma cells. Besides the cytosolic form of Gs subunit (1), three membrane bound forms of Gs were identified with the help of rate zonal centrifugation in sucrose density gradients and identification with specific antipeptide antibodies and cyc-reconstitution assay. The first pool of Gs may be identified with the plasma membrane fragments, the second pool correlates with the endoplasmic reticulum (microsomal) enzyme markers, the third pool was detected in light (low-density membranes) at the top of the gradient. The complete absence of specific adenylyl cyclase and Na,K-ATP-ase activity indicates that this low-density (light) membrane form of Gs is distinct from any plasma membrane fragments.

1. *Ransnas et al.:* Proc. Nat. Acad. Sci. USA 86: 7900-7903, 1989.

III. Cardiac Physiology

ANTIARRHYTHMIC EFFECT OF 7-OXO PROSTACYCLIN (7-OXO PGI_2) ON REPERFUSION-INDUCED ARRHYTHMIAS IN THE ISOLATED RAT HEART. *T. Ravingerová, A. Džurba, N. Tribulová, A. Ziegelhöffer,* Institute for Heart Research, Slovak Academy of Sciences, Bratislava.

An increased intracellular concentration of calcium is knowm to be a powerful arrhythmogenic stimulus in the isolated rat heart upon reperfusion. In the present study, the effect of 7-oxo PGI_2 was investigated on reperfusion arrhythmias in rat hearts 48 h after pretreatment with the drug *in vivo* (50 $\mu g.kg^{-1}$ b.w., i.m). Isolated Langendorff-perfused rat hearts were subjected to 30 min of regional ischaemia (occlusion of LAD) and reperfusion. In the untreated group, reperfusion induced 100 % of both ventricular tachycardia (VT) and ventricular fibrillation (VF), of which 80 % were sustained (lasting > 2 min). In the 7-oxo PGI_2 goup VT and VF were characterized by a rapid onset and rapid cessation (within 2 min of reperfusion), as well as by a lower incidence of VF. The antiarrhythmic action of 7-oxo PGI_2 was unrelated to changes of both heart rate and coronary flow, suggesting a direct action of the drug on the heart. The latter involves stimulation of the sarcolemmal Na-K pump activity thus preventing increased intracellular accumulation of sodium and calcium during ischaemia and reperfusion.

CALCIUM DEPENDENT INDUCTION OF CONTACT SITES (CS) FORMATION IN MEMBRANES OF RAT HEART MITOCHONDRIA IN VITRO. *R. Monošíková, A. Baker[1], F. Sima, A. Ziegelhöffer, N. Tribulová,* Institute for Heart Research, Slovak Academy of Sciences, Bratislava, CSFR and [1]University of Antwerp, Belgium.

Increased metabolic activity of myocytes was found to be associated with enhanced formation of CS in cardiac mitochondria (cM) in which the inner and outer membrane were coming into close contact. The aim of the present study was to contribute to the functional importance of the CS in cM as well as to find out whether intracellular free calcium may attain the level of the metabolic signal which could induce CS formation in cM. Maximal CS formation *in vitro* was observed when isolated rat cM were incubated in presence of 10^{-5} M Ca^{2+} indicating that Ca^{2+} level may represent the signal for CS formation. It is known that membrane bound mitochondrial creatine phosphokinase (mCPK) normally present in dimers may form octamers in CS. Our results demonstrated than mCPK octamer formation has a dual effect: a) the enzyme activity is increased in CS where octamers are localized; b) the total mCPK activity in the mitochondria is decreased. The latter finding is associated with an increased K_M value from 0.098 mM to 0.197 mM. These results indicate that the transfer of energy from mitochondria to cytoplasm in metabolically activated hearts is concentrated to the CS.

DETECTION OF MITOCHONDRIAL CONTACT SITES AND CREATINE KINASE ACTIVITY IN CA^{2+} STIMULATED RAT HEART. *A. Baker[1], W. Jacob[1], N. Tribulová, T. Ravingerová, A. Ziegelhöffer, T. Rada,* Institute for Heart Research, Slovak Academy of Sciences, Bratislava and [1]University of Antwerp, Belgium.

Contact sites (CS) are created by fusion of the inner and outer mitochondrial membrane. They represent a dynamic microcompartment for creatine kinase (CK) activity. It has been reported from EM observations that the frequency of CS is changing according to the energy state of the mitochondria. Isolated rat hearts (Langendorff model) were perfused with Krebs-Henseleit solution containing 1.6 $mmol.l^{-1}$ Ca^{2+}. After 15 min stabilization, hearts were subjected to 1.8, 2.2, 2.6 and 3.6 $mmol.l^{-1}$ Ca^{2+} for a period of 15 min. Haemodynamic parameters were monitored by measuring ECG, heart rate. LVP, dp/dt and coronary flow. At the end of the experiment, the hearts were perfused for EM evaluation and for CK cytochemistry. Stimulation of isolated rat hearts with increased calcium concentrations was manifested by enhanced dp/dt and LVP values. A higher frequency of mitochondrial CS was observed and this was associated with a more frequent reaction for CK. We suppose that the formation of CS is related to the functional activity of Ca^{2+} stimulated rat hearts.

CHANGES IN SARCOLEMMAL ATPase ACTIVITIES IN THE HEART DURING CALCIUM DEPRIVATION PHASE OF THE CALCIUM PARADOX (CAP). *A. Džurba, T. Ravingerová, A. Ziegelhöffer,* Institute for Heart Research, Slovak Academy of Sciences, Bratislava.

Contraversial evidence is available about the participation of Na^+ and the related changes in sarcolemmal transport ATPases, in predisposing cardiac myocytes to CaP during its 1st phase - Ca deprivation. Our previous results revealed that complete development of CaP in rat hearts could be prevented by pretreatment of animals with 7-oxo PGI_2 (PGI_2; 50 $\mu g.kg^{-1}$ i.m.) 48 h prior to Ca deprivation. In the present study, molecular mechanisms of the above effect were investigated. In isolated perfused (Langendorff) control and PGI_2-pretreated hearts the activities of sarcolemmal Na/K-ATPase and Ca-ATPase with low affinity to Ca were studied after 10 min of Ca-free perfusion (Ca depletion). The results revealed a significant decrease in activities of both enzymes in the control group. In the PGI_2 group, the depressed activity of Na/K-ATPase was reversed by PGI_2, suggesting that the enhanced extrusion of Na ions constitutes the molecular basis of the protective effect of PGI_2. Thus, intracellular accumulation of Na due to the depressed activity of Na/K pump during Ca depletion may predispose the rat heart to CaP.

THE EFFECT OF THYROID STATUS ON THE DEVELOPMENT OF CARDIAC SARCOPLASMIC RETICULUM. AN ULTRASTRUCTURAL STUDY. *D. Jarkovská, F. Kolář[1], J. Procházka[1], B. Ošťádal[1],* Institute of Histology and Embryology, First Faculty of Medicine, Charles University and [1]Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The aim of this study was to evaluate the role of thyroid hormones in the maturation of the sarcoplasmic reticulum (SR) of cardiac cells from morphological point of view. Male Wistar rats were rendered hyperthyroid by daily injections of L-triiodothyronine and hypothyroid by including 6-n-propyl-2-thiouracil in the drinking water to mothers from day 2 to day 21 of postnatal life in both cases. The hypothyroid state results in the retardation of SR development, namely by the more seldom occurrence of couplings with the T-system, including classical triads. The T-system is underdeveloped as compared with the controls. In the hyperthyroid myocardium, the density of the SR network is roughly proportional to the degree of hypertrophy of myocardial cells. The couplings of SR with the T-system tubules are richly developed. The transversal T-tubules are often duplicated resulting in the development of pentads. It may be concluded that the thyroid status significantly influences the structural development of cardiac SR.

ENZYME KINETICS AND ACTIVATION ENERGY OF SARCOLEMMAL Ca-ATPase WITH LOW AFFINITY TO CALCIUM IN CONTROL AND ISCHAEMIC RAT HEARTS. *N. Vrbjar, A. Džurba, A. Ziegelhöffer,* Institute for Heart Research, Slovak Academy of Sciences, Bratislava.

Global ischaemia of the rat heart induces a time-dependent diminution in the activity and maximum velocity (V_m) value of sarcolemmal Ca-ATPase with low affinity to calcium. The depression of enzyme activity is maximal in the first 15 min after the onset of ischaemia. After 30 min of ischaemia the V_m diminution slows down, probably due to an adaptational decrease of the K_m value of the Ca-ATPase with low affinity to calcium. The observed phenomenon may be interpreted as a mechanism by which the enzyme attempts to cope with a situation when the supply by ATP is not sufficient. The decrease of the K_m value seems to represent the most important mechanism of the adaptation of the enzyme to changed physiological conditions, since the amount of energy needed by the enzyme to start the hydrolysis of ATP, the activation energy remains unchanged during ischaemia.

CONTRACTILE RESPONSE OF THE NEONATAL RAT HEART TO VERAPAMIL AND RYANODINE. *I. Ošťádalová, F. Kolář, J. Rohlíček, V. Rohlíček, J. Procházka, B. Ošťádal,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The contractile response of isolated perfused rat hearts to verapamil and ryanodine was studied in 1-, 2-, 4- and 7-day-old animals. Changes in developed force and maximum rate of force development were evaluated during a stepwise increase of the verapamil concentration in the perfusion solution (10^{-9} to $3.3x10^{-7}$ $mol.l^{-1}$). The negative inotropic response to the calcium antagonist, verapamil, increases significantly from day 1 to day 4 of postnatal life, suggesting day by day changes in cellular calcium regulation at this period. During further development, the cardiac sensitivity to verapamil decreases in a stepwise manner. The sensitivity to an inhibitor of the calcium release from the sarcoplasmic reticulum (SR) – ryanodine (10^{-6} $mol.l^{-1}$) – was already surprisingly high in 1-day-old animals, indicating that a functionally active SR in the rat heart is present shortly after birth. Our data clearly show that the inotropic responsiveness of the rat heart changes dramatically during the first week of life.

CHANGES OF COLLAGENOUS AND NON-COLLAGENOUS PROTEINS IN THE INFARCTED MYOCARDIUM. *V. Pelouch, N.S. Dhalla[1], R. Sethi[1],* Institute of Physiology, Czechoslovak Academy of Sciences, Prague and [1]Division of Cardiovascular Sciences, St. Boniface Gen. Hospital, Winnipeg, Canada.

Plasticity of the myocardium, resulting from different haemodynamic stimuli and following infarction is an important adaptive process for the maintenance of ventricular function. The aim of this study was to determine how the protein profile in both hypertrophic right (RV) and viable part of the left (LV) ventricles is affected in adult rats after ligation of the left coronary artery (LCA). Animals were tested 2, 4 and 8 weeks after LCA. It was shown that the content of collagen (C) in RV was elevated after the 2nd week, while it took 4 to 8 weeks in viable LVs; the concentration did not change after the 2nd week. The ratio between C and non-C proteins was decreased in RV and LV after the 2nd week. Analysis of hydroxyproline (HYP) in both soluble and insoluble collagen show that the LCA elevated total HYP, but the percentage HYP in soluble C was reduced due to a greater number of cross-linking in the collagen fraction. Our results indicate that the disproportion between synthesis of C and non-C proteins results in remodelling of the ventricular wall and may affect the stiffness and pumping capacity of the diseased heart due to myocardial infarction this way.

THE INCREASE OF PLASMATIC FREE FATTY ACID LEVELS HAS NO INFLUENCE ON THE EXTENT OF EXPERIMENTAL MYOCARDIAL NECROSES. *M. Mráz, A. Vrána[1], E. Faltová[2],* Department of Pharmacology, First Medical Faculty, Charles University, [1]Laboratory of Metabolism, Institute of Clinical and Experimental Medicine and [2]Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The effect of heparin administration on FFA levels and on the extent of heart necrotic lesions induced by isoprenaline (ISO) was studied in several strains of rats (conventional Wistar, hypertriglyceridaemic, ISO-resistant and ISO-sensitive). The extent of necroses was estimated by incorporation of ^{203}Hg into the heart ventricles. The extent of lesions was independent of triglyceridaemia (in hypertriglyceridaemia it was smaller). Heparin administration did not influence the necroses in spite of a marked increase of FFA levels, which was greatly enhanced in hypertriglyceridaemic rats. On the other hand, the necroses were substantially reduced in rat strains with a higher content of glycogen in the heart.

^{125}I-PINDOLOL BINDING ON MYOCARDIAL BETA-ADRENERGIC RECEPTORS IN RATS SENSITIVE AND RESISTANT TO ISOPRENALINE INDUCED HEART DAMAGE. *M. Černohorský, M. Mráz, S. Hynie,* Department of Pharmacology, First Medical Faculty, Charles University, Prague.

In previous experiments performed on the 17th generation of IR and IS rats (strains of rats resistant /IR/ and sensitive /IS/ to isoprenaline-induced myocardial lesions), no clear-cut differences either in the density of beta-receptors or in the dissociation constants were found between IR and IS rats with the use of ^{3}H-dihydroalprenolol as radiologand. In order to prove these findings we used female rats of the 30th generation and ^{125}I-pindolol as radioligand, which has a considerably higher specific activity, in the present study. Under these experimental conditions, and using non-linear regression analysis of specific binding parameters, we were also unable to demonstrate differences between these two experimental groups. Thus, it seems to be possible that the genetic difference in the myocardial resistance of rats to isoprenaline-induced damage is not caused by the differences in beta-adrenergic receptors.

COMPARISON OF ADENYLATE CYCLASE ACTIVITY AND ^{125}I-PINDOLOL BINDING IN HEARTS OF SHR AND NORMOTENSIVE BN.LX RAT STRAINS. *S. Hynie, M. Caisedo, V. Klenerová, P. Šída, M. Pravenec[1], P. Klír[1], V. Bílá, V. Křen,* First Medical Faculty, Charles University, Prague and [1]Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Spontaneously hypertensive (SHR) and normotensive polydactylous BN.lx rat strains are progenitors of two sets of recombinant inbred strains (HXB, BXH) developed for purposes of genetic analysis of hypertension and in the general rat genome as such. In this study we compared the activity of adenylate cyclase (AC) activity and ^{125}I-pindolol (I-P) binding in crude homogenates of rat ventricles of both progenitor strains. AC activity stimulated by norepinephrine, isoproterenol or forskolin did not show any difference in either strains. I-P binding to beta-adrenergic receptors (BAR) showed equal Kd values while maximal binding capacity in the SHR strain was about 17 % lower that in the BN.lx strain. These results indicate that hypertensive responsiveness in SHR cannot be ascribed to alterations of the myocardial BAR-AC system in SHR strains.

DOUBLE SPIRAL STRUCTURE OF THE MEDIA OF CORONARY ARTERIES IN THE DOG. *S. Doležel. S. Bartáková,* Institute of Pathological Physiology, Masaryk University, Brno.

The main trunk of the LAD coronary artery was frozen intravitally by pouring a propane-butane mixture -170 °C cold; five anaesthetized dogs were used for the experiment. A short segment of the artery was lyophilized and embedded in paraffin; 10 μm thin sections were stained with haematoxylin-eosin. The measuring of the horizontal and the ventrical distances between the ends of nuclei of myocytes was used for calculating the angle between the axis of the nucleus and the horizontal plane. This angle determines the pitch of the spiral. In this way, it was found that the nuclei of the media are arranged in two spirals which are twisted in opposite directions. Moreover, the spirals change their positions from the outer layer to the inner layer of the media after passing one half of the circumference of the artery. Probably, this complicated structure counterbalances the twisting force caused by the longitudinal distension of the artery.

A POSSIBLE EXPLANATION OF THE MONOPHASIC ACTION POTENTIAL DEVELOPMENT IN THE HEART MUSCLE. *J. Slavíček,* Department of Physiology, First Faculty of Medicine, Charles University, Prague.

A monophasic action potential (MAP) may be recorded under a suction or contact electrode in the heart muscle. The origin of MAP development is not entirely known. Suction lasting less than 5 min caused a reversible return of MAP to polyphasic ECG without ultrastructural changes (1). In the present work we attempted to prove indirectly the hypothesis about the activation of some stretch-activated channels during the development of MAP by suction or pressure. Some Na, K and Ca current blockers were applied to the tip of the suction capillary electrode before the negative pressure of -40 kPa was introduced in the frog ventricular epicardium. The results showed that the known kationic channel can be influenced by suction and probably possesses some characteristics of stretch-activated membrane channels. However further experiments should be done by the patch clamp technique for obtaining a direct explanation.

1. *Stojan M., Vacek Z., Slavíček J.:* Sborn. lék. 91: 288-293, 1989.

RELATIONSHIP BETWEEN SOME CARDIAC ARRHYTHMIAS AND ECG PARAMETERS DURING RESPIRATORY FAILURE. *Š. Kujaník, M. Mayer[1], I. Linder, I. Bračoková,* Department of Physiology, Faculty of Medicine, Šafárik, University, Košice and [1]Therapeutic Institute of Tuberculosis and Respiratory Diseases, Vyšné Hágy, Slovakia.

The relationship between ventricular (VEB), supraventricular extrasystoles (SVPB), bigeminy (BIG), pairs (VEBP) and run of extrasystoles (VERB), ventricular tachycardia (VT), heart rate (HR), QT interval (QT), irregular rhythm (IR), ST segment elevation (STe) and depression (STd) was studied in 27 respiratory failure male subjects at a sea level of 1150 m during 24 h by Holter monitoring of the ECG by means of simple correlation coefficients. In the control group, simple extrasystoles were rather rare. Very close ($0.9<r<0.99$) and medium close rorrelations ($0.7<r<0.89$) were not found. Relatively close correlations ($0.3<r<0.69$) concerned IR-SVPB, IR-VT, SVPB-BIG, VEB-VEBP, VEB-VEBR, VEB-BIG, VEB-HR, VEB-QT, VEB-QRc, VEBP-VT, VEBP-HR, VEBP-QT, VT-HR, VT-QT, QT-STd. Our findings indicate that there is no close correlation among these parameters but there is a certain relationship between serious arrhythmias and some ECG parameters.

CIRCADIAN RHYTHMS OF THE VENTRICULAR FIBRILLATION THRESHOLD DURING CHANGES IN VENTILATION IN FEMALE WISTAR RATS. *P. Švorc, P. Wilk, J. Murár[1], I. Polubný[2],* Institute of Physiology, Medical Faculty, Šafárik University, Košice, [1]Department of Clinical Biochemistry, Faculty Hospital and [2]KRVP RF Technical University, Košice.

A considerable inter- as well as intraindividual variability of the measured ventricular fibrillation threshold occurred in the course of our experiments. The significant rhythm of ventricular fibrilation threshold ($p<0.05$) was found under conditions of normoventilation as well as during hypoventilation. The maximal values, expressed as the acrophase, were normoventilation -338°, i.e. at 2253 h and in hypoventilation -10°, i.e. at 0040 h respectively with the dark (active) period of the light schedule. The circadian rhythm of ventricular fibrillation threshold was in accordance with the circadian rhythm of rectal temperature. These results demonstrate the fact that electrical stability of the heart is also influenced by the processes which occur in dependence on the oscillation of body temperature and that hypoventilation does not affect the circadian rhythm of electrical stability of the heart.

SPECTROTEMPORAL ANALYSIS OF THE PQ-INTERVAL OF ECG IN MAN. *B. Fišer, N. Horníková, B. Semrád[1],* Department of Physiology, [1]Fourth Department of Medicine, Medical Faculty, Masaryk University, Brno.

Spectrotemporal mapping was recently used for the analysis of ventricular late potentials (1). We used this method for the analysis of the PQ-interval of the signal-averaged high-resolution ECG (XYZ leads) in 14 healthy volunteers (time window 20 ms, resolution 2 ms). The power in the frequency band 0 – 50 Hz was plotted against time. Four distinct peaks were observed in each curve and identified as P1....P4. The mean P4-Q interval was 42.84 ± 9.8 ms. This indicated that P4 corresponded to the His potential. The P3-P4 interval was 37.12 ± 12.08 ms, P2-P4: 70.84 ± 9.84 ms, P1-P4: 103.40 ± 12.31 ms. P3 probably corresponded to the activation of the left atrium, P2 to the right atrium, P1 to the activation of the sinoatrial nodal area. Our spectrotemporal analysis technique increases the diagnostic possibilities of the signal-averaged ECG record.

1. *Lander P., Albert D.E., Berbari E.J.:* J. Electrocardiol. 23: 95-108, 1990.

DETECTION OF MYOCARDIAL INFARCTION DAMAGE BY MONOCLONAL ANTIBODIES AGAINST HUMAN CARDIAC MYOSIN LIGHT CHAINS. *Z. Straková, M. Barančík[1], T. Rada[1], K. Horáková[2], J. Styk[1],* Institute of Molecular Physiology and Genetics, [1]Institute for Heart Research, Slovak Academy of Sciences, and [2]Chemicotechnological Faculty STU, Bratislava.

Myosin is the main component of sarcomeres and after heart damage caused by myocardial infarction its degradation by proteolytic enzymes results in releasing myosin light chains (MLC). The mixture of human cardiac MLC was used as an antigen for the preparation of monoclonal antibodies (MAbs) by hybridoma technology. Using Western blotting, MAbs were specific for MLC II. Cross-reactivity with human skeletal MLC was determined by the ELISA-test. The MAbs isotype was estimated as IgM with kappa light chains. Applying an immunohistochemical method, the MAbs showed good immunoreactivity with the canine heart muscle. In a model of ischaemic reperfusion injury, it was demonstrated that MAbs against human cardiac MLC are suitable and sensitive tools useful for following heart damage, caused by myocardial infarction.

THE CONTENT OF VARIOUS LIPIDS IN THE MYOCARDIUM OF THE LEFT VENTRICLE WITH EXPERIMENTAL INFARCTION AND SUBSEQUENT VOLUME OVERLOAD. *F. Šimko, I. Vozár, A. Fízeľ, A. Fízeľová[1], J. Kyselovič[2], R. Sochorová[1],* Faculty of Medicine, Comenius University, [1]Department of Pathophysiology and [2]Department of Pharmacology, Bratislava.

A model has been introduced in order to study the capacity of adaptation of the heart with left ventricular infarction to subsequent haemodynamic overload represented by aortic regurgitation. Four groups of rabbits were investigated: control, with severe infarction, with aortic insufficiency, with both infarction and subsequent insufficiency. The levels of free fatty acids, triglycerides, cholesterol and cholesterol acetate were investigated in the left ventricular myocardium of these hearts. The content of free fatty acids was decreased in the case of aortal insufficiency. The level of triglycerides and similarly of cholesterol was decreased in all groups of overloaded hearts. The content of cholesterol acetate was not changed in any of the groups with the haemodynamic overload in comparison to control animals.

IV. Neurophysiology

INFLUENCE OF PENTYLENETETRAZOL AND CAFFEINE ON CORTICAL EPILEPTIC AFTERDISCHARGES (ADS) IN RATS. *H. Koryntová[1], P. Tutka[2], H. Kubová[2,3], P. Mareš[1,3],* [1]Department of Pathophysiology, Third Medical Faculty, Charles University, [2]Institute of Physiology, Czechoslovak Academy of Sciences and [3]Department of Pharmacology, Third Medical Faculty, Charles University, Prague.

Cortical epileptic afterdischarges were elicited in rats with implanted electrodes by electrical stimulation of the sensorimotor region. Stimulation series (15 s, 8 Hz) were repeated four times at 10-min intervals, five minutes after the end of the first AD, the drugs were injected intraperitoneally (pentylenetetrazol 10 or 20 mg/kg and caffeine 75 or 150 mg/kg). No drug was injected in control experiments. Eight rats were used for each drug injected, underwent control and two drug sessions at intervals of at least three days. The duration of ADs and motor correlates of ADs were enhanced by both drugs, the EEG pattern of ADs (the incidence of the two types of ADs) was not influenced unequivocally.

INFLUENCE OF ANTICONVULSANT DRUGS ON CORTICAL EPILEPTIC AFTERDISCHARGES (ADS). I. ANTI-cONVULSANTS EFFECTIVE AGAINST PARTIAL SEIZURES.
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Epileptic afterdischarges (ADs) elicited by rhythmic electrical stimulation of the sensorimotor cortex (8 Hz frequency for 15 s) might be used as a model for testing anticonvulsant drugs (1). In a systematic study in adult rats we examined the effects of classical anticonvulsants, with special attention to phenytoin (PHT), carbamazepine (CBZ), phenobarbital (PHB) and primidone (PRI). ADs were always elicited four times at 20-min intervals between the consecutive series, the anticonvulsants being injected i.p. 10 min after the first AD. Three doses of the same drug were tested in the same animal. Best effects were seen with PHB, other drugs were substantially less effective.
1. *Kubová H. et. al.:* Arch. int. Pharmacodyn. 307: 49-59, 1990.

INFLUENCE OF ANTICONVULSANT DRUGS ON CORTICAL EPILEPTIC AFTERDISCHARGES (ADS). II. ANTI-CONVULSANTS EFFECTIVE AGAINST GENERALIZED SEIZURES.
H. Kubová[1,2], M. Mocková[1], M. Lanštiaková[1], P. Mareš[1,2], [1]Institute of Physiology, Czechoslovak Academy of Sciences, [2]Department of Pharmacology and [3]Department of Pathophysiology, Third Medical Faculty, Charles University, Prague.

Cortical ADs in freely moving rats with implanted electrodes consist of a spike-and-wawe rhythm, accompanied by clonic seizures of the head and forelimb muscles. Clonazepam is able to suppress these seizures (1), therefore other anticonvulsants used against human primary generalized epilepsies of the absence type were studied. Both the drugs used in this study were injected intraperitoneally inbetween the first and second ADs out of the four. Valproate (VPA) in doses of 200, 300 and 400 mg/kg was effective; the action of ethosuximide (ESI, 62.5 or 125 mg/kg) was ambiguous. The efficacy of VPA may be associated with its action against generalized tonic-clonic seizures.
1. *Kubová H. et al.:* Arch. int. Pharmacodyn. 307: 49-59, 1990.

ANTICONVULSANT EFFECT OF PROGABIDE DURING ONTOGENESIS IN RATS.
L. Staňková[1], A. Kožuchová[1], P. Mareš[1,2], [1]Institute of Physiology, Czechoslovak Academy of Sciences and [2]Third Medical Faculty, Charles University, Prague.

Motor seizures elicited by pentylenetetrazol (PTZ) might be used for testing the anticonvulsant effect even in immature rats (1). Using this model we studied the action of Progabide (Synthelabo), an agonist of $GABA_A$ receptors, in rats 7, 12, 18, 25 and 90 days old. Progabide in doses from 25 to 150 mg/kg was injected intraperitoneally 10 min before PTZ (100 mg/kg s.c. in all but 18-day-old rats, where 90 mg/kg dose was given). The incidence and latency of minimal and major seizures, patterns and severity of motor seizures were recorded and evaluated. Progabide exhibited a specific action against the tonic phase of major seizures at all developmental stages studied leaving the clonic phase of major seizures as well as minimal seizures unaffected.
1. *Mareš P., Marešová D., Schickerová R.:* Physiol. Bohemoslov. 30: 113-121, 1981.

BICUCULLINE METHIODIDE-INDUCED CORTICAL FOCI IN FREELY MOVING RATS DURING DEVELOPMENT.
S. Soukupová[1], R. Mikolášová[2], H. Kubová[2,3], P. Mareš[1,2], [1]Department of Pathophysiology, Third Medical Faculty, Charles University, [2]Institute of Physiology, Czechoslovak Academy of Sciences and [3]Department of Pharmacology, Third Medical Faculty, Charles University, Prague.

Neocortical foci were induced by local application of bicuculline methiodide (BM) to the sensorimotor cortex of rats 7, 12, 18, 25 day-old and adult animals through an implanted cannula. BM was applied in the amount of 2.5 μl as a 0.5-, 1- or 2-mM solution. EEG focal discharges were recorded in all animals; at first they did not exhibit any motor symptoms, during the 20-min period isolated myoclonic jerks of the contralateral paws appeared progressively. Spontaneous transition into ictal activity, accompanied by clonic seizures of the head and forepaw muscles, was seen in a majority of rats, especially with the 1- and 2-mM concentration of BM. The same developmental changes were described as in immobilized rats.

GLUTAMATE AND BICUCULLINE METHIODIDE ENTER THE BRAIN OF IMMATURE RATS.
P. Mareš[1,2], M. Chino[1,2], H. Kubová[1,3], P. Mathern[1], M. Veliký[1,2], [1]Institute of Physiology, Czechoslovak Academy of Sciences, [2]Department of Pathophysiology and [3]Department of Pharmacology, Third Medical Faculty, Charles University, Prague.

Convulsant action of glutamate and bicuculline methiodide administered intraperitoneally was used as an indicator of the development of the blood-brain barrier. Glutamate in high doses (4mg/kg) induced minimal clonic as well as generalized tonic-clonic seizures in rats 7, 12 and 18 days old, whereas seizures appeared only exceptionally in 25-day-old and adult rats. Bicuculline methiodide was injected to 12-, 18-day-old and adult animals. Motor seizures of both types were induced reliably only in 12-day-old rats with the highest (20 mg/kg) dose, in some 18-day-old rats and never in adult animals. The two substances used gave different results which indicated that the blood-brain barrier does not mature at the same rate for various drugs.

GABA AND GLYCINE ANTIEPILEPTIC EFFECTS DO NOT DEPEND EXCLUSIVELY ON TRANSPORT ACROSS THE BLOOD BRAIN BARRIER.
M. Pohl, P. Mareš I. Lhotková, A. Součková, E. Jelínková, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

GABA or glycine are usually referred to having no antiepileptic effects following systemic administration and this is supposed to be due to their poor transport across the blood brain barrier (BBB). We used an overdose of both neurotransmitters (glycine 2 g/kg, GABA 1 g/kg i.p.) in adult and 12-day-old rats to test this hypothesis. Seizures were induced by strychnine (STR - 4 mg/kg i.p.) or pentamethylenetetrazol (PTZ-100 mg/kg s.c.) with a delay of 30 or 60 min after GABA (glycine) administration. Control animals only received the convulsant. GABA influenced minimal metrazol seizures and STR-induced seizures and the mortality in adult animals. Glycine influenced both PTZ- and STR-induced seizures in adult animals. The effects were present only in the first measured interval. These effects were only weak (if at all) in young animals. Therefore, transport across the BBB does not play the key role in the anticonvulsant effects of GABA and glycine.

PROPENTOFFYLINE (VULM 953) PROTECTION AGAINST RAT BRAIN ISCHEMIC DAMAGE. *M. Fabiánová, P. Noskovič, V. Fáberová,* Drug Research Institute, Modra.

Propentoffyline protection against ischemic and hypoxic brain damage *in vitro* and *in vivo* is well known from the literature. The aim of our study was to investigate the effect of propentoffyline on some biochemical parameters in the rat brain after 5 min global ischaemia (occlusion of four vessels). Propentoffyline was administered for two weeks p.o. (25 $mg.kg^{-1}.day^{-1}$) prior to ischaemia. Animals were divided into three experimental groups and the following parameters were tested in different brain structures. Group 1-cyclic AMP and GMP level 30 min after recirculation; group 2-lipid peroxidation (determined by TBA reactive products) 30 min after recirculation; group ^{3}H-leucine incorporation into brain proteins one hour after recirculation. Our studies have shown that repetitive application of propentoffyline significantly increases cGMP levels, prevents the postischemic increase in concentration of malondialdehyde and increases ^{3}H-leucine incorporation into brain proteins.

THE DEVELOPMENTAL EFFECT OF KETAMINE ON THE NMDA ACTIVATION OF EMBRYONIC MOTILITY. *J. Sedláček,* Institute of Physiology, First Faculty of Medicine, Charles University, Prague.

The hypothesis concerning the primary effect of ketamine by means of the NMDA-receptor complex motivated the investigation of NMDA activation of spontaneous motility in chick embryos after longlasting ketamine pretreatment. 1. A chronic continuous supply of ketamine (7.17 ± 0.37 mg/kg e.w./24 h; day 4 to 12-series C. day 4 to 16-series E) decreased the initial resting motility to 68 % of control values on the average. NMDA (20mg/kg e.w., 50 μl physiological saline) activated embryonic motility to 310.6 % on the average, with a maximum of 373.1 % of resting motility before NMDA administration. 3. In series C, a significant decrease of the NMDA enhancing effect to 214.4 % with a maximum of 268.2 % of the resting motility before NMDA administration. This effect was slightly smaller in series E. These results confirmed the assumption that ketamine intervenes in the development of embryonic spontaneous motility and do not exclude the specific connection of this effect with the development of the NMDA-receptor complex.

ACTIVITY-RELATED CHANGES IN MEMBRANE POTENTIAL AND EXTRACELLULAR K^+ IN THE RABBIT VAGUS NERVE. *A. Chvátal, P. Jirounek[1], J.-L. Balfroid[1], P.C. Brunet[1], J. Svoboda, E. Syková,* Laboratory of Cellular Neurophysiology, Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague and [1]Department of Pharmacology, University of Geneva, Geneva.

Stimulation-evoked changes in extracellular K^+ concentration ($[K^+]_e$) were studied in the rabbit vagus nerve which is composed of unmyelinated densely packed axons of very fine diameter. Using the sucrose-gap technique and K^+-selective microelectrodes, we have measured changes in membrane potential and increases in $[K^+]_e$. Stimulation (15 Hz, 30 s) produced a depolarization of 6.0 ± 0.6 mV and an increase in $[K^+]_e$ from the resting level 5.6 mM to 8.5 ± 0.3 mM (n = 11). This depolarization was followed by post-tetanic hyperpolarization (PTH) and a decrease in $[K^+]_e$ below the resting K^+ level (K^+ undershoot). Low K^+ concentration or ouabain abolished both the K^+ – undershoot and PTH. High Ca^{2+} concentrations enhanced the increase in $[K^+]_e$, but did not affect the K^+ – undershoot and PTH. Our results indicate that the electrogenic Na^+/K^+ pump is involved in the generation of PTH and in the regulation of $[K^+]_e$. The effect of Ca^{2+} suggests the participation of a Ca^{2+}-activated K^+ current in the observed activity-related changes in $[K^+]_e$.

EFFECT OF MONOAMINES AND AMINO ACID TRANSMITTERS ON NA^+, K^+ ATPase ACTIVITY IN CORTICAL ASTROCYTES AND NEURONES IN PRIMARY CULTURES. *I. Hájek, L. Hertz[1].,* Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague and [1]Department of Pharmacology, University of Saskatchewan, Saskatoon, Canada.

The effect of noradrenaline, other adrenergic agonists, biological amines and amino acid transmitters was studied on the specific Na^+, K^+ ATPase in primary cultures of astrocytes and neurones of the mouse cortex. ATPase activity was significantly increased by noradrenaline and by specific beta-ligand isoproterenol in astrocytes, but only by noradrenaline in the neurones. The highest activation occurred $[K^+]$ 6.25 mM. Ligands with alpha$_2$ activity had no effect on astrocytes and exhibited even an inhibitory effect on the neurones. Dopamine and serotonin increased ATPase activity in astrocytes significantly, while their effect on neurones was mostly inhibitory. Glutamic acid activated ATPase both in astrocytes and neurones, GABA had no effect and glycine was found to be a potent inhibitor of ATPase activity in astrocytes.

MECHANISMS OF THE ACTIVITY-RELATED ALKALINE-ACID CHANGES IN THE RAT SPINAL CORD. *E. Syková, A. Chvátal,* Laboratory of Cellular Neurophysiology, Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague.

Activity-related transient changes in extracellular pH (pH_e)were studied by means of ion-selective microelectrodes in neonatal rat spinal cords isolated from pups 2-14 days of age. Stimulation evoked an alkaline shift in 3 to 8-day-old pups, and an acid shift in 10 to 14-day-old pups. The alkaline, but not the acid, shift was blocked by Mg^{2+} and picrotoxin (10^{-6} M). Acetazolamide had no effect on the alkaline shift but blocked the acid shift. The acid shift was blocked, and the alkaline shift enhanced, by Ba^{2+}, amiloride, SITS and after postnatal X-irradiation, a procedure which blocks gliogenesis. These results suggest that the acid shifts are due to acid extrusion from glial cells. Application of GABA evoked an alkaline shift in the pH_e baseline which was blocked by picrotoxin. Activation of GABA-gated anion (Cl^-) channels, which induces a passive net efflux of bicarbonate, can lead to a fall in neuronal intracellular pH and to an alkaline shift in the pH_e.

DIFFUSION CHARACTERISTICS AND EXTRACELLULAR SPACE VOLUME CHANGES DURING ANOXIA IN THE RAT SPINAL CORD. *J. Polák, J. Svoboda, E. Syková,* Laboratory of Cellular Neurophysiology, Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague.

Difusion properties of spinal dorsal horns were studied by quantitative analysis of concentration-time profiles of iontophoretically applied tetramethylammonium (TMA^+). TMA^+ was recorded by TMA^+-sensitive mictoelectrodes (ISM) positioned 150-250 μm from the iontophoretic pipette. Simultaneously, the rise in extracellular K^+ ($[K^+]_e$) was recorded by K^+-ISMs. Three parameters characterizing diffusion were studied: extracellular space volume fraction α, tortuosity λ, and uptake k. The values in normoxic conditions were α = 0.24 ± 0.02, λ = 1.55 ± 0.04, k = 5.7 ± 0.7 x 10^{-3} (n = 9). During anoxia, produced by respiratory arrest, the parameters changed gradually, together with rise in $[K^+]_e$. In about 10-20 min the $[K^+]_e$ rose about 70 mM and the parameters reached final values of α = 0.05 ± 0.01, λ = 2.00 ± 0.08, k = 2.2 ± 0.7 x 10^{-3} (n = 6). These results show that during hypoxia and anoxia the duiffusion of various ions and substances, presumably including neurotransmitters, is impaired.

EXTRACELLULAR SPACE VOLUME IN THE RAT CORTEX DURING DEVELOPMENT AND AFTER CEREBRAL INJURY. *J. Svoboda, E. Syková, A. Lehmenkühler*[1], Laboratory of Cellular Neurophysiology, Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague and [1]Institute of Physiology, Westfalishe-Wilhems-Universität Münster, Münster.

Tetramethylammonium-selective microelectrodes (TMA^+-ISM) were used in diffusion studies with TMA^+, which remains essentially extracellular during the measurements. The extracellular space volume fraction (α) and ECS tortuosity (λ) were examined in the cortex of 2 to 21-day-old rats during development and after cortical damage avoked by X-irradiation on the first postnatal days. In control animals, a significantly larger α value was found in the early postnalal days. In 1 to 3-day-old pups $\alpha = 0.49 \pm 0.02$, in 4-6-day-old $\alpha = 0.37 \pm 0.01$, in 7-9 day-old $\alpha = 0.31 \pm 0.004$ and in 10-12 day-old $\alpha = 0.22 \pm 0.002$ (n=9, 32, 37 and 31, respectively). There was no further decrease of α in older rats. After irradiation, α increased to about 0.50-0.70 in cortical areas which were characteristic of blood-brain barrier damage, inflammation and tissue necrosis. In adjacent cortical areas, characterized by astrogliosis, the α increased to 0.35-0.50. There was no significant change in λ. Our results indicate that the ECS volume is significantly higher in the early postnatal period, reaches normal values during maturation of glia and is significantly impaired by astrogliosis and tissue injury.

ULTRAHISTOCHEMICAL STUDY OF THE ISCHAEMIC RAT BRAIN. *P. Jalč, M. Kolaj, J. Maršala,* Institute of Neurobiology, Slovak Academy of Sciences, Košice.

Upon reperfusion, two circulatory disturbances are known to occur after postischaemia, namely reactive hyperaemia or hypoperfusion. The role of reactive hyperaemia in the opening of the blood-brain barrier and induction of oedema was demonstrated following middle cerebral reperfusion following ischaemia, while breathing 100 % oxygen, also markedly increases lipid peroxidation, whereas a decrease in paO_2 during early perfusion improves metabolic and functional recovery of the brain after ischaemia. The present study has shown that the ultrastructural changes were profound in all the experimental groups and could be classified as irreversible changes. A disturbance of calcium homeostasis was observed only in the group with postischaemic hyperaemia.

STUDY OF PHOSPHOLIPIDS IN SUBCELLULAR FRACTIONS OF RABBIT CNS FOLLOWING A LETHAL DOSE OF PENTOBARBITAL USING NUCLEAR MAGNETIC RESONANCE. *D. Dobrota, Š. Macháč, A. Drgová, V. Mézešová, V. Mézeš, F. Saloň, V. Lombardi*[1], *T. Liptaj*[2], *L. Zalibera*[2], Department of Biochemistry, Jesenius Medical Faculty, Comenius University, Martin, [1]Department of Pathological Anatomy, Institute of Oncology, Bari, Italy and [2]Faculty of Chemistry and Chemical Engineering, Slovak Technical University, Bratislava.

The aim of this work was to evaluate the possible changes of phospholipids in subcellular fractions of the rabbit CNS after administering a lethal dose (LD) of pentobarbital. Fifteen rabbits were decapitated and 15 rabbits were killed by an i.v. injection of LD of pentobarbital (150mg/kg b.w.). Subcellular fractions of the rabbits' CNS were assayed for ^{31}P spectra. No changes were found in ^{31}P spectra of phospholipids in subcellular fractions of the rabbits' CNS euthanized by LD of pentobarbital after comparison with the control group (decapitated rabbits). It is concluded that it is unlikely that general anaesthetics can act by dissolving lipids.

THERE IS A DIFFERENCE IN POTASSIUM EVOKED CATECHOLAMINE CONCENTRATION CHANGES IN THE RETICULAR FORMATION AS COMPARED WITH THE CORPUS STRIATUM OF RAT: A VOLTAMMETRIC STUDY. *J. Pavlásek, C. Mašánová, P. Bielik,* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

In anaesthetized rats application of 5 μl 0.5 $mol.l^{-1}$ KCl to the vicinity of the recording electrode in the reticular formation substantially depressed multiunit activity. This started to reappear in the 7th min after a microinjection and attained its control level in the 9th min. There was a decrease of catechol oxidation current (CA.OC) during the first minute after KCl application to 59 % of the control (mean, n=4). Consecutive recordings (taken at regular 3 min intervals) showed minimal CA.OC value (23 %) in the 7th min. At the end of the investigated interval (24 min) CA.OC ranged from 45 to 80 % of the controls. The reaction in the corpus striatum exhibited a two-phase course. Immediately after a microinjection of KCl (1st min) CA.OC rose to 62 % of the control (n=5). Subsequent recordings (1 min intervals) displayed a drop of CA.OC signal (21-63 %) in the second phase of the investigated interval (3rd-10th min). The relationship between the observed phenomena and a wave of the spreading depression is discussed.

V. Sensory Physiology

FREQUENCY ANALYSIS OF VOCALIZATIONS IN GUINEA-PIGS. *J. Syka, A. Vlková,* Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague.

Guinea-pigs emit several acoustic signals – vocalizations – which serve for interindividual communication as well as for expression of the animal's emotional state. We analysed the frequency content of emitted signals with a Fast Fourier Transformation program (Waterfal, Cambridge Electronics Design) supported by an intelligent terminal and a personal computer. Three basic types of vocal signals were found in guinea-pigs: purr, whistle and chirp. Purr represents series of brief acoustic pulses with a broad band frequency spectrum (dominant frequency less than 1 kHz). Whistle is characterized by three or more frequency components, the frequency of which increases during vocalization. Chirps consist of two or three stable frequency components. Vocalizations produced by electrical stimulation of different brain sites are more simple than spontaneous calls: only the basic frequency component is usually present and its frequency is lower than in corresponding spontaneous vocalizations.

AMPLITUDE ENHANCEMENT OF CORTICAL AUDITORY EVOKED RESPONSES IN AWAKE GUINEA-PIGS AFTER NOISE EXPOSURE. *N. Rybalko, J. Syka, J. Popelář,* Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague.

We have previously shown (1) that the amplitudes of auditory-cortex evoked responses (ACER) in awake guinea-pigs were enhanced after noise exposure, while the evoked responses recorded from the inferior colliculus or from the auditory nerve decreased. In the present study this phenomenon was analysed in more details. Guinea-pigs were exposed to white noise, intensity of which ranged from 105 dB to 125 dB for 30 min or one hour. Thresholds of ACER and their amplitudes increased after the noise exposure proportionally to the exposure intensity. Maximal amplitude enhancement amounted to 500 % of the preexposure level. In some experiments, electrodes were also implanted into the colliculus inferior and onto the visual cortex (to record visually evoked responses). No amplitude enhancement of the evoked responses was observed in these brain structures.

1. *Popelář J., Syka J., Berndt H.:* Hearing Res. 26: 239-247, 1989.

PLASTICITY OF AUDITORY-CORTEX EVOKED RESPONSES IN GUINEA-PIGS AFTER UNILATERAL DESTRUCTION OF THE INNER EAR. *J. Popelář, Y. Cazals[1], J.-P. Erre[1],* Institute of Experimental Medicine, Czechoslovak Academy of Sciences, Prague and [1]Laboratory of Experimental Audiology, INSERM, Bordeaux.

Several authors have reported the reorganisation of the auditory cortex after partial or total deafferentation in developing animals. The aim of our study was to evaluate the changes of evoked responses recorded in the contralateral and ipsilateral auditory cortex (ACER) after unilateral ear destruction in adult guinea-pigs. In control animals, the ACER recorded in the ipsilateral auditory cortex had elevated thresholds and depressed amplitudes at higher frequencies (>1 kHz). After intracochlear injection of sisomicide, which totally destroyed sensory hair cells, ipsilateral thresholds decreased and ACER amplitudes increased rapidly within a few days. The changes of ACER responses after interruption of middle ear ossicles or after mechanical destruction of the cochlea appeared two days later and progressed more slowly. Our results demonstrate that the functional changes of ACER after unilateral deafferentation can occur within a few days in adult animals.

FUNCTIONAL DISTRIBUTION OF INFERIOR COLLICULUS NEURONES IN THE GUINEA-PIG. *J. Aštl, J. Popelář[1], R. Druga[2], J. Syka[1],* ENT Clinic, Charles University. [1]Institute of Experimental Medicine, Czechoslovak Academy of Sciences and [2]Institute of Anatomy, Faculty of Medicine, Charles University, Prague.

Distribution of neuronal responses in the inferior colliculus (IC) was evaluated in anaesthetized guinea-pigs. The isofrequency layers decline in the mediolateral direction within the rostral and central parts of the IC, whereas they decline in a reverse direction in the caudal part. Units with the excitatory-excitatory type of binaural interaction were found more frequently in the low-frequency dorsal part of the IC, whereas neurones with the excitatory-inhibitory type of response were observed more usually in the high-frequency ventral part. Electrical stimulation of the auditory cortex evoked responses in 40 % of IC neurones, the responses were unlike in the rat found also in the central part of the IC. Our results suggest that the functional distribution of neurones in the guinea-pig IC is different from that described in the rat.

AUDITORY BRAINSTEM-EVOKED POTENTIALS IN THE NEWBORN INFANT: A THREE-CHANNEL LISSAJOUS' TRAJECTORY STUDY. *A. Smiešková, J. Magula,* Medical Informatics Reserach Institute, Bratislava.

It is known from experimental and clinical work that separate components evoked potentials in the auditory brainstem (ABEPs) are generated at various levels of the nervous system, in more or less distinct anatomical structures of the auditory pathway (1). ABEPs were recorded from newborns using three orthogonal differential electrode pairs, in addition to the widely used vertex-mastoid derivation. The interpretation of these results is enhanced in relation to the question of ABEP generators, by using three channel Lissajous' trajectories descriptors of ABEP (2), which are more comprehensive than their single-channel counterparts.

1. *Buchwald J.S., Hunag C.M.:* Science 189: 382-384, 1975.
2. *Pratt H., Har' El Z., Golos E.:* Electroenceph. Clin. Neurophysiol. 56: 682-688, 1983.

THE EFFECT OF METAZOSINE HYDROCHLORIDE ON INTRAOCULAR PRESSURE IN RABBITS AND MONKEYS. *J. Čepelík, I.. Helfort[1], S. Hynie, V. Trčka[1],* Department of Pharmacology, First Medical Faculty, Charles University and [1]Research Institute for Pharmacy and Biochemistry, Prague.

The influence of topical administration of an alpha$_1$-adrenergic antagonist, metazosine hydrochloride (MTZ), on intraocular pressure (IOP) and pupil diameter was tested in conscious normal rabbits and monkeys. IOP was assessed with an applanation tonometer and the pupil diameter was measured with a plastic ruler. In rabbits, MTZ decreased pupil diameter moderately and transiently. However, this drug potently antagonized the mydriatic effect of an adrenergic agonist, metoxamine. Topically applied MTZ decreased IOP in both rabbits and monkeys in a dose-dependent manner. The IOP lowering effect of MTZ was not accompanied by any decrease of blood pressure in either of these two animal species. It was concluded that MTZ markedly decreases IOP both in rabbits and monkeys. It is therefore suggested that MTZ should be tested as an antiglaucoma agent.

INTERACTION OF VESTIBULAR AND PROPRIOCEPTIVE INPUTS FOR HUMAN SELF-MOTION PERCEPTION. *F. Hlavačka, T. Mergner[1], T. Schweigart[1],* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava and [1]Neurological University Clinic, Freiburg, Germany.

Human perception of horizontal self(body)-motion in space was studied during various combinations of vestibular and leg-proprioceptive stimuli in the dark. During passive sinusoidal rotations of the trunk relative to the stationary feet (functionally synergistic combination), the perception was almost veridical over the frequency range tested (0.025-0.4 Hz). This finding suggested a dominance of the proprioceptive over the vestibular input, since the perception: a) closely reflected quantitatively (gain, phase and detection threshold) the proprioceptive input (i.e., the subjects' foot-to-trunk perception during proprioceptive stimulation) and b) it clearly differed from self-motion perception during vestibular stimulation. However, when using other combinations, self-motion perception changed in a monotonous way as a function of the two inputs. This indicated that the inputs do interact in a linear manner. The findings are interpreted in terms of a mathematical model.

VI. Physiology of Blood Circulation

AORTIC STRIP CONTRACTION IN SHR AND NORMOTENSIVE BN.1X RAT STRAINS. *N. Gaier, S. Hynie, M. Pravenec[1], V. Bílá, P. Klír[1], V. Křen,* First Medical Faculty, Charles University and [1]Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Isometric aortic contractions were studied in progenitors of two sets of recombinant inbred strains, namely in spontaneously hypertensive (SHR) and normotensive polydactylous BN.1x rat strains. The contractions were studied in the the Krebs-Henseleit medium (pH 7.4) at 37 °C and recorded at a basic tension of 1 g. The maximal response of particular strip to cumulative doses of norepinephrine (NE) was considered as 100 %. EC_{50} of NE in the BN.1x rat strain was about 3-times lower ($3x10^{-9}$) than in the SHR rat strain. The alpha$_2$ adrenergic agonist clonidine (CLO) led to contraction of aortic strips which reached about 50-60 % effect of NE in both rat strains. However, SHR rats were more sensitive in CLO than BN.1x rats. These results suggest that altered aortic responsiveness of SHR, compared to BN.1x, might be related to changes in the proportion or responsiveness of α_1 and α_2 receptors in these strains.

THE CONTROL OF CHOLESTEROL ESTERIFICATION RATE IN THE PLASMA BY THE PROPORTION OF HDL2 AND HDL3 SUBCLASSES IN HEALTHY SUBJECTS AND HYPERTENSIVE PATIENTS. *M. Dobiášová, J. Stříbrná[1], P.H. Pritchard[2], J.J. Frolich[2],* Institute of Nuclear Biology and Radiochemistry, Czechoslovak Academy of Sciences, [1]Institute for Clinical and Experimental Medicine, Prague and [2]University Hospital Lipoprotein Research Group, University of British Columbia, Vancouver.

The relation between the fractional rate of cholesterol esterification (FER_{HDL}) in very low (VLDL) and low (LDL) density lipoproteins depleted the plasma and the particle size distribution of high density lipoproteins (HDL) were studied in a) septuagenaerians who had no clinical signs of atherosclerosis, b) outpatients with essential hypertension and c) control groups of healthy men and women. There was a highly significant increase in FER_{HDL} in patients with hypertension compared to healthy subjects. The HDL of hypertensive patients had a markedly increased relative content of HDL3b, while HDL2B fraction was reduced by over 50 % compared to the other groups. Overall, there was a strong positive correlation between FER_{HDL} and HDL3b and a negative correlation between FER_{HDL}, HDL3a and HDL2b.

THE PROLIFERATIVE RESPONSE TO NORADRENALINE IS DIFFERENT IN MALE AND FEMALE RAT VASCULAR SMOOTH MUSCLE CELLS (SMC) IN CULTURE. *L. Bačáková, M. Baudyšová,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

It was found in rats that the proliferative capacity of vascular SMC is higher in males than in females (1). This phenomenon may be related to a greater susceptibility of men to some vascular diseases. One of its causes might be a different reactivity to hormones in SMC of both sexes. We compared the proliferative response to noradrenaline (10^{-6} M) in culture of male and female SMC (the 11th passage, medium with 10 % of foetal calf serum) derived from the thoracic aorta of Wistar rats (200 g, SPF). In the male SMC, noradrenaline shortened the doubling time (90±1 % of the control) and increased maximum population density (126±8 %). However, in the female SMC, the doubling time was longer (143±20 %) and the maximum population density lower (74±8) than in the control. Similar differences were obtained in conventionally raised rats which, however, were smaller. Thus, the role of hormones upon different growth properties of male and female vascular SMC cannot be excluded.

1. *Bačáková L., Baudyšová M.:* Physiol. Bohemoslov. 39: 449-458, 1990.

DILATION AND HYPERTROPHY OF BRONCHIAL ARTERIES 12 AND 90 DAYS AFTER EMBOLISATION OF THE PULMONARY ARTERY IN THE DOG. *J. Jandík, J. Sedláček, B. Ulybin, J. Mráz, H. de Geest[1],* Faculty of Medicine, Charles University, Hradec Králové and [1]Catholic University, Leuven, Belgium.

The aim of this study was to compare the lumen and the wall thickness of bronchial arteries (BA) 12 and 90 days after experimental embolisation of the pulmonary artery (PA). Embolisation of PA was performed with sterile plastic spheres of 1 mm in diameter in 24 dogs. Twelve dogs were killed after 12 days and the other dogs 90 days after embolisation of the PA. The following morphometric data were measured from histological mounts: 1. diameter of the bronchi (DB); 2. diameter of adjacent BA (DBA); 3. and the wall thickness (WT) of BA. Control morphometric data were obtained from 6 dogs without embolisation of the PA. Indexes DBA/DB and WT/DB were calculated from the morphometric data and then statistically analysed. Twelve days after embolisation of the PA, the BA were dilated and hypertrophic in comparison with those of control dogs ($p<0.01$). More conspicuous dilation and hypertrophy of the BA occurred 90 days after embolisation of the PA in comparison with the control dogs and the dogs 12 days after embolisation ($p<0.01$). At 90 days, dilation was more prominent than hypertrophy.

RESPONSIVENESS OF THE ILIAC ARTERY AFTER LONG-TERM RESTRICTION OF THE BLOOD SUPPLY. *A. Holécyová, O. Hudlická[1],* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava and [1]Department of Physiology, University of Birmingham, Birmingham, U.K.

Dawson and Hudlická (1) described reactive changes in the vascular bed of skeletal muscles induced by long-term limitation of blood suply. The object of this study was the responsiveness of the respective conduit artery, supplying the microcirculatory area, exposed to the limited blood supply. Isometric tension of the right common iliac artery in the rat was monitored 2 and 5 weeks after ligation. The results were as follows: 1. the maximum contraction to noradrenaline decreased, however, the concentration-response curves to the drug were shifted to the left; 2. maximum contraction to KCl decreased and 3. endothelium-dependent (acetylcholine) as well as independent (sodium nitroprusside) relaxations were altered.

1. *Dawson J.M., Hudlická O.:* Cardiovasc. Res. 24: 390-395, 1990.

INSENSITIVITY OF SKELETAL MUSCLE ARTERIOLES TO FLOW-RELATED SHEAR STRESS AFTER MOTOR NERVE STIMULATION. *V. Smieško, M. Cábel, P.C. Johnson[1],* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava and [1]College of Medicine, University of Arizona, Tuscon, U.S.A.

An increase in blood flow velocity (BFV) through arterioles induces their dilation. To investigate whether this control mechanism could participate in functional hyperaemia, an effect of 5-20 s motor nerve stimulation (MNS) on arterioles in the rat cremaster muscle was studied. Arteriolar diameter and red cell velocity were monitored with a dual-slit video microscope system. The MNS induced a significant increase in BFV (by 100-350 %) and dilation of the arterioles (by 10-80 %). The early changes in BFV – started immediately after the MNS – were followed by late phasic changes. During these late BFV changes, arteriolar diameter remained constant or changed in the opposite direction to BFV. It may be concluded that the BFV-dependent control of arterioles appears to be "switched off" after MNS, perhaps due to a temporary insensitivity of the endothelium to the flow-related shear stress.

RNA SYNTHESIS IN THE CORONARY WALL FOLLOWS THE INTRAVASCULAR PRESSURE. *O. Pecháňová, M. Gerová, V. Stoev, M. Kittová[1], I. Bernátová,* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences and [1]Institute of Physiology, Comenius University, Bratislava.

An increase in RNA synthesis with no change in DNA in the coronary wall was found during a pressure overload of the heart lasting 3-4 hours (1). It may be conceivable that the increase of total RNA content is transient and RNA synthesis might oscillate in relation to the pressure load in the artery. In 6 anaesthetised dogs, similarly as in previous experiments (1), mean BP was increased by aortic stenosis for a period of 3 hours from 130.6±7.2 to 164.0±5.9 mm Hg. Then the aortic stenosis was released and after 2 hours of recovery BP reached 92.9±7.3 mm Hg. The total content of RNA in the coronary artery after two hours of recovery was 1.63±0.16 mg/g tissue, a value significantly lower than that found during the period of increased BP (2.53±0.04 mg/g tissue, $p<0.001$).

1. *Pecháňová O. Gerová M., Bárta E., Kožík J.:* Abstracts p. 77, Regional Meeting IUPS, Prague 1991.

THE EFFECT OF COLD STORAGE (4 °C) ON THE STRUCTURE AND REACTIVITY OF THE RABBIT THORACIC AORTA. *F. Kristek, J. Török,* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

Changes in ultrastructure of endothelial cells, smooth muscle cells and nerve terminals (NT) were studied in the rabbit thoracic aorta after 2-8 days stored in cold Krebs solution at 4 °C in a refrigerator. The most prominent changes were found in NT. After 2 days, the axoplasm of NT become pale and the axolemma was interrupted in many places. Vesicles in the varicosities were still present. NT were completely destroyed after 6 days. Contraction of the aorta induced by electrical stimulation at 8 Hz decreased by 53.0±7.2 % after 2 days and was totally abolished after 6 days. Endothelial cells were slightly affected after 2 days (moderately vacuolized mitochondria). Changes proceed with the duration of cold storage. After 8 days, only membranous material was present. The structure of smooth muscle cells was only partially changed after 8 days. Furthermore, the contraction to exogenous noradrenaline was not significantly changed during the whole period studied.

EXCITABILITY OF AFFERENT FIBRES IN THE SCIATIC NERVE UNDER CONDITIONS OF HAEMORRHAGIC HYPOVOLAEMIA. *I. Bračoková, M. Mjrín, S. Kujaník, I. Linder,* Institute of Physiology, Medical Faculty, Šafárik University, Košice.

Character of the blood pressure response during stimulation of afferent fibres of the sciatic nerve in normal cats is dependent on the stimulation parameters. A low intensity current evokes a hypotensive response of blood pressure, whereas the high sensitivity current causes a hypertensive reaction. The breathing changes are variable. The objective of this paper was to analyze the effect of stimulation of afferent fibres of the sciatic nerve under conditions of haemorhagic hypovolaemia induced by 10-30 % of the total blood volume loss in cats. Our study concentrated on the estimation of blood pressure changes and their latent time as well as of breathing, the excitability threshold of the sciatic nerve and threshold of the reversal of the blood pressure response (the change of the hypotensive response to a hypertensive one). Our results show that haemorrhagic hypovolaemia lowers the excitability threshold of the sciatic nerve, it reduces the threshold of blood pressure reversal and shortens the latent time of the onset of haemodynamic responses.

BLOOD PRESSURE AND ITS REACTIVITY IN NEONATES WITH DISTURBED FETO-PLACENTAL CIRCULATION. *D. Andrásyová, E. Kellerová, E. Arendášová, I. Rusňák[1], P. Kleskeň[1],* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences and [1]Department of Gynecology and Obstetrics, Dérer's Hospital, Bratislava.

The study was performed to correlate the blood pressure (BP) and heart rate (HR) of the neonates with the condition of feto-placental circulation (F-P). The BP and HR were investigated in the resting supine and head-up position in a group of 13 neonates with serious disturbances of F-P circulation (documented by pulsed-wave Doppler ultrasound, repetitively in the 2nd and 3rd trimester of foetal life) and in a control group of 28 physiological neonates with verified normal umbilical circulation. In spite of the comparable gestational age of both groups (37-42 weeks), the body weight of neonates (3075±470 g) with disturbed F-P circulation was lower than that of the controls (3530±433 g). The values of resting BP on the 3rd postnatal day were surprisingly identical 65±8/39±7 mm Hg (in controls 66±10/37±7 mm Hg). But newborns with impaired F-P circulation showed higher systolic BP and HR reactivity to head-up body position with a BP increase by 11/4 mm Hg and HR by 13 beats as compared to 2/2 mm Hg and 9 beats per minute in the controls. This "hypertensive type" of reaction was more frequent in 86 % of subjects (68 % in controls). The results of this pilot study outline a BP hypereactivity in neonates with abnormal F-P circulation. Further detailed studies are needed in this important area of perinatal physiology.

THE POSTURAL CHANGES OF BLOOD PRESSURE AND HEART RATE IN PRESCHOOL CHILDREN. *V. Regecová, E. Kellerová, A. Hrabovská[1],* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences and [1]Philosphical Faculty, Comenius University, Bratislava.

The reactivity of blood pressure (BP) and heart rate (HR) to changed body position from the supine to the sitting position and standing was investigated in 1500 children aged 3-7 years. The type of this reaction in the 3-year-old group was mostly (in 75 %) the so-called "hypertonic" group with a significant rise of SBP by 5-10 mm Hg on the average, of DBP by 2-3 mm Hg and HR by 6-12 beats per min, with negative correlation to the corresponding initial values (r=-0.3). The presence of this type of reaction gradually decreased with age to 50 %. In a number of children whose DBP dropped in the upright position, it doubled in the age group of 6 years. The results correspond with our previous observations in neonates and document ontogenetic changes of the orthostatic reaction.

EFFECT OF COOLING ON THE ALPHA-ADRENERGIC RESPONSE IN RABBIT PULMONARY AND MESENTERIC ARTERIES. *J. Török, J. Kolárik,* Institute of Normal and Pathological Physiology, Slovak Academy of Sciences, Bratislava.

Experiments were designed to assess the effects of acute moderate cooling on alpha-adrenergic receptor-mediated contraction in rabbit pulmonary and mesenteric arteries. The arteries were cut into rings, suspended in organ baths, and connected to force transducers for the recording of isometric tension. In quiescent rings, cooling from 37 °C to 24 °C decreased basal tension of both vessels. Cooling depressed the contractile response evoked by exogenous noradrenaline (10^{-6} mol/l) in the pulmonary artery but augmented the response evoked by the agonist in the mesenteric artery. This was due to a significant enhancement of both the phasic and the tonic components of the response. In contrast to the pulmonary artery, the sensitivity of postjunctional alpha-adrenergic receptors to cooling in the mesentric artery was also increased. These results show that cooling affected the alpha-adrenergic response in pulmonary and mesentric arteries differently.

DEMONSTRATION OF THE SIMILAR EFFECTS OF EXTERNAL MAGNESIUM AND CALCIUM ON INTESTINAL AND VASCULAR SMOOTH MUSCLE CONTRACTION. *J. Kyselovič,* Department of Pharmacology, Faculty of Medicine, Comenius University, Bratislava.

The result of the study demonstrate the similar effects of external addition of $CaCl_2$ (1 mmol.l^{-1}) and $MgCl_2$ (1 mmol.l^{-1}) on smooth muscles. The experiments were performed on isolated guinea-pig terminal ileum and rings of the rabbit renal artery that were contracted by acetylcholine (10^{-6} mol.l^{-1}) and noradrenaline (10^{-5} mol.l^{-1}), respectively. Magnesium or calcium was added 1, 3, 5 and 10 minutes before the contraction tests. Application of magnesium before contraction not only demonstrated its relaxant effect on smooth muscle contraction but also potentiation of the fast component of the contractile responses in experiments on vascular and intestinal tissues. A similar modulation of the phasic and the tonic components of contraction induced calcium were also observed.

THE EFFECT OF REMOVING ENDOTHELIAL CELLS ON THE REACTIVITY OF PERFUSED RABBIT VESSELS. *V. Kristová, M. Kriška, E. Hejdová, R. Caňová,* Department of Pharmacology, Faculty of Medicine, Comenius University, Bratislava.

Our previous investigations have shown that removal of endothelial cells by air bubles enhances the contractile responses of isolated rabbit femoral arteries to noradrenaline, angiotensin and histamine (1). The present study was conducted in order to evaluate the influence of endothelial losses on the vascular responses in different types of rabbit peripheral vessels: the isolated ear vascular bed and isolated central ear artery. We evaluated the reactivity to increasing noradrenaline dosed in both ear preparations before and after de-endothelialization by air bubbles. Each preparation was tested for the presence or absence of the functional endothelium by acetylcholine. We found that the reactivity of de-endothelialized rabbit ear vessels was not changed. The results indicate that the reactivity in the two vascular beds after removing the endothelium is different.

1. *Kriška M., Babál P., Kristová V., Caňová R.:* Čs. fyziol. 38: 153, 1989.

EFFECT OF COMBINATION OF VERAPAMIL AND MAGNESIUM ON PRECONTRACTED RABBIT FEMORAL AND RENAL ARTERIES. *V. Hrková, J. Kyselovič,* Department of Pharmacology, Faculty of Medicine, Comenius University, Bratislava.

This study demonstrates the relaxant effects of combination of verapamil ($10^{-9} - 10^{-5}$ mol.l^{-1}) and $MgCl_2$ (1 mmol.l^{-1}) on rabbit femoral and renal arteries precontracted by KCl, noradrenaline or prostaglandin F_{2alfa}. The relaxant responses of the vessels were significantly higher after administration of a combination of verapamil and magnesium than to drugs administered alone. After the addition of $MgCl_2$, an approximately 1000 times lower dose of verapamil was necessary for obtaining the same degreee of arterial relaxation. The increased vasodilatory effect on the drug combination was more pronounced in arteries precontracted by KCl or noradrenaline than in arteries precontracted by prostaglandin F_{2alfa}. This synergistic activity could be explained by the different effects of organic calcium channel blockers and magnesium cations on contraction-relaxation processes of blood vessels.

INFLUENCE OF ENALAPRIL ON PLASMA LIPIDS AND FREE OXYGEN RADICALS IN PATIENTS WITH HYPERTENSION. *V. Soška, J. Siegelová, A. Zechmeister[1], A. Lojek[2],* Department of Clinical Biochemistry, Faculty Hospital, 3rd Department fo Medicine, [1]Department of Anatomy, Faculty of Medicine, Masaryk University and [2]Institute of Biophysics, Czechoslovak Academy of Sciences, Brno.

The aim of the study was to determine the effects of enalapril on free oxygen radicals (OFR) production in the blood and on plasma lipid levels. We examined 10 patients with essential hypertension II (EH) (WHO) and 10 healthy subjects. Hypertensive patients were examined after placebo treatment and then after a 2-month enalapril treatment. Healthy subjects were examined at the same time. Spontaneous and stimulated production of FOR in the blood was assessed by the method of luminol-dependent chemiluminiscence, plasma lipid levels were determined enzymatically. We found a significant decrease in triglyceride levels only after enalapril treatment. We did not find significant differences in FOR production between hypertensives and normotensives. Treatment with enalapril had no significant influence on FOR production in the blood.

CAN A DECREASE IN BLOOD PRESSURE CHANGE THE EXCRETION OF N-ACETYL-B-D-GLUCOSAMINIDASE? *K. Ševela, A. Vašků[1], J. Siegelová[1], L. Zahradníček,* Second Department of Medicine and [1]Institute of Pathological Physiology, Faculty of Medicine, Masaryk University, Brno.

The aim of the study was to detect the nephrotoxicity and/or ischaemic damage during enalapril (E) treatment. Ten patients with nephrogenous hypertension were treated 3 weeks with enalapril (5-80 mg/day). The blood pressure and N-acetyl-b-D-glucosaminidase (NAG) in the urine were measured every three days. NAG was assayed fluorometrically by a modification of the method of Leaback and Walker (1). We found correlations ($r = 0.48$, $n = 70$) between the diastolic blood pressure decrease during three days (mm Hg) and the NAG excretion (nkat per day 1). There was no correlation ($r = -0.26$, $n = 70$) between E and NAG excretion as well as no increase in NAG excretion during E treatment. Our results imply that the ischaemic damage of proximal tubular cells can become manifest when a sudden decrease in blood pressure occurs.

1. *Leaback D.H., Walker P.G.:* J. Biochem. 78: 151, 1961.

CIRCADIAN VARIATIONS OF BAROREFLEX HEART RATE SENSITIVITY IN MAN. *J. Siegelová, B. Fišer[1], J. Dušek, F. Halberg[2], G. Cornellissen[2],* Department of Pathological Physiology, [1]Department of Physiology, Faculty of Medicine, Masaryk University and [2]Chronobiological Laboratories, University of Minnesota, USA.

The aim of the present paper was to study circadian variations of the baroreceptor heart rate reflex sensitivity (BRS). We have used a noninvasive method developed recently (1, 2). Ten healthy volunteers were examined every 4 hours during a 24-hour period. The values ranged from 0.06 to 34 ms/mm Hg during the day and from 2.8 to 83.3 ms/mm Hg during the night. The difference between night and day was seen only in 4 subjects with a higher correlation between heart beat interval and BRS ($r > 0.65$). Population cosinor analysis confirmed the circadian rhythm in systolic and diastolic blood pressure. These findings contradicted the hypothesis that high BRS is a cause of the low pressure during the night.

1. *Peňáz J.:* CS patent, 1969.
2. *Fišer B., Honzíková N.:* Arch. Int. Physiol. Biochim. et de Biophys. 99: A148, 1991.

VII. Metabolism, Digestion and Physiology of Blood

PHOSPHOLIPIDOSIS INDUCED IN ISOLATED MAST CELLS DURING STIMULATION AND INHIBITION. *R. Nosál, K. Drábiková, J. Pečivová,* Institute of Experimental Pharmacology, Slovak Academy of Sciences, Bratislava.

Isolated rat mast cells (IRMC) liberate histamine from intracellular granules during stimulation. Amphiphilic beta-adrenoceptor blocking drugs inhibited stimulated histamine liberation by non-receptor mediated interaction (1,2). Stimulation of IRMC with compound 48/80 resulted in an increased incorporation of ^{32}P into fractions of membrane phosphatidylinositol (PI) and phosphatidylcholine (PC), as well as in increased liberation of archidonic acid (AA) and increased formation of thromboxane (TXB_2) from membrane phospholipids. Metipranolol (MET) increased the incorporation of ^{32}P into PI and decreased it into PC in stimulated IRMC. Moreover, MET decreased 48/80-induced AA liberation and TXB_2 formation in stimulated cells. The results indicate that phospholipidosis may be involved in the inhibition of stimulated IRMC by MET at the membrane phospholipid level.

1. *Nosál R. et al.:* Agent Action 27: 36-38, 1989.
2. *Pečivová J. et al.:* Agent Action 33: 41-43, 1991.

CARNITINE LEVELS AND SEVERAL LIPID PARAMETERS IN THE BLOOD FROM THE UMBILICAL CORD AND FROM VENOUS BLOOD OF NEWBORNS AND THEIR MOTHERS, AND IN MATERNAL MILK. *M. Hromadová, V. Parrák[1], M. Hutiová[1],* Institute of Experimental Endocrinology, Slovak Academy of Sciences and [1]Institute of National Health, Department of Neonatology, Bratislava.

To obtain a more complex picture of fat metabolism in healthy newborns, fundamental characteristics of this metabolism as well as the level of carnitine in the blood of newborns was correlated with corresponsing values from the umbilical cord and in both maternal blood and milk. The concentrations of CHOL, TGL and Apo-B in 4-day-old infants were higher than in the cord blood ($p<0.01$, and $p<0.001$, respectively], but corresponded to only 50 % of those in the blood of the respective mothers. No differences in HDL-chol were observed, whereas the level of Apo-Al in cord blood exceeded that in the blood of newborns [$p<0.05$]. No differences were registered in carnitine levels between cord-newborn blood but the content of this compound in milk was found higher by 100 % in milk that in maternal blood $p<0.001$), as a consequence of the high acylcarnitine content in milk. This work should serve as a basis for comparison with subjects exhibiting altered metabolism, especially in hypotrophic individuals.

INFLUENCE OF DIPYRIDAMOLE AND ADENOSINE MONOPHOSPHATE ON THE LEVEL OF DNA DEGRADATION PRODUCTS IN GAMMA-IRRADIATED MICE. *J. Boháček, B. Hošek, J. Šikulová, M. Pospíšil,* Institute of Biophysics, Czechoslovak Academy of Sciences, Brno.

Adenine nucleotides exert radioprotective effects when given to mice shortly before irradiation and these effects are enhanced by previous administration of dipyridamole (DP). In our contribution, the level of thymidine (Thd) in the plasma and soluble polynucleotides in the thymus and spleen were used as indicators of early cellular damage and of radioprotective effectiveness of DP and AMP. Experiments were carried out on $(CBAx57BL/10)F_1$ mice. DP (2 mg s.c.) was administered 20 min before injection of AMP (5 mg i.p.), AMP was administered 20 min before irradiation (^{60}Co, dose rate 0.38 Gy/min). Thd and polynucleotides were estimated 4 h after irradiation. The elevated level of Thd observed in the plasma of irradiated controls decreased significantly (to 60 %) in animals treated with DP and AMP. The decrease of soluble polynucleotides levels was found in the thymus. The mediator of the radioprotective action of DP and AMP seems to be adenosine, which induces systemic vasodilation, accompanied by hypothermia and circulatory hypoxia.

SHORT- AND LONG-TERM EXPOSURE OF MALE RATS TO ESTRADIOL; EFFECTS OF CATECHOLAMINES AND INSULIN ON LIPOLYSIS IN VITRO. *D. Mišeková, D. Lincová, S. Hynie,* Department of Pharmacology, First Medical Faculty, Charles University, Prague.

The purpose of this work was to compare the short- and long-term effects of estradiol (E) on adrenergically mediated lipolysis in rat epididymal adipose tissue (AT) *in vitro*. Short-term exposure (24 h) of rats to E (s.c. 0.2 $mg.kg^{-1}$) significantly increased the lipolytic effect of *in vitro* applied isoprenaline (ISO) and noradrenaline (NOR). Insulin administered *in vivo* (s.c. 100 $\mu g.kg^{-1}$) or added directly to the incubation medium (10 $\mu g.ml^{-1}$) inhibited the enhancing effect of E on ISO-induced lipolysis but only slightly attenuated the lipolytic effect of ISO in AT of control rats. When rats were treated with E for 7 or 13 days, the enhancing effect of E on adrenergic lipolysis ceased and the lipolytic dose response curve of NOR was found unchanged by insulin applied *in vitro*. The results demonstrate that E has only a transient enhancing effect on adrenergic lipolysis and insulin blocks this effect by a direct action on AT.

THE DEMONSTRATION OF α_2 ADRENERGIC MECHANISM ALSO IN RAT ADRENERGIC LIPOLYSIS IN VITRO. *D. Lincová, D. Mišeková, S. Hynie,* Department of Pharmacology, First Medical Facuty, Charles University, Prague.

From literary data the participation of the α_2 adrenergic mechanism in rat adrenergic lipolysis *in vitro* is still doubtful. We therefore tried to solve this problem by using a selective α_2 adrenergic agonist, tramazolin (T) and to use it under special experimental conditions, namely in animals of different body weight and subjected to various periods of starvation. In control animals, we never found the inhibitory effect of T, while in starving animals the inhibition of lipolysis *in vitro* was always present. Its degree (23-44 % inhibition) was closely related to the duration of starvation and to a higher body weight of the rats. Yohimbin (α_2 adrenergic antagonist) not only antagonized the effects of T, but was also able to enhance the lipolytic effect of noradrenaline. Thus, the β adrenergic receptor stimulated lipolysis seems, under special conditions, to be influenced by α_2 adrenergic drugs also in rats.

NON-ENZYMATIC GLYCOSYLATION OF MYOSIN IN DIABETES AND AGEING. *I. Syrový, Z. Hodný[1],* Institute of Physiology, Czechoslovak Academy of Sciences, Prague and [1]Research Institute of Gerontology, Malacky.

The influence of diabetes mellitus, streptozotocin-diabetes and ageing was investigated on the glycation of myosin from cardiac and skeletal muscles. In the cardiac muscle, and to a lesser extent also in skeletal muscles of the rat, glycation of myosin increases, when 6-, 12- and 29-month-old animals are compared. In human cardiac muscle glycation of myosin also increase with age. Myosin from human diabetic skeletal muscles and also myosin from diabetic rat cardiac muscle is more glycated when compared with control myosin preparations. Ca^{2+} ATPase activity of myosin is lower in diabetic muscles when compared with control muscles.

METABOLIC AND HAEMODYNAMIC CHANGES IN THE PRAGUE HEREDITARY HYPERCHOLESTEROLAEMIC (PHHC) RAT HEART MUSCLE. *S. Edelsteinová, I. Tumová, E. Račanská. D. Ježová[1], B. Kovácsová, P. Švec,* Department of Pharmacology and Toxicology, Comenius University and [1]Institute of Endocrinology, Slovak Academy of Sciences, Bratislava.

Changes indicating the dvelopment of ischaemia in the PHHC rat heart muscle in comparison to a control group were studied. Ischaemic changes were found in the myocardium corresponding to 1.3 degrees according to the Zbinden scale. In ECG records, the ST segment was elevated, the ST/R ratio was 32 %. The heart rate increased slightly from 418 to 447 BPM beats per minute. The level of the atrial natriuretic factor decreased in the left and right atrium significantly, but only slightly in the left ventricle. The cardiac output and stroke volume measured on heart-lung preparations were not influenced. The levels of total cholesterol and lactate increased significantly, whereas in the FFA levels was decreased in the PHHC rat heart muscle. The changes found in the PHHC rat heart muscle could be explained as a result of long-term hypercholesterolaemia. The present model of myocardial mild ischaemia could be used for pharmacological studies of antiatherogenic and/or antianginal drugs.

VIII. Physiology of Respiration, Physical Training and Sport

THE GASTROINTESTINAL ABSORPTION OF ALUMINIUM, INDIUM AND GALLIUM. *A. Mravcová, J. P. Berry[1], F. Escaig[1], A.H. Chafi[1], P. Galle[1],* Institute of Hygiene and Epidemiology, Prague and [1]Laboratoire de Biophysique, Faculté de Médecine, Créteil, France.

The gastrointestinal absorption of aluminium, indium and gallium, the three elements of a homogeneous chemical group III, has been studied in mice by modern microanalytical methods (ion microscopy and X ray microanalysis). It was observed previously that these elements were concentrated in the kidney within the lysosomes (1). Lysosomes have recently been shown to have, apart from their function in organic metabolism, a second major function namely that of actively concentrating mineral elements. Our results confirm the fact that aluminium after intragastric application does not penetrate the mucous membrane of the duodenum. No similar granules were found in case of indium and gallium. Gallium was also detected in the kidney. The advantages of the above microanalytical methods for studying the metabolism of trace elements are discussed.

1. *Galle P.:* C.R. Acad. Sc. Paris, Vol. 292, Série III-91, 1981.

AMINO ACID TRANSPORT THROUGH THE RUMEN EPITHELIUM IN YOUNG AND ADULT SHEEP. *Z. Faixova, J. Várady,* Veterinary University, Košice.

The transport of a triad of amino acids (AAs) glycine-alanine-leucine was studied through the rumen epithelium *in vitro* in 6 suckling lambs and in 8 adult sheep fed molasses and hay. The transport of AAs was measured from the mucous to serous side of rumen epithelium after 60 min incubation. We used three concentrations of AAs on the mucous side: 1500, 150 and 15 μM.50 ml^{-1}. The concentration of AAs was measured on the mucous and serous side of rumen epithelium. We found that at the concentration 1500 μM.50 ml^{-1} the concentrations of Gly ($p<0.01$), Ala ($p<0.05$) and Leu ($p<0.05$) were higher on the serous side of the rumen epitleium in adult sheep as compered with lambs. At the 150 μM.50 ml^{-1} concentration the concentration of Ala was lower ($p<0.01$) in adult sheep in comparison with lambs and Gly and Leu were not significantly changed. At the 15 μM.5 ml^{-1} concentration concentration of Leu was lower ($p<0.001$) in adult sheep when compared with lambs and gly and Ala were not significantly changed. The results showed that higher concentrations of AAs on the mucous side of the rumen epithelium has a more marked effect on the transport of AAs in adult sheep than in lambs.

RED BLOOD CELL AGE-DEPENDENT MODIFICATIONS OF INOSITOL 1,4,5-TRISPHOSPHATE. *H. Hrušová, A. Strunecká, M.P. Piacentini[1], A. Accorsi[1], M. Magnani[1],* Department of Developmental Biology, Faculty of Science, Charles University, Prague and [1]Institute of Biological Chemistry "G. Fornaini" University of Urbino, Italy.

The level of inositol 1,4,5-trisphosphate (Ins 1,4,5P_3) was determined in human red blood cells (RBC) of different ages. RBC were fractionated by discontinuous density gradient centrifugation. Ins 1,4,5P_3 was 290 nM in the 0.3 % low dense (youngest) cells compared to values of 107 nM in the whole RBC population. A progressive increase in Ins 1,4,5P_3 was then observed during RBC aging from values of 63 nM in mature RBC to 128 nM in the oldest cells. These results provide evidence for an increase of Ins 1,4,5P_3 during RBC aging and could contribute to the explanation of the age-dependent loss of deformability and impairment of Ca^{2+} homeostasis during RBC aging.

BREATHING FROM THE EXPIRATORY POSITION OF THE THORAX IN ANAESTHETIZED DOGS: INFLUENCE ON THE O_2-DISSOCIATION CURVE. *J. Hájek, O. Slezáková, A. Maťašeje, A. Kurtanský, M. Orgonášová, M. Kittová, E. Schniererová,* Institute of Physiology, Faculty of Medicine, Charles University, Bratislava.

During ventilation from the expiratory breathing level, partial pressure of O_2 and O_2-saturation decrease, the number of erythrocytes and haemoglobin increase. In 10 mongrel dogs, the factors influencing the O_2-dissociation curve were compared between relaxation and expiratory positions of the thorax: partial pressure of CO_2, pH, body temperature and 2,3-disphosphoglycerate (Sigma Diagnostics USA). Results: partial pressure of CO_2 decreased by 0.33 kPa ($p<0.01$) both in arterial and venous blood; pH increased from 7.35 to 7.40 in arterial blood ($p<0.01$); in most of the dogs, body temprature decreased (on an average by 1 centigrade) and 2,3-diphosphoglycerate increased (on an average by only 0.1 mmol.l^{-1}). Consequently, breathing from the expiratory level resulted in a mild shifting of the O_2-dissociation curve to the left, thus decreasing the utilization coefficient of oxygen.

PHYSIOLOGICAL RESPONSE OF PULMONARY TISSUE TO ADMINISTRATION OF NONAGGRESSIVE FOREIGN BODIES IN THE ALVEOLI. *J. Sedláček, J. Jandík, B. Ulybin, Z. Martinec[1],* Medical Faculty and [1]Pedagogical Faculty, Charles University, Hradec Králové.

The course of pulmonary tissue response to intratracheal administration of denaturated calf plasma labelled with Indian ink was investigated. Our results showed that the response was instantaneous and dependent on the amount of foreign bodies in the alveoli and had the following four phases: 1. instantaneous alveolar macrophage (AM) response, i.e. AM were released into the alveoli, phagocytosis of foreign bodies and subsequent AM transport through the bronchi out of the organism; 2. development of aseptic inflammation (granulocytic response) within 4 to 6 h seen in the alveoli where free nonphagocytized foreign bodies persisted; 3. proliferation, i.e. multiplication of granular pneumocytes with mitotic division in the alveoli in which nonphagocytized foreign bodies persisted; 4. the second AM response within 30-35 h, i.e. new AM were released into the alveoli and phagocytosis of the remained of foreign bodies was usually completed by 3-4 days.

AWAKE PHRENICOTOMIZED RATS DO NOT RESPOND ADEQUATELY TO HYPERCAPNIA OR HYPOXIA. *J. Nacházel, F. Paleček,* Institute of Pathophysiology, Second Medical Faculty, Charles University, Prague.

It has been ascertained that resting ventilation of awake pherenicotomized rats does not differ from that in control rats (1). The aim of this study was to measure the ventilation of phrenicotomized rats during stimulated breathing. Ventilation of six awake phrenicotomized rats and six sham-operated rats was measured in a body plethysmograph for unanaesthetized animals. The ventilation was measured during air breathing and after 5 min of inhalation of 2 or 5 % CO_2 or 10 % O_2 in N_2. Hypercapnia increased minute ventilation in control rats by 21 % and 45 %, respectively. Hypoxia augmented ventilation by 46 %. In phrenicotomized rats, however, ventilation was unaffected by breathing the hypercapnic or hypoxic gas mixture. We conclude that breathing of awake phrenicotomized rats is adequate under resting conditions but the animals do not respond to hypercapnic or hypoxic stimuli.

1. *Nacházel J., Paleček F.:* Eur. Resp. J.: 4 (suppl. 14): P742, 1991.

CHANGES IN RESISTANCE OF AIRWAYS AND LUNG COMPLIANCE DURING HIGH FREQUENCY JET VENTILATION (HFJV). *A. Čalkovská, K. Javorka, M. Petrášková,* Department of Physiology, Jesenius Medical Faculty, Comenius University, Martin.

The experiments were carried out on 14 rabbits ventilated by means of an HFJ ventilator for 3 hours. Animals of the first subgroup were ventilated intact, the rabbits of the second subgroup were ventilated after vagotomy. Parameters of the mechanics of breathing (the resistance of airways - Raw, the dynamic compliance of the lungs - C_{dyn}) were continuously evaluated by a computer system. The HFJV was accompanied by the changes of Raw and C_{dyn} in all animals. The airways resistance increased in the first group by 97.8 ± 14.4 % during HFJV. In the second subgroup, Raw rose by 54.1 ± 11.5 %. The dynamic compliance decreased in the first subgroup by 54.0 ± 5.2 % and in rabbits with vagotomy by 35.5 ± 5.0 %. The results indicate that HFJV deteriorates the parameters of mechanics breathing in healthy rabbits. Cervical vagotomy diminished but did not fully eliminate the changes.

CHANGES IN MECHANICS OF BREATHING AFTER THE ASPIRATION REFLEX. *K. Javorka, M. Petrášková, E. Zelenayová,* Department of Physiology, Jesenius Medical Faculty, Comenius University, Martin.

Parameters of the mechanics of breathing (total lung resistance - R_L and dynamic compliance - C_{dyn}) and values of blood gases were evaluated in 29 anaesthetized cats before and after the aspiration reflex. Attacks of the aspiration reflex were elicited without asphyxia (8 cats) and during two-minute asphyxia (9 cats). The control group consisted of 12 experimental animals. Inspiratory efforts during the aspiration reflex elicited short-term improvement of the studied parameters, an increase of PaO_2, C_{dyn} and a decrease in $PaCO_2$ and R_L. The aspiration reflex in combination with asphyxia was accompanied by opposite response, PaO_2, C_{dyn} decreased and $PaCO_2$ and R_L rose. The changes of C_{dyn} persisted up to the end of the experiment (3 hours). The studied parameters remained unchanged in the controls. The results indicate that changes in the paramaters of mechanics of breathing after forced inspiratory efforts depend on blood gas values.

VASOMOTOR TONE CONTRIBUTES TO VASCULAR CRITICAL CLOSING PRESSURE IN ISOLATED RAT LUNGS. *J. Herget, V. Hampl,* Department of Physiology, Second Medical Faculty, Charles University, Prague.

To evaluate the relative participation of mechanical factors and smooth muscle tone on the magnitude of critical closing pressure in peripheral pulmonary blood vessels we measured the pefusion pressure/flow relationship in 5 isolated blood perfused rat lungs before and after administration of papaverine (100 μM/l). In all cases the pressure/flow relationships were linear ($r \geq 0.98$). The critical closing pressure was calculated as the mean extrapolated intercept for perfusion pressure/flow relationship with the pressure axis. Papaverine reduced ($p < 0.05$) critical closing pressure from 10.9 ± 0.4 mm Hg (mean ± S.E.M.) to 8.8 ± 0.8 mm Hg. The slope of perfusion pressure/flow relationship did not change significantly (0.62 ± 0.03 mm Hg/ml/min before and 0.56 ± 0.05 mm Hg/ml/min after papeverine). We conclude that vasomotor tone contributes to the critical closing pressure even though after inhibition of vascular smooth muscle mechanical factors still contribute to the critical closing pressure.

EFFECTS OF FOCAL COOLING OF MEDULLA OBLONGATA STRUCTURES ON LARYNGEAL RESISTANCE DURING QUIET BREATHING IN CATS. *J. Jakuš, A. Stránsky[1], H. Baráni[1], I. Poliačeк[1], A. Oravec[1],* Institute of Physiology and [1]Institute of Biophysics, Jesenius University of Medicine, Comenius University, Martin.

Experiments were carried out on 12 cats under chloralose anaesthesia to determine the effects of unilateral focal cooling of the nuclei of the dorsal and ventral respiratory groups in the medulla oblongata on the laryngeal resistance and quiet breathing parameters. The results of cold block tests of the respiratory nuclei showed: 1. Compared with the control state, cooling of the nucleus ambiguus (NA) and nucleus retroambigualis (NRA) to 20 °C decreased both the inspiratory ($p < 0.01$, $p < 0.05$, respectively) and expiratory ($p < 0.01$, $p < 0.05$) values of laryngeal resistance, while cooling of the nucleus paraambigualis region (NPA) led to their increase ($p < 0.05$, $p < 0.05$). 2. These findings indicate that the respiratory neurones in the paraambigual and the retroambigual medullary regions possibly contribute to the regulation of laryngeal motoneurone activity.

SEASONAL CHANGES OF ENERGY EXPENDITURE AND MECHANICAL WORK IN ROWING. *J. Heller, R. Dlouhá, J. Novotný, J. Sukop, M. Ulbrichová,* Biomedical and Sports Research Centres, Faculty of Physical Education and Sport, Charles University, Prague.

To assess the seasonal effects of rowing training, a group of junior male (n = 10, age 18 ± 1 years, height 188 ± 4, weight 83 ± 8 kg, body fat 9 ± 2 %) and female (n = 10, age 17 ± 1 years, height 176 ± 3 cm, weight 68 ± 4 kg, body fat 15 ± 3 %) rowing competitors were examined three times a year by a 6-minute test on a mechanical friction braked rowing ergometer. Cardiorespiratory and mechanical parameters (power output, force, pulling distance and frequency) were determined concomitantly. Mean overall energy expenditure increased with training in the course of the year both in men (November: 504 kJ, March: 540 kJ, June 550 kJ) and in women (392, 429 and 444 kJ respectively). However, the work output in men remained constant (340 W) while in women it inceased slightly from 217 to 224 W. A decrease in mechanical efficiency of rowing observed both in men (24.2, 22.9, 22.2 %) and in women (19.9, 18.6, 18.2 %) was related neither to the frequency of rowing nor to the total pulling distance as had been reported previously (1).

1. *Di Prampero P.E.:* J. Appl. Physiol. 31: 853-857, 1971.

COMPARISON OF TWO METHODS FOR ASSESSMENT OF BODY FAT CONTENT. *R. Dlouhá, V. Bunc,* Biomedical Research Centre, Faculty of Physical Education and Sport, Charles University, Prague.

The measurement of body fat content is arousing much interest not only from the aspect of physical performance but also as an indicator of cardiovascular diseases. Direct measurements of fat content are not possible except after death. Variety of indirect methods have been proposed: hydrostatic weighing, NMR, ultrasoud determination, impedance determination, skinfold thickness measurements, etc. The purpose of this study was to compare the estimates of lean body mass (LBM) and percentage of body fat (% BF) as predicted by bioelectrical impedance (BIA - Bodystat 500 analyser) and the sum of skinfolds (SF^1). The experimental group consisted of 13 healthy male subjects; mean age was 17.75 ± 0.4 years, height 178.81 ± 7.98 cm and body weight 72.35 ± 8.86 kg. Mean body fat content expressed as a percentage of body weight, estimated by the two methods was 8.63 % by skinfold thickness and 2.84 % by impedance. These results suggest that electrical impedance measurements cannot be used in lean subjects without correction of theoretical linear regression.

1. *Pařízková J.:* Body Fat and Physical Fitness. Martinus Nijhof B.V. (Medical Division), The Hague, 1977.

CHANGES OF LACTATE CONCENTRATION AT INDIVIDUAL ANAEROBIC THRESHOLD IN TRAINED ATHLETES DURING THE YEAR. *V. Bunc, J. Heller,* Faculty of Physical Education, Charles University, Prague.

Seven long-distance runners and 14 top canoeists were studied during two consecutive training years under field conditions to determine the effect of training on lactate levels (LA) at individual anaerobic thresholds (IAT). The training imposed in both groups of athletes elicited maximal changes in LA at IAT in long-distance runners 4.6 % and in canoeists 11.7 % in the course of the two years. The values of LA concentration at IAT ranges in long-distance runners in range 3.10-3.56 mmol.l^{-1}, these values of LA were attained at 84-87 % VO_{2max}, and in canoeists were found in the range of 4.00-4.53 mmol.l^{-1} at 79-82 % VO_{2max}. The lowest values were found in both groups of athletes during the racing period, the highest during the preparatory period. These data provide evidence that the adaptations of human subjects to physical training resulting in changes of LA concentration at IAT (in the submaximal range of exercise intensities) are dependent on the qualitative content of imposed physical training and that the use of fixed values of LA for AT determination may lead to errors in the training schedule.

THE EFFECT OF ACTIVE TRAINING ON THE PRODUCTION OF OXYGEN FREE RADICALS AND BIOENERGETICS IN THE BLOOD. *J. Kucharská, I. Šeitz*[1], *E. Miklovičová, I. Herychová, J. Krošlák*[1], *T. Badová*[2], *A. Gvozdjáková,* Pharmacobiochemical Laboratory, Faculty of Medicine, Comenius University, [1]Faculty of Natural Sciences. Department of Physical Trainig and [2]Clinic of Haematology and Transfusion, Bratislava.

The effect of active training and psychic stress of basketball players on the production of oxygen free radicals and bioenergetic parameters was studied in their blood. Blood donors served as the control group. Although there was a significant decrease of AMP, ADP and Pi and the concentration of ATP was higher in the basketball players, the levels of total adenine nucleotides were the same as in the control group. Active training increases the phosphorylation potential, energy charge and lipid peroxidation. These results prove the adaptation of bioenergetics to a prolonged increase of physical training. On the other hand, increased production of oxygen free radicals can diminish the performance of sportsmen.

LATERAL PREFERENCES IN CHILDREN. *L. Nerad, N. Vlasáková*[1], Institute of Physiology, Czechoslovak Academy of Sciences and [1]School of Sports and Physical Education, Charles University, Prague.

Motor, postural, eye and ear preferences were studied in 48 young (5 to 10-year-old) female gymnasts. The following items were correlated: handedness, sitting with the legs crossed, arms folded, hands interlocked, shovel holding, wringing, looking back, preferred leg when jumping, eyeded-ness and preferred ear when listening. The subjects were tested on two occasions separated by 7 months. Only the preferred leg and looking back tested on the second occasion approached the rejection of their mutual independence ($p = 0.053$, Fisher's factorial test). The average laterality quotient $(R-L)/(R+L)$ was highest for handedness (0.79, right hand) and lowest for sitting with the legs crossed (0.02). In 14 subjects who participated on both occasions only sitting with the legs crossed and hands interlocked proved to be absolutely stable. The change of other preferences was, however, not statistically significant ($p > 0.05$, McNamara' s test).

HUMAN ADAPTATION TO COLD: SHIFT OF THE SHIVERING THRESHOLD DURING REPEATED WATER IMMERSIONS. *L. Janský, V. Hošek, H. Janáková, B. Uličný, J. Pařízková,* Department of Comparative Physiology, Faculty of Science, Department of Psychology, Faculty of Physical Education and Sports, Charles University, Prague and Department of Physiology, Faculty of Medicine, Masaryk University, Brno.

Repeated exposures (15 times) of young men (23 years old, 76 kg) to cold water (14 °C, immersion up to the chest for one hour, 3 times a week) induce changes in cold sensation and body temperature control. Cold adapted men exhibit a typical hypothermic response. Central and peripheral body temperatures at rest and during cold exposure are lowered. Resting O_2 consumption is also lowered and the metabolic response to cold is delayed. The amount of energy saved during the cold test represents 24 % of the total heat production. These changes are due to a shift of the temperature threshold for induction of cold thermogenesis to lower temperatures. Cold adapted men also show a lower cold sensitivity and do not exhibit the temporary tachycardia during cold exposure. The heart rate at rest and during cold exposure is lowered as well as pulse O_2 consumption. No significant changes in body fat content were observed. Adaptional changes begin to occur already after 5 exposures to cold and persist for at least 2 weeks after the end of the adaptational procedure. Changes in the shivering threshold can thus be used as a measure of cold adaptation in man.

IX. Endocrinology

TRIIODOTHYRONINE INCREASES THE SYNTHESIS OF A SPECIFIC PROTEIN IN THE RAT LIVER AFTER PARTIAL HEPATECTOMY. *J. Knopp, J. Brtko, P. Filipčík,* Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.

The administration of 3,5,3-L-Triiodothyronine (T_3) to rats immediately after partial hepatectomy led to an increase in total polysome content of residual hepatocytes, measured 18 h postoperatively which exceeded that observed after partial hepatectomy in the absence of the hormone. The translational capacity of mRNA isolated from regenerating liver after 18 h was greater than that of mRNA isolated from sham operated animals. T_3 increased the translational capacity of mRNA isolated from regenerating liver, when the hormone was administered immediately after surgery. After 2-dimensional electrophoresis of translated products one of the typical proteins recently characterized by molecular weight of 25 000 pI 6.9 appeared after partial hepatectomy and was significantly increased when T_3 was administered immediately after partial hepatectomy. These results show that the thyroid hormone can enhance some of the characteristic features of liver regeneration involved in the synthesis of proteins typical for the process of liver regeneration.

THE TRIIODOTHYRONINE NUCLEAR RECEPTORS APPEARANCE DURING G1/S PHASE OF THE CELL CYCLE. *P. Filipčík, J. Brtko, J. Knopp, Ľ. Rauová*[1], Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava and [1]Research Institute of Rheumatoid Diseases, Piešťany.

The process of the appearance of thyroid hormone receptors in the cell cycle was studied in mouse leukemia cells L1210. After cell synchronization by 2 nM thymidine for 12 hours followed by 80 nM colcemide for 5 hours the specific binding of triiodothyronine (T_3) to its nuclear receptors was determined 3, 6, 9 and 12 hours after release from the thymidine-colcemide block. Three hours after release from the block, ^{125}I-T_3 specific binding was 11.4 ± 2.5 % of control values measured for an asynchronous population. An upward slope in the progression of T_3 nuclear receptors was found 6 hours (32.3 ± 4.5 %), 9 hours (47.8 ± 5.2 %) and 12 hours (83 ± 4.3 %) after release from the thymidine-colcemide block. The data suggest that a) processes involving T_3 receptor promotion in cell nuclei are operative within the G_1-S phases of the cell cycle, and thus b) the increase in T_3 receptor concentrations in the nuclei is in a positive correlation with the number of cells in the S compartment of the cell cycle.

EFFECT OF PROTEASE INHIBITORS AND SUBSTRATES ON 3,5,3'-TRIIODOTHYRONINE (T_3) BINDING TO RAT LIVER NUCLEAR RECEPTORS. *J. Brtko, J. Knopp, P. Filipčík, M.E. Baker*[1], Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava and [1]Department of Medicine, M-023, University of California, San Diego, California, USA.

The effect of protease inhibitors N-tosyl-L-phenylalanine chloromethyl ketone (TPCK) and carbobenzoxy-L-phenylalanine chloromethyl ketone (ZPCK) at concentrations ranging from $1.5x10^{-6}$ to $1.5x10^{-4}$ mol/l on the specific binding of T_3 to rat liver nuclear receptors was evaluated. Under the conditions examined, both TPCK at $1.5x10^{-5}$ mol/l ($p<0.001$) and ZPCK at $1.5x10^{-6}$ mol/l ($p<0.05$) inhibited T_3 binding to nuclear receptors. In addition, a similar inhibitory effect on the T_3 specific binding to rat liver nucelar receptors at $5.0x10^{-4}$ mol/l by four protease substrates was found. The data suggest that a) both protease inhibitors and the substrates tested inhibit the T_3 specific binding to T_3 nuclear receptors and b) they support the hypothesis that T_3 receptors like other nuclear receptors encoded by c-erbA gene possess a "site" that recognizes both serine protease inhibitors and substrates in its hormone binding domain.

EFFECT OF PRETREATMENT BY STEROID HORMONES ON THE REACTIVITY OF THE LUNG TISSUE TO HISTAMINE. *P. Bánovčin, P. Višňovský,* Department of Pharmacology, Faculty of Medicine, Comenius University, Martin.

The effect of the pretreatment with four different depot steroid hormones on the reactivity of the guinea-pig tracheal and lung smooth muscle preparations to histamine was followed according to previous work (1). The animals (n=8 in each group) received an i.m. injection of triamcinolone (Kenalog, 1 $mg.kg^{-1}$, both sexes) progesterone (Agolutin, 2 mg/kg^{-1}, female guinea-pigs), testosterone (Agovirin, 2 $mg.kg^{-1}$, male) or estradiol (Agofollin, 0.2 $mg.kg^{-1}$, female animals) 5 days prior to the experiments. The reactivity to histamine *in vitro* (10^{-8} to 10^{-3} $mol.l^{-1}$) was depressed in the tracheal smooth muscle after pretreatment with both triamcinolone and progesterone, enhanced by estradiol. The reactivity of the lung strip was depressed after triamcinolone, enhanced by estradiol.

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GLUCOCORTICOID RECEPTOR-BINDING AND TYROSINE AMINOTRANSFERASE INDUCTION IN NORMAL AND ADRENALECTOMIZED RATS. *M. Alexandrová, P. Farkaš,* Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.

The effect of glucocorticoid injection and that of acute stress on rat liver cytosol glucocorticoid receptor (GR) was studied to ascertain whether the agonist-induced GR regulation also takes place in intact rats as it does in adrenalectomized (ADX) ones. Results: 1. Corticosterone ("B", 100 μg/100 g s.c.) and dexamethasone ("Dex", 1 μg/100 g s.c.) induced tyrosine aminotransferase (TAT) activity in both normal and ADX rats, though "B" unlike "Dex" caused GR depletion only in the liver cytosol of ADX animals. 2. To increase the level of endogenous "B" rats were exposed acutely to different stressors: swimming, injection of histamine i.p., vasopressin s.c., heat, immobilization, cold. Each of the stressors provoked a pronounced increase of plasma "B" with subsequent induction of TAT. Depletion of the cytosol receptor was, however, noticed only after swimming and histamine injection. It is concluded that in the presence of adrenal glands cytosol receptors are more resistant to corticosterone-induced depletion.

THE EFFECT OF PRENATAL GLUCOSE STIMULATION ON THE DEVELOPMENT OF SOME FUNCTIONS OF THE ENDOCRINE PANCREAS IN THE RAT. *V. Štrbák, K. Považanová,* Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.

We have developed an original model for prenatal stimulation of the endocrine pancreas in the rat. Throughout the whole pregnancy period tap water is replaced by a 15 % glucose solution. This treatment results in growth retardation of the offsprings which was still present on the 8th postnatal day. Growth of the pancreas was also retarded. Glycaemia in foetuses was similar to that in the controls, but was substantially lower during the first hours after birth. Maturation of the pancreatic TRH system was accelerated (peak on the first instead of the third postnatal day): parallel changes of TRH concentrations and PAM (peptidyl-glycine alpha amidating monooxygenaze which controls the final amidation of the precursor to TRH) activity were noted. It is possible that extrapancreatic TRH was also affected, since we have also found increased plasma TSH levels in the offsprings of glucose treated mothers.

INSULIN RECEPTORS IN HEPATOCYTES AND ADIPOCYTES OF RATS AFTER PERINATAL ADMINISTRATION OF A SINGLE DOSE OF INSULIN. *M. Ficková, L. Macho,* Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.

Elevated hormone concentrations in the perinatal period induce different tissue responsiveness in relation to age and the sex of experimental animals. Insulin in a single dose of 0.2 U administered during the first 24 hours of life did not induce any significant changes in the binding capacity of insulin receptors in adipocytes of male rats aged 60 and 120 days. No changes of insulin receptors binding capacity were observed in the hepatocytes of both male and female rats of the above age groups. Hepatocytes of 60-day-old female rats contained a significantly lower number of insulin receptors as compared with their male siblings. In older animals, such sex difference disappeared. No changes of insulin receptors binding affinity in hepatocytes and adipocytes were observed. The plasma insulin concentration did not differ and was independent of the sex and the age of rats. Perinatal administration of insulin did not induce any changes in binding affinity and the number of insulin receptors in rat hepatocytes and adipocytes. This effect was independent of the sex and age of the experimental animals.

THE ROLE OF THYROID HORMONES IN THYROLIBERIN (TRH) AND PEPTIDYLGLYCIN ALPHA AMIDATING MONOOXYGENASE (PAM) REGULATION IN THE PERINATAL RAT PANCREAS. *K. Považanová, V. Štrbák,* Institute of Experimental Endocrinology, Slovak Academy of Sceiences, Bratislava.

The concentration of pancreatic TRH reaches its peak on the 3rd postnatal day, its level in adult rats is minimal. The mechanisms regulating these changes still remain obscure. PAM (enzyme regulating final amidation of the TRH precursor to TRH) activity alters in parallel with the changes of TRH. To clarify the role of thyroid hormones, the rats were put on a Propylthiouracyl (PTU) containing diet from the 17th day of pregnancy until delivery (hypothyroidism), or received daily an injection of 10 ng/g BW DIMIT (thyroxine analogue passing across the placenta) from the 15th day till decapitation (hyperthyroidism). The effectiveness of treatment was proven by TSH changes. The TRH level in PTU treated animals already peaked in 21-day-old foetuses, while it stayed at the control level in those treated with DIMIT. PAM activity in PTU pups paralleled that of the controls, but was much higher, DIMIT treatment was followed with high foetal PAM activity which disappeared on the 3rd postnatal day. The discrepancy between PAM and TRH levels in the pancrease of perinatal rats suggest a possible role of thyroid hormones in TRH degradation.

THE STUDY OF HYPOTHALAMIC (HT) THYROLIBERIN (TRH) SECRETION IN VITRO. *M. Nikodémová, K. Považanová, V. Štrbák,* Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.

To study the mechanisms regulating HT TRH secretion we developed a modified method using two HT halves incubated *in vitro*. Our goal was to leave the basal HT surface as well as the wall of the third ventricule intact (after careful sagital section) containing undamaged HT nuclei (especially PVN). Halves of HT were incubated in a Krebs-Ringer bicarbonate buffer containing 5 mM glucose and saturated with O_2 and CO_2. After 30 min preincubation and medium exchange, aliquots for TRH RIA were taken at 5 min intervals. The medium was changed after 30 min 3-4times. We found that the TRH released into the medium at the beginning was degraded during the incubation. This could be prevented by addition of 0.03 % of bacitracin. The presence of bacitracin in the medium, however, affected the binding of the labelled ligand in TRH RIA. Addition of the medium containing bacitracin to the standards displaced the standard curve to the left leaving it parallel with the original curve. This effect persisted after methanol extraction. An increase of KCl in the medium (from 4.75 mM to 60 mM) resulted in stimulation of TRH secretion due to depolarization.

A COMPARISON OF THE INFLUENCE OF GONADOTROPINE HORMONES ON THE VOLUME OF OVARIES AND THE NUMBER OF PROMINENT FOLICLES OF EWES IN THE ANOESTROUS AND OESTROUS PERIOD. *V. Eliaš, D. Pavlová, J. Halagan, J. Pošivák[1], J. Várady,* Department of Physiology and [1]Experimental Research Centre, University of Veterinary Medicine, Košice.

The ovarian stimulatory influence of FSH (Folicotropine, Spofa) have been studied in ewes during the anoestrous and oestrous period. The ewes (merino breed) were synchronized by 13 days application of intravaginal sponges (Ageline, Spofa). The experiments were carried out on three groups divided according to the doses of FSH (18, 24 or 30 mg) on the 12-14th day of the experiment. The animals were slaughtered on the fourth day after oestrus had occurred. The ovary volume and number of prominent follicles were estimated macroscopically and statistically evaluated by Student's t-test. The significant increase in volume ($p<0.01$), and in the number of prominent follicles ($p<0.001$) was found after administration of 18 m FSH in the oestral period. The administration of 24 mg of FSH during anoestrous caused a significant increase ($p<0.05$) of ovarial volume only. The higher dose (30 mg) of FSH increased the number of prominent follicles ($p<0.01$).

EFFECTS OF VASOPRESSIN INFUSION ON STRESS-INDUCED CHANGES IN PLASMA RENIN ACTIVITY AND ALDOSTERONE SECRETION. *O. Földes, D. Ježová, Z. Opršalová, B. Lichardus, R. Kvetňanský,* Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava.

The neuroendocrine response during immobilization stress in rats includes increases in plasma renin activity (PRA) and in aldosterone (Aldo) release. Little is known, however, about the mechanisms responsible for these changes. This study was aimed to investigate the influence of intravenous infusion of vasopressin (AVP) in the dose range of 8-80 mg/min/kg on PRA, Aldo, ACTH and noradrenaline levels during immobilization in conscious cannulated rats. The stress-induced rise in PRA was significantly inhibited even by the lowest dose of AVP used and was completely absent in rats treated with higher doses. In contrast, AVP infusion resulted in an increase in Aldo levels under non-stress conditions and failed to modify its elevation during stress. A possitive correlation was found between PRA and plasma noradrenaline concentration indicating that AVP may inhibit PRA via an increase in sympathetic activity. The action of AVP on PRA is mediated through V1 receptors as dDAVP was without any effect. Though PRA is generally considered to control Aldo release, the discrepancy between PRA and Aldo response to AVP infusion shows that, at least under these experimental conditions, secretion of Aldo is predominantly regulated by ACTH.

HORMONALLY STIMULATED OVARIAN ACTIVITY AND HISTOLOGICAL CHANGES IN THE HYPOTHALAMUS AND PINEAL GLAND OF SHEEP IN SYNCHRONIZED OESTRUS. *J. Halagan, A. Staníková, V. Rajtová, B. Pástorová, V. Eliáš,* University of Veterinary Medicine, Košice.

The study was carried out on 78 ewes during oestrous and anoestrous periods. The oestrus was synchronized by chlorsuperlutine (20 mg) vaginal tampons for 12 days. The ovulation rate was stimulated by administration of various doses and combinations of PMSG, HCG, FSH, PGF_2 alpha and Anti PMSG serum. The hypothalami, pineal glands and ovaries were evaluated by light, electron and scanning microscopy. Progesterone (P_4), estradiol (E_2), FSH and LH were monitored by RIA methods. The activation of infundibular ependymal cells was found in PMSG, HCG stimulated or Anti PMSG treated ewes. The most pronounced effects (an increase of neuronal nuclei volume, miniblebs) were found in the nucleus infundibularis inferior. These changes negatively correlated with nuclear volume changes in pinealocytes. The E2 concentration and LH peak, found in ewes with a positive stimulatory response confirmed the histological changes and indicates that ependymal cells, neurones and pinealocytes may be involved in processes of hormonal ovarian stimulation in ewes during controlled oestrus.

X. Physiology of Reproduction

EFFECT OF HORMONAL STIMULATION ON CATECHOLAMINE LEVELS AND ACTIVITY OF MAO IN THE BRAIN OF SHEEP. *B. Pástorová, J. Várady, J. Halagan, A. Staníková, V. Eliáš,* Faculty of Veterinary Medicine, Košice.

The effect of a hormone preparation, used for inducing superovulation in farm animals, on catecholamine levels (dopamine and norepinephrine) and MAO activity was studied in the hypothalamus, pituitary and pineal gland of sheep after oestrus synchronization. In the first experimental group, the animals were stimulated by 1 000 IU PMSG; the second group received 1 000 IU PMSG with Antisergon. Catecholamines and MAO activity were determined by radiochemical methods. Administration of PMSG results in significant changes ($p<0.001$) of catecholamine levels and MAO activity in corpus mamillare, eminentia medialis and the pituitary gland. PMSG in combination with Antisergón does not cause significant changes. It is suggested that the changes in catecholamine concentration after PMSG administration is related to the induced prolonged increase in blood plasma oestrogens affecting adrenergic receptors in the hypothalamus.

ANTIPROTEOLYTIC ACTIVITY IN THE FOLLICULAR FLUID OF EWES AFTER HORMONAL STIMULATION OF OVARIES. *M. Molnárová, J. Arendarčik,* Department of Physiology, University of Veterinary Medicine, Košice.

Merino sheep ($n=29$) in oestrus period responded to treatment with PMSG and Antisergon (As-anti PMSG) after Ageline (chlorsuperlutine) synchronization by changes of antiproteolytic activity (AA) of the follicular fluid (FF) in dependence on the intervals of treatment (12, 24, 48 or 58 hours after PMSG) and on the category of ovarian follicles. AA were measured as inhibition of hydrolysis of chromogenic substrate N-alpha-tosyl-l-arginyl-p-nitroanilide (TAPA) with bovine trypsin. FF of follicles <5 mm had the lowest AA in the group, where As was added 24 hours after PMSG, while in the category 5-10 mm it was in the group at the 48 hours' interval. Follicles >10 mm after treatment with As in intervals 12-58 hours were not present. Stimulation with PMSG alone or the treatment with As 68 hours after PMSG, made the production of follicles >10 mm possible. Changes in AA FF and blood plasma indicate that they participate in the development and ovulation of the follicles.

XI. Higher Nervous Activity and Physiology of Behaviour

BEHAVIOURAL ONTOGENY IN MALE LABORATORY RATS OF THE WISTAR, LONG EVANS AND FISHER 344 STRAINS. *S. Fraňková*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Responses to a novel environment and habituation of exploratory activity was studied in male rats of three strains. The experiments were carried out between the first and the twenty-first month of age. Strain differences were found in the life-span trends of the selected behavioural patterns, in the timing, reaching the ontogenetic maximum, in the rate of declination of spontaneous locomotor activity in the course of aging as well as in ontogenetic characteristics of habituation. "Emotionality" (defecation, urination) was also affected by the age, strain and features of the testing situation.The results are discussed in connection with certain discrepancies in the experimental literature, which might result from the fact that age and strain specifities in behavioural manifestations were not taken into account. Furthermore, problems of the periods in which most marked changes take place were discussed.

AMNESIC EFFECT OF NMDA RECEPTOR ANTAGONISTS ON SOCIAL MEMORY IN MALE RATS. *Z. Hliňák, I. Krejčí, Z. Polívka*, Research Institute for Pharmacy and Biochemistry, Prague.

It has been suggested that NMDA receptors may play a critical role in learning and memory. In the present study, the effect of various NMDA and non-NMDA antagonists was evaluated using the social memory test. Time spent by adults in social investigation of a familiar or a novel juvenile animal was measured during reexposure 30 min after the first exposure. The drugs or saline were injected s.c. immediately after the first exposure. It was found that phencyclidine (1 and 1.5 mg/kg), MK-801 (0.1 mg), CPP (2.5 and 4 mg) and CGS 19755 (4 mg) have an amnestic effect, i.e. social investigation time during reexposure of adults to the familiar juvenile corresponded to that during the first exposure. No evidence of the amnestic effect was obtained if adults were treated with CNQX or NBQX (up to a dose of 10 mg): animals showed a shortening of the time spent in social investigation towards the familiar juvenile during reexposure, similarly as the controls. In summary, while representatives of competitive as well as non-competitive NMDA receptor antagonists interfere with the storage of information received through olfactory cues, representatives of non-NMDA antagonists did not suppress the ability of adults to recognize the juvenile.

IMPACT OF HALOPERIDOL ON EARLY INHIBITORY LEARNING AND MEMORY. *J. Mysliveček*, Institute of Pathophysiology, Medical Faculty, Charles University, Plzeň.

Previously, we have shown that dopamine (DA) influences inhibitory learning (passive avoidance) in newborn rats. If given at various after-learning intervals, it changes 24-hour memory differentially (an optimum possitive effect was noted at the one-hour interval) (1). In attempts to decipher DA mediated mechanisms, we studied the impact of haloperidol, a D_2 antagonist, on 24-hour memory in newborn rats at the age of up to 10 postnatal days. In 1-day-old pups, a moderate positive effect on the 24-hour memory was observed that was not, however, related to the after-learning interval; this was shown by positive retention index (RI) values. In animals about 5-days old and more, haloperidol impaired learning by increasing the number of trials to criterion; if given after learning, even within the retention test (relative to learning), so that negative RIs were obtained. DA administration, either intraperitoneally or into lateral cerebral ventricles, partially counteracted the haloperidol effects.

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IS THE NATURE OF NEURONAL VOCABULARY FRACTAL? *D. Svorad*, Institute of Molecular Physiology and Genetics, Slovak Academy of Sciences, Bratislava.

The vocabulary of neuronal bursting activity (VNBA) shows self-similarity (self-similar sets; S) which means invariance with the respect to the change of measure: an identical or similar pattern is repeated in a continued decreasing detail (3). S also appears as the central feature in fractal sets (2,3). The aim of this study was to compare the S in VNBA and in a fractal (F). - S in VNBA can be described by the Zipf law (4), the description of S in F implies, e.g. the Cantor ternary set (1). The split of an F set can be expressed by the Hausdorff dimension (1), whereas the split of VNBA can be described by the Waring distribution (4) and has only a topological dimension.

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CONDITIONED TASTE AVERSION TO SHORTLASTING ANODIC TONGUE STIMULATION IN RATS. *J.A.W.M. Weijnen, G. Brožek*, Tilburg University, Tilburg and Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Shortlasting (5 ms) weak (20 μA constant current) anodic pulses were applied 1 ms after the onset of every 4th lick in freely licking thirsty rats (24 hours of water deprivation). Gastrointestinal distress was elicited immediately after the 15 min period of stimulated licking by means of an i.p. injection of LiCl (0.15 mol/l, 2 % body weight). The ensuing conditioned aversion was manifested by interruption of spontaneous licking (or lapping) immediately after each stimulated lick. This result is in good agreement with an older report by Weijnen (1) that the tendency of rats to lick a metal spout, delivering low anodic current during the whole period of tongue-spout contact, can be suppressed when associated with subsequent poisoning. The immediate behavioural effect of the electrical taste is compared with the less prompt cessation of licking in the case of aversion to natural tastes.

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PROTEIN KINASE C IN THE NUCLEUS PARABRACHIALIS. EFFECT OF LiCl ADMINISTRATION. *J. Křivánek, L. Nováková*, Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Evidence has been provided that nucleus parabrachialis (NPB) plays a key role in the formation of conditioned taste aversion (1). For the biochemical analysis, NPB was dissected from the frontal section of the rat heads frozen in liquid nitrogen by a method based on our previously published procedure (2). Two 1 mg pieces of NPB were pooled and homogenized in an isolation buffer. The crude protein kinase C (PKC) was separated into the cytosolic and the membrane-bound portion. After partial purification on DEAE, activity was determined by measuring the phosphorylation of histone III-S. One hour, 24 h and 82 h after a single i.p. injection of LiCl (0.15 M, 2 % body weight) rats were sacrificed and PKC activity in the NPB was determined. A statistically significant 16 % raise of the membrane bound PKC (61.5 ± 1.3 % vs 53.0 ± 1.8 %; $p < 0.01$) was found one hour after LiCl injection.

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DIAZEPAM IMPAIRS THE RETENTION OF SPATIAL LEARNING IN THE MORRIS WATER MAZE. *M.P. Arolfo, J.D. Brioni,* National University of Cordoba, Cordoba, Argentina.

The effect of diazepam (DZP) on the acquisition, retention and retrieval of spatial information was studied. Four groups of rats receiving i.p. injections of saline, 0.3, 1.0 or 3.0 mg/kg DZP, respectively, were trained to find a hidden platform in the Morris water maze, receiving three trials per day during four consecutive days. DZP was injected 30 min before the first daily trial. The 0.3 mg/kg DZP rats did not differ from the control group. The higher DZP dosage (1.0 mg/kg) impaired location of the escape platform, specifically in the first trials on days 3 and 4. The 3.0 mg/kg DZP animals were impaired during all four days of training. These results suggest a DZP-induced impairment of retention or retrieval rather than of acquisition of spatial information. In another experiment, retrieval was not impaired in overtrained intact rats receiving DZP before testing. The above results suggest participation of the GABA/benzodiazepine receptor in the consolidation of cognitive processes.

FUNCTIONAL BLOCKADE OF PARABRACHIAL NUCLEI DISRUPTS CONDITIONED TASTE AVERSION LEARNING IN RATS IRRESPECTIVE OF THE NATURE OF THE GUSTATORY CONDITIONED (CS) AND VISCERAL UNCONDITIONED STIMULUS (US). *E. Bielevska, J. Bureš,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The recent finding that acquisition of conditioned taste aversion (CTA) can be disrupted by tetrodotoxin (TTX, 10 ng) injected into the parabrachial nuclei (PBN) of rats after saccharin drinking and before administration of LiCl suggests that PBN play an important role in the CTA mechanism. The universal validity of this assumption was tested for CTAs induced by association of other gustatory CS and/or visceral US. Inactivation of PBN by TTX impaired CTA formation both when LiCl was preceded by ingestion of palatable (0.1 % saccharin, 0.9 % NaCl) or non-palatable (0.0072 % quinine, garlic extract) stimuli which usually elicit strong aversions and have (in case of garlic) an important olfactory component. TTX blockade of PBN also disrupted CTAs induced by association of saccharin or NaCl drinking with subsequent LiCl (127 mg/kg), D-amphetamine (3 mg/kg), carbachol (0.15 mg/kg) or cyclohexemide (1 mg/kg) administration. The results support the conclusion that PBN is a point of convergence of different CS and US activated pathways mediating CTA learning.

TETRODOTOXIN BLOCKADE OF AMYGDALA DISRUPTS CONSOLIDATION OF PASSIVE AVOIDANCE LEARNING IN RATS. *C. Bucherelli, G. Tassoni, J. Bureš*[1], Department of Physiological Sciences, University of Florence, Florence and [1]Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Functional blockade of the parabrachial nuclei (PBN) by local injection of tetrodotoxin (TTX, 10 ng) partially disrupts the consolidation of the passive avoidance reaction (PAR) when the acquisition – TTX delays are 0 or 24 h but not 48 h (1). In the present experiments, TTX blockade has been similarly used to investigate the role of amygdala (AM) in PAR consolidation. Rats were trained in the step-through passive avoidance task. TTX injected immediately after acquisition into both AM (2 x 10 ng) significantly reduced avoidance of the dark compartment in the retrieval test performed two days later. The PAR impairment was comparable to that elicited by PBN injection of TTX. The amnestic effect was significant when the acqusition – TTX delay was prolonged to 90 min but not to 6 h. Thus, reversible blockade of AM disrupts PAR consolidation similarly as the blockade of PBN, but during a shorter time. These results suggest that these two structures play different roles in the formation of PAR engrams.

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LATERALISATION OF DIFFERENT COMPONENTS OF THE PLACE NAVIGATION TASK ACQUIRED BY RATS DURING TETRODOTOXIN (TTX) BLOCKADE OF ONE HIPPOCAMPUS. *A.A. Fenton, J. Bureš,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Functional ablation methods confinnig memory trace formation to one hemisphere have been widely used in research into interhemispheric transfer of visual discrimination learning. So as to use this approach in spatial memory studies, unilateral blockade of the hippocampal formation was produced by injection of 5 ng TTX into the dorsal hippocampus of chronically cannulated rats (n = 20). The animals were trained daily in the Morris water tank under TTX blockade of one hippocampus. Retrieval of the same task was then tested during inactivation of either the ipsi- (I) or contralateral (C) hippocampus. Escape latencies (in seconds) in the final unilateral acquisition block (A), and in the first I or C retrieval block showed that lateralisation was absent after a single 4-trial block (A = 49. I = 40, C = 44), marginal after limited training (9 blocks) leading only to an efficient search strategy (A = 6, I = 9, C = 14), and complete in rats overtrained (15 blocks) to asymptotic target oriented navigation (A = 4, I = 4, C = 12). Poor lateralisation of the early stages of place navigation learning suggests their extrahippocampal implementation.

DIFFERENTIAL ROLE OF THE GUSTATORY CORTEX AND AMYGDALA IN THE ACQUISITION AND RETRIEVAL OF CONDITIONED TASTE AVERSION IN RATS. *M. Gallo, G. Roldan*[1], *J. Bureš*[2], Department of Experimental Psychology and Physiology of Behavior, University of Granada, [1]Instituto Mexicano de Psiquiatria, Mexico City and [2]Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Lesion studies of the role of gustatory cortex (GC) and amygdala (AM) in conditioned taste aversion (CTA) are confounded by the irreversibility of the intervention. Functional ablation methods allow more specific influencing of different phases of CTA acquisiton and retrieval. Tetrodotoxin (TTX) blockade of GC (10 ng TTX) or AM (3 ng TTX) before or after saccharin drinking in rats with chronically implanted intracerebral cannulae showed that GC is indispensable for the initial processing of the taste stimulus but not for the association of the gustatory trace with poisoning. Gustatory signals can by-pass the blocked AM the inactivation of which, however, prevents the gustatory trace-poisoning association. TTX injection into both GC and AM impairs CTA retrieval more than an isolated blockade of either of these structures. It is argued that GC and AM implement processing of the gustatory and visceral signals, respectively, whereas the parabrachial nuclei contribute to consolidation of the CTA engram.

XII. Psychophysiology

PAIN INTENSITY AND PAIN CUTANEOUS THRESHOLD: IS DISPERSION OF THRESHOLD VALUES A VALID PREDICTOR OF PAIN? *P. Knotek, H. Urbancová,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Pain intensity is the best measurable aspect of pain perception. Pain threshold is generally considered as a measure of the sensitivity to pain stimulation. The dispersion of measured values is at present usually considered as a measure of the reliability of this approach. Our result have proved a close correlation between the intensity of pain measured by VAS and pain threshold parameters measured by exposition to radiant heat in a group of 78 patients with back pain. Multiple regression proved 1. relatively close relations between threshold parameters and intensity of back pain (R^2 = 0.17), 2. a moderately higher dependence of pain intensity on the standard deviation than on the average of threshold values and 3. stochastically independent relation between x and s as pain predictors. This dispersion of measured threshold levels is considered as a measure of misleading input information, as a condition of unrealistic central information processing, especially in anxiety.

THE STUDY OF LOAD SITUATIONS OF STUDENTS AT THE BEGINNING OF THEIR UNIVERSITY STUDY. *E. Kukurová, M. Bratská, J. Miklovičová, Z. Herdová,* Institute of Medical Physics and Biophysics, Faculty of Medicine, Comenius University and Department of Psychological Sciences, Faculty of Philosphy, Comenius University, Bratislava.

This study is concerned with the prediction and prevention of the psychic load, especially in students during the first year of their university study. The objective resources and subjective determinants conditioning the rise of psychic load situations in overcoming the temporary period between high school to the university level were examined longitudinally. The research results were analysed and compared by means of various interdisciplinary psychological and biophysical methods, techniques and procedures. The objectives of this project were to discover and to help in overcoming the personality desintegrating resources and in activating the maladaptive structures. These lead the individual towards constructive approaches for overcoming this psychic load.

SLOW TONAL SEQUENCES FOLLOWED BY FINGER TAPPING: STIMULUS ANTICIPATION DISAPPEARS. *J. Mates, U. Müller[1], E. Pöppel[1], T. Radil., M. Indra,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague and [1]Institute of Medical Psychology, LMU, Munich.

When following rhythmic tonal sequences by finger tapping the onset of responses precede the onset of the stimulus. This anticipation (A) is minimal at a very high stimulus repetition rate (interstimulus interval – ISI 300 ms). At ISIs up to about 2000 ms the onset of taps precedes stimulus application by several tens of milliseconds although the subjects are not aware of that. At ISIs between 2000 and 4800 ms, the subjects exhibit advanced and delayed responses with high variability with an increase of the incidence of the subsequent ones. At the longest ISIs, the subjects react with the usual reaction time of about 200 ms and A disappears. Central integration mechanisms with a time constant of about 3 s related to perceptual memory and to the phenomenon of the actual present seem to be involved.

FAST AND SLOW TONAL SEQUENCES FOLLOWED BY FINGER TAPPING: EVENT RELATED POTENTIALS. *T. Radil, L. Maras, Z. Bohdanecký, M. Indra, J. Mates, E. Pöppel[1],* Institute of Physiology, Czechoslovak Academy of Sciences, Prague and [1]Institute of Medical Psychology, LMU, Munich.

Event related potentials (ERP-s) were synchronized either by tone or tapping onset when following a) fast (interstimulus interval 700 ms) and b) slow (4800 ms) rhythmic tonal sequences. In a) the response preceded the stimulus by about 30 ms and b) it was followed by the usual reaction time of about 200 ms. ERPs were of much lower amplitude in a in comparison with b. Tapping added to mere listening decreased the ERP amplitudes both in a and b. The onset of the tapping was preceded by a negative potential shift in a and by a positive one in b. Spontaneous tapping was preceded by a negative shift in b only. The results might be caused by differences in motor programming.

THE LEARNING EFFECT DURING TRACING OF SIMPLE GEOMETRICAL SHAPES. *M. Indra, Z. Bohdanecký,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The aim of the present study was to analyze the influence of some geometrical features of manually traced visual patterns on the tracing performance during a five days time period. The combination of three shapes: square, triangle and circle, presented on a computer screen were traced in a clockwise (or counterclockwise) direction using the right or left hand (employing a standard computer mouse). Ten right-handed volunteers participated in the experiment. The tracing accuracy (TA) expressed in RMS measures and tracing time scores (TT) were compared separately by ANOVA. Square-shape tracings yielded the best TA with the lowest values of TT intervals. On the contrary, during circle-shape tracings the worst TA and the longest TT were obtained. The substantial effect of practice was indicated mostly between the first 2 sessions and less expressed between sessions 2 and 3 with generally higher scores for the circle-shape tracing in comparison with the remaining shapes. No significant differences were found among the last three sessions.

DISTRIBUTION OF KEY TOUCHING TIMES DURING RHYTHMIC FINGER TAPPING. *E. Pöppel, T. Radil[1], J. Mates[1],* Institute of Medical Psychology, LMU, Munich and [1]Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The task of subjects was to "tap shortly and rhythmically upon the response key" according to the regular sequence of tones they were listening to. The aim was to find out whether the preferential key touching time (KT) intervals do exist for tapping, a simple task, apparently preprogrammed by highly stereotyped motor activity, reproducible for a long time. A considerable number (n = 39 200) of KTs were accumulated. Their distribution revealed, besides the mean peak corresponding to approximately 80 ms, three additional suspect peaks of about 50, 130-140 and 170 ms. An exponential fitting procedure was adopted to analyze the empirical distribution of KTs. The results support the hypothesis that timing of tapping is discontinuous, i.e. some KTs are preferential, as its certain discrete temporal parameters would be set in the motor programme controlling tapping.

CUTANEOUS TEMPERATURE INCREASE INDUCED BY HYPNOTIC SUGGESTION OF PAIN. *P. Hájek, T. Radil, B. Jakoubek, K. Kýhos,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Eight patients with atopic eczema and six healthy subjects were subjected to hypnotic suggestions of feeling pain in the area of the shoulders or the palm. An average local increase in skin temperature of 0.6 °C detected by thermovision developed under this condition. In the same patients, the cutaneous pain threshold was increased before the experiments by means of repetitive hypnotic suggestion of analgesia. These subjects reported feeling of no pain subjectively after pain suggestion. The local reaction in skin temperature was the same in both cases. The results suggest a central mechanism determining pain threshold changes in the skin which is independent of local changes in blood flow.

TRACING OF SIMPLE GEOMETRICAL SHAPES AND CORRESPONDING HEART RATE CHANGES. *Z. Bohdanecký, M. Indra,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Previous results indicated that significant heart rate (HR) changes during simple tracking tasks (1) and similarly some HR changes related to tracing of simple geometrical shapes may be expected. Nine volunteers traced segments of square, triangle or circle using a computer "mouse". The experiment was repeated during 5 consecutive days, the HR data were recorded on the second and the fifth day, and were compared. The mean scores revealed the highest values during square presentation, the lowest values characterized circle tracing. The cardiac cycle phase-dependency, expressed by a higher number of instants of segment tracing terminations during the first half of the R-R interval, was found during the second day of the experiment, while this contingency disappeared completely on the fifth day.

1. *Indra M., Bohdanecký Z., Radil T.:* Int. J. Psychophysiol. 5: 161-166, 1987.

ANALYSIS OF NEURITIC DRUSES IN AN IMAGE PROCESSING SYSTEM. *M. Srbecký, M. Ondrejka, P. Černay, L. Chrást, J. Mikula,* Medical Informatics Research Institute, Bratislava.

The software environment for image processing developed in MIRI is aimed at mainly interactive evaluations of (microscopic) images. However, for image analysis of brain tissues with neuritic druses, we suggested an automatic procedure, in which the druses are segmented and classified. The relative frequencies and relative areas of individual classes served as the quantitative parameters. The image and results of the analysis were stored by computer, simultaneously with the text data and references to the related images. The larger set of images is currently processed, in order to verify the reliability and statistical properties of the estimation process from the medical point of view. The basic system consists of software developed in C^{2+} language, a microscope, a CCD camera, PC with a Frame Grabber and a printer. The options are SVGA, extended/expanded memory in the PC and TV monitor.

XIII. Methods

A NEW METHOD FOR MEASUREMENT OF POTENTIAL AND IONIC CHANGES IN THE EMBRYONIC KIDNEY TUBULES IN VIVO. *Z. Zemanová, E. Ujec,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

Accessibility of the chick embryo to direct measurements in its mesonephric kidney during the period between embryonic days 5 and 9 was solved by the construction of a "chick embryo incubation bath". This consisted of an airing chamber covering the egg and of a warmed reservoir with a modified Krebs-Heseleit-Ringer solution with the addition of Tricaine anaesthetics, in which the embryo floats, connected by the umbilical cord with the breathing membranes. The surgical removal of the right body wall made mesonephric nephrons accessible. The mortality rate of operated embryos was 60 % until third day after surgery. Embryos of all age groups survived their stay in the incubation bath for several hours as measured by steady values of the heart rate. A DC differential amplifier together with further measurement devices was used for the registration of both voltage and concentration changes with a single ion-selective double-barrel microelectrode inserted into the tubules.

A COMPARISON OF THE SPECTROPHOTOMETRY METHOD VERSUS FLAMELESS ATOMIC ABSORPTION SPECTROPHOTOMETRY FOR THE DETERMINATION OF SELENIUM LEVELS IN THE BLOOD SERUM OF SHEEP. *M. Prosbová,* University of Veterinary Medicine, Košice.

This report compares the results for the determination of selenium in the blood serum of sheep by the spectrophotometry method using 3,3'-diaminobenzidine and by flameless atomic absorption spectrophotometry using selenium stabilization with nickel -II nitrate. The results indicate that flameless atomic absorption spectrophotometry is more accurate and has the advantage of a simple operation, rapidity, low consumption of chemicals and biological material. In 10 samples of the blood serum the concentration of selenium ranged from 0.109 to 0.142 $\mu g.ml^{-1}$.

DETERMINATION OF THE DISTRIBUTION CONSTANTS OF ANTICANCER AGENTS IMPLANTED INTO THE BRAIN TISSUE BY MEANS OF THE RANDOM WALK MODEL. *P. Kvasnička[3], J. Polák[1], M. Šramka[2],* [1]Institute of Preventive and Clinical Medicine, [2]Derer's Hospital with Policlinics, and [3]Faculty of Mathematics and Physics, Comenius University, Bratislava.

The random walk model was used for determination of the distribution constants (diffusion coefficient, elimination constant) of anticancer agents, which were implanted into the brain tissue (1). The model describes the distribution of the drugs as random walks of drug particles in the three dimensional space. The model is also suitable in the case, when nonhomogeneous space and various shapes and sizes of the capsules are taken into account. In our observations, the diffusion coefficient (1.2×10^{-6} $cm^2.s^{-1}$) and the constant of elimination (2.7×10^{-4} s^{-1}) of carboplatin, which is implanted in a capsule into the brain tissue of rabbits, were determined by the random walk model.

1. *Uffink G.J.M.:* Delft, 1990.

COMPARISON OF TWO APPROACHES TO MEASUREMENTS OF ELECTRICAL IMPEDANCE OF A GLASS MICROELECTRODE DESIGNED FOR EVALUATION OF TEMPERATURE CHANGES IN VARIOUS TISSUES. *F. Rech, I. Dittert, F. Vyskočil,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

A temperature sensitive microelectrode was designed for fast measurements of the temperature at the cellular level. The electrical impedance of the tip of the microelectrode changes with temperature. We designed an impulse system (STEP) for measurement of the above impedance changes. The system is based on a presettable negative input impedance of the current voltage converter. We compared the efficiency of the designed system STEP with the current RAMP method. We found the following advantages of the STEP system: 1. elimination of the danger of high voltage oscillations which could mechanically destroy the electrode, 2. the opportunity of setting the maximum sensitivity according to the interval to the temperatures to be measured.

IMAGE ANALYSIS ROUTINE FOR AN AUTOMATIC COUNTING OF PARTICLES IN A HISTOLOGICAL PREPARATION. *M. Gronychová, R. Palovský,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

We designed an image analysis routine for counting particles within an object, e.g. histological preparation. Digitization of image data was accomplished by a scanner of the desk top publishing system. The particles were grey level segmented, however, preprocessing of the image was necessary to eliminate local grey level changes of the background. This was accomplished by convolution in 3x3 windows. The counting of particles was made possible either by painting individual particles or by their erosion until the last element and counting the elements. The above method is being applied to evaluation of a number of neuronal fibres in the histological preparation of dorsal roots in rats. The results will be compared with stereological estimates of the application of which is simple. Since the design is based solely on general purpose hardware (e.g. IBM PC) and written in the C, it has broad exploitation opportunities especially if the processing time is not the most important phenomenon.

A NEW IBM PC BASED SYSTEM FOR AUTOMATION OF THE MORRIS WATER TANK EXPERIMENTS. *J. Kaminskij*[1], *J. Bureš, L. Nerad, I. Krekule,* [1]Brain Research Institute, Russian Academy of Sciences, St. Petersburg and Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

A new IBM PC based system has been developed for the control and evaluation of experiments in the Morris tank. In comparison with our previous system, this new modification has the following advantages: 1. broader operating conditions with respect to the rat tracking (a white rat on a black background and *vice versa* without resetting the system), 2. self checking and self diagnostics of the video channel, 3. recording of all experimental data on a disk for further processing, 4. custom-made computer-TV interface board is also capable of detecting specific events, i.e. neuronal unit activity, 5. more detailed on-line monitoring of the experiment (event occurrence, time spent etc.), 6. more feedback modalities available, 7. a CCD TV camera also sensitive in the infrared range is employed, 8. user friendly, menu driven software which provides also instructions for the experiment.

SELECTED SURFACE AREA MEASUREMENTS *VIA* A SURFACE RECONSTRUCTED BY A COMPUTER FROM SERIAL TISSUE SECTIONS. *R. Palovský, P. Karen, L. Kubínová, V. Valoušková, I. Krekule,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

We developed a system based on an IBM PC with VGA graphics and a digitizer DG-1 (ČSAV) which makes it possible to 1. reconstruct manually the profiles of serial sections of a tissue, 2. evaluate basic geometrical parameters of the tissue, (e.g. volume, surface area). The volume is estimated stereologically by using the Cavalieri principle, while the surface area is estimated as the sum of polygons (triangles) comprising the computer rendered surface of the tissue. The system is applied to the study of brain grafts especially to the quantification of the graft-host tissue interface which was categorized into 5 classes from neuronal interconnections up to empty cavities. The category of the tissue interface is coded by the colour of a given part of the profile. The software allows interactive editing of the rendered surface which is important in case of tissue branching. The further development of the system is aimed to the rendered surface shading and interactive manipulation (e.g. rotation) with them.

A SEMIAUTOMATIC IMAGE ANALYSIS SYSTEM. *P. Karen,* Institute of Physiology, Czechoslovak Academy of Sciences, Prague.

The developed, IBM PC based, semiautomatic system for image analysis, represents a spin-off of the graphical editor GSE described elsewhere. The system consists of an IBM-PC with EGA or higher graphics and a digitizer (graphical tablet) DG-1 (ČSAV). The cursor of the tablet was enhanced by a LED which makes it possible to put in data (contour of a profile) directly from a microscope which is equipped with a drawing arm. The software allows evaluation of both the primary geometrical parameters (area, perimeter) as well as the secondary ones (e.g. length of projections, axes etc.). Moreover, three-dimensional volume could be reconstructed as a stack of profiles or a wire frame object. The evaluated parameters could be exported in popular statistical packages, e.g. Statgraphics for further processing. The developed system will be adapted for other difitizers, e.g. Summagraphics in the near future.

PERSONAL COMPUTER SOFTWARE SUPPORT FOR A SPORTS MEDICINE DEPARTMENT. *V. Soulek, M. Stork*[1], University Hospital, Hradec Králové and [1]Technical University, Plzeň.

We describe a set of five user programs (SPIRO, TREN, TEP, KONSIL, CENIK) designed for sport medicine practice. The program SPIRO deals with complex functional diagnostic testing of a patient under a physical load. The program TREN determines an optimal, everyday sport training schedule according to a rather complicated algorithm. The program TEP is used for heart rate data processing. These data are loaded either *via* a keyboard or a special, customer-built interface. Both the input data and the results, i.e. the heart rate, are displayed as time functions, distribution tables, histograms etc. The program KONSIL is used for conciliar examinations. For example, it consists of clinical examinations, stress test diagnostics, optional exercise regime, measurement of energy consumption, risk of ICHS evaluation, etc. All the above programs exploit commonly used routines, e.g. print-out of results, graphical presentation, statistics, sorting of data etc. CENIK, the last member of the discussed program set is designed to bill patients and insurance companies for the above examinations.

XIV. Microsymposium on "The Hypetriglyceridaemic Rat"

THE GENETICALLY HYPERTRIGLYCERIDAEMIC, HYPERINSULINAEMIC, NON-OBESE RAT: ITS BASIC METABOLIC CHARACTERISTICS. *A. Vrána, L. Kazdová,* Institute for Clinical and Experimental Medicine, Prague.

We selected, after repeated measurement of triglyceridaemia both under basal conditions (intake of standard laboratory chow) and on the intake of hypertriglyceridaemia-inducing high-sucrose diet (70 cal %) in 60 males and 60 females of a random-bred Wistar strain (VELAZ), parent pairs with high triglyceridaemia. Their offsprings were selected according to the degree of hypertriglyceridaemia and breed. The line of hypertriglyceridaemic (HTg) rats obtained under the above conditions is today inbred (25 generations) and characterized by the following features: 1. Increased basal triglyceridaemia and a particularly marked increased hypertriglyceridaemic resoponse to high sucrose intake. 2. Increased liver synthesis of fatty acids and an increased output of VLDL triglycerides from the liver. 3. Consistent hyperinsulinaemia, mild hepatomegaly and mild hypertension. 4. The above line is not contaminated by obesity.

HYPOCHOLESTEROLAEMIC ACTION OF OMEGA-3 FATTY ACIDS VS. A NICOTINIC ACID ANALOGUE (ACIPIMOX): STUDIES IN HEREDITARY HYPERTRIGLYCERIDEAMIC RATS FED VARIOUS DIETS. *E. Šeboková, I. Klimeš, M. Hermann*[1], *M. Hromadová, A. Mitková,* Institute of Experimental Endocrinology, Slovak Academy of Sciences, Bratislava and [1]Institute of Medical Chemistry, Vienna University, Vienna, Austria.

To assess the role of liver LDL receptors in hypocholesterolaemic (hypo-CH) effect of fish (FO) oil (30 wt % n-3 PUFA) *per se*, or in combination with acipimox (ACI = 0.2 g per 100 g diet), hereditary hypertriglyceridaemic Wistar rats were fed basal or high sucrose (HS) (63 cal %) diet, with or without a FO supplement for 21 days. FO suppressed serum TG concentrations ($p < 0.02$) below the hereditary set limit. CH-lowering action of FO in the serum was also found in the liver ($p < 0.005$). The CH-lowering effect of ACI was followed by an accumulation of CH in the liver ($p < 0.005$); ACI + FO did not change the liver CH content. FO raised the LDL binding capacity of liver plasma membranes (1.8 fold, $p < 0.005$). LDL receptor activity was reduced in the HS + ACI group (by 40 %, $p < 0.05$) and remained unchanged in the HS + FO + ACI fed animals. Thus, FO and ACI exert an opposite influence on CH catabolism *via* hepatic LDL receptors.