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MEDICAL RESEARCH CENTRE

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RESEARCH REPORT

STUDIES ON THE FUNCTION OF THE NERVOUS SYSTEM AND ON MECHANISMS CONTROLLING BASIC FUNCTIONS OF THE ORGANISM

Departament of Neurophysiology
Head: Prof. Witold Karczewski

FUNCTIONAL ORGANIZATION OF RESPIRATORY NEURONAL NETWORK — SOURCES AND MECHANISMS OF RESPIRATORY DRIVE

1. The existence of at least three systems of connections coupling both respiratory half-centres was demonstrated: the main one — 2—4 mm rostrally to the obex is synchronizing rhythms, generated by both symmetrical half-centres. The second one — at the level of cervical spinal cord (C_2—C_6) is transmitting excitations and inhibitions between phrenic nuclei. The third one — at pontine level, is inhibitory to the central inspiratory activity. Experiments with total brain transections (split brain) failed to show any additional crossing connections that would be important for the generation or regulation of respiratory pattern (70).

2. It has been shown that consecutive midline transections of the medulla reduce the respiratory activity, whereas a section at the level of 1—4 mm rostral to obex abolishes high-frequency oscillations (HFO) in the respiratory system (139). It has been found that afferents from thoracic respiratory muscles act ipsilaterally, whereas information from the splanchnic nerve and limb muscles gets crossed and acts exclusively contralaterally. After splitting the medulla this information no longer affects the respiratory pattern (18).

3. Hypoxia (PaO_2 49.0 ±6.6 mmHg) and hypercapnia ($PaCO_2$ 52.0 ±5,7 mmHg) induce an increase in the amplitude and/or in the frequency of inspiratory discharges in both respiratory half-centres. A surprising feature of the „split-respiratory centre" is the possibility of chosing different „strategies" in response to the chemical stimuli: an increase in minute ventilation might be achieved by an increase in amplitude (V_1) without changes in frequency or even with its decrease in one half-centre, and by the classical reaction of increased both frequency and amplitude in the other. Additional transverse section of half of the medulla (hemisection) stops ipsilateral respiratory activity. Stimulation of the vagal input restores the activity. The contralateral half-centre and its responses to all stimuli are not affected by this procedure (51).

4. The effects of pentobarbital, ketamine, chloralose and morphine on Breuer-Hering inflation reflex were studied. Morphine appeared to be the only factor enhancing the B-H reflex. The same reflex was shown to be weakened during transition from rest to locomotion (45, 139). It has been shown that synthetic, double enkephalin given i.v. inhibits breathing. This inhibition being dose-dependent is reversed by naloxone. Ketamine has been found to induce typical apneustic breathing (prolonged inspiratory activity interrupted by short expirations) in cats anaesthetized with chloralose. Chemical stimuli (hypoxia and hypercapnia) restore the normal pattern of breathing.

5. It has been demonstrated that section of the greater petrosal nerve re-establishes normal cerebral circulation lowered by subarachnoid haemorrhage. A prophylactic neurotomy of this nerve also prevents the constriction of cerebral vessels after injection of blood into the subrachnoid space. Preliminary results with vagotomy showed that also this intervention might improve cerebral circulation reduced by subarachnoid haemorrhage.

See the LIST OF PUBLICATIONS:
7, 8, 9, 17, 18, 19, 35, 41, 42, 43, 45, 46, 49, 50, 51, 61, 66, 69, 70, 77, 78, 96, 128, 129, 139, 148.

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ADAPTATION TO PHYSICAL EXERCISE AND CHANGES IN ENVIRONMENT

I. Mechanisms controlling skeletal muscle metabolism

1. Continuing the study on thermal dependence of skeletal muscle metabolism experiments were perfomed on dogs exercising with and without external cooling as well as in resting dogs treated with bacterial pyrogen. Elevated body temperature was found to affect energy metabolism in active muscles, shifting an equilibrium of high energy phosphates to lower values of ATP and creatine phosphate, and accelerating glycolysis. These changes are considered as factors limiting performance time of a long-term physical exercise, during which a substantial elevation in core and muscle temperatures occurs.

2. In cooperation with the Department of Biochemistry, University of Oxford (Great Britain) mechanisms determining muscle insulin sensitivity were studied. A diminished sensitivity to insulin in skeletal muscles of rats with dietary-induced or genetic obesity was found to be improved by adenosine receptor antagonists, whereas the increased insulin sensitivity in muscles taken from the rats exposed to cold was completely reversed by addition of adenosine receptor agonists.
These findings indicate a role of adenosine as an important factor modifying effects of insulin on skeletal muscles in different physiological and pathological states.

3. In studies carried out with human subjects effects of physical exercise on the glucose-stimulated insulin secre-

tion were evaluated. It was found that after heavy, prolonged exercise the increases in blood peptide C and insulin concentrations were significantly diminished, whereas glucose tolerance was either unchanged in comparison with standard resting conditions or even improved. The latter effect was demonstrated in the subjects with impaired glucose tolerance. In some of these subjects an index of insulin extraction, calculated basing on blood peptide C and insulin concentrations, was also increased, indicating an enhanced tissue insulin uptake, apart from the diminished insulin secretion and increased tissue insulin sensitivity to this hormone.

4. In cooperation with the Institute of Experimental Endocrinology, Slovak Academy of Sciences in Bratislava (Czechoslovakia) the relative importance of exercise intensity and duration as well as of the total work output for hormonal responses to exercise was investigated in healthy, untrained human subjects. It was shown that the exercise-induced changes in the plasma catecholamine, cortisol, and testosterone concentrations depend to the greater extend on work intensity than on its duration or the total work output, whereas the magnitude of the changes in blood growth hormone and insulin concentrations is less dependent on exercise intensity.

5. Continuing the studies on the role of sympatho-adrenal system in the control of metabolism investigations were undertaken on the effects of prolonged hiperadrenalinemia on exercise tolerance and skeletal mescle metabolism in rats. Hiperadrenalinemia was produces by implanting sc. tablets releasing adrenaline at a constant rate for several days. The sustained high level of circulating adrenaline led to considerable impairement of working ability, which was accompanied by a decrease in energy substrate stores (including liver and muscle glycogen and muscle creatine phosphate) and insufficient ATP resynthesis in active muscles in spite of accelerated glycolysis.

In another series of experiments the effects of selective and nonselective blockade of beta-adrenergic receptors was studied in dogs performing heavy exercise. The data confirmed a substantial contribution of catecholamines in the control of metabolism in exercising muscles.

II. Adaptation of cardiovascular system to exercise in healthy humans

Continuing investigations on the cardiovascular responses to static exercise changes in systolic time intervals (STI) of the left ventricle and in arterial blood pressure were followed during and after a sustained hand-grip. Comparison of the post-exercise measurements made in the subjects with and without blood flow occlusion indicate that the pressor response to this kind of exercise is initiated by stimulation of „metabolic receptors" in working muscles, while the increases in heart rate and the cardiac muscle contractility are controlled by some other mechanisms.

In the studies with young long-distance runners further data were provided concerning the so called „athlete's heart", using echo-cardiography. The data can be useful for differentation of some physiological, training dependent characteristics of the cardiac function from the pathological changes.

In the longitudinal studies performed with healthy, sedentary human subjects the dynamics of changes in working capacity and cardiovascular system were followed during 3-month aerobic training. A significant improvement of aerobic capacity with a concomittant reduction of the submaximal heart rate and blood pressure as well as an increase in the maximal stroke volume were found already after 3—4 weeks of training, while an enhancement of the cardiac muscle contractility (as evaluated using polycardiography) did not occur before 8 weeks of training. The studies showed also that the cardiovascular response to a static exercise (hand-grip) performed with untrained muscles is reduced by the aerobic leg training.

III. Cardiovascular and metabolic responses to exercise in cardiac patients

The effect of long-term exercise of low intensity on some indices of lipid and carbohydrate metabolism was studied in patients with coronary heart disease. It was found that such type of activity can be recommended for the cardiac patients with metabolic disorders, since it effectively decreases elevated blood triglycerides, cholesterol, and glucose concentrations.

Besides, an influence of isosorbide dinitrate on cardiovascular respones to physical exercise was examined in patients with coronary heart disease and healthy control subjects. In the patients with severe coronary insufficiency a marked reduction of ischemic symptoms, and an improvement of hemodynamics were found. However, the drug often caused and impairement of orthostatic tolerance after cessation of prolonged exercise.

IV. Thermoregulation

A relationship between the amount of sweat secreted and evaporated was determined in healthy men during physical exercise performed under different thermal conditions. It was found that about 70% of sweat is evaporating, and this percentage is only slightly increasing with an increment of ambient temperature. It was also demonstrated that body hydration before exercise lowers the threshold of sweating response, and improves effectiveness of body cooling, thereby reducing an increase in core body temperature.

Changes in body temperatures and heat elimination were compared in lean and obese patients following surgical operation. In obese patients body temperature returned to normal values after a longer delay than in the lean ones.

V. Extracellular electrolytes in the kidney

Further improvements were introduced to the original method enabling assessment of electrolyte concentration in

the renal medullary interstitium from continuous measurements of the electrical admittance of renal tissue. By modifying the geometry ef electrode sets for dog and rabbit kidney the tissue damage secondary to insection of admittance electrodes was minimized while high quality of recordings was maintained.

The results suggesting that furosemide action on the rabbit nephron includes the thin ascending limb of the loop of Henle were supported by providing extensive in vitro tissue calibration data which established a clear correlation between electrical admittance and tissue osmolality or sodium concentration. The superiority of 3500 Hz over 400 Hz measuring frequency was confirmed.

In cooperation with the Department of Physiology and Biophysics of the University of Uppsala, a method was developed for in situ measurements of ionic concentration in picaliter samples of renal tubular fluid. This is another application of conductance or admittance measurements for estimation of electrolyte concentration in physiological experiments and follows our own application of the same approach for tissue studies of the in situ kidney.

See the LIST OF PUBLICATIONS:
10, 11, 12, 13, 14, 15, 16, 21, 22, 23, 24, 36, 37, 44, 52, 53, 54, 59, 72, 75, 76, 79, 83, 84, 85, 86, 87, 88, 89, 90, 93, 94, 95, 100, 116, 117, 118, 130, 145, 147, 156, 158, 180, 181, 182.

Cardiovascular Laboratory
Head: Assoc. prof. K. Herbaczyńska-Cedro

CONTROL MECHANISMS OF CARDIOVASCULAR SYSTEM IN DEFINED PATHOLOGICAL CONDITIONS

In view of clinical data on beneficial effect of prostacyclin (PGI_2) in cerebral ischemic stroke, the influence of PGI_2 on the anoxia — induced damage of nervous tissue was studied in vitro. Pretreatment of the rat cerebellar tissue culture with PGI_2 (50 μg/ml medium) prior to anoxia led to preservation of ultrastructure, prevention of swelling and acidosis. PGI_2 — induced cytoprotein may contribute to therapeutic effectiveness of PGI_2 in brain damage. In an attempt to elucidate cellular mechanisms involved in the stress-induced myocardial injury, products of lipid peroxidation malonolialdehyde (MDA), conjugated double bonds (CDB) and mitochondrial high energy phosphates were measured in the myocardium of pigs subjected to stress of 2 hours immobilization and in that of pigs subjected to coronary occlusion of 15 min duration. Both MDA and CDB were raised in the myocardium of stressed animals and they were significantly increased in ischemic myocardium. Contents of high energy phosphates and the respiratory control index of mitochondria isolated from stressed hearts were diminished.

The results suggest that enhancement of lipid peroxidation in myocardial membranes, reflecting increased generation of oxygen free radicals, contributes to the stress- induced myocardial injury. Catecholamines are considered as factors responsible for an increased free radical load in tissues. High level of lipid peroxidation products was detected in the myocardium of rabbits infused with adrenaline (1 μg/kg/min) for 2 hours. Investigations of hormonal, metabolic and ultrastructural alterations caused by stress are in progress.

See the LIST OF PUBLICATIONS:
60

Laboratory of Experimental Surgery
Head: Assoc. prof. Jerzy Borkowski

CLINICAL AND EXPERIMENTAL STUDIES ON PERIPHERAL CIRCULATION

Research work on the role of transcutaneous electrical stimulation (TES) in the therapy of some vascular diseases (e.g. Raynaud's and Buerger's diseases) were continued, being focused on three problems:

1. estimation of effectiveness of TES in a given patient,
2. role of TES in prevention of some vascular diseases (mainly Raynaud's disease),
3. choice of proper TES parameters for a given patient.

To estimate effectiveness of TES in a given patient visual thermography was used. The effect of TES on cutaneous microcirculation at lower ambient temperatures (by 7 — 9° C) was assessed. Twenty nine patients with Raynaud's disease and syndrome, 21 patients with Buerger's disease and 10 healthy volunteers were examined. A significant increase in temperature of the stimulated area was observed. The temperature elevation was most pronounced in cases with Raynaud's disease and syndrome. Cessation of Raynaud phenomenon, caused by an exposure to low ambient temperature, was noted in 72% of cases.

These results indicate that TES can be satisfactorily used not only in therapy but also in prevention of vascular diseases.

See the LIST OF PUBLICATIONS:
165, 166

STUDIES ON THE STRUCTURE AND BIOLOGICAL PROPERTIES OF THE NERVOUS TISSUE

Department of Neuropathology
Head: Prof. Mirosław Mossakowski

1. Investigations on the pathomechanism of hepatic encephalopathy (HE) were continued with the use of the thioacetamide model. Early stages of HE in this model were found to be characterized by morphological manifestations of functional and metabolic activation of the astroglia, with complete absence of degenerative changes. An increased pinocytic transport in cerebral capillaries was observed; however, the blood-brain barrier appeared undisturbed. ECoG recordings revealed progression of the disturbances, which correlated in time with the growing inefficiency of ammonia detoxication in the brain.

The progression of the pathological process was found to be accompanied by a gradual decrease in the number of GABA receptors on the synaptic membranes and their increased affinity to the ligand, which may be instrumental in the activation of inhibitory neurotransmission.

Activation of astrocytes in the early stages of HE was manifested by hyperpolarization of the astroglial cell membranes, most likely related to the increase of the Na^+/K^+ ATPase activity. This suggests that astrocytes are then capable of more active removal of potassium from the extracellular space of the CNS. An enhanced GABA uptake into the astrocytes was observed, indicating more efficient inactivation of the neurotransmitter. The results taken together may be interpreted to reflect improved buffering function of astrocytes in early HE.

2. Studies on the pathomechanism of ischemic brain damage revealed that indomethacin prevents the selective loss of nerve cells in the CA_1 sector of Amon's horn after complete cerebral ischemia. The cytoprotective effect of this drug depends upon its dose and route of administration. Treatment with prostacyclin PGI_2 was found to speed up the recovery of the bioelectric activity and to render normalization more complete, as well as to extend the time of survival following the ischemic episode. In cases in which the drug was applied, the pathomorphological picture of the brain after ischemia was characterized by the absence of the features of cytotoxic and vasogenic edema and by improved maintainence of the neuronal population. The results point to the vasodilatory, antiaggregating and cytoprotective mechanism of PGI_2 action. In addition, the drug was found to prevent cardiac failure and to normalize systemic blood pressure. Electron microscopic and histochemical studies on tissue cultures subjected to anoxia confirmed the cytoprotective effects of PGI_2.

3. Studies on manganese encephalopathy revealed that chronic intoxication with manganese salts, producing irreversible tissue damage with features of leucoencephalopathy, leads to blood-brain barrier damage as tested with horseradish peroxidase (high MW form). This damage is accompanied by changes in alkaline phosphatase and adenyl cyclase activity in the blood tissue interphase, which are indicative of the impairment of blood-brain barrier mechanisms as well. The results obtained suggest the role of vasogenic factors in the development of structural changes induced by manganese.

4. Investigations on the modelling of the Parkinsonian extrapyramidal system in animals by means of manganese and cobaltous salts confirmed the role of the vasogenic factors. The syndrome was found to develop only when intoxication was accompanied by at least unilateral ligation of the carotid artery. At the biochemical level, both metals were

found to interfere with the dopaminergic system, which was manifested by the decrease of dopamine content in all the CNS structures. A concomittant increase of GABA level was indicative of the imbalance between the two neurotransmitter systems. There was a good correlation between the severity of these biochemical disturbances and the advancement of the extrapyramidal syndrome. The tissue damage following cobalt treatment resembled that observed in manganese encephalopathy, which was characterized by selective impairment of the white matter. Electron microscopic analysis revealed considerable swelling of axons and myelin sheaths damage, which was related to oligodendroglial swelling.

5. Studies on the disturbances of neuromediator system in clinical hyperkineses syndrome included determinations of GABA and homovanilic acid in the cerebrospinal fluid of patients with Parkinson's disease, Parkinsonian syndrome accompanying artero sclerosis and Huntington's disease. The disturbances in the content of both compounds were found to correlate with the severity of the symptoms and the kind of treatment. Serum and cerebrospinal fluid from patients with Huntington's chorea were found to produce in cultures of rat striatum changes identical to those obtained with kainic acid. No such changes were induced by serum or cerebrospinal fluids from patients with Parkinson's disease.

6. Investigations on the effects of ethylnitrosourea on the central nervous system included characterization of the subsequent stages of the induced carcinogenesis. These stages were shown to be related to myelinogenic glia: oligodendrocytes in the central nervous system and Schwann cells in the peripheral nervous system.

7. Studies on the antigenic properties of glial cells have dealt with the cellular localization of glutamine synthetase and the relation of its appearance to the degree of the maturity of astrocytes in tissue culture.

See the LIST OF PUBLICATIONS:
1, 2, 3, 4, 5, 20, 25, 63, 64, 65, 80, 81, 82, 92, 105, 106, 107, 108, 109, 110, 111, 112, 113, 114, 123, 124, 125, 126, 127, 132, 133, 134, 135, 136, 137, 138, 149, 151, 152, 153, 154, 155, 159, 160, 161, 167, 168, 175, 177

Department of Neurochemistry
Head: Assoc. prof. J. Łazarewicz

EFFECT OF HYPERTHERMIA AND HYPERTHERMIA WITH HYPOXIA AND HYPERCAPNIA ON THE SYNAPTIC PROCESSES

Participation of membrane lipid and protein in the regulation of metabolic activity of synaptic endings in hyperthermia

In previous studies on the hyperthermic insult on brain metabolism the experiments were carried out, on the homogenates from different parts of the rabbit brain. It was demonstrated that a short-term hyperthermia (3h at 40—41°C) induces an inhibition of lipid peroxidation reaction. Moreover, the lower accumulation of calcium ions into synaptosomes was found.

The aim of the studies performed in 1984 was to determine both localisation and the role of membrane lipids and proteins in the disturbances of peroxidation processes and in the transport of calcium ions.

Mitochondria, microsomes and synaptosomes from the brain cortex were used for the determination of calcium uptake and for measurements of the content of thiobarbituric reactive substances (TBR-S) and thiol groups.

Short-term hyperthermia decreased the accumulation of Ca^{2+} ions in the mitochondria during the oxidation of glutamate and malate but without any effect on Ca^{2+} uptake in the presence of succinate and rotenone.

The accumulation of calcium ions occurred together with increasing fluorescence of chlorotetracycline. This fluorescence decreases in the mitochondria isolated from the animals submitted to hyperthermia.

The release of calcium from hyperthermic mitochondria was not changed as compared to controls. Calcium uptake into microsomes was decreased by hyperthermia by about 20% but it was activated by about 70% into symaptosomes.

Concomitantly, the additionally accumulated Ca^{2+} ions were released from synaptosomes during the first 3 min.

The content of thiobarbituric reactive substances in synaptosomes was significantly decreased. Similar changes were observed in microsomes but only a slight activation of TBR-S production was found in mitochondria. The content of sulphydryl groups was not changed.

The results indicate a decreased ability to accumulate calcium ions by brain mitochondria and microsomes isolated from the brain submitted to hyperthermia. Moreover, an increased permeability of the synaptic plasma membrane for this ion was observed. Thus, a short-term hyperthermia disturbs calcium homeostasis in c.n.s., which may agree with the kown effect of hyperthermia on calcium deprivation.

It is difficult to exclude the secondary effect of the membrane preparation on the membrane structure and function affected former by hyperthermic insult.

The changes in Thiobarbituric Reactive Substances (TBR-S) observed in subcellular fraction from the rat brain are not exactly the same as observed in the brain homogenate. The activation of peroxidation reaction in mitochondria can be the unspecific effect produced during the preparation of this subcellular fraction.

For better understanding of the hyperthermia effect on the peroxidations reactions the more detailed studies will be performed to elucidate the mechanism of disturbances in peroxidation processes under these conditions.

EVALUATION OF MEMBRANE DISTURBANCES AND SYNAPTIC FUNCTION IN DIFFERENT PATHOLOGICAL CONDITIONS

The modulation of synaptic function by alteration of protein and lipid metabolism

The studies on the membrane lipid and protein composition and metabolism as well as on functional changes of synaptic endings in different patological conditions were continuated.

Investigations on protein metabolism have been concentrated on the neutral protease. This enzyme, activated by Ca^{2+} ions, was isolated and purified from frogs c.n.s. The specificity of substrate and susceptibility of protease to the action of endo and exo inhibitors was determined. The proteolytic action of this enzyme towards the fast transported protein in c.n.s. was demonstrated. The studies on lipid metabolism in the brain concerned localisation and regulation of diacylglycerol kinase. The very wide distribution of this enzyme was demonstrated in the all c.n.s. membranes and in cytosol. The highest activity was found in synaptic vesicles which characterize very low endogenous pool of FFA. Free fatty acids and calcium ion are endogenous inhibitors of diacylglycerol kinase. This enzyme converts diacylglycerol to phosphatidic acid, protecting in this way the membrane structure and function against damaging effect of diacylglycerols. It was demonstrated that free fatty acids, liberated during ischemia, are only partly responsible for the inhibition of arachidonate uptake into phosphatidylinositol of the brain synaptosomes.

These findings suggest that ischemia produces a stable damage of enzyme-acyl-CoA synthetase.

In the following studies on the patogenetic role of free fatty acids in synaptic patology, besides their effect on the neurotransmitter uptake, an activation of GABA and dopamine release was found. It was postulated that depolarization of synaptosomes produced by unsaturated fatty acids

affects membrane potential and increases permeability of plasma membrane for calcium ions.

Lipid peroxydation of fatty acids, activated in vitro by exposure the membranes to 60 μM Fe^{2+} with 200 μM ascorbic acid during 60 sec or 5 min, modifies the lipid comonent of synaptoplasma membrane and leads to an inhibition of GABA, choline and Ca^{2+} uptake and to the stimulation of dopamine uptake (26).

It was demonstrated that taurine, the putative neurotransmitter and neuromodulator liberated in the brain during ischemia, can modify calcium membrane binding and decrease depolarization-dependent calcium uptake into the rabbit synaptosomes. The results suggest that taurine may influence the neuronal membrane excitability by its effect on membrane-calcium interaction and may be involved in the inhibition of Ca^{2+} ion influx into ischemic synaptosomes.

Studying the uptake of neurotransmitters into synaptosomal fraction form brain hemispheres after chronic administration of lithium an increase in GABA release without any effect on the uptake of dopamine, serotonine and noradrenaline was found. Chronic administration of lithium (in a dose of 150 mg/kg b.w. during 6 weeks) stabilized the binding of dopamine to synapticplasma membrane and decreased the amount of binding sites and dissociation constant.

In investigations on the effect of acute intoxication of thioacetamide on the function of cell fraction enriched in astrocytes stimulation of Na^+-K^+ ATPase and GABA uptake was observed. The results suggest the protective role of astrocytes in developing of liver encephalopathies.

The studies on lipid composition of the brain white and gray matter during development of the myelin defficient „pt" rabbit mutant revealed characteristic changes in typical myelin lipids. The lower content of cerebrosides and sulphatides as well as the myelin specific phospholipids (sphingomyelin and ethanolamine plasmalogens) in white matter of „pt" rabbit was demonstrated without any changes in

gangliosides — neuronal markers. The ratio of glycolipids to phospholipids indicates retardation of myelin formation, evidencing severe disturbances of metabolic maturation of oligodendrocytes.

See the LIST OF PUBLICATIONS:
5, 25, 26, 33, 157

Department of Neurosurgery
Head: Prof. Eugeniusz Mempel

THALAMIC SOMATOSENSORY EVOKED POTENTIALS (SEP) AFTER PERIPHERAL STIMULATION AND CORTICAL SEP AFTER THALAMIC Vim STIMULATION

Cortical somatosensory potentials (SEP) evoked by direct electrical stimulation of the VL and VPL thalamic nuclei in cats

Experiments were performed on 24 animals. It was shown that the main stream of efferent projections from VL goes to 6 and 4 cortical areas, whereas projection from VPL goes mainly to areas 4, 3a, 3b and 5 and the densent projection reaches the area 3a and 3b. From the configuration of the recorded cortical SEP's it can be postulated, that the synaptic terminals from each of those nuclei are localised in different cortical layers. Namely in layers 4 and 5 synaptic terminals of VL afferents are localised, whereas in cortical layer 3 and 2 VPL terminals are grouped.

These results have been confirmed by data obtained in neuroanatomical investigations concerning the projection of specific thalamic nuclei to sensory-motor cortex.

The verbal memory processes in patients with involuntary movements treated by thalamotomy

The comparative studies on verbal memory were performed on fivteen patients with extrapiramidal syndrome and with involuntary movements treated by the ventrolateral (VL) thalamotomy. Disturbances of verbal memory in the stage of short-term memory, long-term memory and an impairment of verbal learning were found. Results of an analysis proved that these disturbances are caused by reduced retrieval of verbal information from the memory store in interference conditions. The patients with right thalamotomy had this specific deficit before the operation, but in the group of patients with left thalamotomy these disturbances appeared only after the operation and were maintained for a long period. The obtained results suggest that both the left and right thalamus contribute to the processes of verbal memory.

Examination of speech disorders and other gnostic function of the brain

Two main studies were performed:

a. experimental examination of stereoscopic vision in patients with the brain focal damage and
b. clinical examination of 28 children dyslexia and disgraphia.

Visual, motoric amd auditive disfunctions were revealed in the examined children.

See the LIST OF PUBLICATIONS:
150

Laboratory of Developmental Neuropathology
Head: Prof. Maria Dąmbska

THE DEVELOPMENT OF THE NERVOUS SYSTEM UNDER NORMAL AND DISTURBING ITS STRUCTURAL MATURATION CONDITIONS

An influence of cytostatic drugs on the maturing nervous system was the aim of investigations.

The morphologic changes in the central nervous system (CNS) in children dead from neoplastic disease were studied. The material consisted of 25 brains of young patients aged 1 month to 10 years. All were treated with polychemotherapy according to classical programs. Beside this, the majority underwent also surgical treatment and radiotherapy. The material was divided in groups characterized by similar course and treatment of neoplastic disease. Neuropathological picture in cases treated with cytostatic drugs only and with chemotherapy and x-irradiation were compared with those treated only surgically. In the brains of children with prolonged chemotherapy parenchymal changes in cerebral and cerebellar cortex as far as fibrosis and hyalinisation of vascular walls were observed. The degree of lesions depended on the time of treatment and survival. It was concluded that the treatment with cytostatic drugs, which prolongs the survival time in patients caused not only formation of neoplastic encephalopathy but modified its picture, and most probably influenced its intensity.

Results obtained in the investigations on human autopsy material were supported by those of experimental studies carried out on the rabbit and rat brains. It was found that two cytostaticcyclophosphamid and lomustin (CCNU) induce pathologic changes particularly in perivascular astroglia, leading even to irreversible damage of the nervous tissue.

See the LIST OF PUBLICATIONS:
27, 28, 29, 30, 31, 32, 67, 68, 97, 101, 102, 103, 104, 164, 179

Department of Comparative Neurology
Head: Assoc. prof. Irmina Zelman

EFFECT OF „pt" MUTATION ON VARIOUS LINKS OF CLOSED NEURONAL CIRCUITS

STUDIES ON TRANSPLANTATION AND EXPERIMENTAL SURGERY

Department of Experimental Surgery and Transplantation
Head: Prof. Waldemar Olszewski

REGULATION OF IMMUNE PROTEINS TRANSPORT BLOOD — LYMPH

The parameters of active lymph flow in efferent lymph vessels of leg was examined in 15 healthy volunteers. It was found that an elevation of venous pressure by 50 mmHg is followed in the first 30 min by an increase of mean pressure in lymph vessels by about 50 mmHg. This increase is caused by transposition of total interstitial pressure on walls of the lymph vessels. In most cases a rapid increase of venous pressure was accompanied by a decrease of the amplitude and frequency of lymphatic pulse wave. In some cases the pulse wave disappeared completely, most probably as a result of a sudden decrease of the venous flow and capillary filtration. It seems that a decrease of lymph transport in the acute venous hypertension is caused by a drop of blood flow and as a consequence by the decrease of the capillary filtration and tissue fluid and lymph formation.

Experiments on capillary transport of ticarcillin and clavulonic acid as well as preliminary trials on an enhancement of their transport by sadamin were also performed. It was found that half-time of a ticarcillin elimination is 1.1 h in serum and 1.14 h in lymph while that of clavulonic acid is 0.91 and 1.14 h, respectively. The area under a concentration curve (AUC) for ticarcillin in lymph was 58% of the value for serum and for clavulonic acid 81%. The results show a high ability of these drugs to penetrate into an interstitial space. Sadamin caused an earlier appearance of both drugs in lymph.

An influence of hydrocortizon (one dose 400 mg, intravenously) on immunoglobulin transport in efferent lymph of 8 healthy volunteers was also examined. A peak of immunoglobulin transport from blood capillary to efferent lymph of leg was observed after about 4 h at rest and 1 h during leg movement. A total transport of proteins and IgM m.w. 400000 to the interstitial space was enhanced, which suggests that where was an increase in permeability of intraendothelial pores.

IMMUNOLOGICAL DIFFERENTIATION OF LIVING ORGANISMS

Spontaneous migration of lymphocytes through the lymphoid tissues

A method for storage of human granulocytes at the temperature of liquid nitrogen was established. This method was also adapted for storage of the dog and rat granulocytes. Granulocytes were suspended in a cryoprotective mixture consisting of 12% DMSO, 10% HES and 8% BSA, incubated for 5 min at temp. 277°K and cooled with a speed of 1 K/min to 248°K. This temperature was maintained for 5 min, then lowered to 173°K with a speed od 10 K/min and placed in a liquid nitrogen. Granulocyted storaged for 1 year showed in experiments in vitro about 90% viability, high peroxidase activity and NBT reduction. Distribution of labelled fresh and preserved granulocytes injected intravenously into the rat was similar in both xenogeneic and syngeneic system. An increase in radioactivity in lymph tissues and a limited increase in the liver were also observed. These findings suggest that there is a redistribution of live granulocytes between the lymphatic organs as it is observed in case of lymphocytes.

A further characterization of lymphocytes and Langerhans cells from efferent lymph of humans was also performed. It was found that OKIa1 monoclonal antibody did not

block autotransformation and the response to PHA of lymph cells but blocked mononuclear cells of blood. It may suggest that the signal for activation has been given to Langerhans cells already in vivo. OKT6 did not show any blocking effect on lymph cells while it had a stimulatory effect on blood cells. OKT3 antibody acted similarily. A culture of lymph cells for 72 h both with and without PHA led to an increase of $OKT4^+$ (helper) and decrease of $OKT8^+$ (suppressor) cell number. The blood cells behaved adversely. The percentage of cells cultured for 72 h and labelled with $OKIa1^+$ was lowered both in lymph and blood which suggest a rather decreased expression of Ia antigens. The percentage of $OKM1^+$ and $OKT6^+$ cells remained unchanged. The percentage of $OKT9^+$ cells in lymph was 2%, in blood 0%. After a 72 h culture the percentage of these cells increased more significantly in lymph than in blood. Lymph cells responded several times higher to PHA than blood cells. Lymph cells irradiated with 1500 r presented PHA to autologous cells several times stronger than adherent cells of blood.

A further characterization of natural cytotoxic non-parenchymal liver cells was also performed. A population of these cells was separated on Percoll gradient into 7 layers. The highest percentage of cells with azurophilic granules (LGL), $OKT8^+$ cells and cytotoxic cells towards K562 and YAC-1 tumour target cells was found in layers 2—4. This separation allowed to obtain a population enriched to 40—50% in NK cells. The population of these cells was also separated from the whole population by a panning method using $OX8^+$ was significantly more cytotoxic compared to $OX8^-$ cell population. Blocking of cytotoxicity with d-mannose did not diminish the level of the response to YAC-1 tumour cells which suggests that they belong to the NK cell (natural killer) and not NC (natural cytotoxic) population.

Prolongation of allograft survival

Clinical data show a statistically higher survival rate of renal grafts after 1 and 3 years in individuals who received

both donor — specific and — nonspecific blood transfusion. In own experiments carried out in dogs 3 protocols for blood transfusions before grafting were applied: rising doses of blood and supplementation with oral immuran, 3 doses of blood and immuran, intravenously, and 3 doses of blood infused into the portal system supplemented with oral immuran. The activity of recipient peripheral blood suppressor cells, the level of cytotoxic antibodies and the response of recipient in MLR were examined in vitro. Dogs treated according to the first and second protocols had the survival time similar to those in the control group. However, in the group of dogs which received blood transfusion into the portal system out of 8 graft recipients 3 survived 18, 32 and 60 days. Experiments in vitro did not show any changes in the level of activity of recipient blood suppressor cells compared to donor lymphocytes. There was no correlation between the values of suppression and survival time. No significant increase was observed in the level of cytotoxic antibodies.

In experiments dealing with characterization of donor cell population which causes a prolongation of graft survival, a distribution of donor lymphocytes injected with or without recipient antiserum against donor lymphocytes (enhancement protocol) were examined. It was found that donor lymphocytes injected just before or 24 h before alloserum treatment accumulate in a higher percentage in the liver and spleen than when they are without serum. Particularly high level of radioactivity remained in the spleen. In the lymph nodes the level of radioactivity was significantly lower after lymphocyte and antiserum treatment thah after sole lymphocytes administration. Splenectomy did not cause any changes in the amount of accumulated lymphocytes in the liver, however, enhanced trapping of lymphocytes in the lymph nodes. Injection of alloserum abolished completely the enhanced accumulation of donor lymphocytes in lymph nodes. These preliminary studies showed that alloserum of recipient against donor lymphocytes eliminates a population of lymphocytes accumulating in lymph nodes and leads to an enhanced and prolonged accumulation of these cells

in the liver and spleen. In the animals undergoing such a protocol heart allograft survived indefinitely without additional immunosuppression.

The investigations on the autologous mixed lymphocyte reaction (AMLR) were continued in two directions: an evaluation of suppressor cell function and activation of T lymphocytes in AMLR with non-T cells. The changes in postoperative suppressor cell activity were compared in a group of patients with nonadvanced (stage 0) and advanced tumour. Significant differences were observed between two groups of patients. In patients with advanced malignant disease no increase in the level of circulating monocytes was observed, the response of lymphocytes to PHA did not decrease. Preoperative level of suppressor cell activity in these patients was high and after the operation it decreased only slightly. It is suggested that these differences are due to a limited mobilization and redistribution of lymphocytes in the patients with advanced malignant disease.

In studies on AMLR after the operation it was shown that cholecystectomy causes a decrease or complete abolishement of this reaction on day 1. On day 3 an increase or reappearance of AMLR was noted. It seems that a decrease in the number of circulating mature T lymphocytes in blood may be responsible for the observed decrease. The changes in stimulatory capacity of non-T cells and the suppressive effect of monocytes should also be taken into consideration.

See the LIST OF PUBLICATIONS:
6, 34, 40, 47, 48, 55, 56, 57, 58, 62, 71, 98, 99, 115, 119, 120, 121, 122, 140, 142, 143, 144

OTHER RESEARCH WORKS

Mental Health Department
Head: Dr Z. Juczyński

BIOLOGICAL, PSYCHOLOGICAL AND SOCIAL CONDITIONS OF HUMAN ACTIVITY

1. Social and professional activity of patients after the first myocardial infraction was estimated. Data from 80 patients were collected using a standardized questionnaire interview. It was found, that the most significant negative effects of heart infarct concerns profesional activity (only 57.5% of patients returned to work) and family life. Cultural and social activity both before and after infraction, showed a low degree of involvement. Moreover, it has become evident that a year after the infraction among many advices given by physicians only those concerning alkohol and tobacco consumption were respected. Different kinds of the patients activity appear to be closely related to each other, particularly professional activity which stimulates the other fields of life activity.

2. Investigations on life activity of 100 alcoholics, now aged 40—45 were continued. A particular attention was given to premature mortality, professional degradation, as well as to the degree of help given to them in their working places. The thesis of Drew (1968) concerning the self-limiting nature of alcohol disease has been verified.

3. Studies on social parasitism in adults were conducted in a group of 845 persons avoiding work. Among declared motives of avoiding work — low salar was dominating. Professional instability correlated well with low family stability. Fifty percent of respondents had transgressed or done various delinquencies end, as a result, 84% of them were already imprisoned.

See the LIST OF PUBLICATIONS:
73, 74, 91, 131, 141, 146, 169, 170, 171

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Department for Surgical Research and Transplantation

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Łukomska B.	Karolinska Inst., Stokholm, Sweden
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	Surgical Clinic Univ. of Bonn, GFR
	Dept. Surgery Univ. of Cairo, Egipt
	Dept. Surgery Univ. of Alexandria, Egipt
Orłowska E.	Harvard Med. School, Beth Israel Hosp., Boston, USA (long term visit)
Ryffa T.	Clin. Hosp. Univ. Timisoara, Rumania
Ziółkowska A.	Det Norske Radium Hosp., Oslo, Norway (long term visit)

PARTICIPATION IN INTERNATIONAL SCIENTIFIC MEETINGS IN 1984

25th Intern. Conference on the Biochemistry of Lipids „A Jubilee Conference" 4—7 September, 1984, Antwerp, Belgium
Domańska-Janik K., Strosznajder J.

European Developmental Biology Congress
2—7 September 1984, Southampton, U.K.
Maślińska D.

VIIth Intern. Congress of Histochem. and Cytochemistry,
5—11 August 1984, Helsinki, Finland
Renkawek K., Szumańska G.,

XVIIth Danube Symposia for Neurological Sciences
8—11 October 1984, Moscow, USRR
Dąmbska M., Kida E., Mossakowski M., Weinrauder-Semkow H.,

V Meeting of the Intern. Society for Heart Research
19—22 September 1984, Geneva, Switzerland
Herbaczyńska-Cedro K., Kwiatkowska-Patzer B.

Symposium on occasion of the 10th anniversary of cardiac rehabilitation groups in Cologne
24—25 August 1984, Köln, GFR
Kozłowski S.

I Meeting of the Intern. Soc. for Heart Research (ISHR) Sub-Section of Socialist Countries
24—25 August 1984, Szeged, Hungary
Patzer B.

Cajal Conference on Neurobiology
17—22 September, Madrit, Spain
Maślińska D.

Symposium „Neurogenesis of Central Respiratory: electrophysiological, pharmacological and pathological aspects", (CNRS)
18—22 September, Marseill, France
Karczewski W.

VIth Quadrilateral Symposium of Experm. Surgery
5—7 September 1984, Brno, Czechoslovakia
Grochowicz P., Muranyi M., Ryffa T.

6th European Immunology Meeting
3—8 September 1984, Interlaken, Switzerland
Gałkowska H., Grzelak J., Łukomska B.,

8th Congress of the Intern. Microsurgical Soc.
22—26 August 1984, Pittsburgh, USA
Grochowicz P., Olszewski W.

Meeting of the Biochem. Soc.
17—21 July 1984, Leeds, U.K.
Budohoski L.

X Intern. Meeting of the Transplantation Soc.
26—31 August 1984, Minneapolis, USA
Olszewski W.

Symposium „Function of Neuroglia" (IBRO)
18—24 November 1984, Tibilisi, USRR
Albrecht J., Renkawek K.

IXth European Congress of Cardiology
8—12 July 84, Düsseldorf GFR
Czarnecki W., Herbaczyńska-Cedro K.,

8th European Congress on Electron Microscopy
13—18 August 1984, Budapest, Hungary
Zaręba-Kowalska A.

IUPHAR 9th Intern. Congr. of Pharmocology
29 July — 3 August 1984, London, U.K.
Czarnecki W., Herbaczyńska-Cedro K.

Third Intern. Congr. on Cell Biology
26—31 August 1984, Tokyo, Japan
Strosznajder J., Renkawek K.

ISOTT Meeting of the Intern. Soc. on Oxygen Transport to Tissue 26—30 August 1984, Nijnregen, Holland
Grieb P.

XVIIIth Meeting of the European Soc. for Clinical Investigation
17—19 April 1984, Milano, Italy
Czarnecki W., Herbaczyńska-Cedro K.

16th FEBS Meeting
25—30 June 1984, Moscow, USRR
Domańska-Janik K., Strosznajder J., Śmiałek M.

Vth Meeting of the European Soc. for Neurochemistry
20—25 August 1984, Budapest, Hungary
Łazarewicz J., Rafałowska U., Śmiałek M., Wysmyk-Cybula K., Zalewska T.

XXII Intern. Symp. on Biological Models
24-28 April 1984, Hrubá Skála, Czechoslovakia
Wyrzykowska-Sienkiewicz A.

Jubilee Meeting to celebrate Professor Petri's 70th birthday
6 February 1984, Szeged, Hungary
Olszewski W.

XIX Congress of the Europ. Soc. for Surgical Research
9—11 April 1984, Zürich, Switzerland
Grzelak J., Łukomska B., Olszewski W., Ryffa T.

Congress of the European Soc. for Clinical Respiratory Physiology (SEPCR)
11—15 June 1984, Barcelona, Spain
Budzińska K., Głogowska M., Głowicki K., Gromysz H., Karczewski W., Romaniuk J., Szereda-Przestaszewska M.