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11th Nordic Meeting on Cerebrovascular Diseases and 2nd Biennial Kuopio Symposium on Ischaemic Stroke

Kuopio Music Centre, Kuopio, Finland
August 11-14, 2001

Program
and
Abstracts

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Dear Colleagues and Friends,

Welcome to the 11th Nordic Meeting on Cerebrovascular Diseases, which has been arranged to coincide with the 2nd Kuopio Symposium on Ischaemic Stroke this year. We decided to combine these two events to take advantage of the synergy afforded by such a unique opportunity. We hope that the meeting will be a forum for new and fruitful ideas between clinicians and laboratory scientists, advancing our understanding of cerebrovascular diseases.

The program will cover a wide range of topics, from molecular mechanisms of ischemic stroke to the rehabilitation of stroke patients. One new facet of this joint meeting will be a focus on the importance of good nursing care. Thus, a parallel session has been incorporated to encourage neurology nurses to attend the meeting. We hope that this will be the start of a tradition in the Nordic Meetings.

We also hope you will have an opportunity to relax and stay a few extra days in our beautiful Finnish “summer city” on the shores of Lake Kallavesi.

We warmly welcome you to Kuopio.

Juhani Sivenius
Chairman of the Organizing Committee

Program and Abstracts

Saturday, August 11

Registration at the Kuopio Music Centre
12.00-

Opening Ceremonies
16.00-16.15 Juhani Sivenius, The Organizing Committee
Matti Uusitupa, Rector, University of Kuopio
Kjell Asplund, The Scandinavian Society for Cerebrovascular Diseases

Presidential Lectures (Chairman Juhani Sivenius)
(sponsored by Kuopio University Neuroscience Center)

16.15-16.45 **Ionic Perturbations Underlying Ischemic Neuronal Death**
D.W. Choi (USA)
16.45-17.15 **Prospects of Clinical Stroke Imaging**
S. Warach (USA)
17.15-17.45 **Burden of Stroke in the Nordic Countries**
K. Asplund (Sweden)
17.45-18.15 **Management of Ischemic Stroke**
M. Kaste (Finland)

Get-together Party at the Kuopio Music Centre
19.30-21.30

Sunday, August 12

Pathophysiology of Ischaemic Stroke (Chairman Jari Koistinaho)

9.00-9.30 **Caspases in Brain Ischemia**
R.P. Simon (USA)
9.30-10.00 **Unraveling Neuronal Death Signal Cascades**
V.L. Dawson (USA)
10.00-10.30 Coffee break
10.30-11.00 **Importance of Mitochondrial Pores for the Development of Ischemic Neuronal Damage**
T. Wieloch (Sweden)
11.00-11.30 **Anti-inflammatory Treatment in Stroke Animal Models**
J. Yrjänheikki (Finland)
11.30-12.30 **Short Oral Communications I**
Dividing Neurons in Adult Rat Brains Evince Mitotic Spindles after Photothrombotic Ring Stroke
W.-G. Gu, T. Brännström, R. Rosqvist and P. Wester
NPC-037282, a Novel p38 MAPK Inhibitor, and Aspirin Abolish the Increased Vulnerability to Focal Brain Ischemia in b**APP Transgenic Mice**
M. Koistinaho, M.I. Kettunen, R.A. Kauppinen, D. Liu, L.S. Higgins and J. Koistinaho
Apoptosis-related Pathophysiological Mechanisms in a Photothrombotic Ring Stroke Model in Adult Rats
X.-L. Hu, I.M. Johansson, T. Brännström, T. Olsson and P. Wester
Anti-apoptosis Strategies and Brain Plasticity in Permanent MCA Occlusion Model in Rat
L. Simon, P. Koska, Z. Bori and Z. Nagy

12.30-13.30 **Lunch**

Sunday, August 12

Neuroimaging of Stroke (Chairman David Russell)

13.30-14.00 **Multimodal Imaging of the Ischaemic Penumbra**
W.-D. Heiss (Germany)

14.00-14.30 **Imaging of Hemodynamics and Metabolism in Cerebrovascular Diseases**
W.P. Powers (USA)

14.30-15.00 Coffee break

15.00-15.30 **Metabolic Approach to Human Stroke**
P.B. Barker (USA)

15.30-16.00 **Magnetic Resonance Imaging of Reversibility of Cerebral Ischaemia**
R.A. Kauppinen (Finland)

16.00-17.00 **Short Oral Communications II**

Diffusion and Perfusion Magnetic Resonance Imaging in Patients with Acute Ischemic Stroke
J. Karonen, Y. Liu, R. Vanninen, K. Partanen, J. Nuutinen, J. Sivenius and H.J. Aronen

Standard Stereotaxic Coordinate System for the Pig Brain: Automatic Measurement of Regional CBF and $CMRO_2$ after Transient Middle Cerebral Artery Occlusion (MCAO)
H. Watanabe, F. Andersen, A. Gjedde, The DaNeX Study Group, M. Sakoh and P. Cumming

Magnetic Resonance Imaging of Magnetodendrimer-labeled Neural-Progenitor Cell Transplants
J.M. Hakumäki, J.M. Savitt, L. Ifeanyi, P.C.M. van Zijl, J.D. Gearhart, T. Douglas, T. Dawson, J.A. Frank, V.L. Dawson and J.W.M. Bulte

Non-invasive Monitoring of Intracranial Pressure and Compliance
K. Paulat, R. Brucher and D. Russell

How Does the Medical Care of Stroke Patients Work Today – Tomorrow? Staff Meeting of Health Care Personnel (Chairman Juha Kinnunen) **(Auditorium)**

13.30-14.00 **Seamless Service Chain of Stroke Patients – How It Works in Finland**
J. Kinnunen (Finland)

14.00-14.20 **Patient Care Needs and Nurse Work Load: Study of the Number of Working Hours in Bed Care Units of Specialized Patient Care**
P. Partanen (Finland)

14.20-14.40 **Standard Nursing Questionnaire in Acute Stroke Patient Care**
S. Zielke (Denmark)

14.40-15.00 **Evidence Based Nursing Program for Acute Stroke**
B. Mørch (Norway)

15.00-15.20 Coffee

15.20-15.50 **Update on Stroke Rehabilitation**
P.W. Duncan (USA)

15.50-16.10 **How the Umeå Stroke Center Staff Works to Insure Good and Efficient Stroke Nursing Practice**
L.-A. Andersson and S. Wallin (Sweden)

16.10-16.30 **Study Nurse's Viewpoints**
M. Kuparinen (Finland)

16.30-17.00 **The Future of Health Care**
M. Myllykangas (Finland)

Cruise at Lake Kallavesi (from the Passenger Harbour) (sponsored by Boehringer Ingelheim)
19.30-

Monday, August 13

Prevention and Therapy of Ischaemic Stroke (Chairman Markku Kaste)
(sponsored by Boehringer Ingelheim)

9.00-9.30 **Non-pharmacological Prevention of Stroke – Does It Work?**
K. Asplund (Sweden)

9.30-10.00 **Benefits and Risks of Antiplatelet Therapy in the Secondary Prevention of Stroke**
H.-C. Diener (Germany)

10.00-10.30 Coffee break

10.30-11.00 **New Evidence about Temperature, Blood Pressure and Blood Sugar in the Early Hours after Stroke Onset**
G. Boysen (Denmark)

11.00-11.30 **Drug Treatment of Acute Ischemic Stroke**
N.G. Wahlgren (Sweden)

11.30-12.30 **Short Oral Communications III**

A Decline in 28-day Case Fatality But Still No Change in Stroke Incidence. A Population–based Study in Northern Sweden
B. Stegmayr, Å.B. Johansson and K. Asplund

The Independent Predictive Impact of Admission Body Temperature on Long-term Mortality after Stroke: The Copenhagen Stroke Study
L.P. Kammersgaard, U.J. Weber, H.S. Jorgensen, J. Reith, H. Nakayama, P.M. Pedersen and T.S. Olsen

Differences in Long-term Outcome between Patients Treated in Stroke Units and in General Wards
E.-L. Glader, B. Stegmayr, L. Johansson, K. Hulter-Åsberg and P.O. Wester

Trends in the Incidence and Mortality of Stroke Events in Finland during 1993-1997: The FINSTROKE Study
D. Jakovljevic, J. Sivenius, V. Salomaa, C. Sarti and J. Torppa

The General Assembly of the Scandinavian Society on Cerebrovascular Diseases
12.30-

12.30-13.30 **Lunch**

Post-stroke Complications (Chairman Matti Hillbom)

13.30-14.00 **Perilesional Dysfunction following Stroke**
O.W. Witte (Germany)

14.00-14.30 **Stroke and Depression**
A. Carota (Switzerland)

14.30-15.00 **Stroke and Dementia**
T. Pohjasvaara (Finland)

Posters (Chamber Music Hall) and Exhibition
15.00-17.00

Banquet Dinner at the Scandic Hotel
19.30-

Tuesday, August 14

Stroke Rehabilitation (Chairman Juhani Sivenius)
(sponsored by Brain Research and Rehabilitation Foundation)

9.00-9.45 **Measuring Stroke Outcomes**
P.W. Duncan (USA)

9.45-10.30 **Potential Effects of Drugs on Stroke Rehabilitation**
L. Goldstein (USA)

10.30-11.00 Coffee break

11.00-11.45 **The Role of New Treatment Methods in Acute Stroke**
D.C. Good (USA)

11.45-13.00 **Short Oral Communications IV**

What Determines Early Admission of Patients with Acute Stroke?
T.S. Olsen, B.H. Rasmussen, U. Germer and L.P. Kammersgaard

Recovery of Chronic Visual Field Defect after Computer-Assisted Training: A Case Study
L. Julkunen, H. Hämäläinen and O. Tenovuo

Constraint-induced Movement Therapy for the Affected Upper Extremity in Chronic Stroke Subjects
I.M. Tarkka, J. Sivenius and K. Pitkänen

Thrombolytic Therapy of Ischemic Stroke: A Swedish One-center Experience
H.G. Hårdemark, Ö. Nordmark and A. Terént

Cognitive and Motor Performance Related to the Size of the Infarct in a Rat Embolic Stroke Model
R.S. Rasmussen, E.S. Karnick, K. Overgaard, J. Foss and G. Boysen

13.00-13.45 **Lunch**

Experimental Restoration of Function (Chairman Tim Schallert)

13.45-14.15 **Anatomical Targets of Recovery Process**
T.A. Jones (USA)

14.15-14.45 **Functional Reorganization of the Motor Cortex Following Stroke: The Effects of Differential Motor Training**
J.A. Kleim (Canada)

14.45-15.15 **Current Status of Preclinical Restorative Drug Development**
S.P. Finklestein (USA)

Closing of the Meeting
15.15 Jari Koistinaho

Abstracts of Oral Presentations

IONIC PERTURBATIONS UNDERLYING ISCHEMIC NEURONAL DEATH

D.W. Choi
Center for the Study of Nervous System Injury and Department of Neurology, Washington University School of Medicine, St. Louis, MO, USA

The contribution of cellular calcium and sodium overload to ischemic neuronal loss is well recognized. These events, in large part a consequence of glutamate receptor overactivation, tend to induce cellular necrosis. More recently emerging evidence that ischemic neuronal death may substantially reflect apoptosis brings into consideration possible pathogenetic contributions from three other cationic perturbations: calcium starvation, potassium efflux, and zinc overload. Calcium starvation, reflecting inadequate calcium influx or release from intracellular stores, can trigger or promote neuronal apoptosis, and correcting intracellular free calcium levels back towards normal may represent a strategy for limiting ischemic neuronal apoptosis. Presumably both calcium starvation and calcium overload may occur after a single insult, but in different cells or at different times. Optimal therapeutic manipulation of neuronal calcium levels may thus require delineation of intracellular calcium levels, and identification of the relative risks of necrosis versus apoptosis. Potassium efflux, mediated by several routes including voltage- (mainly the delayed rectifier I_K) and agonist-activated (mainly glutamate receptor-activated) channels, may be another cationic disturbance favoring apoptosis. Blockade of neuronal potassium channels has a risk of potentiating excitotoxicity, but may be a promising strategy for limiting ischemic apoptosis. Lastly, after transient global ischemia or mild transient focal ischemia, excessive zinc influx, originating in nerve terminals and entering vulnerable target neurons through several routes, may cause neurons to undergo either apoptosis (at lower levels of zinc influx) or explosive necrosis (at higher levels of zinc influx). Several strategies for limiting toxic zinc influx may be considered, including extracellular chelation, or the delayed administration of membrane-permeant chelators.

PROSPECTS OF CLINICAL STROKE IMAGING

S. Warach
National Institutes of Health, National Institute of Neurological Disorders & Stroke, Stroke Branch, Section on Stroke Diagnostics & Therapeutics, Bethesda, MD, USA

The clinical questions that direct the stroke physician's diagnostic thought process have changed little over the last century. However, it was with the development of cerebral angiography in the first half of the 20th century and computed tomography (CT) in the second half that a shift began from complete reliance on bedside evaluation and inference to direct visualization of ischemic pathophysiology in the living patient. Rapid technological advances in the last several decades have fostered a more central role of imaging in the care of stroke patients and in clinical research in cerebrovascular disease.

Among the clinical neurodiagnostic methods, magnetic resonance imaging (MRI) is emerging as a leading technology in the diagnosis and care of the acute stroke patient because of the range of structural and physiological measurements possible in a relatively non-invasive and rapid manner and because of the wide dissemination and availability of MRI scanners in clinics and hospitals. The investigation of quantitative tissue hemodynamics and metabolism with PET and MR will be discussed is a special session in this meeting.

The ideal neurodiagnostic imaging exam for stroke must rule out intracranial hemorrhage, detect the presence of ischemic pathology (intracranial and extracranial arterial disease, ischemic parenchymal injury and brain hemodynamics), and be achievable in a brief scanning session. An emergency MRI exam including diffusion weighted (DWI), perfusion (PWI), MR angiography (MRA), susceptibility weighted and T2-weighted imaging meets all these requirements. At many institutions where emergency therapies and early diagnosis are part of stroke care, MRI has already taken on a role to accomplish the diagnostic objectives in acute stroke. Conclusions about the state of ischemic pathophysiology in an individual patient, once based solely on clinical conjecture in the first few hours after onset, may now based on objective data as well. Clinical studies support the sensitivity and specificity of the MRI in early stroke diagnosis, predicting clinical outcome, predicting the fate of the tissue at risk at the earliest time points, and in demonstrating reversal of ischemic injury following successful early reperfusion. A reduction in ischemic lesion volume is highly predictive of clinical recovery.

Imaging in clinical drug development and testing will become ever more important in Phase I and II clinical trials to evaluate brain pharmacokinetics, tissue viability, establish proof of pharmacological principle, and provide surrogate measures of potential clinical effects. We are also seeing the original neuroimaging method in acute stroke – cerebral angiography – taking on increased importance as experimental stroke therapeutics move more aggressively toward applying mechanical and pharmacological therapies intra-arterially.

The future of clinical imaging in stroke will be ever faster, more quantitative and multimodal. The ultimate goal of the rapid assessment of the stroke patient is to define the pathology and the at-risk but salvageable tissue - the optimal target for therapies.

BURDEN OF STROKE IN THE NORDIC COUNTRIES

K. Asplund
Department of Medicine, University Hospital, Umeå, Sweden

The burden of stroke has many dimensions. It may be assessed in relation to who is afflicted: the individual, the family, the health care system, social services and the society at large. In this presentation, the focus is more on the societal effects than on the clinical outcome in individuals.

In an international perspective, stroke mortality is relatively low in the Nordic countries. On the other hand, stroke incidence is intermediate or even high, as in Finland. The discrepancy in international ranking between incidence and mortality rates is explained by a low case fatality in stroke in the Nordic countries. The burden of stroke has a social patterning with a considerably higher risk among people with a low level of education also after adjustment for differences in classical cardiovascular risk factors.

Although stroke mortality rates have been declining somewhat faster in Finland than in its Nordic neighbours, it is still higher in Finland than in the other countries. The decline is explained both by a combination of lower incidence rates (particularly in Finland and Denmark) and lower case fatality (most pronounced in Sweden).

The declining case fatality has resulted in an increasing number of stroke survivors with increasing demands on long-term care and secondary prevention. This development is partly counteracted by an ongoing improvement in functional outcome in terms of primary ADL proficiency.

Of the long-term stroke complications, post-stroke dementia and depression account for the largest number of disability-adjusted life years (DALYs) lost. Data from Finland and Sweden have shown that as many as 15% of stroke patients may develop dementia and 40% develop depression.

Studies of unselected stroke populations in all Nordic countries except Iceland have uniformly shown that approximately a fifth of the survivors are still in institutional care at 3 months after stroke.

The annual societal cost of stroke in Nordic countries has been estimated at 100-150 million SEK per 100,000 inhabitants. Approximately 70% of the costs are for institutional care, of which considerably less than half is attributed to hospital costs during the acute phase. Advanced home care for stroke patients seems to as efficient but also as expensive as hospital care. Recent estimates in Denmark and Sweden have come up with the same total costs during the first year after stroke (approx. 143,000 DKK or 170,000 SEK). Direct comparisons between Denmark and the UK have shown case fatality to be much lower in Denmark at a marginal cost of approx. 250-300,000 DKK per life year gained.

Typically, cost estimates do not include the intangible costs for care by family members. In a Swedish nation-wide survey, more than half of the stroke patients stated that they were in need of support from family members as late as two years after stroke.

MANAGEMENT OF ISCHEMIC STROKE

M. Kaste
Department of Neurology, University of Helsinki, Helsinki, Finland

Introduction. Stroke is the second commonest cause of death, and the loss of quality-adjusted life years caused by stroke globally is bigger than for any other disease. The economic burden is enormous. However, the nihilistic attitude that nothing can be done for stroke patients does not find support in evidence based medicine. The key point is a well-organized care.

Prehospital care. Stroke is a medical emergency. A system of triage, analogous to care for acute myocardial infarction in which the patient contacts the emergency services first, is also needed in acute stroke care to prevent unnecessary delays. Emergency prehospital services should be partners in stroke care. It is essential that stroke patients are admitted quickly to hospitals that have the expertise and resources for acute stroke management.

Emergency room. Guidelines for optimal management of acute stroke should be made to reduce in-hospital delays. Codes as thrombolysis can reduce time to CT, which together with the patient's history and exam is the most important initial diagnostic test. Delays can be avoided if necessary personnel and equipment are available at all times. Diagnostic work-up and therapy should be initiated at the emergency room and finalized at acute stroke unit to which all stroke patients should be transferred from the emergency room. Written protocols supported by checklists are necessary to ensure that all patients receive good clinical care at all times. The use of protocols reduces complications, improves outcome and reduces costs.

Stroke units. Stroke unit care in managing acute stroke patients is highly effective evidence- based medicine. A Cochrane systematic review based on the Stroke Units Trialists´ Collaboration showed a 24% reduction in one year mortality, a 25% reduction in death or dependency, and a 24% reduction in death or institutional care. No systematic increase in length of stay was observed. A few factors are typical for successful stroke units. They include geographically defined ward or part of a ward, which exclusively or nearly exclusively takes care of stroke patients, patients are treated by a multidisciplinary stroke team, the core disciplines of which are medical, nursing, physiotherapy, occupational therapy, speech therapy and social work. Systemic prevention of complications, early rehabilitation, continuous education of the staff, and involvement of the careers of the patient are essential features of the stroke unit care. Observed benefits are not limited to any particular subgroups of patients. Males and females, old and young, severe, moderate or mild strokes benefit from it and the results seem to stay. There is no evidence to exclude old or severe stroke patients from stroke unit care.

Thrombolysis. Intravenous rt-PA within 3 hours after the onset of symptoms in patients with acute ischemic stroke is a highly effective evidence-based treatment. The use of rt-PA is supported by results from randomized controlled trials and meta-analyses. The risk of early fatal and symptomatic intracranial hemorrhage is increased, but these hazards are offset by reduction in the proportion of patients being dead or dependent. According to meta-analyses, for patients treated within 3 hours of ischemic stroke approximately 1 of 10 will be independent, 1 of 14 suffer symptomatic hemorrhage and 1 of 100 fewer may die as a result of the treatment. Overall, the net benefit of rt-Pa given within 3 hours of the onset will result in one more independent survivor for every 10 patients treated. Intravenous rt-PA within 6 hours after the onset of an ischemic stroke seems to be beneficial, but the benefit is smaller and the risks higher. The use of rt-PA up to 6 hours is supported by the results of meta-analyses.

Conclusions. Stroke therapy should rely on evidence-based medicine. According to Helsingborg Declaration, all stroke patients should be treated in dedicated stroke units.

CASPASES IN BRAIN ISCHEMIA

R.P. Simon
Dept Molec & Cell Neurosci, Dow Neurobiology Lab Legacy Res, Portland, OR, USA

While the contribution of apoptosis to ischemic brain injury remains incompletely understood, there is emerging evidence for the involvement of the caspase family of cell death proteases from models of both focal and global ischemia.

We will present data that demonstrates the involvement of the key executioner caspase, caspase-3, in neuronal death following experimental global cerebral ischemia. Our findings demonstrate increased mRNA for caspase-3 and proteolytic activation of caspase-3 from 4-72 h following global ischemia within the vulnerable hippocampus and caudate putamen while neuroprotection could be conferred by the caspase-3 pseudosubstrate z-DEVD-fmk. Subsequently we sought to address the route by which caspase-3 is activated examining the extrinsic, death receptor-linked pathways of caspase-8 and caspase-10. We determined that caspase-10, rather than caspase-8, is activated following global ischemia via the Fas/FADD death receptor-signaling complex.

These data establish the caspase family of cell death proteases as of critical importance in the mechanism by which ischemia induces neuronal death. Further, evidence for the involvement of caspase-10 and the death receptor signaling complex as a possible initiation mechanism in this pathway lends significant insight into the little explored role of death receptor signaling in neurological disease and offers novel targets for therapeutic intervention to mitigate the effects of ischemia on the brain.

UNRAVELING NEURONAL DEATH SIGNAL CASCADES

V.L. Dawson
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Neuronal damage following stroke is thought to stem in part from overexcitation of N-methyl-D-aspartate (NMDA) receptors by glutamate. Neurotoxicity elicited by excess stimulation of NMDA receptors is mediated in large part by activation of neuronal nitric oxide synthase (nNOS) and the production of nitric oxide (NO). Simultaneous production of superoxide anion in mitochondria provides a permissive environment for the formation of peroxynitrite. Peroxynitrite can damage DNA leading to strand breaks and activation of PARP. We have found that PARP activation plays a key role in NMDA excitotoxicity and focal cerebral ischemia in experimental animal models.

Cortical cultures from PARP knockout mice are resistant to neurotoxicity following exposure to NMDA, NO donors or combined oxygen-glucose deprivation (OGD) an *in vitro* model of ischemia. PARP knockout cortical cultures show only modest protection against AMPA or Kainate neurotoxicity. *In vivo,* PARP knockout mice are resistant to intrastriatal injection of NMDA or transient focal ischemia. Replacement of PARP in the knockout mice with viral expression vectors restores sensitivity to excitotoxicity both *in vitro* and *in vivo* and to cerebral infarction following transient focal ischemia.

We have observed that diverse forms of neuronal injury lead to cell death by PARP activation. How activation of PARP results in neuronal cell death is not yet known but could involve decrements in energy, ribosylation of specific signaling proteins and cross-talk between nuclear and mitochondrial proteins. Emerging data will be discussed. The degree of protection observed in PARP knockouts suggests that development of appropriate PARP inhibitors could provide potent therapeutic approaches to the treatment of stroke patients.

ANTI-INFLAMMATORY TREATMENT IN STROKE ANIMAL MODELS

J. Yrjänheikki
A.I.Virtanen Institute, University of Kuopio, Kuopio, Finland and Cerebricon Ltd

The brain is traditionally considered as an immunoprivileged tissue since it is devoid of a lymphoid system and antigen presenting dendritic cells. However, increasing evidence suggests that cerebral ischemia and other neurological disorders elicit an unrestrained inflammation, which maturates in a delayed manner and significantly contributes to the evolution of tissue injury, thus offering a rationale for using anti-inflammatory therapy. Ischemic inflammation is a complex phenomenon characterized by production and interplay of cytokines, chemokines, adhesion molecules, free radicals, and destructive enzymes such as COX-2, inducible NOS and proteinases. In addition to circulating neutrophils and monocytes/macrophages, also resident microglia, astrocytes, endothelial cells and neurons are involved in *in situ* inflammatory reactions. While the inflammatory factors contributing to ischemic injury are well documented, the neuroprotective role of certain inflammatory processes is rather poorly understood. Growth promotion, repair and enhanced functional recovery require active inflammatory machinery, and therefore any intervention against inflammatory reactions needs to be designed with caution.

It is presently well established that leukocytes contribute to the degree of ischemic brain injury. Unlike normal brain microvessels that are devoid of adherent inflammatory cells, brain microvessels from ischemic zones are filled with adherent leukocytes and a distinctive zone of edema surrounds them. Neutrophils are initially the predominant leukocytes at the site of inflammation, followed by infiltration of mononuclear phagocytes. An anti-leukocyte strategy has been shown to provide neuroprotection. It is very effective in experimental models of transient focal cerebral ischemia, but not in permanent focal cerebral ischemia. Whereas systemic depletion of neutrophils and other leukocytes is unlikely to be of clinical importance due to the severe immunosuppression, targeted anti-adhesion strategies might be valuable tools, if one is able to diminish unwanted immune reactions.

Microglia, the resident tissue macrophages of the CNS, are believed to play an active role in brain inflammatory, immune and degenerative processes. Depending on the magnitude of the microglial reaction, on the type of stimulus, and on the concurrence of other local factors, microglia can contribute to host defense and repair, or to the development and progression of cerebral injury. The most characteristic property of microglia is the ability to modify their behavior in response to changes in microenvironment, and to synthesize and secrete a large number of substances. The functional role of activated microglia in evolution of ischemic damage is diversified. When the critical point of activation is crossed, microglia launches the cytotoxic machinery that mainly includes, besides the phagocytic property, inflammatory cytokines, destructive enzymes with ROS and NO production, proteolytic enzymes, and neurotoxins. Reduction of microglial activation in ischemia results in diminished neuronal death and improved functional recovery.

DIVIDING NEURONS IN ADULT RAT BRAINS EVINCE MITOTIC SPINDLES AFTER PHOTOTHROMBOTIC RING STROKE

W.-G. Gu[1], T. Brännström[2], R. Rosqvist[3] and P. Wester[1]
[1]Public Health & Clinical Medicine, [2]Medical Biosciences & Pathology, [3]Cell & Molecular Biology, Umeå University, Umeå, Sweden

Background. Division of mature neurons was initially observed in the adult rat brains [2] that may be associated with the post-stroke cortical neurogenesis [1]. This study aimed to explore the possible appearance of mitotic spindles that should presumptively appear in any kind of cells undergoing cell division.

Methods. 12-weeks old male Wistar rats were subjected to stroke [3] and BrdU delivery. Rats were sacrificed at 24h, 48 h, 72 h and 7 days after stroke. Brain sections were processed for immunohistochemistry or immunofluorescence. Primary antibodies used: the neuron-specific markers Map-2, β-tubulin III, Neu N; the glial marker GFAP; the mitosis-specific markers phospho histone H3 (phos H3), γ-tubulin; DNA synthesis marker BrdU. Various fluorescent secondary antibodies were used to detect the primary antibodies. Nuclear DNA was counterstained with DAPI.

Results. Under confocal microscopy, the mitosis specific markers phos H3 or γ-tubulin were colocalized with BrdU in the same cortical cells in the cortical region-at-risk (i.e. the ischemic penumbra). Phos H3 stained the cell nuclei whereas γ-tubulin marked specifically the spindle assembly of the dividing cells. At 24h, 48h, 72h and 7d after stroke, single or doublet phos H3 immunopositive nuclei were detected in cortical neurons as identified by the mature neuron specific marker Neu N. Similarly, γ-tubulin-positive mitotic spindles, which were mono-polar or bipolar assembled, were detected in the Neu N-positive neurons as examined through three-dimension confocal microscopy at the same time intervals after stroke. Furthermore, dividing cell nuclei, counterstained through DAPI, were observed in these cortical neurons. These nuclei were bipolar stretched between the γ-tubulin-positive spindles and varied in shapes. At 72h after stroke and later, γ-tubulin-positive mitotic spindles accumulated also frequently in the cortical astrocytes as identified with the mature glial specific marker GFAP. Dividing nuclei were frequently observed in these cells.

Conclusion. This study suggests that mature neurons and astrocytes do have the ability to divide and thus generate daughter neurons and glial cells in the adult rat brains after stroke.

Acknowledgements: the authors thank Professors Kjell Asplund and Brant Watson for fruitful discussion and critical comments.

1. Gu, W.G., Brannstrom, T. and Wester, P., Cortical neurogenesis in adult rats after reversible photothrombotic stroke, *J Cereb Blood Flow Metab*, 20 (2000) 1166-73.
2. Gu, W.G., Brannstrom, T. and Wester, P., Division of mature neurons in vivo contributes to cortical neurogenesis in adult rats after reversible photothrombotic ring stroke., *Soc Neurosci Abs, Vol. 26*, 2000, pp. 757.
3. Gu, W.G., Jiang, W. and Wester, P., A photothrombotic ring stroke model in rats with sustained hypoperfusion followed by late spontaneous reperfusion in the region at risk, *Exp Brain Res*, 125 (1999) 163-70.

NPC-037282, A NOVEL p38 MAPK INHIBITOR, AND ASPIRIN ABOLISH THE INCREASED VULNERABILITY TO FOCAL BRAIN ISCHEMIA IN b APP TRANSGENIC MICE

M. Koistinaho[1], M.I. Kettunen[1], R.A. Kauppinen[1], D. Liu[2], L.S. Higgins[2] and J. Koistinaho[1]
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Background. Stroke and Alzheimer's disease share common risk factors and the incidence of dementia is higher than expected in stroke patients. We have previously shown that the mice overexpressing the 751-aa isoform of wild type human βAPP have increased sensitivity to ischemic brain injury. Here we studied whether the increased sensitivity to ischemic injury in βAPP transgenic (tg) mice can be prevented by aspirin (ASA) or by NPC-037282, a selective and potent inhibitor of p38 MAPK.

Methods. The left MCA was permanently occluded in βAPP tg and wild type (wt) littermates. ASA (20 mg/kg), NPC-037282 (10 mg/kg) or their vehicles were administered 4 times to 8-mo-old mice at 4 h intervals beginning 4 or 2 h prior to the MCA occlusion. Infarct volumes were quantified from T2-weighted images obtained by MRI using a 4.7 T horizontal magnet 24 h after the MCA occlusion. Neocortical CBF was measured by laser-Doppler flowmetry. Physiological variables measured before and after the MCA occlusion were not altered by the treatments. Inflammatory responses were evaluated histochemically using F4/80 (a microglial marker) and COX-1 antibodies.

Results. Vehicle treated tg mice had 35-40% larger infarcts than the wt mice. Treatment with ASA or NPC-037282 reduced the infarct size in tg mice by 30% and completely abolished the difference seen between the vehicle treated tg and wt mice. NPC-037282, but not ASA, significantly reduced the infarct size also in wt mice (by 19%). Both ASA and NPC-037282 reduced the staining of ischemia-induced histochemical markers of inflammation. Neocortical CBF was not affected by the treatments.

Conclusions. Inflammation and/or apoptosis mediated through p38 MAPK significantly contributes to the increased ischemic vulnerability in APP751 overexpressing mice.

APOPTOSIS-RELATED PATHOPHYSIOLOGICAL MECHANISMS IN PHOTO-THROMBOTIC RING STROKE MODEL IN ADULT RATS

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Background. Apoptosis, a genetically regulated and energy dependent cell death pathway, has recently been shown to act in concert with necrosis in ischemic cell death in the central nervous system. This study aimed at exploring the temporal and spatial patterns of apoptosis and its relationship with the expression of the anti-apoptotic genes bcl-2, bcl-w and the pro-apoptotic gene bax in a dual set up of a photothrombotic ring stroke model with or without late spontaneous reperfusion.

Methods. The photothrombotic ring stroke model with or without spontaneous reperfusion was induced on male Wistar rats by low (0.9W/cm^2) and high (1.84W/cm^2) laser intensity with otherwise identical experimental conditions. Apoptotic cell morphology was demonstrated by electron microscopy, TUNEL staining, Hoechst 33342, and H&E staining. DNA internucleosomal fragmentation was explored by gel electrophoresis. The protein levels of the Bcl-2 gene family were explored by immunocytochemical staining and western blots and their corresponding mRNA expression were verified by *in situ* hybridization.

Results. Cells within the ischemic cortex exhibited apoptotic morphology as revealed by electron and light microscopy with different means and isolated DNA revealed a ladder. The frequency of apoptotic cells is demonstrated in the figure. Bcl-2 immunoreactivity (IR) was substantially upregulated at 48 –72 hours and Bcl-w IR moderately increased at 10-48 h in the region at risk after low intensity irradiation.

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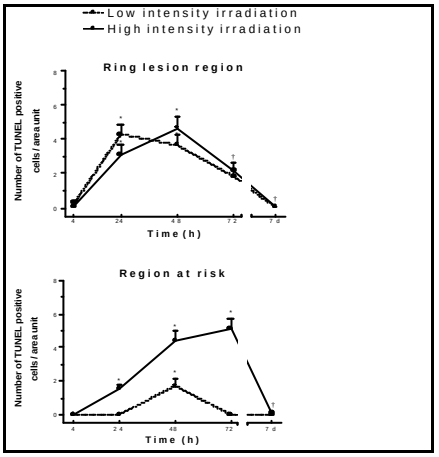

Conclusion. Apoptosis may be an important contributor to neuronal cell death in brain regions with severely reduced blood flow after thrombo-embolic stroke. The upregulation of Bcl-2 and bcl-w in the region-at-risk may play an important role in preventing ischemic cell death in the ischemically challenged penumbra following a photothrombotic cortical stroke lesion.

ANTI-APOPTOSIS STRATEGIES AND BRAIN PLASTICITY IN PERMANENT MCA OCCLUSION MODEL IN RAT

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In focal brain ischemia there are two distinct forms of cell death mechanisms, apoptosis and necrosis. Overexpressing protooncogenes bcl-2 and Bcl-XL could influence cell death. In this study the neuroprotection due to the overexpression of Bcl-2 and Bcl-XL genes were examined in the ischemic penumbra in a permanent middle cerebral artery occlusion model.

Antiapoptotic gene expression has been augmented by an extensively studied drug (-) deprenyl, or with direct gene delivery by a non-replicable adenovirus construct. Infarct volume has been measured on TTC stained brain slices or on MRI images. Number of apoptotic cells were analysed statistically. Plasticity proteins (GAP 43, MAP 2 and Cyclin D1 were visualised by immunohistochemical methods.

Following (-)deprenyl treatment the infarct volume decreased significantly. The number of TUNEL labelled cells averaged in 60 samples was 17$\pm$12 in treated rats, while 28$\pm$25 in control rats (p=0,002). The number of TUNEL-caspase-3 labelled cells averaged in 30 samples was 3$\pm$3 in treated rats, while 8$\pm$6 in control rats (p=0,0003). (-)Deprenyl treatment increased the number of GAP-43-positive cells.

The route of gene delivery influences the effectiveness of the gene therapy in the brain. We examined two ways of administration of recombinant adenovirus vector containing LacZ reporter gene, direct intraparenchimal and intracisternal injection. The LacZ reporter gene expression after the direct intracortical administration has been visualised around the injection site, furthermore the viral spread appeared in the ipsilateral cortex and hippocampus. Microvessel endothelial cells, oligodenrdoglia cells and neurons were transfected. Following the intracisternal virus administration, transduction was significant in the endothelial cells; ependyma cells, in the plexus epithelial cells, furthermore in the CA1 and CA3 hippocampal sectors. Cortical neurons are also transfected focally.

Based on the data of spread of adenovirus vectors, IL-1ra, Bcl-2 Bcl-XL genes were administered into the brain before MCA occlusion. There were no significant decreases in lesion size in either case Decrease in the number of apoptotic neurons were documented after gene delivery. GAP-43, the main indicator protein of neuronal plasticity and regeneration, is upregulated in Bcl-2 and Bcl-XL overexpressing animals. Bcl-2 and Bcl-XL affect apoptosis and necrosis and promote brain plasticity. (Supported by EU grants BMH4-CT-98-3277 and IC-CT-98-0206)

MULTIMODAL IMAGING OF THE ISCHAEMIC PENUMBRA

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Active treatment of acute ischemic stroke can only be successful as long as tissue in the area of ischemic compromise is still viable. Therefore, the identification of the area of irreversible damage, and its distinction from the penumbral zone, i.e., tissue with impaired function but preserved morphology, may improve the estimation of the potential efficacy of various therapeutic strategies. This can be achieved by multi-tracer positron emission tomography (PET) and perfusion-weighted (PW) and diffusion-weighted (DW) magnetic resonance imaging in experimental models. Neuroimaging modalities applied in patients with acute ischemic stroke cannot reliably identify penumbra tissue and detect irreversible damage in the first hours after stroke, when treatment must be initiated to have the potential for success: Multitracer studies for the assessment of flow and irreversible metabolic damage usually are limited in the clinical setting, and arterial blood sampling necessary for quantitative determinations is prohibited under certain circumstances, e.g., when thrombolysis is planned. CT and MRI do not reliably detect irreversible damage in the first hours after stroke, and even DW-MRI may be misleading in some cases; determination of perfusion by PW-MRI yields a poor estimate of the state of tissue. The range of the penumbra can be assessed by combining determinations of flow and benzodiazepine receptor binding by PET of $H_2{}^{15}O$ and ^{11}C-flumazenil (FMZ) and relating flow values and FMZ binding to the final state of the tissue. By this approach, cumulative probability curves can be computed to predict eventual infarction or non-infarction and to define the penumbral range. The computed values are in good agreement with results from other studies proving the validity of the concept of the penumbra which was also demonstrated in several therapeutic studies in which thrombolytic treatment reversed critical ischemia and decreased the volume of the final infarcts. Such neuroimaging findings might serve as surrogate targets in the selection of other therapeutic strategies for large clinical trials.

IMAGING OF HEMODYNAMICS AND METABOLISM IN CEREBRO-VASCULAR DISEASES

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The relative importance of hemodynamic as opposed to thromboembolic mechanisms in the pathogenesis of ischemic stroke remains unsettled. In order to determine what role hemodynamic factors play in the prognosis and treatment of patients with cerebrovascular disease, methods for determining the hemodynamic status of the cerebral circulation, accurately and in awake subjects under normal conditions, must be available. Direct measurements have shown that the degree of arterial stenosis is a poor indicator of the hemodynamic status of the cerebral circulation. Reductions in cerebral perfusion pressure (CPP) distal to a carotid stenosis do not occur until the stenosis is greater than 50%. However, even with stenosis greater than this, the reduction in perfusion pressure is variable among patients and may even be normal with stenosis greater than 90%. Since direct measurements of the intravascular perfusion pressure are impractical, current assessment of local cerebral hemodynamics depends on indirect evidence of compensatory responses to reductions in CPP. These compensatory responses have been determined by direct observation during global reductions in CPP due to systemic hypotension and increased intracranial pressure and are assumed to occur with local reductions in CPP to focal arterial stenosis. When CPP is normal, cerebral blood flow (CBF) is closely matched to the resting metabolic rate of the tissue. Moderate reductions in CPP have little effect on CBF. Vasodilation of arterioles reduces cerebrovascular resistance, thus maintaining a constant CBF. This is known as autoregulation of CBF. During systemic hypotension, blood vessels may show poor response to normal vasodilatory stimuli manifested as a reduced CBF increase to such dilating agents as hypercapnia. Furthermore, with reductions in CPP it is sometimes possible to measure an increase in cerebral blood volume (CBV). This increase is assumed to be a reflection of autoregulatory vasodilation. However, the increase in CBV is not always evident and decreased CBV may be observed with severe reductions in CPP. The mean vascular transit time (MTT=CBV/CBF) is also increased under these circumstances, even more so than CBV. With more severe reductions in CPP, the capacity for compensatory vasodilation is exceeded and autoregulation fails. CBF begins to decline. An increase in the amount of oxygen extracted from the blood by the brain now maintains cerebral oxygen metabolism and brain function.

Practical application of these physiological principles to the study of regional cerebral hemodynamics in patients with cerebrovascular disease has revealed nconsistencies in the actual experimental data. The correspondence between vasodilatory response and CBV is variable. A normal vasodilatory response may occur in the setting of increased CBV. CBF responses to different vasodilatory agents may be normal or impaired in the same patient indicating that these agents do not all measure pre-existing autoregulatory vasodilation. Vasodilatory responses and CBV may be normal or abnormal in patients with increased OEF, contrary to what would be expected. The MTT is a relatively poor discriminator of hemodynamic compromise being abnormal in 2/3 thirds of patients and having poor predictive value for subsequent stroke. Furthermore, increases in the MTT are not specific for reductions in CPP. They also occur with hypocapnia induced reductions in CBF.

We conclude form these discrepant findings that different methods of hemodynamic assessment are not interchangeable even when apparently based on the same pathophysiological response. As a result, it is hazardous to transfer conclusions based on one method to another. Each method must be independently demonstrated to have prognostic or therapeutic value using proper experimental design.

METABOLIC APPROACH TO HUMAN STROKE

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Magnetic resonance spectroscopy (MRS) and spectroscopic imaging (MRSI) allow the evaluation of the metabolic status of the brain in cerebral ischemia and infarction using conventional MRI equipment.

Proton spectra of the brain contain signals from a number of compounds of importance in cerebral ischemia, in particular lactate and N-Acetyl Aspartate (NAA). Lactate is the end-product of non-oxidative glucose metabolism and increases rapidly once blood flow falls below about 20 mL/100g/min. NAA is believed to be predominantly located in neurons and axons in mature brain, and decreases fairly slowly with time after the onset of cerebral ischemia. Based on these signals, proton MRSI therefore has the potential to image ischemic brain regions and assess stroke progression. In the chronic stages of stroke, changes in other metabolites detectable by proton spectroscopy (e.g. creatine, choline-containing compounds) may also be observed. The use of multi-slice spectroscopic imaging techniques (MRSI) is particularly important for stroke assessment, since stroke location may be uncertain at the time of scan, and lesion development is often heterogeneous between the core and periphery.

MRSI can be combined with other MRI techniques such as diffusion and perfusion MRI in a single session which provides information on brain structure, blood flow and metabolism. Potential applications of MRSI in human stroke include selecting patients for thrombolytic or other therapies, and monitoring of treatment efficacy. Multiple technical problems exist in the routine application of MRS or MRSI to acute stroke, however. These include lengthy scan times (unacceptable if an acute intervention such as tPA is being considered), poor spatial resolution and coverage, and head motion artifacts. Other problems involve the interpretation of NAA levels in terms of neuronal viability or loss. Development of fast scanning techniques will eventually allow MRSI to be added to existing stroke imaging protocols, and can also be used to increase brain coverage.

Conventional MRSI may be useful in sub-acute stroke, where short scan times are less critical. Examples of MR-based selection of patients for interventions such as carotid endarterectomy or hypertensive therapy will be presented.

MAGNETIC RESONANCE IMAGING OF REVERSIBILITY OF CEREBRAL ISCHAEMIA

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Both acute and subacute cerebral ischaemia can be explicitly revealed by means of *in vivo* nuclear magnetic resonance (NMR) techniques [1]. NMR spectroscopy (MRS) directly probes cerebral metabolism affected by ischaemia thereby serving as a diagnostic window as well as offering an access to the pathophysiology of the condition. Due to technical constraints, however, MRS has remained a tool for experimental stroke research. Owing to the inherent sensitivity of water diffusion to ischaemia [2], diffusion magnetic resonance imaging (DWI) has been adopted to the clinical imaging protocol of stroke. Combining DWI with T_2-weighted MRI (sensitive to overall water content) [3, 4] or with the bolus-tracking perfusion MRI [5, 6] the accuracy of acute stroke diagnosis has greatly improved. These MRI protocols have also been exploited to predict the long-term tissue outcome [3, 4, 6].

Assigning tissue reversibility in ischaemia *in vivo* remains a challenge for neuroimaging. Yet, tissue classification is becoming utterly important in clinical settings, since novel drug treatment strategies have been introduced [7, 8]. We have addressed the issue of reversibility by means of blood oxygenation level dependent (BOLD) MRI [9-11] and by using novel MR contrasts, such as T_1 in the rotating frame ($T_{1\rho}$) [12, 13] in the rat models of acute stroke. Expression of negative BOLD contrast requires mitochondrial oxidations and thus, only metabolising tissue contributes to the negative BOLD contrast. Interestingly, our data from acute focal cerebral ischaemia indicate that absolute $T_{1\rho}$ MRI may identify irreversible damaged tissue much earlier that either DWI or T_2-MRI [12]. The potentials of using these novel MRI contrasts for brain tissue assignment in acute ischaemic stroke is discussed.

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References

1. Baird, A.E. & S. Warach, J. Cereb. Blood Flow Metab. **18**: 583-609 (1998).
2. Moseley, M.E., *et al.*, Magn Reson Med. **14**: 330-346 (1990).
3. Welch, K.M., *et al.*, Stroke **26**: 1983-1989 (1995).
4. Carano, R.A., *et al.*, J Magn Reson Imag. **12**: 842-858 (2000).
5. Sorensen, A.G., *et al.*, Radiology **199**: 391-401 (1996).
6. Baird, A.E., *et al.*, Ann Neurol. **41**: 581-589 (1997).
7. Hacke, W., *et al.*, Lancet **352**:1245-1251 (1998).
8. Yrjänheikki, J., *et al.*, Proc Natl Acad Sci **22**: 15769-15774 (1998).
9. Gröhn, O.H.J., *et al.*, J Cereb Blood Flow Metab. **18**: 911-920 (1998).
10. Gröhn, O.H.J., *et al.*, J Cereb Blood Flow Metab. **20**: 316-326 (2000).
11.Kavec, M., *et al.*, MAGMA, 12: 32-39 (2001).
12. Gröhn, O.H.J., *et al.*, J Cereb Blood Flow Metab. **20**: 1457-1466 (2000).
13. Kettunen, M.I., *et al.*, Magn Reson Med. 2001. **in press**.

DIFFUSION AND PERFUSION MAGNETIC RESONANCE IMAGING IN PATIENTS WITH ACUTE ISCHEMIC STROKE

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Background. Ischemic stroke is a common cause of death and disability in industrialized countries. Diffusion and perfusion magnetic resonance (MR) imaging techniques are more sensitive than computed tomography in detecting ischemic brain tissue. The purpose of this study was to evaluate the natural course of brain ischemia with diffusion and perfusion MR imaging techniques.

Methods. Forty-nine patients with acute ischemic stroke were studied. Diffusion and perfusion MR imaging was performed within 24 hours of the onset of symptoms. Imaging was repeated on the next day and one week after stroke. A 1.5T whole-body scanner (Magnetom Vision, Siemens Medical Systems) and a head coil were used. Diffusion images of the whole supratentorial brain (slice thickness 5 mm, gap 1.5 mm) were obtained with an echo-planar spin echo sequence with a b-value of 1000 s/mm^2. High signal areas on calculated trace images were considered as infarcted and were measured volumetrically from the slices containing the lesion. Perfusion images were obtained dynamically during a 0.2 mmol/kg intravenous bolus of gadolinium chelate (Magnevist, Schering AG) by using a spin echo echo-planar sequence. Seven slices were covered from the central area of ischemia shown by diffusion images. Three types of perfusion maps – relative cerebral blood volume (rCBV), relative cerebral blood flow (rCBF) and mean transit time (MTT) – were calculated by methods described by Ostergaard et al. (MRM 1996;36:715-736). Regions of decreased perfusion were volumetrically measured from the perfusion maps and the sample of seven slices was compared to the diffusion lesion in the corresponding seven diffusion traces images to calculate three perfusion-diffusion mismatches (rCBV-diffusion, rCBF-diffusion and MTT-diffusion). Correlations of the initial perfusion-diffusion mismatches with the infarct growth during the week were calculated.

Results. Initial MR imaging was performed 11.1 ± 5.6 hours (mean ± SD) after the onset of symptoms. The volume of infarcted tissue depicted by diffusion MR imaging (whole brain) increased from 41 ± 65 cm^3 on day 1 to 65 ± 93 cm^3 on day 2 ($p<0.001$, Wilcoxon matched pairs signed rank test) and to 66 ± 75 cm^3 at one week ($p=0.002$). Measured from seven slices included in perfusion MR imaging, perfusion-diffusion mismatches were: rCBV-diffusion 17 ± 29 cm^3, rCBF-diffusion 48 ± 52 cm^3, and MTT-diffusion 74 ± 76 cm^3 (significant difference between diffusion and perfusion lesions $p<0.001$). The size of perfusion-diffusion mismatch correlated with the infarct growth during the week depicted by diffusion MR imaging (Spearman correlation coefficient r 0.56 – 0.66 depending on the perfusion map used, $p<0.001$). The size of the initial perfusion lesion correlated with the infarct size at one week (r 0.76 – 0.89, $p<0.001$). The mean infarct size at one week (51 cm^3) was closest to the initial perfusion lesion, when measured from rCBV maps (44 cm^3), whereas initial rCBF maps and MTT maps clearly overestimated the infarct size at one week, with perfusion lesion volumes of 75 cm^3 and 103 cm^3, respectively.

Conclusions. Perfusion-diffusion mismatch detected with combined diffusion and perfusion MR imaging in the acute phase is associated with higher risk of subsequent infarct growth. Infarcts often grow after the first few hours. Diffusion and perfusion MR imaging are effective in evaluating the tissue at risk and may have a role in clinical decision making, when acute phase treatments are considered.

STANDARD STEREOTAXIC COORDINATE SYSTEM FOR THE PIG BRAIN: AUTOMATIC MEASUREMENTS OF REGIONAL CBF AND $CMRO_2$ AFTER TRANSIENT MIDDLE CEREBRAL ARTERY OCCLUSION (MCAO)

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Background. The domestic pig is increasingly used as an experimental model for brain imaging studies with positron emission tomography (PET). In order to identify anatomical regions of interest in pig brain, we have created an MR-based Statistical Volumetric Atlas of the Brain of Pigs (Watanabe et al., in press). We now test the usefulness of this atlas for measuring regional changes in CBF and $CMRO_2$ after MCAO in pigs.

Methods. Four anesthetized pigs (40 kg) underwent a series of PET studies to measure CBF and $CMRO_2$ in the baseline condition, during MCAO (2hours), and during 6h of reperfusion. MCAO was produced using the left transorbital approach. Emission recordings were registered to the pig brain atlas and the regional CBF and $CMRO_2$ were calculated automatically in the MCA territory cortex (MCA-c), MCA territory white matter (MCA-w), putamen, caudate, thalamus and cerebellum on the left side.

Results.

CBF	MCA-c	MCA-w	Putamen	Caudate	Thalamus	Cerebellum
Pre	42 +/- 7	38 +/- 5	48 +/- 10	44 +/- 5	46 +/- 9	47 +/- 7
During	27 +/- 11	25 +/- 11	*21 +/- 7	27 +/- 3	41 +/- 8	39 +/- 5
Post 1h	44 +/- 11	42 +/- 11	53 +/- 11	48 +/- 8	49 +/- 4	45 +/- 4
Post 2h	44 +/- 6	40 +/- 5	53 +/- 15	49 +/- 11	52 +/- 5	43 +/- 4
Post 3h	54 +/- 18	*50 +/- 15	*60 +/- 19	56 +/- 15	60 +/- 14	49 +/- 12
Post 4h	48 +/- 12	44 +/- 11	55 +/- 20	49 +/- 12	57 +/- 12	47 +/- 11
Post 5h	47 +/- 13	45 +/- 14	51 +/- 19	49 +/- 13	54 +/- 10	48 +/- 8
Post 6h	40 +/- 7	37 +/- 7	38 +/- 3	39 +/- 8	48 +/- 1	43 +/- 7

(ml/100g/min)

$CMRO_2$	MCA-c	MCA-w	Putamen	Caudate	Thalamus	Cerebellum
Pre	207 +/- 23	181 +/- 16	229 +/- 39	194 +/- 24	225 +/- 47	212 +/- 30
During	*134 +/- 84	*116+/- 77	*95 +/- 61	141 +/- 35	227 +/- 22	191 +/- 42
Post 1h	187 +/- 80	170 +/- 66	159 +/- 57	172 +/- 43	218 +/- 66	210 +/- 40
Post 2h	187 +/- 58	171 +/- 56	153 +/- 33	166 +/- 43	230 +/- 35	197 +/- 36
Post 3h	165 +/- 79	152 +/- 76	144 +/- 89	140 +/- 37	187 +/- 59	162 +/- 61
Post 4h	177 +/- 73	162 +/- 70	*151+/- 71	140 +/- 47	211 +/- 62	188 +/- 65
Post 5h	161 +/- 54	152 +/- 49	*133+/- 34	*137+/- 43	189 +/- 82	193 +/- 61
Post 6h	*116 +/- 34	*108+/-37	*93 +/- 54	*109+/42	151 +/- 32	184 +/- 50

*: significant difference between pre value ($p<0.05$) (μmol/100g/min)

Conclusions. The MR-based statistical volumetric atlas of the brain of pigs was very useful for measuring post-ischemic changes in CBF and $CMRO_2$ in subcortical regions. However, the infarct volume was smaller than the presently defined cortex region. Consequently, the metabolic changes in the cortical MCA territory were underestimated.

MAGNETIC RESONANCE IMAGING OF MAGNETODENDRIMER-LABELED NEURAL PROGENITOR CELL TRANSPLANTS

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Background. Stem cell-based therapies are believed to offer a unique way of curing many debilitating diseases of the central nervous system (CNS), including stroke. Human pluripotent stem cells capable of differentiating into neural cell subtypes have been isolated (1,2) and can be maintained and cultured *in vitro*. We aim at developing a non-invasive technique for tracking neural progenitor (NP) cells *in situ*, and the curative potential of these cells, using an intracellular, superparamagnetic magnetodendrimer label (3) in conjunction with magnetic resonance imaging (MRI) in a mouse model. This combination should allow for excellent temporal and spatial resolution, facilitating research into cell-based therapies of the brain.

Methods. Embryonic stem (ES) cells of mouse 129 strain, derived from the inner cell mass of a mouse blastocyst (4), were cultured and differentiated into NP cells in differentiation media as described previously (5). Cells at 70-80% confluency were labeled with MD-100 magneto-dendrimers at a concentration of 20 µg Fe/ml of culture media for 24 hours, then trypsinized, and washed with phosphate buffered saline (PBS). Cells were then injected into 129 strain male mice (n=3) via stereotactic injection into the right striatum under pentobarbital anesthesia (5-25 x 10^4 cells/injection). Mice were sacrificed after seven days, perfused first with PBS to remove blood, and fixed with 4% paraformaldehyde (PFA). *Ex vivo* imaging of excised brains was performed on a GE Omega 400 microimaging scanner at 9.4T using a custom built coil. A 3D dataset was obtained using a multiple spin echo (SE) sequence (repetition time 3.0 s, echo times 31,41,50 and 59 ms) and a voxel size of (70x80x80 µm) after zero filling.

Results. Mouse NP cells could be efficiently labeled with magnetodendrimers, demonstrated by the presence of numerous positively staining cytoplasmic particles in Prussian Blue staining of replated cells. *Ex vivo* images showed a large hypointense area (T_2-values ranging from 1-20 ms) confined to within 0.5 mm of the injection site and along the injection needle track. Cerebral gross anatomy appeared unremarkable in all other aspects.

Conclusions. Magnetodendrimers allow for efficient labeling of stem cells, and the proliferative capacity and differentiation of the cells appear not to be affected by the label. The particles generate excellent contrast in MRI, and only a few labeled cells may ultimately be required for detection by gradient echo techniques. In therapeutic applications, larger numbers of cells are involved, and SE MRI techniques, which are less susceptible to magnetic field homogeneity and artifacts arising from parenchyma/ bone/air interfaces, can easily be used. We show here that MD-100 labeled NP cell transplants can be readily detected by MRI, allowing for the monitoring of the tissue localization of NP cells, and potential tissue regeneration in the CNS, in much the same fashion as shown earlier in the rat spine (6).

1. Shamblott et al. (1998) PNAS **95**:13726.
2. Thomson et al. (1998) *Science* **282**:1145.
3. Bulte et al. 2000 *Proc. 8th ISMRM*, 2062.
4. Doetschman et al. (1985) *JEEM* **87:**273.
5. Okabe et al. (1996) *Mech. Dev.* **59:**89
6. Bulte et al. (1999) *PNAS* **96:**15256.

NON-INVASIVE MONITORING OF INTRACRANIAL PRESSURE AND COMPLIANCE

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Introduction. Increasing intracranial pressure (ICP) is one of the most dangerous complications following acute stroke. Monitoring of ICP provides the physician with data for rational treatment management but to date this must be carried out by invasive methods which carry a considerable risk of serious complications.

Methods. In this study we have assessed a new non-invasive method for measuring ICP and compliance in 10 healthy controls. ICP variations were assessed by measuring tympanic membrane ICP propagation using a specially designed pressure transducer. Cerebral compliance was measured by simultaneously assessing the Windkessel volume displacement of the cerebral arteries. This is achieved by measuring the cross-section transcranial Doppler power index in the middle cerebral arteries with corrections for the Doppler frequency distributions (frequency corrected power). ICP was increased step-wise using a tilt-table (11-12 measurements in all subjects). This increases cerebral venous pressure which is the hydrostatic pressure relative to the right atrium and is known to be transformed into an equivalent ICP increase due to the valve function of the bridging veins. Compliance was calculated at each ICP value.

Results. The pressure increases measured by tympanic membrane propagation and the calculated ICP increases showed a linear relationship in all 10 subjects. Compliance curves also showed typical decreases in compliance with increasing ICP.

Conclusion. This study has shown that it is now possible to non-invasively monitor intracranial pressure and compliance. This new method should be assessed in extensive clinical studies.

SEAMLESS SERVICE CHAIN OF STROKE PATIENTS – HOW IT WORKS IN FINLAND

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In the presentation the latest development of health care reforms concerning seamless chains of care will be described. Is there really something new, or is it just a new label for stepwise structures. The need for renewed national and regional guidelines and care protocols are urgent also for the ischaemic stroke patients.

Do the stroke patient benefit from the hospital care? Are there some sort of mismatch of care, cure and rehabilitation for patients? This question will be analysed in the presentation, based on unpublished research data (about 2000 cases), in which cost benefit of hospital care of stroke patients is evaluated. The methodology used in the data collection and in the analysis is totally new. The material is collected from Finland.

The results shows that cost benefit of the stoke patient is not always at optimum level.

As a conclusion, it is obvious that more cost benefit research is needed to gather knowledge on real life effectiveness of care of stroke patients. The second conclusion is that in our health care system substantial reallocation of resources should be done before we can justify that the care is seamless from patients' point of view.

PATIENT CARE NEEDS AND NURSE WORK LOAD: STUDY OF THE NUMBER OF WORKING HOURS IN BED CARE UNITS OF SPECIALIZED PATIENT CARE

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Background. Systematically collected data concerning nursing is still at the moment scarce in health care, although nursing personnel forms the biggest group of hospital personnel. Patients' need for nursing care, their acuity levels, are the major factors which influence the workload of the practicing nurses. In Kuopio University Hospital there has been an international factor-type patient classification instrument in use since 1997. Classification of patients needs to be combined with quantification of specific nursing interventions and patient categories by nursing effort required. The purpose of this study was to identify the average nursing care time per patients' acuity levels, and thus validate the PC-instrument. Also, the staff nurses' and head nurses, perceptions on the adequacy of current staffing levels, were sought in this study.

Methods. Data were collected during the period of two weeks in March 1999 by self-observations and questionnaires to nursing personnel. Four wards were selected for the study: general medicine, oncology, gastroenterology, and neurosurgery. A data gathering instrument was developed that quantifies the nursing work into four groups: 1) direct care (12 activities), 2) non-direct care (8 activities), 3) unit-related work (7 activities), and 4) personal time. Work sampling techniques were used to ascertain time allocation to various activities. Quantitative methods were used in data analysis.

Results. The average time distribution between the four basic categories was: direct care activities 41 % (RNs 40%, LPNs 45%); indirect care activities 35% (RNs 39%, LPNs 26%); unit-related activities (not directly concerned with patients) 16 % (RNs 13%, LPNs 21%); personal time 8%. As patient's acuity level growed (more nurse dependent patients), the nurses' time distributed to caring also clearly increased. On average 46 % (variation 35-60%) of the staffing levels on various shifts were evaluated to be to some extent not adequate.

Conclusions. The hospital's patient classification instrument seemed to be valid enough, capable of dividing patients into different groups according their needs for nursing care. Work sampling method with nurses' self-observations proved to work well in this study. Most of the nurses' working time (76%) is patient centred, occupied with the activities that are being done to the patients either directly (patient is present) or indirectly (patient is not present). There are differencies in time distribution between different units, obviously depending on their staffing levels. Nearly every second working shift, and in some units even more, are understaffed.

STANDARD NURSING QUESTIONNAIRE IN ACUTE STROKE CARE

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Background. Patients with acute stroke are at high risk of developing complications as malnutrition, urinary tract infections and constipation. Systematic observation and monitoring are needed to ensure a correct treatment and care. The aim of this project was to implement evidence based clinical guidelines[1] of nutrition and elimination of urine and faeces in nursing practise

Methods. A key point in translating clinical guidelines to clinical practice is a standard questionnaire. Three standards were prepared and they were systematically used to identify risk factors and to evaluate the effect of nursing interventions.

Standard questionnaire of nutrition.
- Clinical swallowing assessment
- Clinical nutritional status

Standard questionnaire of elimination of urine.
- Incontinence history
- Voiding diary

Standard questionnaire of elimination of faeces:
- Incontinence score
- Constipation score

Data were assessed on admission and during hospital stay.

Results. The clinical guidelines and the standard questionnaires have been implemented in stroke units at Bispebjerg University Hospital, Århus University Hospital and Esbønderup Hospital. Since January 2001 and until now selected quality indicators have been registered in a stroke database as a pilot project. It is our plan to establish a full quality assurance protocol every 3 months. Results from spot checks will be presented at the meeting

Conclusions. Although still preliminary our results from spot checks indicate that guidelines and standard questionnaires can be implemented at dedicated stroke unit.

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[1]The clinical guidelines are made in cooperation with Doris Christensen Århus University Hospital and Ingrid Muus Esbønderup Hospital

EVIDENCE BASED NURSING PROGRAM FOR ACUTE STROKE

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WHO Region Europe has in The Helsingborg Declaration on stroke management recommended that patients with acute stroke should have access to care in specialised stroke units or from stroke team. In my presentation I will present some of the key points in a nursing program for acute stroke which has shown to be effective and which will be a part of new guidelines from WHO. High quality nursing performed by nurses with knowledge, competence and interest in stroke patients is essential in acute management of stroke. To improve the outcome for stroke patients it is important to have nursing of high quality.

Meta-analyses show that dedicated stroke units are the most effective way to organize acute stroke care. Most evidence is from units, which focus both on acute treatment and acute rehabilitation. In the University Hospital of Trondheim, Norway, we have such a combined stroke unit and a nursing program which has shown to be very effective. The nursing program in our unit is based on experience from the treatment program which was developed in our unit during a randomised controlled trial on stroke unit care, and from information and data collected by the Stroke Unit Triallists`s Collaboration.

Key functions of acute nursing and care of stroke patients in a stroke unit are:

-The role of the nurses on admission of an acute stroke patient.
-Systematic observation of vital signs and neurological deficits by the nurses including use of acute stroke scales.
-Acute nursing of unstable bed-ridden stroke patients.
-The role of the nurses in the acute medical treatment.
-Nutrition/swallowing/elimination.
-Early stimulation/mobilisation/rehabilitation.
-Co-ordination of the stroke team.
-Nursing in order to prevent common complications.
-Information/psychological support
-Continuity of care.

Acute nursing of stroke patients should be within the frame of a stroke unit and a trained nursing staff is one of the most important factors for evidence based stroke unit care. However, several of the principles of acute nursing of stroke patients can also be applied to other settings if stroke units are not established.

UPDATE ON STROKE REHABILITATION

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The purpose of this presentation is to review the evidence to support therapeutic interventions to enhance post-stroke rehabilitation. Basic science studies that support biological plausibility of therapeutic interventions will be presented. Results from recently completed efficacy studies and the design of ongoing clinical trials of therapeutic interventions will also be reviewed. Finally we will discuss four factors that must be considered to enhance integration of research into clinical practice: generalizability of results, meaningful outcomes, support of combination of interventions, and cost-effective interventions.

HOW THE UMEÅ STROKE CENTER STAFF WORKS TO INSURE GOOD AND EFFICIENT STROKE NURSING PRACTICE

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At Norrland's University Hospital in Umeå, there have been stroke units at various locations, i.e. at the Department of Medicine since 1977 and at the Neurology Department since 1987. However, in September 1999, these stroke units were merged into a single multidisciplinary Stroke Center. All patients with TIA and acute stroke (except SAH) from the Umeå catchment area (app. 150.000 inhabitants) are referred to the Stroke Center. In addition, patents with suspected symptomatic carotid stenosis presumably eligible for carotid endarterectomy as well as young, i.e. stroke patients <45 years old are referred from the Northern Sweden (cathment area of app. 1 million). The Stroke Center has 21 beds and gives care to ca 700 patients each year, i.e., app. two acute admissions each day. The mean duration of stay at the unit is 8.5 days.

The expectations on the Stroke Center from the society are large and a good and safe care is required at minimum costs. The whole hospital health care system has a lack of caring staff, especially nurses and doctors. One general trend is also that personnel at various wards do not stay as long today as in the past. The Swedish National Board on Health and Welfare has recently published national guidelines on stroke. The results will then be followed up by the nation wide unique quality assurance registry stroke patients (Riksstroke).

It is according to these facts that we have organised the caring of stroke so that the work is performed according to documented routines/guidelines that generates an effective and safe caring throughout the entire stay that is from the emergency to the discharge. These documented routines/guidelines are based upon science and clinical experiences. We put great emphasis on a good introduction for our new employees to create security and knowledge. This gives patients/relatives competent persons as caring staff. The material is continuously study and update.

Our goal is to give patients/relatives stroke care that has high quality, is safe and cost effective. We can with the following documented routines/guidelines secure the quality of stroke care: 1) acute medical observational protocol chart (in order to detect early deterioration), 2) nurse care and patient's activity protocol (in order to avoid complications and to grade activity), 3) lists on medical investigations required, 4) daily routines, 5) multidisciplinary care conference, 6) initial supervision routines and specific nursing measures, 7) resuscitation at cardiac stop, 8) memo medical measures, 9) memo specific nursing measures, 10) memo occupational therapy, 11) memo physiotherapy, 12) information to patients/relatives, 13) specific demands of knowledge according to responsibility, 14) introduction programme for new employees and students, 15) study visit frame/information to our interested parties, 16) discharge folder and 17) plastic coated cribs in pocket form for all personnel, new employees, substitutes, students describing: routines/guidelines for the whole stay, resuscitation of heart and lung, multidisciplinary care conference, initial supervision routines and specific nursing measures, and daily routines, that is the content of a typical day at the Stroke Center. Guidelines for: 1) medical investigation/treatment, 2) general and specific care, 3) mobilisation, 4) general and specific rehabilitation, 5) patient- and relative information, 6) multidisciplinary care conference and 7) treatment chain – transfer of information to and co-ordinated treatment planning in municipality and county.

In our opinion the above-presented concept will enable good and efficient stroke care and we also think the concept is possible to implement at all stroke units.

STUDY NURSE'S VIEWPOINTS

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It's self-evident that medical studies has to be done to get new information and new medicines to help neurological patients. Today these trials are often large international studies and well organized by medicine factories. Principal investigator has often co-investigators and a study nurse to help his work. A study nurse can be hired by the hospital, CRO (Contract Research Organization) or the investigator himself. In this presentation I concentrate on ethical responsibility of a study nurse.

In Finland patients are quite willing to take a part in medical trials. The study information to the patient is usually given by the investigator. The most important duty of a study nurse is to make sure that the patient is well informed of the trial. "Informed consent" really means that the patient must be aware where he or she is taking apart. Nothing can be done before you have the informed consent, which is signed by the patient. In a case of neurological patient sometimes the patient himself cannot sign the paper. In this case the investigator must interview the patients spouse or parent. He or she may sign the informed consent, if he thinks it's what the patient should choose himself, if was able to do so. It's important to know that not even a blood test can be taken before the informed consent. Study nurse must also pass this information on wards and stroke units, so that every nurse knows the patients rights.

Another responsibility of a study nurse is to motivate personal in stroke units and wards to take part in trials. Sometimes the personal thinks that it's too much work extra on top of all other work. All the other patients has also to be taken care of! A good trial is always a matter of teamwork. One ethical question might also be time and money. Is the study nurses work done on hospital time, or own time? Where does the money come from? How it feels to be on call for 24 hours…

In my opinion participating in medical trials motivates nurses also to do own nursing studies and develop nursing in that unit or ward. Also it's good way to get latest information from medical field.

THE FUTURE OF HEALTH CARE

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Allthough health services appear to differ from one country to another, there are certain common factors that influence the evolution of systems of health service organisation and health care delivery in all countries. Population ageing, rising patient and health care personnel expectations and the advent of new technologies are obvious in industrialised nations, but these factors affect all the other nations too.

The dilemma in Finland and all the other industrialised countries is that in health care more is unlikely to mean better and is neatly summed up in a phrase: “doing better and feeling worse”. If no boundaries are placed on the public’s seemingly voracious appetite for health care then a country`s whole GNP could be consumed by health care.

In Finland there has been serious problems in health care for many years. In any circumstances we can not afford all the therapies patients might benefit from. The rationing dilemma is worsening all the time. We can not continue this way anymore. We have to start to make choices, doing the right things right.

NON-PHARMACOLOGICAL PREVENTION OF STROKE – DOES IT WORK?

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The ongoing decline in stroke mortality in West Europe is attributed mainly to reduced short-term and long-term case fatality (risk of dying once a stroke has occurred), reflecting improved management in the acute phase and more effective secondary prevention. In contrast, population-based stroke registries have shown that the incidence of stroke has been unchanged with one exception: Finland, where incidence rates have been declining. It seems that primary prevention against stroke has, in general, not been successful to reduce the risk of stroke in West Europe. It can, nevertheless, be speculated that preventive efforts are resulting in less severe strokes, contributing to the reduced case fatality.

Of the methods used in high-risk strategies in individuals, smoking cessation by doctor's advice and nicotine replacement therapy is highly cost-effective, whereas a cost-effectiveness pharmacological interventions in people with hypertension remains to be demonstrated.

A systematic review of controlled community-based multifactorial intervention programs against cardiovascular diseases has been conducted. Ten such program in which the development was compared between an intervention and a control population were identified. Only one of the studies has reported on stroke morbidity (Minnesota Heart), whereas another two have reported on stroke mortality rates (North Karelia and German Cardiovascular). In all projects, the effects on classical cardiovascular risk factors have been studied, and the possible impact on stroke occurrence can be calculated using risk factor equations.

In none of the studies that have included stroke morbidity and/or mortality, has the trends been more favourable in the intervention population than in the control population. The risk factor equation shows a possible modest reduction of the risk of stroke by a few of the programs, but in most cases the development in the intervention and control areas has been the same. The specific effects of multifactorial prevention programs thus seems to have small or moderate effects on the risk of stroke. On the other hand, there have been at least two controlled trials (in Sweden and China) in which community-based programs dedicated to reduce blood pressure have shown effects on stroke risk at the population level.

In the view of the apparent difficulties to document beneficial effects of community-based intervention programs against cardiovascular diseases, methodological issues in the evaluation of such programs will be discussed.

BENEFITS AND RISKS OF ANTIPLATELET THERAPY IN THE SECONDARY PREVENTION OF STROKE

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Patients who suffered a TIA or a first ischaemic stroke have a 10-15% risk of a first or recurrent stroke within the first year. Stroke risk can be reduced by anti-platelet drugs. Acetylsalicyclic acid (ASA) reduces stroke risk compared to placebo by 13-14%. Risk reduction is independent of dose of ASA. The risk of significant GI-bleeding is also not dose-dependent. GI-intolerability, however, increases with the dose of ASA. Therefore low doses of ASA between 50 and 325 mg are recommended for the secondary prevention of stroke. Ticlopidine (2x250 mg) is superior to ASA for stroke prevention. The major serious adverse event of ticlopidine is neutropenia which may occur in up to 0.8% of patients within the first 3 months of therapy. Ticlopidine has been replaced by clopidogrel 75 mg which reduces the combined endpoint of stroke, MI and vascular death by 8.3% compared to ASA alone. Clopidogrel is well tolerated. The combination of ASA (2x25mg) and slow release dipyridamole (2x200 mg) offers an additional benefit of 23% over ASA alone. There is no increase in the risk of servere bleeding complications compared to ASA alone. Dipyridamole may lead to headache at the beginning of therapy. This can be overcome by a slow increase in dose. Neither Dipyridamole alone nor the combination with ASA increase the risk of cardio-vascular events. The treating physician has a selection of therapeutical options in the secondary prevention of stroke. Decisons which drug is used depend on the efficacy, the side effect profile and cost/benefit considerations.

NEW EVIDENCE ABOUT TEMPERATURE, BLOOD PRESSURE AND BLOOD SUGAR IN THE EARLY HOURS AFTER STROKE ONSET

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Acute stroke is a dynamic process where body temperature, arterial blood pressure and plasma glucose may change within the first hours after stroke onset. Researchers have found that pyrexia after stroke onset was associated with market increase in morbidity and mortality (1). High blood pressure on admission has been associated with increased mortality (2) and elevated blood glucose on admission has been associated with poor outcome (3,4). We here report data on a series of 1050 acute stroke patients admitted within 6 hours of stroke onset. In 50% of these patients the first measurements were done within 2 hours of stroke onset.

Temperature. In our prospective observational study (5) we found normal or subnormal temperature in the very early hours after stroke onset. In patients with severe stroke temperature started to increase after 4 – 6 hours; and 8 – 10 hours after stroke onset increased body temperature was related to poor outcome at 3 months. This association, however, was due to the initial severity of the stroke and not to the increase of temperature. In patients with mild stroke there was no change in temperature.

Blood pressure. In acute stroke patients with mild ischemic stroke or with TIA blood pressure declined significantly within the first 4 - 6 hours after admission, at what time a lower blood pressure was associated with a favourable outcome at 3 months. In patients with severe ischemic stroke there was little change in blood pressure over the first 36 hours. Blood pressure did not differ in patients with deteriorating stroke from that in patients without deterioration.

Glucose. In non-diabetic patients hyperglycaemia in the acute phase has been associated with poor outcome. We found that plasma glucose increased during the first hours after stroke onset, more so in patients with severe stroke than in those with mild stroke. This may indicate that elevated blood sugar is a stress response, as found earlier in a Finnish study (6).

1. Hajat C, Hajat S, Sharma P. Effects of poststroke pyrexia on stroke outcome. A meta analysis of studies in patients. Stroke 2000;31:410-14.
2. Carlberg B, Asplund K, Hagg E. The prognostic value of admission blood pressure in patients with acute stroke. Stroke 1993;24:1372-75.
3. van Kooten F, Hoogerbrugge N, Naading P, Koudstaal PJ. Hyperglycemia in the acute phase of stroke is not caused by stress. Stroke 1993;24:1129-32.
4. Weir CJ, Murray GD, Dyker AG, Lees KR. Is hyperglycaemia an independent predictor of poor outcome after acute stroke? Results of a long term follow up study. BMJ 1997;314:1303.
5. Boysen G, Christensen H. Stroke severity determines body temperature in acute stroke. Stroke 2001;32:413-17.
6. Murros K, Fogelholm R, Kettunen S, Vuorela AL, Valve J. Blood glucose, glycosylated hemoglobin, and outcome of ischemic brain infarction. J Neurol Sci 1992;111:59-64.

DRUG TREATMENT OF ACUTE ISCHAEMIC STROKE

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Reperfusion, induced by fibrinolytic drugs in the hyperacute phase after occlusion of a cerebral artery, is today the only specific treatment of acute ischaemic stroke with proven efficacy. Treatment must be initiated within three hours, or at least 6 hours after stroke onset. The risk of early fatal and symptomatic intracranial haemorrhage is increased, but these hazards are offset by reduction in the proportion of patients being dead or dependent. A decision to treat a patient with thrombolysis is presently based on a clinical evaluation and lack of haemorrhage and extensive ischaemic damage on CT scan. The development of MR imaging documenting a perfusion deficit more extensive than an irreversible ischaemic lesion (‘perfusion/diffusion mismatch’) is likely to increase the net benefit of this treatment in the future. During the last two decades, research has also been focussed on finding drugs with neuroprotective effects for cerebral ischaemia, but these efforts have not yet been successful. There is a striking discrepancy between animal stroke models and clinical results for potential neuroprotective agents. Methodological weaknesses in the design of both experimental studies and clinical trials may be part of the explanation for the lack of positive results. It is worrying however that some recent positive experimental results with designs closely mimicking the clinical setting also have failed to reproduced in stroke patients, so the validity of the animal stroke models is challenged. Improved study designs and increased knowledge on the complexity of the pathophysiological interactions during ischaemia may however justify continued efforts to develop neuroprotective classes of drugs. Drugs directed towards relearning of lost neurological functions are in early development. Aspirin has a small but statistically significant preventive effect on early recurrences and this drug is now a part of the evidence-based management of acute ischaemic stroke.

A DECLINE IN 28-DAY CASE FATALITY BUT STILL NO CHANGE IN STROKE INCIDENCE: A POPULATION-BASED STUDY IN NORTHERN SWEDEN

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Background. Over the last decades, stroke mortality has decreased in western countries, whereas is has increased in many of the East European countries. In the stroke component in the WHO MONICA Project, trends in stroke attack rate (recurrent and first ever strokes) and incidence (first ever) as well as case fatality have been followed. The aim of this study was to investigate trends in first ever and recurrent stroke events and 28-day case fatality in different age groups.

Methods. Within the Northern Sweden MONICA study, stroke has been monitored since 1985 in age group 25-74 years in both men and women. The same strict WHO criteria have been used during all years. In the present study, patients with subarachnoid hemorrhages have been excluded. In total 5,584 men and 3,537 women had a first ever stroke and an additional 25% (1,920 men and 1,187 women) had a recurrent stroke during this period, in total 12,228 acute stroke events in a 14-year period.

Results. In men, stroke attack rates in age groups 25-54, 55-65 and 65-74 years varied little over the years. The attack rate in age group 25-74 years varied between 315 and 366 per 100,000 and year, with no significant secular trends totally in age group 25-74); $p=0.67$, or in any of the other age groups. In women the attack rate varied between 239 and 194 with a significant decrease seen in age group 65-74 years ($p=0.014$). The entire reduction came from a decline in women with recurrent strokes ($p=0.001$). No trends over time were seen in first ever stroke in women ($p=0.61$). In all men and women aged 25-74 years, CF declined from 19% in 1985 to 11% in 1998. Logistic regression analyses showed a risk reduction in dying from stroke to 0.55 ($p<0.0001$) the last year compared to the first, with an absolute annual reduction in CF of 3%, both in men and women. Declines in early (0-7 days) and late (8-17 days) CF contributed to similar extent to the overall decline in 28-day CF.

Conclusion. Since 1985, there have been no changes in first ever or recurrent stroke, in any age group in northern Sweden except for in women, where there has been a decreasing trend in recurrent stroke. During the same period case fatality has been almost halved, probably due to advances in stroke management. With more people aging and with decreasing case fatality there will be more people who have had a stroke living in the community.

THE INDEPENDENT PREDICTIVE IMPACT OF ADMISSION BODY TEMPERATURE ON LONG-TERM MORTALITY AFTER STROKE: THE COPENHAGEN STROKE STUDY

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Background. Body temperature is considered crucial in the acute management of stroke patients. Lowering body temperature in the acute stage of stroke may be neuroprotective in man. In this study we sought to investigate the independent impact of admission body temperature on long term mortality.

Methods. In the community-based Copenhagen Stroke Study comprising 371 unselected patients admitted within 6 hours of onset we measured body temperature on admission. Patients were followed mean 84 months after stroke. To evaluate admission body temperature as independent predictor for long-term mortality we build a Cox Proportional Hazards model including admission stroke severity as measured by Scandinavian Stroke Scale (SSS), age, cardiovascular risk factors, and patient demographic characteristics that may be ralated to long-term mortality.

Results. Univariately, patients with hyperthermia (> 37 degr.) acutely had more severe strokes compared to patients with hypothermia, mean SSS score 30.1 vs. 36.1, p=0.001. No differences were found for age, previous stroke, stroke subtype, or cardiovascular risk factors. Mortality rates at 60 months were 58% versus 42% respectively, p=0.006. In the Cox Proportional Hazards model long-term mortality was independently predicted by admission body temperature (OR 1.3 per 1 degr. C increase; 95% CI 1.1-1.6).

Conclusion. Body temperature in acute stroke predicts mortality for patients admitted within 6 hours of onset. This seems to be a long lasting rather than a temporary relation. Intervention by lowering body temperature may improve even long-term outcome in acute stroke patients.

DIFFERENCES IN LONG-TERM OUTCOME BETWEEN PATIENTS TREATED IN STROKE UNITS AND IN GENERAL WARDS

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Background. The long-term beneficial effects of non-intensive stroke unit care (SU) have been proven in several randomized trials. However, there is a question of large-scale applicability in routine clinical practice, of interventions used by dedicated investigators in small randomized trials. The objective of this study was to compare, two and a half years after stroke, patients that had been treated in stroke units and those treated in general wards in routine clinical practice.

Methods. This is a prospective cohort study based on 8194 patients who were included, during the first six months in 1997, in Riks-Stroke, the Swedish National Register for quality assessment of acute stroke. Two years after the event, 5189 patients were still alive, and 5104 were followed up with a postal questionnaire to which 4038 responded.

Results. During the hospital treatment, 5134 patients were treated in SUs and 2400 were treated in general wards (GW). An additional 659 patients were treated in a setting which neither fulfilled the criteria for a SU nor a GW. The following results are presented for the group of patients that was independent in primary ADL functions before the stroke. The patients that were treated in SUs were two years younger than patients treated in GWs and they had less often cerebral haemorrhages and an impaired consciousness at arrival to the hospital. Two years after the stroke, patients in the SU group were more often independent in ADL functions, (OR 1.27, 95% CI 1.52-1.06). Among patients that before the stroke lived at home, patients treated in SUs had a lower case fatality two years after the stroke (OR 0.81, 95% CI 0.72-0.92). These results are valid also after adjustment for differences in case mix. The results for the patients that before the stroke were dependent in ADL functions, showed no differences between patients treated in SUs and in GWs before the stroke and during hospital treatment. Nor were there any differences in outcome two years after the stroke.

Conclusion. Long-term beneficial effects of treatment in stroke units in routine clinical practice, were shown for patients who were independent in primary ADL functions before the stroke. No benefits were shown for patients who were dependent for primary ADL functions before the stroke. Further studies on this group of patients with more detailed outcome measures are needed.

TRENDS IN THE INCIDENCE AND MORTALITY OF STROKE EVENTS IN FINLAND DURING 1993-1997: THE FINSTROKE STUDY

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Background. Stroke continues to be an important public health problem. FINSTROKE is a collaborative stroke register project, which continues the work of the FINMONICA Stroke Register, (a part of the WHO MONICA Project) in Finland. The aim of the present study was to evaluate the 5-year trends (from 1993 to 1997) in incidence and mortality of stroke and it's subtypes in two geographical areas of Finland.

Methods. FINSTROKE is a population-based stroke register, which aimed to collect data on all suspected events of acute stroke that occurred in the population aged 25 to 99 years permanently residing in the monitored areas of the town of Turku, the town of Kuopio and certain adjacent rural communities. During the period 1993-1997, 4890 suspected stroke cases aged 25-74 years, were checked and evaluated for registration. Of these, 4022 fulfilled the register criteria of an acute stroke event. The trends in the incidence, attack rate, and mortality of stroke were estimated from the logarithms of the annual age-standardised event rates using ordinary linear regression, and the confidence intervals of trends were obtained in the usual manner from the standard error of the regression coefficient. Results in this report are based on preliminary analysis of the FINSTROKE data.

Results. According to the official mortality statistics of Finland, mortality from stroke (ICD codes 430-38, I60-I69) has declined from 1991 to 1998 on average by 4.3% per year in men and by 5.6% per year in women. In the FINSTROKE register we observed a significant decline in the mortality of stroke among women aged 25-74 years (-13,29% / year (95% confidence interval [CI], -23.4% to -3.19%, $p < 0.03$). Among men, a declining trend was also observed, but it did not reach statistical significance during the short study period. When subarachnoid haemorrhages, intracerebral haemorrhages and ischaemic strokes were considered separately declining trends were observed both in the incidence and mortality, but usually they did not reach statistical significance at the 0.05% level. The only changes that reached statistical significance during the 5-year study period were the incidence of ICH (-22.63% 95%CI -37.6 to -7.7%) and the occurrence of recurrent ischemic strokes (-23,3% /year 95%CI -45,4 to -1,1%), both among women. Interestingly, our data suggested a substantial reduction of the prehospital delay: the proportion of patients arriving at hospital within 3 hours after the onset of symptoms increased from 20% in 1993 to over 50% in 1997 in both genders.

Conclusions. Data of the FINSTROKE study demonstrated that both the incidence and the mortality of stroke have continued to decline in Finland during 1993 - 1997.

PERILESIONAL DYSFUNCTION FOLLOWING STROKE

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Focal brain ischemia has a considerable impact on the function of perilesional and remote brain tissue. Time course, topography and functional consequences of these functional disturbances will be described: (1) In perilesional and remote brain areas, a hyperexcitability can be observed which is associated with an alteration of the GABAergic neurotransmitter receptor system. Functionally, this alters sensory representations in the brain, it increases brain plasticity, but it also may induce epileptiform discharges. Clinically, post stroke epilepsy can be observed in 5 - 25 % of patients. (2) In the penumbral tissue, the excitatory responses are reduced. This may be associated with a loss of function in these areas. (3) At late stages following stroke, a premature aging of neurotransmitter receptor patterns can be observed indicating that focal brain lesions may have a detrimental effect also on remote brain tissue.

STROKE AND DEPRESSION

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Poststroke depression (PSD) constitutes a challenging field of research in clinical neurology not only because of the possibility of correlating emotions, mood states, and behaviors with neural circuits, but also because PSD has both high incidence (20 to 60%) and a strong negative impact on functional outcome. The etiopathology of PSD is not yet completely understood. The neurocognitive approach is of limited value in such a research. The complexity of behavioral and cognitive symptoms and the marked inter-patient variation in clinical expression limit the use of protocol paradigms in functional neuroimaging. The physiological neural systems processing mood regulation and the neurotransmitters involved are only partially known, and there are no biological markers and experimental models suitable for PSD.

A vast amount of scientific literature has, unfortunately, failed to define anatomo-clinical correlations. Methodological limitations and metanalysis studies strongly suggest that results regarding the role of a left-right and an anterior-posterior gradient may be biased and that lesion location may contribute only a small extent to the risk of developing PSD. Nevertheless, there is sufficient data to retain the role of fronto-subcortical, temporo-limbic, basal ganglia, prefrontal circuits and other limbic-related areas, while a serotonine imbalance is probably the neurotransmettorial or metabolic causal factor of PSD.

Univocal diagnostic criteria (DSM-IV) are actually available in order to diagnose PSD, nevertheless the possibility of subgroups of syndromes (major and minor, predominantly with vegetative signs or with verbal referral, or reacting to psychological or to social factors) makes problematic not only to carry out clinical investigations on large population samples, but also to maximize treatment of individual patients. A major reworking of criteria for case definition is then necessary, placing great emphasis on cognitive, behavioral, somatic or vegetative changes. Other cognitive (i.e. aphasia, anosognosia, and amnesia), behavioral (i.e. motor aprosodia, fatigue, apathy, emotionalism, catastrophic reactions, indifference reaction, and loss of psychic auto-activation), physiological (appetite, physical immobility, dysphagia, slurred speech, and noncommunication), and psychological changes due to the stroke itself should be carefully assessed in patients and not misinterpreted as PSD.

Finally it is important to determine the impact of specific functional deficits (i.e. aphasia, hemiplegia, visual disturbances) that account, at least in part, for the mood change. Poststroke depression and cognitive impairment are strictly correlated and both influence negatively the outcome. There is no definite consensus as to whether cognitive impairment is a consequence of PSD or vice versa and which condition is the main risk factor for developing dementia after stroke (‘‘dementia of depression’’ or ‘‘depression of dementia?).

Psychosocial factors have also great significance. The stroke patient is coping with a serious physical illness that reduces his self integrity in terms of personal vulnerability and low self-esteem and generates possible conflicts with close relationships or other caregivers. Strategies to increase social activity, support and contacts, financial security, job satisfaction, adequacy of living arrangements, and psychiatric care are needed in order to provide the best care. Specific psychiatric or neurological evaluation of the depressive symptoms, including subjective distress and vegetative symptoms, can permit the choice of more specific therapies, such as cognitive or pharmacological approaches or a combination of the two. Pharmacological therapy, when the diagnosis and indication have been defined can provide great benefit and improve the quality of life. The high prevalence of depression accompanying cerebrovascular diseases and the prolongation of disability in depressed people with stroke means that closer liaison is needed between psychiatrists, neurologists, and physicians caring for the elderly.

STROKE AND DEMENTIA

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Stroke and dementia, both independently and in combination, have caused a growing economic and human burden. The prevalence of dementia is larger for stroke survivors. A substantial fraction of demented patients have vascular risk factors and brain changes, regardless of etiology. This holds promise for early prevention and treatment.

SUMMARY POINTS:

The effect of different definitions on prevalence. depending on the composition of cohorts and variation in defining the cognitive syndrome, prevalence rates of poststroke dementia differed from 13.6% to 31.8% at 3 months post-stroke.
Associated clinical findings. in addition to age and the low level of education, different combinations of vascular risk factors (prior stroke, diabetes and non-white race) and stroke features (dysphasia, major dominant stroke syndrome and lacunar stroke) have been associated with poststroke dementia.
Imaging correlates. the radiological correlates of post-stroke dementia vary in different studies, including the following factors: infarct features (type, side, site, number and volume), extent and type of white matter lesions (WMLs) and degree and site of atrophy.
Impact on ADL. cognitive impairment has been shown to be an independent correlate of dependent living post-stroke, after adjusting for age and physical impairments.
Behavioral correlates. depression is the most common neuropsychiatric consequence of stroke, affecting 20-65% of the patients and it is associated with poststroke cognitive impairment.
Prognosis. a significant reduction in survival rates has been reported in stroke patients with dementia.

Alzheimer's disease (AD) and cerebrovascular disease (CVD). the magnitude of groups with mixed dementia, [AD with CVD/Vascular Dementia (VaD)], group has been previously underestimated; its proper diagnostication is a challenge, especially in the older population. Several vascular risk factors and brain changes relate to clinical manifestation of AD.

Challenge. dementia criteria are typically modelled on AD, whereby involvement of the medial temporal lobe results with dense episodic memory impairment. By contrast, patients with vascular lesions have no such predilection. It has been suggested to abandon the "Alzhemerized" dementia concept in the setting of CVD, and substitute it with a broader category of vascular cognitive impairment. Therefore, the challenge is to find the patients at risk for stroke and vascular and cognitive impairment.

MEASURING STROKE OUTCOMES

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Physical function limitations are the most common sequelae of stroke. These limitations have a major impact on stroke quality of life as well as economic burden of the disease. In order to adequately describe physical limitation across the entire range of severity, instruments need to be sensitive to manifestations of limitations all along this continuum. None of the currently used instruments meet this requirement. The most common measure of stroke related physical disability is the Barthel Index, which grades patients on their level of independence in activities of daily living. The Barthel has been shown to suffer from ceiling effects in which patients with the best score nevertheless have substantial disabilities. The Barthel Index discriminates between the more disabled individuals with stroke, but is insensitive to functional differences among less severely disabled individuals. The physical domain of the SF-36 has also been recently used to measure physical function after stroke. In contrast to the Barthel, the SF-36 has floor effects in which patients with the worst score may yet deteriorate further. However, the SF-36 discriminates differences in the less severely disabled individuals with stroke (8). While different instruments could be used for patients at different levels of severity, there are benefits to having a single instrument for all levels of severity.

The Stroke Impact Scale (SIS) is a new stroke specific outcome measure that is comprehensive and psychometrically robust in its assessment of stroke-related outcomes (4, 11). The measure was developed from the perspective of patients, caregivers and health professionals with stroke expertise. The SIS assesses eight domains: Strength, Hand Function, ADL/IADL, Mobility, Communication, Emotion, Memory and Thinking, and Participation (4). Four of these domains (Strength, Hand Function, ADL/IADL, and Mobility) can be combined to produce a composite physical domain. The SIS composite physical domain score was developed to extend the range of function measured by the Barthel ADL index and the physical domain of the SF-36. This presentation will introduce the SIS and compare it to the Barthel Index.

POTENTIAL EFFECTS OF DRUGS ON STROKE REHABILITATION

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Studies in laboratory animals clearly show that the rate and extent of functional recovery after focal brain injury can be modulated by drugs affecting certain central neurotransmitters. These studies also show a complex relationship between drug dose, behavioral experience, and timing in relationship to the stroke and the interval, intensity and duration of interventions. This presents a challenge for the design of clinical trials. Understanding this pharmacology is also important as several of the classes of drugs that impair recovery in laboratory experiments are used in stroke patients to treat coincident medical problems. Preliminary clinical studies suggest that similar drug effects occur in humans recovering from stroke.

THE ROLE OF NEW TREATMENT METHODS IN ACUTE STROKE

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During the past ten years, a number of potential new treatments for enhancing recovery during rehabilitation after stroke have been proposed. These new treatment approaches, as well as older techniques, are being evaluated for effectiveness through randomized clinical trials. This presentation will focus on new techniques to improve motor recovery following stroke.

Efforts to enhance motor recovery are a major focus of stroke rehabilitation. While there is a general acceptance that traditional techniques of physical and occupational therapy facilitate adaptation, whether they promote enduring recovery of motor function remains controversial. Rigorous clinical trials of traditional physical therapy approaches are few. Although the intensity of rehabilitation appears to be important, the timing and duration of therapy programs remains controversial.

New approaches to motor retraining have recently received considerable publicity. These include treadmill training with partial body weight support, constraint-induced therapy (CIT) and robot-assisted training. Studies of treadmill training with body weight support suggest that this approach promotes improvement in important gait parameters, including cadence and stride length. The approach that has received the most attention for upper extremity rehabilitation is CIT. Originally devised as a method to overcome “learned non-use” of deafferented forelimbs in monkeys, positive results obtained in small clinical trials in stroke patients led considerable enthusiasm for this technique. During CIT, the unaffected hand is constrained, forcing the individual to use the stroke-affected upper extremity during repetitive practice of functional tasks. The technique has been shown to be modestly effective in studies of both chronic and acute stroke patients. The benefit is probably related to the intensity of CIT therapy rather than the fact that the unaffected hand is restrained. A large multi-center randomized trial of immediate vs. delayed CIT therapy (the EXCITE study) is currently enrolling 240 patients 3-6 months following stroke who have completed traditional therapy. Another novel approach has been the use of robot-assisted therapy for the stroke affected extremity. Several prototypes and training techniques are currently under study. Finally, there is renewed interest in pharmacological enhancement of motor recovery following stroke. Amphetamine coupled with motor training leads to improvement of motor function in animal stroke models, and small clinical trials demonstrated similar improvement in humans. A randomized multi-center placebo-controlled study of dextroamphetamine in stroke patients undergoing rehabilitation is currently in progress.

The availability of non-invasive techniques to study cortical motor networks has provided new insight into motor recovery following stroke. Functional neuroimaging (PET and BOLD fMRI) measures changes in cerebral blood flow during motor tasks. Transcranial magnetic stimulation (TMS) results in depolarization of pyramidal neurons. Functional neuroimaging and TMS therefore provide related, but complimentary information about motor function. Cross-sectional studies in subjects with chronic stroke have suggested that certain patterns of motor network reorganization are associated with good clinical recovery, but the temporal development of those changes and whether they truly represents the substrate of recovery remain unknown. Preliminary results from a longitudinal study of stroke recovery using TMS and fMRI will be presented. Motor networks may also change in response to specific programs of motor training. Preliminary studies using TMS have demonstrated an expansion of the representational motor map of the stroke-affected hand following intensive training. These findings parallel changes in primates with experimental cortical lesions following motor training.

Innovative new approaches to enhance motor recovery and the availability of non-invasive techniques to assess cortical motor networks in humans makes this an exciting time for scientists and clinicians interested in stroke rehabilitation. Better knowledge of the mechanisms that underlie cortical recovery will likely result in improvements in stroke rehabilitation programs.

WHAT DETERMINES EARLY ADMISSION OF PATIENTS WITH ACUTE STROKE?

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Background. Early admission is a prerequisite for optimal stroke treatment. We studied factors related to delay of admission to hospital.

Methods. The study is prospective and community-based including all patients with stroke/TIA admitted to hospital in a Copenhagen community. The Scandinavian Stroke Scale was used to measure stroke severity. Patients were divided into 3 groups: Admitted directly via ambulance, via general practitioner (GP), via other routes. All patients had a structured interview.

Results. We included 494 stroke patients and 63 TIA patients. Mean age 74 years; male/female ratio 50.4%. Admitted via GP 49%, via ambulance 38%, via other routes 13%. In only 286 patients (51%) reliable information about time of stroke onset was available. In the rest patient or caretaker/relatives were unable to supply this information reliably. Mean delay was 8.6 hours. In patients admitted via GP 16.9 hours, via ambulance 2.4 hours, via other routes 10.2 hours. Of the 286 patients 137 (48%) arrived within 3 hours and of these 65% arrived via ambulance; 170 patients arrived within 6 hours and 58% of these came by ambulance. *Univariately* arrival within 6 hours was significantly related to stroke type/severity, admittance via ambulance, consciousness, aphasia and patients knowledge of the cause of symptoms. *Multivariately* independent predictors of early arrival within 6 hours were: Admittance via ambulance OR 5.6 (95% CI 3.2-9.8; p<0.0001) and knowledge about the cause of symptoms OR 2.6 (95% CI 1.5–4.7, p=0.001). Type/severity of stroke, age, gender, being alone on stroke onset, previous stroke/TIA and pre-stroke function were of no predictive value.

Time from symptoms to admission

	Delay hours (SD)	0-3 hours	0-6 hours	Total
Ambulance	2,4 (4,6)	94/43	110/27	137
via GP	16,9 (29,1)	32/81	47/66	113
Other	10,2 (18,9)	13/23	21/15	36
All	8,6 (20,0)	139/147	178/108	286

Conclusions. Admission via ambulance dialling “112” is encouraged as it seems to be the strongest determinant for early admission. The patient’s knowledge about the cause of symptoms significantly enhances early admission. Our study justifies initiation of stroke information campaigns.

RECOVERY OF CHRONIC VISUAL FIELD DEFECT AFTER COMPUTER-ASSISTED TRAINING: A CASE STUDY

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Background. Stroke is one of the most common causes of neurological disability in adults, causing deficits affecting motor, sensory and cognitive functions. Rehabilitation of motor function is clinical routine but there is a lack of efficient tools for sensory rehabilitation, even though it has been shown that e.g. visual field defects can be restored to some extent. It is also evident that factors such as onset of rehabilitation and localisation of the infarct or intracerebral haemorrhage in the brain affect the outcome. The present study further explores the clinical effects of computer-assisted visual field rehabilitation after chronic stroke.

Methods. Our participant is a 59-year-old male who suffered from a cardiogenic cerebral embolus in March 1998, which resulted in homonymous right upper quadrant visual fiel defect. Acute phase MRI showed multiple haemorrhagic infarctations in the area of left middle and posterior cerebral arteries. Training was conducted using a specially designed computer program, in which multiple stimulae are targeted to the borderline zone between intact and damaged visual fields. Total time of effective training was one hour at a time three times a week for a period of 3 months. Visual field was assessed by automated perimetry (Octopus 2000) using a standard general screening program. Perimetry was obtained before and after training as well as after a follow-up period of 3 months. Two perimetric measurements for both eyes were conducted at each time point.

Results. Mean values of two separate sessions for each eye were compared across the time points. In the right upper quadrant the proportion of the defect had diminished in the right eye from 73.5% pre-training to 55.9% post-training and continued to diminish to 42.6% by follow-up. Meanwhile, the proportion of intact vision had enlarged from 3.0% to 4.4% and subsequently to 8.8% whereas the proportion of relative deficit had enlarged from 23.5% to 39.7% and finally to 48.5%. The same tendency could be observed in the left eye.

Conclusions. This case study provides evidence that chronic visual field defects can be partially rehabilitated and that the resulted improvement can be maintained and even improved after the active training period – probably due to the spontaneous training by the participant after noticing a slight improvement. Absolute defect changing into relative defect accounted for most of the change and some of the relative defect changed into normal vision.

CONSTRAINT-INDUCED MOVEMENT THERAPY FOR THE AFFECTED UPPER EXTREMITY IN CHRONIC STROKE SUBJECTS

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Background. Recently a new rehabilitation approach based on relearning the use of a paralysed hand was described by Taub et al. (1993). The purpose of our study was to investigate the effectiveness of this constraint-induced movement therapy (CIMT) in improving motor abilities in chronic stroke subjects with persistent paralysis of the hand. In addition we aimed to assess if the possible changes remain after the treatment period.

Methods. We performed baseline, pre, post and control testing of the motor abilities of 12 participants of the rehabilitation program. Subjects were chronic stroke subjects (mean age 63 ±10 y, mean time since stroke 3,7 ±2 y) and 5 had left-sided and 7 had right-sided hemiparesis. They volunteered for a two-week long CIMT program. The intervention consisted of a large number of exercises for the affected hand performed under supervision for 7 hours a day for 10 days. Healthy arm was restrained in a sling. Structured upper limb motor test (Wolf-test) with 17 timed tasks was performed 2 weeks before the treatment (baseline), at the beginning, immediately after and 3 months after the CIMT program.

Results. All 12 participants completed 2 weeks of CIMT and were retested at 3 months. Mean sling time for the healthy hand was 79 h. No significant differences were found between the baseline and the beginning tests. Total time for timed tasks reduced significantly from the beginning to the after test (7 min vs. 3,9 min, $p < .001$). At 3 months total mean time was 3,8 min. Total scores for both the functionality and the quality of movement improved significantly from the beginning to the after the treatment. At 3 months functionality and quality scores were slightly improved from the after test scores.

Conclusions. CIMT provides a powerful treatment with lasting effects for well-motivated stroke subjects whose hand and arm function would otherwise remain unchanged.

THROMBOLYTIC THERAPY OF ISCHEMIC STROKE: A SWEDISH ONE-CENTER EXPERIENCE

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Background. After the publication of the National Institute of Neurological Disorders and Stroke (NINDS) trial in 1995, tissue plasminogen activator (tPA) was approved therapy of acute ischemic stroke in USA in 1996. Results of the use of tPA outside clinical trials have mainly been reported from USA, Canada and Germany, and published experiences from other countries are very limited. The aim of this study is to report the safety and outcome of intravenous tPA for acute ischemic stroke in a Swedish University Hospital.

Methods. Sixty consecutive patients, 75% males, with a mean age of 68 (range 35-88) years were treated with tPA outside clinical trials during 76 months, from November 1993 through August 1996, and from August 1997 through April 2001. Ischemic changes on the CT before treatment were graded according to the Alberta Stroke Programme Early CT Score (ASPECTS) criteria. Infarct size on the 24-48 hour CT was measured and hemorrhagic changes according to the European Cooperative Acute Stroke Study (ECASS) criteria and symptomatic hemorrhage according to NINDS criteria were registered. Scandinavian Stroke Scale (SSS) and/or National Institute of Health (NIH) Stroke Scales were registered before and 24 hours after treatment and mortality rates and modified Rankin Scale (MRS) score at 3 months was registered.

Results. Mean time from onset to treatment was 3.2 (range 1.3-6.1) hours, and mean “door to needle” time was 2.0 (range 0.8-5.3) hours. One third of the patients were treated with tPA 1.1 mg/kg and in 52%, treatment was started after 3 hours from symptom onset. Mean NIH and SSS score before treatment as compared to 24 hours after treatment improved significantly ($p<0.01$, respectively). At the 24-48 hour CT, 37 patients (63%) had a non-hemorrhagic infarction, 8 (13%) had a parenchymatous hemorrhage, 7 (12%) had a hemorrhagic infarction and 7 (12%) had no ischemic or hemorrhagic changes. Mean infarct size was 74 (range 0-487) mL. Patients with hemorrhagic changes had larger infarcts than patients without such changes ($p<001$). Mean final infarct size was larger among 8 patients (13%) with an initial ASPETCS score <=7 (mean: 212, range: 25-487 mL) as compared to patients with a score above 7 (mean: 50, range: 0-391 mL), ($p<0.001$). Three patients (5%) had a symptomatic hemorrhage, one requiring surgery, and one patient (2%) had a fatal hemorrhage. At 3 months, 23% of the patients were graded as MRS 0-1, 45% were graded as MRS 0-2, and 18% were dead.

Conclusions. The data on safety and efficacy of the use of tPA in ischemic stroke at our institution is comparable to the results of the NINDS trial and published phase 4 trials adhering to the NINDS protocol. These results were accomplished despite 1/3 of our patients having a higher dose of tPA than was used in the NINDS trial and a treatment start more than 3 hours after symptom onset in more than half of the patients. The ASPECTS score seems to be useful for predicting the development of a large infarct exceeding 1/3 of the MCA territory.

COGNITIVE AND MOTOR PERFORMANCE RELATED TO THE SIZE OF THE INFARCT IN A RAT EMBOLIC STROKE MODEL

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Background. The present study was designed to evaluate cognitive and motor perforin rats after unilateral middle cerebral artery occlusion (MCAO). Rats were trained to obtain solid baseline performance in Montoyas' Staircase Test (ST), which is sensitive to neglect and fine motor behavior, and in Delayed Alternation (DA) using a T-Maze, which is sensitive to gross motor performance, neglect, memory and learning. Studies using a beamwalk test have shown that gross motor performance recovered 2 weeks after MCAO in rats.

Methods. Animals were trained in DA measuring the total time of 20 daily runs, until a criteria of at least 19 correct daily runs in 3 consecutive days was reached. After reaching this DA criteria rats were embolised or sham operated. From day 21 to 28 after the operation all animals were re-tested. The 28^{th} day after surgery brains were fixed and infarct volumes evaluated microscopically.

Results and conclusions. In the present study we found no difference between sham operated and embolized rats in DA gross motor performance. However, embolized rats declined 54 % in ST fine motor performance ($p = 0.001$, Mann-Whitney) and made 134 % more DA errors ($p = 0.007$, Mann-Whitney) than the sham operated animals. When examining all embolized animals infarct volume significantly correlated inversely with ST fine motor performance ($p = 0.0005$, $R = -0.65$, Spearman). Thus, 1 month after embolisation rats showed marked deficits in cognitive abilities and fine motor behavior, but not in gross motor behavior. Furthermore, fine motor performance, but not cognitive abilities, decreased significantly as infarct volume increased.

ANATOMICAL TARGETS OF THE RECOVERY PROCESS

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Damage to the adult brain results in degenerative effects in regions that are connected to the site of principal injury and, at least in some systems, an apparently adaptive neuronal plasticity and restructuring of connectivity in response to this degeneration. Our research on the effects of focal damage to the sensorimotor cortex in adult rats has indicated that lesion-induced neural restructuring varies dependent upon post-injury behavioral changes, including compensatory behavioral changes that animals develop spontaneously and those induced via rehabilitative training. Following unilateral damage to the sensorimotor cortex, a connected region of cortex contralateral and homotopic to the damage undergoes time-dependent and behaviorally-linked neuronal and glial changes including dendritic growth, synaptogenesis and reactive changes in astrocytes. Although the neuronal growth and synaptogenesis are dependent upon post-injury behavioral experiences (e.g., compensatory changes in the use of the forelimb ipsilateral to the lesion) behavioral manipulations alone are not sufficient to reproduce the effects found after the lesions. Experiments which independently manipulate denervation of a neocortical pathway and post-injury behavioral change indicate that partial denervation may cause affected cortical regions to become exceptionally responsive to relevant behavioral changes. These effects are likely to be linked to denervation-triggered neural growth-promoting processes, which may create a fertile milieu for behavioral change. These and related findings suggest that mild axonal degeneration in the neocortex might enable a plastic response to learning which is, perhaps, not normally possible in adult animals. Additional studies indicate that this facilitation of neuronal growth can be capitalized upon using rehabilitative training to promote adaptive dendritic and synaptic restructuring and to enhance behavioral function.

FUNCTIONAL REORGANIZATION OF MOTOR CORTEX FOLLOWING STROKE: THE EFFECTS OF DIFFERENTIAL MOTOR TRAINING

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Background. More than 80% of all ischemic attacks result in motor impairments. Any improvements in motor performance are typically observed within the first 3 months following the insult. Functional compensation by intact brain regions is believed to mediate this recovery. Further, post-stroke rehabilitative training may both promote neural compensation and enhance functional recovery. To test this hypothesis, we used intracortical microstimulation to examine the topography of movement representations within the rat forelimb motor cortex before and after focal ischemic infarct. Further, we examined how skilled versus unskilled rehabilitative training influenced both behavioral recovery and the functional organization of the motor cortex.

Methods. Adult male rats were trained for two weeks on a skilled forelimb reaching task. Immediately following training, intracortical microstimulation was used to derive detailed maps of forelimb movement representations within the motor cortex contralateral to the trained paw. A focal ischemic infarct was then produced via electrocoagulation of surface vasculature within approximately 35% of the wrist representation. The animals were allowed to recover and then placed in one of three rehabilitative training conditions. Animals in the Skilled ReHab Condition (SHRC) were given two weeks of skilled reach training while rats in the Unskilled ReHab Condition (URHC) were given a similar amount of training on a reaching task that did not promote skilled forelimb movements. NonReHab Condition (NRHC) animals received no motor training. Following rehabilitation, all animals received a probe trial on the skilled reaching task to assess motor performance before intracortical microstimulation was again used to examine the topography of movement representations within motor cortex. Finally, unbiased stereological techniques were used to measure changes in neuron structure within perinfarct regions of the cortex from all animals.

Results. All animals exhibited profound reaching impairments immediately following ischemic infarct. Following the rehabilitation period, SRHC animals recovered to near pre-stroke levels while URHC and NRHC animals did not. In comparison to SRHC animals, NRHC rats exhibited a loss of distal and proximal movement representations within perinfarct cortical regions. URHC animals showed a significant loss of distal but not proximal representations. Despite the loss of cortical function within the URHC and NRHC animals, there were no significant differences in neuron density within peri-infarct regions of cortex between the three conditions.

Conclusions. The results show that there is a loss of cortical function that extends well beyond the infarcted region of cortex into intact cortical regions. This loss can be prevented through rehabilitative training and is correlated with improved motor performance. Further, the nature of the neural and behavioral compensation following rehabilitation is dependent upon the nature of the motor training experience.

CURRENT STATUS OF PRECLINICAL RESTORATIVE DRUG DEVELOPMENT

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Neurological recovery to some degree is the rule rather than the exception after stroke. The precise mechanisms of such recovery remain unclear, but may depend on neural reorganization and generation of new neurons from uninjured parts of brain. In recent studies, we have investigated new molecular and cellular treatments that might enhance recovery after experimental stroke. In particular, polypeptide growth factors promote neural sprouting as well as precursor cell proliferation in brain. We found that the intracisternal administration of at least two of these, basic fibroblast growth factor (bFGF) and osteogenic protein-1 (OP-1, BMP-7) enhance sensorimotor recovery of the impaired (contralateral) limbs following unilateral stroke in rats. The effective time window of treatment was as long as 1-3 days after stroke onset. At these time points, growth factor treatment did not reduce infarct volume. In other recent studies, we found that the exogenous administration of immortalized mouse neural stem cells or human umbilical cord blood cells also enhanced sensorimotor recovery after stroke. The combination of growth factors and stem cells may be superior to either treatment alone. The success of stroke recovery-promoting treatments in the clinic will depend on the thoughtful design of clinical trials that can identify and discriminate meaningful recovery in fundamentally distinct functional spheres (e.g., sensorimotor and cognitive function) after stroke.

Abstracts of Poster Presentations

C1

CASE ASCERTAINMENT IN STROKE INCIDENCE STUDIES: EXPERIENCES FROM TWO STROKE REGISTERS IN ÖREBRO

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Background. Data from two different stroke registers were compared in order to assess the completeness of each of them. The effect of selection bias on patient characteristics was also analyzed.

Methods. In Örebro, two different stroke registers were active at the same time; a local population-based stroke register (LSR), and a national hospital-based quality register, also called "Riks-Stroke" (RSR). Data from the registers were compared in three ways: (1) all LSR patients (LSR-A), (2) hospitalized LSR patients only (LSR-H), and (3) RSR patients. In order to attain comparable groups, cases with subarachnoidal hemorrhage, TIA, and recurrent strokes were excluded. Data collection was prospective and took place from February 1st 1999 through January 31st 2000. By the way of linking and matching computer files, the completeness of case ascertainment was evaluated. The effect of selection bias on patient characteristics was analyzed with information emanating from the LSR.

Results. 377 first-ever patients were ascertained in the LSR-A group, 347 in the LSR-H group (92% of LSR-A), and 220 in the RSR group (63% of LSR-H). No patients were missed in the LSR compared to the RSR. Relatively fewer lacunar strokes and more stroke of the undetermined type were found in the LSR-A compared to the LSR-H and RSR groups. Stroke severity (according to the NIH Stroke Scale) and 28-day case fatality was higher in the LSR-A group compared to the LSR-H and RSR groups. Also, patients in the LSR-A group less frequently underwent CT and treatment at acute stroke units and rehabilitation units.

Conclusions. Sometimes hospital-based case ascertainment is used in stroke incidence studies. Even with as high admission rate as in our study (92%), resulting data get skewed when out-patients are not included. This stresses the importance of complete community-based case ascertainment in stroke incidence studies. The purpose of a national quality register such as "Riks-Stroke" is to assure the quality of care, and to improve it by providing feedback. It is an advantage to have knowledge about selection bias when interpreting data to the health care system as a whole.

C1 to C25, B1 to B19 and M1 to M2 refer to the board number of posters at the Chamber Music Hall

ON-LINE CEREBRAL EMBOLUS DETECTION WITH ARTEFACT REJECTION

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Introduction. Cerebral embolus monitoring systems suitable for routine clinical use must have the ability to automatically recognise and differentiate between artefacts and emboli. This has to date proven to be an extremely difficult problem to solve.

Methods. In this study we present a new advancement with regard to the automatic recognition of cerebral emboli and differentiation from artefacts based on a binary decision tree which includes a completely new parameter. This is the ¼ Doppler shift for the maximum power reflection of an embolic event at 2.5 MHz insonation frequency compared to 2.0 MHz. A new multifrequency transcranial Doppler system together with this software was used in this study of 2000 artefacts and 300 embolic events in five heart valve patients. The level of energy increase for event recognition was set at a 5db increase above background Doppler power which lasted for more than 4 msec (sensitivity > 20 dBmsec). The artefacts in 2 healthy controls consisted of 200: tapping the probe, 200: tapping the skull, 200: talking (counting), 200: swallowing, 400: coughing, 200: wrinkling the forehead, 200: clenching the teeth, 400: movement of the skin near the probe.

Results. One thousand nine hundred and ninety-six (99.8%) of the 2000 artefacts (artefact specificity = 99.8%) and 295 (98.3%)of the 300 embolic events (embolus specificity = 98.3%) were correctly classified.

Conclusion. This study suggests that a binary decision tree including ¼ Doppler shift assessment at 2.0 and 2.5 MHz insonation frequencies will greatly improve the ability to carry out automatic differentiation between artefacts and cerebral emboli during monitoring in routine clinical situations.

C3

EVIDENCE-BASED NURSING TO STROKE PATIENTS APPLICATION OF A EVIDENCE-BASED CLINICAL GUIDELINE CONCERNING THE BASIC NEEDS: NUTRITION

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Background. The aim of the project was to evidence-base nursing care of stroke patients and to monitor the quality of nursing practice. The monitoring process is related to basic human needs, as developed and described by Virginia Henderson.
The aim of nursing concerning the basic need nutrition is: preventing weight loss in the acute phase, preventing pneumonia caused by aspiration and keeping the patient's Body Mass Index unchanged or improved by discharge.

Methods. The clinical guideline

Problem. Research show that 50% of stroke patients are malnourished during hospital stay. Dysphagia is the most important factor. About 50% of stroke patients have dysphagia initially, most of the patients get rid of the symptom within a few weeks, but about 10%, mainly patients with brainstem lesions, may have persistent problems.

Recommendations for good clinical practice.
Identifying the risk patient:
Assessment according to screening tool
- Drinking test (B)
- Nutritional state (C)

Nutritional therapy for the risk patient:
- Calculating the patient's need for energy, protein and fluid (C)

Choosing diet:
- Adjusting food consistency in relation to dysphagia (C)
- nasogastric tube feeding within 72 hrs by transient dysphagia
- percutaneous endoscopic gastrostomy tube feeding within 2-4 weeks by expected Persistent dysphagia (B)

Evaluating the patient's need for specific training

Monitoring. Nursing sensitive quality indicators are registered with acute stroke patients in two areas. Study population approximately 1500 stroke patients.
Quality indicators:
- Daily assessment of energy, protein and fluid
- Weekly assessment of body weight
- Eating ability according to Barthel Index or other validated instrument
- Body Mass Index at admission, after 3 weeks, and at discharge
- Events of pneumonia treated with antibiotics during hospitalisation

Results. The clinical guidelines have been implemented in stroke units at Bispebjerg University Hospital, Århus University Hospital and Esbønderup Hospital

SURVIVAL AND RECURRENT STROKES IN PATIENTS WITH DIFFERENT SUBTYPES OF STROKE: A FOURTEEN-YEAR FOLLOW-UP STUDY

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Background. The purpose of this study was to determine the long-term outcome in different subgroups of patients with stroke and predictors of recurrent stroke and death in patients admitted to the Stroke Unit, Department of Neurology, Linköping in 1986.

Methods. This study is a report of the long-term follow-up of 339 out-of 411 patients hospitalised at the Stroke Unit. The first follow-up was in January 1994, the second in 1999 and thereafter the patients have been followed up continuously (the last follow-up was in Jan.2001). This study was a continuation of a previous study with survival analysis until March 1990.

Results. There were 339 patients, 154 men and 185 women with a median age of 74 years (range 23-97). There were 30 patients (8.8%) with intracerebral hemorrhage (ICH), 71 (20.9%) with cardioembolic cerebral infarction (CE), 47 (13.9%) with lacunar infarct (LI) and 191 (56.3%) with atherosclerotic cerebral infarction (ACI). The cumulative probabilities of recurrent stroke rates at one; 5 and 10-year follow-ups were 13.5% with 95% confidence interval (CI), (9.6-17.4); 38.7% (32.6- 44.8) and 53.9% (46.7-61.1). According to the Cox proportional hazard regression analysis each of age, severity of stroke, previous stroke, and systolic blood pressure are of importance in predicting recurrent stroke. 290 patients (85.5%) died during the observation period. Fifty five patients (19%) died of index stroke, 76 (26.2%) of recurrent stroke, 107 (36.9%) of cardiovascular/peripheral vascular disease and 52 (17.9%) of other causes. Fatal index/recurrent stroke occurred statistically more frequently in the cardioembolic infarction group vs non-cardioembolic ($p=0.005$). Thirty nine percent of all vascular deaths after the first year were caused by recurrent strokes. Patients with LI had lower mortality rates vs ICH by log rank tests ($p= 0.0275$); vs CE ($p=0.000$) and vs ACI ($p=0.049$). Cox proportional hazard regression analysis indicated that age, severity of stroke, previous stroke, heart failure, fasting blood glucose (> 6 mmol /L) or history of diabetes were each predictors of mortality.

Conclusions. This study has shown a worse outcome for all subtypes of stroke compared to the normal population and also clearly pointed out independent predictors of recurrent stroke or death at the time of diagnosis. Cardiovascular death is common and the risk of recurrent (fatal) stroke is still high after 10 years follow up.

C5

THE IMPACT OF INFECTIONS AND HYPERTHERMIA OF UNKNOWN ORIGIN ON IN-HOSPITAL MORTALITY

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Background. Clinical as well as experimental data suggest that body temperature has a strong impact on the prognosis and on the size of the brain lesion. Clinical trials with moderate (33°C) as well as mild (35°C) hypothermia have been started. However, recent reports indicate that infections contribute to the complication rate in moderate hypothermia. Some of these infections may be prevalent already on admission to the hospital. The present study is undertaken to elucidate the impact of infectious disorders as well as hyperthermia of unknown origin on in-hospital mortality in patients with ischemic as well as hemorrhagic stroke (IS and HS respectively).

Methods. All patients with a diagnosis of acute stroke (ICD 10, I60-9) and admitted within a week to the University Hospital in Uppsala during 1998, were studied retrospectively. Medical records of these patients were collected and data analysed using the StatView data analysing system. Hyperthermia was defined as a body temperature > 37.5°C. The validity of continuous data with a normal distribution was tested by the unpaired t-test, and for non-parametric data with the Mann-Whitney U test. The relationship between different groups of nominal variables, was tested by arranging the data in contigency tables and using the chi squared test. Multiple logistic regression model was fitted for in-hospital mortality. Explanatory variables included in the model were those with a correlation coefficient ≤ -0.1 or ≥ 0.1.

Results. The screening identified 589 patients with an acute stroke. 517 patients (88%) had an IS and 72 patients (12%) had a HS. There was no significant difference in the distribution of age, gender, motor deficit, blood pressure, blood glucose, body temperature and CRP on admission between the two groups. 276 patients (47%) developed hyperthermia during the hospital stay, 167 patients (28%) were diagnosed with infections. Community based infections were more common than those acquired at the hospital ($p<0.0001$). Hyperthermia of unknown origin was more common in patients with HS than in patients with IS (36% vs. 16%, $p<0.0001$). In the multiple logistic regression model, in-hospital mortality was significantly correlated to mean body temperature during hospital stay, but not to the admission body temperature or stroke type (IS/HS).

Conclusions. Hyperthermia is found in nearly 50% of patients with an acute stroke and contributes to in-hospital mortality. Infections are identified in approximately 50% of these patients. The impact of infections is strongest in patients with ischemic stroke. We propose that infections should be identified and treated as early as possible after the arrival to the hospital.

IN-HOSPITAL MORTALITY IS RELATED TO MEAN BLOOD GLUCOSE CONSENTRATIONS AFTER ACUTE STROKE

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Background. Type 2 diabetes is a major risk factor for stroke and most diabetic patients die in cardiovascular diseases. Epidemiological data strongly indicate that patients with more severe hyperglycemia, measured as HbA1c, more often develop stroke. A recently performed intervention trial in diabetic patients with acute myocardial infarction (DIGAMI) demonstrates a significantly lower 1 year mortality with intensive insulin treatment. The aim of the present investigation was to study the impact of blood glucose, both on admission and the following days, on in-hospital mortality among diabetic and non-diabetic patients with acute stroke.

Methods. All patients with a diagnosis of acute stroke (ICD 10, I60-9) and admitted within a week to the University Hospital in Uppsala during 1998, were studied retrospectively. Medical records of these patients were collected and data analysed using the StatView data analysing system. The validity of continuous data with a normal distribution was tested by the unpaired t-test, and for non-parametric data with the Mann-Whitney U test. The relationship between different groups of nominal variables was tested by arranging the data in contigency tables and using the chi squared test. Multiple logistic regression model was fitted for in-hospital mortality. Explanatory variables included in the model were those with a correlation coefficient ≤ -0.1 or ≥ 0.1.

Results. The screening identified 589 patients with an acute stroke. 117 patients (20%) had diabetes, 6 of them were diagnosed during their hospital stay. 95% were type 2 diabetics (111/117). There was no significant difference in the distribution of age, gender, stroke severity and blood pressure between diabetics and non-diabetics. Diabetics showed a tendency to have more often an ischemic than hemorrhagic stroke compared with non-diabetics ($p=0.09$). Diabetics had 50% higher blood glucose, both on admission and during the following days, than non-diabetics ($p<0.0001$). In-hospital mortality among diabetics was 15% as compared to 11% among non-diabetics ($p=0.22$). In the multiple logistic regression model, in-hospital mortality was significantly correlated to mean blood glucose, admission blood glucose and diabetes.

Conclusions. We demonstrate a strong correlation between blood glucose levels after acute stroke and in-hospital mortality. These results indicate a possibility to improve the prognosis of stroke patients, especially those with diabetes, by lowering blood glucose values.

C7

POST STROKE FATIGUE, A TWO-YEAR FOLLOW-UP OF STROKE PATIENTS IN SWEDEN

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Background. Fatigue is common among stroke patients. This study determines the frequency of fatigue among long term survivors after stroke and what impact fatigue has on the various aspects of daily life.

Methods. This study is based on Riks-Stroke, a hospital-based national register for quality assessment of acute stroke events in Sweden. The register also includes a three-month follow-up. During the first six months of 1997, 8194 patients were registered in Riks-Stroke and 5104 were still alive two years after the stroke. They were traced and followed up by a postal questionnaire which 4023 (79%) responded to. Fatigue is often a part of a depression and therefore the 153 patients that reported that they always felt depressed were excluded from further analyses. Another 218 patients had not answered the question “Do you feel tired” and were also excluded, leaving 3667 patients.

Results. On the question “Do you feel tired?” 366 (10.0%) patients answered that they always felt tired and an additional 1070 (29.2 %) were often tired. Men and women reported fatigue in the same extent. Patients that always felt tired were in average older than the rest of the study population (74.5 vs. 71.5 years, p-value <0.001) and therefore all subsequent analyses were adjusted for age. Among patients that before the stroke were living at home, a greater proportion of patients with fatigue had an institutional living two years after the stroke (OR 2.2, 95% CI 1.63-2.96). Patients that before the stroke were independent in their primary ADL functions and fully conscious at arrival at the hospital were more often dependent after the stroke if they were always tired (OR 3.84, 95% CI 2.88-5.13). These differences were valid also after adjustment for differences in background variables. Patients with fatigue also had more speech problems and they estimated their general health as lower.

Conclusions. Fatigue is frequent, and often severe even late after stroke. It is associated with profound deterioration in several aspects of everyday life, but it usually receives little attention by health care professionals. Intervention studies are needed.

FACTOR V LEIDEN AND PROTHROMBIN GENE MUTATIONS MAY PREDISPOSE TO PARADOXICAL EMBOLISM IN SUBJECTS HAVING A PATENT FORAMEN OVALE

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Backround. In patients with cryptogenic brain infarction, patent foramen ovale has been shown to be a common finding, but its clinical significance as a risk factor for cerebral ischemia is still controversial. The aim of this case-control study was to find out whether prothombotic states and patent foramen ovale co-exist in young patients with cryptogenic brain infarction.

Methods. Coagulation abnormalities in 57 patients with cryptogenic brain infarction and patent foramen ovale and in 104 community-based controls matched by age and sex were determined. We also searched for predisposing factors and triggering events. Stepwise logistic regression was used to test differences in the prevalence of variables between cases and controls.

Results. Coagulation abnormalities were more common in the patients than controls (p=0.012) and multivariate analyses showed that particularly the presence of factor V Leiden and prothrombin G20210A mutation significantly associated with cryptogenic brain infarction (OR, 12.07; 95% CI, 1.31 to 111.2; p=0.028), whereas other coagulation abnormalities did not. Valsalva manoueuvre at stroke onset (p=0.002) and predisposing events such as surgery, traumatic injury, bed rest in hospital and long-distance travel by car etc. associated (p=0.041) with coagulation abnormalities in our patients.

Conclusions. Factor V Leiden and protrombin G20210A mutations were frequent findings in patients with cryptogenic brain infarction and patent foramen ovale. The high frequency of underlying risk factors that predispose to venous thrombosis in our patients supports the concept of paradoxical embolism through patent foramen as an important mechanism behind ischemic stroke of unknown origin.

C9

CARDIORESPIRATORY FITNESS AND THE RISK OF STROKE

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Background. Low cardiorespiratory fitness is considered to be one of the major public health problems. Our aim was to examine the relationship of cardiorespiratory fitness, as indicated by maximal oxygen uptake with subsequent incidence of cerebrovascular stroke. We also compared maximal oxygen uptake with conventional risk factors as a predictor for future strokes.

Methods. Population-based cohort study with an average follow-up of 10.3 years from Kuopio and surrounding communities of eastern Finland. 2312 men with no stroke at baseline participated in the study. Among these men 112 strokes occurred, of which 92 were ischaemic strokes. Maximal oxygen uptake was measured directly during exercise testing.

Results. The relative risk of any stroke in unfit men (maximal oxygen uptake <25.2mL/kg/min) was 2.5 (1.4 - 4.4, p=0.003, p =0.002 for the trend across the quartiles) and the relative risk for ischaemic stroke 2.4 (1.2 - 4.5, p=0.01, p =0.005 for trend across the quartiles) as compared with fit men (maximal oxygen uptake >35.2mL/kg/min) after adjusting for age and examination year. A further adjustment for smoking, alcohol, systolic blood pressure and serum LDL cholesterol did not substantially weaken the association for either any or ischemic strokes. Low cardiorespiratory fitness was comparable with systolic blood pressure, obesity, alcohol consumption, smoking and serum LDL cholesterol as a risk factor for stroke. An adjustment for prevalent coronary heart disease at baseline did not weaken the association significantly.

Conclusions. Our findings show that low cardiorespiratory fitness was associated with an increased risk of any and ischaemic strokes. Maximal oxygen uptake is one of the strongest predictors of stroke comparable to other modifiable risk factors.

SYSTOLIC BLOOD PRESSURE RESPONSE TO EXERCISE STRESS TEST AND THE RISK OF STROKE

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Background. Systolic blood pressure (SBP) during exercise has been found to predict a future diagnosis of hypertension, coronary heart disease (CHD) and cardiovascular disease (CVD) death. However, there are no studies showing a relationship between SBP during exercise test and stroke. The aim of the present study was to study the associations between SBP rise during exercise, percent maximum SBP at two minutes post-exercise and the risk of stroke in population-based sample.

Methods. SBP was measured every two minutes during and after exercise test. The subjects were a population-based sample of 1026 men without clinical CHD or antihypertensive medications and prior stroke at baseline. During an average follow-up of 10.4 years, there were 46 cases of all and 38 ischemic strokes.

Results. Men with SBP rise more than 19.7 mmHg per minute of exercise duration had a 2.3 fold risk of any stroke and a 2.3 fold risk of ischemic strokes, when compared with men whose SBP rise was less than 16.1 mmHg per minute. Similarly percent maximum SBP at two minutes post-exercise (SBP at two minutes recovery divided by maximal SBP) was associated (highest tertile) with a 4.6 fold risk of any stroke and a 5.2 fold risk of ischemic strokes.

Conclusions. SBP rise during exercise and percent maximum SBP at two minutes post-exercise were directly and independently associated with the risk of all strokes and ischemic strokes. Exercise SBP testing may be recommended for as an additional tool in the prediction of future stroke.

C11

”THE GAIT TRAINER” FOR REHABILITATION OF GAIT AND POSTURE AFTER STROKE

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Background. Walking is a complicated task where controlled cyclic movements of different parts of the body are a major challenge to the central nervous system. Restoration of normal gait pattern after stroke or brain trauma is difficult if the patient cannot support his / her weight on the paralyzed lower limb. Paralyzed patients are well motivated to relearn walking because of its impact on the quality of life.

Methods. A new “Gait Trainer” was tested with three severely hemiparetic chronic stroke patients. They trained 9–12 sessions, 10 to 20 minutes each, on the Gait Trainer. A mechanized gait trainer produces gait-like movement. The movement of two foot plates simulates the stance and the swing phases in a nearly physiological manner. The foot is lifted during the swing phase and the natural ratio of 60% / 40% between the two phases is achieved. The velocity is adjusted by the frequency inverter from 0–80 steps / min (0–2km/h), and the step length is 400mm. The Gait Trainer also controls the swing of the center of the mass (-1cm to +1cm) in vertical direction with a rope attached to a patients hip and in horizontal direction through the weight support harness. The harness is easy to put on a patient and it enables stepless body weight reduction from 0% to 100%. The walking ability of the patients were measured before and after the treatment by a gait analyzing system (the GaitRite).

Results. Each patient performed 8000–9000 steps altogether and the walking speed was at the beginning 1km/h and was increased up to 1.5km/h at the end of the treatment. Walking abilities of each of the patients improved after the training period with the Gait Trainer.

Conclusions. The unique characteristics of the Gait Trainer, such as stepless velocity control, adjustable weight support, gait-like walking with swing and stance phases and control of the center of the mass in vertical and horizontal directions allows various rehabilitation paradigms. The Gait Trainer enables severely disabled patients to practice gait-like walking with an assistant of only one therapist.

COMPARISON OF THE SIS-16 VS. THE BARTHEL INDEX - A SECONDARY ANALYSIS OF THE GAIN AMERICAS STUDY

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Objective. The "SIS-16" is a 16-item subscale derived from the 4 physical domains of the Stroke Impact Scale (SIS) using a psychometric technique for item reduction known as Rasch analysis. This study examined performance of the SIS-16 compared to the commonly used disability measure, the Barthel Index (BI).

Method. The 64-item SIS was added as a secondary outcome measure in the GAIN Americas (Glycine Antagonist in Neuroprotection) randomized stroke trial after that trial was underway. Of 931 study subjects eligible for SIS evaluation, 801 were alive at 3-months post-stroke and were administered the BI. Thirty-seven of these subjects (4.6%) were either too ill, aphasic, did not speak English, or had telephone follow-up at Month 3, and so did not complete the SIS questionnaire. The remaining 764 subjects completed both the SIS and the BI, and constituted this study sample. The SIS-16 includes 7 ADL/IADL items, 8 Mobility items, and 1 Hand Function item and has a total score ranging from 0 to 100. The distribution of the scores from the SIS-16 measured at 3 months post stroke was compared to that of the BI. The SIS-16 scores were further evaluated in patients who achieved BI scores of 95 or greater, a level generally regarded as functionally independent. Descriptive statistics were used for the analysis.

Results. The study sample had an average age 68$\pm$12.6 years and 55% men. Stroke severity was classified as minor in 119 patients (NIHSS 2-5), moderate in 365 (NIHSS 6-13), and major in 280 patients (NIHSS >= 14). Distributions of the 3-month BI and SIS-16 scores stratified by baseline stroke severity are shown below.

Baseline			3-month BI			3-month SIS-16	
NIHSS	n	median	% floor (BI=0)	% ceiling (BI=100)	median	% floor (SIS-16=0)	% ceiling (SIS-16=100)
2 – 5	119	100	0	55	8	< 1	11
6 – 13	365	95	2	43	70	1	6
>= 14	280	60	11	24	45	6	5

By 3 months after stroke, 369 patients (48%) had achieved BI of 95 or greater. These patients had a SIS-16 median score of 87.5 (range: 37.5 to 100). The 25th and 75th percentiles were 75 and 95.3, respectively.

Conclusion. Compared to the Barthel Index, the SIS-16 has an excellent distribution with a very low ceiling effect for minor stroke patients and a very low floor effect for patients with major strokes. The SIS-16 may be better able to capture residual stroke disability than the BI.

C13

EVIDENCE-BASED NURSING TO STROKE PATIENTS APPLICATION OF AN EVIDENCE-BASED CLINICAL GUIDELINE CONCERNING THE BASIC NEED: ELIMINATION OF FAECES

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Background. The aim of the project was to evidence-base nursing care of stroke patients and to monitor the quality of nursing practice. The monitoring process is related to basic human needs, as developed and described by Virginia Henderson. The aim of nursing concerning the basic need: elimination of faeces, is establishing the pattern of bowel movements prior to the stroke, maximum score in bowel control and individually adjusted faeces incontinence aid.

Methods. The clinical guideline

Problem. Constipation is often connected with stroke. There is, however, no current convincing evidence for that connection. It stated, that constipation and faecal incontinence in neurological patients may be caused by impaired perception of the urge to empty the bowels. Faecal incontinence is an indicator of stroke severity. The prevalence in a stroke population is uncertain.

Recommendations for good clinical practice.
Identifying the risk patient:
Assessment according to screening tool
- Examining faeces present in rectum by rectal exploration
Constipation/impaction (normalizing):
Evacuation of faeces
- Suppositories bisacodyl/- mineral oil retention enema/phosphate enema
Constipation (prevention):
- Scheduled defecation reflecting patients' previous pattern (A)
- Dietary fibre supplements (5-10 g daily)/ - fluid (30-40 cc/kg. body weight/24 hrs)
- Glycerine suppositories stimulating the patients' previous pattern of defecation (C)
- Magnesium sulphate or citrate
- Training the coordination of pelvis floor muscles (A)
Faecal incontinence:
- Incontinence aid and skin care (C)t
- Training sphincter muscles (C)

Monitoring. Nursing sensitive quality indicators are registered with acute stroke patients in two areas. Study population approximately 1500 stroke patients.
Quality indicators:
- Daily assessment of bowel pattern
- Bowel control according to Barthel Index or other validated instrument
- Constipation treated with mineral oil enema during hospitalisation

Results. The clinical guidelines have been implemented in stroke units at Bispebjerg University Hospital., Århus University Hospital and Esbønderup Hospital

THE PUBLIC`S KNOWLEDGE OF BLOOD PRESSURE AND RISK OF STROKE IN DENMARK

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Background. Hypertension is responsible of near to 50% of all strokes. Data from North-America shows, however, that blood pressure is reduced to normal (140/90 mm Hg) in only ¼ of hypertensive persons. Information about the Dane's knowledge of blood pressure as a major cause of stroke is largely lacking. Prior to a stroke awareness campaign the Danish Stroke and Aphasia Association undertook a telephone interview of 1001 persons representative of the Danish population.

Methods. In April 2001 an established opinion-research institute (Vilstrup Research) interviewed by telephone a random sample of 1780 adults (>15 years of age) about their knowledge of blood pressure. Of these 590 refused to participate and 189 were not at home. Hence, 1001 persons were interviewed.
The following questions were asked: Do you know your blood pressure? Those who answered yes were asked: What is the level of your blood pressure (too high, too low, normal)? Are you on antihypertensive medication? When was the last time your blood pressure was measured? Where did you have your last BP measurement (general practitioner, hospital, self measurement)? What are the symptoms of high blood pressure? What is the risk of high BP? What is a stroke? What causes stroke? The answers were related to age, and sex.

Results. Thirty-nine percent had knowledge of their BP; 31% of the males and 46% of the females; 21% in age group 15-29 years and 57% in age group > 60 years.

- Eleven percent of the 1001 persons received antihypertensive medication; age group 15-44 years 1%, age group 45-59 years 14% and age group > 60 years 29%.
- Thirty percent of the persons who knew that their BP was too high did not receive antihypertensive treatment.
- Fifty-one percent had their blood pressure measured within the last year; 42% in age group 15-44 years and 71% in age group > 60 years. In 18% BP was measured within the last 3 years and in 22% BP was measured more than 3 years ago. Five percent had never had a BP measurement and 4% did not know.
- BP was measured by the general practitioner in 59%, in hospital in 23%. Only 4% measured BP by themselves.
- Sixty-eight percent did not know what a stroke is.
- Forty-seven percent had no knowledge of the risk of hypertension. Thirteen percent knew that high blood pressure might cause cerebral hemorrhage, 24% that it may cause occlusion of cerebral vessels.

Conclusions. The finding that 30% of those known to have high blood pressure were untreated taken together with North-American experience indicate that at least half of the Danes with hypertension are untreated. Many strokes could be prevented by increasing the public's awareness of hypertension as an important risk factor for stroke.

C15

THROMBOLYTIC THERAPY IN ACUTE STROKE: HOW BIG IS THE TARGET POPULATION AND WHO BENEFIT?

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Background. Thrombolysis is approved for the treatment of acute ischemic stroke in North America based on one randomized clinical trial. Two European trials failed to demonstrate benefit of thrombolysis. A Cochrane metaanalysis analyzing all available randomized studies showed, that the combined risk of death and poor functional outcome is reduced significantly in patients receiving thrombolysis within 3 hours. On the other hand, thrombolysis significantly increases the risk of dying as well as the risk of changing an infarct into a hemorrhage. Furthermore, only a minority (2% to 6%) of the stroke population is eligible for treatment.
We investigated how large a fraction of Danish stroke patients would be eligible and benefit from thrombolysis by applying the selection criteria used in North America.

Methods. The population-based retrospective study included 502 unselected patients with acute stroke admitted through a period of eight months 1999/2000 to Gentofte University Hospital. This hospital is the only hospital in an uptake area of about 300 000 inhabitants admitting acute stroke patients. The most important exclusion criteria from the North American stroke trial with rt-PA (recombinant tissue plasminogen activator) for acute ischemic stroke were applied on the Danish cohort. The number of patients who might benefit from thrombolytic therapy was estimated from the North American trial, which reported a 32% relative increase of patients who would have full recovery.

Results. 357 patients (71%) were excluded because treatment could not be instituted within 3 hours; 46 patients (9%) were excluded because the stroke was too mild (Scandinavian Stroke Scale Score > 50). Thirty-nine (8%) would be eligible for thrombolytic therapy. Of these 13 patients (3%) would die irrespective of treatment and 11 (2%) would have experienced full recovery spontaneously. Only five of the patients (1%) would benefit from thrombolytic therapy. In the ideal situation - all patients were admitted in due time – 24 (5%) would have benefited from thrombolytic therapy.

Conclusions. The beneficial effect of thrombolysis is well documented in Cochrane analyses. The risk associated with thrombolytic therapy in acute stroke is, however, well documented as well. Introduction of thrombolytic therapy requires extensive re-organization of acute stroke treatment towards highly specialized and costly acute stroke centers. As documented in this study the number of stroke patients who may benefit from thrombolysis is, however, very small. The target population was 8% and only 1% could be expected to benefit. Practically speaking, therefore, thrombolysis cannot be expected to influence functional prognosis of the stroke population.

Rehabilitation and treatment in stroke units has proven to benefit all stroke patients. Seen in the light of this study establishment of stroke units should still be of the highest priority the more so as the need of stroke units is still not met. Establishment of thrombolysis in the treatment of stroke should still have lower priority.

CASE-FATALITY IN A STROKE UNIT YEARS 1999/2000

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Background. The goal of WHO 2005 is to keep case-fatality of stroke in Europe below 20%. We studied case-fatality (30-days mortality) in a Copenhagen stroke-unit (SU) during 8 months in 1999/2000. We also studied case-fatality of stroke patients in a dedicated SU compared to case-fatality in general medical wards (GMW) of the same hospital.

Methods. From september 1, 1999 to april 30, 2000, 494 pts with stroke were admitted to a Copenhagen hospital with an uptake area of 300 000 inhabitants. All stroke patients in the area were admitted to the same hospital being the only hospital in the area to which stroke patients are admitted. Due to limited capacity 50 pts were treated at general medical wards (GMW). All patients had a structured interview, stroke severity was measured by the Scandinavian Stroke Scale (SSS) and all had an evaluation of their stroke risk profile. Blood pressure, body temperature, and blood glucose were measured in all. Univariate as well as multivariate statistic were entertained.

Results. Mean age was 74 years, 51% were females. Case fatality was 11% in 444 patients admitted to SU vs 46% in 50 patients admitted to GMW ($p<0.0001$). However, patients admitted to GMW had more severe strokes; mean-admission SSS in patients at SU was 42 vs 30 in patients at GMW ($p<0.001$). Furthermore, 11% of patients in SU had hemorrhage vs 32% in GMW ($p<0.0001$). Age, co-morbidity and risk factor profile was the same in patients admitted to SU and GMW. In a multivariate analysis predictors of case-fatality were *stroke severity* (OR 2.6 per 10 points increase in SSS score, 95% CI 1.9 to 3.6), *age* OR 0.4 per 10 years increase, 95% CI 0.2 to 0.7) and *admission to SU* (OR 6.4, 95% CI 1.5 to 28) but gender, smoking, alcohol intake, diabetes, hypertension, atrial fibrillation, co-morbidity, claudication, previous stroke/TIA, stroke subtype or current ASA prophylaxis influenced case-fatality. Case-fatality of the total population of 494 patients (SU and GMW) was 14.6%.

Conclusions. Most probably due to introduction of stroke-units case-fatality of stroke is much lower than the goal of 2005 set by WHO. Case-fatality was predicted by stroke severity, age and whether the patients were admitted to a stroke unit or not. Thus, our study suggests that the goal of case-fatality for stroke patients in 2005 at least in Scandinavia should be much lower than 20%. In this study the relative risk of dying from stroke within in the first months was six-fold higher in the GMW than in the SU. Thus, the study, stresses the life saving benefit of SU treatment.

C17

THE EFFECT OF CUTANEOUS ELETRICAL STIMULATION IN REHABILITATION OF CHRONIC STROKE

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Background. The purpose of this study was to find out if cutaneous electrical stimulation has any effect in functional recovery of chronic stroke measured by functional tests and somatosensory evoked potentials. Subjects were drawn from patients at the Brain Research and Rehabilitation Center Neuron in Kuopio.

Methods. Fifthy-one subjects with chronic stroke received treatment in hand (n=32) or foot (n=19) and additional eight received no-current treatment in hand. Outcomes were assessed before treatment began and at the end of treatment by the same masked raters, with Modified Motor Assessment Scale (MMAS), paretic limb function and skin sensation and somatosensory evoked potentials (SEP).

Results. MMAS ($p<0.01$), paretic hand function ($p<0.01$), upper limb skin sensation ($p<0.01$) and SEP normality classification in paretic upper limb ($p<0.01$) and paretic lower limb ($p<0.05$) improved significantly in the treatment group (n=51) after three weeks rehabilitation. When hand treatment (n=32) and hand placebo (n=8) groups were compared there were a significant improvement in MMAS ($p<0.01$), paretic hand function ($p<0.01$), upper limb skin sensation ($p<0.01$) and SEP normality classification in paretic upper limb ($p<0.001$) only in the treatment group.

Conclusions. Cutaneous stimulation during the rehabilitation period had clear positive effects in the motor performance, limb sensation and the quality of somatosensory evoked potentials in chronic stroke subjects.

ON-LINE DIFFERENTIATION OF SOLID AND GASEOUS CEREBRAL MICRO-EMBOLI USING MULTIFREQUENCY DOPPLER

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Introduction. Although cerebral microemboli may be detected using transcranial Doppler, it is at present extremely difficult to determine if they are composed of gaseous or solid elements. Theoretically this is possible by insonating an embolus simultaneously with two different ultrasound frequencies since the reflected ultrasound power will differ for each frequency depending on embolus type.

Methods and results. A new Doppler instrumentation (EmboDop) has therefore been developed which discriminates between solid and gaseous microemboli by insonating them simultaneously with two different ultrasound frequencies (2.0 MHz and 2.5 MHz).
In an experimental study using a pulsatile closed-loop system, analysis of the reflected Doppler power for the two different ultrasound frequencies showed correct differentiation between solid (N = 61) and gaseous (N = 45) microemboli for 95% of the microembolic events.
Five hundred and sixty-six microemboli in 5 prosthetic heart valve patients and 33 microemboli in 16 carotid stenosis patients have also been analysed. In the prosthetic heart valve patients 474 (83.7%) were gaseous, and 83 (14.7%) solid and in 9 (1.6%) the differentiation was uncertain. In the carotid stenosis patients 30 (91%) of the 33 microemboli were classified as solid and 3 (9%) were uncertain.

Conclusions. These studies have shown that it is now possible to automatically discriminate between solid and gaseous cerebral microemboli which in the future may have important therapeutic consequences in patients with cerebrovascular disease.

C19

FACTORS PREDICTING POOR OUTCOME OF PRIMARY INTRACEREBRAL HEMORRHAGE IN MIDDLE-AGED AND ELDERLY PEOPLE

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Background. Several predictors for death after spontaneous intracerebral hemorrhage (ICH) have been identified. However, predictors of functional outcome in elderly patients with the highest incidence of ICH are not well known. We investigated the prognostic value of clinical and radiological characteristics as well as lifestyle factors in middle-aged and elderly people with ICH.

Methods. Ninety-eight consecutive patients with spontaneous ICH were prospectively examined, and outcome was assessed according to the Glasgow Outcome Scale 3 months after hemorrhage. Odds ratios (ORs) and 95% confidence intervals (CIs) for death and poor outcome, after adjustment for possible confounding factors, were calculated by logistic regression.

Results. The independent predictors for poor outcome after ICH were age (per year) (OR, 1.13; 95% CI, 1.05 to 1.21), prognostic score of the Scandinavian Stroke Scale (SSS-PRG) on admission (per unit) (OR, 0.755; 95% CI, 0.66 to 0.87), heart rate (per beat/min) (OR, 1.04; 95% CI, 1.00 to 1.08) and thalamic hemorrhage (OR, 6.19; 95% CI, 1.10 to 34.9). The independent predictors for death were Glasgow Coma Scale score on admission (per unit) (OR, 0.614; 95% CI, 0.45 to 0.84), and thalamic hemorrhage (OR, 11.1; 95% CI, 1.16 to 105).

Conclusions. The functional outcome in elderly patients with relatively small ICHs is well predicted by the SSS-PRG score. Thalamic hemorrhage seems to predict both poor functional outcome and death. Other factors which associate with poor functional outcome are age and heart rate.

DYNAMIC RADIOTHERMOMAPPING FOR EXAMINATION OF BRAIN NEURO-CIRCULATORY DISEASES

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Introduction. Brain temperature is one of important characteristics of its microcirculation and metabolism dynamics. Monitoring of temperature in different brain areas is important in clinic as for patient state estimation in the acute period of brain pathology so as for the control in the period of rehabilitation. It is known also that the course of different brain pathological processes depends on temperature dynamics. For instance, after brain trauma, ischaemic stroke the brain temperature can increase to hyperthermia which changes the brain state for the worse (Schwab et al, 1997). At the same time mild hypothermia is able to make positive therapeutic treatment (Laptook et al, 1995; Whalen et al., 1997). Methods of brain temperature measurement used now in clinics are invasive or do not reflect completely brain temperature and its dynamics (Cady et al,1995; Gulyaev et al,1984; Millergard, 1995; Stone et al, 1995; Verley et al, 1995; Shevelev et al, 1989). Nowadays the search of adequate noninvasive method of brain temperature monitoring is the urgent task for clinic (Corbett et al, 1995; Laptook et al,1995; Nurse, Corbett, 1994).

Methods. At the Institute of Radioengineering and Electronics Russian Academy of Science the noninvasive method of dynamic multichannal radiothermovision was created especially for monitoring of human brain thermal processes (Selsky et al, 1991). The equipment consists of radiometer with 12 antennae, computer and software. The method permits to measure temperature changes in 12 areas on the depth 2-2,5 sm. from the head surface. The sensitivity of the equipment is 0.1K, the spatial permission is 25 mm sq, the apparatus time of integrating is 2 sec. By this method brain thermomaps can be obtained.

Results. By now the method was applied for examination of brain temperature dynamics in some cases of cerebrovascular diseases (focal microcirculatory disorders and consequences of arterial branch pathology). Reliable difference was shown between temperature dynamics in healthy persons and patients with discirculatory disorders. It was shown that in patients the brain temperature changed in wider ranges; localization of pathological foci could be estimated; specific temporal and spatial temperature dynamics was obtained.

Conclusion. We suppose that the method of dynamic multichannal thermovision can be useful for analyse of different kinds of brain neurocirculatory pathology.

C21

TREATMENT WITH ORAL ANTICOAGULANT AGENTS IN ISCHEMIC STROKE PATIENTS WITH ATRIAL FIBRILLATION IN A POPULATION BASED STUDY

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Background. People with atrial fibrillation (AF) have a 5-fold excess risk of stroke compared to people without AF. Randomized trials have shown that treatment with oral anticoagulant agents (AC) decrease the risk annual risk by two thirds in intention-to-treat analyses.

Methods. Within the Northern Sweden MONICA study, stroke has been monitored since 1985 in the age group 25-74 years in both men and women. The same strict WHO criteria has been used all years. All patients with subarachnoid hemorrhages have been excluded. In total 5,584 men and 3,537 women had a first ever stroke 1985 through 1998. Information about history of atrial fibrillation has been monitored since the start, as well as treatment with oral anticoagulants at onset, while secondary prevention in terms of AC has been followed since 1995. In this presentation, only patients in age group 55-74 are included, because few (3.4%) patients in age group 25-54 years had a known atrial fibrillation at onset.

Results. In age group 55-74 years 683 of 4,844 men (14.1%) and 452 of 3,059 women (14.8%) with ischemic stroke had previous known atrial fibrillation at onset. Of these, 56 men (8.1%) and 39 women (8.5%) were on treatment with oral anticoagulant drugs at onset, with no statistical difference between men and women; p=0.83. Within the first 28 days after onset, 50% of male and 56% of female (p=0.18), were treated with AC. Intracerebral hemorrhage during AC treatment in patients with AF was a relatively uncommon event occurring in 32 of all 603 men (5.3%) and 20 of all 404 (5.0%) women affected by an intracerebral hemorrhage.

Conclusions. Few patients with known atrial fibrillation were treated with oral anticoagulants before the first stroke event in age group 55-74 years, although more than half of the patients with ischemic stroke received oral anticoagulants agents within 28 days after the event. In a population perspective, intracerebral hemorrhage during AC treatment in patients with atrial fibrillation is uncommon, accounting for only 0.6% of all strokes.

LONG-TERM CHANGES IN STROKE INCIDENCE AND SURVIVAL IN SÖDERHAMN, SWEDEN, 1975-1999. A TREND FOR MILDER STROKES

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Background. The factors underlying the decline in stroke mortality are not yet clarified. Trends towards milder strokes may be explained by a shift from one stroke subtype to another (e.g. from intracerebral hemorrhage to cerebral infarction) or a by a shift of stroke severity within a subtype (e.g. from large vessel occlusion to small vessel occlusion). The present study is undertaken in order to elucidate some of these questions. It is based on a register for stroke and TIA in a well-defined population in Sweden. Cerebral infarcts with mild symptoms (RIND) were prospectively registered.

Methods. The population was screened in the periods May 1975 to April 1978, and September 1983 to August 1991. All included patients have been followed until May 1999. The incidence rate for each study year was standardised to the Söderhamn population of 1990 by a direct method. Trends were measured by calculating the correlation coefficients for standardised incidence rates and study year (1975-1991). Survival ratios were achieved by cross-tabulation. The relationship between the survival ratio (dependent variable) and stroke subtype, age, sex and study year was analysed with logistic regression.

Results. 1186 patients with first-ever stroke and 178 patients with first-ever TIA were registered. The incidence rate for all strokes and TIA were not correlated to study year. For stroke subtypes, a significant positive correlation was found between the incidence of cerebral infarction and study year. As regards severity of stroke, there was a significant positive correlation between cerebral infarctions with mild symptoms and study year. The survival ratios, from one up to seven years, increased significantly in all age groups except for the youngest (15-64 years), in which no change was found. For cerebral infarction, the 7-year survival ratio increased from 30.3% to 48.3%. In logistic regression analyses, there was a relationship between survival ratios and stroke subtypes, age and study year.

Conclusions. During the period 1975-1991, there seems to have been a shift towards cerebral infarctions with mild symptoms which possibly reflects relatively less large vessel occlusion and more small vessel occlusion. The long-term survival improved gradually, possibly being a cohort effect or a result of improved post-stroke therapy.

C23

PARTICIPATION IN GROUP ACTIVITIES CHANGES FUNCTIONING: EVALUATED WITH THE CANADIAN OCCUPATION PERFORMANCE MEASURE

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Background. Lately several studies have showed that chronic stroke patients who have participated in an intensive individual rehabilitation program improve in functioning. The aim of this study was to find out if chronic stroke patients experience functional improvement after participation in an intensive group rehabilitation program.

Method. A total of 20 persons participated in a four-weeks intensive rehabilitation program based on group activities. During the program the patients also received individual sessions in occupational and physical therapy twice a week respectively. The Canadian Occupational Performance Measure (COPM) was used to identify problem areas in occupational activities before the training period and to rate the level of performance and satisfaction before and after the training period. The material was analysed by non-parametric statistical tests.

Results. A total of 75 occupational activities were identified as difficulties, of which 64% presented self care, 20% presented productivity, and 16 % presented leisure. The result showed that the patients rated performance significantly higher ($p<0.001$) as well as they rated satisfaction significantly higher ($p<0.001$) compared to before participating in the group rehabilitation program. The patients also rated performance significantly higher ($p<0.01$) three months post participating in the rehabilitation program but not in satisfaction.

Conclusions. This study emphasises that motivated chronic stroke patients experience improvements in functioning after participation in an intensive group rehabilitation program. The COPM showed to be a clinically usable assessment tool in terms of administration, time, and ease of scoring.

THE NORDIC COOLING STROKE STUDY - NOCSS

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Background. Theoretically hypothermia is the most potential neuroprotective treatment today. In experimental animal studies hypothermia confers protection from global as well as focal ischemia. Recent studies in human stroke have shown an association between body temperature on admission and outcome. Hypothermic therapy is since long applied in open heart surgery to protect the patients from brain ischemia. In a Cochrane Library Document *'Cooling therapy for acute stroke'* it is concluded that trials investigating the neuroprotective effect of cooling in humans with acute stroke are needed. In a pilot study of mild hypothermia in 17 patients with acute stroke the cooling procedure was found to be feasible and safe.
We have therefore now initiated *'The Nordic Cooling Stroke Study'* which is the first large, randomized, controlled study of the effect of mild hypothermia in acute stroke patients.

Design. The study is a randomized, prospective, multicenter study comparing two groups:

- The hypothermia group: The patients receive 9 hours of hypothermic treatment within the first 6 hours after the stroke. Discomfort and shivering are reduced by intravenous injections of pethidin. The target temperature is 35° C.
- The control group: Patients admitted within 6 hours post-stroke that, except for the hypothermic treatment and the pethidin injections, are observed and treated just as the hypothermia group.

Methods. Hypothermia is achieved by surface cooling using the convection principle with cold water (Medivance - The ArticSun). Patients are awake during the cooling procedure. Shiverings are controlled by intravenous injections of pethidin (25-50 mg).

Inclusion/exclusion. We expect to include 500 patients in each group from 20 Scandinavian centers. Admission Scandinavian Stroke Scale (SSS) should be from 10 to 44.
Main exclusion criteria:

- Hemorrhagic stroke
- SSS sub-score for consciousness ≤ 2
- Patients with severe aphasia
- Severe cardiac and pulmonary disease

Endpoints. Primary endpoint: Significant improvement of SSS at 3 months post stroke.
Secondary endpoints: Significant improvement of:
NIHSS, patients with Modified Rankin Scale 0-2, survival, Barthel Index, quality of life (SF-36) and patients discharged to their own home at 3 months post stroke.

Status. Inclusion of patients started May, 2001 and is expected to be completed May 2004.

C25

EVIDENCE-BASED NURSING TO STROKE PATIENTS APPLICATION OF AN EVIDENCE-BASED CLINICAL GUIDELINE CONCERNING THE BASIC NEED: ELIMINATION OF URINE

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Background. The aim of this project was to evidence-base nursing care of stroke patients and to monitor the quality of nursing practice. The monitoring process is related to basic human needs, as developed and described by Virginia Henderson. The aim of nursing concerning the basic need: elimination of urine, is the patient's spontaneous passing of urine 4-6 times a day, maximum score of bladder control, individually adjusted continence aid, and prevention of urinary tract infection.

Method. The clinical guideline

Problem Research show that 50% of patients with stroke are incontinent with urine on admission. About 20% of the patients' have persisting urinary incontinence. Prevalence in a general population lies around 10%, increasing with age. The most frequent type of urinary incontinence is urge incontinence. Urinary incontinence is shown to increase the risk of urinary tract infections. In the acute phase of stroke there may be symptoms of urinary retention.

Recommendations for good clinical practice.
Principles of Minimal care are followed during examination and diagnosing
Identifying the risk patient:
Assessment according to standard questionnaire
Retention/post residual urine
Sterile Intermittent Catheterisation (B/C)
Urge/stress incontinens

Intervention: patient unable to participate actively
- scheduled voiding (B)
- incontinence aid

Intervention: patient able to participate actively.
- scheduled voiding (B)
- incontinence aid
- bladder training (C)
- pelvic floor training (C)

Monitoring. Nursing sensitive quality indicators are registered with acute stroke patients in two areas. Study population approximately 1500 stroke patients.

Quality indicators.
- Daily assessment of voiding pattern
- Bladder control according to Barthel Index or other validated instrument
- Events of urinary tract infections treated with antibiotics during hospitalisation

Results. The clinical guidelines have been implemented in stroke units at Bispebjerg University Hospital, Århus University Hospital and Esbønderup Hospital

ENVIRONMENTAL EFFECTS ON FUNCTIONAL RECOVERY AND SPATIAL MEMORY AFTER FOCAL BRAIN ISCHEMIA IN RAT

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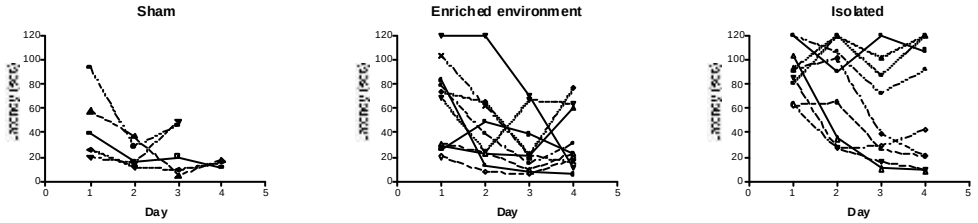
Background. Environmental enrichment has been shown to be a potent inducer of neuronal plasticity, including improved recovery of sensorimotor functions after focal cerebral ischemia. Memory impairment is common after stroke in man and has been shown after MCA occlusion in rats. Effects of enrichment on recovery of memory function are less studied. The plasticity associated transcription factor NGFI-A (egr-1, Krox24) is a possible candidate in the signal transduction pathway mediating the functional improvement induced by environmental enrichment.

Methods. In this study 19 male Sprague-Dawley rats were subject to a 90 min occlusion of the right middle cerebral artery using the intraluminal suture model. Rats were allowed to recover in single cages for two days, and were then randomized either to an enriched environment (n=10) or single cages (n=9). Five sham operated rats were housed in single cages. Sensorimotor performance was tested weekly up to 4 weeks after ischemia by limb placement tests and ability to walk across a rotating pole. Spatial memory was tested in the Morris swim maze five weeks after MCAO. Brain sections (10µm thick) were stained with hematoxylin and eosin and infarct volume was measured using computerized image analysis. Levels of NGFI-A mRNA expression in hippocampal subregions was determined by riboprobe in situ hybridization, analyzed using computerized image analysis.

Results. Rats housed in enriched environment after ischemia showed moderately enhanced recovery of sensorimotor function. Ischemic isolated animals had a significantly longer latency to find the platform compared to rats housed in an enriched environment. However, variability among the isolated rats was considerable. No difference in total infarct volume was found between the isolated and enriched environment groups, or between better or worse isolated rats, but preliminary analyses suggest that poorly performing isolated rats had a more severe injury in the posterior parts of the amygdala. No difference was found in hippocampal NGFI-A mRNA expression.



Conclusions. These data suggests that environmental enrichment may influence cognitive outcome after focal ischemic brain damage. In rats housed isolated after MCA occlusion more severe damage to the amygdala may be of importance for Morris swim maze performance. At this timepoint (five weeks) hippocampal NGFI-A levels appear not to be altered by environmental enrichment.

B2

PROTEOMIC ANALYSIS OF PROTEIN OXIDATION IN RAT BRAIN AFTER TRANSIENT FOCAL CEREBRAL ISCHEMIA

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Background. Oxidative stress after cerebral ishemia is thought to cause severe damages to lipids, nucleic acids and proteins. Although elevation of total protein carbonyls has been previously shown in rat cerebral ischemia, the identity of the proteins that are most sensitive to oxidization has remained unknown. We have applied proteome analysis to study oxidatively damaged proteins in the rat brain after transient focal cerebral ischemia.

Methods. Protein carbonyls were derivatized with 2,4-dinitrophenylhydrazine (DNPH) and then separated in two-dimensional (2D) electrophoresis. Proteins were stained with Sypro Ruby dye and subjected to western blotting using anti-DNP antibody. To further identify the proteins, analysis with 2D electrophoresis and electrospray ionization mass spectrometry (ESI-MS) was applied.

Results. After ischemia elevated oxidation was observed in proteins, which were slightly carbonylated even in physiological state and, in some proteins, which were not oxidatively modified upon normal conditions. The sequence analysis of the peptides derived from oxidatively damaged proteins in the rat brain revealed high homology with L-lactate dehydrogenase, actin, creatine kinase, heat shock protein 70, phosphatidylethanolamine-binding protein and ubiquitin carboxyl-terminal hydrolase.

Conclusions. Proteome analysis can be successfully applied for identification of oxidatively damaged proteins in diseased brain tissues. Cerebral ischemia causes increased carbonylation of proteins. Creatine kinase was identified as one of the targets of protein oxidation in the brain, suggesting that enzymes involved in energy metabolism may be especially sensitive to oxidative modification upon ischemia-reperfusion injury in the brain.
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THE INVESTIGATION OF SOMATOSTATIN INJECTION INFLUENCE ON NEUROLOGICAL DISTURBANCES IN RATS AFTER 12-MIN CARDIAC ARREST

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Background. Recently it has been shown that there are many pathological cascades and disturbances in all areas of the organism in postischemic period. Many of them have a long-term character. It is known, that there are some substances, which demonstrate a positive effects in the early postischemic stage, but not all them have the same effects in delayed period. On this basis it would be expedient to pay attention to the neuropeptides family because of the important role which they play in functioning of the whole organism. One of them - somatostatin - is the inhibitor of many hormones and peptides functions, and has membrane and citoprotection properties in the organism.
The aim of the present study was the estimation of somatostatin influence on the mortality level, recovery of the neurological status and behavior of rats in the postischemic period.

Methods. The cardiac arrest type cerebral ischemia was induced by compression of intrathoraxic main vessels with special hook under the ether anesthesia. The resuscitation of animals was started 12 min after onset of ischemia and included approximately 1-2 min of cardiac massage and 30 min of artificial ventilation with respirator. Somatostatin in dose 0,05 mg/kg was injected subcutaneously 30 min after resuscitation. For two weeks we estimated the recovery of the neurological status and mortality level. Then alterations in central nerve system functioning in behavioral tests with nonstressogenic conditions (“RODEO”), and stressogenic conditions (“Open field”) were estimated.

Results. Somatostatin in single injection did not show significant influence on the mortality level and recovery of the neurological status on the early postischemic stage. In both nonstressogenic and stressogenic conditions animals, subjected to ischemia, showed the increase of emotional reactivity and exploratory reaction - level of horizontal and vertical activity in both tests and crossing arena’s center in the last test were higher as compared with the intact rats. The data of nonstressogenic conditions testing suggest that somatostatin in single injection had no influence on the emotional status of ischemic rats. In stressogenic conditions rats treated by this peptide showed significant decrease of postischemic pathological excitement: parameters of horizontal activity and crossing the center of arena (I and II) were significantly lower than ischemic rat’s parameters. But this decrease was not up to the level of intact rats. It is important, that behavior of intact animals, injected by somatostatin were similar to the intact rats without such injection.

Conclusions. Thus, somatostatin in single injection in early postischemic period seems to possess the compensatory effects on emotional reactivity of rats and their estimation of environmental stimuli significance after cardiac arrest-type cerebral ischemia in delayed stage because of its membrane protection properties.

B4

ON AND OFF-RESONANCE T_{1r} AS NOVEL MRI CONTRASTS IN ACUTE CEREBRAL ISCHEMIA

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Background. In very short time magnetic resonance imaging (MRI) has become a method of choice in imaging of acute stroke. Yet, identification of reversible or irreversible ischemia has proven to be difficult. It has recently been shown that acute increases of $T_{1\rho}$ relaxation time [1] and dispersion of $T_{1\rho}$ relaxation ($d_{T1\rho}$) [2] are sensitive indicators of irreversible neuronal damage in acute phase of cerebral ischemia and reperfusion, also in the cases with ambiguous diffusion change. Here we have studied off-resonance $T_{1\rho}$ relaxation during acute focal ischemia and reperfusion in the rat.
Methods. Male Wistar rats (n=6) were anesthetized with 1 % halothane for 90 minutes middle cerebral artery occlusion (MCAO) using intraluminal thread model [3]. MCAO was followed by reperfusion. Arterial blood gases and pH were analyzed; blood pressure and body temperature were monitored on-line MRI experiments were performed at 4.7 T. $T_{1\rho}$ was quantified using five variable length (10-90 ms) adiabatic spin-lock pulses, in front of a fast spin echo imaging sequence (TR=2.5 s, echo spacing=10 ms, 16 echoes/excitation, 2 averages). Off-resonance $T_{1\rho}$ was quantified with adiabatic pulses in which the adiabatic sweep was truncated when the desired off-resonance frequency was reached. In the on-resonance measurements three B_1-fields (0.4 -1.6 G) and in off–resonance measurements 5 off-resonance frequencies (0-7.5 kHz) with B_1=0.4 G were used. The trace of the diffusion tensor images (D_{av}) were obtained using a spin-echo sequence (TR=1.5 s, TE=55 ms, b-values: 0, 469, 1057 s/mm^2).
Results. In ischemic putamen, D_{av} decreased by 34±6 % if compared to contralateral value of 0.75±0.02mm^2/m during 90 min MCA occlusion as a sign of severe ischemia. No recovery of diffusion coefficient was detected during reperfusion. In normal caudate putamen, $T_{1\rho}$ changed from 72.9±0.8 to 95.0±1.1 ms in B_1 range of 0.4-1.6 G. During ischemia and reperfusion on-resonance $T_{1\rho}$ gradually increased, and the highest relative increase of 17–24 % was detected with the strongest spin-lock field. In ipsilateral putamen, dispersion of the $T_{1\rho}$, $d_{T1\rho}$, was elevated by 46±1 % after 30 min of ischaemia if compared with contralateral putamen. $d_{T1\rho}$ gradually increased both during and after MCAO reaching a value that was 58±5 % higher than in contralateral after 90 minutes of reperfusion. In unexposed putamen $T_{1\rho,off}$ increased from 73.7±0.5 to 328.8±6.7 ms as off-resonance frequency increased from 0 to 7500 Hz. $T_{1\rho,off}$ increased during ischemia 2-6 %, which was of same magnitude as detected with an on-resonance spin-lock with the same B_1-field. However, during reperfusion there was increased sensitivity to post-ischemic alterations with off-resonance approach if compared to on-resonance conditions. The highest sensitivity was reached with the on-resonance frequencies between 1250 and 2500 Hz.
Conclusions. We have characterized on-resonance and off-resonance $T_{1\rho}$ relaxation changes during acute cerebral ischemia. $T_{1\rho}$ dispersion was elevated 46-48 % during 90 minutes of occlusion, which exceeds previously determined tissue MRI contrasts in acute cerebral ischemia. Similar dispersion effect was not detected using off-resonance $T_{1\rho}$ methods during ischemia. The interpretation of the off-resonance $T_{1\rho}$ results is complicated with increased T_1 contribution and off-resonance saturation effects, but may provide insight to the mechanisms underlying $T_{1\rho}$ relaxation alterations in acute cerebral ischemia.
[1] Gröhn et al. MRM, 42:268-276, 1999 [2] Gröhn et al. JCBFM, 20: 1457-1466, 2000
[3] Longa et al. Stroke 20:84-91, 1989

PERMANENT FOCAL ISCHAEMIA IN MICE: EVALUTION OF SPONTANEOUS REGENERATION AND OF BEHAVIOUR

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Background. Focal cerebral ischaemia in mice is a model of stroke often used for testing potential neuroprotective drugs. However, due to the very limited time-window for treatment and regarding many clinical studies with negative outcome, the strategy of neuroprotection does not seem to lead to successful solutions for the treatment of human stroke in the next future. We therefore want to focus on mechanisms of regeneration after an ischaemic insult. For this purpose we started experiments using the model of Middle Cerebral Artery occlusion (MCAO) in mice and post-ischaemic behavioural testing, followed by histological evaluation of the infarct-area.

Methods. For our experiments we chose the model of transcranial permanent electrocoagulation of the MCA in mice. With this technique the animals are not strongly impaired, survive well and can be used for long-term survival studies and behavioural testing. Briefly, male NMRI mice were anaesthetised with tribromethanol (600mg/kg; i.p.). The temporalis muscle was removed by electrical coagulation. A burr hole was drilled into the outer surface of the skull just over the MCA which was then occluded by microbipolar coagulation. Decapitation was performed on day 2 and day 10 post-ischaemia. Sham-operated animals underwent the same procedure except the electrocoagulation. After conventional histological procedure the brain slices were stained with cresyl violet. The behavioural testing battery focussed on locomotion and motor skills and was based on the SHIRPA-protocol. From all paradigms (e.g. spontaneous activity, startle response, grip-strength) locomotion, rearing and running wheel-experiments show the most interesting results and will be presented. Briefly, mice were placed in a glass jar and number of rearings were counted during a 2 min. period. Afterwards, mice were tested in an arena with squares on the bottom and numbers of squares entered during 30 sec were counted. Finally, mice were placed in an running wheel for 1 min and we registered number of rotations.

Results. The infarct volume differed highly significantly between ischaemic animals decapitated on day 2 and on day 10 after MCAO (infarct volume (cubic centimetre) 2d after MCAO: 13.41+/-2.7; 10d after MCAO: 0.33 +/-0.25; $p<0.001$). As expected, there was also a significant difference between MCAO and sham-operation on day 2 post ischaemia (2d after MCAO: 13.41+/-2.5; 2d after sham-operation: 3.52+/-2.95; $p<0.05$); statistics: ANOVA). Numbers of rearings and rotations were increased on day 10 after operation in comparison to naive animals, however, there was no difference between sham-group and MCAO-group. Locomotion did not differ between control-group, sham-operated and MCAO animals.

Conclusions. Mice show a striking ability to spontaneously decrease the infarct area after MCAO to almost zero. This has also severe implications for studies of neuroprotection, as we could show that with this model it is not possible to differentiate between mere acceleration of reduction of infarct volume and real protection. However, a more detailed analysis of this spontaneously occurring regeneration by histological and immunohistochemical techniques will be necessary to understand the mechanisms. By testing locomotion, rearings and behaviour in running-wheel, our experiments revealed that in this model not ischaemic damage seems to change behaviour in these paradigms (as sham-operated animals showed the same effects) but rather the stress of operation might have significant influence.

B6

MITOTIC SPINDLES IN ADULT RAT BRAIN SUGGEST DIVIDING NEURONS AS A SOURCE TO NEUROGENESIS AFTER TRANSIENT MIDDLE CEREBRAL ARTERY OCCLUSION

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Background. Division of mature neurons [1,2] may be associated with post-stroke cortical neurogenesis in adult rat brains [3]. This study aimed to further verify the occurrence of post-stroke nerve cell divisions in the adult rat brains through demonstration of the spindle assembly-a unique cell structure that appears only in cells under mitosis.

Methods. Ten weeks old male Wistar rats were subjected to unilateral middle cerebral artery suture occlusion with reperfusion at 2 h after stroke [3] and BrdU delivery. Rats were killed at 24h, 48h or 72h after stroke (n=3 in ischemic groups, n=2 in sham control). Brain sections were processed for immunohistochemistry or immunofluorescence. Primary antibodies used: the neuron-specific markers Map-2, β-tubulin III, Neu N; the glial marker GFAP; the mitosis-specific markers phospho histone H3 (phos H3), γ-tubulin; DNA synthesis marker BrdU. Various fluorescent secondary antibodies were used to detect the primary antibodies. Nuclear DNA was counterstained with DAPI.

Results. Analysed with three-dimension confocal microscopy, the mitotic markers phos H3 and γ-tubulin colocalized with the mature neuron specific marker Neu N in the same cells in the ischemic boundary cortex at 24-48-72h after MCA-O. The γ-tubulin -labeled mitotic spindles, appeared either as mono-polar or bipolar assembly, which enveloped completely the Neu N image in these cells. The cell chromosomes, visualized through DAPI counterstaining, appeared in various shapes while they were stretched between the γ-tubulin positive mitotic spindles bipolar accumulated. From 48h after stroke, γ-tubulin positive mitotic spindles were frequently observed in astrocytes as identified with the mature glial cell marker GFAP. Similarly, splitting nuclei were commonly seen in these mitotic astrocytes.

Conclusion. These data suggest that cell divisions did occur in neurons as well as astrocytes after MCA occlusion and support the hypothesis that adult neurons may be a cellular resource for post-stroke cortical neurogenesis in the adult brains.

1. Gu, W.G., Brannstrom, T. and Wester, P., Division of mature neurons in vivo contributes to cortical neurogenesis in adult rats after reversible photothrombotic ring stroke., *Soc Neurosci Abs, Vol. 26*, 2000, pp. 757.
2. Jiang, W., Gu, W.G., Brannstrom, T. and Wester, P., Division of mature neurons in vivo contributes to cortical neurogenesis in adult rats after middle cerebral artery occlusion., *Soc Neurosci Abs, Vol. 26*, 2000, pp. 757.
3. Jiang, W., Gu, W.G., Brännström, T., Rosqvist, R. and Wester, P., Cortical neurogenesis in adult rats after transient middle cerebral artery occlusion, *Stroke*, 32 (2001) 1201-1207.

SPONTANEOUS FORELIMB USE AFTER TRANSIENT FOCAL CEREBRAL ISCHEMIA IN RATS TREATED WITH AN a $_2$-ADRENOCEPTOR ANTAGONIST

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Backround. Transient occlusion of the middle cerebral artery (MCA) produces corticostriatal infarcts, which induce sensorimotor deficits in rats. Atipamezole (ATI), a selective α_2-adrenoceptor antagonist, enhances the recovery of sensorimotor function. The present study used the cylinder test to investigate whether atipamezole affects forelimb use during spontaneous exploratory activity following transient MCA occlusion.

Methods. The right middle cerebral artery of male Wistar rats was occluded for 120 min using the intraluminal filament method. Ischemic rats were assigned to equal groups according to the behavioral deficit in the limb-placing test. Sham-operated rats were used as controls. Rats were video-recorded in a transparent cylinder (ø 25 cm) for 10 min before surgery and on postoperative days 6 and 23. ATI (1 mg/kg) or 0.9% NaCl was administered 20 min before the cylinder test on postoperative days 2 through 11, 15, 19, and 23. Exploratory activity was analyzed in a blind manner during a 2-min observation period. Forelimb use was observed during take-off, during the first contact against the wall, during lateral exploration on the wall, and during landing. The independent use of impaired and nonimpaired forelimbs and the simultaneous use of both forelimbs were assessed**.** The number of quarter turns in either direction were also counted.

Results. On postoperative day 6, both ischemic groups made more ipsiversive quarter turns than sham-operated controls. Ischemic controls used the nonimpaired forelimb during take-offs more than did sham-operated controls on day 23 after the operation. There was no difference in impaired forelimb use during take-offs or landings between ATI- or NaCl-treated ischemic rats. ATI increased the use of the nonimpaired forelimb on the wall in sham-operated and ischemic rats. ATI also increased the number of contraversive quarter turns in ischemic rats compared to ischemic controls on postoperative day 6.

Conclusions. These results indicate a significant limb use asymmetry in exploratory behavior of ischemic rats. Ischemic rats might compensate for deficits in weight-shifting movements by increasing the use of the nonimpaired forelimb. ATI increases exploratory activity but not the independent use of the impaired forelimb in ischemic rats.

B8

MR RELAXATION TIME T_{1r} INCREASES IMMEDIATELY UPON GLOBAL CEREBRAL ISCHEMIA IN RATS

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Background. Unambiguous early detection of cerebral ischemia is of great clinical impact. We have previously shown that the MR relaxation time $T_{1\rho}$ increases within 15 minutes of ischemia and it may offer new information from tissue viability in acute stroke [1,2]. Since the flow thresholds for diffusion and $T_{1\rho}$ changes are similar, it is possible that these two MRI parameters are triggered by similar cellular mechanisms [2]. In the present work we have determined the time course of immediate parenchymal $T_{1\rho}$ response to ischemia and studied its temporal interrelationships with diffusion and anoxic depolarization at varying blood glucose levels. In addition, $T_{1\rho}$ response was compared to magnetization transfer (MT) contrast.

Methods. Global cerebral ischemia was induced by cardial arrest in the rats. Interleaved absolute $T_{1\rho}$ (at a B_1=1.6 G) and the trace of the diffusion tensor (D_{av}) images were acquired at 4.7 T using a line-scan sequence with time resolution of 17 s. DC potentials were simultaneously recorded from somatosensory cortex. Finally, MT ratio (MTR) was assessed by measuring signal intensity with and without MT pulse. The time points for the start of the extensive change (t_{ini}) and 50% change of the maximal change (t_{50}) were estimated for each parameter.

Results. $T_{1\rho}$ started to increase about 20 s after cardiac arrest stabilizing to a level that was 7 % higher than in controls by 200 s. Pre-ischemic blood glucose level did not influence the kinetics of $T_{1\rho}$. Extensive drop in D_{av} began at 40 s of ischemia and the rates of diffusion changes were dependent on pre-ischemic blood glucose with significantly lenghtened t_{50} at high pre-ischemic blood glucose levels. The initiation of extensive DC shifts were also blood glucose dependent, but depolarization did not coincide with any MR parameter. A decreasing trend in MTR was observed with kinetics that were independent of other MRI parameters.

Conclusions. Our results show that $T_{1\rho}$ starts to increase within 30 seconds of global cerebral ischemia. Unlike diffusion, $T_{1\rho}$ increase proceeds apparently independent of preischemic blood glucose. It is therefore possible that these MR parameters probe different cellular mechanisms. Interestingly, early changes in MT contrast do not match $T_{1\rho}$ suggesting that change in the cross relaxation of macromolecules may not be involved in the immediate $T_{1\rho}$ contrast. The present data suggest that anoxic delarization has a small role in the early $T_{1?}$ increase in ischemic brain as $T_{1?}$ is apparently independent on DC potential. An increasing trend in the magnitude of $T_{1?}$ change is observed as pre-ischemic blood glucose level increases indicating that acidification may contribute to $T_{1?}$ response possibly through proton exchange [3]. However, the similarity of $T_{1?}$ response between different glycemic states suggests that tissue acidification may not be the main factor causing ischemic $T_{1?}$ increase and other factors such as altered intracellular microsusceptibility may influence cerebral $T_{1?}$.

1. Gröhn et al. MRM, 42, 268-276, 1999.
2. Mäkelä et al. Proc ISMRM 2001, 854.
3. Gröhn et al. JCBFM, 20, 1457-1466, 2000.

HYPOXIA–REOXYGENATION PROVOKES MATRIX METALLO-PROTEINASE-9 EXPRESSION IN CULTURED HUMAN BRAIN CAPILLARY ENDOTHELIAL CELLS

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Background. The pathogenesis of the hemorrhagic transformation in ischemic stroke involves disruption of the integrity of the microvascular beds. Matrix metalloproteinases (MMP-2, MMP-9) have been associated with profound damage of blood-brain barrier in the ischemic lesions. In this paper, MMPs expression and regulation have been analysed in an endothelial cell culture system. Relevant factors in the ischemic brain tissue (hypoxia, thrombin) as triggers of MMP release have been examined.

Methods. Microvascular endothelial cells from human brain are isolated and culture in vitro. The release MMPs are identified with zymography (combined with inhibitor sensitivity study) and Western-blotting with monoclonal anti-MMP antibodies. The amount of MMP is measured with a three-stage assay: binding of MMP from the sample to immobilised specific monoclonal antibody, activation of a recombinant pro-urokinase by the captured MMP and detection of the generated urokinase activity with a chromogenic substrate. The specific activity of the MMP is determined using the rate of total protein synthesis, estimated on the basis of (^{3}H)amino-acid incorporation.

Results. 4 h long hypoxia elevates the level of MMP-9 in the supernatant of the cultures endothelial cells and this early response is matrix-dependent (requirement for collagen). The increase MMP.9 activity persists for periods longer than 24h. Reoxygenation (with 20 µmol/L H2O2 in the post-hypoxic period causes a 6-fold increase in the specific activity of MMP-9 over the normoxic cells and a similar effect is exerted by thrombin (50 nmol/L).

B10

ERK, JNK AND p38 IN FOCAL CEREBRAL ISCHEMIA

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Background. Focal cerebral ischemia activates intracellular signaling pathways, including the mitogen-activated protein kinases (MAPK) ERK, JNK and p38. These MAPK are involved in the regulation of cell survival, proliferation and inflammatory reaction. The roles of MAPK in cerebral ischemia are currently unclear.

Methods. Rats were subjected to focal cerebral ischemia using the filament technique for transient and permanent middle cerebral artery occlusion (MCAO). Rats were sacrificed after 6 hours to 7 days, perfusion fixed and evaluated with immunohistochemistry directed against the activated forms of the three MAPK.

Results. ERK was activated in blood vessels, glia and neurons in both the ischemic and the contralateral hemispheres. Activated JNK was observed in blood vessels and glia in the ischemic hemisphere, while p38 occured in inflammatory cells.

Conclusion. MAPK are activated in different cell types after MCAO in the rat. These findings may be of value for interpreting the patophysiology of cerebral ischemia.

EFFECTS OF LEAD OR CADMIUM POISONING ON MEMORY PROCESSES FOLLOWING TRANSIENT BRAIN ISCHEMIA IN MICE

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Background. Lead or cadmium exposure can lead to neurological dysfunctions including learning and memory deficits. It may be assumed that although transient bilateral clamping of carotid arteries (BCCA) do not cause deficiency signs, it can reveal neuronal disturbances related to memory processes after exposure to additional toxic agents like lead or cadmium. The purpose of our study was to find out whether transient cerebral ischemia induced by BCCA affects the influence of lead or cadmium poisoning on memory retention and learning.

Methods. Albino-Swiss mice were subjected to 30 minutes of bilateral common carotid arteries occlusion by using thread. BCCA occlusion was carried out under ketamine + xylazinum (50mg/ml+20mg/ml) anesthesia. Sham-operated animals had their carotids separated without occlusion. Each test was performed on four groups containing sham and BCCA mice injected with lead acetate or cadmium chloride i.p and sham or BCCA mice treated with vehicle. Learning or memory retention were assessed by means of a step-through passive avoidance test.

Results. Administration of lead acetate in a dose of 29.3 mg/kg (1/20 LD_{50}) after surgery once a day for 10 consecutive days impaired learning in both BCCA and sham-operated mice compare to vehicle treated groups. A single dose of lead 87.9 mg/kg (1/7.5 LD_{50}) given 24h after surgery and 30 min. before the acquisition trial showed memory retention deficits on postsurgical days 1, 7, 14 and 21. The most effect was observed in BCCA with lead group especially on 1, 14, and 21 day suggesting long-lasting consequences for both BCCA and lead exposure. A dose 58.6 mg/kg (1/10 LD_{50}) had not this influence. Cadmium chloride in a single dose 1.4 mg/kg (1/10 LD_{50}) but not 0.7 mg/kg (1/20 LD_{50}) impaired memory retention when injected 30 min. prior to the retention test. These effect was greater in BCCA group. Treatment with cadmium 0.7 mg/kg after surgery daily for 10 days did not impair learning. Similarly, a single cadmium dose 1.4 mg/kg administrated 30 min before the learning trial was ineffective.

Conclusions. Both environmental toxins affected the memory processes. These results suggest that transient brain ischemia enhances susceptibility to lead or cadmium poisoning and their effects on memory retention and learning.

B12

EXPERIMENTAL HEMORRHAGIC STROKE TREATMENT BY BRAIN-DERIVED FACTORS IN RATS

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The novel Cerebral® remedy belongs to the group of brain-derived factors and its therapeutic action in cerebrovascular disorders treatment is in the process of intensive exploration. The efficacy and mechanism of total remedy and revelation of active combounds of Cerebral® action are the goals of the present study.

The total remedy activates the synthesis and secretion of NGF, elevates the survival in acute hemorrhagic stroke conditions and accelerates the recovery of locomotor activity.

The exploration of separate compounds of Cerebral® obtained by HPLC method revealed the active fraction of the remedy. In the experiments of bilateral hemorrhagic stroke of internal capsule anterior limb in rats this fraction gave the best outcome judged by histopathological indices, neurological deficit score, restoration of locomotor, exploratory and skilled forelimb activities.

TROPHINOTROPINE - THE NEW CLASS OF REMEDIES FOR HAEMORRHAGIC STROKE (HS) TREATMENT

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"Cerebralum" is founder of new medicine group Trophinotropine, that open new era in treatment of acute cerebrovascular and chronic organic neurodegenerative diseases.

The Trophinotropin's row new groop founder is new substance – the modulator of neurotropic factors (IL-family), which change in neocortex maintenance of neuropeptides and some AA (GLU, ASP, GLI, ALA) in HS modelling animals. Administration of this medicine was accompanied with 70% decrease of animals mortality with experimental model of bihemispheric HS. Medicine "Cerebralum" is a peptide Trophinotropine with regulative and modulative makes in acute HS brain tissues, and later - neuroactivative make with a good post stroke outcome.

High medical striking of "Cerebralum" and real lack of sight effects in HS treatment provides with entering molecules substances medicine into the terminations of sense smell nerves (N. olfactorius) after "Cerebralum" intranasal administration. Medicine molecules transported by axons in different regions of CNS (limbic midbrain system) with the mechanism of retrograde axonal transport.

"Cerebralum" accelerated progress of structural and functional restoration of alterative neurons (activative effect), have a neurocytoprotective and different pharmacological (trigger and other) properties in post stroke periods.

"Cerebralum" suggest for treatment of sharp period development of HS in the phases of decompensation, essentially, simplified treatment of patients. This medicine was used for treatment of neurological diseases, conditions that was accompanied with blood entered into the brain and cerebro-sipinal fluid and child organic fall ill (child cerebral pulsy), some other organic brain disorders (Kravchenko S.V. et al, 1997).

Polypharmacological actions of "Cerebralum" are realised of new endopharmacological principe of therapy and are scientific basis for receiving in a nearest future the Individual Medicine for concrete patient's treatment in the concrete stage of disease development.

B14

COMPARISON OF MAGNETIZATION TRANSFER AND T_{1r} RELAXATION IN ACUTE CEREBRAL ISCHEMIA OF THE RAT

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Background. We have previously shown that on-resonance $T_{1\rho}$ increases upon acute cerebral ischemia and that absolute $T_{1\rho}$ images provide predictive information from tissue outcome under conditions when diffusion MRI is ambiguous [1,2]. The biophysical mechanisms affecting $T_{1\rho}$ relaxation are not completely understood, yet $T_{1\rho}$ is known to probe water with restricted mobility in a close interaction with immobile macromolecular proton pools. A MRI method known to probe tissue water interface is magnetization transfer (MTC) and thus it would be of substantial interest to directly compare changes in $T_{1\rho}$ and MTC in the focal cerebral ischemia**.**

Methods. Male Wistar rats (n=6, 230-280 g) were anesthetized with 1% halothane for middle cerebral artery occlusion (MCAO). Arterial blood gases and pH were analyzed and both blood pressure and body temperature were monitored on-line. The intraluminal method of Longa [3] was used for induction of MCAO for 95-100 minutes followed by remotely induced reperfusion.
MRI experiments were performed at 4.7T using a Varian ^{UNITY}INOVA console. A Litz-type volume coil and a dual-loop surface coil were used as transmitter and receiver, respectively. In $T_{1\rho}$ dispersion measurements, the same surface coil was used both as receiver and transmitter to yield high B_1 fields. All the MR data was acquired using a linescan spin-echo sequence (TE=15 ms). Measured line (3mm*3mm*35mm, 128 points) was positioned so that it covered both areas in ischemic and normal putamen and parietal cortex. $T_{1\rho}$ was quantified using adiabatic spinlock pulses (SLT=10-90 ms, B1=0.4-1G) and Z spectra were measured with 5s off-resonance (0.5-100 kHz) saturation pulses (B_1=0.025-0.2G). T_2, T_1 and the trace of the diffusion tensor (D_{av}) were also quantified.

Results. Physiological parameters were within normal range. All the animals were ischemic as indicated by D_{av} reduction by $0.2x10^{-3}mm^2/s$ in ipsilateral hemisphere upon MCAO and no recovery of D_{av} was detected during reperfusion. $T_{1\rho}$ measured with spin-lock field of 1G increased in ipsilateral hemisphere by 14-23 % during ischemia and reperfusion relative to the value of 88.9 ms in the contralateral cortex. Interestingly, virtually no changes in Z-spectra were detected between normal and ischemic tissue (Fig.).

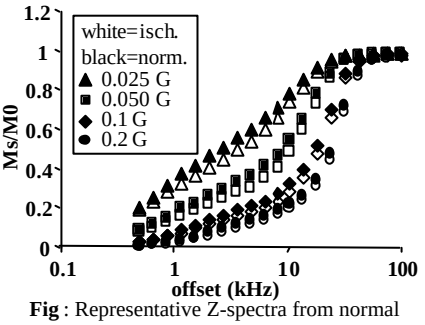

Fig : Representative Z-spectra from normal and ischemic brain tissue 35 min after MCA occlusion.

Conclusions. Our data show that $T_{1\rho}$ is a sensitive indicator of acute cerebral ischemia, a finding consistent with earlier reports [1,2,4]. The plausible changes in MTC between macromolecular and free water pools, as probed by Z spectra, remain small during first hours of MCA occlusion. This indicates that these two MRI methods may probe distinct aspects of water/macromolecule interactions, for instance MTC is known to be mediated predominantly by water molecule exchange pathway while $T_{1\rho}$ may be directly influenced by proton exchange. The latter claim is supported by our recent data from immobilized protein phantoms [5].

References. [1] Gröhn et al. Magn Reson Med 42, 268-276 (1999), [2] Gröhn et al. J Cereb Blood Flow Metab 20, 1457-1466 (2000), [3] Longa et al. Stroke 20, 84-91 (1989), [4] Kettunen et al. Magn Reson Med in press (2001), [5] Mäkelä et al. Proc Int Soc Magn Reson Med 9, 854 (2001)

SELEGILINE COMBINED WITH ENRICHED-ENVIRONMENT HOUSING ATTENUATES SPATIAL LEARNING DEFICIT AFTER FOCAL CEREBRAL ISCHEMIA IN RATS

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Background. Selegiline is an irreversible monoamine oxidase B inhibitor that is suggested to have neuronal rescuing properties. The present study investigated whether selegiline facilitates behavioral recovery after focal cerebral ischemia in rats.

Methods. The right middle cerebral artery of rats was occluded for 120 min using the intraluminal filament method. Selegiline (0.5 mg/kg, SC) was administered 30 min before rats housed in enriched-environment had their daily exercise in a labyrinth containing traversable objects, providing both sensorimotor stimulation and spatial exercise. Selegiline was given once a day, beginning on the second day after induction of ischemia and continuing for 30 days. A limb placing test, a foot-slip test and a modified version of Montoya's staircase test and a water-maze were used for evaluation of behavioral deficits.

Results. The selegeline-treated ischemic rats housed in enriched-environment had significantly shorter escape latencies ($P<0.001$) and path lengths ($P<0.005$) than did the ischemic control group. The rats housed in an enriched-environment that received selegiline were more likely to make attempts to reach food pellets with the affected ($P<0.05$) and nonaffected ($P<0.01$) forelimbs. There was no significant improvement in sensorimotor tasks when selegiline treatment was combined with enriched-environment housing. Selegiline treatment without the enriched-environment was not beneficial in any of the behavioral tests.

Conclusions. The present study showed that selegiline treatment combined with training and housing in an enriched-environment diminished a spatial learning disability of ischemic rats.

B16

BEHAVIOURAL DEFICITS IN FORELIMB-USE AFTER PHOTOTHROMBOTIC LESIONS IN RAT CORTEX

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Background. Following focal brain lesions in the rat, substantial alterations, e.g. in neuronal excitability and receptor density, are observed in the surround of the lesion as well as in remote brain areas. It is also known that an aspiration lesion or a electrolytic lesion in the cortex of the rat leads to a clear deficit if the sensomotorical representation of the limbs are effected (Forgie & Kolb 1995). In the present series of experiments we used a battery of behavioural test to examine whether a persistent functional deficit can be monitored following photothrombotic lesion in the sensorimotor cortex of the rat.

Methods. Focal brain lesion with a diameter of about 2 mm were induced by means of a photothrombosis in the forelimb area (FL-SMC), in the motorcortex (Fr1/Fr2) and more caudal brain areas (Occ2/Par1). In the first test we observed the rearing of rats in a glass cylinder. The movements of forelimbs along the wall were counted. In the second task we counted “false steps” during walking on a grid. The results were determined as score for the impaired limb compared to controls.

Results. Animals with lesions in FL-SMC showed functional deficits for the impaired limb or both tests. In the first week after surgery they either used the healthy limb or both together for motion along the wall. These animals also had a high score of false steps on the grid. Within two weeks the deficit considerably recovered. With the lesion in the parietal area, walking on a grid was not impaired. However, a subtle impairment of limb use developed within the first seven days.

Conclusions. The investigations show (i) that clear motor deficits can be demonstrated with focal brain lesions in the sensorimotor cortex, (ii) there is considerable recovery within the first two weeks following lesion induction; and (iii) animals with lesions in parietal areas may show some changes of limb preference indicating that the lesion has some remote behavioural effects.

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INTRAVASCULAR R_2* AS A FUNCTION OF BLOOD OXYGENATION

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Background. MRI based on BOLD (Blood Oxygenation Dependent) effect is a sensitive tool to probe cerebral hemodynamics. Most BOLD-MRI studies employ T_2* contrast changes, measured with gradient echo sequences. In order to quantify physiological parameters from T_2*-weighted signal changes, intravascular relaxation models should be included. At present, most quantification approaches focus on extravascular T_2* effects (1,2). However, the contribution of veins is large (3,4). Thus, it is important to determine what fraction of signal is of intravascular origin and how the intravascular T_2* changes as a function of oxygen saturation (Y) and relates to change in T_2 (5). Transverse relaxation rate in blood has been shown to follow a hyperbolic behavior, $R_2=A+BY+CY^2$, where Y is oxygenation and A, B, C constants (6). A, B and C have been detailed earlier (5,7-8) in terms of plasma and erythrocyte relaxation rates (A, B) and the susceptibility shift difference between plasma and erythrocytes (C). Here we studied intravascular T_2* using isolated bovine blood samples in order to create a model for oxygenation dependence of gradient echo signal from blood.

Methods. Bovine blood samples of various hematocrit fractions (Hct 0.28, 0.44, 0.64) were circulated in a gas exchange system and a 1.5 T magnet. Temperature was maintained at 37 C, and blood oxygenation was manipulated using mixtures of N_2, O_2, each containing 5% CO_2. To avoid cell precipitation, blood was circulated all the time. For measurements, flow in the sample tube was slowed down to 3.1 ml/min (equals to velocity 6.2 mm/min). Shimming was performed on the fully oxygenated sample and kept unchanged throughout the set of oxygenation levels. Water line widths at half peak height were typically a few Hz for oxy blood. T_2's were measured using spectroscopic CPMG (inter-echo spacing τ_{CPMG} 2 ms and 20 ms) and single-echo (SE) sequences (TE 20-200 ms), using a voxel (≈1 ml) in the middle of the sample tube. T_2* was measured from integrals of gradient echo (GE) profiles across the sample tube (TE 20-80 ms; 10 mm slice).

Results. At all hematocrits, both R_2 and R_2* fitted well to the second order polynomial. The data show that the difference, $R_2'=R_2*-R_2$, increases with deoxygenation, as well as with Hct, indicating that additional field-gradient related relaxation mechanisms influence R_2* as deoxygenation increases. However, R_2' is approximately constant at physiological range of oxygenation, Y = 0.55–1.0. This difference is dependent on the quality of the shim, and is thus expected increase in vivo.

Conclusions. We found that the difference between SE and GE relaxation rates is approximately constant at the normal oxygenation range for veins, capillaries and arteries in blood with normal Hct. This suggests that intravascular BOLD changes in this region are dominated by T_2-relaxation changes and that the use of gradient echoes just adds a constant term in the intravascular transverse relaxation rate. This simplifies modeling intravascular gradient echo data. When total relaxation changes, measured from a parenchymal voxel, are analyzed, extravascular effects must be included.

References. 1) Ogawa S and Lee TM et al. Biophys J 64 1993; 2) Yablonskiy DA and Haacke EM MRM 32:746 1994; 3) Haacke EM NMR Biomed. 7:54 1994; 4) Gati JS et al. MRM 38:296 1997; 5) van Zijl PCM et al. Nat Med 4:159 1998; 6) Wright GA et al. JMRI 1:275 1991; 7) Thulborn KR et al. Biochim Biophys Acta 714:265 1982 8) Bryant RG et al. MRM 13:133 1990

B18

DIRECT MICROGLIAL ACTIVATION IN RESPONSE TO IN VITRO EXCITO-TOXICITY

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Background. Excitotoxicity is a common mechanism in several neurodegenerative diseases and may involve inflammatory responses. Because microglia have been demonstrated to express different glutamate receptors, we studied whether stimulation of glutamatergic receptors could directly trigger microglial activation.

Methods. The pure microglial cultures, harvested from rat mixed glial cultures, were used at 3 days after the harvesting and replating. The cells were exposed to 500μM glutamate, its receptor agonists or 10ng/ml lipopolysaccharide (LPS, a classical inducer of inflammation) for 24h and analyzed for release of nitric oxide (NO) and interleukin-1β (IL-1β. Some cultures were treated with 10μM SB203580, a p38 mitogen activated protein kinase (MAPK) inhibitor, or 10μM PD98059, a p44/42 MAPK inhibitor. Activation of MAPKs was studied by immunoblots.

Results. Glutamate and its ionotropic receptor agonists (NMDA and kainate) triggered NO and IL-1β release, microglial proliferation and changes in morphology into activated phenotype. LPS also increased NO and IL-1β release, but prevented proliferation. Glutamate increased p38 MAPK activity and decreased p44/42 MAPK activity. The inhibition of p38 MAPK prevented the microglial responses induced by glutamate, kainate or NMDA. Also, inhibition of p44/42 MAPK alone or together with p38 MAPK inhibition reduced NO release induced by excitotoxins.

Conclusions. It is concluded that glutamatergic stimulation directly contributes to inflammation by activating microglia through a mechanism, which involves both p38 and p44/42 MAPK pathways.

ASPIRIN PROVIDES A Cdk-5-DEPENDENT PROTECTION AGAINST SUBSEQUENT HYPOXIA/ REOXYGENATION DAMAGE IN VITRO

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Background. Kinase pathway signaling is crucial in triggering ischemic preconditioning, a phenomenon where a single or a series of mild insult(s) provide protection against a subsequent, more severe ischemia. *In vitro* experiments have demonstrated a role for protein kinase C and mitogen-activated protein kinase (MAPK) families in the protection mechanism against a later, damaging insult in cardiac and brain systems.

Methods. We studied the effect of aspirin (ASA) pretreatment on neuronal survival and protein kinase pathways after hypoxia/reoxygenation (H/R) damage in rat spinal cord (SC) cultures. 3 mM ASA was added to the cultures for 24 h before a 20-h hypoxia. PD98059, an inhibitor of the p44/42 MAPK pathway, cyclin-dependent kinase 5 (cdk5) inhibitors roscovitine or olomoucine were added to the cultures at the onset of 4-h-reoxygenation.

Results. ASA pretreatment blocked the hypoxia/reoxygenation –induced lactate dehydrogenase (LDH) release and the decrease in the cell number. Administration of PD98059 at the onset of reoxygenation had no effect on the ASA protection. However, the administration of 5-20 μM roscovitine or 50/100 μM olomoucine at this point decreased significantly the neuroprotection gained by ASA pretreatment. Further, ASA pretreatment increased the amount of Cdk5 protein and the Cdk5 kinase activity 2 h after reoxygenation, and this activation correlated with cell survival, suggesting a critical role for Cdk5 activity at the reoxygenation period.

Conclusions. As our previous studies imply that H/R damage in rat SC cultures does not involve cyclooxygenases, inducible nitric oxide synthase or ASA-sensitive NF-κB modulation, ASA may induce tolerance against H/R damage by activating Cdk-5 pathway.

M1

PRENATAL STRESS AND AMYGDALA NUCLEI VOLUMES

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Background. The amygdala is an essential link between limbic and sensory regions of cerebral cortex and subcortical areas that are responsible for producing emotional and motivational responses to fearful environmental stimuli. On the other hand during development organisms are sensitive to numerous influences; exposure at critical times can permanently affect behaviour and metabolism. In rats, offspring of stressed dams show behavioral disturbances indicative of increased fearfulness. Therefore we might expect that prenatal stress in rats affects the development, and consequently, the morphology and the functioning of amygdala.

Methods. By means of unbiased stereological methods, we analyzed the volume of 4 nuclei of the amygdala (basal, central, intercalated, lateral) of prenatally stressed and control rats at the 7^{th}, 25^{th}, 45^{th} and 120^{th} postnatal day of life.

Results. We observed significant differences in the volume of basolateral and lateral nuclei between prenatally stressed and control rats in almost all analyzed age groups of animals. In younger animals the volume was smaller in prenatally stressed groups, but the final effect in adult animals was partially opposite, showing bigger lateral nuclei in prenatally stressed rats. There was no such differentiation in central and intercalated nuclei. Total brain weights, as well as cross-sectional areas of corpus callosum and whole hemispheres, were not influenced by prenatal stress. This suggests no overall brain growth changes between groups.

Conclusions. Presented results might suggest a slower but longer development of the basolateral complex due to prenatal stress, resulting in a significantly bigger lateral nucleus in the final adult stage. However, they also may be a reflection of more complicated ontogenetic processes caused by prenatal stress.

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DISTANCE CRITERION IN THE MORPHOMETRICAL ANALYSIS OF PERI-VASCULAR AXON TERMINALS

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Background. In order to find the morphological uniqueness of perivascular axon terminals (PAT), their differences from the distant axon terminals (DAT) should be shown. But to make such a comparison a basic criterion has to be established for preliminarily dividing the analyzed axon terminals into perivascular and distant groups. Only axon terminals in direct contact with the basement membrane of the capillary wall should be considered as perivascular ones. But this definition is true only in 3-dimensional space. On electron micrographs, the axon terminals are represented only by their profiles and any profile separated by a thin glial process from the capillary wall may come from an axon terminal adjacent to it beyond the examined section plane. Therefore, one has to arbitrarily establish the distance border, inside which the axon terminal profiles will be considered as PAT. On the other hand, some of the PAT profiles could be located quite far from the capillary wall. Consequently another distance criterion has to be made in order to secure the homogeneity of the population of DAT.

Analysis of distance criterion. The proper choice of the distance criteria is not so simple. If the distance border is small some profiles of PAT will be lying too far and therefore will be rejected from perivascular population. The number and size of rejected profiles, depending on the three-dimensional shape of axon terminals, may differently influence the results of morphometrical analysis. On the other hand, if we establish a much wider distance criterion, our population of PAT profiles will not only comprise most of them, but unfortunately it will also include profiles of more distant terminals. Such heterogeneity may of coarse significantly influence the comparison of PAT and DAT. The shape does not seem to be of importance in the case of DAT, where another problem exists. Considering different orientations of the vessel to the cutting plane, the distance from the capillary wall, measured on the electron micrographs, can be bigger than the actual distance in three-dimensional space. This means that the actual distance border for DAT may be shorter than the admitted criterion.

Conclusions. Presented problems concern a very detailed issue in vascular system studies. However, similar problems have to be considered during analyses of any objects differentiated by distance from other structures. The rightness of a distance criterion in the case of PAT seems to be affected primarily by their three-dimensional shape. For DAT, the most important factor seems to be the orientation of the vessel to the cutting plane.

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