

THEME ONE PLENARY

Oedema and the Blood Brain Barrier

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BRAIN EDEMA-THE “LETHAL ENIGMA” OF TBI

R Bullock, T Marmarou, P Fatouros (Richmond, USA)

Brain Swelling and high ICP kills about 80% of those who die after severe Traumatic Brain Injury (TBI), although the major focus of therapy, in the ICU is control of ICP. Until the causes of this swelling are understood, therapy will be symptomatic, and less effective. Previous studies with cold injury models implicated vasogenic edema as the main mechanism, but newer studies, in patients, and models, now show that early swelling is mainly cytotoxic edema, due to astrocyte swelling. “Ionic leakage” due to steric conformational changes in channels, and failure of Na/K dependent ATPase are major causes, and may reflect mitochondrial failure, and ATP depletion, in neurons, and astrocytes. Aquaporin changes may also be important. Astrocyte swelling also reduces capillary lumina, and impairs local CBF, causing a “vicious cycle”, leading to neuronal death, by early necrosis, or apoptosis. Around focal lesions (contusion, SDH, ICH) early cytokine activation, and endogenous complement activation, leads to blood brain barrier opening, vasogenic edema, and macrophage/monocyte activation, leading to delayed neuron loss, due to apoptosis, and microglial upregulation. Therapies therefore need to be specific, to the causes, and may include ion channel blockers, aquaporin inhibition, and mitochondrial protection, as well as anticytokine, and anticomplement strategies.

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THE PATHOPHYSIOLOGIC BASIS FOR TREATING TRAUMATIC BRAIN EDEMA: NOVEL CONCEPTS

A Marmarou (Richmond, USA)

For several decades, the concept of traumatically induced brain edema and swelling has been focused on the compromise of the blood brain barrier and production of a vasogenic form of edema. It is this concept that has been challenged in the past several years by our laboratories and others. More recent data has indicated that the barrier closes rapidly in traumatic brain injury and the swelling that ensues is primarily due to a cellular edema. The cause of cellular swelling may be due to neurotoxins causing cellular membrane breakdown and subsequent ionic disruption or an energy crisis. The energy crisis may be the end result of reduced cerebral blood flow or in the presence of adequate flow, a mitochondrial impairment. Studies in TBI patients have shown that both may co-exist. Having demonstrated that a cellular edema is mainly responsible for brain swelling, we have focused our efforts in the laboratory in identifying the pathways for water entry. Data has been obtained which obviates an extracellular route for water and solute entry into the cell and we have concluded that excess water enters brain via the astrocytic endfeet. This implicates the Aquaporins, specifically AQP4, which we believe plays a vital role in water regulation following trauma. This presentation will summarize the results of our investigation into the pathways for fluid entry and the means of modulating the AQP4 site to block water entry to prevent brain swelling.

0 - 3**NEW DEVELOPMENTS IN THE PHARMACOLOGIC MANAGEMENT OF POSTTRAUMATIC OEDEMA***R Vink (Adelaide, Australia)*

A major risk factor contributing to increased mortality and morbidity following traumatic brain injury (TBI) is oedema formation. A recent analysis of TBI in children concludes that over 50% of these young patients develop diffuse brain swelling/oedema, resulting in death in almost half of this group [1]. Interventional strategies to date have utilized a variety of physiological and surgical approaches, largely because the mechanisms of oedema formation are unclear and pharmacological approaches undeveloped. Recent efforts in our laboratory, and others, have concentrated on establishing the mechanisms associated with oedema formation following brain injury, and developing appropriate pharmacologic interventional therapies. Our own studies to date have shown that neurogenic inflammation is a major contributor to brain oedema formation. Neuropeptides are released after TBI and the binding of the neuropeptides to their receptors results in an increased blood brain barrier permeability and oedema formation (vasogenic followed by cytotoxic). Attenuating the neurogenic inflammation, either by preventing release of the neuropeptides or blocking receptor binding, profoundly inhibited these events. There was also an improvement in functional outcome, which correlated with a reduction in neuronal cell death. Our results suggest that pharmacological intervention targeting oedema formation may be a promising strategy for oedema management in the near future.

Reference

- [1] H.J. Feickert, S. Drommer and R. Heyer, *J. Trauma* **47** (1999), 33-38.

SESSION 1.1: Inflammation**0 - 4****INFLAMMATION - BLOOD-BRAIN BARRIER - BRAIN OEDEMA***A Baethmann, N Plesnila, J Schulz, D Pruneau, M Stoffel, J Eriskat (Munich, Germany)*

Opening of the blood-brain barrier causing brain edema is particularly threatening in brain trauma and

infarction. As a space occupying process edema raises intracranial pressure and impairs blood flow to the brain already in jeopardy. It is not clear yet, if inflammation plays a causal role in this context. Yet, mediator agents of secondary brain damage have inflammatory properties, as e.g. the peptides of the kallikrein-kinin system (KK). This laboratory has intensely studied the KK-system in brain trauma and ischemia. The requirements defining a mediator agent of secondary brain damage are all valid for the KK-system. Respective conclusions may apply for NO, as inhibition of iNOS attenuates the loss of parenchyma from trauma.

An intriguing question is, whether under these circumstances leukocyte activation in cerebral blood vessels is causally involved in the damage of the blood-brain barrier and brain edema formation. Although, leukocyte-endothelial interactions (LEI) are observed in brain trauma and ischemia, their inhibition not always confers protection. E.g. neutropenia was associated with enhancement rather than inhibition of edema from a focal lesion. Inhibition of LEI most likely is not beneficial in global or permanent focal cerebral ischemia, while more likely in transitory interruption of the focal cerebral flow. Whereas evidence indicates that inflammation occurs in acute brain lesions, it is not clear whether this contributes to the secondary loss of brain tissue, or just represents a response to the necrosis formation in the brain.

0 - 5**INFLAMMATION AFTER TRAUMATIC BRAIN INJURY: RESEARCH MUST GO ON.***MC Moganti-Kossmann (Melbourne, Australia)*

The decade between 1990 and 2000 was particularly fruitful for studies that explored the role of neuroinflammation following traumatic brain injury (TBI), beginning from detection of cytokines in the brain up to the application of therapeutical strategies with the objective to suppress cytokine action. However, from the beginning of the new decade there has been a continuous decrease in the number of publications in this field of neurotrauma research. Why? Is it due to the conflicting results on cytokine function produced in the brain tissue? What is needed to reconcile the controversial data on cytokine function derived by classical studies on animal models and the application of TBI to cytokine knockout mice? Is this a conceptual or experimental related issue? Has the dichotomy intrinsic of inflammation been sufficient to discourage neuroscientists to pursue such avenue of research?

In this talk, the evolution of the neuroimmunology research of TBI will be reviewed and confronted in regard to the various approaches utilised in experimental and clinical studies. As a double-edged sword, inflammation can play its dual role contributing to neuronal survival and neurogenesis but also initiate neuronal cell apoptosis. Research must go on.

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INTERLEUKEN-1 RECEPTOR ANTAGONIST IMPROVES TRAUMATIC BRAIN INJURY OUTCOME

AN Taylor, JK Bando, O Oluwadara, SU Rahman, HE Romeo, MJ Sanders, DL Tio (Los Angeles, USA)

Cytokines mediate secondary injury after brain insult, and interleukin-1 receptor antagonist (IL-1ra) has been found to be neuroprotective. We investigated the effects of IL-1ra on body temperature, locomotor activity, body weight, and neurobehavioral and histologic outcomes in rats after traumatic brain injury (TBI) by cortical contusion. Rats were injected sc with 100 mg/kg IL-1ra (Amgen, Inc., Thousand Oaks, CA) or PBS immediately after TBI (day-0) and until day-6 post-TBI. TBI induced AM and PM hyperthermia, with elevated AM body temperatures returning to pre-TBI levels at day-5, while PM temperatures remained elevated. IL-1ra significantly ($p < 0.05$) reduced TBI-induced hyperthermia at 2–7 hours after AM injections on post-TBI days 0, 4 and 5. Activity was unaffected by IL-1ra. TBI-induced weight loss was significantly ($p < 0.05$) less in the IL-1ra group. IL-1ra enhanced learning of the Morris water maze (MWM): Beyond a significant ($p < 0.05$) reduction in latency to reach the hidden platform between days 1–2 of testing in both groups (daily testing commenced 1 week post-TBI), a significant reduction occurred between days 2–3 in the IL-1ra group but not until days 4–5 in the PBS group. On average, animals in both groups achieved MWM criterion at 7 days of testing. The effect of IL-1ra on the extent of neuronal damage and apoptosis is currently being assessed. These findings suggest that IL-1ra neuroprotection is mediated by its anti-inflammatory action within the first week post-TBI. (Supported by Department of Veterans Affairs Medical Research Service and UCLA Brain Injury Research Center; JKB: UCLA Undergraduate Research Scholar; OO, MacArthur Foundation Fellow, University of Ibadan, Nigeria)

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MATRIX METALLOPROTEINASES ASSOCIATED WITH NEUROGENIC INFLAMMATION AND SUBSEQUENT BRAIN EDEMA FORMATION AFTER TRAUMATIC BRAIN INJURY

Y Katayama, T Kawamata, T Mori (Japan)

The final outcome of traumatic brain injury (TBI) is crucially determined by the extent of secondary damages mediated by excitotoxicity, oxidative stress, apoptosis and inflammatory response. However, the precise mechanisms of these cascades remain unclear. Recent studies have suggested that matrix metalloproteinases (MMPs) increase after cerebral ischemia, TBI, and neurodegenerative disorders. These proteins might play critical roles in inflammatory responses such as degradation of extracellular matrix, disruption of the blood-brain barrier, facilitation of leukocyte infiltration, etc. MMPs are also induced by cytokines which are produced with inflammation. The inhibition of such cascades may therefore, provide therapeutic effects on TBI. In the present study, we focus on polymorphonuclear leukocyte accumulation and MMP-9 upregulation in cerebral contusion, which leads to inflammatory response, subsequent edema formation and secondary cellular damages. The effects of MMP inhibitors on TBI-induced secondary brain damages are also discussed.

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TIME COURSE OF MICROGLIAL RESPONSE TO TRAUMATIC AXONAL INJURY

LM Christian, PD Leclercq, B O'Dwyer, C Smith, D Graham, SM Gentleman (London, Edinburgh, Glasgow, UK)

Traumatic axonal injury (TAI) is a common pathological consequence of severe head injury as a result of road traffic accidents, falls and assaults. It is believed that although there may be some limited physical axonal transection at the time of injury much axonal damage arises as the result of delayed secondary mechanisms. Microglial cells are ubiquitously expressed throughout the CNS and are capable of mounting a rapid response to an array of pathological events. A diffuse microglial reaction following blunt head injury is well documented, and it has been suggested that this population of functionally diverse cells could act as effectors of the secondary damage to axons. A number of previous studies have made qualitative observations of

this cellular response but there is some disparity in the precise timings and in the specificity of this response to damaged axons. In this study we have undertaken a detailed quantitative analysis of the microglial reaction to TAI.

We have studied 90 cases of fatal non-missile head injury with a range of post-injury survival times, supplied by the Institute of Neurological Sciences, Glasgow. Immunostaining was carried out on serial sections with antibodies against amyloid precursor protein (APP), CR3/43 (an MHC class II marker) and CD68. Quantitative analysis of immunostain loads was carried out in the lateral corpus callosum and parasagittal white matter. Initial results suggest that there is a biphasic microglial response associated with TAI. At short survival times, when there was thought to be relatively little microglial response, we observed a profound increase in the number of CR3/43 immunoreactive cells. With longer survival there appears to be a shift towards a predominance of CD68 immunoreactive cells. Furthermore this microglial response appears to persist even in those cases with survival times of many months. Double immunostained sections revealed a close association between the microglial markers and the injured axons

SESSION 1.2: Vascular Injury

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ROLE OF ANGIOGENIC FACTORS IN BLOOD-BRAIN BARRIER BREAKDOWN FOLLOWING BRAIN INJURY

S Nag (Toronto, Canada)

The role of the endothelial specific agents-vascular endothelial growth factors (VEGF) -A and B and the angiopoietins (Ang) 1 and 2, in blood-brain barrier (BBB) breakdown was investigated in the rat cortical cold-injury model, a well characterized model for BBB studies. VEGF-A is a known potent inducer of increased vascular permeability and angiogenesis in non-cerebral vessels while the Angs function as ligands for the endothelial-specific receptor tyrosine kinase, Tie-2. Ang-1 has a major role in vascular stabilization and maturation and a potent anti-leakage effect in non-neural vessels, whereas Ang-2 can act as a Tie-2 antagonist and block the effects of Ang-1. Normal cerebral vessels show constitutive expression of VEGF-B and Ang-1 proteins while VEGF-A or Ang-2 proteins are not detectable. Following injury, BBB breakdown

in lesion vessels is associated with decreased localization of VEGF-B and Ang-1 proteins while there is a concomitant increase in VEGF-A and Ang-2 mRNA and proteins raising the question whether Ang-2 like VEGF-A can induce BBB breakdown? Studies using tracers demonstrate that VEGF-A increases BBB permeability to HRP in the normal rat cortex and the novel finding is that Ang-2 has a similar effect. Thus, VEGF-B and Ang-1 are important in the maintenance of cerebral homeostasis in steady states while VEGF-A and Ang-2 have the opposite effect and promote BBB breakdown following injury. Understanding the mechanisms leading to BBB breakdown have therapeutic implications because of the potential of attenuating BBB breakdown by the delivery of factors that have an anti-leakage effect on cerebral endothelium.

O-10

CHANGES IN BRAIN BARRIER PERMEABILITY FOLLOWING FOCAL TRAUMATIC INJURY TO THE BRAIN

NR Saunders, N Bye, KM Dziegielewska, CJ Ek, MD Habgood, A Potter, C Morganti-Kossmann (Melbourne, Australia)

Barrier mechanisms in cerebral vasculature (blood-brain barrier) and choroid plexus epithelial cells (blood-CSF barrier) determine and control composition of the internal environment of the brain and spinal cord. The underlying mechanism for these barriers is the diffusion restraint provided by tight junctions between cells forming these interfaces. Superimposed on this diffusion restraint is a series of influx and efflux mechanisms in these cells.

Understanding effects of trauma on these processes and their time course allows distinction between damaging and beneficial effects of barrier damage. Also determination of time course of increased barrier permeability may identify a "window" when drugs that do not normally cross the blood-brain barrier might do so. Most published studies have concentrated on passive markers of barrier permeability, particularly horseradish peroxidase (value of which may be limited by toxic effects). Less is known about small lipid insoluble molecules and nothing seems known about effects of injury on influx or efflux mechanisms. Our group is undertaking a systematic study of barrier permeability in a mouse model of cerebral contusion using inert lipid insoluble small and large molecules that can be both visualised and quantified. An essential pre-requisite of

such a study is that reasonably consistent lesions can be made and their size and the spread of any marker estimated (see Habgood et al poster, this meeting).

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ORIGIN, PATHOGENESIS, AND FATE OF
CHRONIC SUBDURAL HEMATOMA

KS Lee (Chonan, Korea)

The origin, pathogenesis, and natural history of chronic subdural hematoma (SDH) are controversial issues. The author will clarify them by a literature review. Chronic SDH has dual origins, from subdural hygromas (SDG), or from acute SDH. Transformation from these lesions requires pre-morbid condition, i.e., sufficient potential subdural space (PSS). SDG is produced by separation of the dura-arachnoid interface, when the PSS is sufficient. SDG usually occurs at the least pressure in the cranium as a lesion of ex vacuo. When the brain remains shrunken, the SDG remains unresolved. Any pathologic conditions inducing cleavage of tissue within the dural border layer at the dura-arachnoid interface can induce proliferation of dural border cells with production of neomembrane. In-growth of new vessels will follow. These vessels are usually fragile, and easy to bleed. Unresolved SDGs become chronic SDHs by repeated microhemorrhages from these vessels, the same mechanism of the hematoma enlargement. When rebleeding exceeds absorption, chronic SDHs enlarge to become symptomatic. Although most victims with acute SDH underwent surgery or died, some patients could be managed conservatively. Since the acute SDH is usually absorbed within a few weeks, only a very few acute SDHs become chronic SDHs, when the PSS is sufficient. When the neomembrane is matured, the neocapillary is no longer fragile. If absorption exceeds rebleeding, the hematoma will disappear. Maturation of the neomembrane and stabilization of the neovasculature eventually result spontaneous resolution. The fate of chronic SDH depends on the pre-morbid status, the dynamics of absorption-expansion, and maturation of the neomembrane.

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MEDICAL THERAPY FOR POST-TRAUMATIC
ICP INCREASE USING THE NOVEL NITRIC
OXIDE SYNTHASE INHIBITOR VAS203

N Terpolilli, K Zweckberger, R Doblhofer, F Tegtmeier, N Plesnila (Munich; Würzburg, Germany)

Uncontrollable increase of ICP is one of the major causes of death following TBI. We investigated whether the novel NOS-inhibitor VAS203 counteracts pathological vasodilatation after TBI and thereby lowers ICP.

For dose finding C57/Bl6 mice subjected to CCI received increasing single doses of VAS203 (1–500 mg/kg i.v. or vehicle; $n = 4$ per group). Other animals received VAS203 or vehicle continuously for 3 h after trauma (25, 50, 100, or 300 mg/kg; $n = 8$ per group). ICP was measured at 3 and 24 h. In a parallel group (300 mg/kg) ICP, rCBF, MAP, and blood gases were monitored continuously for 2 h. Mice were sacrificed 24 h after TBI for quantification of contusion volume. VAS203 reduced ICP at doses < 100 mg/kg, but induced CBF, MAP, and ICP peaks at 100–500 mg/kg. Continuous infusion of VAS203 at doses not influencing CBF and MAP (1.5 or 0.5 mg/kg/min) for 3 h starting 30 min after CCI reduced ICP 3 h after trauma from 14.4 ± 1.9 mmHg (control, mean \pm SD) to 10.7 ± 2.5 and 10.5 ± 1.1 mmHg (-26% ; $p < 0.01$), respectively. In the animals receiving 1.5 mg/kg/min this effect was still present 24 h after trauma (7.4 ± 2.3 mmHg vs. 12.1 ± 2.0 mmHg; -40% ; $p < 0.01$). VAS203 had no effect on contusion volume. Our results demonstrate that the NOS inhibitor VAS203 reduces ICP after experimental brain injury in mice at doses not influencing CBF, MAP, and histopathological outcome. The proposed mechanism of action is the increase of intracranial compliance by mild constriction of pathologically dilated vessels. Accordingly, VAS203 may serve as a future compound for the medical therapy of increased ICP following TBI.

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CHRONIC SUBDURAL HEMATOMA IN THE
ELDERLY

Y Shigemori, Y Katayama, T Mori, T Maeda, T Kawamata (Tokyo, Japan)

Chronic subdural hematoma (CSDH) is predominantly a neurosurgical disease of the elderly. The diagnosis and treatment have already well established, however complications and prognostic factors are not fully

understood in the elderly. In the present study, to clarify the magnitude of the factors what determine prognosis of these patients, we retrospectively analyzed in 697 CSDH patients who had been operated in our hospital from 1976 to 2003. 384 patients (55.1%) were older than 65 years or more, and 210 patients (30.1%) were older than 75 years or more. Advanced age tended to increase the morbidity rate but did not reach statistical significance. 301 patients (78.3%) had good recovery, while 72 patients (18.8%) had unfavorable outcome. 11 patients (2.9%), died and 10 of 11 was due to general complications. Older than 75 years or more patient mobility is clearly poor compared with less than 75 years. Poor prognostic factors were moderate consciousness disturbance ($P < 0.001$) and walking disability persisted over one-week ($P < 0.001$) when patients admitted to the hospital. The elderly patients with CSDH have gradually increased in recent years. The deterioration of existing general diseases caused the poor prognosis for CSDH in the elderly. The present results demonstrated that neurological states at the time of diagnosis contribute to the prognostic factor. These finding suggest that early diagnosis and treatment are important in the elderly patients with CSDH.

SESSION 1.3: Neuronal/Glial Interactions

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ROLE OF GLIA AND EXTRACELLULAR MATRIX IN VOLUME TRANSMISSION

Eva Sykova (Prague, Czech Republic)

Synaptic transmission is an important means of communication in the CNS; however, neurons themselves and neurons and glia also communicate by extrasynaptic "volume" transmission, which is mediated by diffusion in the extracellular space (ECS). The ECS of the central nervous system (CNS) is the microenvironment of neurons and glial cells. The composition and size of the ECS change during neuronal activity as well as during pathological states. Following their release, a number of neuroactive substances, including ions, mediators, metabolites and neurotransmitters, diffuse via the ECS to targets distant from their release sites. Glial cells affect the composition and volume of the ECS and therefore also extracellular diffusion, particularly during development, aging and pathological states such as ischemia, injured tissue, X-irradiation, tumors, demyelination and often in grafted tissue. Our studies also indicate that diffusion in the ECS is affected by

ECS volume inhomogeneities, which are the result of a more compacted space in certain regions. Besides glial cells, the extracellular matrix also changes ECS geometry and forms diffusion barriers, which may result in diffusion anisotropy. ECS size, geometry, and content together with pre- and postsynaptic terminals and glial processes form the so-called "quadrupartite synapse". ECS diffusion parameters affect neuron-glia communication, cell migration, and the movement and accumulation of neuroactive substances in the brain. ECS size and geometry therefore play an important role in extrasynaptic transmission, transmitter spillover, cross-talk between synapses in functions such as vigilance, sleep, depression, chronic pain, LTP, LTD and memory formation, and in cell migration and fate.

0 - 15

CELLULAR AND EXTRACELLULAR MATRIX REACTIONS TO CNS DAMAGE

James Fawcett (Cambridge England)

Whenever the CNS is injured a reactive process is initiated known as glial scar formation which acts as a barrier to the regeneration of damaged axons. The main inhibitory molecules in the glial scar are chondroitin sulphate proteoglycans (CSPGs), most of which have axon growth inhibitory properties, and are upregulated after CNS injury. The glycosaminoglycan chains (GAGs) are responsible for much of the inhibition. The final stage of GAG synthesis is sulfation, which can occur in three positions, mediated by seven sulfotransferases. 6-sulfated GAG, which is particularly inhibitory to axon growth is upregulated after injury. All CSPGs possess GAG chains and these can be removed by chondroitinase. We therefore tested to see whether GAG digestion by chondroitinase would promote axon regeneration *in vivo*. We first treated mechanical lesions of the nigrostriatal tract, and saw regeneration of about 4% of axons back to their target. Next dorsal column lesions of the spinal cord at C4 were treated. Both sensory and corticospinal axons regenerated in treated cords, and there was rapid return of function in beam and grid walking tests. CSPGs are also involved in the control of plasticity by coating many neurones and their dendrites in perineuronal nets. Treatment of the visual cortex of adult animals can reactivate ocular dominance plasticity, which normally terminates at the end of the critical period. The molecules responsible for the construction of these nets around neurones are being investigated.

0 - 16**AQUAPORIN WATER CHANNELS: ROLE IN CEREBRAL EDEMA AND BRAIN WATER BALANCE**

GT Manley, H Watanabe, DK Binder, MC Papadopoulos, AS Verkman (San Francisco, USA)

Aquaporin-4 (AQP4) is the major water channel in the central nervous system. Its expression at fluid-tissue barriers (blood-brain and brain-CSF barriers) throughout the brain and spinal cord suggests a role in water transport under normal and pathological conditions. Phenotype studies of transgenic mice lacking AQP4 have provided evidence for a role of AQP4 in cerebral water balance. Primary cultures of astrocytes from AQP4-null mice have greatly reduced osmotic water permeability compared to wild-type astrocytes, indicating that AQP4 is the principal water channel in these cells. AQP4-null mice have reduced brain swelling and improved neurological outcome following water intoxication and focal cerebral ischemia, establishing a role of AQP4 in the development of cytotoxic (cellular) cerebral edema. In contrast, brain swelling and clinical outcome are worse in AQP4-null mice in models of vasogenic edema caused by freeze-injury, probably due to impaired AQP4-dependent brain water clearance. Pharmacological modulation of AQP4 function may thus provide a novel therapeutic strategy for the treatment of traumatic brain injury, stroke, and other disorders of the central nervous system associated with altered brain water balance.

0 - 17**PERSISTENT CYTOTOXIC OEDEMA AFTER ISCHAEMIC STROKE IN THE CONTEXT OF THE GLIAL - NEURON UNIT: FROM A MAGNETIC RESONANCE VIEWPOINT**

H Rumpel, CL Ling, WEH Lim (Singapore)

This presentation reviews the persistence of cytotoxic oedema after stroke as demonstrated by diffusion-weighted MRI and MR spectroscopy and discusses the possible mechanisms of cooperative volume effects within the glial - neuron unit. The study comprised 22 patients with stroke in the territory of the middle cerebral artery examined only once at a time ranging from

eight hours to six days following the onset of symptoms. The evolution of both the apparent diffusion coefficient and the glial-specific marker myo-inositol were assessed, and the results were compared with the total cellular density by means of the creatine level.

0 - 18**ROLES OF ASTROCYTE REACTIVITY AFTER CRUSH SPINAL CORD INJURY**

JE Herrmann, JR Faulkner, MJ Woo, MD Sislak, MV Sofroniew (Los Angeles USA)

Reactive astrocytes are prominent in the cellular response to spinal cord injury (SCI), but their roles are incompletely understood. We are using different transgenic mouse models to study the functions of astrocyte reactivity after crush SCI. In this study, we compare the effects of ablating reactive astrocytes with the effect of attenuating astrocyte reactivity. To ablate reactive astrocytes, mice expressing a glial fibrillary acidic protein-herpes simplex virus-thymidine kinase (GFAP-HSV-TK) transgene were treated with the anti-viral agent ganciclovir (GCV) after SCI. Moderate crush injuries in control mice caused focal tissue disruption and cellular degeneration, with mild and largely reversible motor impairments. Equivalent moderate crush injuries combined with ablation of reactive astrocytes caused pronounced cellular degeneration, inflammation, and severe, essentially permanent motor deficits. To attenuate astrocyte reactivity, we deleted the STAT3 gene selectively from astrocytes using the Cre-loxP system. STAT3 is an intracellular signal transducer thought to play a role in regulating the response to injury in a variety of cell types. Astrocytes deficient in STAT3 signaling exhibited reduced expression of GFAP and attenuated reactivity after SCI. Moderate crush injuries caused cellular degeneration greater than that observed in non-transgenic mice but less than that in mice with reactive astrocyte ablation. Mice with STAT3 deficient astrocytes exhibited more recovery of motor function after SCI than did mice with astrocyte ablation, but did not recover as well as non-transgenic mice. Taken together, these findings suggest that reactive astrocytes exert more protective functions than non-reactive astrocytes after SCI.