newborn Wistar rats, exposed to LPS at p3, were treated with rhTSG-6 i.p. (four repetitive doses of 2.25mg/kg every 12h, first dose three hours before LPS-injection).

Results Investigations of TSG-6's developmental brain expression showed a linear increase from p1 to p15 on gene level. Additionally, different expression was detected in Cortex, Thalamus and Striatum on gene level at p6. Expression of TSG-6 after LPS treatment (0–24h) was significantly increased on gene level and tendentially on protein level. cCaspase-3, a marker of apoptosis, showed a significant down-regulation of >30% under additional TSG-6 treatment versus sole LPS exposure (n=12–14, p=0,025).

Conclusions TSG-6 Expression is developmentally regulated and increased after LPS exposure. The reduction of activated Caspase-3 demonstrates the neuroprotective potential of exogenous TSG-6 administration in inflammatory-induced developmental brain injury.

310 METHEMOGLOBIN DETERMINES DEGREE OF PRO-INFLAMMATION AND CHOROID PLEXUS APOPTOSIS FOLLOWING INTRAVENTRICULAR HEMORRHAGE IN PRETERM RABBIT PUPS

doi:10.1136/archdischild-2012-302724.0310

S Sveinsdottir, MG Olsson, M Cinthio, M Mögelin, B Åkerström, D Ley. *Lund University, Lund, Sweden*

Background and Aims Inflammation following intraventricular hemorrhage (IVH) has been shown to cause periventricular brain damage. Released free hemoglobin may be an early up-stream initiator of inflammation in the intraventricular space and cause damage to the choroid plexus. We aimed to evaluate release of free hemoglobin, the relationship to inflammation in the intraventricular space and effects on choroid plexus epithelium following IVH.

Methods IVH was induced in preterm rabbit pups. Intraventricular samples of cerebrospinal fluid (CSF) were obtained under guidance of high-frequency ultrasound at 24, 48 and 72h following IVH for measurement of free oxyhemoglobin (OxyHb), methemoglobin (MetHb) and TNF α . Analysis of gene expression (RT-PCR) for TNF α , IL-1 β and heme-oxygenase-1 (HO-1) and immunostaining with electronmicroscopy (EM) was performed in choroid plexus obtained at 72h.

Results Intraventricular CSF concentrations of free OxyHb remained stable whereas those of MetHb and TNF α increased from 24 to 72 h. There was a highly positive correlation between concentrations of MetHb and TNF α in CSF at 72h (r= 0.98, p<0.0001). Levels of mRNA for HO-1, IL-1 β and TNF- α were up-regulated in choroid plexus following IVH as compared to controls (all p<0.01). EM showed choroid plexus epithelium with fragmented microvilli, apoptotic bodies, swollen mitochondria and increased epithelial expression of TNF α following IVH (fig. 1).

Abstract 310 Figure 1

Conclusions Amount of released free Hb, subsequently auto-oxidised to MetHb determines degree of pro-inflammation in intraventricular CSF following IVH. This is associated with severe apoptosis in choroid plexus epithelium.

311 THE SIGMA-RECEPTOR-AGONIST 4-PHENYL-1-(4-PHENYLBUTYL) PIPERIDINE (PPBP) DECREASES NEONATAL EXCITOTOXIC BRAIN INJURY AND DIMINISHES MICROGLIAL CELL ACTIVATION

doi:10.1136/archdischild-2012-302724.0311

K Wegleiter, N Lippert, A Posod, V Neubauer, M Urbanek, U KiechlKohlendorfer, E Griesmaier. *Paediatrics IV, Medical University Innsbruck, Innsbruck, Austria*

The pathogenesis of perinatal brain injury is multifactorial. In animal models of adult brain injury the sigma-1-receptor agonist 4-phenyl-1-(4-phenylbutyl) piperidine (PPBP) was shown to be neuroprotective due to modulation of inflammatory and apoptotic pathways and affection of excitotoxic signaling cascades.

In the current study we evaluated the effect of PPBP in a neonatal animal model of excitotoxic brain injury.

We performed an intracranial injection of ibotenate in CD1 mice on postnatal day 5 (P5) to induce excitotoxic injury. A single injection of vehicle or PPBP, in a dosage of 1, 5 or 50 μ g/g body weight, was applied intraperitoneally one hour after the insult. Primary study endpoints were evaluated 24 (P6) and 120 (P10) hours after the insult by analysis of lesion size, number of cells stained positively for cleaved caspase-3 and isolectin B4.

Systemic application of PPBP resulted in a significant reduction of lesion size in cortical gray matter in all dosages (p<0.05). and underlying white matter in 1 and 50 μ g/g (p<0.05) at P10. At P6 microglial cell activation was significantly diminished by PPBP in cortical gray (p<0,001) and underlying white matter (p<0.05). Lesion size on P6 and number of cells stained positively for activated caspase-3 at P6 was not affected by the treatment (p>0.05).

In this neonatal animal model we were able to demonstrate a beneficial effect of the sigma-1 receptor agonist PPBP. This data suggest that PPBP may be a potential therapeutic approach in neonatal brain injury.

312 INTERMITTENT HYPOXIA: EFFECTS ON BRAIN STEM OF OXIDATIVE STRESS AND NRF2 TRANSCRIPTION FACTOR ACTIVATION IN A RAT PUP MODEL

doi:10.1136/archdischild-2012-302724.0312

M Vento. *Division of Neonatology, University and Polytechnic Hospital La Fe, Valencia, Spain*

Background Apnea of prematurity is a common condition in the neonatal period caused by immature brainstem respiratory neural output. Apneic episodes and resultant intermittent hypoxia are a potential cause of oxidative stress during this vulnerable developmental period.

Objective We tested the hypothesis that chronic intermittent hypoxia alters oxidative metabolism and resultant redox status in the medulla of rat pups.

Design/methods Litters of 10 rat pups and their dams were assigned to one of two groups: normoxia (controls) and intermittent hypoxia (Hx). Exposure occurred from postnatal day 1 for 7 days. Chronic intermittent hypoxia consisted of exposing rat pups and their dams to alternating cycles of nitrogen (N $_2$) and room air (RA): 45 seconds of hypoxia (nadir of 5% O $_2$) was administered every 5 minutes for 8 hours/day. For controls, animals were kept at RA. On the eighth day, brainstems were harvested and quickly snap frozen in liquid nitrogen. Reduced (GSH) and oxidized (GSSG) glutathione, and the GSH precursors -glutamyl-cysteine (-G-cysteine)