inmunostained, were assigned a relative value from 0 (minimal staining) to 4 (most intense inmunostaining). Quantitative morphometric assessment was done on coded slides with 400x magnification and a eye piece with a sample square grid pattern (model CPLW 1018, Zeiss Optical, Hannover Md) and was done following the mathematical model of Weibel. Differences between the groups were determined by one way ANOVA ($p < 0.01$).

Results: The VEGF was significantly decreased in the lungs of rats recovered in hyperoxia, it was correlated with a lower degree of alveolarization. Inhaled NO treatment after hyperoxia neither increase lung VEGF expression nor alveolarization.

Conclusion: The inhaled NO did not improve the changes observed in rat lungs after hyperoxia exposure.

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MATRIX METALLOPROTEINASE 2, TIMP-1 AND TIMP-2 CONCENTRATIONS IN TRACHEAL ASPIRATE FLUID AND PLASMA OF PRETERM INFANTS DEVELOPING BRONCHOPULMONARY DYSPLASIA

M. Gach[1], M. Rybak-Krzyszkowska[2], W. Fendler[3], R. Lauterbach[4], A. Piotrowski[1]

[1]Pediatric Anesthesiology and Intensive Care Department, Medical University of Lodz, Lodz, [2]Department of Obstetrics and Perinatology, Jagiellonian University Medical College, Krakow, [3]Department of Immunopathology and Genetics, Medical University of Lodz, Lodz, [4]Department of Neonatology, Jagiellonian University Medical College, Krakow, Poland

Background and aims: Bronchopulmonary Dysplasia (BPD) is one of the most important complications of prematurity. MMPs are a group of proteolytic enzymes involved in lung development. Imbalance between MMPs and their inhibitors has been implicated in increased inflammation and impaired alveolar differentiation and maturation. MMP-2 is known to play a crucial role in this process. The aim of this study was to investigate the potential association between levels of MMP-2, TIMP-1, TIMP-2 and the development of BPD in preterm infants.

Methods: 27 preterm neonates with a gestational age of ≤32 weeks and birth weight ≤1500g were included in this prospective study. 14 neonates developed BPD according to NIH criteria. TAF aspirates were collected on day 1-2 and 5 after birth. Plasma concentrations were measured in umbilical cord blood samples. MMP-2, TIMP-1 and TIMP-2 concentrations were assayed by ELISA.

Results: In the group of neonates with BPD or death initial levels of examined proteins were significantly higher compared to non-BPD preterms. Additionally, in preterm infants TAF levels of MMP-2, TIMP-1 and TIMP-2 undergo dynamic changes in the first 5 days of live. Significantly lower levels of MMP-2 and TIMP-2 on day 5 compared to day 1-2 were associated with development of BPD or death. Umbilical cord blood plasma concentrations of MMP-2, TIMP-1 and TIMP-2 did not differ between groups.

Conclusions: Determination of TAF concentrations of MMP-2 and TIMP-2 and observation of their changes in the first days of life in preterm neonates is of prognostic value for development of BPD.

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RECOMBINANT HUMAN KGF/PALIFERMIN® AS AN ALTERNATIVE TO GLUCOCORTICOIDS-EFFECTS ON PNEUMOCYTE PROLIFERATION AND GENE EXPRESSION IN NEONATAL RATS

W. Bernhard[1], R. Koslowski[2], H. Fehrenbach[3], C.F. Poets[1]

[1]Neonatology, Eberhard-Karls-University, Tübingen, [2]Physiologic Chemistry, Univ. Dresden, Dresden, [3]Dept. of Experimental Pneumology, Laboratory of Applied Molecular Biology and Immunologyeibniz Center for Medicine and Biosciences, Borstel, Germany

Background: Surfactant decreases surface tension of pulmonary air:liquid interfaces. It mainly comprizes characteristic phosphatidylcholine (PC) species and proteins SP-A to -D. Betamethasone used to accelerate „lung maturation“ is catabolic, while keratinocyte growth factor (KGF), expressed by fibroblasts, is non-catabolic, acts specifically on type-II-pneumocytes and correlates inversely with bronchopulmonary dysplasia incidence.

Aims: To explore the potential of recombinant human KGF (rhKGF, Palifermin®) on neonatal lung development and surfactant metabolism. To contrast rhKGF with betamethasone effects *in vivo*.

Methods: Postnatal rats (d1,d5,d19) were injected with rhKGF (2x5mg/kg), betamethasone (2x1mg/kg) or rhKGF+betamethasone over 48h. Pneumocyte

proliferation was detected using bromodeoxyuridine incorporation. Expression of SP-A to -D, adipocyte triglyceride lipase (ATGL), lyso-PC-acyltransferase (LPCAT) and the ATGL activator CGI-58 were measured with PCR.

Results: RhKGF, betamethasone and combination treatment increased SP-B expression throughout by +97-117%, +42-51% and +93-309% (P< 0.001), respectively. SP-C was increased by +35-48%, +30-44% and +74-108% (P< 0.001) after the respective treatments, while SP-A &-D were unchanged in immature lungs. All treatments increased the expression of ATGL (+50-75%), LPCAT (+15-20%) und CGI-58 (+5-10%) (P< 0.05). However, whereas rhKGF effects were accompanied by increased pneumocyte proliferation, betamethasone blocked both basic (d7) and rhKGF-induced (d7+d21) proliferation.

Conclusion: Short term medication of neonatal rats with rhKGF/Palifermin® increased the expression of genes relevant for surfactant function and metabolism. Actions were identical or superior to those of betamethasone, with combination treatment exerting maximal effects. However, while betamethasone effects were at the expense of lung anabolism/pneumocyte proliferation, rhKGF enhanced proliferation.

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FEMALE SEX STEROIDS MODULATE ALVEOLAR EPITHELIAL SODIUM TRANSPORT

M. Laube[1], E. Küppers[2], U.H. Thome[1]

[1]Neonatology, University of Leipzig, Leipzig, [2]Molecular and Cellular Anatomy, University of Tübingen, Tübingen, Germany

Background and aims: Sodium transport plays a crucial role in alveolar fluid clearance (AFC), where sodium enters the alveolar epithelial cells through apical sodium channels (ENaC) and is extruded by basolateral Na,K-ATPases. Women with acute respiratory distress syndrome have higher AFC and higher survival than males. Female sex steroids are supposedly responsible for these gender-related differences. Therefore, the aim of this study was to analyze the effects of estrogen (E2) and progesterone (P) on epithelial sodium transport.

Methods: Isolated alveolar epithelial cells from 18-19-day gestational age rat fetuses, grown in serum-free media supplemented with E2 and P were studied, involving Real-Time PCR analysis, single-channel patch clamp, Ussing chamber measurements and Western blotting.

Results: rtPCR revealed an increase of α- and β-ENaC in media supplemented with E2 and P, where the effect on α-ENaC was most pronounced in P-rich media. The mRNA-level of γ-ENaC was not altered, but the Na-K-ATPase-β_1 subunit and the CFTR-mRNA expression were increased by E2 and P supplementation. Single-channel patch clamp analysis showed highly-selective and non-selective ENaC in the examined cells. The percentage of responsive patches increased from 45% in non-supplemented media to 91% in the presence of E2 and P. Short-circuit current (I_{SC}) measurement showed that the baseline, amiloride- and ouabain-sensitive I_{SC} were elevated by E2 and P in a dose-dependent manner.

Conclusions: These results demonstrate increased expression of epithelial transport proteins by female sex steroids. Furthermore, vectorial sodium transport was increased. The findings may explain the observed survival advantage.

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HYPEROXIA SUPPRESS ANTIOXIENZYME ACTIVITY IN CULTURED PULMONARY ARTERIAL ENDOTHELIAL CELLS

R.-H. Fu[1], Y.-H. Cho[2], R. Lien[1], P.-H. Yang[1], M.-C. Chiang[1]

[1]Department of Pediatric, Division of Neonatology, [2]Department of Pediatric Health Research, Chang Gung University and Children's Medical Center, Taoyuan County, Taiwan R.O.C.

Background: Hyperoxia is one of the risks to cause BPD or ROP in preterm infants. It has been known to suppress the angiogenesis of vascular endothelial cells. However, the effect of hyperoxia to the antoxienzyme activity of vascular endothelial cells is not very clear.

Objective: To evaluate the effects of hyperoxia to the superoxide dismutase (SOD) and glutathione peroxidase (GPx) activities of cultured pulmonary arterial endothelial cells (PAEC).

Materials and methods: Primary fetal sheep (GA 125 to 140 days) pulmonary arterial endothelial cells (PAEC) were cultured under room air, 60%, and 80% oxygen. NO donor (DETA NONOate) was administrated to the culture medium with the concentrations of 0, 0.01, 0.02, and 0.05mM. The