- 107 "IN VITRO" PROLIFERATION OF ACTIVATED LYMPHOCYTES INFILTRATING SMALL BOWEL IN COELIAC DISEASE. M. Stegagno, M. Cambiaggi, M. Bonamico, P. Mariani, E. Carapella Departments of Child Health and Paediatrics, University "La Sapienza" Rome - ITALY.

The ability of interleukin 2 (IL-2) to stimulate cellular division of activated T lymphocytes allows to propagate "in vitro" the T cells infiltrating inflammatory tissues. We developed a system that allows to propagate relevant amounts of activated T lymphocytes from minute fragments of peroral small bowel biopsies performed for diagnostic purposes. In preliminary experiments on three different patients with gluten enteropathy on a gluten containing diet, the culture of biopsied tissue in the presence of IL-2 allowed us to recover as many as 10^8 lymphocytes. The evaluation of surface markers with monoclonal antibodies showed that these cells were virtually all T lymphocytes (CD3+): the two different T cells subsets, CD4+ (helper / inducer) and CD8+ (suppressor / cytotoxic) were almost equally represented in the two patients challenged with gluten while in the third one, who was at the first diagnostic biopsy, there was a marked predominance of CD8+ cells. This difference could be related to the different phases of the disease in the patients examined. The relevant numbers of cells obtained with our experimental procedure can be used in functional assays to further investigate the pathogenesis of coeliac disease.

- 108 COGNITIVE EFFECT AT 5 YEARS OF AGE IN INFANTS WHO WERE ANEMIC AT 12 MONTHS: A LONGITUDINAL STUDY. T. Walter, I. de Andraca, M. Castillo, F. Rivera, C. Cobo. Hematology and Psychoneurology Units. INTA, U. of Chile, Santiago. Anemic infants from a longitudinal study of iron nutrition who were evaluated with the Bayley Scales of Infant Development showed significantly lower scores than their iron sufficient controls at 12 mo and after therapy for complete correction of iron status. Anemia was determined by random assignment to non fortified diets (Peds. 84:1-17, 1989). At 5½ years of age they were reevaluated with a general intelligence test (1), fine and gross motor abilities (2), psycho-linguistic capabilities (3), visual-motor integration (4) and an educational preschool scale (5). Maternal and environmental variables have been controlled for. Results are:
Former iron status (at 12 mo)	Anemic n=43	p<	control n=29
Current age (months)	66 ± 2.4	NS	66.5 ± 4.8
(1) Stanford-Binet (IQ)	87 ± 8.1	.05	92.4 ± 8.0
(2) Bruininks-Oseretsky (Composite)	39 ± 9	.04	44 ± 8
(3) Illinois (composite quotient)	83 ± 11	.02	89.6 ± 8
(4) Visual Motor (percentile)	31 ± 24	.02	45 ± 24
(5) Woodcock (Score)	86 ± 19	.02	96 ± 11
- Unfavourable effect of iron deficiency anemia on development at 12 mo persists in cognition at 5½ despite timely and adequate iron therapy. Although causality cannot be proven, anemia in infancy may be a marker for disadvantaged cognition in childhood.

- 109 INDUCTION OF DIFFERENTIATION OF A549 HUMAN ALVEOLAR TUMOR CELLS BY BASEMENT MEMBRANE. Paul Stevens¹, Ursula Brands¹, Bernd Rüstow², Michael Obladen¹. ¹ Univ. Children's Hospital, Berlin, FRG; ² Inst. Pathol. Clin. Biochemistry, Charité Hospital, Berlin, GDR. A 549 tumor cells may be of type II cell origin, but do not synthesize surfactant under normal conditions. We cultured this cell line either on tissue culture-treated plastic or on top of an extracellular matrix, obtained from the Englebreth-Holm-Swarm (EHS) tumor. We found that cells cultured on EHS matrix synthesized surfactant protein A (SP-A) as shown by Western blot and by slot blot hybridization with an SP-A cDNA probe (n=5). Compared to cells cultured on plastic, cells cultured on EHS for 3-5 days also contained a higher percentage of 16:0-16:0 phosphatidylcholine (10 vs. 2 %, n=2) and incorporated more ³H-glycerol into this lipid species (10 vs. 2.8 %, n=2). On EHS matrix, the cells became cuboidal, and formed alveoli-like spherical structures surrounding a closed lumen. Also, cell division rate was reduced on basement membrane-type matrix. These results suggest that culture on basement membrane induces expression of type II pneumocyte-specific properties in human A549 cells. This culture model may allow a better understanding of the mechanisms by which cell-extracellular matrix interactions modulate specific gene expression. Supported by DFG Stc 459/1-1.

- 110 THE ANTIBODY (anti-CD 2, anti-CD 3) INDUCED LYMPHOCYTE MITOGENESIS IS DEFECTIVE IN INFANCY AND EARLY CHILDHOOD N. Remy, S. Krusche, T. Grass, M. Oberreit, U. Wahn Univ. Childrens Hospital, FU Berlin, Germany

Recent findings indicate a defective mitogenesis of cord blood lymphocytes as demonstrated by stimulation with the monoclonal antibodies anti-CD 2/2a (alternative pathway) and anti-CD 3 (T-cell-receptor-complex) (Gerli 89). In order to investigate the age dependency of this antibody induced transformation and a possible association with changes of lymphocyte subsets, blood was obtained from healthy subjects: cord blood samples, 5 day old neonates, 4 month old infants, 2-4 year old children as well as adults (n=10 in each group). Lymphocytes were incubated for 96 h with optimal concentrations of anti CD-2/2a and anti-CD 3. In addition lymphocyte subsets CD 2 and CD 3 were determined by flow cytometry. Only cord blood samples were found to have a significantly (p<0.001) reduced percentage of CD 2 and CD 3 positive lymphocytes, whereas from day 5 on percentages were within the range of adults. In contrast, proliferative responses of lymphocytes induced by anti-CD 2/2a and anti-CD 3 were significantly reduced in all pediatric groups (p<0.001) compare to adults. Within the pediatric groups there were no differences in lymphocyte responses. From our data we conclude that in infancy and early childhood there is a functional defect of lymphocytes detectable by stimulation with monoclonal antibodies, which is not related to differences in lymphocyte subsets.

- 111 LUNG EDEMA DECREASES THE RESPONSIVENESS TO EXOGENOUS SURFACTANT.

Mikko Hallman, Veeraiah Chundu, William Cvetnic, Stefan Slivka, University of California, Irvine, Medical Ctr., Department of Pediatrics, Orange, CA.

There is a large variability in the effect of exogenous surfactant in RDS. We have tested the hypothesis that hydrostatic lung edema decreases the responsiveness to surfactant. Respiratory failure was induced to rabbits, wt. 1-1.3 kg, by alveolar lavage (BAL, 10 ml/kg x 4). Thereafter, natural surfactant (NS, 100 mg/kg in 4 ml saline, n=10), saline carrier (n=8), or air (n=8) was administered. The animals were paralyzed and ventilated at constant tidal volume. They received either 1 (low) or 20 ml/kg/h (high) saline i.v. during 155 min. The lung function was measured and BALs were analyzed for phosphatidylcholine (PC), concentration of PC in epithelial lining fluid (PC_{ELF}), protein, and surface activity. Saline carrier to airways decreased the gas exchange and compliance (p<0.01). In animals on low i.v. fluids, NS improved the respiratory function as compared to air-treated controls. However, in animals on high i.v. fluids, NS neither improved the gas exchange nor the compliance, despite the increase in PC (p<0.05) and the decrease in soluble protein (p<0.05) in BAL. The lack of surfactant responsiveness was due to low PC_{ELF} and to surfactant inhibitors. Thus, variability in surfactant responsiveness may be caused by factors affecting lung liquid.

- 112 HYPOXANTHINE (HX) IN VITREOUS HUMOUR (VH) AND PLASMA DURING CONTINUOUS INFUSION OF HX IN NORMOXIC PIGLETS Terje Rootwelt, Jan P. Poulsen, Ola D. Saugstad Inst. for Surg. Res., Dep. of Ped. and Ped. Res. Rikshosp., Oslo, Norway

Hx was infused intravenously into eight newborn piglets (aged 7-14 days) and measured with a HPLC method in VH and plasma before and after 4-12 hours of infusion. The piglets were normoxic and normotensive throughout the experiment. A plateau was reached in plasma after 30-60 min. There was a significant correlation between Hx concentrations in VH and plasma (r=0.64, p<0.008). The first 5-30 min after ending infusion plasma Hx was eliminated quickly with a mean T 1/2 of 23 min (range 13-40 min), then at a much lower rate with a mean T 1/2 of 5 1/2 hours (range 2 1/2- 9 hours).

Conclusion: Hx in VH is lower than in plasma, but reflects plasma concentration fairly well and is therefore at least to some extent derived from plasma. Plasma Hx in normoxia is eliminated more slowly than previously reported in posthypoxic conditions.