

● 217

EVALUATION OF AN IGE-ELISA SPECIFIC FOR BOVINE  $\alpha$ -CASEINS  
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ELISA microtiter plates (Greiner) were coated with highly purified bovine  $\alpha$ -caseins and incubated with diluted sera. Specific IgE was detected by successive incubation with three antibodies, the last one conjugated with horseradish peroxidase. Twelve patients (age 1/2 - 3 years) with positive provocation and skin prick test to bovine caseins were compared with 12 children (age 1/2 - 12 years) with negative tests. The results were compared to CAP-RAST (Code f78, Pharmacia) data. Additionally IgE-immunoblots were made as a qualitative criterion and to distinguish between immunoreactions to the different caseins and the other major cow milk proteins.

67% of the cow milk allergic patients had positive RAST (class > 1) and 92% were positive (OD > 0.1) in the ELISA. In the immunoblot 83% showed distinct reactions to  $\alpha$ -caseins. None of the control group gave positive reactions. Only one child's serum showed a positive reaction to  $\alpha$ -caseins in the immunoblot.

In conclusion, compared with the clinical allergy of the children studied our ELISA proved to have a higher sensitivity than the RAST; specificity of both tests equalled 100%.

● 218

PULMONARY DIFFUSING CAPACITY AT REDUCED ALVEOLAR VOLUME WITHIN THE PAEDIATRIC AGE RANGE. H. Stam, A. v.d. Beek, K. Grünberg, H.A.W.M. Tiddens and A. Versprille.  
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In adults  $D_L$ , i.e. total diffusing capacity of carbon monoxide, increases and  $D_L$  per liter alveolar volume ( $D_L/V_A$ ) decreases with increasing alveolar volume  $V_A$ . The decrease in  $D_L/V_A$  is linear and less steep in older subjects (1). We also determined  $D_L$  and  $D_L/V_A$  at total lung capacity (TLC) and at lung volumes below TLC in 103 normal children with ages ranging from 6 to 20 years (55% and 48%). The major objectives of this study were to examine whether the  $D_L/V_A$  vs.  $V_A$  relationship is also linear in children and whether the slope of this relationship depends on age, sex and height. In all children a linear regression equation was the best mathematical description. The slopes decreased with age as well as height in boys ( $p < 0.05$  and  $p < 0.02$ ) and girls ( $p < 0.01$  for both). From the regression equations of  $D_L$  and  $D_L/V_A$  with  $V_A$  reference values could be calculated at  $V_A$  below TLC to evaluate the diffusion variables in children, who suffer from a restrictive disease.

1. Stam et al., Diffusing capacity dependent on lung volume and age in normal subjects. J. Appl. Physiol. (in press)

▲ 219

MECHANISM OF SP-A-MEDIATED SURFACTANT ENDOCYTOSIS BY TYPE II CELLS.  
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AIM: Surfactant protein A (SP-A) enhances surfactant lipid uptake by type II pneumocytes. In the presence of SP-A internalized surfactant lipids are reported to bypass the degradative pathway and are recycled towards lamellar bodies. We wanted to further clarify the role of SP-A in surfactant lipid endocytosis.

METHODS: A previously described antibody (2H5) against a type II cell membrane protein which stimulates surfactant lipid uptake by type II cells (Pediatr. Res. 1994, 35: 278) was used in parallel with SP-A to study uptake and intracellular fate of liposomes with surfactant-like composition in rat type II cells.

RESULTS: In the presence of 2H5 or SP-A significantly more labeled lipid is internalized in a time- and concentration-dependent fashion by type II cells than in their absence (2H5 2-fold, SP-A 3-fold above control). In cells in solution no difference in the distribution of label in phospholipid classes between control cells and cells incubated with either 2H5 or SP-A was found. In adherent 24 hour-old cells after one hour of incubation 82% of the internalized  $^3H$ -label is still associated with PC in control cells vs. 87% in the presence of 2H5 and 94% with SP-A. Surprisingly, inhibition of coated pit formation (uptake pathway for SP-A) by  $K^+$ -depletion enhanced lipid uptake by type II cells significantly. Also, inhibition of protein kinase C (PKC) (staurosporine 100 nM) enhanced lipid uptake by type II cells in the presence of SP-A.

CONCLUSIONS: SP-A and 2H5 do quantitatively enhance lipid uptake in type II cells. The subsequent intracellular fate of the PC molecule may depend on the type of assay used. Uptake via coated pits and PKC activation are involved in these processes.

● 220

DO PERIVENTRICULAR FLARES (PVF) PREDICT OUTCOME? Ann Stewart, John Wyatt, Ann Lorek, Vincent Kirkbride, Judith Meek, Juliet Penrice, Jenny Baudin, Jan Townsend, Osmund Reynolds, Dept of Paediatrics, University College London Medical School, London, UK.

Interpretation and significance of PVF seen with ultrasound (US) in the brains of newborn infants remain controversial. To find out if PVF predict adverse neurodevelopment, follow up data for PVF without periventricular haemorrhage were analysed from a prospectively scanned cohort (n=725) of very preterm (<33 w) survivors born 1983-89. PVF were defined as non-haemorrhagic echodensities in the periventricular region. Probabilities (p%, 95% CI) were calculated for disabling impairments and for total impairments, with and without disability, identified by neurological and developmental assessments at 1 year of corrected age. Values are given for normal US scans (n=355), PVF without later cyst formation (n=50), PVF with cystic periventricular leucomalacia (n=7, all parieto-occipital) (PVF+PVL) and PVL cysts not preceded by PVF (n=7; 5 frontal, 1 parietal, 1 parieto-occipital), of which 6 were noted aged 1-4 days, and thus were antenatal in origin.

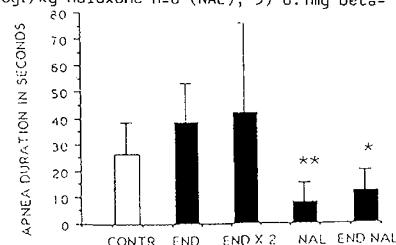
Impairment	Normal scan		PVF		PVF+PVL cysts		PVL cysts	
	n	p%(CI)	n	p%(CI)	n	p%(CI)	n	p%(CI)
Disabling	11	3(1-5)	4	8(2-19)	6	86(42-99)	0	-
Total	48	13(9-16)	11	22(12-36)	7	100(59-100)	0	-

We conclude a) only PVF followed by cyst formation caused a significant excess of neurodevelopmental impairments at 1 year b) cysts acquired before birth were predominantly frontal and not associated with neuromotor or sensory impairments.

▲ 221

NALOXONE INHIBITS THE DURATION OF THE LARYNGEAL CHEMOREFLEX (LCR) ACTIVATED APNEA IN PIGLETS Hanne Storm, Lauritz Stoltenberg, Ola D. Saugstad, Torleiv O. Rognum, Karl L. Reichelt, Department of Pediatric Research and Institute for Forensic Medicine, National Hospital, Oslo, Norway

Beta-endorphin may induce respiratory depression and bradycardia. Infants with apnea and increased level of beta-endorphin immunoreactivity in CSF have been successfully treated with naloxone. Therefore, LCR apnea was activated in five groups of 5-10 days old piglets: 1) untreated n=6 (CONTR), 2) 0.1mg beta-endorphin in cisterna magna (i.c.m.) n=6, (END) 3) 0.2mg beta-endorphin i.c.m. n=6 (END X 2), 4) 100 mikrogr/kg naloxone n=6 (NAL), 5) 0.1mg beta-endorphin i.c.m. and naloxone n=6 (END NAL). Respiration, heart rate, and blood pressure were monitored. We found that 0.1-0.2mg beta-endorphin induced apneas and bradycardia and hypertension. Furthermore, naloxone shortened the induced LCR.



Conclusion:

Naloxone inhibits laryngeal chemoreflex induced apnea.

● 222

IMMUNOPHENOTYPE IN THYMUS AND LIVER OF FETUSES WITH 19-21 WEEKS GESTATIONAL AGE

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We have studied the immune phenotype in thymus and liver, obtained from 7 fetuses with 19-21 weeks gestational age, using the monoclonal antibodies of Russian and American manufacture in reaction of indirect immunofluorescence. The monoclonal antibodies' panels were different for the assay of thymic or liver lymphocyte subpopulations, including CD3, CD4, CD1, CD8, CD38, CD2, HLA-DR,  $\Sigma$ g, Ig A, Ig G, Ig M,  $\kappa$ - and  $\lambda$ -chains' markers, respectively. The number of CD3+ cells in fetal liver turned out to be quite variable, while the number of B cells was rather stable. CD3+ lymphoid subset from the studied fetal thymus was found to possess 39,25 +/- 2,25%, CD4+ reaching 47,2 +/- 2,2% and CD2+ equaling 60,9 +/- 3,62%, which data are quite comparable with such ones from similar subsets of fetuses with higher gestational age (27-39 weeks): 33,0 +/- 5,45%, 50,4 +/- 3,33% and 63,1 +/- 6,20%, respectively. Besides, we have registered the signs of IL-2 synthesis (0,6 +/- 0,11 I.U.). We conclude that the immunophenotype of thymus and liver in human fetuses of 19-21 weeks gestational age is able to perform certain immune response.