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CEREBRAL HAEMODYNAMICS DURING FAILURE OF OXIDATIVE PHOSPHORYLATION FOLLOWING BIRTH ASPHYXIA.

J.S.Wyatt, A.D.Edwards, D.Azzopardi, C.E.Richardson, M.Cope, D.T.Delpy, E.B.Cady, E.O.R.Reynolds. Depts. of Paediatrics, and Medical Physics & Bioengineering, University College and Middlesex School of Medicine, London.

Twelve infants born at 36-42 weeks of gestation who had suffered severe birth asphyxia were studied repeatedly in the first week of life by near infrared spectroscopy (NIRS) and magnetic resonance spectroscopy (MRS). Six infants with normal brains were also studied by NIRS to provide control data. Phosphocreatine/inorganic orthophosphate (PCr/Pi) concentration ratios determined by MRS following asphyxia were frequently normal in the first 48 hours, but subsequently low ratios were always seen. In the first 48 hours mean cerebral blood volume (CBV) was elevated (4.40±SEM 0.38ml.100g^{-1}) compared with the control infants (1.98±0.26ml.100g^{-1}, p<0.001), and CBV response to changes in arterial carbon dioxide tension ($PaCO_2$) was reduced (0.10±0.06 compared with 0.53±0.06ml.100g^{-1}.kPa^{-1}, p<0.001). Cerebral haemodynamics in the asphyxiated infants subsequently returned towards normal.

Thus, following birth asphyxia, elevated blood volume and reduced response to changes in $PaCO_2$ may be detected prior to impairment of oxidative phosphorylation.

Supported by the MRC, Wellcome Trust and Hamamatsu Photonics KK.

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IN VIVO XANTHINE OXIDASE (XO) ACTIVITY IN PREMATURE INFANTS WITH RDS AND IN HYPOXIC STATES OF CHILDREN MEASURED BY URINARY CAFFEINE METABOLITES

Domokos Boda and Ilona Németh, Dept. of Paediatrics - University Medical School, Szeged, Hungary

It has been proven by numerous investigations that in hypoxic and shock states the purine breakdown by XO is the main source of the superoxide-mediated reperfusion injury of cells. Grant et al. have demonstrated that after caffeine intake the molar ratio of methyluric acid (MU) and methylxanthine (MX) measured by HPLC in urine reflects the XO activity in vivo. The results of such measurements ($\bar{x} \pm$ S.D.) in various groups of children were as follows: 15 controls: 1.11 ± 0.52; 17 newborns with RDS first day: 16.9 ± 4.90; third day: 10.5 ± 6.9; 18 critically ill children, in acute phase: 11.5 ± 14.4 (range 1.9 - 50.0); in reparation: 2.3 ± 3.7; in 6 so-called shock-tolerant patients: 0.38 ± 0.25. The MU/MX ratio and the serum uric acid value showed a significant correlation (r: 0,57, n = 44, p < 0,001) in various groups. In conclusion: The XO activity in vivo is extremely increased in severe acute clinical states and - presumably by an adaptive process -, it rapidly decreases in reparation and becomes extremely low in shock-tolerant states. These results explain the favourable effect of allopurinol in experimental and clinical shock states.

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HUMAN FETAL XANTHINE OXIDASE

Kim Vettenranta & Kari O. Raivio, Children's Hospital, University of Helsinki, SF-00290, Finland

Xanthine oxidase (E.C. 1.2.3.2.) has been implicated in the pathogenesis of perinatal postischemic and hyperoxic tissue injuries through its action on the accumulated substrate, hypoxanthine (Hx), and concomitant oxygen free radical production. We measured the activity of xanthine dehydrogenase/oxidase (XOD), and its apparent Km for the main physiological substrate, hypoxanthine, in human fetal liver, intestine, brain, and myocardium. Autopsy samples from 45 fetuses (10-20 gestational weeks), 9 preterm babies (25-28 weeks), and 15 term babies were included in the study. XOD-activity increases in the liver and decreases in the intestine with gestation. During the last trimester the hepatic activity increases almost twofold to a mean of about 0.5 nmol/mg protein/min, while that in the intestine decreases to a third, a mean of about 0.3 nmol/mg protein/min. The apparent Km for Hx is 4.8-5.5 µM in the intestine throughout gestation and in the liver at term, but higher than 30 µM in the liver during the first half of pregnancy. The activity is below 20 pmol/mg protein/min in the brain, and undetectable in the myocardium. Thus, human fetal liver and intestine have substantial enzymatic capacity to convert Hx and/or xanthine to uric acid throughout a gestation. This is a prerequisite but not proof of the involvement of XOD in the pathogenesis of perinatal ischemia-reperfusion injury.

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EXOGENOUS XANTHINE OXIDASE (XOD) DEPLETES ADENINE NUCLEOTIDES FROM ENDOTHELIAL CELLS (ECs)

Kristiina Aalto and Kari O. Raivio, Children's Hospital, University of Helsinki, SF-00290, Finland

Hypoxanthine (Hx) is produced by hypoxic cells.On re-oxygenation the Hx-XOD-reaction generates oxygen free radicals, which are assumed to damage tissues. Since ECs have been reported to contain XOD and are exposed to circulating Hx, they are a likely target for damage.We have studied the effect of Hx and/or XOD on nucleotide metabolism of ECs from human umbilical veins, cultured under standard conditions. Cells in culture wells were labeled overnight with 14C-adenine (100uM), washed and further incubated for 3-6 hr with either Hx (100uM), XOD (40mU/ml) or both. Medium was removed and the cells extracted with perchloric acid. Nucleotides from cells and medium, and Hx, xanthine (X) and uric acid (Ua) from medium were separated by TLC and counted.

In the presence of serum, Hx alone had no effect, but XOD alone caused a 30% fall in cellular nucleotides and an equivalent increase of X+Ua in the medium. In the absence of serum the decrease in nucleotides was 60 to 90%. The combination of Hx + XOD caused a 60-90% (no serum) or 80% (with serum) fall in cellular adenine nucleotides. Thus, endogenous Hx in the presence of exogenous XOD triggers adenine nucleotide catabolism but not vice versa. Serum seems to have a protective role. We conclude that accumulation of Hx alone does not damage ECs, but in the presence of XOD, nucleotide depletion occurs and cellular damage is possible.

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WASHING OUT OF HYPOXANTHINE (HX) IN PRETERM BABIES WITH RDS TREATED WITH NATURAL SURFACTANT (CUROSURF)

O.D. Saugstad, K. Gloppestad, T.R.J. Tubman, H.L. Halliday, S. Øyasæter, T. Curstedt, B. Robertson Dep. Ped., Inst. Surg. Res., Dep. Ped. Res. Rikshospitalet, Oslo, The NICU Royal Maternity Hospital Belfast, Dep. Clin.Chem. Karolinska Hospitalet Stockholm Dep. Ped. Pathol. St. Görans Hospital Stockholm

Plasma HX concentration was monitored in four preterm babies with severe RDS who were treated with natural surfactant (G.A. 28-30 weeks, B.W. 1115-1325 g). FiO_2 before surfactant application ranged between .9-1.0. There was a 262% increase in HX concentration 15 min after surfactant administration (p<.05). After 60 min the HX level reached pretreatment levels in all but one baby. There was no changes in HX concentration in control babies.

minutes	0	5	15	30	60
HX umol/l	4.5 (1.1)	- -	11.8 (9.0)	9.1 (8.3)	6.6 (4.7)
paO_2 kPa	5.9 (1.0)	30.3 (9.5)	14.8 (11.1)	10.1 (6.7)	6.5 (1.4)
FiO_2	.96(.06)	.95(.07)	.64(.12)	.51(.10)	.45(.20)

These data demonstrate a significant washing out of HX after surfactant application. Whether or not this comes from the lungs is not known. Since HX is an oxygen radical generator it is speculated whether this phenomenon plays a role in the clinical situation.

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PULMONARY LIPID DEPOSITS AFTER SURFACTANT SUBSTITUTION FOR NEONATAL RESPIRATORY DISTRESS SYNDROME (RDS).

Hugo Segerer, Martin Vogel[2], Michael Obladen, Tore Curstedt[3], Bengt Robertson[4], Children's Hospital, [2]Institute of Pathology, Free Univ, Berlin (West), Germany; [3]Dept of Clinical Chemistry, Karolinska Hospital, [4]Dept of Pediatrics, St.Göran's Hospital, Stockholm, Sweden.

Ten very low birth weight infants (770 - 1400g) with severe RDS were treated with porcine surfactant (Curosurf) at a median age of 6.7 h. The typical response was immediate improvement of oxygenation. 4 babies could be weaned from the ventilator within 36 h. However, in three patients (birth weights 1300g, 890g, 830g) improvement was only transient; these infants died from pulmonary insufficiency at the age of 9, 9, and 14 days, respectively, in spite of repeated instillation of surfactant within 21 - 64 h after the first treatment.

Histological examination of lung specimens obtained at autopsy revealed, in one case, broadened alveolar septae with interstitial infiltration of lymphocytes and monocytes (representing organizing hyaline membrane disease?); in the two other cases, there was evidence of obliterative bronchiolitis compatible with an early stage of bronchopulmonary dysplasia. In all three cases, Sudan-stained frozen sections showed lipid deposits in macrophages. Such lipid-loaded cells were found in alveolar spaces, alveolar walls, and perivascular interstitial tissue. No similar deposits were seen in infants with RDS who died without receiving surfactant. The reason for treatment failure is not explained by the present autopsy findings, but it seems likely that the accumulation of lipids is secondary to surfactant treatment. The possibility that macrophage function might, in some infants, be hampered by overloading with surfactant lipids, needs to be considered.