19

SYNCYTIOTROPHOBLASTIC CELLS IN THE STUDY OF HUMAN PLACENTAL METABOLISM IN EARLY AND LATE GESTATION

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

In order to study developmental and metabolic characteristics of normal and abnormal placental tissue, especially purine metabolism and effects of oxygen deprivation, we have optimized a method for primary culture of trophoblastic cells. After collagenase dispersion, the cells were enriched by filtration and cultured in Ham's F-10 medium. The cell population contained mainly syncytiotrophoblasts (over 90%), characterized using anti-cytokeratin and -vimentin antibodies, and the viability remained over 90%. The plating efficiency was significantly higher on dishes coated with fibronectin, collagen-IV or laminin, when compared to uncoated dishes. On the former, a higher degree of specialized function was retained, as evidenced by consistently higher chorionic gonadotropin (hCG) secretion. Comparing first trimester and term trophoblast, the following observations have been made: no growth of either first or third trimester syncytiotrophoblast in primary culture; active mesenchymal cell growth in first trimester, but none in term cultures; hCG and pregnancy-specific beta-1-glycoprotein secretion inversely proportional to gestational age; a six-fold increase in alkaline phosphatase, but a 40% decrease in 5'-nucleotidase activity with increasing gestational age, but no difference in the activities of three other key enzymes of purine catabolism and re-utilization; active de novo purine synthesis, but a large inter-placental variation. In conclusion, the trophoblastic character and specialized function in the primary cultures are retained, and they can be used for studies on normal and abnormal placental metabolism.

20

PHOSPHORUS METABOLITES AND INTRACELLULAR pH IN THE BRAINS OF NORMAL AND GROWTH RETARDED INFANTS.

Hamilton, P.A., Hope, P.L., Cady, E.B., Delpy, D.T., Wyatt, J.S., Reynolds, E.O.R. University College London, Depts. of Paediatrics, and Medical Physics and Bioengineering, London, England.

Studies by magnetic resonance spectroscopy were performed at a median age of 5 days on 18 appropriately grown (AGA) and 9 growth retarded (SGA) newborn infants with birthweights below the 3rd centile, to investigate: 1. Maturational changes in cerebral intracellular high-energy phosphorus (P) metabolite concentrations and pH_i during normal gestation, and: 2. Whether any abnormalities were detectable in the SGA infants. The methods used have been described previously (Hope PL et al, Lancet 1984, ii: 366).

Linear regressions were calculated for P-metabolite concentration ratios and pH_i on gestational plus postnatal age for the AGA infants. Phosphocreatine (PCr)/inorganic orthophosphate (P_i) increased from 0.77±0.24 (95% confidence limits) at 28w to 1.09±0.24 at 42w ($p<0.001$). No significant changes were found in adenosine triphosphate (ATP)/total P, PCr/ATP, Pi/ATP, phosphomonoester (PME)/ATP, phosphodiester (PDE)/ATP, or pH_i, though PCr/ATP tended to rise, and Pi/ATP and PME/ATP tended to fall. Values from the SGA infants at 30-38w of gestational plus postnatal age were distributed evenly across the data from the AGA infants.

We conclude: 1. PCr/Pi increased with advancing maturation of the brain, and: 2. Values for P-metabolite ratios and pH_i in the SGA infants did not differ from those in the AGA infants.

21

FETAL MATURATION OF THE SYNTHESIS OF THE COFACTOR OF PHENYLALANINE HYDROXYLASE.

Dhondt J.L., Farriaux J.P. Laboratoire de Recherche de Pédiatrie, CHR, Lille, France.

Since a delayed maturation of tetrahydrobiopterin (THB) synthesis might explain some observations of transient neonatal hyperphenylalaninemia, the aim of this work was to explore the ontogenesis of the biosynthesis of THB (the cofactor of phenylalanine-hydroxylase) by measuring unconjugated pteridines in amniotic fluid (AF). AF have been obtained from a total of 252 women between 12 and 40 weeks of normal gestation. Neopterin (N) and biopterin (B) were measured by a HPLC method after iodine oxidation at pH 1 and sample clean-up on ion-exchange resins.

gestational age	neopterin	biopterin	N/B
12-27 weeks (n=152)	36.6±20.6	13.3± 5.1	2.9±1.4
28-40 weeks (n=100)	192.7±66.4	39.7±16.2	5.3±1.8

At midgestation N and B levels in AF (nmol/l) remained relatively constant between 12 and 27 weeks of gestation. On the contrary, during the third trimester a progressive increase in N and B levels was observed, highly correlated with gestation age and accompanied by an increase of N/B ratio indicating a fetal source of AF pteridines. However, a considerable scatter of values was noticed in preterm AF suggesting existence of great variation in individual maturation process.

22

LOW METABOLIC RATES OF VERY LOW BIRTHWEIGHT (VLBW) NEWBORN INFANTS IN THE FIRST DAYS OF LIFE IN THE NEONATAL WARD.

van Asselt W.A., Geerdink J.J. and Okken A. Div. Neonatology, Dept. Paediatrics, State University Groningen, The Netherlands.

Many studies on metabolic rate of VLBW newborn infants have been performed in the laboratory. Data obtained under such conditions may not fully apply to newborn infants in the neonatal ward. We have studied oxygen consumption ($\dot{V}O_2$) in VLBW infants in the nursery. Birthweight and gestational age of the infants studied so far were 1.350±0.240 kg (mean±SD) and 28.5±1.2 wks respectively. Studies were performed at low and high humidity (water vapour pressure, $P(H_2O)$) while incubator air temperature (Tinc) was not changed. Results:

Age (d)	P_{H_2O} (mmHg)	Tinc (°C)	$\dot{V}O_2$ (ml/kg/min)
3.4	17.4 ± 6.0	35.3 ± 0.4	4.44 ± 0.98
	32.1 ± 1.9	35.3 ± 0.2	4.10 ± 0.77
11.0	19.5 ± 4.0	34.5 ± 0.6	5.20 ± 1.23
	28.2 ± 2.7	34.6 ± 0.6	5.20 ± 0.75
17.0	19.0 ± 1.8	33.5 ± 1.5	6.33 ± 0.76
	25.6 ± 2.1	33.5 ± 1.6	6.63 ± 1.04

It is concluded, that metabolic rate of VLBW newborn infants in the nursery is very low in the first days of life. Despite their presumably large insensible water loss, increasing water vapour pressure in the incubator does not seem to affect their metabolic rate largely.

23

RESPIRATORY WATER LOSS IN RELATION TO ACTIVITY IN FULLTERM NEWBORN INFANTS.

Riesenfeld T, Hammarlund K & Sedin G. Dept. of Paediatrics, University Hospital, Uppsala, Sweden.

In the newborn infant water loss from the skin has earlier been said to contribute 75% to the total insensible water loss. With a method for direct measurement of respiratory water loss (RWL) we have found that this loss contributes more than 50 % to the total insensible water loss at rest in fullterm infants on their first day after birth. The influence of activity on the respiratory water loss has now been studied in ten healthy fullterm infants on their first day after birth.

RWL is measured with an open flow-through system, where air drawn over the face of the infant picks up the expired gas. The flow of mixed gas is measured with a Fleisch flow-head connected to a differential pressure transducer, and the concentrations of water vapour, oxygen and carbon dioxide in the mixed gas and in the ambient air are measured with a Perkin Elmer mass-spectrometer. RWL, oxygen consumption and carbon dioxide production are calculated from the obtained flows and concentrations. During the measurements the infant is placed in an incubator with carefully controlled air temperature, humidity and air flow velocity. The infants are then studied at different activities, from deep sleep to furious crying.

RWL was 4.2 mg/kg min with the infant at rest in an ambient humidity of 50 % and an ambient temperature of 32.5 °C. Oxygen consumption was 5.4 ml/kg min. Both RWL and oxygen consumption increased with increasing motor activity. When the infant was crying RWL increased to 10.8 mg/kg min while oxygen consumption increased to 8.2 ml/kg min.

24

MYOCARDIAL OXYGEN CONSUMPTION (MVO_2) AND SUBSTRATE UPTAKE IN FETAL AND 8-WEEK-OLD LAMBS.

M Dalinghaus, GP Toorop, AM Gerding, IE Mulder, R Berger, JRG Kuipers, AM Rudolph.
Depts of Pediatric Cardiology and Pediatrics, University of Groningen, Groningen, The Netherlands.

We measured myocardial blood flow to the left ventricular free wall and blood glucose, lactate, pyruvate, fatty acid, triglyceride, 8-OH-butyrate, acetoacetate and oxygen concentrations in aorta (Ao) and coronary sinus (CS) of 6 fetal and 12 8-week-old chronically instrumented lambs at rest. We calculated MVO_2 and substrate uptake when Ao and CS concentrations differed significantly. Despite significantly lower fetal Ao oxygen concentrations (3378±737 (SD) vs 5300±589 $\mu mol \cdot l^{-1}$), MVO_2 was not significantly different between the two groups of lambs (454±135 vs 514±194 $\mu mol \cdot min^{-1} \cdot 100\ g^{-1}$). Glucose (30.9±20.3 vs 36.5±52 $\mu mol \cdot min^{-1} \cdot 100\ g^{-1}$) and triglyceride (7.9±5.6 vs 6.6±10.4 $\mu mol \cdot min^{-1} \cdot 100\ g^{-1}$) uptakes were also similar, while the fetal Ao concentrations were significantly lower (920±171 vs 3850±485 and 123±29 vs 319±114 $\mu mol \cdot l^{-1}$, resp.). Besides glucose and triglyceride uptake by the myocardium, the fetuses also showed lactate (108±69 $\mu mol \cdot min^{-1} \cdot 100\ g^{-1}$) and pyruvate uptake (19.8±22.8 $\mu mol \cdot min^{-1} \cdot 100\ g^{-1}$), while the 8-week-old lambs showed only 8-OH-butyrate uptake (22.0 ±20.4 $\mu mol \cdot min^{-1} \cdot 100\ g^{-1}$). In both groups of lambs there was no uptake of free fatty acids. Assuming all substrates are metabolized aerobically they could account for 200% of oxygen uptake by the heart in fetal and 8-week-old lambs. We assume that during these studies the surplus of substrate uptake is stored by the myocardium.