

193

ERYTHROCYTE GTP DEPLETION IN PNP DEFICIENCY
PRESENTING WITH HAEMOLYTIC ANAEMIA AND HYPOURICAEMIA
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A 4-yr old Arab girl, referred for investigation of hypouricaemia and hypouricosuria discovered during studies for Coombs positive haemolytic anaemia, was found to excrete inosine, guanosine, deoxyinosine and deoxyguanosine in place of uric acid. The patient and a 10-month old brother were homozygous for purine nucleoside phosphorylase (PNP) deficiency. Three of the other four surviving siblings were heterozygotes, as were the parents who were first cousins. Another sibling had died of chicken-pox. The baby showed head-lag, the girl spastic diplegia.

As in our previous case, severe red cell GTP depletion - associated with raised dGTP levels and grossly increased NAD levels, were noted. Red cell PP-ribose-P levels and lysate APRT activity were likewise increased. The clinical findings also accord with previous reports. In addition to immunodeficiency, anaemia and neurological complications (ataxia, spastic tetraparesis) have been reported in 30-40% of cases.

The results suggest that patients with severe PNP deficiency may suffer the consequences of two enzyme defects - immunodeficiency due to an inability to metabolise toxic deoxy-nucleosides plus the neurological complications of hypoxanthine guanine phosphoribosyltransferase (HGPRT) deficiency because of a resultant lack of substrates for the latter. Red cell GTP depletion appears to be a consistent finding in inherited disorders associated with neurodevelopmental retardation.

194

A CASE OF LESCH NYHAN SYNDROME WITH DELAYED
ONSET OF SELF MUTILATION: A search for
abnormal biochemical, immunological and cell
growth characteristics in fibroblasts and neurotransmitters in urine.

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A 12 year patient with marked hyperuricemia, tetraparesis, choreoathetosis, dysarthria and mental retardation was recently referred for a sudden development of a rapidly increasing autoaggressive behaviour. Erythrocyte HPRT activity was virtually nil. Lysates of fibroblasts showed values 2.6% of the controls. The hypoxanthine utilisation in intact fibroblasts was 3% in the patient and 35% in his mother's cells. Cell growth in the 8-azaguanine and HAT was almost in the range of classical type and the mother showed heterozygosity.

Surprisingly, the level of dopamine, 5HT and bioppterin was significantly increased in the urine of unmedicated patient. Further biochemical data encompassing neurotransmitters, immunology, purine metabolism and cell selection would be presented.

195

INVESTIGATION OF PURINE UTILISATION IN
CULTURED FIBROBLASTS, AMNIOCYTES AND
CHORIONIC VILLI: AN ATTEMPT TOWARDS UNDERSTANDING
CLINICAL VARIABILITY IN LESCH NYHAN SYNDROME

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HPRT activity, enzyme kinetics, hypoxanthine uptake and cell growth in selection media has been studied in the cultured fibroblasts from a collective of 22 patients with different forms of HPRT deficiency. From the data it becomes apparent that the clinical manifestation can be best correlated to the ability of the cells of the patients to utilise hypoxanthine and growth resistance to its analogue metabolites, thus permitting a worthwhile prognosis. Because of the unreliability of the HPRT assay due to mycoplasma contamination, the assay of the purine utilisation in cultured cells and their selection could give a better and reliable assessment of the inherent mutation. These specific parameters have therefore also been assayed in cultured amniotic and chorionic villi cells to standardise the culture conditions and the laboratory norm values for their potential use in the diagnosis and prenatal monitoring.

196

MYOADENYLATE DEAMINASE DEFICIENCY AND McARDLE'S DISEASE: PLASMA ADENOSINE, INOSINE AND HYPOXANTHINE AFTER ISCHEMIC FOREARM EXERCISE.

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Plasma adenosine, inosine and hypoxanthine concentrations were assayed in 7 controls, 5 myoadenylate deaminase deficient (MADD) patients and 6 McArdle patients before and after ischemic forearm exercise. The MADD patients showed a significantly lower increase of plasma inosine and hypoxanthine following exercise as compared to the controls, indicating diminished adenine nucleotide catabolism in the exercising MADD muscle. In the McArdle patients the increase in plasma inosine and hypoxanthine after exercise did not differ significantly from the values measured in the controls. The plasma adenosine increase was very low in all test groups and there were no significant differences.

197

SYNTHESIS AND TURNOVER OF PURINE NUCLEOSIDE
PHOSPHORYLASE IN HUMAN LYMPHOCYTES
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The synthesis of purine nucleoside phosphorylase (PNP) was examined during phytohemagglutinin stimulated T lymphocyte transformation using a polyclonal rabbit antibody to human erythrocyte PNP. Lymphocytes were pulsed at various times for 3 hours with [³⁵S]methionine and immunoprecipitated lysates were analyzed by SDS-polyacrylamide gel electrophoresis. The labelled 32,500 molecular weight species corresponding to PNP were cut out of the dried gels and counted. PNP synthesis increased greater than 10-fold during the first 12 hours of transformation and continued to a maximum of 30-fold. The rate of synthesis of PNP relative to total protein was 0.04% in unstimulated T lymphocytes and increased to 0.18% 12 hours after stimulation and remained constant at 0.16-0.19% between 40 and 60 hours. These studies identify a 4-5 fold preferential synthesis of PNP during the early stages of T lymphocyte transformation.

In other studies with the B lymphoblast WI-L2, proteins were labelled with [³H]leucine and during further culture in non-radioactive medium the half-lives for total protein and PNP were found to be 14.5 and 14.1 hours respectively.

Supported by the Medical Research Council of Canada Grant MT-6376.

198

ELEVATED ADENOSINE LEVELS IN NEWBORNS. Alf Sollevi,
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Hypoxia enhance tissue adenosine formation during experimental conditions. This study examines the levels of adenosine and its metabolites in umbilical plasma from newborns (15-45 sec after vaginal delivery), a clinical condition known to have low oxygen levels. The study was approved by the Ethics Committee. Adenosine, inosine, hypoxanthine (by HPLC, 1), blood gases and catecholamines (NA and A, by HPLC, 2) were determined in arterial and venous umbilical blood from 30 normal deliveries. pH was 7.25±0.06, Base excess -5.7±2.8 and arterial PO₂ 2.0±0.6 kPa. Plasma NA (5.3-374 nM) and A (0.1-16.9 nM) showed marked elevation. Arterial plasma adenosine 0.61±0.09 μM and inosine 0.67±0.07 μM was similar in venous plasma, corresponding to 4 times higher levels than in adults (1, 3). Arterial hypoxanthine was 0.9 μM higher than the vein (p<0.01). This is the first clinical study demonstrating elevated adenosine and inosine levels during hypoxia and metabolic stress. It is proposed that adenosine may serve as a vascular and metabolic modulator in neonatals.

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