Proc the 2nd Joint Annual Conference of Biomedical Science, Taipei, 1987,p.125.

089. DETERMINATION OF PTERINS IN AMNIOTIC FLUID BY HPLC AND ITS APPLICATION FOR PRENATAL DIAGNOSIS OF ATYPICAL PHENYLKETONURIA CAUSED BY TETRAHYDROBIOPTERIN DEFICIENCY. K.-J. Hsiao, T.-T. Liu*, S.-J. Wu, Z.-L. Yang[1] and M.-L. Yang[1]. Dept. of Medical Research and Obst. & Gyn.[1], Veterans General Hospital; Taipei, Taiwan, R.O.C.

Atypical Phenylketonuria (PKU) due to tetrahydrobiopterin (BH_4) deficiencies may be caused by deficient activity of dihydrobiopterin synthetase (DHBS). Urinary pterin analysis by HPLC is a good diagnostic aid to differentiate variant forms of PKU and to detect the heterozygotes of DHBS deficiency. The determination of pterins in amniotic fluid by HPLC was studied for prenatal diagnosis of PKU caused by BH_4 deficiency. Pterins in amniotic fluid (16-20 weeks of gestation) were oxidized by manganese dioxide or 0.1% iodine at pH 1. After ultrafiltration, the separation of total biopterin (B), neopterin (N) was accomplished by reverse phase (C-18) HPLC and were detected by fluorescence. The reference ranges (n=13) of B (pmol/mL), N (pmol/mL) and total biopterin ratio (B%=B/(B+N)%) in amniotic fluid were 9.8-24.3, 26.1-48.0 and 16.7-36.9, respectively. The analysis of pterins (B=13.4, N=130.3, B%=9.3) in amniotic fluid taken from a pregnancy (19th week) at risk of DHBS deficiency indicate that the male fetus might be a heterozygote. A healthy boy was born prematurely (36th week). Blood phenylalanine level was normal at the ages of 5 days, 2 months and 3 months. The diagnosis was confirmed by urinary pterin profile at the age of 3 months. The results indicate that analysis of pterins in amniotic fluid by HPLC is a good aid for prenatal diagnosis of PKU caused by BH_4 deficiency.

(20)

090. DETERMINATION OF PORPHOBILINOGEN DEAMINASE ACTIVITY IN ERYTHROCYTE FOR THE DIAGNOSIS OF ACUTE INTERMITTENT PORPHYRIA. K.-J. Hsiao, F.-Y. Lee[1], W.-J. Chang, S.-J. Wu* Departments of Medical Research and Internal Medicine[1], Veterans General Hospital, Taipei, Taiwan, R.O.C.

Actue intermittent porphyria (AIP) is an autosomal dominant disorder, in which the basic genetic defect is a deficiency of porphobilinogen (PBG) deaminase. Identification of affected persons and the AIP trait is important for preventive management, because acute episodes may be provoked readily by certain drugs, infections, or steroids. The erythrocyte PBG deaminase activity was determined at 37°C and pH 8.2 by using porphobilinogen as the substrate. The reaction was stopped after 90 min incubation by addition of 12.5 g/dl TCA solution. After centrifugation, the fluorescences (Ex.405nm and Em.596nm) of the supernatant were measured with coproporphrin as the standard. The within-run and between-run imprecision of the assay were 3.8-4.4% (C.V.). The erythrocyte PBG deaminase reference range was estimated to be 30.0-73.7 nmol/hr/ml RBC in Chinese adults (n=133). The pH optimal and Km were 8.2 and 14.0 uM, respectively, for normal controls, one case (female, 32 y/o) presented with unexplained abdominal pain was suspected as AIP clinically. The Watson-Schwartz test of her urine was positive. Her erythrocyte PBG deaminase activity was 23.8 nmol/hr/ml RBC, which was about 50% of the mean of the normal controls. Nine of 13 her family members has an activity below 30.0 nmol/hr/ml RBC. These results indicate that this assay of erythrocyte PBG deaminase could be used for diagnosis of AIP and may be applied to detect AIP trait in the high risk family for genetic consulting.

(21)