77

IDENTIFICATION AND CHARACTERIZATION OF THE HEPATIC TAURINE (TR) TRANSPORTER IN THE RAT. S. Maisel, J. Bucuvalas, C. Schmidt, and R. Dumaswala

Division of Pediatric Gastroenterology and Nutrition, Children's Hospital Research Foundation, Cincinnati, Ohio.

Our objective was to identify and characterize the Na:TR co-transporter on the rat hepatic basolateral membrane (BLM). TR transport by highly purified hepatic BLM prepared from 14d and adult rats was inhibited 40-50% following exposure to the sulf-hydryl (SH) modifying reagent, N-ethylmaleimide (NEM) (250μM). Preincubation of BLM with TR (100μM) prior to exposure to NEM protected TR transport activity. In contrast, phenyl glyoxal (100-250μM) and phenylisothiocyanate (500μM), reagents which modify arginine and lysine amino groups respectively, did not inhibit Na-dependent TR transport. BLM from 14d and adult rats was analyzed by SDS-PAGE to determine if increased TR transport activity in suckling compared to adult rats is due to quantitative differences in the carrier protein. Increased band densities were noted at apparent MW of 72, 38 and 36 kDa in 14d compared to adults. Conclusion: A SH group at or near the TR binding site is essential for Na:TR cotransport by rat hepatic BLM. Preliminary work by our group has demonstrated a human placental 72 kDa protein with 34 and 38 kDa subunits associated with Na:TR cotransport. The increased expression of proteins of similar MW in suckling rat liver suggests that they may be components of the putative TR transporter. Increased ontogenic expression in combination with labeling of a substrate protectable SH group will enable us to specifically identify the hepatic Na:TR transporter.

78

MONOETHYLGLYCINEXYLIDIDE FORMATION: A NOVEL QUANTITATIVE LIVER FUNCTION TEST. HH A-Kader, DA Gremse, D Berkowitz, TJ Schroeder, WF Balistreri. Children's Hospital Cincinnati OH

Lidocaine is deethylated in the liver to form Monoethylglycinexylidide (MEGX) which can be measured by Fluorescent Polarization Immunoassay. In order to determine if MEGX formation can be used as a quantitative liver function test, the MEGX concentration was measured in serum 15 min after lidocaine injection (1mg/kg IV) in 15 controls and in 40 patients with chronic liver disease (CLD). Salivary MEGX concentration was measured (at 15 and 30 min) in 10 controls and in 8 pts with CLD. Serum MEGX concentration was decreased in CLD compared to controls (38±5 vs 106±9 μg/L, X±SE, res-pectively) in an inverse proportion to severity of CLD: low risk for death (J Ped 111:479) 48±6 (n=28); medium risk 27±1(n=2); high risk 13±4 (n=10). Salivary MEGX concentration was decreased in CLD compared to controls (77±19 at 15 min and 155±31 at 30 min vs 103± 7 and 346± 74, respectively , p<0.5). Serum MEGX levels at 15 min correlated with salivary levels at 15 and 30 min (r=0.43 and 0.34 respectively, p<0.05). Conclusion: The serum concentration of MEGX is decreased in pts with CLD in an inverse proportion to disease severity. Salivary MEGX levels, which correlate with serum levels, are also decreased in pts with CLD. Assessment of MEGX after lidocaine injection may be a useful quantitative liver function test.

79

GASTROINTESTINAL MANIFESTATIONS OF CF PATIENTS CARRYING THE ΔF508 MUTATION. P Durie, L-C Tsui, M Corey, E Kerem, B Kerem, J Rommens, H Levison. The Hospital for Sick Children, Toronto, Canada.

The gene responsible for CF has been cloned. In the majority of CF patients a 3 base pair deletion results in the loss of a phenylalanine residue at the 508 position (ΔF) of the putative gene. At least 7 other mutations are predicted, which may account for the variable clinical phenotypes. Our previous genetic analysis showed that patients with or without pancreatic function (PS and PI) possess different mutant alleles. We have further characterized the relationships between the CF gene mutations and various gastrointestinal manifestations in 261 CF patients (Table).

	ΔF/ΔF(50%)		ΔF/Other(41%)		Other/Other(9%)	
	PI	PS	PI	PS	PI	PS
# Patients	133	0	70	33	10	15
# Liver palp	14	–	9	4	0	0
# Abn SGOT	28	–	14	5	0	1
# Abn Alk Phos	17	–	13	2	0	0
# Mec Ileus	19	–	9	0	4	0
Mean Wt Centile	41	–	45	62	39	78

The prevalence of the ΔF allele is ~70%. In addition, regression analysis of FEV1 revealed significantly better lung function in PS patients with a single ΔF or no ΔF chromosome. Mortalities within 9 years were PI patients with ΔF/ΔF (n=9) and ΔF/other (n=7). These data also suggest: (1) Phenotypic variations of intestinal, nutritional and pulmonary disease in CF correlate with the nature of mutations in the gene; (2) ΔF/ΔF confers a severe phenotype; (3) additional mild and severe mutations exist.

80

ILEAL RESECTION/DYSFUNCTION PREDISPOSES TO LITHOGENIC BILE AFTER PUBERTY. JE Heubi, NC O'Connell, KDR Setchell, Children's Hospital Research Foundation, Cincinnati, OH 45229, USA.

Adults with ileal disease or resection have biliary cholesterol supersaturation and are therefore prone to cholesterol cholelithiasis. Our previous studies have shown that bile from children with ileal resection/disease (IR/D) was not supersaturated with cholesterol (Gastroenterol 1982;82:1295). The present study was designed to test the hypothesis that infants with IR/D will develop biliary cholesterol supersaturation after puberty presumably because of the influence of sex hormones in biliary secretion. Five sexually mature subjects (3M, 2F, age 16-19 yr) who had been previously investigated in childhood (ages 4-9 yrs) were studied. Ultrasound examination of the gallbladder performed using real-time sonography revealed gallstones on one subject. Duodenal bile samples, obtained after an overnight fast, were examined for bile acids (BA), phospholipid (PL) and cholesterol (XOL), and the molar fraction (M%) and lithogenic index (LI) calculated. Comparisons are shown between subjects prior to puberty (C), post puberty (PP) and 20 young adult controls [x±SEM, $*p < .001$ or $^{+}p < .005$ vs. Ileal disease (PP)]:

	XOL (M%)	PL (M%)	BA (M%)	LI
Ileal Disease (PP)	22.0±4.8	19.9±1.6	58.1±3.9	3.1±0.7
Ileal Disease (C)	3.1±2^{+}	17.1±1.8	79.4±2.0^{+}	0.6±1^{+}
Adults	5.7±.5*	16.5±1.6	78.2±1.9^{+}	1.1±.1*

Conculsions: Children with ileal resection/dysfunction are not at risk for cholesterol gallstone formation during childhood; however, biliary cholesterol supersaturation after puberty predispose them to cholesterol cholelithiasis.

81

SUCCESS OF LIVER TRANSPLANTATION IN FULMINANT HEPATIC FAILURE (FHF) OF CHILDHOOD. Vargas JH, McDiarmid S, Brill J, Harrison R, Semnoni H, Busuttil, Ament ME. Department of Pediatric and Surgery, University of California, Los Angeles, USA.

Prior to development of orthotopic liver transplantation (OLT), mortality of FHF in pediatric patients was 80-85%. OLT has been a major factor in reducing mortality from this condition. Thirty pediatric patients, median age 4 yrs (range 3 mo. to 17 yrs.) presented with FHF from hepatitis of unkown etiology (NANB) 20; Hepatitis A virus (HAV) 4; Hepatitis B virus (HBV) 1; drug toxicity (acetaminophen and erythromycin) 2; metabolic disease (Wilson's disease and neonatal hemochromatosis) 2, and Chronic Active Hepatitis (CAH) 1. 19 were females and 5 were <1 year of age. All presented with hyperbilirubinemia, coagulopathy, and hypoproteinemia. 16/30 had ascites, 15/30 were in hepatic coma stage I-II, 12/30 in stage III and 2/30 in stage IV. 19/30 received one or more exchange transfusions with transient improvement of coagulation parameters. Twelve were successfully transplanted and 14 (46%) died. One patient with HAV died post-operatively with irreversible neurological damage but normal graft function; transplantation was done while in stage IV coma. 3 patients survived without transplantation, (1, HBV and 2, NANB) and only one showed moderate liver dysfunction with severe architectural damage and regeneration histologically. The presence of ascites was a good clinical predictor for fatal outcome. OLT substantially reduces mortality from FHF and should be considered in any patient who progresses from Stage I.

82

SUCCESSFUL TREATMENT OF PRURITUS IN SEVERE CHOLESTASIS SYNDROMES WITH RIFAMPIN. Vargas JH, Ament ME. Department of Pediatrics, Div. of Gastroenterology and Nutrition. University of California, Los Angeles. USA

Pruritus is, without a doubt, the most frequent and uncomfortable symptom of severe cholestasis. Its etiology and physiopathology are still unclear but are believed linked to bile salts. Therapy has traditionally consisted of administration of phenobarbitol, antihistamines, binding resins, plasmapheresis and emollients without reduction in symptoms. We report results of therapy with rifampin (an agent that reduces bile acid uptake) in a series of pediatric patients with severe cholestasis syndromes who had failed to respond to classic treatments (cholestyramine and phenobarbitol). Twenty one children, 11 males in a group representative of all pediatric ages, (0-1 yr=5, 1-3 yrs=5, 3-6 yrs=5, and 6-17 yrs=6), with severe cholestasis and pruritus secondary to Biliary Atresia (11), Familial Cholestasis (3), Biliary Hypoplasia (2), Neonatal Hepatitis (2), Cystic Fibrosis (1), Hemangioendothelioma post-extensive resection (1) and Cholestasis of unknown etiology (1), were given a course of 5 mg/Kg/ day of Rifampin, orally. A net improvement, with disappearance of pruritus within 7 days of treatment, was observed in 50% of the patients. The rest of the patients improved, with decreased itch-severity scales (subjective parameters by parent or child), to the point of total control in all cases except one, within a range of 1-28 weeks of therapy. No change or deterioration of liver chemistries or synthetic function, blood counts, or cholilglycine levels were observed during a period from 4 weeks to 2 years of therapy. No complications were recorded and the medication was well tolerated. Rifampin is a safe and effective drug to treat severe pruritus in cholestatic infants and children.