

- 554 PRECURSOR FORMS OF RAT SMALL INTESTINAL LACTASE. Ricardo O. Castillo, Linda K. Kwong, Philip Sunshine, Richard Quan, Gary M. Gray, and Kenneth K. Tsuboi. Stanford University School of Medicine, Departments of Pediatrics and Medicine, Stanford, CA.

High molecular weight precursor forms of various intestinal brush border enzymes have been demonstrated by pulse-labelling of intestinal explants maintained in culture. In the present study we have examined *in vivo* precursor forms of rat intestinal lactase. Fasted infant rats were injected (IP) with [<sup>35</sup>S]-methionine and killed after 0.5, 2, 4, and 18 H. The short fast was necessary to maximize incorporation of label. Lactase forms were isolated from Triton X-100 extracts of the intestinal homogenates by immunoprecipitation techniques using highly selective antisera. The isolated enzyme forms were separated by SDS-electrophoresis and examined by fluorography. Labelling within 30 min occurred in precursor forms of apparent mol. wts of 170, 125, and 90 Kd. A further precursor form of 200 Kd appeared within 2 H. All labelled precursor forms were no longer evident by 4 H. Apparent labelled brush border forms of lactase of 220, 130, and 95 Kd were evident by 2 H and were still present after 18 H. Using similar methodology, we have examined precursors of sucrase-isomaltase, with confirmation of existence of mature and immature precursor forms of 250 and 240 Kd respectively. Demonstration of lactase precursor forms determined *in vivo* provides confirmation of similar *in vitro* findings. These studies are consistent with the initial intracellular synthesis of high molecular weight precursor forms of lactase with subsequent transfer to brush border sites.

- † 555 DIETARY NEEDS IN BONE MINERALIZATION OF CHILDREN. Gary M. Chan, Gurmail Gill, Renae McInnes. Department of Pediatrics, University of Utah, Salt Lake City, UT.

There has been very little data about the dietary factors which may affect children's bone mineral status. We prospectively studied 88 white healthy children aged 3 to 16 years (mean age 8.7 years). There were 46 boys and 42 girls in our study. During a 3 month study, weight and height were recorded, and at least two 24-48 hour dietary histories were averaged for intakes of calcium, phosphate, magnesium, sodium, protein, and total calories. Bone mineral status was measured by photon absorptiometry on the child's distal non-dominant radius. Blood was drawn for serum calcium, phosphorus, magnesium, alkaline phosphatase, and 25-OH vitamin D. We found that the child's age ( $r=0.84$ ,  $p<0.001$ ), weight ( $r=0.86$ ,  $p<0.001$ ), and height ( $r=0.84$ ,  $p<0.001$ ) were correlated to bone mineral status. Dietary calcium intakes were correlated to the children's bone mineral status ( $r=0.31$ ,  $p<0.01$ ). Children ingesting more than 1000 mg calcium daily had higher bone mineral status (8%) than those ingesting less ( $t$  test,  $p<0.03$ ). Dietary protein ( $r=0.25$ ,  $p<0.03$ ) was also associated to bone mineral status; children ingesting more than 50 g protein had higher bone mineral status (12%) than those taking less ( $p<0.01$ ). We found no significant relation between bone mineral status and dietary phosphate, magnesium, sodium, and total calories. All serum determinations were within normal ranges for children and none correlated with the child's dietary intakes or bone mineral status. In summary, our study has shown that age, weight, height, calcium and protein intakes have important effects on children's bone mineral status.

- 556 IN UTERO CORTISONE ALTERS THE INTESTINAL SIALYL- AND FUCOSYLTRANSFERASE ACTIVITIES IN NEWBORN RATS. Shu-heh W. Chu, Ilene Ely and W. Allan Walker. Harvard Med. Sch., Children's Hosp. and Mass. Gen. Hosp., Dept. Pediatrics, Boston, MA 02115

Changes in intestinal surface carbohydrates, such as sialic acid and fucose, may influence the binding of membrane receptors to bacteria/toxins or antigens. We have recently reported (Biochim. Biophys. Acta 883, 496, 1986) that the activities of intestinal sialyltransferase (ST) and fucosyltransferase (FT) are reciprocally related and modulated by cortisone during postnatal development. These glycosyltransferase changes may provide an enzyme basis for the shift from sialylation to fucosylation of the intestinal surface during maturation. To further study whether prenatal treatment of cortisone may affect the activities of these two enzymes, cortisone (20 mg/100 g body weight) was injected *i.p.* in pregnant rats daily beginning at 17 days of gestation. The newborn rats (<24 hr old) were sacrificed and ST and FT were assayed in the membranous (105,000 x g pellet) fraction of small intestine, using asialofetuin as an exogenous acceptor. The results show that the activity of ST was decreased by 50% in the newborn rat exposed to cortisone in utero, while the activity of FT in the cortisone-treated group was induced 2-3 fold compared to the control. This study indicates that cortisone seems able to act as a signal in utero to modulate the expression of intestinal ST and FT and suggests that the cortisone-induced changes in intestinal glycosyltransferases may play a role in the maturation of the mucosal barrier in neonates. (Support by NIH grant DK37521).

- 557 IMMUNOGENICITY OF CAMPYLOBACTER PYLORIDES (CP) OUTER MEMBRANE PROTEINS. Steven J. Czinn, Vera F. Hupertz, Howard Carr, Pauline Labrozzi and Stephen C. Aronoff. (Spon. by William T. Speck) Case Western Reserve University School of Medicine, Rainbow Babies and Childrens Hospital, Department of Pediatrics, Cleveland, Ohio.

Studies in a variety of gram negative bacteria have shown that outer membrane proteins play a major role in the adherence of the pathogen to the host epithelium, in resistance to phagocytic killing mechanisms, and as potential antigens. CP is a gram negative rod associated with a subset of pediatric and adult patients with gastritis and peptic ulcer disease. It has been shown that CP contains three immunogenic proteins with approximate molecular weights of 14-21 KD, 33KD and 55-70 KD (J. Med. Microbiol 22:57, 1986). The current study explored the hypothesis that these antigenic proteins of CP are contained within the outer membrane. Outer membrane proteins from a pediatric isolate of CP were extracted and purified using ultracentrifugation and sequential washings with sodium laurel sarcosinate. These were analyzed by SDS PAGE and shown to contain six major proteins with relative molecular weights of 66KD, 40KD, 31KD, 28KD, 27KD and 16KD. This is in marked contrast to *C. jejuni* which contains two major outer membrane proteins with molecular weights of 63KD and 45KD. Mice were intraperitoneally immunized on Day 1 and 14 with sublethal ( $10^3$  CFU) doses of CP and sacrificed on Day 17. The serum was examined for antibody production specific for CP outer membrane proteins by immunoblot (Western blot) analysis. There appeared to be three major reactive protein bands of relative molecular weights of 66KD, 27KD and 16KD. These results suggest that the three immunogenic proteins of CP are present in the outer membrane. The role of these CP-specific antibodies, [specific for CP], in protecting the host from chronic CP infection is now being investigated.

- 558 PLASMA AMINO ACID CONCENTRATIONS IN CHILDREN RECEIVING LONG TERM TOTAL PARENTERAL NUTRITION. Anders K. Dahlstrom, Marvin E. Ament, Stuart A. Laidlaw, Joel D. Koppel. Pediatric Departments, UCLA Medical Center, Los Angeles, Karolinska Institute, Stockholm, Harbor Medical Center, Torrance.

Plasma amino acid (AA) concentrations were analyzed using ion exchange chromatography in 15 children aged 4-65 months who received total parenteral nutrition (TPN) as their only source of nutrition (7 pts, Gp I) or who ingested 30-70% of their total energy requirements orally and received the remainder intravenously (8 pts, Gp II). The pts in Gps I and II received TPN for 25.6±14.4 SD and 25.7±19.1 months, respectively. All pts in Gp I and 4 in Gp II had short bowel syndrome (SBS); 4 in Gp II had pseudoobstruction syndrome. Gp I had decreased plasma concentrations of total essential AA, branched chain AA, the semi-essential AA (cysteine and tyrosine) and the urea cycle AA. Gp II children had notably decreased plasma concentrations of branched chain AA and tyrosine. Both gps had significantly increased concentrations of glycine and decreased concentrations of taurine. The children with cholestatic liver disease had the lowest plasma concentrations of the branched chain AA. The plasma concentrations of the individual AA in the children reflected the composition of the AA given in the TPN solutions (Travasol® 8.5%), indicating that children receiving long term TPN will develop abnormalities in their plasma AA concentrations despite receiving adequate amounts of AA in their TPN solutions. This condition could be explained by the unbalanced AA solutions the children received, but other factors that could interfere with their AA metabolism were significant liver disease and SBS with minimal enteral intake.

- 559 LACK OF FUNCTIONAL IMMATURITY OF THE GLUCOSE TRANSPORT SYSTEM FOLLOWING SMALL BOWEL RESECTION IN THE RAT by Steven H. Erdman, Jung H. Y. Park, Carter J. Grandjean, Michael H. Hart and Jon A. Vanderhoof. Department of Pediatrics, University of Nebraska College of Medicine, Omaha, Nebraska.

Despite the mucosal hyperplasia that follows massive small bowel resection, changes in absorptive function are thought not to parallel changes in mucosal mass. This is described as "functional immaturity" of the new enterocytes. We approximated unidirectional glucose uptake using an inert glucose analogue, 3-O-Methylglucose (3MG). The study included 24 160 gram male Sprague-Dawley rats, half of which underwent 80% jejunoileal resection, the remainder being sham-operated. Six animals of each group were studied at post-op day 3 and six at post-op day 10. The unidirectional 3MG uptake was determined utilizing intestinal rings incubated for 3 minutes in KRb buffer at various concentrations of 3MG. The J value in resected animals is shown below in both duodenum and ileum at 10 mM concentration. Value x1000±SEM

	DUODENUM	ILEUM
Day 3	9.00±0.78	7.60±0.94
Day 10	8.55±1.00	7.27±0.86

Despite a marked increase in mucosal mass, there was no significant difference in 3MG uptake between day 3 and day 10. No functional immaturity of the enterocytes at day 10 was apparent, suggesting that the marked increase in mucosal mass following resection is accompanied by the corresponding increase in glucose absorption.